Poster Session 2

Acid-base and ion transport

Su001 ❋ ♦ SPAK KNOCKOUT MICE EXPRESS GITELMAN'S SYNDROME AND IMPAIRED VASOCONSTRICTION

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Introduction and Aims: SPAK [STE20 (sterile 20)/SPS1-related proline/alanine-rich kinase] interacts with WNK [With-No-Lysine (K)] kinases 1/4 to regulate Na^+-(K^+)-(2)Cl^- contransporter [N(K)CC]. Polymorphisms in the STK39 gene encoding SPAK have been identified as hypertension susceptibility genes in humans. Mutations in WNK1/4 and N(K)CC can cause hyper/hypotesnion and dyskalemia in humans.To elucidate the physiologic role of SPAK *in vivo*, we generated SPAK-null mice by disrupting *Stk39* gene.
Methods: Blood pressure, aortic contractility, blood and urine electrolytes and biochemistries, and relevant protein expression in the kidneys and aortic tissues were examined.
Results: $SPAK^{+/-}$ mice exhibited hypotension without significant electrolyte abnormalities while the $SPAK^{-/-}$ mice not only exhibited hypotension but also recapitulated Gitelman's syndrome (GS) with hypokalemia, hypomagnesemia, and hypocalciuria.

Blood Pressure (BP), blood and urine biochemistries in SPAK-null mice

SPAK genotype (n)	+/+ (10)	+/– (16)	–/– (10)
BP (mmHg)			
Systolic	109.5±4.5	95.6±3.6*	93.2±4.1**
Diastolic	76.2±5.6	62.1±4.6*	59.3±6.2**
Mean	87.2±5.6	74.0±4.5*	71.2±3.5**
Weight (gram)	23.8±2.5	23.6±3.3	23.6±3.3
Plasma			
Aldosterone (pg/ml)	570±158	707±226	1006±394**
Na+ (mmol/l)	156±3	158±3	157±3
K+ (mmol/l)	4.2±0.3	4.0±0.3	3.6±0.2**
Cl- (mmol/l)	114±2	113±3	109±2**
Total Ca2+ (mg/dl)	9.6±0.2	9.5±0.4	9.7±0.3
Mg2+ (mg/dl)	2.5±0.1	2.3±0.2	1.9±0.2**
Cr (mg/dl)	0.16±0.05	0.17±0.04	0.16±0.04
Urine (ml/day)	1.5±0.4	1.5±0.7	1.6±0.4
Na+ (μmol/day)	97±21	85±25	89±16
K+ (μmol/day)	125±8	124±25	142±22
Cl- (μmol/day)	85±10	92±11	89±15
FENa (%)	0.21±0.07	0.23±0.08	0.22±0.09
FEK (%)	15.4±3.7	17.6±6.8	22.6±4.2**
FEMg (%)	4.8±0.9	5.2±0.8	6.1±0.7**
Ca2+/Cr (mg/mg)	0.19±0.03	0.17±0.05	0.12±0.02**

Cr: creatinine; FENa, FEK and FEMg represent the fractional excretion of Na+, K+ and Mg2+, respectively. **denotes p<0.05 when —/- vs. +/+; *denotes p<0.05 when +/– vs. +/+.

In the kidney tissues of $SPAK^{-/-}$ mice, the total and phosphor (p-)NCC expression was markedly decreased but that of p-OSR1, total NKCC2 and p-NKCC2 was significantly increased. In aortic tissues, both $SPAK^{+/-}$ and $SPAK^{-/-}$ mice had impaired response to phenylephrine (a selective α_1-adrenergic agonist) and bumentanide (a NKCC1 inhibitor). While total NKCC1 expression was increased, p-NKCC1 was decreased.
Conclusions: SPAK-null mice have defects of NCC in kidneys and NKCC1 in blood vessels, leading to hypotension through renal salt wasting and vasodilatation. SPAK may be a promising target for anti-hypertensive therapy.

Su002 ★ CROSS-REGULATION BETWEEN NATRIURETIC PEPTIDE AND RENAL DOPAMINERGIC SYSTEMS DYSFUNCTION IN PUROMYCIN AMINONUCLEOSIDE-INDUCED NEPHROTIC SYNDROME

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Introduction and Aims: The mechanism responsible for the primary sodium retention in nephrotic syndrome (NS) is suggested to be the combination of a blunted natriuretic response to atrial natriuretic peptide (ANP) and an enhanced Na^+,K^+-ATPase activity in the cortical collecting duct. The ANP resistance appears to result from the activation of the phosphodiesterase type 5 (PDE5) responsible for the catabolism of cyclic guanosine monophosphate (cGMP), the second messenger of ANP. Currently, it is well recognized a synergistic correlation between renal dopaminergic (RDS) and natriuretic peptide (NPS) systems. In the animal model of puromycin aminonucleoside-induced NS (PAN-NS) was reported a blunted RDS evidenced by decreased urinary DA output and insensitivity of natriuresis to dopamine 1 receptor (D1R) agonist. The aim of the present study was to evaluate the interaction between the NPS dysfunction and blunted RDS activity in PAN-NS rat model by assessing the effects of zaprinast (PDE5 inhibitor) and sch-23390 (D1R antagonist) intravenous (iv) perfusion during volume expansion (VE) on urinary sodium and cGMP excretion.
Methods: Male Sprague-Dawley rats (Harlan, Spain) weighing 150g received a intraperitoneal injection of PAN (150 mg/kg, n=15) or vehicle (NaCl 0,9%, n=9). Seven days after the injection of PAN or vehicle (control group, Ct), the animals were submitted to VE and iv perfusion of zaprinast (Z, 100mg/kg bw/min), zaprinast plus sch-23390 (S, 30mg/kg bw/min) or the vehicle (V, 0.9% NaCl) creating six experimental groups: CtV; CtZ, CtZS, PANV, PANZ and PANZS. The infusions started at a rate of 5ml/kg bw/h for 120 min (basal period), followed by VE period (50 ml/kg bw/30 min, 5% of rat's weight) for 30 min; thereafter, the infusion was reduced to basal rate for another 30 min (recovery period, R-EV). Quantification of cGMP urinary levels were determined with a commercially EIA kit (R&D Systems).
Results: During all perfusion periods, PANV rats presented reduced urinary sodium and cGMP excretion in comparison to CtV animals. After Z perfusion, PANZ-rats presented a significantly increase in urinary sodium and cGMP excretion in comparison to PANV rats whereas the addition of S to Z, attenuated the natriuresis and the increase in the urinary cGMP excretion observed in PANZ group to levels similar of those observed in PANV group. These changes were more marked in VE and R-VE periods. The urinary sodium and cGMP excretion in Ct rats were not altered after the perfusion of Z or Z+S during all protocol periods.
Conclusions: Our results show that, in PAN-NS, the natriuresis induced by NPS requires D1R activation. This provides evidence for a close interaction between NPS and RDS activities in NS.

Su003 HIGH OSMOTIC STRESS PROMOTES THE UPREGULATION AND PHOSPHORYLATION OF ZONULA OCCLUDENS-1

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Introduction and Aims: Tight junction molecules (TJ) form a barrier between adjacent epithelial cells and thus mediate the cells' ability to develop membranes that constitute the boundaries of different compartments within the body. Membranes with selective ion and water passage are especially important for the electrolyte and water homeostasis in the kidney. Due to their role in the urinary concentration process, renal medullary cells are exposed to an extraordinarily high osmotic stress.Therefore, we were interested in the question of how mouse inner medullary collecting duct

cells (mIMCD3) manage to maintain their cell-cell contacts, despite high osmolality-induced cell shrinkage.
Results: We found that the Zo-1, MUPP1 and cortactin mRNA expression was upregulated in an osmolality-dependent manner. Using western blot analysis, immunoprecipitation and immunofluorescence, we show that the Zo-1 protein is upregulated, hyperphosphorylated and linked to the actin cytoskeleton in response to high osmotic stress. In mIMCD3 cells, which were exposed to high osmolality, the rearrangement of the actin cytoskeleton resulted in the colocalization of actin fibres with Zo-1. Urea, which generates high osmolality, but no transcellular gradient, did not induce changes in Zo-1 protein expression or actin rearrangement.
Conclusions: This data indicates that Zo-1 is a response protein to high inner medullary osmolality and that extracellular stressors can promote Zo-1 protein expression, tyrosine phosphorylation and cytoskeleton association.

Su004 PPAR-γ AMELIORATES AMPHOTERICIN B-INDUCED TUBULAR EFFECTS

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Introduction and Aims: Amphotericin B (AmB) nephrotoxicity is characterized by reduction in glomerular filtration rate (GRF) and tubular toxicity manifested by urinary waste of potassium and magnesium, renal tubular acidosis and reduced urinary concentration capacity. Rosiglitazone (RSG) is an agonist of PPAR-γ with renal tubular effects, increasing the expression of some membrane transporters.
Methods: This study evaluates the effects of RSG in AmB-induced nephrotoxicity. Rats were fed with normal diet supplemented or not with RSG (92mg/kg of food) and treated with AmB (5mg/kg BW i.p.) for 4 days. Four groups were studied: control (normal diet and saline only); AmB (normal diet and Amb i.p.); RSG (diet with RSG and saline) and AmB + RSG (diet with RSG and AmB i.p.). In the last day of treatment the rats were housed in metabolic cage for 24h urine and blood sample collection. Clearance creatinine (ClCr), blood gas analysis, urinary pH and serum/urinary Na, Cl, K, Mg, osmolality were measured. Transtubular potassium gradient (TTKG) was calculated.
Results:

Biochemical parameters

	Urine output (mL/day)	ClCr (mL/min/kg)	serum HCO3 (mEq/L)	Serum K (mEq/L)	Serum Mg (mg/dL)
Control	10,6±1,5	1,20±0,09	29,0±0,7	4,3±0,2	1,85±0,1
RSG	9,2±1,3	1,02±0,12	29,6±1,0	4,5±1,0	1,76±0,09
AmB	19,3±2,2a	0,69±0,06b	19,1±1,3c,d	3,2±0,2a	1,43±0,08a
AmB+RSG	11,7±1,0	0,87±0,11	25,1±1,5	4,5±0,3	1,9±0.05

Data are shown as mean±SEM. a: p<0,01 vs. other groups; b: p=0,02 vs. control; c: p<0,001 vs. control and RSG; d: p<0,01 vs. AmB+RSG
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Urinary Parameters

	Urinary pH	Urinary Osm (mOsm/kgH2O)	TTKG	UMgV (mg/day)
Control	6,67±0,13	814±51	8,9±0,3	0,76±0,07
RSG	6,86±0,17	838±44	7,6±0,3	0,82±0,11
AmB	7,05±0,07d	474±45e	13,6±0,4e	1,66±0,25f,g
AmB + RSG	6,4±0,14	768±33	8,8±0,5	1,02±0,06

Data are shown as mean±SEM. c: p<0,001 vs. control and RSG; d: p<0,01 vs. AmB + RSG; e: p<0.001 vs. other groups; f: p<0.01 vs. control and RSG; g: p<0.05 vs. AmB + RSG.

Conclusions: Although RSG has no significant effect on AmB-induced GFR reduction, there was a completely protection in all features of tubular toxicity, including renal tubular acidosis just as urinary loss of potassium and magnesium.

Su005 CELLULAR MECHANISMS OF RENIN SECRETION

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Introduction and Aims: Active renin is exclusively secreted into the circulatory system by the renal juxtaglomerular epitheloid cells (JG-cells). The release process of renin vesicles is assumed to be similar to other endocrine cells, although morphological evidence for this is rather limited. It is well established that an increase of cAMP in JG-cells stimulates renin release and that increased Ca2+-concentrations are paralleled by inhibition of release, what stands in contrast to classical secretion processes, in which exocytosis is stimulated by Ca2+. This unusual behaviour of renin cells is known as "calcium paradoxon of renin release." The mechanisms by which cAMP stimulates and Ca2+ inhibits secretion in renin producing cells remain largely obscure. The aim of our work is to obtain more direct information about the cellular events triggering the release of renin. For this purpose we have a) analyzed the structure of renin storage vesicles and b) we have analyzed the the components of the classic exocytosis machinery in JG cell.
Methods: a) The structure of renin storage vesicles was analyzed by 3-dimensional electron-microscopical reconstruction of single JG- cells with corresponding secretory vesicles. Reconstructions of single JG cells were made with 70nm thick serial slices of kidney tissue. Slices were photographed and reconstructed with the AMIRA program.
b) The exocytosis machinery was analyzed by immunohistochemical identification of v-SNAREs, t-SNAREs, munc- and rab-proteins. For positive controls we used tissue of classical exocrine glands (pancreas, adrenal gland, thyroid gland).
Results: We could not yet identify a protein involved in the classic exocytosis process, such as vamp2, 3 and 8, munc 18 or snap25. Notably, we also did not detect synaptotagmins in JG cells. 3-dimensional reconstructions of renin cells revealed no exocytotic events. Most of the renin containing vesicles appeared to be interconneted to mesh-like structures.
Conclusions: The absence of important classical exocytosis proteins in JG cells and particularly the absence of synaptotagmin, which acts as "calcium paradoxon of renin release." in classical endo- and exocrine cells, indicate that the release mechanism of JG-cells is significantly different to mechanisms of classical secretory cells. The mesh-like appearance of renin containing vesicles strengthen this assumption.

Su006 INTERACTION OF PROLYL HYDOXYLASE INHIBITORS WITH ORGANIC ANION TRANSPORTERS

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Introduction and Aims: In response to reduced oxygen supply, mammalian cells activate the transcription factor Hypoxia-Inducible Factor 1 (HIF-1). Target genes for HIF-1 have important roles in many physiological and pathological events such as angiogenesis, vascular remodelling, erythropoiesis, glucose utilization, cell proliferation, and tumor progression. Whereas HIF-1β is constitutively expressed, HIF-1a is rapidly degraded under normoxic conditions by hydroxylation by the prolyl hydroxylases 1, 2, and 3 (PHD1, 2, 3). The activity of all PHDs requires Fe^{2+}, ascorbate, and alpha-ketoglutarate (alpha-KG). Accordingly, analogues of alpha-KG or other dicarboxylates have been used as PHD-inhibitors, which are able to stabilize HIF-1 even in the presence of oxygen. Since PHDs are located within the cell, inhibitors of PHDs have to be translocated across the plasma membrane of the cell to reach their target. Because of their dicarboxylate-like structure, alpha-KG, 2,4-diethyl-pyridine dicarboxylate (2,4-DPD), *N*-oxalylglycine and fumarate, which have been demonstrated to interfere with prolyl hydroxylation, could be candidate substrates of the sodium-dependent dicarboxylate transporter 3 (NaDC3), and/or of the organic anion transporters 1, 3, and 4 (OAT1, 3, 4).
Methods: To test for this hypothesis, either HEK293 cells stably transfected with human OAT1, 3, 4, or NaDC3 were used to test for the impact of alpha-KG, *N*-oxalylglycine, 2,4-DPD or fumarate on these transporters in cis-inhibition and trans-stimulation experiments with the prototypical substrates *p*-aminohippurate (PAH), estrone sulfate (ES) and succinate for OAT1,3, 4 and NaDC3, respectively.
Results: Whereas *N*-oxalylglycine, 2,4-DPD, and fumarate did not interact with OAT3 and OAT4, uptake of labelled PAH by OAT1 was significantly inhibited by alpha-KG, *N*-oxalylglycine and 2,4-DPD with K_is of 1.1, 320 and 40 microM, respectively. Fumarate was not a substrate for OAT1. Fumarate and alpha-KG inhibited succinate uptake by NaDC3 with a K_is of 65 and 46 microM, respectively.

Conclusions: In terms of the central role of HIF in many biological pathways, regulators of HIF are of interest for pharmaceutical approaches. Because OAT1 has a broad tissue distribution, HIF inhibitors with dicarboxylate-like structures can be sufficiently taken up into cells by OAT1 to regulate HIF-1a degradation by the PHDs. Since the distribution of NaDC3 is restricted to the kidneys, the liver, and the brain, the action of fumarate and alpah-KG is limited to PDHs located in cells of these organs.

Su007 GLUT2 IS DOWN-REGULATED TO PRODUCE RENAL GLYCOSURIA IN CYCLOSPORINE-TREATED RATS

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Introduction and Aims: Renal glycosuria has been proposed as an index of cyclosporine nephrotoxicity. Glycosuria may also be derived from glucose intolerance induced by cyclosporine administration. GLUT2 is the main renal glucose transporter upregulated by hyperglycemia, but whether it is affected by cyclosporine administration is not clear.
Methods: Cyclosporine was subcutaneously injected to male Sprague-Dawley rats at a daily dose of 25 mg/kg (n = 6) for 2 weeks (Experiment I) and 7.5 mg/kg (n = 6) for 6 weeks (Experiment II). Biochemical data were obtained from urine and plasma samples, and immunoblot analysis and immunohistochemistry were carried out to see if the renal expression of GLUT2 is altered by cyclosporine treatment.
Results: In Experiment I, cyclosporine treatment caused a remarkable increase in urine volume and a decrease in urine osmolality. In Experiment II, cyclosporine treatment still had an increasing tendency in urine volume but no difference in urine osmolality. However, urinary excretion of glucose was remarkably elevated by cyclosporine administration in both animal experiments. In renal cortical homogenates, the GLUT2 protein abundance was significantly decreased by cyclosporine treatment. The result of GLUT2 immunohistochemistry was compatible with that of immunoblot analysis. Notably, plasma glucose levels at the end of each animal experiment were not affected by cyclosporine administration.
Conclusions: We demonstrate that, in contrast to diabetes mellitus, the expression of GLUT2 protein is decreased by cyclosporine administration in rat kidney. Downregulation of GLUT2 may have a role in producing renal glycosuria in cyclosporine-treated rats.

Su008 REGULATION OF THE SAT1 (SLC26A1) SULFATE/BICARBONATE TRANSPORTER IN KIDNEY DURING METABOLIC ACIDOSIS AND SULFATE LOADING

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Introduction and Aims: The SAT1 anion exchanger (SLC26A1) is expressed on basolateral membranes of the renal proximal tubule cells and the liver and is able to mediate transport of anions such as sulfate, chloride, bicarbonate, or oxalate. In the kidney this transporter has been implicated into the basolateral release of reabsorbed sulfate into blood. We have previously observed in microarray and proteome studies that 2 and 7 days of NH_4Cl-induced metabolic acidosis increased SAT1 mRNA and protein abundance in mouse kidney. The aim of the present study was to investigate the *in vivo* regulation of SAT1 in rat kidney and liver.
Methods: Rats were divided into 4 groups with different diets that were given into the food to dissect the regulation by acidosis or sulfate repletion/depletion. Group 1 received 100 mM NaCl, group 2 100 mM Na_2SO_4, group 3 100 mM NaCl plus NH_4Cl 3%, and group 4 100 mM Na_2SO_4 plus NH_4Cl 3%. All animals were treated for 7 days and placed in metabolic cages at the end of the study for 24 hours. Realtime rt-PCR, Western blotting and immunohistochemistry were performed to study the regulation of SAT1.
Results: Addition of NH_4Cl to the diet induced similar metabolic acidosis in both groups with increased urinary excretion of ammonium as expected. NH_4Cl together with Na_2SO_4 as well as dietary sulfate alone stimulated urinary sulfate excretion. Addition of sulfate to the diet reduced mRNA expression of the renal NaSi (SLC13A1) sulfate transporter but had no effect on renal SAT1 mRNA and protein abundance. NH_4Cl-loading with NaCl reduced NaSi mRNA but did not alter SAT1 mRNA when compared to NaCl or Na_2SO_4 alone. Similarly, combined Na_2SO_4 and NH_4Cl loading decreased NaSi mRNA but again did not affect renal SAT1 mRNA. In kidney, NH_4Cl treatment enhanced SAT1 protein abundance 3 to 4 fold. In contrast, in liver, Na_2SO_4 or NH_4Cl alone increased SAT1 mRNA whereas the combination of both treatments has surprisingly no effect. In line with the mRNA results SAT1 protein abundance was increased in Na2SO4 treated animals. However, induction of acidosis rather reduced SAT1 protein abundance in liver.
Conclusions: Thus, regulation of SAT1 by sulfate and acidosis may be organ-/cell-specific. The exact role of SAT1 in the transport of sulfate and other substrates in both organs requires further elucidation.

Su009 RENAL DYSFUNCTION IN AMERICAN CUTANEOUS LEISHMANIASIS (ACL)

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Introduction and Aims: Leishmaniasis is an infectious, non-contagious zoonotic disease for which humans are incidental hosts. Various types of kidney injury have been reported in cases of visceral leishmaniasis. The renal dysfunction seen in ACL has been attributed to the use of antimonials. To determine whether ACL itself can cause renal dysfunction, we evaluated ACL patients prior to the initiation of treatment.
Methods: This was a controlled, observational prospective study, conducted in the city of Barbalha, state of Ceara, Brazil, between July 2008 and August 2009. The study group comprised 37 patients diagnosed with ACL based on clinical, epidemiological, serological and histopathological criteria, as well as on Montenegro skin test results. Prior to treatment, glomerular and renal tubular function were tested, and the results were compared with those obtained for 8 control subjects. Urine and plasma osmolality (Uosm and Posm) were tested before and after administration of DDAVP (intranasal, after a 12-h fast). Plasma bicarbonate (Pbic), urinary pH (UpH) and plasma pH (PpH) were evaluated before and after oral administration of $CaCl_2$ (acidification test). The following urinary exosomes were quantified: AQP2; NHE3; NKCC2; and H-ATPase.
Results: Mean age was 35.6±12 years in the study group and 29±5 years in the control group. The study group comprised 19 men and 26 non-whites. All patients had cutaneous ACL (mean disease duration, 31±22 days). Of the 37 patients, 27 had a single skin lesion, 7 had 2-4 lesions, and 3 had >4 lesions. None of the patients presented glomerular dysfunction (plasma creatinine, 0.81±0.16 mg/dl; ClCr, 109±31 ml/min). Urinary concentrating defect was identified based on the post-test U/Posm ratio (<2.8) in 27 patients (77%) and post-test Uosm (<700 mOsm/kg) in 22 (63%). There was no statistical difference between the pre- and post-test osmolality values (539±43 vs. 618±34 mOsm/kg). Urinary expression of AQP2 was significantly lower in patients than in controls (99.5±0.5 vs. 38.5±12%, p=0.006), whereas that of NKCC2 was significantly higher (102±2.5 vs. 147±12%, p=0.02). Urinary acidification defect (post-test UpH >5.45) was detected in 17 patients (46%; p=0.006 vs. controls). Pre-test Pbic was <21 mEq/L in 12 patients (32.5%), and pre-test PpH was <7.35 in 14 (38%). Expression of NHE3 was significantly higher in the patients than in the controls (100±0.6 vs. 176±15%, p=0.015), as was that of H-ATPase (98±0.2 vs. 190±8%, p=0.04).
Conclusions: It is likely that the urinary concentrating defect observed in patients with ACL is caused by downregulation of AQP2 expression, and that the increased NKCC2 expression represents a compensatory mechanism. The greater expression of NHE3 and H-ATPase might represent another compensatory mechanism, since ACL patients also presented urinary acidification defect.

Su010 SALT INDUCED RECRUITMENT PATTERN OF RENIN PRODUCING CELLS IN KIDNEYS LACKING BOTH AT1a RECEPTORS AND CONNEXIN40

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Introduction and Aims: Salt induced recruitment of renin producing cells along afferent arterioles by reversible metaplastic transformation of vascular smooth muscle is a well known and physiologically relevant phenomenon, which is not yet understood at the cellular level. Recruitment of renin producing cells along afferent arterioles by modulation of salt intake is markedly enhanced in mice lacking ANGII-AT1a receptors. In contrast, mice lacking the gap junction protein connexin 40 (Cx40) show renin producing cells in the periglomerular and peritubular interstitium. In order to examine the relative relevance of ANGII-AT1a receptors and of Cx40 for the recruitment pattern of renin producing cells, we generated Cx40/AT1a double knockout mice (Cx40AT1a -/-), subjected these animals to different salt diets and determined the renal recruitment pattern of renin producing cells.
Methods: Kidney sections of Cx40AT1a -/- mice were stained immunohistochemically for renin and α-smooth muscle actin. Three mice respectively were maintained on chow with either high (4% NaCl), normal or low (0.02% NaCl) sodium content for 10 days.
Results: Compared to wildtyp mice Cx40AT1a -/- mice displayed increased numbers of renin producing cells in the walls of afferent arterioles as characteristical for AT1a deficient mice. In addition AT1aCx40 -/- featured renin producing cells located in the interstitial space as characteristical for Cx40 deficient animals. Modulation of salt intake by feeding either a high or low salt diet did not influence the number of renin producing cells in the afferent arterioles, but strikingly lowered respectively increased the number of renin expressing cells in the periglomerular and peritubular tissue.
Conclusions: These findings suggest that – independent of salt intake – lack of AT1a receptors maintains a higher number of renin expressing cells in afferent arterioles, possibly by attenuating the developmental postnatal downregulation of renal renin expression. Our data, moreover, indicate that AT1a receptors are not involved in the aberrant renin expression induced by lack of Cx40. Conversely, Cx40 appears to be highly relevant for the recruitment of renin producing cells along afferent arterioles. Finally, our data suggest that the hitherto unknown cellular factors mediating the effect of salt intake on renin expression are not only active in afferent arterioles, but also in the periglomerular und peritubular interstitium.

Su011 ALTERED FUNCTION OF PORE-FORMING PURINERGIC RECEPTORS IN CKD PATIENTS – IMPACT ON INTRACELLULAR CALCIUM HOMEOSTASIS

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Introduction and Aims: The ATP-sensitive $P2X_7$ receptor is a cation channel activated by high concentrations of extracellular ATP, and responsible for multiple processes, including calcium influx. Transient stimulation with ATP causes a rapid opening of the channel for small anorganic cations (Ca^{2+}). Sustained stimulation induces pore forming for large organic cations entry. The aim of this study was to investigate the participation of pore-forming purinergic $P2X_7$ receptors in intracellular calcium homeostasis regulation in early-stage chronic kidney disease (CKD).
Methods: The study involved 22 healthy volunteers and 22 CKD stage 2-3 patients. Intracellular calcium concentration ($[Ca^{2+}]_i$), intracellular calcium reserves and the capacitative calcium entry were measured using Fluo-3 AM fluorimetry in peripheral blood mononuclear cells (PBMCs). To determine the function of a cation channel $P2X_7$ receptors, a specific inhibitor (KN-62) and a specific agonist of these receptors (BzATP) were used. The $P2X_7$ pore function was evaluated by confocal microscopy using a fluorescent indicator ethidium bromide. Results are expressed as means ± S.E.M.
Results: In PBMCs of CKD patients, $[Ca^{2+}]_i$ (126±1.4 vs 103±1.3 nmol/l; $p<0.001$), the calcium concentration of intracellular reserves (138±9 vs 113±8 nmol/l; $p<0.02$) and the capacitative calcium entry (75±6 vs 36±3 nmol/l; $p<0.001$) were significantly increased when compared with healthy subjects. BzATP caused a sustained increase in $[Ca^{2+}]_i$ in both groups, but the effect was significantly smaller in CKD patients (21±2 vs 51±5 nmol/l; $p<0.001$). KN-62 reduced $[Ca^{2+}]_i$ in CKD patients from 126±1.7 to 114±3.9 nmol/l ($p<0.01$), but had not effect in healthy subjects. The effect of KN-62 on BzATP-gated calcium influx was also attenuated in CKD patients in comparison with healthy subjects ($p<0.001$). In CKD patients, the permeability of ethidium bromide through $P2X_7$ pores was distinct from the permeability in healthy volunteers, too.
Conclusions: Presented results demonstrate that the calcium signaling pathway is defect in PBMCs of CKD stage 2-3 patients. Purinergic pore-forming $P2X_7$ receptors are involved in altered intracellular calcium signaling and permeation of $P2X_7$ pores.
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Su012 PRE AND POSTNATAL UNDERNUTRITION CHANGES PROXIMAL TUBULAR SODIUM TRANSPORTERS

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Introduction and Aims: Chronic undernutrition compromises distal nephron ability to reabsorb sodium. It was investigated whether proximal tubule handling of sodium contributes to the loss of sodium in rats submitted to undernutrition from prenatal up to juvenile age.
Methods: Control group (C) was male Wistar rats maintained on standard diet, while urdernourished group (U) was maintained on a multideficient diet from conception, throughout lactation (maternal diet) and after weaning up to age of 60 days. Part of the animals, C (n=14) and U (n=14), was used to investigate urinary concentrating ability. Another part of the animals, C (n=25) and U (n=21), was used to obtain cortical renal homogenate and membranes. To investigate concentrating urinary ability, fasting animals were submitted to water overload, 5% (v/w), and placed in metabolic cages during 4 h. Proximal tubular (Na^++K^+) ATPase (n=7) and ouabain-insensitive Na^+-ATPase (n=7) activities were evaluated, in vitro, by measuring ATP hydrolysis. The content of cholesterol in cortical homogenate and in plasma membranes (n=6) was measured using colorimetric method. Results are mean ± SEM. Statistical analysis was performed using unpaired Student's *t*-test.
Results: U group showed body mass lower than C group, at birth (4.1±0.2 vs. 6.3±0.2 g, respectively, $p<0.05$), and also, at age of 60 days (87±4 vs. 345±12 g, respectively, $p<0.05$). Urinary Na^+ excretion (162±23 vs. 97±11 μEq/100 g/4 h, respectively, $p<0.05$) and free-water clearance (1.79±0.07 vs. 0.96±0.05 mL/100 g/4 h, respectively, $p<0.05$) were higher in U compared to C group. The (Na^++K^+) ATPase activity was lower (193±16 vs. 326±16 nmol Pi x mg^{-1} x min^{-1}, respectively, $p<0.05$), while the oubain-insensitive Na^+-ATPase activity (258±25 vs. 112±10 Pi x mg^{-1} x min^{-1}, respectively, $p<0.05$) was higher in U than in C group. The content of cholesterol in cortical homogenate (0.058±0.005 vs. 0.060±0.002 mg/mg protein, respectively) was similar between groups, while in membranes (0.045±0.015 vs. 0.100±0.018 mg/mg protein, respectively, $p<0.05$) it was lower in U than in C group.
Conclusions: Urinary sodium excretion was increased and concentrating ability was compromised in U group. The low content of cholesterol in membranes was not induced by undernutrition *per se* (see value in homogenate), it may be correlated with the decreased (Na^++K^+) ATPase activity. This enzyme is responsible for the bulk reabsorption of Na^+ in the proximal tubule and, in addition it has a signaling role that affects membrane cholesterol, while the ouabain-insensitive Na^+-ATPase is responsible for the fine tuning of Na^+ and depends on hormones. The increased activity of ouabain-insensitive Na^+-ATPase in proximal tubule of undernourished rats may reflect an hormonal scenario to preserve Na^+, while the reduced activity of the (Na^++K^+) ATPase sums up to distal loss of sodium.

Su013 CELECOXIB INHIBITS P-GLYCOPROTEIN EXPRESSION THROUGH COX-2 DEPENDENT MECHANISM IN HK-2 HUMAN TUBULAR CELLS

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Introduction and Aims: P-glycoprotein (Pgp) is a transmembrane glycoprotein encoded by the ABCB1 gene which transports a wide range of compounds from the inside of the cell to the outside, leading to a decrease in intracellular drug concentration. Pgp is located on the apical side of cells of several excretory organs. In the kidney, Pgp is expressed mainly in the proximal tubule and in a minor extension in the thick limb of Henle's loop, collecting ducts and glomerular mesangium. Several drugs including calcium channel blockers, immune suppressants, cardioactive glycosides, antibiotics and antineoplastics are actively transported by Pgp. In some cancer cell lines it was shown that regulation of ABCB1 expression was dependent on COX-2 activity. In this study we investigated the effect of the COX-2 inhibitor celecoxib (CEL) in modulating Pgp expression and activity in HK-2 human tubular cells.
Methods: HK-2 human tubular cells were cultured in presence of the selective COX-2 inhibitor CEL added to medium at different concentrations (5-50 μM) for 48-72 hours. COX-2 and Pgp expression were assessed by Western Blot (72 hrs) and SQRT-PCR (48 hrs). Calcein AM assay was used to study Pgp transport function.
Results: HK-2 cells exhibited COX-2 expression as demonstrated by the presence of a relative immunoreactive band on WB. Treatment of HK-2 cells with CEL (25 μM) resulted in reduction of Pgp expression to 41% of controls. RT-PCR showed a decline of ABCB1 gene expression to 68% of controls. Pgp dependent transport was also affected by CEL since calcein-AM intracellular accumulation test showed a dose dependent retention of the probe to 264% of controls. HK-2 cells exposed to celecoxib (25 μM) resulted more sensitive to Cyclosporin A toxicity (51.5% viability of controls).
Conclusions: The results of this study demonstrated that HK-2 tubular cells constitutively express COX-2. Regulation of Pgp expression was dependent on COX-2 activity. Treatment with CEL, a specific COX-2 inhibitor, down-regulated Pgp expression and activity. COX-2 inhibitors may enhance tubular cytotoxic effect of drugs like CsA that are substrates of Pgp.

Su014 EARLY POSTNATAL α-TOCOPHEROL MODULATES PRENATAL UNDERNUTRITION-INDUCED CHANGES ON PROXIMAL TUBULAR SODIUM TRANSPORT IN RATS

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Introduction and Aims: Prenatal undernutrition increases renal oxidative stress and suppress ouabain-insensitive Na^+-ATPase responsiveness to Angiotensin II (AngII). This second sodium pump of basolateral membrane of proximal tubule has been correlated with hypertension. We evaluated whether α-tocopherol administered during lactation leads to reprogramming of prenatal undurnutrition-programmed changes on this enzyme.
Methods: Rats were obtained from Wistar dams fed with standard (C) or multideficient diet (M) during gestation and orally submitted to corn oil (V, 1 mL/kg BW) or α-tocopherol (T, 350 mg/kg BW) treatment during lactation. At age of 90 days, the offspring (CV, n=10; MV, n=7; CT, n=12; MT, n=12) was sacrificed to collect renal proximal membranes. Na^+-ATPase activity was evaluated by measuring furosemide-sensitive ATP hydrolysis. PKA and PKC activities were assayed by measuring incorporation of γ-phosphoril from γ^{32}P-ATP into histone using specific inhibitors. Enzymes activities were corrected for protein content. AngII receptors, AT_1R and AT_2R, PKA catalytic subunit and PKC isoforms (α, ε,λ, and ζ) expression were evaluated by immunoblotting. Results are mean± SEM. Statistical analysis was performed using Student-Newman-Keuls.
Results: The CV group showed increased Na^+-ATPase activity in presence of AngII 10^{-14} M (AngII: 140±8 vs Basal: 105±6 nmol/mg/min, $p<0.01$). MV though had presented higher Na^+-ATPase activity than CV group (152±14 vs 105±6 nmol/mg/min, respectively, $p<0.01$), was unresponsive to AngII effects on this enzyme (AngII: 156±9 vs Basal: 152±14 nmol/mg/min). MV also presented, increased expression of AT_1R (56%, $p<0.01$) and AT_2R (86%, $p<0.001$), increased PKA activity (6.7±1.2 vs 1.7±0.1 pmol/mg/min, respectively, $P<0.001$), but lowered PKC activity (0.94±0.10 vs 0.34±0.02 pmol/mg/min, respectively, $p<0.01$) when compared to CV group. This reduced PKC activity was accompanied by lower expression of PKCε (24%, $p<0.01$). Different from MV, MT group showed Na^+-ATPase activation by AngII (AngII: 130±11 vs Basal: 84±4 nmol/mg/min, $p<0.01$). Moreover, MT group presented the same Na^+-ATPase, PKC and PKA activities as CV group, as well as, the same expressions of PKC, PKA, AT_1R and AT_2R. CT group presented increment in Na^+-ATPase (191%, $p<0.001$) and PKC (166%, $p<0.001$) activities.
Conclusions: The unresponsiveness of the ouabain-insensitive NA^+-ATPase to AngII, in adult prenatal malnourished rats, was allied to a reduced PKC activity that could be determined by dimerization between AT_1R and AT_2R, which downregulates their activities or even, it could be due to the observed increment in PKA activity. α-Tocopherol administered during lactation recovered, in adult prenatal malnourished rats, the expression of AngII receptors, as well as, the PKA and PKC activities and the responsiveness of the ouabain-insensitive Na^+-ATPase to AngII.

Su015 OXIDATIVE STRESS PATHWAYS IN THE PATHOGENESIS OF RENAL FIBROSIS: THE ROLE OF THE VASOACTIVE COMPOUND ANGIOTENSIN II (ANGII) AND THE PLATELET DERIVED GROWTH FACTOR (PDGF)

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Introduction and Aims: Profibrogenic agonists such as ANGII and PDGF have been implicated to stimulate oxidative stress (OS) by enhancing the production of reactive oxygen species (ROS). As a consequence, they contribute in the development and acceleration of renal damage by sustaining cell growth, inflammation and excessive tissue remodelling and accumulation.
Despite the vast knowledge about the mechanisms and the significant progress in many aspects of renal fibrosis research, still associated changes in protein expression which may help maintain and control the fibronic and proliferative state of the cell receive little intension.
In our study, we aimed to investigate the influence of both ANGII and PDGF on protein expression in two established human renal cell lines derived from normal kidney.
Methods: In order to identify new molecular factors and pathways potentially associated with OS and renal fibrosis, we performed differential proteomics analysis. Proteins extracted from control and treated cell groups were profiled by two dimensional gel electrophoresis. Using the Delta2D software (Decodon GmbH) more than 2000 different protein spots were focalized in the pH range 5-8. Differently expressed proteins were processed and identified using mass spectrometery combined with data bank search.
Results: Incubation of cells with ANGII or PDGF mediated cell proliferation. However, the proliferation rate in the fibroblastic TK173 cells was more pronounced compared to the epithelial HK2 cells.
The expression of more than 70 and 40 proteins was altered in the TK173 and HK2 cells respectively under each stress condition. Fibrogenesis protein markers (e.g. FN1, VIM, CO1A1, CO6A1, NPM1) were highly up regulated in the treated fibroblast compared to the epithelial cells. Interestingly, comparing the expression of proteins involved in the OS pathways in both cell lines revealed similar behaviour where several OS marker proteins (e.g. PRDX, TXN, GLRX3, SOD, STP1, HYOU1, GPX1) were highly up regulated. Nevertheless, the OS triggering factor ANGII evoked the regulation of more proteins than its corresponding PDGF. Diverse groups of proteins from which, cytoskeletal proteins, proteins affecting metabolism, proteins involved in signal transduction and that functioning in translation and RNA processing were also identified and served as landmarks for comparison.
Conclusions: Our results support the view that cellular adaptation to OS is accompanied by modulation of coordinated cellular and molecular events.

Further, strengthen the fact that proteomics analysis is a valuable tool that may facilitate the determination of novel fibrosis key proteins.

Su016 WARNING: LIFE-THREATENING HYPERKALEMIA DUE TO RENIN-ANGIOTENSIN SYSTEM + ALDOSTERONE INHIBITION IN UNDIAGNOSED TYPE IV RENAL TUBULAR ACIDOSIS

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Introduction and Aims: Life-threatening hyperkalemia has been increasingly reported in patients with even mild chronic renal failure and type IV renal tubular acidosis (RTA) treated with drugs which inhibit on renin-angiotensin-aldosterone system (RAAS).
Aim of the study was to evaluate prevalence and clinical features of patients with type IV RTA who were admitted at our hospital for hyperkalemia.
Methods: To evaluate entity of the problem and define predisposing risk factors we analysed patients who were admitted at our Emergency Department for hyperkalemia (serum potassium > 5,5 mEq/L) between November 1st 2007 and April 30th 2009 and focused on a subgroup of patients in whom type IV renal tubular acidosis was diagnosed. These latter patients were compared with a group of normokalemic patients as for the correlation curve between potassium fractional excretion (KFe) and creatinine clearance.
Results: Iperkalemia accounted for 0,5% of admission to Emergency Department at our Hospital in the examined period (492 pts out of 120.000) and it occurred in patients with drugs active on RAAS in 40% of cases. Type IV RTA was diagnosed in 40 pts, 24 male and 16 female. Their mean age was 75 years-old, they all had type 2 metabolic acidosis (normal anion gap), urinary pH < 6 and serum potassium levels exceeding 6 mEq/L in 75% of cases. Serum creatinine (sCr) value was 2,4 mg/dL and tended to underestimate severity of chronic renal failure, as shown by corresponding MDRD value of < 30 ml/min in 69% of male and 58% of female. Pathogenesis of RTA could be attributed to: primary hypoaldosteronism (2/40), hyporeninemic hypoaldosteronism (2/40) and chronic nephropathies (36/40) such as nephrosclerosis and diabetic nephropathy. 85% of pts was on therapy with at least one drug acting on RAAS and in 40% of cases and association of two such drugs was present. Hyperkalemia-related symptoms were more frequent in elderly patients (mean age 78), with higher potassium levels (7,5 vs 6,4 mEq/L, p=0,001), lower sodium levels (131 mEq/L vs 139 p=0,001), lower creatinine clearance and bicarbonate levels (16,8 vs 20,8 mmol/L, p=0,008) as compared with asymptomatic patients. Patients with RTA had a significantly lower KFe when compared with 115 patients with several levels of renal function, especially in the area of advanced renal failure (creatinine clearance of less than 20 mL/min) in which the expected compensatory rise in K excretion was impaired.
Conclusions: Type IV RTA is a frequent underlying condition in patients with chronic renal failure admitted for hyperkalemia, especially when this is not proportionate to the degree of renal failure. Diagnosis of RTA is especially important due to the widespread use of drugs active on RAAS in elderly patients and the consequent risks of life-threatening hyperkalemia.

Su017 RETINOIC ACID BLUNTED THE EFFECTS OF VEGF ON ENDOTHELIAL PERMEABILITY AND FENESTRAE-ASSOCIATED PV-1 PROTEIN EXPRESSION VIA VEGFR-2

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Introduction and Aims: Retinoic acid (RA), a vitamin A derivative, regulates cell differentiation and improves kidney function, albuminuria and blood pressure in experimental glomerulonephritis. In afferent arteriole vascular endothelial growth factor (VEGF) may increase endothelial fenestration, permeability and the expression of fenestrae-associated PV-1 protein. We hypothesized that RA may alter the effects of VEGF on endothelial cells by modulating the activation of VEGFR-2.
Methods: Human umbilical vein endothelial cells (HUVEC) in culture were treated with 1 and 10 μM of RA. The mRNA and protein of PV-1 was measured with real-time PCR and western blot. 40 kDa large FITC-labeled dextran was used to follow the changes in endothelial permeability. The activation (phosphorylation) of VEGFR-2 at tyrosine 1175 was detected with western blot.
Results: 10 μM of RA decreased the mRNA and protein levels of PV-1 and reduced the permeability of endothelial monolayer. This effect was most pronounced after 48 hours. Furthermore RA had beneficial effect on cell viability demonstrated with MTT assay. 30 minutes pre-incubation of HUVEC with RA blocked the endothelial permeability induced by 100 ng/ml VEGF and blunted the increase in mRNA and protein levels of PV-1. By inhibiting the phosphorylation of tyrosine 1175, RA pretreatment prevented the activation of VEGFR-2 after VEGF treatment.
Conclusions: In summary, we demonstrated that RA decreased permeability and PV-1 expression and blocked the effect of VEGF on endothelial permeability. Our results suggest that RA may modulate the endothelial permeability effect of VEGF which at least is partly induced by PV-1 protein. These effects of RA are proceeded through inactivation of VEGFR-2. Further studies are needed to clarify if this mechanism is involved the regulation of afferent arteriolar fenestration and the beneficial effect of RA in experimental glomerulonephritis.

Su018 COX-2 INHIBITION LIMITS P-GLYCOPROTEIN DOWN-REGULATION INDUCED BY ALBUMIN IN HK-2 HUMAN TUBULAR CELLS

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Introduction and Aims: P-glycoprotein (Pgp) is a transmembrane glycoprotein which transports a wide range of compounds from the inside to the outside of the cell leading to a decrease in intracellular drug concentration. Pgp is located on the apical side of cells of several excretory organs. In the kidney, Pgp is expressed mainly in the proximal tubule and in a minor extension in the thick limb of Henle's loop, collecting ducts and glomerular mesangium. Pgp expression is regulated by several factors, including the activity of COX-2 that, as shown by various studies, appears involved in renal physiology. Stimuli able to induce COX-2 expression, such as ROS or cytokines, induce Pgp overexpression as demonstrated in some cancer cell lines. Here we investigated the effect of the COX-2 inhibitor celecoxib (CEL) in modulating Pgp expression and activity in HK-2 human tubular cells exposed to albumin overload, a condition that significantly down-regulates Pgp.
Methods: HK-2 human tubular cells were cultured in the presence of albumin (15 mg/mL). CEL was added to the medium at different concentrations (5-50 μM) for 48-72 hours. COX-2 and Pgp protein expression were assessed by Western Blot (WB). Semi-quantitative RT-PCR was performed to study ABCB1 gene expression.
Results: HK-2 cells exhibited COX-2 expression as demonstrated by the presence of a relative immunoreactive band on WB. Albumin down-regulated Pgp expression (WB) up to 49% of controls. Treatment of HK-2 cells with CEL (25 μM) resulted in reduction of Pgp expression at protein (41% of controls) and mRNA (61% of controls) levels. In cells exposed to albumin CEL partially restored Pgp expression (WB) to 61% of controls.
Conclusions: The results of this study demonstrated that HK-2 tubular cells constitutively express COX-2. In HK-2 tubular cells regulation of Pgp expression was dependent on COX-2 activity. Both albumin and CEL, a specific COX-2 inhibitor, down-regulated Pgp expression. However CEL reduced the Pgp down-regulation induced by albumin. This result suggests that in HK-2 tubular cells COX-2 inhibition hinders the albumin down-regulation of Pgp probably by acting on a different signaling pathway.

Su019 COMPLEMENT 3 IS INVOLVED IN CHANGING THE PHENOTYPE OF HUMAN RENAL TUBULAR EPITHELIAL CELLS

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Introduction and Aims: Complement activation contributes to tissue injury in various forms of glomerulopathy and is characterized by deposition of complement components, which accelerates the progression of chronic renal damage. We recently reported that complement 3 (C3), a critical component of the complement system, is associated with the synthetic phenotype of mesangial cells. It is possible that C3 stimulates human renal tubular epithelial cells to come true epithelial to mesenchymal transition to, in turn, induce interstitial fibrosis. We investigated the role of C3 in the epithelial to mesenchymal transition of human renal tubular epithelial cells.
Methods: Cultured HK-2 were treated with 50ng/ml IFN-γ, 10ng/ml TNF-α and 0.1μM C3a for 24 hours. siRNA target human C3 was transfected to HK-2. After 24h transfection of C3 siRNA, HK-2 were stimulated with 10 mg/ml TNF-a for 24 h.The expression of C3, α-SMA and E-cadherin mRNA and protein were analysed by RT-PCR and Western Blot. The expression of α-SMA and E-cadherin were also detected by indirect immunofluorescence.
Results: Cultured human renal tubular epithelial cells (HK-2) expressed C3 mRNA and protein, and levels were increased in response to IFN-g and TNF-a. Exogenous C3a increased expression α-smooth muscle actin (α-SMA) mRNA and protein, a marker of myofibroblasts, and decreased the expression of E-cadherin mRNA and protein, which are markers of renal tubular epithelial cells. Small interfering RNA (siRNA) targeting C3 increased expression of E-cadherin mRNA and protein, and decreased expression of α-SMA mRNA and protein in HK-2.
Conclusions: These results indicate that C3 causes human renal tubular epithelial cells to convert to myofibroblasts, suggesting that C3 may play an important role in epithelial to mesenchymal transition of human renal tubular epithelial cells in renal interstitial fibrosis.

Su020 DIRECT EFFECT OF METHYLPREDNISOLONE ON RENAL SODIUM AND WATER TRANSPORT VIA THE PRINCIPAL CELLS IN THE KIDNEY

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Introduction and Aims: Glucocorticoids influence renal concentrating and diluting ability. We tested the hypothesis that methylprednisolone treatment increased renal water and sodium absorption by increased absorption via the aquaporin 2 water channels (AQP2) and the epithelial sodium channels (ENaC), respectively.
Methods: The effect of methylprednisolone was measured during fasting in a randomized, placebo-controlled, single-blinded cross-over study of 15 healthy humans. The subjects received a standardized diet on day 1, fasted during day 2, and received methylprednisolone 500 mg IV on day 3. The effect variables were urinary excretions of aquaporin2 (u-AQP2), urinary excretion of the β-fraction of the epithelial sodium channel (u-$ENaC_\beta$), cyclic-AMP (u-cAMP), prostaglandin E_2 (u-PGE_2), free water clearance (C_{H2O}), fractional excretion of sodium (FE_{Na}), and plasma vasopressin (p-AVP), angiotensin II (p-Ang II), aldosteron, atrial- and brain natriuretic peptid (p-ANP, p-BNP).
Results: Methylprednisolone treatment increased u-AQP_2, u-$ENaC_\beta$ and p-AVP significantly, but did not change u-cAMP, C_{H2O} and FE_{Na}. P-ANP increased during methylprednisolone treatment, but after the increase in u-AQP2 and u-$ENaC_\beta$. U-PGE_2, P-Ang II and p-BNP were unchanged. Heart rate increased and diastolic blood pressure fell.
Conclusions: Methylprednisolone increased u-AQP2 and u-ENaC. Neither the vasopressin-c-AMP-axis, nor changes in the renin-angiotensin-aldosterone system nor the natriuretic peptide system seem to bear a causal relationship to the increase in either u-AQP2 or u-ENaC. Most likely the effect is mediated via a direct effect of methylprednisolone on the principal cells. The lack of decrease in urinary output and sodium reabsorption most likely can be attributed to the diuretic and natriuretic properties of the increased secretion of ANP.

Su021 PROTEIN ENRICHED DIET INCREASES WATER ABSORPTION VIA THE AQUAPORIN2 WATER CHANNELS IN HEALTHY HUMANS

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Introduction and Aims: According to animal experiments, a protein enriched diet increased renal absorption of sodium and water. We wanted to test the hypotheses that a protein enriched diet would increase the expression of the aquaporin2 water channels and the epithelial sodium channels in the distal part of the nephron using biomarkers for the activity of the two channels.
Methods: We performed a randomized, placebo controlled crossover study in 13 healthy humans to examine the effect of protein enriched diet on renal handling of water and sodium during baseline condition and during hypertonic saline infusion. We measured the effect of protein enriched diet on urinary excretions of aquaporin2 (u-AQP2), the β-fraction of the epithelial sodium channels (u-$ENaC_\beta$), free water clearance (C_{H2O}), fractional excretion of sodium, and vasoactive hormones.
Results: During baseline conditions, u-AQP2 increased and C_{H2O} decreased during protein enriched diet, whereas u-$ENaC_\beta$ was unchanged, although urinary sodium excretion increased. During hypertonic saline infusion, the response in the effect variables did not deviate between protein enriched and normal diet. Plasma concentrations of angiotensin II and aldosterone increased as well as pulse rate. Vasopressin in plasma was unchanged, and prostaglandin E_2 fell during protein enriched diet.
Conclusions: Protein enriched diet increased water absorption via increased transport via the aquaporin2 water channels. The increased u-AQP2 might be due to a reduced prostaglandin level. The increase in renal sodium excretion seems to be mediated in another part of the nephron than the epithelial sodium channels.

Su022 ALTERED RENAL REGULATION OF ACID-BASE TRANSPORTERS IN RATS WITH GLYCEROL-INDUCED ACUTE KIDNEY INJURY

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Introduction and Aims: The present study was aimed to investigate whether the renal tubular acid-base transporters are altered in the glycerol-induced acute kidney injury.
Methods: Male Sprague-Dawley rats were injected with 50% glycerol in normal saline (7 mL/kg, i.m.) after water deprivation for 12 hours. They were sacrificed at 24 hours after the glycerol injection. Control rats were injected with normal saline. The protein expression of Na,K-ATPase α1-subunit, type 3 Na^+/H^+ exchanger (NHE3), type 1 $Na^+{:}HCO_3^-$ (NBC1), H^+-ATPase and anion exchanger-1 (AE1) was determined in the cortex of the kidney by semiquantitative immunoblotting and immunohistochemistry.
Results: In experimental rats, creatinine clearance was decreased. Plama pH and bicarbonate concentrations were not changed. However, urine pH and bicarobonate concentrations were significantly decreased. In experimental rats, the protein expression of Na,K-ATPase α1-subunit was decreased in the cortex of the kidney. The expression of NHE3 and NBC1 was also decreased. Immunolabeling of Na,K-ATPase α1-subunit, NHE3 and NBC1 in the proximal tubule was decreased. However, the protein expression of H^+-ATPase and AE1 was increased in the cortex. Immunolabeling of H^+-ATPase and AE1 was also increased in the cortical collecting duct (CCD).
Conclusions: Glycerol treatment results in the downregulation of Na,K-ATPase α1-subunit, NHE3 and NBC1 in the proximal tubule, which may contribute to impaired proximal tubular reabsorption of bicarbonate. In addition, upregulation of H^+-ATPase and AE1 in CCD may play a compensatory role in the maintenance of acid-base homeostasis.

Su023 METABOLIC EVALUATION AND RISK OF RELAPSE IN A LARGE COHORT OF PATIENTS WITH KIDNEY STONES

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Introduction and Aims: Nephrolithiasis is a common condition in the western countries. The pathogenesis of the stones is the result of an imbalance between the presence of promoting factors and inhibitors of lithogenesis and the lithogenic risk is the results of the balance between these factors.The aim of the study was to perform the metabolic evaluation in a large cohort of patients with kidney stones recruited in 2 different Italian centres, in North (Centre A – Turin) and South (Centre B – Bari) of Italy, to estimate the lithogenic risk and the possible factors responsible for the risk of relapse of nephrolithiasis.
Methods: We evaluated the metabolic profile of 1624 patients who suffered from kidney stones [922 from Centre A (431 M, 491 F; mean age 49.7±16.7 years) and 702 patients from Centre B (302 M, 400 F; mean age 46.5±22.8 years)]. In 1153 patients we evaluated the urine state of supersaturation (β) with calcium oxalate (βCaOx), calcium phosphate (βbsh) and uric acid (βAU) using a validated software (LITHORISK). In 299 patients followed for at least 2 years the risk of relapse and the possible covariates were analyzed by Kaplan-Meier method and multivariate Cox proportional hazard method.
Results: There were no significant differences between the Centre A and Centre B, except for diuresis (22.8±8.5 dL/24h vs 17.4±6.7 dL/24h; $p<0.001$), oxaluria (30.1±12.9 mmol/24h vs 26.3±10.5 mmol/24h; $p<0.001$) and uricuria (479.9±177.6 mg/24h vs 540.4±202.2 mg/24h; $p=0.002$). Urinary saturation estimation (β) showed a Calcium Oxalate β (β CaOx) higher than Calcium Phospate β (βbsh) and Uric Acid β (β AU). Diuresis influenced all urinary saturation states (β), particulary Calcium Oxalate (βCaOx) and Uric Acid (βAU). βCaOx and Calcium Phosphate β (βbsh) directly correlated with Calciuria. Oxaluria influenced βCaOx less than calciuria. βbsh (directly) and βAU (inversely) correlated with pH. In the long term follow-up (mean follow-up 110±84 months) 76/299 (25.4%) patients had a relapse. The probability of relapse in these patients was 14%, 28%, 36%, 46% at 5, 10, 15, 20 years respectively. At the multivariate analysis only baseline BMI (HR 1.12; CI 95% 1.04-1.22) and modification at two years of diuresis (HR 0.91; CI 95% 0.85-0.95) and urinary specific gravity (HR 1.48; CI 95% 1.12-2.06) were independent predictors of the risk of relapse.
Conclusions: All urinary saturation states were influenced mainly by urinary volumes. In the long term follow-up, modification of diuresis and urine specific gravity reduced the risk of relapse. Moreover Body Mass Index may be a novel predictor of relapsing kidney stones.

Su024 RECURRENT DEEP INTRONIC MUTATION (c.1670-191C>T) IN THE *SLC12A3* GENE RESPONSIBLE FOR TAIWAN ABORIGINES WITH GITELMAN'S SYNDROME

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Introduction and Aims: Deep intronic mutations undetected by the traditional analysis of genomic DNA may be responsible for Gitelman's syndrome (GS) caused by mutations in *SLC12A3* gene encoding the thiazide-sensitive NaCl cotransporter (NCC). This study was to evaluate if deep intronic mutations in the *SLC12A3* were detected in some Taiwanese GS patients with only one heterozyous or undetectable *SLC12A3* mutation.
Methods: Thirty-two patients with GS (M/F = 23/9, age 27±9) including 9 negative *SLC12A3* mutation and 23 heterozygous *SLC12A3* mutations were enrolled. None of them had a large deletion in *SLC12A3* and *CLCNKB* mutation. RT-PCR for mRNA transcripts extracted from blood leukocytes in two patients and a normal control was performed. The corresponding introns of genomic DNA was sequenced in all patients and 100 normal controls if deep intronic mutations were detected. The NCC expression in the DCT in the renal tissue was evaluated.
Results: mRNA transcript showed that an additional pseudoexon 238bp between exons 13 and 14 due to a homozygous point mutation in the intron 13 (c.1670-191C>T) was identified in these two patients. The pseudoexon created by this deep intronic mutation contained a new premature truncated codon (PTC, TGA)-containing transcript. Apical NCC expression in the DCT of renal tissue was markedly diminished in one patient with this homozygous mutation. This mutation was undetected in 100 healthy subjects but identified in the other 5 of 32 GS patients (7/32, approximately 20%). Of note, all GS patients carrying this mutation were Taiwan aborigines.
Conclusions: Deep intronic mutations in *SLC12A3* can be identifed with RNA-based approach in GS patients with heterozygous or negative *SLC12A3* mutation. Deep intronic point mutation (c.1670-191C>T) in the intron 13 with an inclusion pseudoexon leads to defective NCC expression and may be recurrent "hot spot" for GS in Taiwan aborigines and Austronesians outside Taiwan.

Su025 CALCIC NEPHROLITHIASIS, A DOUBLE-BLIND RANDOMIZED STUDY, 2008-2009

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Introduction and Aims: It seems to be evident that reducing the recurrence of lithiasis with a medical treatment is possible, orientated in a selective way to those patients suffering recurrent calcic urolithiasis on therapy not only to control those physiologic, metabolic or physic – chemical abnormalities inducing calculi formation but using products such as Renalof, which destroys and disintegrates the calculus in the urinary system.
Goals: To evaluate the efficacy and safety of the use of Renalof in patients with Calcic urolithiasis, to diminish and/or eliminate the calcic lithiasis.
Methods: A controlled clinical essay was carried out; a phase III randomized study using two parallel groups double blind with placebo. Patient selection was determined by patients' age from 18 to 65 years who were metabolic active carriers of renal-uretheral lithiasis smaller than 2 cm. Each patient was assigned a treatment schedule including a natural product developed by the Spanish Laboratory Catalysis SA, with a dose of 1 capsule three times daily during three months, according to the randomized list done by the Biostatistics department of the above mentioned Laboratory. A monthly clinic, radiologic, CT scan and ecographic follow up was done and a metabolic study at the beginning and at the end of the treatment were carried out, recording all the adverse effects.
Primary evaluation criteria: Size and number of calculus: Favourable outcome was considered when a reduction of the size or elimination of the calculi showed by ultrasound, urinary tract X ray or abdominal CT scan at the end of the study and secondary evaluation criteria of Metabolic studies and physic-chemical.
Results: 110 patients randomly received, a natural product, Renalof (n=52) and Placebo (n=58). Calculus diminished in a 7,7% in the Renalof treated group while 0% in the Placebo group three months after treatment; Confidence Interval (CI) 95%: 0,01-0,17 ($p<0,001$), A 78,8% of calculus were eliminated with CI: 0,60–0,88 meaning a 86,5% of response at the end of the third month using Renalof while only a 5,2% in the Placebo group. In both groups hypercalciuria was the most important disorder, when comparing both groups after treatment there was a 50% reduction of this disorder in the Renalof group while a minimal variation was noted in the Placebo group CI: 0,01–0,21, ($p=0,04$). Crystallization Risk Index and Low Urinary Output diminished at the end of the third month after treatment (53,8% and 11,5%) in the Renalof group. The mean number of Renal Colic suffered by patients of both groups pre treatment was 3,7±1,6 in Renalof's and 3,1±1,4 in Placebo's group while three months after treatment it was 0,4±1,3 in Renalof's and 2,0±1,3 in Placebo's ($p=0,001$). In this study there was a reduction in the sick leave days due to colics in the group treated with Renalof, with a mean at the third month of 0,9±2,7 while a 5,7±5,1 in the Placebo's group ($p=0,001$).
Conclusions: Renalof is an efficient and safe product to treat renal calcic calculus. No unexpected adverse effects were seen with its use.

Disclosure: I am thankful to Labarotorio Catalysis SA, of Spain, to facilitate products Renalof a with the major the security, seriousness and responsibility.

Su026 UROLITHIASIS AND BETA THALASSAEMIA MAJOR

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Introduction and Aims: Patients with beta-thalassaemia major (BTM) are subject to renal complications, including nephromegaly from extramedullary haemopoeisis and tubular dysfunction from haemosiderin deposition. They are also prone to urolithiasis, and this is thought to be due to chronic haemolysis leading to high urinary uric acid excretion and uric acid stones. We interrogated our renal stone database in order to compare patients with BTM and kidney stones with the other stone forming patients.
Methods: 13 patients with BTM were compared with the other 432 stone forming patients on the database. The probability of stone formation (PSF) was calculated according to the mathematical model of Robertson (Front Biosci, 2003). Data were analysed using Graphpad Prism software and statistical significance was calculated using the unpaired t-test. Values expressed are means ± S.E.M.
Results: As expected, BTM patients were significantly younger than non-BTM patients (31.4±3 vs. 47.2±1 years p=0.0001). However, there was no significant different between the serum creatinine measurements of BTM and non-BTM patients (69±3 vs. 92±2 mmol/L ns, respectively). Stone composition data was unavailable for 2 of the BTM patients, and all identified stones in this group were calcium oxalate or mixed calcium oxalate/phosphate.
The BTM patients had significantly higher serum bicarbonate measurements than the non-BTM patients (28.6±0.5 vs. 26.4±0.2 mmol/L p=0.01) although there was no significant difference in urine pH (6.18±0.1 vs. 6.15±0.1, ns).
Serum uric acid was significantly lower in the BTM patients (270±34 vs. 340±5 μmol/L, p=0.01), however the 24-hour urinary uric acid was significantly higher in BTM patients (4.1±0.4 vs. 3.3±0.1 μmol/L, p=0.02). 24-hour urinary calcium was significantly higher in BTM patients (9.5±1.8 vs. 5.4±0.2 mmol/L, p<0.0001), despite the fact that they had a slightly lower dietary calcium intake than non-BTM patients (23.3±2 vs. 24.6±1 mmol/day, ns, normal range 24-26 mmol/day)
Serum phosphate was higher in BTM than non-BTM patients (1.2±0.1 vs. 1.02±0 mmol/L, p=0.001), as was 24-hour urinary citrate (3.4±0.4 vs. 2.5±0.1 mmol/L, p=0.03).
The PSF in BTM and non-BTM patients was very low for uric acid stones (0.7×10^{-6} ± 0 vs. 0.04±0, ns), and mixed uric acid and calcium oxalate stones (0.5×10^{-4}±0 vs. 0.04±0, ns).
The PSF was significantly higher in BTM patients for the formation of calcium oxalate (0.57±0.1 vs. 0.3±0.01, p=0.005) and mixed calcium oxalate/phosphate stones (0.6±0.1 vs. 0.3±0.01, p=0.005).
Conclusions: Despite a higher concentration of urinary uric acid in the BTM patients, these data do not support the commonly held view that they are more likely to form uric acid stones. They are hypercalciuric and have an even higher tendency to form calcium oxalate and mixed calcium oxalate/phosphate stones than the other stone forming patients on the database. Resorptive bone disease is prevalent in these patients, which may be genetic, or related to hypogonadism. We suspect that this, and the consequent hypercalciuria is the cause of urolithiasis in these patients, rather than uric acid stone formation consequent to chronic haemolysis.

Su027 SERUM POTASSIUM LEVELS ON ADMISSION AND INFARCT SIZE IN PATIENTS WITH ACUTE MYOCARDIAL INFARCTION

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Introduction and Aims: Little data exist on the relationship between potassium level and infarct size after ST-segment elevation acute myocardial infarction (STEMI). We investigated the influence of potassium levels on infarct size measured with single photon emission computed tomography (SPECT) in patients with STEMI after mechanical reperfusion.
Methods: The study included 598 patients. Potassium measurements at baseline and 2 SPECT examinations, at baseline and 7-14 days after intervention, were performed. Infarct size in the 7-14 days SPECT and salvage index were the primary outcome analyses.
Results: Tertiles of baseline potassium were: <4.05 mEq/L (1st tertile), 4.05-4.37 mEq/L (2nd tertile) and > 4.37 mEq/L (3rd tertile). In potassium 1st, 2nd and 3rd tertiles, the infarct size in the 7-14 days SPECT (median [25th-75th percentiles]) was 9.0% [2.0%-21.8%], 10.0% [3.5%-22.0%] and 12.0% [5.0%-25.5%] of left ventricle (p=0.026); salvage index was 0.50 [0.26-0.84], 0.56 [0.26-0.81] and 0.40 [0.23-0.75] (p=0.09).

Scintigraphic Data of Study Participants

Characteristic	Lower tertile (n=199)	Middle tertile (n=196)	Upper tertile (n=203)	P value
Initial perfusion defect (% of LV)	23.0 [12.1; 37.6]	23.0 [13.6; 40.0]	25.0 [14.0; 41.0]	0.33
Perfusion defect at 7 to 14 days (% of LV)	9.0 [2.0; 21.8]	10.0 [3.5; 22.0]	12.0 [5.0; 25.5]	0.026
Absolute myocardial salvage (% of LV)	13.0 [6.0; 24.0]	13. 0 [6.0; 23.2]	12.0 [7.0; 23.0]	0.85
Salvage index	0.50 [0.26; 0.84]	0.56 [0.26; 0.81]	0.40 [0.23; 0.75]	0.09

Data are medians [25th; 75th percentiles] or number of patients (%). LV indicates left ventricle.

Patients with anterior infarction in upper potassium tertile had greater infarct size compared with patients in lower potassium tertile (p=0.049). After adjustment in multivariable analysis, potassium was an independent correlate of infarct size in the 7-14 days SPECT (p=0.05).
Conclusions: In patients with STEMI, higher baseline potassium levels are associated with a larger scinitgraphic infarct size.

Su028 STEROID INDUCED HYPERCALCIURIA; INCIDENCE, COURSE AND ASSOCIATED FACTORS

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Introduction and Aims: Steroid induced calciuria is a well known phenomenon but its charcteristics and details are not well defined. We aimed to investigate effects of corticosteroids on urinary calcium excretion and some related factors.
Methods: Thirty two adult patients (22 female, 10 male) who were planning to take corticosteroids at least 10 mg/day as methyl prednisolon or its equivalent at least for 30 days ore more were included in the study. Four patients died during the study, three patients who gave up their follow up and three patients who should not stop corticosteroids were excluded from the study. As parameters, 24-hours urinary calcium, sodium, urinary calcium/creatinin ratio in spot sample, urinary deoxypyridinoline and serum calcium, phosphorus, alkalen phosphatase, albumin, creatinine, osteocalcin and parathormone levels were studied before corticosteroid treatment, after the first week, within second month and after the cessation of the treatment.
Results: The mean 24-h urinary calcium excretion was 98,7±88,1 mg/day before the treatment which increased to 182,2±158,6 mg/day at the first week and 196,9±167,8 mg/day in 2nd month and later decreased to 118,9±90,2 mg/day after cessation of corticosteroid. The paired-samples T- test revealed significant increase at the first week and 2nd months compared to baseline (p<0,001). There is no relationship between age and body mass index with the calcium excretion. The mean deoxypyridinoline levels were 80,8±64,0 nM before the treatment, 81,5±64,1 nM at first week, 139,9±109,2l nM in 1st-2nd months and 111,2±79,7 nM after cessation of corticosteroid. The difference was significant in 2nd month compared to baseline. The mean serum calcium levels were 8,6±1,15 mg/dl before the treatment, 8,8±0,9 mg/dl at first week, 9,2±0,7 mg/dl in 2nd month and 9,5±0,6 mg/dl after cessation of corticosteroid, the differences

were significant between baseline and 1st-2nd months or after the cessation (p<0,05). There is no significant change detected in parathormone, osteocalcin, phosphorus, creatinin, and alkalen phosphatase levels.
Conclusions: We demonstrated that corticosteroid induced hypercalciuria onsets after the begining of the treatment at the first week and continues till the cessation of corticosteroid. It was accompanied by an increased urinary excretion of deoxypyridinolines at second month.

Su029 A COMPARISON OF METABOLIC VARIABLES AMONG OBESE AND NON-OBESE KIDNEY STONE FORMERS

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Introduction and Aims: Kidney stone disease is common worldwide and the incidence is growing. Increasing prevalence of obesity, metabolic syndrome and type 2 diabetes have been linked with this increase. Studies also suggest that these insulin resistant states may be associated with increased risk of uric acid stones. Several metabolic abnormalities are thought to be responsible for this association, including lower urinary volume, acidic urinary pH, hyperuricosuria, hypercalciuria, hyperoxaluria and hypocitraturia. We studied metabolic differences between obese and non-obese stone formers.
Methods: We investigated the kidney stone database for the stone patients attending the stone clinic between November 97 and November 09. Those patients with a recorded BMI were divided into two categories – BMI less than or equal to 25.0 kg/m^2 (BMI<25 group) and BMI greater than 30.0 kg/m^2 (BMI>30 group). Serum creatinine, uric acid and bicarbonate were compared along with 24 hour urinary volume (U.Vol), pH (U.pH), calcium (U.Ca), oxalate (U.Ox), citrate (U.Cit), uric acid (U.UA), magnesium (U.Mg), sodium (U.Na) and potassium (U.K). We also compared the stone composition and probability of stone formation (PsF) for these groups. Data were processed using 'SigmaPlot' and statistical significance calculated using Mann-Whitney Test. Results were expressed as mean ± S.E.M.
Results: A total of 1654 patients were included, 664 (40.1%) had BMI <25 and 336 (20.3%) had BMI >30. Serum creatinine levels were significantly higher in BMI>30 group when compared with BMI<25 group (95.6±2.38 vs. 88.6±1.56 umol/L p<0.001) as was uric acid (371.0±6.0 vs. 301.5±4.0 umol/L p<0.001). Serum bicarbonate levels on the other hand were significantly lower in those with BMI >30 (26.5±0.1 vs. 27.8±0.2 mmol/L p<0.001). Significantly higher levels of U.Ox (0.364±0.0 vs. 0.348±0.1 mmol/L p<0.001), U.Cit (2.7±0.1 vs. 2.4±0.0 mmol/L p=0.004), U.UA (3.9±0.07 vs. 3.19±0.04 mmol/L p<0.001), U.Na (188±4.0 vs. 150±2.5 mmol/L p<0.001) and U.K (71±1.3 vs. 68.4±1.09 mmol/L p=0.037) were also seen in those with BMI >30. U.pH was significantly lower (5.974±0.072 vs. 6.382±0.023 p<0.001) in those with BMI >30. Differences in U.Vol, U.Mg and U.Ca were not significant. Stone composition data (N=468) showed 4% of stones to be composed of uric acid in those with BMI <25, whereas the proportion of uric acid stones was 27.8% in those with BMI >30. 1.7% individuals with BMI <25 had increased uric acid stone risk (PsF_{UA} >0.5) as compared to 10.2% of those with BMI >30.
Conclusions: There is an over-representation of individuals with high BMI (>25) in our database, suggesting increased risk of kidney stones in obesity. Increasing BMI not only increases the overall risk of stone formation but also skews the risk factors in favor of uric acid stones. This greater risk is primarily due to raised U.UA, U.Ox, U.Na and lower U.pH. U.Ca was not a significant additional factor. Higher U.Cit and U.K levels are clearly insufficient in mitigating this increased risk. These patients also have worse renal function which puts them at a higher risk of prospective renal dysfunction.

Su030 HYPERKALEMIA IN HEMODIALYSIS PATIENTS NOT ALL IS SAID

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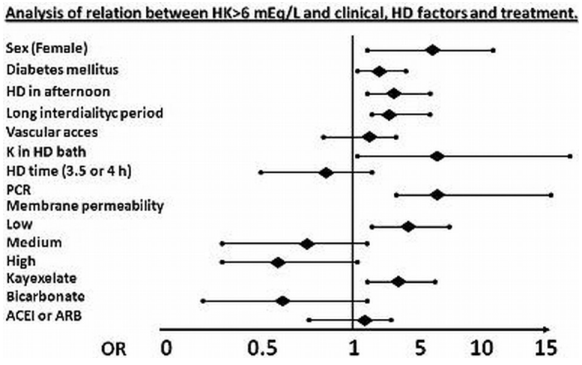
Introduction and Aims: In dialysis patients, mortality due to hyperkalemia (HK) is high, 10% of hemodialysis patients had predialysis plasma potassium concentrations > 6.0 mEq/L, moreover 24% of ESRD patients requiring an emergency dialysis due to HK. In view of the above our aim was to identify key risk factors associated with HK in hemodialysis patients.
Methods: Prospective study during 2009 year, included 20 patients with biochemical monitoring monthly for 12 months. Recording was made for biochemical, clinical and HD parameters (long or short period of interdialytic HD, time of HD (morning or afternoon), membrane permeability (low, medium and high), days of HD (M-W-S or T-T-F), kT/V, HD time, K concentration in bath, protein catabolic rate (PCR)) and treatments. We excluded subjects who were hospitalized or died during the study. Statistical analysis was performed using SPSS 17, we use mean and percentages, chi square test, T-student and risk estimates. Finally, we perform logistic regression analysis and considered as a possible explanatory variable from the regression those that were statistically significant with a p value <0.05.
Results: Patients with a K>6 mEq/L were associated with being female (p=0.001, OR: 5.88 [95% CI 2.72-12.71]), diabetes (p=0.03, OR: 1.96 [95% CI 1.06-3.62]); afternoon HD (p: 0.0002, OR: 3.4 [95% CI 1.71-6.76]); long interdialytic period (p=0.001, OR: 2.8 [95% CI 1.47-5.33]), low membrane permeability (p=0.0001, OR: 3.9 [95% CI 1.85-8.21]); K in HD bath of 2.5 mEq/L (p=0.02, OR: 7.66 [95% CI 1.01-58.12]), treatment with kayexelate (p: 0.0002, OR: 3.41 [95% CI 1.71-6.76]), PCR (p: 0.0004, OR: 6.91 [95% CI 3.05-15.43]), pH (p: 0.009), glycemia (p: 0.04), but was unrelated with kT/V, BMI, albumin, bicarbonate in blood or therapy, ACEI, ARBs, HD time. Logistic regression analysis shows the association between a K>6 mEq/L with sex, DM, PCR, long interdialytic period and HD the days T-T-F.



Logistic regression analysis for HK>6 mEq/L

Variables	B	t	Sig.	IC 95% UL	IC 95% LL
Constant	-1.45	10.75	0.001		
Diabetes	1.41	12.61	0.0003	1.88	8.94
Long period	1.35	12.40	0.0004	1.82	8.17
Days T-T-F	0.92	5.01	0.025	1.12	5.67
PCR	1.38	6.45	0.011	1.37	11.54
Sex	0.95	3.32	0.068	0.93	7.21

Conclusions: Rational treatment and prevention of HK in patients with ESRD requires the knowledge of the risk factors, so we concluded that the days of HD (T-T-F), long interdialytic period, DM, PCR and sex appears to influence the levels of serum predialysis potassium and are relate with K>6 mEq/L. Therefore it is important to modify the risk factors that are possible in patients who are high risk.

Cell signalling

Su031 HYPOXIA-INDUCIBLE PROTEIN 2 (HIG2) IS A NOVEL LIPID DROPLET PROTEIN AND A SPECIFIC TRANSCRIPTIONAL TARGET OF HYPOXIA-INDUCIBLE FACTOR (HIF)-1

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Introduction and Aims: Hypoxia-inducible protein 2 (HIG2) is a 63 amino acid peptide that has been implicated in canonical Wnt signalling, both as target and activator, and is abundantly expressed in renal clear cell carcinoma. The potential link between hypoxia and an oncogenic signalling pathway prompted us to analyze HIG2 function and regulation in detail.
Methods: HIG2 expression was analysed by RNase protection, RT PCR, immunoblot assays and immunohistochemistry. Promoter analyses, gelshift assays and siRNA studies served to characterize the mechanisms underlying the hypoxic induction of HIG2. HeLa clones with tetracyclin-inducible HIG2 overexpression were generated to study HIG2 function, and its subcellular localization was determined by immunofluorescence. Intracellular lipids were stained with the fluorescent neutral lipid dye Bodipy 493/503 and lipid accumulation was quantified by flow cytometry.
Results: We found that HIG2 was markedly upregulated by hypoxia and chemical inducers of hypoxia-inducible factors (HIF) in all cell types and mouse organs investigated. Promoter analyses showed that HIG2 is a direct and specific target of HIF-1, but not responsive to HIF-2. Surprisingly, HIG2 overexpression neither stimulated proliferation, nor did it activate canonical Wnt signalling, and secretion of HIG2 was not detectable. Instead, we show that endogenous and overexpressed HIG2 decorates the phospholipid monolayers of cytosolic lipid droplets, whose number and size increase upon hypoxic inhibition of fatty acid β-oxidation. Accordingly, HIG2 colocalized with known lipid droplet proteins such as adipophilin/ADRP and TIP47. Normoxic overexpression of HIG2 was sufficient to increase neutral lipid deposition in HeLa cells as well as expression of vascular endothelial growth factor and cytokines such as macrophage migration inhibitory factor. In renal clear cell carcinomas with constitutive HIF-1 activity due to loss of the von Hippel-Lindau tumor suppressor protein HIG2 lined the empty spaces representing washed-out lipid droplets. Furthermore, HIG2 could be detected by immunohistochemistry in atherosclerotic plaques in macrophage foam cells and in hepatocytes in particular cases of fatty liver disease, indicative of regional hypoxia.
Conclusions: Thus, HIG2 is a ubiquitously inducible, specific HIF-1 target gene and stimulates the intracellular storage of neutral lipids under conditions of reduced oxygen supply, which may have important clinical implications in atherosclerosis, fatty liver disease and metabolic syndrome.

Su032 FIBROBLAST MIGRATION AND PROLIFERATION IS REGULATED BY N-RAS THROUGH ERK AND AKT ACTIVATION

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Introduction and Aims: Interstitial renal fibrosis, the common end-point of progressive kidney disease, is a complex process involving not only derangements in both the synthesis and degradation of extracellular matrix (ECM), but also cell infiltration, accumulation of activated myofibroblasts and increased deposition of ECM. Cellular migration and proliferation are important processes during the development of fibrosis. The small GTPase Ras is crucial for the TGF-beta1- induced epithelial-to-mesenchymal transition. There are three isoforms of Ras GTPases: H-, N- and K-Ras. Previous experiments of our group showed an increase in ECM accumulation in H-Ras and N-Ras (H,N-ras-/-//-/-) mice embrionary double knock-out (KO) fibroblasts, and two of the main Ras-induced pathways, Raf – mitogen activated protein kinase (MAPK) Erk and phospho-inositol 3 kinase (PI3K) – Akt seem to mediate this ECM synthesis. In this study we assessed the involvement of the N-Ras isoform in fibroblast migration and proliferation. We also studied the role of Erk and Akt pathways in these processes.
Methods: For this purpose, we analyzed fibroblast proliferation and migration in embrionary fibroblasts obtained from knockout (KO) mice for N-Ras (N-ras-/-) isoform. Proliferation was analyzed by immunostaining of the nuclear antigen Ki67, by fibroblast nuclei staining with crystal violet and by PCNA western-blot. Fibroblast mobility was evaluated by the half-closure time of in vitro induced wounds in fibroblast monolayers, and by trans-well migration of fibroblasts through a Boyden chamber.
Results: The lack of N-Ras isoform led to a reduction in fibroblast proliferation and migration. TGF-beta1-induced proliferation was reduced in N-ras-/- fibroblasts and pre-treatment with U0126, a MEK inhibitor, dismissed TGF-beta1-induced proliferation in control fibroblast whereas treatment with LY294002, a PI3K inhibitor, decreased TGF-beta1-induced proliferation in both control and KO fibroblast. We also observed that Akt inhibition reduced both wound healing and fibroblast migration through Boyden chamber, whereas Erk inhibition dismissed migration through 8 micrometers-pore membranes only in N-ras-/- fibroblasts; however, there were no changes in wound closure time after Erk inhibition.
Conclusions: All these results show the involvement of N-Ras isoform in fibrotic processes as fibroblast proliferation and migration. Our data suggest that N-ras isoform is regulating the proliferation and cell migration through Erk and Akt: both pathways seem to modulate the TGF-beta1- induced proliferation in fibroblasts; N-Ras and Akt may be involved, at least in part, in fibroblasts mobility.

Su033 REGULATORY T CELLS (Treg) AND INTERLEUKIN 17 PRODUCING T CELLS (TH17) IN IgA NEPHROPATHY

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Introduction and Aims: IgA nephropathy (IgAN) is a glomerular disease characterized by a dysregulation of the immune system leading to an abnormal immune response to mucosal antigens. In these patients, we recently reported an increased expression of Toll-like receptor-4 (TLR4) in peripheral blood mononuclear cells (PBMC), which was correlated with proteinuria levels. Recent reports have focused interest on the role of the balance between interleukin (IL)-17 producing T cells (TH17) and regulatory T (Treg) cells in immune-mediated diseases. We aimed this study at investigating Treg/TH-17 producing cells balance in patients with IgAN, looking at possible correlation with TLRs expression, as marker of innate immunity.
Methods: PBMC were isolated from 28 patients with IgAN (median age 39.6, IQ range 19.1-65.0 years), 82% males, e-GFR 92.6±46.5 ml/min, median proteinuria 0.2 g/day (IQ range 0.10-0.40), and from 10 healthy controls (HC).
The research protocol included the measurement in Taqman of mRNA expression of regulation-associated genes of Treg (Foxp3), TH17-related factors (IL-17 and retinoid orphan nuclear receptor (RORc), and TFG-beta1 which modulates the differentiation of TH17. Moreover, mRNAs encoding for TLR 2, 4 and 4 were measured and normalized to Abelson murine leukaemia viral oncogene homologue 1 (Abl).
Results: The transcriptional level of Foxp3 was significantly lower in patients with IgAN in comparison to HC (0.82±0.30 vs 1.05±0.37, p=0.041), while those of IL-17 and of its regulatory factor RORc were slightly, but not significantly increased (IL-17 1.17±1.07 vs 1.05±0.41 in HC, RORc 1.25±0.85 vs 1.14±0.71 in HC). A significant correlation was found between IL-17 and RORc mRNAs values ($p<0.0001$). Transcriptional levels of TGF beta1 were similar to controls in patients with IgAN and were directly correlated with RORc (p=0.0015) and IL-17 (p=0.031) mRNAs. Patients with proteinuria > 1 g/day (median 1.62 g/day) had Foxp3 levels

lower than those in phase of clinical inactivity (proteinuria < 1 g/day, median 0.18 g/day), p=0.06.
ROR-C level displayed a significant inverse correlation with e-GFR values (p=0.06).
Of interest, a significant inverse correlation was found between Foxp3 and TLR4 expression (p=0.022).
Conclusions: In conclusion, these results suggest that in patients with IgAN there is a functional defect in Tregs which appears to be correlated with signs of hyperactive innate immunity. TGF beta 1 in these patients seems to favour the expansion of TH17 cells. Some trend correlation between circulating TH cell subset balance and known risk factors, as levels of proteinuria, suggests a potential interest in further investigation in this area.

Su034 CD40 CROSS-LINKING INDUCES MIGRATION OF RENAL TUMOR CELL THROUGH NFAT ACTIVATION AND β1 INTEGRIN RIORGANIZATION

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Introduction and Aims: CD40 crosslinking play an important role in regulating cell migration, adhesion and proliferation in renal cell carcinoma (RCC). CD40/CD40L interaction on RCC cells activates different intracellular pathways, however the molecular mechanisms leading to cell scattering are not clearly defined. Aim of our study was to investigate the principal intracellular factors activated by CD40 ligation and their specific involvement in RCC cell migration.
Methods: RCC cell lines were isolated from kidney tissue samples of patients affected by RCC and subsequently stimulated with CD40L.
Results: We found that CD40-CD40L interaction induced cell proliferation through a cytoskeleton reorganization and integrin b1 distribution, while did not affect apoptosis. Interestingly, CD40 ligation did not activate the pathway involving phosphatidylinositol 3-kinase (PI3K), Akt and p70 ribosomal S6 kinase, while increased the phosphorylation of extracellular signal-regulated kinase (ERK), c-Jun NH(2)-terminal kinase (JNK) and p38 MAPK. Furthermore, CD40 crosslinking activated different transcriptional factors on RCC cell lines: AP-1, NFkB and some members of the NFATs family. In particular, the specific inhibition of NFAT factors by cyclosporine A, completely blocked RCC cell motility induced by CD40 ligation.
Conclusions: These findings support the hypothesis that CD40 ligation induces cell scattering through cytoskeleton reorganization and activation of different intracellular signalling pathways, in particular the NFATs family. These factors could represent a potential therapeutic target in the setting of patients with metastatic RCC.

Su035 INVOLVEMENT OF ENDOPLASMIC RETICULUM STRESS IN DECREASED INTEGRIN α3β1 EXPRESSION AND APOPTOSIS OF PODOCYTES BY HUMAN SERUM ALBUMIN

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Introduction and Aims: The β1 integrin undergoes N-glycosylation from endoplasmic reticulum (ER) to Golgi apparatus. This transition represents as a regulation of integrin maturation and function for cell surface expression. In the ER, the most prevalent, incompletely glycosylated form of β1 has a mass of 105 kDa (p105). Mature β1 that forms β1-6 branching of N-linked oligosaccharides within medial/trans Golgi has a mass of 125 kDa (p125). We found in our previous report that the amount of daily protein loss was negatively correlated with integrin α3β1 expression of podocytes. Here we investigated whether albumin overload induced ER stress could decrease the expression of integrin α3β1 and cause apoptosis in podocytes.
Methods: Podocytes were exposed to medium alone or in the presence of 10 mg/ml endotoxin-free human serum albumin (HSA) at 37°C. At the end of incubation, expression of integrin α3 subunit and β1 subunit mRNA and protein in cell lysates were analyzed by RT-PCR and Western blotting. To investigate the effect of albumin overload on ER stress, GRP78, CHOP, p-eIF2α and eIF2α were analyzed by Western blotting. The uptake of albumin was detected by immunofluorescence in confocal microscopy and Western blotting. The flow cytometric assay was performed to examine apoptosis.
Results: We found that podocytes can uptake albumin after one hour of albumin loading at 37°C. GRP78 level was increased after one hour of albumin loading. p-eIF2α/eIF2α level was increased 6 hours after albumin loading. CHOP level was increased 12 hours after albumin loading. The mRNA expression of integrin α3 and β1 subunits was no change after albumin loading. The protein level of α3 integrin was decreased one hour after albumin loading. The p105 and p125 protein levels of β1 integrin also became to be deceased one hour after albumin loading. Apoptosis was increased 24, and 48 hrs after albumin loading.
Conclusions: In this study, we found that albumin uptake can induce ER stress in podocytes. ER stress may be associated with decreased protein expression of α3 and immature (p105) and mature (p125) β1 integrin of podocytes. ER stress may induce apoptosis of podocytes. Anyway, proteinuria may cause decrease of integrin α3β1 protein expression and apoptosis by ER stress.

Su036 TGF-β1-INDUCED ANCHOR SITES IN CELL JUNCTIONS DETECTED BY AFM

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Introduction and Aims: Intracellular contractile forces are essential for all kinds of motility, from locomotion of whole organisms down to the migration of single cells. Force generating actomyosin not only controls the migration of immune or tumor cells, but also steers stem cell differentiation. Hence, mechanical forces might also be important for epithelial to mesenchymal transdifferentiation (EMT), a process linked to fibrosis and loss of organ function as in diabetes, inflammation or tumor progression.
Methods: In the current study, the EMT of TGF-b1-stimulated proximal tubule epithelium (NRK-52E cells) is analyzed by atomic force microscopy (AFM). AFM performed stiffness measurements and showed true, three dimensional height images. AFM as well as immunofluorescence antibody staining were used to show morphological changes and force generation during EMT. ECIS (Electric Cell-Substrate Impedance Sensing) measurement was applied to determine the electrical resistance.
Results: AFM showed an detailled cell surface compared to contrast phase microscopy. Furthermore AFM was able to detect compensatory nodular protrusions at cell junctions, where stress fibers of neighboring cells perform a kind of “handshake”. An elevated cellular stiffness, which indicates a higher isometric tension was shown in living cells underoing EMT. The phosphorylated form of the myosin light chain (pMLC) was used as an indicator for force generation and showed an increased assembly along stress fibers in cells undergoing EMT. These forces lead to a disassembly of the adherence junction protein N-Cadherin. Additionally, the transepithelial resistance showed only a moderate loss. Since inhibitors of force generation were effective to block the nodular protrusions, tension has a main influence on the process of epithelial transdifferentiation.
Conclusions: The nodular protrusions may serve as anchor sites for tensile force along stress fiber formations.

Su037 ROLE OF CALRETICULIN (CRT) BY REGULATING CALCIUM HOMEOSTASIS IN THE OSMOTIC STRESS ADAPTATION OF THICK ASCENDING LIMB OF HENLE'S LOOP (TALH) CELLS

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Introduction and Aims: Calcium binding chaperones of endoplasmic reticulum (ER) lumen such as CRT, and GRPs are generally induced as stress response proteins to protect the cell against various toxic insults. They are also involved in intracellular calcium homeostasis due to accumulation of large amounts of calcium without an excessive increase in the free ER intra-luminal calcium concentration. Especially, CRT, a calcium buffer in the ER, is assumed to regulate the intracellular calcium homeostasis and ER Ca^{2+} storage capacity.
Our former studies demonstrated that epithelial cells of thick ascending limb of Henle's loop (TALH) normally exposed to variable osmotic stress conditions showed a significant down-regulation of calcium binding ER proteins especially, CRT as a part of their reaction to NaCl osmotic stress. Moreover intracellular free calcium concentration quantified using fura-2/AM fluorescent dye showed a time dependent increase in intracellular free calcium concentration under NaCl stress. Parallel to increase in intracellular free calcium concentration, RT-PCR analysis showed a significant decrease of CRT mRNA in a time dependent manner under hyper-osmotic stress. The aim of our study is to further investigate the role of CRT in TALH cells under NaCl stress.
Methods: To investigate the role of CRT in terms of its calcium binding nature, we treated TALH cells with heparin, to block the IP3R mediated calcium release, and exposed to hyper-osmotic stress with NaCl. Western blot analysis was done to analyse the protein expression. MTT assay was performed to check the percent viability of cells. To further evaluate the role of CRT in osmotic stress, we over-expressed wild type and mutant (without its calcium binding domain) CRT separately, with subsequent NaCl stress.
Results: In contrast to NaCl stress alone, our data showed that the CRT expression was not altered in cells treated with NaCl combined with heparin. However, cell viability assay showed significant increase in cell death upon exposition of heparin treated cells to NaCl stress compared to NaCl or heparin treatment separately. Furthermore, MTT assay performed with transfected cells showed a significant decrease in viability (%) of wild type CRT expressing cells when treated with NaCl while cells over-expressing mutant CRT showed no significant difference of viability compared to non-transfected cells.
Conclusions: It is concluded that CRT expression significantly impacts the survival of TALH cells under osmotic stress by regulating calcium homeostasis.

Su038 ERP57 INHIBITION AND REDUCTION OF PROTEIN DISULFIDE BRIDGE REDUCE THE ECM SYNTHESIS AND EXCRETION IN TRANSFORMED RENAL INTERSTITIAL FIBROBLAST

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Introduction and Aims: ER-stress proteins ERP72, ERP57 and PDI as members of disulfide isomerase family, they catalyze the formation and breakage of disulfide bonds between cysteine residues within proteins as they fold. For the synthesis and correct folding of ECM proteins especially collagen IV, LAMA1/2, and FIN the disulfide bridges play a central role. Without formation of these disulfide bridges the proteins are misfolded and degraded via the proteasomal pathway.
Methods: We performed differential proteomics analysis with TGFβ-1 transformed TK173 cells (renal interstitial fibroblast) and the non-treated control. Differential two dimensional gel electrophoresis combined with mass spectrometry analysis were used to screen the protein expression alteration in transformed cells. Western blot and Immunohistochemistry were used to confirm the data in tissue sections. PDI activity and inhibition assay were used to highlight the role of ERP57 in ECM synthesis and accumulation.
Results: The proteomics analysis revealed that the expression of more than 30 proteins was altered in TK173 treated with TGFβ-1. Interestingly a large part of the identified proteins could be classified in three categories: The first category grouped the proteins that have been described to be involved in fibrogenesis (e.g. FIN, ACTA2, VIN, VIM, DES, KRT), the second protein group were involved in oxidative stress pathway and the third category involved protein markers of the ER-stress pathway (GRP78, GRP94, ERP57, ERP72, PDI and CALR) which were highly up-regulated in FKIF cells. The up-regulation of ER-stress proteins as fibrosis marker was also investigated in human tissues. RT-PCR and Immunohistochemistry staining of kidney biopsies from patients suffering from kidney injuries showed an increase in the expression of the investigated proteins correlating with the progression of fibrosis. The evaluation of kidneys with signs of renal interstitial fibrosis revealed cells in the tubulointerstitial space in the medualla and the cortex positively stained for ERP57, ERP72 and GRP78, CALR. Moreover there was a clear correlation between the grade of fibrosis and the expression level of the ER-stress proteins. An inhibition of the protein disulfide isomerases with bacitracin significantly affected the ECM synthesis, secretion and accumulation in TGFβ1 transformed TK173. Our study shows an important role of PDI, ERP72 and ERP57 in the transformation of TK173 to myofibroblast.
Conclusions: Our data reveal a central role of protein disulfide isomerases in correct folding and stabilisation of ECM proteins.

Su039 NERVE GROWTH FACTOR (NGF) AND NGF-RECEPTORS EXPRESSION IN IMMATURE HUMAN PODOCYTE (IHP) CELL LINE

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Introduction and Aims: The podocytes are highly specialized cells with foot processes bridged by the slit diaphragm, establishing the selective permeability of the glomerular filtration barrier. This structural integrity is also supported by several factors (cytokines, adhesion molecules and growth factors). Among these, the NGF, a protein belonging to the neurotrophin family, involved in development, survival and function of nervous cells, could play an important role. NGF induces its biological effects through the activation of two distinct receptor (NGF-R): the high affinity tyrosine kinase A (TrkA) receptor and the low-affinity NGF-R (p75). Our recent data have demonstrated that NGF and NGF-R are expressed in the human kidney and that their levels are altered in a wide spectrum of renal disorders. Despite a suggestive role of NGF could be hypothesized considering the common biological features between podocyte and neural cells, no evidences on the exact role played by the neurotrophin in maturation, structural changes and functional activity of podocyte are available.
Aim of the study was to characterize NGF/NGF-R system on IHP. For this purpose we used a conditionally IHP cell line maintained in a proliferative and undifferentiated state at permissive conditions.
Methods: The IHP, obtained from human kidney specimen and transfected with the tsSV40 gene construct, was cultured in RPMI-1640 medium supplemented with Fetal Bovine Serum, Penicillin/Streptomycin and Insulin-Transferrin-Selenium at 33°C. Cells were grown to 80% of confluence and then trypsinized and reseeded. After 24 hours NGF and NGF-R podocyte expression was evaluated with RT-PCR, Western Blotting assay (WB), Immunofluorescence microscopy (IFm) and Transmission Electron Microscopy (TEM).
Results: IHP constitutively express NGF, trkA and p75 mRNA as evidenced by RT-PCR. Analysis of proteins levels by WB revealed a band at 140 kDa which corresponds to TrkA, whereas other different detectable bands at 75 kDa, 50-55 kDa and 25-30 kDa, corresponding to p75 and its fragments, respectively, were observed. These fragments derive from a proteolitic reaction, since no alternative splicing was observed. Similar results were obtained with IFm and TEM of p75. In particular TEM showed positive

NGF-R staining and revealed that IHP express p75 full-length protein on cytoplasmatic membrane while its cytoplasmatic fragment was visible in the perinuclear region and on nuclear membrane.
Conclusions: Our findings indicate that IHP constitutively express mRNA for NGF and NGF-R. TrkA receptor and full-length p75 are expressed on cytoplasmatic membrane whereas other isoforms of p75 are localized on nuclear membrane and in the perinuclear region. The latter localization could suggest the p75 involvement as transcriptional factors in the maintenance of a proliferative state of immature podocytes. Further experiments are in progress to better clarify the role of NGF and its receptors in the functional activity of both immature and mature podocytes.

Su040 ISOFORM-SPECIFIC FUNCTIONS OF RHO-KINASES IN RENAL TUBULAR EPITHELIAL CELLS

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Introduction and Aims: Experimental inhibition of the Rho-kinases ROCK1 and 2 has shown promising effects in kidney disease, cardiovascular disorders and neuroregeneration, and is therefore currently subject of several preclinical and clinical trials. The two kinase isoforms ROCK 1 and 2 are the main downstream mediators of the small GTPase RhoA, which is crucial for cytoskeletal organisation. Until now, studies on ROCK inhibitors did not focus on specific qualities of ROCK 1 and 2 despite recent evidence that there are isoform-specific functions. This fact is not only due to the high homology between the two kinases, it also reflects the lack of isoform-specific inhibitors.
Methods: We used a siRNA-based approach to selectively knockdown ROCK isoforms in human primary tubular epithelial cells and in the renal epithelial cell line HKC-8. Thereby we were able to investigate isoform-related changes in cytoskeletal organisation and gene expression. The results were compared to cells treated with Y-27632, an inhibitor of both ROCK isoforms or new ROCK 2 specific inhibitors.
Results: We observed changes in organisation and distribution pattern of F-actin, focal adhesions and the cell adhesion molecules E- and N-cadherin under selective knockdown or inhibition of ROCK 1 and 2. Knockdown of ROCK 1 led to a strong increase in cortical F-actin and decreased focal adhesions, whereas selective ROCK 2 knockdown or ROCK 2 specific inhibition caused F-actin disorganisation and discrete invaginations in the cellular membrane. Inhibition of both isoforms with Y-27632 heavily modified membrane shape and totally disrupted F-actin and focal adhesion organisation.
Gene expression of two profibrotic proteins, Connective Tissue Growth Factor (CTGF) and Fibronectin, was strongly inhibited by the ROCK inhibitor Y-27632. A partial reduction was observed upon interference with ROCK2 whereas downregulation of ROCK1 was not sufficient to inhibit protein expression.
Conclusions: In conclusion, we found evidence that there are specific functions of both ROCK isoforms in renal epithelial cells. Consequently, isoform-specific ROCK inhibition needs further investigation in disease models.

Su041 EXPRESSION OF NKG2D LIGANDS IN HUMAN RENAL CELL LINES

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Introduction and Aims: The MHC class I-related Chains A and B (MICA and MICB) and UL-16 binding proteins (ULBP1-5) are distantly related homologs of MHC class I proteins that are induced upon cellular distress conditions. MICs and ULBPs are recognized by the NKG2D activating receptor, which activates NK cells and costimulates effector T-cell subsets, leading to cytotoxic lysis of the stressed target cells. The metalloproteinase ADAM-17 and the chaperon ERbP-5 are able to shed MIC A/B from the cell membrane. Recently a role in graft versus host disease and transplant rejection could be shown.
Methods: Three human renal cell lines, mesangial cells, podocytes and proximal tubular epithelial cells (HK2), were analysed for expression of NKG2D-ligands. MIC A/B expression was tested by real-time PCR and FACS-analyses. The presence of ADAM-17 and ERbP-5 was confirmed by real-time PCR. The amount of soluble MIC A/B was measured by ELISA-experiments. In addition, to examine the interaction between renal cells and CD8+ T lymphocytes cytotoxicity-assays were performed.
Results: Mesangial cells (HMC) and podocytes expressed only low amounts of MIC A/B-RNA whereas HK2 cells showed a marked expression. Surface expression of NKG2D-ligands was analyzed by flow cytometry using monoclonal antibodies to MICA/B and ULBP1–3 and significant differences in NKG2D-L expression patterns among the cell lines and inducing stimuli could be observed. In HMC ULBP3 protein was most prominent. In podocytes ULBP2, ULBP3 and MICA was comparable high. In contrast, in HK2 cells MICA was the most prominent protein. In the supernatant of all cell lines a significant release of soluble MICA was noted. HK2 cells also released considerable amounts of MIC B. Stimulation with LPS induced the synthesis of MIC A protein in HMC only. Investigation of ERbP-5 and ADAM17 showed a higher expression in HK2 compared to HMC and podocytes. In cytotoxicity-assays only HMC were lysed, in contrast podocytes and HK2 cells which were not easily affected. The specific lysis could be significantly blocked using antibodies directed against NKG2D and MIC A/B.
Conclusions: In conclusion, we show for the first time a functional expression of NKG2D ligands by renal cells which might play a role for local activation of infiltrating immune cells into damaged kidney tissues.

Su042 mTOR-SIGNALING PATHWAY IS INSTRUMENTAL FOR CARDIOPROTECTIVE EFFECTS OF β-ESTRADIOL IN REVERSING MALADAPTIVE MYOCARDIAL HYPERTROPHY

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Introduction and Aims: Targeting the mTOR pathway in the prevention of myocardial hypertrophy secondary to uremia or for regression of already established myocardial hypertrophy after transplantation could represent a novel therapeutic option. The underlying molecular mechanisms and especially the influence of estrogen remain sparcely investigated. Aim of our study was therefore to elucidate how β-estradiol (E2) may modulate PI3K/Akt signaling in response to physiologic and pathologic hypertrophic stimuli and to determine consequences of mTOR inhibition in the context of the "female cardiomyocyte".
Methods: Female HL-1 cardiomyocytes were treated with "physiologic" (IGF-1) and "pathologic" (ET-1) stimuli in the presence or absence of E2 or mTOR inhibitor rapamycin. Cell size was determined by immunocytochemistry and FACS-analysis. Signal transduction was assessed by immunoprecipitation with anti-mTOR polyconal antibodies and westernblotting using phospho-specific antibodies against Akt to monitor TORC2-activity, Erk and p70S6K to monitor TORC1-activity. Taqman was applied to investigate genomic changes.
Results: E2, IGF-1 and ET-1 induced phosphorylation of Akt and p70S6K in a time-dependent manner. Long-time pretreatment with E2 lead to a reduced basal phosphorylation of p70S6K downstream of mTOR. E2 reversed the increase in p70S6K-phosphorylation upon ET-1-stimulation, whereas co-treatment of E2 with IGF-1 increased p70S6K-phosphorylation. mTOR-complex formation with raptor and rictor was increased by E2-cotreatment. ANP-mRNA-expression increased significantly with E2-cotreatment and most markedly with additional ET-1-stimulation. Rapamycin inhibited cardiomyocyte hypertrophy and p70S6K-phosphorylation by IGF-1 and ET-1 irrespective of E2-cotreatment, whereas positive feedback loop towards Akt-phosphorylation was differentially regulated by rapamycin dependent on the presence or absence of E2.
Conclusions: Rapamycin effectively inhibits female cardiomyocyte hypertrophy irrespective of the presence or absence of E2. However, E2 differentially modulates TORC1 and TORC2 activities dependent on the nature of the hypertrophic stimulus and rapamycin treatment. This could impact different cellular functions like apoptosis and autophagy which are under current investigation.

Su043 RHO-KINASE INHIBITORS INCREASE RENAL ENDOTHELIAL CELL MIGRATION

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Introduction and Aims: Defective microcirculation is a prominent feature in chronic renovascular disease. Damage of renal microvessels and their eventual loss (microvessel rarefaction) influence the progression of renal injury. There is experimental evidence in non-renal endothelial cells that Rho/Rho-kinase activation is involved in vessel destabilization and retraction. Therefore, it was the aim of the present study to investigate the effect of Rho-kinase inhibitors in renal microvascular endothelial cells. To address different aspects of the angiogenic process, different in vitro models of migration, sprouting and tube formation were analyzed.
Methods: A murine glomerular endothelial cell line, glEND.2 was used for all studies. Chemotaxis of the cells was determined using a transmigration Boyden-chamber assay with FCS or fibronectin as chemoattractants. To analyze sprout formation, cells were incubated overnight in hanging drops in order to form defined cell spheroids consisting of 400 cells per spheroid. Spheroids were then embedded in collagen gels and sprouting was quantified after 3 days. Furthermore, endothelial cells were allowed to migrate out of cell spheroids on glass plates coated with different extracellular matrices. Tube formation was analyzed in cells cultured on matrigel. Cell-cell-contacts and the cytoskeleton were visualized by immunocytochemical detection of VE-cadherin and F-actin fibers, respectively. Cell proliferation was assessed by BrdU incorporation.
Results: Inhibition of Rho-kinases by H1152 strongly reduced cell spanning F-actin fibers and loosened cell-cell contacts as determined by VE-cadherin immunofluorescence. VE-cadherin protein levels, however, were not influenced by Rho-kinase inhibitors implicating that the protein was internalized and not degraded. These alterations in cell morphology differentially affected the motility of glEND.2 cells: Migration of endothelial cells out of the spheroid into the collagen gel or on fibronectin-coated plates was increased. Furthermore, formation of structures on matrigel was improved by the Rho-kinase inhibitor. By contrast, no effect of Rho-kinase inhibition was observed in the chemotaxis assay, which primarily detects single cell migration. These effects were mimicked by a specific inhibitor of the Rho-kinase isoenzyme ROCK2.
Conclusions: Comparing different assays of endothelial cell migration and structure formation, our data indicate that cell-cell contacts as formed in spheroids are essential for Rho-kinase inhibitors to increase endothelial cell motility. Furthermore, the capacity of the cells to form structures on matrigel was strongly enhanced. These data suggest that direct effects of Rho-kinase inhibitors on endothelial cells may contribute to their renoprotective effects.

Su044 K-RAS ISOFORM MODULATES EXTRACELLULAR MATRIX SYNTHESIS AND PROLIFERATION IN FIBROBLASTS

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Introduction and Aims: Chronic renal insufficiency is characterized by an excessive accumulation of extracellular matrix (ECM) in the interstitium and/or in the glomerular mesangium. Accumulation of ECM and proliferation are some of the main mechanisms involved in fibrosis. Transforming growth factor beta 1 (TGF-beta 1) has a relevant role in the origin and maintenance of glomerulosclerosis and tubule-interstitial fibrosis (TIF). Ras proteins are small GTPases with prooncogenic effect that act as transducers of extracellular signals that regulate cell survival, growth and differentiation. There are three main Ras isoforms: H, N and K-Ras. Previous studies from our laboratory showed that TIF is associated to increases in Ras and Erk and also demonstrated that H- and N-Ras modulate several fibrotic processes. TGF-beta and Ras signaling pathways are close related: TGF-beta 1 overcomes Ras mitogenic effects and Ras counteracts TGF-beta signaling. We have studied the role of K-Ras isoform and the involvement of the MAPK-Erk pathway in TGF-beta 1-induced ECM synthesis and proliferation.
Methods: For this purpose, we have generated and characterized knockout (KO) mouse embrionary fibroblasts for K-Ras isoform (K-ras-/-) obtained from mating of K-Ras heterozygote mice (K-ras+/-) (KO mice for K-Ras are not viable, and embryos die at day 11 post coitum). ECM synthesis, measured by the expression of fibronectin and collagen type I, and phospho-Erk expression were evaluated by western blot. Fibroblast proliferation was analyzed by crystal violet assay.
Results: ECM synthesis was increased in K-Ras KO fibroblasts in basal conditions. Treatment with TGF-beta 1 induced a significant increase in fibronectin and collagen type I expression. Erk activation (phospho-Erk expression) was similar in both K-ras-/- and control fibroblasts, but its expression was decreased after TGF-beta 1 treatment in KO fibroblasts. Cell proliferation is reduced in K-Ras KO fibroblasts with respect to controls.
Conclusions: This is the first study that shows the involvement of K-Ras isoform in fibrotic processes. Our findings show that K-Ras is involved in ECM synthesis and fibroblast proliferation, two of the main mechanisms involved in fibrosis. Moreover, K-Ras isoform seems to be necessary for TGF-beta-induced Erk activation in mouse embrionary fibroblasts.

Su045 IMPACT OF GENDER AND SOLUBEL GUANYLATE CYCLASE STIMULATION ON RENAL RECOVERY FOLLOWING RELIEF OF UNILATERAL URETERAL OBSTRUCTION

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Introduction and Aims: Pharmacological stimulation of soluble guanylate cyclase (sGC) and subsequent cGMP production have recently shown to be renoprotective. Estrogen has been found to promote endothelial-dependent NO effects. In this study, the specific sGC stimulator Bay 41-8543 was administered to male and female rats after relief of unilateral ureteral obstruction (UUO) in order to analyze the effects of gender and enhanced NO/cGMP signalling on renal disease severity and restoration.
Methods: Age-matched male (M) and female (F) Sprague-Dawley rats underwent UUO. After 5 days, obstruction was relieved and the animals were randomly assigned to UUO and UUO+Bay 41-8543 (10 mg/kg body weight/day) (n=10 per group) for 7 days. Then, effects on plasma cGMP levels, systolic blood pressure and indices of histological renal disease severity were determined. The symbol * indicates $p<0.05$.
Results: UUO rats showed elevated systolic blood pressure and marked tubular atrophy, tubular apoptosis, tubulointerstitial macrophage infiltration and fibrosis. Renal injury was more severe in M versus F (tubular diameter 41±0.3 vs 36±0.5 μM*; tubulointerstitial volume 19.3±0.5% vs. 15.4±0.7%*; matrix protein expansion 10.3±1% vs. 6.9±0.9%* and collagen IV deposition 5.4±0.5% vs. 3.8±0.5%*). Bay 41-8543 increased significantly plasma cGMP levels (M: +104%*, F: +412%*). This went along with significant reductions in systolic blood pressure (UUO+Bay 41-8543 vs. UUO, M: 112±4 vs 145±8 mmHg*, F: 117±3 vs 145±8 mmHg*), tubular diameter (M: -20%*, F: -17%*), tubular apoptosis (M: -67%*, F: -76%*), tubulointerstitial macrophage infiltration (M: -65%*, F: -50%) and fibrosis (tubulointerstitial volume M:-38%*;F: -29%; matrix protein expansion M: -60%*, F: -16%; collagen IV expression M: -52%*, F: -19% and alpha-smooth muscle actin expression M: -69%*, F: -49%).
Conclusions: Renal disease after relief of UUO is more severe in male than in female animals. The sGC in female UUO-rats is more sensitive to stimulation and produced more cGMP in response to Bay 41-8543. Enhancing NO/cGMP signalling with Bay 41-8543 significantly accelerated restoration of renal architecture following relief of ureteral obstruction in both gender.

Su046 THE S1P1 AGONIST AUY954 ACTIVATES eNOS VIA AKT-MEDIATED PATHWAY

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Introduction and Aims: In the mammalian cardiovascular system, nitric

oxide (NO) plays a pivotal role in the maintenance of vascular homeostasis. The endothelial isoform (eNOS) is a key determinant of blood pressure or other fundamental response in the vascular wall. Sphingosin-1-phosphate (S1P) is a bioactive sphingolipid which regulates diverse physiological processes by serving as ligand to G-protein coupled S1P receptors and recently identified as eNOS activator, whereas the S1P1 receptor play a key role. A newly developed mono-selective S1P1 modulator – AUY954 – should be investigated in this study on its potential to active eNOS.
Methods: Human umbicial vein endothelial cells (HUVEC) were used. S1P receptor mRNA was detected via RT-PCR. Phosphorylation of eNOS was detected by Western Blot technique. Akt phosphorylation was measured using Luminex™ technology. The production of NO was measured in DAF2-DA labeled cells.
Results: AUY954 [0.1 – 10 μmol/L] led to a significant and dose-dependent increase in NO liberation in HUVEC detected by fluorescence increase in DAF2-DA labeled cells. This effect could be blocked by L-NAME [100 μmol/L], an inhibitor of NO synthases. AUY954 significantly increase Ser1177 phosphorylation time- and dose-dependently. S1P1 and S1P3 receptor are the most prominent S1P receptors in HUVECs and only a weak expression of S1P2 was detectable. So, we next investigated a specific, non-selective inhibitor of S1P1/3 receptor, VPC23019 which significantly block the AUY954-induced NO liberation. Cay10444, a selective S1P3 antagonist doesn't have any effect. This indicates a S1P1 specific effect. Furthermore, AUY954 is able to stimulate Akt phosphorylation in a significant and time-dependent manner. The co-stimulation of AUY954 with a specific Akt inhibitor blocked the NO production in DAF2-DA labeled HUVECs.
Conclusions: Here, we showed that the selective S1P1 receptor agonist AUY954 is a very potent activator of endothelial NO synthase via a Akt-mediated process which results in increased NO liberation of human endothelial cells. Therefore, eNOS/NO activation by AUY954 might play a role in cardiovascular response by promoting endothelial cell survival, proliferation and migration.

Su047 EPOα AND DARBEPO-MEDIATED ERYTHROPOIETIN RECEPTOR DESENSITIZATION/RESENSITIZATION

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Introduction and Aims: Anemia associated with chronic kidney disease (CKD), is primarily a consequence of inadequate erythropoietin (EPO) production by the kidneys. Epoetin alfa (EPOα) and darbepoetin (DarbEPO) are two EPO receptor agonists indicated for the treatment of anemia in patients with CKD. The clinical efficacy of the drugs is tightly dependent on the functional responsiveness of surface membrane EPO receptors: the stimulation by EPO induces the receptor tyrosine phosphorylation and the subsequent activation of STAT5 transcription factors. The intracellular signalling activated by EPO receptor stimulation are rapidly regulated by desensitisation mechanisms which control receptor responsiveness to agonists.
In the present work we investigated the effects of the two different epoetin derivatives, EPOα and DarbEPO in HUVEC cells: in particular the effects of different concentrations of both agonists at different cell exposure times on the kinetics of receptor desensitisation and resensitisation were investigated.
Methods: EPO receptor-mediated STAT5 phosphorylation was investigated by using an ELISA kit (RayBio® Cell-Based Stat5 (Tyr694), RayBiotech Inc., USA). Concentration and time-dependence STAT-5 phosphorylation assays, EPO receptor desensitisation/resensitisation experiments and EPO receptor immunoblotting analysis were conducted.
Results: We demonstrated that both agonists activated STAT5 phosphorylation in a concentration and time dependent manner with similar kinetics.
Following receptor exposure to both agonists for different times, EPO receptor underwent to desensitisation with a maximal effect after 60 minutes. The comparison of desensitisation kinetics did not show any significant differences in EPOα or DarbEPO-induced desensitisation, demonstrating the two drugs have a similar pattern in receptor regulation.
The recovery of receptor responsiveness following desensitisation was strictly dependent on agonist concentration and on the time of receptor agonist exposure (desensitisation time). Actually, the receptor functional recovery appeared to be faster when the receptor was activated by low agonist concentrations (1U/ml vs 3U/ml) and desensitised for short time (18 min). The comparison between resensitisation kinetics obtained with the two agonists clearly demonstrated that EPO receptor resensitisation following cell treatment with EPOα occurred with a faster kinetic than that obtained with DarbEPO. Finally, when DarbEPO was used in desensitisation experiments at high concentration (3U/ml), receptor functioning was completely lost and resensitisation of the signal did not occur.
Conclusions: These results suggest that the two epoetins differently regulate receptor resensitisation, demonstrating that the type of molecule as well as the time of receptor stimulation are crucial points in the control of receptor functional activity. If these data can explain clinical differences such as efficacy and stability of response, deserves further investigation.

Su048 GROWTH FACTOR DEPENDENT REGULATION OF MESENCHYMAL STEM CELL DIFFERENTIATION IN VASCULOPATHY AND FIBROSIS

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Introduction and Aims: Circulating bone marrow derived mesenchymal stem cells (MSC) may contribute as precursors to tissue fibrosis and vasculopathy in damaged native and transplant kidneys. We sought to investigate the impact of growth factors (GF) involved in fibrotic and neointimal lesion formation on MSC differentiation along fibroblast and smooth muscle like lineages.
Methods: MSC from patients with systemic sclerosis, a model disease characterized by fibrosis and vasculopathy, were incubated with CTGF, b-FGF, PDGF-BB, and TGFβ. The boyden chamber assay was used to analyze GF directed migration. GF induced proliferation was measured by BrdU incorporation. Distinct differentiated cell types after exposure to GF were identified by phase contrast microscopy, western blot analysis for the expression of vascular smooth muscle and fibroblast marker proteins, measurement of L-type Ca-channels to confirm a vascular smooth muscle cell (VSMC) like phenotype, and the SirCol-assay and qRT-PCR for collagen production to further characterize fibroblastic differentiation. Signal transduction analyses were carried out by western blotting.
Results: Each GF exerted characteristic effects on MSC. Migration and proliferation were stimulated most by PDGF-BB. CTGF had no detectable effect on differentiation of MSC. While b-FGF and PDGF-BB decreased expression of smooth muscle marker proteins and increased levels of fibroblast markers TGFβ had the opposite effect. Only b-FGF induced functional L-type Ca-channels in contrast to PDGF-BB and TGFβ that abrogated nimodipine sensitive Ca influx. Secretion of collagen was reduced by b-FGF and increased by PDGF-BB and TGFβ. This was reflected on the mRNA level by upregulation of col1a1 transcripts. We detected unique signal transduction signatures of the GF explaining the observed phenotypic differences.
Conclusions: We propose that b-FGF promotes a phenotype similar to proliferative VSMC. PDGF-BB and TGFβ induce fibroblast and myofibroblast differentiation, respectively. Our results suggest that the interplay of several GF coordinates the contribution of circulating MSC to fibrotic lesions and vasculopathy by controlling homing, proliferation and differentiation of MSC distinctively. Detailed knowledge of these mechanisms may lead to tailored therapeutic strategies in fibrotic and vascular renal diseases targeting GF signaling.

Su049 THE SERUM AND GLUCOCORTICOID-REGULATED KINASE 1 IN HYPOXIC RENAL INJURY

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Introduction and Aims: The serum- and glucocorticoid-inducible kinase 1 (SGK1) is a serine threonine protein kinase activated through the phosphatidylinositol 3-kinase (PI3-kinase) pathway and counteracts apoptosis. Protein expression and activation of SGK1 are increased in various models of cell stress such as hyperosmotic shock, ultraviolet radiation, heat shock, oxidative stress and hyperglycemia. The present study explored the role of SGK1 in hypoxia/ischemia induced apoptosis in the kidney.
Methods: Human embryoinic kidney (HEK) 293 cells were exposed *in vitro* to hypoxia/reoxygenation (H/R), and mRNA and protein levels of SGK1 were assessed. Furthermore, to analyze the role of SGK1, we investigated the impact of SGK1 overexpression on H/R induced apoptosis. *In vivo*, SGK1 expressions were analysed in renal ischemia/reperfusion (I/R) injury in rats, and apoptosis was assessed in *sgk1-/-* knock-out mice in comparison with wild-type mice.
Results: *In vitro*, H/R of HEK293 cells increased SGK1 transcript levels, SGK1 protein abundance and SGK1 phosphorylation. H/R injury further enhanced the percentage of apoptotic cells, an effect significantly blunted by prior SGK1 overexpression. In *vivo* renal I/R injury increased significantly SGK1 transcript levels and SGK1 protein abundance. I/R induced apoptosis, an effect significantly more pronounced in gene targeted mice lacking SGK1.
Conclusions: In conclusion, SGK1 is markedly up-regulated and counteracts apoptosis following H/R *in vitro* and ischemia *in vivo*. The kinase participates in the machinery fostering cell survival under those stress conditions.

Su050 DECREASED CONTENT OF HIF-1 AND VEGF-A ARE RESPONSIBLE FOR CAPILLARY RAREFACTION IN LOCOMOTOR BUT NOT POSTURAL MUSCLES OF RATS WITH CKD

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Introduction and Aims: Chronic kidney disease (CKD) negatively impact skeletal muscles. It could depend on poor muscle microcirculatory network and catabolic state mediated by cytokines, acidosis and decreased physical activity. Hypoxia inducible factor 1 (HIF-1) increases expression of genes engaged in angiogenesis, carbohydrate metabolism and transformation of glycolytic muscle fibers. HIF-1 contents were found to be constitutively higher in glycolytic muscles compared with oxidative ones. Our aim was to establish the association between capillary density, HIF-1 and VEGF-A mRNA and proteins content in functionally different skeletal muscles of rats with CKD.
Methods: Male Wistar rats weighing 350 g were divided into groups: CON (n=8) was sham-operated, while CKD5/6 (n=12) underwent subtotal nephrectomy. After 6 weeks blood was taken for lab tests. Muscle samples from locomotor, mainly glycolytic, the gastrocnemius muscle (MG) and postural, mainly oxidative, the longissimus thoracis muscle (ML) were stained for alkaline phosphatase to differentiate the capillaries. The capillary density (CD) and capillary per fiber (C:F) indexes were calculated. mRNA expressions of HIF-1 and VEGF-A were detected by RT-PCR. Protein expressions of HIF-1 was detected by Western blotting (WB).
Results: are shown in Tables 1 and 2.

Table 1. Baseline characteristic of groups

	CON	CKD5/6	P
Hemoglobin [g/dl]	14,1±1	12,2±1,7	<0,05
BUN [mg/dl]	28,07±4,0	63,8±17,8	<0,001
Haptoglobin [mg/ml]	0,7±0,4	1,63±0,6	<0,05
MCP-1 [pg/ml]	292±114	609±255	<0,05
AGE [AU]	4,6±0,9	5,1±0,6	NS

BUN - blood urea nitrogen, MCP-1 - monocyte chemotactic protein-1, AGE - advance glication end product.

Table 2. The capillary density and capillary per fiber indexes in muscles

	Gastrocnemius muscle			Longissimus thoracis muscle		
	CON	CKD5/6	p	CON	CKD5/6	p
CD [n/mm^2]	1105±302	581±206	<0,001	629±160	560±190	NS
C:F [n/n]	3,66±0,75	1,97±0,55	<0,001	2,49±0,57	1,99±0,57	<0,05

Compared with CON, HIF-1 mRNA (1 vs 0,48, p<0,05) and VEGF-A mRNA (1 vs 0,31, p<0,05) were significantly lower in MG from CKD5/6. There was no similar differences in ML. WB confirmed lack of HIF-1 protein contents in MG of CKD 5/6 group.
Conclusions: Decreased expression of HIF-1 and VEGF-A in glycolytic muscle fibers besides microinflammatory state and disuse might be a key player which is responsible for the process of capillary rarefaction in chronic kidney disease.

Su051 THE DINUCLEOTIDE UP4A INDUCE THE PRODUCTION OF REACTIVE OXYGEN SPECIES IN VSMCS VIA NADPH OXIDASE

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Introduction and Aims: There is a growing body of evidence that nucleotides and purinergic signaling play a crucial role in the development of vascular diseases such as atherosclerosis. Uridine adenosine tetraphosphate (Up4A), an endothelium-derived dinucleoside polyphosphate showed inflammatory promoting effects. The inflammatory response is triggered by NADPH oxidase by influencing the production of chemokines. Here, we investigated the effect of Up4A on activation of NADPH oxidase and the production of reactive oxygen species in vascular smooth muscle cells.
Methods: Rat vascular smooth muscle cells (VSMCs) were used for experiments. MCP-1 and Nox1/4 expression was measured by real-time PCR. Rac1 phosphorylation was measured by ELISA. Translocation of p47phox was detected by Western Blot technique in membrane fraction and cytosolic protein fraction of the cells. H2O2 production was examined by loading VSMCs with 5,6-chloromethyl-2',7'-dichlorodihydrofluorescein-dieacetate-acetylester (CM-H2DCFDA).
Results: Previous work proposed that the MCP-1 expression in VSMCs is controlled by the intracellular redox status. Thus, we investigated the effect of tiron, a vitamin E analog, which is able to significantly diminish the Up4A-induced MCP-1 expression (48±23% decrease, n=5). Furthermore, incubation of CM-H2DCFDA-labeled VSMCs with Up4A resulted in a significant and dose-dependent increase in DCF fluorescence intensity (n=6). Thrombin (8 IE/mL) was used as positive control. NADPH oxidase is composed of different subunits. The expression of Nox1 is increased after Up4A stimulation of the cells, whereas Nox4 expression is decreased. The activation of NADPH oxidase requires phosphorylation of Rac1 and translocation of p47phox to the plasma membrane. Up4A induced Rac1 phosphorylation in a significant and time-dependent manner. P47phox amount in membrane fraction increased time-dependently after Up4A stimulation, whereas the amount in the cytosolic protein fraction decreased, respectively.
Conclusions: In this study we could show that Up4A is influencing the vascular ROS production in a NADPH oxidase dependent manner. Therefore, the endothelial-derived factor Up4A is not only a potent vasoconstrictor but in addition a potent inductor of pro-inflammatory response in the vascular wall.

Su052 TGFβ1 SIGNALLING INDUCES DIFFERENTIATION OF VASCULAR SMOOTH MUSCLE CELLS BY Smad2/3-DEPENDENT UPREGULATION OF THE TRANSCRIPTION FACTOR Snail1

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Introduction and Aims: In arteriosclerosis, neointimal proliferation as well as neointimal fibrogenesis is associated with both growth factor-induced differentiation of smooth muscle cells and endothelial-to-mesenchymal transition. Previously we have shown that TGFβ1 reduces migration of vascular smooth muscle cells (VSMCs) during intimal fibrogenesis which correlates with upregulation of the cell-cell adhesion molecule N-cadherin. We now investigated the transcriptional pathway of TGFβ/N-Cadherin signalling in primary human VSMC in vitro.
Methods: We used different pharmacological inhibitors to modulated TGFβ signalling to define the role of TGFβ1 in the regulation of N-cadherin during differentiation of smooth muscle cells. Furthermore, we investigated the expression of different transcription factors during intimal fibrogenesis. Cultured primary human VSMCs were treated with TGFβ1. Pharmacological inhibitors against the kinase of the ALK5 TGFβ receptor type I (TβR1) were used to modulate TGFβ1 signalling. N-cadherin, β- and α-catenin concentrations as well as the activation of the PI3K-Akt signalling pathway was estimated by Western blotting.
Results: TGFβ1-treatment of cultured human VSMCs reduced their migratory activity as determined in transwell migration assays. This reduced migration correlated with increased concentrations of N-cadherin on mRNA level and on protein level as well as enhanced amounts of N-cadherin, β- and α-catenin complexes as analysed by immunoprecipitation. The TGFβ1 induced increase of N-cadherin was sensitive against inhibition of the ALK5 TGFβ receptor pointing to a signalling pathway which involves a TGFβ1-indued activation of the TGFβ receptor kinase. Activation of the TβR1 resulted in phosphorylation of the TGFβ1 effector proteins Smad2 and Smad3 which correlated with elevated expression of the transcription factor Snail1. Inhibition of TβR1 by pharmacological inhibitors eliminated Snail1 upregulation and subsequently reduced N-cadherin expression. The mRNA of other transcription factors, which have been shown previously to be involved in TGFβ1-induced activation of myofibroblasts, such as ZEB1, Sip1, Slug, Twist or E47, were not altered. Furthermore, we could demonstrate that the small GTPase RhoA was activated by TGFβ1. This induction of RhoA-GTP was supported by homophilic ligation of N-cadherin molecules which were increased by TGFβ1.
Conclusions: Our results show that TGFβ1 induces the differentiation of primary human vascular smooth muscle cells by Smad2/3-dependent upregulation of the transcription factor Snail1 which results in increased expression of cell-cell adhesion protein N-cadherin and the assembly of N-cadherin/catenin complexes. Our data suggest a role for TGFβ1 in promoting intimal fibrogenesis and atherosclerotic plaque formation by differentiation of vascular smooth muscle cells and induction of myofibroblast-dependent processes.

Su053 THE EFFECT AND MECHANISM OF C3-C3A-C3AR ON RENAL TUBULE EPITHELIAL MESENCHYMAL TRANSITION

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Introduction and Aims: The aim of this study is to investigate the effect and the mechanism of C3-C3a-C3aR pathway on renal tubular epithelial mesenchymal transition (TEMT).
Methods: Renal proximal tubular cell line HK-2 were cultured in vitro and divided into six groups: control group,TGF-β1 positive control group,10nM anaphylotoxin C3a group,50nM C3a group,100nM C3a and 50 nM C3a+1μM C3a receptor inhibitor (C3aR, SB290257)group. The expressions of E-cadherin,α-SMA,TGF-β1,CTGF,Col-Iand β-catenin were detected by immunohistochemistry, Western blot and RT-PCR. And scanning electron microscope (SEM) was used to observe the cellular morphological changes.
Results: Normal HK-2 cells showed the classic cobblestone shape, and SEM showed a loss of apical-basal polarity and microvilli of the cells stimulated by C3a, and the cells become elongated and invasive. Immunohistochemistry, RT-PCR and Western blot showed that C3a induced the loss of the epithelial marker E-cadherin and increased the expressions of α-SMA, TGF-β1, CTGF, Col-Iand β-catenin in dose-dependent and time-dependent manners. While C3aRA can partial block the decrease of E-cadherin and increase of α-SMA and TGF-β1.
Conclusions: C3a can induce tubular epithelial-to-mesenchymal transition, TGF-β1, β-catenin pathway may participate in the mechanism. And C3aRA can partial block the effect of C3a on renal tubular cells.

Su054 MYCOPHENOLIC ACID INHIBITS THE PHOSPHORYLATION OF NF-kB AND JNKs AND CAUSES A DECREASE IN IL-8 RELEASE IN H_2O_2 TREATED HUMAN RENAL PROXIMAL TUBULAR CELLS

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Introduction and Aims: Ischaemia-reperfusion injury is a common occurrence in renal transplantation and may affect the long-term survival of the allograft. Oxidative stress may play a crucial role in this, with reactive oxygen species formed during reperfusion causing direct cellular damage as well as activating pro-inflammatory signalling pathways.
Methods: We used a well known human proximal tubule cell line named HK-2 incubated with hydrogen peroxide (H_2O_2) for up to 2 hours, after pretreatment with (or without) different concentrations of mycophenolic acid (MPA). In addition, tissue lysates were prepared from whole Sprague Dawley (SD) rat kidneys (obtained from rats treated with mycophenolate mofetil for 4 days) that had been subjected to ischaemia (30 minutes) followed by reperfusion (5 minutes and 24 hours). Lysates from cells and from kidney tissue were subjected to Western blot analysis. Interleukin-8 (IL-8), was measured in the cell culture supernatant by using a conventional enzyme-linked immunosorbent assay (ELISA) kit. Cell viability was measured by the known ability of viable cells to reduce MTT (3-(4,5-Dimethyl-2-thiazolyl)-2,5-diphenyl-2H-tetrazolium bromide).
Results: HK-2 cells were subjected to H_2O_2 stress that resulted in phosphorylation of c-jun N-terminal kinases (JNKs) and the transcription factor NF-κB at Ser276, both of which have been associated with inflammation. IL-8 production also increased upon H_2O_2 stimulation. Pre-incubation of the cells with MPA resulted in reduced phosphorylation of both JNKs and NF-κB, and reduced IL-8 release in H_2O_2-stimulated HK-2 cells. MPA also reduced the H_2O_2-induced phosphorylation of p38 MAP (Mitogen-Activated Protein) kinase, the extracellular-signal regulated kinase 1/2 (ERK1/2), Akt/PKB kinase and the transcription factor CREB (Cyclic AMP Response Element Binding protein). In SD rat kidneys subjected to ischaemia-reperfusion an increase in both phospho-JNK1/2 and phospho-NF-κB (at Ser 276) was observed, which was reduced in the kidneys obtained from mycophenolate mofetil (MMF)-treated rats.
Conclusions: These results suggest that MPA may inhibit pro-inflammatory responses in the kidney by inhibiting activation of pro-inflammatory molecules in both the kidney and human renal proximal tubular cells subjected to oxidative stress.

Su055 RESVERATROL DECELERATES SENESCENCE AND PROTECTS AGAINST ADVERSE FUNCTIONAL CHANGES IN HUMAN PERITONEAL MESOTHELIAL CELLS IN VITRO

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Introduction and Aims: It has recently been shown that senescent human

peritoneal mesothelial cells (HPMCs) accumulate within omental tissue in vivo, and the fraction of these cells may increase during organismal aging. The features which senescent HPMCs exhibit, including deterioration of growth capacity, increased oxidative stress and DNA damage, hypertrophy, and up-regulated release of aniogenic and fibrotic cytokines, point to the adverse effect of the senescence on long-term maintenance of peritoneum integrity and function in dialysed patients. Despite these data, no effective strategies to cope with detrimental impact of senescent HPMCs have yet been developed. This study was designed to examine whether resveratrol, an active ingredient derived from grapes and red wine, may reduce the rate of HPMC senescence, and thus protect the peritoneal membrane against premature collapse.

Methods: Studies were performed on primary omental HPMCs (n=5), passaged until senescence in control medium and in medium containing 0.5 uM resveratrol. The expression of the proliferative antigen PCNA was determined by immunocytochemistry. Cell cycle distribution was assessed using flow cytometry. The expression of senescence marker – SA-beta-Gal was performed by cytochemistry. Cell apoptosis (subG1 fraction) was recorded using PI staining with flow cytometry. The generation of reactive oxygen species (ROS) was determined using H_2DCFDA. The magnitude of oxidative DNA damage (8-OH-dG level) was assessed with a competitive immunoassay. Antioxidative cell protection (SOD activity) was determined using an oxidase assay. Production of angiogenesis and fibrosis mediators (VEGF and TGF-beta1) was assessed by ELISA.

Results: The following results show the effect of resveratrol with respect to the control group in late-passage HPMCs. Replicative cell lifespan increased by 67±8% ($P<0.02$); the activity of SA-beta-Gal decreased by 26±11% ($P<0.03$); the expression of PCNA increased by 16±3% ($P<0.05$); the fraction of replicatively active cells in the S phase of the cell-cycle increased by 7±8% ($P<0.05$); the fraction of apoptotic cells decreased by 21±3% ($P<0.04$); the production of ROS decreased by 25±11% ($P<0.02$); the level of 8-OH-dG decreased by 28±7% ($P<0.02$); the activity of SOD increased by 13±2% ($P<0.05$); the secretion of VEGF decreased by 31±13% ($P<0.03$); the secretion of TGF-beta1 decreased by 9±12% ($P<0.05$).

Conclusions: Collectively, these results indicate that resveratrol has the ability to counteract the senescence-induced deterioration of HPMC growth and function, mostly as a result of decreasing oxidative stress, cell apoptosis and DNA damage. These findings in combination with beneficial effect of resveratrol on the release of angiogenic and fibrotic factors may make it a promising candidate as supplement to peritoneal dialysis fluids.

Su056 INHIBITORS OF MITOCHONDRIAL RESPIRATION INHIBIT TGFβ-INDUCED UPREGULATION OF PAI-1 AND FIBRONECTIN EXPRESSION

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Introduction and Aims: The anti-diabetic drugs metformin and phenformin have been shown to inhibit plasminogen activator inhibitor-1 (PAI-1) secretion in liver cells. Recently, inhibition of mitochondrial respiratory chain complex I has been shown as one of the cellular target of these drugs, accompagnied by an upregulation of AMP-activated kinase (AMPK) activity.

In previous work, we have shown that in renal fibroblasts, phenformin inhibits TGFβ signalling and blocks TGFβ induced PAI-1 and fibronectin (FN) expression through upregulation of the inhibitory SMAD7. In this study, we analyzed the effect of established inhibitors of mitochondrial oxidative phsophorylation (OxPhos) (rotenone, oligomycin, D942) and of ciglitazone and troglitazone, who recently have also been characterized as inhibitors of mitochondrial function, on cellular signaling pathways and TGFβ effects in renal fibroblasts and tubular epithelial cells.

Methods: Rat renal fibroblasts (NRK49F) and tubular epithelial cells (NRK52E) were treated with various inhibitors at non-toxic doses and TGFβ effects were analyzed by western blotting of FN and PAI-1. Signaling pathways were analyzed by wetern blotting using phosphor-specific antibodies for AMPK, ERK, p38-MAPK, and Akt. Expression of SMAD7 was analyzed by RT-PCR. Expression of PPARgamma was analyzed by RT-PCR and western blotting.

Results: D942 and oligomycin, but not rotenone activated AMPK and blocked TGFβ induced PAI-1 and FN expression in both 49F and 52E cells. Blockade of TGFβ effects was associated with an upregulation of SMAD7 mRNA.

Pre-treatment of cells with high glucose reduced both AMPK activation and the inhibition on TGFβ effects.

Treatment of cells with ciglitazone and troglitazone similarly activated AMOK and blocked TGFβ effect.

Exchange of glucose in the medium for galactose (which forces the cell to use only mitochondrial OxPhos to generate ATP) leads to increased baseline AMPK activation, reduced induction of PAI-1 and Fn after TGFβ treatment and is associated with increased toxicity of both ciglotazone and troglitazone.

Neither 49F nor 52E cells express PPARgamma receptor, as analyzed by RT-PCR and western blotting using 2 different rabbit monoclonal antibodies.

Conclusions: Our data suggest that mild inhibition of mitochondrial function followed by activation of AMP-kinase leads to inhibition of TGFβ effects, presumably through activation of SMAD7 expression.

Endogenous activation of AMPK by treatment with galactose showed similar effects, suggesting that the inhibition of TGFβ effects is due to modulation of mitochondrial function. Interestingly, both glitazones show a similar pattern of signalling (AMPK) and anti-fibrotic effects. We could not detect PPARgamma receptor in these cells, suggesting PPARgamma independent effects of glitazones.

Our data suggest that the functional status of mitochondria is involved in regulating TGFβ signalling in renal cells.

Su057 PROTEOME ANALYSIS OF URINE SAMPLES FROM PATIENTS WITH BALKAN ENDEMIC NEPHROPATHY

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Introduction and Aims: The complexity of the pathogenesis of the Balkan endemic nephropathy (BEN) makes its discovery and understanding very difficult. The limited value of the established markers demands analysis of new molecular parameters having a potential to be used for an early diagnosis and in prevention. Proteomics is the large-scale analysis of the proteins of biological samples and involves all aspects of protein analysis from the determination of amino acid sequence to the identification of functional partners and ultimately assignment to regulatory pathways. Protein markers for kidney and other diseases of major clinical importance (cancer, diabetes, atherosclerosis etc) have been detected in urine, and in many cases have been approved to be utilized in the clinic practice. Urine, excreted by the kidneys and indirectly reflecting kidney's function, can be easily and largely collected, therefore it could be an ideal substrate for biomarker research.

Methods: To improve visualization/detection of proteins and identification of Balkan endemic nephropathy (BEN) related biomarkers in urine, we combined the two-dimensional difference gel electrophoresis (2D-DIGE) and mass spectrometry (MS). Urine samples from healthy volunteers from BEN regions (32), BEN patients with proteinuria less than 150 mg/L (30), BEN patients with proteinuria more than 150 mg/L (30), German patients with acute renal failure (ARF) (26), patients with type 2 diabetes (DM) (30) and healthy volunteers from Germany (40) were analysed. The proteins were chloroform/methanol precipitated and analysed with two-dimensional electrophoresis (2-DE) and mass spectrometry. For several selected proteins the 2-DE results were confirmed by Western blot analysis.

Results: Comparative analyses of the different patients groups allowed the identification of proteins discriminating BEN with low and high proteinuria from the other groups. Among this proteins: β2-microglobulin (B2M), α1-microglobulin (AMBP), β-2-glycoprotein 1 (APOH), protection of telomeres protein 1 (POT-1), superoxide dismutase [Cu-Zn] (SOD1), mannose-binding lectin 2 (LMAN2) and lithostathine (REG1A) were found to be highly excreted in urine from BEN-patients in comparation to other groups.

Conclusions: Our study highlights several proteins in urine from BEN patients as potential prognosis/diagnosis markers which can provide new insights in the patho-mechanisms of the diseases.

Su058 NEUTROPHIL GELATINASE-ASSOCIATED LIPOCALIN (NGAL) CORRELATES *IN VIVO* WITH THE SEVERITY OF RENAL CANCER

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Introduction and Aims: Our knowledge on renal cancer biology is still lacking. This can represent a problem because today the incidence and prevalence of this neoplasia are steadily increasing. If on the one hand epidemiological studies have reported associations between cigarette smoking, obesity and genetic assessment and the future development of renal cancer, on the other hand it seems that also several endogenous molecules, such as hormones, cytokines and stress proteins, may play a key role. The identification of new protagonists in such a contest may thus become important to improve the management of this neoplasia. Recent studies have underlined that NGAL, a small kidney stress protein well-known by nephrologists for its predictive value in the early diagnosis of AKI, represents also a fundamental key factor involved in the biology of different tumours such as breast, ovaric, thyroid and gastointestinal cancers. With the present study we aimed at evaluating the possible relationships between NGAL and human renal cancer.
Methods: NGAL expression was evaluated by immunoblot analyses in 27 cases of surgically removed renal tumors (17 clear cells carcinomas (CCCs), 5 papillary carcinomas (PCs), 2 urothelial carcinomas, 2 oncocytomas, 1 CCC with sarcomatoid differentiation). The staining intensity (IS) was graded as (0) negative, (1) weak, (2) moderate, (3) strong. The area of staining positivity (ASP), recorded as percentage of neoplastic positive cells, was assessed by following values: 0 ($\leq$ 10%), 1 (11-25%), 2 (26-50%), 3 (51-75%), 4 (>75%), Then, an intensity-distribution (ID) score was generated for each case by multiplying the values of IS and ASP. Tubule cells were used as positive control for NGAL expression.
Results: The presence of NGAL resulted positive in 25 specimens with variable ID scores, the expression being particularly localized within the cytoplasm and all over the plasmatic membrane.
Very interestingly, even a primary peritoneal metastatis of CCC also showed a strong NGAL immunoreaction. Only two specimens did not present a detectable NGAL expression: a low-grade renal cell carcinoma and a low-grade renal cell carcinoma with sarcomatoid differentiation.
The NGAL staining intensity resulted strictly and directly correlated to the grade of neoplasia (Fuhrman classification; R: 0.475, $p<0.001$), as well as the NGAL staining positivity (R: 0.512, $p<0.001$) and the intensity distribution (R: 0.612, $p<0.001$).
Conclusions: As previously reported for other human neoplasias, the present study shows for the first time that renal tumors also notably express NGAL. This observation seems to regard almost every histologic subtype: NGAL may thus play a central role in the biology of this neoplasia. In the specimens analyzed, NGAL expression is also strongly correlated with the severity of the neoplasia. Furthermore, the analysis of primary metastasis demonstrates that these cells also can produce this protein. Further studies are thus mandatory to clarify the potential applications of NGAL in the diagnosis, prognosis and clinical evaluation of patients affected by renal neoplasias.

Su059 PRECONDITIONING OF ER STRESS ATTENUATED TGF-β-INDUCED EMT IN RENAL TUBULAR CELLS

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Introduction and Aims: Epithelial-to-mesenchymal transition (EMT) of renal tubular cell is known to be a key process of renal fibrosis, however there are still controversies regarding the mechanism of EMT. Endoplasmic reticulum (ER) stress is a cellular stress pathway induced by the accumulation of unfolded proteins in the ER which is initially a defense mechanism of cells against various stressful environments whereas prolonged activation of ER stress induced cell apoptosis. ER stress in the kidney is reported to play a role in ischemia/reperfusion injury with a demonstration of protective effect of ER stress preconditioning in renal functional recovery. However, there is still limited knowledge on the pathophysiologic role of ER stress in the progression of various kidney disease with few in-vitro experimental data using renal tubular cells. To investigate the role of ER stress on renal disease progression, we examined the effect of ER stress preconditioning on EMT induced by transforming growth factor-β (TGF-β), a key cytokine of renal damage.
Methods: Preliminary experiment to determine the optimal preconditioning condition of HK-2 cells with tunicamycin (TM) or thapsigargin (TG) which induced the expression of two ER stress chaperones (GRP78/94 and eIF-1α) without an evidence of cell apoptosis was performed. EMT was evaluated with comparing the expression of epithelial cell marker, E-cadherin and mesenchymal cell marker, α-SMA. Senescence of HK-2 cells was assessed by senescence-associated (SA) β-gal staining. TGF-β-induced phosphorylation of ERK after ER stress preconditioning was also examined by Western blotting.
Results: Exposure of HK-2 cells to TGF-β (10 ng/ml) for 48 to 72 hours resulted in a changes in cell morphology from cobble stone appearance to elongated, fibroblastoid morphology. TGF-β also decreased E-cadherin expression with an increase in α-SMA from 48 hours of stimulation. Treatment of HK-2 cells with 0.1 μg/ml of TM or 0.1 μM of TG for 6 hours resulted in an enhanced expression of GRP78/94 with a phosphorylation of eIF-1α. Preconditioning with TM or TG protected HK-2 cells from TGF-β-induced changes in cell morphology and reversed the changes in the expression of E-cadherin and α-SMA by TGF-β. TGF-β-induced cell senescence was also ameliorated by pre-treatment with TM or TG. TGF-β induced ERK phosphorylation was inhibited by ER stress preconditioning.
Conclusions: In conclusion, our finding suggests ER stress preconditioning protects renal tubular cells from phenotyphic transformation, which can be a potential therapeutic target to prevent the progression of chronic kidney disease. Further studies are necessary to verify the role of ER stress preconditioning in animal model of chronic kidney disease.

Su060 SIGNAL TRANSDUCTION IN ENDOTHELIAL AND ERYTHROID PROGENITOR CELLS: LOW VERSUS HIGH DOSES OF EPOETIN BETA REVEALS DIFFERENT ACTIVATION OF THE PRO- SURVIVAL AKT PATHWAYS IN THESE CELLS

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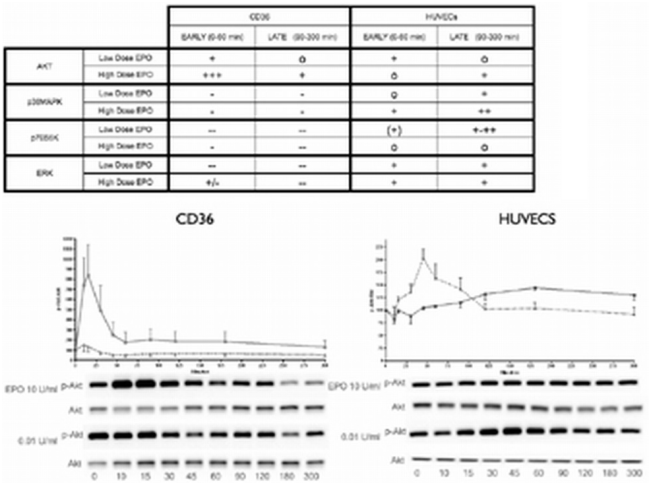
Introduction and Aims: In the haematopoietic system, the principal function of erythropoietin (EPO) is the regulation of RBC production. Consequently, recombinant human EPO have been widely used for treatment of anaemia in chronic kidney disease (CKD). However, several studies have been recently published on the tissue-protective, nonhaematological effects of EPO that prevent ischaemia-induced tissue damage in several organs including the kidney. Because this tissue-protective action of EPO is not the result of correction of anaemia-related tissue hypoxia, we analysed potential molecular pathways involved in both, endothelial (HUVEC) and erythroid progenitor cells (CD36 cells).
Methods: Four days after the last medium change we stimulated the HUVEC or CD36 cells with 0.01 or 10 U/ml Epoetin beta and analysed the activation of different signal transduction pathways (AKT, p38MAPK, ERK, p70S6K, Caspase-3, FoxO1, CDK2, STAT5) at baseline or at 10, 15, 30, 45, 60, 90, 120, 180, 300 minutes.
Results: We confirmed previous results indicating that in CD36 cells Akt signal transduction is activated in a dose dependent manner. Interestingly, in HUVECs low doses of EPO activated Akt more pronounce and much earlier in comparison to higher doses. We currently investigate whether the instant and pronounced activation of Akt may also result in an activation of eNOS in ECs. Recent work clearly demonstrated that the effects of EPO on thrombocytes depend on the presence of an intact eNOS driven NO formation of the endothelium. It can therefore be speculated that an increase in haematocrit related to EPO treatment is not associated with an increased risk for thrombosis as long as endothelial NO production serves as a compensatory mechanism.
Conclusions: Previously we could demonstrate that chronic low-dose therapy with EPO conferred vascular and tissue protection, preserved renal function, and improved survival in a classic remnant kidney rat



model characterized by severe endothelial damage, progressive vascular sclerosis, and ischemia induced tissue fibrosis. We interpret these findings as evidence that low dose EPO treatment prevented the "firststrike" injury at the endothelial cell level and, in addition, reduced apoptotic cell death and preserved tissue integrity in the critical phase of tissue damage and destruction.
Our current findings indicate, that these effects may be mediated, at least in part, by a rapid activation of the pro-survival Akt pathway.

Su061 FLUVASTATIN ATTENUATES HIGH GLUCOSE- INDUCED FIBRONECTIN EXPRESSION BY INHIBITING THE SIGNAL PATHWAY OF RHO-KINASE IN HK-2 CELLS

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Introduction and Aims: To explore the effect of Rho-kinase signal pathway in renal interstitial fibrosis of diabetic nephropathy (DN) and the mechanism of fluvastatin in the prevention of renal interstitial fibrosis of DN.
Methods: Human renal proximal tubular epithelial cells (HK-2 cells) were cultured in vitro. Rho-kinase activity wasexpressed as phosphorylation of myosin-phosphatase target 1 (p-MYPT1). The level of p-MYPT1 and fibronectin (FN) stimulated by high glucose was determined by Western blot at the time of 0h, 6h, 12h, 24h, 48h. Same marks were detected when high glucose cultured HK-2 cells treated with different concentrations (10^{-7}, $10^{-6}$and 10^{-5} mol/L) of fluvastatin for 12h and when treated by lysophosphatidic acid (LPA) further.
Results: High glucose enhance the expression of p-MYPT1 and FN at the time of 6h, 12h, 24, 48h compared with the time of 0h in culured HK-2s. The increase of FN expression stimulated by high glucose was in time-dependent fashion and the increased level of p-MYPT1 reached the peak at 12h. Fluvastatin decreased high glucose-mediated level of p-MYPT1 and FN in dose-dependent manner. The inhibitory effect of fluvastatin on up-regulation of p-MYPT1 and FN stimulated by high glucose was reversed by LPA.
Conclusions: Rho-kinase may be one of the initiation signals of renal interstitial fibrosis of DN. Fluvastatin can prevent the development of renal interstitial fibrosis of DN by inhibiting Rho-kinase signaling pathway.

Su062 C5A REGULATES TUBULAR EPITHELIAL MYOFIBROBLAST TRANSDIFFERENTIATION IN VITRO

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Introduction and Aims: Growing evidence indicated that the activation of complement system was involved in the development of tubulointerstitial injury in clinical and animal studies. But the mechanism is not fully understood. We hypothesize that complement induce the tubular epithelial-myofibroblast transdifferentiation (TEMT) during the tubulointerstitial fibrosis. This study investigate the effect of anaphylotoxin C5a on TEMT in vitro.
Methods: Renal proximal tubular cell line HK-2 were cultured in vitro and divided into six groups: control group,TGF-β1 positive control group,10nM anaphylotoxin C5a group,25nM C5a group,50nM C5a and 50 nM C5a+1μM C5a receptor inhibitor (C5aR) group. The expressions of E-cadherin,α-SMA,TGF-β1,CTGF,Col-Iand β-catenin were detected by immunohistochemistry, Western blot and RT-PCR. And scanning electron microscope (SEM) was used to observe the cellular morphological changes.
Results: Normal HK-2 cells showed the classic cobblestone shape, and SEM showed a loss of apical-basal polarity and microvilli of the cells stimulated by C5a, and the cells become elongated and invasive. Immunohistochemistry, RT-PCR and Western blot showed that C5a induced the loss of the epithelial marker E-cadherin and increased the expressions of α-SMA, TGF-β1, CTGF, Col-Iand β-catenin in dose-dependent and time-dependent manners. While C5aRA can partial block the decrease of E-cadherin and increase of α-SMA and TGF-β1.
Conclusions: C5a can induce tubular epithelial-to-mesenchymal transition, TGF-β1, β-catenin pathway may participate in the mechanism. And C5aRA can partial block the effect of C5a on renal tubular cells.

Su063 ALTERED RENAL REGULATION OF mTOR PATHWAY AND AUTOPHAGY IN RATS WITH UNILATERAL URETERAL OBSTRUCTION

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Introduction and Aims: The present study was aimed to investigate the role of the mammalian target of rapamycin (mTOR) pathway and autophagy in the pathogenesis of tubulointerstitial injury in obstructed kidney in rats with unilateral ureteral obstruction.
Methods: Male Sprague-Dawley rats were unilaterally obstructed by the ligation of left proximal ureters for 7 days. Control rats were treated in the same way, except that no ligature was made. The protein expression of Akt, mTOR and transforming growth factor (TGF)-β1 was determined in the kidney by semiquantitative immunoblotting. The protein expression of microtubule-associated protein light chain 3 (LC3) and anti-apoptic factor Bcl-2 was also determined.
Results: In the obstructed kidneys, the protein expression of Akt was significantly increased in the cortex and outer stripe of outer medulla (cortex/OSOM) and inner medulla compared with the controls. The expression of mTOR was increased in the cortex/OSOM although did not changed in the inner medulla. The expression of TGF-β1 was increased in the cortex/OSOM and inner medulla. The expression of LC3 was increased in the inner medulla althogh unaltered in the cortex/OSOM. The expression of Bcl-2 was decreased in the cortex/OSOM and inner medulla.
Conclusions: In the obstruced kidney, the upregulation Akt/mTOR pathway may play a role in the renal fibrosis by increasing abundance of TGF-β1. In addition, the decreased abundance of Bcl-2 may contribute to the upregulation of LC3, which may play a role in the pathogenesis of the tubular apoptosis.

Su064 EGCG INHIBITS PMA-INDUCED MCP-1 IN HUMAN ENDOTHELIAL ECV 304 CELLS VIA BLOCKING P38 MAPK AND NF-kappa B

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Introduction and Aims: Monocyte chemotactic protein-1 (MCP-1) is a potent chemoattractant for monocytes and plays a key role in various inflammatory responses, including atherosclerosis, In this study, we examed the effect of (-)-epigallocatechin-3-gallate (EGCG), a major green tea catechin, on the expression of MCP-1 in human endothelial ECV304 cells.
Methods: We used Human endothelial ECV304 cells, and analyzed with Northern blot, Western blot, transient transfection and luciferase assay and electrophoretic mobilty shift assay.

Results: EGCG markedly inhibited the phorbol 12-myristate 13-acetate (PMA)-induced MCP-1 mRNA and protein levels in a dose-dependent manner. EGCG was also found to reduce the MCP-1 transcriptional activity. The upregulation of MCP-1 by PMA was significantly inhibited by blockade of P38 MAPK and NF-kappa B, but not by blockade of extracellular signal-regulated kinase and c-Jun N-terminal kinase pathway. Furthermore, The PMA-induced P38 MAPK and NF-kappa B activation were obviously attenuated after pretreating ECV304 cells with EGCG. The conditioned media from the endothelial ECV304 cells treated with PMA could remarkably stimulate the migration of THP-1 monocytes and this effect was partially abrogated by MCP-1 neutralizing antibodies.
Conclusions: EGCG may exert an anti-inflammatory effect in endothelial cells by controlling MCP-1 expression, at least in part, mediated through the suppression of P38 MAPK and NF-kappa B activation.

Su065 RUBOXISTAURIN ATTENUATES TGF-β-INDUCED EMT IN RENAL TUBULAR CELLS

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Introduction and Aims: Activation of protein kinase C-β (PKC-β) has been implicated in the pathogenesis of diabetic nephropathy and Ruboxistaurin mesylate, a selective inhibitor of PKC-β has been completed phase II and III trials in diabetic nephropathy and retinopathy. Although high glucose is known to activate the PKC-β, subsequent up-regulation of TGF-β transcription, PKC-β localizes not only upstream, but also downstream of TGF-β signaling. Recently, PKC-β inhibitor was reported to attenuate the progression of non-diabetic nephropathy, remnant kidney model. However, it is not known the renoprotective mechanism of ruboxistaurin in non-diabetic nephropathy. In this study, we examined whether PKC-β inhibitor affect the TGF-β induced epithelial to mesenchymal transition (EMT), a crucial event in renal fibrosis of non-diabetic nephropathy.
Methods: EMT was evaluated by morphologic transformation of NRK-52E and HK-2 cells under inverted microscopy as well as a quantitative analysis of the expression of the epithelial marker, E-cadherin and the mesenchymal marker, α-smooth muscle actin (α-SMA) protein, by western blot analysis after stimulating of TGF-β (10 ng/ml) with or without pre-treatment of Ruboxistaurin (1μM). ERK1/2 and p38 MAPKinase activation in TGF-β (10 ng/ml)-stimulated cells were examined by western blotting. The effects of pre-treatment with Ruboxistaurin (1μM) on TGF-β-induced EMT were determined.
Results: TGF-β (10 ng/ml) induced a morphological transformation from cuboidal and cobble stone appearance to spindle shaped scattered fibroblast-like cells at 48hrs, which was inhibited by pre-treatment of Ruboxistaurin (1 μM). TGF-β down-regulated E-cadherin and up-regulated α-SMA expression at 48 hours, which were significantly attenuate by pre-treatment of Ruboxistaurin (1 μM). TGF-β (10 ng/ml) significantly increased phosphorylation of ERK1/2 and p38 MAPK from 5 minutes of stimulation. Although phosphorylation of ERK1/2 was blocked, phosphorylation of ERK1/2 was not blocked by pre-treatment of Ruboxistaurin (1 μM).
Conclusions: In conclusion, Ruboxistaurin inhibited TGF-β induced EMT in renal tubular cells. These result suggests that Ruboxistaurin might be considered as a potential therapeutic strategy in non-diabetic kidney disease.

Su066 PHOSPHORYLATION OF MOESIN IS INVOLVED IN TGF-b1 INDUCED EPITHELIAL-TO-MESENCHYMAL TRANSITION IN HUMAN TUBULAR EPITHELIAL CELLS

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Introduction and Aims: TGF-β is known for its profibrogenic effects during renal fibrosis. This study aims to focus on phosphorylated proteins during process of TGF-β induced EMT in kidney.
Methods: Human tubular epithelial cells (HK-2 cells) were divided into TGF-β1-treated group or control group. Phosphorylated proteins of both groups were extracted using Phosphoprotein purification kit and differentially mass labeled using isobaric tags for relative and absolute quantitation (iTRAQ) 4-plex reagent kit (Applied Biosystems). Peptide and protein identifications were performed 2D-nano-HPLC-MS/MS. Validation of isobaric tagging was confirmed by western blot.
Results: HK-2 cells treated by TGF-β1 exhibited increased expression of a-SMA and reduced expression of E-Cadherin by real-time PCR and western blot. Such features were consistent with characteristic of EMT. The morphological changes of HK-2 cells also confirmed the EMT process. By iTRAQ-2D-nano-HPLC-MS/MS, we identified phosphorylated moesin was up-regulated in TGF-β1-treated group. Western blot confirmed the differential expression of phosphorylated moesin. Furthermore, our results demonstrated that TGF-β1 could induce phosphorylation of moesin in a time-dependent and dose-dependent manner. Such manner was induced by Erk1/2 signaling pathway. Blocking Erk1/2 by Erk inhibitor PD98059 could also block phosphorylation of moesin in HK-2 cells.
Conclusions: Our results suggested TGF-β1 could induce phosphorylation of moesin by activating Erk1/2 signaling pathway during TGF-β1-induced EMT.

Su067 DEXAMETHASONE INDUCES APOPTOSIS OF OSTEOBLASTS IN CULTURE BY INTERFERENCE WITH CELL ADHESION COMPLEX

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Introduction and Aims: Changes in mineral metabolism and bone structure are an almost universal finding in chronic renal failure. Those alterations may be manifested in several types of bone diseases, known as renal osteodystrophy. Lesion of bone tissue can become more severe with administration of glucocorticoids. Various pathogenetic mechanisms have been investigated in order to explain the damage to bone cells caused by corticosteroids, such as those related to interference with cell adhesion. In this context, integrin-linked kinase (ILK), a scaffold protein linked to β1-integrin subunit cytoplasmic domain, participates in survival and cell death signaling pathways. There are sparse reports on the expression of ILK in osteoblasts.
Methods: Primary cultured human osteoblasts were treated with dexamethasone in 10-9M (physiological) and 10-6M (pharmacological) dosage for 24 and 48 hours. Cell viability (MTT assay), adhesion (crystal violet assay) and apoptosis (flow cytometry) were analyzed. In addition, expression and immunolocalization of β1-integrin and ILK were analyzed by western blot and immunofluorescence.
Results: A decrease in cell viability and adhesion was observed in both treated groups. This reduction was higher in pharmacological-treated groups when compared with the physiological-treated groups. There was a slight reduction of apoptotic cells in groups treated with the physiological dose and significant increase in apoptotic cells treated with the pharmacological dose in comparison to control group. The ILK expression was slightly increased, while β1-integrin significantly decreased in all treated groups, when compared to controls. The two proteins were co-localized in cell membrane in all groups.
Conclusions: Our results suggest that dexamethasone causes decrease in osteoblasts viability due to anoikis caused by reduction in β1-integrin expression. The decrease in β1-integrin expression probably influenced ILK activity, reducing its activation and inhibiting its anti-apoptotic effect.
Disclosure: FAPESP #2007/54403-8

Renal development and cystic diseases

Su068 ★ PROTEOMICS MAPPING OF EMBRYONIC KIDNEY DEVELOPMENT: IDENTIFICATION OF RELEVANT PROTEINS DURING THE NEPHROGENESIS

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Introduction and Aims: The kidney plays an important role in the whole-body homeostasis, regulating acid-balance, electrolyte concentrations, extracellular fluid volume, and regulation of blood pressure. Nephrogenesis, or kidney development, describes the embryologic origins of the kidney. It implies a highly controlled series of morphogenetic and differentiation events which are well characterized morphologically, but less understood concerning their molecular mechanisms. To elucidate kidney embryonic development and to identify proteins that are regulated during the initial steps of nephrogenesis, large-scale proteome analysis of the embryonic rat kidney from different developmental stages was performed.
Methods: Embryonic kidneys from rats were isolated at different stages of development: E14, E16, E19 (days after fertilization) and P1 (newborn). Histological and proteomic analysis were performed of each embryonic stage. The embryonic kidney proteome was investigated combining 2D gel electrophoresis and mass spectrometry. Comparative analysis between the proteomes of the different embryonic stages was performed using the 2D-DIGE (fluorescence difference gel electrophoresis) technology and Western blot analysis.
Results: Out of the 977 excised protein spots, 685 protein spots were identified, of which 305 proteins were non-redundant. 106 of the identified proteins were common in all developmental stages. 84% of the proteins found in E14 and E16 and 70% of the proteins found in E14 and E19 were identic, whereas only 38% of the proteins found in E14 compared to P1 overlapped. An interaction network of the identified proteins was created using STRING functional protein association network. DAVID bioinformatics was used to categorize the proteins.
Conclusions: We present the first comprehensive proteomics study of four different stages of the embryonic development of the rat kidney. The proteins identified to be differentially expressed during the embryonic development, will elucidate the molecular mechanisms underlying renal embryonic development.

Su069 ★ ABERRANT SPLICING OF THE *PKD1* PRE-MRNA DUE TO SYNONYMOUS MUTATIONS IN AUTOSOMAL DOMINANT POLYCYSTIC KIDNEY DISEASE

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Introduction and Aims: Autosomal dominant polycystic kidney disease (ADPKD) is the most common inherited kidney disorder and an important cause of end-stage renal disease. ADPKD is characterized by the progressive, bilateral development and enlargement of focal cysts in both kidneys that usually result in end-stage renal disease. ADPKD is caused by mutations in the *PKD1* or *PKD2* genes. Most patients present mutations in *PKD1*, and a considerable number of these mutations are nucleotide substitutions within the coding sequence that have been classified as missense or synonymous mutations. Recent studies have suggested that a significant fraction of these mutations can be harmful by affecting pre-mRNA splicing. The aim of our study is to determine the functional consequences of *PKD1* missense and synonymous mutations at the mRNA level.
Methods: *PKD1* minigenes containing exons 2-3, 11, 23, 24-25-26 and 37-38-39 were constructed. Mutations were introduced by site-directed mutagenesis and confirmed by DNA sequencing. These constructs were transfected into COS7 or HEK293T cells. pre-mRNA splicing analysis was performed by RT-PCR using RNA isolated from cultured cells. The web-based tool NNSplice was used to predict the impact of mutations on RNA splicing. ESEfinder and Rescue-ESE web-based resources were used to evaluate the effect of mutations on putative ESE sequences.
Results: Seven missense mutations, L845S, R2765C, R2985G, Q3016R, R3105W, L3731Q, Q3751R; and three synonymous variants, G109G, L3754L and R3753R were analysed. The results showed that synonymous variants G109G and R3753R altered *PKD1* pre-mRNA splicing. G109G creates a donor splice site in exon 3, leading to an anomalous mRNA lacking 34 bp at the 3' end of this exon. R3753R activates a cryptic donor splice site upstream in exon 39. This yielded an altered mRNA lacking 16 bp of the 3' end of this exon. At protein level, both mutations predict a new reading frame that results in a premature stop codon. The rest of mutations showed no effect in pre-mRNA splicing.
Conclusions: Our results indicate that synonymous variants G109G and R3753R act as splicing mutations. These represent the first *PKD1* synonymous mutations that lead to altered mature RNA. The evaluation of the effects of synonymous mutations at RNA level might have important implications for suitable genotype-phenotype correlations.
Disclosure: This work was supported by grant PI 07/1037 from Fondo de Investigación Sanitaria (Spain).

Su070 MATERNAL SALT INTAKE – BLOOD PRESSURE AND TARGET ORGAN MORPHOLOGY IN THE OFFSPRING

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Introduction and Aims: An adverse environment during fetal development has life-long consequences: hypertension, increased cardiovascular risk, renal malfunction. We studied, using stereology, whether high salt intake in pregnancy modifies structure of the kidney as well as structure of vasculature and blood pressure in the offspring.
Methods: Sprague-Dawley rats were fed low (0.15% NaCl), medium (1.3%), or high (8.0%) salt diets during pregnancy and weaning. The offspring were weaned at 4 weeks of age and subsequently received a standard rodent diet. Blood pressure was measured by telemetry and albuminuria by a rat specific ELISA up to 52 weeks of age. Nephron number was determined by a design-based stereology at 12 weeks of age.
Results: The nephron number was significantly lower in offspring of dams on low (males: 19100±3700, females: 19000±2500) and high (m: 12100±600, f: 12800±2800) compared to medium salt intake (m: 32400±2500, f: 28500±6000). In male offspring of dams on both low and high salt intake albumin excretion increased at 6 months of age and was higher than in offspring of dams on medium salt intake.
Systolic, diastolic, and mean arterial pressures were not significantly different between the offspring until 12 weeks of age. After 7 months of age systolic blood pressure was higher in offspring of dams on low (123±8 mmHg) and high (124±7) compared to medium (116±5) salt intake.
There was no significant difference in vascular geometry at 7 weeks postnatally. At 12 weeks, however, wall thickness of central (aorta, carotid, iliac) and muscular (mesenteric and even intrapulmonary) arteries was significantly higher in offspring of mother rats on high salt diet irrespective of the post-weaning diet compared with other groups.
Conclusions: Both too high and too low salt intake in pregnant rats predisposes their offspring to increased renal risk due to reduced nephron number, increased albuminuria, hypertension and thickening of the wall of vessels even those not exposed to high blood pressure.

Su071 RENIN EXPRESSION AND RENAL DEVELOPMENT WITH A VIEW TO CYCLOOXYGENASE-2 ABSENCE

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Introduction and Aims: The renin distribution pattern varies during kidney

development. Renin appears first at embryonic day 15 within the A. arcuatae, from where it migrates along the developing arterial tree to the proximal vessels, to be finally localized at its classical juxtaglomerular position in adulthood. The factors influencing this shift of renin producing cells are still unknown. The last few years there has been growing evidence indicating that cyclooxygenase-2 (COX-2) might have an impact on renin expression. The aim of our study is to investigate if and to what extend missing COX-2 abundance does influence the renin expression pattern in the maturing mouse kidney.
Methods: – 3d-renal arterial tree reconstructions
– Immunohistochemical researches on COX-2 localization and renin expression in perfused COX-2 +/+ adult and maturing kidneys
– Real time PCR measurements of COX-2 and renin mRNA expression during murine nephrogenesis
Results: The renin expression levels are lowered in COX-2 knockout mice, but the expression pattern does not differ from wild type neither in adulthood nor during kidney development.
The COX-2 Knockout mice showed a thickened compressed arterial tree and small glomeruli localized directly beneath the capsule with nearly no intermediate subcapsular space.
In wild type mice COX-2 could be found in the nephrogenic zone namely in developing early tubules for example in s-shaped bodies. In all examined developmental stages COX-2 was present at the macula densa region.
Conclusions: In summary with the previous data from 3d reconstructions of COX-2 -/- kidneys, we conclude that COX-2 seems not to be essential for the correct positioning of renin producing cells during murine nephrogenesis. It remains unclear whether the decreased renin expression in COX-2 -/- mice is a direct consequence of the absent COX-2 or could be affected by the malformed subcapsular kidney structure, particularly by the abnormal glomeruli.

Su072 CONDITIONED MEDIA OF VASCULAR ENDOTHELIAL AND TUBULAR EPITHELIAL CELLS LEAD IN VIVO GLOMERULOGENESIS USING TUBULAR EPITHELIAL CELLS OR MESENCHYMAL STEM CELLS

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Introduction and Aims: A kidney is one of the most difficult organs to regenerate. A few reports using a fertilized egg, an embryo or embryo's tissues have done, but their methods seem to be hard to apply for human nephrogenesis. We have attempted in vivo glomerulogenesis under the concept of cellular interaction between vascular endothelial (VEC) and tubular epithelial cells (TEC) through their conditioned media (CM) perhaps including their releasing factors.
Methods: First, we studied the interaction between VEC; murine MILE SVEN 1 (MS1) and TEC; Madin-Darby canine kidney cell (MDCK) or human mesenchymal stem cell (hMSC) on the cell proliferation in 2-dimentional (-D) culture and morphological changes in 3-D culture. CM of MS1 and MDCK were collected at the passage after 2-time medium change without supplements and FBS, and were concentrated at 10 times (10X). The 10X MS1- or MDCK-CM (1 ml) was added to the basal medium (10 ml) in 2-D culture. Each CM (30%) was mixed in the gel of Collagen-I (60%) in 3-D culture. Second, we tried the differentiation of hMSC to cytokeratin-positive epithelial cell (hMSC-EC) by the addition of CM for 4 weeks. Finally, we investigated in vivo glomerulogenesis using MDCK, hMSC or hMSC-EC with both CM (each 15%). Twelve weeks after the implantation into the subcutaneous space of nude rat, we picked up the implanted tissues and performed microscopic studies.
Results: 10X MS1-CM promoted the proliferation ($p<0.0001$) and morphological change (tubulogenesis) of MDCK, whereas 10X MDCK-CM acceralated the proliferation ($p<0.0005$) and morphological change of MS1, compared with the other cell. On the other hand, hMSC was turned into cytokeratin-positive cell (hMSC-EC) only by the culture with 10X MDCK-CM. However, 10X MS1-CM, not 10X MDCK-CM, promoted the cell proliferation ($p<0.001$) and morphological change. Finally, many glomeruli with PAM staining-positive capillary walls were observed in the implanted tissues of MDCK, hMSC or hMSC-EC. Tubulogenesis is also certified. There were no evident differences among these 3 tissues.
Conclusions: This is the first report of glomerulogenesis with some factors, that is, without the use of natural developmental course. These findings show 'cross-talk' between VEC and TEC beyond species. These methods seem to be applied for nephrogenesis in human without the ethical problems and rejection/immunosuppression.

Su073 EFFECT OF SIROLIMUS ON KIDNEY VOLUME GROWTH AND RENAL FUNCTION IN AUTOSOMAL DOMINANT POLYCYSTIC KIDNEY DISEASE – RESULTS OF THE SUISSE ADPKD TRIAL

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Introduction and Aims: Autosomal dominant polycystic kidney disease (ADPKD) is a slowly progressive chronic kidney disease that leads to end-stage renal failure by the age of 50 yrs in approximately 50% of the affected patients. Based on evidence for effectiveness of sirolimus in various animal models of PKD we conducted a prospective clinical trial to examine the effect of sirolimus on cyst volume progression in young patients (mean age 32±7 yrs, 71% males) with documented ADPKD and GFR>70 ml/min.
Methods: After a 6-month run-in phase we randomized 100 patients 1:1 to receive either sirolimus 2 mg/day or no treatment and examined total kidney volume (TKV) progression in both groups after 18 months of treatment.
Results: Baseline characteristics were similar in both groups, including blood pressure (131±15/84±12 in the sirolimus group vs. 132±16/82±12 mm Hg in the control group), TKV (1099±682 vs. 1120±632 cm^3) and GFR (109±32 vs. 110±38 ml/min).The sirolimus treatment was well tolerated and no adverse event related drop outs occurred.
Conclusions: The effect of sirolimus on the change of TKV during the 18-month treatment phase as well as the effect on eGFR will be reported at the meeting. The results of our trial will allow assessing whether sirolimus represents an effective and useful treatment to retard progression of ADPKD. (NCT00346918)

Su074 MEDULLARY CYSTIC KIDNEY DISEASE TYPE 1: CLINICAL AND BIOCHEMICAL FINDINGS

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Introduction and Aims: Medullary Cystic Kidney Disease Type 1 (MCKD1) is an adult type, hereditary, chronic interstitial nephritis leading to End Stage Renal Disease (ESRD) in the 4^{th} to 7^{th} decade of life. MCKD1 is a heterogeneous disease in several clinical aspects. The aim of this study was to present demographic, clinical and biochemical findings in 7 large families with established MCKD1 and find out whether one or more can lead to early diagnosis.
Methods: 59 living individuals (30 M, 29 F) among 108 MCKD1 gene carriers (confirmed by linkage analysis) were studied and compared with 59 age, sex and renal function matched controls. MCKD1 individuals and controls underwent interview, clinical examination, blood and urine tests. Creatinine clearance was calculated using Cockroft–Gault formula, corrected for body surface area of 1.73 m^2. Renal function was classified according to Chronic Kidney Disease (CKD) classification of The Kidney Disease Outcomes Quality Initiative. Fractional Excretion of Sodium (FE_{Na}) and Fractional Excretion of Urate (FE_{urate}) were calculated after a 24-hour abstinence of sodium and 12-hour overnight water deprivation. Proteinuria was defined as Protein/Creatinine ratio>0,2. For the CKD classification and median ESRD age calculation for MCKD1 group we also used data from 22 transplanted MCKD1 patients. χ^2 test was used for the comparison between the groups.
Results: Among the 81 affected living individuals that we were able to examine, 15 presented with normal renal function (18.6%), 37 with mild

or advanced CKD (45.6%) and 29 with ESRD (35.8%), treated either by dialysis or already transplanted. Median age of ESRD was 50 years old (range: 32-79). Interfamiliar variation according to Median age of ESRD was found. Proteinuria was not a prominent feature. The vast majority of individuals (84%) had protein/creatinine ratio < 0.2. Hypertension was found in 39 MCKD1 patients (66%) and 35 control (59%), a non statistically significant difference (p=0.446). Among MCKD1 patients, hypertension was more common in women (72%) than in men (63%) (p=0.455). Blood tests revealed hyperuricemia in 32 (54%) MCKD1 patients and 29 (49%) controls, a non statistically significant difference (p=0.461). Between MCKD1 gene carriers, hyperuricemia was more frequent in men (69%) than in women (38%), a statistically significant difference (p=0.0178). Among hyperuricemic patients 75% had a decreased FE_{urate}. An inverse correlation between FE_{Na} and degree of renal failure was detected.
Conclusions: MCKD1 is a heterogeneous disease in several clinical aspects, including clinical appearance, age of onset, and rate of progression to ESRD. None of the symptoms was found to be characteristic for MCKD1. Heterogeneity of symptoms complicates the clinical diagnosis, particulary in the early stage when renal cysts are not usually present. Genetic breakthroughs are expected to help in more accurate and earlier diagnosis.

Su075 PERINATAL PRIMING OF RENIN PRODUCING CELLS

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Introduction and Aims: The amount of daily salt intake inversely affects the number of renal renin producing cells, which can reversibly switch between a smooth muscle and a renin secretory phenotype by metaplastic transformation. The cellular signals and pathways underlying positive or negative recruitment of renin producing cells are only poorly understood. In this context it is known that the sensitivity of renin expression towards modulations of salt intake is strikingly enhanced in mice lacking ANGII-AT1a receptors.
To narrow down possible reasons for the altered salt sensitivity in AT1a deficient mice, we have firstly examined as to whether this effect is due to the actual absence of AT1a receptors in the adult organism or is the result of an altered neonatal reprogramming of kidney development, which is known to occur after neonatal inhibition of the renin angiotensin system.
Methods: For this aim we have analyzed renin expression (number of renin producing cells, renin mRNA) in kidneys of four groups of adult mice (age 8-10 wks):
A) wt (Sv129) mice treated with high or low salt diet for one week
B) wt mice treated with high or low salt diet in combination with the AT1 receptor blocker losartan (30mg*kg-1*d-1) for one week
C) wt mice, treated with losartan (30mg*kg-1*d-1) from day pp1-pp5, and treated with high or low salt diet in combination with the AT1 receptor blocker losartan for one week
D) AT1a deficient mice
Results: The salt sensitivity of renin expression of the different groups was D=C>>B>A, indicating that neonatal blockade of the renin angiotensin system produces a similar salt sensitivity of renin expression as does genetic ablation of the AT1 receptors.
Conclusions: The mechanisms underlying this special striking reprogramming phenomenon of kidney development are currently under investigation.

Su076 EARLY POSNATAL α-TOCOPHEROL MODULATES PRENATALLY PROGRAMED RENAL OXIDATIVE STRESS IN RATS

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Introduction and Aims: Increased oxidative stress in the kidneys is involved in the pathogenesis of hypertension. Maternal undernutrition increases placental oxidative stress, and programs increased renal oxidative stress in the adult offspring. We evaluated whether α-tocopherol, administered during lactation, prevents the increment of renal oxidative stress induced by prenatal undernutrition.
Methods: Female Wistar rats were fed, during pregnancy, with a standard diet, the control group (C), or with a multideficient diet, the malnourished group (M). During lactation, all dams were fed with a standard diet. Additionally, part of them received vehicle (V, corn oil, 1 mg/kg BW), the CV (n=6) and MV (n=5) groups, while another part received α-tocopherol (T, 350 mg/kg BW), the CT (n=7) and MT (n=5) groups. Maternal and offspring diet intake and weight gain were evaluated. Oxidative stress was evaluated measuring the levels of thiobarbituric acid reactive substances (TBARS) in maternal livers, after weaning, and in the adult offspring kidney (CV, n=8; MV, n=13; CT, n=14; MT, n=6), at age of 90 days. TBARS values are expressed as the levels of malonildialdehyde (MDA) corrected for tissular weight. Data are mean ± SEM. Statistical analysis was performed using the Student-Newman-Keuls test.
Results: During gestation, weight gain of malnourished dams was lower than that of control dams (83±7 vs. 108±5 g, respectively, $p<0.05$). During lactation, weight gain was the same for MV and CV dams (12±4 vs. 8±4, respectively), but it was higher in MT than in CT dams (36±8 vs. 8±6, respectively, $p<0.01$). Hepatic TBARS was higher in MV than in CV dams (6.8±0.3 vs 5.6±0.4 nmol MDA/g, respectively, $p<0.05$), while CT and MT dams showed lower TBARS levels (~25%, $p<0.05$) than CV and MV dams. The birthweight of prenatal malnourished rats was lower than that of control rats (4.9±0.1 vs. 6.3±0.1 g, $p<0.001$). At weaning, the weight of MV was lower than of CV (57±2 vs. 64±1, respectively, $p<0.001$), while it was the same for MT and CT groups (61±1 vs. 64±1, respectively). At age of 90 days, MV and MT offspring had body weight lower than their respective control groups (307±10 vs. 343±5 and 323±5 vs. 360±7 g, respectively, $p<0.05$). TBARS levels were higher in the kidney of MV compared to CV offspring (16.1±0.6 vs 13.7±0.4 nmol MDA/g, $p<0.01$), while MT presented the same values of TBARS as CT and CV offspring (10.8±0.5, 8.5±0.5 and 13.7±0.4 nmol MDA/g, respectively).
Conclusions: It was remarkable that malnutrition-induced elevation in maternal hepatic oxidative stress persisted up to the weaning. In the offspring, early postnatal treatment with α-tocopherol led to a reprogramming of the prenatal malnutrutrion-programmed increment in renal oxidative stress.

Su077 THERAPEUTIC POTENTIAL OF THE NOVEL DUAL PI 3-KINASE/mTOR INHIBITOR IN THE Han:SPRD RAT MODEL OF PKD

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Introduction and Aims: Both the mammalian target of rapamycin (mTOR) and Akt pathway were shown to be aberrantly activated in the polycystic kidney disease (PKD). Blocking the over activated mTOR pathway in animal models of PKD halts cyst growth incompletely. NVP-BEZ235, a novel dual phosphatidylinostiol 3-kinase (PI3K)/mTOR inhibitor can inhibit the catalytic activity of these kinases by binding to the ATP binding cleft of these proteins. In order to evaluate the cellular responses of NVP-BEZ235 in a model of PKD, we analyzed the effect and the mechanism of NVP-BEZ235 on proliferation and apoptosis in renal primary tubular epithelial cells derived from Han;SPRD rats (Cy/+ TECs).
Methods: The effect of NVP-BEZ235 (c= 0.01nM to 1000nM) on Cy/+ TECs proliferation was assessed by cell counting, MTS and BrdU assays. Induction of apoptosis was detected by western blotting (caspase 3 cleavage) and cytofluorimetry (distribution of annexin V). Expression and activation of proteins, targets either mTORC1 (Thr308p Akt, Thr37/46p 4EBP, Thr421/Ser424p S6K, Ser235/236p S6) or mTORC2 (Ser473p Akt, Thr32p Foxo3a, Ser21/9p GSK3) and of the mitogen-activated protein kinases (MAPK) signaling pathway were evaluated by western blotting by using phosphor-specific antibodies.
Results: NVP-BEZ235 reduced total cell numbers, cell viability and DNA synthesis of Cy/+ TECs without inducing complete inhibition at 1000nM,

which is the highest concentration tested in these studies. Cleavage fragments of caspase 3 could not be detected and the proportion of PI-/Annexin V+ and PI+/Annexin V+ cells treated with NVP-BEZ235 remained substantially unchanged. The phosphorylation of the downstream effectors of mTORC1 (Thr37/46p 4EBP, Thr421/Ser424p S6K, Ser235/236p S6) were prevented at a concentration lower than the one blocking upstream regulators of mTORC1 (such as Thr308p Akt). NVP-BEZ235 suppressed phosphorylation of Akt at Ser473 site, the readout of mTORC2 activity whereas the phosphorylation status of other downstream effectors of the mTORC2 (Thr32p Foxo3a, Ser21/9p GSK3) remained mostly unchanged. On the contrary, phosphorylation of the MAPK ERK1/2 increased (indicating activation) upon NVP-BEZ235 treatment in a dose-dependent way.
Conclusions: NVP-BEZ235 inhibited the proliferation of Cy/+ TECs incompletely in the concentration range tested and apoptosis was not observed in vitro. A partial lack of sensitive of mTORC1 upstream regulators and mTORC2 downstream effectors, as well as an activation of the MAPK signaling pathway occurs in parallel with these effects. Our data suggest that additional studies in PKD models are warranted to further assess the therapeutic potential of NVP-BEZ235 in this disease.

Su078 ADPKD TO BE ADDED TO THE LIST OF CONDITIONS WITH ELEVATED FGF23 LEVELS AND APPARENT RENAL LEAK OF PHOSPHORUS IN PATIENTS WITH NORMAL GFR

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Introduction and Aims: Fibroblast growth factor 23 (FGF23) and parathyroid hormone (PTH) serum levels have been shown to rise following progressive loss of glomerular filtration rate (GFR) and counteract phosphorus retention in patients with chronic kidney disease (CKD). At CKD stage 1 these levels are meant to still be comprised within respective normal ranges but this is difficult to demonstrate. To address the issue we studied 2 cohorts of patients with slowly progressive CKD whose early diagnosis is given by the nature of the disease: autosomal dominant polycystic kidney disease (ADPKD) and diabetes mellitus type 2 (DM2).
Methods: Serum levels of FGF23 were measured in patients with ADPKD (n=100) and DM2 (n=26), and results compared with those obtained in 20 healthy volunteers, all with similar eGFR (in ADPKD 101±19 vs. DM2 101±26 vs. volunteers 101±14 ml/min, mean ± SD).
Results: In ADPKD, FGF-23 levels were 163±33 RU/ml, significantly higher than in DM2 (40±56) and in healthy volunteers (28±22) (ANOVA, P<0.0001). After stratification of eGFR in <90, 90-120, >120 ml/min for ADPKD and healthy volunteers, respectively, the difference remained highly significant in each of the strata for FGF23, whereas neither PTH, nor 25(OH)D, nor 1,25(OH)2D levels differed between both groups at each strata. After selection of subjects with microalbuminuria or of subjects with hypertension the respective difference in FGF23 levels remained highly significant between ADPKD and DM2. The FGF23 levels were correlated neither with age nor with eGFR over the range studied for these parameters (18 to 41 years and 66 to 152 ml/min, respectively). Prevalence of subjects with serum phosphorus< 0.87 mM was 38% in ADPKD, 27% in DM2 and 5% in healthy volunteers (χ^2, P=0.01). TmP/GFR was lower in ADPKD (0.81 mM/GFR) than in healthy volunteers (0.88 mMol/GFR, P=0.04; not measured in DM2), the correlation between serum phosphorus and TmP/GFR was highly significant both in ADPKD (r=0.92, P<0.0001) and in healthy volunteers (r=0.93; P<0.0001).
Conclusions: The finding of elevated FGF23 levels in ADPKD with normal renal function, normal PTH and apparent renal leak of phosphorus is a newly described feature of ADPKD. The origin of the disorder and the consequences of this disturbance remains to be elucidated.

Su079 NEUTROPHIL GELATINASE-ASSOCIATED LIPOCALIN (NGAL): GENOTYPE-PHENOTYPE CORRELATIONS IN AUTOSOMAL DOMINANT POLYCYSTIC KIDNEY DISEASE

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Introduction and Aims: Autosomal dominant polycystic kidney disease (ADPKD) is the most frequent genetic cause of chronic kidney disease (CKD) and renal failure in adults. It is associated with large inter- and intra-familial variability, which can be explained to a large extent by its genetic heterogeneity and modifier genes.
ADPKD is caused by mutations of PKD1 and PKD2 genes, accounting for 85% and 15% of cases respectively.
NGAL, a small 25-kD protein massively released from kidney tubular cells after harmful stimuli, is one of the most promising next-generation biomarkers in clinical nephrology. Recent studies also suggest a possible role for NGAL in CKD and it may be involved in the pathophysiological process of these diseases, such as polycystic kidney disease and glomerulonephritis. Our aim was to evaluate the relationship of NGAL and severity of CKD in 10 ADPKD families with confirmed mutations in PKD1.
Methods: We measured NGAL, creatinine (Cr), urea in 12 ADPKD patients on dialysis (ADPKD-RRT: 5 on pd, 7 on hd), 16 of their relatives not on dialysis (ADPKD-No RRT), 15 wild-type relatives (WT) and 30 healthy controls (Cont).
4ml of whole blood in EDTA tubes were collected from each subject. A point-of-care testing device and test (Triage®MeterPro, Biosite Inc, San Diego, CA) was used to measure plasma NGAL. Creatinine and urea were measured according to standard methods in the routine clinical laboratory. The eGFR was calculated with the 4-variable standardized-MDRD formula (http://mdrd.com).
Results:

	ADPKD-RRT	ADPKD-No RRT	WT	P
Age	60±12	46±13	57±9	0.009
NGAL (pg/mL)	581 (534,638)	62 (60,105)	60 (60,72)	<0.001
Cr (mg/dL)	9.3 (6.9,10.7)	1.2 (1.0,2.0)	0.8 (0.8,0.9)	<0.001
Urea (mg/dL)	133 (110,142)	52 (34,78)	28 (24,38)	<0.001
eGFR (ml/min/1.73 m^2)	N/A	64 (38,72)	79 (71,89)	0.003

Plasma NGAL values in ADPKD-RRT were significantly higher than Cont (median 61.7 pg/ml, p<.001). However NGAL in ADPKD-No RRT and WT were both similar to Cont (p>.05 for both).
ADPKD-RRT patients had significantly higher NGAL, creatinine and urea levels compared to ADPKD-No RRT and WT. ADPKD-No RRT patients were significantly younger, had higher urea and Cr levels, and lower eGFR compared to WT relatives. NGAL levels were similar between these 2 groups. NGAL correlated well with Cr (r=.81, p<.001), urea (r=.76, p<.001) and inversely with eGFR (r=-.79, p<.001). Among ADPKD-No RRT patients, 6 had CKD while 10 had eGFR≥60 ml/min/1.73m^2. NGAL levels were significantly higher in CKD group (median 138 vs 60 pg/ml, p=.01).
Conclusions: Ours is the first to study NGAL in the context of genotype-phenotype. In ADPKD, NGAL was higher in patients already on RRT compared to their affected relatives not on RRT. In these relatives, NGAL was higher in those with CKD. Wild-type relatives had normal NGAL levels.

Su080 RENAL SONOGRAPHIC APPEARANCE OF MEDULLARY CYSTIC KIDNEY DISEASE TYPE 1

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Introduction and Aims: Medullary cystic kidney disease type 1 (MCKD1) is a progressive tubulointerstitial nephropathy with autosomal dominant inheritance, leading to End Stage Renal Disease (ESRD) in the 4th to 7th decade of life. Renal cysts are not always present, especially in early stages and thus are not considered essential for diagnosis. The aim of this study was to present the sonographic findings among MCKD1 gene carriers and evaluate their significance for disease diagnosis.
Methods: 55 MCKD1 gene carriers (28 M, 27 F) underwent abdominal ultrasound and were compared with 55 age, sex and renal function matched controls. Cysts were classified as cortical, medullary and corticomedullary. Creatinine clearance was calculated using Cockroft–Gault formula, corrected for body surface area of 1.73 m^2. Renal function was classified according to Chronic Kidney Disease (CKD) classification of The Kidney Disease Outcomes Quality Initiative. Comparison between the groups was performed using χ^2 test or Fisher's Exact Test accordingly.
Results: Cysts were present at 28 (51%) and 19 (34.5%) patients of MCKD1 and control group respectively (p=0.83). When GFR<60, cysts were present at 68.2% and 59% of the MCKD1 and control group respectively (p=0.531). For GFR>60 the respective values were 39.4% and 18.2% (p=0.057). No statistically significant difference was detected when comparing the number of cysts between the 2 groups (p=0.182). The presence and the number of cysts at each stage of CKD between the 2 groups was (p=0.357, p=0.87, p=0.699, p=1.0, p=0.505) and (p=0.501, p=0.330, p=0.536, p=1.0, p=0.325) respectively, not statistically significant. Among 28 MCKD1 gene carriers with cysts, 21% had medullary, 35.7% cortical and 42.8% corticomedullary cysts, while for the 19 controls with cysts the respective values were 10.5%, 73.7% and 15.8% (p=0.037). For GFR<60, 13.3% of the MCKD1 group and 7.7% of the control group had medullary, 53.3% and 69.2% had cortical and 33.3% and 23% had corticomedullary cysts (p=0.686). Respective values for GFR>60 were, 30.8% and 16.7%, 15.4% and 83.3%, 54% and 0% (p=0.013). By classifying cyst to cortical and non cortical, for GFR<60, cortical cysts were present in 53.3% and 69.2% and non cortical cysts in 46.6% and 30.8% of MCKD1 and control group respectively (p=0.390). For GFR>60 the respective values were 15.4% and 83.3% for cortical and 84.6% and 16.6% for non cortical cysts (p=0.04). The presence of cysts to one or both kidneys between the 2 groups was not statistically significant (p=0.212).
Conclusions: The prevelance of non cortical cysts is significantly higher in MCKD1 gene carriers with GFR>60 than in age, sex and renal function controls. Therefore, their presence in such patients should raise the suspicion of MCKD1. Larger series are needed to establish whether the presence of cyst is a diagnostic clue.

Su081 KIDNEY VOLUME QUANTIFICATION FROM mri WITHOUT INJECTION OF CONTRAST MEDIUM IN ADPKD PATIENTS

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Introduction and Aims: In patients with autosomic dominant polycystic kidney disease (ADPKD), kidney volume (KV) growth is assumedly the best marker predicting the disease progression and the risk of renal insufficiency. A reliable estimation of kidney volume is needed to achieve a quantitative monitoring of kidney growth. KV has been mostly evaluated through imaging techniques, computed tomography (CT) and magnetic resonance imaging (MRI) using contrast medium (CM) by manually tracing left and right kidney contours on each acquired plane; the volume is then computed taking into account the slice thickness and applying the disc summation method. The use of CM is potentially nephrotoxic and the manual procedure is tedious and time-consuming. In this study we present a fast and automatic method for KV assessment from MRI acquired without the injection of contrast medium.
Methods: T2-weigheted SPIR sequences were used to acquire axial images (TR=1200 ms TE=5 ms) on 15 patients (age: 24/60 years) with ADPKD and normal renal function (serum creatinine < 1.3 mg/dl or a creatinine clearance >70 ml/min/1.73 m^2). In all patients the diagnosis of ADPKD was made with echography investigation based on Ravine criteria. The MRI datasets were analyzed using custom software, which allows automatic kidney contour detection, starting from two initial seed points manually selected in the left and right kidneys on one plane. The segmentation procedure is based on a region-based method and exploits the fact that pixels with the same semantic information have similar gray values. The rough kidney contours resulting from the preliminary region growing application are then optimized by applying morphological operators and curvature motion. The area inside each contour is computed and the disc summation method is applied to calculate right and left KVs. Linear regression and Bland-Altman analyses were used to compare our automated volume estimates versus measurements obtained by manually tracing kidney contours.
Results: The time required to analyze each data set was less than 4 minutes. KV ranged between 188/2900 ml (mean: 772±600 ml). Linear regression analysis between automatic and manual measurements resulted in excellent correlation coefficient and regression slope (r=0.99, y = x-29.6). Bland-Altman analysis showed a small bias (-26.2 ml) reflecting a systematic error of -3.4%. The 95% limits of agreement were relatively narrow (49 ml) providing additional support to the tight agreement between the two techniques.
Conclusions: This preliminary study shows the feasibility of automatic segmentation of kidneys. It provides the basis for fast, automated assessment of kidney volume which is potentially clinically useful for assessment of kidney volume growth.

Genetic diseases and molecular genetics

Su082 PROGNOSTIC INDICATORS OF RENAL DISEASE PROGRESSION IN FABRY DISEASE

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Introduction and Aims: Renal dysfunction is a common and serious manifestation of Fabry disease (FD), an X-linked metabolic disorder caused by deficiency of α-galactosidase A activity. Longitudinal renal data from the Fabry Registry were analyzed to better understand the natural progression of renal disease over time.
Methods: Data from 2,850 adult Fabry Registry patients were examined to evaluate longitudinal renal function in untreated patients with FD. A total of 462 untreated patients (121 males and 341 females age ≥18 years as of July 2009) who had 2 or more estimated glomerular filtration rate (eGFR) values over a span of ≥12 months during the natural history period and 1 or more corresponding urinary protein to urinary creatinine ratio (UP/Cr) values reported were included in these analyses. The CKD-EPI equation was used to estimate GFR.
Results: Most males (86 of 121, 71%) had more rapidly progressing renal disease than the normal adult population (defined as eGFR loss of more than 1 mL/min/1.73m^2 per year), whereas fewer females (133 of 341, 39%) had this condition. Patients with more rapidly progressing renal disease had significantly higher mean UP/Cr values than patients with slower progression (1.5 versus 0.2 for males and 1.4 versus 0.5 for females,

$p<0.0001$). Renal function in males declined more rapidly with higher UP/Cr, with the steepest declines observed in 30 males with UP/Cr >1.5. Compared to males, eGFR slope declined more slowly in females, with the steepest declines observed in 85 females with UP/Cr >1.2. Regression models of eGFR slope indicated that UP/Cr is the most important factor in renal disease progression in Fabry males. In addition to UP/Cr, lower baseline eGFR levels and lower endogenous α-galactosidase activity were also associated with faster renal disease progression in Fabry females.

Conclusions: Untreated male and female adult patients with FD who have significant proteinuria lose renal function more quickly than those with little or no urinary protein excretion. Urinary protein excretion and eGFR levels should be closely monitored in all Fabry patients, regardless of other signs or symptoms. Understanding the natural history by age, gender and baseline kidney function and proteinuria are essential for interpreting the effects of enzyme replacement therapy (ERT) on kidney function outcome. To date, these analyses include the largest number of women with FD for whom longitudinal renal data are available.

Disclosure: The Fabry Registry is sponsored by Genzyme Corporation. AO, DPG, JPO, BV, SW, CW, and DGW serve on the Genzyme-sponsored Fabry Registry Boards of Advisors. SW and DGW have served as a paid consultant to Genzyme Corporation. JPO and SW have received speaking fees from Genzyme Corporation. JPO, AS, SW, and DGW have received travel and research support from Genzyme Corporation.

Su083 NEPHROCYSTIN-4 CONTROLS PROTEIN-PROTEIN INTERACTIONS AND SUBCELLULAR LOCALISATION OF NEPHROCYSTIN-1

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Introduction and Aims: Nephronophthisis is the most common genetic cause of end stage renal failure during childhood and adolescence. The most frequent mutations affect the NPHP1 gene encoding for the protein Nephrocystin-1. While less frequent, mutations in NPHP4 lead to a very similar clinical presentation as mutations in NPHP1. Interestingly, both NPHP1 and NPHP4 are evolutionarily conserved and can be found in the nematode C. elegans, in which they play a crucial role for the maintenance of both ciliary function and morphology. However, the function and regulation of both proteins so far are poorly understood.

We have previously shown that Nephrocystin-1 interacts with the tyrosine kinase Pyk2 and gets phosphorylated by Pyk2 at three defined residues. This phosphorylation is controlled by Nephrocystin-4. We now see that the tyrosine phosphorylation of Nephrocystin-1 increases its affinity to the the trans-golgi network transporter protein PACS-1. This interaction is controlled by Nephrocystin-4. As it has previously been shown that the interaction with PACS-1 is important for the right loalization of Nephrocystin-1 at the ciliary base we investigated whether knockdown of Nephrocystin-4 affects Nephrocystin-1 localization in ciliated human epithelial cells. Indeed knockdown of Nephrocystin-4 changes the subcellular localization of Nephrocystin-1 to a more golgi-oriented pattern. Together these data point to a role of Nephrocystin-4 in cellular signalling pathways upstream of Nephrocystin-1.

Su084 DIAGNOSTIC PROCEDURES FOR HEREDITARY RENAL HYPOURICEMIA

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Introduction and Aims: Primary hereditary renal hypouricemia is a inherited disorder characterized by defective renal urate reabsorption with severe complications such as exercise-induced acute renal failure and nephrolithiasis. The known major causes are: defects in the SLC22A12 gene, which encodes the human urate transporter 1 (hURAT1), and also impairment of the recently identified voltage urate transporter (URATv1), encoded by SLC2A9 gene. Diagnosis is based on two biochemical markers: hypouricemia and increased fractional excretion of uric acid (FE-UA). Therapy involved alkalization of urine, drinking plenty of water, and avoidance of strenuous exercise. To date, the cases with mutations in hURAT1 gene have been reported in East Asia only. 19 loss of function mutations have been described in over one hundred Japanese patients. The number of Japanese patient is unique worldwide. Hypouricemia is sometimes overlooked, therefore we have set up the complete investigations for this disorder.

Methods: The patients (with repetitive serum uric acid lower than 60 μmol/l and FE-UA higher than 43%) were selected for molecular analysis of SLC22A12 and SLC2A9 genes from the group of 569 Czech hypouricemic cases. These hypouricemic individuals were found from 3500 blood and urine samples. Serum and urinary UA and creatinine were determined. The sequence analysis by automated DNA sequencer (Applied Biosystems 3100) of the coding region of SLC22A12 and SLC2A9 genes were performed after informed consent.

Results: The proposed scheme for the investigation was as follows. As the first step – secondary causes of hyperuricosuric hypouricemia were excluded. The estimation of: 1) serum uric acid, 2) excretion fraction of UA, 3) and analysis of hURAT1 and URATv1 genes follow then. Using this flow chart we were able to find three transitions G366R, T467M, R477H and one deletion A416_L418del in SLC22A12 gene in 8 Czech patients. In addition, sequence analysis of SLC2A9 gene revealed three unpublished transitions G216R, D281H N333S; one insertion with frame shifting change p.[I118HfsX27] and four sequence variants G25R,V282I, R294H, P350L in two heterozygotes, five compound heterozygotes and two homozygotes. Sequence variants p.[I118HfsX27] and N333S were not detected in 300 control alleles (150 samples) by restriction assay. Three patients were suffering from acute renal failure and urate nephrolithiasis.

Conclusions: Our estimation of the defect in URATv1 gene gives further evidence that SLC2A9 is a causative gene of renal hypouricemia and supports the important role of SLC2A9 in regulation of serum urate levels in humans. Hereditary renal hypouricemia is still unrecognized condition and probably not wide spread in Japan and Korea only. This disorder belongs to differential diagnosis of the causes of exercise-induced acute renal failure and nephrolithiasis. (Supported by grant VZMSM0021620806, Czech Republic).

Su085 ★ THE WILMS TUMOR SUPPRESSOR PROTEIN (WT1) SPLICEVARIANTS WT1(-KTS) AND WT1(+KTS) SHOW DISTINCT INFLUENCES ON DIFFERENT CONTROL LEVELS OF THE RENIN GENE EXPRESSION

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Introduction and Aims: Renin plays a crucial role in the control of various physiological processes such as blood pressure and body fluid homeostasis. Clinical studies indicate that Renin gene expression is controlled by the WT1 splicevariants WT1(-KTS) and WT1(+KTS). We investigated the underlying mechanisms through which they influence the Renin gene expression.

Methods: To investigate the influences of the WT1 splicevariants on Renin gene expression we used bioinformatic tools as well as molecularbiological techniques such as reporter gene assays, siRNA knockdown experiments, qPCR and Western blotting. For these experiments different cell lines and tissues were used. The effects of WT1 overexpression on Renin gene expression was investigated in several transiently and stable transfected cell lines. The co-expression of Wt1 and Renin in kidney sections could be shown by using the immunostaining technique. To investigate the Renin mRNA localisation in WT1 stable transfected cells we used ultracentrifugation and qPCR.

Results: Using bioinformatics tools, we initially predicted that a WT1-binding site exists in a regulatory region about 12 kb upstream of the Renin promoter; this was confirmed by reporter gene assays and gel shift experiments in several cell lines. Co-expression of Wt1 and Renin proteins was

found in rat kidney sections, mouse kidney blood vessels, and a cell line derived from the juxtaglomerular apparatus that produces Renin. Knockdown of WT1 protein by siRNA significantly increased the cellular Renin mRNA content, while overexpression of WT1(-KTS) reduced Renin gene expression in stable and transiently transfected HEK293 cells. A mutant WT1(-KTS) protein found in Wilms' tumors failed to suppress Renin gene reporter activity and endogenous Renin expression. Overexpression of both WT1 variants in human kidney cells decreases the Renin protein level. Reporter gene studies demonstrate that the WT1 splice variants modulate the Renin promoter activity either directly (+KTS form) or through interactions within a regulatory region [hRENc] (both variants). The WT1(+KTS) variant is able to interact with the Renin mRNA-UTRs in contrast to the WT1(-KTS) form. We showed that the Renin mRNA undergoes a translocation into a translational inactive compartment after overexpression of the WT1(+KTS) variant.
Conclusions: The posttranscriptional influence of WT1(+KTS) may be responsible for a translational arrest of Renin synthesis, which explain the decrease of the Renin protein following WT1(+KTS) overexpression. Both variants of the Wilms Tumor Suppressor Protein inhibit the Renin synthesis through distinct and different mechanisms and this may explain findings in patients with WT1 gene mutations of increased plasma Renin and hypertension.

Su086 CLINICOGENETIC ANALYSIS OF KOREAN PATIENTS WITH GITELMAN'S SYNDROME

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Introduction and Aims: Gitelman's syndrome (GS) is autosomal recessive disorder characterized by hypokalemic metabolic alkalosis, hypomagnesemia, and hypocalciuria. However, clinical features were recently reported to be diverse. Although gene analysis of *SLC12A3* encoding NaCl cotransporter (NCC) is known to be the best way to diagnose GS, mutations in *CLCNKB* encoding ClC-Kb can also be the cause of GS. We investigated the clinical features, usefulness of thiazide test, and genetic variations of Korean patients with Gitelman's syndrome.
Methods: In this study, 24 Korean GS patients (16 males and 8 females, age 28±13.1 yr) were enrolled. All of the subjects denied factitious vomiting and use of diuretics or laxatives. Their clinical history, biochemical features, results of thiazide test, and mutation analysis of *SLC12A3* and *CLCNKB* were compared. All coding regions, including intron-exon boundaries, of *SLC12A3* and *CLCNKB* were analyzed.
Results: All the subjects had hypokalemia, metabolic alkalosis, normal blood pressure. Prominent hypocalciuria was observed in 20 patients. Nine of the 24 patients did not have hypomagnesemia. Thiazide test was adequately performed in 20 patients and the results of 19 out of the 20 patients were consistent with GS. *SLC12A3* gene mutations were detected in 21 patients (9 compound heterozygous, 7 heterozygous, and 5 homozygous mutations) and not detected in 3 patients. Mutations in *CLCNKB* were identified with *SLC12A3* mutations in 2 patients (1 heterozygous; W530L, 1 homozygous; W610X) and without *SLC12A3* mutations in 1 patient (homozygous; E199X). The patient who had simultaneous homozygous mutation in *CLCNKB* and heterozygous *SLC12A3* mutation showed more severe clinical presentation (early onset, mild growth retardation, polyuria, normocalciuria) but no distinct phenotypic variation was identified in the other patient with both heterozygous *CLCNKB* and heterozygous *SLC12A3* mutation. The patient with only homozygous *CLCNKB* mutation showed reduction of distal fractional chloride reabsorption (DFCR) in response to thiazide by more than 50%, normocalciuria and normomagnesemia. Some GS patients may have normomagnesemia. The diagnosis of GS primarily can depend on the clinical criteria and thiazide test. Gene analysis of *SLC12A3* is complementary and may misleading due to intrinsic limitation of the analysis method. Identification of *CLCNKB* mutation should also be considered.
Conclusions: In the diagnosis of GS, clinical criteria and genetic analysis of *SLC12A3* are mutually complementary and identification of *CLCNKB* mutation should also be considered.

Su087 ★ NEW AND *DE NOVO TRPC6* MUTATIONS IN CHILDREN WITH STEROID-RESISTANT NEPHROTIC SYNDROME

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Introduction and Aims: Mutations in the *TRPC6* gene, encoding a member of the transient receptor potential (TRP) superfamily of ion channels, have been reported in eight families with adult-onset autosomal dominant focal segmental glomerulosclerosis (FSGS) and three cases of non-familial FSGS. In the present study, we investigated the role of TRPC6 mutations in Italian patients with early onset steroid-resistant nephrotic syndrome (SRNS) and familial adult-hood onset of FSGS.
Methods: *TRPC6* mutation analysis was performed by PCR and direct sequencing in 33 Italian patients with sporadic SRNS and in 3 Italian family with adult-onset autosomal dominant FSGS. A normal control group of 100 healthy donors was also screened. All patients were previously analysed for *NPHS1*, *NPHS2*,*ACTN4* and *WT1* genes. A comparative analysis in five different species of TRPC6 amino acid sequences was done by ClustalW software. Moreover, the expression levels of TRPC6 and nephrin proteins were evaluated by indirect immunofluorescence on renal biopsies from patients with *TRPC6* mutations and from control subjects.
Results: Three missense substitutions (c.374A>G_p.N125S; c.653A>T_p.H218L; c.2684G>T_p.R895L) were identified in four cases of early onset SRNS. p.N125S mutation, previously described in a 42-year-old female with adult-onset FSGS, was found in two siblings, resulted heterozygotes also for a mutation (p.R831C) in the *NPHS1* gene (digenic inheritance). pH218L mutation, described for the first time in this report, was found in a 18 year-old boy, who developed a severe form of SRNS at the age of 8 years. p.R895L, the first *de novo* mutation described in *TRPC6* gene, was detected in a female child with unusual clinical features: collapsing glomerulosclerosis in childhood and rapid progression to end stage renal disease. Both healthy parents, wild type for detected mutation, were analyzed by microsatellite markers to ensure paternity and maternity. Moreover, sequence alignments showed a high evolutionary conservation of TRPC6 amino acid substitutions and indirect immunofluorescence on renal biopsies from patients with TRPC6 mutations revealed up-regulated expression levels of TRPC6 and a faint and irregular distribution along the capillary loops for nephrin protein. No mutations were found in familial adult-onset FSGS patients.
Conclusions: We show that TRPC6 mutations are not so rare in children with SRNS (4/33) and describe for the first time a *de novo TRPC6* mutation in a severe form of childhood collapsing glomerulosclerosis, suggesting that *TRPC6* gene may contribute to early and late-onset SRNS with a very variable penetrance and in different ways: hereditary or de novo mutations.

Su088 OVEREXPRESSION OF LOX-1 IN BIOPTIC TISSUE OF ANCA-ASSOCIATED GLOMERULONRPHRITIS PATIENTS

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Introduction and Aims: The renal outcome of patients with ANCA-associated glomerulonephritis is highly variable. Previous investigations have demonstrated that the renal function and prognosis of this disease on long-term follow-up correlate with the active and chronic histological lesions in renal bioptic specimens.

The aim of the present retrospective study was to investigate inflammatory factors which correlate with the progressivity of ANCA- associated glomerulonephritis expressed by a histometric method in renal bioptic specimens.
Methods: Bioptic samples from 10 ANCA-associated glomerulonephritic patients were investigated; the mean age was 55.2 years (27-79). 7 were female, with a disease distribution of 3 Wegener's granulomatosis, 2 renal-related vasculitis and 5 microscopic polyarteritis as determined by immunofluorescence and electronmicroscopy. The activity and chronicity indices of the biopsies were evaluated according to a standardized scoring protocol. A maximum of 35 points were available for acute lesions, and 22 points for fibrosing/sclerotic lesions. ANCA positivity and the cytoplasmic and perinuclear staining patterns were determined by an indirect immunofluorescence technique and ELISA was performed for antibodies to PR3 and MPO. For gene expression analysis quantitative real-time was carried out with gene-specific primers and SYBR Green protocol. Relative expression ratios were normalized to two housekeeping genes, tubulin and hypoxanthine phosphoribosyltransferase. Relative changes in expression of the LOX-1 gene from bioptic samples were investigated.
Results: We found that LOX-1gene was up-regulated in 6 of the 10 bioptic samples. We have investigated the correlation of the relative expression of the LOX-1 gene in the bioptic samples and the samples in which the activity index was ≥ 15, indicating the significant activity of the disease, the LOX-1 gene was overexpressed. No correlation could be detected between the LOX-1 expression and the chronicity index. These results correlate well with the recent findings of Hu et al. (Kidney International 2010; 76, 521), who found that the deletion of LOX-1 attenuates renal injury.
Conclusions: Our results support the hypothesis that LOX-1 is an important modulator in the development of renal damage, and that this factor possibly relates with the pathogenetic processes of ANCA-associated glomerulonephritis.

Su089 UROMODULIN IS EXPRESSED IN RENAL PRIMARY CILIA AND *UMOD* MUTATIONS RESULT IN DECREASED CILIARY UROMODULIN EXPRESSION

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Introduction and Aims: *Uromodulin* (*UMOD*) mutations are responsible for three autosomal dominant tubulo-interstitial nephropathies including medullary cystic kidney disease type 2 (MCKD2), familial juvenile hyperuricemic nephropathy (FJHN), and glomerulocystic kidney disease (GCKD). Symptoms include renal salt wasting, hyperuricemia, gout, hypertension and end-stage renal disease. MCKD is part of the "nephronophthisis-MCKD complex", a group of cystic kidney diseases. Both disorders have an indistinguishable histology and renal cysts are observed in either. For most genes mutated in cystic kidney disease, their proteins are expressed in the primary cilia/basal body complex. We were interested if UMOD protein was expressed in the primary renal cilia of human renal biopsies and if mutant UMOD would show a different expression pattern compared to that seen in control individuals.
Methods: In order to identify patients with *UMOD* mutations we performed mutation analysis of the *UMOD* gene in affected patients. We obtained kidney biopsies of two affected patients and compared the number of cilia and ciliary UMOD expression with kidney biopsies of patients with other tubulo-interstitial kidney diseases and healthy individuals. We studied ciliary UMOD expression in cell culture by immunofluorescence and electron microscopy. Mutant UMOD contructs were transfected in IMCD3 cells and analyzed for ciliary UMOD expression. Co-localization of other ciliary expressed proteins as kinesin family member 3A (KIF3A) and nephrocystin-1 with UMOD was studied.
Results: We identified seven novel and three previously published *UMOD* mutations. We demonstrate that UMOD is expressed in the primary cilia of renal tubules, using immunofluorescent studies in human kidney biopsy samples. The number of UMOD positive primary cilia in UMOD patients is significantly decreased compared to healthy individuals and patients with other tubulo-interstitial kidney disease. Mutant *UMOD* constructs lack ciliary UMOD expression in cell culture. Additional immunofluorescence studies confirm ciliary expression of UMOD in cell culture. Ciliary expression of UMOD is also confirmed by electron microscopy. UMOD localization at the mitotic spindle poles and colocalization with other ciliary proteins such as nephrocystin-1 and KIF3A is demonstrated.
Conclusions: Our data adds UMOD to the group of proteins expressed in primary cilia where mutations of the gene lead to cystic kidney disease.

Su090 COMMON GENETIC VARIANTS ARE ASSOCIATED WITH CHRONIC KIDNEY DISEASE STAGE 3B: THE CKDGen CONSORTIUM

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Introduction and Aims: Chronic kidney disease (CKD) is associated with an increased risk of end-stage renal disease, cardiovascular disease, and all-cause mortality. Compared with the commonly used definition of CKD as estimated glomerular filtration rate (eGFR) of <60 ml/min/1.73m^2, these associations are more pronounced for Stage 3b CKD (defined as GFR<45 ml/min/1.73m^2). Studies assessing the genetic contribution to eGFR have been conducted but genome-wide association (GWA) studies investigating novel genomic loci in association with Stage 3b CKD have not yet been performed. Taking advantage of the availability of several studies with GWA markers and renal phenotype information in the CKDGen consortium, we performed a meta-analysis of GWA data for Stage 3b CKD in 53,503 Caucasian samples from 12 population-based studies.
Methods: GFR was estimated from calibrated serum creatinine with the abbreviated MDRD Study equation in each study. Individuals were classified as cases if eGFR<45 ml/min/1.73m^2 or as controls if eGFR>60 ml/min/1.73m^2. Genetic association was assessed for $\sim$2.5 million genotyped and imputed single nucleotide polymorphisms (SNPs) per participant in each study. Results were pooled using a fixed-effects meta-analysis with inverse-variance weighting, with a minor allele frequency filter of 5% for studies with <50 cases. To assess the between-study heterogeneity a random-effect meta-analysis was repeated for SNPs with p-value$\leq 1\times10^{-5}$ in the fixed-effects analysis. According to the I^2 statistic heterogeneity was classified as null to low ($I^2\leq25\%$), moderate ($25\%<I^2\leq50\%$), high ($50\%<I^2\leq75\%$), and very high ($I^2>75\%$). This classification enabled us to quantify to which extent significant results could be considered as common genetic variants and which ones were instead population-specific findings.
Results: Overall, 1083 cases (range: 22-199 per study) and 52,420 controls (range: 637-21,940 per study) were analyzed. We identified 20 novel loci associated with Stage 3b CKD with null to low evidence for heterogeneity of effect size and direction: 10 loci with p-value$\leq5\times10^{-6}$ and 10 with p-value$\leq5\times10^{-5}$. Three additional loci were associated with Stage 3b CKD with p-value$\leq5\times10^{-5}$ but with moderate between-study heterogeneity. Moreover, we validated the *UMOD* locus, which we previously found associated with eGFR and CKD, as associated with Stage 3b CKD (OR=1.35, p-value=3.3×10^{-6}).
Conclusions: Genome-wide association for Stage 3b CKD is feasible in population-based studies. Our results await external replication and suggest that novel loci can be uncovered using this more extreme phenotype.

Su091 THE INTERLEUKIN-6 GENE PROMOTER POLYMORPHISM –174 IS PREDICTIVE OF ATHEROSCLEROTIC EVENTS IN OVERWEIGHT TRANSPLANTED PATIENTS

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Introduction and Aims: Chronic inflammation plays a pivotal role in the pathogenesis of atherosclerosis. We hypothesized that combining overweight and a greater genetic capacity to produce IL-6 predicted by *IL-6* gene promoter polymorphism at position –174 (G/C) may allow to identify individuals exhibiting higher IL-6 and C-reactive protein (CRP) concentrations and carrying a higher risk of atherosclerotic events (AE).
Methods: The occurrence of AE was analyzed with respect to body mass index, *IL-6* gene promoter polymorphism at position *–174 (G/C)* and other relevant risk factors, retrospectively in 217 renal transplant recipients, and, prospectively, in 132.
Results: Circulating IL-6 concentrations were closely related to BMI (r=0.55, p= 0.0005). In overweight patients, serum IL-6 concentration was found to be significantly lower in C carriers than in GG patients (4.2 [1.0-5.1] vs 7.3 pg/ml [4.4-100]; p=0.025). Overweight GG patients also had higher CRP levels than other categories of patients (4.9±2.3 vs 3.8±2.7; p=0.015). The incidence of AE was higher in overweight GG patients than in other individuals (29.5% vs 10.1%; p=0.0003).

Table 1. Incidence of atherosclerotic events (AE) in the retrospective, prospective and overall cohort according to IL-6-174 genotype and body mass index (BMI)

	All patients	Overweight patients carrying the GG genotype	Other patients	p
Retrospective cohort (n= 217)	14.7% (n=32)	33% (n=9)	12% (n= 23)	0.006
Prospective cohort (n= 132)	11.8% (n=17)	33% (n=3)	9% (n=12)	0.031
Overall cohort (n=349)	14% (n=49)	29.5% (n=12)	10.1% (n=35)	0.0003

In multivariate analysis, overweight-GG had an increased risk to develop AE (HR 2.96 [95% CI 1.09-8.04], p=0.034 in the retrospective cohort, and HR 2.99 [95% CI 0.92-9.33], p=0.069 in the prospective).

Table 2. Risk factors of cardiovascular events in the retrospective, prospective and overall cohort (multivariate analysis expressed in Hazard Ratio)

	HR	95% CI	P
Retrospective cohort			
Age > 45 y.o	1.07	[1.03–1.12]	0.001
Overweight + IL-6 GG genotype	2.96	[1.09–8.04]	0.034
Prospective cohort			
Age > 45 y.o	1.07	[1.03–1.12]	0.001
Past history of CVE	3.88	[1.09–11.35]	0.042
Overweight + IL-6 GG genotype	2.99	[0.92–9.33]	0.069
Overall cohort			
Age >45 y.o	1.86	[1.03–1.09]	0.0005
Male gender	2.83	[1.22–6.55]	0.016
Overweight + IL-6 GG genotype	3.08	[1.33–7.13]	0.009

Conclusions: All these data are consistent with a role for both genetic (*IL-6* gene promoter polymorphism) and environmental determinants of inflammation (white adipose tissue mass) in the development of AE in renal transplanted patients.

Su092 CALCIMIMETIC R-568 EFFECTS ON ACTIVITIES OF THE CALCIUM-SENSING RECEPTOR R990G POLYMORPHISM

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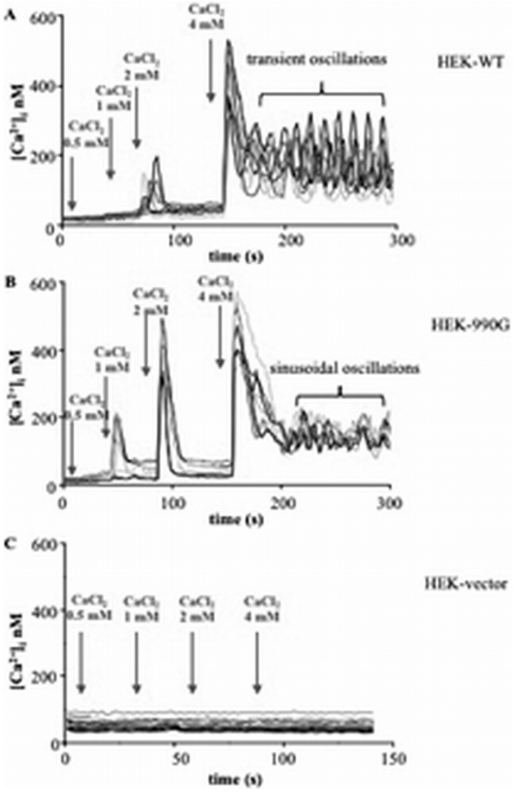
Introduction and Aims: Our previous studies found that the R990G polymorphism of Calcium-Sensing Receptor (CaSR) gene causes a gain of function and resulted associated with hypercalciuria in nephrolithiasis and primary hyperparathyroidism. In the present study we investigated CaSR signaling after stimulation by the extracellular calcium ([Ca2+]o) and the calcimimetic R-568.
Methods: HEK-293 cells were stably transfected with wild type or R990G polymorphic CaSR (HEK-WT and HEK-990G cells, respectively) and spontaneous intracellular calcium oscillations were measured by Fura-2 (specific calcium-dye) in fluorescence microscopy. Time course of [Ca2+]i was analyzed during stimulations with increasing [Ca2+]o (0.5, 1, 2, 4 mM CaCl2) in presence of R-568 (0.01, 0.05, 0.1 μM) or PLC, SERCA and PKC inhibitors or PKC activator. ERK 44/42 phosphorylation levels were measured by western blot.
Results: We found different oscillatory patterns stimulated by [Ca2+]o: transient in HEK-WT and sinusoidal in HEK-990G cells, indicating a different signaling response.
The pre-incubation with SERCA or PLC inhibitors completely prevented oscillations in both cell lines, confirming an involvement of the IP3 pathway. A PKC activator also inhibited oscillations in both cell types, while a PKC inhibitor blocked oscillations in the HEK-WT cells, but not in HEK-990G cells, suggesting that in this case the process was independent from PKC. The 990G allele was also associated with higher CaSR sensitivity to the short-term activation of the p44/42 ERK pathway. HEK-990G cells stimulated with increasing R-568 concentrations showed lower R-568-EC50



than HEK-WT cells, and lower Ca-EC50 in the presence of R-568. The calcimimetic stimulation induced a left shift of the oscillatory events at 2 mM [Ca2+]o instead of 4 mM [Ca2+]o, with a larger percentage of sinusoidal oscillating cells in HEK-990G cells. Also, oscillatory pattern seemed to be influenced by the calcimimetic, shifting from transient to sinusoidal type in both cell lines.
Conclusions: Our findings indicate that cells transfected with 990G allele were more sensitive to [Ca2+]o and to calcimimetic R-568 in the IP3 pathway and provided new evidence for differences in signaling.

Su093 MULTIPLE NEW GENETIC LOCI FOR RENAL FUNCTION ARE ASSOCIATED WITH EGFR ACROSS THE SPECTRUM OF HYPERTENSION AND DIABETES: THE CKDGen CONSORTIUM

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Introduction and Aims: Diabetes and hypertension are key risk factors for chronic kidney disease (CKD). It is uncertain whether genetic loci that underlie estimated glomerular filtration rate (eGFR) in population-based studies are also associated with renal function among patients with diabetes and hypertension. We therefore tested whether genetic risk variants for CKD and eGFR are also associated with eGFR in subsets of patients with and without diabetes and hypertension in the CKDGen Consortium.
Methods: We assessed whether each of 16 novel risk SNPs, identified as associated with eGFR in the overall sample, was associated with eGFR among strata of diabetes (defined as fasting plasma glucose≥126 mg/dl or diabetes treatment) or hypertension (defined as systolic blood pressure≥140 mm Hg, diastolic blood pressure≥90 mm Hg, or hypertension treatment). Fixed effects meta-analyses with inverse-variance weighting were applied to data from 67,093 Caucasian participants of 20 population-based studies (AGES, Amish, ARIC, ASPS, BLSA, CHS, ERF, FamHS, FHS, KORA-F3/F4, Korcula, MICROS, NSPHS, ORCADES, RS-I/II, SHIP, Vis, WGHS). A z-score test was applied to test for heterogeneity of the SNP effect across strata of diabetes and hypertension.
Results: Overall, 4884 participants (7%) had diabetes and 25,383 (38%) had hypertension. Each of the 16 validated SNPs was associated with eGFR in individuals without diabetes (p-value range: 3.8×10^{-17}-0.034). Among individuals with diabetes 9 SNPs were associated with eGFR (p-value range: 1.3×10^{-5}-0.046), with the strongest signals in *UMOD* and *SHROOM3*. These 2 loci also showed some evidence of heterogeneity between diabetics and non-diabetics (p-value=0.023 in both cases). Among participants without hypertension 15 SNPs were associated with eGFR (p-values$\leq2.3\times10^{-5}$), with similar results in the hypertensive group. Only the SNP at the *UMOD* locus was significantly heterogeneous (p-value=0.0018) across strata of hypertension: the effect was twice as strong in those with hypertension compared to those without.
Conclusions: We have identified several genomic regions, associated with eGFR in population-based cohorts, that are also associated with renal function among participants with diabetes and hypertension. These findings suggest that the use of a broad phenotype definition including individuals with and without major non-genetic risk factors for kidney disease allows for the identification of common genetic risk variants important across the spectrum of kidney disease.

Su094 CCR5Δ32 POLYMORPHISM IN PATIENTS WITH CHRONIC PERIAORTITIS (RETROPERITONEAL FIBROSIS)

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Introduction and Aims: Chronic periaortitis (CP) is a rare disease hallmarked by a retroperitoneal periaortic tissue which often entraps the ureters and thus causes acute renal failure. Once thought to be due to advanced atherosclerosis, CP is now considered an autoimmune/inflammatory disease. CP includes non-aneurysmal and aneurysmal forms, known respectively as idiopathic retroperitoneal fibrosis (IRF) and perianeurysmal retroperitoneal fibrosis (PRF). CCR5, a receptor for different chemokines, mediates the recruitment of inflammatory cells into inflamed tissues. The *CCR5* gene has a 32-base-pair deletion (CCR5Δ32) polymorphism, which is associated with autoimmunity. We investigated whether this polymorphism confers susceptibility to CP.
Methods: We enrolled 100 consecutive CP patients and 180 age- and sex-matched healthy controls. DNA was isolated from blood samples using standard techniques and stored at 4°C until analysis. Genotyping was performed with a PCR-based allelic discrimination assay using primers and allele-specific probes. The frequencies of the alleles and genotypes among patients and controls were compared by the chi-square test with the values predicted by the assumption of Hardy-Weinberg equilibrium in the sample. Odds ratios (ORs) and 95% confidence intervals (95% CIs) were calculated. Corrected *P* values (P_{corr}) were calculated by multiplying the *P* value by the number of alleles compared.
Results: Of the 100 CP patients, 82 had IRF and 18 PRF. The CCR5Δ32 allele was significantly more frequent in CP patients than in the controls (P_{corr} = 0.03, OR 2.8 [95%CI 1.2-6.4]). The distribution of allele and genotype frequencies of the CCR5Δ32 polymorphism did not differ significantly between IRF patients and controls. However, the CCR5Δ32 allele was significantly more frequent in PRF patients than in the controls (P_{corr}=0.0002, OR 10.0 [95% CI 3.7-27.3]). We investigated CCR5Δ32 polymorphism association with CP, IRF and PRF stratifying on the presence/absence of established atherosclerotic disease (coronary, cerebrovascular or peripheral); the significant association with CCR5/Δ32 allele was preserved and increased in CP patients without atherosclerotic disease compared to healthy controls (16.4% versus 5.6%, P = 0.012, OR 3.3 [95% CI: 1.4-8.1), but not in those with atherosclerotic disease. Similarly, the significant association with CCR5Δ32 allele was preserved in PRF patients without atherosclerotic disease compared to healthy controls (66.7% versus 5.6%, P = 0.00001, OR 34.0 [95% CI: 7.4-156.3]).
Conclusions: CCR5Δ32 polymorphism is strongly associated with susceptibility to CP, particularly its perianeurysmal form, PRF. This association is independent of atherosclerotic disease. Further studies are warranted to explore the functional consequences of this polymorphism and its role in the pathogenesis of CP.

Su095 GLOBOTRIAOSYLSPHINGOSINE ACTIONS ON HUMAN GLOMERULAR PODOCYTES: IMPLICATIONS FOR FABRY NEPHROPATHY

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Introduction and Aims: Transforming growth factor-β1 (TGF-β1) and the macrophage inhibitory factor receptor CD74 link the metabolic disorder with tissue injury in diabetic nephropathy. Fabry disease is an X-linked lysosomal glycosphingolipid storage disorder resulting from a deficient activity of α-galactosidase A that leads to proteinuric renal injury. However, the link between the metabolic abnormality and renal injury is poorly

characterized. Globotriaosylsphingosine (lyso-Gb3) was recently identified as a bioactive molecule accumulating in Fabry disease. We hypothesized that lyso-Gb3 could modulate the release of secondary mediators of injury in glomerular podocytes. *Aim:* To study the biological activity of lyso-Gb3 in cultured human podocytes.
Methods: Real time PCR and Western blot was used to detect lyso-Gb3 dose- and time-increased TGF-β1, extracellular matrix proteins and CD74 expression. RNA analyses was used to text whether paricalcitol prevented the increase in TGF-β1, CD74 and extracellular matrix induced by lyso-Gb3.
Results: In human podocytes lyso-Gb3 dose- and time-dependently increased the expression of TGF-β1, extracellular matrix proteins and CD74. TGF-β1 mediated lyso-Gb3 effects on extracellular matrix production. Vitamin D receptor activation with paricalcitol prevented the increase in TGF-β1, CD74 and extracellular matrix induced by lyso-Gb3.
Conclusions: Lyso-Gb3 may have a role in glomerular injury in Fabry disease by promoting the release of secondary mediators of glomerular injury common to diabetic nephropathy. These effects are prevented by paricalcitol, raising the issue of vitamin D receptor activation as potential usefulness as adjunctive therapy in Fabry nephropathy.
Disclosure: Alberto Ortiz, consultant, Genzyme.

Su096 NPHS2 GENE MUTATIONS IN RUSSIAN CHILDREN WITH STEROID-RESISTANT NEPHROTIC SYNDROME

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Introduction and Aims: Autosomal recessive steroid-resistant nephrotic syndrome (SRNS) is caused mainly by a mutation in NPHS2 gene. The aim of this study is to determine frequency and spectrum NPHS2 gene mutations in Russian children with SRNS.
Methods: We examined 37 children from 36 different families with steroid-resistant nephrotic syndrome for mutations in NPHS2. Median patients age was 3 years (range 3 month – 15 years). Their renal biopsies showed focal segmental glomerulosclerosis, minimal change disease or mesangial proliferation.
The mutational analysis of NPHS2 was performed by DNA sequencing. Genomic DNA was extracted from leukocytes, and all 8 exons were analyzed.
Results: Two recessive podocin mutations were present in 16, 2% (6 of 37) of children with SRNS. Compound heterozygous mutations of NPHS2 were identified in 3 and homozygous mutation (E87X) in 3. Four patients (two of them are sibs) from 6 had the same nonsense mutation in NPHS2: E87X. Missense mutation R138Q was detected in two patients, one of them had this mutation heterozygously in compound with E87X, and another one had it in compound with V165X. One patient was compound heterozygotes for the R229Q variant and missense mutation T315I. The median age at onset of the disease was 3 years (range 8 month – 15 years) in children without mutations compared with 10, 5 month (range 3 month – 7 years) in patients with mutations. Among 6 nephrotic children with NPHS2 mutations, 4 were before 1 year of age at diagnosis.
Conclusions: NPHS2 mutations are responsible for some cases of SRNS in Russian children. The E87X mutation appears to be common in Russia. Podocin mutations associated with earlier onset of the disease.

Su097 FAMILIAL MEDITERRANEAN FEVER: CLINICAL AND GENETIC CHARACTERIZATION OF GEORGIAN AND AZERBAIJANI FAMILIES

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Introduction and Aims: Familial Mediterranean Fever (FMF) is an autosomal recessive hereditary disease, characterised by recurrent attacks of fever, serositis and arthritis, which primarily affects non-Ashkenasi Jews, Armenians, Arabs and Turks. A small number of FMF cases are also described in other ethnic groups. very few data are available on the presence and genetic spectrum of FMF in Georgian and Azerbaijani patients.
The purpose of our study was to find FMF cases in ethnically Georgian and Azerbaijani patients through genetic testing; to investigate distribution of FMF gene mutation in these ethnic groups.
Methods: 59 patients from 59 non-related families, ethnical Georgians, 7 Azerbaijani patient and 1Kurdish with clinically suspected diagnosis of FMF, mean age 19 year (1-65 year), 30 male, 29 female, underwent molecular genetic studies using polymerase chain reaction. We also registered clinical manifestations, severity of disease, treatment and its efficacy (using standardised questionnaire) and correlated them with mutation.
Results: FMF gene mutations were found in 56 patients; in two cases mutation was not found. The M694V Mutation was predominant; it was present in 33 patients (55, 9%). Other identified mutations were: E148Q, V726A, E167D, M680J, M680I, M694V, V726A, E251K, R76H, 31 patients (52, 5%) were homozygous M694V/M694V, 26 patients were compound heterozygotes for M694V and other mutation. Family history of FMF was positive only in 9 (16.1%) cases.
Most frequent clinical symptom was abdominal pain (55.9%), fever (47, 5%), and Arthralgia (18, 6%). Renal function was deteriorated in 7 (11, 9%) cases, 1 patient was on haemodialysis, renal biopsy (RB) was done in 3 cases, Amiliodosis was found in all RB. Treatment with colchicine was performed in 24 cases (40, 7%).
In Azerbaijani patients predominant gene mutation was M694V.
Conclusions: FMF is present in ethnical Georgians and Azerbaijani. Most frequent mutation is M694V.

Su098 A LARGE DELETION IN PKD2 ASSOCIATED WITH AUTOSOMAL DOMINANT POLYCYSTIC KIDNEY DISEASE AND DEXTROCARDIA

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Introduction and Aims: Genetic inactivation of Pkd2 in mice has been associated with abnormal left-right asymmetry. In human, PKD2 mutations are associated with 15% of autosomal dominant polycystic kidney disease (ADPKD) cases, but no perturbation of left-right asymmetry has been described in patients so far. In situs inversus, the internal organ arrangement is mirror image of normal anatomy, termed situs solitus. Heterotaxia is a group of disorders characterized by misplacement of one or more organs in relation to the left-right axis. Dextrocardia is characterized by displacement of the axis of the heart to the right side of the chest. To date no association between ADPKD2 and abnormal left-right asymmetry has been described.
Methods: DNA was extracted by standart procedure. A point mutation in PKD1and PKD2 gene was screened by high resolution melt analysis on a LightCycler 480 (Roche Applied Systems). The product of PCR with abnormal melting curve were sequenced using classical sanger technique. Deletion of exons were searched by genomic quantitative PCR (qgPCR) in PKD2 and ABCG2 gene. Large deletion was identified by classical FISH analysis on interphasic and metaphasic nuclei.
Results: Two patients from two unrelated families exhibited autosomal dominant polycystic kidney disease associated with left right laterality disorders. The first patient is a 70 years old man presenting a typical ADPKD associated with an isolated dextrocardia. The second patient is a 64 years old female presenting a typical ADPKD associated with a situs inversus. The pace of progression of chronic renal failure in the both families is in favor of a mutation in PKD2 gene. No other member of the families has heterotaxia.
We identified the molecular anomalies responsive of ADPKD. We did not identify no mutation in PKD1 gene in the both patients.Patient 1 has a large deletion (80kb) of PKD2 gene and ABCG2 confirmed by qgPCR and FISH analysis. Patient 2 has a duplication of the third exon of PKD2 responsive of a lost of expression of normal polycystin 2. The two genomic anomalies are clearly pathogen and cosegregate with ADPKD in the families.
Conclusions: We report here the first molecular anomalies in PKD2 associated with left-right asymmetry disorder in humans with ADPKD.

Lost of expression of polycystin-2 in zebrafish and mouse is associated with laterality disorders. We confirmed in humans the critical role of PKD2 in laterality. This is the first description of a clear phenotypic difference between ADPKD1 and ADPKD2. Further more our data pave the way for systematic screening of PKD2 in humans presenting an anomaly in left-right asymmetry.

Su099 ENPP1 GENE POLYMORPHISMS ARE ASSOCIATED WITH LEFT VENTRICULAR (LV) MASS AND SYSTOLIC FUNCTION IN END STAGE RENAL DISEASE (ESRD) PATIENTS

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Introduction and Aims: The plasma cell antigen 1 (PC-1), the product of ENPP1 gene, is a membrane glycoprotein up-regulated by inflammation which inhibits calcification and insulin-sensitivity. These mechanisms are altered in end stage renal disease (ESRD) and potentially involved in LV hypertrophy (LVH) and LV dysfunction.
Methods: Therefore, we investigated the relationship of 10 single nucleotide polimorphisms (SNPs), representative of the haploblock structure of ENPP1 gene, with LV mass and systolic function (by echocardiography) in an ethnically homogeneous series of 238 Caucasian dialysis patients. Three (rs1974201, rs9402349, rs858342) out of ten SNPs were associated with indicators of LV mass and geometry (LVMI, RWT and MWT) and function (mwFS).
Results: Univariate and multivariate analysis, according to a dominant model, showed that the mutated C alleles of rs1974201 and rs9402349 SNPs were inversely related to RWT and MWT (Table). Furthermore, the C allele of rs1974201 SNP resulted also directly linked with mwFS (Table). The mutated G allele of rs858342 SNP was unrelated to RWT and MWT but significantly associated to LVMI on univariate (r=-0.13, P=0.04) and multivariate (β=-0.14, P=0.008) analyses.

		RWT: Relative wall thickness	MWT: Mean wall Thickness	mwFS: Midwall fractional shortening
rs1974201	Crude (r, P)	-0.20(0.002)	-0.22(0.001)	0.19(0.004)
	*Fully adjusted (β, P)	-0.18 (0.004)	-0.17(0.002)	0.14(0.01)
rs9402349	Crude (r, P)	-0.17(0.01)	-0.19(0.003)	0.08(0.24)
	*Fully adjusted (β, P)	-0.13(0.03)	-0.15(0.007)	0.03(0.62)
rs858342	Crude (r, P)	-0.03(0.63)	-0.09(0.18)	0.04(0.55)
	*Fully adjusted (β, P)	-0.05(0.39)	-0.11(0.05)	0.06(0.28)

*Adjusted for Framingham risk factors, anti-hypertensive treatment, Hb, Albumin, Ca*P, cardiovascular comorbidities, dialysis vintage, treatment modality, ADMA, CRP, Homocysteine.
Data are correlation coefficient (r), standardised regression coefficient (β) and P value.

Since the inverse relationships of the C alleles of rs1974201 and rs9402349 SNPs with the muscular component of left ventricle suggest a lower risk of concentric LV hypertrophy/remodelling, we tested this hypothesis in a multivariate logistic regression analysis. In this analysis the odds of concentric LV geometric pattern was about 50% lower in patients carrying the mutated allele in rs1974201 (odds ratio: 0.42, 95% CI: 0.23-0.76, P=0.004) and in rs9402349 (odds ratio: 0.51, 95% CI: 0.27-0.97, P=0.04) than in those without.
Conclusions: Polymorphisms in ENPP1 gene are independently associated with LV remodelling, LVH and LV dysfunction in ESRD. These results offer a genetic basis to the hypothesis that the biological product of this gene (PC-1) is implicated in cardiomyopathy in this population.

Su100 RELATION BETWEEN ABCG2 DYSFUNCTIONAL MUTATIONS AND URATE HANDLING IN PATIENTS WITH ASYMPTOMATIC HYPERURICEMIA OR GOUT

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Introduction and Aims: Hyperuricemia is an inducer for gout and kidney stones and has been recently reported to be associated with metabolic syndrome and accelerate the progression of renal and cardiovascular disease. Uric acid is the end product of human purine metabolism and is mainly excreted from the kidney in urine and in part from the intestine. Thus, serum uric acid levels are determined by the balance of urate amount produced via purine metabolism and urate excretion amount from kidney and intestine. A urate transporter 1 (URAT1) encoded by *SLC22A12*, and glucose transporter 9 (GLUT9), encoded by *SLC2A9*, have been identified as urate reabsorption transporters. These are major transporters to regulate serum uric acid levels because both of the deficiencies result in hypouricemia. However, urate secretion transporters regulating serum uric acid levels have not been identified. ATP-binding cassette, sub-family G, member 2 (ABCG2) is a multispecific transporter expressed on the apical membrane of a variety of tissues including intestine, liver and kidney and mediates efflux of xenobiotics. ABCG2 has polymorphic reduced functionality or nonfunctional variants. In this study, we clarify the role of ABCG2 in uric acid handling.
Methods: Using membrane vesicles prepared from ABCG2-expressing human embryonic kidney 293 (HEK293) cells, transport assays were performed with isotope-labeled [14C]urate.
We have investigated roles of ABCG2 in urate excretion in detail through analysis of urate handling in 193 asymptomatic or gouty outpatients.
Results: We show that ABCG2 is a high-capacity urate secretion transporter. Furthermore, we demonstrate that dysfunctional genotype combinations result in hyperuricemia and markedly increase gout risk.
Conclusions: ABCG2 is a major urate secretion transporter and ABCG2 defects would account for a part of hyperuricemia with with a genetic predisposition.

Su101 SEARCHING FOR GENES INVOLVED IN IDIOPATHIC CALCIUM NEPHROLITHIASIS: CGH ARRAY ANALYSIS IN AFFECTED MEMBERS Of A THREE GENERATION FAMILY

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Introduction and Aims: The importance of hereditary factors in idiopathic calcium nephrolithiasis (ICN) has emerged from several studies of familiar renal stones and from twin studies. Computer programs predicted the best inheritance fit with a model of single gene co-dominant model/polygenic model. In order to identify some of the major susceptibility genes involved in ICN, we analysed by CGH arrays the genome of a patient whose clinical and familiar characteristics are so peculiar to make him a good candidate to be analysed with this type of approach.
Methods: MV, 49 years old, since 19 he has formed calcium oxalate and calcium phosphate renal stones; he expelled about 275 calculi both spontaneously and by lithotripsy (he underwent 33 treatments). The metabolic phenotype revealed intermittent hypercalciuria, and phosphaturic tubulopathy. He was resistant to all pharmacological treatments so far adopted. MV belongs to a family with consanguinity (his parents are second cousins and both affected by nephrolithiasis). In this family nephrolithiasis is transmitted in an apparently dominant fashion, with females more frequently affected

than males but males being more severely affected than females. CGH arrays has been performed in the proband and in eight family members to detect the presence of Copy Number Variants (CNVs) of the genome. CNVs have emerged as a major force, shaping both genetic and phenotypic variation, including the continuum of etiological events underlying monogenic to complex diseases.
Results: A duplication of 308 kb in the Xq22.2 chromosome has been detected in the proband. It was not present in CNV data base, it was inherited from the affected mother and was present also in the maternal affected aunt and cousin. We investigated the effect of this mutation on candidate genes both within and far away the duplicated Xq region. We focused our attention on CLDN2, MORC4, NUP62CL, PRPS1 and NCX1 genes. NCX1 is not on X chromosome, but it might be affected via a position effect mechanism mediated by FXYD8 ion transport regulator gene. For each gene, leucocyte mRNA levels were evaluated by Real Time PCR. The study was conducted on two mutated males, in two not mutated family members and in two normal controls. We found that, although within the pedigree both healthy controls and mutated samples seem to share a similar pattern of gene expression, a striking change in expression occurs for NCX1 (an increase of 396% and 82% in respect to normal leucocytes and to normal familiar samples). NCX1 encodes for the Na^+/Ca^{2+} exchanger which has been shown to play a very important role in calcium omeostasis, finely regulating the calcium reabsorption in the kidney.
Conclusions: Our results evidence for the first time that a disregulation of NCX1 could have a role in the pathogenesis of nephrolithiasis.

Su102 NEPHROLITHIASIS IN AUTOSOMAL DOMINANT POLYCYSTIC KIDNEY DISEASE

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Introduction and Aims: Nephrolithiasis is more prevalent in patients with autosomal dominant polycystic kidney disease (ADPKD) than in general population. The diagnosis of lithiasis in these patients is hindered by the distorted anatomy of the polycystic kidneys and the frequent occurrence of parenchymal and cystic wall calcifications. Morphologic examination of urinary stones that points to specific lithogenic factors has been seldom reported in ADPKD.
Methods: A retrospective study of ADPKD patients in a referral center in Paris, France was performed. Medical files of 208 ADPKD patients referred to our center between 1998 and 2008 were analysed to assess kidney stones frequency by radiological methods and to understand its characteristics using morphological, infra-red spectrophotometry and chemical stone analysis. Cyst wall calcifications detected by radiographic procedures were excluded.
Results: We have found that during this period of time 29 (13.9%) ADPKD patients had experienced nephrolithiasis including those who had a clinical manifestation of colic pain and/or ultrasonography, native roentgen, CT scan detection of kidney stones. Kidney stones could be analysed in 9 patients. The most common type of stone was urico dependent in 6 (66.6%) patients and oxalo dependent in 2 (2.22%) patients. One patient had a combined urico-oxalo dependent lithiasis and in this patient the stone nucleus was uric acid (type IIIb). Superficial morphology of the stones was III b in 6 cases and I b in 4 cases. Morphology of the stone nucleus section was IIIb in 6 cases and IVa in 2 cases. Infrared spectophotometry analysis confirmed uric acid in the nucleus of 7/9 stones. In one case, whewelite (monohydrated calcium oxalate) was the main component of the stone (60%), whereas uric acid represented only 30% but was the only component found in the nucleus. Mean value for urine pH was 6.5±0.8, and mean value for urinary uric acid was 2.28±0.66mmol/L suggesting that increased urinary uric acid was not a major determinant of stone formation.
Conclusions: Our data provide evidence that ADPKD is associated with a high proportion of uric acid stones. The underlying pathophysiology of uric acid stone formation in ADPKD remains to be clarified.

Su103 MUTATIONAL ANALYSIS OF THE NPHS2 GENE IN ADULT PATIENTS WITH FOCAL SEGMENTAL GLOMERULOSCLEROSIS IN CZECH REPUBLIC

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Introduction and Aims: Focal segmental glomerulosclerosis (FSGS) is a frequent cause of nephrotic syndrome (NS) in adults. About 50% of patients are resistant to immunosuppresive therapy and progress to chronic renal failure. NPHS2 mutations are the cause of idiopatic NP in about 20-30% of steroid-resistant children. NPHS2 mutations are described less frequently in adults.The aim of the study was the mutational analysis of the NPHS2 gene in adult patients with FSGS in Czech Republic.
Methods: 17 patients (9 females, 8 males) with steroid-resistant FSGS and 10 patients (5 females, 5 males) with steroid-sensitive FSGS were studied. Renal biopsy with the histological finding of FSGS was performed in the years 2004-2008. The mean age of patients at the time of renal biopsy was 44.9±18.3 years. The mean age of the onset of NS was 39±20.7 years. Control group was formed by 100 healthy individuals. 8exons and intron-exon boundaries were amplified by polymerase chain reaction (PCR) with published primers followed by direct sequencing on ABIPrism 810 sequencer.
Results: No mutation was found in patients with steroid-sensitive NS in spite of one R229Q polymorphism in one patient. Mutations were established in 3 steroid-resistant patients. Heterozygous mutation/polymorphism (P20L-already described mutation and R229Q polymorphism) was found in the patients with onset of NS at 57 years. R229Q polymorphism in connection with other mutation was already described in NS. FSGS and severe hypertensive changes were in renal biopsy. Immunosupressive therapy without effect on proteinuria was finished after 6 months, the advanced chronic renal failure is present nowadays in this patient. P20L and G97S mutations were found in two other patients. G97S is probably a new missense mutation which was not present in 100 healthy individuals of the control group. R229Q polymorphism was in 4 patients with FSGS (4/27-1 steroid-sensitive patient, 3 steroid-resistant patients), in 14.8%. This polymorphism was established in 10 individuals of control group (10%).
Conclusions: NPHS2 mutations are a rare cause of FSGS in adults. A new G97S missense mutation was described. R229Q polymorphism is frequent in patients with FSGS.
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Su104 MORBUS FABRY: A CELL CULTURE MODEL FOR PODOCYTE DAMAGE

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Introduction and Aims: Fabry disease is an X-chromosomal disease leading to a defective alpha-galactosidase A (a-gal A) function. This leads to accumulation of sphingolipids, in particular globotriaosylceramide (Gb3) in various tissues including the glomerular podocyte. Patients with Fabry disease develop proteinuria and, in later stages of the disease, end-stage renal insufficiency. Although renal insufficiency is one of the major causes of a reduced life expectancy in Fabry patients the pathophysiological mechanism leading to renal damage is unclear. Histological observations show that besides massive endothelial damage there is a pronounced accumulation of Gb3 in glomerular podocytes. This effect might play a major role in podocyte damage. The role of Gb3 accumulation in podocytes cannot be investigated in model organisms due to a different glycosphingolipid metabolism in other species, especially in the mouse.
Methods: We established a human podocyte cell culture model for Fabry disease by reducing cellular a-gal A activity by RNA interference (RNAi).

To generate stable cell lines with markedly reduced a-gal A activity we transduced human podocytes with different small hairpin RNA (shRNA) constructs against human a-gal A by lentiviral gene transfer.
Results: Correct function of our shRNA constructs was tested in vitro by a luciferase reporter assay. We confirmed reduced a-gal A expression and function by western blot, qPCR and enzymatic assays. The generated podocyte cell lines did not show an overt phenotype with no visible morphological changes in short-term cell culture. Increased Gb3 accumulation studies, functional studies and elucidation of long-term effects on protein expression of slit diaphragm components are currently underway.
Conclusions: This new cell culture model will be a valuable tool for elucidating the cause of podocyte damage in Fabry disease. It will provide us with new insights into pathophysiological mechanisms of podocyte malfunction in patients with Fabry disease.

Su105 IMPACT OF THE PERSISTENT STATE OF ALBUMINURIA ON THE ASSOCIATION OF THE ACE I/D POLYMORPHISM WITH DIABETIC NEPHROPATHY IN TYPE 1 AND TYPE 2 DIABETES

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Introduction and Aims: Several genetic association studies have defined diabetic nephropathy based on the presence of persistent albuminuria. Less well established, however, is the relevance of non-persistent albuminuria. We therefore investigated whether the association of ACE I/D polymorphism with diabetic nephropathy was impacted by the presence of persistent or non-persistent states of albuminuria.
Methods: We previously reported an association of the ACE I/D polymorphism and diabetic nephropathy in a meta-analysis of 53 studies comprising 17,791 diabetic subjects[1]. These studies were grouped into those that required the renal status of both cases and controls to be confirmed using at least three urine samples (Group A, n = 34 studies) and other studies which did not and thus would contain patients with non-persistent albuminuria (Group B, n = 14). A small handful of studies that specified persistency in cases but not controls were not analyzed further (Group C, n = 5).
Results: There was no obvious publication bias. As previously reported[1], the II genotype was protective against diabetic nephropathy (pooled OR = 0.78, 95%CI=0.70-0.87). In subgroup analyses according to the type of diabetes, the pooled OR for Group A was similar to Group B in type 2 diabetes, suggesting that the ACE I/D was indeed associated with both persistent and non-persistent states of albuminuria. Likewise, ACE I/D was also associated with non-persistent albuminuria in Group B (OR = 0.53, 95%CI=0.38-0.74) among studies with type 1 diabetes. Most unexpectedly, no association was observed in Group A in type 1 diabetic studies (OR = 0.99, 95%CI=0.81-1.21) where the more stringent case definition would presumably have facilitated the detection of an association. In analyses of the clinical characteristics, cases from Group A had significantly longer diabetes duration than cases from Group B ($P<0.001$) while controls were similar between the two groups in the type 1 studies. Cases and controls from both groups A and B had similar clinical characteristics in studies with type 2 diabetic patients.
Conclusions: Our study has highlighted the importance of a more nuanced approach in defining renal phenotypes when conducting genetic association studies on diabetic nephropathy.
Reference:
1. Ng DPK, Tai BC, Lim XL (2008). Is the presence of retinopathy of practical value in defining cases of diabetic nephropathy in genetic association studies? The experience with the angiotensin-I converting enzyme I/D polymorphism based on 53 studies comprising 17,791 subjects. Diabetes 57, 2541-2546

Su106 NEW INSIGHTS IN NEPHROPATHIC AMYLOIDOSIS ASSOCIATED TO A MUTATION IN THE FGA GENE: THROMBOSIS, BLEEDING AND VASCULAR EVENTS

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Introduction and Aims: Fibrinogen amyloidosis (AFib) is considered predominantly a renal disease characterized by variable penetrance, distinctive histological appearance, proteinuria, and progressive renal impairment. Recent reports suggest that patients (pts) with AFib are at increased risk of other organ failure, but the pattern of that involvement has not been well characterized.
Methods: Over the past 6 years (yr) we collected data on the natural history of AFib pts and some of their pre-symptomatic relatives (psr), with the same amyloidogenic mutation in fibrinogen (E526V), and from a restricted geographical area (Braga district in north Portugal). Baseline data on clinical manifestations, causes of death and histological involvement on a cohort of 25 AFib pts (14 female, 11 male) and 22 psr were analysed.
Results: Renal involvement led to diagnosis of AFib in the 25 pts, median age at presentation was 62 yr (34 to 77 yr). Hypertension was universal and diagnosed at a median age of 58 yr (34 to 71yr). Twenty one pts (84%) were not diagnosed with AFib until after they had received renal replacement therapy. Twenty two pts progressed to dialysis after a median time from presentation of 50 months. One homozygous patient received a kidney transplant in 1996, and was without clinical recurrence 14 yr later. Renal amyloid deposits were mainly present in glomeruli, but arterioles were also involved. In other tissues, vascular amyloid was also present, including vessel walls in the abdominal fat, and large and small vessels in the mesentery and colon; large deposits were present in one ruptured spleen. Clinically significant extra-renal manifestations were mainly characterized by vascular and gastrointestinal diseases. Nine pts (36%) had cerebral vasculopathy (2 hemorrhagic, 7 ischemic). Two pts had severe coronary disease and peripheral arteriopathy. Eight pts (32%) had peptic ulcer disease. Plasma fibrinogen levels were normal, except low levels in the homozygous. Seventeen pts died. Stroke was the leading cause of death. One patient died of mesenteric ischemia.
Family history of nephropathy was present in 72% of propositi. Eight of the psr presented hypertension at median age of 43 yr (32-63 yr), 1 had ischemic stroke, 1 underwent coronary bypass and 2 had intermittent claudication.
Conclusions: Our findings are consistent with a systemic nature of AFib amyloidosis but the penetrance of the disease in Portugal appears to be higher. Cerebrovascular disease was an important cause of morbidity and mortality. Variation in the fibrinogen gene FGA, but not fibrinogen level, appears to be associated with cardiovascular events. Considering the number of psr further collaborative studies are needed.

Su107 GENETIC BASIS OF CYSTINURIA

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Introduction and Aims: Cystinuria is a hereditary renal disease characterized by a large concentration of cystine in the urine, and it leads to the formation of kidney stones. The symptoms of cystinuria include hematuria, flank pain, renal colic, obstructive uropathy and urinary tract infection. Cystinuria is caused by a mutation in the SLC3A1 and SLC7A9 genes and has an autosomal recessive pattern of inheritance. Until now, about 40 mutations in the SLC3A1 and 30 in the SLC7A9 have been described in the literature. The mutation detection rate in cystinuric patients is about 70–80%. The aim of this study was to classify phenotypically cystinuria patients and to identify the novel variants of the cystinuria genes.
Methods: In this study, we performed direct sequencing of entire coding regions of the SLC3A1 and SLC7A9 genes in 34 unrelated individuals who had been referred to our laboratory with a clinical diagnosis of cystinuria. These patients were of different ethnic groups.

Results: Mutations were found in 20 of the 34 patients studied, and 6 of the mutations were novel. Twelve mutations were found in the SLC3A1 gene, and 8 in the SLC7A9 gene. Overall, the mutation detection rate was about 88%. Four novel mutations (C285Y, T624A, R301Q and IVS6+3delT) were identified in the SLC3A1 gene, and 2 novel mutations (422fs486X and R326G) were identified in the SLC7A9 gene. One mixed cystinuria family transmitted mutations in both genes. There were homozygous and heterozygous mutations.
Conclusions: In conclusion, the mutations found in these patients almost prove the clinical diagnosis of cystinuria and help to classify the diagnosis into the categories of heterozygous and homozygous cystinuria.

Su108 INFLUENCE OF LYMPHOCYTE POLARIZATION ON ATHEROSCLEROTIC EVENTS AFTER RENAL TRANSPLANTATION

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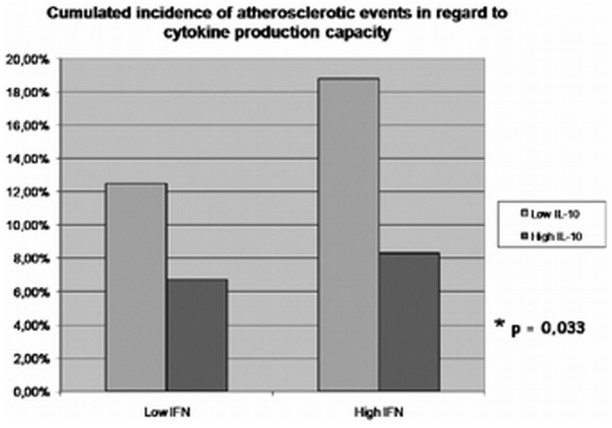
Introduction and Aims: Atherosclerosis is an inflammatory disease involving all actors of immunity. Nevertheless, the relevance of these immune mechanisms in immunodepressive patients remains to be determined. The aim of this work was to analyze relationships between T lymphocyte polarization toward a pro- or anti-inflammatory profile and atherosclerotic events in renal transplanted patients (RT).
Methods: The population studied was consecutively recruited. Two hundred and thirty nine RT were included. The inclusion date corresponds to the renal transplantation date. We studied the IL-10 (([IL-10-592 C/A; IL-10-819 C/T; IL-10-1082 G/A]) and IFN-γ ([IFN-γ (CA) repeat]) gene promoter polymorphisms. The relationship between the polymorphisms and the cytokine production capacity in response to PHA was studied. Moreover we studied for thirty nine patients the T lymphocytes secretion capacity of both cytokines in response to viral antigens from CMV (peptide pp65 pool) and EBV (peptide EBNA 3A pool) according to the existence of a cardiovascular history.
Results: Thirty eight patients (16%) had an atherosclerotic complication during the following period (9,7±4,2 years). The 2/2 [IFN-γ (CA) repeat] genotype had a production capacity of IFN-γ significantly higher after polyclonal stimulation ($p<0.05$). For IL-10 polymorphisms, we observed a trend toward a higher IL-10 production associated with GCC/GCC haplotype. Patients were divided in high and low producers of IFN-γ and IL-10, determining four groups. The cumulated incidence of atherosclerotic events was lower in high IL-10 producers than in low producers (7,4% vs 18,5%, p=0.033) and was independent from the IFN-γ production capacity. In low IL-10 producers, the cumulated incidence of atherosclerotic events was higher in high IFN-γ producers (24,5% vs 14,5%, p=0.033).



Endlessly, the IL-10 production capacity in response to viral antigens was higher in atherosclerotic patients.
Conclusions: These results suggest that a type 1 pro- inflammatory response is continuous in atherosclerotic process of the RT and that the evolution of the atherosclerotic lesions depends on the capacity to generate an anti-inflammatory response.

Su109 RENALASE GENE POLYMORPHISMS AND HYPERTENSION IN END STAGE RENAL DISEASE PATIENTS

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Introduction and Aims: Renalase is a novel flavin adenine dinucleotide-dependent amine oxidase that is secreted by the kidney, circulates in the blood and regulates cardiac function and systemic blood pressure. Renalase plasma levels are substantially reduced in patients with chronic kidney disease. Insufficiency of renalase may explain a high occurrence of hypertension among patients with ESRD and an increased risk for cardiovascular events in this group.
The aim of the study was to assess the relationship of two renalase gene polymorphisms with hypertension in dialysed patients.
Methods: Groups of 143-278 dialysed patients were examined. The patients were divided into 2 subgroups: with hypertension (ESRD HY+) and without hypertension (ESRD HY-). All subjects were genotyped for two renalase gene polymorphisms (rs 2576178 and rs 10887800) by polymerase chain reaction (PCR) and subsequent cleavage with restriction endonucleases (Msp I and Pst I, respectively).
Results: The genotype distribution and allele frequencies of rs 2576178 renalase polymorphism were compared in the following subgroups of patients: ESRD HY+(n=200) and ESRD HY- (n=169). The genotype distribution were similar between those two groups. There was a significant difference in the frequency of the G allele carriers (0.28 in HY+ and 0.22 in HY-), p=0,039. G allele carriers were associated with 1.5 times higher risk of hypertension (OR=1.55; 95%CI: 1.023-2.357).
Genotype distribution and allele frequencies of rs 10887800 renalase polymorphism were compared in the following subgroups of patients: ESRD HY+(n=278) and ESRD HY- (n=143). The genotype distribution were similar between those two groups.
G allele carriers occurred with significantly higher frequency in HY+ patients (0.46 vs 0,37 in HY- subgroup) (OR=1.76; 95%CI: 1.159-2.667).
Conclusions: Our results suggest for the first time an association between analysed renalase gene polymorphisms and hypertension in dialysed patients. It may be an important step in better understanding of cardiovascular pathophysiology and perhaps also in providing optimal treatment and better prognosis for patients with chronic kidney disease.

Su110 SAA1 GENE POLYMORPHISMS IN BELARUSSIAN PATIENTS WITH AMYLOIDOSIS SECONDARY TO RHEUMATOID ARTHRITIS

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Introduction and Aims: Secondary (AA) amyloidosis in rheumatoid arthritis (RA) is considered to be extensively determinated by genotype characteristics. Serum amyloid A protein (SAA) is synthesized by liver under inflammatory conditions. In spite of several polymorphism described in the SAA1 gene, particular investigator's attention is attracted to two single nucleotide polymorphisms localized in the third exon of this gene: – 2995C/T and 3010C/T. The combination of these polymorphic variants determines three SAA1 haplotypes – α, β and γ. The influence of the γ/γ genotype as well as γ haplotype itself on the development of secondary amyloidosis in patients with RA in Asian population (including Japanese) is described in number of studies. *Aims.* In the present study we compared the influence of the SAA1 gene allele polymorphism in AA-positive RA patients with those in AA-negative RA. All patients are Belarusian citizens.
Methods: Native DNA was extracted from leucocytes of blood samples obtained from 21 AA-positive RA patients (1st group) and 27 AA-negative

RA patients (2nd group). To amplify a segment of the SAA1 gene including the polymorphic sites -13T/C, 2995C/T and 3010C/T it was genotyped by polymerase chain reaction (PCR) with subsequent restriction enzyme digest analysis. Statistical analyses of genotype and allele frequency comparisons of the various single nucleotide polymorphisms between groups were performed using the chi-square test.

Results: Genetic polymorphism of the SAA1 gene in Belarusian AA-positive RA patients (1st group) and AA-negative RA patients (2nd group) was determined. An odds-ratio (OR) calculated for the α/α genotype was 23.6, and the 95% confidence interval was – 95%CI (1.7-38.8). It was shown that the SAA1 α/α genotype dominated in both groups and consisted 95.2% (1st group) and 45.8% (2nd group), respectively. The SAA1 α/β and α/γ genotypes were revealed only in the 2nd group; one patient of the same group had β/β genotype. Therefore, according to obtained data SAA1 α/α (allele variants 2995T and 3010C) are the genetic risk factor for the development of secondary amyloidosis in Belarusian patients with rheumatoid arthritis. The -13T allele frequency of the SAA1 gene (polymorphism -13T/C) was 13% in the 2nd group; among AA-positive RA patients (1st group) this allele was absent. In both groups there were no -13T/T homozygotes. Consequently, the -13T allele has no influence on the manifestation of AA-amyloidosis in Belarusian patients with RA.

Conclusions: In contrast to Japanese data, our results revealed that in Belarusian citizens (Caucasians) *SAA1* α/α isotype was the most amyloidogenic. Relative risk of secondary amyloidosis in RA patients significantly increases in α/α genotype. Presence of the -13T allele in *SAA1* gene allele had no influence on the risk of AA-amyloidosis development in investigated Belarusian patients with RA.

Su111 AN ANALYSIS OF 60 CHINESE PATIENTS WITH GITELMAN'S SYNDROME

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Introduction and Aims: Gitelman's syndrome (GS) is an autosomal recessive renal disorder characterized by hypokalemia, hypomagnesemia, metabolic alkalosis and hypocalciuria. GS is caused by inactivating mutations in SLC12A3 gene, which encodes the thiazide-sensitive sodium chloride cotransporter (NCCT). We analyzed 60 patients with GS. This is a systematic evaluation of the largest group of Chinese GS patients.

Methods: 60 Chinese GS patients from 41 unrelated families were included in our study. We analyzed main symptoms and laboratory data: plasma and urinary electrolyte, plasma renin activity and the concentration of aldosterone, glomerular filtration rate (GFR), 24-hour urinary protein etc.

Results: The study group consisted of 60 patients (41 males and 19 females,average age 31.5±13.3 yrs). The average age of their fist visit were 24.6±12.4 yrs. eight patients had no symptoms. The most common symptoms were fatigue (43/60), muscle weakness (42/60), nocturia (27/60), salt craving (20/60), polydipsia (19/60), muscle cramps (20/60) and palpitation (22/60) by investigation. However, early symptoms (occurred during 6-10 yrs) were nocturia and polydipsia. 13 patients had a history of enuresis at school age. Biochemical data of patients were shown in the table. All the patients had hypokalemia, alkaline urine, and hypomagnesemia. 59 patients had hypocalciuria, 9 patients had proteinuria (>150mg/24h), and two of them had renal insufficiency with GFR 64ml/min and 37 ml/min respectively.

Conclusions: The phenotype of GS is highly heterogeneous in terms of age, nature/severity of the biochemical abnormalities and clinical manifestations. Long-time tubulointerstitium injury probably due to hypokalemia or other unknown mechanism, sometimes leads to a decrease in GFR, even causes an evolution to end-stage renal disease.

Su112 A NOVEL c: 3622_3623insGTGT MUTATION OF *PKD1* GENE IDENTIFIED IN A CHINESE FAMILY CAUSING AUTOSOMAL DOMINANT POLYCYSTIC KIDNEY DISEASE

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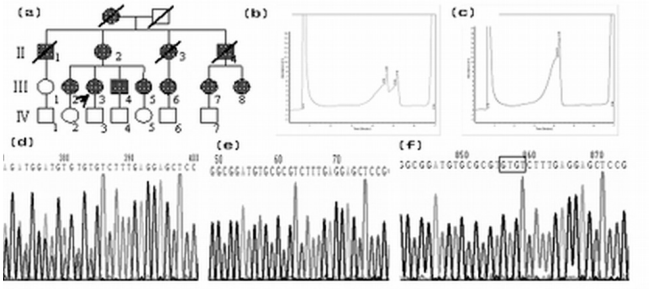
Introduction and Aims: Autosomal dominant polycystic kidney disease (ADPKD) is a common genetic renal disease characterized by numerous and gradually enlarged fluid-filled epithelial cysts in bilateral kidneys. The disease has heterogeneity, and the gene *PKD1* (16p13.3)and *PKD2* (4q21.2) are identified accounting for 85% and 14% of the cases, respectively. In the present study, we found a new c: 3622_3623insGTGT mutation of *PKD1* gene in a proband. Further, we researched to evaluate the mutation is the pathogenesis of the family with ADPKD.

Methods: The clinical diagnosis of the available members in this family was according to renal ultrasound criteria by York Pei *etc. Genomic DNA was extraced from peripheral blood cells. All 46 exons of the *PKD1* gene were analyzed by PCR, following with DHPLC and DNA sequencing. The 15 exons of *PKD2* gene were proceeded with PCR and direct sequencing. 100 genomic DNA extracted from 100 unrelated normal persons were used as control.

Results: The pedigree of this family was shown as fig.1a.



[Fig 1](a) Pedigree based on imaging diagnosis. According to the renal ultrasound criteria by York Pei *etc, NO II (1~4) and III(2~8) could be defined as ADPKD. (b, c) Mutant heteroduplexes are visualized as an earlier elution peak or shoulder on the homoduplex peak. (b) Aberrant elution peak of DHPLC obtained from analysis of exon fragment 15b in the proband; (c)Normal elution peak of DHPLC found in control. (d) DNA sequencing of the fragment 15b of the proband; (e) Fragment 15b in the wild-type gene sequence; (f) Insertion of GTGT in exon 15 of *PKD1* confirmed by monodonal sequencing.

* York P, Obaji J, Dupuis A, et al. Unified Criteria for Ultrasonographic Diagnosis of ADPKD. *J Am Soc Nephrol* 2009, 20:205-212

The proband, female, 50 yr, was presented with blepharedema, lumbodorsal megalgia and hypertension and was diagnosed with polycystic kidneys and polycystic liver in age 26. Her serum creatinine was 134 μmol/L and none abnormalities were reported by the test of urinary sediments. II_2, III_2, III_4, were with nephrolithiasis, and III_4 as well as the proband was with polycystic liver too. Clinical examination revealed that the families all had normal serum creatinine, uric acid and blood urea nitrogen except the proband. II_1 and II_3, died of cerebrovascular disease at age 68 and 71, were described with polycystic kidneys and nephrolithiasis; II_4 was died of ESRD at 72 years old. The blood sample of II_2 (75 yr) was unavailable and the results of her health examination was reported one year ago. Analysis of DHPLC was performed to extronic fragments of *PKD1* and an abnormal elution-peak was tested in G15b of exon 15 of the samples of $III_{2\sim8}$, IV_2 (19 yr) and IV_5 (15 yr) (fig. 1b). Then those extronic fragments of *PKD1* and *PKD2* were analysized by DNA sequencing and a 4 bp insertion mutation was found in the same fragment in *PKD1* from those above sufferers in accordance with the analysis of DHPLC (fig. 1d).The others in the family without renal disease as well as normal controls have no this mutation (fig. 1c & e). Finally, the *c: 3622_3623insGTGT mutation of PKD1 gene* was confirmed by monoclonal sequencing (fig. 1f.

Conclusions: we found a novel insertion c:3622_3623insGTGT unreported in the PKDB and Human Gene Mutation Database (HGMD), and combined with the family history, we suggested the insertion is a novel pathogenic mutation leading to ADPKD.

Su113 DO THE HEREDITARY FACTORS OF THROMBOPHILIA PARTICIPATE IN PATHOGENESIS OF DIARRHEA-POSITIVE HEMOLYTIC-UREMIC SYNDROME IN CHILDREN?

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Introduction and Aims: Despite a great progress achieved in understanding of hemolytic-uremic syndrome (HUS) pathogenesis in the recent time, the factors contributed to the onset of HUS in 5% children with diarrhea are not fully discovered. Hereditary thrombophilia could play a role in renal microvascular thrombosis in HUS patients.
Aim: to investigate coagulation blood system genes polymorphisms in children with diarrhea-positive hemolytic-uremic syndrome (HUS D+).
Methods: 48 children with HUS D+ aged from 6 months to 16 years (18 girls and 30 boys) were examined. We investigated coagulating blood system genes polymorphisms: genes coding methylentetrahydrofolatreductasa (MTHFR C677T), Leidens' factor V (F5), prothrombin (PGT G20210A), plasminogen activator inhibitor 1 (-675 4G/5G PAI-1), fibrinogen (-455 G/A FGB), platelets' fibrinogen receptor (L33P ITGB3), using molecular genetics methods (PCR-mass spectrometry, PCR-restriction analysis).
Results: Polymorphism of examined genes was found in all HUS D+ children. The only gene polymorphism was found in 12 patients (25%), 8 of them had polymorphism of gene PAI-1. In most of the patients (36; 75%) multigene form of thrombophilia was discovered: 2 genes polymorphisms were found in 16 children (33,3%), 3 genes – in 13 cases (27%), 4 and more genes polymorphisms – in 7 children (14,6%). Mutation of genes PAI-1 (in 87% of patients), MTHFR (62,5%), FGB (58,3%) were more frequently. 17 (35,4%) patients were homozygous carriers of 4G allele PAI-1, 10 (20,8%) – of T allele MTHFR, and 4 (8,3%) – of A allele FGB. Heterozygous carrier of 4G PAI-1, T MTHFR, and A FGB alleles were 26 (54,1%), 19 (39,5%) and 24 (50%) patients, respectively. Leiden mutation (F5) was found in 1 patient, heterozygous form of PGT gene polymorphism – in 3 cases. Multigene form of thrombophilia was characterized by combination of MTHFR and PAI-1 genes polymorphisms in 9 patients (18,8%), combination of 3 mutant alleles (MTHFR, PAI-1, FGB) in 8 patients (16,6%). In 6 cases (12,5%) combination of 4 mutant genes (MTHFR, PAI-1, FGB, ITGB3) was found.
Conclusions: High frequency of coagulating blood system genes polymorphisms in HUS D+ patients suggested of hereditary thrombophilia role as a risk factor of renal thrombotic microangiopathy in children with diarrhea.

Su114 A TRANSPLANT IN VON GIERKE'S DISEASE

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Introduction and Aims: Type 1a glycogen storage disease (GSD 1a), or von Gierke's disease, is a rare autosomal recessive disease, and it is caused by a deficiency of glucose-6-phosphatase, which leads to glycogen accumulation in the liver, kidney and intestinal mucosa. Clinical manifestations include: hypoglycaemia, growth retardation, hepatomegaly, lactic acidaemia, hyperlipidaemia and hyperuricaemia; long-term complications include renal disease, gout, osteoporosis, pulmonary hypertension, short stature, hepatocellular adenomas which may undergo malignant transformation. We describe the management and the clinical course of a GSD1a patient who underwent simultaneous liver-kidney transplantation (SLKT).
Methods: A woman was diagnosed GSD 1a when she was 2 years old. Molecular analysis of the glucose-6- phosphatase gene revealed R83C/Q347X mutations.
The clinical course was complicated by hepatomegaly, short stature, lactic acidaemia, hypoglycaemia, osteoporosis and elevated level of uric acid, cholesterol and triglycerides. Frequent meals prevented hypoglycaemia but metabolic control remained poor. When the patient was 15 and 22 years old resection of 2 hepatic adenomas was done and at 23 the patient developed hypertension and renal failure.
At 30 a kidney biopsy was performed and focal glomerulosclerosis was found; before biopsy a intravenous desmopressin and two units of packed red cells reduced the bleeding time, typically elevated in GSD1a. The patient was pre-uremic (creatinine clearance 13 ml/min). She had 2 hepatic adenomas (a magnetic resonance imaging abdomen was performed). Eight months later she became uremic but refused to start dialysis. After the next six months the patient underwent SLKS from a 11 year old donor who had died from a head injury. The immunosuppressive regimen included steroids, tacrolimus prolonged release and mycophenolate mofetil. No acute rejection episode was observed. Laboratory results normalized on postoperative day 10. One month later transplant was complicated by cytomegolovirus infection which was treated with valganciclovir. Seven months post transplant the patient was in good condition with both grafts functional: creatinine 1.1 mg/dl; cholesterol 106 mg/dl; triglycerides 145 mg/dl; SGOT 27 UI/L; SGPT 25 UI/L; HCO_3 24 mMol/L.
Results: In our patient affected by GSD1a, SLKS has solved liver and renal disease.
Conclusions: There aren't clear indications when to perform SLKS in GSD1a. In this patient we decided to perform SLKT because of the risk of hepatocellular adenomas' malignant transformation secondary to immunosuppression and to offer the best quality of life. In our opinion uremic patients with GSD 1a have to be considered for SLKT. To our knowledge this is the fifth case of SLKS in GSD1a described in literature.

Hypertension

Su115 TOLL-LIKE RECEPTOR 4 DISTRIBUTION AND FUNCTION IN HYPERTENSIVE CARDIOMYOPATHY

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Introduction and Aims: Toll-like Receptors (TLRs) are important contributors of the innate immune system. TLR4 is involved in cardiovascular disease. Low-grade inflammation is a key characteristic of hypertensive cardiomyopathy. The study objective was to investigate whether TLR4 contributes to hypertension and hypertensive cardiomyopathy in different hypertensive rat models.
Methods: Cardiac TLR4 was determined by Western Blot in young, adolescent and adult spontaneously hypertensive rats (SHR) and normotensive Wistar Kyoto rats (WKY; 4, 8 and 16 weeks of age). Secondly, TLR4 was assessed in another hypertensive model using the NO-synthase inhibitor Nω-nitro-L-arginine- methyl ester hydrochloride (L-NAME) in WKY. Cardiac tissue of SHR and WKY was stimulated for 4 hours with the endogenous TLR4 ligand heparansulfate. Thereafter, the proinflammatory mRNA pattern was assessed (TNFα, Il-6 and MCP-1). Finally, the effect of ACE and renin-inhibition on TLR4 density was investigated in the L-NAME model.
Results: TLR4 was present in all animals, but adult SHR showed more TLR4 than WKY. Similarly, L-NAME induced hypertension and cardiac hypertrophy showed enhanced TLR4 expression. This was associated with a pronounced TLR4 staining of hypertrophied cardiomyocytes. In SHR, stimulated cardiac tissue resulted in aggravated TNF-α and Il-6 mRNA response as compared to WKY. Finally, effective antihypertensive treatment did not reduce TLR4 density in L-NAME treated WKY.
Conclusions: Hypertensive cardiomyopathy is associated with enhanced cardiac TLR4 density. In SHR, the endogenous TLR4 ligand heparansulfate exaggerated the inflammatory response of cardiac tissue. Thus, TLR4 may participate in hypertensive cardiomyopathy. As TLR4 density remains unaltered by ACE-inhibition, this effect may be independent of the renin-angiotensin system.

Su116 URINARY ALBUMIN EXCRETION RATE AMONG NON-DIPPER HYPERTENSIVE PATIENTS IS CLOSELY RELATED WITH THE PATTERN OF NON-DIPPING

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Introduction and Aims: Blunted nocturnal hypotension (non-dipping), as detected by ambulatory blood pressure monitoring, in patients with hypertension has been associated with increased urinary albumin excretion rate (UAER). However, the relationship between 24-hour UAER and the pattern of non-dipping (isolated systolic non-dipping, isolated diastolic non-dipping and both systolic and diastolic non-dipping) is not fully known.
Methods: Medical history, physical examination, laboratory analysis, and office and ambulatory blood pressure measurements were performed. 24-hour urine specimens were collected to determine creatinine clearance as well as UAER.
Results: Totally, 158 non-diabetic essential hypertensive patients (male/female; 54/104, mean age; 46.9±10.7) were included; 104 patients were dippers, 54 patients were non-dippers. Among non-dippers, 14 patients were isolated systolic non-dippers, 7 patients were isolated diastolic non-dippers, and 33 patients were both systolic and diastolic non-dippers. Among non-dipper patients, 17 had microalbuminuria and among dipper patients 9 had microalbuminuria (P< 0.0001). The median UAER of dippers were lower when compared to non-dippers (5.25 mg/day vs.23 mg/day, P <0.0001). The median UAER of isolated systolic non-dippers, isolated diastolic non-dippers and both systolic and diastolic non-dippers were 8.45 mg/day, 7.7 mg/day and 25.5 mg/day, respectively (P: 0.001). Subgroup comparison of patients revealed that UAER was higher in both systolic and diastolic non-dippers when compared to dippers (P <0.0001), when compared to isolated systolic non-dippers (P: 0.001) and when compared to isolated diastolic non-dippers (p: 0.017).
Conclusions: In conclusion, not only non-dipping itself, but the pattern of non-dipping may be related with UAER in non-diabetic essential hypertensive patients.

Su117 DIAGNOSTIC VALIDITY OF 64 OSCILLOMETRIC BLOOD PRESSURE MEASURING DEVICES DESIGNED FOR HOME USE

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Introduction and Aims: Lacking accuracy of blood pressure (bp) measuring devices for home use has repeatedly been stated. Between 2001 and 2008 we tested 64 devices (41 designed for the upper arm, ua, 23 for the wrist. wr) according to the 'test seal' of the German Hypertension League DHL. For details see:http://www.hochdruckliga.de/prufsige.htm. We calculated the sensitivity (ss), specificity (sc), and the Youden index (ss + sc -1) for the detection of high blood pressure (syst. >135 and/or diast. >85 mm Hg) by these devices.
Methods: Briefly, all devices are tested in 96 persons equally distributed for sex, age and bp categories. Wrist devices are additionally tested in another 20 persons with diabetes above 56 years of age. Three trained testers take 12 sequential bp measurements of each test person, one tester by reading the test device two testers by control measurement (method: Riva Rocci and Korotkoff with mercury manometer) i.e. by simultaneously auscultating the sounds and separately documenting the results. For statistical calculation more than 39000 bp measurements taken from 1589 test persons were analysed by descriptive methods and multivariate regression analysis.
In comparison to standard bp measurement most devices show significant deviations with increasing and relevant underestimation of high systolic and diastolic bp values as well as some overestimation of low bp values (results submitted to ESH congress 2010).
Results: True positive ratio (sensitivity), true negative ratio (specificity), and Youden-Index for the detection of elevated blood pressure values by different types of home blood pressure measuring devices see table.
Conclusions: Confining the information from home devices for blood pressure measurement to whether blood pressure is elevated or not on average one in four measurements may give false positive and one in ten measurements false negative results. The Youden-Index summarizes this diagnostic validity of a device in a single figure. Differences between upper arm and wrist instruments are small. Not only for physiological but also for technical reasons home blood pressure measurement cannot substitute for office measurement or ABPM.
Disclosure: Financial support has been accepted from different producers of blood pressure measuring devices.

Table of results

Devices (n=64)	Sensitivity		Specificity		Youden-Index	
	range	median	range	median	range	median
Upper arm	0.52–0.85	0.72	0.82–0.98	0.93	0.50–0.72	0.64
Wrist	0.60–0.94	0.76	0.72–0.98	0.89	0.51–0.75	0.63

Su118 ADDITIVE ANTIHYPERTENSIVE EFFECTS OF AT2 STIMULATION AND AT1 INHIBITION IN ANAESTHETISED RATS – MEDIATORY ROLE OF RENAL MEDULLARY VASODILATATION?

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Introduction and Aims: Angiotensin II vasoconstricts via AT1 and vasodilates via AT2 receptor activation. Selective ligands of both receptor types are needed to study their relative importance or possible interaction in the control of normal or elevated arterial blood pressure (BP); the knowledge of the role of AT2 is still limited.
Methods: In this study effects of a newly synthetized (A. Lipkowski) tripeptide AT2 agonist (LKP) on BP and intrarenal haemodynamics (left renal artery Transonic probe, RBF; cortical-, outer- and inner medullary laser-Doppler fluxes, CBF, OMBF, IMBF) were determined in anaesthetized Wistar rats, receiving background intravenous infusion of norepinephrine (NE) which raised BP to 130±2 mmHg.
Results: LKP infusion (120 μg/kg/h i.v.) decreased BP (-4%, p<0.01), which was associated with a 9% increase in OMBF (p<0.05). Although NE-induced hypertension was expected to inhibit endogenous angiotensin II synthesis, additional AT1 inhibition (losartan, 1 mg/kg i.v.) further decreased BP and increased OMBF, and tended to increase RBF. The renal excretion tended to decrease in these experiments.
Conclusions: The findings indicate that the levels of AT1 and AT2 activity can independently influence arterial pressure in acutely hypertensive rats. A striking association of BP decrease with an increase in OMBF suggests that perfusion of the renal medulla has a direct causative role in the control of arterial pressure, unrelated to changes in body fluid volume.

Su119 INHIBITION OF NEURONAL NOS PREVENTS THE PARADOXICAL INCREASE IN BLOOD PRESSURE WHICH FOLLOWS ACUTE RENAL DENERVATION IN WISTAR RATS

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Introduction and Aims: Interaction of the effects of renal sympathetic nerve activity (RSNA) and different sodium intake on arterial blood pressure (BP) and renal haemodynamics and the role of nNOS derived NO in the mechanism of this interaction were examined in Wistar rats fed standard (STD), low-sodium (LS, 0.15% Na) or high-sodium (HS, 4% Na) diet.
Methods: These animals were anaesthetized and underwent, at 0.5-h intervals, right- and left renal denervation using an acute non-invasive technique. Throughout experiments BP and left kidney perfusion of the cortex, outer- and inner medulla (CBF, OMBF, IMBF; laser-Doppler fluxes) were recorded.

Results: Surprisingly, in untreated rats sequential bilateral renal denervation progressively and significantly increased BP by 6%, 12% and 19% in rats on STD, LS and HS diets, respectively. There was a 9% and 18% post-denervation increase in CBF in STD and LS rats, respectively, contrasting with no change in HS rats. The changes in medullary perfusion (OMBF, IMBF) were minor or highly variable. Inhibition of nNOS (pretreatment with N-propyl-L-arginine (L-NPA), 1 mg/kg/h i.v.) significantly attenuated the post-denervation increase in BP in LS rats whereas in STD and HS rats BP decreased significantly.
Conclusions: Thus, the paradoxical post-denervation increase in BP was most pronounced in HS rats and was abolished by elimination of NO derived from nNOS. We speculate that abundance of NO (overexpressed nNOS, especially under HS), buffers vasoconstrictor RSNA influence. After denervation the NO not utilized for this buffering may undergo transition to constrictor radicals (e.g. peroxynitrites), hence an increase in BP. Blockade of nNOS prevents this increase.

Su120 PREECLAMPSIA: MARKER FOR FUTURE RISK OF END STAGE RENAL DISEASE (ESRD) AND CARDIOVASCULAR DISEASE

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Introduction and Aims: Preeclampsia is a disorder of gestation characterized by hypertension and proteinuria. It complicates 5-8% of pregnancies and represents 15-20% of maternal death and one of the most important causes of preterm birth and perinatal death. Preeclampsia is also a marker for future risk of End Stage Renal Disease (ESRD) and for morbidities and death due to cardiovascular disease. This is, at least in part, related to hypertension that causes importants metabolic and vascular alterations.
The aim of this study is to evaluate blood pressure and proteinuria since delivery, aiming to identify patients with, both short and long term, high risk to develop renal failure and cardiovascular diseases.
Methods: This retrospective analysis was conducted over 77 pregnancies (without hypertensive disorders before pregnancy) complicated by preeclampsia. The women (mean maternal age 34.3 years ±4.8) was selected between January the 1st 2007 and April the 30st 2009.
Results: Grouping the population according to the delivery date, it results that 45.4% (35 women) delivered in 2007, 40.3% (31 women) in 2008 and 14.3% (11 women) in 2009. In November 2009, we contacted all patients, (respectively 2 years, 1 year and 6 months from the delivery date), measured blood pressure and recorded the antihypertensive therapy history. All patients were subjected to urinalyses to analyze the persistence of proteinuria since the date of delivery.
We found that a significant percentage of patients that didn't follow any therapy, had borderline or high blood pressure (PA≥ 130/80 mmHg): 22.9%, 29% and 18.2% of the patients of 2007, 2008 and 2009 respectively. We measured the percentage of patients that in the follow-up kept values of proteinuria ≤ 30 mg/dl and blood pressure ≥ 130/80 mmHg: 2.9%, 16.1% and 9% respectively 2 years, 1 year and 6-9 months from the delivery time. We found a direct correlation, statistically significative, between the values of the proteinuria during pregnancy and in the follow-up ($p<0.05$).
Conclusions: Results from our study that women keep blood pressure levels border-line or high and proteinuria are already a significant percentage one or two year after delivery.
This results show that is very important making women aware that they should have more frequent health check-ups, should be advised that life-style and dietary changes may delay progression of the renal and cardiovascular disease.

Su121 THE AAA-REGISTRY: REAL LIFE ANALYSIS OF BLOOD PRESSURE CONTROL IN 15.000 HYPERTENSIVE PATIENTS IN GERMANY CLASSIFIED BY COMORBIDITIES

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Introduction and Aims: Epidemiological data of the relationship between blood pressure control and chronic renal failure are scare. In an observational prospective study patients were eligible for documentation in whom the physician had decided to modify the antihypertensive therapy. This included treatment with the direct renin inhibitor aliskiren or an ACE inhibitor or an angiotensin receptor blocker or an agent not blocking the renin-angiotensin system, alone or on top of an existing drug regimen.
Methods: Overall, nearly 15.000 hypertensive patients were recruited by participating physicians (General Practicioners N= 361, Internists N=219, Cardiologists N= 151, Nephrologists N= 18, Diabetologists N=86). At baseline casual, systolic and diastolic blood pressure was on average 155/90 mmHg, and in the subset of patients with 24-hour ambulatory blood pressure average ambulatory BP was 145/85 mmHg (N=5823). Data was analyzed at baseline and at 1 year (data not yet available). Serum creatinine and thereby estimated glomerular filtration rate (Cockcroft Gault) was available in every patient, but in only 1.264 (8.4%) albuminuria had been measured.
Results: The prevalence of chronic renal failure < 60 ml/min/1.73 m^2 was N= 4566 (30.9%), with the following distribution: Stage IIIa (GFR 59-45) 60.1%, stage IIIb (GFR 44-30) 28.0%; stage II (GFR 30-15) was 5.5% and stage V (GFR < 15) 5.4%, respectively.
The prevalence of chronic renal failure (stage ≥ 3) was increased in diabetic patients and those with existing cardiovascular disease (see table). Treatment blood pressure goals below 140/90 mmHg has only been found in 16.1% at the beginning of the observational period with chronic renal failure and the recommended strict blood pressure goal < 130/80 mmHg was only found in 6.9% patients with chronic renal failure.
Conclusions: Thus, we conclude, the prevalence of eGFR < 60 among hypertensive patients treated in Germany is high and their blood pressure poorly controlled. Further analysis of baseline data, in conjunction with 24-hour ambulatory blood pressure values will be analysed and available at the time of the meeting.

Prevalence of chronic renal failure stage 3 and higher related to comorbidities

	eGFR < 60 ml/min/1.73^2	eGFR > 60 ml/min/1.73m^2
Total	30.9% (4566)	69.1% (10234)
Diabetes mellitus	30.9% (1410/4564)	26.1% (2673/10229)
Cardiovascular disease	46.3% (2113/4564)	24.1% (2465/10229)
– Coronary or heart	29.8% (1360/4564)	16.6% (1700/10222)
– Peripheral artery disease	9.6% (434/4501)	4.5% (448/10051)
– Stroke	7.1% (319/4502)	3.2% (317/10056)

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Su122 OSTEOPROTEGERIN LEVELS ARE INCREASED IN HYPERTENSIVE PATIENTS

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Introduction and Aims: Hypertension is the second major cause of end stage renal disease, after diabetes, being responsible for 25–30% of all reported cases. Osteoprotegerin (OPG), also known as osteoclastogenesis inhibitory factor, is a cytokine, member of the tumor necrosis factor

(TNF) receptor superfamily which can inhibit osteoclastic activity, and it is produced at the cardiovascular system. Its levels are higher in both aorta and renal arteries. Clinical studies have shown that serum OPG concentrations are increased in patients with cardiovascular disease and may serve as an independent risk factor of cardiovascular events. Moreover, serum OPG is increased in diabetic patients, but to this date there are no studies relating plasma OPG levels and hypertension.
Methods: We have studied OPG serum levels by an ELISA immunoassay in 245 patients (median age: 55 years; 63% male): 139 hypertensive, 16 non-hypertensive diabetic, 36 diabetic with hypertension and 54 healthy controls matched by age and sex. Systolic (SBP), diastolic (DBP) and pulse blood pressure (PBP) were also monitored, as well as the circadian blood pressure pattern, left ventricular hypertrophy (by Cornell index) and cardiovascular risk.
Results: As expected, OPG levels were increased in diabetic patients, but there is also a significant increase in serum OPG in hypertensive patients. Serum OPG is clearly reduced in patients with higher values of DBP (measured both in rest and activity, and during 24 h); the same correlation was observed separately in hypertensive or diabetic patients. Moreover, OPG is increased in patients with higher levels of SBP and PBP. On the other hand, serum OPG levels were significantly higher in patients with riser circadian pattern than in patients with dipper, non-dipper and extreme dipper patterns. There are also strong correlations between increased OPG serum levels and prognosis of cardiovascular risk in both hypertensive and diabetic patients, and between increased OPG levels and left ventricular hypertrophy.
Conclusions: Our data show for the first time that OPG levels are clearly increased in hypertensive patients, as well as there is a strong relation between serum OPG and SBP, DBP, PBP, and circadian blood pressure pattern; our findings also strongly support the role of OPG as an indicator of cardiovascular risk and of the presence of diabetes.

Su123 ANGIOTENSIN-(1-7) MODULATES p38 ACTIVITY IN RENAL VASCULAR REACTIVITY IN apolipoproteinE (apoE) DEFICIENT MICE

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Introduction and Aims: Increased vascular reactivity to angiotensin (Ang) II in atherosclerotic vessels could contribute to development of hypertension by altering blood flow in resistance arteries. Recent studies suggested that mitogen activated protein kinases (MAPK) may not only influence inflammatory processes but may also be involved in regulating vascular reactivity. Thus, the aim of our study was to investigate the role of the MAPKs p38 and ERK 1/2 in regulating Ang II induced renal vasoconstriction in apoE deficient mice (apoE -/-). Especially under western diet, apoE-deficiency causes elevated serum cholesterol and triglyceride levels leading to atherosclerosis and increased oxidative stress. Moreover, we examined whether chronic treatment of Ang-(1-7), an endogenous opponent of Ang II acting via the Mas receptor, might influence these effects.
Methods: 12 weeks old apoE -/- and wild type (WT) mice on western diet were treated via osmotic minipumps either with saline or Ang-(1-7) (82μg/kg/hr) for 6 weeks. Vascular reactivity was tested in the model of isolated perfused kidney (n=5-10).
Results: Ang II induced renal pressure response was significantly increased in apoE -/- compared to WT mice. Chronic Ang-(1-7) treatment attenuates Ang II induced pressor response in apoE -/- mice. Pre-treatment with PD 98059 (5μmol/L), an ERK 1/2 inhibitor, had no effect on Ang II mediated vascular reactivity. In contrast, p38 inhibition by SB 203580 (5μmol/L) attenuates Ang II induced renal pressor response in apoE -/- but not in WT mice.
In order to underline the importance of p38, renal cortex homogenates were analyzed for pospho-p38 to total-p38 ratio, a marker of p38 activity (n=5). WT phospho-p38 to total-p38 ratio was considered "baseline" and set to 1.00±0.08. Compared to WT-levels, apoE -/- showed an increase in this ratio to 2.83±0.48, while treatment with Ang-(1-7) in apoE -/- mice restored this ratio almost to WT levels (1.29±0.19).
In addition, we measured isoprostane-8 excretion, a marker for oxidative stress, in 24h-urine samples from these mice (n=9-10). Isoprostane-8 to creatinine ratios did not show significant differences between WT- and ApoE-KO mice on western diet (in ng/mg: 3.56±1.38 vs. 3.25±1.43). Ang-(1-7) markedly reduced total isoprostane-8 excretion and isoporstane-8 to creatinine ratio in ApoE-KO mice on western diet (in ng/mg: 1.79±0.58), indicating a reduced abundance of reactive oxygen species (ROS).
Conclusions: Our data clearly demonstrated a crucial involvement of p38 on Ang II induced augmented renal vascular reactivity in a mouse model of atherosclerosis. Moreover, chronic Ang-(1-7) treatment seems to attenuate this effect by decreasing phospho p38 levels. This counter regulatory mechanism of Ang-(1-7) might be due to reduced oxidative stress and reduced abundance of ROS. Further in vivo and in vitro studies are necessary to elucidate the underlying mechanisms and clarify the potential as a therapeutic agent.

Su124 ANGIOTENSIN AT2 RECEPTOR STIMULATION FAILS TO ALLEVIATE BLOOD PRESSURE (BP) INCREASE INDUCED BY HIGH SALT DIET IN NORMAL WISTAR RATS

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Introduction and Aims: Selective ligands of both Ang II receptor types are needed to study their relative importance or possible interaction in the control of normal or elevated arterial blood pressure (BP); the knowledge of the role of AT2 is still limited.
Methods: We examined if a newly synthetised (A. Lipkowski) agonist of vasodilator AT2 receptors (LKP) would affect increase in BP (7 mmHg) which developed during a 10-day exposure of Wistar rats to high-salt diet (HS, 4% Na w/w).
Results: With LKP treatment (48 mg/kg/24 h orally, beginning from day 3 of HS diet) BP increased more (31 mm Hg) than in untreated rats. With combined treatment: LKP + AT1 receptor antagonist (oral losartan (Los), 15 mg/kg/24 h; gift from Adamed Company, Pienków, Poland), BP increased 19 mm Hg. At the end of studies the response of the renal blood flow (RBF, renal artery Transonic probe) to intrarenal infusion of acetylcholine (Ach, 5-10 μg/kg/h) or norepinephrine (NE, 10-30 μg/kg/h) was determined. In untreated HS rats intrarenal Ach increased RBF 17%, whereas in HS+LKP and HS+LKP+Los groups it decreased RBF 1 and 2%, respectively (NS). LKP treatment did not modify decreases in RBF after NE.
Conclusions: We conclude that stimulation of AT2 receptors did not effectively oppose the increase in systemic peripheral vascular resistance and BP elevation, which follows increased Na intake in Wistar rats. At high AT2 activity the renal vascular bed lost its ability to dilate, which suggests a state of substantial basal vasodilation; the ability to constrict was preserved. This suggests that intrarenal microvasculature is more responsive to AT2 stimulation compared to other peripheral vessels, as it is also more responsive to stimulation of vasoconstrictor AT1 species.
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Su125 QUANTIFICATION OF PROTEINURIA IN MILD PREECLAMPSIA WITH RANDOM ALBUMIN CREATININE RATIO

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Introduction and Aims: There has been considerable discussion regarding the best way to measure daily urinary excretion of protein in preeclampsia. Collection of 24-hours urine samples is still a burden, alternatively, random spot urine albumin creatinine ratio (ACR) has been used for some time as an accurate representation of the 24-h urine collection. The aim of this study was to evaluate the correlation between albuminuria measured as ACR and amount of protein in 24-hour urine samples in women with pre-eclampsia and significant albuminuria.
Methods: 80 hypertensive pregnant women of more than 20 weeks ges-

tational age were enrolled in this study. All had positive urinary test strip for proteinuria of +1 or +2. Women with a concurrent diagnosis of upper urinary tract infection, chronic hypertension (hypertension before pregnancy and persistent elevation of blood pressure before the 20th week of gestation), diabetes mellitus and pre-existing renal disease were excluded. 24 hours urine collection for proteinuria was done for all patients. In addition two urine samples (5 ml each) were collected for measurement of urine albumin creatinine ratio; one was in the morning before starting 24 hours urine collection, and the other one was taken during daytime of 24 hour urine collection. First voided urine samples were discarded.

Results: Two of 80 were excluded because of incorrect sampling of urine for ACR measurement. The morning systolic blood pressure varied from 140 to 158 mmHg (mean, 150 mmHg) and the diastolic blood pressure varied from 90 to 105 mmHg (mean, 114 mmHg). The total volume of urine produced during the 24-hour collection varied between 600 and 3000 ml with mean value 1580 ml. The mean total protein was 1961.46±1683 mg/24h, and the ACR in random samples was 781.31±1041, while in the morning sample ACR was 886.43±1180.9. There was a statistically significant positive correlation between 24 hours urinary protein and urine albumin/creatinine ratio in both daytime random urine sample and morning urine sample. Test result variables and ROC curve showed that the best cutoff point for ACR in daytime random urine sample was 262.5 mg/dl with a sensitivity of 85.5% and a specificity of 81.8%, the positive predictive value was 96.7% and negative predictive value was 47.4%. Meanwhile the best cutoff point for ACR in morning sample was 240 mg per mg/dl with a sensitivity of 94.2% and a specificity of 63.6%, the positive predictive value was 94.2% and negative predictive value was 63.6%.

Conclusions: Albumin/Creatinine ratio is not an ideal diagnostic test but it can be used as a screening test for the detection of significant proteinuria in early preeclampsia.

Su126 PULSE PRESSURE AND KIDNEY FUNCTION IN GHANAIAN VILLAGES

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Introduction and Aims: Pulse Pressure (PP) is a better predictor of mortality in the elderly, in diabetics and in patients with end-stage renal disease, than Systolic (SBP) or Diastolic BP (DBP). However, the relationship between PP and kidney function in normal population is unknown, and has not been investigated in Africans.

Methods: The study investigated the relationship between PP and Glomerular filtration rate [GFR]; in 944 40-75 year olds from 12 villages [6 rural; 6 semi-urban] Ashanti, Ghana; also the relationship between SBP and DBP, and GFR. GFR was measured as the mean of two Creatinine clearances [CrCl] based on 24 hour urine collections, adjusted for body surface area. Data were analysed using SPSS v.16 (Chicago, SPSS Inc). Results are presented as mean (SD) and relationships between variables using regression coefficients with 95% confidence intervals.

Results: The clinical characteristics of the population were: age 55(11) [mean(SD)] years, females 62%, rural village-dwellers 52%, diabetes 1.5%, body mass index (BMI) 21(4) kg/m^2, haemoglobin 12(2)g/dl, cholesterol 147(39)mg/dl, 24hour CrCl 84(23)ml/min/1.73m^2. 29% of the population were hypertensive [BP >140/90mmHg], and while SBP and DBP were not high [125/74 (26/14) mmHg], PP was raised at 51(17) mmHg.

PP increased with age by 0.55 (0.46 to 0.64) mmHg per year and with cholesterol by 0.08 (0.05 to 0.10) mmHg per mg/dl. SBP also increased with age by 0.66 (0.51 to 0.80) mmHg per year and with cholesterol by 0.16 (0.11 to 0.20) mmHg per mg/dl.

GFR decreased both with increasing PP [-0.19 (-0.27 to -0.10) ml/min/1.73m^2/mmHg] and with increasing SBP [-0.09 (-0.14 to -0.03) ml/min/1.73m^2/mmHg] but there was no significant correlation with DBP [-0.04 (-0.15 to 0.06)]. After adjusting for SBP the relationship between GFR and PP became steeper [-0.31 (-0.50 to -0.12) ml/min/1.73m^2/mmHg] while GFR increased as SBP increased after adjusting for PP but the relationship was not significant [0.09 (-0.03 to 0.21) ml/min/1.73m^2/mmHg].

GFR decreased with age [-0.77 (-0.89 to -0.65) ml/min/1.73m^2/year] and increased with BMI [0.78 (0.44 to 1.11)] and was higher in females [85(23) v 82(22); p=0.051]. GFR was not associated with either cholesterol or history of diabetes.

PP was higher (53(17) v 49(15) mmHg; p<0.001) in the semi-urban participants while GFR showed little difference [85(24) v 83(21) [semi-urban v rural] ml/min/1.73m^2].

Using multivariate regression analysis that included PP, age, gender, BMI, only increasing age [-0.75 (-0.88 to -0.62)] and decreasing BMI [0.50 (0.17 to 0.82)] were associated with decreased kidney function.

Conclusions: PP was high overall, and higher in the semi-urban participants; it increased with age. Both PP and SBP showed an inverse relationship to GFR, the relationship being stronger with PP than with SBP. DBP showed no relationship with GFR. When the data were adjusted for age and BMI, PP lost its statistical association with GFR.

Su127 FEASIBILITY OF BLOOD PRESSURE TELEMONITORING IN PATIENTS AFTER RENAL TRANSPLANTATION

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Introduction and Aims: Arterial hypertension is an important cardiovascular risk factor and can deteriorate graft function and possibly trigger acute rejections in patients after renal transplantation. Telemonitoring has potential to be an effective instrument for improving blood pressure management in patients with essential arterial hypertension. Aim of this study was to assess the feasibility of blood pressure telemonitoring in patients after renal transplantation.

Methods: In 7 patients from our renal transplantation outpatient clinic with hypertension and stable graft function blood pressure telemonitoring (I.E.M. Stabil-O-Graph) was tested for the period of 12 weeks. Patients were asked to measure their blood pressure and heart rate twice daily, after getting up in the morning and before going to bed in the evening. Data were transmitted automatically and directly after the measurement via modem or cell phone and were immediately accessible online for patient and physician (I.E.M. e-health service). Patients were regularly contacted via telephone. On the basis of medical history and the transmitted blood pressure data the antihypertensive therapy was modified as needed.

Results: From the 7 patients included 4 were male und 3 female (mean age 56,3 years; range 47 to 68 years). At onset of telemonitoring chronic kidney disease (CKD 5) had been known for 9,2 years on average (range 1,9 – 17,4) and patients had received a kidney transplant between 0,5 and 12,3 years prior (mean 4,7). Before telemonitoring all patients presented an "office blood pressure" > 130/80 mmHg being under antihypertensive therapy of 2,8 drugs on average. Patients transmitted on average 13,1 sets of data out of 14 scheduled per week (range 10,2 – 14). In the course of the monitoring 4,3 modifications of the antihypertensive regime were undertaken on average. There was a significant decrease of the arterial blood pressure measured telemetrically between beginning and end of the study (week 1: 151,4/93,7 mmHg; range 173 – 129/102 – 86 vs. week 12 134,3/83,9 mmHg; range 142 – 125/93 – 75; Wilcoxon signed rank test, p<0,02).

Conclusions: Telemonitoring is a promising tool for the blood pressure surveilance of patients after renal transplantation. The beneficial effect may partly be due to more frequent changes in type or dose of antihypertensive medications, but also more accurate and timely information.

Su128 ALISKIREN IN HYPERTENSIVE PATIENTS WITH CHRONIC KIDNEY DISEASES

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Introduction and Aims: Present guidelines for treatment of hypertension emphasize not only on lowering blood pressure, but also on the ability to protect against subclinical organ damadge.

Aliskiren, a direct inhibitor of renin at the site of its activation is a new antihypertensive drug, present in the last two years. Several trials show its good potential for renal protection.

The aim of our study was to investigate the role of aliskiren in treating hypertension in proteinuric patients and its influence on urinary protein loss and kidney function.
Methods: We present 28 patients treated with aliskiren for 6 months – 15 female and 13 male, aged 33 to 79 – 12 diabetics, 9 with Glomerulonephritis, 7 with ischemic nephropathy. All patients had a long hystory of hypertension and chronic kydney disease – stage III 19 patients – mean eGFR 47,1±0,48ml/min and stage IV 9 patients – mean eGFR 25,9±0,51ml/min. All patients had been treated with different antihypertensive agents up to the start of the study and continued the same therapy during the investigation.
15 patients received 150mg Aliskiren and 13–300mg.
Proteinuria, serum albumin, serum creatinine level, eGFR and serum potassium were measured at 1st, 3rd and 6th month.
Results: Urinary protein loss was reduced from mean 1,59±0,09 to mean 1,16±0,08 g/d in all patients. In diabetics protein loss was reduced from mean 1,82±0,08 to 1,52±0,07 g/d; in patients with glomerulonephritis from mean 1,42±0,04 to 0,97±0,03; in patients with ischemic nephropathy from mean 0,92±0,05 to 0,59±0,03g/d.
23 of the patients reduced the other antihypertensive medications during the trial. 2 remained only with aliskiren.
All patients showed stable renal function during the trial. Mean eGFR at the begining was 49±1,1ml/min and 47±0,9ml/min. at the end of the 6th month. No major side effects were seen.
Conclusions: Patients treated with Aliskiren show a good control of high blood pressure. Aliskiren reduces proteinuria without significantly reducing GFR. These results appear promising for the treatment of hypertensive proteinuric patients and especially diabetics.

Su129 NICORANDIL PREVENTS PROGRESSION OF RENAL DAMAGE WITHOUT CHANGE OF BLOOD PRESSURE IN SPONTANEOUSLY HYPERTENSIVE RATS

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Introduction and Aims: ATP-sensitive potassium channel opener (KCO) has cardioprotective effects in human and animals. Furthermore, recent studies have demonstrated that KCO also has renal protective effects in acute renal failure models such as ischemia-reperfusion model or anti Thy-1 model. But currently renoprotective effects of KCO in chronic renal failure models are hardly investigated. In the present study, the renal protective effect of nicorandil, an anti-anginal KCO with prognosis improvement by cardioprotection, was investigated in renal failure model using spontaneously hypertensive rat (SHR).
Methods: Male SHR and Wistar-Kyoto rat (WKY,11 weeks old, n=8) were used. SHR was divided into two groups; control (SHR-C, n=8) and nicorandil group (SHR-N, n=8). Nicorandil (15 mg/kg/day) was chronically administrated in drinking water for 20 weeks. At the end of administration, systolic blood pressure was measured by a tail cuff method and renal function was determined by plasma and urine parameters (plasma creatinine, urinary protein, N-acetyl-β-D-glucosaminidase (NAG) and β2-microglobulin).
Results: Early stage of hypertensive renal failure was showed in SHR at the end of administration. Significant hypertension was appeared in SHR compared with WKY (SHR-C 207.5±12.0, WKY 136.1±7.3 mmHg, mean±SD, $p<0.01$) with cardiac hypertrophy. In SHR-C, urinary protein was greater than in WKY (SHR-C 33.0±10.0, WKY 5.5±0.9 mg/day, $p<0.01$). β2-microglobulin and NAG in SHR-C were also higher than in WKY. Administration of nicorandil significantly decreased in urinary protein (SHR-N 21.7±7.9, $p<0.05$ vs SHR-C), but did not change in systolic blood pressure (SHR-N 201.8±14.2 mmHg) and heart weight (SHR-C 3.0±0.07, SHR-N 2.9±0.1 g/kg). Neither β2-microglobulin nor NAG, the markers for tubular damage, were changed by nicorandil (β2-microglobulin: SHR-C 60.8±13.1, SHR-N 52.6±8.2 micro-g/day, p=0.15, NAG: SHR-C 0.34±0.06, SHR-N 0.36±0.04 U/day, p=0.48). These results suggested that nicorandil might suppress proteinuria through other mechanism than protecting renal tubule.
Conclusions: Nicorandil prevents progression of proteinuria in SHR without change of blood pressure. This result suggested that nicorandil might have preventive effect in hypertensive nephropathy by some mechanisms other than hypotensive action.

Su130 RAT RENAL ARTERY RELAXATION induced BY WINE POLyPHENOL RESVERATROL

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Introduction and Aims: Resveratrol, a stilbene polyphenol found in grapes and red wine, has recently been found to produce vasorelaxation in endothelium-dependent and endothelium-independent manner. The aim of this study is to determine the mechanism(s) of relaxation produced by resveratrol in the Wistar rat renal artery (RA).
Methods: RA rings were precontracted with phenylephrine. In order to assess the endothelial integrity of the preparation we have used acetylcholine. Endothelium was removed mechanically by rubbing with a steel wire. Failure of arteries to relax to acetylcholine was considered to indicate a state of endothelial denudation. For detection of K^+ channels in smooth muscle a peptide-specific antibodies in immunoperoxidase were used. Resveratrol effect on renal haemodynamics was also studied in vivo in anestetized rats. After abdominal incision, renal artery preparation was utilized and ultrasonic flowprobe, 1RB (internal diameter = 1mm) was placed for renal blood flow (RBF) determination (Transonic System Inc., Ithaca, New York). For detection of K^+ channels in smooth muscle a specific K+ channel antagonist and peptide-specific antibodies in immunoperoxidase were used.
Results: Resveratrol produced concentration-dependent relaxation of RA rings with endothelium and without endothelium (The EC_{50} were: 10 and 15 microM, $P < 0.05$). Methylene blue and L-NAME did not antagonize the resveratrol-induced relaxation of RA rings with endothelium. In order to analyze the contribution of different types of K^+ channels in resveratrol-induced relaxation in the RA, various K^+ channel blockers were used. The relaxation of RA was not blocked by glibenclamide, a selective ATP-sensitive K^+ channel blocker, and tetraethylammonium, a non selective blocker of calcium-dependent K^+ channels. 4-aminopyridine, blockers of voltage-dependent K^+ (K_v) channels, antagonized resveratrol-induced relaxation of RA in a noncompetitive manner. Margatoxin, highly selective blockers of Kv1.1-1.6 channels shifted the concentration response curves induced by resveratrol to the right without significant inhibition of maximal responses. Kv1.3 channels were detected in endothelium but not in the smooth muscle of RA using peptide-specific antibodies in immunoperoxidase. Bolus injection of resveratrol resulted in a mild to moderate increase of RBF in vivo.
Conclusions: Here we have shown that resveratrol produces relaxation of the rat renal artery by activation of voltage-gated K^+ (K_V) channels. It is likely, that endothelial Kv1.3 channels are involved in relaxation of RA produced by resveratrol. However, further experiments with smooth muscle peptide-specific antibodies are necessary to be performed.

Su131 ORAL MAGNESIUM SUPPLEMENTATION REDUCES AMBULATORY BLOOD PRESSURE IN PATIENTS WITH MILD HYPERTENSION

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Introduction and Aims: Accumulating evidence implicates a role of magnesium in the pathophysiology of essential hypertension. Previous studies evaluating the antihypertensive efficacy of magnesium supplementation gave contradictory results. This study aimed to investigate the effect of oral magnesium supplementation on 24-hour blood pressure (BP) and intracellular ion status in patients with mild hypertension.
Methods: A total of 48 patients with mild uncomplicated hypertension participated in the study. Among them, 24 subjects were assigned to 600 mg of pidolate magnesium daily in addition to lifestyle recommendations for a 12 week period and another 24 age- and sex-matched controls

were only given lifestyle recommendations. At baseline and study-end (12 weeks) ambulatory BP monitoring, determination of serum and intracellular ion levels and 24-hour urinary collections for determination of urinary magnesium excretion were performed in all study subjects.
Results: In the magnesium supplementation group small but significant reductions in mean 24-hour systolic and diastolic BP levels were observed (-5.6±2.7 vs -1.3±2.4 mmHg, P<0.001 and -2.8±1.8 vs -1±1.2 mmHg, P=0.002, respectively), in contrast to the control group. These effects of magnesium supplementation were consistent in both daytime and nighttime periods. Serum magnesium levels and urinary magnesium excretion were significantly increased in the intervention group. Intracellular magneium and potassium levels were also increased, while intracellular calcium and sodium levels were decreased in the intervention group. None of the intracellular ions were significantly changed in the control group.
Conclusions: The present study suggests that oral magnesium supplementation is associated with a small but consistent ambulatory BP reduction in patients with mild hypertension.

Su132 COFFEE HAS A FAVORABLE EFFECT ON RENAL ARTERY RESISTIVE INDEX IN HUMAN HYPERTENSION

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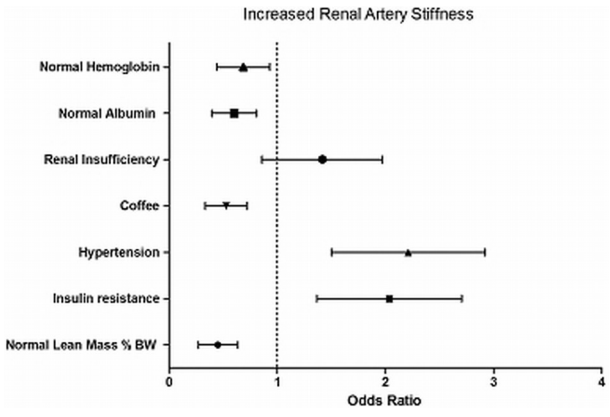
Introduction and Aims: The relationship between nutrition, hypertension and atherosclerosis is known, even dissociated from overt protein malnutrition. Cardiovascular impact and underlying mechanisms of several nutrients are recognized; among them, the action of coffee is still debated and cardiovascular effect of caffeine has been investigated without definite results. The aim of the study is to investigate if coffee habits, and/or quantity of coffee consumption, have any relationship with Renal Resistive Index (RRI), a hallmark of Arterial Stiffness (AS). The relationship of AS with nutritional status, assessed by body composition and serum albumin, insulin resistance (assessed by HOMA) and renal function assessed by glomerular filtration rate (GFR), is concurrently investigated.
Methods: This study was done with 221 consecutive patients, without diabetes, cancer, liver, heart disease, severe renal insufficiency (GFR<30) referred to our day hospital for clinical non-invasive assessment and nutritional counseling: 124 essential hypertensive and 97 non-hypertensive patients were eligible.
Results: By Multiple Linear Regression Fat Free Mass, HOMA (positive relationship) and number of cups of coffee/day (negative relationship) account for 17.2% of the variance to RRI.

Multiple Linear Regression to Renal Resistive Index

Predictors	R 0.414	R2 0.172	F 5.147	sig <0.0001	β	p
Cups of coffee/day, n					-0,182	0,025
Fat Free Mass, %					-0,144	0,063
GFR					-0,024	0,752
Albumin, g/dl					-0,173	0,029
Hemoglobin, g/dl					-0,176	0,034
HOMA					0,158	0,038

Weighted Least Squares Regression - Weighted by Age.



Odds ratios show that lower risk to increased RRI is associated with higher serum albumin, higher hemoglobin and Fat Free Mass; greater risk is associated with hypertension, greater Insulin resistance (HOMA≥ 3.0) and moderate renal insufficiency; coffee habits, assessed by number of cups/day, reduce risk.
Conclusions: Coffee use is inversely associated with the degree of RRI. Habitual coffee users appear to have a risk protection to increased RRI, while lower serum albumin, insulin resistance and moderate renal insufficiency are associated with a greater RRI. The concurrent effect of coffee habits and nutritional state could be important also in the evaluation and reliability of clinical trials with drugs and/or therapeutic regimens, particularly when addressed to modify artery stiffness and/or metabolic profiles.

Su133 ELEVATED PULSE PRESSURE IS ASSOCIATED WITH INCREASED CARDIIOVASCULAR EVENTS IN PATIENTS ON MAINTENANCE DIALYSIS

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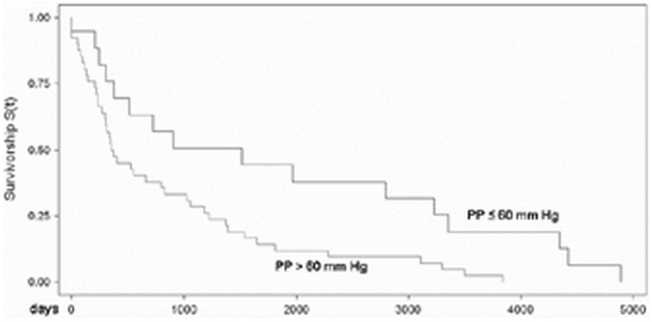
Introduction and Aims: Hypertension in dialysis is common, predominantly systolic, and associated with high pulse pressure (PP). In patients on haemodialysis, PP is a better predictor of mortality than systolic BP (SBP) or diastolic BP (DBP), however its association with cardiovascular events is not established. This study aimed to investigate the association of PP with major adverse cardiovascular events (MACE), i.e. myocardial infarction, stroke, amputation, unstable angina and angiogram in dialysis patients.
Methods: Data were collected on 247 patients undergoing dialysis between 1 January and 30 June 2008 in St Georges Hospital; including demographics, laboratory parameters and MACE since the initiation of dialysis; from patient's notes and electronic records. Analysis was performed using Statistix v7.
Results: The clinical characteristics were, age 64±14 years, 41% females, 38% diabetes, haemoglobin 11±2 g/dl, albumin 31±5 mg/dl, calcium 2.2±0.2 mmol/l, phosphate 1.4±0.4 mmol/l, parathyroid hormone 46±43 pmol/l, SBP 137±22 mm Hg, and DBP 70±19 mm Hg. PP was high 67±19 mm Hg and increased with increasing age (r=0.3, p<0.001). DBP (r=-0.3, p<0.001) decreased with increasing age. SBP did not change with age.
Patients with PP>60 mm Hg had significantly higher MACE than patients with PP≤60 mm Hg (p=0.03). On Kaplan-Meier analysis PP>60 mm Hg was associated poorer MACE free survival (log rank p=0.02).



PP above 60 mmHg was associated with poorer CV event free survival

In a multivariable Cox proportional hazards model analysis PP was the only predictor of MACE, adjusted for age, diabetes and smoking.

VARIABLE	COEFFICIENT	STD ERROR	Z	P value	REL RISK
Age	-0.01505	0.01227	-1.23	0.2203	0.99
PP	0.01379	0.00663	2.08	0.0375	1.01
Diabetes	0.03748	0.25715	0.15	0.8841	1.04
Smoking	-0.04180	0.74745	-0.06	0.9554	0.96

Proportional hazards model showing PP as an independent predictor of MACE

Conclusions: The data demonstrates that PP in dialysis patients was high and elevated PP was associated with higher rates of major adverse cardiovascular events in patients undergoing dialysis.

Su134 BIOMARKERS OF OXIDATIVE STRESS AND LIPID PROFILE IN PATIENTS WITH HYPERTENSIVE NEPHROPATHY

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Introduction and Aims: Etiopathogenetic mechanisms of hypertensive nephropathy are very complex. We wanted to get an insight into the role of oxidative injury, i.e. the disturbed balance of oxidative and antioxidative activity and change of lipid profile in its etiology. That is why the basic aim of this study is determination of biomarkers of oxidative injury and lipid status parameters in patients with hypertensive nephropathy.
Methods: Our study included 283 patients of both sex; 185 men and 98 women, averagely aged 62,6±8,76 years, with hypertensive nephropathy diagnosed according to the clinical and labaratory parameters and 57 healthy subjects in control group (40 men and 17 women, averagely aged 60,5±5,21 years. We determined the lipid status (total cholesterol -TC, LDL and HDL fraction of cholesterol -LDL-C; HDL-C and triglycerides -TG), serum xantine oxidase (OX), serum cathalase (CAT) and malondialdehyde (MDA). Parameters of lipid status, CAT and OX were determined on BIOSYSTEMS A 25 analyser, while MDA values were determined using modified method with tiobarbitur acid (TBA). Sectional study was performed and, in statistical processing, descriptive statistical analysis was utilized.
Results: The results obtained demonstrate that there had been a significant increase of oxidative activity measured on the basis of serum xantine oxidase values (9.73±0.41), compared to the control group (6.57±0.62; $p<0.01$). We also registered a significant decrease of serum cathalase values (23.5±2.7), an important parameter to assess the antioxidative activity, compared to controls (36±4.2; $p<0.01$). In this group, there was a significant increase of lipid peroxidation activity MDA (15±1.7), in comparison with controls (10.2±1.1; $p<0.01$). These changes indicateed outstanding oxidative disbalance in the patients with hypertensive nephropathy. Monitoring of the lipid parameters demonstrated pronounced hypercholesterolemia (7.21±1.74 vs. 4.8±1,1, $p<0.01$). HDL cholesterol fraction was moderately reduced (0.97±0.32 vs. 1.2±0.35, $p<0.05$) while LDL fraction, as the most atherogenic marker of the lipid status, demonstrated a statistically significant increase (4.1±1.1 vs. 3.1±0.6, $p<0.05$). There was also a slight increase in TG fraction 2.8±0.55, $p<0.05$.
Conclusions: This suggests the important roles of chronic oxidative stress and disturbed lipid metabolism in hypertensive nephropathy changes. The results may be of importance in prevention of occurrence or progression of changes in hypertensive microvascular ischaemic nephropathy.

Su135 INFLUENCE OF ARTERIAL STIFFNESS AND 24h-AMBULATORY BLOOD PRESSURE MONITORING ON RENAL LESIONS IN MORBIDLY OBESE PATIENTS WITH MILD OBESITY-RELATED GLOMERULOPATHY

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Introduction and Aims: Obesity and morbidly obesity are health problems of epidemic proportions and are a risk for many conditions as common as type 2 DM and hypertension. Kidney disease is also associated with obesity although this relationhsip is scarcely documented. The aim of this study is to evaluate the influence of 24-hour ambulatory blood pressure monitoring (24h-ABPM) and arterial stiffness on renal lesions.
Methods: 50 morbidly obese patients with normal renal function and biopsy proven early stages of obesity-related glomerulopathy. 24h-ABPM was recorded. Ambulatory arterial stiffness index (AASI) was defined as 1 minus the regression slope of diastolic on systolic blood pressure during 24h-ABPM.
Results: Age 39.98±9.75 years; BMI 52.68±8.9 kg/m^2; 89.5% nocturnal hypertension; 10.5% diurnal hypertension; 60% non-dipper. *Association between AASI and renal lesions:* mesangial cell proliferation (OR: 4.356;95%CI: 1.105-17.167; p=0.035) and glomerulomegaly (OR: 4.40; 95%CI: 1.204-16.140; p=0.025) but not other glomerular or vascular renal lesions were associated with AASI in the multivariant logistic regression model. *Association between 24h-ABPM and renal lesions:* Patients with higher diurnal, nocturnal and 24h- DBP had more vascular lesions than those with lower levels ($p<0.05$). In the multivariate model, only renal arteriolosclerosis was associated with diurnal (OR: 1.141; 95%CI: 1.017-1.279; p=0.024), nocturnal (OR: 1.101; 95%CI: 1.009-1.202; p=0.031) and 24h-DBP (OR: 1.139; 95%CI: 1.020-1.272; p=0.021). Any glomerular lesions were associated to 24h-ABPM in the multivariate logistic regression model. Age but not AASI or any 24h-ABPM measurements was associated with renal arteriosclerosis.
Conclusions: In morbidly obese patients with early stages of obesity-related glomerulopathy: 1. AASI could be an independent risk factor for some glomerular lesions but not for vascular lesions; 2. High DBP but not SBP in the 24h-ABPM could be an independent risk factor for renal arteriolosclerosis; 3. Age was the only risk factor for renal arteriosclerosis.

Su136 ELSARTAN (BV6): THE NEW LIPID-SOLUBLE SARTAN FOR TRASDERMAL USE

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Introduction and Aims: Previous experimental in vitro and in vivo studies in mices and rabbits showed that BV6 succesfully inhibits the hypertensive reaction from angiotensin II (AII) per.os., IV, sc and transdermal administration.This study was designed in order to evaluate the effectiveness of transdermal use of BV6.
Methods: Mices of 300 gr were used. Their blood pressure calculated with CODA II device of Kent Scientific. Hypertensive responce of 50mmHg was steadily reproduced with sc administration of AII at a dose of 50μg/kg.
The previous day the back from the experimental animals were shaved and pealed. In the clean skin of 7 of them (group BV6) a mixture of 50μg/kg BV6 in an excipient of 30% azones, 30% ethylic alcohol,30% propyloglycol and 10% of water was placed.In another group of 4 (placebo group) only excipients (without BV6) was placed.The hypertensive reaction after the administration of 50μg/kg AII was measured at the beginning and after 4 and 8 hours, respectively.
Results: The following table includes the results of the arterial blood pressure of the experimental animals in mmHg.The values are as a mean value ± standart deviation.

Table 1. Results

	Blood pressure baseline	1st bolus AII (t = 0)	2nd bolus AII (t = 4h)	3rd bolus AII (t = 8h)
Placebo (n=4)	101±22	148,5±28	154±32	155±26
BV6 (n=7)	107±12	150±14	131±19	122±20

Conclusions: The new lipid soluble sartan BV6 can succesfully penetrate the dermis to the systematic circulation of the experimental animals and inhibit the hypertensive responce of AII. Further experiments are required in order to procede to clinical trials.

Su137 DOES TELMISARTAN DECREASES MICROINFLAMMATION IN OBESE PATIENTS WITH ARTERIAL HYPERTENSION?

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Introduction and Aims: There are several lines of evidence suggesting that angiotensin II receptor type 1 antagonists posess anti-inflammatory properties. The aim of the present study was to estimate changes of plasma concentrations of selected proinflammatory cytokines after long-term antihypertensive treatment with telmisartan in obese hypertensive patients.

Methods: Thirty four previously untreated obese adults with arterial hypertension were enrolled into the study. A daily dose of telmisartan (40 or 80 mg/d) was adjusted to achieved blood pressure values below 130/80 mmHg. Plasma concentrations of hsCRP, TNFα, IL-6, IL-8 were estimated before and after 6-months telmisartan therapy.
Results: Twenty five patients completed the study. Telmisartan therapy was followed by significant ($p<0.001$) decrease of both systolic blood pressure by 14.2% and diastolic blood pressure by 19.6%. hsCRP and IL-8 concentrations also decreased significantly by 19.2% (5.1±2.9 vs 4.1±1.9 mg/L, p=0.02) and by 28.9%, (3.8±2.1 vs 2.7±1.9 ng/mL, p=0.03), respectively. Plasma concentrations of IL-6 and TNFα also tended to decrease, but the observed changes did not reach statistical significance (3.5±2.4 vs 3.4±2.3 pg/mL and 4.4±2.0 vs 4.1±1.0 pg/mL, respectively).
Conclusions: 1. Telmisartan in monotherapy reduces microinflammation in obese patients with arterial hypertension. 2. Such additional potential benefit of telmisartan therapy may contribute to the slowing of atherosclerotic process and development of cardiovascular complications in obese hypertensive patients.

Su138 ENDOTHELIAL DYSFUNCTION 10 YEARS AFTER PREECLAMPSIA

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Introduction and Aims: Recent studies have shown that women who have had preeclampsia have increased risk of end-stage renal disease and cardiovascular disease. Endothelial dysfunction is central in the pathophysiology of preeclampsia, and might be the unifying link between preeclampsia, cardiovascular disease and renal disease. We wanted to explore this further, by investigating whether preeclampsia was associated with measures of endothelial dysfunction several years after the preeclamptic pregnancy.
Methods: We used data from the Medical Birth Registry of Norway and the Norwegian Population Registry to identify women living in or around Bergen who had preeclampsia in their first pregnancy 9 to 11 years ago. Women with diabetes, rheumatic disease, essential hypertension or renal disease before first birth were excluded, as were those who had later preeclamptic pregnancies. Based on these criteria, 120 women were randomly picked and invited to participate in the study. 120 randomly picked controls, matched on age, year of first birth and municipality were also invited.
We measured blood pressure, weight, height and calculated BMI and waist/hip ratio. Blood samples were drawn for standard laboratory analyses and analyses on biomarkers for endothelial dysfunction. Flow-mediated dilation of the brachial artery (FMD) was used for measuring endothelial function.
Results: Results are based on data from the first 44 women, 26 cases and 18 controls. The study is currently ongoing, and we expect to be finished before June 2010. The results presented in this abstract are therefore temporary. Mean BMI for cases was 26.1 (SD = 5.7) and 26.2 (SD = 5.2) for controls (not statistically different). Mean systolic blood pressure was 121 mmHg (SD = 21 mmHg) for cases, 114 (SD = 11) for controls (p=0.26). Mean diastolic blood pressure was 72 mmHg (SD = 16 mmHg) for cases, 67 (SD = 9) for controls (p=0.27). Mean HOMA-IR (homeostasis assessment of insulin resistance) was 1.8 (SD = 3.9) for cases, 0.97 (SD = 0.58) for controls (p=0.42). Mean flow-mediated dilation was 6.6% (SD = 2.9%) for cases, 7.9% (SD = 3.4%) for controls (p=0.21).
Conclusions: Women who had had preeclampsia in their first pregnancy 10 years ago had a slightly higher blood pressure as well as impaired flow-mediated dilatation as compared to women who had a normal first pregnancy. There was also a tendency towards more insulin resistance in women who had had preeclampsia. These differences were not statistically different, but they are likely to become significant when we have investigated more women and completed the study.

Su139 MODEST SALT REDUCTION IN IMPAIRED GLUCOSE TOLERANCE AND TYPE 2 DIABETES LOWERS BLOOD PRESSURE AND URINARY ALBUMIN EXCRETION

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Introduction and Aims: Reducing salt intake lowers blood pressure, irrespective of whether blood pressure levels are normal or raised. Tight control of blood pressure in diabetics lowers the risk of strokes, heart attacks and heart failure and slows the progression of diabetic kidney disease. Despite the high cardiovascular risk of these patients and theoretical reasons for increased salt sensitivity in these patients, the current knowledge of the role of salt in regulating blood pressure in diabetes is limited.
Methods: We therefore carried out a randomized controlled crossover study of placebo or slow sodium, each for 6 weeks in 26 diet controlled type 2 diabetics and 20 individuals with IGT with untreated normal or high normal blood pressure. We measured the effect of a modest salt reduction on clinic and ambulatory blood pressure and urinary albumin excretion.
Results: 24h urinary sodium was 165±9mmol/24h on slow sodium and 117±10mmol/24h on placebo, with a reduction in urinary sodium of 49±9mmol/24h, equivalent to 2.9g/day salt. This modest salt reduction significantly lowered SBP from 135.5±2.0mmHg to 131.2±1.9mmHg, a fall of -4.2±1.5mmHg ($p<0.01$). DBP was also reduced from 81.3±1.1mmHg to 79.7±1.2mmHg, a reduction of -1.7±0.9mmHg, with borderline significance (p=0.055). This effect was also evident in ambulatory blood pressure monitoring with a reduction in mean day SBP by -3.3±0.9mmHg, mean night BP by -4.3±1.2/2.3±0.9 mmHg and mean 24h BP by -3.3±0.9/1.8±0.8mmHg. ACR was reduced from 0.73 (IQR 0.5-1.5) mg/24h on slow sodium to 0.64 (IQR 0.3-1.1) mg/24h on placebo (p=0.014). There was therefore a 12% reduction in ACR with this modest reduction in salt intake.
Conclusions: This modest salt reduction causes significant and clinically relevant falls in blood pressure in type 2 diabetes and impaired glucose tolerance, where initial blood pressure levels are normal or high normal. Furthermore other additional benefit was found in the small reduction in albumin creatinine ratio. These findings support the recommendation to reduce salt intake in diabetes as recommended in hypertension guidelines to less that 6g/day.

Su140 INCREASED ENDOGLIN LEVELS IN HYPERTENSIVE AND DIABETIC PATIENTS

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Introduction and Aims: Hypertension is the second most common cause of chronic renal disease, next to diabetes. Renal hypertension puts stress and increases pressure on the kidney. Endoglin is a cytokine, member of the receptor system of transforming growth factor-beta 1. Several authors suggest that endoglin, which is constitutively expressed in endothelial cells, may be linked to vascular alterations or even fibrosis. Endoglin plays a critical role in both vascular development and endothelial normal functions. Previous studies from our group show that endoglin levels are altered in animal models of diabetes in vivo. However, there are no studies concerning the role of endoglin serum levels in hypertensive or diabetic patients.
Methods: We have studied endoglin serum levels by an ELISA immunoassay in 288 patients (median age: 55 years; 62% male): 159 hypertensive, 22 non-hypertensive diabetic, 42 diabetic with hypertension and 65 healthy controls matched by age and sex. Blood glucose, glycated hemoglobin, systolic (SBP), diastolic (DBP) and pulse blood pressure (PBP) were also monitored, as well as the circadian blood pressure pattern, left ventricular hypertrophy (by Cornell and Sokolow index) and cardiovascular risk.

Results: Serum endoglin levels were increased in patients with higher values of blood glucose (>160 mg/dl) and higher percentage of glycated hemoglobin (> 6%). Endoglin levels were also increased in hypertensive and diabetic patients with higher levels of SBP and PBP. Moreover, serum endoglin was significantly higher in diabetic patients with extreme dipper circadian pattern than in patients with dipper, non-dipper and riser patterns. On the other hand, serum endoglin was also higher in hypertensive or diabetic patients with left ventricular hypertrophy, and also in diabetic patients with elevated cardiovascular risk.
Conclusions: Our study suggests for the first time a strong correlation between the presence of diabetes and higher levels of endoglin. Moreover, our data shows the close relation between serum endoglin and SBP, PBP and circadian blood pressure pattern in diabetic patients; our findings also strongly support the role of endoglin as an indicator of cardiovascular risk.

Su141 BLOOD PRESSURE AND SERUM INTER-CELLULAR ADHESION MOLECULE 1 (S-ICAM-1) CHANGES AFTER LOW FRUCTOSE DIET IN PATIENTS WITH CHRONIC KIDNEY DISEASE (CKD) AND DIFFERENT CLINIC BLOOD PRESSURE PROFILE

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Introduction and Aims: Increased fructose consumption is associated with hyperuricemia, vascular damage and could lead to hypertension. In this study we tested the hypothesis that lower daily fructose intake may improve blood pressure and diminish the inflammatory state in subjects with chronic kidney disease based on whether they were dippers (BP falling at night) or nondippers (BP does not fall during sleep).
Methods: We included 28 patients (age 59,96±14,63 years old) with non-diabetic CKD; eGFR $_{MDRD}$ 46,89±12,54 ml/min/1,73m^2. 24-hours ambulatory blood pressure monitoring (ABPM), blood and urine collection were performed before and after 6-weeks low fructose diet (LFD) without changing of total energy requirement. On base of first result of ABPM the patients were divided into two groups: 20 dipper patients and 8 non-dipper patients. Baseline age, kidney function, serum uric acid and fructose consumption were similar between groups.
Results: The mean daily fructose consumption was reduced from 59±22 grams before LFD to 12±3 grams during 6-week period of LFD (p<0,001). The effect of LFD on blood pressure and some laboratory parameters is presented in table (results: mean ± SD).

Characteristic of dippers group

	Before LFD	After LFD	p
Systolic BP [mmHg]	129,9±14,2	124,3±7,6	0,061
Diastolic BP [mmHg]	80,7±10,70	75,2±8,10	0,019
eGFR [ml/min/1,73m^2]	47,90±12,7	48,50±12,8	ns
Serum uric acid [mg/dl]	7,12±1,47	6,77±1,44	ns
Urinary uric acid excretion [mg/day]	531±294	474±253	ns
hsCRP [mg/L]	4,11±4,13	2,42±3,18	0,001
sICAM-1 [ng/ml]	247,48±52,55	233,90±53,23	ns

Characteristic of non-dippers group

	Before LFD	After LFD	p
Systolic BP [mmHg]	132,1±12,1	132,5±14,7	ns
Diastolic BP [mmHg]	75,9±8,20	76,9±6,2	ns
eGFR [ml/min/1,73m^2]	46,2±13,5	47,0±15,9	ns
Serum uric acid [mg/dl]	6,90±0,80	6,09±0,97	ns
Urinary uric acid excretion [mg/day]	453±195	459±292	ns
hsCRP [mg/L]	5,03±6,65	5,48±6,35	ns
sICAM-1 ng/ml	258,54±76,23	231,25±37,45	ns

Correlation in dippers group:
a) delta systolic BP with delta eGFR (r = 0,50 p=0,04) and delta urinary uric acid excretion (r = 0,51 p=0,02)
b) delta diastolic BP with delta urinary uric acid excretion (r = 0,44 p=0,05) and delta s-ICAM (r=0,40, p=0,09).
In non-dippers group was found positive correlation between sICAM and: systolic (r=0,71, p=0,05) and diastolic BP (r = 0,75; p=0,03) after LFD.
Conclusions: Low fructose diet reduces BP and inflammation in subjects with CKD with a dipper BP profile. Lack of improvement of blood pressure control in non-dippers group could be related to persistent inflammatory state.

Acute kidney injury – basic research 2

Su142 INVOLVEMENTI OF DDAH-ADMA AXIS IN ACCELERATED RENAL INJURY IN ACUTE ISCHEMIA-REPERFUSION INJURY

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Introduction and Aims: Recent evidence suggests that injury to the renal vasculature may play an important role in the pathogenesis of ischemic acute kidney injury (AKI). Since nitric oxide (NO) is a vasodilator and known to play an important role in the maintenance of renal microvasculature and its flow and may exert a protective role against AKI, it is conceivable that asymmetric dimethylarginine (ADMA), an endogenous inhibitor of NO synthase, could enhance tubulointerstitial injury during AKI by decreasing NO bioavailability. However, the association between ADMA and AKI remain to be elucidated.
Methods: To determine the role of ADMA in AKI, eight-week-old male C57BL/6J (wild) mice and DDAH-1, a key enzyme for ADMA degradation, transgenic (Tg) mice were used in this study. Ischemia- reperfusion (IR) injury were performed in wild mice with (n=8) or without (n=11) ADMA infusion (0.01mg/kg/min) and Tg mice (n=5). Sham operated mice (n=5) were used as control. Renal function and morphology of acute renal injury were compared after IR. Tissue or plasma levels of ADMA were measured by HPLC. ADMA-related related enzymes such as DDAH and PRMT, an enzyme for ADMA synthesis, were measured by western blot analysis.
Results: Western blot analysis revealed significant decreases in renal DDAH expression levels and increases in PRMT expression levels during IR injury associated with increased ADMA levels in wild mice. Compared with IR-treated wild mice, ADMA infusion markedly enhanced tubular injury and increases in BUN levels, whereas these changes were significantly attenuated in IR-treated DDAH Tg mice.
Conclusions: These results strongly suggest that the active involvements of ADMA-DDAH axis in the pathogenesis of IR injury. ADMA-DDAH axis could be a novel target for patients with AKI.

Su143 ★ INHIBITION OF p53 BY PIFITHRIN-α PROTECTS AGAINST SEPSIS-RELATED ACUTE KIDNEY INJURY

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Introduction and Aims: Acute kidney injury (AKI) is a common clinical problem, typically associated with factors such as sepsis, hypovolemia, heart failure and radiocontrast use. The pathophysiology of sepsis-induced AKI involves ischemic and toxic injury to the renal tubule epithelium, resulting in necrosis and apoptosis. In addition, the oxidative stress occurring during sepsis can induce renal damage, leading to high mortality. Sepsis is also a transcriptional activator of p53, a tumor suppressor protein that modulates cellular stress responses and regulates the cell cycle, controlling the activation of Bax (apoptosis) and p21 (proliferation). We investigated the role of pifithrin-α (PIF-α), a specific inhibitor of p53, in sepsis-related AKI, using a cecal ligation and puncture (CLP) model.
Methods: Wistar rats were randomly divided into three groups: control (n=5); CLP (n=8); and CLP+PIF-α (n=8). CLP+PIF-α rats received PIF-α

via three i.p. injections (each 2.2 mg/kg) as follows: 24 h before CLP; 1 h before CLP; and 24 h after CLP. We measured mean arterial pressure (MAP), inulin clearance (CIn) and fractional interstitial area (FIA) at 24 h after CLP. In addition, kidney tissue samples were submitted to immunohistochemical staining for p53, Bax, ED1 (monocytes/macrophages) and proliferating cell nuclear antigen (PCNA), as well as to immunoblotting for cytoplasmic p21.
Results: Treatment with PIF-α improved the GFR and MAP. After CLP, the FIA increased in the CLP group but not in the CLP+PIF-α group. In the renal cortex samples from the CLP+PIF-α group, the p53 staining score was lower and there was less infiltration by monocytes/macrophages. In addition, PIF-α improved p21 expression, resulting in fewer PCNA-positive cells.

Variable	Control	CLP	CLP+PIF-α
CIn (mL/min/100g BW)	0.84±0.04	0.46±0.06**	0.67±0.10^{+}
MAP (mmHg)	108.0±2.08	91.75±3.65***	103.43±3.43^{++}
p53 (score)	0.56±0.07	0.80±0.05**	0.52±0.06^{++}
Bax (score)	0.48±0.01	0.59±0.01	0.49±0.01
ED1 (+cells/grid field)	4.52±0.25	8.3±0.12***	4.87±0.66
PCNA (+cells/grid field)	1.27±0.27	3.08±0.25**	1.79±0.36^{+}
p21 (% of control)	98.96±2.11	59.67±1.66***	82.77±3.62^{+++}
FIA (%)	5.44±0.88	12.55±0.72***	8.97±0.32***$^{,+++}$

**p<0.01 vs. control; ***p<0.001 vs. control; $^{+}$p<0.05 vs. CLP; $^{++}$p<0.01 vs. CLP; $^{+++}$p<0.001 vs. CLP. Data are expressed as mean±SEM. Scores range from 0 to 4. Grid field, 0.087 μm^2.

Conclusions: Our results show that PIF-α treatment protected the renal function of animals presenting sepsis-induced AKI, and that it did so by suppressing p53 and inducing p21, thereby reducing cell proliferation. Our data provide a novel description of the mechanisms by which sepsis causes kidney injury. (Supported by FAPESP-06/56320-0 and CNPq).

Su144 ★ THE ROLE OF INNATE IMMUNITY IN THE ACUTE RENAL FAILURE TRIGGERED BY THE ISCHEMIA AND REPERFUSION INJURY (IRI)

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Introduction and Aims: AKI is a major problem in clinical. Its pathogenesis is quite complex but it is known that the AKI is caused by an intense inflammatory process. The innate immune system provides a variety of pattern recognition receptors (PRR) such as Toll-like receptors (TLR) and NOD-like receptors (NLR) which have the task of identifying the presence of foreign pathogens and/or endogenous ligands released during an inflammatory process of tissue damage or necrotic cells. The aim of our study was the involvement of PRR: TLR and NLR and molecules that are part of the complex inflammasomes: NALP3, ASC and Caspase-1 in renal dysfunction during the of ischemia and reperfusion.
Methods: The knockout (KO) for TLR-2, TLR-4, MYD88, NOD-1, NOD-2,Nod-1/2, ASC and caspase-1 animals were subjected to 45 minutes of ischemia and 24h of reperfusion. Blood and renal tissue were collected at different times for biochemical analysis, histology, gene and protein expression.
Results: First, we observed in TLR-2, TLR-4, MYD88, NOD2, ASC and caspase-1 KO showed a significant protection of renal function when compared to WT animals, with low levels of creatinine and serum urea and with lower percentage of NTA.

Mean serum creatinine (SD) of animals WT and KO for innate receptors

Serum creatinine (mg/dl) 24h after IRI						
WT	MYD88 KO	NOD-1 KO	NOD-2 KO	NOD-1/2 KO	ASC KO	Caspase-1 KO
3,04±0,91	0,87±0,17	2,97±0,73	0,79±0,2	0,97±0,32	0,72±0,18	0,86±0,21

Furthermore, the analysis of pro-inflammatory cytokines showed that these animals had lower levels of IL-1b, IL-6, TNF-alpha, and chemokine MCP-1. Even more interesting was that we showed that TLR-2 and 4 are important for the interaction between innate and adaptive immunity through its influence on Th1 (pro-inflammatory)/TH-2 (anti-inflammatory) balance of CD4 + T cells. We observed that gata-3 was up-regulated mainly 24h after reperfusion, in the KO animals (TLR-2: 3.05±1.36; TLR-4: 2.94±1.58) while the WT, it was down-regulated (0.55±0.10). Interestingly, T-bet expression was blocked in the KO animals and up-regulated in the WT. We observed the same with IL-4, IL-10 and INF-γ.

Gene expression (mRNA) of cytokine in the WT and KO animalssubjected or not to IRI 24h subjected to IRI

	mRNA of renal tissue after IRI						
	WT	MYD88 KO	NOD-1 KO	NOD-2 KO	NOD1/2 KO	ASC KO	Caspase-1 KO
IL-1β	8.23	1.95	2.38	1.37	1.17	1.33	2.40
IL-6	6.78	1.74	1,57	1.22	1.35	1.47	1.17
TNF-α	6.96	0.99	1.36	1.01	1.01	2.14	0.91
MCP-1	5.47	0.57	0.98	1.4	1.18	1.1	0.77

Conclusions: In summary, we observed that the IRI involves the activation of different PRR as initiators of the inflammatory response and may influence the Th1/Th2 balance of immune response resulting in severe renal dysfunction.

Su145 ★ IFNδ SIGNALING PATHWAY ON MESENCHYMAL STEM CELL IS CRUCIAL TO THEIR RENOPROTECTIVE ACTIONS

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Introduction and Aims: Renoprotective role of mesenchymal stem cells (MSC) is well known. In acute injury models, MSC mainly repair by paracrine way, inducing immunomodulation, proliferative and anti-apoptotic responses. However, how MSC act is greatly unknown. In a inflammatory milieu, these cells can be activated at damaged kidney, however its role in renoprotection is still unexplored. Thus, we analyzed the necessity of IFNg receptor signaling on MSC to achieve renoprotection.
Methods: MSC were isolated from adipose tissue (AdSC) of IFNg Receptor knockout animals and from C57Bl6 as control. These cells were characterized by cytometry and differentiation assays. Ischemia reperfusion injury experimental model were achieved in C57Bl/6 male by clapping both renal pedicles for 45 min. After 4h, 2.105 AdSC and AdSC from IFNgR knockout (AdSC IFNgR-KO) animals were administrated intraperitoneally, and animals sacrificed 24h after surgery.
Results: Functional analysis demonstrated a significant urea and creatinine serum levels reduction in AdSC-treated animals, but no protection were seen with AdSC obatained from IFNgR-KO. NTA analyses correlate with these data. Further analyses on kidney inflammatory response demonstrated that IL-6 was higher expressed in AdSC IFNgR KO-treated animals compared with AdSC treatment. On the other hand, IL-4 mRNA expression was higher in AdSC-treated animals compared to untreated, and lower in AdSC IFNgR-KO-treated animals.
Conclusions: These results indicated that IFNg receptor signaling is necessary to full MSC repair response, but it is not essential, since AdSC from IFNg-KO leads to a mild amelioration of AKI symptoms. More studies on this field are necessary to understand the biological role of MSC at regenerative repair on cellular therapy. FAPES and CNPq.

Su146 ★ THE FUNCTION OF POXIMAL TUBULAR SUPPRESSOR OF CYTOKINE SIGNALLING 3 IN ACUTE KIDNEY INJURY

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Introduction and Aims: Acute kidney injury (AKI) is a major clinical

problem associated with high morbidity and mortality. Currently, therapies for treatment of AKI are unsatisfactory. In an attempt to identify new therapeutic targets, we found that suppressor of cytokine signalling 3 (Socs-3) is up-regulated after ischemia reperfusion (IR) injury in mouse proximal tubules. Socs-3 is a major intracellular negative regulator of the JAK/STAT and other receptor tyrosine kinase pathways, and therein it regulates many important cytokines and growth factors involved in cell injury and repair. Socs-3 has been implicated as a regulator of inflammation and regeneration in different organ systems but its role in the kidney has not been addressed.
Methods: To elucidate the role of Socs-3 in AKI we used the *loxP* system to create a proximal tubular conditional knockout mouse (Socs-$3^{\Delta sglt2/\Delta sglt2}$) by mating a Socs-$3^{fl/fl}$ mouse with a sglt2-cre mouse. Bi-lateral clamping of the renal pedicels was used to induce IR injury and AKI in Socs-$3^{\Delta sglt2/\Delta sglt2}$ and Socs-$3^{fl/fl}$ control mice. Renal function was monitored and kidneys were analysed by immunohistochemistry, quantitative RT-PCR and immunoblot.
Results: IR injury on Socs-$3^{\Delta sglt2/\Delta sglt2}$ and control mice revealed no significant difference in mortality or kidney function measured by serum urea and creatinine. However, analysis of post-ischemic kidneys revealed marked differences in inflammation and repair processes. At 72 hours post clamping, when Socs-3 expression peaks in wild-type mice, Socs-$3^{\Delta sglt2/\Delta sglt2}$ mice had significantly more proximal tubular cell proliferation as shown by Ki-67 staining. Kidneys from Socs-$3^{\Delta sglt2/\Delta sglt2}$ mice also had significantly higher expression of inflammatory markers such as interleukin-1 beta, greater transforming growth factor-β expression, and significantly more infiltrating leukocytes.
Conclusions: Our data illustrates the complex downstream effects of proximal tubular Socs-3 expression. While an increased Socs-3 level might attenuate post-ischemic inflammatory processes, it also suppresses reparative mechanisms such as epithelial proliferation. We speculate that the lack of functional differences in our model was due to these ambivalent functions of Socs-3. Further studies are therefore being performed to address acute phase differences and possible long-term effects of renal Socs-3 expression.

Su147 ★ ALISKIREN PROPHYLAXIS ON EXPERIMENTAL CONTRAST-INDUCED NEPHROPATHY MODEL

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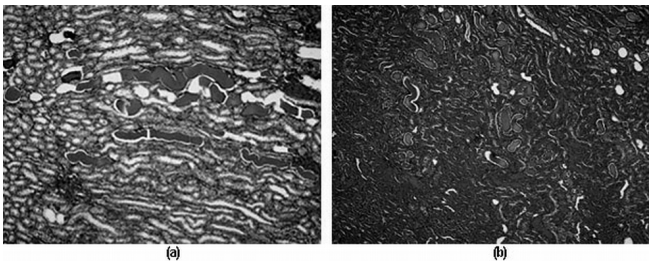
Introduction and Aims: Renal medullary hypoxemia is believed to play an important role in the pathophysiology of contrast-induced nephropathy (CIN). Vascular endothelial growth factor (VEGF) plays an important role in response to medullary hypoxemia by providing cellular adaptation to the contrast-induced hypoxic stress. Aliskiren, which inhibits renin, angiotensin I and angiotensin II biosynthesis, improves dye induced renal vasoconstriction. We hypothesized that aliskiren pretreatment may confer protection against CIN through the inhibition of renin-angiotensin system.
Methods: Thirty-two Wistar-albino rats were divided into 4 groups (n=8 each); control group, aliskiren group, CIN group, CIN+aliskiren group. Aliskiren (50 mg/kg/day) was given by oral gavage for 5 consecutive days prior to induction of CIN. CIN was induced at day 4 by iv injection of indomethacin (10 mg/kg), L-NAME (10 mg/kg) and meglumine amidotrizoate (6 ml/kg). Renal functional parameters, histopathologic findings and VEGF expression were evaluated.
Results: Mean serum creatinine levels were found significantly lower in CIN+aliskiren group compared to CIN group (0,52±0,12 mg/dl vs 1,01±0,43 mg/dl, p=0.009) and was similar to the control groups (p>0.05). Mean creatinine clearance (CrCl) in CIN+aliskiren group (0,90±0,37 ml/min) was significantly higher than CIN group (0,57±0,37 ml/min, p=0.046), and was significantly lower than the control groups (1,42±0,50 ml/min in group 2, p=0,028; 1,92±0,67 ml/min in group 4, p=0,003). The mean scores of tubular necrosis (p>0.05), proteinaceous cast (p>0.05), medullary congestion (p>0.05) and VEGF expression (p>0.05) in CIN+aliskiren group, were not significantly different from the CIN group (Table 1).

Comparisons of the histopathologic and immunohistochemical findings between the study groups

	Control group (n=8)	Aliskiren group (n=8)	CIN group (n=8)	CIN + aliskiren group (n=8)
Tubular necrosis	0.25±0.46$^{\sigma}$	0.28±0.48$^{\sigma}$	2.50±0.53	2.75±0.46$^{\sigma}$
Proteinaceous cast	0.37±0.51$^{\sigma}$	0.14±0.37$^{\sigma}$	2.87±0.64	3.00±0.53$^{\sigma}$
Medullary congestion	0.75±0.88$^{\sigma}$	0.85±0.37$^{\sigma}$	3.50±0.53	3.00±0.53$^{\sigma}$
VEGF	0.50±0.53$^{\sigma}$	0.42±0.53$^{\sigma}$	2.62±0.51	2.37±0.51$^{\sigma}$

$^{\sigma}$ CIN + aliskiren group vs control group and aliskiren group, p<0.05. Evaluation: no changes (0), mild (1), moderate (2), severe (3), very severe (4). VEGF: Vascular endothelial growth factor



Conclusions: Aliskiren prophylaxis improved deterioration of CIN induced renal functions but was not able to prevent contrast induced histopathological changes. Further studies are needed to delineate the pathophysiological mechanisms that aliskiren prevents contrast induced renal functional deterioration.

Su148 RENAL AND VASCULAR EFFECTS OF Phalaris canariensis IN NORMOTENSIVE AND SPONTANEOUSLY HYPERTENSIVE RATS

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Introduction and Aims: Hypertension is a disease highly prevalent throughout the world and it is associated with three chronic diseases namely; stroke, kidney disease, and heart disease. Popularly the birdseed Phalaris canariensis (Pc) has been used as an antihypertensive agent. The aim of this study was to evaluate the effect of the aqueous extract of Pc (EAPc) on arterial pressure and renal function of normotensive Wistar rats and spontaneously hypertensive rats (SHR). Also it was evaluated whether EAPc treatment would be able to prevent the development of hypertension in SHR, by administrating EAPc soon after weaning, i.e. before the establishment of hypertension.
Methods: Adult male Wistar rats, SHR and young (3 weeks old) SHR were used. Adult animals were divided into groups control (receiving water) and treated with EAPc (100 mg/kg/day, p.o.) for 30 days. After this period treated group was divided into 2 sub-groups: treated for another 30 days (EAPc 60) and a group which EAPc treatment was interrupted and animals received only water for 30 days (EAPc 30). Young (three weeks old) and non-hypertensive SHR, received EAPc treatment during 30 days.
Results: The administration of the EAPc for 60 days produced significant reduction in mean arterial pressure (MAP, mmHg) in both adult groups, Wistar (122±2 vs 103±4, p<0.05) and SHR (210±1 vs 171±, p<0.05). However, the interruption of treatment was followed by a gradual return of MAP to the baseline levels in both groups Wistar (117±1) and SHR (193±3). Young SHR group was normotensive after weaning (122±2) but became hypertensive after 30 (161±3) and 60 days (195±4). EAPc treatment during 30 and 60 days minimized the increase in MAP (133±2 and 148±4, respectively). The discontinuation of the treatment caused an increase in the MAP similar to observed in untreated group. There was no significant change in plasma parameters among groups. Fractional excretion of Na+ (FENa) was lower in untreated SHR and EAPc treatment restored FENa to control levels.
Conclusions: The results suggest that Pc has potential antihypertensive effect in SHR, without inducing any significant risk of nephrotoxicity and it may be use as an alternative and/or adjuvant treatment option to reduce blood pressure. The antihypertensive mechanism may be related to tubular transport of sodium.

Su149 FAS LIGAND-MEDIATED FRATRICIDE ON RENAL TUBULAR CELLS *IN VIVO*

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Introduction and Aims: Acute kidney injury (AKI) has been shown to be mediated, at least partly, through Fas-dependent apoptosis. Both tubular cells and infiltrating immune cells might provide the disease-causing source of Fas Ligand (FasL) in AKI as investigated by a mouse model of cisplatin induced nephrotoxicity (CIN).
Methods: Initially, we demonstrate that treatment of mice with the FasL inhibitory monoclonal antibody MFL3 restores kidney function after CIN. In order to investigate the functional relevance of FasL expression on tubular cells we treated SCID-Beige mice with MFL3 after induction of CIN and isolated whole proximal tubule segments to treat them with cisplatin and MFL3 or cisplatin alone.
Results: Whereas all wildtype mice died from CIN within 5 days, SCID-Beige mice exhibit a significant survival benefit. Interestingly, six days after cisplatin application, SCID-Beige mice developed renal failure as quantified by highly elevated serum creatinine levels. 55,6% mortality rate was registered in SCID-Beige mice at day 10. Treating SCID-Beige mice with MFL3 completely protected from lethal CIN until day 16. Incubation of complete proximal tubule segments with cisplatin resulted in 85% apoptotic cells after 6 hours which could be prevented by coincubation with MFL3 in the complete absence of immune cells.
Conclusions: We conclude that infiltration of FasL positive immune cells accelerates and deteriorates CIN. For the first time, we demonstrate that FasL-mediated fratriced on renal tubular cells is of functional relevance in the pathogenesis of CIN, even in the absence of immune cells.

Su150 PROTECTIVE EFFECT OF HIGH VOLUME HEMOFILTRATION ON SEPTIC PLASMA-INDUCED TUBULAR EPITHELIAL AND ENDOTHELIAL CELL INJURY

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Introduction and Aims: The mechanisms of sepsis-induced acute kidney injury (AKI) are not only related to hypoperfusion but also to a detrimental activity of circulating mediators on kidney cells. We previously demonstrated that septic plasma induce apoptosis and functional alterations of tubular and glomerular epithelial cells. Sepsis is also associated with endothelial dysfunction that contributes to the development of AKI. The aim of this study was to evaluate the efficiency of High Volume Hemofiltration (HVHF) to remove from septic plasma soluble mediators involved in AKI.
Methods: We selected 5 patients with sepsis-associated AKI (IVOIRE study, inclusion in RIFLE criteria) and we collected their plasma at different time points (1, 6, 12 and 72 h) after the start of HVHF (70 ml/kg/h restitution fluids, polyethersulfone membrane 1.9 m^2, 1/3 pre-dilution). We evaluated the effects of plasma on leukocyte adhesion, apoptosis (TUNEL, caspase ELISA) and functional alterations of tubular epithelial (trans-epithelial electrical resistance, albumin uptake) and endothelial cells (angiogenesis assay). We evaluated plasma levels of mediators involved in inflammation and apoptosis (TNF-alpha, Fas-Ligand, CD40-Ligand and von Willebrand Factor ELISA).
Results: Septic plasma collected after 1h HVHF induced leukocyte adhesion on endothelial and tubular cells through the up-regulation of ICAM-1 and CD40. On endothelial cells, septic plasma induced a cytotoxic effect associated with an impairment of angiogenesis. On tubular cells, septic plasma induced a pro-apoptotic effect via Fas (CD95) up-regulation and caspase activation and several functional alterations such as the loss of cell polarity, the altered expression of endocytic receptors and tight junction molecules and the impaired ability to internalize albumin. In comparison to plasma obtained after 1h HVHF, leukocyte adhesion as well as endothelial and tubular injury was significantly reduced incubating cells with plasma collected after 6, 12 and 72 h HVHF. Moreover, the effluent fluid collected after 1h HVHF induced a dose-dependent cell injury, suggesting the potential removal of detrimental mediators by HVHF. In accordance to in vitro data, plasma concentrations of TNF-alpha, Fas-Ligand, CD40-Ligand and von Willebrand Factor decreased after 6h and then remained stable after 12 and 72 h HVHF.
Conclusions: Septic plasma induce a direct injury of tubular and endothelial cells by favouring leukocyte adhesion, by triggering apoptosis and by altering different biological functions. These effects are related to the presence of circulating inflammatory mediators that could be removed by HVHF. However, the protective effect of HVHF is limited at the first hours of treatment, suggesting that unexplored mechanisms (membrane adsorption, polarization, etc.) may interfere with convective-driven solute removal.

Su151 RENOPROTECTIVE EFFECTS OF TERLIPRESSIN ON EXPERIMENTAL CONTRAST-INDUCED NEPHROPATHY MODEL

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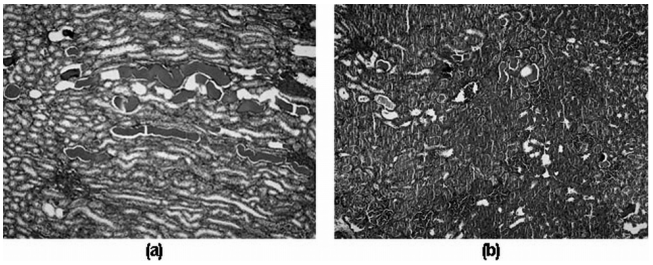
Introduction and Aims: The reduction in renal blood flow and direct toxic action on renal tubular cells are considered to be involved in the pathophysiology of contrast-induced nephropathy (CIN). Up-regulation of hypoxia inducible factor (HIF) and HIF target genes including vascular endothelial growth factor (VEGF) play an important role in the response to regional renal hypoxia. Terlipressin, an agent that improves intrarenal vasodilatation was used as a prophylactic regimen in experimental CIN model. We hypothesized that terlipressin may prevent CIN due to its renal vasodilatation effects.
Methods: Thirty-two Wistar-albino rats were divided into 4 groups (n=8 each); group 1 (terlipressin with CIN model), group 2 (terlipressin), group 3 (CIN model), group (control). Terlipressin (0.017 mg/kg/min for 3 minutes) was given just before the CIN protocol. CIN was induced by iv injection of indomethacin (10 mg/kg), L-NAME (10 mg/kg) and meglumine amidotrizoate (6 ml/kg). Renal functional parameters, renal histopathologic findings of CIN and immunohistochemical VEGF expression were evaluated on rats.
Results: It was found that mean serum creatinine value in group 1 (0,53±0,16 mg/dl) was significantly lower than group 3 (1,01±0,43 mg/dl, p=0.027) and was similar to the control groups (p>0.05). Mean creatinine clearance (CrCl) value in group 1 (1,07±0,31 ml/min) was significantly higher than group 3 (0,57±0,37 ml/min, p=0.021) and was similar to the

Comparisons of the histopathologic and immunohistochemical findings between the study groups

	Group 1 (n=8)	Group 2 (n=8)	Group 3 (n=8)	Group 4 (n=8)
Tubular necrosis (mean±SD)	1.37±0.5†	0.12±0.35	2.50±0.53†	0.60±0.89
Proteinaceous cast (mean±SD)	2.25±1.28$^{\sigma}$	0.12±0.35	2.87±0.64$^{\sigma}$	1.00±1.22$^{\sigma}$
Medullary congestion (mean±SD)	2.25±0.88†	1.12±0.35	3.50±0.53†	1.40±1.34
VEGF (mean±SD)	1.00±0.53†	0.50±0.53	2.62±0.51†	1.00±1.22

†Group 1 vs group 3, p<0.05, $^{\sigma}$ group 1 vs group 3 and 4, p>0.05. Evaluation: no changes (0), mild (1), moderate (2), severe (3), very severe (4). VEGF: Vascular endothelial growth factor



control groups ($p>0.05$). The mean scores of tubular necrosis ($p<0.05$), medullary congestion ($p<0.05$) and VEGF expression ($p<0.05$) were significantly higher in group 3 compared to the other groups (Table 1). Treatment with terlipressin, attenuated the development of all these lesions (Fig. 1).
Conclusions: Attenuations of histopathologic lesions and reducing VEGF expression with terlipressin suggested that these effects could be due to improved renal medullary blood flow and reversed radiocontrast-induced medullary hypoxemia via vasopressin-1 receptor agonist effect.

Su152 EXPRESSION OF KIDNEY INJURY AND DEGENERATION BIOMARKERS IN RESPONSE TO RAAS ACTIVATION BY LOW SALT DIET

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Introduction and Aims: The NT-CVD-Consortium (BMBF/NGFN-Transfer Alliance: New Tools for the prevention of Cardiovascular Diseases and Disorders in Chronic Kidney Disease) conducts preclinical and clinical studies for the identification and qualification of early and sensitive biomarkers that are correlated with pro-fibrotic or pro-inflammatory processes that ultimately result in tissue remodeling, loss of tissue function and end organ degeneration.
Methods: A rodent model (male Crl(Wi)BR-Wistar rats) was used to monitor the expression profiles of tissue injury and degeneration biomarkers responding to adaptive alterations in kidney and heart tissues under physiological changes of osmolality and endogenous aldosterone regulation by employing a diet containing a reduced sodium chloride content (0,02% w/w NaCl). Longitudinal gene expression profiles were determined by Affymetrix gene chip and RT-PCR analyses in renal cortex and myocardium during the first 10 days of dietary sodium chloride reduction versus control groups (0,2% w/w NaCl). In parallel, plasma level of aldosterone and markers of tissue remodeling and kidney injury marker were measured by ELISA.
Results: While a series of tissue degeneration markers were rapidly and transiently induced by the exposure to the low salt diet, mineralocorticoid receptor (MR) regulated genes and other genes were significantly induced only after 5 – 10 days of the low salt diet application. This finding corresponded to gradually increased plasma aldosterone levels which augmented the expression of PAI1, SGK1, GILZ, ZBTB16 and other MR regulated genes. Genes referred to as tissue injury or degeneration markers like pro-fibrotic osteopontin, tissue-PAI1, TIMP1, MMP9 or extracellular matrix components e.g. collagen-1A1 were also induced in renal cortex under this dietary change. Other markers indicating integrity of kidney tissue function like KIM-1 (HAVCR1), NGAL (LCN2), CDKN1A e.g. were transiently yet significantly induced.
Conclusions: Kidney injury and tissue remodeling markers are being induced rapidly in renal cortex even by moderate physiological changes in response to a diet of reduced sodium chloride content. The secreted kidney tissue remodeling biomarker candidates are also being analyzed in plasma and urine samples from patients with defined chronic or acute kidney dysfunction for further translational kidney injury biomarker qualification.
Disclosure: This abstract was possible thanks to a grant given by Bundesministerium für Bildung und Forschung / Nationales Genomforschungsnetz NGFN-Transfer to a Consortium of Bayer Schering Pharma, Charité, ExCorLab and University Hospital Essen.

Su153 EFFECTS OF cGMP PHOSPHODIESTERASE INHIBITOR ON EXPRESSION OF eNOS AND VEGF IN RATS WITH CYCLOSPORINE A INDUCED NEPHROTOXICITY

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Introduction and Aims: Cyclosporine-A (CsA) inhibits the differentiation of T cells by suppressing calcineurin and thus blocks the activation of the IL-2 promotor gene which prevents the rejection of transplanted kidney. However, long term CsA use must be limited because CsA induced nephrotoxicity develop. The mechanism of CsA induced nephrotoxicity have been suggested vasoconstriction due to the reduction of nitric oxide (NO), and tissue fibrosis caused by the elevation of TGF-beta, VEGF. The cyclic guanosine monophosphate (cGMP) phosphodiesterase (PDE)-5 inhibitor (Udenafil, Zydena®), active in the NO/cGMP pathway, with its physiological function induced by cGMP, has been reported to ameliorate renal injury by the increase of cGMP. In this study, in a rat model of CsA-induced nephrotoxicity, the administration of the PDE-5 inhibitor was studied to determine whether it ameliorated nephrotoxicity and altered the expression of eNOS and VEGF.
Methods: A right nephrectomy was performed in Sprague-Dawley rats (N=30, 200-250g, male). The Ischemia group (N=6), had clamping of the left renal pedicle for 45 minutes (IR) and was maintained for 28 days. The Udenafil group (N=6), after IR, was treated with 10 mg/kg udenafil orally for 28 days. The CsA group (N=6), after IR and then 15 mg/kg cyclosporine-A was injected subcutaneously for 28 days. The CsA with udenafil group (N=6), after IR received 15 mg/kg cyclosporine-A injected subcutaneously together with the oral administration of 10 mg/kg udenafil for 28 days. After the collection of blood on day 28, the remaining kidney was resected surgically and assessed by hematoxylin-eosin staining, immunohistochemical staining, western blot, real time-PCR was used to assess degree of renal injury and the expression of eNOS and VEGF.
Results: The comparison with the Ischemia group and the CsA group, after the administration of udenafil, showed that the creatinine was significantly decreased ($p=0.002$, $p=0.002$, respectively). The H&E staining, comparing the Ischemia group and the CsA group, showed that the level of loss of the nuclei in the proximal tubules was significantly decreased after the administration of udenafil ($p=0.004$, $p=0.002$, respectively). The immunohistochemical staining showed that eNOS stained strongly in the Udenafil group and the CsA with udenafil group compared to the other groups. the western blot of eNOS showed that it was not changed in the Ischemia group; however, the expression of the protein was decreased in the CsA group, and increased in the Udenafil group. For the western blot of VEGF, the expression of the protein was reduced only in the CsA with udenafil group. The eNOS mRNA assessed by real time-PCR was decreased in the CsA group compared to the other groups ($p=0.000$). VEGF mRNA was decreased in the CsA group, and after the administration of udenafil, it showed a tendency to decrease more ($p=0.003$, $p=0.005$, respectively).
Conclusions: cGMP PDE inhibitor ameliorated kidney injury in a rat model of CsA-induced nephrotoxicity. The mechanism appears to be associated with an increase of eNOS and reduction of VEGF.

Su154 AN IN VITRO SYSTEM WITH CULTURED TUBULAR EPITHELIAL CELLS IDENTIFIES ALPHA-ENOLASE AS A URINARY MARKER OF GENTAMICIN-INDUCED ACUTE KIDNEY INJURY

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Introduction and Aims: Nephrotoxicity is the main side effect of the aminoglicoside antibiotic gentamicin, which is mainly characterized by tubular damage. It is estimated that some degree of renal damage occurs in 10-25% of patients treated with this drug, a situation that is especially concerning in critically ill patients. An important determinant for the clinical handling of nephrotoxicity is an early detection. In this sense, it is necessary to identify new, sensitive markers for a useful and incipient diagnosis at the population level. For these purposes, urine is an ideal body sample. First, because it is easy to obtain in a non-traumatic manner; and second, because it is in direct contact with the tissues affected by nephrotoxins, and it is thus expected to contain pathophysiological information in the form of biomarkers. Beside filtered molecules (e.g. proteins) differentially handled by damaged tubuli, another prospective source of urinary markers is posed by renal cells injured or altered by gentamicin, most prominently epithelial tubular cells. In order to identify new markers of gentamicin's nephrotoxicity we established an in vitro system with cultured tubule cells in which to study the effect of gentamicin in the composition of the secretasome.

Methods: We used a human tubule cell line (HK2). Cells were treated with different concentrations of gentamicin (or vehicle as control) during 1-4 days. Proliferation/viability was assessed by the MTT method to check the level of cytotoxicity. After a selected time of treatment, a differential proteomic analysis of the culture media was done by means of two-dimensional (2D) gels followed by MALDI-TOF for protein identification in selected spots. Western blot analysis of culture media was used to further validate the presence of selected proteins. Furthermore, the presence of the proteins identified was studied in the urine of rats treated with gentamicin, whose renal function and morphology was assed by biochemical parameters (serum creatinine, BUN, creatinine clearance, proteinuria, NAG excretion), and histological studies and Western blot of renal and urinary markers of damage, respectively.
Results: Proteomic studies on culture medium from HK2 cells treated with gentamicin or vehicle show that alpha-enolase secretion is significantly enhanced by this antibiotic, even under a non-cytotoxic scenario. Western blot studies further confirmed the proteomic result. Interestingly, alpha-enolase also appeared increased in the urine of rats treated with gentamicin, which suffered an overt renal failure.
Conclusions: Our results indicate that this in vitro approach might be useful to identify novel urinary markers associated to drug nephrotoxicity, which originate in particular cell types subject to the influence of a determined drug under variable scenarios of toxicity, some of which might be related to early stages of nephrotoxicity. This system also offers ethical advantages that might help to reduce the use of experimental animals.

Su155 DENDRITIC CELL MODULATION PARTIALLY MEDIATES BENEFICIAL EFFECT OF MESENCHYMAL STEM CELL IN ISCHEMIC ACUTE KIDNEY INJURY

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Introduction and Aims: There is great promise in the application of mesenchymal stem cells (MSCs) for acute kidney injury (AKI). Recently, differentiation-independent mechanisms such as paracrine growth factors, immunomodulation were suggested to mediate the beneficial effect of MSC. Especially MSCs have shown anti-inflammatory action through tolerogenic dendritic cells (DCs) or $CD4^+$ $CD25^+$ Tregs in other injury models, but the mechanisms by which MSCs work in AKI, are not well understood yet. Therefore, the purpose of this study was to examine the beneficial effects of MSCs focusing on DCs or Tregs in ischemia/reperfusion induced AKI.
Methods: C57/BL6 mice underwent bilateral ischemia (32min). MSCs were administered before I/R injury. Twenty four hours after reperfusion, biochemical, histological kidney damage was assessed along with tissue cytokines and chemokine by cytometric bead array. Percent of DCs or Tregs, and their phenotype were examined in kidney and spleen by flow cytometry. We performed additional studies with CD11c-DTR mice in vivo, and MSC-splenocyte coculture was also performed with or without liposome clodrenate (LC) pretreatment to examine the role of DCs.
Results: Pretreatment with MSCs attenuated I/R induced kidney injury and also infiltration of inflammatory cells with simultaneous decrease in tissue proinflammatory cytokines. MSCs treatment reduced $CD11c^+$DCs and their expression of CD80 in spleen, while spleen $CD4^+FoxP3^+$ Tregs increased. Systemic depletion of $CD11c^+$DCs using CD11c-DTR mice resulted in partial loss of the beneficial effect by MSCs treatment. In vitro, MSCs also suppressed proliferation of splenocyte, while LC pretreatment partially reversed this effect.
Conclusions: These results suggest that beneficial effect of MSCs in I/R injury might be partially mediated by dendritic cell modulation or increased Treg and further studies identifying tolerance induction mechanisms might be useful in developing various strategies that ultimately improve prognosis of AKI.

Su156 VASCULAR AND BONE EFFECTS OF LANTHANUM CARBONATE IN UREMIC APOLIPOPROTEIN E DEFICIENT MICE

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Introduction and Aims: The rapid progression of atherosclerosis and vascular calcification in chronic kidney disease (CKD) is favored by the associated disorder of mineral and bone metabolism (MBD). The purpose of the study was to compare the effect of the phosphate binder lanthanum (La) carbonate with that of sevelamer-HCl on arterial calcification and atherosclerosis and also on bone structure and turnover in mice with chronic renal failure (CRF).
Methods: After surgical creation of CRF or sham operation, respectively, 12 week-old female apolipoprotein E deficient (apoE-/-) mice were randomized to one non CRF group and three CRF groups and fed for a duration of 8 weeks with standard diet (Harlan 2918) alone (non CRF and CRF group), or with same diet supplemented with either 3% lanthanum carbonate (La3%; CRF group) or 3% sevelamer-HCl (Sev3%; CRF group). At sacrifice, blood was sampled for serum biochemistry as well as proteomic analysis and aorta and femoral bones were removed.
Results: Compared with unsupplemented control CRF mouse groups, both the La3% and Sev3% supplemented CRF mice displayed a decrease of serum phosphorus, a reduction of arterial calcification at both intima (plaque) and media (non-plaque) of aortic root, and less atherosclerosis in thoracic aorta.

	Non CRF 2918 a	CRF 2918 b	CRF 2918 + La3 (%) c	CRF 2918 + Sev3 (%) d	p
Plaque calcification (%)	4.97±0.8	21.36±2.7 a	7.37±2.1 b	10.14±2.3 b	<0.001
Non-plaque calcification (%)	4.87±1.1	15.6±3.2 a	6.38±1.7 b	7.36±0.7 b	<0.001
Thoracic aorta plaque lesions (%)	0.70±0.1	2.52±0.3 a	0.99±0.3 b	0.88±0.2 b	<0.001
Collagen I expression in plaques (% of lesion surface)	7.5±2.8	21.9±6.1 a	6.8±1.4 b	6.04±1.9 b	< 0.01
Mice with positive nitrotyrosine staining (%)	10	80 a	50	30.8 b	< 0.01

There was a reduction of aortic collagen I expression with either phosphate binder, and in nitrotyrosine expression in response to sevelamer-HCl. Proteomic analysis showed that different detected peptide peaks were significantly modified by CRF state and at least one peak by Sev3% supplementation. Finally, mineral apposition and bone formation rates were increased in CRF mice compared with non CRF mice. This increase was corrected by Sev3%, but not La3%.
Conclusions: Both La carbonate and sevelamer-HCl retarded the progression of vascular calcification and atherosclerosis in uremic apoE-/- mice. These effects could be mainly due to the control of hyperphosphatemia by the two phosphate binders, and likewise the reduction of arterial collagen I expression. The effect of La carbonate differed however from that of sevelamer-HCl in that it did not appear to exert its beneficial vascular effects via changes in oxidative stress or bone remodeling in the present model.
Disclosure: This work was generously funded by a grant from Shire.

Su157 CONDITIONATED MEDIUM FROM MESENCHYMAL STEM CELLS IS RENOPROTECTIVE WITHOUT INDUCING IMMUNOMODULATION

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Introduction and Aims: Paracrine effects of Mesenchymal Stem Cells (MSC) are one possible mechanism that explains repair after MSC treatment in acute kidney injury. Recently, it was shown MSC secrete microvesicles

that help kidney repair. Here, we analyzed the the role of Conditionated Medium (CM) in repairing acute kidney injury.
Methods: MSC were isolated from femurs and tibias of Wistar rats, and characterized by FACS analysis and differentiation assays. The CT was obtained by serum depriving MSC cultures for 48h. Male Wistar were subjected to ischemia reperfusion injury, by clamping both pedicles for 60 min. After 6h of reperfusion, animals were treated with 10 mL of concentrated CM.
Results: Serum creatinine and urea analyses showed significant reduction in CM-treated animals, when compared to untreated animals (SCr IR = 3.28±0.8 mg/dl vs. IR+CM = 1.59±0.6 mg/dL). No differences were seen between CM and MSC treatment (IR+CM = 1.59±0.6 mg/dL vs. SCr = 1.29±0.29 mg/dL). NTA analysis also demonstrated that CM-treated animals had lower levels of necrosis and higher levels of regeneration. However, immunomodulation was not seen at kidney tissue. mRNA expression of IL-4, IL-1b and TNF-a demonstrated no differences between CM-treated and untreated animals. IL-10, an anti-inflammatory cytokine, was slightly increased in CM-treated animals, but no significant difference was observed.
Conclusions: The improvement seen on functional parameters correlates with proliferative and regenerative properties, suggesting a role for reduced apoptosis.

Su158 ACTIVATION OF NF-kB IN IMMORTALIZED HUMAN MESANGIAL CELLS (ihMCs) IS STIMULATED BY URIC ACID (UA): ROLE OF ANGIOTENSIN II (AII) AND ENDOTHELIN-1 (ET-1)

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Introduction and Aims: Hyperuricemia is associated with increases of cardiovascular risk and renal disease. Mesangial cells regulate glomerular filtration rates through the release of hormones and vasoactive substances mediated by transcription of proinflammatory genes such as NF-kB. This study evaluates the signaling pathway of UA in ihMCs.
Methods: IhMCs were treated with UA (10 mg/dl) for 8 and 24 h alone and simultaneously with losartan (10^{-7} M) or PD 123319 (10^{-7} M), AT1 and AT2 blockers, respectively. AII and ET-1 were assessed using the ELISA technique. Pre-pro endothelin and NF-kB mRNA were evaluated by the RT-PCR technique. Nitric oxide was measured by chemiluminescence technique.
Results: UA (10 mg/dl) for 8 and 24 hours significantly increased mRNA of NF-kB (0.28±0.09 to 7.56±2.68 arbitrary units) in 8 hours and (1.24±0.47 to 9.02±1.16 arbitrary units) in 24 hours. This effect was significantly reversed when ihMCs were pre-incubated with blockers of the AII receptors (Losartan: 9.02±1.16 to 0.84±0.44 arbitrary units) and (PD: 9.02±1.16 to 3.63±0.58 arbitrary units) in 24 hours. UA (10 mg/dl) for 24 significantly increased AII protein syntheses (0.19±0.01 to 0.24±0.01 pg/mL) compared to control situation. For 8 h, ET-1 expression and protein production was significantly increased (1.06±0.16 to 3.49±1.08 arbitrary units) and (56.72±5.23 to 70.80±7.28 pg/mL) respectively. Interestingly, after 24 hours pre-pro ET-1 mRNA expression was significantly decreased (1.15±0.28 to 0.15±0.02 arbitrary units) compared to control situation. This effect was significantly reversed when ihMCs were pre-incubated with blockers of the AII receptors (Losartan: 0.15±0.02 to 4.17±0.78 arbitrary units) and (PD: 0.15±0.02 to 3.30±0.72 arbitrary units) in 24 hour. Nitric Oxide did not undergo significant changes.
Conclusions: Our results suggested that UA triggers reactions including transcription NF-kB, increase and AII that induced the decrease in ET-1 production. The latter mechanism could be related the long term effects of UA on renal function and chronic kidney disease.
Disclosure: This work was aided by grants from Conselho Nacional de Desenvolvimento Científico Tecnológico (CNPq), Financiadora de Estudos e Projetos (FINEP), Fundação Oswaldo Ramos (FOR), Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP), Associação Beneficente de Coleta de Sangue (COLSAN) and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES).

Su159 APPLICATION OF NMR-BASED METABONOMICS IN THE STUDY OF RAT RENAL ISCHEMIC-REPERFUSION INJURY

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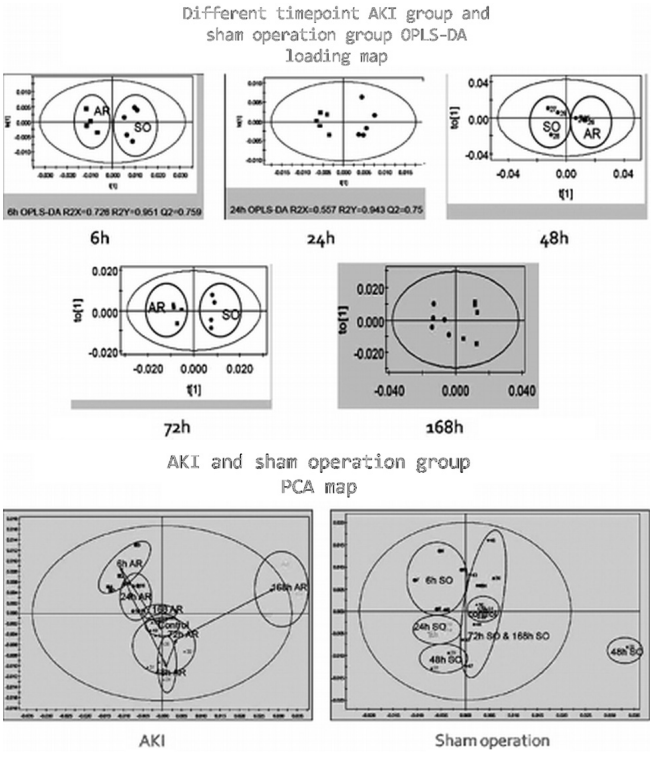
Introduction and Aims: The kidney is an organ with high metabolism and sensitive to ischemic injury. There is obvious change of renal metabolism after ischemic injury. Few studies has been carried on the metabolism change of ischemic/reperfusion kidney with NMR method. So we hope to estabolish the metabolic response to renal ischemic/reperfusion injury.
Methods: Male Sprague-Dawley rats (weight 200-230g) were subjected to either clamp of both sides of renal artery, sham operated (SO) surgery, or no treatment (n=5/group) and samples collected over 6 hours, 24 hours, 48 hours, 72 hours and 168 hours.We utilized ^{1}H NMR and ^{31}P NMR spectroscopy of kidney. And the morphological change of renal tissue were evaluated by PAS and HE staining and transmission electron microscope.
Results: A number of renal metabolic perturbations were observed in I/R rats compared with SO animals, including elevated isobutyrate, lactate, Sn-glycero-3-phosphocholine and hippurate while betaine, taurine, creatine and NADP were reduced.

The major change of metabolism product

Metabolites	24 hr
Isobutyrate	↑↑
3-hydrothybutyrate	↑↑
Lactate	↑↑
Creatine	↓↓
Sn-glycero-3-phosphocholine	↓↓
Taurine	↓↓
NAD+; NADP+	↓↓

↓↓ or ↑↑ means $p<0.001$ AKI vs sham operation.

Scores plot of pattern recognition analysis were capable of distinguishing I/R from SO.



The ATP level of renal tissue was decreased in the I/R group.The observed metabolic perturbations were consistent with the morphological change of kidney including the damaged membrane of renal tubular cell and the decreased density of mitochondria in tubular cells.
Conclusions: The observed changes in ^{1}H and ^{31}P NMR spectroscopy and morphology confirm the decreased energy metabolism and suggest the

protection of mitochondria may be a strategy for preventing and treatment of AKI.

Su160 FC RECEPTER GAMMA CHAIN DEFICIENCY IS RELATED TO THE DEVELOPMENT OF LIPOPROTEIN GLOMERULOPATHY

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Introduction and Aims: Lipoprotein glomerulopathy (LPG) is a unique renal disease characterized by intraglomerular lipoprotein thrombi. In human, several novel apolipoprotein E (apoE) mutations and the mutation-related dyslipidemia were identified in patients with LPG. On the other hand, Kanamaru et al. (J Am Soc Nephrol 13: p1527, 2002) reported LPG developed in chronic graft-versus-host disease (GVHD) induced Fc receptor gamma chain (FcRγ)-deficient mice. We attempted to clarify which has a principle role in development of LPG, dyslipidemia or FcRγ insufficiency.
Methods: Human apoE3 gene was introduced by adenovirus-mediated gene transfer into 20 week-old ApoE knockout mice (AKO), apoE and FcRγ double knockout mice (DKO) and wild type mice (WT), respectively. Blood sample were collected every other week and kidneys were dissected after 3 weeks of apoE3 introduction. Formalin-fixed specimens were followed by postfixation in osmium tetroxide for lipid fixation before embedding in paraffin. The sections were stained with oil red O. In addition, alcohol-fixed specimens were also embedding in paraffin, and were immuno-stained with CD68. High performance liquid chromatography (HPLC) was performed for analyzing lipoprotein profiles.
Results: In histlogical examination, LPG-like lesions were observed in DKO and AKO (Figure 1), and the glomerular lipoprotein thrombi positive rate was significantly higher in DKO than that of AKO (DKO 38.5±20.4%, AKO 2.0±2.0%). The number of CD68 positive cells in glomerulus were less in DKO than that of AKO (DKO 1.6±0.1, AKO 5.5±1.1). In serum lipid analysis, serum TC levels of AKO and DKO were significantly higher than that of WT at baseline, but there were no significant difference between AKO and DKO. Hypercholesterolemia of both mice were improved to normal level after apoE3 administration. While, serum TG levels of both mice revealed similarly transiently increase after viral transfection. In HPLC analysis of serum from mice after apoE3 introduction, elevation of TG-IDL fractions were observed in AKO and DKO, and showed similar pattern in both groups.

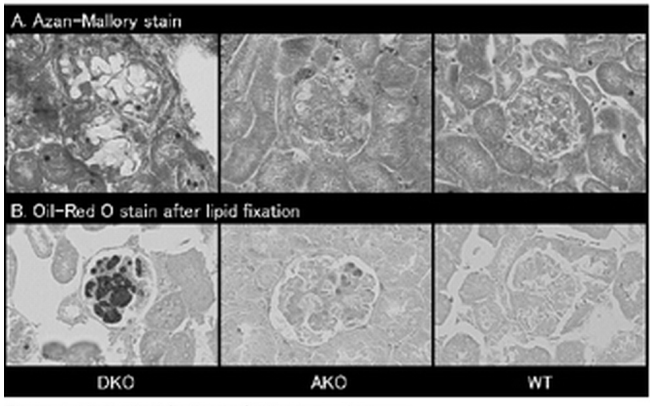

Figure 1. Histlogical findings of kidney specimen.

Conclusions: These results suggest that FcRγ impairment plays more important role than dyslipidemia for development of LPG, and the mechanism may be related to the function failure of macrophages.

Su161 ROLE OF MESENCHYMAL STEM CELLS IN FOLIC ACID-INDUCED ACUTE KIDNEY INJURY

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Introduction and Aims: Despite recent advances in treatment, acute kidney injury (AKI) continues to be responsible for a high mortality rate among hospitalized patients. As so, mesenchymal stem cells (MSC) have been considered in literature as a promising therapy for AKI due to its immunomodulatory potential.
For this study, we analyzed the role of these cells in AKI induced by high doses of folic acid (FA).
Methods: MSC were obtained from male C57Bl/6 mice (6 weeks) bone marrow and cell population was expanded in Mesencult cell media® (Stem Cell Technologies). Before administration of these cells, MSC were characterized by immunophenotyping for CD34, CD45, CD73 and CD105, and also by differentiation assays. Experimental model was achieved by administration of FA (200mg/kg body mass) and animals were sacrificed after 48h. After 24h of FA administration, 5.10^5 MSC were administrated for each animal intraperitoneally. Serum and kidney tissue samples were collected 48h after FA treatment for kidney function evaluation, morphologic study and PCR analysis (TNF-a, IL-1b, IL-6).
Results: MSC-treated animals demonstrated reduced serum creatinine and urea levels that correlated with acute tubular necrosis (ATN). This functional amelioration was accompanied by reduction of IL-6 (MSC-treated animals 2.45±1.88 vs. untreated animal 27.30±10.24), IL-1b (MSC treated animals 0.64±0.50 vs. untreated animal 2.62±1.14) and TNF-a (MSC treated animals 1.75±0.27 vs. untreated animal 8.45±0.25) mRNA expression at kidney tissue in MSC-treated animals when compared to untreated animals. Immunohistochemistry analysis also showed that MSC–treated animals had increased cell proliferation by PCNA staining.
Conclusions: This study demonstrated that MSC immunomodulation and proliferative potential correlate with functional improvement in AKI achieved by AF. Support: FAPESP, Complex Fluids INCT and CNPq.

Su162 β2-ADRENOCEPTOR MODULATES THE ORGAN RESPONSE TO RENAL INFECTION INDUCED BY *ESCHERICHIA COLI*

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Introduction and Aims: Renal infections elevate the risk of sepsis and are important causes of septic shock and multiple organ failure. The objective of the present study was to test the hypothesis that functional β_2-adrenoceptor (β_2-AR) activation modulates the organ response to renal infection induced by *Escherichia coli* (*E.coli*) administration.
Methods: Four-week-old Wistar rats pre-treated with β_2-AR agonist, terbutaline (27.4 ng/kg) and/or the β_2-AR antagonist, ICI118,551 (3.14 μg/kg) were injected into the right kidney with a 0.05ml sample of 4.7×10^9 CFU of *E. coli* per kg of body weight or with an equal volume of saline, and then 1, 2, or 3 days later, blood and kidneys were collected to assay cytokines, renal function and the β_2-AR signaling system.
Results: The rat renal infection model pre-treated with β_2-AR agonist had no effect on an elevation in interleukin-6 (IL-6), Growth-Related Oncogene/keratinocyte-derived cytokine (GRO/KC), and Granulocyte-macrophage colony-stimulating factor (GM-CSF) in the right kidney, and serum levels of their respective proteins and nitric oxide (NO), whereas *E-coli*-induced renal tumour necrosis factor-α (TNF-α) production was suppressed and creatinine clearance rate (Ccr) was maintained over the course of the infection. Conversely, treatment of the rat model with the β_2-AR antagonist resulted in a decrease of Ccr and serum NO and greater increases in serum TNF-α associated with an elevation of the right renal TNF-α and a reduction of the right renal Gsα and cAMP levels. However, the increases in *E.coli*-induced renal GM-CSF, GRO/KC and IL-6 expressions were not changed by pre-treatment of the β_2-AR antagonist. On the other hand, the

inhibition of β_2-AR activation impaired the clearance of endotoxin from the kidney and was associated with a raised mortality rate.
Conclusions: The renal β_2-AR activation modulates TNF-α responses in the infected kidney, changes serum levels of TNF-α and NO. Importantly, the blockade of a renal β_2-AR signaling cascade leads to renal dysfunction and a higher rate of mortality.

Su163 SEPSIS-INDUCED ACUTE KIDNEY INJURY AND β_2-ADRENOCEPTOR GENE THERAPY

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Introduction and Aims: Septic shock following surgery, trauma, burns, or severe infection is a common cause of acute kidney injury (AKI), resulting in a high mortality rate. Recently, we reported in a rat model that an injection of adenoviral construct containing the human β_2-adrenoceptor gene (adeno-β_2-AR gene) prevented the progressive renal inflammation associated with Gram-negative sepsis. The present study was undertaken to explore whether the utilization of adeno-β_2-AR gene after the onset of sepsis could potentiate the β_2-AR signaling systems in the kidney and provide the kidney with protection against *Escherichia coli* (E.coli)-induced AKI.
Methods: An endotoxemic rat model of AKI was induced by renal artery occlusion plus subcutaneous injections of E.coli in four-week-old Wistar rats and was then injected intraperitoneally with the adeno- β_2-AR gene.
Results: The β_2-AR gene delivery resulted in a recovery of glomerular filtration rate and the renal β_2-AR signaling system which were depressed 24 hours after the onset of sepsis. Moreover, while the adeno-β_2-AR gene delivery had no effect on a recovery of cytokines and C-reactive protein in the systemic circulation, except for nitric oxide (NO) and angiotensin II (Ang II), the addition of the β_2-AR gene significantly depressed ($P<0.01$) the expression of renal cannabinoid-1 (CB-1) receptor, CD14, toll-like receptor 4 (TLR-4), and tumor necrosis factor (TNF)-α gene. Furthermore, the β_2-AR gene transfer also improved the survival of the rats exposed to sepsis-induced AKI.
Conclusions: A renal-specific over-expression of β_2-AR resulting from the gene delivery appeared to modulate the renal dysfunction and inflammation associated with sepsis through the cAMP-PKA, CB-1, and CD-14-TLR-4 – TNF-α pathways in the kidney. Moreover, the defense mechanisms following β_2-AR activation by the gene delivery included the modulation of NO and Ang II in the systemic circulation. Adeno-β_2-AR gene administration has the potential of being used as a therapeutic agent for AKI following the onset of Gram-negative sepsis.

Su164 INDOXYL SULFATE STIMULATES REACTIVE OXYGEN SPECIES LEAKAGE FROM RAT KIDNEY MITOCHONDRIA AND AST-120 INHIBITS THIS PROCESS IN EXPERIMENTAL ACUTE KIDNEY INJURY

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Introduction and Aims: Free radicals both initiate and contribute to the progress of AKI but there is no direct evidence of free radical production in the injured cells. We have succeeded to identifying free radical leakage (O2- and OH radicals; ROS) from mitochondria (Mito) directly using an EPR spin trapping technique. Indoxyl sulfate (IS) is known one of the uremic toxins and inhibits kidney SOD activities both normal and CKD model rats (Am J Nephrol 2008:28; 446). To clarify the role of Mito stress on development of AKI, we have investigated ROS leakage from Mito and immunostaining of survivin, an inhibitor of apoptosis protein, in the kidney using AKI model rats.
Methods: Ischemic AKI was induced in 5 SD male rats and divided into the following two groups: AKI control (C) and AST-120 treated (K). Sham operated rats were used as a normal group (N). After 24 hours induction, kidneys were isolated and Mito fractions were prepared by fractional centrifugation. Serum and intra cellular IS levels were measured. Free radicals were detected by the EPR spin trapping method using CYPMPO as a spin trapping agent. ROS adducts were recorded every 1 min for 20 min. Peak height (PH) of ROS were accumulated and leakage rates (PH/min) were calculated. SOD activities (U/mg protein) in the Mito and survivin expression in the kidney were measured.
Results: Levels of serum Cr and IS were significantly lower in the K group compared to the C group. ROS leakage rates were significantly higher in the C group and lower in the K group (N=352±34, C=440±64, K=327±25). Mito SOD activities were lower in the C group (N=9.7±2.4, C=3.1±0.6, K=6.0±28). Intracellular IS levels were higher and survivin area (%) were lower in the C group (IS;N= 16.2±2.0, C=36.8±6.8,K=12.0±9.3, Survivin; N=ND, C=0.56±0.12, K=0.89±0.06). There was significant positive correlations revealed in the ROS leakage and intracellular IS levels, and negative correlations between ROS and survivin.
Conclusions: Mito functions were maintained and apoptosis was inhibited by AST-120.

Su165 ELEVATED FIBROBLAST GROWTH FACTOR 23 EXPRESSION LOCALIZES TO METAPHYSEAL OSTEOBLASTS IN AN EARLY MODEL OF CHRONIC KIDNEY DISEASE WITH SECONDARY HYPERPARATHYROIDISM AND IS LOST AFTER CALCIMIMETIC R-568 TREATMENT

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Introduction and Aims: Renal osteodystrophy (ROD) is a complication in chronic kidney disease-mineral and bone disorder (CKD-MBD) patients with secondary hyperparathyroidism (sHPT). FGF23 is a bone-derived phosphate regulating hormone whose possible role and gene regulation in ROD bone is unknown. Recently, we showed that two week calcimimetic treatment in an early CKD rodent model with sHPT suppressed serum PTH and FGF23 levels and bone FGF23 mRNA (by ribonuclease protection analysis, RPA). This study examined the sites of expression of FGF23 mRNA by in situ hybridization (ISH) in subtotal 5/6 nephrectomized (Nx) rats.
Methods: Rats were placed on a high phosphate diet (HPD 1.2%P) immediately after 5/6 Nx surgery. Three weeks post-Nx, animals were given p.o. vehicle (Nx-veh) or 30 mg/kg R-568 (Nx-R-568) daily for 14 days (n=10/group). Vehicle-treated sham-operated (sham-veh) rats served as controls. Blood was drawn before and 4 hours post-dose on days 0, 7 and 14. On day 14, femurs were harvested, processed for histology; and ISH (n=3-7/group) and histomorphometry (n=10-11) performed.
Results: Nx-veh rats demonstrated elevated serum BUN, creatinine, iPTH, and FGF23; increased osteoid surface (OS/BS%), 32.6±11.9 vs 12.5±5.1 ($P<0001$), higher bone formation rate (BFR, mm^3/mm^2/d), 0.76±0.17 vs 0.45±0.16 ($P<0.01$), and osteoblast surface (Ob.S/BS%), 32.9±12.1 vs 12.7±4.7 ($P<0001$) as compared with sham-veh. ISH localized FGF23 mRNA to growth plate chondrocytes and osteoblast-like cells near the growth plate in normal, sham-veh and Nx-veh. One sham-veh rat expressed low levels of FGF23 transcripts in eosinophils. In addition, all Nx-veh rats expressed FGF23 mRNA in osteoblast-like cells lining metaphyseal trabecular bone (3/3) (not seen in normal or sham-veh rats); and, higher eosinophil FGF23 transcript levels compared with sham-veh rats. Interestingly, FGF23 transcripts were not detected in osteocytes of any group in this early CKD rat model, whereas sclerostin expression was readily detectable in control osteocytes. After 14 day calcimimetic treatment, FGF23 transcripts were not detected in Nx-R568 rat bones. Failure to detect FGF23 transcripts paralleled calcimimetic-induced declines in serum PTH and FGF23, whereas histomorphometric high bone turnover, seen in Nx-veh rats, persisted in Nx-R568 rats after 14 day calcimimetic treatment.
Conclusions: Elevated FGF23 transcript levels in trabecular osteoblasts and eosinophils of CKD-MBD rats were reduced to undetectable levels after 14 day calcimimetic treatment without significant static or dynamic histomorphometric changes. Whether calcimimetics suppress FGF23 expression in bone by acting indirectly (eg. via PTH lowering) or directly on bone

cells is not known. More studies are needed to define precise mechanisms associated with these changes.
Disclosure: All authors are full time employees of Amgen Inc.

Su166 CONDITIONAL KNOCKOUT OF THE VON-HIPPEL-LINDAU PROTEIN PROTECTS FROM ACUTE KIDNEY INJURY

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Introduction and Aims: We hypothesize that silencing of the Von-Hippel-Lindau (VHL) protein confers protection against acute kidney injury (AKI), since VHL-deficient cells exhibit normoxic activation of hypoxia-inducible factors (HIF), which promote hypoxia adaptation.
Methods: We generated a new mouse strain (Pax8-VHL-KO) with conditional knockout of VHL in all nephron segments, as well as in roughly 20% of hepatocytes. First, KO mice were characterized in terms of renal function, morphology, capillary density (immunohistochemistry for caveolin-1), activation of HIF (immunohistochemistry) and of HIF target genes (immunohistochemistry, rtPCR, in situ hybridization). Second, we subjected KO and wildtype mice to rhabdomyolysis-induced AKI, which recently was demonstrated to cause hypoxic damage to proximal tubules. Tubular injury, renal function, HIF activation, and HIF target genes were compared.
Results: Transgenic animals showed robust activation of both HIF-1α and -2α in all nephron segments, as well as in roughly 20% of hepatocytes. VHL silencing induced up-regulation of renal VEGF mRNA, up-regulation of EPO mRNA in hepatocytes, down-regulation of renal EPO mRNA, renal angiogenesis, and polyglobulia. The latter was subsequently corrected by repeated blood collections, in order to exclude secondary changes. Histology of kidneys and liver, as well as renal functional tests were unremarkable until 9 mo, the end of the observation period. Especially, no tubulointerstitial fibrosis and no cysts were apparent. At 24 h after AKI Pax8-VHL-KO kidneys had persistently enhanced HIF-1α and activated cell protective HIF target genes in proximal tubules, namely heme oxygenase-1, glucose transporter-1 and carbonic anhydrase-IX. At 48 h after injury Pax8-VGL-KO mice had better renal function and less proximal tubular damage, including necrosis.
Conclusions: We conclude that 1) in vivo VHL-KO activates both HIF-1α and -2α, 2) VHL-KO ameliorates AKI associated with tubular hypoxia, potentially through tubular survival genes and/or renal angiogenesis.

Su167 ASYMMETRIC DIMETHYLARGININE LEVEL VARIES CONSIDERABLY IN URINE BUT NOT IN BLOOD DURING ARF OF RAT

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Introduction and Aims: Asymetric dimethylarginine (ADMA) is a strong endogenous regulator of NO synthesis and one of the most intriguing risk markers for both cardiovascular (CV) complications and renal disease progression (Kielsteien, Zoccali, 2007). Its isomer symmetric dimethylarginine (SDMA) has been shown to be an excellent marker of renal function. It was shown that chronic renal failure is accompanied with some increase of ADMA level and much more pronounced accumulation of SDMA in the blood. Only poor information exists about the methylarginines in acute renal failure (ARF). The aim of this study was to observe the changes of arginine, ADMA and SDMA in blood and urine during glycerol induced ARF of rats.
Methods: Wistar rats (n=18, 250±10g) were placed in metabolic cages 72h prior to the study. Water and chow were given *ad libitum*. ARF was induced by intramuscular injection 10ml/kg of 50% glycerol (or saline in control group) bilaterally into hind limbs after 24h water deprivation. Rats got free access to water immediately after injection. Rat urine and blood were analyzed on 1st and 3rd days after glycerol injection. The concentration of arginine, ADMA and SDMA were measured by HPLC with fluorescence detector after solid phase extraction according to Teerlink method (2002, 2007). Concentrations of creatinine and urea were determined as well.
Results: 24h after glycerol injection clear oliguria was observed accompanied with the rise of plasma creatinine (+500%, t=8.03, p<0.001) and urea (+700%, t=7.75, p<0.001) levels and decrease of their urinary excretion. GFR decreased from 1.26±0.08 to 0.04±0.01ml/min, t=-15,18, p<0,001. Urine ADMA level was extremely low in control animals, so it was not detected at any time of the experiment. However, at the first day of ARF ADMA has appeared in urine (0.7±0.15μmol/l). This day concentration of SDMA in urine did not differ from baseline (8.64±2.59 vs. 8.24±2.6μmol/l). Plasma concentration of ADMA did not differ as well from control value (0.78±0.07 vs. 0.68±0.05μmol/l). In contrast plasma SDMA concentration was found significantly higher then control one (1.62±0.12 vs. 0.26±0.03μmol/l, t=6,51, p<0.001). No change of arginine was found either in urine or in blood. 72 hr after glycerol injection poliuria has been registered with almost two times increased urine volume as compared to control. GFR that time was 0.18±0.08ml/min. Additional increase of urine concentration of ADMA (2.03±0.31μmol/l) was observed. Thus ADMA excretion achieved values that were close to SDMA order. No significant changes were observed in plasma ADMA (0,59±0,16μmol/l) and SDMA (1,82±0,24μmol/l) levels as compared to the 1st day of ARF.
Conclusions: It was found in this experiment, that no ADMA excretion exists in normal intact rat. The excretion appeared in ARF state and increased from oliguria to poliuria. This excretion did not influence on blood ADMA level. In contrast, decreased SDMA excretion raised plasma level of SDMA. The reason of such difference is of great interest in respect of mechanisms of isomers processing in kidney. General effects of glycerol ARF described in literature (Ayer et al., 1971) can not explain such phenomenon.
Disclosure: The work was supported by RFBR grant 08-04-00951a.

Su168 ENALAPRIL REDUCES TOLL-LIKE RECEPTOR (TLR) 2 AND TLR4 EXPRESSIONS IN EXPERIMENTAL UNILATERAL URETERAL OBSTRUCTION

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Introduction and Aims: The toll-like receptor (TLR) family serves an important regulatory role in the innate immune system, and recent evidence has implicated TLR signaling in the pro-inflammatory response of a variety of endogenous and exogenous stimuli including experimental unilateral ureteral obstruction (UUO). Activation of TLR2by angiotensin II is reported recently. We examined whether enalapril as an inhibitor of renin-angiotensin system modulates the renal expression of TLR2, TRL4 and its ligands and attenuates renal injury process of experimental UUO in mice.
Methods: Female C57BL/6 mice were divided into 4 groups; sham group, enalapril treated sharm group, control UUO group, and enalapril treated UUO group. We started to administer enalapril via drinking water (100mg/L)1 day before UUO surgery and continued until harvest of kidneys 5 days after surgery. We evaluated the levels of renal mRNA expressions of TLR2, TLR4, Myd88, TNF-α, MCP-1,TGF-β,and α-SMA, by real-time RT-PCR. We also evaluated renal TGF-β and α-SMA protein expressions by immunohistochemistry.
Results: The levels of mRNA expression of TLR2, TLR4 and Myd88 in UUO kidneys were significantly increased compared to sham operated group (all, p <0.05). The mRNA levels of TLR2, TLR4 and Myd88 of enalapril treated UUO group were significantly lower than those of control UUO group (all, p <0.05). The renal mRNA expressions of TNF-α, MCP-1, TGF-β, and α-SMA in enalapril treated UUO group were significantly lower than those of control UUO group (all, p <0.05). Enalapril also significantly reduced the immunostained area of TGF-β and α-SMA in UUO kidneys (p <0.05).
Conclusions: The results of present study suggest that enalapril attenuates renal injury process of experimental UUO in mice and at least in part, suppression of TLR2 and TLR4 may be involved in this mechanism.

Su169 SEX-SPECIFIC CHANGES OF THE RENAL CYP-EICOSANOID PROFILE LEAD TO GENDER DIMORPHISM IN EXPERIMENTAL ACUTE KIDNEY INJURY

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Introduction and Aims: Sexual dimorphism in renal injury is evident in clinical and experimental settings. Incidence of perioperative acute renal failure is lower in women, with the exception of cardiac surgery. In renal transplantation female recipients have a better graft and patient survival compared with males. Similar findings were obtained in animal models for renal ischemia. However, the mechanisms of sex related differences remain undefined. In a previous study we found that inhibition of the generation and action of the arachidonic acid (AA) metabolite 20-HETE conferred sex-specific protection in experimental acute kidney injury of male rats.
We hypothesized that females are protected against acute ischemic injury by changes in cytochrome P450 (CYP450) dependent AA metabolism resulting in a shift towards higher levels of vasodilatory and cytoprotective EETs.
Methods: Renal injury was induced in uninephrectomized male and female rats through 45 minutes of left renal artery clamping. We included additional groups treated 5 minutes prior to ischemia with intrarenal injection of the specific EET-antagonist 14,15-EEZE. Organs were harvested 2 days after reperfusion. Vehicle-treated animals served as controls. In further experiments blood and organ samples were taken immediately after warm ischemia and 2 hours post reperfusion to determine changes in the CYP-eicosanoid profile with liquid chromatography-mass spectrometry (LC-MS/MS).
Results: Female rats developed less extensive functional and structural impairment and postischemic inflammatory response than male rats. 14,15-EEZE administration in female rats reversed the protective phenotype and caused a similar loss of renal function, tissue inflammation and tubular apoptosis as in male rats. Preliminary data for tissue levels of AA metabolites indicate that 45 min of warm ischemia induce a massive release of 20-HETE in both sexes accompanied by a moderate release of EETs that are rapidly hydrolyzed by the soluble epoxid hydrolase (sEH) to DHETs in males.
Conclusions: Our results suggest that a distinct, time dependent pattern of changes in the renal CYP-eicosanoid profile is causally responsible for sexual dimorphism during acute kidney injury. Combined utility of 20-HETE-antagonists, EET-agonists, and sEH-inhibitors may offer new sex-adapted therapeutic strategies for ischemic acute renal failure.

Su170 EFFECTS OF IV IRON PRODUCTS ON PATHOGENESIS OF PULMONARY EDEMA

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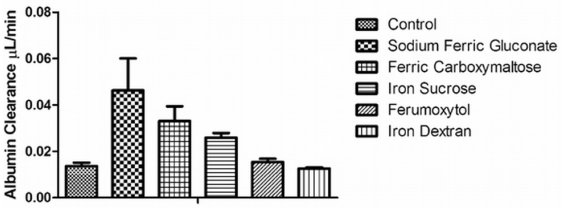
Introduction and Aims: Fluid retention and pulmonary edema are major complications associated with significant morbidity, mortality and hospitalization costs in maintenance HD patients. IV iron induces oxidative stress and may affect intracellular reactive oxygen species (ROS) generation, but is unknown if this affects lung permeability.
Methods: A lung permeability model was used in the 1st experiment. Rat lung microvessel endothelial cell (RLMEC) monolayers were cultured and treated with iron sucrose (IS), sodium ferric gluconate (SFG), ferumoxytol (FMX), low MW iron dextran (ID) and ferric carboxymaltose (FCM) at 0.05 mg/mL for 24 hours and permeability was determined by clearance rate of Evans Blue-labeled albumin between the luminal and abluminal compartments between 10 and 60 mins. Experiments were conducted in quadruplicate. ROS were quantitated in a 2nd experiment. RLMEC treated with iron products (0.05 mg/mL) for 24 hours were incubated with dihydroethidium (a fluorescent probe for ROS) (10 μM, 0.5 h at 37°C). Sonicated cell suspensions were added to microplates (100 μl/well) in quadruplicate. Fluorescence was measured using excitation and emission wavelengths of 490 and 605 nm.
Results: Results: Incubation of RLMECM with SFG, IS and FCM induced greater endothelial barrier dysfunction compared to controls, FMX and ID ($p<0.05$ for all comparisons, Figure 1)



Endothelial permeability after treatment with FMX and ID was not significantly different from control. All IV iron products induced ROS production, with 14, 22, 44 and 85% increases in detected O_2^- for SFG, ID, FMX and IS, respectively vs. untreated controls ($p<0.05$ for FMX and IS vs. controls).
Conclusions: Albumin clearance indicated highest RLMEC permeability for SFG, IS and FCM. All IV iron products induced ROS production. More data are needed to explore intracellular free iron generation and disruption of intracellular signaling. These data indicate that IV iron produces oxidative stress leading to potential for lung injury and adverse clinical outcomes.

Su171 CAFFEIC ACID PHENETHYL ESTER PROTECTS AGAINST AMPHOTERICIN B INDUCED NEPHROTOXICITY IN RAT MODEL

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Introduction and Aims: The present study was conducted to investigate whether caffeic acid phenethyl ester (CAPE) has a protective effect on Amphotericin B induced nephrotoxicity in rat model.
Methods: Thirty-two Wistar-Albino rats were randomly divided into four groups: (I) control group (n = 9) received 10ml/kg/day normal saline intraperitoneally (i.p.); (II) CAPE group (n = 9) received 10 μmol/kg CAPE via i.p. route; (III) Amphotericin B group (n = 7) received 10ml/kg/day normal saline i.p. during study and 50 mg/kg Amphotericin B once on the second day and (IV) Amphotericin B plus CAPE group (n = 7) received 10 μmol/kg CAPE i.p. during study and 50 mg/kg Amphotericin B once on the

Abstract Su171 – Table 1

	Urea (mg/dl)	Creatinin (mg/dl)	MDA (nmol/g protein)	SOD (U/g protein)	KATALAZ k/g protein	NO (nmol/g protein)	Histopathological damage (grade)
I - Control group	21.4±3.4	0.58±0.10	5.10±0.82	0.054±0.008	1.29±0.14	0.21±0.03	0 (0-0)
II - CAPE group	18.7±2.6	0.51±0.03	5.34±0.81	0.052±0.007	1.30±0.16	0.20±0.01	0 (0-1)
III - Amphotericin B group	73.5±39.2	1.61±1.13	6.64±1.38	0.085±0.007	0.84±0.15	0.29±0.04	3 (2-3)
IV - Amphotericin B + CAPE group	88.5±29.1	1.54±0.74	5.55±0.81	0.063±0.006	1.13±0.23	0.23±0.03	1 (1-3)
P values							
I-II	0.80	0.81	0.59	0.50	0.96	0.33	0.66
I-III	0.0001	0.002	0.003	0.0001	0.0001	0.0001	0.0001
I-IV	0.0001	0.004	0.34	0.02	0.06	0.18	0.0002
II-III	0.0001	0.001	0.01	0.0001	0.0001	0.0001	0.0006
II-IV	0.0001	0.003	0.66	0.006	0.06	0.03	0.001
III-IV	0.22	0.83	0.04	0.0001	0.005	0.002	0.0002

second day. Kidneys were used for histopathological examination and the measurements of malondialdehyde (MDA) and nitric oxide (NO) levels and enzyme activities including catalase (CAT), superoxide dismutase (SOD).
Results: The activity of SOD, MDA and NO levels were increased and catalase activity decreased in amphotericin B group compared to control group. However, administration of CAPE before amphotericin B decreased the activity of SOD, MDA and NO levels whereas increased catalase enzyme activity. Histopatological damage was prominent in group III compared to group.
Conclusions: CAPE treatment seems to be effective adjuvant agent for the prevention of amphotericin B nephrotoxicity in rat model.

Acute kidney injury – clinical studies 2

Su172 STATINS PROTECT KIDNEY IN PATIENTS UNDERGOING CARDIAC SURGERY IN CARDIOPULMONARY BYPASS

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Introduction and Aims: Statins, administered before cardiac surgery, have been demonstrated to reduce perioperative mortality, stroke and atrial fibrillation. Preoperative statin therapy also reduces the systemic inflammatory response associated with cardiopulmonary bypass. However, no data are available about the potential protective effect on renal function in cardiosurgery. We have performed a prospective observational study to evaluate the possible role of statins on perioperative kidney function in patients undergoing major cardiac surgery.
Methods: All patients referred to our cardiosurgery unit between January 2009 and January 2010, to undergo major cardiac surgery, were included in the study.
Renal function was evaluated by MDRD formula before surgery, at the end of surgery, after 24 hours, after 48 hours, at the discharge.
Two-ways ANOVA for repeated measures was used to compare the two groups. The administration of statins was used as independent variable.
Comparisons were considered significant if $p<0,05$.
Results: We included 72 patients, 41 of them on statin regimen. Basal and same intra-operative characteristics are reported in table 1.

Table 1. Basal and same intra-operative characteristics of patients

	Statin	No statin	P
Age years	64±9	64,9±10,1	>0,05
Hypertension n (%)	22 (53,7)	27 (54,8)	>0,05
Diabetes n (%)	21 (51,2)	12 (38,7)	>0,05
Euroscore	4,6±2,39	4,3±3	>0,05
Ejection Fraction %	47,68±8,89	48,1±9,2	>0,05
Extracorporeal circulation time min	98,8±31,6	105±37,64	>0,05
Cross clamp time min	61,9±26,08	64,74±23,8	>0,05
Basal creatinine clearance ml/min	92,68±26	87±24,96	>0,05

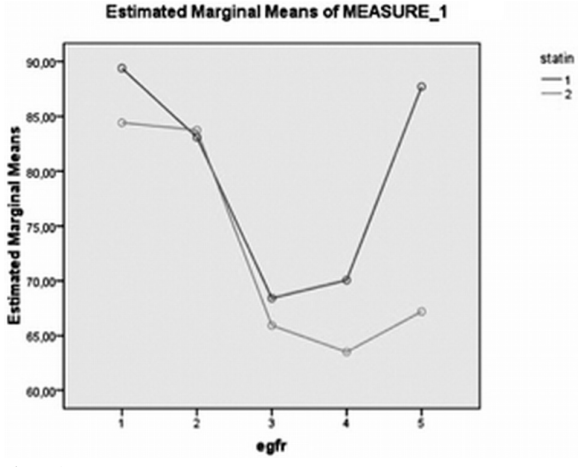

Figure 1

During the perioperative period, creatinine clearance presented a significant reduction from basal value after the end of cardiac surgery, at 24 hours, at 48 hours without difference in both groups, but at the time of discharge, patients that assumed a statin, presented a significantly higher glomerular filtrate rate ($p<0,05$) (Figure 1).
Only two patients died, one in every group.
Conclusions: Our data show that statins have a protective effect on renal function after cardiac surgery probably because their effects on systemic inflammatory status and endothelial dysfunction.
Other prospective randomized studies are necessary to verify these results.

Su173 SURVIVAL AND RENAL FATE AFTER SEVERE ACUTE ON CHRONIC RENAL FAILURE IN CRITICALLY ILL PATIENTS TREATED WITH SLED

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Introduction and Aims: Pre-existing chronic kidney disease in critically ill patients with acute kidney injury (AKI) has been attributed as favorable towards survival but as a risk factor for dialysis dependency at hospital discharge.This effect was thought to be related to an earlier referral into renal care in these patients (Khosla et al. (PICARD), JASN 2009). However, data on this specific group of patients are rare and need to be expanded. Our study aimed to contribute descriptive data on the outcome and renal recovery of critically ill patients in need for renal replacement therapy suffering from acute on chronic renal failure.
Methods: We performed a single-center prospective observational study from 2003 to 2006.All patients with acute on chronic renal failure (AoC-RF), treated by SLED on one of the 8 Intensive Care Units of the Medical School Hannover were eligible for enrolment. Severity of illness, hospital survival, length of stay and renal recovery were monitored, added by several physiological and laboratory variables. Data from patients with dialysis dependent acute kidney injury without pre-existing kidney disease were used as control. Statistical testing utilized t-test, ANOVA and bivariate Pearson correlation (significance: $p<0.05$).
Results: Participants: N=572 patients with AoC-RF vs. n=157 with AKI. Baseline demographics: AoC-RF patients were older (57 vs. 50,7y, $p<0.001$) and thinner (BMI 22.4 vs. 25.3, $p<0.001$). Gender distribution was equal, with males dominating either group. Baseline kidney function (calculated as eGFR from the lowest available creatinine during the hospital stay before initiation of renal replacement therapy (RRT)) was significantly impaired in AoC-RF (132 µmol/l [CKD III°] vs. 87 µmol/l [normal]). Severity of disease was equal at initiation of RRT (SOFA-score 14 vs. 14.1, n.s.). Modality of RRT was equal. Outcome: In-hospital death was more frequent in the AoC-RF group than in the AKI-group (59% vs. 51%, $p<0.001$). Renal recovery (no need for further RRT at hospital discharge) was diminished after AoC-RF compared to AKI (81% vs. 97% of survivors, $p<0.001$). Variables that significantly discriminated survivors from non-survivors in the AoC-RF group at initiation of RRT were: Presence of severe sepsis/SIRS/MODS, malignoma, heart- or lung-Tx, immunosuppressive therapy, low MAD, low platelets, higher creatinine, anuria, increased CRP, lactate, bilirubin and SOFA, lower Horowitz-score and need for respiratory assist. S-creatinine passed as an independent predictor of survival ($p=0.047$).
Conclusions: Our findings in part contrast to recent reports from the PICARD group: The data demonstrate, that acute on chronic renal failure is attributable to an even further enhanced risk of mortality and decreased renal recovery in critical ill patients during the length of the hospital stay.
Perspective: Further analysis by multivariate regression analysis will be used to identify independent risk factors.

Su174 ACUTE KIDNEY INJURY IN CHILDREN AFTER CONGENITAL HEART DISEASE SURGERY

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Introduction and Aims: The Acute Kidney Injury Network (AKIN) criteria are a new staging system for acute kidney injury (AKI). However, data about its use in children are sparse. We aimed to evaluate the AKIN criteria and mortality in children after congenital heart disease surgery.
Methods: We evaluated 130 patients admitted to the Intensive Care Unit after congenital heart.Eighty three patients were male (63.8%) and mean age was 451,7±665 days; seventy (53%) patients were less than one year of age.
Results: According to the AKIN criteria, AKI occurred in 33,8% of all patients (44). Of these, 61,4% (27 patients) met AKIN stage 1 criteria, 18,1% (8 patients) AKIN stage 2 and 20,5% (9 patients) stage 3. Four patients was submitted to renal replacement therapy. Overall hospital mortality was 5% (8 patients) and stratification by AKIN category was 37.5% (3 patients) for stage 1 (p=0.023), 12.5% (1 patient) for stage 2 and 12.5% (1 patient) for stage 3. Using logistic regression, significant independent risk factors for AKI were surgery duration (each minute of surgery increase AKI odds ratio 1.01, 95% confidence interval 1.0002- 1.0226, p=0.047), bypass time (each minute increase AKI odds ratio 1.02, 95% confidence interval 1.0045- 1.043, p=0.0157), and longer vasopressor use time (odds ratio 3.28, 95% confidence interval 1.0061 – 1.321, p=0.039). Per ml of blood transfusion the odds ratio for AKI was 1,03 (95% confidence interval 1.0186-1.058, p=0.0001). Using Cox regression, AKI was independently associated with longer intensive care unit stay (odds ratio 3.05, 95% confidence interval 2.36-4.354, p= 0.000), duration of ventilation (odds ratio 7.8, 95% confidence interval 5.8-9.8, p=0.000) and mortality (odds ratio 6.17% confidence interval 1.14-33, p=0.0345).
Conclusions: We observed a high incidence of AKI following congenital heart surgery with the introduction of AKIN criteria.

Su175 QUALITY OF LIFE OF PATIENTS WITH ACUTE RENAL FAILURE

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Introduction and Aims: Quality of life (QoL) has not been a priority goal for patients with acute *renal failure (ARF),* that show high level of mortality in Brazil and abroad.
Aim: To evaluate QoL in patients with acute renal failure in two public hospitals.
Methods: *Study design:* Case-control study. *Setting:* Hospital São Paulo-UNIFESP and Hospital dos Servidores do Estado –HSPE. *Study participants:* Cases and controls were selected from among all patients followed at ICU in the period between 2008 and 2009. All forms were examined to check inclusion criteria. Patients were randomly selected, but all inclusion and exclusion criteria were verified in an initial interview prior to their inclusion in the study. *Cases* were patients at ICU with a confirmed diagnosis of ARF, with no chronic comorbities, needed renal therapeutic support. The *control group* consisted of patients at ICU with renal acute or chronic disease and we also rejected anyone suffering from any chronic diseases. From 578 patients initially identified, 272 subjects (136 cases/136 controls) were matched according to age and gender. *Main outcome measures:* QoL was evaluated by a Portuguese version of the SF-36 Health Survey, a generic health survey constructed to yield a profile useful in understanding population differences in physical and mental health.
Results: Average (D.P.) SF-36 dimension scores for cases and controls respectively were: *Role Limitations due to physical problems* 41.5 (35.9)/41.6 (33.6), *Physical Function* 27.0 (40.2)/29.2 (41.5), *Bodily pain* 50.8 (29.8)/50.8 (28.4), *General Health Status:*51.4 (22.9)/59.0 (23.0), Vitality: 51.5 (27.5)/60.2 (24.3), *Social function* 49.3 (31.1)/56.8 (28.2), *Role-Emotional Function* 59.9 (45.2)/69.1 (41.9) e *Mental health* 65.3 (24.8)/71.9 (21.6).We also detected any statiscally significant difference between cases and controls related to the domains: General Health Status, Vitality, Social function and Mental health ($p<0.05$). There is, also, significant difference between cases and controls as to death incidence (<0.001); the percentage of death among cases was 49.5% and among controls was 16.5%.
Conclusions: Our data showed that patients with ARF with high levels of mortality had QoL substantially impaired, with significant differences compared to controls as to the domains: General Health Status, Vitality, Social function and Mental health.

Su176 APPROPRIATE HEMODIALYSIS PRESCRIPTION IN ACUTE KIDNEY INJURY AND SURVIVAL: THE CARE ORGANIZATION POINT OF VIEW

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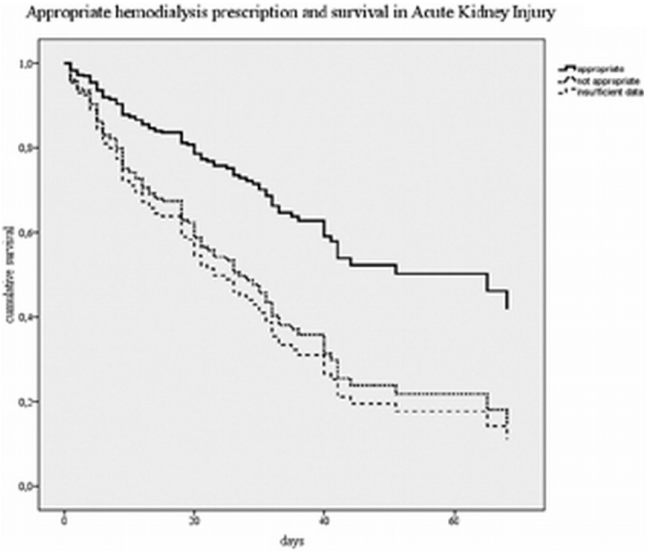
Introduction and Aims: Mortality in patients with Acute Kidney Injury (AKI) who require hemodialysis is extremely high, in intensive care unit (ICU) as well as out of ICU. We wanted to assess the mortality rate and homogeneity of dialysis prescription in AKI patients admitted to a large university-affiliated hospital. Nephrology staff wasn't exclusively dedicated to critically ill patients, consulting was done from all the nephrologist of the Unit, and dialysis was carried out from technicians employed in out-patients dialysis and from the technicians employed in nephrology in-patients care.
Methods: This is a prospective cohort observational on a period of 3 years. 259 patients were studied, 175 male and 84 female, mean age 70.2±15.4 yrs. They performed a total of 1533 HD and 5687 HDhours.
Results: Mean APACHE II score was 22.67±6.49, with higher values in IUC patients (25.44±0.04 vs 21.63±5.48, $p<0.001$). RIFLE was 2.75±0.54, without differences between ICU/non-ICU. Total mortality was 45.6% (IUC=68.6%; non-ICU=37%, $p<0.01$). Thrice/weekly HD was prescribed in 5.6% cases, daily HD in 94.4%. Standard bicarbonate dialysis (sHD) was prescribed in 60.6% patients and Slow Efficacy Daily Dialysis (SLEDD) in 39.4%. The number of hours/session were higher for SLEDD than sHD (4.8±2.7 vs 2.7±0.7 hrs/session), but 68,6% of patients in SLEDD were treated less than 8h/session. Urea Reduction Rate was measured in 83.8% of patients, and only 28.6% of them had an URR ≥ 40%. URR was lower for ICU patients than non-ICU (38.5±1.8% vs 46.6±1.1%, $p<0.001$). We have estimated as appropriate dialysis prescriptions those who fulfill both the criteria of Italian Guidelines for HD beginning and URR≥40%. Only 27% resulted as appropriated; 56,8% resulted non appropriated and 16,2% not interpretable, due to absence of URR data. Survival was conditioned from the appropriate dialysis prescription: mortality was 28% in patients who received an appropriate prescription, whereas in other groups mortality was 52% for inappropriate prescription, and 50% for not interpretable data. After



correction for possible covariates, the Cox analysis resulted in a Hazard Ratio of death for inappropriate dialysis prescription of 2.91 ($p<0.001$, CI 95% = 1.56-5.42).

Conclusions: A decrease of mortality in patients with AKI seems to be possible when an appropriate prescription and monitoring of hemodialysis, in terms of the time of initiation and the dose of dialysis, was made. A dedicated staff to critically ill patients, nephrologists and dialysis technicians, appears to be an adjunctive value to the patient's care, because it's necessary to ensure the respect of guidelines, and the adequateness of dialysis dose prescription. Time to intensive nephrology seems to be arrived in hospitals with a large emergency activity.

Su177 HIGH PERMEABILITY DIALYSIS MEMBRANE ALLOWS EFFECTIVE REMOVAL OF MYOGLOBIN IN ACUTE KIDNEY INJURY DUE TO RHABDOMYOLYSIS

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Introduction and Aims: Rhabdomyolysis is the syndrome arising from the loss of integrity of skeletal muscle and release of contents of muscle into the extra-cellular fluid. Myoglobinuria occurs only in the context of rhabdomyolysis. Myoglobin is a dark red 17.8-kDa protein that is freely filtered by the glomerulus, enters the tubule epithelial cell through endocytosis and is metabolized. It appears in the urine only when the renal threshold of 0.5 to 1.5 mg of myoglobin per deciliter is exceeded. The clinical manifestations of this disorder occupy a diverse spectrum, from asymptomatic elevation of the serum creatine kinase concentration to acute kidney injury (AKI) requiring renal replacement therapy in the setting of multiorgan failure. It has been suggested that rhabdomyolysis from all causes leads to 5-25% of all cases of acute kidney injury (AKI).

The objective of this study was to test the ability of myoglobin removal of a novel, high permeability polysulphone dialyzer in acute kidney injury due to rhabdomyolysis.

Methods: Extended dialysis was performed using a single pass batch dialysis system and a novel polysulphone high-flux dialyzer (effective surface area 1.8 m^2; inner lumen 220 μm; wall thickness 35 μm; allowing elimination of substances with a molecular weight of up to 30kDa).

Samples were collected at pre-filter and post-filter sites, as well as from the collected spent dialysate. The dialyzer clearance was calculated from concentrations before and directly after the dialysis membrane, the blood flow and the ultrafiltration rate. The total amount of the myoglobin removed was measured directly as the whole dialysate was preserved.

Results: The use of the extended dialysis (ED) with the Genius® dialysis system, utilizing the novel polysulphone high-flux dialyzer (Ultraflux® AV 1000 S), was associated with a median myoglobin clearance of 82.0 mL/min (range: 52.4 – 126.3 mL/min), resulting in a median myoglobin removal of 188.4 g/d (range: 124.2 – 355.7 g/d).

Urea clearance values with 200 mL/min blood flow and 200 mL/min dialysate flow rates were found to be 163±17 ml/min. Creatinine clearance values were determined: 145±20 ml/min at a dialysate flow of 200 Lml/min. For phosphate, we found a plasma clearance of 160±19 ml/min at 200 mL/min dialysate flow rate. β2-microglobulin clearance values measured 97±15 ml/min at 200 mL/min dialysate flow. Vitamin B12 clearance values were determined: 64±4 ml/min at a dialysate flow of 200 Lml/min.

Conclusions: Extended dialysis with high flux, high permeability membrane allowed effective elimination of myoglobin with a clearance of myoglobin that surpassed all previously reported dialysis techniques. This membrane may be advantageous in preventing acute kidney injury or avoiding complete loss of kidney function in patients with rhabdomyolysis. Further studies are needed to determine whether improving renal recovery or mortality in patients with AKI due to rhabdomyolysis is possible.

Disclosure: The dialysis filter used in this study was provided courtesy of Fresenius Medical Care, Bad Homburg, Germany. JTK received speaker fees and research support from Fresenius Medical Care Germany and the Else Kröner-Fresenius-Foundation (P63/06//EKMS 06/03).

Su178 PROPHYLACTIC CONTINUOUS VENOVENOUS HEMOFILTRATION CAN PREVENT WORSENING OF THE KIDNEY FUNCTIONS AFTER PERCUTANEOUS CORONARY INTERVENTION IN PATIENTS WITH ADVANCED CHRONIC KIDNEY DISEASE

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Introduction and Aims: Contrast nephropathy (CIN) is associated with increased risk of in hospital morbidity and mortality. Patients with chronic kidney disease (CKD) are at higher risk of CIN. The purpose of this study is to find out whether post procedure continuous venovenous hemofiltration (CVVH) can prevent CIN in CKD patients after coronary angiography.

Methods: Patients with stage III and IV CKD scheduled for coronary angiography between January 2004 and December 2005 were enrolled in the study. CVVH was done after the procedure. Serum creatinine and calculated GFR (c-GFR) were estimated before the procedure, repeatedly during hospitalization, at discharge, and 15 days after the procedure. The incidence of CIN was calculated.

Results: 98 patients were enrolled in the study. 52 (53.1%) were males, the mean age was 60.7±10.99 years. Pre procedure serum creatinine was 411.29±79.94 μmol/l, GFR 18.04±4.26 ml/min. All patients underwent post procedure CVVH for 21.34±2.12 hrs. The mean time interval between the procedure and the start of CVVH was 44.34±18.77 min. The mean serum creatinine at discharge was 403.58±8 8.39 μmol/l, and 422.54±88.86 μmol/l 15 days after the procedure. Mean GFR was 18.52±4.61 ml/min, and 17.62±4.27 ml/min at discharge, and 15 days after the procedure respectively. One patient (1.02%) developed CIN that required repeated CVVH during hospitalization and ended up on regular hemodialysis. The in hospital mortality was 0%.

Conclusions: CVVH is effective in preventing CIN after coronary angiography in CKD patients.

Su179 STAGE 3 ACUTE KIDNEY INJURY (AKI) IN A NEPHROLOGY UNIT – PROGNOSTIC FACTORS ARE DIFFERENT TO THOSE IN AN ITU SETTING

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Introduction and Aims: Acute Kidney Injury (AKI) is common, however there remains a lack of evidence regarding the factors affecting outcome, especially in a general nephrology setting. The heterogeneous nature of AKI has meant that patients have previously predominantly been studied in isolated settings such as critical care, but development of the Acute Kidney Injury Network classification for AKI allows for improved comparison between different patient groups. The aim of this study was to determine the factors that influence mortality from stage 3 AKI in patients presenting to a single-centre nephrology unit.

Methods: 228 patients with stage 3 AKI were referred to a single nephrology unit between 1st January 2006 and 31st December 2006. Data were collected prospectively on all patients and included patient demographics and co-morbidities along with factors previously reported to influence the outcome of AKI including aetiology of AKI, cardiac co-morbidity, urine output and the development of multi-organ failure or sepsis. Patients were followed up for one year from presentation by using computer records, and were complete for 198 patients. Potential prognostic factors were tested using a multivariate Cox proportional Hazards model.

Results: Of the 228 patients, 146 patients (64%) were male and 199 (87.3%) were Caucasian. The mean age at presentation was 65.3 years. Acute Tubular Necrosis due to either ischaemia, sepsis or a combination of both was the commonest cause of AKI (98 patients, 43.0%), followed by obstruction (23 patients), pre-renal failure (20 patients), rapidly progressive glomerulonephritis (19 patients) and myeloma (17 patients). 154 patients (67.5%) received renal replacement therapy. Overall in-patient mortality was 29.8% (68 patients) with a higher mortality rate in the group receiving renal replacement therapy (35.1%, 54 patients). 1-year mortality rose to 50% (99 patients) overall, and 57.9% (77 patients) in the group that received renal replacement therapy. Age (HR = 1.04, $p<0.0001$), malignancy (HR

= 2.87, p<0.0001) and multi-organ failure (HR = 3.92, p<0.0001) were found to be significant independent predictors of patient survival. Cardiac events, urine output and sepsis were not significant prognostic indicators of survival.

Conclusions: The incidence, outcome and mortality of AKI in this study are similar to those reported in the literature and confirm that AKI confers an excess in both short- and medium-term mortality. Patient age and the presence of malignancy or multi-organ failure were independent predictors of survival for patients with AKI, whilst cardiac events were only found to be correlated with with age and multi-organ failure and were not independently prognostic for mortality. In contrast to other studies predominantly performed in critical care populations, urine output and sepsis did not affect survival. This study highlights the difficulty of applying data from distinct patient subsets to a more generalised nephrology population and supports the need for further research in this area.

Su180 MASS BALANCE AND MEASURED SOLUTE CLEARANCE IN CONTINUOUS VENO-VENOUS HAEMODIALYSIS (CVVHD)

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Introduction and Aims: Recent outcome studies have utilised the effluent flow rate (EFR) as a measure of dose in the different continuous renal replacement therapies. Kinetic assumptions that equate EFR with solute clearances in these slow therapies may not, however, hold true in the acute setting. The present study aimed to i) assess cross-dialyser mass balance by comparing urea nitrogen (UN) removal calculated from blood- and dialysate-side measures in CVVHD, and ii) examine potential explanatory factors for any shortfall in dialyser performance.

Methods: CVVHD utilised multi-component technology with renal technical staff replacing individual components, including dialysers, when required, to facilitate ongoing treatment. From initiation of therapy to a maximum of 7 days we performed simultaneous daily measurements of: dialyser fluid effluent UN (FUN), BUN at the dialyser inlet (BUNi) and outlet, and *Transonic* Qb. UN flux was calculated on the blood-side using conventional correction factors for the water content of erythrocytes (0.72) and plasma (0.93) and was compared to the dialysate-side UN flux. Dialyser performance was assessed using the FUN:BUN ratio (FBR) where FBR = FUN/(BUNi/0.93) applying the plasma water correction to BUNi. Potential explanatory factors for a shortfall in FBR (<<1) were examined.

Results: Seventy-eight measurements were performed in 15 patients: median [range] number of measurements/patient = 5 [2 – 7]. Circuit patency was maintained with unfractionated heparin infusions (45 measurements) or a saline flush strategy (33). Excluding 3 outliers, we found no mass balance error with median blood- and dialysate-side UN removal within 2% of each other. The mean (SD) FBR was 0.89 (0.13). Low FBR was not explained by anticoagulation status, dialyser longevity, by a Qd within the measured range of 1400 – 1600 ml/hr or by a clinically relevant *Transonic* Qb of 150 – 250 ml/min. Application of an upward correction factor of 1.3 to the prescribed EFR would have reflected target solute clearances in most cases with CVVHD functioning within usual treatment parameters.

Conclusions: The usual assumptions about water and erythrocyte water content hold true in the acute setting with no evidence of a mass balance error. Shortfalls in dialyser performance cannot be explained by the usual factors but upward correction of the dialysate effluent flow rate can better reflect desired solute clearances with CVVHD working within the usual treatment parameters.

Su181 COMBINATION OF URINARY KIDNEY INJURY MOLECULE-1 AND INTERLEUKIN-18 AS EARLY BIOMARKER FOR THE DIAGNOSIS AND RISK STRATIFICATION OF ACUTE KIDNEY INJURY FOLLOWING CARDIOPULMONARY BYPASS SURGERY: A PROSPECTIVE NESTED CASE-CONTROL STUDY

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Introduction and Aims: The role of urinary biomarker for early diagnosis and risk stratification of acute kidney injury (AKI) has not been well described. This prospective nested case-control study demonstrated the diagnostic and prognostic values of combining urinary kidney injury molecule-1 (uKim-1) and interleukin-18 (uIL-18) for AKI after cardiopulmonary bypass surgery (CPB).

Methods: One hundred and twenty-two adult subjects undergoing cardiopulmonary bypass surgery were investigated. Thirty were diagnosed with AKI by the RIFLE criteria. Sequential urine samples of each subject enrolled were measured for Kim-1 and IL-18 levels normalized by urinary creatinine. The relationships between AKI occurrence or development and the increased levels of the two biomarkers were analyzed in a preliminary study including 75 matched subjects. Furthermore, the diagnostic and prognostic performances of uKim-1, IL-18 and combined for AKI were evaluated in the whole cohort by receiver operating characteristic curve.

Results: Increased level of uKim-1 was associated with AKI occurrence while that of uIL-18 was related to progressive AKI. The patients with higher uIL-18 level were more prone to develop longer AKI. As demonstrated by area under the receiver operating characteristic curve, the performance of uKim-1 for diagnosis of AKI at 6 and 12 hour after surgery was 0.881 and 0.882 respectively, and the performance of uIL-18 and serum creatinine for progressive AKI was 0.872 and 0.907. Combining the two biomarkers, the area under the curve for predicting progressive AKI at 6h and 12h was 0.883 and 0902.

Conclusions: Urinary uKim-1 and uIL-18 are complementary biomarkers of AKI after CPB. Combination facilitates the early diagnosis and prognosis assessment.

Su182 UREA KINETIC MODELS FOR RENAL REPLACEMENT DOSE PRESCRIPTION AND FOR EFFECTIVELY DELIVERED DOSE EVALUATION IN ACUTE KIDNEY INJURY

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Introduction and Aims: Dose prescription in AKI is still a controversial issue, either regarding dose itself, or suggested time for initiation and withdrawal. To this aim we suggest a simple urea kinetic model for both standard prescription and evaluation (UKMAKJ), and a more complex one for precise evaluation of the effective dialysis delivered dose, based on the direct dialysis quantification on urinary and effluent fluids (DDQAKJ).

Methods: The prescription model UKMAKJ considers the wKT/V parameter (whose value=6 is considered as the minimum desired target) in place of the standard coefficient in ml/h/kg of body weight, and takes account of residual renal function; filtration fraction (FF) and maximum filtration rate (MFR) are considered too to avoid excessive ultrafiltration.

The evaluation model DDQAKJ is conversely based on urea nitrogen measurement on blood samples, equilibrated (e) and corrected for plasma water (pw), on urine (Eu) and effluent (E) values, according to the following equations (iteration between Eq. 1 and 2):

Eq. 1: eG = (pwBUNo2*Vo2 – pwBUNe*eVt+Euid)/(tid-30)

Eq. 2: eVt = (E + Eu – eG*(t+30) – Qf*t*pwBUNo)/(pwBUNo-pwBUNe)

Eq. 3: eK = ((E -Eu)*ln(pwBUNo/pwBUNe))/(t*(pwBUNo – pwBUNe))

Results: See tables. The average values for UKMAKJ have evidenced infusion rate of about 2.7 Lt/h, corresponding to wKt/V = 7.25±4.57; this, additioned to renal value, gives a total value of 8.61±4.41, higher than target value = 6.

Table 1. Main results of the CVVH parameters used for daily dose prescription by UKMAKJ model

n	Qr ml/h	Predilution %	FF ratio	wKrt/V renal	wKt/V CVVH	wKT/V total
24	2731±657	49.4±30.1	16.1±10	1.36±1.48	7.25±4.57	8.61±4.41

Table 2. Main results of the DDQAKJ model, based on the direct dialysis quantification

n	eVt ml	eG mg/min	nPCR gr/kg/24h	eK ml/min	wKT/V index	SR/G index
4	39132±3890	8.87±1.57	1.48±0.28	53.4±19.6	6.17±1.55	1.13±0.18

By DDCAKJ eVt resulted 39.1±3.8 Lt; in a selected case Vt = 38975 ml was measured in overt overhidratation, while resulted=32704 after five days of sustained fluid removal. The eG and nPCR higher than normal suggested hypercatabolism. The effective wKt/V = 6.1±1.55 was only sligtly higher than its target.
A ratio value higher than the unit between urea nitrogen removal and generation suggests adequate depuration, as compared to metabolic production.
Conclusions: Our results have confirmed the usefulness of UKMAKJ for simple prescription to achieve an adequate depurative dose, without the risk of excessive ultrafiltration that might compromise extracorporeal circulation. The DDCAKJ model allowed us to measure urea distribution volume, a parameter helpful to assess the patient hydration status by comparison with anthropometric values; furthermore, an effective Kt/V may be obtained by measuring real clearance and volume; urea generation rate (G) and nPCR are moreover useful to assess protein metabolic status, that is often compromised in AKJ. Finally, the ratio between urea removal and its generation might represent a new index of adequacy, when values ≥1 are obtained.

Su183 EVALUATION OF PROGNOSTIC MODELS IN ACUTE KIDNEY INJURY PATIENTS REQUIRING DIALYSIS

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Introduction and Aims: General and specific severity scores for patients with Acute Kidney Injury (AKI) may have limitations to estimate the risk mortality depending on the diversity of methods, on the moment of data recording, and on the patient's care setting.
Aim: Here we prospectively evaluated four general score system (APACHE II, SAPS, SOFA, and LODS) and one specific model (RIFLE) in the ability to predict mortality of 70 critically ill patients who developed acute kidney injury, admitted in the intensive care unit (ICU), and requiring mechanical ventilation and renal support.
Methods: We prospectively evaluated these models also in 174 patients who developed AKI and were admitted into non-ICU medical departments. Patients with obstructive acute renal failure were excluded from the study. All data were recorded at the moment of nephrology consulting.
Results: Mortality in IUC patients was 68,6%, male gender 71.4%, patient's age= 69.1±14.4 yrs, RIFLE=2.74±0.53, APACHE II=25.44±8.04. Mortality in non-ICU patients was 37.9%, male gender 65.5%, patient's age = 70.13±16.05 yrs, RIFLE=2.64±0.58, APACHE II=21.57±5.56. APACHE II was singnificantly lower in non-ICU than ICU patients ($p<0.001$), RIFLE did not differ between the two care settings. In ICU patients mortality was 80% in the case of AKI class RIFLE III, and lower for class RIFLE II and I (41.7% and 33.3%, $p<0.01$). In non-ICU patients mortality was 36% in the case of AKI class RIFLE III, 50% class RIFLE II, and 30% class RIFLE I (p=ns). In ICU patients ROC curves demonstrated the largest area for RIFLE score (0.67, p=0.019, CI 95%=0.52-0.82).
In non-ICU patients RIFLE score had the lowest area, and largest area was represented for LODS score (0.73, $p<0.001$. CI 95%=0.65-0.80).
Conclusions: Determination of RIFLE score in critically ill ICU patients, at the moment of nephrology consulting or dialysis beginning, represents a reliable estimation of survival possibilities. RIFLE class III value is accompanied by higher mortality. The lower mortality at low RIFLE score let us to suppose that early treatments determined on the basis of RIFLE can be necessary to improve prognosis. On the contrary, in non-ICU patients RIFLE score is not representative of the risk of death. In this case the evaluation of multiple organ dysfunction, as that obtained with LODS score system, is helpful in the determination of the risk and may represent an instrument on decision making algorithm.

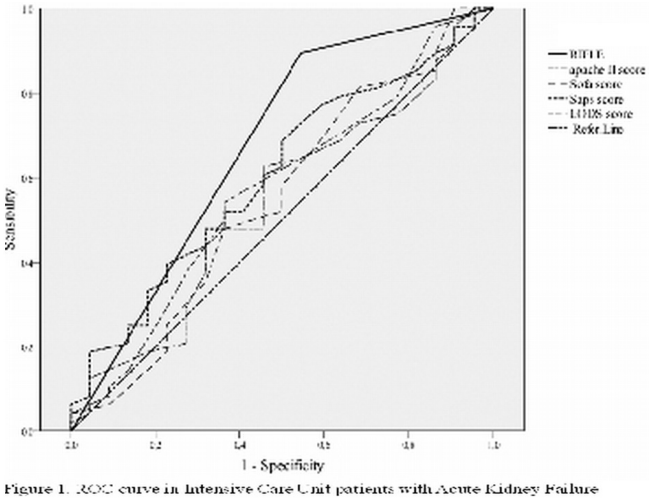

Figure 1. ROC curve in Intensive Care Unit patients with Acute Kidney Failure

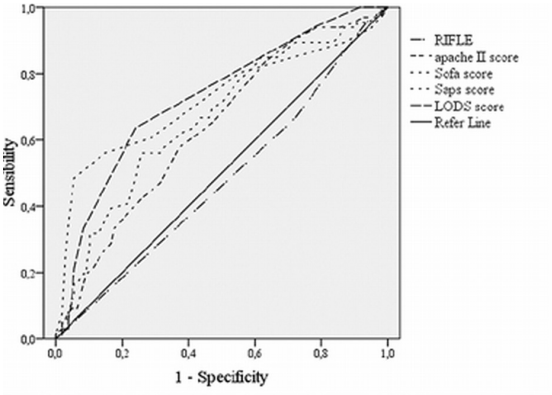

Figure2. ROC curve in NON-Intensive Care Unit patients with Acute Kidney Failure

Su184 RENAL FUNCTION IN PATIENTS WITH ACUTE PANCREATITIS

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Introduction and Aims: Acute kidney injury (AKI) can often be present in patients with acute pancreatitis (AP). The aim of this study was to assess the presence and the risk factors of AKI in patients with AP.
Methods: We performed a cross-sectional study in a gastro-enterology clinic (non-intensive care unit) during a 36 months period; 188 patients with AP were admitted (mean age 54.03 + 17.45;66 female,122 male). AP was caused in 84 patients (44.68%) by gallstones, in 52 (27.65%) by alcohol intake, while in the remaining 52 (27.65%) the cause of AP was diverse (hypertriglyceridemia, post-ERCP, unknown). The following data was obtained: medical history, abdmonial ultrasound, serum creatinine, serum lipase, AST, ALT, glycemia, leucocyte count, C-reactive protein (CRP-at 48 hours). AKI was diagnosed using Acute Kidney Injury Network criteria. Statistical analysis of the data was performed using Open Epi 2.1.1 and Win Stat for Microsoft Excel programs.
Results: AKI was present in 55 patients (29.25%).The risk of developing AKI was not increased by the presence of gallstones – 24/84 showed AKI (OR= 0.94, CI=95%,0.4744-1.858, p=0.49) or by alcohol intake- 11/52 (OR=0.56, CI=95%,0.2374-1.248,p=0.06), but the risk was increased in non alcoholic- non billiary pancreatitis (OR=1.79,CI=95%,0.858- 3.732,p=0.04). We found no statistically significant correlation between baseline creatinine and the other biological parameters (follow-up creatinine, serum lipase,

AST, ALT, total bilirubin, CRP). There was only a correlation of serum lipase with AST (R=0.19,p=0.006) and ALT (R=0.23,p=0.0008).
Severe AP was diagnosed using RANSON criteria (≥3) or the presence of pancreatic necrosis in 16 (8.5%) patients, 6 of them presented AKI (OR=1.50,CI=95%,0.4247-4.859,p=0.22). None of the RANSON criteria was a risk factor for AKI in our patients: AST (>250U/l)- 40 patients (OR=0.89, CI=95%, 0.3703-2.052, p=0.39), leukocytosis (>16,000/mmc)- 22 patients (OR=0.54,CI=95%,0.1265-1.822,p=0.15), hyperglycemia (>200mg/dl)- 28 patients (OR=1.17,CI=95%,0.4339-2.966,p=0.35). Increased CRP >150mg/l represents a prognosis marker of AP, was present in 36 patients and is a risk factor for AKI (OR=4.2, CI=95%,1.649-11.03,p=0.0005).
In 50 (26.59%) patients the presence of peripancreatic fluid collections was found sonographically and represents a risk factor for AKI (OR=1.9, CI=95%, 0.9278-4.086, p=0.02).From the associated diseases some represented risk factors for AKI in patients with acute pancreatitis: cardiovascular disease (OR=2.1,CI=95%, 1.007-4.496,p=0.01), arterial hypertension (OR=1.61,CI=95%,0.8158-3.194, p=0.07) and especially preexisting chronic kidney disease (OR=3.8,CI=95%, 1.367-11.03,p=0.002); while obesity, dyslipidemia and diabetes mellitus were not risk factors for AKI in patients with AP.
Conclusions: AKI is an important complication in AP even in non-intensive care units, especially in patients with previous CKD or cardiovascular disease and in the presence of peripancreatic fluid collections and increased C-reactive protein.

Su185 PROXIMAL TUBULAR CELLS ACQUIRE THE PROGENITOR MARKERS CD24 AND CD133 DURING TUBULAR REPAIR

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Introduction and Aims: Acute tubular necrosis is followed by regeneration of the injured renal tubular epithelial cells. It is has been shown that proximal tubuli are regenerated by intrinsic cells. Nonetheless, it is still unresolved whether intratubular stem/progenitor cells or the entire cell population contribute to the regeneration.
Methods: In this study we identified a cell population localized in the proximal tubuli expressing the progenitor markers CD24 and CD133 using immuno-electron microscopy and immunofluorescent stainings on normal human kidney sections and on biopsies showing acute tubular necrosis (ATN).
Results: Within normal human kidney CD24 positive cells were scattered as single cells or in small groups of 2-4 cells throughout the proximal tubule and co-expressed the stem cells marker CD133. The morphology of CD24 positive cells differed from that of normal proximal tubular cells and contained less cytoplasma, less mitochondria and the brush border was absent. In contrast to normal proximal tubular cells, the cells expressed CD24, CD133, CXCR4, and other markers that are associated with epithelial dedifferentiation.
In ATN biopsies, the number of CD24 positive tubular cells was much higher compared to normal human kidneys. To establish whether the CD24 positive cells were regenerative, we co-stained for the proliferation marker Ki-67. We observed that in both normal human kidneys and in the ATN biopsies the majority (∼85%) of the Ki-67 positive cells was also CD24 positive.
Conclusions: CD24 positive tubular cells represent a novel distinct subpopulation of cells, which participate in the regeneration after acute tubular necrosis.

Su186 EXTENDED DAILY ON-LINE HIGH VOLUME HAEMODIAFILTRATION IN SEPTIC MULTIPLE ORGAN FAILURE: OUTCOME AND COSTS

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Introduction and Aims: The outcome of patients with septic multiple organ failure (MOF) remains poor. There are experimental and clinical data indicating a beneficial effect of high volume haemofiltration. Delivering a high volume therapy is only cost-effective using on-line devices because of high costs for additional solution bags in conventional continuous renal replacement therapy (CRRT). In a pilot study we investigated feasibility and effectiveness of extended daily on-line high volume haemodiafiltration (HDF) with technically maximum convective volume in patients with septic MOF.
Methods: We included 21 consecutive critically ill patients with septic multiple organ failure having an extremely poor prognosis (SAPS II over 50, APACHE II over 25). RRT was applied with extended daily haemodiafiltration for 6 to 23 h using the AK 200 Ultra dialysis machine (Gambro AB, Sweden) in the ultracontrol predilution mode. Dialysate and substitution fluid were prepared on-line. Patients underwent 289 treatments.
Results: The mean convective volume was 17,8 l/h and 208 ml/kg/h respectively, mean treatment time was 10.25 hours/day.17 of 21 patients survived 28 days (81%). 12 patient survived by day 60, 11 patients by day 90 (52%). The predicted survival probability of patients with APACHE II (mean 33,6) and SAPS II (mean 68,6) scores is 19%. Haemodynamics (mean arterial pressure, noradrenaline dose) improved significantly during the treatment procedures. Material costs per treatment did amount to 35 Euro being comparable to the costs for a haemodialysis session in a chronic patient.
Conclusions: Extended daily on-line HDF using maximum convective volume seems to improve the outcome of septic multiple organ failure, especially in the early phase. The investigated mode of treatment proved to be feasible and highly cost-effective compared to conventional CRRT.

Su187 EFFECT OF OBSTRUCTIVE SLEEP APNEA ON MARKERS OF ACUTE KIDNEY INJURY

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Introduction and Aims: Obstructive sleep apnea (OSA) is a relatively common condition causing recurrent hypoxia and hypercapnia. An acute rise of arterial blood pressure caused by ischaemia during apneic episodes may contribute to the generalized endothelial damage. We postulated that the frequent apneic episodes during sleep can result in repeated renal ischaemia-reperfusion injuries, which thereby may lead to acute subclinical kidney injuries eventually resulting in chronic kidney disease (CKD).
The aims of the study were: 1. to assess the influence of sleep apnea syndrome on kidney function and the excretion of early urine markers of acute kidney injury, arterial stiffness and central aortic pressure and 2. to evaluate whether the treatment of sleep apnea with continuous positive airway pressure (CPAP) could induce a possible recovery from kidney injury.
Methods: To this prospective, interventional pilot study we recruited 13 males with polysomnography-based diagnosis of sleep apnea syndrome and GFRMDRD> 60ml/min/1,73 m^2. Mean age was 49,6±9,0 years, BMI 37,7±6,1 kg/m 2 and WHR 1,10±0,05. Central blood pressure in the aorta and PWV was noninvasively measured with ECG-synchronized tonometry (PulsePen, DiaTecne s.r.l., Milan, Italy) at baseline. Serum creatinine concentration, lipids, hsCRP and urine AKI markers: cystatin C, NGAL, L-FABP were measured both before and after polysomnography. All patients underwent polysomnography (diagnostic night) and when the

diagnosis of OSA was confirmed were later treated with the continuous positive airway pressure (CPAP). After 6 to 8 weeks of CPAP during the second polysomnographic assessment all the measurements were repeated according to the same scheme as at baseline.

Results: The results of all measurements are presented in table 1

Table 1

	Polysomnography 1 (diagnostic night)		Polysomnography 2 (therapeutic night)	
	before	after	before	after
Serum creatinine (mg/dl)	1,07±0,12	1,01±0,11	1,08±0,13	1,04±0,1
PWV (m/s)	10,7±1,7	–	9,0±1,9	–
Systolic BP central	115,4±15,9	–	115,4±12,3	–
Diastolic BP central	75,8±16,5	–	75,3±8,7	–
hsCRP (µg/ml)	6,2±3,9	5,7±3,6	5,1±3,5	5,9±4,1
Cystatin C (ng/ml)	85,4±36,1	121,3±34,7	95,2±46,9	111,0±40,8
L-FAB P (ng/ml)	1,9±1,3	2,6±1,8	2,5±3,4	2,7±2,8
NGAL (ng/ml)	9,0±8,4	12,5±11,6	9,5±12,7	18,6±19,8
Total cholesterol (mg/dl)	222,2±49,9	211,5±45,4	210,7±51,0	200,4±41,6
Triglicerides (mg/dl)	361,9±317,2	182,2±92,7	384,2±277,4	166,1±65,5
LDL cholesterol (mg/dl)	125,0±33,8	134,9±35,3	119,2±37,7	124,9±38,0
HDL cholesterol (mg/dl)	41,6±8,0	40,5±8,6	39,9±7,1	40,5±6,5

There were no significant changes of all the parameters during the study.

Conclusions: In this pilot study we were not able to confirm the hypothesis that the sleep apnea syndrome induces a kidney injury and its treatment with CPAP may improve kidney function.

Su188 SUSTAINED LOW-EFFICIENCY DAILY DIALYSIS WITH ACETATE-FREE DIALYSATE IN PATIENT WITH ACUTE KIDNEY INGURY AFTER CARDIAC SURGERY

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Introduction and Aims: Sustained low-efficiency daily dialysis (SLEDD) have demonstrated favourable results in critically ill patients with acute kidney injury (AKI). However, hemodynamic complications during SLEDD have been reported from 15% up to 27%. The accumulation of acetate containing in bicarbonate dialysate (3 mmol/l) in blood of patients may be a possible cause. The purpose of this study was to determine the safety of SLEDD with the acetate-free dialysate (acetate is replaced by hydrochloric acid).

Methods: A prospective study included 36 patients with AKI after cardiac surgery. Twenty-two patients were on vasopressor support, 14 patients on mechanical ventilation. 16 patients have had arrhythmia, and 12 ones– acute myocardial infarction. Patients were classified in 2 groups: in group A (n=18) we used SLEDD with acetate-free dialysate, and group B (n=18) – SLEDD with bicarbonate dialysate. Mean age was 57±4.2 years (A) and 53±3.1 years (B), mean APACHE II score was 23.2±2.4 and 21.4±1.9, respectively. We used a dialysis machine, the 5008 (Fresenius Medical Care). Mean blood flow was 150 ml/min, dialysate flow at 100-200-300 ml/min, mean treatment session time was 8 hours, number of procedures 2.6±0.3. Biochemical data, episodes of hypotension and arrhythmias were studied. Episodes of hypotension were defined as the need for resuscitative intravenous fluids and/or to increase inotrope dose.

Results: The level of urea has significantly decreased (14.8±1.3 vs 26.7±2.4 mmol/l from to (A) and 12.6±1.6 vs 24.8±2.2 mmol/l from to (B); $p<0.01$), and the creatinine has reduced (126±12 vs 292±18.7 mkmol/l from to (A) and 112±10.8 vs 308±18 mkmol/l from to (B); $p<0.01$). In both groups, the stable control of electrolytes and acid-base levels was achieved. At the same time, the use of acetate-free dialysate resulted to significant declines hemodynamic complications (4.5% vs. 22.4%).

Intradialytic hemodynamic complications

Parameter	Group A	Group B
Number of patients	18	18
Number of procedures	44	49
All complications	2*	11*
Episodes of hypotension	1	7
Arrhythmia	1	4

*$\chi^2=6.18$; p=0.046

Conclusions: SLEDD with acetate-free dialysate is favourable method comparing SLEDD with bicarbonate dialysate in patients with AKI after cardiac surgery.

Su189 ACUTE KIDNEY INJURY REQUIRING DIALYSIS IN CRITICAL CARE UNIT IN A DEVELOPING COUNTRY

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Introduction and Aims: In developing countries, little is known about renal replacement therapy (RRT) for acute kidney injury (AKI) in critically ill patients. The aim of this study is to describe characteristics of patients, clinical practice of renal support and outcome in intensive care unit (ICU) in a developing country.

Methods: Patients who underwent RRT for AKI from May 2003 to July 2008, in 4 ICUs in our institution were included in this retrospective study. Patients with end stage renal disease or younger than 18 were excluded. We have considered: patients demographics, indications of RRT, number of dialysis session, comorbidties, APACH II score for illness severity, mechanical ventilation, use of vasoactive drugs, and mortality rate.

Results: 105 critically ill patients admitted during the study period were treated with RRT, with a mean age of 56,13 16,8 (19-85) years, 65 were males and 40 females, all of them received intermittent hemodialysis. The total number of dialysis sessions was 284, and the mean number was 3,7 2,9 (1-11), mean length of session was 225,22 75,16 (60-290) min. Majority of the cases were from medical ICU accounting for 67%, followed by 30,2% from surgical ICUs (including cardiothoracic surgery ICU) and 2,8% were from burn ICU. The most common comorbidities were Type II diabetes mellitus in 46 (44%) and hypertension in 35 (33,4%). Sepsis was a contributing factor to AKI in 60 patients (57,14%), hypovolemia in 30 (28,5%), and cardiogenic shock in 9 (8,5%). The average of APACHE II score was 25,86 (11-35), 70 (66,7%) patients were ventilated, 63 (60%) were under vasoactive drugs and 88 (83,8%) were oliguric. The most common indication for initiation of dialysis was hyperkalemia in 51 (48,5%) of the cases, followed by severe acidosis 35 (33,3%) and acute pulmonary edema 20 (19%). ICU mortality was 68,5%, increased to 95,2% when more than 2 organs were involved.

Conclusions: Our experience suggests that indications for inititation of RRT in ICU are not greatly different from that in industrialized countries, the big difference is in high mortality rate among our patients.

Su190 URINARY NEUTROPHIL GELATINASE-ASSOCIATED LIPOCALIN IS AN EARLY PREDICTIVE BIOMARKER OF ACUTE KIDNEY INJURY FOLLOWING CONTRAST ADMINISTRATION IN PATIENTS WITH DIABETES – RELATED CHRONIC KIDNEY DISEASE

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Introduction and Aims: Diabetes-related chronic kidney disease is an important risk factor for the development of acute kidney injury (AKI) after contrast agents administration. Currently, AKI diagnosis is established using serum creatinine but it may take be delayed several days. Therefore, we hypothesized that neutrophil gelatinase-associated lipocalin (NGAL) produced in tubule cells in response to injury is a predictive biomarker of AKI after contrast administration which could be useful for early diagnosis in patients with diabetes-related chronic kidney disease (CKD).

Methods: We prospectively enrolled 46 patients (mean age 54 years, range 40–81) with diabetes – related CKD undergoing imagistic examinations with contrast administration. Serial urine samples, obtained at baseline and at multiple time points after contrast administration, were analyzed for NGAL expression in a double-blind fashion using ELISA. The primary endpoint was AKI, defined as an increase in serum creatinine levels >50% from baseline.

Results: AKI was found in 12 pts (26.1%), but diagnosis by serum creatinine was possible only 36–72 hours after contrast infusion. There was no correlation between the GFR and uNGAL levels (r = – 0.348, p=0.4) before contrast administration. NGAL urine levels showed no difference from baseline in the remaining pts without AKI. At a cut-off value of 150ng/ml, the 4-hour urine NGAL revealed the highest sensitivity and specificity (83.3% and 88.2% respectively) in predicting AKI. The area under the ROC curve of NGAL to predict AKI was 0.921 at 4 h after contrast infusion. Both 4- and 6-hour urinary NGAL measurements proved comparably useful biomarkers. By multivariate analysis, NGAL urine concentration (R^2=0.52, p<0.0001) at the 4-hour time point emerged a powerful independent predictor of AKI. Patient demographics and contrast volume were not predictive of AKI.
Conclusions: Increased NGAL urine concentration is an early predictor of AKI following contrast administration. Using this biomarker of renal dysfunction, earlier therapeutic intervention may be possible, particularly in pts at high risk for renal insufficiency.

Su191 CONTINUOUS VERSUS INTERMITTENT RENAL REPLACEMENT THERAPIES IN CRITICALLY ILL PATIENTS: IS THERE ANY DIFFERENCE?

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Introduction and Aims: Acute Kidney Injury (AKI) is a major clinical problem in critically ill patients, with impact on morbidity and mortality. Approximately 5% of such patients require renal replacement therapy (RRT). Continuous renal replacement therapy (CRRT) is often the preferred choice over intermittent renal replacement therapy (IRRT), mainly because hemodynamic stability. A new approach in renal replacement therapy is the slow extended dialysis (SLED), which combines the advantages of CRRT and intermittent hemodialysis. This technique offers a valuable alternative to the classical dialysis strategies in the intensive care patient. However, controversy still exists as to what constitutes optimal RRT in this setting. The aim of this study was to compare CRRT with SLED, to establish if any of these techniques is superior to each other in critical care patients with AKI.
Methods: We performed a prospective observational study in one Intensive Care Unit (ICU), from January 2007 to December 2008. For each patient, we collected demographic data; SAPS II and SOFA scores at the admission; criteria for the initiation of RRT; hemodynamic and ventilation parameters; fluid balance and outcomes. AKI was defined using the RIFLE classification.
Results: Eighty-six patients were enrolled (male gender: 57%; age 57.0±16.2 years): 59 received CRRT, 16 received SLED and 11 were treated with mixed RRT schedules (SLED, CRRT). We didn't observed significant differences among groups concerning admission SOFA and SAPS II scores or in the criteria for the initiation of RRT. However, patients who received CRRT needed higher dosage of noradrenalin (1.28±1.59 μg/kg/min vs 0.51±0,70 in SLED patients and 0.26±0.36 in mixed RRT patients, p=0.016) and began RRT more frequently on *Failure* (66.1 vs 37.5% in SLED patients and 81.8% in mixed RRT patients, p=0.042). Mortality rate was also higher in this group (72.9 vs 31.2 in SLED patients and 36.4% in mixed RRT patients, p=0.002).
On a second analysis, all the confounding factors were eliminated (patients previously on hemodialysis programs, who received mixed RRT schedules and with a RRT length lesser than 3 days) and 52 patients were included: 44 received CRRT and 8 received SLED. Patients treated with CRRT had more hemodynamic instability at admission, but SOFA and SAPS II scores were similar. The evolution of the hemodynamic and ventilation parameters was different among the groups on the first 3 days of RRT: despite the more negative fluid balance in patients on CRRT, there was not a concomitant favorable evolution of the PaO2/FiO2 ratio. Mortality was also higher in the CRRT patients (75 vs 25%).
Conclusions: Our data does not support that CRRT is superior to SLED in critically ill patients. Despite the differences in hemodynamic stability at initiation of RRT, organ dysfunction and severity scores were similar among the groups. Indeed, patients on SLED did better than patients on CRRT, with a lower mortality rate and a more favorable evolution of ventilation parameters. More prospective studies directly comparing these two techniques will help defining the role for each modality in the ICU.

Su192 BLOODLESS CARDIAC SURGERY IN JEHOVAH'S WITNESSES: INCIDENCE OF ACUTE KIDNEY INJURY AND MORTALITY COMPARED WITH A CONTROL GROUP

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Introduction and Aims: Acute kidney injury (AKI) is one of the most serious complications occurring after cardiac surgery (ACS) and is associated with increased mortality. Cardiac surgery often requires blood transfusion but some patients, such as Jehovah's Witnesses refuse to use blood products. The objective of this study is to evaluate the incidence of AKI (defined by RIFLE and AKIN) and mortality in Jehovah's Witnesses undergoing cardiac surgery compared with a control group.
Methods: All Jehovah's Witnesses undergoing cardiac surgery in a Madrid University Hospital, between January 2003 and July 2009 were retrospectively evaluated (n=67). All these patients refused any form of blood transfusion by signing a special consent. Data were prospectively collected. We evaluate the incidence of AKI using RIFLE and AKIN classification and in-hospital mortality. The statistical package SPSS (version 15 for windows) was used. Crude and adjusted dates for age, type of surgery and baseline renal function (CKD< ó >60 ml/min) are expressed.
Results: 67 patients (mean age 62.2±10.9 years; 53.7% females; mean Cleveland Score 3.66±1.6 and SRI score 2.13±0.93, 83.6% valve surgery and 16.4% coronary artery bypass) were evaluated. Mean baseline creatinine 1.06±0.29 mg/dl and maximum creatinine 1.13±0.32 mg/dl.

Table 1. Studied group

	Jehovah's Witnesses	Control Group	p value
Age	62.19	65.95	0.281
Type of surgery			
Coronary artery bypass	16.4%	30%	0.022
Valve	83.6%	70%	
Baseline renal function			
MDRD >60 ml/min	67.2%	68.5%	0.89
MDRD <60 ml/min	32.8%	31.5%	

The incidence of AKI defined by AKIN classification in Jehovah's Witnesses was 13.4% (n=9) and in the control group 16.6% (n=100) (p=0.603). Defined by RIFLE the incidence was 9% (n=6) in Jehovah's Witnesses and 7.2% (n=43) in the control group (p 0.619). Mortality in Jehovah's Witnesses was 10.4% (n=7) and in the control group 8% (n=48) (p 0.481). Crude and adjusted dates were not statistically significant.
Mortality was 10.4% in Jehovah's Witnesses and 8% in control group (p 0.48).

Table 2. Results

	Jehovah's Witnesses	Control group	Crude p	Adjusted p
AKIN	9 (13.4%)	100 (16.6%)	0.603	0.360
RIFLE	6 (9%)	43 (7.2%)	0.619	0.640
Mortality	7 (10.4%)	48 (8%)	0.481	0.36

Conclusions: The Jehovah's Witnesses group seems to be quite similar to the control group: 1. We found no significant differences in AKI incidence (defined by RIFLE or AKIN) between Jehovah's Witnesses and the control group. 2. Inhospital mortality was not different in the two groups.

Su193 TIME IS TUBULE; CONSEQUENCES OF DELAY IN ACUTE KIDNEY INJURY REFERRAL

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Introduction and Aims: Acute Kidney Injury [AKI] is common in hospitalised patients and may account for 2- 3% of all admissions and is associated with severe consequences. The recent NCEPOD UK (National Confidential Enquiry into Patient Outcomes and Death) report on AKI quoted 21% of patients in the study had a delay in referral to a nephrologist.

Abstract Su193 – Table 1

Referral Delay	AKI stage			In hospital Outcome	2 Year Outcome
	Pre-referral	At Referral	Post-Referral		
<1 day, n=27 (20%)	Stage 1, n=8 (30%); Stage 2, n=5 (18%); Stage 3, n=14 (52%)	Stage, 1 n=8 (30%); Stage 2, n=5 (18%); Stage 3, n=14 (52%)	Stage 1, n=7 (26%); Stage 2, n=5 (18%); Stage 3, n=15 (56%)	Death 8 (30%); Dialysis Dependant 0; Full Recovery 13 (48%); Partial Renal Recovery 5 (19%); No Renal Recovery 1 (4%)	Dialysis Dependant 0; Death 16 (59%)
≥1 day, n=108 (80%)	Stage 1, n=59 (55%); Stage 2, n=16 (15%); Stage, 3 n=33 (30%)	Stage, 1 n=50 (46%); Stage 2, n=15 (14%); Stage 3, n=43 (40%)	Stage 1, n=42 (39%); Stage 2, n=13 (12%); Stage 3, n=53 (49%)	Death 25 (23%); Dialysis Dependant 1 (1%); Full Recovery 44 (41%); Partial Renal Recovery 25 (24%); No Renal Recovery 13 (12%)	Dialysis Dependant 4 (4%); Death 61 (57%)

The aim of our study was to determine whether a delay in referral by more than 1 day was associated with worsening of the AKIN staging or poor outcome and to evaluate prognostic factors for AKI mortality.

Methods: Hospitalized patients referred to the nephrology service over six months from July 2007 to January 2008 were studied. Serial Serum Creatinine values obtained at 48 hour intervals, prior to and throughout admission were used to estimate the AKIN stage prior to referral, at referral and post referral. Statistical analysis was performed on their demography and outcome at discharge and up to 2 years thereafter. Multivariate analysis was performed on various prognostic factors.

Results: Among the 135 patients referred with AKI, the median age was 74 (range 25- 96) and males numbered 81 (60%). Ninety nine patients (73%) had known CKD.

Twenty patients out of 75 (27%) referred late progressed from AKI stage 1 or 2 to Stage 3 whereas only 1 in 13 (8%) patients referred early showed similar progression (p value =0.12). Thirty nine patients out of 108 (36%) referred late had poor outcomes (Death, Dialysis dependence and no renal recovery) on discharge against 9 of 27 (33%) referred early with similar outcomes (p value=0.83). *Presence of sepsis, odds ratio 2.6, p value< 0.05 and use of diuretics, odds ratio 3.0, p value< 0.05 were strong independent predictors of 1 year mortality overall.*

Conclusions: Early referral within 24 hours of AKI is associated with less patients progressing to higher AKI stages and having poor outcomes at discharge (a trend towards statistical significance), but the mortality outcomes at 2 years are not different likely due to the fact that the more than half of the early referrals belonged to AKI stage 3 at referral. More importantly this study emphasizes the overwhelming need for attention to the treatment of sepsis and avoidance of diuretics in at risk population which can potentially minimize poor outcome in AKI.

Su194 COMPARING CYSTATIN C WITH AKIN CRITERIA FOR INTENSIVE CARE UNIT AKI

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Introduction and Aims: Recent epidemiological studies suggest a significant association of small increases in serum creatinine (sCr) with adverse outcomes. AKIN group sought to increase the sensitivity of AKIN criteria to AKI by recommending that a small change in serum creatinine. Recent several studies revealed that serum cystatin C (cysC) is more accurate to GFR than sCr.

To investigate the incidence and outcomes of ICU AKI by comparing AKIN criteria to small increment of cysC (= or > 0.3 mg/L).

Methods: This was a prospective cohort study of 274 consecutive incident patients to ICU. Clinical data including urine output, sCr, cysC and outcomes were collected up to 3 months. Kaplan-Meier curve was used to determine 90-days survival. Mortality was adjusted by *Cox's proportional hazard model.* SPSS, 14.0.

Results: AKI was developed in 84 (30.7%) patients by AKIN criteria. In AKIN group, 42 patients (50%) had stage I, 8 (9.5%) had stage II, and 34 (40.4%) had stage III. 13 patients with increased cysC, did not define AKI by AKIN criteria (Only cysC group). Overall 90-days mortality was 20.8% and stratified by each group was 5.7% for control group, 28.6% for Only cysC, 33.3% for AKIN stage I, 62.5% for stage II, 70.6% for stage III (p<0.001). On Kaplan-Meier curve, each group were associated with 90-day survival by stages. On Cox's analysis, AKIN criteria and increment of cysC was associated with mortality. Only cysC gruop has worse prognosis than control group in spite of no AKIN-diagnosed AKI. (HR 4.4 P=0.027).

Conclusions: Small increment of cysC is associated with increased mortality in ICU patients who are not diagnosed AKI by AKIN criteria. CysC is superior to AKIN criteria for predicting AKI and outcomes.

Su195 SERUM URIC ACID LEVEL AS A MARKER FOR MORTALITY AND ACUTE KIDNEY INJURY IN PATIENTS WITH ACUTE PARAQUAT INTOXICATION

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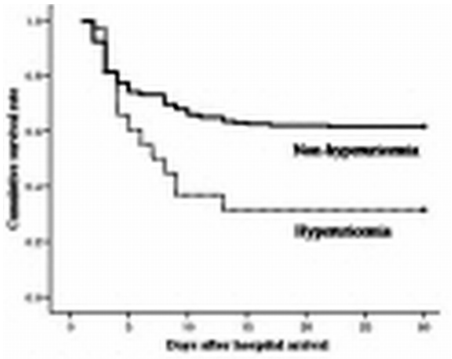
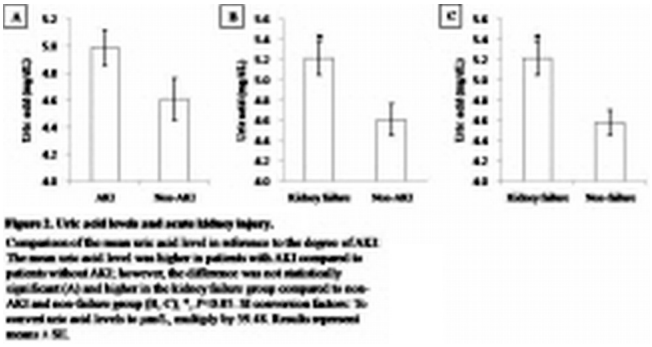
Introduction and Aims: Paraquat (PQ) is a non-selective herbicide that generates reactive oxygen species *in vivo*. Uric acid emerged as a marker of oxidative stress, and may enhance ROS-mediated injury in acute PQ intoxication. Therefore, we investigated the association between uric acid levels and mortality and acute kidney injury (AKI) in the present study.

Methods: From January 2007 to December 2008, patients who arrived at our hospital with acute PQ intoxication (*n*=247) were included in the study. Patients were divided into two groups (hyperuricemia vs non-hyperuricemia) based on uric acid levels. Mortality and acute kidney injury were analyzed in reference to uric acid level.

Results: *Uric acid as a marker for mortality:* A total of 247 patients were included in the study. The crude mortality rates were 68.3% in the hyperuricemia group, 38.3% in non-hyperuricemia group, and 43% of the study patients died. The hyperuricemia group had a lower cumulative survival rate than the group without hyperuricemia [Figure1]. Hyperuricemia significantly increased the risk of mortality after adjustments for age, gender,





and the estimated amount of PQ ingestion. The adjusted odds ratio for mortality was 3.67 (95% CI, 1.35-9.98) with hyperuricemia.
Uric acid as a marker for AKI: The mean uric acid level of the AKI group was higher than in the non-AKI group; however, this difference was not statistically significant [Figure 2A]. The failure group was significantly higher when compared to non-AKI and non-failure group [Figure 2B, 2C]. After adjustments for age, gender, and the estimated amount of PQ ingestion, hyperuricemia still was significantly associated with an increased risk of kidney failure (OR, 3.30; 95% CI, 1.33-8.20).
Conclusions: Baseline serum uric acid level might be a good clinical marker for patients at risk of mortality and AKI after acute PQ intoxication.

Su196 BONE-MORPHOGENIC PROTEIN-7 LEVEL DECREASES SIGNIFICANTLY IN PATIENTS WITH CONTRAST-INDUCED NEPHROPATHY

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Introduction and Aims: Previous studies have demonstrated that endogenous Bone morphogenic protein-7 (BMP-7) level is reduced in acute kidney injury and administration of exogenous BMP-7 has a beneficial effect on kidney function. Even preventive management, contrast-induced nephropathy (CIN) is still the third cause of acute deterioration of kidney function is hospitalized patients. The aim of this study is to test the hypothesis that BMP-7 level might have a role in the development of CIN.
Methods: We enrolled 45 consecutive adult patients with a baseline serum creatinine $\geq$1.4 admitted for coronary angiography. We measured serum BMP-7 levels before and 48 hours after coronary angiography. The primary end point was the development of CIN, defined as an increase in serum creatinine concentration $\geq$ 0.25 mg/dl increase or 25% over the baseline value within 72 hours from contrast exposure.
Results: Overall, CIN occurred in 8 (17%) patients. The concentrations of serum BMP-7 levels were significantly decreased in the CIN group compared to baseline (488.6$\pm$56.8 vs. 356.4$\pm$24.8, p=0.01), in contrast, the concentration of BMP-7 levels were not changed in patients without CIN compared to baseline (444.6$\pm$54.6 vs. 440.0$\pm$53.9, p=0.09).
Conclusions: BMP-7 level significantly decreases in CIN in patients after coronary angiography. Therefore, BMP-7 might be a dignostic biomarker for diagnosis of CIN and BMP-7 might be a promising agent for treatment of CIN.

Lab methods, progression & risk factors for CKD, nutrition in CKD, renal diseases (except GNs and cystic diseases) 2

Su197 METABOLOMIC ANALYSIS OF UREMIC TOXINS IN CHRONIC RENAL FAILURE RATS BY LIQUID CHROMATOGRAPHY/MASS SPECTROMETRY

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Introduction and Aims: The uremic syndrome is considered to be caused by retention of metabolites in the body, due to kidney dysfunction, which are normally excreted into urine. The retention metabolites responsible for the uremic syndrome are called uremic toxins. Recently, the research on uremic toxins arouses interest, even if the problem is an old matter, thanks to the application of new analytical methodologies.
We applied the metabolomic analysis of comprehensive small-molecular metabolites using liquid chromatography/quadrupole ion trap time-of-flight mass spectrometer (LCMS-IT-TOF) to identify and quantify uremic toxins in the serum of chronic renal failure (CRF) rats.
Methods: Five-sixths of the normal kidney mass was removed from 6-week-old male Sprague-Dawley rats to make animal models of CRF.
After analysis of CRF and normal rat serum using LCMS-IT-TOF, principal component analysis (PCA) was done to make ranking list of metabolites associated with CRF rat. Identification of these metabolites was confirmed by comparing m/z, fragment ion patterns and retention times with those of authentic compounds.
For quantification of uremic toxins, selected reaction monitoring (SRM) analysis was carried out using a triple quadrupole mass spectrometer (API4000) equipped with an electrospray ionization source.
Results: A profiling software tool was used to create arrays of mass intensity and retention time pairs which consist of 7241 ions in positive ion mode and 7475 ions in negative ion mode in all the samples. There were 461 positive ions and 423 negative ions, of which peak intensity was statistically (p value $<$ 0.05) higher in CRF rat serum than in normal rat serum.
PCA showed indoxyl sulfate was the first principal metabolite in negative ion mode which differentiates CRF from normal, followed by phenyl sulfate, hippuric acid and p-cresyl sulfate, although hippuric acid was only identified in positive ion mode.
We measured the serum levels of these metabolites and demonstrated these serum levels were increased according to decline in renal function.
Conclusions: The metabolomic analysis using LC-MS and PCA is useful to select and identify serum metabolites that differentiate CRF from normal. By using the method, indoxyl sulfate was found to be the most principal metabolite, followed by phenyl sulfate, hippuric acid and p-cresyl sulfate.

Su198 ★ THORACIC AORTA INVOLVEMENT IN CHRONIC PERIAORTITIS (RETROPERITONEAL FIBROSIS): IDENTIFICATION OF A NEW DISEASE SUBSET

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Introduction and Aims: Chronic periaortitis (CP), also known as retroperitoneal fibrosis, is a rare disease, characterised by a fibro-inflammatory tissue surrounding the abdominal aorta which often causes obstructive renal failure. CP is considered by some authors a systemic autoimmune disease. Anecdotal reports have shown that the thoracic aorta and its major branches may also be involved. We explored the frequency and patterns of involvement of the large thoracic arteries in the largest series of CP patients considered so far, and compared the clinical characteristics and outcome of patients with and without thoracic involvement.
Methods: We enrolled 52 consecutive CP patients who, at the time of diagnosis of CP, underwent imaging studies considered to be appropriate to assess the inflammatory involvement of the thoracic vessels, such as chest angio-CT/MRI, whole-body ^{18}F-fluorodeoxyglucose (FDG) positron emission tomography (PET) or CT-PET. All patients also underwent clinical examination and routine laboratory tests including erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP). They were all treated with prednisone (PDN) for one month and then continued with tapering PDN and/or other drugs (e.g. tamoxifen, methotrexate); 90% of them were enrolled in two randomised controlled trials. Remission was defined as the absence of symptoms and the normalisation of ESR and CRP. The median follow-up was 28 months.
Results: CP involved the abdominal aorta and the iliac arteries in all patients. Twenty-three patients (44%) also showed thoracic vessel involvement: 17/23 (74%) had thoracic periaortitis, which developed around an aneurysmal thoracic aorta in 7 patients and also involved the epiaortic vessels in 6 patients; 6/23 (26%) had a thoracic aortic aneurysm without periaortitis. Histological examination of the thoracic aortic wall, available in 3 patients, showed a pattern similar to that found in retroperitoneal periaortic biopsies, with fibrosis and lymphomonocytic inflammation, organised in lymphoid aggregates with a core of CD20+ cells and a periphery rich in CD4+ cells. Compared with those without thoracic involvement, patients with thoracic

disease showed a predominance of female gender (52% vs 17%, *p=0.016*), a more advanced age at diagnosis (median, 65 vs 52 years, *p=0.001*) and a higher frequency of systemic symptoms (91% vs 62%, *p=0.002*). After the first month of PDN, remission was achieved in 88% of patients with and 100% of those without thoracic involvement (*p=0.038*); in addition, remission was more sustained in the latter group *(p=0.049)*.
Conclusions: This is the first study showing that in a large proportion of patients CP also involves the thoracic aorta and its major branches, with patterns ranging from periaortitis to thoracic aortic aneurysm. This subset of patients has distinct clinical features, such as female predominance, a more advanced age at diagnosis, a higher frequency of systemic symptoms and a higher risk of relapse.

Su199 INVOLVEMENT OF SERUM INDOXYL SULFATE IN EXACERBATION OF RENAL ANAEMIA IN NON-DIALYSIS CKD PATIENTS: A CROSS-SECTIONAL STUDY

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Introduction and Aims: Role of uremic toxins in exacerbating renal anaemia is still unclear. In CKD patients, serum indoxyl sulfate, a uremic toxin, is markedly accumulated and was shown to accelerate the progression of renal dysfunction and vascular disease. We conducted a retrospective observational study to assess the involvement of serum indoxyl sulfate in renal anaemia in non-dialysis CKD patients.
Methods: Study design: Cross-sectional study. Setting & Participants: Consecutive 505 non-dialysis CKD patients were recruited from single outpatient nephrology clinic at referral hospital from February through June 2007. Patients with infection, chronic inflammatory disease, under treatment with steroids or immunosuppressant were excluded. Predictors: serum indoxyl sulfate. Outcomes and other measurements: Relationship between predictor variables and serum hemoglobin was assessed using multiple linear regression analysis. Factors considered potential confounders were age, sex, diabetic status, estimated GFR (eGFR), albumin, CRP, interleukin-6, intact-PTH, transferrin saturation, ferritin, erythropoietin use, and AST-120 administration.
Results: Participants were predominantly man (54.3%), and non-diabetic (79.8%); participant mean age was 63.4 years [SD, 15.8]. Mean eGFR was 43.5 ml/min/1.73m^2 [SD, 26.9], mean hemoglobin was 12.3 g/dL [SD, 1.9], and median indoxyl sulfate was 0.30 mg/dL [25th-75th percentiles, 0.14-0.63]. To examine the effects of indoxyl sulfate on serum hemoglobin levels, multiple regression analyses were performed. Sex, age, diabetic status, eGFR, indoxyl sulfate, albumin, EPO use, transferrin saturation, and ferritin were selected as explanatory variables by the stepwise method. The goodness-of-fit of this model was high (R^2 = 0.52). In multiple regression analysis, log-transformed indoxyl sulfate was significantly and negatively associated with serum hemoglobin level adjusted for covariates. Standardized partial regression coefficient for indoxyl sulfate and eGFR were -0.19 ($p<0.001$) and 0.22 ($p<0.001$), respectively. Variance inflation factors of this model were less than 2.7, indicating absence of multicollinearity problem.
Conclusions: After adjusted for eGFR, indoxyl sulfate was independently and negatively correlated with serum hemoglobin level. These results suggested that accumulation of indoxyl sulfate may exacerbate renal anaemia in non-dialysis CKD patients.

Su200 IMPACT OF ELEVATED INTACT PARATHYROID HORMONE ON MORTALITY AND RENAL DISEASE PROGRESSION IN PATIENTS WITH STAGE 3 AND 4 CHRONIC KIDNEY DISEASE

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Introduction and Aims: Investigating the impact of elevated Intact Parathyroid Hormone (iPTH) levels on mortality and renal replacement therapy (RRT) in patients with stages 3 & 4 Chronic Kidney Disease (CKD) identified by using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation.
Methods: A retrospective cohort analysis from January, 1996 to September, 2007 was conducted in the GE Centricity database with 5,170 CKD Stage 3 & 4 patients using the CKD-EPI equation to estimate their glomerular filtration rate (eGFR). The patients' highest iPTH level was used to define the index date and cohort. Cohorts were separated by iPTH levels and followed for 1-year. Mortality and RRT events, used as a composite endpoint, were evaluated among cohorts at pre-defined iPTH levels. Crude rate ratios (CRR) were calculated using Poisson regression and adjusted rate ratios (ARR) were calculated using Poisson regression after adjusting for age, gender, & estimated eGFR.
Results: Baseline characteristics showed that as the iPTH levels increased, the mean age, number of females and eGFR decreased. Patients with iPTH $<$ 50 were defined as the reference group. Mortality and RRT ARR for iPTH 51-110 was 1.00 (Confidence Interval (CI) 0.07-1.00), iPTH 111-199 was 1.20 (CI 0.89-1.86), iPTH 200-299 was 1.73 (CI 1.40-2.07), iPTH 300-399 was 2.50 (CI 2.11-2.88), iPTH 400-499 was 3.18 (CI 2.77-3.58), and iPTH $\geq$ 500 was 2.25 (CI 1.90-2.59). Below are results using the CKD-EPI equation.
Conclusions: It was determined that iPTH levels greater than 50 pg/mL up to 400-499 pg/ml in patients with Stage 3 & 4 CKD are associated with an escalating combined risk of mortality or RRT in patients with stage 3 and 4 CKD identified by utilizing the CKD-EPI equation. This study demonstrates the importance of monitoring CKD Stage 3 & 4 patients for secondary hyperparathyroidism. Further studies are needed to determine the potential impact of treatment of secondary hyperparathyroidism.
Disclosure: Drs. Marx, Khan, Sterz, and Audya are employees of Abbott, the manufacturer of paricalcitol. Drs. Asche, Jaewhan, and Unni are employees of University of Utah contracted by Abbott Laboratories to conduct this analysis.

Abstract Su200 – Table 1

Index PTH Level	n=8,972	Patient Years	Per 100 Patient Years				
			RRT Events	Mortality Events	All Events	CRR* (95% CI)	ARR* (95%CC)
PTH <50	2,349	2,525	0.67 (17)	8.60 (217)	9.27 (234)	Reference	Reference
PTH 51-110	2,816	3,033	1.09 (33)	9.49 (288)	10.58 (321)	1.01 (0.17-1.00)	1.00 (0.07-1.00)
PTH 111-199	1,769	1,889	2.33 (44)	13.66 (258)	15.99 (302)	1.11 (0.83-1.40)	1.20 (0.89-1.86)
PTH 200-299	816	877	3.99 (35)	16.19 (142)	20.18 (177)	2.17 (1.85-2.49)	1.73 (1.40-2.07)
PTH 300-399	397	462	5.63 (26)	19.49 (90)	25.12 (116)	3.04 (2.68-3.41)	2.50 (2.11-2.88)
PTH 400-499	267	278	7.56 (21)	20.15 (56)	27.70 (77)	3.79 (3.41-4.17)	3.18 (2.77-3.58)
PTH ≥500	558	604	8.45 (51)	16.73 (101)	25.17 (152)	2.93 (2.62-3.25)	2.25 (1.90-2.59)

*CRRs calculated using Poisson regression and ARRs calculated Poisson regression adjusted for age, gender and eGFR using the CKD-EPI equations.

Su201 THE BIOIMPEDANCE UTILITY IN THE EVALUATION OF THE HYDRATION STATUS IN NON-DIALYSIS CKD PATIENTS

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Introduction and Aims: Although overhydration is involved both in car-

diovascular and nutritional complications of patients with chronic kidney disease not on dialysis (ND-CKD), currently there is no validated method to clinically evaluate the hydration status. The utility of bioimpedance spectroscopy using a portable whole body device (BCM, Fresenius Medical Care), has been investigated in dialysis patients with promising results and might be also useful in ND-CKD patients.
To evaluate the hydration status of ND-CKD patients using BCM.
Methods: We evaluated the hydration status using bioimpedance and echocardiography (inferior vena cava diameter). Overhydration was defined as >1 liter by BCM and an inferior vena cava diameter >11mm/m^2. The nutritional status was evaluated clinically by Subjective Global Assessment (SGA) and using BCM (lean and fat total mass, body cellular mass).We investigated 142 ND-CKD patients (M/F 49/51%, mean age 63±15years, 45% over 65 years), with vascular nephropathies (58%) and glomerulonephritis (27%) as main causes of CKD (only 7% diabetic nephropathy), 49%, 7%, 28%, 15% in stage 5, 4, 3 and 2 CKD, respectively. High blood pressure was common (83% of patients), ACEIs were administrated in 44% and diuretics in only 23%. The nutritional status was good (73% SGA="A"; 50% had a serum albumin >4g/dL) and 60% of patients had a PCR>10mg/L.
Results: Most of the patients (88%) were overhydrated; mean overhydration estimated by BCM was 3±2L (0-10L). Overhydration was negatively correlated with GFR (r^2=0.2; p<0.0001) and was positively associated to hypertension (r^2=0.02, p=0.04) or IVC diameter (r^2=0.66, p<0.0001). Meanwhile, overhydration was also related to nutritional parameters, negatively with lean total mass (r^2=0.14, p<0.0001) and SGA "A" vs. "B" (r^2=0.04, p=0.01). In multivariable analysis, in a model which explained 66% of variation in hydration status, only inferior vena cava diameter made a significant contribution (p<0.0001).
Conclusions: In patients with advanced CKD and a high prevalence of hypertension in which diuretics were underused, overhydration was associated with lower GFR, hypertension and protein energy wasting (PEW) according to bioimpedance measurements but not to SGA. BCM and inferior vena cava diameter seem to be useful in the evaluation of the hydration status of ND-CKD. BCM seems superior to SGA in the PEW diagnosis in overhydrated ND-CKD.

Su202 ★ COMPARISON OF ESTIMATING GFR FORMULAS IN ELDERLY

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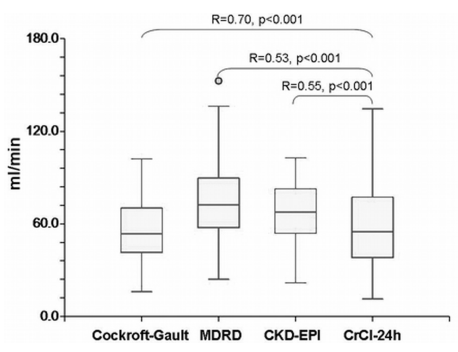
Introduction and Aims: Despite currently use, 24 hours creatinine clearance (CrCl24H) is difficult to employ to estimate GFR in elderly. Although many easy formulas are currently available to estimate GFR in adults, their use in elderly is not validated. We conducted this study to better understand the relationship between estimating CrCl24H and GFR formulas in elderly.
Methods: Design: Cross-sectional study. We enrolled 72 patients with age 65-100 years who did not have polycystic kidney disease. Renal function was estimated by CrCl24H, Cockroft-Gault, MDRD and CKD-EPI formula. Renal dysfunction was considered as CrCl24H < 60 ml/min. We classified estimated GFR (eGFR) according to best cut-off. Spearman correlation was used to correlate estimating GFR formulas.



Results: The mean age was 79.9±6.9 years, male sex 44.4%, diabetes 29.2%, hypertension 80.6%. serum creatinine was 0.98±0.42 mg/dl, CrCl 52.0±28.0 ml/min, Cockroft-Gault 56.2±20.2 ml/min, MDRD 77.3±29.4 ml/min, CKD-EPI 67.6±19.8 ml/min, body surface area (BSA) 1.70±0.22 m^2. Renal dysfunction was present in 41 pts (56.9%). The highest correlation was found between CrCl24H and Cockroft-Gault formulas (figure 1).
Sensibility (Se), specificity (Sp), positive (PV+) and negative (PV-) predictive value of eGFR for renal dysfunction are reported in Table 1. ROC curve analysis show that discriminatory power of Cockroft-Gault < 52.3 ml/min (AUC 0.86, p<0.001) was higher than discriminatory power of MDRD < 86.9 ml/min (AUC 0.71, p<0.001) and CKD-EPI< 64.9 ml/min (AUC 0.70, p<0.001).

Diagnostic tests of three estimating GFR formula for renal dysfunction

Patients with:	Se (%)	Sp (%)	PV+ (%)	PV– (%)
Cockroft-Gault < 52.3 ml/min (n=34)	78.0	93.5	94.1	76.3
MDRD < 86.9 ml/min (n=50)	87.8	54.8	72.0	77.3
CKD-EPI < 64.9 ml/min (n=26)	53.7	87.1	84.6	58.7

Conclusions: In elderly, Cockroft-Gault formula is a stronger predictor of renal dysfunction than MDRD and CKD-EPI formulas.

Su203 SIGNIFICANCE OF HYPERURICEMIA AS A RISK FACTOR FOR PROGRESSION OF NON-PROTENURIC LOW eGFR IN GENERAL HEALTH CHECK POPULATION ON THE COHORT STUDY DURING FIVE YEARS

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Introduction and Aims: The recent years, there are some reports referring to the fact that uric acid is related as the predictor to hypertension, metabolic syndrome (Mets) and chronic kidney disease (CKD). CKD is defined as the abnormality of urine, especially proteinuria and/or low eGFR (eGFR<60mL/min/1.73m^2). In the general health check population, non-proteinuric low eGFR individuals were not observed scarcely. But the pathogenesis and the risk factor for the progression of low eGFR were not clarified in this group. So we evaluated the risk factor for non- proteinuric low eGFR by retrospective cohort study during 5 years.
Methods: In general population 71574 subjects (Male;50327, Female;21247) who had the first health check at Toranomon Health Management Center during 1985-2005, 6617 individuals (Male 4986;age 45.1±7.8, Female 1631;47.5±7.8) was selected for their first health check without low eGFR and had been rechecked every year during 5 years. The value of eGFR was calculated by using the formula for Japanese people.
The prevalence and risk factor to low eGFR who had not proteinuria at the first health check, was evaluated. As the related factor to low eGFR, body mass index (BMI), blood pressure, triglyceride (TG), HDL-cholesterol (HDL-C), fasting plasma glucose (FPG), and serum uric acid (SUA) were examined by the multivariate cox proportional hazard model. Moreover cumulative incidence of low eGFR was studied during 5 years on the quartile of uric acid level at the first check for participants who had not low eGFR.
Results: In the first health checked subjects, low eGFR individual was observed in 11.1% in male and 8.7% in female and in low eGFR individuals, 88.9% showed in male and 91.3% in female showed non-proteinuria.
In the multivariate logistic regression analysis for male, odds ratio (OR) for BMI (>23kg/m^2), systolic blood pressure (>123mmHg), TG (>107mg/dl), HDL-C (<49mg/dl), FPG (>93mg/dl),and SUA (>5.9mg/dl) were as follows respectively,1.34 (p<0.001),0.99 (p=0.736), 1.03 (P=0.497), 1.23 (p<0.001), 0.87 (p=0.001), 2.79 (p<0.001) after adjusted age. That is obesity, low HDL-C, and high uric acid were significant risk factors in male. In female, OR for SUA (>4.2mg/dl) and FPG (>88mg/dl) were 2.75 (P<0.001) and 0.86 (p=0.032), respectively and others showed no significant. The risk of Mets to low eGFR was 1.28 (p<0.001) in male but in female no significance result was observed. The OR for SUA was the highest. So, SUA was the strongest risk factor of low eGFR in male and in female.
By the cohort study, Kaplan-Meier curves for cumulative incident rate of low eGFR on quartile of uric acid levels at the first check without low eGFR, Q4 (SUA>6.5mg/dl) showed the highest hazard ratio (HR): 1.531 (95%CI 1.15-2.03)with significance (P<0.0034) compared to Q1

(<5.1mg/dl). The HR for Q2 (5.2-5.8mg/dl) and Q3 (5.9-6.5.mg/dl) were 1.260 (0.935-1.699) and 1.298 (0.957-1.762) after adjusted age and serum creatinine, respectively.
Conclusions: High uric acid is an independent predictor for non- proteinuric low eGFR in male and female.

Su204 CanPREDDICT: CANADIAN STUDY FOR THE PREDICTION OF DIALYSIS, DEATH AND CARDIOVASCULAR RISK IN CKD PATIENTS

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Introduction and Aims: The CanPREDDICT Study is a prospective observational cohort study of 2500 persons known to nephrologists across 26 centers in Canada. The primary objectives CanPREDDICT are to 1) describe the levels and temporal evolution of 5 known biomarkers representing inflammation (CRP, IL-6), cardiac injury (Troponin I), heart failure (ProBNP), and vascular health (ADMA); 2) establish whether any of these biomarkers, singly or in combination, can accurately discriminate between patients at high and low risk of renal or cardiovascular outcomes; 3) identify novel urine and serum biomarkers of cardiac or renal disease progression using genomic, proteomic, and metabolomic techniques.
Methods: We used multivariate logistic analysis, adjusting for age, sex, race and GFR level, to determine traditional laboratory parameters and biomarkers associated with history of DM, IHD and CHF at baseline.
Results: 2542 CKD pts with eGFR 15-45 ml/min were enrolled during 2008 and 2009. The mean age of the cohort is 68yrs, the median eGFR is 28ml/min (20% < 20mil/min, 38% 20-29mil/min and 41% 30-45mil/min), 62% are male and 90% are Caucasian. The prevalence of DM is 48%; 34% pts. have history of Ischemic Heart Disease (IHD) and 27% history of Congestive Heart Failure (CHF).
Table 1 presents biomarkers associated with comorbidities at baseline:

	Diabetes	Ischemic HD	Congestive HF
Variables	OR (95% C.I.)	OR (95% C.I.)	OR (95% C.I.)
Age (5 yrs)	1.02 (0.97-1.06)	1.22 (1.17-1.29)	1.19 (1.11-1.28)
Sex (Male vs. Female)	1.25 (1.01-1.57)	1.69 (1.36-2.09)	0.86 (0.63-1.18)
Race (Other vs. Caucasian)	1.26 (0.90-1.76)	0.90 (0.63-1.27)	0.71 (0.44-1.14)
Diabetes	n/a	1.94 (1.58-2.38)	ns
eGFR (5 ml/min)	1.10 (1.01-1.17)	1.04 (0.98-1.11)	1.17 (1.07-1.28)
Phosphate (0.1 mmol/L)	1.11 (1.06-1.16)	ns	ns
iPTH (log pmol/L)	ns	ns	1.22 (1.01-1.47)
Albumin (g/L)	0.96 (0.94-0.99)	ns	ns
Hemoglobin (5g/L)	0.94 (0.90-0.97)	ns	ns
IL6 (>LLD- 6.0pg/ml vs. <LLD)	1.56 (1.20-2.03)	1.35 (1.04-1.75)	ns
IL6 (>6.0 pg/ml vs. <LLD)	2.17 (1.66-2.83)	1.42 (1.09-1.84)	ns
Troponin I (>LLD vs. <LLD)	1.61 (1.28-2.02)	1.79 (1.44-2.23)	2.36 (1.70-3.28)
ProBNP (1000 pg/ml)	ns	1.08 (1.04-1.12)	1.18 (1.10-1.26)

The addition of biomarkers to multivariate models with traditional parameters improved *c*-statistic for DM (63% to 67%), IHD (69% to 73%) and CHF (67% to 75%) models.
Conclusions: With limited resources and increasing identification of CKD through laboratory testing and heightened awareness, methods which will facilitate the identification of high risk populations are essential so that appropriate interventions can be tested and implemented. Despite recognition that not all CKD progresses, there is currently no reliable method by which to predict patients at high risk for progression of CKD and CVD. CanPREDDICT results will help to improve our understanding of referred patients; represents an important national study for the community.
Disclosure: The study is supported by Jensen-Ortho Inc. grant.

Su205 ASSESSING GLOMERULAR FILTRATION RATE IN HOSPITALIZED PATIENTS. A COMPARISON BETWEEN MDRD-4 MS AND FOUR CYSTATIN-C BASED EQUATIONS

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Introduction and Aims: Estimation of glomerular filtration rate (GFR) is central to the evaluation and management of hospitalized patients but at present no method exists which would be applicable to all patients. It requires an accurate method for its measurement. Serum creatinine and urinary creatinine clearance have important restrictions, and neither the MDRD-4-ID-MS equation nor equations based on cistatin C have been formally validated on hospitalized patients. The aim of our study was to analize the accuracy, bias and precission of the MDRD-4 ID-MS equation and of four equations based on CsC.
Methods: We studied a representative sample of 3114 hospitalized patients. We collected demographic, clinical and biochemical data of each patients. We measured serum creatinine and cystatin levels in the first three days after being admitted to the hospital. We analized the accuracy, bias and precission of the MDRD-4 ID-MS equation and four equations based on Cystatin, using iohexol clearance as a gold standard.
Results: MDRD-4 ID-MS and equation 3 based on CsC (including both creatinine and cistatin levels) significantly overestimated the GFR. In both cases the error arose from the total estimated muscular mass which depended on the nutritional state of the patients, after adjustment for age, sex and IMC. In patients with stable renal function, GFR < 90 ml/min/1.73 m, without malnutrition, reduction of corporal surface nor loss of muscular mass, MDRD-4 ID-MS provided an adequate estimation of the GFR in all age groups. In the presence of malnutrition, loss of body surface or muscular mass, the cistatin equations, age and sex provided more accuracy in the estimations of GFR.
Conclusions: These results suggest that in hospitalized patients it seems appropriate to use equations based on CsC, age and sex for the estimation of GFR, which, in spite of being affected by factors not related to the GFR, are much less dependent on the nutritional state and the muscular mass of the patient.

Su206 ★ N-ACETYLCYSTEINE (NAC) PROTECTS RENAL FUNCTION IN AGING RATS BY DOWNREGULATING p53 AND UPREGULATING KLOTHO

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Introduction and Aims: Increased oxidative stress play a major role in the process of cellular aging. Senescence is associated with progressive atherosclerosis, the underlying of vascular and the principal cause of morbidity and mortality worldwide. Depletion of inorganic phosphate (Pi) reserves occurs frequently in aged animals and can result in diminished bone mineralization and osteoporosis. This altered Pi balance results from a reduction in intestinal Pi absorption and an elevation in renal Pi excretion. Upregulation of the protein p53 is observed in the kidney of aging male rats. The Klotho protein is an antiaging hormone which confers protection against oxidative stress and regulates mineral metabolism. Previous studies have shown that the antioxidant NAC protects the kidney against the progression of chronic renal failure. NAC also improved the activity of lipoprotein lipase, reducing blood lipids.
The aim of this study was to evaluate the effects of NAC on renal function, serum cholesterol, urinary phosphate excretion, p53 and Klotho expression in kidney tissue of aging rats.
Methods: Normal 8 mo-old male Wistar rats were treated or not with NAC (600mg/L in drinking water) and monitored for 16 months, after which clearance studies were performed in two groups (n=6 each): control (C) and NAC. We measured inulin clearance (GFR); arterial blood pressure (BP), serum thiobarbituric acid reactive substances (TBARS, a marker of oxidative stress), serum cholesterol, proteinuria and urinary phosphate excretion. In addition, immunohistochemical staining for p53 and Western blotting for Klotho protein were performed in kidney tissues.
Results: At baseline, the groups presented similar values for body weight (BW), creatinine clearance (C=0.34±0.02; NAC=0.35±0.02 ml/min/100gBW), proteinuria (C=11±2; NAC=10±1 mg/day), TBARS (C=3.2±; NAC=3.1±0.4 nM/mL) and cholesterol (C=63±7; NAC=74±12 mg/dL). Results at 2 years of age are illustrated in Table 1.
Mean daily water intake, BW and BP were similar in both groups. p53 score was 0.78±0.10 in control rats and was decreased in NAC group (0.36±0.05, p<0.01). The Klotho expression was increased by NAC (C=1.16±0.28; NAC=1.87±0.13).

Table 1

Aged (2yrs)	Control	NAC
GFR (ml/min/100gBW)	0.32±0.01	0.42±0.03**
TBARS (nM/mL)	10.6±0.7	8.4±0.3*
Proteinuria (mg/day)	20.2±2.6	12.5±1.9*
Serum cholesterol (mg/dL)	155±22	98±3*
Urinary phophate (mg/day)	34.5±3.0	17.7±5.3*

*p<0.05; **p<0.01.

Conclusions: NAC treatment significantly improved GFR while decreased TBARS, proteinuria, serum cholesterol, and urinary phosphorus excretion. p53 was downregulated by NAC treatment while Klotho expression was upregulated in NAC-treated rats, suggesting that the protective effect of NAC was mediated by these two proteins. These findings have significant clinical implications for renal protection against senescence, hyperlipidemia and mineral metabolism.

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Su207 OBSTRUCTIVE SLEEP APNEA IN NON-DIALYSIS CHRONIC KIDNEY DISEASE PATIENTS

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Introduction and Aims: Recently, obstructive sleep apnea (OSA) has been widely recognized as an important risk factor for cardiovascular disease. Although a high prevalence of OSA was reported in ESRD patients, there is little information regarding OSA in non-dialysis CKD patients. The aim of this study was to examine OSA in non-dialysis CKD patients and elucidated the relationship between the prevalence of OSA and renal function.

Methods: *Study design:* Cross-sectional study.

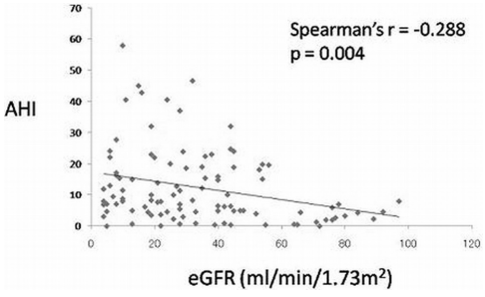
Setting & Participants: Consecutive patients who were hospitalized mainly for CKD educational program or renal biopsy from June 2009 to November 2009.

Predictors: Age, sex, body mass index (BMI), diabetes mellitus and eGFR calculated from serum creatinine using the formula for Japanese.

Outcomes: Prevalence and the degree of OSA.

Measurements: Apnea-hypopnea index (AHI) measured by type 3 (cardiorespiratory) monitor devices (Morpheus; Teijin Pharma Ltd.).

Results: One hundred patients (men 68%), median (interquartile) age 66.0 (56.5-75.0) years, BMI 23.1 (21.1-25.2) kg/m^2, eGFR 28.5 (15.3-45.0) ml/min/1.73m^2 and 31% with diabetes mellitus were enrolled. Overall, 67% of the subjects were suffering from OSA (mild (5≤AHI<15): 34%, moderate (15≤AHI<30): 24%, and severe (30≤AHI): 9%). In multivariate logistic regression analysis, lower eGFR was significantly associated with increased prevalence of OSA adjusting for age, sex, BMI and diabetes mellitus (adjusted odds ratio 10.5, 95% confidence interval 1.2 to 108.7). Moreover, AHI inversely correlated with eGFR (figure) and this relation was maintained after adjusting for covariates (p=0.03).



Conclusions: Our data demonstrated that the prevalence of OSA was also high in non-dialysis CKD patients and suggested the vicious cycle between the decline of GFR and OSA.

Su208 ESTIMATION OF GLOMERULAR FILTRATION RATE IN TYPE 2 DIABETIC PATIENTS USING EQUATION BASED ON BOTH SERUM CYSTATIN C AND CREATININE

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Introduction and Aims: The estimation of Glomerular Filtration Rate (GFR) by the Modification of Diet in Renal Disease (MDRD) equation, which is based on serum creatinine (Scr), has recognised limitations. Serum cystatin C (Scys) has been proposed as a potential replacement for Scr in GFR estimation. So, equations using Scys alone or in combination with Scr provide an alternative method. We compared the MDRD equation to a Scyst-Scr based formula for GFR estimation in patients with type 2 diabetes.

Methods: We studied 270 participants with type 2 diabetes, of whom 113 (42%) were men, with the following characteristics, mean (SD): age 65 (10) years, BMI 30.8 (5) kg/m^2, HbA1c 6.8 (1.4)%. GFR was measured using plasma clearance of 51Cr-EDTA (mGFR). In parallel, GFR was estimated twice, using the MDRD formula (MDRDGFR) and the Stevens equation which is based on both Scr and Scyst [cyst/crGFR: 177.6* SCr−0.65 * CysC−0.57 * age−0.20 * (0.82 if female)]. Estimated GFR results were compared with isotopic GFR by means of two-tailed, paired t tests and by Levene's test for equality of variance. Bland-Altman plots were obtained. All calculations were performed using SPSS 17.00. p<0.05 was taken to indicate statistical significance.

Results: MGFR was 70.2 (19.9) ml.min^{-1} 1.73 m^{-2}. MDRDGFR was 89.0 (24.9) ml min^{-1} 1.73 m^{-2} and cyst/crGFR was 80.2 (25.1) ml min^{-1} 1.73 m^{-2} (p<0.05 for difference from mGFR). Bland-Altman plots showed that 95.2% and 96.1% of estimations for MDRDGFR and cyst/crGFR respectively, lie within the ± 1.96SD of the mean difference. Bias (mean difference between estimated GFR and mGFR) was 18.6 and 10.0 ml min^{-1} 1.73 m^{-2} for MDRDGFR and cyst/crGFR respectively (p<0.05 for difference in bias between MDRDGFR and cyst/crGFR). Precision (SD of the bias) was 17.6 and 16.5 ml min^{-1} 1.73 m^{-2} for MDRDGFR and cyst/crGFR respectively (NS difference in precision between MDRDGFR and cyst/crGFR). Accuracy 10% (proportion of estimated GFR results within 10% of mGFR) was 29.6% and 44,6% for MDRDGFR and cyst/crGFR respectively (p<0.05 for difference in accuracy 10% between MDRDGFR and cyst/crGFR). Accuracy 30% (proportion of estimated GFR results within 30% of mGFR) was 75.9% and 88.9% for MDRDGFR and cyst/crGFR respectively (p<0.05 for difference in accuracy 30% between MDRDGFR and cyst/crGFR).

Conclusions: Cystatin C-creatine based formula was less biased and more accurate to within 10% and 30% of isotopic GFR than MDRD. These results support further evaluation of cystatin C for estimation of GFR in patients with type 2 diabetes.

Su209 ASPIRIN RESISTANCE IN PATIENTS WITH CHRONIC KIDNEY DISEASE

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Introduction and Aims: Patients with chronic kidney disease frequently undergo thrombotic cardiovascular events, but the relationship between these increased thrombotic events and aspirin resistance is poorly defined in chronic kidney disease patients. We hypothesized that an increased incidence of aspirin resistance would be associated with high prevalence of thrombotic vascular events in chronic kidney disease patients who had received aspirin and investigated the incidence of, and factors associated with, aspirin resistance in patients with chronic kidney disease.

Methods: Between December 2008 and August 2009, 91 chronic kidney disease patients who had taken aspirin alone or aspirin plus clopidogrel

daily for ≥ 7 consecutive days were included. Chronic kidney disease was defined as estimated glomerular filtration rate < 60 ml/min/1.73 m^2 using the abbreviated Modification of Diet in Renal Disease formula. Aspirin resistance was measured using the VerifyNow Aspirin Assay and compared with patients with normal kidney function (N = 111). Aspirin resistance based on the VerifyNow Aspirin Assay was defined as an ARU ≥ 550 in a patient taking aspirin.

Results: Aspirin reaction unit was 462±61 U in the chronic kidney disease patients compared with 434±56 U in patients with normal kidney function (P = 0.002). Twenty-three of 202 patients (11%) were found to be aspirin resistant based on the criterion of aspirin reaction unit (≥ 550). The prevalence of aspirin resistance was significantly higher in the chronic kidney disease patients than in patients with normal kidney function (18% vs. 6%, P = 0.012). The presence of chronic kidney disease (odds ratio [*OR*] 3.17, 95% confidence intervals [*CI*] 1.24–8.09, P = 0.016), lower hemoglobin (*OR* 0.74, 95% *CI*, 0.60–0.92, P = 0.007), lower serum albumin (*OR* 0.38, 95% *CI* 0.17−0.86, P = 0.021) and the use of angiotensin-converting enzyme inhibitors (*OR* 2.89, 95% *CI* 1.20−6.99, P = 0.018) were associated with aspirin resistance in the univariate logistic regression analysis. However, only the presence of chronic kidney disease and the use of angiotensin-converting enzyme inhibitors remained significant after multivariate analysis (chronic kidney disease, adjusted *OR* 2.96, 95% *CI* 1.04–8.41, P = 0.042; angiotensin-converting enzyme inhibitors, adjusted OR 3.39, 95% CI 1.34–8.60, P = 0.010).

Conclusions: The incidence of aspirin resistance was higher in patients with chronic kidney disease than in patients with normal kidney function. In addition, the presence of chronic kidney disease and the use of angiotensin-converting enzyme inhibitors were significant predictors for aspirin resistance. Thus, although aspirin is highly effective at reducing cardiovascular events in patients with normal kidney function, it may be less beneficial for patients with chronic kidney disease and the high incidence of aspirin resistance in patients with chronic kidney may contribute to their high incidence of thrombotic events.

Su210 MATRIX METALLOPROTEINASES (MMPs) AND ITS INHIBITORS IN CHRONIC GLOMERULONEPHRITIS (CGN): ASSOCIATION WITH CLINICAL ACTIVITY AND PROTEINURIC REMODELING OF RENAL TUBULOINTERSTITIUM

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Introduction and Aims: Recent studies show that renal function reduction in CGN associates with tubulointerstitial fibrosis (TIF). The dysbalance between MMPs and its inhibitors is widely discussed as a main mechanism of extracellular matrix (ECM) accumulation and TIF formation. But contribution of proteolysis disturbances (not only defective ECM degradation) to proteinuric remodeling of tubulointerstitium (TI) is poorly studied.

The aim of our study was to estimate urinary excretion, TI expression of MMPs, its inhibitors, to reveal associations with CGN clinical activity, TI inflammation and TIF.

Methods: We examined 86 CGN pts: (I group) 23 pts with moderate proteinuria (PU); (II group)-26 pts with nephrotic syndrome (NS) without renal failure (RF); (III group)-22 pts with high PU and transient RF, (IV group)–15 pts with PU and permanent RF (CRF). 12 healthy subjects were investigated as controls. MMP-2, MMP-9, tissue inhibitor of MMP type 2 (TIMP-2) expression were evaluated in 62 renal biopsy specimens. Cellular inflammatory infiltration (CII), TIF area were assessed by morphometria. Urinary level of MMP-2, MMP-9, TIMP-2, PAI-1, TGF-β1 were measured by ELISA.

Results: MMP-2, MMP-9, TIMP-2 were expressed in renal tissue of CGN pts. We observed intensive TI expression of named factors in CGN with high clinical activity (II and III group). Exactly in these pts we revealed significant urinary MMPs and TIMP-2 elevation. MMPs, TIMP-2 renal expression and urinary levels did not associate with CGN morphological type. There were positive correlations between MMP-2, MMP-9 urinary level and PU (r=0,76 and r=0,49 p<0,001) and interstitial CII (r=0,79 p<0,05). Interstitial MMP-9 and TIMP-2 expression in NS directly correlated with CII (r=0,67 p<0,001 and r=0,72 p<0,001).These data reflect MMP/TIMP system involvement into the inflammation (including PU induced) perhaps through regulation of cytokines, growth factors degradation. MMPs urinary excretion and renal expression significantly decreased in CRF. There were indirect correlations between urinary MMP-2 and serum Cr value (r=-0,83 p<0,05), urinary MMP-9 and TIF area (r=-0,58 p<0,05). Urinary PAI-1 greatly increased in CGN progressive forms, especially in CRF. There were correlations: negative-between urinary MMPs and PAI-1, direct-between urinary PAI-I and TGF-β1 (r=0,77 p<0,05) and TIF degree (r=0,61 p<0,01). Our results confirmed the role of impaired proteolysis in the TIF formation, CGN progression. In pts with RF we revealed massive MMP-2 depositions in the proximal tubular cells, which directly correlated with PU (r=0,57 p<0,05), TIF degree (r=0,52 p<0,05), urinary TGF-β1 (r=0,51 p=0,05). These data may reflect PU induced epithelium changes, which characterized by proteolysis of its cytoskeleton, basal membrane with following epithelium transformation into myofibroblasts.

Conclusions: We suggest that MMPs and its inhibitors disturbances play an important role in the TI inflammation, epithelial transdifferentiation, ECM accumulation and thus contribute to TIF formation and CGN progression.

Su211 CLINICAL SIGNIFICANCE IN THE LOCATION OF CORONARY ARTERY STENOSIS DETECTED BY CORONARY CT ANGIOGRAPHY IN PATIENTS WITH CHRONIC KIDNEY DISEASE

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Introduction and Aims: Coronary artery disease is common in chronic kidney disease (CKD). It has reported that pathologic mechanism could be various in calcification of the vascular tree. We evaluated renal function and coronary artery calcium scores according to the location of coronary artery stenosis in CKD patients.

Methods: We enrolled 224 CKD patients who underwent 64-slice multidetector row computed tomography angiography, and had estimated glomerular filtration rates (eGFR) over 40 ml/min/1.73m^2. Subjects were categorized into 3 groups; patients with no stenosis (NS), patients with proximal stenosis (PS), and patients with distal or diffuse stenosis (DS).

Results: Forty-one (18%) patients had significant (>50%) diameter stenosis (17 in PS group vs. 24 in DS group). PS group had lower renal function and higher proportion of proteinuria than DS or NS group (eGFR: 69.9±16.5 in PS vs. 83.0±18.9 in DS vs. 83.1±18.1 mL/min/1.73m^2 in NS, P=0.016; proteinuria: 21.4% in PS vs. 9.5% in DS vs. 2.9%, P=0.007). In contrast, DS group had higher levels of coronary artery calcium score (CACS) than PS or NS group (911±423 vs. 328±100 in PS vs. 70.1±17.2 in NS, P < 0.001). Atherosclerotic plaques were identified in 152 (67.8%, 1.43 segments/subject) individuals. Proportion of non-calcified plaques was higher in the PS group than in the DS group (P=0.012).

Conclusions: Our results showed that proximal coronary artery stenosis may be associated with poor renal function and non-calcified plaques, and that distal or diffuse stenosis may be associated with higher CACSs. It suggests that different therapeutic approach in the location of coronary artery stenosis could be needed in CKD patients.

Su212 EL-MINIA EQUATION: A MODIFIED EQUIATION FOR IMPROVEMENT OF ESTIMATION OF GLOMERULAR FILTRATION RATE IN EGYPTIAN PATIENTS WITH CHRONIC KIDNEY DISEASE

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Introduction and Aims: Estimation of GFR is limited by differences in creatinine generation among ethnicities. CKD-EPI equation was developed for estimation of GFR in white and African Americans, but evaluation of its validity in El-Minia Governorate patients with chronic kidney disease (CKD) indicated that modification of this equation is necessary for application in El-Minia patients with CKD. *Aim:* is to modify CKD-EPI equation on basis of data from El-Minia CKD patients and compare the performance of the modified CKD-EPI equation (*El-Minia Equation*) with original CKD-EPI

equation and other published equations using measurement of GFR by isotopic GFR (iGFR) as a reference value.

Methods: The study included 285 CKD patients; 182 were males, age 46±12 years, body weight 85±19 kg, body surface area (BSA) 2±0.2 m^2; BMI 29±6 kg/m^2. creatinine 1.9±0.9 mg/dl, BUN 31±13 mg/dl. iGFR 44±19 ml/min/1.73m^2, as BSA represents body mass better than simple body weight so we assumed that the performances of CKD-EPI equation could be improved in El-Minia patients by adding a corrective factor that was extracted from BSA estimated by Mosteller formula, so our suggested formula is:

$eGFR\ (ml/min/1.73m^2) = (CKD\text{-}EPI) \times (BSA)^{0.1}$

Other equations shown in table below were used for comparison with El-Minia equation.

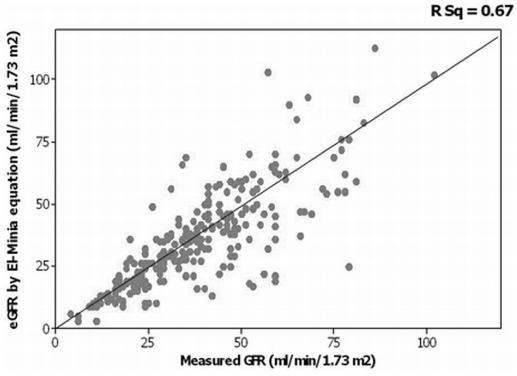
Results: El-Minia equation gave the best performance in eGFR considering error range between ±10%, ±30%, ±50%, and analysis by r^2 showed also that it is the best one for El-Minia Patients with CKD.

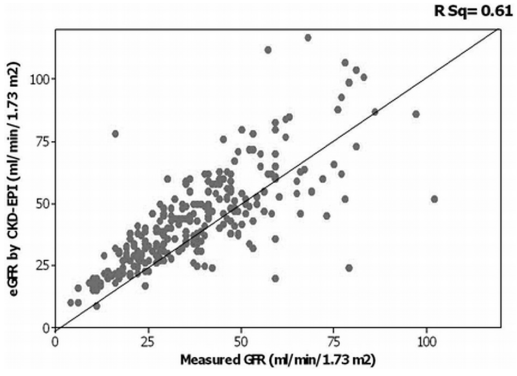
iGFR (measured GFR) and eGFR by El-Minia equation and other equations

	Mean	Range	Median
iGFR (ml/min/1.73m^2)	44±21	8-139	39
eGFR El-Minia equation (ml/min/1.73m^2)	44±19	8-130	38
eGFR CKD-EPI (ml/min/1.73m^2)	41±18	7-123	35
eGFR MDRD (ml/min/1.73m^2)	42±18	9-117	38
eGFR Walser (ml/min/1.73m^2)	44±19	7-116	40
eGFR Mayo Clinic (ml/min/1.73m^2)	47±25	9-144	39
eGFR Nankivell (ml/min/1.73m^2)	49±16	12-115	46
eGFR Cockroft-Gault (ml/min/1.73m^2)	51±20	12-139	46

% of prediction error in all equations

	Within ±10%	Within ±30%	Within ±50%	R2
El-Minia Equation	43	77	93	0.67
CKD-EPI	23	54	79	0.61
MDRD	19	50	73	0.55
Walser	18	48	72	0.58
Mayo Clinic	17	46	63	0.55
Nankivell	15	36	65	0.56
Cockcroft-Gualt	14	34	51	0.55





Conclusions: El-Minia equation is the best one for monitoring kidney functions in Egyptian CKD patients and could be applied in clinical practice, at least in El-Minia patients with CKD.

Su213 COMPARISON OF ESTIMATING GFR FORMULAS TO PREDICT CARDIOVASCULAR EVENTS IN ISCHEMIC HEART DISEASE POPULATION

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Introduction: Glomerular filtration rate (GFR) is a known prognostic factor of cardiovascular (CV) events. Cockroft-Gault (CG), MDRD and the CKD-EPI formulas are used to estimate GFR. However, the prognostic performance of each GFR formula in patients with high CV risk is not well investigated.

Aims: To compare the prognostic performance of each GFR formula to predict CV events in patients with high cardiovascular risk.

Design: Longitudinal study with a follow-up of 730 days. The end-points were: (1) all cause mortality, (2) major cardiac and cerebrovascular events (MACCE) (all-cause mortality, cerebrovascular accident, myocardial infarction and repeated revascularization/coronary bypass) and (3) all CV events (MACCE, angina, acute pulmonary oedema, renal failure and arrhythmia).

Setting: Division of Cardiology, patients affected by ischemic heart disease. SUBJECTS: 999 patients underwent coronary angiography.

Methods: GFR was estimated using the CG, MDRD and CKD-EPI formulas. A standard setting of CV risk factors (age, sex, diabetes and smoking) was used to create a baseline model (Model A). A second model was created for each GFR estimating formula (Model A + GFR). -2 Log Likelihood was used to compare the prognostic performance of each model.

Results: Kaplan-Meier estimates of the rates of all cause mortality, MACCE and all CV events at 2 years were respectively 3.9%, 20.4% and 30.3%. The analysis of -2 Log Likelihood statistics showed that the addition of GFR as estimated by Cockroft-Gault, MDRD and CKD-EPI formula did not improve the prognostic performance of the basic model for predicting all-cause mortality, MACCE and overall CV events (see Table 1).

-2 log Likelihood analysis

	All cause mortality	MACCE	All CV events
Model A	460.22	2585.60	2754.41
Model A + Cockroft-Gault	460.13	2585.12	2753.66
Model A + MDRD	460.18	2585.51	2754.25
Model A + CKD-EPI	459.45	2585.22	2753.81

Conclusions: Among patients with high CV risk, the addiction of GFR did not increase the prognostic performance of standard CV risk factors for all cause mortality, MACCE and all CV events.

Su214 GERIATRIC NUTRITIONAL RISK INDEX AND MENTAL STATUS IN GERIATRIC PATIENTS UNDER HEMODIALYSIS TREATMENT

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Introduction and Aims: Malnutrition is common among hemodialysis (HD) patients. Nutritional screening tools may be useful to identify those patients at nutritional risk among HD patients. A new tool Geriatric Nutrition Risk Index (GNRI) was recently proposed to screen malnutrition in this population. Although much progress has been made in recent years in identifying the causes and pathogenesis of malnutrition in HD patients, the effect of aging and associated mental status changes are not thoroughly investigated.

We aimed to assess the relationship between mental status and degree of malnutrition in HD patients.

Methods: A total of 82 geriatric patients (52 female, 30 males, mean age 74±12 years, range 60-87) were investigated between November 2007 and November 2009. The mean duration of HD was 117.60±73.23 months (range, 14-289 months). We have performed mini-mental state examination (MMSE) test and GNRI test. GNRI scores were categorized as: severe risk (<82), moderate risk (82-91), low risk (92-98) and no risk (above 98). Laboratory, clinical and demographic data were obtained from medical records.

Results: GNRI score was correlated with MMSE (r=0.33, p<0.005), blood urea (r=-0.30, p=0.012), serum creatinine (r=-0.23, p=0.049), total protein (r=-0.43, p<0.001), albumin (r=-0.81, p<0.001), C-reactive protein (r=0.28, p=0.019), hemoglobin (r=-0.25, p=0.034), serum iron binding capacity (r=0.23, p=0.048). According to GNRI score, patients were further divided into 4 groups: Group 1 (GNRI>98, n= 11), group 2 (GNRI=98-92, n=17), group 3 (GNRI<92-82, n=25), group 4 (GNRI<82, n=16). Distribution of mean MMSE values laboratory and metabolic parameters were given in table.

Parameter (serum)	Group 1	Group 2	Group 3	Group 4	P value
MMSE	25,8±2,3	25,9±1,9	23,5±4,0	20,3±3,5	<0.005
Creatinine (μmol/L)	9,6±2,1	8,2±2,0	8,4±2,6	7,4±2,1	=0,007
Potassium (mmol/l)	5,2±0,8	5,3±0,7	5,8±0,9	5,9±0,6	=0,006
Fosfor (mmol/l)	5,7±1,5	5,9±0,9	6,0±0,8	6,1±1,0	<0.001
Total protein (g/l)	7,0±0,6	6,9±0,4	6,4±0,5	6,2±0,9	<0.001
Albumin (g/dl)	4,2±0,2	3,6±0,4	3,5±0,3	3,0±0,5	<0.001
C-reactive protein (mg/l)	5,8±2,4	7,3±4,3	12,6±10,7	13,1±11,8	=0.019
Hemoglobin (g/l)	11,5±1,0	10,4±1,2	10,1±0,9	9,5±1,3	=0.034
Ferritin	87,5±35,2	65,4±38,2	54,7±25,1	35,6±15,4	=0,001

Conclusions: The GNRI is calculated by a very simple equation in which only 3 nutritional variables-serum albumin,heigh and body weight-are involved and the patients at high risk of malnutrition can be referred by numbers. Lowest nutritional scores were noted in patients with decreased cognitive function. Malnutrition is associated with decreased mental function in geriatric HD patients. Assessment of nutritional status and treatment of malnutrition, especially in the elderly HD patients, might prevent cognitive dysfunction in this population.

Su215 PREVALENCE OF ABNORMAL *IN VITRO* CLOSURE TIME (CT) USING PLATELET FUNCTION ANALYZER-100 (PFA-100) IN CHRONIC KIDNEY DISEASE (CKD) PATIENTS AND ANALYSIS OF FACTORS AFFECTING IT

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Introduction and Aims: PFA-100 system evaluates platelet function by determining the time to occlusion of an aperture in a membrane coated with collagen and epinephrine (CEPI) or collagen and ADP (CADP) as citrated whole blood flows.
This study was performed to evaluate prevalence of abnormal *in vitro* closure time using Platelet Function Analyzer-100 in patients with CKD and to analyze factors associdated with it.
Methods: The cross-sectional study was performed on 356 patients with CKD (Stage I 30, II 36, III 30, IV 56, V 204, chronic hemodialysis (CHD) 131, chronic peritoneal dialysis (CPD) 73). CEPI-CT, CADP-CT, CBC, serum creatinine (Cr), blood urea nitrogen (BUN) were measured. Estimated glomerular filtration rate (eGFR) was calculated with modified MDRD equation. We already reported that the normal range of CEPI-CT was 82-182 sec and CADP-CT 62-109 sec in Koreans (Ann Clin Lab Sci 38:247, 2008).
Results: Table 1 shows prevalence of abnormal CT. In predialysis patients, both CEPI-CT and CADP-CT were correlated with BUN (r=0.464, p<0.001, r=0.230, p=0.004), but not with Cr or eGFR. In CHD, CEPI-CT was not correlated with BUN or Cr. In CPD, there were significant correlations with CEPI-CT and BUN or Cr (P<0.001 and 0.005). In both CHD and CPD patients, there were no correlations between CADP-CT and BUN or Cr. Both CEPI-CT and CADP-CT did not correlate with age, gender, hemoglobin, or platelet.

Table 1. the prevalence of of abnormal CT in CKD patients

STAGE	I	II	III	IV	V	CHD	CPD
CEPI-CT	13.3% (4/30)	8.3% (3/36)	16.6% (5/30)	32% (18/56)	53% (109/204)	58% (76/131)	45% (33/73)
CADP-CT	13.3% (4/30)	8.3% (3/36)	6.6% (2/30)	21% (12/56)	50% (101/204)	50% (65/131)	49% (36/73)

Conclusions: We reported prevalence of abnormal CT in predialysis CKD patients for the first time. In CHD and CPD patients, prevalence of abnormal CT were similar with those previously reported. CT was correlated with BUN in predialysis CKD patients.

Su216 CLINICAL BENEFITS OF SEVELAMER TREATMENT ON PRE-DIALYSIS PATIENTS WITH STAGE III-IV CHRONIC KIDNEY DISEASE AND WITH SIGN AND SYMPTOMS OF SECONDARY HYPERPARATHYROIDISM

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Introduction and Aims: Cardiovascular pathologies, chronic inflammatory condition and secondary hyperparathyroidism (hyPTH) are the most frequent complications emerging at the very first stage of chronic kidney disease (CKD). At the same time, the clinical evidence shows that patients classified in Stage I, according to the National Kidney Foundation (NKF), are affected by mineral disorders characterised by parathormone (PTH) blood level alterations and increase of phosphoremia. The fibroblast growth factor (FGF23) is a new sensitive biomarker being increasingly used to monitor the efficacy of pharmacological treatments aimed to prevent the cascade of events leading to calcium/phosphorus dismetabolism. We studied the clinical efficacy of chlorohydrate sevelamer, a calcium- and aluminium-free phosphate chelant agent, on echocardiographic and serum parameters in stage III and IV of CKD patients with signs and symptoms of early secondary hyPTH and cardiovascular comorbidities.
Methods: 170 CKD patients (100M/70F, mean age 50.7±4.3 years, 100/170 [58.8%] with anemia at the baseline visit) were studied. Other secondary hyperPTH modifying drugs were excluded from the study. The efficacy of a 12-month therapy with a daily 1600 mg oral dose of sevelamer has been evaluated by laboratory exams (mainly FGF23, PCR, phosphoremia, calcemia), cardiovascular echocardiographic exams (e.g.: EF, LVEDV) and degree of mitral valve calcification (Wilkins score) at 6 and 12 months after baseline.
Results: After 6 months of treatment, a mean reduction of 15.2% (p<0,0001) of serum phosphate (mg/dl), 19.1% (p<0,0001) of FGF23 (RU/ml) and 31.6% (p<0,0001) of PCR (mg/dl) and a dramatic improvement of cardiovascular tissue deposition (79.3% mean reduction of Wilkins score: p<0,0001) were observed in all patients. By contrast, hemodynamic exams improved slightly (EF 3.1%, LVEDV -2.0%). The degree of improvement of serum phosphorus, FGF23, PCR, EF and Wilkins score after 6 months of sevelamer therapy was comparable among anemic and not anemic subgroups.
Conclusions: The treatment for hyperphosphataemia in stage III-IV of CKD patients has typically focused on the use of oral calcium-based phosphate binders, taken at mealtimes, which bind dietary phosphate. Sevelamer, a non-calcium, non-magnesium, aluminium-free agent, recently become available for use as phosphate binder in patients with end-stage renal disease, has accumulated a robust body of evidence on the effects on all-cause of cardiovascular mortality. Our study confirms published data on serum mineral levels control efficacy of this phosphate binder drug, and draws the attention on its role in reducing chronic inflammation markers (FGF23, PCR) and the sclerotic process of cardiovascular apparatus (Wilkins score).

Su217 SEVERE HYPOPROTEIC DIET SUPPLEMENTED WITH KETOANALOGUES IN PREDIALYZED PATIENTS WITH CHRONIC KIDNEY DISEASE

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Introduction and Aims: The reduced dietary protein intake has been reported for more than a century to improve many uremic symptoms and even to slow down the rate of decline in renal function, thus postponing the initiation of renal replacement therapy (RRT).
A prospective, open-label, parallel, randomized controlled trial aimed to assess the effect of a severe hypoproteic diet supplemented with

ketoanalogues (SVLPD) for 48 weeks on the progression of CKD, as well as on certain metabolic disorders of chronic kidney disease (CKD).
Methods: All the 1524 CKD patients admitted in a Nephrology deparment in 2-years enrollment period were evaluated for selection criteria. Only 155 non-diabetic CKD patients with an estimated glomerular filtration rate (eGFR) < 30mL/min/1.73m^2 (MDRD4 formula), proteinuria < 1 g/g urinary creatinine, good nutritional status and anticipated good compliance to the diet were randomly assigned in two groups. Group I (n=83) received SVLPD (0.3 g/kg per day vegetable proteins and ketonalogues, 1 cps/5 kg per day). Group II (n=82) continued a conventional low mixed protein diet (0.6 g/kg per day). Nitrogen waste products retention, calcium-phosphorus and acid-base disturbances were set as primary efficacy parameters, while "death" of the kidney or of the patient and the eGFR were secondary efficacy parameters. The nutritional status and the compliance to the diet were pre-defined as safety variables. There were no differences between groups in any parameter at baseline.
Results: Only in the SVLPD group serum urea significantly decreased (123.1±37.9 versus 169.2±51.0 at baseline, mg/dL) and significant improvement in: serum bicarbonate (23.4±2.3 versus 17.9±1.3 mEq/L), serum calcium (8.8±0.4 versus 7.8±0.2 mg/dL), serum phosphates (4.5±0.9 versus 6.1±1.9 mg/dL) and calcium-phosphorus product (39.6±8.1 versus 47.6±9.0 mg^2/dL^2) were noted after 48 weeks. No patients' death was registered in any group. Significantly lower percentages of patients in Group I required RRT initiation (6 versus 27%). After 48 weeks, eGFR did not significantly change in patients receiving SVLPD (16.8±4.1 versus 18.9±4.6 at baseline, mL/min per 1.73 m^2), but significantly decreased in controls (13.4±5.1 versus 18.6±4.3, mL/min 1.73 m^2). Patients receiving SVLPD had a lower mean eGFR decline: – 2.7 (-5.2,-0.6) versus -5.2 (-5.3,-2.9) mL/min per year in controls. The compliance to the keto-diet was good in enrolled patients. No significant changes in any of the parameters of the nutritional status and no adverse reactions were noted.
Conclusions: The SVLPD seems to be effective in selected predialyzed CKD patients ameliorating certain manifestations of CRF (the toxin retention, acid-base and calcium-phosphorus metabolism disturbances) and in delaying the RRT initiation, without any negative influence on the nutritional status.

Su218 URINARY EXCRETION OF CYTOCHROME C (Cyc) IS AN EARLY MARKER OF TUBULAR DISFUNCTION IN HIV-INFECTED PATIENTS TREATED WITH NUCLEOTIDE REVERSE TRANSCRIPTASE INHIBITOR (NRTI) TENOFOVIR

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Introduction and Aims: Some NRTI drugs, such as TDF, may cause renal damage and proximal tubular disfunction. Abacavir (ABV) is less commonly nephrotoxic. A selective mitochondrial toxicity of TDF has been hypothesized, but no definitive data are available. Renal damage and decreased renal function may occurr after years of therapy. Aim: to evaluate an early marker of tubular injury in patients treated with TDF, in comparison with ABV-including regimens.
Methods: We enrolled 99 patients shifted from thymidinic backbones to either TDF or ABV-including backbones (respectively 72 and 27). Patients underwent 24-hour urine collection before the shift (T0), after 1 (T1), 3 (T2), 6 (T3) and 12 (T4) months. Samples were analyzed by ELISA for concentration of Cyc as marker of mitochondrial proximal tubular toxicity. Data were corrected for urinary creatinine (UCr) (ng/g). Serum (SPhos) and urinary phosphate (UPhos) were also evaluated. Statistical analysis was performed by parametric or non-parametric ANOVA (Kruskal-Wallis) with post-test comparison among all time points in each treatment group; urinary levels between groups were compared by the Mann-Whitney test.
Results: Patients under ABV regimen showed non-significant increase of urinary Cyc. In the group of TDF-treated, Cyc excretion increased significantly in the first 3 months from the shift, but decreased progressively up to 12 months (T2 vs T0, $p<0.01$). Cyc levels of TDF group were significantly higher than levels of ABV group at T1 and T2. Interestingly, UPhos did not vary in ABV-patients, but increased significantly up to 12 months after the shift to TDF. The higher UPhos values in TDF group, compared with ABV patients, were more evident at 6 and 12 mo. of treatment. SPhos levels and percentage of hypophosphatemic patients were comparable in both groups at all time points.
Conclusions: These findings confirm an involvement of the tubular mitochondria, during therapy with TDF. We hypothesize that although Cyc excretion decreased after three months, a subclinical tubular injury persisted during TDF therapy. The tubular disfunction, manifested by increased UPhos, in some cases, and in concomitance with other factors, may evolve to overt renal tubular damage and decreased renal function. Therefore, increased urinary Cyc excretion may be considered an early laboratory marker of TDF-induced renal toxicity.

Su219 CORRECTION FACTOR FOR e-GFR CALCULATED WITH SCHWARTZ FORMULA TO MATCH WITH MDRD IN ADOLESCENTS

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Introduction and Aims: Estimated glomerular filtration rate (e-GFR) in children and adults is mostly calculated by Schwartz' (e-GFR= k*L/Scr) and MDRD formulas. For adolescents transferred to adult care e-GFR value is calculated by MDRD with frequent discrepancies. Enzymatic creatinine assays, (isotope dilution traceable international standard calibration) is the goldstandard for MDRD and new k for this method was calculated for Schwartz' without solving discrepancies. Aim of this work was to compare e-GFR results obtained by Schwartz' and MDRD in subjects in transition age (15-19 years) to adult nephrologists care.
Methods: We analyzed 582 Caucasian subjects 320 males and 262 females aged 15 to 19 years. e-GFR was calculated using enzymatic serum Creatinine and both Schwartz' formula using the recently published k of 0.413 and MDRD were applied.
Results: In males median e-GFR-Schwartz' was 73.19 ml/min (IQR 53.13-86.78) and MDRD 101.8 ml/min (IQR 17.88-123.9) (Wilcoxon rank test $p<0.0001$; Bland Altman Bias 29.80, SD 17.04, 95% CI -3.59 to 63.20). In females median e-GFR-Schwartz was 79.99 ml/min (IQR 66.95-94.05) and MDRD 92.38 ml/min (IQR 75-110.4), (Wilcoxon rank test $p<0.0001$; Bland Altman Bias 11.72, SD 8.81, 95% CI -5.55-29.00). The two formulas strictly correlated ($r2= 0.77$; $p<0.0001$), but in males a significant increase of e-GFR-MDRD (mean 40% ± 19.5) was observed vs Schwartz' (95% CI slope 0.58-0.61, Y intercept when X=0: 8.72 to -14.24). In females the discrepancy was lower (mean difference of 13%±9) (95%CI slope 0.75-0.78, Y intercept when X=0: 7.24 to 10.27). Assuming a linear mathematical model we obtained for males a new k of 0.58± 0.08 for e-GFR-Schwartz for age 15 to 19 years that made e-GFR-Schwartz comparable to e-GFR-MDRD

Abstract Su218 – Table 1

	T0	T1	T2	T3	T4	ANOVA (p)
ABV (Cyc/UCr)	52.3 (24.1–160.2)	47.2 (17.1–336.1)	262.5 (65–558)	301.2 (36.5–561)	270.3 (16–816)	NS
ABV UPhos	668±220	728±155	691±119	593±130	540±189	NS
TDF (Cyc/UCr)	109.3 (24.3-463)	294.1 (57.3-645)	448.8 (170-952)	223.8 (40.2-561)	159.5 (36.7-626)	0.0052
TDV UPhos	626±183	741±219	700±175	780±342	697±152	0.0001
Cyc/UCr (p)*	NS	0.0201	0.0186	NS	NS	
UPhos (p)*	NS	NS	NS	0.0461	0.0219	

CyC/U Cr (ng/g) median (IQ range); UPhos (mg/g U Cr) mean ± SD; *TDV vs ABV.

(Bland Altman test (Bias 0.14, SD of bias 9.7; CI 95% -18.9 to 19.2). This new k in males was not influenced by height, BSA and age in the period 15-19 years. In males aged 15-19 years e-GFR-Schwartz calculated with k=0.413 can be corrected to obtain e-GFR analogous to MDRD by multiplication by 1.415. (Linear regression r2=0.95, 95%CI slope 0.83-0.87, Y intercept when X=0 12.43 to 16.98). In females at the same age (15-19) we obtained a k of 0.47±0.04 and a correction factor of 1.146 to be applied to the Schwartz' formula with k=0.413. (Bland Altman: bias 0.31, SD of bias 6.3, 95% CI limit from -12.0 to 12.7).

Conclusions: In conclusion we demonstrated a significant difference in e-GFR obtained by Schwartz' and MDRD in the transition age: to allow comparison of the two methods and correct interpretation of data in adolescent investigated in future by MDRD, we propose the adoption of a new k for Schwartz' formula of 0.58 in males from 15 years old and to correct e-GFR calculated with Schwartz' (k=0.413) by 1,415; in females we propose the adoption of k= 0.47 and the correction factor of 1.146 for Schwartz' calculated on k=0.413.

Su220 CENTRAL FAT DISTRIBUTION IS MORE CLOSELY RELATED WITH KEY RISK FACTORS THAN ELEVATED BMI IN PATIENTS WITH CKD STAGE 3

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Introduction and Aims: Body Mass Index (BMI) as a marker of obesity is an established risk factor for chronic kidney disease (CKD) and other co-morbidities such as cardiovascular disease (CVD) and diabetes. BMI is indiscriminate in including fat, muscle and bone mass in its measurement and can overestimate obesity. Waist-to-hip ratio is emerging as a more important risk factor as it is a marker of central fat distribution.

We studied BMI and waist-to-hip ratio in a cohort of patients with CKD stage 3 and compared the results with other known risk factors for CKD progression and CVD.

Methods: 1277 patients with estimated GFR 59-30ml/min/1.73m^2 were recruited from primary care practices. Each participant underwent clinical assessment, including measurement of BMI, waist-to-hip ratio and pulse wave velocity, as well as urine and serum biochemistry tests.

Results: The mean (SD) BMI was 28.8±5 kg/m^2 and mean (SD) waist-to-hip ratio was 0.91±0.09. 35% of the cohort were classified as obese (BMI > 30 kg/m^2) of which 69% were female. 88% of the cohort had central fat distribution (waist to hip ratio >0.8 for females and >0.9 for males) of which 57% were female. Univariate analysis revealed significant correlations between BMI, waist-to-hip ratio and several risk factors.

Risk Factor Correlations with BMI and Waist-to-hip ratio

	BMI		Waist to Hip Ratio	
	r	p value	r	p value
BMI (kg/m^2)			0.14	<0.001
Age (years)	-0.13	<0.001	0.15	<0.001
Uric Acid (μmol/L)	0.17	<0.001	0.10	<0.001
Pulse Wave Velocity (m/s)	-0.12	<0.001	0.12	<0.001
Urine Albumin:Creatinine (mg/mmol)	0.01	NS	0.10	<0.001
eGFR (mL/min/1.73m^2)	-0.01	NS	-0.19	<0.001

Central fat distribution was associated with a higher mean uric acid, pulse wave velocity, albumin:creatinine ratio and a lower mean eGFR (P = <0.05). Obesity defined by BMI was associated with a higher mean uric acid and a lower mean pulse wave velocity (P = <0.001).

Multivariable linear regression analysis identified waist-to-hip ratio, age, systolic blood pressure, diabetes, uric acid but not BMI as independent determinants of albumin:creatinine ratio (P = <0.005). Multivariable linear regression analysis also identified age, diabetes, systolic blood pressure, diastolic blood pressure, BMI (inversely) and waist-to-hip ratio as independent determinants of pulse wave velocity (P = <0.001).

Conclusions: Waist-to-hip ratio is closely related to more key risk factors in CKD stage 3 patients than BMI. Central fat distribution as represented by waist-to-hip ratio may be of greater importance as a risk factor in CKD than BMI and reliance on BMI alone may therefore underestimate the associated risk.

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Su221 INFLUENCE OF PROTEINURIA ON THYROID HORMONE PARAMETERS IN PATIENTS WITH CHRONIC RENAL FAILURE

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Introduction and Aims: Chronic renal failure (CRF) has great impact on thyroid hormone metabolism resulting in low T3 concentrations in patients with low GFR. The degradation of Thyroxin (T4) to T3 and reverse T3 (rT3) is carried out by dejodinase enzymes, which are selenoproteins. Thus, CRF might affect the degradation pathways of thyroid homones in a certain manner. So far, (subclinical) Hypothyroidism has been reported in patients with the nephrotic syndrome. These observations have been explained by urinary losses of thyroid hormone binding proteins such as albumin and TBG (Thyroxine Binding Globulin). However, data on the prevalence of thyroid dysfunction in patients with proteinuria are missing. Therefore, we carried out a prospective study evaluating the influence of various degrees of proteinuria on thyroid hormone parameters in patients with different stages of renal insufficiency (CKD stages 1-5)

Methods: 180 patients with various degrees of renal insufficiency (CKD stages 1-5) not yet on dialysis were prospectively investigated. Besides clinical data, renal function parameters (serum creatinine, GFR, serum albumin) and proteinuria (albuminuria) as well as the following thyroid hormone parameters were measured: TSH, T4, fT4, T3, fT3,TBG and (rT3).

Results: According to albumin excretion patients could be subdivided into: albuminuria <300 mg/gm creatinine (n=79), 301 mg -3gm (n=80) and > 3 gm/gm creatinine (n=21).

T3 and T4 were positively correlated with serum albumin (r= 0.18; p<0.01 and r=0.19; p<0.01 respectively), however not correlated to albuminuria.

TSH, fT4 and fT3 were neither correlated to serum albumin nor to albuminexcretion.

rT3 (normal: 0.18-0.45 ng/dl) was significantly lower in patients with albuminuria > 3 gm/gm creatinine (0.29 ng/dl ±0.08 (mean ± SD) vs. 0.38±0.12); p<0.001). rT3 was negatively correlated to albuminuria (r=-0.28; p<0.0001).

Renal function was strongly correlated with decreasing T3 levels with advancing renal failure (r=0.001) and also with decreasing T4 and fT4 concentrations (r= 0.18; p<0.02) respectively. rT3 showed a negative correlation with renal function (r= -0.19; p<0.02). TSH was not affected by renal function.

Conclusions: In contrast to previous studies proteinuria seems to have only minor effects on thyroid hormone parameters such as T4, fT4 and T3. Only rT3 is inversely correlated to albuminuria with significantly lower rT3 values in patients with albuminexcretion > 3gm/gm creatinine. This suggests a direkt influence of proteinuria on the dejodinase activities, which are the major contributor to the degradation of thyroid hormones.

Decreasing renal function is associated with corresponding lower T3, T4 and fT4 concentrations in patients with CRF not yet on dialysis.

TSH is neither affected by proteinuria nor by renal function.

Su222 UTILITY OF THE MODIFICATION OF DIET RENAL DISEASE (MDRD) FORMULA IN THE POPULATION THE FOLLOW UP OF KIDNEY DONOR

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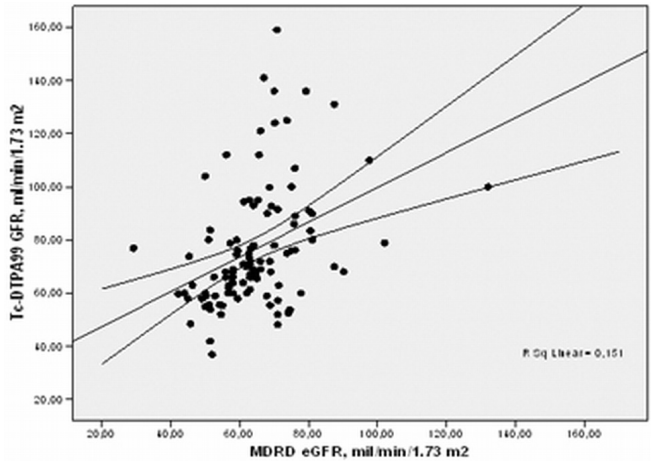
Introduction and Aims: Based on important limitations for the measuring of the glomerular filtration rate (mGFR) by means of isotopic techniques, different formulas have been developed as a result of important epidemi-

ological studies which attempt to mathematically relate diverse variables for the estimation of the renal function. More recently, the most innovative MDRD formula for estimating the eGFR was elaborated. The validation of the MDRD simplified has been studied in several areas; however its utility has not been validated in subjects with unilateral nephrectomy due to renal donation.

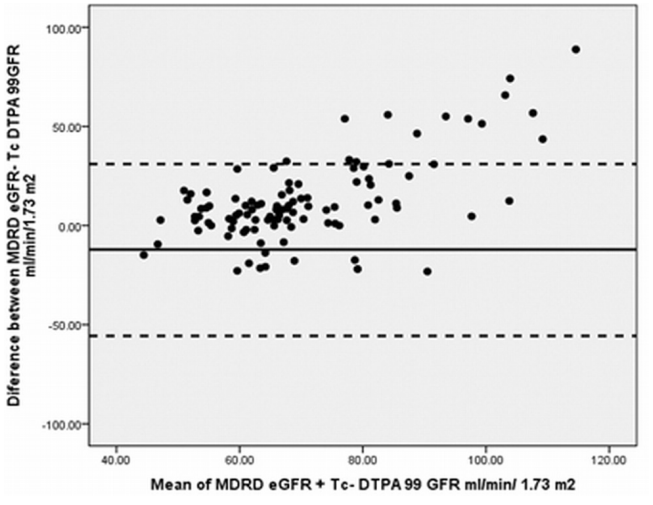
Aim: To evaluate the utility of the MDRD equation as a test for eGFR compared with the mGFR for the Tc-DTPA99^{M} scintigram in kidney donors.

Methods: Adult kidney donors of ≥18 years were included, of either sex or renal donation time. Patients with HAS, DM and pregnant patients were excluded. Estimated GFR values by the MDRD formula were compared with the mGFR by renal scintigram using the Tc-DTPA99^{M}. All SCr values were measured by the Jaffe method (alkaline picrate reaction).

Results: 104 donors were studied, average age 40±10, 61% women, average donating time 33±13; the average GFR by means of the MDRD was 64.4±13.5 and by scintigram 76.5±22.8 ml/min/1.73 m^2, respectively. The relation between the mGFR by scintigram Tc-DTPA99^{M} and the estimated GFR by the MDRD formula and their variability coefficient (r2) are depicted in Figure 1. The correlation coefficient (r) between Tc-DTPA99^{M} and the MDRD formula was 0.38 (p<0.0001).



Agreement: Bland–Altman plots for the differences of eGFR and mGFR against the combined mean values are depicted in Figure 2. The average of the differences between the two tests is -12.09. The limits of agreement for MDRD formulas were -55.21- 31.03ml/min/1.73 m^2.



Conclusions: The MDRD formulas had serious limitations for correct GFR estimation in kidney donors.

Su223 DIFFERENT PREDICTORS OF ADIPONECTINEMIA IN DIFFERENT STAGES OF DIABETIC NEPHROPATHY

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Introduction and Aims: Adiponectin is an anti-iflammatory and antiatherogenic cytokine, increasing with the severity of renal disease. Diabetic patients start out with lower adiponectin levels than controls; increase in adiponectin parallels albuminuria, inflammation and decrease in GFR. Recent experimental data suggest that overexpression of adiponectin might be secondary to early renal damage and could have a protective role.

Methods: 4 cohorts of type 2 diabetic patients were included: normoalbuminuric (n=59), microalbuminuric (n=20), proteinuric (n=29) and dialysis (n=28) patients. Clinical and laboratory data, antropometric measurements, urinary albumin/creatinine, and total adiponectin (ELISA) were obtained; creatinine clearance (Clcr) was estimated (abbreviated MDRD formula). Adiponectin levels were compared to those of 21 age and sex matched healthy controls and of 10 nondiabetic patients on dialysis.

Results: Excepting differences in albumin excretion and higher age in macroproteinuric patients, the three predialysis cohorts were similar and were considered together when compared to dialysis patients. Adiponectin was highest in nondiabetic dialysis controls -39.04±10.11μg/ml, followed by diabetic dialysis patients -21.50±3.25μg/ml, healthy controls -10.34±1.67μg/ml, and predialitic patients -8.00±1.14μg/ml; differences were significant–p=0,0001. Dialysis patients had lower hemoglobin (Hb)-11.19±0.32 vs13.89±0.22 g/dl, p=0.0001; albumin -3.75±0.33 vs 4.64±0.04 g/dl, p=0.0001; and HbA1C -6.79±1,38 vs 7.67±0.21%, p=0.01; but higher CRP -2.26±0.60 vs 0.67±0.20 mg/dl, p=0.002 than the predialysis group. In *normoalbuminuric* patients adiponectin correlated negatively to age-p=0.04 and BMI -p=0,04; BMI remained significant predictor of adiponectin in multiple regression -p=0,03. In *microalbuminuric* patients simple regression disclosed a positive link to CRP -p=0.0008 and negative one to Hb -p=0.04. In multiple regression only CRP predicted adiponectinemia -p=0.004. In *macroproteinuric* patients adiponectin correlated negatively to Clcr -p=0.002, albumin -p=0.001, Hb -p=0.01 and triglycerides -p=0.03, and positively to HDL cholesterol -p=0.01, but the strongest correlation was a positive link to albuminuria -p<0.0001). HDL cholesterol -p=0,04 and albuminuria remained the only predictors of adiponectin in multiple regression -p=0.03. In the *dialysis* patients, adiponectin correlated negatively to CRP -p=0.03, serum triglycerides -p=0.01 and BMI -p=0.03 and directly to HDL cholesterol -p=0.05. In multiple regression triglycerides -p=0.01, BMI -p=0.04 and HDL cholesterol -p=0.04 remaind predictors of adiponectin.

Conclusions: Our data outline the different factors influencing adiponectinemia in different stages of diabetic nephropathy. CRP is most closely linked to adiponectin in early kidney damage, suggesting that inflammation could influence adiponectin rise in these patients. With more advanced renal disease albuminuria (reflecting severity of renal injury) becomes the main predictor of adiponectin levels. In dialysis patients adiponectin levels seem to be predicted mainly by metabolic parameters and BMI.

Su224 PLEIOTROPIC EFFECTS OF VITAMIN D IN STAGE 3 CHRONIC KIDNEY DISEASE. EFFECT ON INSULIN RESISTANCE

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Introduction and Aims: Disturbances of carbohydrates metabolism are common in patients with Chronic Kidney Disease. Possible pathogenetic mechanisms involve unresponsiveness of insulin receptor and/or a blunt response of the beta cell at the stimulus of hyperglycemia. It is known that vitamin D has an important role for in the endocrine functioning of the pancreas, particularly in the insulin release process. The aim of our study was to investigate the effect of vitamin D treatment on insulin resistance in patients with CKD stage 3.

Methods: We included in our study 37 (M/W 25/12) non diabetic patients with CKD of stage 3 (eGFR: 30-59 ml/min/1,73 m^2 calculated with the

Modification of Diet in Renal Disease MDRD formula). In all patients, chronic kidney disease-mineral and bone disorder (CKD-MBD) was established. All patients were treated at the beginning with dietary phosphorous restriction, phosphate binding agents, calcium supplements. If iPTH values were persistently over 70 pg/ml, administration of 1alpha-hydroxyvitamin D3 at a dose of 0,25 μg in a single daily dose was initiated. Fasting glucose concentration, insulin levels, peptide C, HbA1, iPTH, Ca, P and insulin resistance evaluated by HOMA (Homeostasis Model Assessment) index were measured before (T0) and 12 weeks after (T1) initiation of treatment with vitamin D. In all patients were performed an oral glucose tolerance test (OGTT).

Results: Plasma levels of fasting glucose (103±2,3 mg/dl T0 vs 101±2,4 mg/dl T1, p:n.s.) and insulin levels 9,5±1,6 mU/ml T0 vs 9,9±1,8 mU/ml T1, p:n.s) were comparable before (T0) and after treatment with vitamin D (T1). HOMA index was statistically higher in time T0 that T1 (3.4±0.3 vs 2,58±0.4,p=0.017) indicative that insulin resistance was improved. Mean glucose plasma levels 120 minutes after administration of 75 gr of glucose was 153±25 mg/dl T0 vs 134±29 mg/dl at T1 (p=0,024).

Conclusions: According to our data, treatment with vitamin D in patients with CKD stage 3 has beneficial effect on insulin resistance assessed with HOMA index. Further randomized controlled large scale studies are needed in order to assess long term benefits in morbidity and mortality of those patients.

Su225 EFFECTS OF BETA-ADRENERGIC ANTAGONISTS IN PATIENTS WITH ADVANCED CHRONIC KIDNEY DISEASE: A SYSTEMATIC REVIEW AND META-ANALYSIS

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Introduction and Aims: There is a substantial burden of cardiovascular disease and mortality in people with advanced chronic kidney disease (CKD stages 3-5). The role of beta-adrenergic antagonists' (beta-blockers) in dialysis patients is uncertain, so this systematic review aimed to study the effects of beta-blockers on mortality, cardiovascular events, hemodynamic measures, and surrogate cardiovascular markers.

Methods: The Cochrane Controlled Trial Registry, MEDLINE and EMBASE were searched for randomised controlled trials (RCT) with at least 3 months follow up in patients with CKD stages 3 to 5. Summary estimates were obtained using a random effects model and heterogeneity measured using I^2.

Results: Eight trials involving 4,156 participants met the inclusion criteria. Of these, 4 placebo-controlled trials were performed in patients with symptomatic congestive heart failure (1 trial in 114 dialysis patients and *post hoc* analyses of 3 trials in 3,019 pre-dialysis patients). Treatment with beta-blockers reduced the risk of death (risk ratio 0.69, 95% confidence interval 0.60-0.79, P<0.001; test for heterogeneity I^2 0%, P=0.676) in patients with heart failure. There is no data on effects of beta-blockers on mortality and clinical outcomes in CKD patients on dialysis. Summary statistics for other outcomes including heart rate, blood pressure and left ventricular ejection fraction are not reported due to high-level heterogeneity in available data. There is paucity of data on safety and tolerability of beta-blockers in advanced CKD.

Conclusions: Treatment with beta-blockers improves survival in patients with advanced CKD and heart failure but there is paucity of data in patients on dialysis. Adequately powered RCTs are needed in CKD patients receiving dialysis to confirm the beneficial effects of beta-blockers.

Su226 DETERMINANTS OF SKIN AUTOFLURESENCE IN CKD STAGE 3 PATIENTS

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Introduction and Aims: Tissue advanced glycation end products (AGE) accumulation is a measure of cumulative metabolic stress. Assessment of tissue AGE by skin autofluoresence (AF) correlates well with cardiovascular (CV) outcomes in diabetic, transplant and dialysis patients and may be a useful marker of CV risk in earlier stages of CKD.

Methods: 1277 patients with estimated GFR 59-30ml/min/1.73m^2 were recruited from Primary Care Practices. Detailed medical history was obtained and each participant underwent clinical assessment as well as urine and serum biochemistry tests. Skin AF was assessed as a measure of skin AGE deposition using a cutaneous AF device (AGE Reader®, DiagnOptics, Groningen, The Netherlands).

Results: Univariate analysis revealed significant correlations between AF readings and several potential risk factors for CV disease (CVD) (Table 1).

Table 1. Significant Correlations with skin AF

	r	p value
Age (years)	0.220	<0.0001
Calculated GFR (mL/min/1.73m^2)	-0.227	<0.0001
Total Cholesterol (mmol/L)	-.0.175	<0.0001
Hb (g/dL)	-0.227	<0.0001
Albumin (mmol/L)	-0.120	<0.0001
Glucose (mmol/L)	0.134	<0.0001
Diastolic BP (mmHg)	-0.154	<0.0001
PWV (m/sec)	0.105	<0.0001
Waist:Hip Ratio	0.109	<0.0001
IMD Score (Indices of Multiple Deprivation)	0.098	0.001
Alcohol (units/week)	-0.080	0.005
MAP	-0.077	0.007
Uric Acid (mmol/L)	0.070	0.013
HDL Cholesterol (mmol/L)	0.067	0.019

Skin AF readings were also significantly higher among males, diabetics, patients with microalbuminuria, periodontal disease, a history of smoking or evidence of self reported cardiovascular disease.

Multivariable linear regression analysis identified independent determinants of higher skin AF (Table 2; R^2=0.17 for equation).

Table 2. Independent determinants of higher skin AF

	β	p value
Age (years)	0.119	<0.0001
Diabetes	0.147	<0.0001
Haemoglobin (g/dL)	-0.159	<0.0001
Calculated GFR (mL/min/1.73m^2)	-0.105	<0.0001
Periodontal Disease	0.094	0.001
IMD score (Indices of Multiple Deprivation)	0.082	0.002
Ever smoked	0.086	0.002
Previous CVD (self reported)	0.070	0.010
Female gender	-0.071	0.017

Conclusions: Increased skin AF is independently associated with multiple cardiovascular risk markers in CKD 3. Long-term follow up will be conducted to assess the value of skin AF as a predictor of cardiovascular risk in this population.

Disclosure: This study has been funded in part by an unrestricted educational grant from Roche.

Su227 INCREASE IN CIRCULATING RETINOL BINDING PROTEIN-4 LEVEL PREDICTS ENDOTHELIAL DYSFUNCTION IN PEOPLE WITH CHRONIC KIDNEY DISEASE AT ALL STAGES

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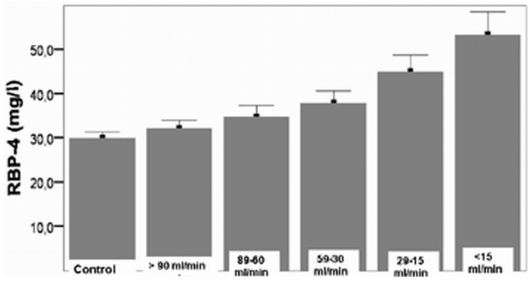
Introduction and Aims: Endothelial dysfunction, the critical step in the mechanism of atherosclerosis, starts in the early stages of CKD. People with CKD have increased blood level of retinol binding protein-4 [RBP-4] which is known to be involved in atherosclerotic process. In the present study, we investigated whether there is an association of circulating RBP-4 level to endothelium dependent vascular dilation [EDVD] in all stages of CKD starting from healthy controls.

Methods: A total of 240 participants were examined. The patient group consisted of 200 individuals with CKD at five stages, and there were about 40 cases in each group. The number of healthy controls was 40. Blood RBP-4 level was determined along with EDVD, high sensitivity CRP [hsCRP], homeostasis model assessment–insulin resistance [HOMA-IR index], and demographic parameters.
Results: RBP-4 blood level was found to increase in parallel with the stage of the disease p=0.000.



Logistic regression analysis revealed that circulating RBP-4 concentration successfully predicted the level of EBVD β: –0.523; p=0.000.
Moreover, the pronounced association was stronger in the advanced stages.
Conclusions: The results indicate that RBP-4 blood level successfully estimates the presence of endothelial dysfunction in CKD. RBP-4 measurement may be a noninvasive and sensitive tool for detecting endothelial function in both clinical research and patient follow-up.

Su228 NUTRITIONAL STATUS IS AN INDEPENDENT ASSOCIATE OF SERUM BETA2-MICROGLOBULIN LEVEL IN CHRONIC HEMODIALYSIS PATIENTS

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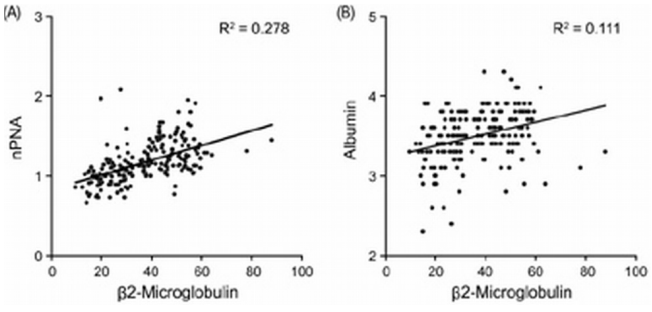
Introduction and Aims: Beta2-microglobulin (β_2-M) has been considered a surrogate marker of putative middle-molecule uremic toxins, which are difficult to dialyze using low-flux membranes. This cross-sectional study evaluated the correlation of serum β_2-M to nutritional parameters in chronic hemodialysis (CHD) patients.
Methods: Laboratory parameters, including β_2-M, albumin, prealbumin, creatinine, blood urea nitrogen (BUN), high-sensitivity C-reactive protein (hs-CRP), lipid battery, KT/V, and normalized protein nitrogen appearance (nPNA), were measured in 201 CHD patients (102 men and 99 women, mean age 63±12 years). Clinical data such as age, sex, duration of hemodialysis, and presence of cardiovascular disease and diabetes mellitus, were also recorded.
Results: The mean serum β2-M concentration was 37.1±14.4 ug/mL. On univariate analysis, β2-M was positively correlated with nPNA (R = 0.528, P < 0.001), creatinine concentration (R = 0.335, P < 0.001), albumin concentration (R = 0.334, P < 0.001), duration of hemodialysis (R = 0.284, P < 0.001), BMI (R = 0.191, P = 0.007), hs-CRP concentration (R = 0.18, P = 0.011), and BUN concentration (R = 0.152, P = 0.031) and negatively correlated with HDL-C concentration (R = -0.148, P = 0.037).



On multiple regression analysis, nPNA (P < 0.001), duration of hemodialysis (P < 0.001), creatinine concentration (P < 0.001), albumin concentration (P = 0.006), BUN concentration (P = 0.011) and HDL-C concentration (P = 0.038) were independent associates of serum β2-M concentration.

Table 1. Multiple regression analysis of factors associated serum β2-microglobulin concentration

Variable	B	β	t-value	p-value
nPNA (g/kg per day)	24.315	0.425	6.737	< 0.001
HD duration (months)	0.053	0.206	3.583	< 0.001
Creatinine	1.322	0.239	3.417	< 0.001
Albumin	7.643	0.169	2.794	0.006
HDL-C	-0.139	-0.120	-2.094	0.038
BUN	-0.133	-0.177	-2.571	0.011

B = unstandardized coefficients, β = standardized coefficients, dependent variable = β2-microglobulin; nPNA, normalized protein nitrogen appearance; HDL-C, high-density lipoprotein cholesterol.

Conclusions: Our results show that nutritional status is an independent associate of serum β2-M concentration in CHD patients.

Su229 DETECTION OF EARLY IMPAIRMENT OF GLOMERULAR FILTRATION RATE: SERUM CREATININE VERSUS LOW-MOLECULAR WEIGHT PROTEINS

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Introduction and Aims: Serum creatinine is considered an efficient marker of GFR impairment only at CKD stages ≥3b (GFR<45 mL/min/1.73 m^2), while serum concentrations of low molecular weight proteins (LMWP) have been proposed as a more sensitive marker of GFR impairment.
Aim of this study was to assess the accuracy of serum levels LMWP (cystatin C, Cys; beta2-microglobulin, β2M; retinol-binding protein, RBP; beta-trace protein, BTP) as indicators of impairment of GFR, in comparison with serum creatinine (Creat) and urinary excretion of albumin (U-Alb).
Methods: Two hundred and ninety-five adult CKD patients (F 137, M 158), affected by different kidney diseases with various impairment of renal function (S-Creat 0.40-12.1 mg/dL), participated to this study. GFR was measured as the renal clearance of 99mTc-DTPA. Creat, Cys, β2M, RBP, BTP and U-Alb were measured with standard laboratory methods.
Results: Serum concentration of all markers increased with the reduction of GFR. The slight increases found at CKD stage 2 resulted already statistically significant versus the values found in the group of CKD at stage 1. Statistical significance was higher for S-Creat, S-β2M and S-BTP than for S-Cys and S-RBP. Some differences among the different markers were found according to the gender of patients. In any case, a very high correlation was found between GFR (99mTc-DTPA), and serum Creat (r=0.9254), Cys (r=0.9361), β2M (r=0.9381), and BTP (r=0.9251). The lowest correlation was that of RBP (r=0.6917). In general, the criterion values to screen the same GFR impairment were higher for male than for female patients. Furthermore, to screen patients with mild impairment in GFR, one must use criterion values lower than those necessary to screen patients with a more advanced impairment in GFR. All serum markers showed a similar accuracy as indicators of different degrees of GFR impairment, except RBP, which resulted significantly less accurate.U-Alb was inadequate as indicator of any level of GFR impairment.
Conclusions: Serum levels of LMWP are probably not more sensitive or accurate than serum creatinine as an early indicator of GFR impairment. U-Alb gives no information on GFR impairment.

Su230 BONE MINERAL STATUS AND INFLUENCE OF ALFACALCIDOL ON MINERAL METABOLISM PARAMERERS OF PATIENTS WITH CHRONIC KIDNEY DISEASE (CKD) STAGES 3 AND 4

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Introduction and Aims: The abnormalities in bone histology and laboratory parameters of Pi-Ca metabolism, known as CKD-MBD, can be observed

early in the course of the kidney disease. Aims of our study were to investigate associations between vitamin D status and BMD and to identify significant predictors of deterioration of BMD and influence of alfacalcidol on mineral metabolism parameters of this population.
Methods: The study examined the frequency and severity of the skeletal changes in 103 predialysis CKD patients (age 57±13,1 years) with mean glomerular filtration rate (GFR) 26,1±1,2 ml/min. BMD by DXA and mineral metabolism parameters were measured. Serum 25(OH)D levels were estimated in 43 patients, which have not administered any forms of vitamin D earlier. 19 patients with CKD stages 1 and 2 were control group.
Results: Low BMD was noticed in 89% of patients. That is 3 times above in general white population with the same demographic data. Among women ≥ 50 years the osteoporosis of spine was observed 2 times more often, than in males of the same age. Inverse relationship was obtained between parathyroid hormone (iPTH) and BMD of proximal forearm (r=-0,301; p=0,007). Independent predictors low BMD were iPTH level, body mass index and age. Metabolic acidosis correlated with BMD index of vertebra (r=0,278; p=0,005) and hips (r=0,340; p=0,001). High turnover MBD estimated in 47,6% of patients, low turnover MBD – in 22,3% and the mixed form – in 30,1%. Adenoma of parathyroid gland was revealed in 7 patients (6,8%). Decreased serum 25(OH)D was detected in 74,4% of patients with CKD stages 3 and 4 and in 84% of patients with CKD stages 1 and 2. Deterioration of BMD was more profound in patients with deficiency and insufficiency of 25(OH)D levels and include 31,2% of osteoporosis and 68,8% of osteopenia compared with 27,3% of osteoporosis and 36,4% of osteopenia in patients with normal vitamin D status. Serum 25(OH)D level was not directly associated with BMD, but it was independent predictor of iPTH elevation. Patients with deficiency 25(OH)D had significantly lower serum level of calcium and higher serum levels of phosphorus and iPTH. Treatment with alfacalcidol (mean dose 0,30±0,11 μg/d) during 4 months was accompanied by the tendency to a gain mineral density of lumbar vertebras in 1,9%, of forearm–in 2,2%, of hips–in 1,6% and significant decline iPTH (p<0,03). In patients without treatment serum iPTH has increased (p<0,05).
Conclusions: Vitamin D deficiency very common in patients with all stages of CKD. Decline of BMD begin at an early stage of CRF. In CKD stages 3 and 4 population deficit of vitamin D may indirectly affect on BMD of these patients. Alfacalcidol dose from 0,25 to 1,0 μg/d for treatment hyperparathyroidism in patients with CKD stages 3 and 4 has appeared adequate, allowing to normalize serum calcium and iPTH and retard decrease BMD loss.

Su231 CLOCK DRAWING TEST AND MORTALITY IN HAEMODIALYSIS PATIENTS, REVERSED EPIDEMIOLOGY IN 5 YEARS OBSERVATION?

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Introduction and Aims: Nonverbal cognitive tests, particularly clock drawing test (CDT) have been presented in the last years as independent mortality predictors. On the other hand, the high rate of cognitive dysfunctions has only recently been recognized in haemodialysis patients, also by our observations. Severe cognitive impairment has been described as connected with higher mortality rate. Mini Mental Status Examination (MMSE) was usually used as a basic test for these evaluations, but still some tests were not frequently mentioned, CDT in particular. Due to this fact, we performed a 5-year observation of our haemodialysis patients to find out if there is a possible link between cognitive dysfunctions and mortality for both verbal and nonverbal tests.
Methods: 102 HD patients (65 male/37 female) aged from 23 to 82 years (avg. 57,5±14,1). We performed commonly used tests for cognitive functions: MMSE, Labyrinth Test, Trial Making Test type A and B (TMT A&B) and Letter-Cross-Out Test. Furthermore, Beck's Depression Inventory was used in all the patients. We also analyzed normal clinical and biochemical parameters as age, gender, education level, arterial blood pressure, EF, ECG, hemoglobin, urea, kt/v and CRP, cholesterol.
Results: The impaired cognitive function was diagnosed in the majority of patients in at least one of the tests: MMSE, TMT, CDT, BDI, Labyrinth and Letter-Cross-Out Test.During the 5-year observation, 52 (53%) our patients died. This group of patients had significantly worse results compared to those who survived in MMSE, TMT A, Labyrinth, and Memory Test, but not in CDT.
Furthermore, the patients who died during the observation period had initially showed significantly higher CRP level, greater left atrium dimensions, but smaller ejections fractions and HDL cholesterol. We performed Cox regressions analysis and we constructed a mortality risk model including 6 parameters such as age, hemoglobin, CRP, CDT, TMT B, and EF (χ^2= 71,7631; df = 6; p<0,00001).

Cox regressions analysis

	Exponent Beta	P
Age	1.104425	0.00001
Hgb	0.782592	0.020921
CRP	1.006587	0.027868
CDT	0.671853	0.007731
TMT B	0.996771	0.020233
EF	0.948432	0.000174

While factor beta determines the strength and direction of dependency and Wald's statistics defines what portion of the risk of death is explained by each parameter. For example, increasing CRP by 1 mg/dl increases the risk of death by 0.1% and CDT worse by 1 level is connected with the decrease of mortality risk by 33%. The interpretation of our findings must be very cautious because it was carried out in a single center with a small number of patients. However, we cannot exclude that the causes leading to impaired nonverbal test results are different in haemodialysis and other patients.
Conclusions: 1. Mortality in haemodialysis patients is connected with impaired cognitive functions. 2. Impairment of nonverbal function, however, seems to be protective at least in our observation.

Su232 RENOVASCULAR EFFECT OF ADDUCIN GENES ON ARTERIES RESTENOSIS AND RENAL FUNCTION

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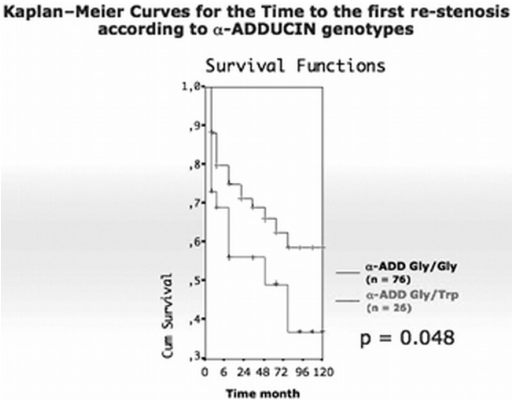
Introduction and Aims: The role of α-adducin Gly 460 Trp polymorphism on endothelial remodelling and apoptotic reaction in both experimental models and patients with coronary artery lesions has been described. Aim of our study was to investigate the predictive power of α and β adducin (ADD1, ADD2) genes polymorphism on re-stenosis in patients with renal artery stenosis (RAS) underdoing PTA/Stenting.
Methods: We enrolled 126 patients with renal artery stenosis who underwent PTA/Stenting in our Hospital. Mean age 73.5 years (range 37-91 years),mean eGFR 59 ml/min (range 7-165 ml/min/1.73)with a mean follow up of 60 months (range 6-120 months). Our outcomes were: renal function, blood pressures, and time free from re-stenosis according to α and β adducin polimorphisms.
Results: Our data show that ADD1 polimorphism has a predictive role on



time to re-stenosis: patients with α adducin polymorphism Gly/Trp have a higher percent of re-stenosis (p=0,048 as Kaplan -Meier as figure show). Besides our study focused on renal function (eGFR) and sistolic blood pressure (SBP) according to β adducin polymorphism (ADD2 CT-TT/ ADD2 TT). We found out that patients with RAS carrying the ADD2 CT-TT genotype have a long lasting eGFR improvement with smart significativity (p=0,014) after covariation for time of re-stenosis; on the other hand systolic blood pressure control wasn't substantially different in the two groups.
Conclusions: Our study shows a longer benefit of PTA in the subgroup of patients with α adducin Gly-Gly and an improvment in eGFR in patients with β adducin CT/TT. Besides patients carrying the ADD1 Trp allele have a higer percent of restenosis may related to both its vascular and hypertensive effects.

Su233 PHOSPHATE LOAD AND SERUM PHOSPHATE LEVELS AS DETERMINANTS OF DECLINE IN GLOMERULAR FILTRATION RATE IN ADVANCED CHRONIC KIDNEY DISEASE

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Introduction and Aims: High serum phosphate (P) has been shown to be associated with a more rapid decline in renal function in patients with chronic kidney disease (CKD). Plasma level of FGF-23, a surrogate of the overall burden of P loading, is a predictor of the progression of renal disease. However, it is not known whether direct estimates of P load independently relate to the progression of renal disease.
The aim of the present study was to determine the association of serum phosphate and total urinary phosphate excretion in the progression of renal failure, adjusted for other potential confounders.
Methods: This retrospective follow-up study included 184 patients (69±12 years, 93 females) with CKD stage 3, 4 or 5 pre-dialysis (mean eGFR = 15.2±5.6 ml/min/1.73 m^2). The rate of decline in renal function was calculated as the difference between final and baseline eGFR (MDRD-5) divided by the follow-up time (ml/min/month). Median follow-up time was 300 days. Urinary excretion of proteins, urea, and P were measured in 24 h urine samples, and total urinary P excretion and P excretion corrected for protein equivalent of nitrogen appearance (PNA) were calculated. Multivariate linear regression analysis was used to assess the relation between the rate of decline of renal function and the study covariates. Apart from demographics, body mass index, smoking habit and initial eGFR, the following continuous covariates were included in the analysis as time-average values: mean arterial blood pressure, serum P, calcium, albumin, bicarbonate, haemoglobin, PTH, total urinary protein and P excretion.
Results: Mean variation of eGFR was -0.20±0.38 ml/min/month. In addition to the highly expected association between serum P levels and renal function or phosphate load, total urinary protein excretion was positively associated with serum P levels (R=0.38; p<0.0001). Baseline serum bicarbonate levels were associated negatively with serum P, and patients treated with diuretics had significantly higher serum P levels than those non-treated.
In the multivariate analysis, serum P levels were associated with the rate of decline of renal function adjusted for the rest of study covariates (beta = -0.431; p<0.0001). However, neither total urinary P excretion nor P excretion corrected to PNA was associated with the rate of decline of renal function.
Conclusions: In conclusion, although high serum P levels were associated with a more rapid decline of renal function in advanced CKD patients, estimates of P load were not useful to predict the progression of renal disease.

Su234 PREDICTION OF GLOMERULAR FILTRATION RATE FROM SERUM CYSTATIN C: COMPARISON OF IMMUNE NEPHELOMETRIC AND TURBIDIMETRIC METHODS

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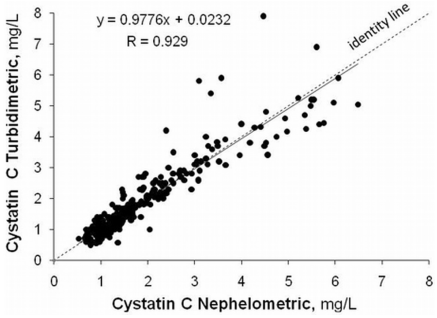
Introduction and Aims: The aim of this study was to compare the analytical performance of two different methods, proposed for the measurement of serum cystatin C (Cys) and to predict glomerular filtration rate (GFR).
Methods: Three hundred-sixty six adult renal patients (194 females and 172 males) affected by chronic kidney disease (CKD) with different degree of functional impairment (serum creatinine 0.4-10.5 mg/dL) partecipated to this study. GFR was determined as the renal clearance of ^{99m}Tc-DTPA. Serum concentration of cystatin C (Cys) were determined with a nephelometric method, and for comparison with a recent immune-turbidimetric method.
Results: A very high linear correlation was found between the two measurements of Cys (r=0.929). The mean difference CysTurb-CysNeph was 0.02±0.43 mg/L (NS).



A high logarithmic correlation was also found between Cys and GFR (correlation coefficients r were 0.92 for Cys Neph and 0.94 for Cys Turb). Point on the regression line between Cys and GFR were higher than upper reference limits when GFR was ≤64.5 (CysNeph) and ≤ 60.0 mL/min/1.73 m^2 (Cys Turb). On the basis of the relationship of Cys Neph and Cys Turb with GFR we developed two formulas to predict GFR from Cys. A good agreement was found between predicted GFR and measured GFR: the mean differences were -0.7 mL/min/1.73 m^2 (Cys Neph) and -0.6 mL/min/1.73 m^2 (Cys Turb). The accuracy of the 2 methods to evaluate a moderate impairment of GFR (GFR<80 and <60 mL/min/1.73 m^2), evaluated with ROC plots, resulted quite similar.
Conclusions: In conclusion, the immune-turbidimetric method to measure serum cystatin C seems adequate, at least like the immune-nephelometric method, to predict GFR and its impairment in CKD.

Su235 IS CARNOSINASE 1 GENE (*CNDP1*) POLYMORPHISM ASSOCIATED WITH CHRONIC KIDNEY DISEASE PROGRESSION? RESULTS OF FAMILY-BASED STUDY

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Introduction and Aims: Recent studies have shown that the carnosinase 1 gene (*CNDP1*), which encodes the serum secreted carnosinase, is strongly associated with diabetic nephropathy. The aim of the study was to investigate the role of the D18S880 microsatellite polymorphism of *CNDP1* gene in the development and progression of chronic kidney disease (CKD) of non-diabetic aetiology.
Methods: We applied the family-based study design using transmission/disequilibrium test. The study population consisted of 109 patients with CKD stages 3-5 caused by chronic glomerulonephritis (GN) or tubulointerstitial nephritis (IN) and their 218 healthy parents. The *CNDP1* gene (D18S880) polymorphism (PCR and DNA sequence analysis) and serum carnosinase activity (fluorometric method) were determined in all subjects. Serum carnosinase activity was also measured in 20 healthy controls.
Results: No significant differences were observed in the transmission of the alleles of the *CNDP1* gene polymorphism from parents to the children in the whole group of patients or in the IN subgroup. In the GN subgroup, the 5 allele was transmitted significantly more often than the 6 allele. There was no association between that polymorphism and the loss of glomerular filtration rate (fast *vs* slow progression of the disease). Serum carnosinase activity was significantly higher in the CKD patients than in the controls (148.75±19.49 and 140.98±11.04 nmol histidine/ml/min, respectively; p<0.01). We found

no association between that serum protein value and polymorphism of its encoding gene.
Conclusions: This study showed no association between the *CNDP1* polymorphism and increased risk for development of CKD caused by chronic tubulointerstitial nephritis. However, the polymorphism can influence on CKD caused by chronic glomerulopathy. The progression rate of CKD does not depend on this polymorphism. The increased serum carnosinase activity in the CKD patients may suggest its role in the pathomechanism of the disease.

Su236 CLINICAL UTILITY OF ESTIMATED GFR GRAPH IN PATIENTS WITH CHRONIC KIDNEY DISEASE

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Introduction and Aims: The estimated glomerular filtration rate (eGFR) graph plotted at every hospital visit is a useful clinical tool to visualize the progression rate of chronic kidney disease (CKD). The objective of this study is to measure a distribution of eGFR slope and to explore the clinical factors associated with the slope.
Methods: In this retrospective cohort study, 119 patients with eGFR of 15-50 ml/min/1.73m^2 were enrolled between 2004 and 2006. The patients who developed acute kidney injury or treated with steroid during observation were excluded. The eGFR was estimated using the abbreviated MDRD formula modified for Japanese. The eGFR graph during the study period was plotted in the individual patient and the eGFR slope of the linear approximation curve was calculated. The relationship between baseline variables and the eGFR slope was investigated using regression analysis.
Results: The median follow-up period was 2.95 years, the age was 65.4±13.6 years, and 79.0% of the patients were male. The eGFR slope ranged widely from -4.38 to 24.31 ml/min/1.73m^2/year with a mean ± SD of 3.38±4.18 ml/min/1.73m^2/year. The eGFR slope was significantly correlated with urinary protein to creatinine ratio in multiple regression analysis (R^2 = 0.45, P < 0.001). The eGFR slope was significantly larger in patients with diabetic nephropathy than in the others (7.19±5.35 ml/min/1.73m^2/year [n = 27] vs. 2.26±2.98 ml/min/1.73m^2/year [n = 92], P < 0.001). In nonprogressive patients with eGFR of -0.37±1.29 ml/min/1.73m^2/year [n = 38], cardiovascular risk factors such as diabetes (P = 0.005), hypertension (P = 0.027), hyperlipidemia (P = 0.005) was fewer than in progressive patients.
Conclusions: The eGFR slopes have a large distribution and correlate with urinary protein to creatinine ratio. Combination of eGFR graph and urinary protein to creatinine ratio may be useful in understanding the progression rate of CKD.

Su237 CAN TIME-AVERAGE PROTEINURIA BE PREDICTED IN IgA NEPHROPATHY BY OTHER POTENTIAL RISK FACTORS?

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Introduction and Aims: Risk factors for IgA nephropathy (IgAN) include reduced renal function (decreased e-GFR) hypertension, and proteinuria at renal biopsy. However, while the first two factors are related to irreversible sclerotic changes, proteinuria can be modified by several approaches, such as using glucocorticoids or angiotensin-antagonist therapies. For that reason, proteinuria exposure during follow-up, time-average proteinuria (TA-proteinuria), may be a more precise expression of this risk factor for progression of IgAN. Specifically, the patients with TA-proteinuria <1g/day exhibit less disease progression than those with higher values (JASN, 2007;18:3177).
Methods: In this study, we assessed the prognostic significance of TA-proteinuria in a cohort of 62 patients with IgAN, with a median follow-up of 4 years (interquartile range, IQ, 0.27-1.67), who showed median TA-proteinuria of 0.74 g/day (IQ 0.27-1.67) and a rate of e-GFR decline of –0.75±6.1 ml/min/1.73m^2/year. We also assessed whether a serum marker of IgAN or measures of oxidative stress predicted TA-proteinuria. The marker of IgAN was serum levels of galactose-deficient IgA1 (Gd-IgA1), measured by ELISA with *Helix aspersa* lectin (HAA) and expressed relative to standard Gd-IgA1 myeloma protein (%HAA) or as total amount of Gd-IgA1 (U/ml). Measures of oxidative stress were serum levels of advanced oxidation protein products (AOPPs), determined by spectrophotometry, and serum levels of free sulfhydryl groups of albumin (SH-Alb), assayed using iodoacetamide cyanine.
Results: TA-proteinuria inversely correlated with e-GRF slope (r=-0.348, p=0.026), AOPPs levels (r=0.49, p<0.0001) and %HAA (r=0.29, p=0.024). When dichotomizing TA-proteinuria (≤1 g/day = low; >1 g/day = high), using binary logistics we found at univariate analysis that serum levels of AOPPs and %HAA significantly predicted TA-proteinuria (B=0.025, p=0.002 and B=0.048, p=0.03 respectively), but SH-Alb and Gd-IgA1 did not. At multivariate analysis, in a model including %HAA, AOPPs, and SH-Alb values (significance of the model p=0.004), %HAA and AOPPs were independent predictors of TA-proteinuria (B=0.046, p=0.048 and B=0.031, p=0.002), while SH-Alb was not quite (B=-0.23, p=0.078). ROC analysis confirmed that AOPPs significantly predicted TA-proteinuria (AUC 0.77; sensitivity 0.70 and specificity 0.79, p<0.0001); the best cut-off value was 121.5 μMol/L. Patients with AOPPs >121.5 μMol/L had a significantly higher TA-proteinuria when compared with those with AOPPs <121.5 μMol/L (1.41±0.74 vs 0.69±0.82, p=0.001).
Conclusions: Thus, an oxidative stress marker, serum AOPPs level, was the best predictor of TA-proteinuria during follow-up in patients with IgAN.

Su238 ATORVASTATIN IMPROVES TUBULAR STATUS IN NON-DIABETIC PATIENTS WITH CHRONIC KIDNEY DISEASE – PLACEBO COTROLLED, RANDOMIZED, CROSS-OVER STUDY

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Introduction and Aims: Inhibition of the renin-angiotensin-aldosterone system (RAAS) with angiotensin converting enzyme inhibitors (ACEI) and/or angiotensin II subtype 1 receptor antagonists (ARB) constitutes a strategy in the management of patients with chronic kidney disease (CKD). There is still no optimal therapy that can stop progression of CKD. Statins e.g. atorvastatin (ATO) has been reported as a promising strategy in this field. We evaluated the influence of ATO (40mg/day) added to RAAS blockade.
Methods: In a placebo-controlled, randomized, cross-over study we evaluated the influence of ATO added to RAAS blockade on proteinuria and surrogate markers of tubular injury in 14 non-diabetic patients with proteinuria [0,4 – 1,8 g per 24 hours] with normal or declined kidney function [eGFR 55-153 ml/min]. Subjects entered the 8 weeks run-in period during which the therapy using ACEi and/or ARB was settled with blood pressure below 130/80 mmHg. Next, patients were randomly assigned to one of two treatment sequences: ATO/washout/placebo or placebo/washout/ATO. Clinical evaluation and laboratory tests were performed at the randomization point and after each period of the study.
Results: The ATO therapy significantly reduced urine excretion of α1–microglobulin (p=0.033) and N-acetyl-β-D-glucosaminidase (NAG) (p=0.038) as compared to placebo. There were no differences in proteinuria, blood pressure, eGFR and serum creatinine between the ATO and placebo.

Conclusions: Atorvastatin treatment was safe and improves tubular status in non-diabetic patients with CKD.

Su239 RELATIONSHIP BETWEEN MARKERS OF INFLAMMATION AND SERUM PHOSPHATE DURING THE COURSE OF CHRONIC KIDNEY DISEASE

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Introduction and Aims: Levels of serum phosphate (SP) are associated with progression of renal disease, development of vascular calcifications and cardiovascular events in Chronic Kidney Disease (CKD). Inflammation is a key factor in atherosclerosis and is a highly prevalent in CKD. We sought to determine whether changes in markers of inflammation predict SP changes independent of CKD progression or modify the relationship between progression of CKD and SP.
Methods: Consecutive patients referred to a large CKD program between 2001 and 2006 were eligible for the study if they were followed for 2 yrs and their GFR at the time of referral was between 15 and 60 ml/min (CKD stage 3-4). Patients with GFR < 60 ml/min receive routine serial measurements of SP, serum Ca, PTH, albumin, C reactive protein (CRP), creatinine, urea and 24 hrs urinary creatinine, urea and protein excretion. Protein intake was calculated from urea excretion. ANCOVA was used to model SP at follow-up end as a function of change in CRP and its baseline values, controlling for GFR decline, associated mineral metabolism derangements, and markers of nutrition, including protein intake and serum albumin.
Results: There were 227 patients (mean age 70 yrs, mean BW 74 kg, 66% male, 28% diabetics) satisfying the eligibility criteria. During the study GFR decreased from 32.5±11 to 29.9±14 ml/min (P< 0.01). SP and PTH increased from 3.4±0.6 to 3.6±0.8 mg/dl (P<0.001) and from 88±55 to 101±60 pg/ml (P<0.001), respectively. Serum Ca (9.5±0.5 and 9.35±0.5 mg/dl) and estimated protein intake (70±19 and 70±22 g/day) did not change. In 2 years 68 patients (30%) progressed to a worse CKD stage; the proportion of subjects with SP >4.3 mg/dl increased from 10% to 16%; and the proportion of patients with CRP >5 mg/L from 45% to 55%. Baseline SP and CRP correlated with GFR (–0.40 and –0.20). No correlation was found between SP and CRP. Changes in SP (-039) but not changes in CRP during follow up correlated with variations in GFR. Multivariate analysis (r^2 0.47) showed that the main predictors of SP levels after 2 yrs were the decline in GFR, female gender, protein intake and calcitriol supplements. Serum albumin and CRP did not show any independent or modifying effect.
Conclusions: 1) An inflammatory state is frequent in CKD stage 3 and 4, but does not seem to affect SP levels during the course of CKD; 2) GFR decline, protein intake and calcitriol therapy are the main predictors of SP levels during the course of CKD. Protein and phosphate intake restrictions and careful use of calcitriol are critical measures to control SP levels in CKD stage 3 and 4.

Su240 HIGH PREVALENCE OF HYPOGONADISM IN MALE PATIENTS WITH CHRONIC RENAL FAILURE

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Introduction and Aims: Chronic hemodialysis treatment is associated with a high prevalence of low testosterone levels in male patients. Moreover, clinical features of hypogonadism are simillar to those of chronic renal failure (CRF). There are also reports on elevated prolactin levels in dialysis patients contributing to low testosterone levels by lowering gonadotropin levels. However, data on the prevalence of hypogonadism in patients with moderate renal insufficiency and in patients not yet on dialysis are very rare. Therefore, we prospectively investigated sex hormone status in a large group of male patients with CRF.
Methods: 280 male patients with CRF and 47 male patients on hemodialysis were prospectively investigated. Clinical data such as etiology of renal diseesae, prevalence of diabetes mellitus, BMI and medication were studied. Serum creatinine and GFR were measured as well as serum testosterone, sex hormone binding globulin (SHBG), as the major binding protein, and prolactin. Patients were subdivided according to the CRF stages 1-5.
Results: Decreasing GFR was associated with a significant fall of testosterone (r= 0.25; p<0.0001); 34% of all 280 patients (stage 1-5) had testosterone deficiency (< 10 nmol/l). In patients wth stage 4 and 5 (GFR <30 ml/min; n= 49) the prevalence of hypogonadism was 48% and in the 47 hemodialysis patients 49%. There was no difference in testosterone concentrations between those two groups; stage 4 and 5: 10.6±5.3 nmol/l (mean ± SD) vs 10.5±5.1 in the hemodialysis group.
Testosterone was negatively correlated with BMI (r= -0.16; p<0.007) and with age (r= -0.31; p<0.0001). SHBG, however, was negatively correlated with GFR (r=-0.23; p<0.0001, decreaed with increasing BMI (r= – 0.28; p<0.0001) and was positively correlated with age (r= 0.34; p<0.0001).
Prolactin levels (normal <16 ng/ml) were sigificantly higher in hemodialysis patients: 14.6±15.7 ng/ml vs. 11.2±15.7; p<0.015 in patients with stage 4 and 5. Moreover, increasing prolactin conc. were associated with lower testosterone levels in the dialysis group (r= -0.32; p<0.027), but not in the patients in stage 1-5 (r= -0.1; ns.)
Conclusions: 1/3 of patients with CRF (stage 1-5) have testosterone deficiency. Decreasing renal function is associated with decreasing testosterone levels. There is a very high prevalence of hypogonadism (48%) in CRF stage 4 and 5 which is not different from hemodialysis patients. However, the major testosterone binding protein (SHBG) increases with falling renal function. Prolactin is only a relevant contributor to hypogondism in the dialysis patients cohort.

Su241 PREGNANCY IN A PATIENT WITH FANCONI-BICKEL SYNDROME UNDERGOING DAILY HAEMODIALYSIS TREATMENT

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Introduction and Aims: Fanconi-Bickel syndrome (FBS) (glycogenosis type XI, GSD XI) is a rare genetic disease inherited AR pathway. This disease is caused by a mutation of a gene coding membrane protein which is a transporter of a glucose GLUT2. FBS syndrome is characterized by an accumulation of glycogen in liver, disturbed transport in proximal tubular cells of a nephron and improper metabolism of a glucose and galactose. So far there is no description of pregnant patient with FBS who delivered a healthy baby. We didn't found a case report of a FBS patient suffering from ESRD and being hemodialysed.
Methods: Patient S.D aged 32y, in whom first symptoms of FBS such as polyuria and polydypsia appeared when she was 2 months old. Urine examination showed glucose and protein presence, and she was diagnosed with Fanconi-de Toni-Debre syndrome. She was physically retardated and had an osteodystrophy. Aged 4 she was diagnosed with bilateral kidney stones, since then the creatinine level was about 2 mg/dl. When she was 17 years old she had a liver biopsy and glycogenosis type XI was recognized. Patient height is about 135 cm. Her psychological and social development is normal.
Since 2004 patient is under supervision of the Outpatient Clinic of Dept of Nephrology in Szczecin, Poland. During 5 years of supevision her creatinine level incresed from 2.17 up to 2.7 mg/dl. We found two mutations within GLUT2 (SLC2A2) in our patient: a 1-bp- deletion in exon 1 and a splice site mutation at the acceptor site of intron 8. Both parents are heterozygous as expected.
Results: In 2009 patient became pregnant, what was confirmed in 8 Hbd. Due to increase of creatinine level eGFR (MDRD)= 21 ml/min/1,73 m^2 we started hemodialysis treatment 6x a week, 4 hours per session.
In 10 week of pregnancy patient was diagnosed with cholestasis. In 12 HBD there was a prenatal diagnostics performed, and increased risk of a trisomia of 21 chromosome was found. Between 16/17 week of the pregnancy she had an amniocentesis. The analysis of cariotype confirmed normal male cariotype of fetus- 46XY. In 23 week of pregnancy a hydramnion was found. The number of hours of hemodialysis was increased up to 5 hours per session.

In 34 week of pregnancy our pregnant patient with FBS started to complain of fatigue and dyspnea at rest. Obstetricians decided to terminate the pregnancy. Patient had a caesarian section. She delivered a healthy male newborn, weight 2540 g, and was discharged home in a good condition 4 days after the delivery. Up to now she is under supervision of our Outpatient Clinic. Her results are stable, creatinine and urea level are similar to those before pregnancy. Currently she does not demand hemodialysis therapy. Development of a child is normal.

Conclusions: This case report is to reveal that carefull and thorough supervision allows patients with rare tubular deffects and ESRD to become pregnant and have healthy children.

Su242 FETUIN-A MAY NOT BE ASSOCIATED WITH INFLAMMATION IN CHRONIC KIDNEY DISEASE STAGES 3 & 4

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Introduction and Aims: Fetuin-A, a 62kDa glycoprotein, is synthesized in the liver. Fetuin-A is a potent circulating inhibitor of ectopic calcification, but is also thought to be a negative acute-phase reactant. Low circulating fetuin-A concentrations have been associated with aortic stiffening and adverse cardiovascular outcomes in both pre-dialysis and dialysis chronic kidney disease (CKD). This association is potentially confounded by inflammation. However no significant relationship was present between plasma fetuin-A concentration and CRP in a cohort study of CKD stages 3 & 4. We therefore examined the relationship between plasma fetuin-A and high sensitivity CRP and interleukin-1β (IL-1β) in this cohort.

Methods: 133 stable outpatients with CKD stages 3 & 4 were enrolled in a prospective study of cardiovascular risk. Fetuin-A and IL-1β were measured by ELISA (Biovendor, CZ) and RnD Systems, Europe Ltd. (UK) respectively. hsCRP was measured by cardiophase assay (Siemens, UK).

Results: Mean MDRD eGFR was 32 (±10.9 ml/min/1.73m^2). Age 69 (±11.5 yrs). Male:female 103:30. Mean fetuin-A was 0.24 (±0.078 g/l). Median hsCRP was 2.18mg/l (IQR 4.46 mg/l), median IL-1β was 2.78pg/ml (IQR 1.12 pg/ml). Fetuin-A did not correlate with either log hsCRP (r=0.077, p=0.379), or log IL-1β (r=0.091, p=0.295). As expected, log hsCRP correlated with log IL-1β (r=0.181, p=0.038).

Conclusions: In this cohort of stable pre-dialysis CKD patients fetuin-A was not associated with markers of inflammation. Variation of plasma fetuin-A concentration in this setting may therefore be better explained by factors other than the negative acute phase response.

Su243 EVOLUTION OF RENAL FUNTION IN 1009 CARDIAC TRANSPLANT RECIPIENTS: IMPACT OF DIFFERENT IMMUNOSUPPRESSIVE REGIMENS

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Introduction and Aims: Achieving efficacy while maintaining safety is key after heart transplantation (HTx). However, chronic kidney disease (CKD) is an increasing complication with substantial morbidity and mortality and seems to be related to immunosuppressive regimens (ISR) used. Lowering immunosuppression (IS) can be an option to improve renal outcome but bears the risk of increasing acute rejections (AR). Here, we retrospectively analyzed the impact of 6 ISR on evolution of renal function (RFct) relative to efficacy outcome.

Methods: 1009 HTx recipients (HTxR) from 3 randomized, multicenter trials received one of the following ISR: SD-CsA/AZA (n=214); SD-CsA/MMF (n=84); SD-CsA/h-EVR (n=211); SD-CsA/l-EVR (n=209); SD-CsA/TDM-EVR (n=100); RD-CsA/TDM-EVR (n=191) [SD=standard dose; RD=reduced dose; h-EVR=3.0 mg/d; l-EVR=1.5 mg/d; TDM-EVR=C_0 3-8 ng/mL]. Incidence and severity of AR and evolution of eGFR were analyzed. HTxR were also stratified according to CKD stages.

Results: HTxR had similar baseline (BL) characteristics. With EVR based IS more patients were free of AR and also less early AR occurred than with MMF or AZA. The SD-CsA/TDM-EVR group showed the best efficacy profile. Comparing RFct, HTxR on RD-CsA/TDM-EVR showed the slowest decline in RFct (MDRD) from BL to M6 compared to the other ISR, but still showed a higher absolute change than HTxR treated with MMF or AZA (-7.8 vs -4.9 and -6.5mL/min). HTxR with BL GFR 30-59 mL/min showed less decline in RFct over time and less differences between the ISR compared to HTxR with better (60-89mL/min) or good RFct at BL (>90mL/min) (Table 1).

	SD-CsA/AZA	SD-CsA/MMF	SD-CsA/ l-EVR	SD-CsA/ h-EVR	SD-CsA/ TDM-EVR	RD-CsA/ TDM-EVR
No AR at M6 (%)	13.6	13.1	20.6	24.2	28.0	23.6
Total AR ≤Day 90 (%)	81.8	78.6	74.2	71.6	67.0	72.3
Change in GFR, BL to M6 (MDRD, mL/min)						
All HTxR	-6.5	-4.9	-14.4	-16.2	-15.6	-7.8
HTxR with BL GFR ≥90	-38.5	-43.3	-40.6	-53.9	-27.5	-31.3

Conclusions: While controlling AR is the main goal of IS post HTx it is often accompanied by a rapid decline in RFct, therefore risks and benefits of different ISR need to be weighed. Best efficacy outcome was observed for SD-CsA/TDM-EVR. However, RFct was worse when EVR was combined with SD-CsA than with RD-CsA. EVR combined with CsA dose reduction resulted in improved renal outcome while providing better AR protection than AZA or MMF based ISR. HTxR with good RFct at time of HTx benefited most from CsA reduction.

Disclosure: Medical Scientific Advisor, Novartis Pharma AG.

Su244 OVERWEIGHT, OBESITY AND METABOLIC DISORDERS – RISK FACTORS IN CHRONIC KIDNEY DISEASE (CKD)

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Introduction and Aims: In the past three years there has been an alarming rise in the prevalence of CKD and also, the absolute risk of death appeared to increase exponentially with renal function decreasing. We conducted a retrospective study to evaluate whether obesity, overweight and metabolic disorders have an impact on the progression rate of non-diabetic CKD.

Methods: We reviewed 60 non-diabetic CKD patients' medical records, which were in our evidence for the past 2 years. Various demographic, clinical and bioumoral parameters were retrospective collected from the patients' database.

Patients were categorized in normal weight (BMI=18.5-24.9 kg/m^2), overweight (BMI=25-29.9 kg/m^2) and obese (BMI>30 kg/m^2). Multivariate regression analysis was used for eGRF reduction per year, as a dependent variable to evaluate the impact of obesity on CKD progression.

Results: All patients included in our study were in stage 3 CKD (mean eGRF=41.5 ml/min/1.73m^2) at the first presentation. Baseline mean arterial blood pressure (BP) was significantly higher in overweight and obese than normal weight patients (p_1=0.009 and p_2=0.003). In follow-up, mean arterial BP was also higher in overweight (p=0.003) and obese (p=0.004) compared with normal weight CKD patients. Mean cholesterol and triglycerides levels were significantly higher in obese (p=0.042).

The progression of CKD based on eGRF per year (>1ml/min/1.73m^2) was observed in 85% obese, 75% overweight and 55% normal weight patients, but we haven't noticed any significantly difference in the progression rate between the three groups.

In multivariate regression analysis adjusted for age, BP, proteinuria, dyslipidaemia, smoke, baseline BMI was an independent predictor of CKD progression. 10% of obese patients have lost weight but this didn't result in a slower decline in eGFR. However there was a lower proteinuria level in these patients.

Conclusions: Baseline BMI are strongly and independently associated with faster CKD progression based on the annual rate of eGFR fall.

Su244bis A BALANCE STUDY COMPARING THE ABSORPTION OF DIETARY PHOSPHORUS WITH LANTHANUM CARBONATE OR SEVELAMER CARBONATE IN HEALTHY ADULT VOLUNTEERS

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Introduction and Aims: Use of dietary restriction in patients with chronic kidney disease (CKD) as a means to control serum phosphorus presents a challenge to maintaining adequate dietary protein intake. Data from patients on dialysis suggest that increasing protein intake while reducing serum phosphorus is associated with a lower risk of mortality compared with concurrent increases or decreases in both parameters, or a decrease in protein intake with an increase in serum phosphorus. Therefore, highly effective phosphate binders may be desirable to treat elevated serum phosphorus while allowing increased protein intake in patients with CKD on dialysis.
Methods: This randomized, open-label, crossover study compared the absorption of dietary phosphorus after single doses of lanthanum carbonate or sevelamer carbonate. Both treatments are noncalcium phosphate binders used to reduce serum phosphorus in patients with CKD. Healthy volunteers underwent 4 treatment periods, separated by 7–14-day washouts. After fasting, the gastrointestinal tract was cleaned prior to ingestion of a standardized meal; 10 hours later rectal effluent was collected. Volunteers completed periods with meal alone, meal plus lanthanum carbonate (1000 mg of lanthanum) and meal plus sevelamer carbonate (2400 mg) in a randomly assigned sequence. Participants fasted in the fourth period. Inductively coupled plasma-optical emission spectroscopy was used to analyse the phosphorus content of duplicate meals and rectal effluent. Safety and tolerability were assessed and adverse events monitored.
Results: Net phosphorus absorption was analysed using a mixed effect linear model in the pharmacodynamic population (N = 18). Data are least squares mean ± standard error. Phosphorus absorption from the meal was 281.7±14.1 mg (9.1±0.5 mmol). Phosphorus absorption was significantly lower with lanthanum carbonate than with sevelamer carbonate (156.0±14.2 versus 221.8±14.1 mg [5.0±0.5 versus 7.2±0.5 mmol]): the difference was –65.8 mg (−2.1 mmol) (95% CI: –96.0, –35.5 [–3.1, –1.1], $p<0.001$). Lanthanum carbonate bound 135.1±12.3 mg (4.4±0.4 mmol) of phosphorus while sevelamer carbonate bound only 63.2±12.3 mg (2.0±0.4 mmol), a difference of 71.9 mg (2.3 mmol) (95% CI: 40.0, 103.8 [1.3, 3.4], $p<0.001$).
Conclusions: More than twice as much phosphorus was bound by a single 1000 mg dose of lanthanum carbonate than a single 2400 mg dose of sevelamer carbonate. These data suggest that in clinical practice, lanthanum carbonate may be a more effective phosphate binder than sevelamer carbonate. Use of highly effective phosphate binders may help to achieve sustained control of serum phosphorus while enabling an increase in dietary protein intake.
Disclosure: This study was funded by Shire Pharmaceuticals. LP, PM, AR and RP are employees of Shire Pharmaceuticals.

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Su245 RENAL SCARS AFTER ACUTE "NON COMPLICATED" PYELONEPHRITIS. RELATIONSHIP WITH THE INITIAL LESION: LONGITUDINAL ANALYSIS AT MAGNETIC RESONANCE

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Introduction and Aims: Acute pyelonephritis (APN) is a severe infectious disease, in the short term for the risk of sepsis, and in the long term, for the risk of developing kidney scars, also in the absence of morphologic and systemic predisposing factors ("non complicated" APN). In the literature, the prevalence of kidney scars after an episode of APN varies widely (30 to 80%), also according to imaging tests employed. Few recent studies assess their incidence; the relationship with the initial lesion was not studied sofar. Aim of this study is analyze the incidence and clinico radiological correlates of kidney scars in a cohort of "non complicated" APN patients, studied in the same settings by Nuclear Magnetic Resonance (MR) at diagnosis and during follow-up.
Methods: Retrospective review by two "blind" operators of the MR images and films of the patients with diagnosis of acute non complicated pyelonephritis followed in the same setting in the period 2007-2009, and who had at least two MR controls performed in the same setting.
In the period of study, treatment consisted in long-term intravenous antibiotic therapy, of 2-8 weeks, followed by one month of oral therapy; duration of intravenous therapy was tailored upon imaging data, according to the type of the original lesion(s), and to the eventual evolution; as a rule, i.v. therapy lasted until healing (or scar) of the lesion at imaging.
The type (abscessual versus simple) number (per kidney) of lesions and type of the eventual scar (with or without distortion of the renal profile) were analyzed in relation with the clinical and laboratory presentation.
Results: Overall the data on 39 patients were reviewed (all females, median age 32 years). The total number of lesions recorded was 105; 10 patients had single lesions, 29 multifocal; the median value of the maximum diameter of the lesions was 12.5 mm. Twenty-three lesions were abscessual. The median number of MR studies per patient was 3 (2-5), at 2-10 weeks from diagnosis. The total number of scars recorded was 16, in 9 patients (23%); of the scars, 7/16 caused distortion of the renal profile, or measured over 1 cm.
None of the clinical and laboratory parameters at diagnosis (C-reactive protein, fever, WBC, duration of symptoms, previous therapy) correlated with the initial lesions or with the outcome in renal scars.
Conversely, the initial size of the lesions (dichotomized at 1 cm) showed a significant correlation with the outcome in renal scar (p=0.01), while the presence of abscessual lesions showed a suggestive, albeit non significant trend (p=0.06).
Conclusions: In spite of long term antibiotic therapy the prevalence of kidney scars after acute non complicated APN remains relatively high (23% of the patients, considering small scars; 14% considering only scars causing profile distortion). The relationship with imaging data at presentation warrants further attention, and the higher risk in large or abscessual lesions indirectly underlines the importance of early diagnosis.

Su246 LITHIUM NEPHROTOXICITY, WHEN IS THE TIME FOR NEPHROLOGY CONSULTATION?

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Introduction and Aims: Lithium is a widely used drug for treatment of bipolar mood disorder (BMD). However nephrologists are familiar with the side effect of this drug, but the late referral of these patients for nephrology consult has a negative impact on patient's kidney management. In order to evaluate the prevalence of renal abnormalities in BMD patients on lithium therapy a study planned in one of our psychotherapy university centers.
Methods: Study group were consisted of 43 patients with BMD who were on lithium therapy for more than 3 months (range: 3- 312months). Lithium dosages were in the range of 300-1500 mg daily. Inclusion criteria were negative history of diabetes mellitus, hypertension or renal disease before lithium therapy. After taking patient's consent data extracted by patient's interview and medical charts. Then patient's blood sample and urine were sent for blood urea, serum creatinine, urine albumin to creatinine ratio, urine osmolarity and sediment evaluation. Patient's GFR were calculated by Cockcraft Gault formula and data analyzed by SPSS program version 16.
Results: They were 26 females and 17 males with the mean age of 36.5±12.5 years. Polyuria was the most prevalent complains in 8 (18.6%) patients. Urine sediment abnormalities including pyuria and hematuria, urine albumin to creatinine ratio> 30mg/g, urine osmolarity <350 mos/l were reported in 9 (21.4%), 8 (21.6%)and, 6 (14.3%) sequentially. On the basis of patient's GFR 24% of patients were classified in stage 2 and 2.4% in stage 3 chronic kidney disease (CKD).
Abnormal urine sediments were more common in females ($P<0.01$) and low urine osmolarity had an inverse correlation with treatment duration ($P<0.05$) in our study.
Conclusions: We conclude that more than 20% of our patients on lithium therapy had some degree of renal involvement. Since most of psychiatrists

are not familiar for early diagnosis and management of CKD patients, we suggest that all patients on lithium therapy should have early referral to CKD clinics. Educational program for both psychotherapist and patient is mandatory in this field.

Su247 EPIDEMIOLOGICAL DATA REGARDING BALKAN ENDEMIC NEPHROPATHY IN RELATIONSHIP WITH THE PLIOCENE COAL ETIOLOGICAL HYPOTHESIS

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Introduction and Aims: Balkan Endemic Nephropathy (BEN) is a disease encountered in Romania, Bulgaria, Serbia, Croatia and Bosnia. The vicinity of pliocene coal deposits generated the pliocene coal etiological hypothesis of BEN, which sustains that pliocene hydrocarbons (polycyclic aromatic hydrocarbons, aromatic amines etc.) from coal in the BEN area are leaching into the groundwater; toxic substances could thus contaminate the drinking water in springs/wells and could exert nephrotoxic and carcinogenic actions. The aim of this paper was to analyze the pliocene coal hypothesis in the BEN region, where miners were most exposed to these substances, by drinking water from the wells/springs adjacent to the mines.
Methods: We investigated the prevalence of miners among dialyzed patients from the BEN region. Further, an epidemiological analysis regarding GFR and proteinuria of persons over 50 from the BEN villages: Hinova, Bistrita, Livezile (mine now closed-pliocene coal), and Husnicioara (mine still functioning-pliocene coal) was conducted. At the same time, we investigated persons from Motru (city of miners from the non-endemic region; the mine does not contain pliocene coal), and persons from Drobeta Turnu Severin (a city near the endemic zone). Thirty-four miners (mean age: 56.7±6.42) from BEN villages were also analyzed. Statistical analysis (unpaired t-test) was performed using OpenEpi.
Results: In the Dialysis centers from Drobeta Turnu Severin 165 patients are undergoing hemodialysis, 79 of them being BEN patients, and 5 of the BEN patients being former miners.
The GFR was lower in persons from the endemic villages Bistrita and Hinova than in persons from Drobeta Turnu Severin ($p=0.003$; $p=0.0004$). Further, persons from the endemic village of Husnicioara (mine still functioning) had a higher GFR than persons from Drobeta Turnu Severin or from Livezile ($p=0.0055$ and $p=0.01$ respectively), but a lower GFR than persons from Motru (where a non-pliocene coal mine is functioning) ($p<0.001$). The GFR and proteinuria for the different villages are presented in Table 1.
The mean GFR in the thirty-four miners was: 94.13±26.58 ml/min/1.73m^2.
Three miners (8.82%) had a GFR<60 ml/min/1.73m^2.
No former miner had tumours of the urothelium.
Conclusions: Our data do not favour the toxic hypothesis regarding the relationship between BEN and pliocene coal. It is likely that BEN etiology is multifactorial.

Su248 PROPENSITY SCORE ANALYSIS TO EVALUATE ASSIGNMENT OF PATIENTS TO HEMODIAFILTRATION AS INITIAL MODALITY OF CHRONIC DIALYSIS

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Introduction and Aims: Several studies investigated the factors related to the outcomes of patients starting with hemodialysis (HD) or hemodiafiltration (HDF) as treatment modality. In an observational study design the factors associated with assignment to HD or HDF could also be determinants of the outcomes, because the patients are not assigned to the treatment through a randomization method. Few studies evaluated this issue, in particular through data derived from an area-based registry. To evaluate the factors associated to assign a patient to HDF as initial modality of chronic dialysis treatment, we conducted a study using the propensity score analysis, that balances two non-equivalent groups on observed covariates.
Methods: Retrospective cohort study of 3957 patients undergoing chronic dialysis (CD) with HD or HDF, notified to Lazio Dialysis Registry (Italy) from 1-1-2004 to 31-12-2008. We performed a propensity score analysis to estimate the factors associated to HDF as initial modality of treatment. We considered for analysis baseline information: age, gender, education level, type of vascular access, self-sufficiency degree, diagnosis of nephropathy, presence of comorbidities (coronary heart disease, congestive heart failure, stroke, vasculopathy, diabetes, chronic lung diseases, chronic liver diseases, cancer, hypertension), haemoglobin level, albumin level, calcium level, creatinine level, type of dialysis facility (public vs. private).
Results: The prevalence of HDF as first treatment modality among the subjects who started CD in Lazio region was 3.8%. A higher probability of initiation with HDF compared with HD was found for patients with 1 year age increase (OR=1.04; 95%CI: 1.03-1.05), with 1g/dl haemoglobin level increase (OR=1.19; 95%CI: 1.07-1.34), with 1mg/dl creatinine level increase (OR=1.06; 95%CI: 1.00-1.12), with a catheter and not an arterovenous fistula as first vascular access (OR=1.81; 95%CI: 1.13-2.91), with congestive heart failure as comorbidity (OR=1.89; 95%CI: 1.17-3.06), with a higher educational level (OR=1.36; 95%CI: 0.97-1.91).
Conclusions: We observed a lower prevalence of HDF as first treatment modality among patients undergoing chronic dialysis than among all patients present in chronic dialysis (3.8% vs. 19.2%), according to the data of Lazio Dialysis Registry at 31-12-2008. Our study suggests that assignment to HD or HDF is influenced not only by a clinical evaluation, but also by a proxy of socioeconomic status, such as level of education. This finding could be partially explained as a preference for patients with a higher education level to be treated with HDF, a treatment modality that several studies reported associated with a better quality of life, independently from effect on survival. The role of factors different from clinical ones should be clarified to explain the high proportion of switch from HD to HDF during chronic dialysis history, in a region where private facilities (about 50%) has a threshold (about 20%) to treat patients with HDF modality.

Su249 EFFECT ON SURVIVAL OF SWITCHING TO HEMODIAFILTRATION (HDF) IN A COHORT OF PATIENTS STARTING CHRONIC DIALYSIS WITH HEMODIALYSIS (HD)

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Introduction and Aims: Several studies compared the effect on survival of chronic dialysis (CD) patients according to the choice of hemodialysis (HD) or hemodiafiltration (HDF) as first CD modality. However, few studies evaluated the role of switch from HD to HDF on survival. The aim of the study is to evaluate the factors influencing the time to switch from HF or HDF and the effect of switching on patients' survival in a cohort of patients all starting CD with HD.
Methods: Retrospective cohort study of 3804 patients who started CD with HD as treatment modality. The subjects were notified to Lazio Dialysis Registry (Italy) from 1-1-2004 to 31-12-2008. We performed a multiple Cox model to estimate hazard ratios (HRs) of time to switch, and another Cox model to estimate HRs of time to death; in the latter we include

Abstract Su247 – Table 1

Village/City	Bistrita	Hinova	Livezile (mine now closed-) pliocene coal)	Husnicioara (mine still functioning-) pliocene coal)	Motru (non-endemic region; mine does not contain pliocene coal)	Drobeta Turnu Severin (non-endemic region)
Number of persons	224	243	326	77	256	254
Gender distribution (F/M)	125/99	149/94	183/143	35/42	124/132	150/104
Mean age	62.25±8.69	64.91±10.49	67.48±9.84	65.85±7.65	61.1±8.42	62.22±8.80
Mean GFR (ml/min/1.73m^2)	70.12±16.62	69.41±15.46	76.63±16.63	81.68±15.87	100.01±27.45	74.98±19.14
Number of persons with proteinuria	18 (8.03%)	18 (7.4%)	0	2 (2.59%)	2 (0.78%)	18 (7.08%)

the "modality of dialysis" as a time-dependent covariate (before and after switch). We included in both models the baseline covariates associated to the outcome with a $p<0.10$.

Results: The median follow-up time was 20.6 months. Three hundred-nine subjects (8.1%) changed from the initial HD to HDF. We found a higher hazard to switch from HD to HDF when patients were 1 year younger (HR=1.03; 95%CI: 1.02-1.04), men (HR=1.37; 95%CIs: 1.06-1.76), self-sufficiency (HR=1.39; 95%CI: 1.03-1.86), had a fistula as first vascular access (HR=1.44; 95%CI: 1.06-1.96), a 1 g/dl haemoglobin level decrease (HR=1.14;95%CI1.05-1.23), a 1 g/dl albumin level decrease (HR=1.26; 95%IC: 1.01-1.57). We did not observe a difference for survival of subjects who switched from HD to HDF (HR=1.22; 95%CIs: 0.88-1.69), compared to patients who remain in HD during all study period, taking also into account age, gender, type vascular access, diagnosis of nephropathy, presence of comorbidities. We found a risk to death for men (HR=1.26; 95%IC: 1.10-1.43) and for patients with 1 year old increase (HR=1.03; 95%IC: 1.02-1.04), with a catheter as first vascular access (HR=1.41; 95%IC: 1.23-1.61), without self-sufficency (HR=1.65; 95%IC: 1.43-1.90), with a coronary heart disease (HR=1.16; 95%IC: 1.01-1.33), with a vasculopathy (HR=1.24; 95%IC: 1.07-1.45), with a cancer (HR=1.23; 95%IC: 1.02-1.47), with a 1 g/dl albumin level decrease (HR=1.40; 95%IC: 1.23-1.59), a 1 mg/dl creatinine level decrease (HR=1.07; 95%IC: 1.04-1.10), a 1 g/dl haemoglobin level decrease (HR=1.15; 95%IC: 1.10-1.20).

Conclusions: We found that several demographic and clinical conditions are associated with time to switch and that survival of subjects who switched from HD to HDF is not different compared to patients who remain in HD during all study period. These results seem to suggest that the choice to switch from HD to HDF modality is not associated with better prognosis for HD switching patients. As HDF has higher costs than HD for national health service, our findings suggest the need of further cost/effectiveness evaluation to support the choice of HDF as alternative treatment modality to HD.

Su250 INCIDENCE OF CHRONIC KIDNEY DISEASE IN A POPULATION REFERRED TO A CENTRAL LABORATORY FOR BLOOD ANALYSIS

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Introduction and Aims: Estimated glomerular filtration rate (eGFR) based on the MDRD formula has replaced serum creatinine for assessment of chronic kidney disease (CKD). Studies estimating the overall incidence of CKD are mainly derived from population-based health surveys.

We investigated the incidence of CKD based on eGFR in a population of patients referred from general practitioners for measurement of serum creatinine at a County Hospital in Norway.

Methods: The central laboratory has routinely reported eGFR from January 2007 based on the MDRD formula. Serum creatinine was measured by an enzymatic method. SPSS version 15 was used for the statistical analysis.

Results: There were 57 343 eGFR measurements performed in 36 178 individuals during 2007. In people with multiple eGFR measurements the average of the first and last eGFR was used. Individuals with eGFR> 60 ml/min per 1.73 were excluded.

In all, 4957 (13%) (3211 women and 1746 men) had eGFR <60 ml/min per 1.73 m^2. Median age for women was 79.0 years (SD 12.1) and for men 77.0 years (SD 11.6). Aged ranged from 18 to 100 years. Thirty-seven (0.1%) had eGFR <15 ml/min per 1.73, (stage 5). Two hundred and eighty-seven (0.8%) had eGFR 15-29 ml/min per 1.73, (stage 4) and 4447 (12.3%) had eGFR 30-59 ml/min per 1.73, (stage 3).

Conclusions: The incidence of CKD stage 3 and 4 in a patient population referred to a central laboratory for blood analysis is 3 to 4 times higher than reported from a Norwegian population-based health survey. Number of patients with CKD is substantially higher than patients registered with the diagnosis of CKD at the hospital registry. Our findings suggest that there is a need for guidelines for referral and follow-up of individuals with severe, but not recognized CKD.

Su251 PARICALCITOL TREATMENT IN CKD PATIENTS WITH SECONDARY HYPERPARATHYROIDISM IS ASSOCIATED WITH FEWER INFECTION-RELATED EVENTS WHEN COMPARED WITH NO VITAMIN D RECEPTOR [VDR] ACTIVATOR TREATMENT

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Introduction and Aims: We aimed to assess infection-related medication use, outpatient services, hospitalizations and costs in pre-dialysis CKD patients with SHPT receiving paricalcitol compared with no VDR activation treatment.

Methods: MedStat, I3 and IMS databases containing 2,524 patients with CKD and SHPT were analyzed to compare matched cohorts of patients treated with paricalcitol versus no VDR activator treatment. 1,262 patients per cohort were matched using propensity scoring of age, gender, Charlson Co-morbidity Index, and pre-index total cost. Multivariate analyses adjusting for baseline age, gender, CKD severity, insurance, physician specialty, region, pre-index co-morbidities, and pre-index costs were used to explore differences.

Results: Multivariate analyses demonstrated lower all-cause and infection-related out-patient visits, hospitalizations and total costs in paricalcitol cohort.

Paricalcitol vs No VDR activator	Parameter Estimate	Utilization/Cost Ratio	95% Confidence Interval	
Infection-related Medications	0.02	1.02	0.97	1.08
Infection-related Outpatient Visits	-0.06	0.95	0.91	0.98
Infection-related Hospitalizations	-0.55	0.58	0.46	0.73
Infection-related Total Costs	-0.52	0.60	0.48	0.74
All-cause Medications	0.17	1.19	1.18	1.20
All-cause Outpatient Visits	-0.05	0.95	0.94	0.95
All-cause Hospitalizations	-0.22	0.80	0.76	0.85
All-cause Total Costs	-0.10	0.91	0.87	0.95

Conclusions: Although infection-related medication use was no different between cohorts, paricalcitol was associated with significantly fewer infection-related outpatient services and hospitalizations, as well as lower total costs compared with no VDR activator treatment. Further studies may be needed to confirm these real-world findings.

Disclosure: Drs. Sterz, Khan, Marx, and Audya are employees of Abbott, the manufacturer of paricalcitol. Drs. Frye, Deering, and Harshaw are employees of EPI-Q, Inc., an independent health outcomes research firm contracted by Abbott Laboratories to conduct this analysis.

Su252 SERUM CYSTATIN C CONCENTRATION IN SUBJECTS OLDER THAN 65 YEARS – POLISH POPULATION PRELIMINARY DATA

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Introduction and Aims: Serum cystatin C concentration is a new index of excretory kidney function which enable estimation of glomerular filtration rate. The cystatin C-based equations do not include age, unlikely those routinely used, creatinine-based (Cocroft-Gault and MDRD equations). The assessment of sensibility and specificity of glomerular filtration rate estimation (eGFR) based on cystatin C equations in elderly was not performed in population based studies.

Aim: The estimation of frequency of chronic kidney disease – CKD (stage 3-5) in older than 65 years.

Methods: Study was performer in 113 subjects in age 55-60 y. and 565 in age 65-101 y. PolSenior study population. Subjects in age 65-101 y. were divided into three subgroups (65-75; 76-85 and over 85 y.). Serum cystatin C concentration was assessed by enzyme-linked immunoabsorbent method. eGFR was estimated cystatin C-based Hoek' equation.

Results: Serum cystatin C concentration was increasing in subsequent age subgroups (0.78 ± 0.17mg/ml in 55-60 y. subgroup, 0.94 ± 0.30mg/ml in 65-75 y. subgroup, 1.06 ± 0.26mg/ml in 76-85 y. subgroup and 1.22 ± 0.35mg/ml in older than 85 y. subgroup). The frequency of subjects with eGFR<60

ml/min (cystatin C-based equation) was increasing with age, from 0.9% in 55-60 y. subgroup through 8.5% in 65-75 y. subgroup and 23.0% in 76-85 y. subgroup to 41.1% in over 85 y. subgroup. In the oldest subgroup the prevalence of subjects with eGFR<60ml/min estimated with cystatin C-based equation was significantly higher than estimated in subjects with MDRD formula.
There was a strong correlation between serum cystatin C concentration and age (r=0.480, p<0.001) in the whole group of analysed subjects. Serum cystatin C concentration was not related to BMI (r=0.026 p=0.51).
Conclusions: The estimation of glomerular filtration rate with cystatin C-based equation causes the increased frequency of chronic kidney disease diagnosed in very old subjects.
Disclosure: Research relating to this abstract was funded by the Polish Ministry of Science and Higher Education number PBZ-MEIN-9/2/2006

Su253 FACTORS RELATED WITH COGNITIVE FUNCTION IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Although end-stage renal disease has been associated with cognitive impairment, factors related with cognitive impairment are not well-established. The objective of this study was to assess the factors related with the cognitive function as measured by Standardized Mini Mental State Examination (SMMSE) in hemodialysis (HD) patients.
Methods: All patients were receiving 4-hour HD 3 times weekly with standard bicarbonate dialysis. Dialysis adequacy was assessed by spKt/V. Sociodemographic, clinical, laboratory variables were recorded. Severity of depressive symptoms and health-related quality of life were assessed by Beck Depression Inventory and Short Form-36 (SF-36), respectively. Cognitive assessment was performed by using SMMSE. Self-reported sociodemographic characteristics of patients included age, sex, living status, educational level, marital status, economic status, presence or absence of sleep disturbance, previous renal transplantation history, presence of diabetes mellitus and presence of coronary artery disease.
Results: Totally, 138 patients (male/female; 73/65, mean age; 52.7±13.1 years, median HD duration: 48 months) were included; 46 patients were diabetic, 43 patients had coronary artery disease. Mean SMMSE score was 26.4±5.7. Independent parameters related with SMMSE score were analyzed with stepwise linear regression analysis; advanced age (Beta:-0.269, P: 0.008), male gender (Beta: +0.215, P: 0.016) and C-reactive protein levels (Beta-0.231, P: 0.007) were independently associated with SMMSE scores. Interestingly, SF-36 scores and Beck Depression Inventory scores were not related with SMMSE scores.
Conclusions: Cognitive function is associated with advanced age, gender and inflammation in HD patients. Unlike what expected, cognitive function is not related with depression and health-related quality of life in HD.

Su254 CKD IN PRIMARY CARE:BASELINE DATA FROM THE RENAL RISK IN DERBY (R^2ID) STUDY

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Introduction and Aims: The majority of patients with CKD3 are managed in primary care and there are few data regarding their characteristics. Moreover, previous studies have largely excluded patients >75years, who make up a large proportion. We undertook a detailed study of these patients with the long-term goal of defining their risk of cardiovascular disease and CKD progression.
Methods: To date 1277 participants with estimated GFR 59-30ml/min/1.73m^2 (on at least 2 occasions 3 months apart) have been recruited from Primary Care Practices. Adult patients of any age were invited. Detailed medical history was obtained and each participant underwent clinical assessment as well as urine and serum biochemistry tests. Skin AF was assessed as a measure of skin AGE deposition using a cutaneous AF device (AGE Reader®, DiagnOptics, Groningen, The Netherlands) and carotid to femoral arterial pulse wave velocity (PWV) was measured with a Vicorder® device. Information was also collected regarding dietary habits.
Results: Table 1 shows the baseline characteristics.

Table 1. Baseline characteristics

	Total Cohort n=1277	CKD 3a n=988	CKD 3b n=284	No Diabetes n=1060	Diabetes n=217	<75 yrs n=666	≥75 yrs n=611
Age (yrs mean ± SD)	73 ±9	72 ± 9	76 ± 9*	73 ± 9	72 ± 8	66 ± 6.6	80 ± 4*
Female (n [%])	769(60)	624(63)	144 (51)*	649 (61)	120 (55)	434(65)	335(55)*
Ethnic group white (n [%])	1244(97)	962(97)	277 (98)	1037(98)	207 (95)	642(96)	602(99)
Smoking- current (n [%])	58 (5)	39 (4)	18 (6)	45 (4)	13 (6)	42 (6)	16 (3)*
Smoking previous (n [%])	640 (50)	479(49)	160 (56)*	523 (49)	117 (54)	308(46)	332(54)*
BMI <25.0 (n [%])	274 (22)	208(21)	65 (23)	251 (24)	23 (11)	128(19)	146(24)
BMI 25.0-29.9 (n [%])	551 (43)	431(44)	116 (41)	466 (44)	85 (39)	267(40)	284(47)
BMI>30.0 (n [%])	452 (35)	349(35)	103 (36)	343 (32)	109 (50)*	271(41)	181(30)*
Central Obesity (n [%])	1111(88)	854(87)	253 (91)	910 (87)	201 (94)*	564(86)	547(90)
DBP(mmHg; ± SD)	73 ±11	74 ±11	71 ±12*	74 ±11	69 ±11*	75 ±11	71 ±11*
SBP (mmHg; ± SD)	134 ±18	134 ±18	136 ±20	134 ±18	136 ±20	131 ±18	137 ±19*
PWV (m/sec; mean ± SD)	10 ±2.1	9.9 ±2.1	10.3 ±2.2	9.9 ±2.1	10.6 ±2*	9.3 ±1.8	10.7±2.2*
Urinary ACR (mean ± SD)	4.9 ±37	2.4 ±12	13.5 ±74*	3.6 ±29	11.3 ±62*	6.1 ±49	3.6 ±14
Microalbuminuria (n [%])$^\delta$	199 (16)	114(12)	85 (30)*	140 (13)	59 (27)*	91 (14)	108 (18)
eGFR (mL/min/1.73m^2)	53 ±10.6	57 ±8	38.5 ±5*	54 ±10.5	50 ±10.5*	55±10.3	50 ±10.2*
Uric acid (mmol/L)	383 ±90	365 ±90	442 ±94*	380 ±90	397 ±89*	375±89	391±90*
Haemoglobin (g/dL)	13.2±1.4	13.3±1.4	12.7±1.5*	13.3±1.4	12.6±1.5*	13.4±1.4	13 ±1.4*
Cholesterol (mmol/L)	4.8 ±1.2	4.9 ±1.2	4.6 ±1.2*	5 ±1.2	4 ±0.8*	5 ±1.3	4.7±1.2*
Previous CVD (n [%])	277 (22)	197 (20)	79 (28)*	220 (21)	57 (26)	110 (17)	167 (27)*
ACEI /ARB use (n [%])	825 (65)	614 (62)	209 (74)*	648 (61)	177 (82)*	415 (62)	410 (67)

*p=<0.05 versus compara tor group $^\delta$ Albuminuria above microalbuminuria threshold

Conclusions: Patients with CKD in primary care, in Derbyshire, are predominantly elderly, female and overweight or obese. The majority fall into CKD stage 3a and significant albuminuria is uncommon. BP was generally well controlled but PWV was elevated. Patients with CKD 3b, diabetes and those over 75 years evidenced a higher risk profile. This study affords a unique opportunity to study CKD 3 and its consequences in patients generally excluded from other studies. Complete baseline data will be presented at the meeting.
Disclosure: This study is funded in part by an unrestricted educational grant from Roche.

Su255 PREVALENCE OF CHRONIC KIDNEY DISEASE AMONG TYPE 2 DIABETIC PATIENTS IN RUSSIA

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Introduction and Aims: The aim of this study was to estimate prevalence of cronic kidney disease (CKD) among type 2 diabetic (T2DM) patients in Russia.
Methods: A random sample of 3,728 adult patients with T2DM was screened. Levels of HbA1c, total cholesterol, triglycerides, creatinine and urinary albumin excretion (UAE) rate were assessed. The test of UAE was considered to be positive if urinary albumin concentration exceeded 20 mg/l (for microalbuminuria -MicroA) and above 200 mg/l (for macroalbuminuria – MacroA) in three urine samples. Glomerular filtration rate (GFR) was estimated using formula the Cockroft-Gault. Statistical analyses were carried out using the STATISTICA 6. Results were considered significant if p<0.05. Results are presented as median and quantiles [25%; 75%].
Results: Prevalence of normalalbuminuria (NormalA) was 53.7%, MicroA – 36.9%, MacroA – 9.4%. Prevalence of stage 2 of CKD (GFR 60-89 ml/min per1.73m^2) was 23.7%, stage 3 of CKD (GFR 30-59 ml/min per1.73m^2) – 6.3%, stage 4 of CKD (GFR 15-29 ml/min per1.73m^2) – 0.2%. GFR<60 ml/min per 1.73m^2 was 4.3% among patients with NormalA, 7.1% – with MicroA (from them 40 patients (41.2%) without diabetic retinopathy), 16.9% – with MacroA. Patients with Micro- and MacroA in comparison with patients with NormalA were older (59 yrs [53;67], 60 yrs [54;68], 58 yrs [52;65] respectively), with longer duration DM (8 yrs [4;14], 11 yrs [6;18], 6 yrs [3;12]), had higher levels of systolic BP (160 mmHg [140;170], 170 mmHg [150;184], 150 mmHg [130;160] respectively) and HbA1c (8,7%[7.3;10.2], 8,9% [7.6;10.4], 8,0% [6.6;9.6] respectively), p<0.001. We compared two groups of patients: 1st – GFR<60 ml/min per1.73m ^{2} (242), 2nd – GFR>60 ml/min per1.73m ^{2} (3469). 1st group was older (1st. – 70 years [65;74], 2nd – 57 years [52;65], p=0.001), more often a female (1st – 75.2%, 2nd – 69.4%, p=0.001), with longer duration DM (1st – 13 yrs [6;20], 2nd – 7 yrs [3;13], p=0.0001), had higher levels of systolic BP (1st – 155 mmHg [140;178], 2nd – 150 mmHg [140;170], p=0.007) and cholesterol (1st – 5.3mmol/l [4.6;6.1], 2nd – 5.1 mmol/l [4.3;5.8], p=0.001) and lower BMI (1st – 26 kg/m^2 [23.9;29.2], 2nd – 30,7 kg/m^2 [27.6;34.2], p=0.001). Levels of HbA1c:1st – 8,2% [6.8;9.5], 2nd – 8,4% [6.9;10], p=0.15.

Conclusions: 46.3% of T2DM patients had pathological urine albumin excretion. UAE was strongly correlated with age of patients, duration of T2DM, levels of HbA1c and systolic BP. 4.3% of patients with NormalA and 41.2% of patients with MicroA are required additional inspection for the purpose of revealing of no diabetic causes of CKD.

Su256 MEASUREMENT OF RENAL FUNCTION ADDS SIGNIFICANTLY TO A RELIABLE PREDICTION OF SURVIVAL IN PALLIATIVE CARE PATIENTS

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Introduction and Aims: An accurate assessment of prognosis is one of the most challenging questions in palliative care medicine. Various clinical scoring systems are used for survival prediction such as the palliative prognostic score (PPS). Impaired renal function is a known risk factor for patient survival, however the influence of kidney function on short term survival of palliative care patients has not yet been studied.
Methods: Therefore we prospectively analyzed all patients admitted to our palliative care unit from Jan 1st to Dec 31st 2008 with respect to the association of markers of renal function with survival compared to currently used scoring systems. In addition to clinical routine measurements serum creatinine (S-Crea), the estimated glomerular filtration rate using the simplified MDRD-formula (eGFR), urine protein (U-Prot) detected by dipstick semi-quantitatively, the Karnofsky-Index (KI) as well as the PPS were determined at admission. The association of these parameters with patient survival was tested comparing the group of patients who died on ward (group 1) with those who could be discharged (group 2) using the Wilcoxon- and Median-Test as well as based on censored data linear rank statistics (Kaplan-Meier estimates and Wilcoxon scores).Their predictive value was calculated by stepwise logistic regression analysis.
Results: Altogether 308 consecutive patients (mean age=70.2±12.9 years, 58.4±49% male, mean stay on ward=8.3±6.6 days) could be included in the study. The mean parameter values at admission were as follows: S-Crea=1.5±1.6 mg/dl, eGFR=86.2±65.8 ml/min and percentage of patients with significant U-Prot=35.2±47.9%. The median KI and PPS were 40% (Range: 10-90%) and 30-50% (Range:<30%->70%), resp. Group 1 differed significantly from Group 2 in S-Crea (x=1.9±2.0 vs. 1.1±0.9 mg/dl, p=0.0007), eGFR (x=76±67 vs. 96±64 ml/min, p=0.0004), KI (x=20 (Range: 10-70) vs. 50 (Range: 20-90)%, p<0.0001) and PPS (x<30 (Range:<30->70) vs. 30-70 (Range:<30->70)%, p<0.0001), whereas U-Prot failed to reach statistical significance (Wilcoxon- and Median-Test, resp.). In the age adjusted survival analysis the same parameters showed a significant association (S-Crea p=0.0406, eGFR p=0.0266, KI p<0.0001 and PPS p<0.0001). Stepwise logistic regression shows only PPS (p<0.0001) and eGFR (p=0.0072) as independent predictors for short term survival. None of the other parameters met the 0.05 significance level for entry into the model.
Conclusions: The determination of eGFR provides significant additional information for assessing the prognosis of palliative care patients. The combination of eGFR with the PPS allows a robust prediction of short term survival time.

Su257 THE EFFECT OF MULTIDISCIPLINARY PREDIALYSIS EDUCATION ON CONTROL OF HYPERTENSION, SMOKING RATE, SALT INTAKE, AND THE USE OF NEPHROTOXIC DRUGS: A SINGLE CENTER EXPERIENCE

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Introduction and Aims: Existing data suggest that multidisciplinary predialysis education programs may improve the medical care of patients and has significantly impact of clinical and quality of life outcomes. The aim of the study was to assess the influence of multidisciplinary predialysis education on control of hypertension, smoking, salt intake and the use of nephrotoxic drugs in predialysis patients.
Methods: A total of 108 predialysis chronic kidney disease patients with an estimated glomerular filtration rate between 11 and 30 mL/min/1.73 m^2 were included to the study. Patients were followed-up for 6 months according to the cigarette smoking status, salt intake, hypertension and the use of the nephrotoxic drugs. All patients and their families received at least 3 times pre-dialysis education seminars, and optimal medical care based on the NKF/DOQI guidelines.
Results: The cigarette smoking rate decreased from 25% to 6% (p<0.05), use of non-steroidal anti-inflammatory drugs decreased from 56% to 2% (p<0.05), use of nephrotoxic antibiotics decreased from 35% to 1% (p<0.05), use of iodinated contrast agents without prophylactic medications decreased from 27% to 3% (p<0.05), the use of excessive salt intake decreased from 40% to0%, the control of hypertension increased from 15% to 85% (p<0.05).
Conclusions: Our data support that an efficient standardized multidisciplinary predialysis education decrease the incidence of smoking and excesive salt intake and the use of nephrotoxic medications and also it helps to control the hypertension in chronic kidney disease patients.

Su258 CARDIORENAL RISK IN NON-DIALYSIS CKD (CKD-ND) PATIENTS FOLLOWED IN THE NEPHROLOGY SETTING: A MULTICENTRIC PROSPECTIVE COHORT ANALYSIS

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Introduction and Aims: Cardiorenal risk associated with CKD-ND has been extensively evaluated in cohorts of patients derived from general population while remaining ill-defined in patients regularly followed by nephrologist. We evaluated the competing risk of mortality versus ESRD and the relationship between terapeutic goals and outcome in a large cohort of CKD-ND patients prevalent in Italian renal outpatient clinics.
Methods: We considered eligible all the consecutive pts with diagnosis of CKD and estimated GFR <60 ml/min/1.73m^2 consulting the 26 participating clinics from Sep 2002 to Mar 2003. We excluded pts with kidney transplant, changes of GFR >30% in the previous 6 months, and follow-up in the clinic <6 months. For renal survival analysis, pts were followed until ESRD, death or March 31st 2009.
Results: We studied 1248 pts (57% males, age 67±14y, diabetes 28%, previous cardiovascular disease-CVD 32%) with estimated GFR 31±14 mL/min/1.73 m^2 (stage III: 49%, stage IV: 36%; stage V: 15%) and 24h proteinuria 1.04±1.56 g. Blood pressure (BP) was 139±18/81±10 mmHg, cholesterol (CHO) 199±40 mg/dL, serum phosphorus (sP) 3.9±0.8 mg/dL, haemoglobin (Hb) 12.5±1.8 g/dL. Mean number of antihypertensive drugs was 2.2±1.1; CEI or ARB were prescribed in 72% pts, statin in 22%, ESA in 13%. Salt intake was <6 g/d in 22% pts and protein intake was <0.8 g/kg/d in 54%. During follow-up, that lasted a median of 57 mo, renal death occurred in 663 pts (363 ESRD, 222 pre- and 78 post-ESRD deaths), with a annual incidence rate of 8.3% for ESRD and 5.7% for death. Competing risk analysis demonstrated that risk of ESRD accounted for the large majority of the risk of the composite outcome in stage 4 and 5 while risk of death overcame risk of ESRD in stage 3. Table shows the results of Cox analysis (HR and 95%CI) stratified for centre and CKD stage and adjusted for gender and smoking.

	ESRD	Death
Age (5 years)	0.93 (0.90-0.98)	1.56 (1.46-1.66)
BMI (g/kg2)	0.97 (0.95-0.99)	0.99 (0.97-1.02)
Diabetes	1.16 (0.91-1.48)	1.31 (1.02-1.70)
CVD	1.30 (1.02-1.65)	1.38 (1.08-1.76)
ESRD (time-dependent variable)	–	1.52 (1.08-2.14)
BP≥130/80 mmHg	1.09 (0.77-1.55)	0.71 (0.49-1.03)
Hb <11 g/dL	1.26 (0.98-1.62)	1.44 (1.09-1.90)
Cholesterol >190 mg/dL	0.96 (0.77-1.19)	0.96 (0.76-1.22)
sP >4.5 mg/dL	1.46 (1.14-1.86)	1.14 (0.83-1.58)
Uric acid >7mg/dL	0.92 (0.73-1.17)	1.25 (0.97-1.60)
Proteinuria >0.5 g/24h	1.90 (1.47-2.46)	1.38 (1.08-1.78)

Conclusions: The majority of the CKD-ND patients followed in Italian renal clinics advanced to ESRD with higher competing risk of death only in CKD stage 3. The predictive role of risk factors specific of CKD is predominant; in particular, treatment of proteinuria, anemia and hyperphosphatemia are identified as potential areas of improvement in nephrology clinical practice.

Su259 AGONIST-STIMULATED VASODILATION CORRELATES WITH LEVELS OF ANTINUCLEAR ANTIBODIES IN PATIENTS WITH SYSTEMIC SCLEROSIS

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Introduction and Aims: Abnormalities of both microvascular structure and function are key features of systemic sclerosis (SSc). We investigated endothelial function in the skin microcirculation of patients with SSc compared with healthy controls as well as the putative association of agonist-stimulated vasodilation with serological markers of inflammation and activation of the immune system in patients with SSc.
Methods: We used transdermal iontophoresis and Laser Doppler Scanning (MoorLDI V.5.0, Axminster, UK) to assess changes in skin blood flow in 8 patients with SSc (age 56.3±3.2 years) and in 10 age-matched, healthy controls. Acetylcholine 1% (ACh) and sodium nitroprusside 0.01% (SNP) were used to evaluate endothelium-dependent and endothelium-independent vasodilation, respectively. ACh was delivered using an anodal current (7 x 0.1 mA for 20 seconds (s), followed by 1x 0.2 mA for 20 s) and SNP was applied using a cathodal current (2 x 0.1 mA for 20 s, followed by 1 x 0.2 mA for 20 s). Subjects were blinded as to the succession of the substances. Levels of high-sensitive CRP, TNF-alpha, interleucin-6 (IL-6), endothelin-1 (ET-1) and antinuclear antibodies (ANA) were determined. Blood flow is expressed in arbitrary perfusion units (PU). Data are presented as mean±SEM. Statistical significance was assessed using t-test and Pearson correlation analysis.
Results: ACh-induced vasodilation was reduced in patients with SSc (SSc: 190±14 PU vs. controls: 270±13 PU, $P<0.001$) while vasodilation to SNP did not differ significantly between groups (SSc: 199±26 PU *vs.* controls: 240±33 PU, $P>0.05$). ACh-induced vasodilation was negatively correlated with ANA-levels in patients with SSc ($r= -0.66$, $P<0.001$), a weaker negative correlation was also seen for ANA and SNP-mediated vasodilation ($r= -0.35$, $P<0.01$). No correlation was demonstrated for the markers of inflammation and vasodilatory responses.
Conclusions: Our investigations in patients with SSc indicate that activation of the immune system as represented by elevated ANA may directly reflect impairment of microvascular function in SSc. At present these data have to be regarded as preliminary and studies in larger numbers of patients are needed to determine the clinical relevance of our results.

Su260 PREGNANCY MAY BE THE OCCASION FOR EARLY CKD DIAGNOSIS: ON 35 NEW DIAGNOSES OVER 150 PREGNANCIES IN THE SAME SETTING (2002-2010)

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Introduction and Aims: Chronic kidney disease (CKD) is at the same time a well known challenge for pregnancy and an often difficult differential diagnosis versus preeclampsia. In fact, CKD is frequently asymptomatic, and often diagnosed only if specifically searched for. In particular in the low risks groups, such as young woman, diagnostic occasions may lack and severe, potentially treatable diseases may go unrecognised.
Aim of the study was to evaluate the prevalence and the main clinical and outcome features of the new diagnosed of CKD performed during pregnancy in women referred to our Outpatient unit dedicated to pregnant CKD patients, or hospitalised in the Materno-foetal Unit.
All cases were followed, and all the diagnosed were posed by the same nephrology gynaecology team.
Methods: Prospective analysis of CKD cases referred to the Unit, January 2002– January 2010. Start of observation: referral to the unit. End of observation: 1 month after delivery. In the period, 165 patients with CKD were referred, 54.5% in the first trimester, 35.5% in the second and 10% in the third trimester.
Results: Over 156 pregnancies, 39 patients had a diagnosis of renal disease during pregnancy. Excluding 4 cases with acute pyelonephritis and without predisposing factors, CKD was diagnosed in 35 new patients.
The diagnosed were glomerular diseases in 10/35 patients (2 dropped from follow-up after delivery; 7 underwent a renal biopsy: IgA nephropathy in 4, membranous nephropathy in 3, one is under study at present); in 6 cases CKD stage 4-5 was diagnosed; in all the first clue was either a small for gestational age baby or alteration in biochemical tests during pregnancy. Of note, an interstitial nephropathy was diagnosed in 5 cases (nephrocalcinosis, with hydro-electrolyte disorders in 3 cases), persistent urinary anomalies in 4 cases, and urological anomalies in the remaining 10.
Referral often occurred late (23% in the first trimester, 60% in the second and 17% in the third). At referral, median creatinine was: 0.7 mg/dl, median proteinuria: 0.3 mg/24 ore; median GFR: 107 mL/min. The mean age of the new CKD diagnoses was 30 years; 29 women were Caucasian.
30 women delievered; 18 babies were premature, 12 at terms. all the babies are presently well, in spite of frequent need for cesarean sections (54%) and low birth weight (mean 2478.2±871.9 g), as a marker of intrauterine growth retardation or small for getational age. 25 woman are presently on regular nephrological follow-up.
Conclusions: Pregnancy is a precious occasion for the early diagnosis of CKD. The higher awareness of this condition, in particular in the differential diagnosis with preeclampsia, is precious tool for ensuring appropriate and timely interventions.

Su261 THE CHANGES IN RENAL FUNCTION AFTER MYOCARDIAL INFARCTION

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Introduction and Aims: Chronic kidney disease (CKD) is a risk factor for cardiovascular diseases, however how cardiovascular events affect the progression of CKD is largely unknown. This study investigated the effect of myocardial infarction on the annual changes in renal function.
Methods: We compared the annual decline in eGFR in 138 myocardial infarction patients (67 years, male 70%, diabetes 41%) with those in matched general population and longitudinally evaluated the sequential changes in eGFR between before and after cardiac event in 10 patients.
Results: The baseline eGFR in cardiac patients was 73.4±22.9 mL/min/1.73m^2 and the 1-year decline in eGFR after myocardial infarction was significantly greater than that in control (-9.0±13.0 vs. -1.9±9.3 mL/min/1.73m^2/year, mean ± SD, $p<0.001$). The background of cardiac patients (age, gender, diabetes, proteinuria, ARB or statin treatment) did not significantly affect the changes in eGFR. The mean eGFR decline between 1-year before and 3-year after myocardial infarction were -1.7, -7.0, -5.9 and -4.6 mL/min/1.73m^2/year.
Conclusions: This result indicates that the eGFR decline was significant and persistent after myocardial infarction, suggesting that careful follow-up of renal function is necessary after cardiac events.

Su262 SIMPLE RISK STRATIFICATION STRATEGY FOR PATIENTS WITH STAGE 3-5 CKD

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Introduction and Aims: Population-based studies have reported outcomes and risk factors for patients with chronic kidney disease (CKD), defined primarily by decreased estimated glomerular filtration rate (eGFR). They are characterized by old age, low proteinuria level, and stage 3 CKD. However, many patients referred to nephrologists are younger and have overt proteinuria and advanced CKD. This study evaluated the association between outcomes and those factors among referred CKD patients.

Methods: We retrospectively reviewed 461 referred patients with stage 3-5 CKD from January 2004 to December 2007. Key outcomes were death and end-stage renal disease (ESRD). Patients were followed from the time of first serum creatinine measurement to December 2008.
Cumulative mortality was estimated using the Kaplan-Meier method and compared using the log-rank test. Cumulative probability of ESRD was estimated using the competing risk method and compared using the Gray test in the presence of death as a competing risk. Univariate and multivariate regression analysis was conducted to evaluate the association between baseline characteristics and ESRD, using Cox model for death and Fine and Gray model for ESRD. Then the outcomes of death before RRT and ESRD were re-analyzed stratifying all patients by age, overt proteinuria, and CKD stages.
Results: The median age of subjects was 67.0 years, and median follow-up was 2.4 years. Overt proteinuria was present in 56.3% of subjects. For stage 3, 4, and 5 CKD, cumulative mortality and probability of ESRD at 3 years was 10.7 and 6.1%, 12.5 and 27.9%, and 16.5 and 79.4%, respectively. Using regression model, age was a determinant for death, whereas overt proteinuria was independently associated with ESRD. Among stage 3 CKD patients over 65 years without overt proteinuria, the incidence of death before renal replacement therapy (RRT) was 3.0/100 patient-years and none of them had ESRD (Figure). In patients with advanced CKD and overt proteinuria, the incidence of ESRD was substantially higher than that of death before RRT.

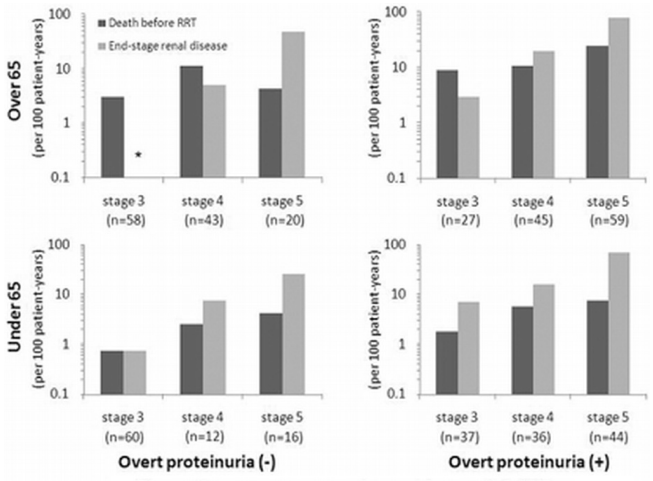

Figure. Outcomes among patients with stage 3-5 CKD stratified by age and overt proteinuria

Conclusions: In the present study, we showed different backgrounds and outcomes in CKD patients referred to nephrologists compared to patients included in population-based studies. Advanced CKD and overt proteinuria were common among referred patients. Proteinuria and age contributed differently to CKD outcomes. Stratification of heterogeneous CKD patients by those factors and CKD stage could be a simple strategy for the selection of patients who should be intensively treated by nephrologists.

Su263 VASCULAR CALCIFICATIONS AND CARDIOVASCULAR RISK FACTORS IN CHRONIC KIDNEY DISEASE

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Introduction and Aims: Vascular calcifications is a common finding in patients with chronic kidney disease (CKD) and are associated with arterial stiffness and mortality. The aim of this study was to evaluate vascular calcifications by two simple plain X-ray scores in a cohort of patiens with CDK stage 4 and 5 non in dialysis and its realtionship with traditional and non traditional cardiovascular risks factors.
Methods: We studied 177 patients, mean age was 63±13 years, 61.6% were men, mean Glomerular Filtration Rate (GFR-MDRD4) was 20.8±5.6 mL/min, 80% CKD stage 4 and 20% CKD stage 5, 65% had diabetes, mean body mass index was 31.9±25, 50% had BMI>30, 45.8% of the patients had previous cardiovascular diseases (23.2% coronary artery disease, 16.4% cerebrovascular disease and 25.4% peripheral arterial disease). All subjets underwent a plain X-ray of pelvis, hands and lateral lumbar column and semicuantitave vascular calcification score of abdominal aorta, iliacs, femoral, radial and interdigital arteries were calculated as described by Adragao (SVCS) and Kauppila. 24 hours ambulatory blood pressure monitoring (ABPM) was also registred. Assesment of biochemical parameters included lipid perfil, mineral metabolism, serum albumin, 25 hydroxyvitamin D, PCR and Homocysteine.
Results: Only 23.7% of patients had no vascular calcifications according Adragao score and 55.4% had more than 3 points. Only 13% of the patiens had no vascular calcifications according Kauppila score, and 41.2% had more than 12 points.
Table 1 shows the results of diferent variables according to SVCS Adragao and Kauppila scores:

Table 1

	SVCS<=3	SVCS>3	Sig.	kauppila<=12	kauppila>12	Sig.
Age	58.2	67.2	p<0.001	57.9	69.8	p<0.001
% Diabetes	38.5	87.5	p<0.001	51.9	83.6	p<0.001
Prev. Vasc. Dis.%	26.9	62.6	p<0.001	32.7	64.4	p<0.001
Haemoglobin	12.2	11.6	p<0.05	12	11.8	n.s.
Albumin	4.1	3.9	p<0.005	4	4	n.s.
25 hydroxi vit D	25.3	18.9	p<0.001	23.9	19	p<0.005
PCR	0.43	0.65	p<0.05	0.5	0.6	n.s.
Homocisteine	20.2	23.1	p<0.05	20.8	23	p=0.083
Pulse pressure	54.6	71.7	p<0.001	57.8	71.8	p<0.001

No differences were seen regarding BMI, calcium, phosphate, iPTH, total cholesterol or triglycerides. On multivariate analysis age, diabetes, 25 hydroxyvitamin D level and pulse pressure in ABPM were related with vascular calcification scores.
Conclusions: Vascular calcifications are highly prevalent in CKD patients prior to dialysis initiation and they are related with some cardiovascular risks factors like age, prevalence of diabetes, hypertension and previous cardiovascular diseases.
Low levels of 25 hydroxyvitamin D seems to be a risk factor for the development of vascular calcifications. We could not see a relationship with other alterations of bone and mineral metabolism parameters.

Su264 NGAL (NEUTROPHIL GELATINASE-ASSOCIATED LIPOCALIN) AS A MARKER OF KIDNEY FUNCTION IN THE ELDERLY

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Introduction and Aims: Prevalence of chronic kidney disease is about 10-16%, mainly in the elderly. According to K/DOQI GFR below 60 ml/min is associated with the increased risk of cardiovascular morbidity and mortality. Since creatinine is an unreliable parameter describing kidney function, the search for new and sensitive marker of kidney function is under way. NGAL (neutrophil gelatinase-associated lipocalin) has been recently prove useful in the quantitation of chronic kidney disease. The aim of the study was to assess prevalence of chronic kidney disease using estimated GFR/creatinine clearance according to different formulae in 991 consecutive patients with normal serum creatinine with stable coronary artery disease in relation to age (below and over 65 years).
Methods: We assessed kidney function according to the simplified MDRD, Cockcroft-Gault, CKD-EPI formulae. Additionally, we assessed NGAL and cystatin C in 400 patients (200 over 65 years of age and 200 younger than 65, they were sex- and creatinine-matched).
Results: In the group of patients below 65 years with normal serum creatinine according to Cockcroft-Gault formula stage 3 CKD was found in 31%, according to MDRD in 5%, and according to CKD-EPI in 5%. Normal eGFR i.e. 90-120 ml/min was found in 20% (Cockcroft-Gault formula), 40% (MDRD) and 39% (CKD-EPI).

In the group of elderly patients (over 65 years) with normal serum creatinine according to Cockcroft-Gault stage 3 CKD i.e. GFR 30-59 ml/min was found in 61%, stage 4 CKD in 3.5% patients. Normal eGFR was found in 6.9% of patients older than 65 years. According to MDRD stage 3 was diagnosed in 17%, while normal eGFR in 22% of patients older than 65 years. According to CKD-EPI stage 3 was diagnosed in 19% of patients over 65 years of age, stage 4 in 1%, and normal eGFR in21% of this population. In the population of 400 patients, where NGAL was assessed found that patients over 65 years of age had significantly lower eGFR (MDRD, CKD-EPI, Cockcroft-Gault formulae) than their younger counterparts despite identical serum creatinine. They also had significantly lower hematocrit, despite similar Hb, lower platelet count, higher serum fibrinogen, higher systolic and lower diastolic blood pressure, significantly higher serum NGAL and cystatin C, but similar urinary NGAL. Serum NGAL correlated with age, hematocrit, leukocyte, platelet and erythrocyte count, eGFR (any formula), serum creatinine, fasting glucose, HbA1c, fibrinogen, NYHA class, SBP, diabetes duration. In multiple regression analysis kidney function (either eGFR or serum creatinine), cystatin C and systolic blood pressure (beta value 0.18, p=0.05) were predictors of serum NGAL.

Conclusions: In our study we found a very high prevalence of CKD up to 61%, on the basis of estimated GFR/creatinine clearance, in elderly patients with coronary artery disease and normal serum creatinine. NGAL could be a sensitive marker of kidney function, particularly in elderly patients with other risk factor for kidney damage i.e. hypertension.

Su265 INSULIN RESISTANCE IN PATIENTS WITH CHRONIC KIDNEY DISEASE TREATED WITH PERITONEAL DIALYSIS, HEMODIALYSIS, OR TRANSPLANTATION

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Introduction and Aims: Uremia can cause insulin resistance and thereby lead to hyperinsulinemia, glucose intolerance, and dyslipidemia. Atherosclerotic cardiovascular complications are more frequent in people with chronic kidney disease (CKD) than in people with normal kidney function. Insulin resistance can contribute to these complications. This study investigated whether insulin resistance in patients with CKD differed according to type of renal replacement therapy.

Methods: The study included 31 patients on haemodialysis (HD), 36 patients on peritoneal dialysis (PD), 36 transplanted patients (Tx), 25 patients with chronic kidney disease (CKD) stage 3-4, and 31 healthy controls. Fasting blood glucose and insulin levels were assessed in all participants. Homeostasis model assessment (HOMA) scores were calculated by using formula; [(insülin (μU/ml) x glukoz (mg/dl)/405]. Individuals who have equal to or more than 2.7 of HOMA score were accepted as to have insulin resistance.

Results: There was no different between groups acording to mean age (yr): 46.7$\pm$15.3 in HD group, 43.6$\pm$14.6 in PD group, 36.2$\pm$11.8 in Tx group, 49.4$\pm$16.3 in CKD group and 35.6$\pm$8.0 in control. HOMA indices are 5.1$\pm$2.9 in HD group, 9.3$\pm$6.8 in PD group, 5.4$\pm$3.1 in Tx group, 4.7$\pm$1.9 in CKD group and 2.5$\pm$1.2 in control. HOMA score in PD group was higher significantly than other groups ($p<0.001$). Rates of insuline resistance in patients groups were markedly more than control group (92.8% in HD, 96.8% in PD, 86.1% in Tx, 83.3% in CKD, 35.5% in control; $p<0.001$).

Conclusions: Prevalance of insulin resistance has been increased in patients who diagnosed CKD and underwent renal replacement therapy.
Especially PD group has been shown the highest rate of insuline resistance.

Su266 FACTORS INFLUENCING THE LOW PREVALENCE OF CENTRAL VEIN CATHETER USE IN OUR HAEMODIALYSIS UNIT: AN EIGHT YEARS DATA

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Introduction and Aims: The arteriovenous (AV) fistula consists the gold standard for the nephrologists as it is the optimal vascular access for chronic haemodialysis patients (HDpts). It is generally accepted that the preferred access for the HD is a mature native AV fistula, followed by AV grafts and then the central venous (CV) catheters. Data from different countries or even from center to center are controversial, regarding HD unit's policy and other specific or unspecified factors, that might influence the prevalence of certain type of vascular access for HD in the clinical practice. On the other hand CV catheters in HDpts have been associated with a number of side effects like thrombosis and higher rates of hospitalization due to access related infections, compared to HDpts with AV fistulae or even AV grafts.
The purpose of our study was to determine factors and attitudes that might influence the prevalence of certain type of vascular access for our HDpts, as our policy for years has been an optimal patient selection for specific access modalities and institution of an early access intervention.

Methods: Data sheet were retrospectively analysed only for 463 chronic HDpts (202 males and 261 females, age 69$\pm$11.87 years) treated in our unit between 2002 and 2009. Patients who were receiving HD for acute kidney injury or acute intoxication or they will be treated with PD, were excluded from the study. Data were collected from the patients' records and included: 1. the type of vascular access being used for HD 2. vascular access history 3. time on HD 4. factors or attitudes for a CV catheter use as vascular access for HD.

Results: There were 463 consecutive chronic HDpts who started dialysis in our center over the aforementioned period. AV fistula was used in 72.78% (n=337), in 17.92% AV grafts (n=83) and CV catheters only in 9.30% (n=43). It should be mentioned that the vast majority of the AV fistulas (96.15% n= 324) referred to mature access which could be cannulated with two needles and deliver a high blood flow. A binary logistic regression analysis was performed to determine which variables were independent predictors of CV catheter use. It was found that male gender, patient refusal, older age, peripheral vascular disease, and shorter time in HD were the main predictors that influenced the vascular access creation, but only in the first 3 months of HD initiation.

Conclusions: Our study indicates that there are specific factors and attitudes that might influence the use of CV catheters as a vascular access for HD which we should take into account for optimal patient selection. The relatively very low prevalence of CV catheter use in our unit compared to the literature data, is due mainly to our continuous policy to encourage and persuade the patients for the advantage of an early AV fistula creation, 6-8 months before the HD estimated initiation.

Su267 REVERSING THE PANDEMIC OF ESKD IN THE ELDERLY WITH 'REHABILITATION' – CAN IT BE DONE?

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Introduction and Aims: The new incident elderly patients with treated end stage kidney disease (ESKD) has been rising in the U.K. The highest take on rate has been reported to be over 400 per million population (PMP) per year in the 75-79 age band compared to an average take on rate of 110 PMP. As a large tertiary renal unit in the north of England serving over a million population, with 350 haemodialysis (HD) and 80 peritoneal dialysis (PD) patients, we serve a large number of elderly patients with advanced kidney disease (CKD 5). The increase co-morbidity and functional decline in the elderly kidney patients are massive but interval rehabilitation and improve co-ordination with geriatric care may reverse this trend.

Methods: The data of all patients attending our renal department were retrieved from our renal computer (PROTON). The CKD stages of the patients have been assigned by the responsible physicians. We were able to study the prevalence of our patients and the darbepoetin alfa usage in different CKD stages (3 – 5), including conservative management (CM) patients according to age.
We have observed a small number of advanced kidney disease patients whom we have been able to delay dialysis following a period of in-patient therapy and targeted rehabilitation.

Results: There was a total of 2414 CKD patients with CKD 3-5 including 60 patients coded for conservative management (CM). Approximately half of our CKD patients and all CM patients were over 70 year old. One third of the prevalent HD patients and 12% of the PD patients were over 70 year old. There were 450 CKD patients receiving darbepoetin alfa including 250 patients who were over 75 (and including all CM patients).

Twelve months ago, two elderly obese type 2 diabetic patients with AV fistula were successfully rehabilitated, following admission with general fluid overload, and had avoided otherwise long term haemodialysis so far.

Conclusions: It is estimated that elderly patients with ESKD would continue to increase in the next two decades. In those elderly patients who went onto require dialysis, unfortunately, functional decline and mortality are high but it would appear that in some patients, interval rehabilitation could be successful in delaying long term dialysis and reversing functional decline. The provision of interim assessment to detect pre-frailty, along with an in-patient rehabilitation program with goal setting have been shown to benefit elderly kidney patients and reduce long term health care cost (Jassal, AJKD 2007).

It is crucial that we improve the co-ordination with our geriatric colleague to provide integrated care to elderly kidney patients. Like the U.S.A., training in geriatric medicine should be promoted and included in the training of nephrology fellow in Europe.

Su268 THE DEVELOPMENT OF LIMITED-CARE DIALYSIS CENTERS IN FRANCE

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Introduction and Aims: Limited care haemodialysis units (LCU), conceived as an alternative to in-centre haemodialysis (CH) was introduced in 2002 by decree within the French health care system. This study reports on its impact on evolution of ESRD treatment modalities in France from 2005 to 2008. It is based on data from the French REIN registry that is used here to monitor the impact of public health policy on the actual management of ESRD.

Methods: Patients characteristics and treatments modalities are described each year on December, 31 for all patients treated in one of the 12 districts that were contributing exhaustive data to the French REIN registry from 2005. Changes in treatment modalities are reported for patients under renal replacement therapy (RRT) on December, 31 2006 with their treatment modality one year before and their status and treatment one year later.

Results: a) Evolution of treatment modalities: During the study period, the number of patients under RRT rose 4.7% annually, with a persistent increase of in-centre haemodialysis (mean yearly variation +3.9%), a stagnation of peritoneal dialysis (+0.3%) and a decrease in out-centre haemodialysis (mean yearly variation -2.0%). The development of LCU revealed to be slower than expected, with an mean yearly variation of +24.2% instead of +67%. The number of patient living with a functioning transplanted kidney increased (yearly mean variation +6.6%). The percentage of patients treated in LCU increased from 5.4 to 9.9% of dialysed patients.

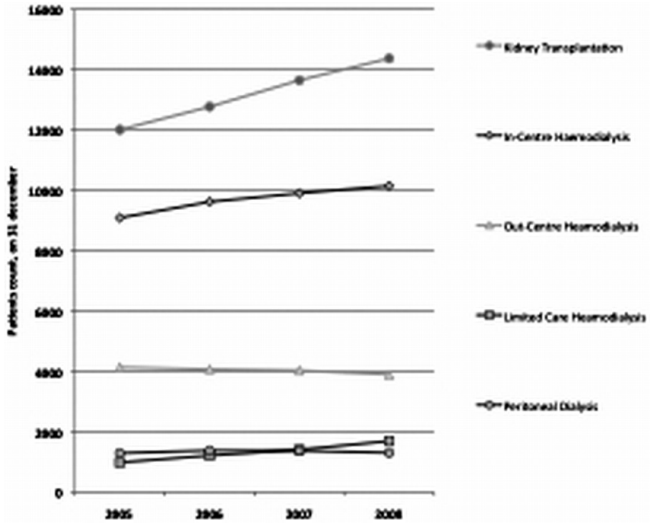

Figure 1. Evolution of RRT modalities, treated patients count on 31 December.

b) Origin and Outcome of patients treated in LCU on December 2006: Among patients who were treated in LCU on December 2006, 499 moved to LCU during year 2006 of whom 44% were incident patients who started a RRT, 31% were treated by in-center haemodialysis and 21% by out-center haemodialysis on December 2005.

Among patients who were treated in LCU on December 2006, 342 discharged from LCU during year 2007 of whom 35% died, 28% were treated by in-center haemodialysis, 23% by transplantation and 11% by out-center haemodialysis on December 2007.

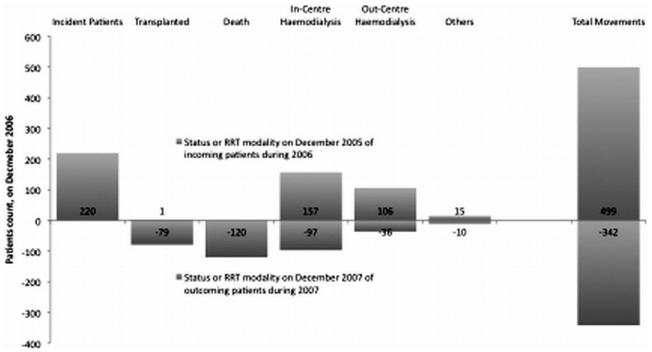

Figure 2. Origin and outcome of patients treated in LCU on December 2006.

Conclusions: The introduction of LCU in France, slower than scheduled in regional healthcare management scheme, appeared to be mainly supplied by incident patients, less sickest in-center patients but also by the sickest out-centre patients. The increase of kidney transplantation was another determinant of the decrease of out-centre dialysis in France.

Su269 THE DEMOGRAPHIC AND CLINICAL PROPERTIES OF PATIENTS WITH CKD STAGE 5 NOT RECEIVING RRT: 1,854 PATIENTS FROM 6 UK RENAL CENTRES

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Introduction and Aims: The demographic and clinical properties of patients reaching chronic kidney disease stage 5 prior to starting renal replacement therapy have not previously been described in a large multicentre study of UK patients. The aim of this study was to describe the baseline demographics of this population.

Methods: All adult patients reaching CKD stage 5 (defined as two eGFR results $<$15ml/min/1.73m^2, more than 90 days apart with no intervening eGFR results $\geq$15 and not having received RRT) during 2006 and 2007 at six UK renal centres were included in the study. Baseline clinical and demographic data including: age, gender, ethnicity, primary renal disease and socioeconomic deprivation were extracted direct from the centres' IT systems. Patients with missing ethnicity or PRD data were excluded from the respective analyses.

Results: 1,854 patients reached CKD stage 5 according to the above definition in the 6 centres during the two-year period. The median age at which CKD stage 5 was reached was 69.8 years. The age group breakdown was: <45 years (12.3%), 45-64 (26.5%), 65-74 (26.1%) and 75+ (35.0%). The male: female ratio was 1.30:1. The ethnicity breakdown was: White (86.2%), South Asian (9.2%), Black (3.5%) and Other (1.1%) (after excluding 372 patients (20.0%) with missing ethnicity data). The proportion of primary renal diseases is shown in table 1 after excluding 395 patients (21.3%) with missing PRD data.

Table 1. Primary renal disease amongst patients reaching CKD stage 5

PRD	%
Glomerulonephritis	9.9
Diabetic nephropathy	24.8
Reno-vascular disease and hypertension	11.1
Polycystic kidney disease	9.2
Pyelonephritis	11.3
Diagnosis uncertain	27.1
Other diagnosis	6.6

Conclusions: This is the first large, multicentre study of the epidemiology

of patients reaching CKD stage 5 (prior to RRT) in the UK. This analysis describes the baseline clinical and demographic properties of this cohort.

Su270 SERUM CYSTATIN C IS A SIMPLE TOOL TO PREDICT INCIDENT CHRONIC KIDNEY DISEASE IN PATIENTS RECEIVING HEMAOPOIETIC STEM CELL TRANSPLANTATION

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Introduction and Aims: Chronic kidney disease (CKD) is a significant cause of morbidity and mortality in survivors receiving allogeneic hematopoietic stem cell transplantation (HCT). We assessed if an early elevation in serum cystatin C (Cys) during the post-HCT period is useful to predict incident CKD following HCT.
Methods: We performed a retrospective cohort study in 33 consecutive individuals (15 males and 18 females) receiving allogeneic HCT at the Komagome hospital from January 2007 to July 2009. Mean patient age was 42.4±11.5 years. Estimated glomerular filtration rate (eGFR) was determined based on serum creatinine (Cr) using the MDRD equation of K/DOQI guideline. Mean eGFR at baseline was 91.6±24.8ml/min/1.73m^2. Incident CKD was defined as eGFR < 60 ml/min/1.73m^2 at the time of 6 months following HCT. Cys level was measured before and 1 month after HCT. Multivariate logistic regression analysis was performed to find factors associated with incident CKD, adjusted for demographic and laboratory confounders. The ability of variables for predicting incident CKD was tested using area under the receiver operating characteristic (ROC) curve analysis.
Results: The incidence of CKD was 42%. While serum Cr levels showed no significant change between before and 1 month after HCT [0.65±0.15 mg/dl (before) and 0.74±0.31 mg/dl (1 month after HCT, P=0.13)], Cys levels significantly increased from 0.73±0.17 mg/L to 1.25±0.65 mg/L (p<0.0001). Multivariate logistic regression analysis showed that the incidence of CKD was significantly associated with Cys at 1 month after HCT (OR 284.3, 95%CI 2.15-1341133, p=0.018). The area under the ROC (AROC) curve analysis showed that Cys at 1 month after HCT could be a simple predictor for incident CKD with moderate accuracy (AROC=0.765;cut-off value 1.09 mg/L, p=0.0347).
Conclusions: Cys level at 1 month after HCT could be a simple tool for early identification of HCT patients at risk for incident CKD.

Su271 THE KNOWLEDGE AND SELF-DECLARED HEALTH-PROMOTING BEHAVIOUR WITH RESPECT TO BREAST CANCER AMONG HEMODIALYZED WOMEN

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Introduction and Aims: The frequency of occurrence of tumour among chronic renal failure patients undergoing renal replacement therapy is considerably higher than in the overall population. Breast cancer is one of the most common tumours among women. Chronic renal failure and renal replacement therapy contribute to the fact that both the patients' and medical stuff's attention is mainly focused on the basic illness and some possible complications. Pro-health activity and the interest in other diseases including cancerous ones are commonly significantly decreased. The aim of the research is to investigate the scope of knowledge and self-declared health-promoting behaviour about breast cancer among hemodialyzed women.
Methods: The research method used was a questionnaire conducted among 230 women at the age of 50, 8±12 years treated on the basis of repeated dialysis in 24 dialysis outpatient facilities in the south of Poland (DW). The questions concerned the knowledge about breast cancer, the ability of breast self-examination and also health-promoting behaviour following the doctrines of the Polish Gynecologist Association (PTG) and recommended anti-tumour prevention. The controlled group comprised 138 healthy women at the age of 49,3±0,8 (CG).
Results: Out of 16 questions concerning this knowledge, the percentage of women answering at least half of them correctly was significantly smaller in the group of dialyzed women (DW) than in the controlled one and amounted respectively to 37,4% versus 68,8% (p<0,0001). The mean number of correct answers was also much smaller in this group, respectively 7,1±3,5 vs. 10,0±3,0 p<0,0001 than in the CG. The percentage of women following the recommendations of PTG concerning the prevention and early diagnostics of changes in the mammary gland (breast self-examination, ultrasonography, mammography, visiting a gynecologist) is also considerably lower among DW than in the case of CG and amounts respectively to 23, 4% vs. 52, 2% (p<0,0001). Breast self-examination is not performed by a considerably higher number of dialyzed women (44, 4%) in comparison to healthy ones (20, 3%). Among hemodialyzed women, better results of the analysed parameters concerned the women with higher education. However, one does not notice the differences resulting from the time of dialysis therapy, place of living (city or village) or a subjective evaluation of life satisfaction.
Conclusions: Hemodialyzed women differ from the women belonging to the overall population with respect to their knowledge and self-declared health-promoting behavior concerning breast-tumour prevention.

Su272 LEFT VENTRICULAR HYPERTROPHY IS THE STRONGEST RISK FACTOR OF PROGRESSION TO DIALYSIS IN NON DIABETIC CHRONIC KIDNEY DISEASE

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Introduction and Aims: The role of cardiovascular factors in predicting renal outcome of patients with chronic kidney disease (CKD) has not been extensively elucidated. Aim of this prospective study was to evaluate the impact of left ventricular hypertrophy (LVH) on general and renal outcome in non diabetic patients with CKD.
Methods: We studied 144 patients (99 men; age 62±14 yrs) with stage 3-4 CKD, who had baseline assessment of left ventricular mass index (LVMi) by echocardiography, estimated glomerular filtration rate (eGFR) by 4-variable MDRD equation, 24-hr blood pressure profile by ABPM, and 24-hr proteinuria. Combined endpoint was progression to ESRD requiring dialysis, or death within 5 years.
Results: Forty-nine patients (34%) progressed to dialysis, 24 (17%) died, 57 (39%) were dialysis-free after 5 years, and 14 were lost to follow-up. Cox proportional hazard analysis showed that increased LVMi (HR 1.24, 95% CI 1.16-1.31 for each 10 g/m^2 increase, P<0.001) together with eGFR (7% risk increase for each 1 mL/min reduction, P<0.001) and 24-hr proteinuria (HR 1.21, 95% CI 1.02-1.45, P=0.03) were the significant predictors of the combined endpoint, whereas the HR for the risk of dialysis was 1.45 (95% CI 1.26- 1.66) for each 10 g/m^2 greater baseline LVMi, and 0.89 (95% CI 0.84- 0.94) for each 1 mL increase in eGFR (both P<0.001). When evaluating the predictive role of LVMi on outcome using AUC-ROC curves, the overall performance of the model including LVMi was superior to the model without LVMi for both the combined endpoint (AUC 0.92, 95% CI 0.86-0.98, and AUC 0.68, 95% CI 0.56-0.80, respectively), and the endpoint of progression to dialysis (AUC 0.923, 95% CI 0.85 to 0.99 and AUC 0.69, 95% CI 0.51 to 0.87, respectively).
Conclusions: LVH proved to be the strongest predictor of the risk of progression to dialysis in non diabetic CKD, whereas the role of both proteinuria and elevated blood pressure resulted negligible. Regardless of whether it is a simple marker or a pathogenetic factor, LVH encompasses all factors possibly affecting renal and general outcome in CKD patients.

Su273 ALBUMINURIA IN SCHISTOSOMA MANSONI-INFECTED INDIVIDUALS

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Introduction and Aims: Immunological response has been correlated with increased endothelial oxidative stress that may lead to microalbuminuria. In its turn, microalbuminuria has been associated with later development of cardiovascular disease. We investigated whether patients with hepatosplenic schistosomiasis, infected with Schistosoma mansoni (SM) parasite, show microalbuminuria.
Methods: Albuminuria, obtained from spot urine indexed to urinary creatinine, was investigated in 54 subjects showing hepatoesplenic fibrosis due to SM infection, the SM group and, in 30 control subjects, the C group. Inclusion criteria required nonhypertensive, nondiabetic and nonsplenectomyzed subjects. Evaluated individuals showed the clinical characteristics seen in Table 1. Urinary albumin and creatinine were measured by nephelometry (Behring) and standard colorimetric method, respectively. The protocol was approved by the local Ethical Committee. Values of albuminuria are expressed as mean ± SD.
Results: From the 54 evaluated patients, 3.7% (1 man and 1 woman) presented microalbuminuria (values between 30 and 299 mg/g creatinine). Similarly, among the control individuals, 6.6% (1 man and 1 woman) showed microalbuminuria. On the other hand, 29.6% of SM group (9 men and 7 women) had undetectable albuminuria, while 66.6% of SM group (22 men and 14 women) showed normal levels of albuminuria (Table 1).

Some Clinical Characteristics of the Evaluated Individuals

	SM Men	C Men	SM Women	C Women
Age, y	48.8±10.9 (n=32)	38.8±9.0 (n=7)	42.0±13.6 (n=22)	41.6±13.3 (n=23)
Mean arterial pressure, mmHg	98.8±8.4 (n=32)	97.2±8.9 (n=7)	90.1±12.2 (n=22)	89.2±9.1 (n=23)
Albuminuria, mg/g creatinine	5.1±3.4 (n=22)	5.7±3.4 (n=6)	5.1±3.4 (n=14)	18.9±6.2 (n=22)

Conclusions: Contrary to the assumption that immunological response could lead to microalbuminuria, our data shows that the most advanced stage of Schistosoma mansoni infection was not correlated with microalbuminuria.

Su274 NEPHROCALCINOSIS: AN UMBRELLA TERM, A NOT-SO-RARE DISEASE

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Introduction and Aims: Nephrocalcinosis is a rare, heterogeneous disease; the term describes the deposition of Calcium salts in the renal parenchyma, distinct from nephrolithiasis. The diagnosis is mainly based upon radiological grounds; CT scans and ultrasounds in experienced hands are the methods of choice. As the disease is often asymptomatic in particular in its early phases, and diagnosis requires experienced radiologists, it prevalence may be underestimated. Aim of the study is to describe the main clinical, radiological and laboratory features of patients with nephrocalcinosis, referred in December 2007-January 2010 in a new Nephrology Outpatient facility.
Methods: Review of the clinical charts with reference to reasons for referral, clinical situation at referral, main diagnostic clues; in our setting, prospective collection of the data of patients with diseases labelled as "rare" according to the National definitions is performed. The patients are categorised at diagnosis.
Results: In the period December 2007- January 2010, 1670 patients were referred to the new Unit. The diagnosis of nephrocalcinosis was done in 14; in 2 further cases a familiar form of nephrocalcinosis was diagnosed at family screening (16 in total).
The prevalence of the diagnosis of nephrocalcinosis in our unselected series of cases was thus of about 1%. The diagnosis of nephrocalcinosis was posed in all but one cases in our Unit. In one case, a child who had been diagnosed as affected by renal tubular acidosis in infancy, a diagnosis of nephrocalcinosis was further done for her mother and sister, after family screening in the study setting. All but one patient, affected by medullary sponge kidney, were females, with a median age of 39 years (9-68). At referral, 3 patients had renal function impairment (GFR<60 mL/min), one had proteinuria over 0.3 g/day (median: creatinine 0.8 mg/dL; GFR 73 mL/min). As for the main subcategories, 3 patients were affected by medullary sponge kidney (nephrocalcinosis was diagnosed by imaging data in our setting; two had a previous diagnosis of polycystic kidneys, one of renal cysts); 5 were affected by Sjogren syndrome (2 referred by the local rheumatologists for urinary anomalies, one for stone disease, and two from other settings); 2 had a history of diuretic abuse (one was referred with acute renal failure and severe electrolyte imbalance); 6 had complex tubular defects, in 4 in the context of familial or congenital syndromes. Interestingly, in two cases the diagnosis was posed during pregnancy (refractory oedema and renal function reduction). In all patients the diagnosis was leaded by imaging techniques; while CT scan was chosen as confirmatory technique, in 12 cases however, ultrasounds in experienced hands were diagnostic.
Conclusions: Nephrocalcinosis is a rare, but not exceptional diagnosis, being found in about 1% of the cases referred to a new-born Nephrology Unit. The strict cooperation with the Radiologists and Rheumatologists, aware of this differential diagnosis, is fundamental for diagnosis.

Su275 PREVALENCE OF VITAMIN D DEFICIENCY AMONGST ETHNIC SUB-POPULATIONS IN SOUTH LONDON

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Introduction and Aims: Vitamin D deficiency in common in the general population and may be a potential risk factor for cardiovascular disease. The prevalence of 25 hydroxyvitamin D [25 (OH)D] deficiency in south London is unknown. This cross-sectional study aimed to investigate the prevalence of Vitamin D deficiency in an ethnically diverse south London population.
Methods: Data were obtained on serum 25 (OH)D levels, parathyroid hormone levels, alkaline phosphatase and corrected calcium from all patients age 18 and over from December 2008 to November 2009 tested at a single centre in south west London. Serum 25 (OH)D levels were measured using direct electrochemiluminescence immunoassay (Elecsys 2010 analyzers, Roche Diagnostics). The ethnicity of the corresponding patients was ascertained by searching the hospital electronic database. Patients were classified into three main ethnic groups including Caucasian South Asian and Afrocarribean.
Results: A total of 7247 patients were analysed. The mean vitamin D levels were 45.9±18 nmol/L. 43% were 25(OH)D deficient [25 (OH)D< 37.5 nmo/L], 46% were vitamin D insufficient [25 (OH)D >37.5 and <75 nmol/L] and 11% were vitamin D sufficient [25 (OH)D≥75 nmo/L]. There was no difference in 25 (OH)D levels between males and females

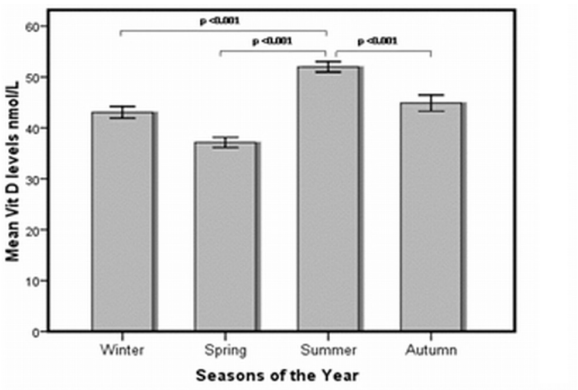

Figure 1: Vitamin D levels show a seasonal trend with highest levels in summer months (June-August)

(45.1±24.1vs. 44.9±25.8 nmol/L,p=0.756). Mean vitamin D levels was lowest in South Asians 32.7±18.9, followed by Afrocarribeans 39.6±20.3 and Caucasians, 43.8±25.4 nmol/L (all p<0.001). There was an anticipated seasonal trend with summer months showing the highest mean 25 (OH)D levels and spring showing the lowest mean 25(OH)D levels.
Vitamin D showed an inverse relationship with serum PTH and Alkaline phosphatase.

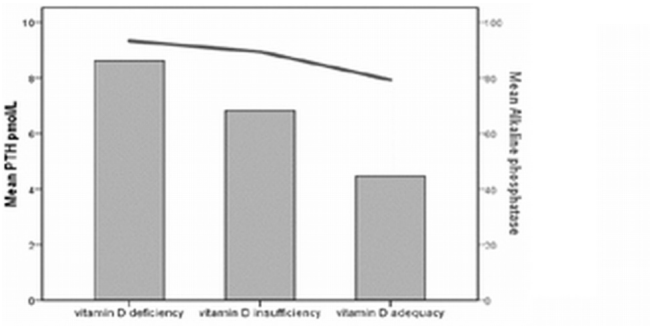

Figure 2: Vitamin D shows an inverse relationship with serum PTH and alkaline phosphatase

Conclusions: Our study revealed hypovitaminosis D is highly prevalent (89%) amongst the general population in south London with lowest levels observed in South Asians. Hypovitaminosis D may be associated with an increase in bone turnover as evidenced by an increase in serum alkaline phosphatase levels.

Su276 THE CHALLENGES OF MANAGING CARDIOVASCULAR RISK FACTORS IN YOUNGER PATIENTS WITH CHRONIC KIDNEY DISEASE STAGE III AND IV

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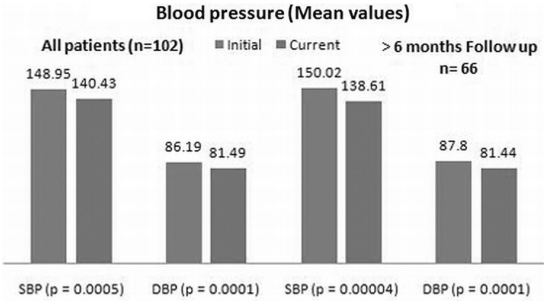
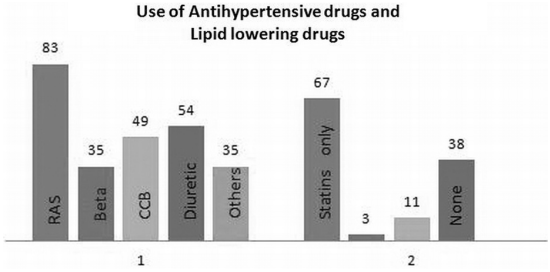
Introduction and Aims: Patients with Chronic Kidney disease (CKD) carry a higher risk of cardiovascular morbidity and mortality. Death from cardiovascular disease is more common than needing dialysis at all stages of CKD. Our aims are *1.* To assess the prevalence of cardiovascular risk in young adult population with CKD stage III and IV, and *2.* To assess management of these risk factors.
Methods: We collected demographic, clinical, co-morbid, drug and lab data on patients aged ≤ 65yrs with CKD stage III and IV attending our hospital Nephrology clinic over a 6 week period from the first week of November 2009. Results were compared with Renal Association standards. Student-t test was used to calculate significance.
Results: Data was collected on 118 patients of whom 34 belonged to CKD stage IIIa, 42 to stage IIIb and IV. Fifty percent of the study population (n=59) were aged 56 to 65 years, 46.6% (n=55) were females, 40% (n=47) were diabetics, and 31 patients (26.5%) of patients had evidence of cardiovascular disease.
82% of patients (n=89/109) had a blood pressure above the renal association target of <130/80. Comparing with presenting BP there was a positive trend of improved BP control with duration of follow up.





Data on cholesterol was available on 83 patients of whom 59 (71%) had a cholesterol ≥ 4.0 with a mean of 5.37. Mean cholesterol of the whole population was 4.40. 68% (n=81) of patients were on anti-lipaemic drugs with 11 patients on 2 agents.
Proteinuria quantification was known in 78 patients of whom 55 (70.5%) were significant. 23 (29.49%) of these had either Proteinuria > 1gram per day. Anti-platelet agent was prescribed for 44 (38.6%)patients.
BMI data was available on 80 patients of which 77.5% (n=62) were obese. On comparing with presenting BMI there was a statistically significant increase in mean BMI from 30.32 to 30.97 (p<0.005). Only 3 patients stopped smoking during nephrology follow up and 19 (20%) were still smoking.
Conclusions: There were a significant proportion of young patients with CKD whose cardiovascular risk factors have not been successfully addressed. We found that documentation of current risk factors and steps to address them were inadequate. However, there was a significant improvement in BP with nephrological follow up which should translate into a more favourable cardiovascular outcome.

Su277 WAIST CIRCUMFERENCE MODIFIES THE RELATIONSHIP OF ADIPOSE TISSUE CYTOKINES LEPTIN AND ADIPONECTIN WITH ALL-CAUSE AND CARDIOVASCULAR MORTALITY IN END STAGE RENAL DISEASE (ESRD)

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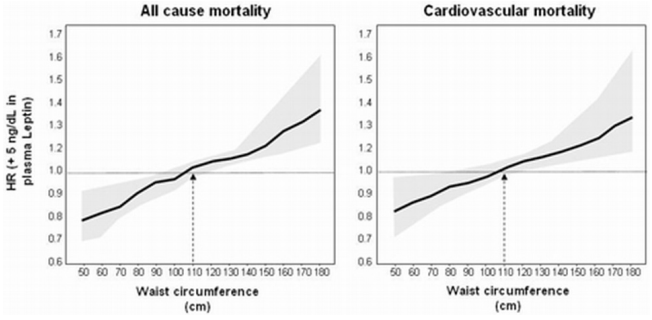
Introduction and Aims: The relationships between adipose tissue cytokines Leptin and Adiponectin with clinical outcomes have been scarcely studied in end stage renal disease (ESRD) patients and remain highly controversial.
Methods: Since central obesity is an important modifier of the effect of various risk factors for clinical outcomes, we tested the hypothesis that waist circumference (WC) modifies the link of these cytokines with overall and cardiovascular (CV) death in a cohort of 537 hemodialysis patients.
Results: Leptin and Adiponectin were inversely related to each other and robustly associated with waist circumference (all P<0.001). During the follow-up (average: 29 months, range 1-47 months) 182 patients died, 113 of them of CV causes. In analyses adjusting for all potential confounders there were strong interactions between plasma leptin and WC in relationship to both, all-cause (P<0.001) and CV death (P=0.002). Accordingly, a fixed excess in plasma leptin signalled a gradually higher risk for all-cause and CV mortality in patients with a large WC but an opposite effect in those with relatively small WC (see figure).



An interaction between adiponectin and WC for all-cause (P=0.01) and CV mortality (P=0.01) emerged only in models excluding the leptin-WC

interaction, suggesting that that these adipokines share a common pathway conducive to adverse clinical events in ESRD patients.

Conclusions: The predictive values of Leptin and Adiponectin for all-cause and CV death in ESRD patients appear critically dependent on WC. Overall these findings generate the hypothesis that intervention studies in ESRD patients with abdominal obesity and deranged adipokines levels may translate into better clinical outcomes.

*Alati Giovanni, Ascoli Giuseppe, Barreca Eleonora, Bovino Margherita, Bruzzese Vincenzo, Chiarella Salvatore, D'Agostino Francesco, De Gaudio Maria, D'Anello Emanuela, Fabiano F. Salvatore, Fornaciari Cesare, Foscaldi Aldo, Franco Corrado, Galati Domenico, Grandinetti Francesco, Gullo Maurizio, Lucà Nicodemo, Maimone Igina, Mannino M. Letizia, Martire Vincenzo, Mazza Giuseppe, Mellace Agazio, Minisci A. Francesco, Montera Dario, Pantaleone Procopio, Plutino Domenico, Pugliese Antonio, Reina Anna, Roberti Rita, Rizzuto Giuseppe, Rocchetti Vincenzo, Santangelo M. Grazia, Scida Giuseppe, Sellaro Arcangelo, Senatore Massimo, Vardè Carmela.

Su278 A PATIENT'S PERSPECTIVE ON HOME HEMODIALYSIS: RESULTS OF SEQUENTIAL PROVINCIAL SURVEYS

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Introduction and Aims: In 2004, the British Columbia Renal Agency launched a province-wide independent HD program, which now includes both home hemodialysis (HHD) and independent nocturnal in-centre hemodialysis. The program has experienced more than 500% growth since 2004 with 142 patients dialyzing independently as of May 2009. In total, we have successfully trained 263 patients. This volume speaks to the effectiveness of an integrated program. To ensure growth and development, an ongoing assessment of patient attitudes and concerns toward independent therapies are assessed via surveys every 2 years, gauging the interest in the program. The current study describes the results of the survey used in 2006 and 2008.

Methods: All registered individuals with CKD within the Province of BC were mailed a survey with a return envelope. Reponses were centrally collected at the Renal Agency. There were 3 surveys depending on which of 3 groups the patient was registered with (CKD, PD, HD). Response was voluntary. Surveys were made available in English, Chinese and Punjabi.

Results: Table 1: Expression of interest in learning more about independent therapies.

Expression of interest in learning about independent therapies

Year	Surveys (n)	Response (%)	CKD (%)	PD (%)	HD (%)	Overall (%)
2006	2788	46	27	10	10	12
2008	3009	44	37	29	19	25

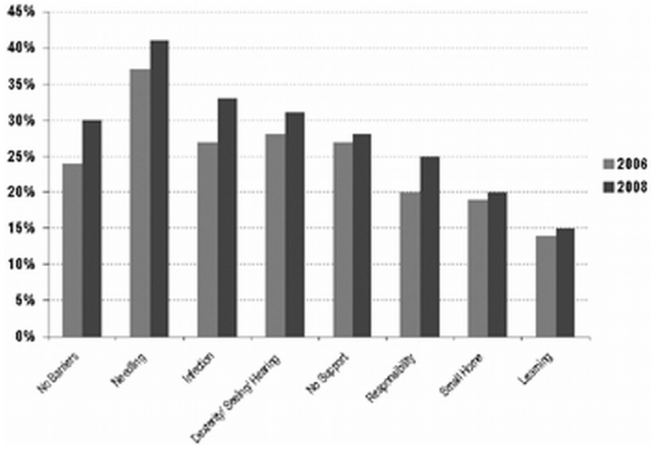

Figure 1. Identified barriers to independent hemodialysis.

Conclusions: The trends of our 2 surveys show that interest in Independent HD is growing in both our CKD and established dialysis patients, with the CKD population expresing the greatest amount of interest. The 2008 data shows that patient characteristics associated with interest in HHD included younger age (62±14 yrs vs. 67±14 yrs, p<0.001) and a history of diabetes (33% vs. 20% p=0.03), while gender no longer seems to play a significant role. More patients do not perceive any barrierse to independent dialysis (30% in 2008 vs. 24% in 2006). Of identified patient perceived barriers, fear of self-needling and infection remain the largest concern. Given the cost-savings of independent therapies, further resource allocation is justified to address the perceived barriers to maximize program growth. It should be noted that despite being administered in both Punjabi and Chinese, the response rate from Punjabi and Chinese speaking patients was very low (<1% and 25% respectively) This may represent a specific area to address in our recruitment strategy.

Disclosure: Dr. Michael Copland sits on an Advisory Board with Baxter Global Healthcare.

Su279 DIRECT MEDICAL COST OF PERITONEAL DIALYSIS: RESULTS FROM A MEXICAN COHORT

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Introduction and Aims: Costs for treating patients with end-stage renal disease (ESRD) have grown remarkably. Most of the costs are estimated from the perspective of payers; however, direct ESRD costs from the perspective of physicians are not accurately known. *Aim:* To estimate direct medical costs for peritoneal dialysis (PD) and compare them according to the type of dialysis [continuous ambulatory peritoneal dialysis (CAPD) *vs* automated peritoneal dialysis (APD)].

Methods: A cohort of consecutive patients initiating PD (Jan-Dec 2008) was retrospectively analyzed. Only direct medical costs were considered [from the Mexican Institute of Social Security (IMSS) health system perspective], including: out-patient clinic/emergency care, dialysis procedures, medications, laboratory tests, hospitalization, and surgery. All costs were converted into US dollars (1 USD=12.84 Mexican pesos). The Inverse Probability Weighting method (IPW) was used for total mean cost (TMC) estimation and resource utilization.

Results: Forty-one patients were evaluated, age was 52±18.0 years and 39% were women. The PD-related costs are summarized in the following table:

Costs per patient year (USD)

	All N41	CAPD N22	APD, N19
Total mean costs	14,898 (13,785-16,010)	14,088 (12,606-15,570)	15,834 (14,107-17,561)
Out-patient/Emergency care	422 (357-490)	397 (324-469)	451 (323-579)
Dialysis procedures	6,279 (6,053-6,566)	5,639 (5,580-5,697)*	7,027 (6,944-7,109)
Medications	1,733 (1275-2195)	1,617 (1,068-2,167)	1,867 (1,052-2,681)
Laboratory tests	90 (68-112)	89 (67-111)	91 (49-132)
Hospitalization	5,231 (4,397-6,066)	5,198 (4084-6313)	5,270 (3,892-6,664)
Surgery	1,142 (952-1,332)	1,153 (869-1437)	1,129 (853-1406)

*p<0.05 vs APD

The largest part of costs was caused by dialysis procedures (CAPD 40%, APD 44%) followed by hospitalization (CAPD 37%, APD 33%), medication and surgery. Dialysis procedures were significantly higher in APD than CAPD.

Conclusions: There was no significant difference between APD and CAPD costs; however, dialysis procedures had higher cost in APD than in CAPD. The largest cost in both modalities was caused by dialysis procedures followed by hospitalization.

Su280 THE COST-EFFECTIVENESS ESTIMATION FOR ADDITIVE LANTHANUM CARBONATE THERAPY OF JAPANESE DIALYSIS PATIENTS WITH HYPERPHOSPHATEMIA

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Introduction and Aims: Lanthanum carbonate (LC) is a new, non-calcium-based phosphate binder and possibly useful for dialysis patients who can't control hyperphosphatemia with conventional treatment. On the other hand, LC is expensive medication and our previous modeling study suggested that LC might not be cost-effective as the first line therapy for hyperphosphatemia in dialysis patients. However, the cost-effectiveness of LC as the second line therapy has not been elucidated due to lack of published clinical data on LC as the second line therapy in Japan. Therefore we investigated clinical effect of LC as the second line therapy and assessed its cost-effectiveness from the Japanese perspective.
Methods: We enrolled 136 patients and investigated change in phosphate, calcium, parathyroid hormone, and the dose of calcium containing phosphate binder, sevelamer, vitamin D analogues and cinacalcet before and after lanthanum carbonate treatment as the second-line in prospective 16-weeks study. Based on the study data, a state transition model was developed to evaluate the health benefits and costs associated with of LC as the second-line therapy. The potential impact of reducing hyperphosphatemic events by LC was analyzed by using the database of the Japanese Society for Dialysis Therapy registry and the other published sources. Uncertainty was explored through sensitivity analysis.
Results: After 16 weeks of treatment, serum phosphate levels decreased from 7.0±1.1 mg/dL to 5.7±1.3 mg/dL. The dose of calcium containing phosphate binder and sevelamer decreased. Calcium, parathyroid hormone, and the dose of vitamin D analogues and cinacalcet were not changed. Compared to conventional treatment, the LC treatment as the second-line incurs average additional lifetime costs of €18,294 (JPY2,378,220) per person, while it confers an additional 0.543 quality adjusted life years (QALYs), which resulted in an incremental cost-effectiveness ratio (ICER) of €33,695 (JPY4,380,350) per QALY gained. Applying a cost-effectiveness threshold of €33,600 (£30,000) per QALY, probabilistic sensitivity analysis showed that LC as the second line therapy has a probability of 0.515 of being cost-effective compared with conventional treatment.
Conclusions: Our modeling study suggested that the use of LC as the second-line therapy would be cost-effective in Japan. The validity of modeling, however, must be investigated by the future clinical studies, regarding the long-term evidence on morbidity and mortality of the LC treatment.
Disclosure: Isao Kamae: Grants/Research Support: Bayer, Yakuhin, Lyd. Scientific Advisor or Membership: Bayer Schering Pharma AG. Masafumi Fukagawa: Grants/Research Support: Kyowa Hakko Kirin; Chugai. Scientific Advisor: Bayer, Japan

Su281 DIAGNOSTIC RELEVANCE OF PYURIA IN DIALYSIS PATIENTS WITH URINARY VOLUME FOR DETECTING URINARY TRACT INFECTION

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Introduction and Aims: Urinary tract infections (UTIs) are common in dialysis patients, are associated with an increased rate of complications. Diagnostic accuracy of pyuria in the dialysis patients whose urinary volume were decreased, to identify UTIs, has not been adequately defined. So diagnostic performance of pyuria to dialysis patients by urinary volume per day is the object of this study.
Methods: The study included 78 (34F, 44M, 41 diabetes mellitus [DM], 37 non-DM cases) stable hemodyalsis, peritoneal dialysis patients with diuresis ranged from <100, 100∼400, >400 ml per 24 h. We assembled a historical cohort of patients who did not have symptoms of the UTIs with urinalysis and urine culture data. We valuated the diagnostic performance and plotted receiver operating characteristic curves for different cutoff values of pyuria (5∼10, 10∼30, >30, white blood cells per high-power field (WBC/HPF).
Results: Rate of bacteriuria was 44%. E. coli is the most common bacterial strain in infected patients. The patients with DM had more increased incidence of bacteriuria than those with non-DM. Urinary WBC significantly increased with decrease in the urinary volume.
At each urinary volume per days, presence of pyuria (5∼10WBC/HPF, 10∼30WBC/H PF, >30 WBC/HPF) has 83%, 79%, 74% sensitivity and 46%, 49%, 55% specificity. But even the presence of >30 WBC/HPF has low specificity (55%) and positive predictive values (67%). So pyuria of dialysis patients with decreased urinary volume is poor diagnostic performance in the identification of a positive urine culture at any urine WBC indices.
Conclusions: Because of low specificity and positive predictive value in dialysis patients, Urine culture should be needed to determine further treatment.

Su282 AMINOTRANSFERASE ACTIVITY AS A POOR PREDICTOR OF LIVER DISEASE PROGRESSION IN HEMODIALYSIS PATIENTS WITH CHRONIC HEPATITIS C

Pavlina Dzekova-Vidimliski, Galina Severova-Andreevska, Lada Trajceska, Vladimir Pusevski, Gulsen Selim, Saso Gelev, Vili Amitov, Sonja Dzikova, Aleksandar Sikole. *University Clinic of Nephrology, Skopje, Macedonia, The Former Yugoslav Republic of*

Introduction and Aims: Lower aminotransferase activity in dialysis patients makes the assessment of the natural history of hepatitis C virus (HCV) infection difficult. The aim of the study was to determine the risk factors associated with the aminotransferase activity in hemodialysis patients with chronic hepatitis C.
Methods: Eighty hemodialysis patients (51 men and 29 women) with chronic hepatitis C were enrolled in the study. The patients were followed from January 2004 to January 2008. Serum levels of aminotransferases were measured monthly with standard automated analyzers. The presence of HCV antibodies was tested by ELISA- third generation assay, HCV RNA was determined by reverse transcriptase polymerase chain reaction (RT-PCR), and HCV genotype was analyzed by RT-PCR followed by hybridization of amplified products. Ultrasound examination with measurement of morphological parameters (liver size, morphology, surface, echogenicity, and spleen volume) and hemodynamic parameters (portal vein diameter and portal vein mean flow velocity) was done in all patients.
Results: According to the serum levels of alanine aminotransferase (ALT), during the follow-up, patients were divided into two groups. The first group consisted of 34 chronically HCV infected patients with persistently normal levels of ALT (28.8±7.3 U/L) The second group included 46 chronically HCV infected patients with elevated levels of ALT (62.8±27.0 U/L). Genotype 1 was the dominant genotype in both groups (78 patients, 97.5%). Patients with elevated ALT levels were characterized with significantly shorter dialysis duration (p=0.048) and significantly shorter duration of HCV infection (p=0.005) compared to the patients with persistently normal levels of ALT. The values of measured ultrasound parameters were not significantly different between the two groups. The univariate analysis identified higher serum level of direct bilirubin (p=0.044), shorter duration of dialysis (p=0.048), and shorter duration of HCV infection (p=0.005) as potentional predictors of elevated serum ALT levels in dialysis patients. After stepwise logistic regression, none of the potential predictors was independently associated with elevated ALT levels.
Conclusions: Serum aminotransferase levels are poor predictors of liver disease progression in hemodialysis patients with chronic hepatitis C. Further studies should be conducted in order to identify non invasive indicators of the disease progression in uremic patients with hepatitis C.

Su283 ASSOCIATION OF TIME-VARYING ALBUMIN AND PHOSPHATE LEVELS AND ITS INTERACTION WITH ALL-CAUSE MORTALITY IN INCIDENT DIALYSIS PATIENTS – THE INVOR STUDY

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Introduction and Aims: Hypoalbuminemia and hyperphosphatemia have been shown to be strong predictors of mortality in chronic dialysis patients. There are only limited data evaluating concurrently the relationship between these changing parameters and survival outcomes over a long-term observation period in incident dialysis patients.
Methods: We followed 235 incident dialysis patients (mean age±SD: 61.7±14 years; m/f: 146/89) in a prospective single-centre cohort study (INVOR-Study: Study of *IN* cident Dialysis Patients in *VOR* arlberg) applying a time-dependent Cox regression analysis using all measured laboratory values for up to more than seven years. In the Cox-model adjustments were made for gender, age, dialysis modality, hemoglobin, CRP, calcium and PTH. Additionally the interaction effects of albumin and phosphate values were determined including effects of varying albumin values at fixed phosphate levels and vice versa.
Results: During a median follow-up of 35.1 months 82 patients died (35%). Albumin was found to be inversely associated with the risk of all-cause mortality (HR [95%CI] 0.24 [0.15, 0.37]; p<0.00001), whereas increasing phosphate levels were associated with an increased mortality risk (HR [95%CI] 1.58 [0.99, 2.54]; p=0.057). In an additional model including an interaction term, albumin and phosphate showed a significant interaction effect (p=0.047). In this model high albumin levels were still significantly associated with a reduction in mortality risk (HR [95%CI] 0.08 [0.02, 0.26]; p=0.000025), whereas the phosphate effect alone was attenuated (p=0.126), emphasizing its interacting effect. For fixed albumin values at the median and higher (≥4 g/dL) increasing phosphate levels were significantly associated with an increased mortality risk, while this association was not found at lower albumin. Analyzing varying albumin values at simultaneously fixed phosphate quantiles, we found a linear inverse association between albumin and mortality with attenuation of this effect at very high phosphate levels.
Conclusions: Time-varying albumin and phosphate values interact in their association with all-cause mortality in incident dialysis patients. The lowest risk is found with simultaneously low phosphate and high albumin values, whereas risk is increased with either concurrent low phosphate and low albumin values or high phosphate and high albumin values. Epidemiological studies and therapeutic guidelines aiming for target values should consider this significant interplay.

Su284 SHORT-TERM IMPACT OF A NUTRITION EDUCATION PROGRAM USING FEEDING BEHAVIOR TRANSTHEORETICAL MODEL ON HYPERPHOSPHATEMIA OF HEMODIALYSIS PATIENTS

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Introduction and Aims: Evaluate the impact in a short period of time of a nutrition educational program on hyperphosphatemia offered to hemodialysis patients, using the transtheoretical model of feeding behavior.
Methods: The study included 108 patients undergoing hemodialysis in a satellite clinic in <u1:City u2:st="on"><u1:place u2:st="on">São Paulo</u1:place></u1:City> city, showing changes in levels of phosphorus, being 39.8% women and 60.2% men, mean age 53±15.9 years, mean BMI 24.25±4.2 kg/m^2, mean time of dialysis 7.4 (3 – 74) months. In order to perform nutritional intervention, we used the transtheoretical model, also called the stage model of behavior change. According to this model, changes in behavior related to health occur have five stages: pre-contemplation, contemplation, decision, action and maintenance. Each stage represents a temporal dimension of behavior change, i.e., it shows when change occurs and assess the patient's level of motivation. In order to classify individuals, we used the Ling and Horwath survey (2000) before and after intervention. After we have identified the stages of change related to food, we have presented a lecture and a nutrition dynamic concerning phosphorus in June 2009. During the lecture, we used a flip chart with information and illustrations about chronic renal disease, hyperphosphatemia and phosphate binder. During nutrition dynamic, we used pictures of food and asked the participants if images were appropriate or not and why. We also asked how they should eat the phosphate binder. Serum concentrations of phosphorus were assessed before and after intervention. At the end of the lecture, we delivered an illustrated brochure on the topics covered.
Results: There was a significant decrease in serum phosphorus of 6.4±1.3 to 5.5±1.5 mg/dL, P<0.001. However, we observed a statistically significant decrease in patients classified action (7.5±1.8 to 5.9±1.7 P<0.001), maintenance (6.6±0.8 to 5.6±1.2 P<0.001) and relapse (6.7±0.9 to 5.6±1.7 P 0.01) when these topics were correlated to the stages of feeding behavior.
Conclusions: Investigating feeding behavior aims to increase the effectiveness of nutritional interventions, not only to provide information, but to achieve a change in eating behavior. This is a slow process, so it must be continuous. A medium and long term approach could be more effective in achieving a higher adherence to treatment in all phases of feeding behavior.

Su285 CAN CYSTATIN C BE USED TO ESTIMATE RESIDUAL RENAL FUNCTION IN PATIENTS ON MAINTENANCE HAEMODIALYSIS?

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Introduction and Aims: In dialysis patients residual renal function (RRF) has been found to have significant influence on mortality and morbidity. However, RRF determination is sometimes difficult to perform due to the need for a 24 h urine collection to measure creatinine and urea clearance. Thus, a cystatin C based equation was proposed to estimate RRF. An external validation of this formula in dialysis patients is still missing.
Methods: 169 patients on chronic haemodialysis (62 female) were included in the study. Cystatin C serum concentrations prior to the dialysis session were determined by fully automated latex-enhanced immunonephelometric technique on a Behring Nephelometer II (Dade Behring, normal values <0.96 mg/l). RRF was determined by the mean creatinine and urea clearance. eGFR was calculated using the following formula: eGFR=-0.77+21(1/Cystatin C). Mean difference (bias) and correlation coefficient were calculated and Bland and Altman plots were used for analysis.
Results: Mean RRF was 2.4 (95%CI: 1.9-2.9) ml/min/1.73m^2 Serum Cystatin C concentration was 5.7 (95%CI: 5.5-5.9) mg/l at average. Mean eGFR was 3.2 (95%CI: 2.9-3.4) ml/min/1.73m^2. Thus, RRF was overestimated by 0.81 ml/min at average (p<0.001). eGFR correlated well with RRF (r=0.72; p<0.0001). However, Bland and Altman plots showed an overestimation in patients with low or even zero RRF and an underestimation in patients with RRF > 5 ml/min resulting in limits of agreement of 4 ml/min/1.73 m^2 above and 5.6 ml/min/1.73 m^2 below the measured RRF.
Conclusions: Despite some drawbacks estimation of RRF in dialysis patients using a cystatin C based approach seems to be reliable in daily clinical work. This tool may be helpful to guide prescription of medication and dialysis dose.

Su286 HEALTH RELATED QUALITY OF LIFE AND ADHERENCE TO PHOSPHATE-BINDING MEDICATION IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Health related quality of life (HRQOL) is affected by chronic kidney disease (CKD). Pure control of phosphorus levels has

been associated with poorer outcomes in these patients. The aim of the study was to assess adherence to phosphate-binding agents and its correlation with HRQOL in prevalent hemodialysis (HD) patients.
Methods: HRQOL was estimated using a validated translation of the Kidney disease quality of life – short form (KDQOL-SF) questionnaire, which has been used previously in ESRD patients, while adherence to treatment was assessed using the Medication Adherence Report Scale (MARS) which consists of 10 separate nonadherence-related behaviours. Behaviour is scored on a 4-point scale (1-4: always, often, sometimes, or never true, respectively), high scores (>18) indicating good adherence. Based on their adherence, patients were divided in two groups: group 1 consisted of high adherers (MARS>18) and low adherers (MARS ≤18). T-test and Mann-Whitney U-test were used to assess possible differences between groups as appropriate. Correlation of adherence to treatment with basic laboratory parameters was estimated with the help of Spearman correlation coefficient (cc).
Results: A total of 73 patients out of 85 completed the questionnaires, (response rate 85.9%), their mean age was 62.2 years. Group1 consisted of 30 patients (41%), while group2 of 43 patients (59%). Even though differences were not proven statistically significant, group 1 scored higher than group 2 in all three major KDQOL-SF categories: in physical component summary (50.6 vs 42.0), as well as in mental component summary (55.8 vs 50.5) and in kidney disease component summary (55.4 vs 52.5). Over 25% forgot to take their medication sometimes, often or always, while 24% forgot to take phosphate-binders at mealtimes. Intentional nonadherence was observed, with 17% altering their dose, 16% deciding to miss a dose and 15% taking less medication than instructed. Adherence to treatment was found negatively correlated with age (correlation coefficient = -379, p<0.001) and positively with urea (cc = 0.270, p=0.002), creatinine (cc = 0.396, p=0.001), phosphorus (cc = 0.308, p=0.008) and serum albumin levels (cc =0.341, p=0.003). Nevertheless, there were found no significant differences between groups regarding these laboratory parameters neither between high and low responders. Age was the only significant difference between groups, with high adherers being significantly younger (58.9 vs 67.0, p=0.008).
Conclusions: About a 40% of patients reported low adherence to phosphate-binding medication. Age was found to be the major determinant of adherence to treatment. Health related quality of life was not proven significantly related to adherence to phosphate binders treatment. Nevertheless, even though didn't reach statistical significance, patients showing higher adherence to treatment scored higher in all major components of health related quality of life.

Su287 A THREE YEARS EVOLUTION OF QUALITY OF LIFE (QoL) AND CLINICAL PROFILES OF HAEMODIALYSIS OUTPATIENTS IN FRANCE

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Introduction and Aims: Aging and comorbidities in haemodialysis patients are a tremendous challenge for health-care delivery systems in western countries. In order to improve the organisation of care and care processes in haemodialysis facilities, we compared clinical and quality of life characteristics of chronic haemodialysis patients populations within a three year period in 5 chronic haemodialysis outpatient facilities in a 2.5 million inhabitants area in France.
Methods: Data were collected from the 300 chronic haemodialysis patients followed-up within the 5 participating outpatient facilities, 150 at baseline assessment and 150 at 3 years. Assessment included social, demographic and medical data, as well as QoL (SF36), cognitive impairment (MMSE) and depression (BDI). The questionnaires were administered in an interview format by a trained psychologist.
Results: Quality of life was found to be impaired in the baseline population, but all components were found to be much more lowered in the population studied 3 years later, especially BP (pain increased, p=0.01), General Health (GH, p<0.01), Social Functioning (SF, p<0.01) and Mental Component Scale (MCS, p=0.02). Social and demographic characterisitics and particularly age of HD patients were found to be similar as well as depressive symptoms and level of cognitive impairment. But we observed a significant increase in comorbidities, especially diabetes (from 34.7 to 46.0%, p = 0.05) and peripheral vascular diseases (from 22.0 to 32.6%, p=0.04). The various QoL domains were negatively correlated with depression and cognitive disorders at baseline and at 3 years. While the physical components (PF: Physical Functionning, GH, PCS: Physical Component Scale) were correlated with several comorbidities at baseline, only GH was correlated with diabetes at 3 years. When considering separately diabetic and non diabetic patients, QoL at baseline was much impaired in diabetic patients in comparison with non diabetic patients (PF p<0.01, GH p=0.03, PCS p=0.03, SF p=0.05), but at 3 years QoL was similar in both populations. Indeed non diabetic patients decreased significantly various QoL domains during the studied period (BP p=0.003, VT p=0.01, SF p<0.01).
Conclusions: QoL was impaired in the baseline haemodialysis outpatients population, but the impairment was worse in the population studied 3 years later within the same facilities. One reason would be an increase in comorbidities, especially diabetes and peripheral vascular diseases. A second explanation was an alteration of QoL in non diabetic patients with an increase of pain and a decrease in vitality. Clinical and QoL profiles of patients are changing over time. Consequently, care processes and objectives have to be adapted in order to better take into account these clinical profiles, but also patient's pain and depressive symptoms.

Su288 THE SURVIVAL AND COMBINATION OF VITAMIN D AND PHOSPHORUS BINDER IN DIALYSIS

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Introduction and Aims: Treatment with either vitamin D or phosphorus binder alone improves the survival of dialysis patients. This multicenter observational study investigated whether the combination of these drugs was related with the prognosis in Japanese dialysis patients.
Methods: A total of 466 patients were registered and followed up to five years. Group survival was compared using Cox-proportional hazard analysis, according to their use of vitamin D and phosphorous binder.
Results: During the follow-up period, 132 deaths (63 cardiovascular and 69 non-cardiovascular deaths) occurred. Compared with the patients with concomitant use of vitamin D and phosphorus binder (n=269), all-cause mortality was higher in other groups (using neither drug, phosphorus binder alone, and vitamin D alone, n=197), (HR 2.09, 95%CI 1.26-3.48, *P*=0.005) after adjustment with possible confounders. The advantage in patients with concomitant use of both drugs was significant both in cardiovascular and non-cardiovascular mortalities. Subgroup analysis showed that the beneficial effect of concomitant use of these drugs was lost in patients with high serum calcium (>10 mg/dL), or phosphate (>6.0 mg/dL) levels.
Conclusions: The combination of vitamin D and phosphorus binder was significantly related with survival in dialysis patients with well controlled serum calcium and phosphate levels.

Su289 DEVELOPMENT OF A NOVEL QUESTIONNAIRE EVALUATING THE DISABILITY OF UPPER EXTREMITIES IN PATIENTS UNDERGOING MAINTENANCE HEMODIALYSIS

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Introduction and Aims: Maintaining the ability to perform activities of daily living (ADL) is essential for hemodialysis (HD) patients to prevent deterioration of their quality of life (QOL), because the disability of ADL is known as one of the factors that deteriorate the QOL. However, the disability of upper extremities in ADL has not been fully evaluated in HD

patients, because no specific scale on it was provided yet. The aim of the present study was to develop a novel questionnaire evaluating the disability of upper extremities in HD patients (QDUE-HD).

Methods: Sixty-five outpatients who were undergoing maintenance HD three times a week at the HD center, 24 males and 41 females with a mean age of 66±10 years and a mean duration on HD of 9.2±7.5 years, were recruited for the present study. On selecting the items of the QDUE-HD, the original questionnaire consisting of 37 items was examined in which 19 items were referred from the Disability of the Arm, Shoulder and Hand questionnaire and 11 items from the Activities of Daily Living Test. The remaining 7 items indicated the questionnaire concerning the activities that HD patients felt unable or extremely difficult to do using their upper extremities. The questionnaire measured the degree of perceived difficulty, when HD patients performed some activities using their upper extremities. Each item was rated from 1 to 5 points indicating impossibility, severe difficulty, moderate difficulty, mild difficulty and ease, respectively. In the QDUE-HD, the items that more than 40% of HD patients rated at 1 or 5 points were excluded from the 37 items of original questionnaire based on their floor and ceiling effects, respectively. The principal factor analysis with promax rotation was performed to determine significant factors as a category and to group statistically significant items of the QDUE-HD into them.

Results: Of 37 items, 2 and 24 items showed the floor and ceiling effects, respectively. The factor analysis was performed in the remaining 11 items, and identified 2 factors as a category. The first and second factors of the QDUE-HD showed eigenvalues of 6.13 and 1.51, respectively. The first factor of the QDUE-HD consisted of 6 items indicating "gardening or doing yard work", "gardening or doing yard work", "gardening or doing yard work", "gardening or doing yard work", "gardening or doing yard work" and "gardening or doing yard work", and was named "gardening or doing yard work". The second factor of the QDUE-HD consisted of 5 items indicating "gardening or doing yard work", "gardening or doing yard work", "gardening or doing yard work", "gardening or doing yard work" and "gardening or doing yard work", and was named "gardening or doing yard work".

Conclusions: The QDUE-HD developed in the present study had 2 factors that were related to light works and holding activities. Moreover, the QDUE-HD seemed to be a useful clinical tool evaluating the disability of upper extremities in HD patients.

Su290 PREVALENCE OF NEPHROGENIC SYSTEMIC FIBROSIS (NSF) IN DIALYSIS PATIENTS: THE Pro-FINEST STUDY

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Introduction and Aims: NSF is a cutaneous and systemic disorder characterized by widespread tissue fibrosis, first described in 2000. It has been suggested that gadolinium-based contrast agents (GBCA) may be responsible for NSF in patient with renal insufficiency and particularly those on dialysis. The Pro-FINEST study (etude Prospective sur la FIbrose Nephrogenique SysTemique) is a national study endorsed by the French Drug Agency (afssaps), and the French Societies of Nephrology, Dermatology, and Radiology. It aims at investigating the prevalence of NSF after a Magnetic Resonance Imaging (MRI) examination, with or without GBCA, in chronic dialysis patients. Secondary objectives are to determine clinical and biological characteristics of NSF-affected patients and to identify risk factors.

Methods: The study is based on a patient form consisting of 3 sections: Section 1, filled by the nephrologist, to collect patient demographic and dialysis data (technique, duration, centre); Section 2, filled by the radiologist, to gather MRI examination data (injection: Yes/No, GBCA used, volume injected); Section 3, filled by the patient and sent back to our department 4 months after the MRI at the latest, to report dermatological events (Yes/No, characteristics, dermatologist consultation: Yes/No). Further investigations, including a dermatological evaluation, are planned in case a patient reports any dermatological manifestation. When a NSF diagnosis is confirmed in one patient, an ancillary study is to be performed with collection of additional biological and clinical patient data, with a random selection of 4 patients of the same gender, the same dialysis technique, in the same centre, without any dermatological events after having received one MRI with the same GBCA (if injected).

Results: The study started in January 2009. 247 dialysis centres agreed to participate. 195 patients have been included (1 year): mean age 63.7 years, 60.5% were men. 70 forms have been received: 62.9% of the patients received GBCA; most of them (90.9%) received Gadoterate. Three patients reported dermatological manifestations. Diagnoses were hemodialysis-related prurit in one case, one localized skin reaction to a trinitrine patch, and in the third case, neither the nephrologist nor the dermatologist to whom the patient had been referred to, retrieved any evidence of NSF. So far thus, no case of NSF has been observed.

Conclusions: After 1 year of investigation, no case of NSF has been reported in 195 dialysis patients among whom the majority received a GBCA (Gadoterate).

Su291 SEX-SPECIFIC ASSOCIATION OF TIME-VARYING HEMOGLOBIN VALUES WITH MORTALITY IN INCIDENT DIALYSIS PATIENTS – THE INVOR-STUDY

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Introduction and Aims: Previous studies in dialysis patients showed an association between hemoglobin levels and all-cause mortality, however, without addressing sex differences.

Methods: We followed 235 incident dialysis patients (61.7±14.0 years, m/f: 146/89) in a prospective single-center cohort study (INVOR-Study – Study of INcident Dialysis Patients in VORarlberg) applying a time-dependent Cox regression analysis using all measured laboratory values for up to more than seven years. In total, 12,242 hemoglobin measurements with a median of 47 (range 3-270) per patient were available to evaluate the impact of hemoglobin levels and their variability on all-cause mortality in a sex-stratified analysis. Nonlinear P-splines were used to allow flexible modeling of the association with mortality.

Results: We observed an inverse relationship between increasing hemoglobin values and decreasing risk of mortality. The linear component of the nonlinear spline was highly significant for both, men (p=0.00005) and women (p=0.0000000052). The nonlinear component was also significant but less pronounced than the linear component. The inverse relationship was clear to see up to hemoglobin values of 12-13 g/dL in women, which reached a plateau for higher values of hemoglobin. For men an inverse trend was observed but clearly attenuated when compared to women. After adjustment for additional parameters of inflammation and malnutrition as well as diabetes, the linear component was more significant in women (p=0.0018) than in men (p=0.023).

Conclusions: This study applied the first time a time-dependent Cox regression analysis over a long-term observation period of several years using all available measurements. Besides the methodological advantages our data indicate a sex-specific linear as well as nonlinear effect of hemoglobin levels on all-cause mortality, which was markedly more pronounced in women.

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Su292 EARLY MORBIDITY AND MORTALITY IN CHRONIC RENAL PATIENTS INCLUDED IN DIALYSIS

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Introduction and Aims: Late referral to nephrologist of chronic renal failure patients (pts) can lead to urgent unprepared dialysis start, with increased morbidity and mortality.
We assess morbidity and mortality in pts emergently included in dialysis versus pts in whom dialysis could be timely planned.
Methods: We studied 250 pts (males = 131, females = 119, mean age = 52,9±15,2 years with range between 17 – 84 years) admitted in our center for dialysis initiation. At hospital admission, all patients were routinely investigated for uremia complications. Survival rates were evaluated at 30 and 90 days after first dialysis.
Results: In 147 cases (group 1) was necessary rapid start of dialysis, in 140 pts (95,2%) using central venous catheter and in 7 pts (4,8%) using peritoneal catheter. Chronic dialysis (group 2) was started in 28 cases (27,2%) by native arteriovenous fistula, and in 75 pts (72,8%) by peritoneal catheter. Mean interval between the time of referral and start of dialysis was 8,1±15,4 months in group 1 and 15,4±25,3 months in group 2 (p=0,006).
30 days survival after dialysis initiation was significantly lower (95% CI) in group 1 (84,2%) compared to group 2 (99%); the difference was also significant (95% CI) about the 90 days survival: 76% in group 1 versus 98% in group 2.
Complications were significantly more frequent (95% CI) in group 1 compared to group 2: pericarditis was present in 31% versus 14,1% pts, cardiac failure in 54,2% versus 25,3% pts, cardiac rhythm disturbances in 33,8% versus 13,1% pts, pleural effusion in 39,4% versus 11,1% pts, pulmonary infections in 19,7% versus 7,1% pts, neurological disorders in 36,6% versus 7,1% pts, digestive problems in 48,6% versus 34,3% pts, hemorrhagic syndrome in 28,3% versus 7,4% pts, electrolytic disturbances (hyponatremia and/or hyperkalemia) in 52,1% versus 38,8% pts, acidosis in 58,5% versus 21,6% pts.
As to the mean values of the biological parameters, we found statistically significant differences between the group 1 and group 2 on the hemoglobin level (7,8±1,7 g/dl versus 8,9±1,8 g/dl, p<0,0001), serum creatinine (13,4±6 mg/dl versus 10,1±3,7 mg/dl, p<0,0001) and natremia (132,8±7,5 mEq/l versus 135,8±6,7 mEq/l, p=0,009). No significant difference were noticed between the group 1 and group 2 concerning the albumin concentration (3,2±0,9 g/dl versus 3,7±0,6 g/dl, p=0,1), calcium level (7,5±1,5 mg/dl versus 7,8±1,3 mg/dl, p=0,21), serum phosphate (6,8±2 mg/dl versus 6,5±1,7 mg/dl, p=0,45), kalemia (5,2±1,2 mEq/l versus 5±1,1 mEq/l, p=0,009) and bicarbonate level (14,4±6,9 mmol/l versus 17,5±4,8 mmol/l, p=0,1).
Conclusions: In our study we found a significantly higher morbidity and mortality in pts emergently included in dialysis treatment, compared to pts in whom replacement therapy was appropriate considered. Hemodialysis using central venous catheter was the preferred method as imperative solution for renal substitution. In pts referred earlier to nephrologist, increased the proportion of cases included in peritoneal dialysis.

Su293 EFFICACY AND SAFETY OF PEGYLATED INTERFERON MONOTHERAPY IN HEMODIALYSIS PATIENTS IN CROATIA

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Introduction and Aims: Hepatitis C virus (HCV) infection is a frequent complication among the long-term dialysis patients. The aim of the present study was to evaluate the efficacy and to report side effects of pegylated interferon alpha-2a (PEG-INF 2a) treatment in hemodialysis patients.
Methods: We retrospectively reviewed charts of 16 HCV-RNA positive hemodialysis patients. Patients were planned to receive 135 μg of PEG-INF 2a weekly for 48 weeks. Virological response was followed after 12 weeks of treatment (early virological response – EVR), after 24 weeks, at the end of treatment (ETVR), and at week 72 (sustained virological response-SVR).
Results: There were 11 male and 5 female patients aged 34-62 years (average 51.06) treated with dialysis for 6-28 years (average 19.25 years). Thirteen patients had HCV genotype 1b, 2 patients 3a and one patient had genotype 2a. Biopsy was performed in 3 patients. Eleven patients completed 48 weeks of treatment, with EVR and ETVR obtained in 9 and 10 patients, respectively. SVR was recorded in 9 patients (out of 14, while 2 patients have recently finished their treatment). Treatment was discontinued in 3 patients because of the side-effects and in two patients because of the inefficiency with significant side-effects (both genotype 1b), while one patient received 90 μg from the week 30 to the end of treatment because of cytopaenia. The most common side-effect that occurred in 12 patients was anemia requiring transfusion in one patient, and increased weekly dose of erythropoietin (4.000-12.000 IU (average 10.933 IU)) in other patients. A flu-like syndrome was documented in 6, myalgiae in 4, and arthralgiae in 5 patients. Rectorrhagia, endocarditis and severe cough were recorded in one patient each.
Conclusions: According to our experience PEG-INF 2a has limited efficacy in dialysis patients. Significant proportion of patients discontinued treatment because of side-effects. SVR did not significantly differ from the ETVR. Additional studies with different duration of PEG-INF 2a treatment and long-term follow up are needed to determine the optimal length of treatment of HCV infection in dialysis population.

Su294 ROLE OF INTRA DIALYTIC PARENTERAL NUTRITION IN MALNOURISHED PATIENTS ON MAINTENANCE HEMODIALYSIS: RANDOMIZED CONTROLLED STUDY

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Introduction and Aims: Poor nutritional status and muscle wasting is common in Indian patients on hemodialysis and is associated with increased morbidity and mortality.
Methods: Effect of intradialytic amino acid supplementation on nutritional status and body composition. Methods: Prospective randomized study on 22 ESRD patients divided into two groups: Intradialytic parenteral nutrition (IDPN) group and controls (not on IDPN) with 11 patients in each group. IDPN group was given aminoacid infusion containing 25 grams of protein and 200 calories, twice weekly during dialysis over a period of 2 months. Body composition was evaluated by bioelectrical impedance analyzer. BIA (BMI, fat, fat free mass (FFM), protein, glycogen, body cell mass (BCM) muscle mass, extracellular mass (ECM), extracellular solids (ECS) and water compartments) and anthropometric measurements were taken at baseline, one moth and two months. Serum albumin, prealbumin homocystein, C reactive protein (CRP), transferrin, lipid profile were done at baseline. Baseline, one month and two months valuesW were compared within each group and between two groups.
Results: Subjective global assessment showed patients had moderate to severe malnutrition (Score 3-5). All the patients had low serum albumin and prealbumin. Homocystein and CRP levels were high. On comparing two groups significant difference was observed after 1 and 2 months of supplementation in weight (p=.048), phase angle (p=0.013) BMI (p=0.36) FFM (p=0.021), BCM (p=0.018), ECM (p=0.026) Protein (p=0.48), Muscle mass (p=0.50) Glycogen (p=0.011), ECS (0.021). Within IDPN group significant difference was observed in various parameters (FFM, Fat, BCM, ECM, Protein, Muscle mass, calcium and glycogen) between baseline, month one and month two suggesting improvement in nutritional status with IDPN. All the patients were malnourished and felt weak at the start of the study. Patients on IDPN showed significant improvement in appetite, general condition and weakness. They felt more energetic and could get back to work after four weeks of supplementation.
Conclusions: Despite small sample size and short duration of study, patients in IDPN group showed significant improvement in general well being suggesting beneficial effect of supplementation during dialysis.

Su295 ADIPONECTIN LEVELS CAN BE USEFUL IN STRATIFICATION OF CHRONICALLY HAEMODIALYSED PATIENTS

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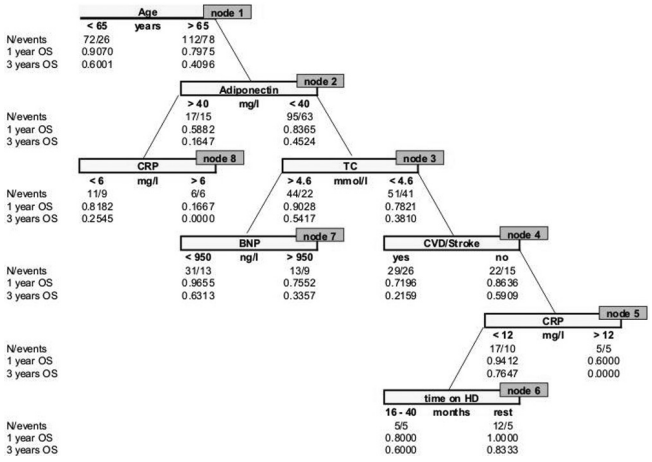
Introduction and Aims: End-stage renal disease (ESRD) patients on chronic haemodialysis (HD) treatment suffer from highly increased overall mortality. Simple algorithms for differentiating high-risk individuals within this group can be useful for improving HD patients' survival rates. Adiponectin (ADPN) is adipocyte-derived hormone that was associated with both positive and negative outcomes. Natriuretic peptide type B (BNP) is a marker of heart failure – the most common cause of death in ESRD patients. We aimed to prove, whether adiponectin and BNP measurement, in addition to traditional nutrition status and inflammation markers, can be useful in selecting high-risk HD patients.

Methods: We analysed total ADPN levels by ELISA method, BNP by immunochemical chemiluminiscence method, C-reactive protein by ultrasensitive method (CRP) and albumin, hemoglobin, total cholesterol (TC), HDL cholesterol by routine methods in blood samples of 184 patients (123 males), median (interquartile range) of age was 68 (59 – 74) years. For multivariate statistical analysis, classification and regression tree (CART) was used. Predictors in each node of CART were found by stepwise multivariate Cox regression model, only predictors with $p<0.05$ were included. In addition to the above mentioned analytes, age, sex, time on HD, body-mass index, smoking, presence of diabetes and history of cardiovascular disease or stroke (CVD/Stroke) were tested for inclusion in the final model. All analyses were performed by SAS Release 8.02 software.

Results: The resulting CART is depicted in the following picture *(N = number of patients in appropriate group; 1 year OS and 3 years OS = 1 and 3 years overall survival; in simple words, reading from top to bottom is algorithm of subsequent questions that leads to the most efficient selection of high-risk patients)*:



In our model, ADPN levels with cut-off value 40 mg/l were the 2nd most important factor in differentiating high-risk patients. Furthermore, BNP concentrations (with cut-off value 950 ng/l, the 7th most important factor) appeared to help with risk stratification of HD patients. From more traditional predictors, age, TC, CVD/Stroke, CRP and time on HD proved their usefulness.

Conclusions: Measurement of ADPN and BNP levels in ESRD patients treated by chronic HD can be useful in differentiating high risk patients. Surprisingly, some of more traditional survival factors (albumin, hemoglobin) did not prove its value in this setting.

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Su296 ECONOMIC BENEFITS OF ANAEMIA MANAGEMENT USING ESAs AND INTRAVENOUS IRON SUPPLEMENTATION IN NON-DIALYSIS-DEPENDENT-CKD PATIENTS

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Introduction and Aims: In non-dialysis-dependent chronic kidney disease (NDD-CKD), erythropoiesis-stimulating agents (ESAs) are widely used for anaemia treatment. However, ESA usage has come under pressure due to safety concerns. By optimising iron stores and iron bioavailability, ESA efficiency can be maximised and their initiation delayed. Ferric carboxymaltose (FCM) is a dextran-free, innovative intravenous (i.v.) iron preparation that allows the administration of a single, high dose of up to 1000 mg iron in approximately 15 minutes without the necessity of a test dose. Initiating FCM prior to ESA therapy may lead to significant cost savings. This study aimed at establishing the cost impact of adding i.v. FCM to ESA-based anaemia treatment in NDD-CKD patients, from a third party payer perspective in Switzerland.

Methods: We combined, by calculating weighted averages, data from several clinical studies showing that i.v. iron supplementation reduces ESA dose and/or delays ESA initiation in NDD-CKD patients. On this basis, cost savings due to reduced ESA use were estimated and a cost neutral price (i.e. the average cost of spared ESAs per iron-treated patient) for i.v. iron was determined. According to this definition, if the costs of purchasing and administering i.v. FCM are less than the cost neutral price, FCM will be cost saving.

Results: In our model, ESA initiation was avoided in 55% of NDD-CKD patients, resulting in an overall ESA dose-sparing effect of 25%. The annual cost-neutral price of i.v. FCM was €61, assuming an ESA regimen of 4000 IU per week with yearly ESA costs of €2429, compared with a recommended ex-factory FCM price of €25 per 100 mg iron, plus a pro rata administration cost of €2 per 100 mg (assuming a dose of 1000 mg per iron administration). In the clinical studies, typically, an initial loading dose of 1000 mg iron was found to be needed, followed by a 150 mg monthly maintenance dose. Based on this observed dosing, in our model we determined the annual cost savings of FCM, using a total dose of 2650 mg i.v. iron per patient in the first year (including loading dose), at a cost of €743, and 1800 mg per patient per year in subsequent years, at an annual cost of €510 (administration costs included). In both the initial and maintenance phases, i.v. FCM resulted in ESA sparing, saving €1613 per patient per year. Therefore using i.v. FCM would result in a net cost saving per patient of €870 in the first year and €1103 annually in subsequent years.

Conclusions: From the perspective of Swiss statutory health insurance system, managing anaemia in NDD-CKD patients using i.v. FCM may be considered cost-saving. Key elements of these economic benefits included ESA sparing and low administration costs. By reducing the use of ESAs overall, the costs saved outweighed the costs of buying and administering FCM.

Su297 RISKS OF MALIGNANCIES BEFORE AND AFTER DIALYSIS IN ESRD PATIENTS – RESULTS FROM TAIWAN NATIONAL HEALTH INSURANCE DATA ANALYSIS

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Introduction and Aims: Past studies had proved that patients who received renal transplantation had higher risks of malignancies but less studies focus on long term dialysis patients. We aimed to estimate the risks of malignancies in dialysis patients compared to general population during the period before and after dialysis.

Methods: Patients who received long term dialysis in 1999-2004 were considered as dialysis group (DG). Control group (CG) was randomly selected from dataset of general populations from National Health Insurance data by matching birth year and sex. We defined index date, date of stating dialysis for DG, and the first day of the year compared to case's starting dialysis year for CG. Time was backward calculated from the index date to the first date of malignancy diagnosed or Jan. 1 of 1997 for before dialysis period and end of 2004 for after dialysis period. All subjects who occurred malignancies (ICD9: 140-208) were considered as events in those periods, otherwise was censored. Cox regression model was used to compare risks of malignancies between DG and CG.
Results: The crude incident rate of malignancy was 14.42‰ in DG, 6.53‰ in CG before dialysis period; 16.40‰ in DG and 9.96‰ in CG in after dialysis period. After adjusted by age, sex, dialysis year, and geographic, the risks of all malignancies, liver cancer, renal cancer, transitional cell carcinoma, upper urinary tract cancer, and bladder cancer were higher in DG group than in CG group, both in the period before and after dialysis.

Hazard ratio for malignancies compare to dialysis and control group before and after dialysis

	Before dialysis		After dialysis	
	Hazard Ratio (95% CI)	p-value	Hazard Ratio (95% CI)	p-value
All malignancy (140-208)	2.25 (2.09, 2.43)	<0.001	1.66 (1.57-1.75)	<0.001
Liver cancer (189 or 189.0)	2.06 (1.61, 2.62)	<0.001	1.78 (1.54-2.07)	<0.001
Renal cancer (189 or 189.0)	11.91 (5.54, 25.61)	<0.001	9.65 (6.22-14.99)	<0.001
Transitional cell carcinoma (188 or 189.1, 189.2)	5.73 (4.72, 7.96)	<0.001	7.74 (6.04-9.92)	<0.001
Upper urinary tract cancer (189.1 or 189.2)	15.79 (6.44-38.71)	<0.001	10.78 (6.35-18.28)	<0.001
Bladder cancer (188)	4.47 (3.13-6.40)	<0.001	7.16 (5.41-9.47)	<0.001

Full Model adjusted by age, sex, dialysis year, and geographic

Conclusions: Dialysis patients had higher risks of malignancies, especially the renal and urinary tract cancers, than the general populations even during the period before dialysis. It suggests that carefully monitoring possibility of malignancy is necessary in care of CKD patients.

Su298 IS IT NECESSARY TO CHANGE THE STRATEGY IN THE METHOD OF B HEPATITIS VACCINATION IN HAEMODIALYSIS PATIENTS?

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Introduction and Aims: The prevalence of hepatitis B virus infection in haemodialysis patients has decreased in the last years. Because the risk of viral transmission has not been eliminated completely the vaccination continues playing a fundamental role in prevention. The rate of inmunization in this population, with the conventional recombinant HBV vaccine intramuscularly administrated is very changeable and in occasions limited.
We compared the inmunogenicity, defined as the increase of the titer of HBV surface antibody (anti-HBs) to 10 UI/L or greater, of a intradermal method of vaccination with the one of the conventional intramuscular route of administration.
Methods: The intradermal group (ID) received a 20 mcg dose every two weeks during 3-6 months, in comparation with the standard intramuscular (IM) double dose, at 0, 1, 2 and 6 months.
82 haemodialysis patients were analyzed, with a follow-up period of 8 months. The titer of anti-HBs was determinated at the beginning of the study, at 5 and 8 months. The variables were analyzed with the statistical program SPSS 14.0.
Results: 53 of 82 patients analyzed (64.6%) had received the standard (IM) double dose recombinant vaccine before the beginning of the study with an average time from the last dose of 15.9 ±. 16.8 months.
46 patients (86.8%) were not immunized, 7 (13.2%) achieved a titer greater than 10 UI/L. The remaining, 29 patients (35.4%) had not received previous vaccination and 5 of them showed antibodies.
We administrated the ID vaccine in all those not immunized, a whole of 69 patients. After completing the initial cycle ot treatment (3 months), 42 patients (60.8%) obtained high level of anti-HBs, opposite to 27 (39.1%) that were not immunized, the above mentioned received the second cycle of ID vaccination for another 3 months. 18 of these 27 patients (66.6%) achieved immunitation. 9 (33.8%) did not respond. After completing 3 and 6 months of ID treatment every two weeks, 60.8% and 86.9% of patients, respectively, developed high titer of antibodies. In contrast with the percentage (13.2%) of the IM group that were immunitated, the difference was statisticly significant (p 0.01).
No patient presented adverse prominent reactions related to the form of administration.
Conclusions: The Response limited to the standard method of VBH vaccination in the dialysis population forces to look for other strategies. In our experience, with the intensive ID vaccination it reaches a high percentage of immunization. We believe that it might be a suitable option, of easy application and without prominent complications.

Su299 THE RISK OF DEATH IN MAINTENANCE HEMODIALYSIS PATIENTS CAN BE PREDICTED BY MEANS OF ELECTRICAL BODY IMPEDANCE

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Introduction and Aims: Malnutrition is a major determinant of long-term outcome of maintenance haemodialysis (MHD) patients. Severe malnutrition is accompanied by alterations in body weight and body mass index (BMI). However, the major limitations of BMI are its poor sensitivity.
The aim of this study was to evaluate the efficiency of electrical body impedance measurement (BIA) to indicate alterations in body composition, and to predict clinical outcome of MHD patients.
Methods: BIA has been performed during a two year-time period, by means of a single frequency (50 kHz, 0.8 mA) impedance analyzer (ST-BIA, Akern), in 126 MHD patients (F46, M80), aged 22-90 years (mean 65.1), dialysis vintage 0.5-36 years (mean 6.2), body weight 36.3-135.8 kg (mean 76.0). Twenty-two patients died during this time period. Forty-six patients were re-examined at 2 years interval. Serum albumin (ALB) and dialytic efficiency (single pool Kt/V) were measured in all patients.
Values of resistance (R), reactance (Xc), phase angle (PA), body cell mass (BCM), and of extra-cellular volume as percentage of total body water (ECW%) were directly measured by the impedance analyzer.
Results: The cross sectional analysis of the 126 MHD patients demonstrated a statistically significant positive correlation between ALB and PA, Xc, BCM. Age and ECW% correlated negatively with ALB. No correlation was found between BMI or sp Kt/V and ALB.
The longitudinal study of 46 patients demonstrated no significant variation in ALB and sp Kt/V, a slight, but statistically significant reduction in BMI ($p<0.05$) and a higly significant reduction in PA, Xc, BCM, while ECW% significantly increased.
Significantly lower values of PA, Xc and BCM and significantly higher values of ECW% were found in the 22 patients deceased within 2 years from the first examination in comparison with still alive MHD patients, while no difference was found for ALB and sp Kt/V values.
Conclusions: BMI seems inadequate to assess nutritional status in MHD. Data derived from BIA, in particular PA, Xc, BCM and ECW% are significantly correlated with ALB, which is a validated marker of malnutrition in MHD patients. Furthermore, BIA seems even more sensitive than ALB to predict the risk of death in MHD patients.

Su300 EFFECT OF NUTRITIONAL STATUS AND COMORBIDITY INDEX ON QUALITY OF LIFE IN HEMODIALYSIS AND PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Research of quality of life (QoL)) is applied to estimate the efficiency of treatment, to selection the optimal variants of therapy, including methods of renal replacement therapy.
The aim of the study was to evaluate QoL parameters of patients with

end-stage renal disease treated by hemodialysis and peritoneal dialysis and to analyze comorbidity index and nutritional status.

Methods: We carried out a comparative study from 2008 to 2009 years in Botkin's hospital in Moscow. 185 patients have been investigated. The first group (97 patients) received treatment by hemodialysis, 56 men and 41 women, their average age 59,3±11,5 years. Period of treatment by hemodialysis on the average made 57,2±11,0 months. The second group included 88 patients on PD, 52 men and 36 women, aged 58,2±13,2 years. Time of treatment by a peritoneal dialysis on the average made 29,6±3,9 months. Research was made with the account of social (the questionnaire Kidney Disease Quality of Life Short Form and the hospital scale of A.S. Zigmond an estimation of alarm and depression) and the general laboratory data (albumin, C-reactive protein (CRP), KT/V, hemoglobin). We also estimated comorbidity index of M.E. Charlson and nutritional status: body mass index (BMI), fat mass (FM) and lean body mass (electrical bioimpedance).

Results: At patients on PD the QoL parameters authentically above, then at patients on HD ($p<0,05$): role emotional functioning 47,4±13,0 vs 37,0±17,0; social functioning 53,3±12,1 *vs* 44,3±13,2; burden of kidney disease 30,6±12,6 vs 19,8±16,3; effects of kidney disease 60,1±10,2 vs 52,5±8,4; work status 14,8±8,4 vs 5,0±3,0. The total amount of points according to the scale QoL negatively correlated at all investigated patients with Charlson comorbidity index (r = – 0,54) and CRP (r = -0,76). Directly correlation with nutritional status: albumin (r = +0,32), total protein (r = +0,63), FM (r = +0,44), lean body mass (r =+0,75). At the analysis of level of affective frustration it has been revealed: at patients on PD it was observed authentically ($p<0,05$) higher level of anxiety 8,3±2,7 vs. 6,6±3,6; and the estimation of propensity to depression was lower, than at patients on HD 7,4±3,9 vs. 8,2±3,5.

Conclusions: Thus, QoL and depression parameters depend on types of renal replacement therapy, comorbidity index and conditions of nutritional status.

Su301 PERITONEAL DIALYSIS TREATMENT IN PATIENTS WITH SEVERE MENTAL RETARDATION. IMPORTANT MEDICAL AND ETHICAL CONSIDERATIONS

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Introduction and Aims: In Hungary, renal replacement therapy (RRT) is available for everyone whose kidney function progresses to end stage renal disease. Hungarian Healthcare Law declares that "All patients have the rights for medical treatment without any discrimination". As all human lives have equal values, people with severe mental retardation must not be discriminated. Nevertheless, providing dialysis treatment for severely mentally retarded patients is challenging due to their inability to cooperate. The major questions are, which modality of RRT is the most beneficial, whether they can achieve adequate dialysis by CAPD, how frequent the complications are, and whether their quality of life can be improved by initiating dialysis.

Methods: Among the 46 CAPD patients in our PD unit, there are 3 severely mentally retarded young males. All three patients have difficulties in expressing themselves verbally, which make their cooperation even more difficult. For two of them, who live in the same long-term care facility, the 18-member staff had been trained by us to perform CAPD. These dedicated caregivers precisely make the bag exchanges, care for dietary restrictions, fluid balance, and are in charge of the daily medications.

Results: N.I. is a 27 years old male, who was born with multiple developmental abnormalities. He has developed ESRD in 2003. Difficulties in planning RRT were, that – besides his small body size (130 cm, 29 kg) -, he has an even smaller abdominal space due to severe thoracic gibbosity, and that he has been excreting some urine by uretero-cutaneostoma. His mental age is about 5 years old. In spite of these difficulties, Tenckhoff catheter was successfully inserted, and regular CAPD has been performed without any difficulties, with a weekly KT/V of 2.4. He has been cooperative in taking medications and dietary restrictions. His peritonitis rate – 1/11.5 months – is reasonable, considering the proximity of the uretero-cutaneostoma. B.L., a 36 years old male has autism. ESRD had developed due to malignant hypertension, and CAPD has been initiated for 10 months. His weekly KT/V is 2.2, he has had peritonitis twice. Our third patient, H.D., a 23 year old male has oligophrenic imbecility with symptomatic epilepsy. He has sometimes been aggressive, especially when dietary and fluid restrictions have to be kept. His CAPD treatment has been performed by his mother since 2006 without any technical problems. He is anuric, his weekly KT/V is 1.5, and the peritonitis rate is 1/26 months. The caregiver persons have judged all three patients' quality of life good.

Conclusions: Based on our observations, severely mentally retarded patients can be successfully treated by CAPD. Their treatments can be organized in long-term care facilities, provided that dedicated caregivers are well trained for this modality. Alternatively, one of the family members may care for CAPD at the patient's home. The quality of life of this special patient population can be preserved in spite of their multiple severe disabilities.

Su302 PREGNANCY IN HAEMODIALYSIS PATIENTS

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Introduction and Aims: Pregnancy in patients on maintenance haemodialysis is rare. The frequency of conception has been reported as 0.3% to 1.4% with only a 30-50% rate of successful delivery. This study investigated the pregnancy outcome in patients on chronic haemodialysis over the past 30 years in our department.

Methods: 7 pregnancies in 6 women undergoing chronic haemodialysis during the period from 1975 to 2007 in our department were retrospectively reviewed.

Results: All patients were already on haemodialysis when they became pregnant. They were aged of 27 to 35 years (mean= 32.8 years). Pregnancy was obtained 2,75 years (1 -5 years) after start of haemodialysis. Maternal complications were hypertension in 2 cases and anaemia in 6 cases. Foetal complications were intrauterine growth retardation in 5 cases and premature delivery in 4 cases. Two pregnancies delivered live newborns and 4 ended with intra-uterine fetal demise or neonatal death. The mean birth weight was 1570g (1000 – 2250g). No fetal anomaly was detected.

Conclusions: Foetal complications are frequent and serious in haemodialysis patients. The special attention was paid on the necessity of multi-disciplinary collaboration and increasing dialysis delivery for successful delivery. After renal transplantation the chances of becoming pregnant are much higher as well as the chances of a successful outcome.

Su303 USING DISCRETE EVENT SIMULATION TO PREDICT THE NEED FOR RENAL REPLACEMENT THERAPY RESOURCES

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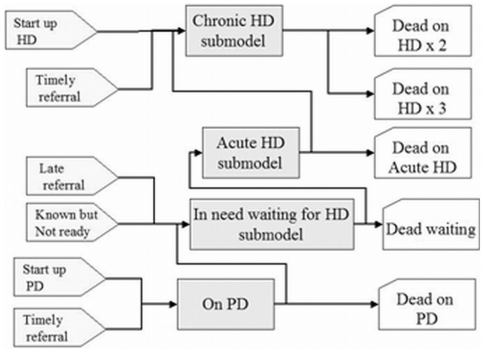
Introduction and Aims: Barbados, like the rest of the Caribbean, has seen a rising burden of end-stage renal failure (ESRF). The HealthCare System in Barbados is socialized and all treatment is provided free for its citizens. Hence the socio-economic burden of renal replacement therapy (RRT) on society is significant.

The aim of this study was to assess the feasibility of building a discrete event simulation (DES) model to assess the impact of possible healthcare system interventions on the need for RRT capacity.

Methods: We have previously shown that DES can be a very useful tool[1]. Using the ARENA simulation software (V 12.00 CPR9 Rockwell Automation Technologies Inc, USA) we built a simulation model with a core as proposed by Davies & Roderick[2].

By using a sub-modeling technique, we added several sub-models see figure.

1. The module 'Chronic HD'. The patient is scheduled for 3 times, 3 hours weekly HD if slots are available, otherwise reduced to 2 times, 4 hours weekly for as many patients as needed to free slots. If capacity is full, no new patients are accepted. It is expected that 2 times weekly incorporates a greater mortality and morbidity than the 3 times weekly option.
2. The module 'In need waiting for HD' simulates patients in immediate need for RRT who are waiting to receive treatment.



3. The module 'Acute HD' simulates the increased mortality and morbidity during the first 3 months.
4. An entry point for patients starting HD as timely referrals. They enter the 'chronic' HD module directly.
5. Patients with HIV are currently only accepted into the PD program.
6. The programs scheduling module was employed detailing the available dialysis resources.

Results: The simulation was simulating 5 years with 100 replications. With the present number of patients on dialysis and present rate of admissions, the system predicted a larger capacity than perceived by the staff. The capacity was only utilized to 80.2% of its scheduled capacity. The simulation reached a steady state with a population on RRT of approximately 220. However, it also showed the occurrence of peaks where the current resources would not meet demand, as seen by one period had 15 patients dying while waiting on treatment. Organizational and cultural barriers are believed to be major contributors to the non-optimal use of the resources.

Conclusions: Simulation of RRT can be a useful tool in helping accessing the optimal amount of resources needed and in testing the effect of planned interventions.

References:

1. Lassen Nielsen A, Hilwig H, Kissoon N, Teelucksingh S. Discrete event simulation as a tool in optimization of a professional complex adaptive system. Stud Health Technol Inform 2008; 136:247-252.
2. Davies R, Roderick P. Predicting the future demand for renal replacement therapy in England using simulation modelling. Nephrol Dial Transplant 1997; 12(12):2512-2516.

Su304 PREDICTORS OF QUALITY OF LIFE IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Compromised quality of life (Qol) continues to be a significant problem for patients receiving haemodialysis. This study aimed to assess the factors influencing the quality of life in patients on maintenance dialysis.

Methods: The Qol, using (Sort Form – 36 Health Survey) SF-36, in 136 HD patients, has been evaluated. The socio-demographic data was collected (age, gender, socio-economic status, education level, marital status, family support and presence of sleep disturbance). Dialysis practice variables included: dose, vintage, single pooled – kt/v, Protein catabolic rate, ultrafiltration (UF), UF as percentage of dry body weight and UF rate as percentage of dry body weight per hour. Nutritional and inflammatory state were measured by subjective global assessment (SGA), body-mass index (BMI) and malnutrition – inflammation score (MIS) determined by the method of Kalantar et al. Laboratory values including Hemoglobin, Albumin, Creatinin, C-reactive protein, Cholesterol and mineral metabolism indices as Phosphorous and Parathormon were evaluated. Clinical findings on presence of Diabetes, Coronary artery disease, Arrhythmia, Chronic interdialytic hypotension, IDH, Hepatitis C and thromboses of fistulas were noted. Univariate and multivariate regression analysis were performed.

Results: Women scored lower in all Qol scores, but insignificantly. We found that older age, lower social status, sleep disturbance and poor family support are associated with lower Qol scores. Diabetic patients scored significantly worse on the physical health dimension scales. No significant association was found for the Hg levels, probably due to the optimal values found in the vast majority of our patients. A strong association was found between SF 36 scales measuring all the physical and mental health concepts and the albumin level. Those associations didn't hold after adjusting for the confounding factors. We found inverse, but insignificant correlation between all the SF 36 scores with BMI and CRP. The presence of Hepatitis C had no influence on the Qol. Dialysis timing and vintage showed no associations to the Qol scores, but the low dialysis adequacy independently worsened the Mental component score. The IDH correlated independently with all SF-36 dimensions except the social functioning scale. The patients having IDH had significantly lower values for albumin, and higher values for UF, UF as percentage of dry body weight, UF rates per hour and higher rates of thromboses of fistulas. They also had higher presence of chronic interdialytic hypotension. In the multiple regression model, the age, family support, sleep disturbance, creatinine, SGA, DM and IDH were the strongest independent predictive markers for the Physical component score. For the Mental component score age, family support, adequacy, creatinin, MIS and IDH were the strongest predictors.

Conclusions: The Quality of life is predicted by the age, dialysis adequasy, nutritional status, family support, sleep disturbance and comorbidites. Efforts should be done to prevent the frequency of the hypotonic episodes causing IDH to improve the Qol in hemodialysis pts.

Su305 EARLY NEPHROLOGY REFERRAL INFLUENCES ON MORTALITY OF DIALYSIS PATIENTS IN DEVELOPING COUNTRY

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Introduction and Aims: Despite improvements in dialysis care, mortality of patients with end-stage renal disease (ESRD) remains high. One factor that has thus far received little attention, but might contribute to morbidity and mortality, is the timing of referral to the nephrologist.To evaluates the influence of early referral to nephrology of the patients with chronic renal failure in the mortality of the patients who start dialysis.

Methods: There were included in the study the patients who started dialysis from January 2005 to December 2005. Patients who started dialysis after acute renal failure were excluded. Clinical and analytical data were determined for each patient at the start of dialysis and during the follow-up. Early (ER) and late referral (LR) were defined by the time of first nephrology encounter greater than or less than 3 months respectively, before dialysis initiation. Mortality analysis and global and annual during the first five years of follow-up survival analysis (using Cox proportional hazards regression) were carried out.

Results: Of the 186 patients, 95 (51.1%) were referred late and 91 (48.9%) were early referred. There were no differences in gender, cause of ESRD and mean follow-up between ER and LR patients. However, there were significant differences in comorbid factors between these two groups (ER vs LR): age (57.1±13 vs 52.8±15.6; p=0.03), diabetes (48.4% vs 32.6%; p=0.02), hypertension (86.7% vs 67.4%; p=0.002), arterite of inferior member (6.6% vs 4.2%; p=0.016) and Charlson score (5.2±2.1 vs 4.4±2; p=0.018).

Compared to LR, ER patients were more likely to have lower level of serum creatinine (855.5±288.4 vs 948.1±309.9;p=0.04) and higher uricemia (508.5±166.5 vs 478.6±167.3; p=0.04), but, not significant difference concerning albuminemia, hematocrit, and proportion of temporary catheters at the start of dialysis.

The overall mortality rate for the 186 patients was 23.7% at the end of the study. The survival rate for ER versus LR group at 1 year, 2 years, 3 years, 4 years, and 5 years were respectively 88.1% vs 84.1%, 82.1% vs 79.6% and 73.7% vs 66.9%.In multivariate analysis by cox model regression, only the age > 50 years (p=0.0015), diabetes (p=0.01), hypertension (p=0.03), vascular cerebral accident (p=0.025), arterite of inferior member (p=0.003), Charlson score (p<0.001) and serum creatinine level > 950 μmol/l at the start of dialysis (p=0.005) were significant predictors factors of mortality. Nevertheless, in successive models fitting after 5 years of follow-up the variable ER not influenced in any way the mortality rate 5 years later.

Conclusions: It is concluded that early referral to the nephrologists in developing country don't influenced short and long-term mortality. Pre-ESRD care of patients treated by nephrologists was also less than ideal, can explicated by lack of the means available. Meanwhile, pre-ESRD educational efforts need to target patients, generalists, and nephrologists.

Su306 UPDATE ON ESRD IN FRANCE: REIN REGISTRY

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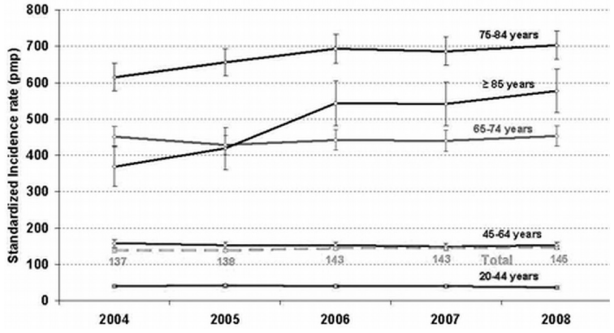
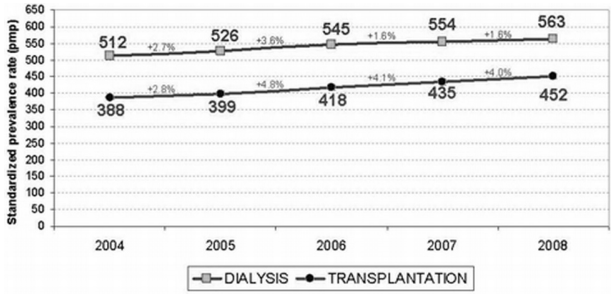
Introduction and Aims: The French REIN registry covers now the entire country. We present key figures of the RRT among 20 regions in 2008. Trends in incidence and prevalence rates of dialysis and transplantation in 9 regions that contributed to the registry between 2004 and 2008, will also be shown.
Methods: The French REIN registry intended to include all ESRD patients on RRT either dialysis or transplantation living in metropolitan France or in overseas districts. The details of its organizational principles and quality control are described elsewhere (NDT 2006). To compute standardized rates, we adjusted by the direct method, for age and gender, using the french population of the considered year as the reference.
Results: In 2008, 8,033 patients with ESRD living in 20 french regions, started renal replacement therapy: median age was 70.0 years; 3.6% had a preemptive graft. The overall crude annual incidence rate of ESRD was 146 pmp, with significant differences in sex and age-adjusted incidence across regions (105 to 422 pmp).
At initiation, more than one patient out of two had at least one cardiovascular disease and 40% diabetes (91% Type 2 diabetes); 19% were dependent, partially or totally, for transfers.
Since 2004, incidence rates among patients under 75 years remained stable while it increased for patients over 75 years.



Between 2004 and 2008, median age for the incident patients increased, more among patients without diabetes (+2 years) than among those with diabetes (+0.9 years).
On December 31, 2008, 57,875 patients were living in the 20 regions, on dialysis (HD 50%; PD 4%) or with a functioning graft (46%). Median age was 70.0 years and 54.1 years for the patients on dialysis and on transplantation respectively.
Among the 9 regions, the gap between the prevalence rates of dialysis and transplantation was constant before 2006. Since 2007, it seems that this gap tends to decrease.
In the 2002-2008 cohort of 34,198 incident patients, the overall one-year survival rate was 83% and 50% at 5 years. Survival remained above 50% at 2 years in patients older than 75 at RRT initiation.



Among the patient under 60 years, the median time to be on the waiting list for a renal graft is 15 months and the median time to be transplanted is 34 months.
Conclusions: After 7 years, in France, the REIN registry appears to be a promising shared support tool for the evaluation of professional practices and policies, for decision support in planning regional public health care and for providing pertinent epidemiologic data that can be used to improve the knowledge of the ESRD.

Su307 β_2 MICROGLOBULIN CLEARANCE AND QUALITY OF LIFE – A COMPARISON OF HIGH-FLUX HAEMODIALYSIS & ONLINE HAEMODIAFILTRATION

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Introduction and Aims: Online haemodiafiltration (HDF) is more expensive than high-flux haemodialysis (HD), but achieves better clearance of middle molecules and has been linked to improved haemoglobin (Hb) values.
It is not clear whether HDF also leads to improved quality of life. We assessed patients' quality of life before and after conversion to HDF, using the Kidney Disease Quality of Life-Short Form (KDQOL-SF).
Methods: We used KDQOL-SF version 1.3 as a self-assessment tool. 51 patients on haemodialysis were recruited into the study.
The questionnaire included 43 items on End-stage Renal Disease (ESRD) targeted areas, and a 36-item health survey. Patients were asked to fill out the questionnaire again after 4 months of HDF.
Pre- and post-dialysis β2M and Hb levels were measured pre- and post-conversion to HDF, and Aranesp dosages recorded.
Questionnaires contained a mix of 'Yes, No' and numerical responses, which were then transformed into a 1-100 range, with higher transformed scores always reflecting better quality of life. These scores represented the percentage of total possible score achieved. Items in the same scale were averaged together to create final scores.
All scores were then added to form an 'Overall Health Score'. These scores, and blood results were compared to see if there was any difference in the quality of life, β_2M clearance, Hb levels, and Aranesp dose.
Results: 34 patients completed the study. Male female ratio was 1:1.4 with average age of 61.6 (29-88) years. Scores from the questionnaires are summarized and compared in tables 1 and 2. The Overall Health Score on HD was 56.8 compared with 55.8 on HDF (p=NS).

Table 1. ESRD-targeted areas

SCALE	HD	HDF
Symptoms/problems	65.5	65.7
Effects of kidney disease	64.7	60.5
Burden of kidney disease	42.5	39.6
Work status	20	20
Cognitive function	61.3	61.7
Quality of social interaction	76.3	74.7
Sexual function	71.5	70.5
Sleep	61.3	66.3
Social support	70	72.9
Dialysis staff encouragement	83	77
Patient satisfation	52	51

Table 2. 36-item health survey (SF-36), & Overall Health Score

SCALE	HD	HDF
Physical functioning	48.6	47
Role – physical	34.5	24.3
Pain	55.4	61.9
General Health	53.9	55.2
Emotional well-being	58.7	55
Role – emotional	51.1	46.5
Social function	51.7	46
Energy/fatigue	57.4	57.8
Overall Health Score	56.8	55.8

Hb values were 11.6 and 11.5 g/dl, and average Aranesp requirements were 30.7 and 35 ug/week before and after switchover to HDF.
Although there was no difference in pre-dialysis β_2M levels (27.1 vs. 27.2 mg/l) on HD and HDF, the β_2M reduction ratio was significantly greater with HDF (mean ± SD: 61.2±7% vs. 72.9±4%. $p<0.0001$)
Conclusions: HDF did not result in any improvement in quality of life of dialysis patients as assessed by the KDQOL-SF. As expected, the β_2M reduction ratio was significantly better on HDF. There was no reduction in Aranesp dose requirements.
Disclosure: The development of this questionnaire was supported in part by an unrestricted grant from Amgen to RAND, a subgrant from the University of Arizona to RAND, and a grant from Baxter Healthcare Corporation.

Su308 MEDICAL PROBLEMS AND QUALITY OF LIFE (QoL) IN ELDERLY PATIENTS ON DIALYSIS. RETROSPECTIVE ANALYSIS AND SURVEY

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Introduction and Aims: According to the WHO registry the average human life span in the second half of the 20th century increased by 20 years, resulting in 10% of the world's population now being over 60 years of age. By the year 2050 this proportion is estimated to increase to 30%. A strong relationship between the increase of life expectancy and a rise in chronic kidney disease (CKD) prevalence has been reported. The number of elderly patients with CKD at stage 5 is expected to be higher in the future for many reasons, eg. type II diabetes mellitus (DM) or because of improved diagnostic methods. Thirty % of dialyzed patients are over 65 years of age. Their medical problems and QoL become more and more important for nephrologists.
Methods: The first part of the study was a retrospective analysis of the medical histories of 40 patients on dialysis, who were older than 60 years. Cardiovascular, osteomuscular, malignancies, diabetes, infections, psychiatric dialysis-related and other diseases were addressed. The second part worked at a specific questionnaire answered by 45 dialyzed patients at the aged 60-89 years. It consisted of 25 questions related to demographics, QoL, disease acceptance, socio-economic aspects and assessment of renal replacement therapy (RRT).
Results: A complete set of information was obtained from all patients. In the retrospective analysis 70% had chronic heart failure, 68% had hypertension and 28% had various types of malignancies. Every 3rd patient in this group had DM with its complications. Urinary tract (48%) and respiratory system (33%) infections were found to be the most common. 35% of this group had various psychiatric disorders including dementia (25%) and depression (10%).In the second part of the study, 40% of patients had medium and 26% higher education level. Twenty eight patients (62%) declared to be financial independent this might have an influence on QoL even though they required dialysis. Despite age, 15% of the responders were still professionally active, especially men; only 22% declared an active social life. Almost half of the questioned patients entirely accepted their kidney disease and the necessity of RRT, although 31% didn't. This part will need particular attention by the medical professionals.
Conclusions: Many new medical and QoL problems have emerged as a consequence of the increased availability of RRT for elderly patients in recent decades. These problems need to be addressed. Our study will hopefully enable a more precise description of these problems, resulting in better treatment and acceptance of the disease, providing our patients with a better chance to live longer and better lives.

Su309 NUTRITIONAL STATUS IN DIALYSIS PATIENTS: TUNISIAN EXPERIENCE

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Introduction and Aims: Malnutrition is common in dialysis patients and closely related to morbity and mortality. Therefore, nutritional status needs to be assessed in these patients by using several methods. In fact, nutritional management of dialysis patients plays a central role in nephrological practice.
Methods: In 97 prevalent diaysis patients (62 men, mean age 50,02±13,48 years), 85 in hemodialysis (HD) and 12 in peritoneal dialysis (PD), we evaluated the nutritional status by anthropometrics [post dialysis weigt (PW), hight (H), body mass index (BMI)], two subjective assessement scores [subjective global assessement (SGA), mini nutritional assessement (MNA)], biochimecal tests [predialysis serum albumin, prealbumin, transferrin,urea, creatinine, total cholesterol, triglyceride, bicarbarbonate, phosphorus, calcium, hemoglobin, high sensitivity CRP (CRP-hs)], protein equivalent of nitrogen appearance (nPCR), bioelectrical impedance analysis (BIA) to estimate body composition [person body fat (%BF), exta-cellular fluids (%ECF)] and quality of dialysis by kt/v and urea reduction ratio (URR).
Results: Age is found to be negatively correlated with serum prealbumin (p=0,03) and hemoglobin (p=0,000). Mean length of dialysis is associated with significantly reduced prealbumin (p=0,01), cholesterol (p=0,03). Compared to PD, HD is positively associated with significantly %MG (p=0,03), haemoglobin (p=0,006), albumin (p=0,005), nPCR (p=0,000) and creatinine (p=0,03). In the other side, patients in PD have higher cholesterol compared to HD patients (0,04). Caloric intake is correlated negatively with BMI (p=0,000), %MG (p=0,003) and with cholesterol (p=0,02) (no correlation with prealbumin and albumin). Protidic intake is correlated negtively with BMI (p=0,001), %MG (p=0,01) and cholesterol (p=0,05). nPCR is positively correlated only with creatinine (p=0,005) and urea (p=0,000). Even though CRP-hs negatively correlated with albumin (p=0,000) and prealbumin (p=0,004). Finaly The A-categorie SGA (compared to B and C-categorie) is associated with significantly advancing age (46,7 versus 54; p=0,007), caloric intake (28,6 versus 23,9; p=0,04), protidic intake (1,15 versus 0,9 g/kg/day; p=0,02), hemoglobin (10,3 versus 9,1g/dl; p=0,003) and prealbumin (352,9 versus 312,2; p=0,02). Among general factors, advancing age; mean length of dialysis; modality of dialysis; SGA score; CRP-us; nPCR and dietary intake influence significantly nutitional status.
Conclusions: Effort should be focused on the most efficient ways to maitain nutritional status in these patients based on anthropometrics, biochimecal tests and subjective assessement scores.

Su310 AMBULATORY CAPACITY IS A MAJOR DETERMINANT OF QUALITY OF LIFE IN MAINTENANCE HEMODIALYSIS PATIENTS

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Introduction and Aims: Quality of life (QoL) is invariably low in patients on maintenance hemodialysis (HD). However, the factors which determine QoL are incompletely understood. The aim of this study was to assess the characteristics of a Swiss HD cohort with regard to both physical and mental QoL.
Methods: Prospective dynamic maintenance HD cohort study (*monitor!* trial) assessing a wide range of clinical, laboratory and anthropometrical parameters. 122 patients were stratified by the median of the physical and mental composite of the SF-12 QoL instrument at time of inclusion into the study and were analyzed for their baseline characteristics as well as for living status and cumulative length of hospital stay during follow-up.
Results: See Table 1. Bivariate correlation analysis revealed significant associations between both physical and mental QoL with age, lean body mass, mineral mass, 3 min walk, step count, serum 25-OH-Vit. D and selenium concentrations. By multivariate regression analysis 3 minute walking distance was the sole independent variable predictive for both physical and mental QoL.
Conclusions: Assessing a broad array of clinical, epidemiological and biochemical parameters physical capacity turned out to be the major determinant of both physical and mental QoL in a maintenance HD

Abstract Su310 – Table 1

	Physical QoL			Mental QoL		
	Low	High	P	Low	High	P
Age, yr	69.4±11	63.5±15	0.012	68.2±13	64.7±14	0.139
Lean body mass, kg	44.7±9	51.5±9	0.002	46.5±8	49.9±9	0.077
# Comorbidities, N	1.71±1.1	1.34±1.0	0.053	1.77±1.1	1.29±1.0	0.011
Body mineral mass, g	1858±649	2226±690	0.012	2018±508	2078±862	0.684
3 min walk, m	172±86	198±56	0.053	170±69	201±73	0.018
24-h step count, N	4767±5743	8522±8822	0.017	4939±6551	8269±8273	0.035
Hb, g/dl	12.3±3	12.7±1	0.352	12.5±3	12.5±1	0.885
25-OH Vit. D, µg/L	17.1±12	25.0±21	0.026	17.5±11	24.0±20	0.063
IL-6, ng/l (<3.3)	10.8±13	6.2±5	0.007	10.3±13	6.7±5	0.033
Selenium, µmol/L (0.89-1.9)	0.64±0.2	0.76±0.3	0.009	0.64±0.2	0.76±0.3	0.009
NT-pro-BNP, ng/L (<400)	14564±17720	9557±15200	0.092	12645±16293	11622±17078	0.726
QoL, physical composite (>50)	14.2±14	45.2±7	0.000	18.3±19	40.7±11	0.000
QoL, mental composite (>50)	25.4±26	52.9±9	0.000	20.7±21	57.1±5	0.000

population. Serum vitamin D concentrations and bone mineral mass may be causally interrelated with these findings.

Disclosure: The study presented within this abstract/presentation has been supported by an unrestricted scientific grant from Roche Pharma Switzerland, Fresenius Medical Care Switzerland, and Genzyme Switzerland.

Su311 HEALTH RELATED QUALITY OF LIFE IN HEMODIALYSIS PATIENTS WITH VARIOUS TYPES OF ARTERIAL CALCIFICATION

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Introduction and Aims: At present, there is a lack of studies investigating health-related quality of life (HRQoL) in hemodialysis (HD) patients based on the presence/absence of arterial calcification (AC). The aim of our study was to evaluate whether the presence of AC may impact the score of HRQoL questionnaires in HD patients.

Methods: In a cross-sectional study on 75 HD patients (48 men; mean age 53.4±12.9 years; HD duration 120.7±75.1 months) we evaluated the presence of arterial intima (AIC) and media calcifications (AMC) using plain radiograms of the pelvis. The scales for mental component summary (MCS) and physical component summary (PCS) were derived from eight different subscales originally developed for the short form health survey (SF-36): physical functioning, role-physical, bodily pain, general health, vitality, social functioning, role-emotional and mental health. A higher score implicates a better HRQoL. We compared PCS and MCS scores among the groups of patients with various type of AC (group without AC, AIC and AMC) presence on radiograms.

Results: The groups did not differ (ANOVA, chi square) significantly in variables that may affect the HRQoL of HD patients, such as age, gender, hemoglobin, serum albumin and dialysis doses. Patients with presence of AMC (n=27; 36%) had significantly lower PCS (41.7±27.6 vs 56.9±26.1; p=0.006) score, as well as significantly lower MCS (45.6±25.7 vs 52.1±23.9; p=0.014) score in comparison with the group of patients with absence of AC (n=23; 30.7%). On the other hand, patients with the presence of AIC (n=25; 33.3%) had significantly higher PCS (46.8±25.1 vs 41.7±27.6; p=0.039) score in comparison with the group of patients without AC. There was differences in both, PCS and MCS scores, among the group of patient with presence of AIC and AMC. Also, no statistical difference was found in MCS score between the patients with AIC compared to the group with absence of AC.

Conclusions: There was significant association between the lower HRQoL scores and the presence of AIC and AMC on plain radiograms of the pelvis in HD patients. In order to improve the quality of life in HD patients, a prevention of AC development is an important prerequisite.

Su312 EPIDEMIOLOGY OF END STAGE RENAL DISEASE IN EL-MINIA GOVERNORATE, UPPER EGYPT: DATA OF THE YEAR 2008

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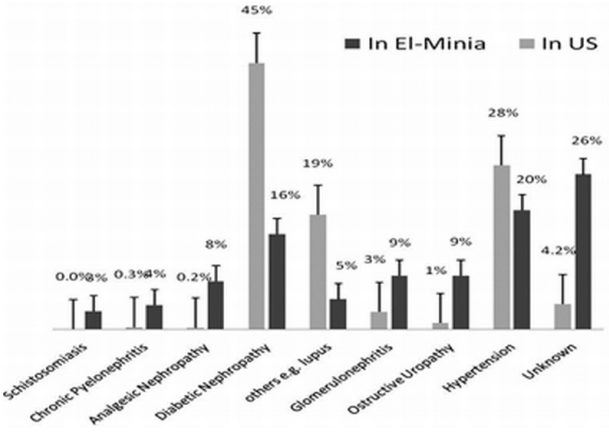
Introduction and Aims: Hemodialysis (HD) remains the main treatment line for most patients with end-stage renal disease (ESRD) in El-Minia Governorate, Egypt. Patients with ESRD are usually listed in the annual registry of El-Minia university hospital which report, demographics, family history, risk factors for ESRD, environmental exposure to toxins, work conditions, social history, and causes of death. Aim is to ascertain prevalence, etiology and risk factors for ESRD during year 2008 also to find some ideas that may explain causes of difference in epidemiology of ESRD in El-Mina Governorate and epidemilology of the US.

Methods: El Minia governorate is one of Egyptian governorates which are 29 governorates, it is located to the south of cairo by 143 Miles, it is formed of 9 districts its area is 32,279 square kilometer that is 3.2% of the total area of Egypt its population is 4.4 millon. Patients on renal replacement therapy (RRT) in El-Minia Governorate was 1729 patient, all were offered to participate in this study medical records and interview of the patients were the source of information.

Main causes of ESRD in El-Minia Governorate in comparison to US

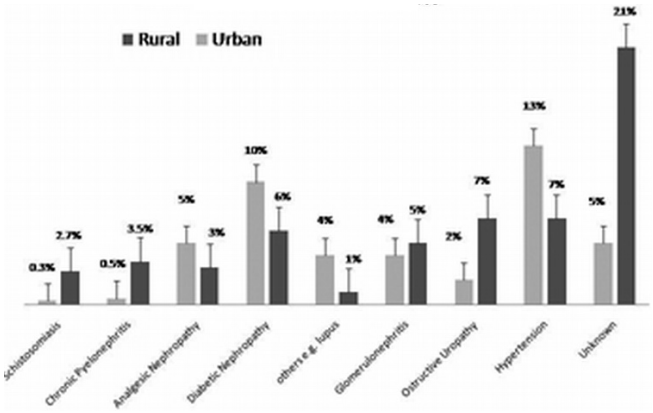
Cause	In El-Minia	In US*
Unknown	266 (26%)	4.2%
Hypertension	204 (20%)	27.5%
Chronic Glomerulonephritis	92 (9%)	2.9%
Obstructive uropathy	92 (9%)	1%
Analgesic Nephropathy	82 (8%)	0.2%
Schistosomiasis	31 (3%)	–
Chronic Pyelonephritis	41 (4%)	0.3%
Diabetic nephropathy	163 (16%)	44.7%
Others	49 (5%)	19.2
Total	1020 (100%)	536877 (100%)

*Prevalence of ESRD in US, Data from USRDS.org.



Causes of ESRD in El-Minia Governorate in comparison to US.

Results: The prevalence of ESRD was 392 per million population (PMP). RRT modality was HD in 1677 (97%), PD in 17 (1%) and Renal Tx in 35 (2%) patients. out of the total 1729 patients under RRT only1020 of them included in this study (59%). The mean age was 48±13 years, median 44 years, range (21 to 78 years). Gender distribution, males were almost as twice as females (62% Vs 38%), patients lived in the rural areas are more than those lived in urban areas (56% and 44% respectively).The etiology of ESRD is shown in the table. The death rate among them during this year 2008 was 194/1000.



Etiology of ESRD in rural and urban areas in El-Minia Governorate.

Conclusions: Epidemiology of ESRD in El-Minia Governorate has an annual increase of 25 new ESRD than year 2007 and is still different from epidemiology of ESRD in the US, thrifty gene theory may be involved as a cause for the different etiology of ESRD between El-Minia Governorate and the US, so region specific intervention is recommended to control the epidemic of ESRD in the world.

Su313 CLINICAL PROFILE AND OUTCOME OF END STAGE RENAL DISEASE PATIENTS PRESENTING TO TERTIARY CARE CENTER IN A DEVELOPING COUNTRY

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Introduction and Aims: To study demographic and clinical data of end stage renal disease (ESRD) patients presenting to maintenance haemodialysis (MHD) at a government funded tertiary care centre in a developing country.
Methods: A prospective analysis of all new ESRD patients attending to haemodialysis at our centre from 2004 to 2007 (three years) has been done.
Results: Results: There were 237 new ESRD patients, who required haemodialysis during three year period. There were 153 males (mean age: 44.85±12.99 yrs) and 84 females (mean age: 44.99±13.56 yrs) in this cohort. Diabetes mellitus (31.22%) was the most common cause of ESRD, followed by chronic glomerulonephritis (24.89%) and hypertension (15.61%). 65.40% patients had emergency haemodialysis, while the rest had planned initiation of maintenance haemodialysis. Most common indication of dialysis was uraemia (59.07%) followed by fluid overload (16.45%) and metabolic acidosis (5.90%). Most of the patients had hemoglobin less than 10g/dL (92.4%) at presentation. Only 10.1% of patients have received intravenous iron therapy and recombinant human erythropoietin (rhEPO) therapy before initiation of dialysis.
Arteriovenous fistula (AVF) was secured in 29.8% of patients at presentation. The number of patients with HBV and HCV infection at presentation were 6 (2.53%) and 8 (3.37%) respectively. One patient had both HCV and HBV infection at presentation. The number of patients who had sero-converted to HBV and HCV infection during dialysis were 6 (2.53%) and 19 (8.01%) after 3.66±2.51 months and 7±5 months respectively. Two (0.84%) patients had sero-converted to both HBV and HCV infection during dialysis. There was no correlation between blood transfusion or duration of haemodialysis and HBV or HCV seroconversion rates.
Primary failure of the AVF was the most common complication of vascular access. Catheter related infection was present in 13.55% of patients, who were on catheter. Methicillin resistant staphylococcus (25%) was the most common organism. Most common complication of haemodialysis was hypotension (29%) followed by vomiting (12%). Most common infection in dialysis patients was urinary tract infection (37.14%), followed by pneumonia (21.42%) and tuberculosis (21.42%).
Of the 237 patients who were initiated on haemodialysis in these three years, 103 (43.45%) were lost to follow up. Renal transplantation was opted by 9.7% patients and continuous ambulatory peritoneal dialysis (CAPD) in 20.25%. The rest (8.86%) continued on MHD. At our centre there were 42 (17.72%) deaths over three year period, out of which 36 (15.1%) were in first 90 days.
Conclusions: The information of the practice of haemodialysis, its population characteristics or outcomes in India was scant. As the data was from a government funded tertiary care centre, it reflected the true status of Indian ESRD patient.

Su313bis A SWEDISH COST-EFFECTIVENESS ANALYSIS OF RENVELA FOR THE TREATMENT OF HYPERPHOSPHATEMIA IN PATIENTS WITH PRE-DIALYSIS CHRONIC KIDNEY DISEASE

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Introduction and aims: To determine the cost-effectiveness of Renvela vs. standard care for the treatment of hyperphosphatemia in patients with pre-dialysis CKD from the social economic perspective in Sweden.
Methods: The analysis was based on a health state transition model that simulated the natural history of a cohort of pre-dialysis CKD patients, quantified the clinical and economic impact of treating hyperphosphatemia with Renvela vs. standard care and projected the incremental cost per life year (LY) gained and incremental cost per quality-adjusted life year (QALY) gained. All patients started the model in the 'Alive without Dialysis' health state with a defined set of characteristics (e.g. age, CKD stage, baseline serum phosphate) based on a clinical study of Renvela in pre-dialysis CKD patients (Ketteler 2008). Each model cycle, patients in the 'Alive without Dialysis' state could transition to 'Alive with Dialysis' or 'Dead'. Patients in the 'Alive with Dialysis' state could also transition to 'Dead'. Standard care was based on a natural history study of pre-dialysis CKD patients in Sweden (Evans 2005) and included calcium-based binders and no pharmacologic treatment. Probability for transitioning between health states in the standard care arm, up to 6 years, was based on data from Evans. The survival impact of treating all patients with Renvela was captured by applying hazard ratios for mortality among pre-dialysis patients (Kestenbaum 2005) to the survival curves from Evans. Based on Kestenbaum, each 0.323 mmol/l increase in serum phosphate in the model was associated with an estimated 33% increased risk of death. Outcomes beyond 6 years were extrapolated using a Weibull regression model. Utility weights and decrements, resource utilization and Swedish unit costs were derived from published literature and validated by expert opinion. The base case analysis was conducted for a 10-year time horizon and excluded costs associated with standard care and dialysis. Costs (expressed in 2009 Swedish kronor [SEK]; 1 SEK=0.0976 Euros) and outcomes beyond 1 year were discounted at 3% annually.
Results: Compared with standard care, Renvela resulted in greater LYs (5.80 per patient for Renvela vs. 4.58 for standard care), QALYs (4.31 per patient for Renvela vs. 3.45 for standard care) and incremental costs (398,854 SEK per patient for Renvela vs. 117,898 for standard care). From the social economic perspective, Renvela was associated with an incremental cost per LY gained of 230,280 SEK and an incremental cost per QALY gained of 324,188 SEK. The results were most sensitive to the perspective, overall survival, inclusion of dialysis costs, Renvela treatment duration, and utility estimates.
Conclusions: This analysis demonstrates that Renvela, a non-absorbed and calcium-free phosphate binder, represents a cost-effective alternative to standard care for the treatment of hyperphosphatemia in pre-dialysis CKD patients in Sweden.
Disclosure: Funding for this research was provided by Genzyme Corporation, Cambridge, MA, USA.

Diabetes – clinical studies

Su314 EXPOSURE-RESPONSE (ALBUMINURIA REDUCTION) RELATIONSHIP OF PARICALCITOL IN PATIENTS WITH DIABETIC NEPHROPATHY IN THE VITAL STUDY

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Introduction and Aims: Paricalcitol (Zemplar) is a selective vitamin D receptor activator approved for prevention and treatment of secondary hyperparathyroidism associated with chronic kidney disease. The VITAL study was designed to test the effectiveness of paricalcitol for the reduction of residual albuminuria in patients with type 2 diabetic nephropathy receiving stable ACEi or ARB treatment. The purpose of this analysis was to evaluate paricalcitol pharmacokinetics (PK) and the relationships between paricalcitol plasma concentration (exposure) and reduction in albuminruia (clinical response).

Methods: This multinational, randomized, placebo-controlled, double-blind study enrolled patients (N=281) with urinary albumin/creatinine ratio (UACR) 100 to 3000 mg/g. Patients received either placebo or paricalcitol (1 or 2 μg) once daily (QD) for 24 weeks. Dose reductions of study drug from QD to three times-a-week (TIW) were allowed based on safety. Blood samples for determination of paricalcitol plasma concentrations were collected at weeks 4, 8, 12, 16 and 20. Population PK and exposure-clinical response models were built using a non-linear mixed effects analyses (NONMEM software). The relationship between change from baseline UACR (primary efficacy variable) and paricalcitol concentrations was assessed using mixed effects models (variations of linear, exponential and Emax) that accounted for the paricalcitol concentration-time profile, time-course of placebo and paricalcitol effect.

Results: Concentration-UACR data from 273 patients were analyzed; 94 patients were on placebo; 76 patients were on 1 μg QD; 50 patients were on 2 μg QD; 13 patients converted from 1 μg QD to 1 μg TIW; 40 patients converted from 2 μg QD to 2 μg TIW. Mean observed change from baseline in UACR at week 24 for placebo, 1 and 2 μg QD doses were 5%, 7% and 19%, respectively.

Paricalcitol PK was characterized by a 1-compartment model with lag time and first order elimination; clearance was 2.2 L/h and volume of distribution was 43 L.

Exposure-response analyses were performed using individual observed concentration-UACR data over the entire study duration, thereby accounting for observed dose reductions, time-course of placebo and paricalcitol effect. A statistically significant linear relationship was identified between paricalcitol concentration and UACR response after accounting for differences in baseline between treatment groups ($p<0.001$). Exposure-response analyses predicted a 24% and 17% reduction in UACR for patients on 2 μg QD and 1 μg QD, and a 15% and 13% reduction in UACR for patients on 2 μg TIW and 1 μg TIW, respectively, compared to a 10% reduction for patients on placebo at 24 weeks, consistent with observed data.

Conclusions: In patients with diabetic nephropathy, paricalcitol at doses of 3 to 14 μg/week, when added to stable ACEi or ARB therapy, effectively lowers albuminuria by 13-24% based on exposure-response analyses. Exposure-response modeling is an effective method to validate dose ranging effects on primary outcomes in clinical trials and should be considered for validation purposes in early phases of drug development.

Disclosure: All authors are employees of Abbott Laboratories. This study was funded by Abbott.

Su315 BARDOXOLONE METHYL (BARD) IMPROVES KIDNEY FUNCTION IN PATIENTS WITH TYPE 2 DIABETES (T2D) AND CHRONIC KIDNEY DISEASE (CKD)

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Introduction and Aims: BARD inhibits NF-κB and induces Nrf2, activating >250 antioxidant genes to decrease reactive oxygen species (ROS) which are elevated in T2D and CKD. ROS increase NF-κB, angiotensin II and other inflammatory mediators which impair kidney function and glycemic control. A Phase 2a open-label study with 2 cohorts (Stratum 1 and 2) was performed to investigate the effects of daily oral BARD in patients (pts) with T2D and CKD (serum Cr: men 1.5–3.0; women 1.3–3.0 mg/dL). The primary endpoint was change in estimated glomerular filtration rate (eGFR; 4–variable MDRD). Additional measures of kidney function included BUN, serum phosphorus, uric acid, and magnesium.

Methods: Stratum 1: 60 pts were randomized to 25, 75, or 150mg BARD for 28 days; Stratum 2: 20 pts received 25mg for 28 days and then 75mg for 28 days. Demographics were similar in Stratum 1 and 2: age (62 & 64 yr, respectively), T2D duration (19,16 yr) and diabetes control (HbA1c 7.6%,7.8%) and extrarenal complication rate (42,50%), HTN (98%,100%), ACE &/or ARB use (78%,75%). At baseline eGFR (mL/min/1.73m^2) was 35.6 in Stratum 1 and 30.3 in Stratum 2.

Results: As reported previously, BARD produced significant dose- and time-dependent changes in eGFR; 89% of pts in each stratum exhibited an eGFR increase. The table below summarizes improvements observed in eGFR and other measures of kidney function. Importantly, changes in renally excreted solutes were significantly correlated with the observed eGFR changes, consistent with improvement in kidney function. Renal function improvements were of a similar magnitude in patients with Stage 3 and Stage 4 CKD. BARD was well-tolerated, with no treatment-related SAEs.

Table 1

	Stratum 1 (n=60)		Stratum 2 (n=18)	
	Change from Baseline	Correlation eGFR Change	Change from Baseline	Correlation eGFR Change
eGFR (mL/min/1.73m^2)	+6.73±0.82** (+18.88%)	N/A	+7.22±0.85** (+23.80%)	N/A
BUN (mg/dL)	-4.90±1.34** (-11.69%)	-0.46**	-4.94±1.60 (-13.16%)	-0.66*
Serum Phosphorus (mg/dL)	-0.28±0.08** (-7.16%)	-0.30†	-0.07±0.10 (-1.88%)	-0.42
Uric Acid (mg/dL)	-1.03±0.17** (-12.41%)	-0.27†	-0.92±0.21† (-12.59%)	-0.71*
Magnesium (mg/dL)	-0.19±0.02** (-8.92%)	-0.39*	-0.22±0.06** (-10.48%)	-0.44

†P≤0.05, *P≤0.01, **P≤0.001.

Conclusions: This study demonstrates clinically and statistically significant improvements in eGFR that correlate with improvements in multiple independent measures of kidney function. These effects, observed in patients with Stages 3 & 4 CKD, persisted without plateau at 56 days of BARD treatment. A 12-month randomized, blinded, placebo-controlled Phase 2b study is underway.

Su316 RENAL ARTERY STENOSIS IS PREDICTIVE OF CARDIOVASCULAR EVENTS IN PATIENTS AFFECTED BY ISCHEMIC HEART DISEASE

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Introduction and Aims: The presence of atherosclerotic renal artery stenosis (ARAS), even when incidentally discovered, is independently associated with a higher cardiovascular (CV) risk. This study was planned to know the impact of ARAS on CV morbility and mortality in patients (pts) with suspected coronary artery disease (CAD).

Methods: A cohort of 1298 pts affected by ischemic heart disease, consecutively enrolled from 2006 for one year, undergoing coronary and renal arteriography, were studied at baseline and followed up for 2-5 years. Major CV events (MACCE), defined as myocardial infarction, stroke, death from CV causes, coronary artery revascularization, pulmonary oedema, were collected. Cox regression analysis was used to investigate the effect of several risk factors on time to the first MACCE.

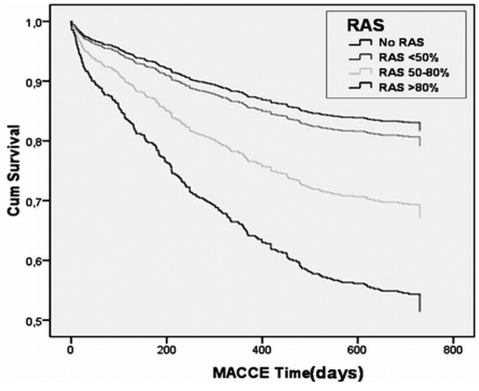
Results: The age of the study cohort was 64±10 yrs (M 927 F 371); sCr 1.00±0.29 mg/dl, eGFR (MDRD) 81±23 ml/min; Diabetes 36%, Hypertension 87%, Hyperlipidemia 75%. 70/1298 pts (5.4%) had ARAS≥50%. The presence of Peripheral Vascular Disease, eGFR (< 67 ml/min/1.73 m^2), age (< 66 yrs), hyperlipidemia, CAD severity (≥ 2 coronary vessel), and pulse pressure were identified as independent clinical predictors of RAS≥50%

[ROC AUC 0.79 (95% CI: 0.73-0.85, P<0.001]. FoIlow up data at 2 years were available in 1087 pts (ARAS≥50% in 58 pts). At least one MACCE was present respectively in 152 out of 809 pts (19%) without ARAS, in 57 out of 220 pts (26%) with ARAS <50%, in 21 out of 52 pts (40%) with ARAS ≥50-80% and in 4 out of 6 pts (67%) with ARAS > 80%. In a multivariate Cox regression model, including the variables identified by the univariate Cox analysis (i.e.P<0.10), only age, the presence of ARAS, and the severity of CAD significantly predicted the time to the first MACCE, while diabetes, eGFR, hyperlipidemia or blood pressure control did not.

Cox regression analysis

Variables	Pts (n)	HR	CIs (95%)	P
Age (1yr)	1087	1.01	0.99-1.03	0.066
RAS				0.005
No RAS	809	1		
RAS <50%	220	1.16	0.84-1.59	
RAS ≥50-80%	52	1.97	1.24-3.13	
RAS >80%	6	3.29	1.20-9	
CAD				<0.001
No CAD	59	1		
no sign CAD	188	2.71	0.82-8.96	
1-vessel CAD	299	3.06	0.95-9.83	
2-vessel CAD	264	4.74	1.49-15.15	
3-vessel CAD	190	6.29	1.96-20.16	
LMCA	87	7.21	2.19-24	

Further, the risk of MACCE increased with the severity of ARAS.



Conclusions: The data showed that the prevalence of ARAS≥50% is quite low in a cohort of pts affected by CAD. However, the presence of ARAS is significantly related to the risk of MACCE, thus supporting that the early diagnosis is useful to better chararacterize the CV risk profile of pts affected by ischemic heart disease.

Su317 PROXIMAL TUBULE DYSFUNCTION AND NOT ENDOTHELIAL DYSFUNCTION EXPLAINS EARLY DIABETIC NEPHROPATHY IN PATIENTS WITH TYPE 2 DIABETES MELLITUS: A CROSS-SECTIONAL STUDY

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Introduction and Aims: Currently, there is debate as to whether early diabetic nephropathy (DN) in type 2 diabetes mellitus (DM) may be attributed to the glomerulus or to the proximal tubule (PT). The aim of the study was to clarify this hypothesis in normoalbuminuric patients (p) with type 2 DM by evaluating PT dysfunction and endothelial dysfunction in two vascular beds, the kidney and the brain, both affected by microangiopathic modifications.

Methods: A total of 68 normoalbuminuric type 2 DM p with no microangiopathic complications and no history of cerebrovascular disease were enrolled in a cross-sectional study [25 males, 43 females, mean age 58.10±7.12 y]. All p were assessed concerning urinary albumin: creatinine ratio (UACR), urinary α1-microglobulin, plasma asymmetric dymethylarginine (ADMA), serum creatinine (SC), GFR, C-reactive protein (CRP), fibrinogen (F), HbA1c, cholesterol, triglycerides, haemoglobin; pulsatility index-PI, resistance index-RI in the internal carotid artery-ICA and middle cerebral artery-MCA, intima-media thickness-IMT in the common carotid artery-CCA; cerebrovascular reactivity (CVR) was evaluated in the MCA through the breath-holding test (BHT) by utilizing transcranial Doppler ultrasound (% increase in mean flow velocity-MFV after hypercapnia as compared to basal MFV).

Results: There were increased levels of plasma ADMA in 12p (17.5%) and urinary α1-microglobulin in 24p (35.7%). The cerebral haemodynamics indices correlated with plasma ADMA (R2=0.57; P<0.01), CRP (R2= 0.78; P<0.0001), F (R2=0.85; P<0.0001), duration of DM (R2=0.53; P< 0.01), HbA1c (R2=0.57; P<0.01), urinary α1-microglobulin (R2=0.723; P< 0.0001). There was no correlation between plasma ADMA and UACR (R2= 0.019; P=0.261), urinary α1-microglobulin (R2=0.004; P=0.59), fibrinogen (R2=0.008; P=0.53), CRP (R2=0.006; P=0.52), GFR (R2=0.008; P=0.46).

Conclusions: In p with type 2 DM the increase in urinary alpha1-microglobulin occurs before the stage of albuminuria, thus leading to the assumption that early DN is related to PT dysfunction and not to endothelial dysfunction. In these p endothelial dysfunction plays a pivotal role in the brain vasculature, while its involvement in the development of early DN remains questionable.

Su318 ROLE OF INSULIN RESISTANCE IN THE OUTCOME OF ADVANCED CHRONIC KIDNEY DISEASE PATIENTS

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Introduction and Aims: Insulin resistance (IR) increases significantly the risk for cardiovascular disease (CV) in the general population. IR is a common metabolic disorder in patients with chronic kidney disease (CKD). However, the influence of IR on the evolution of CKD patients has scarcely been studied.

Methods: This study aims to determine whether IR relates with the progression of CKD, the development of new CV events, or all-cause mortality of non-diabetic patients with CKD stage 4 or 5 not yet on dialysis. The study group consisted of 365 non-diabetic patients (63±16 year, 169 females) with GFR < 30 ml/min. The degree of IR was estimated by the Homeostasis Model Assessment parameter (HOMA) calculated according to the formula: plasma fasting insulin (mU/ml) x plasma fasting glucose (mmol/l)/22.5. The outcome measures were: progression of CKD (composite of initiation of dialysis or doubling of baseline serum creatinine level), new cardiovascular events, and all-cause mortality. Unadjusted and multivariable-adjusted relative risks were calculated for HOMA either as a continuous or qualitative variable (tertiles), using Cox proportional hazards models.

Results: Mean HOMA value (±SD) was 4.28±2.07. HOMA values were correlated significantly with body mass index (beta = 0.36; p<0.0001), plasma triglycerides (beta = 0.21; p<0.0001), plasma albumin (beta = 0.14; p=0.007), and serum phosphate (beta = 0.12; p=0.031). Progression of CKD was observed in 234 patients (64%) with a median pre-dialysis follow-up of 542 days. Patients with HOMA values within the lower tertile (<3.13) showed a slower progression of CKD than that of the rest of study patients (log rank 4.16, p<0.05). In adjusted models for age, sex, baseline GFR, body mass index, and proteinuria, HOMA values within the lower tertile entered as an independent variable in the best predictive equation for progression of CKD (HR 0.71, p<0.012). Fifty-one patients developed a new CV event and 103 patients died during the study period (median follow-up of 1103 days). HOMA did not relate to the development of new CV events or all-cause mortality in unadjusted or adjusted models for age, sex, comorbid index, plasma albumin, and C-reactive protein.

Conclusions: In conclusion, progression of renal disease was slower in those non-diabetic CKD patients with low HOMA values; however, HOMA values did not relate to the development of new CV events or all-cause mortality.

Su319 POSSIBILITIES OF DECELERATION OF DIABETIC NEPHROPATHY PROGRESS: RESULTS OF 12-MONTHLY SUPERVISION

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Introduction and Aims: Diabetic nephropathy is one of the major "microvascular" complications of diabetes and is associated with increased cardiovascular mortality. It is the most common cause of end-stage renal disease requiring dialysis in the world. That why to slacken speed of the progression of diabetic nephropathy and prolongation of predialysis period are important. The aim of the current study was to estimate influence of the combined therapy by angiotensin-converting enzyme inhibitors (ACEI) or angiotensin receptor blockers (ARB) and sulodexide on rates of diabetic nephropathy progressing in comparison with therapy by only ACEI or ARB.
Methods: We investigated 90 patients with stage 2 chronic kidney disease (CKD) caused type 2 diabetes. All patients had arterial hypertension, which corrected with antihypertensive therapy. A systolic blood pressure was 134±12,1 mm Hg, a diastolic blood pressure was 77±9,2 mm Hg. All patients were divided on two groups of method randomization. I group (45 patients) – they received sulodexide and ACEI or ARB. II group (45 patients) – they received only ACEI or ARB. Sulodexid appointed in a doze 600 LU intramuscularly during 10 day and in a doze 500 LU per os during three months. Data are presented as mean±standard deviation at normal distribution or median (25:50 percentile) at not normal distribution.
Results: Results of treatment (changes of urinary protein excretion (proteinuria), glomerular filtration rate (GFR)) were analyzed in 12 months. For a year the decline of the level of urinary protein excretion took place in both groups: in 1 group – from a 1,6 (0,9:2,2) g per 24 hours to 0,25 (0,1:0,9) g per 24 hours ($p<0,001$); in 2 group – from a 1,5 (0,7:2,1) g per 24 hours to 0,65 (0,3:1,7) g per 24 hours ($p=0,021$). Difference in the values of proteinuria in the investigated groups in a year was significant ($p=0,021$). At the analysis of index of decrease of urinary protein excretion for a year next results are got: in a 1 group decrease presented 1,0 (0,65:1,22) g per 24 hours for a year, and in 2 group – 0,4 (0,2:0,6) g per 24 hours for a year ($p=0,001$). The decline of GFR for a year presented in a 1 group from 74,4±6,9 mL/min/1,73m^2 to 74,3±8,2 mL/min/1,73m^2 ($p=0,9$); in 2 groups – from 71,6±5,7 mL/min/1,73m^2 to 67,3±6,4 mL/min/1,73m^2 ($p<0,05$). A difference in the values of indexes of GFR in the investigated groups through one year was significant ($p=0,004$). At the analysis of index of decrease GFR for a year next results are got: in a 1 group decrease presented 0,35 (0,2:2,9) mL/min/1,73m^2 for a year, and in 2 group – 3 (3:4) mL/min/1,73m^2 for a year ($p<0,001$).
Conclusions: Our data indicate therapy with sulodexide and ACEI or ARB in combination is more effective that therapy only with ACEI or ARB for decline of the level of urinary protein excretion and slacken speed of the progression of diabetic nephropathy.

Su320 EFFECT OF CHROMIUM SUPPLEMENTATION ON MICROALBUMINURIA IN TYPE 2 DIABETES

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Introduction and Aims: Introduction: Microalbuminuria can reflect the progress of microvascular complications and may be predictive of macrovascular disease in type 2 diabetes. Many studies have shown the changes of chromium in Type 2 diabetic patients, but its role in the development of diabetic nephropathy is insufficiently studied. Aim: This study evaluates effect of chromium supplementation on renal function in type 2 diabetes.
Methods: The research encompasses 36 patients with type 2 diabetes mellitus DM, divided into groups with bad and very good glycemic control and duration of diabetes mellitus over 5 years. In course of 60 days the patients receive a daily addition of 30 μg chromium picolinate. Prandial glycemia, glycated hemoglobin A_1c, immunoreactive insulin, serum chromium, creatinine clearance and urinary microalbumin levels are measured.
Results: The addition of chromium increase it's serum levels, leads to significant reduction of hyperinsulinemia, prandial glycemia and glycated hemoglobin A_1c. The microalbuminuria in patients significantly decreased from 265,4±2,11 mg/d to 149,5±3,25 mg/d. This reduction is most pronounced in group with good glycemic control.
Conclusions: Supplementation with chromium picolinate benefits glycemic control, enhances insulin sensitivity and reduces urinary albumin excretion in type 2 diabetic patients. It improves carbohydrate metabolism and renal function in type 2 diabetes. These observations suggest that chromium supplementation may retard deterioration of glomerular function in type 2 diabetes mellitus probably through recovering renal chromium concentration.

Su321 PREVENTION OF MICROALBUMINURIA: PREDICTORS FOR A GOOD RESPONSE TO OLMESARTAN TREATMENT (ROADMAP TRIAL)

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Introduction and Aims: Microalbuminuria (MAU) is an early sign of diabetic nephropathy and increased cardiovascular risk. We investigated whether early treatment with an angiotensin receptor blocker (ARB) in diabetic subjects with normal albumin excretion delays the occurrence of MAU and analysed subgroups that would benefit most from treatment.
Methods: We studied 4,447 subjects with type 2 diabetes and at least one additional cardiovascular risk factor in a randomized, double-blind, multicentre, controlled, and event-driven (MAU) trial. They received either 40 mg olmesartan medoxomil (OM) or placebo (Pb) od. for a median duration of 3.2 years. In both groups, additional antihypertensive treatment (except ACE inhibitors or ARBs) was used to reach the target BP of <130/80 mmHg.
Results: Baseline UACR, eGFR, blood pressure and cardiovascular disease (CVD) risk profiles were comparable in both groups. The SBP/DBP control was excellent during the entire study with an average of 125.7/74.3 mmHg in the OM group and 128.7/76.2 mmHg in the Pb group. During the double blind period, 178 (8.2%) subjects in the OM group and 210 (9.8%) subjects in the Pb group developed MAU (time-to-onset: HR: 0.770; 95.1% CI: 0.630 to 0.941, p: 0.0104). To identify factors influencing the response to OM treatment, explorative post-hoc subgroup analysis using the corresponding median at baseline as a cut-off were performed. This analysis revealed that the treatment effect on time to onset of MAU was better in subjects with a SBP >135 mmHg than with SBP values ≤135 mmHg. The treatment effect in subjects with DBP < or > 80.3 mmHg was comparable. A baseline HbA1c ≤7.3% and an eGFR ≤83.79 were predictors for a better response to OM treatment. Furthermore, less than 5% of patients with a baseline UACR ≤4 mg/g developed MAU during the study and the rate was similar between the OM and Pb treated patients. In contrast, 13.9% in the OM group and 18.1% in the placebo group with a baseline UACR >4 mg/g developed MAU (time-to-onset: HR: 0.75, 95% CI: 0.60 to 0.96, p-value= 0.02).
Conclusions: In subjects with type 2 diabetes olmesartan showed a significant 23% risk reduction regarding time to onset of microalbuminuria. Patients with a baseline eGFR ≤83.79, or an UACR >4mg/g benefit most from olmesartan treatment.
ClinicalTrials.gov ID no.: NCT00185159
Disclosure: All authors received honoraria from Daiichi-Sankyo.

Su322 IMPACT OF OLMESARTAN WITH OR WITHOUT ACE INHIBITOR ON RENAL AND CARDIOVASCULAR PROTECTION IN TYPE 2 DIABETES WITH OVERT PROTEINURIA

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Introduction and Aims: The cardiorenal effects of ACEI, ARB or the combination in type 2 diabetes remain controversial and need further evaluation.

Methods: In a randomized, double-blind, placebo-controlled study for 3.2 years, we examined the effects of olmesartan, an ARB, on primary outcome of doubling of serum creatinine (SCr), endstage renal disease (ESRD), and death in type 2 diabetic patients with overt nephropathy. Secondary outcome included cardiovascular outcomes, changes in renal function and proteinuria. The effects of olmesartan on the outcomes were analyzed separately with or without ACEI.
Results: 282 received olmesartan and 284 received placebo in addition to conventional antihypertensive therapy,73.1% patients treated with ACEI. In patients not taking ACEI, time-averaged difference of BP was lowered by olmesartan compared to placebo (SBP; 6.4mmHg, DBP; 4.0mmHg, $p<0.001$). In patients taking ACEI, BP was not significantly different between the 2 groups during study (SBP; 1.0mmHg, DBP 0.7mmHg). Proteiniria was significantly decreased by treatment with or without olmesartan. The hazard ratio (HR) for primary renal composite outcome in the olmesartan group was 0.97 (p=0.791) and the HR was 1.02 (p=0.852) after adjustment for BP. Compared to placebo, the HR for primary outcome in the olmesartan group were 0.84 (p=0.436) in patients not receiving ACEI (n=152) and 1.02 (p=0.891) in those receiving ACEI (n=414). The composite secondary cardiovascular outcome occurred in 40 olmesartan-treated patients (14.2%) and 53 placebo-treated patients (18.7%) with a HR of 0.73 (p=0.126). HR was decreased to 0.64 (p=0.039) after adjusting for unbalanced distribution of history of CVD at baseline. The trend of risk reduction in cardiovascular outcome was not influenced by co-treatment with ACEI. The HR in olmesartan group without ACEI and with ACEI were 0.56 (p=0.174) and 0.77 (p=0.290), respectively. The median yearly rate of change of 1/SCr was -0.071 (interquartile range (IQR); -0.148 to -0.032)dL/mg/year in the olmesartan and -0.089 (IQR; -0.151 to -0.044)dL/mg/year in the placebo group (p=0.044). In patients not treated with ACEI, the rate of change of 1/SCr was -0.075dL/mg/year (olmesartan) and -0.121 dL/mg/year (placebo). In patients treated with ACEI, the rate of changes of 1/SCr were -0.070dL/mg/year (olmesartan) and -0.080dL/mg/year (placebo).In total, 73 (25.9%) of olmesartan-treated patients and 64 (22.5%) of placebo-treated patients were discontinued before study completion due to adverse events.
Conclusions: In type 2 diabetic patients with nephropathy, olmesartan reduced proteinuria, slowed decline of renal function and improved cardiovascular outcome, but did not further improve renal outcomes. The renal and cardiovascular outcomes by olmesartan were not affected by the co-treatment with ACEI substantially if blood pressure was adjusted.
Disclosure: Dr Imai has received consulting and lecture fees and grant support from Daiichi-Sankyo, Kyowa-Hakko-Kirin, Banyu, Kaken, Chugai, Takeda, Shionogi, Dainippon-Sumitomo, Astelas, Novartis. Dr Chan has received consultancy, lecture fees and grant support from Daiichi-Sankyo, MSD, Astra Zeneca, Bristol Myers Squibb, GSK, Pfizer and Bayer. Dr Ito has received consultancy, lecture fee, and research grant from Daiichi-Sankyo, Novartis, Astelas, Banyu and Pfizer. Dr Haneda received research grant from Daiichi-Sankyo. Dr Makino received consultancy, lecture fee and research grant from Daiichi-Sankyo, Astelas, Banyu, Kyowa-Hakko-Kirin, Taked, Chugai, Novartis, Boehringer Ingelheim, Dainippon-Sumitomo, and Kowa.

Su323 COMBINED THERAPY WITH RENIN ANGIOTENSIN SYSTEM AND CALCIUM CHANNEL BLOCKERS IN TYPE-2 DIABETIC HYPERTENSIVE PATIENTS WITH PROTEINURIA: EFFECTS ON TWEAK, PTX3 AND FLOW MEDIATED DILATATION

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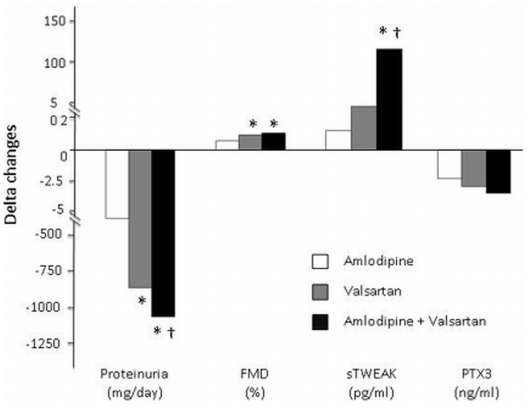
Introduction and Aims: soluble TWEAK (sTWEAK) and long pentraxin-3 (PTX3) concentrations have been associated with endothelial function in patients with chronic kidney disease. We hereby tested the hypothesis that the improvement in endothelial function after initiation of AII receptor blocker (valsartan), calcium channel blocker (amlodipine) therapy or a combination of both is directly linked to the normalization of these molecules.
Methods: Patients with history of coronary artery disease, smokers and those taking statins or renin-angiotensin blockers were excluded because of the effect of these factors on ED. 108 diabetic CKD stage I patients with hypertension (56% males, 46.7±5.3 years) were allocated to a 12-week intervention with amlodipine (10 mg/d), valsartan (160 mg/d) or their combination. Serum levels of sTWEAK, PTX3 and flow-mediated dilatation (FMD) were studied during the interventions.
Results: All treatment strategies effectively increased FMD and reduced proteinuria, confirming a more prone reduction with the combined therapy. These improvements were also followed by significant PTX3 reductions. Both valsartan alone and in combination with amlodipine achieved significant incremental raises in sTWEAK plasma levels.



More importantly, the changes observed in sTWEAK (b=0.25, P=0.006) or PTX3 (b=-0.24, P=0.007) plasma levels were independently associated with the improvement in ultrasonographically measured FMD.
Conclusions: Our study shows that treatment with amlodipine, valsartan or their combination improves FMD and normalizes proteinuria, PTX3 and sTWEAK in diabetic CKD stage I patients with hypertension. In addition, the improvement in FMD was associated with both PTX3 and sTWEAK normalization. We therefore identify two surrogate biomarkers of endothelial health with potential as therapeutic targets (NCT: The study was registered in clinicaltrials.gov as NCT00921570)

Su324 A NOVEL BIOMARKER PANEL FOR DIABETIC NEPHROPATHY DIAGNOSTICS AND STAGING

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Introduction and Aims: Diabetic nephropathy (DN) is the leading cause of kidney failure, accounting for more than 40% of new end-stage renal disease (ESRD) incidents. There is an increasing need to effectively monitor renal dysfunction in diabetic patients to pinpoint opportune time to initiate therapy for potential reversal or slow-down of disease. The aim is to develop a non-invasive diagnosis method that would give a good predictive value of the presence of DN in diabetic patient. Such test might be used to estimate the extent of DN as an alternative to renal biopsy.
Methods: Serum and urine samples were collected from healthy individuals and diabetic patients with and without different DN stages at participating medical centers of the Taiwan Renal Biomarker Consortium. Using differential proteomics strategies, a series of urinary and serum biomarkers were discovered which can clearly distinguish diabetic individuals with and without DN and can assign individuals to specific stages of DN. Five of these markers have been validated with ELISA in a large sample set. In order to improve the sensitivity and specificity with combination markers,

we have integrated the above protein markers with other clinical parameters to develop diagnostic algorithms for scoring DN.

Results: The overall diagnostic performance was evaluated by area under curve (AUROC) based on a combination of 5 protein markers. After expanded verification in 300+ subjects with varying degrees of DN, these biomarkers in their best combination can properly identify individuals at early stage nephropathy with high sensitivity and high specificity. The diagnostic accuracy using AUROCs in predicting differential stages of DN, Stage 2, Stage 3, Stage 4 and Stage 5, in our patient population is 0.94, 0.98, 0.98 and 0.91, respectively. More interestingly, the Score based on combination markers linearly correlates with disease stages and can be further developed for staging application.

Conclusions: In summary, we have successfully developed diagnostic algorithms with urinary and serum markers, which have been validated in 300+ samples and demonstrated to be applicable to DN staging. These markers together constitute a method to minimal-invasively diagnosis diabetic nephropathy and monitor disease progression.

Su325 VITAMIN D DEFICIENCY IS ASSOCIATED WITH SUDDEN CARDIAC DEATH, COMBINED CARDIOVASCULAR EVENTS AND MORTALITY IN DIABETIC HEMODIALYSIS PATIENTS

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Introduction and Aims: Dialysis patients experience an excess mortality, predominantly of sudden cardiac death (SCD). Accumulating evidence suggests a role of vitamin D for myocardial and overall health. This study investigated the impact of vitamin D status on cardiovascular outcomes and fatal infections in haemodialysis patients.

Methods: 25-hydroxyvitamin D (25[OH]D) was measured in 1109 diabetic haemodialysis patients who participated in the German Diabetes and Dialysis Study (4D Study) and were followed-up for a median of 4 years. By Cox regression analyses, we determined hazard ratios (HR) for pre-specified, adjudicated endpoints according to baseline 25(OH)D levels: SCD (n=146), myocardial infarction (MI, n=174), stroke (n=89), cardiovascular events (CVE; n=414), death due to heart failure (n=37), fatal infection (n=111) and all-cause mortality (n=545).

Results: Patients had a mean age of 66±8 years (54% male), and mean 25(OH)D of 18.0±9.8 ng/ml. Patients with severe vitamin D deficiency (25[OH]D $\leq$ 10ng/ml) had a 3-fold higher risk of SCD compared to those with sufficient 25(OH)D levels >30ng/ml (HR 3.0; 95% confidence interval 1.4-6.4). Furthermore, CVE and all-cause mortality were strongly increased (HR 1.8, 95% CI 1.2-2.7, and HR 1.7, 95% CI 1.2-2.5, respectively), all persisting in multivariate models. There was a trend for higher risks of stroke and fatal infection, while MI and deaths due to heart failure were not significantly affected.

Conclusions: Severe vitamin D deficiency was strongly associated with SCD, CVE and mortality, and by trend with stroke and fatal infection in diabetic haemodialysis patients. Whether vitamin D supplementation decreases adverse outcomes, requires further evaluation.

Su326 EVALUATION OF RENAL FUNCTION BY MEANS OF DIFFUSION TENSOR MR IMAGING (DTI) AND ITS RELATIONSHIP WITH CKD STAGES

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Introduction and Aims: Diffusion Tensor MR Imaging (DTI) is an MR imaging technique used to show molecular diffusion. The apparent diffusion coefficient (ADC) combines the effects of capillary perfusion and water diffusion in the extracellular space. DTI provides information on perfusion and diffusion simultaneously. Fractional anisotropy (FA) is a quantitative value that provides information on direction of water molecules flow within a tissue. Aim of this study was to evaluate feasibility of DTI in assessment of renal function in two groups of patients-group A (CKD 1-2) and group B (CKD 3-4).

Methods: Ten CKD 1-2 (3M, 7F, age 50±18 ys) and 10 CKD 3-4 (4M, 6F, age 58±10 ys) patients (pts) were enrolled in the study. The underlying nephropathies were: GROUP A: chronic pielonephrites 30%, diabetic nephropathy 20%, nephroangiosclerosis 20%, medullary kidney cystic disease 20%, glomerulonephrites 10%; GROUP B: nephroangiosclerosis 30%, glomerulonephrites 30%, chronic pielonephrites 20%, diabetic nephropathy 10%. All pts underwent morphological MR evaluation of kidneys with transverse T1 and T2 weighted acquisition, followed by transverse fat-saturated echo-planar DTI during breath-holding (with a *b*-value 500 mm^2/sec). ADC and FA in cortex and medulla were evaluated. Cockcroft and Gault formula was used for glomerular filtration rate (GFR) assessment. T-test was performed for statistical analysis.

Results: In group A, GFR was 76±12 ml/min, while in group B was 33±10 ml/min. In group A, ADC was higher in cortex than in medulla (2.55±0.09 and 2.10±0.08×10^{-3} mm^2/sec), while FA was higher in medulla than in cortex (0.195±0.03 and 0.280±0.04 mm^2/sec). In group B, ADC and FA were distributed as in group B (ADC 2.0±0.04 and 1.80±0.05 mm^2/sec; FA 0.130±0.03 and 0.160±0.04), but with statistically significant lower values ($p<0.05$). Our finding was the result of the water molecules flow, that in medulla has a radial direction due to the presence of tubular structure.

Table 1. Results

Group	N	ADC (×10^3 mm^2/s		FA	
		Cortex	Medulla	Cortex	Medulla
A	10	2.55±0.09	2.10±0.08	0.195±0.03	0.280±0.04
B	10	2.0±0.04	1.80±0.05	0.130±0.03	0.160±0.04

Data epressed as mean ± SD.

Conclusions: DTI of kidneys is feasible with excellent cortex-medulla differentiation. According to literature data, we found lower ADC values in patients with moderate-severe renal failure, than in pts with mild degree of renal impairment. In group B, the reduction in FA is more significant than the reduction in ADC, and this is particularly evident in medulla. Furthermore, FA may play an important role in detecting early renal damage, even before the onset of renal function impairment. More data are needed to confirm our preliminary results.

Su327 URINARY B2-MICROGLOBULIN EXCRETION REFLECTS THE SEVERITY OF RENAL DAMAGE IN TYPE 2 DIABETICS AND MAY HELP TO IDENTIFY DIABETIC GLOMERULOSCLEROSIS FROM NON-DIABETIC NEPHROPATHIES

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Introduction and Aims: About one half of Type 2 diabetic patients with renal insufficiency have kidney lesions other than classical diabetic glomerulosclerosis (DN) at renal biopsy, and this possibly implies the adoption of different therapeutic approaches. The increasing incidence of diabetes and its complications heralds a further increase of ESRD patients over the next few years, but the costs and invasiveness of renal biopsy ask for the development of new strategies able to reliably identify DN. We previously demonstrated the presence of a urinary protein pattern able to differentiate DN from non-diabetic chronic kidney disease. Among the mass peaks differently expressed in DN, we selected a 11700 m/z protein, previously identified as urinary β2-microglobulin. We aimed to ascertain

whether urine β2-microglobulin levels would help identify DN and would correlate with the severity of renal damage.
Methods: Urine samples collected from 169 patients [20 healthy subjects (HS), 20 normoalbuminuric (NAD) and 18 microalbuminuric (MICRO) diabetics and 111 patients with biopsy-proven nephropathy (54 DN, 20 IgA Nephropathy (IgAN), 24 Membranous Nephropathy (MN) and 13 benign nephroangiosclerosis (NASB)] were analysed by CM10 proteinChip array and SELDI-TOF/MS. The mass peak of ∼11.700 m/z was highlighted into all the mass spectra and a comparative analysis among HS, NAD, MICRO and DN and between DN and non-diabetic nephropathies was performed. Further, urine concentration of β2-microglobulin was measured by ELISA in a proportion of NAD, MICRO and DN patients. Finally, we sought to correlate β2-microglobulin excretion with the histological score recorded at biopsy examination.
Results: SELDI analysis revealed a significant increase of β2-microglobulin excretion in diabetic patients with biopsy-proven DN and renal insufficiency. In contrast, we were unable to find any difference between HS and diabetic patients without renal insufficiency, regardless of their albumin excretion rate. Urine β2-microglobulin evaluation by ELISA strictly confirmed the above findings, thus supporting the semi-quantitative value of the former approach. The median intensity of β2-microglobulin peaks turned out to be significantly higher in DN than in all non-diabetic nephropathies, but in NASB. Finally, higher β2-microglobulin levels were significantly associated with a higher global sclerosis score, and were independent from albumin excretion rate.
Conclusions: Urinary β2-microglobulin excretion seemingly correlates with the severity of renal damage in Type 2 diabetics and is independent from the rate of albumin excretion. Then, it would distinguish DN from MN and IgAN, but not from NASB.

Su328 FACTORS OF RENAL INTERSTITIAL FIBROSIS IN TYPE 2 DIABETICS WITH RENAL ARTERY STENOSIS AND IN TYPE 1 AND 2 DIABETICS WITH DIABETIC NEPROPATHY

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Introduction and Aims: to estimate renal interstitial fibrosis and endothelial dysfunctions factors in type 2 diabetics patients (T2DP) with renal artery stenosis or diabetic nephropathy (DN) and in type 1 diabetics with DN.
Methods: We studied 33 T2DP without renal pathology (I group), 33 T2DP with renal artery stenosis (II group), 24 T2DP with DN (III group) and 30 diabetic type 1 patients with DN (IV group). Patients with T2DP were invited to undergo multispiral computer tomography or selective angiography of renal arteries to define the presence of renal artery stenosis (renal artery stenosis more than 60%). Transforming growth factor (TGF-β 1), monocyte chemotactic peptide-1 (MCP-1), regulated on activation normal T-cell expressed and secreted (RANTES), matrix metalloproteinase 9 (MMP-9),vascular endothelial growth factor (VEGF), inhibitor activator of plasminogen (IAP-1), asymmetric dimethylarginine (ADMA), factor Willebrand (FW), homocystein (HCYST) were measured in blood. Glomerular filtration rate (GFR) was calculated by the MDRD equation.
Results: Studied parameters in groups are shown in Table 1. In groups with renal pathology TGF-β 1, MCP-1, RANTES, IL-6, MMP-9, HCYST, were negative correlated with GFR
Conclusions: DN in type 1 and 2 patients and renal artery stenosis in T2DP were associated with significant increase of mediators of fibrosis and endothelial dysfunctions factors which is known to have a predictive role in the development of the tubulointerstitial expansion and reduction of renal function.

Table 1

Parameter	I group (n=33)	II group (n=33)	III group (n=24)	IV group
TGF-β 1 ng/ml	40,6	121***	80,6**	65***
	[8,6; 81]	[11,9; 78]	[31,7; 128]	[18,3; 104]***
MMP-9 ng/ml	123	141***	258***	269***
	[88,5;149]	[102; 176]	[156; 319]	[160;362]
MCP-1 pg/ml	127	198*	282	268***
	[60;175]	[59; 262]	[220; 346]	[169;345]
RANTES pg/ml	21972	22896	28316*	39509**
	[8747; 32747]	[10494; 27193]	[14604; 35758]	[17882; 72576]**
HCYST mkmol/L	9,8	13,5*	73,8	13,5
	[7,8; 11,7]	[8,9; 14,3]	[8,3; 75,2]	[8,9;14,3]
FW UE/ml	0,74	0,9***	10,1*	0,76***
	[0,191;1,9]	[0,79; 1,14]	[0,2; 1,3]	[0,13;0,85]
IAP-1 ng/ml	40,8	61,9*	48,2	58,9**
	[13,9; 55]	[31,7; 81]	[17;69]	[25; 87]
ADMA mkmol/l	1,42	14,1**	1,7	–
	[0,02; 1,46]	[0,02; 1,4]	[0,4; 1,6]	
VEGF pg/ml	182	284**	203	184
	[49; 263]	[140; 386]	[61,9; 223]	[81; 259]

Results are Me [25%; 75%] *p<0,05, **p<0,01, ***p<0,0001 vs group I.

Su329 THE INCREASED RISK OF POST TRANSPLANT DIABETES MELLITUS IN PERITONEAL DIALYSIS TREATED KIDNEY ALLOGRAFT RECIPIENTS

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Introduction and Aims: The post transplant diabetes mellitus (PTDM) belongs to the common metabolic complications in the kidney allograft recipients appearing in 4–30% patients. The occurrence of PTDM contributes significantly to the elevated cardiovascular morbidity after kidney transplantation and increased risk of chronic transplant dysfunction. Pathogenesis of PTDM embraces both impairment of insulin secretion and peripheral resistance. The aim of the present investigation was the complex analysis of the factors influencing the PTDM development. Under particular consideration was the modality of dialysis treatment – peritoneal dialysis (PD) versus hemodialysis (HD).
Methods: A retrospective cohort of 377 consecutive out-patients who underwent renal transplantation in our institution between I. 2003 and XII. 2005 were analyzed. PTDM was diagnosed according to current American Diabetic Association/World Health Organization criteria. The study group encompassed 356 kidney allograft recipients with graft functioning above 12 months. 48 recipients (13.5%) were diabetic already during dialysis. Therefore, the final analysis included 308 patients.
Results: In the study group 72 patients (23.4%) developed PTDM after renal transplantation; 55 HD patients and 17 PD patients. PTDM incidence at 3, 6, 12 months was 15.9%, 22.1%, 23.4%. The mean interval from transplantation to the onset of the PTDM was 3.08±2.73 months. The following factors exerted significant impact on PTDM appearance: older recipient age ($p \leq 0.001$), hypertensive nephropathy (p=0.0048; OR=2.452), higher body mass index (BMI) at the transplantation day (p=0.045), treatment by peritoneal dialysis (p=0.0319; OR=2.044), older donor age (p=0.032).
Conclusions: 1. Evaluating the risk of PTDM occurrence more attention should be paid to the stratification of the predisposing abnormalities existing during dialysis period. 2. The treatment by peritoneal dialysis was disclosed as the independent, yet not reported risk factor for PTDM development.

Su330 FOCAL SEGMENTAL GLOMERULOSCLEROSIS: ALTERNATE DAYS STEROID THERAPY

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Introduction and Aims: Alternate day steroid therapy (1) is widely believed to be effective in patients of focal segmental glomerulosclerosis (FSGS) patients over the age 60 years, because of an increased susceptibility to the immunosuppressive effects of corticosteroids and altered glucocorticoid kinetics in the elderly. We report results, FSGS patients of all age groups, who, were treated with alternate day steroids.
Methods: All renal biopsy proven primary FSGS patients, except collapsing type, were treated with prednisolone 2 mg/kg/alternate day for 3 months followed by slow tapering till 6th month and simultaneous initiation of levamisole 2.5 mg/kg/alternate day for eighteen months. Only those patients

who completed this schedule from 2002 to December 2009 were included in the study.
Results: Total number of patients: 110; 54.45% of patients were males; Mean age: 28.1 years
Table 1 shows the biochemical parameters of FSGS patients at initiation, 6 months and 8 months of therapy.

Table 1. Biochemical parameter of FSGS patients at initiation and of the therapy

	At initiation of therapy	At the end of 6 months	At the end of 18 months
S. creatinine (mg/dL	1.6±0.8	1.4±0.7	1.2±0.4
Serum proteins (g/dL)	3.6±1.8	5.8±2.6	5.2±2.8
Serum albumin (g/dL)	1.1±0.5	3.8±1.8	3.2±1.5
24 hour urine protein (g)	1.1±0.5	0.6±0.04	0.4±0.03

Number patients achieved complete remission: 64 (58.18%)
Number of patients achieved partial remission: 23 (20.90%)
Conclusions: The alternate day prednisolone plus vermisole, therapy appeared to be successful in achieving remission in majority number of patients of all age groups of patients.
Reference:
1. Nagai R, Cattran DC, Pei Y: Steroid therapy and prognosis of focal segemental glomerulosclerosis in the elderly. Clin Nephrol 42; 18-21,1994

Su331 PENTOXIFYLLINE EFFECT ON DECREASING PROTEINURIA IN TYPE 2 DIABETIC PATIENTS IN A RANDOMIZED-CONTROLLED TRIAL

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Introduction and Aims: Pentoxifyline potentially inhibits the inflammatory cell proliferation and extracellular matrix accumulation. Human studies have suggested it's antiproteinuric effects in glomerular disease. In addition to angintensin blockers,it appears to reduce proteinura in diabetic nephropathy. This benefit have been studied in some trials around the world but not in Iran.
Methods: In a randomized placebo- controlled study, 56 patients (38 female and 18 male mean age 56.43 + 9.25y.) with type II diabetic nephropathy and estimated proteinuria greater than 500 mg/day and creatinine clearance > 60 ml/min, who were treated with angiotensin blockade agents for 3 months or longer, were screened in nephrology and endocrine clinics during May 2008 until April 2009. They were randomly assigned to 1200 mg daily pentoxifylline or placebo in addition of standard treatment. 24 hour urine protein were assessed before and after trial.
Results: Patients were randomly designed in two groups A&B. Pentoxifylline decreased median range of proteinuria from 1662.68 to 678.45 mg/day (mean diff: 61.44%) (P=0.000), compared with 1093.71 to 803 mg/day in the control (mean diff: 19.65%) (P=0.005), the difference between groups was 41.7% (P= 0.000).
Conclusions: Pentoxifylline added to angiotensin blockade agents decreased proteinuria in patients with type II diabetic nephropathy. According to this great difference,we strongly recommend to add this modality to the conventional treatment of type II diabetic nephropathy with macroproteinuria.

Su332 SERUM URIC ACID LEVELS AT THE APPEARANCE OF OVERT NEPHROPATHY IN TYPE 2 DIABETES IS AN INDEPENDENT PREDICTOR FOR PROGRESSION OF RENAL DYSFUNCTION

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Introduction and Aims: Hyperuricemia has recently been reported to be a risk factor for hypertension, chronic kidney disease, end stage renal failure and diabetes.
Whether uric acid has a causal role in the progression of overt diabetic nephropathy is not clarified.
So, historically and prospectively, we assessed the predictive role of serum uric acid levels in renal dysfunction from the appearance of overt nephropathy in type 2 diabetic individuals.
Methods: This retrospective cohort study consisted of 290 patients (231 men and 59 women, mean age 61.9 years) followed from the onset of overt nephropathy in type 2 diabetes. We examined the correlation between serum uric acid levels at the appearance of overt nephropathy (Urinary albumin/creatinine ratio >300mg/gCr) and later progression of renal dysfunction by serum creatinine (Cr) doubling.
The independent association of serum uric acid levels with progression of renal dysfunction was assessed by Cox proportional-hazards models and adjusted for conventional risk factors and several potential confounders.
And, the proportional risk of Cr doubling was represented with the Kaplan-Meier survival curves by uric acid tertile (>368μmol/l[6.2mg/dl], 303-368[5.1-6.2], <303[5.1] for men, >303μmol/l[5.1mg/dl], 244-303[4.1-5.1], <244[4.1] for women)at the appearance of overt nephropathy.
Results: During a median follow-up of 5 years, 85 of 290 patients developed Cr doubling.
In univariate analysis, current smoking, hemoglobin A1c (HbA1c) and uric acid levels were significantly associated with increased risk of Cr doubling (hazard ratio [HR] 2.17, [95%CI 1.40-3.43], 1.32, [1.19-1.46], 1.25, [1.06-1.47] per 1mg/dl increase in uric acid level, p=0.005). After adjustment for age, gender, body mass index, current smoking, hypertension, dyslipidemia, diabetes duration, HbA1c, medications usage (anti-hypertensive, lipid lowering, anti-platelet drugs and anti-hyperuricemia), estimated glomerular filtration rate, the association of serum uric acid with Cr doubling remained statistically significant (HR 1.36, [1.14-1.62], p=0.0005). Patients in the highest tertile of serum uric acid levels had a significantly higher risk rate than the lower two tertiles (log-rank test, p=0.0007)
Conclusions: Serum uric acid levels at the appearance of overt nephropathy in type 2 diabetes is an independent predictor for later progression of renal dysfunction.

Su333 ALISKIREN REDUCES SYMPATHETIC NERVE ACTIVITY IN CHRONIC KIDNEY DISEASE PATIENTS. INDIRECT EVIDENCE THAT IT IMPROVES KIDNEY OXYGENATION

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Introduction and Aims: Hypertensive chronic kidney disease (CKD) patients often have sympathetic hyperactivity. Kidney ischemia seems to be important in the pathogenesis of sympathetic hyperactivity. In this study, we evaluated the effect of chronic treatment with aliskiren on blood pressure and sympathetic activity in CKD patients. Effects of short term oxygen administration on sympathetic activity were also analyzed.
Methods: In ten CKD patients (8 males, aged 44±11 years, GFR 57±22 ml/min per 1.73 m^2) blood pressure and sympathetic activity (quantified by assessment of muscle sympathetic nerve activity, MSNA) were assessed while taken off antihypertensive medication and during chronic (6 weeks) aliskiren 300 mg/day. Secondly, the change in MSNA during 10 minutes of 100% oxygen administration by non-rebreathing mask was measured under both conditions.
Results: Aliskiren lowered supine systolic and diastolic blood pressure from 147±10 to 120±8 and from 96±7 to 83±7 mmHg respectively, (both $P < 0.05$). MSNA was reduced from 36±8 to 26±8 bursts/minute ($P = 0.01$). In untreated condition, oxygen lowered MSNA from 36±8 to 31±7 bursts/minute ($P = 0.03$), while during chronic aliskiren no change was observed (26±8 to 26±9 bursts/minute).
Conclusions: In conclusion, in hypertensive CKD patients, aliskiren effectively lowers blood pressure and MSNA. Short term oxygen administration lowers MSNA in untreated CKD patients, but has no effect during chronic aliskiren. (Clinical trial government identifier number: NCT00719316).

Su334 SERUM 25(OH) VITAMIN D CORRELATES INVERSELY WITH URINARY MONOCYTE CHEMOATTRACTANT PROTEIN-1 BUT NOT WITH URINARY TRANSFORMING GROWTH FACTOR-β1 IN TYPE 2 DIABETIC PATIENTS WITH CHRONIC KIDNEY DISEASE

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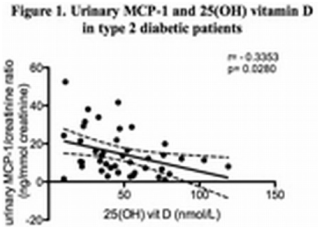
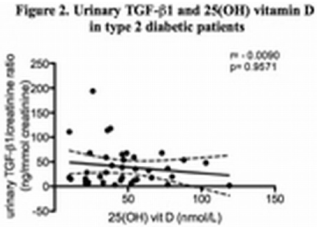
Introduction and Aims: Anti-inflammatory effects of active vitamin D have been demonstrated in vitro as well as in vivo models. A previous study reported on an inverse association of 1,25(OH)2 vitamin D with markers of renal inflammation, such as urinary monocyte chemoattractant protein-1 (MCP-1) in several types of kidney disease. Growing evidence also indicates that vitamin D analogues may have renoprotective effects in patients with CKD. However, it remains unclear, whether these "nonclassical" actions are related to the local production of active vitamin D or circulating 1,25(OH)2D produced by the kidneys. In a prospective study, we have investigated the serum levels of 25(OH)D, 1,25(OH)2D, urinary MCP-1, urinary transforming growth factor-β1 (TGF-β1) and albuminuria, and their potential association to each other in type 2 diabetic patients. A longitudinal follow up of these patients will also provide the opportunity to investigate the changes of those parameters during the replenishment of 25(OH)D.
Methods: Type 2 diabetic patients with CKD stage 2-4 attending the Hammersmith Hospital diabetic/renal outpatient clinics were enrolled in a prospective study. Serum 25(OH)D, 1,25(OH)2D and urinary albumin-creatinine ratio (ACR) were measured for a baseline. Urinary MCP-1 and TGF-β1 were measured by commercially available ELISA kit. Patients with insufficient levels of 25(OH)D (< 80 nmol/L) were replenished according to the hospital standard management guidelines. Regardless of the treatment, all patients were monitored at 2 and 4 months after the enrolment and the same parameters as for the baseline were measured.
Results: 43 type 2 diabetic patients with CKD 2-4 were enrolled and 19 patients were monitored at 2-month follow-up. At the baseline, urinary MCP-1 correlated inversely with serum 25(OH)D level (figure 1, Spearman correlation, r= -0.3353, p=0.0280), but not with urinary TGF-β1 (figure 2, r= -0.0090, p=0.9571) and ACR (r= 0.1129, p=0.4711). There was no correlation of these parameters with 1,25(OH)2D. Interim analysis of 16 treated patients at 2-month follow-up did not show any clear correlation between the changes in urinary MCP-1 and the increase in 25(OH)D levels.





Conclusions: This is the first report showing an inverse correlation of serum 25(OH)D with urinary MCP-1 in type 2 diabetic patients. The ongoing enrolment of further patients and the longitudinal follow-up of these patients will provide the opportunity to investigate the impact of 25(OH)D replenishment on inflammatory and profibrotic cytokines in this patient population.

Su335 ASSOCIATION BETWEEN LEFT VENTRICULAR DIASTOLIC FUNCTION AND GLOMERULAR FILTRATION RATE IN ASYMPTOMATIC TYPE 2 DIABETIC PATIENTS

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Introduction and Aims: Recent studies have shown that hemodynamic changes influence glomerular filtration rate (GFR) in patients with heart failure with normal ejection fraction. The association between subclinical left ventricular (LV) diastolic dysfunction and GFR is not clear yet. This study aims to assess an association between GFR and LV diastolic function (LVDF) in asymptomatic type 2 diabetics. LVDF is widely assessed on the basis of parameters E' and E obtained by tissue Doppler and by conventional pulse Doppler echocardiography respectively. E' is early diastolic mitral annular velocity and reflects myocardial relaxation (impairment <7,5 cm/s in patients aged over 60 years). E is transmitral flow in early diastole. E/E' ratio correlates with LV end-diastolic pressure measured invasively (pathologic value>15, physiologic value <8, borderline 8-15).
Methods: 82 type 2 diabetics (mean age 61±6 years, 34% female) with blood pressure <130/85 mmHg (with or without pharmacologic intervention), LV ejection fraction > 55%, and negative myocardial perfusion SPECT, without overt diabetic glomerulopathy and with mean serum creatinine 89,5±19,2 umol/l were included. Parameters of LV diastolic function E' (cm/s), E (cm/s) and E/E' ratio, NT-proBNP (pg/ml) and eGFR-MDRD (ml/s/1,73 m^2) were obtained. Linear regression was used to analyse the associations between eGFR and other parameters.
Results:

Correlation in whole cohort

eGFR-MDRD (N=82)	E'	E/E'	NT-proBNP
r	0,074	-0,230	-0,357
R2	0,005	0,053	0,127
p	0,508	0,038	0,001

Correlations in subgroups E' < or ≥ 7,5 cm/s

	E' < 7,5 cm/s (N=41)			E' ≥ 7,5 cm/s (N=41)		
eGFR-MDRD	E'	E/E'	NT-proBNP	E'	E/E'	NT-proBNP
r	0,171	-0,348	-0,360	0,144	-0,169	-0,366
R2	0,029	0,121	0,130	0,021	0,029	0,134
p	0,286	0,026	0,021	0,371	0,291	0,019

Other results *i) in whole cohort:* E' versus E/E' (r=-0,562, p<0,0001), E/E' versus NT-proBNP (r=0,317, p=0,0037); *ii) in the subgroup E'<7,5 cm/s:* E' versus E/E' (r=-0,429, p=0,0051), E/E' versus NT-proBNP (r=0,323, p=0,0397); *iii) in the subgroup E' ≥7,5 cm/s:* E' versus E/E'(r=-0,235, p=0,1395), E/E' versus NT-proBNP (r=0,297, p=0,0597).
Conclusions: *1.* The inverse relationship of eGFR with E/E' and simultaneously with NT-proBNP was only found in case of myocardial relaxation impairment in type 2 diabetics. NT-proBNP is probably not influenced only by GFR but also by LV diastolic dysfunction in this group. *2.* The inverse relationship of eGFR with E/E' in patients not presenting with clinical signs of heart failure suggests the possibility of direct association between asymptomatic diastolic dysfunction and GFR. *3.* The inverse relationship of NT-proBNP with eGFR in absence of significant correlation between NT-proBNP and E/E' in type 2 diabetics without LV relaxation impairment is probably predominantly influenced by degree of GFR impairment.
Disclosure: This study was funded by a research grant IGA MZ CR NR/9520-3.

Su336 THE RELATION BETWEEN SMALL AND LARGE CEREBRAL VASCULAR LESIONS AND CHRONIC KIDNEY DISEASE – PRELIMINARY RESULTS

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Introduction and Aims: The relation between small and large cerebral vessel disease and chronic kidney disease (CKD) is a debated subject. In this paper we try to bring some data to the debate, from the point of view incident neurological patients.
Methods: 123 consecutive patients admitted to a neurology department, needing cerebral MRI investigation have been included in the study (44 female, 79 male, average age 61.73±11.50 years). Personal medical data have been retrieved from their hospital files and their GP evidences. Patients have been evaluated for cardiovascular disease (CVD), diabetes mellitus (DM), hypertension (HT), dyslipidemia, chronic kidney disease (CKD) and hsCRP. MRI images have been evaluated for stroke (ischemic – IS and hemorrhagic – HS), silent cerebral infarction (SCI), white matter hyperintensities (WMH). Data have been processed by SPSS 16 data

analysis software system using the t-test and the Fisher exact test (as appropriate) and for correlation the Pearson test.
Results: On MRI imaging IS was identified in 41.4% of the cases, HS in 6.5%, SCI in 21.9%, and WMH in 6.5%. DM was present in 15.44%, CVD in 26.82%, HT in 53.65%, dyslipidemia in 49.59% and CKD in 34.95% of the cases. Elevated hsCRP (>3mg/l) was evidenced in 52.84% of the patients.
CKD patients were significantly older (68.5±10.6 vs. 59.1±10.8 years p=0.0001)and presented significantly higher prevalence of DM (30.2% vs. 13.7% – p=0.035), of HT (83.7% vs. 50% -p=0.0005) and of CVD (46.5% vs. 16.2% p=0.0003). The prevalence of elevated hsCRP was also significantly higher in these patients (74.4% vs. 56.2% p=0.04) but the prevalence of large (IS) and small (HS, SCI, WMH) cerebral vessel lesions did not significantly differ.
IS was directly correlated with the presence of CVD (p=0.030), HT (p=0.01), and inversely with eGFR values (p=0.049) but not correlated with DM, and levels of cholesterol, triglycerides and hsCRP. Cerebral small vessel lesions were correlated with presence of HT (p=0.03) and with cholesterol levels (p=0.022) and inversely correlated with eGFR (p=0.0001) and not correlated with DM, CVD, triglycerides, and hsCRP.
There was a strong positive correlation between large and small vessel cerebral lesions in the studied group.
Conclusions: The prevalence of CKD in cerebrovascular disease patients needing MRI was high and CKD patients were older, with more DM, HT and CVD. It seems that both small and large cerebral vascular (MRI) lesions were inversely correlated with the eGFR levels and HT. Small and large vessel lesions were strongly associated in the studied group.

Su337 ANTI- AND PRO-INFLAMMATORY CYTOKINES IN PATIENTS WITH DIABETIC NEPHROPATHY

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Introduction and Aims: The increased level of some from proinflammatory cytokines such as interferon-gamma (INF-γ) and interleukin (IL)-1 correlates with progress diabetic nephropathy. At the adequate inflammatory answer proinflammatory cytokines play a local protective role, however excessive and generalized their products result in development of organ disfunctions. At the same time there is activating of synthesis of antiinflammatory cytokines on principle of feedback. Different in the origin cytokines are interconnected and co-operate inter se. Therefore a simultaneous estimation of level of a few cytokines can be very informing. Aim is to estimate role of pro- and antiinflammatory cytokines for patients with chronic kidney disease (CKD) stage 1-3 caused type 2 diabetes mellitus.
Methods: We have analyzed the changes in level of serum IFN-γ, IL-1, IL-10 and transforming growth factor β (TGF-β) in 75 patients with diabetic nephropathy (20 patients with stage 1 CKD, 27 patients with stage 2 CKD, 28 patients with stage 3 CKD). As the control parameters of these cytokines of 25 healthy people (norma) has been studied.
Results: The results have shown (see the Table 1) the increase in levels of investigated cytokines in comparison with the norm. The significant difference has been revealed in the levels of such cytokines as IFN-γ, IL-1 and IL-10 for patients with stage 1, 2 and 3 CKD. Significant difference between the index of TGF-β1 for patients with stage 1, 2 and 3 CKD was not (p=0,167), but a tendency took place to the increase to the index with progress of disease.

Serum levels of cytokines (pg/ml) in patients with diabetic nephropathy depending on the stage of CKD

	CKD, st. 1-3	CKD st. 1	CKD st. 2	CKD st. 3	Norm
IFN-γ	136,1±23,4*	113,1±8,6*	138,0±18,9*	150,9±21,9*	20,2±2,2
IL-1	145,2±20,0*	124,6±11,8*	142,4±17,1*	162,5±9,3*	94,9±2,8
TGF-β	100,3±24,2*	91,9±26,7*	101,8±21,1*	105,0±24,3*	56,6±4,3
IL-10	87,9±20,7*	101,6±16,1*	87,4±17,6*	78,6±21,6*	19,9±3,1

Data is presented as M±SD, p<0,001 in comparison with the norm.

Conclusions: There is significant increase of synthesis of proinflammatory cytokines (IFN-γ, IL-1) at progress of diabetic nephropathy. Thus synthesis of antiinflammatory cytokine IL-10 remains significant increased, but with progress of disease his synthesis reduced. The certain changes of investigated cytokines shows that immune mechanisms take part in progress of diabetic nephropathy through enchancement of processes of inflammation.

Su338 CIRCULATING ANGIOGENIC GROWTH FACTORS (SERUM ANGIOPOIETIN-1 AND ANGIOPOIETIN-2) IN PATIENTS WITH DIABETIC KIDNEY DISEASE

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Introduction and Aims: Angiopoietin-1 (Ang-1) promotes supporting and stabilizing of endothelial cells, limits permeability effects of vascular endothelial growth factor (VEGF). In contrast, angiopoietin-2 (Ang-2) is antagonist of Ang-1 and offers vessel regression in absence of VEGF. Up-regulation of Ang-2 is suspected in destabilization of glomeruli endothelium which might play a role in development of increasing glomerular permselectivity in proteinuric renal damage, notably diabetic kidney disease. The aim of our study was to investigate the levels of circulating Ang-1 and Ang-2 and their relations with renal function deterioration markers – glomerular filtration rate (GFR), albumin-to-creatinine ratio (ACR), hemoglobin (Hb) and with main angiogenic factor – VEGF.
Methods: We studied 78 DM patients (pts) (39 type 1 and 39 type 2, 31 males, 47 females). Their mean clinical data: age – 47,6±16,5 years, diabetes duration – 16,6±9,0 years, GFR was calculated by MDRD formula -82,8±21,1 ml/min/1,73 m^2, Hb -132,5±20,0 g/l. Of these, 41 pts had normoalbuminuria (NAU), 15 pts had microalbuminuria (micro-AU), 22 pts had macroalbuminuria (macro-AU). The groups did not differ in mean values of age, DM duration, BMI, $HbA1_c$, CRP, cholesterol, triglycerides and systolic/diastolic BP. The only differences between group were in the levels of Hb and GFR. Anemia was defined by NKF/KDOQI criteria for chronic kidney disease (CKD) pts and WHO classification for pts without CKD.The prevalence of anemia in total group was 30.8% (24 pts). We measured serum Ang-1, Ang-2 and VEGF with ELISA technique. Pts with GFR<15 ml/min/1,73 m^2 were not included.
Results: The mean levels of Ang-1 were similar in pts with NAU, micro-AU and macro-AU. Also its values was not different in pts with and without anemia.We observed the significant elevation of Ang-2 in pts with macro-AU compared to pts with NAU (919,2±603,4 pg/ml *vs* 491,2±217,5 pg/ml, respectively; p<0,001). Pts with micro-AU did not demonstrate the elevation of Ang-2 levels compared to pts with NAU, also both in pts DM type 1 and 2. We noted that in anemic pts the serum concentration of Ang-2 was higher than in pts without anemia (897,7±568,3 pg/ml vs 513,6±256,9 pg/ml, respectively; p<0,001).The serum levels angoipoietins correlated with some parameters: Ang-1 positively correlated with Hb (r=0,24; p<0,05) and VEGF (r=0,38; p<0,001); Ang-2 was negatively associated with GFR (r=-0,38; p<0,001), Hb (r=-0,41; p<0,001) and positively with ACR (r=0,42; p<0,001), VEGF (r=0,25; p<0,05). We did not find association of angiopietins with $HbA1_c$.
Conclusions: Increasing of serum levels of Ang-2 in proteinuric diabetic pts and their direct relations with albuminuria degree may presumably reflects their excessive production in the renal tissue, particularly in glomerulies and the role of Ang-2 in genesis of proteinuria. The positive association of serum VEGF levels with circulating Ang-1 and Ang-2 levels might be an attempt to compensate for depletion of interstitial capillaries and hypoxia in chronically failing diabetic kidney.

Su339 CORRELATION BETWEEN KIDNEY ECHOGRAPHIC DATA AND FUNCTION IN ELDERLY. STRATIFICATION FOR DIABETES

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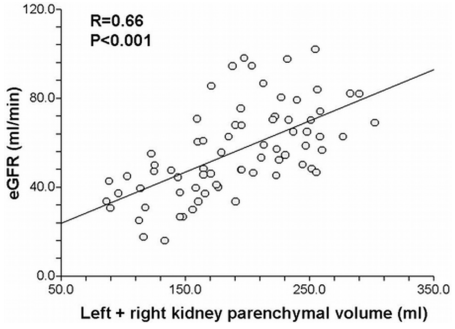
Introduction and Aims: In elderly, few data are available about the correlation between kidney echographic data and function. The relationship between GFR and kidney size is poorly understood in ageing. *Aim:* To evaluate, in a population with age > 65 years, the correlation between

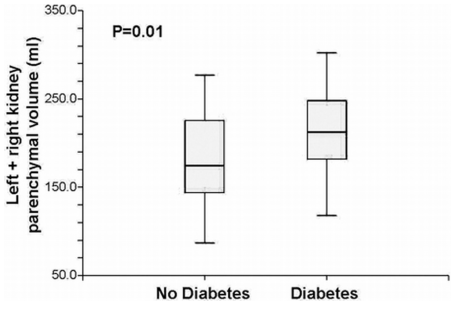
estimated GFR (eGFR) and echographic data in diabetic and not diabetic patients.

Methods: *Design:* Cross-sectional study. We studied 144 kidneys measured by ultrasound in 72 patients with age 65-100 years who did not have autosomal polycystic kidney disease. eGFR was reported according to Cockroft-Gault formula. The sum of left and right kidney longitudinal diameter (L+R-long), kidney volume (L+R-vol), kidney parenchymal volume (L+R-par) and kidney parenchymal section area (L+R-ASMP) were analyzed. Spearman correlation and unpaired t test were used in this study.

Results: The mean age was 79.9±6.9 years, male sex 44.4%, diabetes 29.2%, and hypertension 80.6%. serum creatinine was 0.98±0.42 mg/dl, eGFR 56.2±20.2 ml/min, body surface area (BSA) 1.70±0.22 m^2.

eGFR was correlated to L+R-ASMP (R=0.61, p<0.001), L+R-long (R=0.50, p<0.001), L+R-par (Figure 1, R=0.66, p<0.001) and L+R-vol (R=0.68, p<0.001) in whole population. Patients with diabetes have higher L+R-ASMP (57.3±10.0 cm^2 vs 50.0±11.6 cm^2, p=0.01), L+R-vol (301.8±78.2 ml vs 252.7±67.5 ml, p=0.02) and L+R-par (Figure 2, 214.0±48.5 ml vs 180.6±53.5 ml, p=0.01).





Conclusions: A correlation between kidney echographic data and function is found in elderly. Patients with diabetes have higher kidney volume.

Su340 CIGARETTE SMOKING COULD BE A POTENTIAL CAUSE OF DIABETIC NODULAR GLOMERULOSCLEROSIS

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Introduction and Aims: Nodular glomeruloclerosis, glomerulomegaly, thickening of glomerular basement membrane and arteriolar hyalinosis are present both in diabetic nephropathy with nodular glomerulosclerosis and in idiopathic nodular glomerulosclerosis, which suggests a common mechanism. Chronic smoking is known as a risk factor in the former and a potential causative factor in the latter disease.

Methods: A retrospective analysis of all native renal biopsy specimens (n=890) available in the Renal Pathology Laboratory at our clinic from 2002 to 2009 was performed.

Results: The data revealed significantly more smokers (10 out of 11) among patients with diabetic nephropathy and nodular glomerulosclerosis (DNP + NGS) compared to the random selected patients (4 out of 10) with diabetic nephropathy without histological signs of nodular glomerulosclerosis (DNP non-NGS, p=0.024). Between the two group of patients (DNP + NGS and DNP non-NGS) no significant difference was found in the age (56±14.7 vs. 53.7±7.7years, p=0.875), body mass index (30.5±5.6 vs. 31.9±4.1 kg/m^2, p=0.578), duration of diabetes mellitus (12.11±5.67 vs. 8.89±6.51 years, p=0.279), prevalence (89% both) and duration (10.43±8.92 vs. 12.25±13.55 years, p=0.573) of hypertension, serum total cholesterol (7.5±5.3 vs. 6.9±2.2mmol/l, p=0.781), serum triglyceride (4.5±4.4 vs. 2.8±1 mmol/l, p=0.344), serum creatinine (225.6±136.2 vs. 180.6±88.1, p=0.479), estimated (MDRD) renal function (39.51±24.58 vs. 45.24±26.92ml/min, p=0.685) and the renin-angiotensin system blocker treatment (100% vs. 75%, p=0.155) at the time of the kidney biopsy.

Conclusions: Our results show, that chronic cigarette smoking could contribute to the nodular glomerulosclerosis seen in diabetic nephropathy patients.

Su341 ASSOCIATION OF CONICITY INDEX AND LIPID PROFILES IN PRE-DIALYSIS CHRONIC KIDNEY DISEASE

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Introduction and Aims: Dyslipidaemia and pattern of the fat distribution may accelerate atherosclerosis and the progression of renal disease. Abdominal fat deposition was represented by means of the conicity index (CI), an anthropometric estimate that models the relative accumulation of abdominal fat as the deviation of body shape from a cylindrical towards a double-cone shape. We examined the influence of markers of cardiovascular disease in terms of inflammation, lipid profile and body fat distribution on the progression of renal disease in patients with stable CKD stage 3-5. We also compared the diagnostic sensitivity of CI in MeS with other indices of fat distribution.

Methods: We studied 104 pre-dialysis chronic kidney disease patients between 2008 and 2009 (64 male 62%, 39 female 38% age 64,6±14,7 years). GFR was estimated (44,62±14,38 mL/min/1,73 m^2) based using MDRD study formula. GFR values estimated at initial period and at the end of the 12 months follow-up and retrospectively recorded. Patients were stratified into three group as group 1 loss of GFR ≥20%, group 2 loss of GFR 10 to 20%, group 3 patients were stable renal fonctions or GFR change <10% at the end of 12 months. Anthropometric measurements (height, weight, circumferences of waist and hip) and body mass index (BMI), WHR and CI were subsequently computed. Renal resisitive index (RRI) was measured using Doppler ultrasound. Metabolic syndrome (MeS) was defined in accordance with the National Cholesterol Education Program (Adult Treatment Panel III) criteria for all patients.

Results: CI was strongly correlated with total cholesterol (r=0,37,p<0,00), LDL (r=0,53, p<0,00), CRP (r=0,21, p<0,05). serum potassium (r=0,216, p<0,02), MeS (r=0,22, p<0,00); BMI and WHR were not associated with these parameters. CI, serum cholesterol, LDL, alkaline phosphatase, ALT, LDH, proteinuria, microalbuminuria, resistive index values were significantly lower in group 3. In regression analysis, only RRI was found as independent variable (β=0,68, p<0.01).

	Group 1 N=26	Group 2 N=51	Group 3 N=26	P
CI	1,5±0,1	1,3±0,1	1,2±0,1	<0.01
BMI	27,7±4,0	27,7±4,4	26,5±3,9	<0.01
WHR	0,9±0,1	1,02±0,2	0,9±0,2	=0.33
T. cholesterol	214,5±45,5	185,5±54,0	163,1±48,0	<0.01
LDL	144,6±24,5	119,5±34,5	92,8±26,0	<0.05
Alkaline phosphatase	189,0±72,9	165,1±73,9	163,5±66,5	<0.05
Alanine aminotransferase (ALT)	18,3±6,1	17,8±7,4	13,9±4,5	<0.05
Hemoglobin	11,1±1,3	11,8±1,7	12,1±1,6	<0.01
Proteinuria	1631,0±1002,0	1476,7±962,0	386,6±112,7	<0.01
Microalbuminuria	813,6±100,4	756,1±632,4	211,2±41,7	<0.05
Resistive Index	0,73±0,05	0,69±0,08	0,67±0,087	<0.01

Conclusions: Presence of MeS significantly influenced renal progression in CKD. CI is associated with a higher prevalence of inflammation and cardiovascular risks than other antropometric measurements. CI had the greatest association with GFR thus CI is a beter marker than WHR or BMI in CKD for evaluation of metabolic status, therefore can be substantial in the criteria of MeS.

Su342 COMBINATION OF ANGIOTENSIN CONVERTING ENZYME INHIBITORS WITH MINERALOCORTICOID ANTAGONISTS IN DIABETIC NEPHROPATHY

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Introduction and Aims: Notwithstanding the fact that diabetes mellitus is a low renin state, it has been shown that intrarenal renin-angiotensin system is hyperactivated in this condition. Not only angiotensin II, but aldosterone also has sclerotic effects on different structures of the kidney. The blockade of the renine angiotensin system is an important therapeutic strategy in reducing cardiovascular and renal disease. However the therapeutic response achieved with only one blocker of the renine angiotensine system although efficacious, is limited. We tried to evaluate the effects of an ACE-I (enalapril) combined with a mineralocorticoid antagonist (spironolactone), in order to improve the renoprotective effects of this condition.
Methods: We studied in a placebo controlled trial 62 patients with diabetic nephropathy who had arterial hypertension and proteinuria (> 300 mg/l). Our patients (pts) received enalapril 20 mg/d. We randomly assigned these pts to placebo, losartan (100 mg/d), or spironolacton (50 mg/d) for 72 weeks. In all pts, it was measured proteinuria creatinine, urea and electrolytes.
Results: In comparison with placebo group, proteinuria decreased by 42,3% (95% CI, p=0.004) in the group assigned to spironolactone and by 21,2% (95% CI, p=0.3) in the group assigned to losartan. Blood pressure effects did not differ between the groups. Also, dietetic protein and sodium intake as well as creatinine clearance were similar in all groups. Hyperkalemia was increased similarly between losartan and spironolacton group.
Conclusions: The combination of ACE-I with spironolacton was more renoprotective than ACE-I plus losartan in our pts with diabetic nephropathy.We believe that other long term and large scale studies will support our results.

Su343 INSULIN RESISTANCE IS A LASTING BIO-MARKER IN NON-DIABETIC ESSENTIAL HYPERTENSION PATIENTS AFTER EFFECTIVE ONE-YEAR LIFESTYLE CHANGES

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Introduction and Aims: Little information on lasting effect of concurrent dietary and lifestyle interventions in hypertensive patients is available. The objective of this study is to investigate if lifestyle intervention and effective blood pressure control influence insulin resistance (IR) assessed as HOMA-IR, and Renal Artery Stiffness assessed as Renal Resistive Index (RRI), after one year interventional program in overweight-obese patients.
Methods: 315 patients: 156 hypertensive and a control group of 159 patients without arterial hypertension and/or any history of arterial hypertension were studied by US measurements of Renal Resistive Index, Insulin Resistance (by HOMA), and extensive laboratory investigations. One year program aimed at lifestyle modification, diet and physical activity changes, was arranged.
Results: At post-interventional assessment 156 hypertensive patients have HOMA-IR still abnormal and significantly higher in comparison with a control group of 159 patients, not different according to age, body weight and BMI. Lower salt/lower calories Mediterranean diet, physical activity increase and smoking withdrawal counseling were provided.

Post-interventional differences between hypertensive patients vs. normal BP patients

	Hypertensive patients	Normal BP patients	p
Age, years	50.72±7.56	49.42±9.39	0.225
Systolic Blood Pressure, mmHg	130.99±6.49	118.74±5.36	<0.0001
Diastolic Blood Pressure, mmHg	81.41±5.27	75.60±4.60	<0.0001
BMI, kg/m^2	28.29±4.35	27.50±4.31	0.108
Fat Mass %	34.92±10.36	35.35±10.12	0.767
HOMA-IR	2.57±1.79	2.13±1.62	0.026
Creatinine, mg/dl	0.86±0.18	0.82±0.17	0.074
Total Cholesterol, mg/dL	204.99±39.11	205.02±42.35	0.996
HDL Cholesterol, mg/dL	52.55±14.12	53.92±16.50	0.433
Triglycerides, mg/dL	122.24±53.41	117.15±78.70	0.503
LDL Cholesterol, mg/dL	128.39±32.89	128.37±37.70	0.996
Renal Resistive Index	0.62±0.06	0.60±0.04	0.009

Values are averages ± SD.

By multiple stepwise regression to HOMA-IR, BMI and Cholesterol-HDL are the best predictors confirming the close relationship of IR with persisting overweight. RRI, as a sign of renal artery stiffness, improves in EH patients after lifestyle interventions.No significant predictor to RRI is identified in the present series of patients.
Conclusions: The persistence of insulin resistance can be tentatively assumed as a steady sign of the cluster of disease encompassed by metabolic syndrome, lasting also after extended lifestyle intervention in essential hypertension. As a consequence, conceivably, there is still a major indication to more intensive or targeted dietary interventions also in pharmacologically treated patients responding to therapy.

Su344 TARGETTING PARATHYROID HORMONE LEVEL IN NON-DIALYSED DIABETIC CHRONIC KIDNEY DISEASE PATIENTS: DOES METABOLIC SYNDROME MATTER?

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Introduction and Aims: Type 2 diabetic patients have lower intact parathyroid hormone (iPTH) levels when compared to non-diabetics. Patients with metabolic syndrome (MetSyn) have increased iPTH levels than normal subjects. We hypothesized that type 2 diabetic patients with MetSyn might have higher iPTH levels compared to patients without MetSyn.
Methods: Study had an observational design and 90 type 2 diabetic patients with chronic kidney disease were recruited (male/female: 42/48). After an overnight fasting, each patient underwent the following procedures: (a) determination of full blood count, clinical chemistry profile, lipid profile, serum creatinine, and iPTH; (b) determination of 24-hour urinary albumin excretion on the same day. The presence of MetSyn was determined according to Adult Treatment Panel (ATP) III criteria.
Results: Sixty-one patients had MetSyn. As going from stage 1 to stage 5 chronic kidney disease, iPTH levels increased significantly (Ptrend: 0.003). Serum iPTH was negatively correlated with estimated glomerular filtration rate as measured by Modification of Diet in Renal Disease (MDRD) formula (r:-0.305, P: 0.003), serum calcium (r: -0.249, P: 0.02) and hemoglobin (r: -0.228, P: 0.034) and was positively correlated with urinary albumin excretion rate (r: +0.233, P: 0.030). Diabetic patients with MetSyn had lower high-density lipoprotein cholesterol (P: 0.024), and higher body mass index (P: 0.031), systolic blood pressure (P: 0.028), fasting plasma glucose (P: 0.022), HbA1c levels (P: 0.021), iPTH (P: 0.026) and triglyceride (P <0.0001) than patients without MetSyn. After adjusting for potential confounders, logPTH was higher in patients with MetSyn compared to patients without MetSyn among type 2 diabetic patients with chronic kidney disease (P: 0.032).
Conclusions: MetSyn might influence iPTH levels in type 2 diabetic patients. We think that whether MetSyn should be taken into account in determining target iPTH levels in type 2 diabetic patients with chronic kidney disease remains to be questioned.

Su345 EFFECT OF THIAZOLIDINEDIONES ON ALBUMINURIA AND PROTEINURIA IN DIABETES: A META-ANALYSIS

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Introduction and Aims: Due to the major clinical and economic burden of diabetic nephropathy, new therapeutic tools to delay its progression are needed. Recent studies suggest that thiazolidinediones have renal benefits. This systematic review and meta-analysis aimed to evaluate the effect of thiazolidinediones on urinary albumin and protein excretion in patients with diabetes mellitus.
Methods: A systematic literature search of MEDLINE/PubMed, EMBASE and Cochrane CENTRAL databases was performed to identify randomised controlled trials published until September 2009 comparing the effect of rosiglitazone or pioglitazone to placebo or other antidiabetic drugs. Two investigators independently extracted data on study and patient characteristics, study quality and outcomes of interest. Weighted mean differences

(WMD) and standardized mean differences (SMD) for the change in the levels of urinary albumin or protein excretion between the thiazolidinedione and control groups were calculated using random-effect models.
Results: Of the 171 originally identified studies, 15 studies (5 with rosiglitazone and 10 with pioglitazone), involving 2860 patients were included in the analysis. In subjects with baseline normo- or microalbuminuria the proportional change between thiazolidinedione and control groups in urinary albumin excretion measured by time-specified urine collections was -64.77% (95% CI: -75.6% to -53.94%) and the change in albumin to creatinine ratio was -24.78% (95% CI: -39.59% to -9.96%). Overall, in subjects with normo- and microalbuminuria thiazolidinedione treatment was associated with a significant reduction in urinary albumin excretion (SMD: -0.63 units of SD, 95% CI: -0.83 to -0.43). Similarly, thiazolidinediones were associated with a significant reduction in urinary protein excretion (SMD: -1.11 units of SD, 95% CI: -1.79 to -0.42).
Conclusions: Treatment with thiazolidinediones significantly reduces the levels of urinary albumin and protein excretion in patients with diabetes. This finding calls for clinical trials with hard renal outcomes to elucidate a possible beneficial effect of thiazolidinediones on diabetic nephropathy.

Su346 MORTALITY IN PATIENTS WITH DIABETIC NEPHROPATHY IN MOSCOW COUNTY

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Introduction and Aims: Aim of the study was to analyze mortality rate, standardization mortality ratio, life expectancy, survival rate in patients with diabetic nephropathy (DN) of Moscow county.
Methods: Data for 168 270 diabetes mellitus (DM) patients who live in Moscow County is in DM register, DN was registered among 10 953 of them. Mean mortality rate, life expectancy and survival rate were examined during 2004-2008 yrs. Age adjusted standardization mortality ratio (SMR) was calculated as ratio of observed and expected deaths. Survival analyses by the Kaplan-Meier method was also used.
Results: Mortality rate of T1DM and DN (age adjusted) was 4000 per 100,000 person-years. It was two times higher than in T1DM without DN (SMR -1,93). Mortality rate of T2DM and DN (age adjusted) was 6296,5 per 100,000 person-years. T2DM patients with DN are more at risk of death than their peers with T2DM without DN (SMR = 1,99). Patient with T1DM and DN have shorter life expectancy than patient with T1DM without DN.(48,74±1,17 v.s. 50,98±1,06 yrs). There was no difference between T2DM patients with or without DN. Life-analyses by the Kaplan-Meier method indicated cumulative survival rates of 77,2% % at 5 years (2004-2008 yrs) after DN diagnosis. It was higher in males than females (87,9% v.s. 61,8%). Among T2DM patients survival rate at 5 years after diagnosis was 69,7% (females-71,7%, males-62,3%).
Conclusions: Among diabetic patient with DN age –adjusted mortality rate remains to be high compared with diabetic patients without DN. Life expectancy among DM patients with DN is compatible with life expectancy among DM patients without DN due to long period between time of DM and DN onset. Cumulative survival rates is higher in males than females with DN, probable, due to high risk of progression of DN during pregnancy.

Su347 EVALUATION OF RENAL FUNCTION PARAMETERS AMONG THE POPULATION WITH TYPE 2 DIABETES IN THE REGION OF MADRID. DIMA-2 STUDY

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Introduction and Aims: To analyze renal function parameters in a population with type 2 diabetes attending primary care offices at the region of Madrid, Spain.
Methods: This is an epidemiological, observational, multicenter study. Data on clinical history is collected. Inclusion criteria are:
– Population is older than 18 years
– More than 6 months since diagnosis of type 2 diabetes
– Informed consent signature
Results: 1020 patients (mean age 67.0±11.6 years, male 50.5%) were studied. At the baseline study, according to clinical history data, percentage of patients with a estimated glomerular filtration rate (eGFR) < 60 ml/min was 8,2% and prevalence of microalbuminuria was14,2%. However, on analyzing biochemical data, the prevalence of eGFR < 60 ml/min is 24% (according to MDRD-4) and 29,9% (Cockroft-Gault). Patients with urine albumin excretion higher than 30 mg/24h were 20,5%. Percentaje of patients with serum creatinine (SCr) ≥1.3 mg/dL in males and ≥1.2 mg/dl in women was 11,4%. If analyzing on gender, a significant difference in SCr and eGFR (MDRD-4) is observed, worse in women, as expressed at the table. On evaluating the population whether followed or not by the endocrinologist, a significant difference is observed in microalbuminuria level, higher among patients attended both in primary care physician and endocrinologist (27.2±86.2 vs 59.4±136.7mg/24h;p= 0.019). There is also a significant difference on analyzing the percentage of patients with a pathological SCr level, worse if receive double follow-up (10.0 vs 15.7%; p=0.024)
Conclusions: Percentage of type 2 diabetic patients with renal function derangements is high. However, perception of those alterations is much lower among their primary care physicians.

Variable (unit) n	Male, Mean ± sd	Women, Mean ± sd	p
Creatinine (mg/dL)	1.14±0.89	0.96±0.55	<0.001
eGFR Cockroft (ml/min/1.73 m^2)	74.4±24.5	72.0±25.1	0.136
eGFR MDRD-4	74.8±21.9	69.8±20.9	< 0.001
Albumin/creatinine ratio	41.1±108.2	69.8±20.9	0.179

eGFR: estimated glomerular filtration rate.

Su348 RELATIONSHIP OF VASCULAR ENDOTHELIAL GROWTH FACTOR AND ERYTHROPOIETIN CIRCULATING LEVELS IN ANEMIC AND NON-ANEMIC DIABETIC PATIENTS

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Introduction and Aims: Vascular endothelial growth factor (VEGF) production is regulated by oxygen tension with markedly increased VEGF gene expression under hypoxic conditions. The mechanisms stimulating VEGF expression are similar to those of erythropoietin (EPO) and closely related from derivation of hypoxia-inducible factor-1 (HIF-1) to response to hypoxia (anemia). The aim of our study was to investigation the changes in circulating levels of diffusible $VEGF_{165}$ and EPO in diabetes mellitus (DM) patients (pts) with or without anemia and association of circulating VEGF with EPO levels.
Methods: We studied 78 DM pts (39 type 1 and 39 type 2, 31 males, 47 females). Their mean clinical data: age – 47,6±16,5 years, diabetes duration – 16,6±9,0 years, $HbA1_c$ -9,0±1,7%, glomerular filtration rate (GFR) was calculated by MDRD formula -82,8±21,1 ml/min/1,73 m^2, hemoglobin (Hb) -132,5±20,0 g/l. Of these, 41 pts had normoalbuminuria (NAU), 15 pts had microalbuminuria (micro-AU), 22 pts had macroalbuminuria (macro-AU). The mean values of age, DM duration, BMI, CRP, $HbA1_c$, cholesterol, triglycerides and systolic/diastolic BP between groups of pts with NAU, micro-AU and macro-AU were not significant. These groups had differences only in the levels Hb (pts with macroAU (12,2±1,7 g/dl) between pts with NAU (14,0±1,8 g/dl; p<0,001) and micro- AU (13,0±1,9 g/dl; p<0,05)) and GFR (pts with macro-AU between pts with NAU (p<0,00001) and micro-AU (p<0,05)). Anemia was defined as Hb < 13,5 g/dl in men and < 12,0 g/dl in women by the definition of anemia by NKF/KDOQI for pts with chronic kidney disease (CKD) and Hb < 13,0 g/dl in men and < 12,0 g/dl in women by the WHO classification of anemia for pts without CKD. The prevalence of pts with anemia was 30,8% (24 pts), without -69,2% (54 pts). We measured serum VEGF and EPO with ELISA technique. Pts with GFR<15 ml/min/1,73 m^2 were not included.
Results: In pts with anemia and without anemia the mean serum levels of VEGF and EPO were similar (VEGF – 478,0±300,7 pg/ml vs 622,0±507,7

pg/ml, respectively, p=NS; EPO -6,3±6,4 mIU/ml vs 8,2±8,1 mIU/ml, respectively, p=NS). There are no significant differences between groups of pts with NAU, micro-AU and macro-AU in serum levels of VEGF and EPO. We did not find correlation of Hb with serum VEGF. Instead, we observed the presence of physiological inverse association between Hb and EPO in pts with GFR ≥60 ml/min/1,73 m^2 (r=-0,32; p<0,05). We found significant negative association between levels of VEGF and EPO in group of anemic pts (r=-0,46; p<0,05) and their positive relations in non-anemic pts (r=0,34; p<0,01).

Conclusions: The circulating levels of VEGF and EPO are similar in DM pts with anemia and without anemia. The association of EPO with Hb, but not VEGF, was observed only in pts with normal and mild decreasing of GFR (GFR≥60 ml/min/1,73 m^2). However, the negative linkage or misbalance in relations between VEGF and EPO in anemic may reflect the behavior of VEGF in hypoxia which contributes development of endothelial dysfunction, pathological angiogenesis compared to normoxia condition.

Su349 THE ACTIVITY OF THE INFLAMMATORY ENZYME C2GNT, Core-2 [β]1,6 N-ACETYLGLUCOSAMINYLTRANSFERASE ARE RAISED IN PATIENTS WITH DIABETIC NEPHROPATHY

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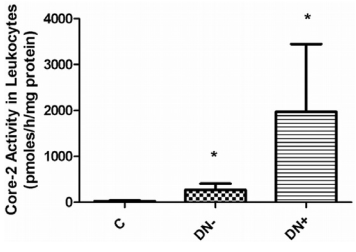
Introduction and Aims: Diabetic nephropathy (DN) is the main cause of end-stage renal diseases (ESRD), but the exact mechanisms leading to the development and progression of renal injury are not yet fully understood. There is substantial evidence suggesting that chronic sub-clinical inflammation plays a role in the pathogenesis of diabetic nephropathy, therefore we examined if the inflammatory enzyme, core 2 b-1, 6-N-acetylglucosaminyltransferase (C2GNT) is associated with DN. We have already established the role of C2GNT in diabetic retinopathy (Chibber et al. Current Diabetes Review 2007) and neuropathy (Spruce et al. Diabetic Med 2004).

Methods: We determined the activity of C2GNT in polymorphonuclear (PMN) leukocytes of patients with Type 1 and Type 2 diabetes with and without diabetic nephropathy.

Whole blood was drawn and collected in EDTA tubes. Polymorphonuclear (PMN) leukocytes and plasma were isolated after density gradient centrifugation over Histopaque. The cells were lysed for the measurement of the C2GNT activity assay (Ben-Mahmud et al. Diabetes 2004).

Results: The activity of C2GNT was significantly higher in PMNs of patients with diabetes as compared to age-matched healthy control subjects [267.4±139.3 *vs.* 22.10±15.05 pmoles/h/mg protein, P<0.05]. Furthermore, the C2GNT activity was significantly higher in patients with nephropathy (DN+) compared to patients with no nephropathy (DN+) [1973±1475 *vs.* 267.4±139.3 pmoles/h/mg protein, P<0.05].



Conclusions: Higher activity of C2GNT in leukocytes supports the important role of inflammation in diabetic nephropathy.

Su350 FOLIC ACID TREATMENT DOES NOT IMPROVE RENAL ENDOTHELIAL FUNCTION AND ALBUMINURIA IN HYPERTENSIVE PATIENTS WITH TYPE 2 DIABETES

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Introduction and Aims: We have previously demonstrated increased oxidative stress within the renal circulation of hypertensive patients with type 2 diabetes, and further found a relationship between renal endothelial dysfunction and albuminura. In the current study, we tested whether antioxidant treatment with folic acid would improve renal vascular nitric oxide (NO) activity and lower albuminuria.

Methods: In a randomized, double-blind, cross-over trial, 16 hypertensive patients with type 2 diabetes and microalbuminuria despite antihypertensive therapy with inhibitors of the renin-angiotensin system were treated with either placebo or folic acid (FA) 5 mg/day over 4 weeks. Renal NO activity and oxidative stress were assessed as the response of renal plasma flow (RPF) to NO synthase inhibition with N(G)-monomethyl-L-Arginine (L-NMMA; 4.25 mg/kg i.v.) and to vitamin C (3g i.v.), respectively. In conjunction, albuminuria, 24h blood pressure (BP) and systemic oxidative stress, as ratio of reduced to oxidized glutathione levels in whole blood (GSH/GSSG) and as total antioxidant capacity of plasma (TAC), were determined.

Results: FA did not affect the RPF responses to L-NMMA (-56±50 ml/min during FA vs -71±47 ml/min during placebo, p=0.18) and vitamin C (+99±98 ml/min vs +122±102 ml/min, p=0.65), and had no effect on albuminuria (48±73 mg/g creatinine vs 62±99 mg/g creatinine, p=0.24). Further, 24h BP (mean BP: 99±9 mmHg during FA vs 102±9 mmHg during placebo, p=0.38) and systemic parameters of oxidative stress (GSH/GSSG ratio: 46±49 vs 86±122, p=0.23; TAC: 0.11±0.05 vs 0.12±0.08, p=0.44) did not change with FA treatment.

Conclusions: In hypertensive patients with type 2 diabetes, FA treatment on top of antihypertensive therapy failed to improve NO activity and oxidative stress within the renal vasculature and had no effect on albuminuria. This might be related to the concomitant treatment with inhibitors of the renin-angiotensin system, which may already have reduced oxidative stress. Further, no effects on 24h BP and systemic markers of oxidative stress were noted. Although the relatively small sample size of our study has to be acknowledged, antioxidant treatment with FA does not seem warranted at this stage of diabetic nephropathy.

Su351 PARICALCITOL INDUCED IMPROVEMENT IN INSULIN RESISTANCE IN PATIENTS WITH CHRONIC KIDNEY DISEASE AND METABOLIC SYNDROME

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Introduction and Aims: Metabolic syndrome (MS) is a mosaic of disorders that include obesity, dyslipidemia, hypertension and intolerance in glucose. The common cause of these disorders is the tissue resistance in insulin action. Patients with chronic kidney disease (CKD) present a high prevalence of insulin resistance (IR) and MS, which are associated with a high risk for diabetes, cardiovascular disease (CVD) and increased all-cause mortality. The aim of the study was to evaluate the influence of oral administration of paricalcitol in the IR of CKD patients with MS.

Methods: We monitored paricalcitol therapy in patients with CKD stage 2 -4 and secondary hyperparathyroidism (sHPT) in a 30-week observational study. The patients were recruited from 2 nephrological centers between 2009 and 2010. Patients with prior administration of vitamin D or its analog, hyperphosphatemia (>6 mg/dL), or hypercalcemia (>10mg/dL) were excluded. All the patients were treated with paricalcitol aiming the normalization of serum levels of the intact parathormone (iPTH) according to the K/DOQI guidelines for the levels of iPTH at different stages of CKD. Fasting plasma insulin (If), glucose (Gf), iPTH, calcium (Ca), phosphorus (PO4) and CaxPO4 product, were measured monthly. IR was evaluated by using the homeostasis model assessment of insulin resistance

index (HOMA-IR). The data were expressed as mean ± SD. Statistical significance was set at $p<0.05$.
Results: The study included 24 patients (16 M/8 F), age 55±11 years; eGFR 44 mL/min (range 17-66). Analyses of the data at the start versus end of the study showed significant changes at the serum levels of If (μU/mL) 25.7±2.6 vs 20.75±3.1, p=0.003, the HOMA-IR 6.3±0.7 vs 4.9±0.6, p=0.001 and of the iPTH (pg/mL) 146.5±30.7 vs 77.5±15.6, $p<0.003$.
LIMITATIONS: The relatively small sample size.
Conclusions: Paricalcitol resulted in a significant reduction in insulin resistance in patients with chronic kidney disease and metabolic syndrome.

Su352 LOWERING OF HEMATOCRIT IN PATIENTS WITH NORMAL RENAL FUNCTION TREATED WITH COMBINED THIAZOLIDINEDIONES (TZD) AND RENIN-ANGIOTENSIN SYSTEM INHIBITORS

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Introduction and Aims: Inhibition of renin-angiotensin-aldosterone system (RAAS) with ACEi or ARBs can cause a minor decrease in hematocrit (almost 1%) in general population. Nevertheless an important drop in Hct has been noticed in patients with Chronic Renal Disease (CRD), COPD and polycytaemia, Chronic Heart Failure (CHF), Renal transplant and high attitude population. The use of TZD has been related with Na and water retention, result in worsening of CHF and lowering Hct due to overhydration.The intention of this study as to evaluate the effect of TZD and ACEi/ARB combined treatment in diabetic patients with normal renal function.
Methods: Medical reports of 48 patients were reviewed before and after one year of treament with the following drugs; (a) other drugs than TZD and ACEi/ARB (control group, n=12) (b)TZD without ACEi/ARB (group TZD, n=12), (c) ACEi/ARB without TZD (group ACEi/ARB, n=12), (d) combination of TZD and ACEi/ARB (TZD+ACEi/ARB, n=12). Patients with heamatopoetic, malignant hepatic and renal diseases were excluded. The results are presented as mean value ± SEM. ANOVA, x-square and t-test were used concerning the statistical analysis.
Results:

	Control group (n=12)	TZD group (n=12)	ACEi/ARB (n=12)	TZD + ACEi/ARB (n=12)
ΔHct	+0,36±1	-0,1±0,88	-0,9±0,5	-4,6±0,4
P	NS	NS	NS	0,000

Conclusions: Combined TZD plus ACEi/ARB treatment in diabetic patients with normal renal function could provoke in the longterm a statistically significant drop of Hct. In contrast either drug individually could provoke small and non-statistically significant drop of Hct with no overt clinical implication.
The underlying mechanism of statistically significant Hct (RBC mass/plasma) drop could be the combined effect of inhibition of erythropoesis caused by ACEi/ARBs, with salt and water retention due to TZDs. In conclusion the resulting anemia with the above combined treatment should be in the differential diagnosis of the attending physician.

Su353 PHENOTYPIC AND GENOTYPIC PROFILE OF UROPATHOGENIC E. Coli COULD PLAY A MAJOR ROLE IN "PRIMARY" ACUTE PYELONEPHRITIS

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Introduction and Aims: Acute "primary" pielonephritis (APPN) is increasingly recognised in young otherwise healthy patients without classical predisposing factors. In this setting intrinsic pathogenic features of infecting microrganisms seems to play a major role.
Aim of the work was to detect genotypic and phenotypic features of E.Coli involved in "primary" forms of acute pyelonephritis (APN).
Methods: We analysed strains of E.Coli isolated in urine of a sample of 118 patients (110 female, 8 male) patients with APPN who were admitted at our Unit between January 2004 and June 2009. These strains were compared with those isolated during routine laboratory activity at our hospital.
Results: Philogenetic study has shown a higher prevalence of B2 group in pyelonephritogenic strains of E Coli compared with the ones isolated from urine which were cultivated at our laboratory in the setting of routine activity (81% vs 62%); genotypic study has shown a higher expression of factors of virulence with statistically significant differences for 2 specific factors of virulence: pathogenicity island ("PAI") and the system of iron acquisition "FyuA". 21% of blood cultures turned out to be positive and E. Coli was isolated in 53% of cases. In 5 patients in whom E. Coli was isolated both from blood and urine the same strain was involved. 40% of patients started therapy with a single type of drug, usually quinolones (67% of cases), while the most frequent association is based on quinolones and penicillins (42%). Mean duration of therapy, which was modified in 53% of cases because of inadequate response, was 34±18 days.
Conclusions: APPN is a new nosological entity characterised by the absence of traditional predisposing risk factors and a significanlty higher prevalence of pyelonephritogenic strains of E.Coli endowed with with specific genetically determined factors of virulence, such as "island of pathogeneticity" ("PAI") and the system of iron acquisition "FyuA". Phenotypic and genotypic a definition of uropathogenic E.Coli is a promising tool for a tailormade and rational management of APPN, which still represents an unmet challenge for the nephrologists.

Su354 SPIRONOLACTONE EFFECT ON PREVENTING PROTEINURIA IN CHRONIC KIDNEY DISEASE

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Introduction and Aims: Aldosterone may contribute to progressive kidney disease. Although angiotensin – converting enzyme inhibitors (ACE) and angiotensin type 1 receptor antagonists (ARB) suppress the rennin – angiotensin system, these agents do not adequately control plasma aldosterone level. Hence, administration of aldosterone receptor antagonist may provide additional renal benefits to the ACE inhibitors and ARBs.
Methods: We evaluated the short- term (8 Weeks) effect of spironolactone on proteinuria in 80 patients with chronic kidney disease (CKD) already receiving ACE inhibitors or ARBs.
Results: Spironolactone (25mg/d for 8 weeks) decreased proteinuria from 2796/1 to 1857/4 after 8 weeks ($P < 0.001$). Four weeks after discontinuation of spironolactone therapy, proteinuria returned to close to baseline values.
Conclusions: This study showed that spironolactone may effectively reduce protenuria in patient with CKD. Prospective randomized trials are necessary to confirm the efficacy and safety of antagonists of aldostrone on proteinuria and progression of CKD.
Disclosure: This study was supported by Ahwaz Jondishapour University of Medical Sciences.

Glomerular diseases 2

Su355 ★ PEXIVAS: DESIGN OF A RANDOMISED CONTROLLED TRIAL OF PLASMA EXCHANGE AND GLUCOCORTICOID DOSING IN SEVERE ANCA ASSOCIATED VASCULITIS

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Introduction and Aims: The current outcomes of ANCA associated vas-

culitis (AAV) are frequently poor for those presenting with severe renal disease or pulmonary haemorrhage and drug-related toxicity contributes to mortality and morbidity. A scientific rationale for plasma exchange has emerged with demonstration of the pathogenicity of ANCA and PLEX has been shown in the recent MEPEX trial, and in a meta-analysis of all PLEX trials in vasculitis, to improve renal recovery in severe renal vasculitis. However it is unclear whether this benefit is sustained and whether PLEX reduces mortality or is effective in pulmonary haemorrhage. The expense and potential complications of PLEX demand stronger evidence before its use can be routinely recommended. Current glucocorticoid (GC) dosing is a major contributor to severe adverse events, a particular concern in those with uraemia or respiratory failure. Recent optimisation of cyclophosphamide regimens and demonstration of the efficacy of rituximab has re-focused attention on whether GC dosing can be safely reduced without compromising efficacy.

Primary Objectives: 1) To determine the efficacy of PLEX in addition to immunosuppressive therapy and GC at reducing death and end-stage renal disease (ESRD). 2) To determine whether a reduced dose GC regimen is non-inferior to a standard dose GC regimen with respect to death and ESRD, as well as being superior with respect to adverse events (especially infections). Secondary and Exploratory Objectives include the effects of the two interventions on disease activity, quality of life, cost-effectiveness, disease-related damage, long-term renal function, vasculitis biomarkers.

Methods: 500 Patients with new or relapsing severe AAV (renal vasculitis with recent eGFR< 50 mls/min or/and pulmonary haemorrhage), >=15 years of age, from 60 centres in 15 countries will be recruited over 5 years and followed-up for a maximum of 7, minimum 2 years. Randomisation will be performed by a central computerised facility at the Birmingham Clinical Trials Unit. PEXIVAS is an open label RCT and the 500 patients will be compared by dividing them into 4 groups with 125 in each, receiving treatment as follows: 1) PLEX + standard dose GC 2) PLEX+ reduced dose GC 3) no PLEX + standard dose GC 4) no PLEX + reduced dose GC. All patients will receive induction therapy, usually cyclophosphamide, with rituximab as a permitted alternative and maintenance therapy with azathioprine. Immunosuppressive and GC dosing will be determined by the protocol for the first 12 months after trial entry.

Results: are awaited.

Conclusions: PEXIVAS aims to clearly determine both the role of PLEX in severe AAV and whether GC dosing can be safely reduced. The results will be of potential benefit to other vasculitis presentations and the use of PLEX in other immune-mediated disorders. The collaborative nature of the trial, stretching over three continents will promote parallel vasculitis research and wider introduction of best practice in vasculitis management.

Su356 ★ GENE POLYMORPHISMS CONTRIBUTING TO HYPERTENSION IN IgA NEPHROPATHY (IgAN)

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Introduction and Aims: IgAN is one of the common glomerulonephritides, and the age of onset is young: chiefly in 10-20 years. Some long-term observational studies have demonstrated that the 10 years renal survival rates were approximately 80%, and still now, many IgAN patients progress to end-stage renal disease. Hypertension has been identified as one of the critical risk factors for renal progression in chronic kidney disease patients, and both genetic and environmental factors play key roles in hypertension, but these mechanisms are unknown. Identification of the genetic factor contributing to hypertension may help to provide a therapeutic strategy. The aim of the present study is to identify the gene polymorphisms associated with hypertension in IgAN patients who were enrolled in our present study: Polymorphism REsearch to DIstinguish genetic factors Contributing To progression of IgA Nephropathy (PREDICT-IgAN).

Methods: *Design and setting:* Multicenter cross-sectional study.

Patients: Among 1132 patients aged ≥15years who were diagnosed as IgAN by renal biopsy in Osaka University Hospital, Osaka General Medical Center and Osaka Rosai Hospital between 1990 and 2005, 429 patients who visited these hospitals between April 2006 and March 2008, were participated in PREICT-IgAN. 240 patients aged 15-50 years with ≥0.25g/day, were enrolled in the present study.

Outcome: Hypertension was defined as ≥140mmHg of systolic blood pressure, ≥90mmHg of diastolic blood pressure, or use of antihypertensives at renal biopsy.

Independent variables: A hundred gene polymorphisms, mainly single nucleotide polymorphisms (SNPs) and clinical characteristics at diagnosis: age, gender, BMI, eGFR, urinary protein, total cholesterol, uric acid and smoking status.

Statistics: We assessed associations between hypertension and gene polymorphisms with the minor genotype with ≥10% of frequency and Hardy-Weinberg equilibrium (dominant and recessive models), using χ^2 test. Family-wise error rates were controlled by the method of Bonferroni. We identified gene polymorphisms associated with hypertension even after adjustment for clinical relevant factors in multivariate logistic regression analysis.

Results: Baseline characteristics: age 33 (24-43) years (median (interquartile range)), male 40.4%, systolic and diastolic blood pressure 122±17/76±14mmHg (mean±SD), eGFR 79±27mL/min/1.73m^2, urinary protein 0.7 (0.4-1.3)g/day, use of antihypertensives 19.7%, hypertension 36.3% and smokers 25.8%. Among 28 candidate gene polymorphisms, *CD14*C159T (CC vs. CT/TT P=0.028) and *ACE*D/I (DD vs. DI/II P=0.034) were significantly associated with hypertension. Multivariate logistic regression models revealed that *CD14*C159T (CC vs. CT/TT Odds Ratio 3.69[95%CI 1.71-7.97]) and *ACE*D/I (DD vs. DI/II 4.07[1.647-10.06]) were independently associated with hypertension.

Conclusions: *CD14*C159T and *ACE*D/I contributed to hypertension in IgAN patients.

Su357 THE EFFECT OF PROTEASOME INHIBITION IN PODOCYTES AND TUBULAR CELLS

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Introduction and Aims: Systemic lupus erythematosus (SLE) is a multisystem inflammatory autoimmune disease that may affect multiple organs. SLE involves the formation of immune complexes which results in a constellation of joint, skin and renal pathology. In over 60% of SLE patients the kidney is participated. The renal manifestations of SLE are extremly heterogeneous and may affect all renal compartiments, including glomeruli, tubules, interstitium and blood vessels.

The treatment with proteasome inhibitors (PI) seems to be a new therapeutic option. As shown in previous studies this kind of therapy can prolong life of SLE-mice and stop proteinuria. The renal morphology of mice treated with proteasome inhibitors is nearly normal.

The aim of the present study was to investigate wheter the benefical effects of PI is due to specific effects on podocytes and tubular epithelial cells. Especially markers for podocytes (nephrin, WT-1), the cytoskeletal (actin, ZO-1) and proteins like HIF1-α (Hypoxia -Inducible Factor) which are regulated via the proteasome were investigated.

Methods: Differentiated K8-cells (immortalized mouse podocytes from immorto-mouse; P. Mundel) were cultured with 5% serum from lupus mice without or with 10nM PI for 16h. As a control K8-cells were also cultured with 5% serum from non-lupus mice without or with 10nM PI for 16h. For immunfluorescence studies the cells were grown on cover slips an fixed at the end of the treatment. The podocytes were stained with nephrin and actin. HKC-8 (human derived renal proximal tubular cell line) were grown and then treated with 10nM, 20nM, 50nM and 100nM PI for 24h. Whole-cell lysates were prepared and subjected to SDS-polyacrylamide gel electrophoresis. A 24h hypoxia cell lysate and an untreated cell lysate served as control. Immunoblotting was performed using a HIF1-α antibody.

Results: Nephrin-expression of podocytes was destroyed by serum from lupus mice. In combination with a PI expression was stabilized. Serum from non-lupus mice had no effect on nephrin-expression. This in vitro result validate our in vivo findings in MRL/lpr-mice with SLE-nephritis

that nephrin-expression is nearly normal after treatment with a proteasome inhibitor.
Podocytes incubated with serum of lupus mice and a proteasome inhibitor showed a rearrangement of the structure of the actin-filament and an upregulation of actin-expression.
Podocytes incubated with non-lupus serum and PI showed a rearrangement in the structure of the actin-filament.
In renal proximal tubular epithelial cells PI was found to stabilize HIF1α-expression.
Conclusions: The above mentioned data argue for direct and specific effects of PI on podocytes and proximal tubular epithelial cells in addition to systemic effects on long-living plasma cells.

Su358 ★ PLA2R SPECIFIC AUTOANTIBODIES AS DIAGNOSTIC TOOL IN PATIENTS WITH MEMBRANOUS NEPHROPATHY (MN)

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Introduction and Aims: Background
Idiopathic membranous nephropathy (IMN) is the leading cause of nephrotic syndrome in Caucasian adults. One third of patients show progressive loss of renal function and may reach end-stage renal failure. Recently it was shown that up to 70% of patients with IMN exhibit autoantibodies (AB) directed against the M-type phospholipase A2 receptor (PLA2R). The PLA2R is in the kidney exclusively expressed on glomerular podocytes and the AB binding to this receptor might be involved in disease induction. Detection of AB in patients with IMN may delineate those patients which need immunosuppressive therapy in order to reduce proteinuria and protect loss of renal function. AB titration could further be a tool to monitor therapy effectiveness in these patients.
Methods: In 9 patients with biopsy proven MN, PLA2R-specific AB in serum were measured by indirect immunofluorescence (IIF). A cDNA encoding full-length PLA2R isoform 1 was used for transient transfection of HEK293cells. Cells were fixed and used as substrates for IIF. Antibody titers of follow-up samples from 9 patients with IMN were determined. Following AB measurement, in addition to supportive therapy (1 patient with ACE-inhibitor, 4 patients with ACE-inhibitor + angiotensin receptor blocker; 1 patient with angiotensin receptor blocker + direct renin-inhibitor; 7 patients with diuretics; 7 patients with statins; 4 patients with anti-coagulation-therapy) or immunosuppressive therapy (4 Patients with glucocorticoids, 1 patient with glucocorticoids + CNI) all patients were treated with rituximab and AB-Titers as well as 24h-proteinuria were measured after 1 week, 4 weeks and every 3 months afterwards.
Results: PLA2R-specific AB were detected in serum of 5 from the 9 patients tested. Following rituximab immunosuppressive therapy, there was a decrease in the PLA2R-AB levels in three patients within 1 week. In the other two cases the levels were constant over 3 months following rituximab therapy. Proteinuria decreased in patients which were positive for PLA2R-specific antibodies from 10046±2754 mg/24h to 7328±5756 mg/24h over a period of 3 months. The effect was greatest in patients who had a decrease in AB-Titer. No significant decrease of proteinuria was detected in patients where PLA2R-specific antibodies were undetectable.
Conclusions: In biopsy proven MN, 56% of the patients exhibited anti-PLA2R AB in the serum. These Patients had a better response to therapy (i.e. proteinuria) than patients negative for anti-PLA2R antibodies. These results, in a small collective, indicate that PLA2R AB in MN may help to guide therapeutic decisions. This has to be proven, however, in a large cohort of patients.

Su359 PLASMA EXCHANGE FOR RENAL VASCULITIS: A META-ANALYSIS

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Introduction and Aims: Plasma exchange (PLEX) may be effective adjunctive treatment for anti-neutrophil cytoplasm antibody associated vasculitis (AAV) with renal involvement. We performed a systematic review and meta-analysis of randomized control trials (RCTs) of PLEX for AAV.
Methods: We searched electronic databases (Medline, EMBASE, and CENTRAL), bibliographies, and contacted experts to identify relevant RCTs. Two reviewers independently selected RCTs comparing standard care with standard care and adjuvant PLEX in adults with AAV or idiopathic rapidly progressive glomerulonephritis. Two investigators abstracted data on study, patient, treatment and outcomes. The primary outcome was the composite of end-stage renal disease or death.
Results: We identified 9 trials including 387 patients. In a fixed-effects model the pooled relative risk of ESRD or death was 0.80 for patients treated with adjunctive PLEX compared to standard care alone (95% confidence interval [CI] 0.65 to 0.99; p=0.04). No significant heterogeneity was detected (p=0.45; I2=0%). The effect of PLEX did not differ significantly across the range of baseline serum creatinine values (p=0.74) or number of PLEX treatments (p=0.83). The relative risk for ESRD alone was 0.64 (95% CI 0.47 to 0.88; p=0.006) while the relative risk for death alone was 1.01 (95% CI 0.71 to 1.4; p=0.98). Although the primary result was statistically significant by conventional measures, there are insufficient patients studied to make reliable conclusions. Three trials included patients with pulmonary hemorrhage but there was insufficient detail to analyze this subgroup.
Conclusions: PLEX may reduce ESRD and death in patients with renal AAV but further evidence is required to advocate widespread use or accurately identify patients most likely to benefit given the limited data available and the procedure's cost and invasiveness.

Su360 IMMUNOABSORPTION FOR RELAPSES OF FOCAL SEGMENTAL GLOMERULOSCLEROSIS IN THE RENAL ALLOGRAFT

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Introduction and Aims: To assess the effect of immunoabsorption for relapses of Focal Segmental Glomerulosclerosis (FSGS) in the renal allograft
Methods: Patients were entered if were kidney transplants recipients with FSGS as primary disease. Initial treatment included daily sessions of immunoabsorption, followed by gradual tapering until discontinuation. Complete remission was defined a reduction in proteinuria to <500 mg/day. Partial response in patients presenting with nephrotic range proteinuria was defined as a reduction of ≥50 percent, and ideally to less than 3.5 g/day.
Results: Eighteen renal allograft recipients with FSGS as primary disease were included. Mean age at transplantation was 30.9 (±11.88) years and they were on dialysis for 37 (±42) months. Seven grafts were from deceased and 11 from living related donors, with a mean age 46.9 (±14.89) years. In 12/18 (66.7%) patients, FSGS relapsed 1.3 (±2.22) months post transplantation with a mean urine excretion of 7.2 (±3.9)g/day. A mean of 18.5 (±7.8) sessions of immunoabsorption were performed per patient. All patients responded to therapy, 7 (58.3%) achieved complete remission and 5 (41.6%) partial remission within 2.58 (±1.44) months. 24 hour urine protein excretion was found significantly declined [1.7 (±0.52)g/day, (p=0.001)] 3 months post treatment initiation. Nine of the twelve patients who relapsed (75%) had a new flare of FSGS upon tapering immunoabsorption. These patients received a new course of immunoabsoprtion sessions, [mean total 105 (±95.9) sessions per patient]. Four of them (44.4%) entered complete sustained remission, 1 (11.2%) partial remission and 4 (44.4%) reached ESRD. During a follow up time of 48.8 (±27.3) months, 7 (58.3%) of the relapsers remain in complete continuous remission and 11/18 (61.1%) of the grafts are functioning
Conclusions: Immunoabsorption can be an effective treatment for relapses of FSGS in renal allograft recipients.

Su361 SIGNIFICANCE OF ANTI-C1q AUTOANTIBODIES IN LUPUS NEPHRITIS

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Introduction and Aims: Autoantibodies (Ab) against C1q (anti-C1q) have received much interest in the recent years. Anti-C1q Ab have been found in a number of autoimmune diseases. A series of studies reported that serum anti-C1q Ab were common in sera from patients with systemic lupus erythematosus (SLE) and were associated with renal involvement.
The aim of this study was to assess the anti-C1q Ab in the serum from 54 patients with lupus nephritis with active and nonactive form of the disease. This study also aims to look for correlations with other clinical and laboratory parameters and with renal pathological characteristics and disease activity. The prognostic significance of anti-C1q Ab was evaluated.
Methods: Sera from 54 patients (49 females and 5 males) with lupus nephritis were collected from peripheral blood at the time of renal biopsy and after mean 64,2 months (from 56 to 77 months). All the sera were stored at -70 ^{0}C until use. All patients were tested by enzyme linked immunosorbent assay using a modification of the method of Wisnieski and Jones. Whole C1q was purified from human plasma by the method of Tener et al. All the patients fulfilled the 1997 ACR revised criteria for SLE. The disease activity was assessed by SLEDAI. Renal histopathology was classified according to the 2003 revised criteria for glomerulonephritis in SLE by the ISN/RPS.
Results: 43 patients were reported as having positive baseline serum anti-C1q Ab. The prevalence of anti-C1q antibody in patients with diffuse proliferative renal lesions (class IV) was significantly higher than in patients with non-diffuse proliferative renal lesions (class II +III) and those with membranous lesions (class V). There was a positive correlation between the presence of anti-C1q Ab and SLEDAI (r=0,71; P<0,001), activity indices (r=0,53, P<0,001), proteinuria (r=0,49, P<0,01), glomerular leucocyte infiltration (r=0,38, P<0,001), kariorrhexis and fibrinoid necrosis (r=0,59, P<0,001), endocapillary hypercellularity (r=0,41, P<0,001). The correlation with SLICC damage index (r=0,46) showed only a bordering significance (p=0,052), which indicates a poor and marginal prognostic value of those tests for the prediction of a severe progressive disease. Levels of the C1q antibodies correlate inversely with levels of other complement system (C3 and C4).
Conclusions: Anti-C1q Ab closely associated with diffuse proliferative lesions. The anti-C1qAb could be used as a marker for disease course monitoring and for determine the treatment strategy and the prognosis.

Su362 OUTCOME OF IDIOPATHIC MEMBRANOUS NEPHROPATHY AND NEPHROTIC SYNDROME IN THE MODERN ERA

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Introduction and Aims: We aimed to analyse the long-term outcome of patients with idiopathic membranous nephropathy (IMN) and nephrotic syndrome (urine protein:creatinine ratio [uPCR] >300mg/mmol, serum albumin <35g/L) in the era of evidence-based anti-proteinuric and immunosuppressive therapies and to compare immunosuppressive regimens.
Methods: 96 adults with nephrotic syndrome secondary to IMN, diagnosed between 01/03/1997 and 01/09/2008, were identified from the electronic patient record. Baseline and follow-up uPCR, eGFR and medication were recorded. Outcome was assessed as time to death, renal replacement therapy (RRT), partial remission (PR) (uPCR 30-300mg/mmol), complete remission (CR) (uPCR<30mg/mmol) and, in those achieving remission, time to relapse (uPCR >300mg/mmol), by Kaplan Meier analysis, censored at 01/09/2009.
Results: At diagnosis, mean age was 61.3 (±15.2) years, 74% were male, 54.2% had eGFR> 60ml/min and median uPCR was 934mg/mmol (IQR 619-1332). 93.8% received inhibitors of the renin-angiotensin axis, 79.2% statins, 69.8% antiplatelet agents and 38.5% immunosuppression. Median follow up was 944 days (IQR 507-2169). Actuarial 1 and 5 year patient survivals were 89.3 and 79.3% respectively. 1 and 5 year RRT rates were 1.1 and 13.9%. 1 and 5 year PR rates were 22.8 and 80.2%. 1 and 5 year CR rates were 0 and 30.0%.
37 patients (38.5%) received immunosuppression; 11 received a maximum of 6 alternating months of corticosteroid and cyclophosphamide/chlorambucil (Ponticelli regimen), 26 received ciclosporin (usually with low dose prednisolone) and 8 (21.6%) were treated with both immunosuppressive regimens successively. The median time from diagnosis to starting immunosuppression was 244 days (IQR 134.5-435). The 1 and 5 year actuarial PR rates in patients who received immunosuppression were 19.4% and 78.7% respectively and the CR rates were 0% and 23.2% respectively with no difference between the regimens. In immunosuppressed patients, 50% relapsed within 5 years (compared with 10.2% in non-immunosuppressed patients who achieved remission). There was no significant difference in relapse rates between cyclophosphamide and ciclosporin based regimes. Ciclosporin was associated with a longer duration of treatment (median 425 days vs. 122 days) and more dose adjustments, but was less likely to be discontinued due to side-effects.
Conclusions: In the era of evidence-based antiproteinuric and immunosuppressive therapy, approximately 80% patients will achieve at least partial remission within 5 years. In patients selected for immunosuppression we found no difference in remission rates comparing the Ponticelli regimen and ciclosporin, but ciclosporin appeared better tolerated.

Su363 EFFECTS OF MIZORIBIN COMBINED WITH PREDNISOLONE FOR THE TREATMENT OF IDIOPATHIC MEMBRANOUS NEPHROPATHY WITH STEROID RESISTANT NEPHROTIC SYNDROME

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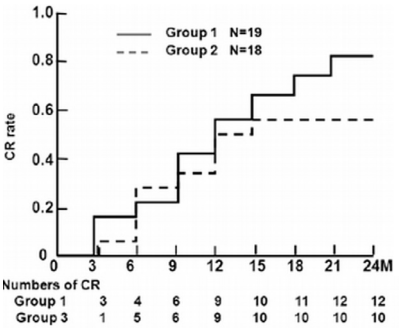
Introduction and Aims: Recently, mizoribine (MZR), one of purine synthesis inhibitors, has been used as a safe immunosuppressive agent for renal transplantation and glomerular diseases in Japan. In steroid resistant nephritic syndrome (SRNS), however, the optimal dose and administration times remain controversial issues. Therefore, we designed a randomized controlled trial for comparing the effects between once-a-day and routine three times-a-day administrations of MZR in combination therapy with prednisolone for idiopathic membranous nephropathy (IMN) with SRNS.
Methods: Patients with SRNS diagnosed as IMN by renal biopsy were registered from renal centers in Japan between 2004 and 2007. The cases were divided prospectively and randomly into 2 groups. Combined administration of PSL and MZR was continued for 2 years. PSL was initially prescribed at 40mg/day for 4 weeks and tapered to 30mg/day when MZR started. In Group 1, MZR was given once a day after breakfast at 150mg. In Group 2, MZR was given three times a day at 50mg each. Accordingly, a total dosage per day was the same in both groups. Biochemical data including 24 hours urine protein (UP) were assayed every 3 months. Remission status of nephrotic syndrome and renal function were indicated by UP and creatinine clearance, respectively.
Results: Nineteen and 18 cases were enrolled in Groups 1 and 2, respectively. By 2 years, 12 cases in Group 1 and 10 cases in Group 2 achieved complete remission (CR), but relapses occurred in one of Group 1 and 3 of Group



2. Logistic regression showed that administration times per day influenced CR ratio at final observation ($p<0.05$). Non-parametric analysis by Kaplan-Meier method showed that cumulative probability of CR was higher in Group 1 (figure), although that is not significant. Six in Group 1 and one in Group 2 were withdrawn from the protocol mainly by patients' removal. No patients suffered from fatal complications and renal dysfunction.
Conclusions: MZR is a safe and beneficial medicine for the treatment of IMN with SRNS. In particular, one-a-day administration may provide more favorable effects compared with a routine three times-a-day administration.
Disclosure: This study is supported in part by the Kidney Foundation, Japan.

Su364 A DOUBLE-BLINDED PROSPECTIVE RANDOMISED STUDY ON THE EFFICACY OF CORTICOSTEROID PLUS CYCLOPHOSPHAMIDE OR FK506 IN IDIOPATHIC MEMBRANOUS NEPHROPATHY PATIENTS WITH NEPHROTIC SYNDROME

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Introduction and Aims: Idiopathic membranous nephropathy (IMN) is the most common cause of adult-onset nephrotic syndrome (NS) and the management of IMN is still controversial. The aim of this study was to compare the efficacy and the drug safety of cyclophosphamide to FK506 with corticosteroid in IMN patients.
Methods: From 2006 Nov to 2008 Jan, twenty-four patients, which had severe NS (admission 24-hour urinary protein excretion >5 g or Albumin < 25g/L), or had renal dysfunction, with primary IMN were enrolled to this study prospectively.
Results: Thirteen patients received cyclophosphamide therapy combined with corticosteroid therapy (CTX group, M9/F4, average age 54.6±13.5 ys), while the other eleven patients were treated with FK506 plus corticosteroid (FK506 group, M6/F5, average age 55.0±13.5ys). Two groups did not differ with respect to their laboratory features at the time of admission. After 2 years, only two patient of the FK506 group were dropped out, other 22 patients were followed up. As the result, 5 of 13 patients of CTX group (38.5%) achieved complete remission (CR) and 8 of them (61.5%) had part remission (PR). On the other hand, 9 of 11 patients (81.8%) of FK506 group achieved CR and 1 of 11 patient of FK506 group (9.1%) achieved PR who existed after 12weeks' treatment. There was significant difference between the two groups (CTX and FK506) in CR rate.($P<0.05$) FK506 reduced proteinuria of IMN patients earlier (about 3 months) and more quickly than CTX. The average proteinuria in CTX group after 3months' treatment was 2.4±1.7g/24h, while which was 1.4±1.1g/24h in FK506. After 9 months, the average proteinuria was 1.5±1.2g/24h in CTX and 0.3±0.29 g/24h in FK506 group. The average albumin was 26.0±6.0g/L and 31.4±5.9 g/L respectively in CTX and FK506 group after 3 months. Furthermore, CTX group had more adverse events, including urinary tract infection (CTX 3 pts), pulmonary infection (CTX 2 pts), liver dysfunction (CTX 2 pts), hyperglycemia (CTX 2 pts, FK506 3pts).
Conclusions: These results suggest that long-term corticosteroid combined with FK506 therapy is more beneficial for controlling proteinuria and increase serum albumin than corticosteroid combined with CTX in patients with MN. Moreover, the drug safety of FK506 was better than CTX.

Su365 ALTERATIONS IN CELLULAR IMMUNE PARAMETERS AFTER RITUXIMAB ADMINISTRATION IN PATIENTS WITH ANCA-ASSOCIATED VASCULITIS AND SYSTEMIC LUPUS ERYTHEMATODES

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Introduction and Aims: Rituximab (RTX), an anti-CD20 monoclonal antibody, has recently emerged as a novel therapy for autoimmune diseases, including ANCA-associated vasculitides (AAV) and systemic lupus erythematodes (SLE). RTX leads to selective transient depletion of CD20+ B cell subpopulation and likely affects also T cell functions but the exact mechanisms remain to be elucidated. Evolution of cellular immune parameters after rituximab administration in AAV and SLE patients has been prospectively followed-up in this study.
Methods: From December 2004 to January 2010, RTX was administered to 8 patients with AAV (M/F ratio 7/1; median age 39 yrs; 6x refractory AAV, 2x new diagnosis; repeated administration in 1 patient) and to 6 patients with refractory SLE (M/F ratio 1/5; median age 35 yrs; repeated administration in 3 patients). Using flow cytometry, the following markers were regularly assessed in peripheral blood cells: surface molecules (CD3, CD4, CD8, CD19, CD20, CD45RA, CD80, CD86, CD28, HLA-DR) and intracellular cytokines (IFNgamma, TNFalpha, interleukin (IL)-2 and IL-4 in CD3+ cells and IL-10 and IL-12 in monocytes).
Results: At least partial remission was achieved in all patients with SLE and 5 patients with AAV. Long-term B cell depletion lasting 4-30 months (median 10.5 months) was observed in all patients. The length of B cell depletion tended to be higher in AAV patients (median 15, range 8-30 months) than in SLE (median 6, range 4-18 months). Baseline levels of HLA-DR+ (active) CD3+ T cells were increased in both AAV and SLE. However, while in SLE patients the numbers of HLA-DR+ CD3+ cells at 3 months after RTX remained stable, a significant increase in HLA-DR+ CD3+ cells (10.7% of lymphocytes at administration vs 25.7% at 3 months, $p<0.05$) was observed in AAV patients. The numbers of CD4+CD45RA+ naive T cells tended to increase after RTX in SLE (25.4% of lymphocytes at baseline vs. 31.0% at 3 months), but decreased in AAV (26.7 vs. 19.5%). An increase in IL-10 production at 3 months after RTX administration seemed to be associated with a favourable treatment outcome in both SLE and AAV patients.
Conclusions: Our results suggest different rituximab effects on T cells in patients with AAV and SLE. The increase in the production of IL-10 with its immunosuppressive and immunoregulatory potential might indicate a positive outcome of RTX treatment in autoimmune diseases.

Su366 CLASS IV-S VERSUS CLASS IV-G LUPUS NEPHRITIS

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Introduction and Aims: The new ISN/RPS classification of lupus nephritis divides diffuse proliferative lupus nephritis into two subcategories with predominantly segmental proliferative lesions (class IV-S) and those with predominantly global proliferative lesions (class IV-G). This study explores the validity of this distinction and possible differences in pathogenesis between the 2 types of lesions
Methods: A retrospective analysis of biopsy-proven 231 cohort of patients with lupus nephritis using ISN/RSP classification was performed. Clinical data were available on all patients selected.
Results: The prevalence of Class IV was 27,27%. Of patients with class IV lupus nephritis, 41 had class IV-S and 22 had class IV-G. The mean age was 33,87±10,39 years. The serum creatinine levels (185,2±138,7 μmol/l vs. 114,0±64,14 μmol/l), proteinuria (5,54±4,69 g/24 h vs 3,22±2,26 g/24 h) and diastolic blood pressures (104,12±10,45 mmHg vs. 96,42±13,12 mmHg) were significantly greater in the IV G group, but haemoglobin was significant lower (102,8±13,64 g/l vs. 115,9±12,48 g/l). Duration of systemic lupus erythematosus were similar in the 2 groups (mean 45,73±22,13 months). Histologically combined lesions with segmental endocapillary proliferation and fibrinid necrosis were more frequent in the IV-S class lupus nephritis. The percentage of glomeruli with cellular crescents also was greater in the IV-S group (28,24% vs 23,88%), but the difference was not significant. No significant difference was detected in outcomes in the 2 groups after follow ups of 145,2±76,87 months.
Conclusions: There are definite clinical and morphologic differences between class IV-S and IV-G lesions. Data suggest that class IV-G lesions behave as an immune complex disease, however, in class IV-S lesions, the presence of proportionally greater glomerular fibrinoid necroses suggest that these lesions may have a different pathogenesis.

Su367 NEUTROPHIL GELATINASE-ASSOCIATED LIPOCALIN (NGAL) AS A BIOMARKER FOR RENAL INJURY IN CHRONIC GLOMERULONEPHRITIS (CGN)

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Introduction and Aims: NGAL is a part of the lipocalin protein family and is a 23 kDa low molecular weight protein secreted by various types of human cells. Its physiologic role seems complex, implying cell growth and differentiation, a bacteriostatic immune effect and a role in cellular iron-transport pathways. By virtue of its small size NGAL is freely filtered by the renal glomeruli without being reabsorbed, and can therefore be measured in the urine. NGAL is strongly expressed in renal tubules in animal models of renal ischemic injury. Later clinical studies demonstrated role of urinary and plasma NGAL as a new early and sensitive biomarker of acute kidney injury. Role of NGAL in chronic kidney disease is not clear yet. The aim of our study was to estimate excretion of NGAL in patients with CGN and its possible association with clinical activity, degree of GFR decrease and duration of the chronic kidney disease.

Methods: 77 CGN patients were studied: age (M (95% CI)) 39 (36; 43), M/F ratio 36/41, proteinuria 2.2 (1.6; 2.9) g/day, nephrotic syndrome in 24%, GFR by Cockroft-Gault formula adjusted for body surface area (eGFR) 73 (65; 82) ml/min/1.73 m^2. Urinary NGAL level was measured by ELISA (R&D Systems).

Results: In total group mean urinary NGAL level was 8.8 (6.7; 11.0) ng/ml. Urinary NGAL level correlated with systolic blood pressure (Rs=0.23, $p<0.05$), number of antihypertensive drugs used (Rs=0.25, $p<0.05$), interventricular septal thickness (Rs=0.46, $p<0.05$), proteinuria (Rs=0.45, $p<0.0001$), serum albumin level (Rs= -0.52, $p<0.0001$). There was no correlation of urinary NGAL and eGFR. But in patients with 4-5 CKD stage urinary NGAL level was significantly higher vs 1-2 CKD stage patients. At the same time in 4-5 CKD stage patients combination of high proteinuria and sever hypertension was found.

Urinary NGAL level in different CKD stages

	1-2 stage CKD	3 stage CKD	4-5 stage CKD
Systolic BP, mm Hg	132 (127;137)	144 (136; 151)	148 (130; 166)
Proteinuria, g/d	2.5 (1.6; 3.4)	1.6 (0.7; 2.5)	2.6 (1.0; 4.2)
eGFR, ml/min/1.73 m^2	94 (85; 103)	45 (41; 49)	18 (11; 26)
Urinary NGAL	7.1 (4.7; 9.6)	10.0 (5.9; 14.0)	14.2 (2.8; 25.5)*

*$p<0.05$ vs 1-2 CKD.

In multiple regression analysis proteinuria was the only independent factor for urinary NGAL.

Conclusions: Urinary NGAL level in CGN patients can be considered an integrative biomarker of chronic kidney injury. It correlates with main factors of clinical activity and progression of CKD; association with proteinuria is the strongest. It can be proposed that urinary NGAL level reflects proteinuric tubulointerstitial remodeling in CGN.

Su368 CIGARETTE SMOKING ABROGATES A RENOPROTECTIVE EFFECT OF RENIN-ANGIOTENSIN-ALDOSTERONE SYSTEM (RAAS) BLOCKADE IN PATIENTS WITH IgA NEPHROPATHY

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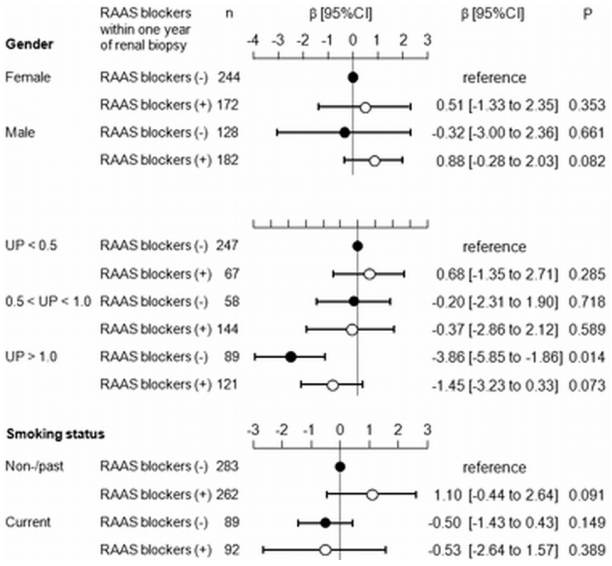
Introduction and Aims: Although renin-angiotensin-aldosterone system (RAAS) blockade has been regarded as one of the best evidence-based treatments of IgA nephropathy (IgAN), individual responses to RAAS blockade were diverse and, therefore, predictors of effectivenss of RAAS blockers should be identified. The present study assessed predictive power for renoprotective effect of RAAS blockers of gender, urinary protein, and smoking status.

Methods: Multicenter retrospective observational cohort study in three nephrology centers in Osaka, Japan. *Participants:* 726 patients diagnosed with IgAN by renal biopsy between 1992 and 2005 with ≥ six months of follow-up period. *Covariates:* Baseline characteristics at renal biopsy (age, gender, BMI, blood pressure, use of antihypertensives, eGFR, urinary protein, uric acid, total cholesterol, and smoking status (non-/past vs. current smokers)) and therapeutic interventions (RAAS blockers and corticosteroid). *Outcome:* ΔeGFR (mL/min/1.73m^2/year) calculated based on eGFR at renal biopsy and the end of observation before initiating renal replacement therapy (RRT). *Statistics:* Interactions between RAAS blockers and potential predictors of effectiveness of RAAS blockers (gender, urinary protein, and smoking status) were assessed in multivariate linear regression models.

Results: Baseline characteristics at renal biopsy: age 37±13 years, male 42.7%, systolic/diastolic blood pressure 122±17/75±13 mmHg, use of antihypertensives 18.9%, eGFR 79±24 mL/min/1.73m^2, urinary protein 0.8±1.0 g/day, current smokers 24.9%. Therapeutic interventions: RAAS blockers 48.8%, corticosteroid 30.0%. During 7.6±4.4 years of observational period 38 patients initiated RRT. Multivariate model demonstrated that urinary protein (per g/day, β -1.40 [95%CI -2.07 to -0.72, P=0.013) and current smokers (β -1.07 [-1.79 to -0.35], P=0.024) were significantly associated with ΔeGFR. Interestingly, smoking status was identified as significant predictors of effectiveness of RAAS blockers (P=0.027 for interaction (RAAS blockers*current smokers)), along with urinary protein (P=0.007 for interaction (RAAS blockers*urinary protein)), whereas not gender (P=0.621 for interaction (RAAS blockers*gender)). Effectiveness of RAAS blockers was blunted in current smokers and the patients at lower level of urinary protein, especially, less than 1g/day.



Conclusions: Smoking status predicted renoprotective effect of RAAS blockers in IgAN, along with urinary protein.

Su369 THE CLINICAL UTILITY OF PROTOCOL BIOPSY IN SYSTEMIC LUPUS ERYTHEMATOSUS

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Introduction and Aims: Renal involvement is common in systemic lupus erythematosus (SLE). Clinical and laboratory analyses are poor predictors of class and disease activity. The situation is further complicated as several case reports documented transformation or progression from one class to another when a second biopsy is performed. The aims of this study are to

systemically assess the response to therapy and examine the change in the classes of SLE nephritis based on a protocol kidney biopsy.
Methods: Patients with Lupus Nephritis were advised to undergo a second kidney biopsy at end of maintenance therapy. All renal biopsies were reclassified according to the new criteria set by the International Society of Nephrology and the Renal Pathology Society Working Group (ISN/RPS 2004 Classification). The pathologists were blinded to the clinical data and to the sequences of the kidney biopsies.
Results: Seventy three patients were included in the study. The mean age at renal disease onset was 28.5±10.4 years and 10 of the patients were males. Histological examination of the initial renal biopsy specimen showed Class II lupus nephritis, 7 patients; Class III lupus nephritis, 26 patients; Class IV lupus nephritis, 27 patients; Class V lupus nephritis, 12 patients and Class VI lupus nephritis, 1 patient. Class V lupus nephritis was mixed with proliferative in 6 patients. The second kidney biopsy showed transformation to another histological pattern in 61% of the cases. Twenty one patients (27%) showed histological improvement and nineteen patients (26%) demonstrated histological deterioration. The activity index at the time of the second biopsy showed significant correlation with only serum complement-3 and anti-double stranded DNA levels (P value < 0.001 and 0.003, respectively). Proteinuria and urine red blood cells correlated weakly with activity index (r =0.26, P =0.04; r =0.25, P =0.04, respectively). However, there were no correlation between the activity index and the serum creatinine (r =0.11, P =0.37).
Conclusions: Protocol biopsy in SLE nephritis appears to have clinical utility, as the majority of the patients had a change in their histological findings on the second biopsy and a poor correlation was noted between the clinical and biochemical parameters on one hand and the histological activity of the disease on the other hand.

Su370 RENAL BIOPSY FINDINGS IN VERY ELDERLY PEOPLE (>80 YEAR-OLD): DATA FROM SPANISH REGISTRY GLOMERULONEPHRITIS

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Introduction and Aims: Nowadays, it is more and more frequent to look after patients with advanced age. The findings obtained from renal biopsies in very elderly people need to be expanded with data obtained from large scale registries. The purpose of this study is to know the prevalence of biopsied renal diseases in patients older than 80 years.
Methods: We have reviewed renal biopsies in from Spanish Registry Glomerulonephritis.
Results: From 1994 to 2008, 382 renal biopsies have been registered in patients older than 80 years, which constitutes 2.4% of the total biopsies, with a considerable increase in the last years (2007: 40%, 2008: 3.2%). There is predominance of males through the period of study (ratio M/F 1.3). The prevalence of hypertension is 58% and the median age 82 years. The median of creatinine and proteinuria at the moment of renal biopsy are 3.6 mg/dl and 2.8 g/d, respectively. More than 50% of the cases have creatinine greater than 3 mg/dl and proteinuria greater than 3 g/d. The main clinical syndrome was acute kidney injury (42.5%) followed by nephrotic syndrome (31.2%), chronic renal failure (10.6%), nephritic syndrome (8.7%), asymptomatic urinary abnormalities (5.7%), macroscopic haematuria (1.1%) and hypertension (0.3%). Renal vasculitis and crescentic glomerulonephritis type 3 are the main renal diseases (21.7%) whereas other renal disorders are less frequent: membranous nephropathy (10.5%), amyloidosis (8.9%), acute interstitial nephritis (6.5%), minimal change disease (6%), remaining crescentic glomerulonephritis (6%) and nephroangiosclerosis (5.8%); diabetic nephropathy is only 3.9%. When we match clinical syndromes against renal histopathology, we found that the first cause of acute kidney injury is renal vasculitis and crescentic glomerulonephritis (33%) whereas nephrotic syndrome is due mainly to membranous nephropathy (32%) and minimal change disease (17%). Moreover, we have found that the renal diseases mentioned above are significantly more frequent in patients older than 80 years compared with elderly people who are between 65 and 80 years.
Conclusions: We conclude that in Spain the percentage of very elderly people in whom renal biopsy is performed is increasing, because of acute kidney injury of uncertain origin and nephrotic syndrome. The renal histopathological findings show the predominance of autoimmune glomerulonephitis whose diagnosis, even at advanced age, could modify the management and prognosis.

Su371 ASSOCIATION BETWEEN THE INTERLEUKIN-1β GENE (IL1B) C-511T POLYMORPHISM AND THE RISK OF PROGRESSIVE IgA NEPHROPATHY

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Introduction and Aims: Interleukin-1 (IL-1) has been previously shown to modulate mesangial cell proliferation and extracellular matrix synthesis, that both are commonly observed in IgA nephropathy (IgAN). IL-1 cluster genes are, therefore, potentially involved in the pathogenesis of DN. The IL-1β gene (IL1B) contains a polymorphism in position -511 (C/T), which is located in the promoter region and seems to modulate IL-1β gene transcription. In order to investigate the association between this polymorphism and IgAN, a case-control study was performed in a Caucasian population.
Methods: Study population consisted of 121 unrelated patients with IgAN diagnosed histologically. Progressive IgAN was defined as persistent proteinuria (>2000mg/24h) and/or renal function impairment (serum creatinine>1.5mg/dl). Progressive IgAN was present in 67 subjects (cases) and non-progressive in 54 subjects (diseased controls). Genotyping was performed using polymerase chain reaction – restriction fragment length polymorphism assay. The comparison of genotype distributions was performed by chi-squared test. Additive (i.e. contrast of homozygotes), recessive, dominant and co-dominant model two by two association was tested by the Fisher's exact test, and expressed as an odds ratio (OR) with corresponding 95% confidence intervals (95% CI).
Results: There was a significant difference in the genotype distribution between the cases with progressive IgAN versus diseased controls (p=0.041). Further analyses showed that the risk of progressive IgAN is significantly enhanced in IL1B T-allele carriers (dominant model) and in heterozygotes (co-dominant model): OR=2.56 (1.22-5.36) (p=0.016) and 2.27 (1.09-4.75) (p=0.043) respectively. However, non-significant results were obtained by testing the recessive (p=0.77) and the additive (comparison of homozygotes; p=0.23) models.
Conclusions: The findings provide evidence for the C-511T IL1B gene polymorphism as a genetic trait contributing to an increased risk for progressive disease in Caucasian patients with IgAN.

Su372 PREDICTIVE VALUE OF URINARY ALBUMIN-TOTAL PROTEIN RATIO FOR PROGRESSIVE RENAL DISEASE IN PATIENTS WITH HEMATURIA AND MILD PROTEINURIA

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Introduction and Aims: Many patients with microscopic hematuria have glomerular disease, most commonly IgA nephropathy (IgAN) or thin basement membrane nephropathy (TBMN). TBMN is considered as benign disease and other glomerular disease is considered as progressive renal disease. However, differential diagnosis between progressive renal disease and benign disease is difficult especially in patients with mild proteinuria. Thus, we evaluated the value of urinary albumin-total protein ratio (APR) as a predictor for progressive renal disease in patients with hematuria and mild proteinuria.

Methods: We measured spot urine albumin-creatinine ratio (ACR) and protein-creatinine ratio (PCR) in patients with hematuria and 24-h urinary total protein less than 1,000 mg/day. According to pathologic findings, normal pathology and TBMN were considered as a benign disease (Ben-Group). Progressive renal disease was defined as glomerular disease other than TBMN (Pro-Group).
Results: Among the 108 patients, 71 (65.7%) were diagnosed as IgAN, 16 (14.8%) as normal, 11 (10.2%) as TBMN, 5 (4.6%) as focal segmental glomerulosclerosis, 5 (4.6%) as membranous nephritis, respectively. Of them, 27 (25.0%) were classified as Ben-Group and 81 (75.0%) as Pro-Gorup. Mean 24-h urinary total protein and APR were significantly increased in the Pro-Group compared with Ben-Group (465.5±284.1 mg/day vs. 190.5±165.6 mg/day, $p<0.001$; 0.66±0.30 vs. 0.30±0.20, $p<0.001$, respectively). The frequency of progressive renal disease was significantly higher in the patients with proteinuria $\geq$ 500 mg/day and APR $\geq$ 0.65 (94.9% vs. 63.8%, OR 10.5, 95% CI 2.3-47.4, $p<0.001$; 94.1% vs. 57.9%, OR 11.6, 95% CI 3.2-41.8, $p<0.001$, respectively). In the multivariate analysis, 24-h urinary total protein and APR were independent predictors of progressive renal disease (RR 6.2, 95% CI 1.3-29.8, p=0.022; RR 8.0, 95% CI 2.1-29.8, p=0.002, respectively). In the subgroup analysis, predictive value of APR $\geq$ 0.65 for progressive renal disease was significant in the patients with proteinuria < 500 mg/day (90.9% vs. 51.1%, OR 9.6, 95% CI 2.0-45.7, p=0.001), but not in the patients with proteinuria $\geq$ 500 mg/day (96.4% vs. 90.9%, $p>0.05$).
Conclusions: APR and 24-h urinary total protein should be considered in the differential diagnosis of patients with hematuria and mild proteinuria. APR seems to be useful especially in the patients with hematuria and minimal proteinuria.

Su373 IMPACT OF EVIDENCE-BASED THERAPY ON THE OUTCOME OF GLOMERULONEPHRITIS

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Introduction and Aims: The therapy of glomerulonephritis is based on widely accepted standards since numerous meta-analysis and several Cochrane Systematic Reviews have been published. However, the impact of such therapy standards on the prognosis of glomerular diseases is not known.
Methods: Between Oct 2002 and Dec 2008 all patients with abnormal urine findings and/or decreasing renal function of unknown cause were referred for renal biopsy (see corresponding abstract #450847). In a collaboration of out-patient nephrologists with a major teaching hospital, all patients received treatment recommendations according to evidence-based therapy guidelines based on Cochrane Systematic Reviews. Patient charts were systematically reviewed and patients were re-examined for follow-up until Nov 2009. Cox-Regression analysis was performed to identify independent prognostic factors.
Results: Two hundred patients with primary or secondary glomerular diseases were identified. Complete follow-up data were available from 196 patients with 324 therapeutic interventions. The mean follow-up was 2.8±2.0 years. Among all patients, 37% remained unchanged ill, 13% died, 17% had progressing renal disease, while 19% had a complete and 14% a partial remission. Proteinuria declined in primary glomerulonephritis (5.6 g/d vs 1.3 g/d, $p<0.001$). The highest rates of remission were observed in minimal change disease (83%) and membranous nephropathy (50%). Survival was lowest in MPGN and secondary rapid-progressive glomerulonephritis (33% and 50%, respectively). 70 (22%) interventions were complicated by adverse events resulting in treatment cessation in 25 cases. Cox univariate analyses identified the following parameters to improve outcome: Histology, no tubulointerstitial fibrosis, primary glomerulopathy, absence of hypertension at presentation, diabetes, ischemic heart disease, no diuretics or insulin, serum creatinine < 175 µmol/l, RR < 160 mmHg, age < 60 ys, prednisolone, cyclosporine A or azathioprine, and follow-up by 24 hr urine. In a multivariate forward Cox regression analysis, tubulo-interstitial fibrosis had a hazard for the combined end-point of death, dialysis and progression of renal failure of 4.4 (1.8 – 10.6) while intensive follow-up by regular 24 hr urine collections reduced the risk to 0.3 (0.1 – 0.7), treatment with prednisolone had a hazard of 0.3 (0.1 – 0.9), and cyclosporine A/azathioprine therapy a hazard of 0.2 (0.02 – 1.4).
Conclusions: In a multivariate model of standardised glomerulonephritis therapy the presence of tubulointerstitial fibrosis was associated with death or progressive renal disease, while prednisolone-based therapy regimens and intensified nephrological follow-up resulted in a significant delay of endstage-renal failure. This results should direct future health care policies because glomerular diseases account for approximately 30% of the dialysis population.

Su374 IMPACT OF BODY MASS INDEX ON CLINICAL AND HISTOLOGICAL PARAMETERS OF IGA NEPHROPATHY

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Introduction and Aims: In the present study we evaluated the influence of body mass index (BMI) on clinical and histological parameters of IgA nephropathy.
Methods: We studied n=178 patients with biopsy proven primary IgA nephropathy, followed up for a mean of 5.9±6.0. The rate of deterioration of renal function was estimated by the slope of the curve of reciprocal serum creatinine against time. According to the BMI at the time of renal biopsy, patients were divided into the following groups: normal BMI (<25, n=104), overweight (25-29.99, n=54) and obese (≥30, n=20). The biopsies of 72 patients were analysed by the same pathologist. The degree of glomerular sclerosis, tubulointerstitial fibrosis and vascular hypertrophy was evaluated.
Results: Age, initial renal function, proteinuria, blood pressure under treatment and the number of antihypertensive agents taken at the time of renal biopsy were similar among normal, overweight and obese patients (ns). Furthermore, there was no significant difference in the degree of glomerular sclerosis, tubulointerstitial fibrosis or vascular hypertrophy between patients with different BMI (ns). The rate of progression however differed significantly between patients with normal BMI (-0.092±0.116), overweight (-0.188±0.366) and obese patients (-0.211±0346 $dl*mg^{-1}$ $*year^{-1}$, p=0.020). Similarly, BMI as a continuous variable correlated with the rate of deterioration of renal function (r=0.232, p=0.002).
Conclusions: An increased BMI is associated with a faster decline of renal function in patients with IgA nephropathy.

Su375 CLINICAL AND PATHOLOGICAL PROGNOSTIC INDICATORS OF IGA NEPHROPATHY IN IRANIAN PATIENTS

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Introduction and Aims: Immunoglobulin A (IgA) nephropathy is the most common cause of primary glomerulonephritis with slow progression to end-stage renal disease (ESRD) in up to 40% of patients.
Methods: A retrospective cohort study of patients with biopsy proven IgA nephropathy was performed in our center from 1997 to 2008. We tried to determine the clinical and pathological data that were associated with the prognosis of the disease. Clinical characteristics at the time of renal biopsy

and follow-up were reviewed. Severity of histology was quantified as grade 1-3.
Results: There were 70 IgA nephropathy patients and 46 were men. The average age of patients at biopsy time was 39±12.1 years. During the median 23.5 (6-130) months of follow-up, 10 patients progressed to ESRD and no patient died. Median time of progression to ESRD was 107 (62-152) months. The renal survival was 94% at one year, 91% at three years and 88% at five years.

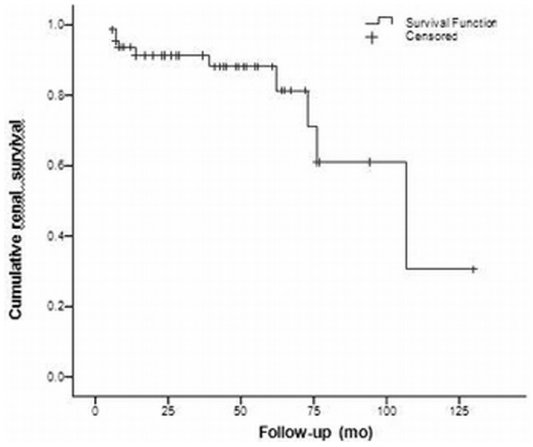

Figure 1. Kaplan-Meier renal survival curve of 70 patients with IgA nephropathy.

A higher histological grade of IgA nephropathy was associated with higher baseline age (P: 0.003), higher mean arterial pressure (P: 0.01), greater serum creatinine (<0.001), more 24-hour urine protein excretion (0.002) and higher number of ESRD events (<0.001). Odds ratio of ESRD events for patients older than 50 years was 13.5 (CI: 95% 2.9-61.7, P: 0.001), for every unit increase in serum creatinine (mg/dL) was 3.7 (1.6-8.6, P: 0.003) and for daily proteinuria more than 3 g/24h was 13.1 (2.5-69.2, P: 0.002). We had no ESRD event in patients with grade I pathology, and odds ratio for grade III versus II was 12 (2.2-64.5, P: 0.004). ESRD events also were more common in male and hypertensive patients but not at significant level.
Conclusions: Although the number of studied patients and median time of follow-up in our study is limited, we showed that kidney biopsy and risk stratification of different factors at baseline in IgA nephropathy are useful for predicting the prognosis and probably appropriate intervention.

Su376 THE PATTERN OF BIOPSY PROVEN PRIMARY GLOMERULAR DISEASES INCIDENCE IN NORTHEASTERN PART OF POLAND IN YEARS 1983-2008

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Introduction and Aims: Primary glomerular disease is one of the major causes of end stage renal disease all over the world. There is evidence, that its incidence varies according to the region and time period included in the study. Thus reports from different parts of the world are needed to get the full knowledge of its epidemiology.
Methods: All records of adult kidney biopsies performed between 1983 and 2008 in our unit were retrospectively analyzed. Data were divided into 3 time frames and evaluated with regard to incidence and time trends of primary glomerular diseases.
Results: One thousand three native adult biopsies were analyzed. Primary glomerulonephritis was found in 64% of native kidney biopsies. The most common diagnoses were: FSGS (28%), IgA nephropathy (25%), membranoproliverative glomerulonephritis (12%), membranous nephropathy (11%), crescentic glomerulonephritis (9%), non-IgA nephropathy (8%), minimal change disease (7%) and fibrillary glomerulonephritis (0.6%). There was a significant increase in percentage of FSGS ($\chi 2$=8.78, p=0.0124) and IgA nephropathy ($\chi 2$=29.96, p=0.0001), together with decrease in proportion of membranous nephropathy ($\chi 2$=17.47, p=0.0002), minimal change disease ($\chi 2$=15.07, p=0.0005) and non-IgA nephropathy ($\chi 2$=46.93, p=0.0001). The frequency of other primary glomerular diseases remained constant during the study period.
Conclusions: In contrast to other European registries, FSGS was the most common nephropathy in our region and its frequency increased over the time. The causes of this finding remain unknown and need further investigation.

Su377 SERUM RATIO OF SOLUBLE TRIGGERING RECEPTOR EXPRESSED ON MYELOID CELLS-1 TO CREATININE IS A USEFUL MARKER OF INFECTIOUS COMPLICATIONS IN MPO-ANCA-ASSOCIATED RENAL VASCULITIS

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Introduction and Aims: The contribution of infections to the mortality of ANCA-associated vasculitis patients is important and should induce early and careful control of these events. However, the differentiation of infection from active vasculitis is often difficult. The triggering receptor expressed on myeloid cells-1 (TREM-1) is a recently discovered as a member of the immunoglobulin superfamily. A soluble form of TREM-1 is being investigated as a clinical marker to distinguish sepsis from non-infectious inflammatory conditions. To distinguish active vasculitis and infectious complications, a serum-soluble TREM-1 was investigated.
Methods: Soluble TREM-1 in serum obtained from 33 patients with MPO-ANCA-associated vasculitis was measured by an enzyme-linked immunosorbent assay. Twenty-one samples were from active vasculitis patients, twenty-one samples from inactive vasculitis patients and ten samples from inactive vasculitis with infectious complications. Serum-soluble TREM-1 was also measured in eight patients with acute pyelonephritis and 8 patients with chronic kidney disease (CKD).
Results: Among patients with MPO-ANCA-associated vasculitis, the serum levels of soluble TREM-1 were higher in patients with infectious complications than in patients with inactive vasculitis (P = 0.006). However, there was no significant difference between patients with infectious complications and patients with active vasculitis. On the other hand, there was significant correlation between serum levels of soluble TREM-1 and serum creatinine levels among all patients (r = 0.559, P < 0.0001). Among patients with MPO-ANCA-associated vasculitis, the serum soluble TREM-1/creatinine ratio was higher in patients with infectious complications than in active vasculitis and inactive vasculitis (P = 0.0039, P = 0.0033, respectively). This ratio in vasculitis patients with infectious complications was also higher than that in CKD patients (P = 0.0006), but not significantly different from that in active pyelonephritis patients. There was no significant difference in the ratio among patients with active vasculitis, inactive vasculitis, and CKD patients. On receiver operating characteristic curve analysis, 77.0 μg/mg creatinine of serum TREM-1 as the lower limit had a sensitivity of 68.4% and a specificity of 86.0% to differentiate patients with infection from those without infectious patients.
Conclusions: The serum levels of soluble TREM-1 in patients with MPO-ANCA-associated vasculitis without infectious condition may depend on their renal function. The ratio of serum soluble TREM-1/creatinine may be a useful marker of infectious complications.

Su378 IMPACT OF THE FUNCTIONAL C-344T POLYMORPHISM OF ALDOSTERONE SYNTHASE (CYP11B2) GENE ON PRIMARY GLOMERULONEPHRITIS

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Introduction and Aims: In the last years aldosterone has been identified as an important mediator of renal injury. In the present study we evaluated the influence of the C-344T polymorphism of aldosterone synthase (CYP11B2) gene, associated with serum aldosterone levels and the development of arterial hypertension, on the clinical course of chronic primary glomerulonephritis.

Methods: We studied n=251 patients with biopsy proven primary glomerulonephritis (IgA nephropathy: n=127, focal segmental glomerulosclerosis: n=71, membranous glomerulonephritis: n=53) followed up for 6.2±5.3 years. According to the slope of the curve of reciprocal serum creatinine against time (≥ or< -0.1 dl * mg^{-1} * $year^{-1}$) group A (slow progressors, n=162) and group B (fast progressors, n=89) were defined. One hundred healthy volunteers were analysed as controls. Aldosterone synthase gene C-344T polymorphism was determined by PCR amplification. Aldosterone serum levels were determined in n=57 of our patients at the time of renal biopsy.

Results: The aldosterone synthase genotype correlated to the aldosterone serum levels (CC/CT: 107±70, TT 243±323 pg/ml, p<0.05). The genotype distribution did not differ significantly between our study and control populations (patients: CC/CT genotypes: 71.3%, TT: 28.7%; controls: CC/CT: 69%, TT: 31%, ns). Age, initial renal function, proteinuria and blood pressure was similar among patients with different genotypes (ns). The C-344T polymorphism was associated with the progression of primary glomerulonephritis as shown by the genotype frequencies in group A (slow progressors; CC/CT: 64.8%, TT: 35.2%) and group B (fast progressors; CC/CT: 83.0%, TT: 17.0%, p=0.002). There was also a significant difference in the actual rate of progression as estimated by the slope of the curve of reciprocal serum creatinine (CC/CT: 0.182±0.45, TT: 0.076±0.09 dl * mg^{-1} * $year^{-1}$, p=0.009).

Conclusions: Our results indicate that the functional C-344T polymorphism of the aldosterone synthase gene is an important marker of progression in patients with chronic primary glomerulonephritis.

Su379 INSULIN RESISTANCE AND RENAL INJURY IN PATIENTS WITH OBESITY AND VARIOUS POLYMORPHISM OF GENE PPARγ2

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Introduction and Aims: Obesity is a multifactor disease. Both genetic and environmental factors are implicated in its etiology. Recently increasing attention has been directed to candidate genes that participate in developing insulin resistance (IR), a key factor involved in the pathogenesis of obesity-associated nephropathy. Aim: To estimate the influence of polymorphism of gene PPARγ2 on IR, to determine the contribution of IR to the development obesity-associated nephropathy.

Methods: 64 patients were examined (44 M and 20F) with a mean age 48,9±10,9 years. All patients were divided into three groups by body mass index (BMI): I (n=35) with BMI 25,0-34,9 (mean 31,6±2,14 kg/m^2), II (n=14) with BMI 35,0-39,9 (mean 37,1±2,17 kg/m^2), III group (n=15) with morbid obesity (mean BMI 46,0±6,01 kg/m^2). The concentration of insulin was measured, HOMA-IR, identified polymorphism Pro12Ala of the PPARγ2 gene.

Results: First, the distribution of polymorphisms of the gene was estimated in the examined patients. In the first group, polymorphism 12Pro12Pro was identified in 46%, 12Pro12Ala-54%; in the second group 12Pro12Pro-84%, 12Pro12Ala-16%, in the third group-12Pro12Ala-41%, 12Pro12Pro-59%. The comparison of the groups of patients based on polymorphism of the gene PPARγ2, reveals that carriers polymorphism 12Pro12Pro show a higher level of HOMA-IR (p<0,007) and insulin (p<0,05). Then we assessed the influence of IR on the functional parameters of kidneys. In the first group the microalbuminuria (MAU) was identified in 9 patients (25%), proteiuria in 12 patients (34%),CKD in 6 patients (17%); in the second group the MAU was identified in 7 patients (50%), proteinuria-in 5 (35%), increasing creatinine in 4 (28%),CKD-in 7 (50%). In the third group-the MAU- in4 (26%), proteinuria in 5 (35%), increasing creatinine in 5 (35%),CKD in 6 (40%). The results of the one-way ANOVA-test demonstrated that an increasing BMI is accompanied by a statistically significant insulin increase (I-12,2±5,64;II-14,09±10,47;III-14,7±7,5p<0,005), increasing HOMA-IR (I-3,2±1,64; II-4,9±3,29;III-4,5±2,06 p<0,005), besides a tendency was revealed towards growing MAU (I-19,4±17,26;II-20,6±21,9;III-37,87±19,06 p>0,05), proteinuria (I-0,11±0,33; II-0,11±0,22; III-0,5±1,04p>0,05), creatinine (I-0,99±0,23 II-1,14±0,2, III-1,05±0,4 p>0,005). The following correlations were identified: in group 1: insulin and creatinine r=0,35 p=0,04; HOMA-IR and proteinuria r=0,36 p=0,038; HOMA- IR and MAU r=0,42 p=0,043; On the basis of the multiple linear regression analysis, it was established that insulin (β=0,36, p<0,05) and HOMA-IR (β= -0,37, p<0,05) are independent determinants proteinuria; HOMA-IR (β=0,097,p<0,05) independent determinants of creatinine.

Conclusions: Pro/pro polymorphism of gene PPARγ2 prevailed in patients with morbid obesity and it was associated with high HOMA index. Insulin level and HOMA index are independent determinants of proteinuria. IR presence can be viewed as a factor of progressing obesity-associated nephropathy.

Su380 URINARY EXCRETION OF AQP2 INCREASE IN IGA NEPHROPATHY: INVOLVEMENT OF KALLIKREIN-KININ SYSTEM

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Introduction and Aims: IgA nephropathy is the most common form of primary glomerulonephritis worldwide. Renal biopsy is essential for the diagnosis of IgAN and in some cases repeated biopsy is performed to evaluation of treatments. Clinical manifestation include proteinuria, in some cases edema, and hypertension. Anti-hypertensive drugs have been useful administrated to lower blood pressure and slower renal damage. Not all patients affected by IgAN respond significantly to the treatment with the angiotensin converting enzyme inhibitors (ACEi). Therefore, the search of urinary biomarkers to predict the clinical response to ACEi therapy would be very useful. We have demonstrated that the IgAN patients unresponsive to the inhibition of renin-angiotensin system (RAS), excreted a very low level of kininogen compared with healthy subjects and responder patients. We have also demonstrated that bradykinin, which derived from kininogen, counteracts the water channel AQP2 translocation at the plasma membrane of collecting duct principal cells.P2 is also excreted in the urine and can be considered a useful urinary biomarker of altered renal functionality. Our pourpose is to evaluate the urinary AQP2 in IgA nephropathy and its possible correlation to kallikrein-kinin system players.

Methods: AQP2 and Bradykinin ELISA (enzyme linked immunoassay) tests were used to analyzed both urine from biopsy proven IgA nephropathy patients and healthy individuals.

Results: We found that urinary excretion of AQP2 was higher in IgAN patients compared to healthy subjects, and higher levels were found in IgAN unresponsive to ACEi treatment. Indeed, urinary bradykinin was significantly lower in IgAN not responder compared to responders.

Conclusions: These data suggest that an increase in AQP2 excretion may be a useful biomarker associated to IgA nephropathy. Moreover low urinary bradykinin levels found in non responders with respect to responders, with urinary kininogen and AQP2 levels may represent a potential panel of predictive biomarkers for the clinical response to therapeutic treatment with ACEi.

Su381 CLINICAL COURSE OF MEMBRANOUS NEPHROPATHY WITH C4d GLOMERULAR DEPOSITS

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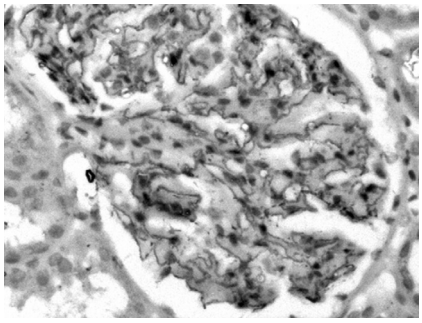
Introduction and Aims: Since 1989 when Kusunoki described C4d renal deposits in membranous nephropathy (MN) their role in disease progression has not been analysed. We have retrospectively evaluated paraffin embedded kidney biopsies performed in our Unit between 1995 and 2008, from 15 consecutive MN patients (7F, 8M).

Methods: Kidney sections were stained using a rabbit polyclonal anti-human C4d antibody. Antigen was retrieved by 13 minutes autoclave incubation in citrate buffer (0.01 M, pH 6.0, 250°C). Paraffin sections were incubated with 1:30 PBS dilution of primary antibody for 1 hour at 37°C. Positive control was a graft with humoral kidney rejection and intense C4d staining.

Results: At the time of biopsy mean age was 57±17 years (mean±SD). Laboratory findings at the time of biopsy: serum creatinine (SCr) 1.26±0.63 mg/dl, proteinuria (UPr) 6.17±5.91 g/day. Five (2F, 3M) out of 15 MN biopsies showed global and diffuse C4d glomerular capillary staining (figure). Interstitial C4d deposits were not observed. No treatment difference between C4d +ve and C4d –ve MN patients was found. No significant SCr and UPr variation was observed between the two groups at time of biopsy and after 6-month follow-up. At last follow-up (mean, 2.54 years; range, 1-8 years) a significantly higher UPr was found in C4d+ve compared to C4d-ve MN patients.

Clinical and laboratory findings of C4d+ve and C4d-ve MN patients

	Basal	Last follow-up	P vs. C4d -ve
C4d +ve; N=5 (M±SD)			
Creatinine, mg/dl	1.14±0.71	1.87±1.61	NS
Proteinuria, g/day	7.38±5.06	5.53±4.92	0.047
Time from biopsy, years	–	2.50±3.00	NS
C4d -ve; N=10 (M±SD)			
Creatinine, mg/dl	1.32±0.62	1.49±0.70	–
Proteinuria, g/day	5.57±6.46	1.02±1.73	–
Time from biopsy, years	–	2.64±2.50	–



Conclusions: Banff 2008 report on criteria for renal allograft rejection pointed out: "C4d in the glomerular capillaries was an independent risk factor for late (>6 month) graft failure". Our data suggest that glomerular capillary deposits of C4d in native MN might be a negative prognostic factor for UPr after 1-year follow-up. We are aware that this work has limitations: it is a retrospective study, small biopsy series, relatively short term follow-up. In this respect relationship between worsening of UPr and glomerular C4d deposits might be considered a valuable clinical association rather than a causal finding.

Su382 CYSTATIN C CONCENTRATION IS CORRELATED WITH THE DISEASE ACTIVITY IN RHEUMATOID ARTHRITIS PATIENTS

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Introduction and Aims: Rheumatoid arthritis (RA) is a chronic, systemic, inflammatory disease. Renal involvement worsens the course of RA and increases mortality. Estimation of renal function is essential to optimize the choice of treatment in accordance with disease activity and possible renal damage. Renal function may be assessed with different methods including serum creatinine (SCr), estimated glomerular filtration rate (eGFR) using creatinine-based formulas as well as serum cystatin C (Cys-C). Cys-C is used as an endogenous marker for GFR, more precise than creatinine.
The aim of the study was to investigate the impact of RA activity on the renal function status assessed with different methods.

Methods: The study population consisted of 140 patients with RA (111 women and 29 men); the mean (SD) age was 50,3 (10,9) years; the mean (SD) disease duration 120,6 (89,8) months. Disease Activity Score in 28 joints (DAS28) was calculated as a primary outcome measure. Health Assessment Questionnaire (HAQ) was a functional status measure. High disease activity (DAS28≥ 5,1) was observed in 42 patients (30%); long-term RA (duration at least 10 years) in 64 patients (45,7%). Several routine and metabolic laboratory markers were determined in all patients. Measures of renal function were Cys-C and SCr concentrations, creatinine-based eGFR calculated by Cockroft and Gault (CG) and Modification of Diet in Renal Disease (MDRD) formulas.

Results: The mean (SD) Cys-C concentration was 0,75 (0,19)mg/l, the SCr concentration was 0,71 (0,23) mg/dl, eGFR by CG 110,5 (37,8) ml/min/1,73 m^2, and MDRD 109,5 (34,5) ml/min/1,73 m^2). Increased Cys-C level (≥ 1 mg/l) was observed in 17 patients (12,1%) and SCr (≥ 1 mg/dl) in 11 patients (7,9%). Stage 3 of chronic kidney disease (eGFR < 60) by CG was observed in 5 patients (3,6%) and by MDRD in 4 patients (2,9%). Cys-C levels correlated positively with SCr (R=0,3; p=0,0006), negatively with eGFR by CG (R= -0,3; p=0,001) and MDRD (R= -0,3; p=0,0002).

Results: The mean (SD) Cys-C concentration was significantly higher in patients with high disease activity in comparison with low/moderate RA activity [0,9 (0,46) vs 0,73 (0,19) mg/l, p=0,002]. Cys-C level was higher in patients with long-term RA than in patients with RA < 10 years [0,82 (0,38) vs 0,74 (0,22), p=0,05]. Patients treated currently with biologic agents had lower Cys-C levels than those treated with conventional modifying drugs [0,71 (0,17) vs 0,8 (0,34), p=0,06]. Serum Cys-C showed significant correlations with markers of disease activity [DAS28 (R=0,28; p=0,0008), ESR (R=0,49; p=0,00001), CRP (R=0,28, p=0,0009)] and functional status [HAQ (R=0,3; p=0,0006), VAS patient's global assessment of disease activity (R=0,26; p=0,02; morning stiffness (R=0,17; p=0,04)].

Conclusions: Cystatin-C seems to be a sensitive indicator of mild impairment of renal function. In patients with RA serum Cys-C concentrations were correlated not only with other parameters of renal function, but were associated with parameters of the disease activity. In patients with RA impairment of renal function could be expected in association with high disease activity.

Su383 TACROLIMUS VERSUS INTRAVENOUS CYCLOPHOSPHAMIDE AS THE INDUCTION THERAPY FOR SEVERE LUPUS NEPHRITIS (LN): A META-ANALYSIS OF RANDOMIZED CONTROLLED TRIALS

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Introduction and Aims: Treatment of lupus nephritis (LN) remains unsatisfactory. Meta analysis of randomized controlled trials (RCTs) was performed to compare the efficacy and safety of tacrolimus (FK506) with cyclophosphamide (CYC) as the induction therapy for severe LN.

Methods: We searched PubMed Database, the Cochrane Central Register Of Controlled Trials (CENTRAL) database, EMBASE and Wanfang Database for RCTs that compared FK506 with CYC for LN treatment. The published languages and years were not limited. The trials searched were evaluated for eligibility and quality, and then the data were extracted and analyzed for remissions, side effects in induction therapy.

Results: Three RCTs with 110 biopsy-proven LN class III-V or IV+V patients, providing the data for FK506 versus CYC as the induction therapy for severe LN were analyzed. Compared with CYC after 6-month induction therapy, FK506 significantly increased the complete and overall remission rate (relative risk (RR) = 1.85, 95% confidence interval (CI) 1.12-3.05, P=0.02 and RR = 1.36, 95% CI 1.01-1.85, P=0.04). The incidence of hyperglycemia was higher than intravenous CYC (odds ratio (OR) = 7.83,

95% CI 1.54-39.70, P=0.01). There was no significant difference between FK506 and CYC induction therapies in the adverse effects of gastrointestinal symptoms, elevating aminotransferase, elevating Scr, hypertension, infection and leucopenia.
Conclusions: A 6-month course of FK506 has higher efficacy than pulsed intravenous CYC as the induction therapy in severe LN without significant difference in the side effect except hyperglycemia. Further large scale, high-quality RCTs for comparing the FK506 regimen with other immunorepressive regimens are needed.

Su384 CLINICAL INVESTIGATION OF FAMILIAL FOCAL SEGMENTAL GLOMERULAR SCLEROSIS IN CHINA

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Introduction and Aims: To discuss the characteristics of familial FSGS patients through pedigree investigation in China.
Methods: From January, 2006 to December, 2007, pedigree investigation was taken for all the non-secondary FSGS patients admitted in our department. 19 patients were found with family history. Clinical screening were taken for their family members, type IV collagen and a- galactosidae activity were also tested to exclude Alport syndrome and Fabry disease. The clinical characteristics of familial and sporadic FSGS patients were compared.
Results: During the above period, totally 213 patients were diagnosed non-secondary FSGS confirmed by renal biopsy. Among them 19 patients had family history, accounting for 8.9 percent. Of the 19 cases, there were ten men and nine women and the gender construction has no significant difference compared with sporadic cases (M: F = 100:94, P> 0.05); The average 24-hours urine protein level of familial FSGS patients was 1.6±1.6g, compared with 1.8±1.7g that in sporadic FSGS patients, with no significant difference (P> 0.05); The average serum creatinine level of familial FSGS proband was 167.5±166.6umol/L, while 198.4±190.6umol/L in sporadic FSGS patients, with no significant difference (P> 0.05). Among the 19 FSGS families, five had two members confirmed as FSGS by renal biopsy, seven families had one cases of renal biopsy confirmed FSGS and more than one cases with ESRD, others had one cases of renal biopsy confirmed FSGS and one or more relatives with proteinuria or unexplained renal insufficiency.
Conclusions: Familial FSGS has a high incidence of 8.9 percent in China. With respect to clinical manifestations there was no significant difference between sporadic patients and familial ones, so it was easy for missed diagnosis. Careful pedigree investigation can significantly improve the diagnostic rate. Paying attention to familial FSGS screening will also contribute to the further study of genetic renal diseases.

Su385 EFFICACY OF CO-ADMINISTRATION OF MIZORIBINE AND PREDONISOLONE FOR IDIOPATHIC MEMBRANOUS NEPHROPATHY

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Introduction and Aims: Membranous nephropathy is one of common causes of primary nephrotic syndrome in adults, however, the efficacy of treatment with corticosteroids and/or immunosuppressive agents, such as cyclophosphamide or cyclosporine, had not been established. Mizoribine (p-INN: 4-carbamoyl-1-beta-D-ribofuranosyl imidazolium-5-olate; MZB), which is an antibiotic agent produced by the soil fungus Eupenicillium brefeldianum in Japan, inhibits purine nucleoside synthesis. MZB has been used for immunosuppressive therapy for organ transplantation and IgA nephropathy in childhood with less myelo-suppression. We investigated the effect of MZB on proteinuria with adult idiopathic membranous nephropathy.
Methods: Fifty patients of biopsy-proven idiopathic MN (63±15 year old, M/F = 31/19, UP = 5.1±2.3 g/day, serum albumin = 2.9±0.8g/dl, serum creatinine = 0.97±0.40 mg/dl, follow-up period: 22±14 months) from April 2006 to September 2009 were subjected. The patients were divided into 3 groups by treatment (MP; oral MZB and oral predonisolone, n = 19, PSL; oral predonisolone only, n = 16, C; control group without MZB nor predonisolone, n = 15). MZB or predonisolone was started from 100 mg/day (2 mg/kgBW/day), or 30 mg/day (0.5 mg/kgBW/day), respectively. Both of MZB and predonisolone was decreased in dose monthly and finished to be administered after 12 months. The pre-treatment physical status did not differ between the groups. Angiotensin II blocker was co-administrated in 35 patients (68%).
Results: There was no difference in proteinuria between the groups at kidney biopsy. Urinary protein decreased in all 3 groups, however, only MP group showed significant lower urinary protein level than C group after treatment ($p<0.05$). Magnitude of decreasing UP were -91%, -70% and -49%, in MP, PSL and C group, respectively. Serum albumin increased both in MP and PSL groups. During the follow-up periods, remission (urinary protein < 1.0 g/day) rates in group MP, PSL or C were 84%, 56% and 33%, respectively. MP group showed significantly higher incidence in remission than C group ($p<0.01$). There was no significant difference in change of proteinuria between PSL group and C group. Serum creatinine levels and blood pressure did not change without any difference between the groups.
Conclusions: Treatment with only oral administration of corticosteroids has not been considered efficacious to induce remission for membranous nephropathy. Co-administration of MZB with corticosteroid may be effective for improving proteinuria in primary membranous nephropathy.

Su386 URINARY ANGIOTENSINOGEN (UAGT) IN IGA NEPHROPATHY PATIENTS

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Introduction and Aims: Intrarenal renin-angiotensinogen system (RAS) plays a crucial role in the pathogenesis of hypertension, diabetes and chronic kidney disease. It was reported that urinary angiotensinogen (uAGT) is parallel to the value of intrarenal angiotensin II and provides a specific index of intrarenal RAS status.
The present study was performed to find the role of uAGT in accordance with possible application as a prognostic marker in patients with IgA nephropathy (IgAN).
Methods: A total of 40 patients (mean age 36.7 years old, male:female=14:26) were included and a random morning urine sample was obtained. The uAGT concentrations were measured with human ELISA kits.
Results: Mean blood pressure in patients was 119/71 mmHg, amount of proteinuria was 1.11 g/gCr and uAGT was 50 ng/mgCr. The uAGT levels were significantly positively correlated with the amount of proteinuria (r^2=0.203), diastolic blood pressure (r^2=0.101) and the degree of tubulointerstitial inflammation (r^2=0.76).
When patients were grouped by their uAGT levels of above (Group A) and below 100 ng/mgCr (Group B), group A showed significantly lower eGFR (65 vs 87 mL/min/1.73 m^2) and higher amount of proteinuria (2.91 vs 0.78 g/gCr) and degree of hematuria (3.5 vs 2.8 grade) compared to group B. All patients received angiotensin blockers among them 55% of patients received steroid therapy or other immunosuppressants. After 3 and 6 months, group A showed also significantly decreased eGFR but there were no significant differences in the amount of proteinuria and the degree of hematuria between two groups.
Conclusions: In IgAN, uAGT is thought to be correlated with proteinuria, diastolic blood pressure and degree of tubulointerstitial inflammation. Also patients with higher levels of uAGT above 100ng/mgCr showed significantly decreased renal function persistently before and after therapy. In conclusion, higher levels of uAGT may indicate poor renal function but additional research with a prolonged duration and a larger patient group is needed for further confirmation.

Su387 ELEVATED SERUM LEVELS OF APRIL AND BAFF IN MPO-ANCA-ASSOCIATED RENAL VASCULITIS

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Introduction and Aims: Two closely related cytokines of the tumor necrosis factor (TNF) superfamily, a proliferation-inducing ligand (APRIL) and The B-cell activation factor belonging to the TNF-family (BAFF) were identified as a novel TNF family ligand and have proven to be a key factor in the selection and survival of B cells. Both BAFF and APRIL are expressed as a cell surface protein and released into the circulation after cleavage by furin protease. Abnormal levels of both BAFF and APRIL have been observed in patients with autoimmune diseases, such as systemic lupus erythematosus (SLE), rheumatoid arthritis, and Sjögren's syndrome. Overexpression of BAFF in mice leads to autoimmunity with (SLE)-like symptoms, while BAFF-deficient mice lack mature B cells. Although, under normal levels of BAFF concentration, non-self-reactive B cells survived and autoreactive B cells were deleted, a higher concentration of BAFF contributed to the survival of autoreactive B cells and elevation of autoantibody production. To analyze the association between the production of autoantibodies and BAFF in ANCA-associated vasculitis, the serum levels of BAFF and APRIL were investigated in MPO-ANCA-associated renal vasculitis.
Methods: APRIL and BAFF in serum obtained from 37 patients with MPO-ANCA-associated vasculitis were measured by an enzyme-linked immunosorbent assay. Twenty-three samples were taken from active vasculitis patients, 24 from inactive vasculitis patients and 13 from inactive vasculitis with infectious complications. Serum APRIL and BAFF were also measured in 20 patients with chronic kidney disease (CKD).
Results: The serum APRIL in vasculitis patients with active vasculitis (54.47±51.50 ng/ml) was higher than that in CKD patients (13.92±15.60 ng/ml, P = 0.0002), but there was no difference in serum APRIL levels among active-vasculitis, inactive-vasculitis (35.06±25.32 ng/ml) and infectious patients (31.76±25.47 ng/ml). Among patients with MPO-ANCA-associated vasculitis, the serum BAFF was higher in active-vasculitis patients (4065.7±2189.7 pg/ml) than in inactive-vasculitis (1514.7±746.6 pg/ml) and infectious patients (1693.9±793.6 pg/ml) ($P < 0.0001$, $P < 0.0001$, respectively). Although the serum BAFF in active-vasculitis patients was also higher than that in CKD patients (1342.5±733.0 pg/ml, $P < 0.0001$), there were no significant differences in serum BAFF among inactive-vasculitis, infectious and CKD patients. There was no significant correlation between serum levels of APRIL and serum ANCA titers, but there was a significant correlation between serum levels of BAFF and serum ANCA titers ($r = 0.465$, $P = 0.0004$).
Conclusions: Excessive BAFF production in patients with MPO-ANCA-associated vasculitis may be one of the principal factors for autoimmune B cell tolerance, result in MPO-ANCA production.

Su388 A CLINICOPATHOLOGY CORRELATION OF LUPUS NEPHRITIS: A RETROSPECTIVE ANALYSIS OF RENAL BIOPSIES

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Introduction and Aims: The optimal treatment of lupus nephritis (LN) is uncertain, but it varies with the type and severity of the disease that is present as well as the ethnicity of the patient. Despite being a common renal manifestation, there are limited data about the outcome of lupus nephritis in our Saudi population. Here we describe our center experience with lupus nephritis in terms of histological classification, clinical characteristics and renal outcome.
Aims: To classify all renal biopsies of lupus nephritis in Saudi patients presenting from 2000 until 2006 at our hospital, according to the modified World Health Organization (WHO) classification, and to correlate the histological findings with the clinical features at the time of presentation and their renal outcome.
Methods: Between January 2000 and December 2006, a total of 1098 biopsies in our hospital were reviewed by a histopathologist with the assistance of a nephrologist.
Results: a total of 1098 renal biopsies were reviewed, 80% were native biopsies while 20% were biopsies of transplanted kidneys. Of the native kidneys, the most common histopathology diagnosis was lupus nephritis with 36% of all native biopsies followed by FSGS (16%). In the lupus nephritis patients, we found that male to female ratio was 1:8, mean age was 25.44±10.69 years, mean creatinine at the time of biopsy was 124.78±103.38 umol/L, mean urine protein 3.68±3.88 g. Class IV is the most common presenting class of lupus nephritis while the heaviest proteinuria was present in class IV+V. More than 40% of class IV and class V present with nephrotic syndrome while more than 30% of class 6 have active urinary sediments. Most of the patients received treatment with ACEI or ARB. Cyclophosphomide was the main induction therapy and Cellcept was the main maintenance therapy for class IV. There was a good post treatment response in all classes in term of urinary protein and dsDNA antibody reduction.
Conclusions: In Saudi population at KFSH, the most common presenting renal pathology is lupus nephritis. Their presentation with hematuria and renal insuffeciency was similar to the presentation of other populations. The predominant histological type was WHO class IV. Patients of this class were more commonly associated with microhematuria, elevated proteinuria, and renal insufficiency. Most patients received IV cyclophosphomide with good renal outcome.

Su389 MANNOSE BINDING LECTIN: SERUM LEVEL, GENE POLYMORPHISM IN SLE PATIENTS AND ITS RELATION TO THE DEVELOPMENT OF LUPUS NEPHRITIS

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Introduction and Aims: Systemic lupus erythematosus (SLE) is an autoimmune disease in which the complement system plays a crucial role in its pathogenesis. Mannan -binding lectin (MBL) is a recognition molecule of the lectin pathway of complement activation. The presence of several polymorphisms at the promoter and coding regions of the MBL-2 gene determines alterations at MBL serum concentration. MBL variant alleles that lead to low serum levels and/or functional deficits of MBL are postulated to contribute to the susceptibility of SLE. Moreover, the influence of MBL variation on antibody production and renal involvement in SLE patients remains controversial.
Objectives: MBL serum level and genotypes were studied in our SLE Egyptian patientswith evaluation of its role in auto antibodies production and lupus nephritis development.
Methods: MBL genotypes and serum level were screened in a case control study included 30 SLE patients as well as 30 healthy controls. MBL polymorphism at exon 1 codons 54 and 57 was detected by PCR using sequence-specific priming (SSP) and serum MBL level was determined by ELISA technique.
Results: There was predominance of AA genotype (80%) in control group. Genotype frequencies of MBL variants in patients with SLE showed significant differences when compared with controls (AA 53.3% vs 80%, P=0.03, OR = 0.29 and AØ+OØ46.6% vs 20%, P = 0.03, OR = 3.5, respectively). Serum MBL in our SLE patients (900 ng/ml was significantly lower than that of the control group (2750 ng/ml, P = 0.00) with positive correlation with low MBL genotypes. SLE patients with mutant alleles were more likely to produce anti dsDNA (92.8% vs 75%, OR = 4.3) and anti-Smith (35.7% vs 18.7%, OR = 2.3). Patients carrying MBL-low genotypes have an increased risk of development of lupus nephritis than those carrying MBL-high genotype (64.7% vs 35.2%, P = 0.02, OR= 2.4).

Conclusions: MBL gene polymorphism associated with low MBL serum levels that were found with significantly increased frequency in our SLE patients may be one of the genetic factors that determine the susceptibility to develop lupus nephritis.

Su390 ADAMTS13 ACTIVITY IN BIOPSY PROVEN THROMBOTIC MICROANGIOPATHY (TMA) PATIENTS WITH THROMBOPHILIA

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Introduction and Aims: Deficit of ADAMTS13 (Von Willebrand factor cleaving metalloprotease) is known for its causative relation to thrombotic thrombocytopenic purpura (TTP).Varying degrees of its deficit are also found in other microangiopathic syndromes (HUS, APS, DIC). The reduction of ADAMTS 13 activity in different glomerular nephropathies (LN, DN, CGN) has been demonstrated recently. The aim:to assess ADAMTS 13 activity in morphologically proved TMA patients with nephropathy and thrombophilia.
Methods: We evaluated ADAMTS13 activity in plasma from 20 patients (6 F, 14 M, mean age 30,0±11 years) with nephropathy and thrombophilia (primary APS -1 pts, secondary APS in SLE – 4 pts and hereditary thrombophilia – 15 pts.

Hemostasis genes polymorphisms

Polymorphism	Genotype	n (%)
MTHFR C677T	CT	7 (37%)
	TT	3 (16%)
FV Leiden	GA	1 (5,3%)
PTG G20210A	GA	1 (5,3%)
FGB -455G/A	GA	10 (56%)
	AA	3 (17%)
ITGB3 L33P	LP	5 (33%)

All of them had biopsy proved TMA: in combination with morphological features of CGN in 9 pts and lupus nephritis -4pts or as the sole manifestation in 1 PAPS pts and 6 pts with multigenic thrombophilia. No one had symptoms of TTP. 18 (90%) pts had impaired renal function and arterial hypertension (sCr 3,2±2,4 mg/dl; GFR 61±38ml/min; mean BP 181/106 mmHg). Acute TMA was found in 2 (20%) pts: in combination with LN-IIIcl. in 1 pts. and membranous GN -in 1pts. Other 18 (80%) patients had chronic TMA: in combination with LN-IIIcl- 1pts, LN- IVcl – 2pts., with IgA- nephropathy -8pts and FSGS-1pts. The ADAMTS13 activity was measured by fluorescence resonance energy transfer assay (Pepta-Nova GmbH, Germany) in the plasma of pts and 14 healthy volunteers. Reference interval from 14 healthy volunteers was 94%-113%.
Results: No case of severe ADAMTS13 deficiency was found among the patients (average level was 97% [57%-123%] and it did not differ from healthy control subjects (p>0,1). The ADAMTS13 activity<93% was found in 8 (40%)pts. The lowest ADAMTS13 activity was found in two acute TMA patients (56% and 76%). Other 6pts had ADAMTS13 activity level in limits 81%-92%.
The ADAMTS13 activity in reference intervals (111%-97%) was in 9 (45%) pts and 3pts (15%) had ADAMTS13 activity level above upper reference values (115%-123%). ADAMTS13 activity directly correlated with platelet counts (r=0,5, p=0,012). In patients with ADAMTS13 activity <90% Scr level was higher (4,0±2.4mg/dl vs 2,7±2.4mg/dl, p>0,05). Partial or complete recovery of renal function observed among 10 (83%) pts with ADAMTS13 activity >93% in compared with 3 pts (42%) with ADAMTS13 activity < 93% (p=0,06)
Conclusions: We suppose that decrease of ADAMTS13 activity in absence of HUS/TTP can present in TMA patients with thrombophilia. The combination of multigenic HT and decrease of ADAMTS13 activity can cause superimposed TMA in patients with various nephropathies. TMA may be the only manifestation of renal involvement in hereditary thrombophilia pts.
Disclosure: This research is supported by Russian Foundation for Basic Reseach (RFBR) grant.

Su391 EFFECT OF ALISKIREN IN PROTEINURIA OF PATIENTS WITH PRIMARY GLOMERULONEPHRITIS

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Introduction and Aims: Aliskiren has been studied mainly for its role in the proteinuria of diabetic nephropathy. We investigated the antiproteinuric effect of aliskiren in patients with primary glomerulonephritis (GN).
Methods: We studied prospectively 12 patients (7 men/5 women) mean age 60 (34-72) years, with GN. All patients for a trimester before the initiation of the study had constant 24h urine protein >1 gr, stable eGFR >30 ml/min/1,73m,^{2} well controlled arterial blood pressure with antihypertensive treatment that included an angiotensin-converting enzyme inhibitor (ACEI) or an angiotensin II receptor blocker (ARB). Without any change in their medication, patients were put for 3 months on 150mg of aliskiren daily. Every 2 weeks 24h urine protein secretion, eGFR, serum potassium and mean arterial pressure (MAP) were evaluated. Plasma renin activity (PRA) was measured at the beginning and at the end of the study period.
Results: The addition of Aliskiren induced a mean reduction of proteinuria by 16,3% (from 2146,8±1203,3mg/24h to 1445,3±1095mg/24h) (p=0,011). This significant reduction of proteinuria wasn't accompanied by significant changes in mean eGFR (beginning: 68,53±25,75 ml/min/1,75 m^2 / end: 65,07±23,52 ml/min/1,75m^2), and MAP (beginning: 90,7±9,3 mmHg / end: 90,56±9 mmHg), while PRA reduced significantly (beginning: 2,65±2,02 ng/ml / end: 1,51±1,45 ng/ml p=0,0O2). Hyperkalemia >5,5 mEq/lt or hypotensive episodes were not observed during the study.
Conclusions: In patients with primary GN who receive ACE or ARB, the addition of Aliskiren reduces proteinuria significantly and is well tolerated. This antiproteinuric effect of aliskiren seems to be independent of MAP or eGFR alterations.

Su392 ELEVATED PLASMA LEVELS OF PRORENIN IN PATIENTS WITH UNTREATED PRIMARY CHRONIC GLOMERULONEPHRITIS

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Introduction and Aims: Elevated plasma levels of prorenin were detected for the first time in patients with diabetes mellitus and were considered as an early indicator of development of diabetic kidney disease. The role of prorenin acting trough specific (pro)renin receptors in the kidneys (angiotensin II-independent pathway) in the development of glomerulosclerosis was recently demonstrated. The aim of this study was an evaluation of plasma concentration of prorenin in untreated patients, without immunosuppressive and antihypertensive drugs, with biopsy–proven primary chronic glomerulonephritis (GN).
Methods: Fifty five untreated patients (31 women, 24 men) aged 39.9±14.8 years with primary GN in stages 1–3 of chronic kidney disease and 20 healthy persons (10 women, 10 men) aged 36.8±9.3 years participated in the study. Prorenin and renin levels in plasma were measured by ELISA and radioimmunological methods, respectively, and 24-hour urinary protein excretion was evaluated.
Results: Significantly higher prorenin levels (p<0.001) in patients with GN (median 0.768 ng/ml) when compared to healthy controls (median 0.379 ng/ml) were found. There was no difference in renin levels between both analyzed groups. The ratio of renin to prorenin in patients' plasma with GN was significantly lower (p<0.001; median 0.014) than in healthy individuals (median 0.029). In patients with GN plasma prorenin concentrations were significantly correlated with 24-hour urinary protein excretion (r=0.406, p=0.002).
Conclusions: In untreated patients with primary GN elevated plasma levels of prorenin are associated with increased protein excretion, suggesting the role of prorenin in glomerular damage.

Experimental pathology and renal histopathology

Su393 ★ PARIETAL CELLS PROLIFERATE IN RESPONSE TO VEGF OVER-EXPRESSION

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Introduction and Aims: Parietal cells play a so far unrecognized role in various glomerular diseases. Recently, we have shown that cellular crescents are primarily derived from parietal cells. The molecular mechanisms that result in activation and proliferation of parietal cells are largely unknown.
Methods: Murine vascular endothelial growth factor (VEGF)164 was over-expressed in adult doubly transgenic Pax8-rtTA/(tetO)7VEGF mice for 2 and 4 weeks by administration of doxycycline (DOX) as described previously (Hakroush et al., AJP, 2009). Some of the mice were followed up for an additional 4 weeks after DOX administration was terminated.
Results: VEGF serum levels were significantly increased in experimental mice. Within the first 2 weeks, capillary enlargement and glomerular hypertrophy was observed. After 4 weeks of DOX administration, significantly higher numbers of parietal cells were observed in some glomeruli. Parietal cell proliferation continued strikingly when the mice were followed for an additional 4 weeks after termination of DOX administration. Classical cellular crescents were observed in about 20% of glomeruli. Within the remaining glomeruli, all parietal cells were positive for alpha-SMA indicating cellular activation.
Conclusions: We provide the first in vivo evidence that parietal cells are activated by systemic overexpression of VEGF, establishing a reproducible non-immunogenic model for crescentic nephritis.

Su394 TUBULAR Gb3-DEFICIENCY BLOCKS VEROTOXIN-MEDIATED RENAL-FAILURE BUT NOT CEREBRAL THROMBOTIC MICROANGIOPATHY

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Introduction and Aims: Verotoxins are one of the etiological agents of the hemolytic-uremic syndrome (HUS) and thrombotic thrombocytopenic purpura (TTP). In vitro experiments have implicated the glycosphingolipid globotrihexosylceramide (Gb3, CD77) as the receptor mediating the verotoxin toxicity. Aims: (1) To explore whether deletion of Gb3 will protect against verotoxin cytotoxicity in vivo. (2) To define the leading pathophysiologic mechanism causing the death of wild-type mice after verotoxin administration. (3) To explore additional mechanisms of verotoxin toxicity in tissue specific glycosphingolipid (GSL) deficiency models.
Methods: Generation of mice with global Gb3-deficiency and tissue-specific Gb3-deficiency in renal tubular cells, platelets and endothelial cells. Administration of verotoxin 2 followed by survival analysis, organ histology and biochemical analysis of blood and urine.
Results: After administration of verotoxin 2 all wild-type (WT) mice died between day 2 and 4 because of renal tubular dysfunction (potassium retention, sodium and water loss) with ensuing uremia, but mice with global GSL-deficiency survived and did not show biochemical or histological alterations. Renal tubular GSL-deficiency fully protected the mice from uremia and water loss. However 4 to 7 days after verotoxin injection these mice demonstrated cerebral thrombotic microangiopathy leading to death in 50%. This could be attenuated by additional GSL-depletion in endothelial cells, but not in platelets.
Conclusions: Gb3 was the exquisite mediator of verotoxin toxicity. In the murine verotoxin-toxicity model the dominant renal pathophysiologic mechanism was toxicity towards the tubular epithelium with resultant sodium and water depletion followed by kidney failure. Gb3 (CD77) mediates cerebral thrombotic microangiopathy.

Su395 ★ NILOTINIB ATTENUATES THE PROGRESSION OF CHRONIC RENAL FAILURE IN 5/6 NEPHRECTOMIZED RATS

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Introduction and Aims: Nilotinib is a second-generation tyrosine kinase inhibitor that demonstrates a 30-fold increase in activity against Bcr-Abl, and a similar level of activity against the PDGF receptor (PDGFR) and c-Kit when compared to imatinib, a compound that has been previously shown to exhibit therapeutic benefits in animal models of renal disease, including cryoglobulinemic membranoproliferative glomerulonephritis (Iyoda M, et al. JASN 2009) and nephrotoxic serum nephritis (Iyoda M, et al. KI 2009). In the current study, we investigated the role of nilotinib in the progression of established renal failure.
Methods: Adult male Sprague Dawley rats were subjected to 5/6 nephrectomy (n=36) or laparotomy (sham-operated, n=9). Rats with 5/6 nephrectomy were then administered either nilotinib (45mg/kg, n=18) or vehicle (n=18) via daily oral gavage from 2 weeks after surgery, and for a period of 8 weeks. Blood pressure (BP), proteinuria (U-P), serum creatinine (Cr) and body weight (BW) were measured periodically. Renal morphological investigations were performed at sacrifice. In vitro, we used renal fibroblasts (NRK49F) and primary mesangial cells. Cells were pretreated with nilotinib or medium alone, and collagen type I synthesis and PDGFRβ phospholyration induced by angiotensin II or PDGF-BB were analyzed by real-time RT-PCR and immunoblotting.
Results: BP and BW were comparable between the two treatment groups throughout the study. Following 6 and 8 weeks of treatment, serum Cr levels in the nilotinib-treated rats were significantly lower than that of the vehicle-treated rats (8 weeks: 1.04±0.15 vs. 0.76±0.03 mg/dL, $p<0.05$; 0.27±0.01 mg/dL in sham-operated rats). When compared to vehicle treatment, nilotinib-treated rats demonstrated reduced U-P at 1 week after treatment. This reduction was maintained over the course of the study (8 weeks: 118.20±21.50 vs. 54.67±9.02 mg/day, $p<0.01$; 41.93±1.64 mg/day in sham-operated rats). Nilotinib treatment also resulted in a decrease in remnant kidney hypertrophy (2.09±0.06 vs. 1.90±0.08 g, $p < 0.05$), in addition to reduced scores of glomerulosclerosis (semiquantitative score (SEMQ-S) (0-4): 1.57±0.30 vs. 0.60±0.20, $p<0.01$) and tubulointerstitial damage (SEMQ-S (0-5): 3.20±0.42 vs. 1.57±0.23, $p<0.01$). Renal cortical mRNA for collagen type I (collagen type I/GAPDH mRNA: 33.40±6.90 vs. 14.60±2.20, $p<0.01$), TGF-β (TGF-β/GAPDH mRNA: 7.40±0.90 vs. 4.67±0.57, $p<0.01$), fibronectin (fibronectin/GAPDH mRNA: 11.87±1.53 vs. 8.48±0.77, $p<0.05$), and PAI-1 (PAI-1/GAPDH mRNA: 8.56±2.04 vs. 4.76±0.71, $p<0.05$) were also significantly decreased in the nilotinib treated group. In vitro, nilotinib blocked collagen type I/GAPDH mRNA production induced by angiotensin-II in renal fibroblasts and mesangial cells. Nilotinib also decreased collagen type I/GAPDH mRNA levels and prevented PDGFRβ phosphorylation induced by PDGF-BB in mesangial cells.
Conclusions: Nilotinib treatment significantly attenuates renal fibrosis in vivo and in vitro. Our results suggest that nilotinib may prove useful in limiting the progression of chronic renal disease to end-stage renal failure.

Su396 ★ FIBRINOGEN PROMOTES KIDNEY FIBROSIS BY ACTIVATING RENAL INTERSTITIAL FIBROBLASTS THROUGH AN ICAM-1 DEPENDENT MECHANISM

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Introduction and Aims: Fibrinogen is the main protein of the blood coagulation system and there is growing evidence that its function extends far beyond blood clotting. Studies in animal models have identified fibrinogen as an important player in inflammation, tissue repair and regeneration. Here, we studied the role of fibrinogen in the development of renal

tubulointerstitial fibrosis using the unilateral ureteral obstruction (UUO) model in mice lacking fibrinogen.
Methods: UUO was performed on fibrinogen$^{-/-}$ and fibrinogen $^{+/-}$ mice which were sacrificed at different time-points. Their kidneys were removed and analyzed using histology, immunohistochemistry and immunoblot. For *in vitro* studies we stimulated rat renal fibroblasts with fibrinogen to test for MAPK activation by immunoblot and for cell proliferation by BrdU uptake. Blocking and stimulation experiments using antibodies and specific peptides were performed *in vitro* to analyse underlying signalling pathways.
Results: In UUO kidneys from fibrinogen $^{+/-}$ mice we found a massive deposition of renal fibrinogen, which was absent in fibrinogen$^{-/-}$ kidneys. Renal fibrosis developed in both groups of mice but fibrinogen deficiency was associated with a significantly attenuated renal tubulo-interstitial damage. We also found reduced collagen deposition and a significant reduction of interstitial alpha smooth muscle actin after 14 days of obstruction. These changes were associated with a significant decrease of S100A4 positive interstitial fibroblasts.
We therefore tested the effect of fibrinogen on rat kidney fibroblasts *in vitro* and found that exposure to fibrinogen resulted in rapid ERK activation and significantly increased cell proliferation. ERK activation was significantly attenuated by antibody-based ICAM-1 blockade; this blockade also blunted the mitogenic effect of fibrinogen. This provided strong evidence that these effects were mediated by binding of fibrinogen to ICAM-1 which was strongly expressed on the surface of renal fibroblasts. Additional experiments showed that the fibrinogen gamma chain sequence (117-133) was responsible for the observed fibrinogen-ICAM-1 interaction.
Conclusions: In summary, we demonstrate that: (1) fibrinogen is an important activator of renal interstitial fibroblasts in the UUO model and *in vitro*, and (2) that fibroblast activation is mediated by fibrinogen binding to ICAM-1. Thereby, our data introduce fibrinogen as a previously unrecognized promoter of renal fibrosis and suggest that inhibition of fibrinogen-ICAM-1 dependent activation of fibroblasts could be a potential therapeutic target.

Su397 PDGF-C MEDIATES GLOMERULAR CAPILLARY REPAIR

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Introduction and Aims: Glomerular endothelial cell injury is a key component of a variety of glomerular diseases. Factors involved in glomerular endothelial cell repair are promising therapeutic agents for the treatment of such diseases. Platelet-derived growth factor (PDGF)-C has proangiogenic properties, however, nothing is known about such functions in the kidney.
Methods: We therefore investigated the consequences of either PDGF-C infusion or inhibition in rats with mesangioproliferative glomerulonephritis, which is accompanied by widespread glomerular endothelial cell damage. We also assessed the role of PDGF-C in a mouse model of thrombotic microangiopathy as well as in cultured glomerular endothelial cells.
Results: PDGF-C infusion in nephritic rats significantly reduced mesangiolysis and microaneurysm formation, whereas glomerular endothelial cell area and proliferation increased. PDGF-C infusion specifically upregulated glomerular fibroblast growth factor-2 (FGF-2) expression. Vice versa, antagonism of PDGF-C in glomerulonephritis specifically reduced glomerular endothelial cell area and proliferation and increased mesangiolysis. Similarly, PDGF-C antagonism in murine thrombotic microangiopathy aggravated the disease and reduced glomerular endothelial area. In conditionally immortalized glomerular endothelial cells, PDGF-C was mitogenic and induced a 27-fold upregulation of FGF-2 mRNA. PDGF-C also exerted indirect pro-angiogenic effects, since it induced endothelial cell mitogens and pro-angiogenic factors in mesangial cells and macrophages.
Conclusions: These results identify PDGF-C as a novel, potent proangiogenic factor in the kidney that can accelerate capillary healing in experimental glomerulonephritis and thrombotic microangiopathy.

Su398 THE KIDNEY PHENOTYPE OF α8 INTEGRIN CHAIN DEFICIENT MICE IS HIGHLY DEPENDENT ON THE GENETIC BACKGROUND

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Introduction and Aims: The α8 integrin chain is a matrix receptor for fibronectin, vitronectin, tenascin C, osteopontin and nephronectin. It is specifically expressed in glomerular mesangial cells and contributes to maintain the integrity of the glomerular capillary tuft. In α8 integrin-deficient mice bred on a mixed genetic background (C57BL/6x129SV), however, only minor glomerular alterations were observed, although the total renal mass in these mice is reduced to about 50%. Thus, we backcrossed these mice into the C57BL/6 and 129SV strains to evaluate renal alterations on a purebred background.
Methods: Mice were backcrossed eight generations with C57BL/6 and 129SV, respectively. Heterozygous breeding pairs were then mated. The offspring was genotyped after weaning. At an age of 10 weeks, the kidneys were evaluated for histological and physiological parameters of renal damage.
Results: In the C57BL/6 colony, mice homozygous for a deletion of the α8 integrin chain (α8-/-) did not survive to weaning (7 litters from 3 breeding pairs). In contrast, α8-/- mice from the 129SV colony survived in a mendelian distribution (17 wildtype, 69 heterozygous, 26 homozygous). Similar to the findings in the mixed genetic background, renal mass was reduced to 50%, but in the 129SV backcross glomerular size was considerably increased compared to wildtype litters and microaneurysm formation as well as glomerulosclerosis was observed in α8-/- mice from the 129SV colony (glomerulosclerosis index: 0.768±0.142 in α8-/- versus 0.209±0.017 in α8+/+, $p<0.05$). Albumin excretion was higher in α8-/- mice than in wildtype litters (6.74±1.29 mg/g creatinine in α8-/- versus 3.01±0.76 in α8+/+, $p<0.05$). Moreover, the expression of podocyte markers, i.e. WT-1, nephrin and podocin was reduced in α8-/- mice.
Conclusions: α8 integrin chain in mice leads to glomerular alterations which are unmasked by backcrossing on a pure 129SV background. Thus, genetic modifiers strongly affect the renal phenotype of α8 integrin chain deficient mice.

Su399 MICROVESICLES DERIVED FROM ENDOTHELIAL PROGENITOR CELLS ACCELERATE GLOMERULAR HEALING IN EXPERIMENTAL ANTI-THY1.1 GLOMERULONEPHRITIS

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Introduction and Aims: The loss of glomerular endothelial cells is associated with glomerulosclerosis and progression of renal failure. Strategies aimed to improve glomerular angiogenesis have been proposed to limit renal injury. Endothelial progenitor cells (EPC) are stem cells with angiogenic activity. EPC release microvesicles (MV), biologically active particles able to trigger angiogenesis via RNA transfer. The aim of this study was to evaluate whether EPC-MV accelerate glomerular healing in anti-Thy1.1 glomerulonephritis.
Methods: EPC were isolated from peripheral blood of healthy volunteers. MV were separated by ultracentrifugation and characterized for protein and RNA content. Wistar rats were treated as follows: 1) saline; 2) 400 μg anti–Thy 1.1; 3) anti–Thy 1.1 + 250 μg/ml EPC-MV; 4) anti–Thy 1.1 + 250

μg/ml EPC-MV pre-treated with 1U/ml RNase. Rats were sacrificed at day 4, 7 and 14. Proteinuria and plasma creatinine were evaluated. In vitro, we studied the effects of EPC-MV on glomerular endothelial cells, podocytes and mesangial cells. In selected experiments, MV were pre-treated with RNase or produced by EPC engineered to knock-down Dicer, the enzyme essential for microRNA intracellular production.
Results: EPC-MV express different adhesion molecules on their surface and are enriched for microRNA, small non coding RNA modulating gene transduction. When injected in anti-Thy1.1-treated rats, EPC-MV induced a significant decrease of proteinuria and ameliorated renal function (day 4, 7). Functional data correlated with the histological findings of decrease of mesangiolysis, microaneurysms and leukocyte infiltration. Electron microscopy and immunohistochemistry confirmed that EPC-MV preserved glomerular integrity (maintenance of RECA, nephrin and synaptopodin expression). When MV were pre-treated with RNase their effect was significantly reduced, suggesting the relevance of RNA transfer from MV to target glomerular cells. In vitro, EPC-MV were internalized in glomerular endothelial cells, podocytes and also in mesangial cells through integrin/selectin-mediated mechamisms. EPC-MV sustained endothelial cell proliferation, migration, angiogenesis and reduced leukocyte adhesion. EPC-MV preserved functional integrity of podocytes as suggested by the maintenance of nephrin expression and cell polarity and by the decrease of albumin leaking in an inflammatory microenvironment. In mesangial cells, EPC-MV reduced complement-mediated injury and leukocyte adhesion. All these biological effects were significantly reduced by MV pre-treatement with RNase or using MV released by EPC previously subjected to Dicer knock-down.
Conclusions: EPC-MV accelerated glomerular healing in anti-Thy1.1 glomerulonephritis. This effect may be ascribed to their internalization and mRNA/microRNA transfer into different resident glomerular cells.

Su400 SYSTEMATIC ANALYSIS OF A HUMAN RENAL GLOMERULUS-SPECIFIC GENE EXPRESSION LIBRARY (REGGEL) IDENTIFIES AXON GUIDANCE AS A RELEVANT PATHWAY IN GLOMERULAR BIOLOGY

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Introduction and Aims: The majority of renal diseases leading to end-stage renal diseases (ESRD) are initiated by glomerular alterations. In the last decade several genes were identified with key relevance for glomerular function. Several of these genes show a preferential or specific mRNA expression in the renal glomerulus. To facilitate the identification of gene products and mechanisms important in regulating renal glomerular structure and function a human renal glomerulus-specific gene expression library (REGGEL) was generated.
Methods: Gene expression profiles from human glomeruli and tubulointerstitium obtained from allografts of living donors (n=6) using Affymetrix HG-U133A arrays were studied and compared. Database for Annotation, Visualization and Integrated Discovery (DAVID) as well as the Kyoto Encyclopedia of Genes and Genomes (KEGG) database were used for gene ontology and pathway analysis. Results were validated using qRT-PCR on an independent cohort as well as *in vitro*. Protein levels were analyzed *in vitro* and *ex vivo* by Western Blot and immunofluorescence.
Results: Comparison of gene expression profiles from human glomeruli with the tubulo-interstitial compartment resulted in 677 genes with prominent overrepresentation in the glomerulus. 15 genes with known glomerular overexpression (e. g. PDPN, PLA2R1, MYH9, NPHS1, -2, WT1) served for validation and were all found in the novel REGGEL. We compared our dataset with published libraries and found the most comprehensive coverage of established glomerluar enriched genes in REGGEL. The mRNA expression for several novel glomerular-enriched genes was verified by qRT-PCR. Pathway- and biological process analysis tools identified processes previously not reported to be of relevance in glomeruli. As an example axon guidance was an overrepresented pathway in the REGGEL genes. We analyzed the expression of the axon guidance molecules neuritin (NRN1) and roundabout receptors ROBO1 and -2 on mRNA- and protein level in renal biopsies and *in vitro*. Microarray-analysis and qRT-PCR for ROBO2 on cohorts of DN, FSGS and pretransplant biopsies revealed a significantly lower expression of ROBO2 mRNA in established DN compared with controls.
Conclusions: REGGEL will help to identify novel genes with a glomerular overrepresentation. It can be used for a systematic analysis of molecular mechanisms and gene networks involved in glomerular biology.

Su401 B1 RECEPTOR BLOCKAGE ROLE ON PHYSIOPATHOLOGY OF FOCAL AND SEGMENTAL GLOMERULOSCLEROSIS

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Introduction and Aims: Focal and segmental sclerosis (FSGS) is one of the most important causes of end stage renal failure. The role of bradykinin (BK) in the physiopathology of inflammatory renal injury has been just recently unveiled.
B1 receptor of BK (B1RBK) has been associated with tissue inflammation and renal fibrosis. Thus, we hypothesize that B1RBK could have an important role in podocyte injury in Adriamycin-induced FSGS. In order to study our hypothesis, we modulated B1RBK with specific modulators in a mouse model adriamycin-induced FSGS.
Methods: FSGS was induced in mice by a single intravenous injection of Adriamycin in a dosis of 10 mg/kg.
Podocyte injury was analyzed by protein (western blot) and gene expression (real time PCR) of podocyte related molecules such as nephrin, podocin and alpha-actinine 4. Urinary albumin and protein:creatinine ratio were measured to estimated the functional podocytopathy. In order to modulate B1BK receptor were used a specific antagonist Des-Arg9-Leu8-BK (DALBK) and agonist Des-Arg9-BK (DABK).
Results: Adriamycin injection induced body weight loss, albuminuria and glomerulosclerosis, a phenomenon associated with up regulation of mRNA for B1 receptor in renal tissue. The treatment with B1 receptor antagonist des-arg$_9$-Leu$_8$-BK (DALBK) reduced albuminuria ($P<0.05$) levels and inhibited glomerulosclerosis ($P<0.05$). Moreover, Q-PCR analysis revealed that Adriamycin treatment induced ($P<0.05$) a down regulation of podocin, nephrin, a-actinin 4.
A delay treatment with DALBK could still induce a podocyte protection measured by mRNA ($P<0.05$) and Western blot protein expression ($P<0.05$). Conversely, the use of a B1RBK agonist (des-arg$_9$-BK) aggravated the renal dysfunction and furthermore suppressed the levels of podocyte-related molecules ($P<0.05$).
Conclusions: Our data reveal an important role for B1RBK on FSGS development, and suggest that modulation of kinin signaling through B1 antagonists can be an alternative approach in FSGS management. Financial support FAPESP e CNPq.

Su402 PROGRESSION OF RENAL AND MYOCARDIAL FIBROSIS IS DETERMINED BY GENETIC BACKGROUND IN ALB/TGF-β1 TRANSGENIC MICE

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Introduction and Aims: We have previously described the molecular mechanism of renal fibrosis in alb/TGF-β1 transgenic mice maintained on inhomogeneous (C57Bl6 x CBA F2) genetic background (Mozes et al, JASN 10:271, 1999). These mice are characterized by elevated plasma levels of TGF-β1 (TGFb) and renal fibrosis. Among the transgenic mice we observed

a severe phenotype with proteinuria and glomerulosclerosis, and a mild phenotype without proteinuria. We hypothesized that the inhomogenous genetic background may influence the progression of renal fibrosis in TGF-β1 transgenic mice.

Methods: C57Bl6-alb/TGF-β1 (B6-TGFb) transgenic mice were generated as previously described (Nephrol Dial Transplant 2007, 22 (S6): 103 (FP255)). To test our hypothesis we generated F1 hybrid mice by breeding the B6-TGFb transgenic males with BalbC, CBA and FVB/N females (BalbCxB6-TGFb F1, CBAxB6-TGFb F1 and FVBxB6-TGFb F1, n=8-14/group). As negative controls, wild type F1 hybrids were also generated using B6 wild type males.

Results: Survival and the progression of renal disease was dramatically influenced by the genetic background (median survival in weeks: BalbCxB6-TGFb 3.57 (n=79), CBAxB6-TGFb 2.57 (n=28), FVBxB6-TGFb 2.3 (n=35) and B6-TGFb 27.7 (n=91), p=0.0001).

Due to the short survival, plasma, urine and organ samples were analyzed at 2 weeks of age (n=8-14/group). The elevated plasma levels of TGFb remained unaltered in all transgenic hybrid strains (plasma TGFb in ng/mL: BalbCxB6-TGFb 57±1.3, CBAxB6-TGFb 44.5±19, FVBxB6-TGFb 38.5±13 vs. B6-TGFb 57.6±17). Wild type controls had 5-10 ng/mL plasma TGFb levels. Urine protein/creatinin ratio (PC) was elevated in all TGFb hybrids but not in B6-TGFb mice (PC: BalbCxB6-TGFb 14±5, CBAxB6-TGFb 12±3, FVBxB6-TGFb 11±6, compared to B6-TGFb 5±1, p=0.025). Hybrid TGFb mice had severe glomerulosclerosis at the age of 2 weeks as compared to B6-TGFb mice (score: BalbCxB6-TGFb 1.5±0.1, CBAxB6-TGFb 2.4±0.1, FVBxB6-TGFb 1.6±0.4, B6-TGFb 0.1±0.1, p=0.041). Fibronectin staining of the kidneys showed similar strain differences.

Apart of renal fibrosis, B6-TGFb mice develop a mild degree of myocardial fibrosis and cardiomegaly at 6 months of age, therefore we investigated the influence of genetic background on myocardial histology. Relative heart weight was similar in all transgenic strains at 2 weeks of age (mg/g: BalbCxB6-TGFb 9.2±1.3, CBAxB6-TGFb 12±0.6, FVBxB6-TGFb 8.6±0.5, B6-TGFb 11.4±0.6). Fibronectin expression was lower in B6-TGFb mice (positive area %: BalbCxB6-TGFb 8±2, CBAxB6-TGFb 10±1, FVBxB6-TGFb 13±4, B6-TGFb 5±1, p=0.036). Arterial wall thickness in myocardium was increased in BalbCxB6-TGFb and FVBxB6-TGFb mice, as compared to B6-TGFb and CBAxB6-TGFb mice (wall/diameter %: BalbCxB6-TGFb 37±0.5, CBAxB6-TGFb 25±0.3, FVBxB6-TGFb 39±0.3, B6-TGFb 24±0.4, p=0.032).

Conclusions: Our data demonstrate that genetic background influences the development and progression of TGF-beta1 induced renal and myocardial fibrosis. We wish to further investigate the molecular mechanism responsible preventing fibrosis in C57Bl6 mice in our model.

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Su403 SYSTEMIC INFUSION OF ATRIAL NATRIURETIC PEPTIDE (ANP) IN RATS INCREASES GLOMERULAR PERMEABILITY IN A BIMODAL FASHION

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Introduction and Aims: Plasma volume overload, in conjunction with e.g. congestive heart failure, is associated with an increased release of ANP, which may be partially responsible for the microalbuminuria seen in heart failure. The present study was performed to investigate the effects of systemic ANP infusion on the glomerular permeability to macromolecules in rats.

Methods: In anaesthetized Wistar rats (250-280g) the left urether was cannulated for urine collection while simultaneously blood access was achieved. Rats were continuously infused with ANP (0.5 or 1 mg/min/kg; n=7+10) and with polydisperse FITC-Ficoll-70/400 (mol.radius 13-90Å) and ^{51}Cr-EDTA for 2 h. Plasma and urine samples were taken during control and at 5, 15, 30, 60 and 120 min of ANP infusion and analyzed by high performance size exclusion chromatography (HPLC) and by gamma radiation detection for determination of glomerular sieving coefficients (θ) for Ficoll, and for assessing GFR from the plasma to urine clearance of ^{51}Cr-EDTA, respectively.

Results: Despite a 25% reduction in blood pressure during the ANP infusion (1 mg/min/kg), GFR remained largely unchanged (0.69±0.06 ml/min/g kidney at 60 min*vs.* 0.72±0.03 ml/min/g kidney during control). ANP caused a rapid (within 5 min), bimodal increase in glomerular permeability. θ to high MW Ficolls (mol. radius 50-80Å) thus increased within 5 min to reach a maximum at 15 min, after which θ returned to near control at 30 min, to again increase moderately at 60 and 120 min. Thus, θ for $Ficoll_{70Å}$ increased from $2.14 \times 10^{-5} \pm 3.05 \times 10^{-6}$ to $3.88 \times 10^{-4} \pm 1.33 \times 10^{-4}$ at 15 min ($p<0.001$; n=10) and then reversed to $4.74 \times 10^{-5} \pm 1.23 \times 10^{-5}$ at 30 min, to again increase to $8.88 \times 10^{-5} \pm 1.76 \times 10^{-5}$ ($p<0.05$ *vs.* contr.; n=6) at 120 min.

Conclusions: Systemic ANP infusion in rats caused a rapid, but bimodal, increase in glomerular permeability, with an early, partly reversible permeability peak, later followed by a more long-lasting and moderate increase in glomerular permeability. The glomerular sieving pattern observed for polydispersed Ficoll was found to mainly reflect an increase in the number of "large pores" in the glomerular filter without any primary changes in "small pore" radius or in the charge-selective properties of the barrier. The increases in glomerular permeability induced by ANP may, at least partly, explain the microalbuminuria observed in patients with heart failure and volume overload.

Su404 Glepp 1 INTERACTS WITH NEPHRIN AND PODOCIN

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Introduction and Aims: Glepp1 (Glomerular epithelial protein phosphatase 1) is a receptor protein-tyrosine phosphatase expressed in the apical membrane of podocyte foot processes. Glepp1 knockout mice show a renal phenotype when challenged. In several proteinuric kidney diseases (i.e. IgA nephropathy, FSGS), Glepp1 expression is reduced and has been implicated as a marker of podocyte injury in these diseases. However, the molecular function of Glepp1 in podocytes has not been determined. The integrity of the slit diaphragm depends on tyrosin phosphorylation of the nephrin c-terminus via src-family kinases. The tyrosine phosphatase Glepp1 could be of relevance in this context. The aim was to characterize the molecular function of Glepp1 in podocytes and its influence on the glomerular slit diaphragm.

Methods: HEK293T cells expressed the cytoplasmic tail of Glepp1 and either nephrin or podocin and other cDNA plasmids as indicated. After cell lysis, co-immunoprecipitation with subsequent westernblot analysis was performed.

Results: The cytoplasmic tail of Glepp1 interacts with nephrin and podocin. The interaction domain of nephrin with Glepp1 maps to the aminoacids 1160-1215 of the nephrin c-terminus. There was no interaction of Glepp1 with beta-arrestin1, beta-arrestin2 and Grb2 and Neph 1. In immunofluorecence Glepp1 induces cytosceletal rearrangement and enrichment of Glepp1 in cell adhesions.

Conclusions: The integrity of the glomerular slit diaphragm is regulated by src-kinase mediated tyrosine phosphorylation of the nephrin C-terminus. However, the role of tyrosine dephosphorylation at the nephrin C-terminus has not yet been determined. We assume that Glepp1 is a relevant tyrosine phosphatase at the glomerular slit diaphragm. Through interaction with nephrin and podocin, Glepp1 may play a role in the development and course of proteinuric kidney disease.

Su405 HETEROZYGOUS DISRUPTION OF K-RAS GENE PROTECTS AGAINST APOPTOSIS AND PROLIFERATION INDUCED BY URETERAL OBSTRUCTION IN MICE

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Introduction and Aims: Ras proteins are membrane-associated molecular switches which regulate cell growth, differentiation, proliferation and apoptosis through interactions with a large number of target proteins, such as phosphatidylinositol-3 kinase (PI3K)/Akt and Raf/Erk signalling pathways. Among the different ras isoforms, K-ras is the only to be essential

for normal mouse development as K-ras deficient embryos die before birth. K-ras has been reported to play a role in stimulated proliferation of renal fibroblasts. The aim of the study is to reveal the possible role of K-ras isoform in renal changes induced by unilateral ureteral obstruction (UUO) in mice.
Methods: For this purpose we have used mice heterozygous for a null mutation of the K-ras gene (*K-ras*$^{+/-}$). Studies were performed in obstructed (O) and non-obstructed (NO) kidneys obtained after 3 and 15 days of UUO to analyze early and long-term changes in ras and downstream signaling pathways activation, interstitial fibrosis, apoptosis and proliferation, using Western blot, immunohistochemistry and ELISA techniques.
Results: Compared with NO kidneys, O kidneys showed an increased H-ras, Erk1/2 and Akt activation, and increased expression of vimentin, α-SMA (both markers of fibrosis), cleaved caspase 3, PARP (both markers of apoptosis), and PCNA (marker of cell proliferation). O kidneys from *K-ras*$^{+/-}$ mice showed a higher H-Ras activation than those form *K-ras*$^{+/+}$ mice although no differences were observed in Erk1/2 and Akt activation. After UUO, vimentin expression was lower in O kidneys from *K-ras*$^{+/-}$ than in O kidneys from *K-ras*$^{+/+}$ mice, whereas no differences were observed in α-SMA expression. O kidneys from *K-ras*$^{+/-}$ mice showed also a lower expression of cleaved caspase 3 and PARP, and a lower expression of PCNA than in O kidneys from *K-ras*$^{+/+}$ mice.
Conclusions: These results suggest that K-ras activation plays a major role in UUO-induced apoptosis and proliferation and also regulates vimentin expression.

Su406 AT1-RECEPTOR BLOCKER INHIBITS THE ANGIOTENSIN II ENHANCEMENT OF THE INTERACTION BETWEEN NEPHRIN AND BETA-ARRESTIN2

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Introduction and Aims: Microalbuminuria is a marker for glomerular impairment and is regarded as the strongest predictor for cardiovascular events. In a mouse model for hypertensive nephrophathy, angiotensin II (Ang II) leads to proteinuria. The underlying, molecular mechanisms have not yet been determined.
We postulate that Ang II stimulation modulates the interaction between nephrin and beta-arrestin2 and thereby nephrin endocytosis.
Methods: HEK293T cells expressing the rAT1-receptor, the cytoplasmic tail of nephrin and beta-arrestin2 were stimulated with Ang II. After cell lysis, co-immunoprecipitation of the cytoplasmic tail of nephrin with subsequent westernblot analysis was performed. The same experiment was conducted with candesartan (100nM), genistein (200mirkoM), BAPTA-AM (20mikroM) and calphostinC (1mikroM).
Results: In HEK293T cells, Ang II enhances the interaction of the cytoplasmic tail of nephrin with beta-arrestin2. This effect is dosage dependent and increases with time. The Ang II modulation seems not to be mediated by tyrosine kinases, calcium-dependent enzymes or PKC. Candesartan, a specific AT-1 receptor blocker, inhibits the Ang II modulation of nephrin and beta-arrestin2.
Conclusions: Ang II modulates the interaction of nephrin and beta-arrestin2. An AT-1 receptor blocker inhibits the Ang II effect. The enhancement of the nephrin-beta-arrestin2 interaction may lead to increase in nephrin endocytosis and proteinuria. The Ang II effect on slit diaphragm proteins might help to understand the underlying mechanisms of the Ang II-mediated microalbuminuria.

Su407 PERIOSTIN: A NOVEL GLOMERULAR MATRICELLULAR PROTEIN INDUCED IN PROGRESSIVE HUMAN GLOMERULONEPHROPATHIES

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Introduction and Aims: Matricellular proteins such as osteopontin or thrombospondin are known to be involved in the pathogenesis of chronic nephropathies. To identify additional matricellular genes contributing to the progression of glomerular diseases we analyzed a unique data set of glomerular gene expression profiles from patients with proteinuric glomerulonephropathies (GN).
Methods: Gene expression was studied by microarrays (Affymetrix HG-U 133A and 133-Plus 2) and real-time RT-PCR. Immunohistochemistry was performed on routine kidney biopsies.
Results: In a recently established human renal glomerular gene expression library (REGGEL) we found periostin (POSTN), a matricellular molecule, to be constitutively expressed in glomeruli. To study its expression in GN gene expression profiles of microdissected glomeruli from diseased individuals (n=45) and living allograft donors (LD, n=32) were analyzed. The mRNA for POSTN was found to be induced in progressive diseases such as focal-segmental glomerulosclerosis (FSGS) and membranous GN (MGN) but not in minimal change disease (MCD). Real-time RT-PCR on additional glomerular samples confirmed the POSTN mRNA induction in progressive GN (lupus nephritis (LN) 12.9±33.8, $p<0.001$, FSGS 2.3±1.9, $p<0.05$, MGN 1.9±1.9, $p<0.1$, MCD 2.8±4.9, n.s. compared to LD). The glomerular and tubulointerstitial expression of POSTN showed also a negative correlation with renal function in a larger set of biopsies (r=-0.18, p=8.1E-03, r=-0.47, p=6.9E-14, respectively; n=221). To further study the expression of periostin in the kidney immunohistochemistry was performed: In pretransplant biopsies periostin showed a discrete positivity in the glomerular tuft, surrounding the vascular pole and along the Bowman's capsule; no expression in normal tubulointerstitium was detected. In routine kidney biopsies periostin showed a prominent mesangial positivity in LN and MGN and to a lower extent FSGS. It was also found positive in areas with interstitial fibrosis. Co-immunofluorescene for smooth muscle actin and periostin showed no overlap but a clear proximity suggesting mesangial cells as the source of glomerular periostin. This was further supported by the detection of periostin by western blot on a mesangial cell line.
Conclusions: Periostin, currently studied in detail in bone and heart pathology, is constitutively expressed in healthy glomeruli. It is a novel matricellular molecule linked to progression of glomerular diseases and renal failure. The glomerular function and the underlying pathomechanism in disease will be further studied.

Su408 INTERLEUKIN 17 – A NEW PLAYER IN ACUTE AND CHRONIC KIDNEY INJURY

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Introduction and Aims: Overexpression of pro-fibrotic and pro-inflammatory cytokines, such as TGF-β and IL-6, and extracellular matrix accumulation are hallmarks of acute and chronic glomerular injury. A connection between TGF-β signaling and the inflammatory cytokine IL-17 has recently been proposed. This study analyzed the expression patterns of TGF-β and IL-17 in rat models of acute anti-Thy1 glomerulonephritis (aGN), in streptozotozin-induced diabetic nephropathy (STZ), hypertensive nephropathy (HN) and chronic anti-Thy1 glomerulosclerosis (cGs). The functional crosstalk of IL-17 and TGF-β was studied in renal cell culture.
Methods: aGN was induced in male Wistar rats by i.v. injection of anti-Thy1 OX-7 antibody. Tissues and glomeruli were harvested at the following days: d0.5/d1 (representing injury phase), d5/d10 (matrix expansion phase) and d15/d20 (resolution phase). PBS-injected animals served as controls (con). Cytokine expression was analyzed at molecular levels (mRNA) and in

immunofluorescence. STZ was induced by single i.p. streptozotocin injection in male spontaneously hypertensive stroke prone rats. HN was induced by uninephrectomy of male Wistar rats, followed by a 2/3 nephectomy 2 weeks later. cGs was induced by uninephrectomy of male Wistar rats, followed by an single i.v. injection of anti-Thy1 OX-7 antibody. Tissues were harvested at week 12 after induction. For *in vitro* experiments NRK 52E cells were used. Cytokine expression was analyzed at molecular levels (mRNA) and in immunofluorescence.

Results: Induction of aGN was characterized by marked proteinuria with highest values in the matrix expansion phase (d5 113±12 mg/d; 3.9-fold vs. con; $p<0.001$), afterwards declining towards normal levels. The histological glomerular matrix score peaked at d5 (+3-fold vs. con; $p<0.001$), in parallel with highest TGF-β and IL-17 mRNA expression (+2.25-fold; +6.50-fold vs. con; $p<0.05$). STZ, HN, cGs showed significantly increased proteinuria at week 12 (STZ 121±33 mg/d; 4.5-fold; HN 341±100 mg/24h; 26.0-fold; cGs 435±87 mg/24h; 17.4-fold vs. con; all $p<0.01$). In kidneys, glomerular TGF-β and IL-17 protein expression were dramatically up-regulated in diseased rats compared to controls (STZ: TGF-β 7.3-fold; IL-17 4.7-fold; HN: TGF-β 7.9-fold; IL-17 25.0-fold, cGs: TGF-β 2.5-fold; IL-17 14.2-fold; all $p<0,001$). In vitro, IL-17 was secreted by NRK 52E at basal levels, up-regulated after exposure to 25 mM glucose (+7-fold) and reversed in the presence of a specific TGF-β receptor blocker (SB 431542). Stimulation of cells with TGF-β or IL-6 amplified IL-17 expression by 2.0-fold. Co-administration of TGF-β and IL-6 led to dramatically enhanced IL-17 secretion by more than 4000-fold.

Conclusions: The present study documents a time-dependant expression of the new pro-inflammatory cytokine IL-17 in acute, anti-Thy1 glomerulonephritis. IL-17 is constitutively expressed in glomerular cells and highly induced under inflammatory conditions. In STZ, HN and cGs rats, IL-17 and TGF-β are co expressed by glomerular cells. The down regulation of IL-17 expression by TGF-β receptor antagonism *in vitro* points a new and central regulation of IL-17 by TGF-β signaling pathway.

Su409 PROTECTIVE EFFECTS OF CLOPIDOGREL IN A MOUSE MODEL OF SELECTIVE MICROVASCULAR INJURY

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Introduction and Aims: We have established a selective endothelial injury model in the mouse kidney and have previously shown that platelets are central mediators of endothelial injury. The present study was undertaken to investigate whether the clinically established P2Y12 blocker clopidogrel can sufficiently reduce platelet activation and subsequent reduce kidney injury.

Methods: Selective endothelial injury was induced in left kidneys of 14 C57Bl/6 mice. Starting the day before disease induction, mice were treated either with clopidogrel ((clopi), 20mg/kg bw) or placebo via oral gavage once daily until the day of sacrifice. Mice were sacrificed on day 3. After assessment of tail bleeding times, urine, blood and renal tissues were harvested for further analysis. Tissue sections were evaluated by PAS and AFOG staining and immunohistochemistry.

Results: Renal injury was present in all left sided kidneys, whereas contralateral kidneys demonstrated no injury. Bleeding times of clopi treated mice were significantly enhanced compared to placebo controls ($p<0.05$) demonstrating treatment efficacy. PAS staining showed significantly reduced glomerular injury in the clopi group ($p<0.05$) and by AFOG staining we detected a reduced glomerular fibrin thrombus formation ($P<0.05$). As evaluated after staining for CD31 and MECA32, clopi treated mice demonstrated more intact peritubular endothelium ($p<0.05$). Glomerular capillaries were preserved as shown by a higher capillary length per glomerular cross-section in clopi treated mice ($p<0.05$). In parallel, glomerular cell proliferation was higher in the clopi treated mice group ($p<0.05$).

Conclusions: Clopidogrel treatment is sufficient to reduce platelet activation and endothelial injury in a mouse model of renal microvascular injury. Thereby, subsequent thrombotic microangiopathy is reduced and the glomerular repair response (as indicated by cell proliferation) is enhanced. Clopidogrel is a protective treatment option in this experimental disease model.

Su410 THE EFFECTS OF DUAL BLOCKADE WITH ANGIOTENSIN RECEPTOR AND ENDOTHELIN RECEPTOR BLOCKERS ON THE PROGRESSION OF CHRONIC EXPERIMENTAL KIDNEY DISEASE

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Introduction and Aims: The renin-angiotensin and endothelin systems may both play a role in the pathogenesis of chronic kidney disease (CKD). Recent studies on the effectiveness of endothelin receptor blockers (ERB) in angiotensin II-induced end-organ damage are conflicting. We investigated the effects of dual blockade with angiotensin receptor blocker (ARB) losartan and ERB sitaxentan in comparison with ARB and ERB treatment alone to the rate of progression of experimental CKD.

Methods: Wistar rats matched for age and body weight underwent 5/6 nephrectomy (5/6NPX) and were subsequently treated until 12 weeks with the losartan (180 mg/L in the drinking water) or sitaxentan (30 mg/kg/day) or the combination of the two drugs. Untreated animals served as controls. Physiological parameters (body weight, blood pressure, 24h albuminuria), morphology were investigated and QPCR was performed for the transcription of intrarenal molecular biomarkers of chronic inflammation. For quantification of rat/CCL2/MCP-1, beta-actin (NF-kB, TGF-beta) mRNA we used a SYBR Green real-time quantitative RT-PCR method based on the TaqMan fluorescence method with the ABI Prism 7000 Sequence Detection System.

Results: Results showed that the degree of hypertension was significantly higher in untreated animals (177.8±4.9) compared with treatment groups ARB (102.3±4.9), ERB (132.2±5.2). The dual treatment was associated with further reductions in blood pressure (101.5±3.4) and urinary protein excretion where the lowest proteinuria degree was found (1.8±0.81 mg/24h). Also, the combination of treatments provided superior effects on the degree of glomerulosclerosis. CCL2/MCP-1 and NF-kB renal abundance were all increased in the remnant kidney of the rats without treatment and were reduced significantly by dual treatment that was comparable with ARB or ERB treatment.

Table shows QPCR transcription differences between study groups using Mann-Whitney test

	CCL2/MCP-1
Healthy vs 5/6NPX	p=0.0039
Healthy vs ERB	p=0.0014
Healthy vs ARB	p=0.1317
Healthy vs ARB+ERB	p=0.5098
5/6NPX vs ARB	p=0.0022
5/6NPX vs ERB	p=0.0166
5/6NPX vs ARB+ERB	p=0.0023
ARB vs ERB	p=0.0851
ERB vs ARB+ERB	p=0.0559

Conclusions: Combined ARB and ERB treatment as well as drugs alone prevented the increase in systemic blood pressure, albuminuria and CCL2/MCP-1 gene expression. These data confirm that ET-1 is a contributory mediator of kidney damage in CKD and suggest that dual treatment had an additional beneficial effect on renal morphology and proinflammatory cytokine expression, which was comparable in magnitude to that of ARB or ERB treatment alone.

Su411 THE POSSIBLE EFFECT OF TOLL-LIKE RECEPTOR 4 ON THE EARLY STAGE OF RENAL FIBROSIS

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Introduction and Aims: Ureteral obstruction (UO) is a common clinical problem, which can activate a maladaptive inflammatory response and leads to the more severe renal injury and fibrosis. Toll-like receptor 4 (TLR4) has been reported to play a role in the pathogenesis of liver fibrosis (Seki E, et al. Nature Med, 2008, 13(11), 1324-32), which led us to hypothesise that

TLR4 might be also involved in the development of renal fibrosis due to UO.
Methods: Male C3H/HeN (wild type) and C3H/HeJ (TLR4 defective) mice aged 8-10 weeks were anesthetized by halothane inhalation and unilateral UO (UUO) performed. Kidney samples were collected at 7 and 14 days after UUO. The tissue was embedded in paraffin and then sectioned and stained with modified Masson Trichrome (Weigert-hematoxylin free). To investigate the degree of fibrosis (i) a method involving histological analysis of the percentage of renal fibrotic area was used (Adobe Photoshop) (ii) soluble type I collagen was extracted from renal tissue and assayed using the Sircol soluble collagen kit (Sircol Co.,UK).
Results: The degree of renal fibrosis is significantly lower ($p<0.05$) in C3H/HeJ ($0.665\pm0.05\%$) than in C3H/HeN mice ($5.29\pm0.684\%$) at 7days after UUO. However, by 14 days post-UUO no significant difference between groups was observed. Type1 collagen was significantly higher in the C3H/HeN than in the C3H/HeJ mice at 7 after UUO.
Conclusions: These results suggest that the TLR4 pathway may be involved in the pathogenesis of the early stages of renal fibrosis during uretral obstruction.

Su412 INDOXYLSULFATE-INDUCED EPITHELIAL-TO-MESENCHYMAL TRANSITION AND APOPTOSIS OF RENAL TUBULAR CELLS AS A NOVEL MECHANISM OF PROGRESSION OF RENAL DISEASE

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Introduction and Aims: Indoxyl sulfate, one of the uremic toxins, is regarded as an important substance in the progression of chronic kidney disease (CKD). The level of indoxyl sulfate is elevated in CKD, and indoxyl sulfate lowering therapy is known to improve renal function or delay the initiation of dialysis. Epithelial-to-mesenchymal transition (EMT) and apoptosis of renal tubular cells are known to play a critical role in the development and aggravation of CKD. However, the effects of indoxyl sulfate on EMT and apoptosis of renal tubular cells have not been determined. In this study, we investigated whether indoxyl sulfate per se induced EMT in cultured rat and human proximal tubular cells with an exploration of potential intracellular signaling pathways responsible for indoxyl sulfate-induced EMT. We also studied the effect of indoxyl sulfate on apoptosis of renal tubular cells.
Methods: The effects of indoxyl sulfate on cell proliferation and cytotoxicity were assessed by direct cell counting and LDH assay respectively, in NRK-52E cells. EMT was evaluated by morphologic transformation of NRK-52E and HK-2 cells under inverted microscopy as well as a quantitative analysis of the expression of the epithelial marker, E-cadherin and the mesenchymal marker, α-smooth muscle actin (α-SMA) protein, by western blot analysis. Cell apoptosis was assessed by flow cytometric analysis after annexin-V and PI double staining. ERK1/2 and p38 MAPKinase activation in indoxyl sulfate-stimulated cells were examined by western blotting. The effects of pre-treatment with PD98059 and SB203580 (MAPKinase inhibitors) and probenecid (an organic anion transporter inhibitor) on indoxyl sulfate-induced EMT or apoptosis were also determined.
Results: Indoxyl sulfate significantly inhibited cell proliferation in a dose-dependent manner from a concentration of 6.25 μg/ml. Indoxyl sulfate induced a morphological transformation from cuboidal epithelial cells to spindle shaped scattered fibroblast-like cells in HK-2 cells and NRK-52E cells. Indoxyl sulfate down-regulated E-cadherin and up-regulated α-SMA expression at 48 hours, which were blocked by a pre-treatment of probenecid. Indoxyl sulfate also induced apoptotic cell death in NRK-52E cells from a concentration of 25 μg/ml. Pretreatment of NRK-52E cells with ERK1/2 or p38 MAPKinase inhibitors, PD98059 or SB203580 was not associated with a reversal of indoxyl sulfate-induced EMT, whereas ameliorated indoxyl sulfate-induced apoptosis of NRK-52E cells.
Conclusions: The result of the present study suggested the phenotypic transformation and apoptosis as a novel mechanism of indoxyl sulfate-induced renal damage. Although indoxyl sulfate activated ERK1/2 and p38 MAPKinase in renal tubular cells, it was not responsible for the indoxyl sulfate-induced EMT, but was related to apoptosis of renal tubular cells.

Su413 ONCOSTATIN M INHIBITS BASAL AND TGF-β1-INDUCED MATRICELLULAR PROTEIN EXPRESSION IN HUMAN PROXIMAL TUBULAR CELLS

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Introduction and Aims: Matricellular proteins (MP) are non-structural secreted glycoproteins that help cells to control their surrounding extracellular matrix. MPs modulate cell function by interacting with components of the extracellular matrix, with cell surface receptors, growth factors, cytokines and proteases. As expression of various MPs is increased in renal tubulointerstitial fibrosis, we studied MP expression in human proximal tubular cells (hPTC) stimulated with the pro-fibrotic cytokine transforming growth factor-β1 (TGF-β1) when compared with the multifunctional cytokine oncostatin M (OSM).
Methods: Human proximal tubular (HK-2) cell (PTC) culture, RNA isolation, real-time PCR, Western blot.
Results: Real-time PCR analysis of quiescent hPTC stimulated with 10 ng/ml TGF-β1 revealed a time-dependent induction of four different MPs, namely of connective tissue growth factor (CTGF), thrombospondin-1 (TSP-1), tenascin-C (TNC), and SPARC (secreted protein, acidic and rich in cystein) when compared with unstimulated controls. While TGF-β1-stimulated mRNA expression of TNC (10.1-fold), TSP-1 (7.5-fold), and SPARC (2.1-fold) was highest after 24 h (n=3), strongest induction of CTGF mRNA (4.2-fold) and protein was detected after 3 h and 6 h, respectively (n=3). In contrast, 10 ng/ml OSM did not only inhibit basal mRNA expression of all four MPs after 12 h and 24 h of incubation (n=4) but also almost completely blocked TGF-β1-induced MP mRNA expression (n=4). This inhibitory OSM effect did not depend on the sequence of ligand administration. Simultaneous addition of TGF-β1 and OSM abolished TGF-β1-mediated induction of all four MPs in hPTC after 24 h (n=4). Identical results were obtained when OSM was added either 5 min prior or 5 min past TGF-β1 incubation (n=4).
Conclusions: We conclude that the pro-fibrotic cytokine TGF-β1 represents a strong inducer of CTGF, TSP-1, TNC, and SPARC mRNA expression in hPTC. In contrast, the IL-6 family member OSM is a potent inhibitor of basal and TGF-β1-induced expression of these four MPs. It is tempting to speculate that, by blocking MP expression in hPTC, OSM might have the potential to act as an anti-fibrotic cytokine.

Su414 K-RAS4A MODULATES MYOFIBROBLASTS ACCUMULATION, APOPTOSIS AND PROLIFERATION AFTER UUO IN MICE

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Introduction and Aims: Ras proteins are membrane-associated molecular switches which regulate cell growth, differentiation, proliferation and apoptosis through interactions with a large number of target proteins, such as phosphatidylinositol 3-kinase (PI3K) and Ras/Raf/MEK-extracellular signal-regulated kinase (ERK1 and ERK2). Previous studies from our Laboratory have demonstrated a role for ras activation in interstitial fibrosis and apoptosis induced in mice by unilateral ureteral ligation (UUO). The three functional ras genes present in mammals, H-ras, K-ras and N-ras show a very similar structure and function. K-ras gene encodes two protein isoforms, K-ras4A and K-ras4B, of 189 and 188 residues respectively by alternative splicing and differs significantly at the C termini which is the region involved in membrane association. The purpose of this study has been to assess the possible role of K-ras4A on renal fibrosis after UUO.
Methods: The study was performed in mice deficient for K-Ras4A (*K-Ras4A*$^{-/-}$), partially defficient in Ras4A (*K-Ras4A*$^{+/-}$) and control mice (*K-Ras4A*$^{+/+}$). Obstructed (O) and non obstructed (NO) kidneys were removed 3 and 15 days after UUO. Activation of Ras and its effectos (ERK1/2 and AKT), extracellular matrix markers (fibronectin and y collagen I), myofibroblasts markers (α-SMA and vimentin), apoptosis (cleaved

caspase 3) and proliferation (PCNA), were asessed by western blot and immunohistochemistry.
Results: After UUO, O kidneys from K-Ras4A$^{-/-}$ mice shows a higher pERK/ERK ratio (142±11 vs 97±9 arbitray units; p: 0.004), higher expresion of α-SMA (162±11 vs 117±9 arbitray units; p: 0.003), vimentin (144±9 vs 99±9 arbitray units; p: 0.005), cleaved caspase 3 (165±12 vs 109±9 arbitray units; p: 0.004), and PCNA than O kidneys from K-Ras4A$^{+/+}$ mice. Histological studies were in agreement with data from western blot.
Conclusions: K-Ras4A modulates myofibroblasts accumulation, apoptosis and proliferation after UUO in mice in a way different to other ras isoforms, such H-ras. Specific ras inhibition could be assessed as an useful tool to prevent renal fibrosis.

Su415 UBIQUITIN C-TERMINAL HYDROLASE L1 (UCH-L1) INDUCES POLYUBIQUITIN ACCUMULATION IN PODOCYTES IN RAT MEMBRANOUS GLOMERULOPATHY

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Introduction and Aims: UCH-L1, a key protease in the ubiquitin-proteasome pathway, regulates the intracellular pool of monoubiquitin and is associated with neurodegenerative diseases and cancer. Recently, we described the podocytic upregulation of UCH-L1 in patients with glomerular injuries. UCH-L1 expression correlated with increased podocytic ubiquitin content in membranous nephropathy. We therefore seeked to understand the role of UCH-L1 in the podocytic accumulation of polyubiquitinated proteins in response to antibody mediated podocyte injury.
Methods: We investigated the regulation of UCH-L1 and its role in the accumulation of ubiquitin in a rat model of membranous glomerulonephritis, the passive Heymann Nephritis and in cultured podocytes overexpressing UCH-L1 by Western Blot, Real-Time PCR, proteasomal activity assays, immunohistochemistry and immunofluorescence.
Results: Early in disease (day 4) protein degradation through the ubiquitin-proteasome-system is induced. This induction preceeds protein degradation through the lysosomal pathway. UCH-L1 is upregulated by protein stabilization which is triggered by decreased glomerular monoubiquitin levels. In response to increasing UCH-L1 expression, proteasomal activity and monoubiquitin levels recover. 28 days after injury, podocytes start accumulating polyubiquitinated proteins comparable to podocytes in human membranous nephropathy. UCH-L1 over-expression in cultured podocytes results in polyubiquitin accumulation. Concomitant inhibition of proteasomal degradation in UCH-L1 over-expressing podocytes or in passive Heymann Nephritis rats leads to ubiquitin aggregate formation, a process observed in rats with persisting UCH-L1 expression 1 year after disease induction and in some cases of human membranous glomerulonephritis. The inhibition of UCH-L1 hydrolase activity 14 days after disease induction decreases glomerular polyubiquitin accumulation day 28.
Conclusions: These data indicate, that persistent UCH-L1 upregulation in podocytes during proteinuric disease could be responsible for the accumulation of polyubiquitinated proteins, a process that might explain irreversibility of podocyte injury.

Su416 PEROXISOME PROLIFERATOR-ACTIVATED RECEPTOR-α AGONIST BAY PP1 ATTENUATES RENAL FIBROSIS IN RATS

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Introduction and Aims: There are no data on the role of peroxisome proliferator-activated receptor-α (PPAR-α) in the development and progression of kidney fibrosis.
Methods: Therefore, we examined the effect of two PPAR-α agonists, fenofibrate and BAY PP1, in a rat model of tubulointerstitial fibrosis: unilateral ureteral obstruction (UUO). We also analyzed the effects of BAY PP1 *in vitro* in renal interstitial fibroblasts and tubular epithelial cells
Results: Compared to unobstructed kidneys, five days after obstruction renal PPAR-α mRNA expression was reduced by 94%. Compared to vehicle treated rats with UUO, BAY PP1 significantly ameliorated the renal cortical expression of collagen type I, III, IV and fibronectin. The number of proliferating tubulointerstitial cells (mitotic figures, PCNA positive cells) and of PDGFR-β/PCNA double-positive cells was significantly lower in BAY PP1 treated rats. Treatment with fenofibrate had no effect on these parameters. The infiltration with monocytes/macrophages was not reduced by either treatment. In vitro, BAY PP1 had no direct effect on the proliferation or expression of fibrosis markers in rat renal fibroblasts. Conversely, rat tubular cells treated with BAY PP1 produced less collagen, fibronectin, TGF-β_1, and the conditioned media of these cells reduced proliferation of fibroblasts.
Conclusions: In conclusion, the PPAR-α agonist BAY PP1, but not fenofibrate, reduced renal fibrosis in rats with UUO by affecting the cross-talk between tubular cells and fibroblasts. These data suggest that novel PPAR-α agonists could be an important treatment option in the early stages of renal fibrosis.
Disclosure: This study was supported by a financial research grant from Bayer Schering Pharma AG, Wuppertal, Germany.

Su417 HEPATOCYTE GROWTH FACTOR (HGF), UROKINASE PLASMIN ACTIVATOR (UPA), UROKINASE RECEPTOR (UPA-R) AND KIDNEY REGENERATION IN NEPHRECTOMISED RATS

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Introduction and Aims: Hepatocyte growth factor (HGF) has been shown to be a factor that possesses mitotic, kinetic and morphogenic properties. Prior activation of plasmin-plasminogen system is required for HGF activation. In addition, plasmin itself appears to play a part, not only in inodolysis, but also in processes that are implicated in the degradation of matrix, denovo carcinogenesis and metastases. Urokinase has been shown to be the main activator of plasminogen in the kidney.
The aim of the study was to investigate kidney regeneration and expession of HGF in nephrectomised rats.
Methods: In Ex-Breeders rats, which have been unilaterally nephrectomised, we used Northern blot analysis in order to study expression of urokinase mRNA and Western blot analysis to study expression of urokinase (uPA), urokinase receptor (uPA-R) and HGF receptor (c-Met). In order to study kidney regeneration we used BRDU infusion.
Results: The results of our study have shown that urokinase mRNA is expressed both in kidney's cortex and medulla from day 0 to day 13th and then disappears. Moreover, maximum urokinase mRNA expression in cortex appears to take place around day 6th, while in medulla maximum

expression appears at day 0 and day 3rd. Kidney regeneration follows an incremental course from day 0 until day 7th when it peaks, and afterwards still exists bur is reduced gradually until day 15th. Urokinase and urokinase receptor are also expressed both in kidney's cortex and medulla only until day 7th. HGF – receptor (c-Met) is expressed from day 0 and peaks at day 7th.

Conclusions: Conclusively, our study has shown that in kidney too, activation of urokinase receptor and urokinase precedes the activation of HGF and maximum expression of HGF coincides with maximum regenerative capacity of the kidney.

Su418 DOES EXPOSURE OF PREGNANT RATS TO CIGARETTE SMOKE CONDENSATE INFLUENCE PODOCYTES NUMBER AND GLOMERULAR STRUCTURE IN THEIR OFFSPRING?

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Introduction and Aims: Disturbances of fetal development caused by maternal smoking may increase the risk of chronic kidney disease and hypertension in the adult life of their offspring. Podocytes are highly specialized glomerular cells with very restrictive regeneration potential. The aim of this experimental study was to assess the impact of exposure of pregnant rats to cigarette smoke condensate on kidney morphology, including quantitative measurement of glomerular cell numbers in their offspring.

Methods: Sprague Dawley rats on day 10^{th} of pregnancy were randomly allocated into two groups (5 animals in each group). Rats from the first group received twice daily on oral mucosa cigarette smoke condensate (CSC) containing nicotine and rats from the second one solvent only until delivery. Glomeruli number per left kidney, mean glomeruli volume, and glomerular cellularity were measured by stereological methods.

Results: At 12 weeks of age significantly higher systolic blood pressure was found in offspring exposed to CSC during the fetal period (n=54) compared to controls (n=51) (122±7 vs 116±8 mmHg; $p<0.001$), respectively. Offspring of mother rats exposed to CSC did not differ from the control offspring with respect to body or kidney weight, albuminuria, serum creatinine concentration and number of glomeruli per left kidney, respectively. In contrast, offspring of mother rats exposed to CSC during pregnancy were characterized by significantly lower mean glomerular volumes (1.15±0.27 vs 1.44±0.26 10^6 μm^3; $p<0.0001$) and calculated per one glomerulus lower number of podocytes (101±13 vs 143±19; $p<0.001$), cells within mesangium (328±45 vs 436±94; $p<0.001$) and endothelial cells (189±43 vs 229±41; $p<0.01$), respectively.

Conclusions: Exposure of pregnant rats to cigarette smoke condensate containing nicotine is related to a decrease of podocyte number and changes of glomerular structure in their offspring which may influence on the future kidney function and development of hypertension.

Su419 ENDOTHELIAL PROGENITOR CELL-DERIVED MICROVESICLES IMPROVE NEOVASCULARIZATION IN A MURINE MODEL OF HINDLIMB ISCHEMIA

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Introduction and Aims: Neovascularization is an important key in rescue tissue after ischemia. It is known that stem cells and endothelial progenitor cells (EPCs) contribute to neoangiogenesis during ischemia. The mechanisms by which EPCs are able to induce a recovery in the damaged tissues are not well understood. Nevertheless, some authors have hypothesized that the release of paracrine mediators by stem cells in injured tissues might be sufficient to induce functional and regenerative events. Recently, we demonstrated that microvesicles (MVs) derived from EPCs are able to activate both *in vitro* and *in vivo* an angiogenic program in human endothelial cells after the incorporation of MVs and transfer of mRNA from EPC. In this study we aimed to investigate whether EPC-derived MVs are able to induce neoangiogenesis and enhance recovery in a murine model of hindlimb ischemia (HLI).

Methods: Severe combined immunodeficient (SCID) mice were divided into three groups (n= 5/group) and treated with 100 mg/proteins of MVs or RNase-inactivated or vehicle alone (control). Hindlimb ischemia was made by ligation and resection of the left femoral artery from the proximal end to the saphenous artery. All treatments were administered intravenously immediately after HLI. Mice were sacrificed at day 7 after HLI and tissues processed for histology. Capillary density was determined in 5 μm-thick frozen sections from the calf muscle stained with anti-mouse CD31 antibodies. Tissue samples were collected at the end of the experiments and the entity of muscle damage/regeneration was evaluated by morphometric and by western blot analysis with eNOS, miogenin, Akt and p-Akt antibodies. The limb perfusion was evaluated by laser Doppler blood perfusion imager.

Results: Quantitative analyses showed that the capillary density in the sections taken from the ischemic limbs was significantly increased in the group treated with EPC-derived MVs compared to the control group (24.4±11.2 vs 10.5±5.4, $p<0.001$); RNase-inactivated MVs abolished the effect induced by EPC-derived MVs (14.7±3.4 vs 10.5±5.4, ns). In addition, we found an increased vascular density in non-ischemic limb in the group treated with EPC-derived MVs compared to the control mice (14.6±7.8 vs 8.2±2.8, $p<0.01$). Morphological and functional studies indicated an enhanced limb perfusion in mice treated with MVs.

Conclusions: The results of the present study indicate that treatment with EPC-derived MVs improves neovascularization in ischemic tissues. Inactivation of RNA shuttled by MVs abolished the angiogenic effects, suggesting that RNA transfer from the MVs to the injured tissues account for this biologic effect.

Su420 DIFFERENT EXPRESSION OF APELIN BY PODOCYTES IN FOCAL AND SEGMENTAL GLOMERULOSCLEROSIS AND MINIMAL CHANGE GLOMERULOPATHY

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Introduction and Aims: Apelin is a vasoactive peptide which is known to directly induce contraction of smooth muscle cells and also described as an angiogenic cofactor for VEGF.

Recently we found glomerular expression of Apelin and its receptor APJ exclusively in podocytes, which suggests an autocrine effect. To investigate the role of Apelin and APJ in podocytes, their expression in renal biopsies with podocyte injury and in normal controls was examined.

Methods: 36 formalin fixed and paraffin embedded biopsies were examined: 12 minimal change disease (MCD), 10 focal segmental glomerulosclerosis (FSGS), 3 IgA glomerulonephritis, 4 membranous glomerulonephritis, 4 diabetic nephropathy and 3 normal controls.

Immunhistochemistry for Apelin and APJ was scaled as positive or negative. The relative expression levels of Apelin and APJ mRNA compared to normal controls were quantified by real time PCR in laser micro-dissected glomeruli.

Results: All normal controls (3/3) and most of the FSGS biopsies (8/10) had positive Apelin immunostaining of podocytes, while most MCD biopsies (9/12) were negative ($p=0.0102$).

In contrast relative Apelin mRNA levels were significantly higher in glomeruli of MCD (mean 1.10) compared to FSGS (mean 0.72; $p=0.0409$), while there was no significant difference between normal controls and either MCD or FSGS.

Real time PCR and immunostains of APJ showed similar results in all specimens examined.

Conclusions: Podocytes in most cases of FSGS seem to retain the normal Apelin immunostaining while it is lost in most cases of MCD.

This contrasts with significantly increased mRNA levels of Apelin in MCD compared to FSGS.
Taken together these data could possibly indicate more rapid secretion of Apelin by podocytes in MCD compared to FSGS, making it undetectable by immunostaining in MCD and preserving the normal Apelin immunoreactivity of podocytes in FSGS.
Ongoing studies examine the autocrine effects of Apelin on podocytes and possible mechanisms of impaired Apelin secretion.

Su421 TUBULOINTERSTITIAL FIBROSIS: POTENTIAL MECHANISMS OF INDUCTION AND REPAIR

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Introduction and Aims: Increased protein filtration and tubular leading to increased production of profibrotic molecules, including the transforming growth factor-beta (TGF-β1), which is a potent inducer of the epithelial-mesenchymal transition (EMT). This mechanism plays a key role in the tubulointerstitial fibrosis. Evidences indicate that the bone morphogenetic proteins (BMP) play a critical role in the repairing processes of the damaged renal tissue, however the activity of BMP is controlled by certain classes of molecules termed BMP antagonists, such as gremlin.
Our aim was to evaluate the role of gremlin in the induction of EMT and reversal of renal fibrosis by an in vitro analysis using immortalized human proximal tubular cells (HK-2) stimulated with TGF-β1 and in in vivo model of tubulointerstitial fibrosis induced by unilateral ureteral obstruction (UUO).
Methods: HK-2 cells were exposed toTGF-β1 (1 ng/ml) for 24 hours. UUO was induced in adult male Wistar rats and renal function and proteinuria were evaluated after 7 days. Gene expression of EMT markers (α-SMA, E-cadherin and fibroblast specific protein 1- FSP1) was analyzed by real time RT-PCR in both HK-2 cells and in the obstructed kidney. The expression of fibrogenic molecules (TGF-β1), and fibrosis markers (fibronectin and collagen) was further analyzed in renal tissue by immunohistochemistry and western blot. The expression of the repair protein BMP-7 and its antagonist gremlin was also evaluated.
Results: HK-2 cells stimulated with TGF-β1 showed increased expression of α-SMA and fibronectin and a decrease in the expression of E-cadherin indicating that the cells were undergo to EMT. Expression of gremlin was increased in HK-2 cells. UUO provoked severe proteinuria and tubulointersticial fibrosis evidenced by a significant increase in the expression of fibronectin and collagen, associated with an upregulation TGF-β1, FSP1 and α-SMA also indicating the presence of EMT in this model in vivo. It was observed an increase in the expression of BMP-7 and gremlin suggesting that, in spite of activation of a potential repair mechanism, the affectivity may be under gremlin control.
Conclusions: TGF-β1 was able to induce EMT in HK-2 cells being a suitable model to study EMT in vitro. Strategies to control BMP-7 and gremlin expressions may improve the endogenous repair mechanisms after the establishment of the tubulointerstiticial fibrosis.

Su422 DETERMINATION OF CD34+ ENDOTHELIAL PROGENITOR CELLS IN PATIENTS ON HEMODIALYSIS AND CARDIOVASCULAR DISEASE

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Introduction and Aims: Cardiovascular disease (CV) is the main cause of morbidity and mortality of patients with chronic kidney disease on hemodialysis. Inflammation, oxidative stress and endotelial disfunction are responsible for these CV disease. Endothelial progenitor cells (EPC) are cells derived from the bone marrow that are thought to be involved in the maintenance and reparation of the vascular endothelium. CD34+ is a surface marker of these EPC. It is known that patients on hemodialysis treatment have a lower count of CD34+. This fact is considered to be responsible for the progression of CV.
The aim of this study was to determine CD34+ EPC in blood sample by flow citometry and establish a possible relation between these cells, main CV risk factors and dialysis parametres of the patients on hemodialysis in our hospital.
Methods: 39 patients on hemodialysis. 65,2% men. Mean age: 65,5±16,3 years old. Months on dialysis: 20 (1-94). We analyzed demographics characteristics, pathological antecedents, dialysis characteristics and treatments. We studied anemia, inflammation markers, nutritional parametres and other cardiovascular risk factors. An echocardiogram was performed on all patients.
Results: Mean CD34+ is 1,53 (0,35-4,13)cells/μL. 86,7% Hypertension (21,7% IECAS); 39,1% dyslipidemia (26,1% statins); 34,8% diabetes mellitus. Vascular access: 58,7% arteriovenous (AV) fistula. Patients with AV fistula have more CD34+ than patients using catheter for hemodialysis (CD34+ EPCs – FAVI 1,76±0,73; CD34+ EPCs – catheter 1,5±1,04; p=0,04). Hemoglobin 11,1 (7,2-13,2)g/dl, medium dose EPO/kg/week 274,9±195,8 units. Serum albumin 3,7 (2,3-4,6)g/dl. Positive correlation between CD34+ EPCs and albumin (r=0,33; p=0,04) was found. Patients with dyslipidemia have lower CD34+ than patients without (p=0,048). Patients who take statins have lower CD34+ EPCs (p=0,012). No correlation was found between CD34+ and other cardiovascular risk factors or echocardiogram analysis.
Conclusions: Patients with dislypemia and those who receive statins have less quantification CD34+. Patients with worse nutritional status (low serum albumin) and those who use a tunelized catheter for dialysis have statistically lower CD34+ EPC than those with AV fistula.

Su423 PUROMYCIN AMINONUCLEOSIDE INCREASES PODOCYTE PERMEABILITY BY THE MODULATION OF ZO-1 VIA OXIDATIVE STRESS

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Introduction and Aims: Puromycin aminonucleoside (PAN)-induced nephrosis is a well-described model of human idiopathic nephritic syndrome because PAN injection into rats results in increased glomerular permeability with the characteristic ultrastructural changes in glomerular epithelial cells (GEpC; podocytes) similar to human nephrosis. We investigated the role of zonula occludens (ZO)-1 and oxidative stress on PAN-induced podocyte phenotypical changes and hyperpermeability *in vitro*.
Methods: Rat GEpCs were incubated in media containing various concentrations of PAN. The phenotypical changes of ZO-1were analyzed by confocal imaging, Western blotting, and PCR. We also examined *in vitro* permeability and oxidative stress level.
Results: Morphological assessment revealed that *in vitro* PAN not only induced the ultrastructural changes of GEpC, such as shortening and fusion of microvilli, but also separated the intercellular gaps and linear ZO-1 resulting in increased intercellular permeability. Oxidative stress level after PAN treatment was markedly higher than that of basal level. PAN induced the inner cytoplasmic translocation of ZO-1 protein and also reduced ZO-1 protein amount and mRNA expression in a dose-dependent manner. These phenotypical changes of podocyte caused by PAN were augmented by the antioxidative effect of vitamin C.
Conclusions: We suggested that the glomerular hyperpermeability caused by intercellular ZO-1 disturbances via oxidative stress would be the mechanism of proteinuria in experimental PAN-induced nephrosis.

Su424 MORPHOLOGY OF BALKAN ENDEMIC NEPHROPATHY – AN UPDATE

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Introduction and Aims: Balkan endemic nephropathy (BEN) is chronic tubulointerstitial kidney disease, which is morphologically presented with diffuse intensive interstitial fibrosis associated with extreme tubular atrophy in contrast to very good preserved glomeruli. The most remarkable morphological characteristic of BEN is that interstitial fibrosis develops without any tubulointerstitial inflammatory process.
At the Institute of Pathology, Faculty of Medicine of the University of Belgrade in the period from 1994 to 2009, in the total number of 9543 autopsies there was only one case of BEN after long term dialysis. In addition, in the period from 2000 to 2009 there were 2082 kidney biopsies diagnosed. Only two of these biopsies were sent as BEN but they were not morphologically confirmed. Since BEN is frequently associated with upper urinary tract transitional cell carcinoma (UUT-TCC), in the present study we analysed the morphology of the renal tissue from nephrectomised patients with UUT-TCC from endemic and non-endemic regions.
Methods: A total of 256 patients with pathologically confirmed UUT-TCC underwent surgical treatment between 1998 and 2009 at the Clinical centre of Serbia in Belgrade, which is a primary national reference centre for urological malignancies. Renal tissues free from tumour invasion obtained by nephrectomy due to TCC of ureter or renal pelvis were morphologically re-evaluated in order to find cases with histological characteristics of BEN. The further task of the present research was to analyse each nephrectomised kidney tissue and make pathomorphological diagnosis without any knowledge whether patient originates from BEN or non-BEN areas. Dimension of nephrectomised kidneys and renal morphological changes in these two groups of patients from endemic and non-endemic regions were statistically assessed.
Results: Multiple sections of kidneys associated with upper urothelial carcinoma were examined in order to find histological changes characteristic for BEN. Out of 215 UUT-TCC patients with reliable data, there were 83 (38,6%) patients from BEN areas and 132 (61,4%) from non-endemic areas of Serbia. Only 5 patients with UUT-TCC with birth and permanent residence in BEN regions had morphological picture similar to BEN. There were no statistically significant differences in morphology, as well as in kidney dimensions between the renal tissue obtained from the patients with UUT-TCC originating from endemic and from non-endemic settlements.
Conclusions: In the last decade, the incidence of BEN and UUT-TCC in BEN regions of Serbia appears to be decreasing. According to our study performed on routine pathological work based on autopsies, routine kidney biopsies and renal tissue obtained by surgical removal of the kidney due to UUT-TCC, we could clearly conclude that BEN today is more clinical and epidemiological than morphological entity.

Su425 MYOFIBROBLASTS INVOLVEMENT IN TUBULAR BASEMENT MEMBRANE REMODELING IN TYPE II DIABETIC NEPHROPATHY

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Introduction and Aims: Diabetic nephropathy is always accompanied by tubulointerstitial damage. The mechanisms and the cells involved in the tubulointerstitial lesions are not entirely clarified. The early phase is characterized by tubular epithelial cell injury and interstitial inflammation. The damaged tubules may regenerate or undergo necrosis or apoptosis. There is some evidence that bone marrow derived fibroblasts play a crucial role in progressive kidney interstitial scaring.
Methods: Kidney biopsies of 23 type II diabetic patients (48% males; 70% with age <60 years; high blood pressure in 61%, proteinuria ≥3.5 g/day in 39% and eGFR <60mL/min in 74%) were examined in electron microscopy and immunofluorescence for a-smooth muscle actin (α-SMA) presence. Diabetic nephropathy was diagnosed using morphologic criteria, e.g. thickened glomerular basement membranes, and glomerular sclerosis either nodular or diffuse.
Results: In optic microscopy, a few tubules were hypertrophic, some atrophic, and some showed a much thickened, double layered tubular basement membrane (TBM).
In electron microscopy, the inner layer of TBMs in contact with the basal pole of epithelial cells was almost normal or slightly lamellated, while the outer layer was larger and sinuous. In between these two layers we found spindle-like cells with the ultrastructural characteristics of myofibroblasts. Some of these cells were in good condition, some were apoptotic or necrotic. In immunofluorescence, α-SMA positive elongated cells were found in the interstitial area, somehow placed around tubules and glomeruli.
The locations of α-SMA positive cells and of the corresponding spindle cells in EM were almost the same, which suggests that these cells were myofibroblasts.
Conclusions: Myofibroblasts seems to be involved in the synthesis of the TBM and extracellular matrix specific proteins, leading to TBM thickening by generating a second layer and finally to renal tubulointerstitial fibrosis.

Su426 HYPOXIA-INDUCIBLE FACTOR-1 (HIF-1) AS A BIOMARKER OF CHRONIC ISCHEMIC TUBULOINTERSTITIAL INJURY IN CHRONIC GLOMERULONEPHRITIS (CGN): ASSOCIATION WITH CLINICAL ACTIVITY AND PERITUBULAR CAPILLARY RAREFACTION

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Introduction and Aims: The aim of the study was to estimate accumulation of HIF-1 alpha active subunit (HIF-1a) in renal tubulocytes in CGN patients and to analyze its association with CGN clinical activity, degree of nephrosclerosis and GFR loss, degree of peritubular capillary rarefaction and expression of angiogenic factors.
Methods: 69 patients were studied: age (M (95% CI)) 34 (31; 37), M/F ratio 44/25, proteinuria 4.8 (3.8; 5.7) g/day, nephrotic syndrome in 52%, GFR by Cockroft-Gault formula adjusted for body surface area (eGFR) 82 (73; 91) ml/min/1.73 m^2.
The intensity of immunohistochemical staining was estimated by semiquantity method (0- no staining, 6 – very intensive and diffuse staining): for HIF-1alpha as a marker of tissue hypoxia, Vascular Endothelial Grouth Factor (VEGF) as the main stimulator of angiogenesis, for Thrombospondin-1 (TSP) as an important antiangiogenic factor, Pax-2 as a marker of renal tubulocytes injury and their dedifferentiation to progenitor-like cells, and CD34 (as a marker of peritubular capillary density) in the renal tubulointerstitium.
Results: The intensity of staining for HIF-1a was associated with proteinuria and arterial hypertension (Table 1).

Table 1. The frequency of intensive staining for HIF-1a (>3) in renal tubular cells in patients with/without arterial hypertension and massive proteinuria

	With proteinuria≥3 g/d	With proteinuria<3 g/d
With hypertension	43%	6%
Without hypertension	18%	0%

Pearson Chi-square=13,15, df=3, p<0.01.

No correlations of HIF-1a tubular expression with eGFR and degree of nephrosclerosis were found. In patients with eGFR<60 ml/min/1.73 m^2 significant decrease of peritubular capillary density (measured by staining for CD34) was revealed which was associated with interstitial TSP elevation (Table 2). Staining for HIF-1a did not correlate with CD34.
Staining for HIF-1a correlated with Pax-2 both in patients with proteinuria ≥3 g/d (Rs=0.73, p<0.0001) and in patients with proteinuria <3 g/d (Rs=0.59, p<0.001).
Conclusions: In CGN with decreased eGFR the significant peritubular capillary rarefaction is developed which is associated with antiangiogenic

Table 2. Association of tubular staining for HIF-1a, VEGF, TSP, and CD34 with eGFR

	HIF-1a	VEGF	TSP	CD34
eGFR ≥60 ml/min/1,73 m^2	2.95 (2.56; 3.34)	3.44 (3.09; 3.79)	1.73 (1.42; 2.04)	5.42 (5.16; 5.68)
eGFR <60 ml/min/1,73 m^2	2.74 (2.31; 3.17)	3.59 (3.05; 4.13)	2.09* (1.78; 2.40)*	4.38# (7.73; 5.23)#

*p<0.05, #p<0.001.

factor TSP expression without compensatory VEGF production. Staining for HIF-1a as a marker for tubular hypoxia can be revealed in early stage of CGN; it is associated with CGN activity and with Pax-2 expression as a marker for tubular injury.

Su427 INCIDENCE AND PREVALENCE OF GLOMERULAR DISEASE IN NORTHERN GERMANY

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Introduction and Aims: Knowledge of possible numbers of patients suffering from chronic disease possibly requiring cost-intensive permanent treatment is mandatory for future health care plans. Despite some regional biopsy registries no valid data about the exact epidemiology of glomerular diseases in Germany exist because all publications are hampered by their retrospective character and lack of completeness.
Methods: In an unique co-operation of out-patient nephrologists with a single major teaching hospital serving a population of approx. 400,000 in the capital of Schwerin and surrounding counties, all patients with abnormal urine findings and/or decreasing renal function of unknown cause were referred for renal biopsy between October 2002 and December 2008. The drop-out rate is guessed to be less than 5%. All biopsies were analysed according to international standards and traditional epidemiological and renal parameters were collected for comparison with the micro-census of the state Mecklenburg-Lower Pommerania of the year 2008. We present the first valid figures for incidence and 7 year prevalence of glomerular disease in Germany.
Results: In 222 patients, 251 renal biopsies were performed. The annual biopsy rate was 64 ppm (range 46.2-87.2), one of the highest in Europe, indicating an almost complete regional survey of patients. Glomerulopathy was diagnosed in 32 per 100,000 population with a mean age of 51±17 years (range 17 – 89). Sixty three percent were male. The prevalence of primary or secondary glomerulonephritis was 29 per 100,000 with two third of primary diseases. The annual incidence for histological subgroups is given in table 1.

Annual incidence of glomerulopathies in Northern Germany

Year	MCN	FSGS	MesGN	MGN	RPGN	LN	ANCA
2003	1.5	7.6	13.7	3.0	6.1	1.5	4.6
2004	1.5	9.2	16.9	6.9	0	1.5	1.5
2005	4.7	18.7	26.5	3.1	7.8	3.1	9.3
2006	3.1	3.1	17.3	4.7	0	4.7	1.6
2007	1.8	11.0	27.6	5.5	9.2	1.8	9.2
2008	6.4	17.7	19.3	8.0	6.4	4.8	6.4
Mean	3.2	11.2	20.2	5.2	4.9	2.9	5.4

MesGN, mesangioproliferative glomerulonephritis incl. IgA nephropathy; LN, lupus nephritis; ANCA, vasculitic nephropathy.

In 125 patients, information about tubulointerstitial fibrosis was reported and in 45% of the biopsies fibrosis was 30% and more.
Conclusions: The prevalence of glomerular disease in North Germany is 32 per 100,000 population which is comparable to other European countries with a public financed centralised health care system. However the rate of significant tubulointerstitial fibrosis on renal biopsy as an indicator for poor response to specific therapy is high, indicating late referral for diagnostic evaluation of renal syndromes.

Su428 PHOSPHOLIPID SCRAMBLASE 1: A MARKER OF MESANGIAL CELLS?

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Introduction and Aims: Phospholipid scramblases (PLSCR) constitute a family of 4 membrane proteins with a number of cell functions. PLSCR1 was originally identified as a strong candidate protein responsible for the bi-directional redistribution of phospholipids between the two leaflets of the plasma membrane that is observed during cell apoptosis or cell activation. More recently PLSCR1 was also identified as a transcription factor. Finally, we and others, have demonstrated that PLSCR1 can be a signaling intermediate that is capable to increase the intensity of signaling pathways during cell activation. Considering its potential role in cell apoptosis and in cell activation, dysregulation of PLSCR1 function could be involved in renal diseases. Therefore, as a first step, expression of PLSCR1 was investigated in the kidney.
Methods: Expression of PLSCR1 in kidneys was analyzed by immunohistochemistry of frozen kidney biopsies from wild-type and PLSCR1-KO mice. Immunohistochemical analysis of rat kidney biopsies were performed with two different monoclonal antibodies raised against rat PLSCR1. Human primary mesangial cells were analysed by immunoblotting of cell lysates with a polyclonal antibody raised against human PLSCR1, and by immunoprecipitation with an anti-human PLSCR1 monoclonal antibody.
Results: Immunohistochemical analysis of mouse kidney biopsies revealed a strong expression of PLSCR1 in glomeruli while no significant expression was observed in tubules. No expression of PLSCR1 was observed in samples from PLSCR1-KO mice, demonstrating the specificity of the staining. Anti-PLSCR1 staining of glomeruli was confirmed in rat biopsies with two different monoclonal antibodies raised against rat PLSCR1. This expression was further confirmed in human primary mesangial cells with another couple of antibodies both in whole cell lysate and after specific immunoprecipitation.
Conclusions: In the kidney, PLSCR1 is observed exclusively in mesangial cells by immunohistochemical analysis of kidney biopsies in non-pathological situations. Its potential role in the activation of these cells and in glomerulonephritides is discussed.

Su429 NEPHRIN EXPRESSION IN LUPUS NEPHRITIS: CORRELATION WITH SEVERE GLOMERULAR LESIONS

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Introduction and Aims: Lupus nephritis (LN) has been associated with immune complex deposition in the subepithelial (predominantly class V) and subendothelial (predominantly proliferative class IV) glomerular capillary wall as well as endocapillary proliferation and inflammation. Immune-mediated podocyte injury in LN is thought to be induced by local release of proinflammatory cytokines and T helper cell infiltration, as a result of complement activation. Nephrin is a transmembrane receptor molecule located at the podocyte slit diaphragm, that has been identified as an essential protein for the maintenance of podocyte and glomerular barrier integrity. In order to investigate the degree of podocyte involvement in glomerular injury in LN, we studied the expression of nephrin in various classes of LN.
Methods: A three-step immunohistochemical method was applied to paraffin-embedded tissue sections from 56 patients with lupus nephritis (7 class II, 19 class III, 18 class IV, 12 class V) and 10 control samples of normal kidney tissue surgically removed from patients with renal cell carcinoma. Patient's clinical data (hematuria proteinuria, serum creatinine) at the time of biopsy was also recorded. The immunohistochemical staining was evaluated by using a semi-quantitive method. Statistical analysis was performed using Fisher's exact test and one-way ANOVA.
Results: Control samples showed intense staining of nephrin along the glomerular basement membrane (4+), while in the LN tissue samples

a reduction in staining intensity was observed(1+ to 3+). Nephrin stain intensity varied significantly among the different LN classes ($p<0,05$). In particular, the intensity of staining was mainly reduced in LN class III and IV in areas of severe endocapilary proliferation, necrotizing lesions and crescents. On the contrary, in class II and V LN, a mild staining decrease was observed. Nephrin staining was also reduced in cases of class V LN with coexisting class III lesions, especially in areas of endocapilary proliferation. An inverse correlation between Nephrin staining and proteinuria was found in patients with severe disease activity (LN class III and IV). The remarkable reduction of staining observed in these patients, correlated with higher levels of proteinuria. ($p=0,039$).

Conclusions: The intensity of nephrin staining was found significantly decreased in LN class III and IV, especially in patients with severe disease activity and increased proteinuria. These results suggest that podocyte injury, as expressed by nephrin down-regulation, is probably induced by strong complement activation and local release of proinflamatory cytokines in LN with severe histology lesions.

Su430 CORRELATION OF HISTOPATHOLOGIC PARAMETERS IN BIOPSY PROVEN POLYOMAVIRUS NEPHROPATHY WITH PERIPHERAL BLOOD VIRUS LOAD AND RENAL FUNCTION

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Introduction and Aims: Polyomavirusnephropathy (PVN) is an incremental and serious infection after renal transplantation, leading to graft loss in about 50%. On the 10th Banff conference on allograft nephropathy a new classification of PVN based on that one reported by Drachenberg et al. In 2005 has been proposed and encouraged for further evaluation (Sis B. et al, AJT Jan. 2010). Aim of the present study was to correlate histopathologic parameters included in the revised classification system with serum creatinine and peripheral blood viral load (PBVL).

Methods: Starting January 2008, all biopsies were routinely stained immunohistochemically for SV40 LargeT-antigen indicating PVN. At the same time points, blood was tested by quantitative polyoma BK virus PCR. 24 biopsies from 19 patients with biopsy proven PVN were reevaluated for the following paramters: Interstitial fibrosis/tubular atrophy, tubulitis, interstitial infiltrate, cortical/medullar area affected by PVN (%), tubules with viral replication in cortex/medulla (%), presence of cytopathic effect, capillaritis, viral replication in medulla/cortex present. Results were correlated with PBVL and serum-creatinine.

Results: PBVL was 2500 to >100000000 copies/ml (mean 4920412 copies/ml), serum-creatinine was 118 to 347 μmol/l (mean 212 μmol/l). The area affected of PVN in the cortex was 0-100% (mean 45%), in the medulla 0-100% (mean 54%); both correlated with PBVL ($p=0.0122$, $r=0.523$; $p=0.0111$, $r=0.734$ respectively). The proportion of tubules affected by PVN in the cortex was 0-30% (mean 9.1%) and 0-50% in the medulla (mean 16.7%); both correlated with PBVL ($p=0.052$, $r=0.775$ and $p=0.007$, $r=0.748$ respectively). PBVL was higher, when PVN was present also in proximal tubular epithelial cells. PBVL did not correlate with interstitial fibrosis/tubular atrophy, tubulitis, interstitial infiltrate, presence of cytopathic effect or capillaritis; none of the parameters was associated with serum-creatinine.

Conclusions: Impairment of transplant function as reflected by serum creatine seems to be multifactorial and not solely related to PVN associated damage. Histopathologic findings like the proportion of cortical and medullar area and proportion of tubules affected by viral replication reflect the BK viral load and should be included in a new classification of PVN, as it has been proposed.

Su431 THE INFLAMMATORY INFILTRATE: A POTENTIAL CLUE TO DIFFERENTIATE BK NEPHROPATHY FROM T-CELL MEDIATED REJECTION IN KEDNEY TRANSPLANT?

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Introduction and Aims: It is notoriously difficult to differentiate T-cell mediated rejection from polyoma virus infection in the context of kidney transplantation. Although stainings for HLA-DR, C4d and polyoma virus can sometimes be of great assistance,the definite decision may be problematic. Therefore, it was investigated whether the composition of the inflammatory infiltrate, with special regard to Treg, can give a hint towards the derivation of the inflammation.

Methods: 18 renal biopsy specimens with immunohistochemically proven polyoma virus infection were compared with 22 specimens with interstitial T-cell mediated rejection. Immunohistochemical stainings with antibodies against CD3, CD8, c-maf, FoxP3, CD20 and CD21 were performed to characterize the infiltrate, which was subsequently quantified. Statistical analysis was performed.

Results: Comparing the subtypes of lymphoid cells, there was in general a predominance of T cells (CD3) over B cells (CD20). Th2 (c-maf) cells usually outnumbered cytotoxic T cells (CD8). Treg (FoxP3) were the smallest subgroup of T cells with only few scattered cells in the infiltrate. No differences were observed in the composition of the infiltrates in BK nephropathy compared to T-cell mediated rejection.

Conclusions: The characterization of the inflammatory infiltrate does not serve as a discrimination aid to differentiate BK nephropathy from T-cell mediated rejection. Independent of the antigen-stimulus the infiltrate is dominated by T cells. The number of Treg is very small and only few scattered cells are found.

Su432 COST-EFFECTIVENESS OF PERCUTANEOUS RENAL BIOPSY WITHOUT HOSPITALIZATION

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Introduction and Aims: Renal biopsy is the gold standard method for the study of many renal diseases. Devices under guidance with ultrasound have allowed incorporate another measurement in our routine practice even in advanced chronic renal disease.

Hypothesis: Percutaneous renal biopsy is a safe method for outpatients and can reduce the hospitalization.

Objectives: Evaluate the feasibility and safety of an outpatient renal biopsy protocol, and measuring hospital costs-efficiency of the patients who enter of programmed form to performed a renal biopsy.

Methods: A prospective study was developed. Entrance at 9:00 A.M. to the short stay hospital. Automatic needle of 16G was used. Blood pressure, heart rate, hemoglobin, hematocrit and diuresis were monitored for 6 hours postbiopsy. Possible complications (hematuria, hypotension, shock) were evaluated and used to measure the effectiveness. Discharged home at 20:00 PM if no contraindication was present. Medical direct cost were registered.

Results: During an 18 month period, 51 consecutive patients were underwent outpatient renal biopsy. Thirty-three 33 (65%) biopsies of own kidneys and 18 (35%) biopsies of renal grafts. Description of the sample: mean age 46±13. Sixty-one % male. Serum creatinine before biopsy 1.5±0,9 mg/dL and 1.7±1.0 after biopsy and, hemoglobin before biopsy 13.2±1.9 g/dL and after biopsy 12.2±2.1 g/dL (no significant differences). Thirty-five (75%) patients achieved the objective. There were some minor complications: 1 hematuria, 1 hiperkaliemia, 2 intrarenal haematoma and 1 local haematoma. The outpatient renal biopsy protocol supposed reduction in one third of the cost and half of complications (effectiveness).

Conclusions: Percutaneous renal biopsy is safe and permit a reduction of the postbiopsy hospitalization (effectiveness) and the cost minimization. It can result in significant savings cost without exposing the patients to an increased risk of complications.

Su433 IMMUNOHISTOCHEMICAL EVALUATION OF CIC-5 IN GLOMERULI OF PATIENTS WITH MEMBRANOUS GLOMERULONEPHRITIS

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Introduction and Aims: ClC-5 and Megalin are part of the same macromolecular complex involved in the re-uptake of albumin in proximal tubuli. In a previous study we have demonstrated at gene expression level the up-regulation of ClC-5 in the glomerular compartment of IgAN and diabetic nephropathy with mild proteinuria. It is well known that in Heymann nephritis the pathogenetic antigen is megalin which is expressed also in podocytes. The aim of this work was to further explore the relationship between glomerular ClC-5 and proteinuria by studying its expression in human Membranous Glomerulonephritis (MG).
Methods: Expression of glomerular ClC-5 was investigated by immunohistochemistry using polyclonal antibodies anti ClC-5 (Santa Cruz) and morphometric analysis (Image Pro-Plus software Media Cybernetics) in 14 renal biopsies of patients with MG. As controls we used 4 cortical tissues obtained from sites remote from tumor bearing renal tissue.
Results: We found that: 1) ClC-5 protein was present both in control and MG glomeruli; 2) the immunohistochemical signal was significantly higher in MG glomeruli ($p<0,0018$) and mainly localized in podocytes; 3) glomerular ClC-5 expression did not correlate with proteinuria (1,92-8,32 g/24h) or serum creatinine and Transmission Electron Microscopy (TEM) stages; 4) podocytes observed by TEM revealed the presence of numerous small vesicles whose relation with proteinuria and ClC-5 will be studied.
Conclusions: Our study demonstrates that ClC-5 is localized in podocytes and that it is up-regulated in MG, thus confirming its role in proteinuric glomerular diseases. Furthermore, it suggests that ClC-5 might have a role in podocyte endocytosis of proteins.

Su434 FIBRILLARY GLOMERULOPATHY IN A PATIENTS WITH MULTIPLE MYELOMA

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Introduction and Aims: Glomerulopathies with congo red-negative organized immunoglobulin (Ig) deposits are rare diseases and are not thoroughly studied. According to researches, fibrillary glomerulopathy (FG) and immunotactoid glomerulopathy are diagnosed much more frequently in patients with monoclonal gammopathy than in renal diseases not related to hematological disorders.At monoclonal gammopathy the combination monoclonal immunoglobulin deposits and fibrils is quite frequently observed.There is hypothesis that fibrils are formed from amorphous deposits, i.e.FG is the further stage of monoclonal immunoglobulin deposits disease (MIDD).
Aims of the research; to study a role of fibrillar deposits in pathogenesis of renal failure in patients with multiple myeloma (MM).
Methods: Twenty MM patients with severe renal insufficiency have been examined. Before treatment renal biopsy was tested by light, immunofluorescent and electron microscopy.In one case after achievement complete remission the repeat renal biopsy was performed.
Results: In four patients (20% of cases) fibrils with thickness of 12-16 nm were found in glomerular basement membrane. Congo red stain was negative. In all patients it was observed the combination of fibrillar deposits with other renal diseases (for two patients myeloma cast nephropathy, in three – MIDD). In one case after chemotherapy was achived the complete remission with normal urine tests and renal function. Before treatment the patient had monoclonal Ig Gk in his serum and he excreted IgGk into the urine. In renal biopsy it was observed immunofluorescent stain IgG, IgA, IgM and kappa light chains in glomerular basement membrane and kappa light chains in the mesangium.After achivement of remission and stopping of secretion of a paraprotein only fine staining along glomerular basement membrane for IgM and C3 were found out. Staining for kappa and lambda light chains were not found. A full resorbtion of granular deposits with formation electronic-transparent cavities in the absence of changes in fibrillar deposits were obsedved.
Conclusions: 1. Fibrillary glomerulopathy in MM patients with severe renal failure was diagnosed in 20% of cases. It testifies to high frequency of similar renal disease at the given contingent of patients.
2. In the most cases FG is combined with MIDD. At achievement complere remission it is possible full resorption of granular deposits. Fibrils do not undergo a reverse process.
3. Fibrillar deposits doas not influence a clinical picture of disease. After the resorption of granular deposits full clinical remission in the absence of changes from outside fibrillar adjournment is possible.
4. Accumulation of fibrils can be a manifesstation of "misdirected regeneration" targeting at restoration of integrity of glomerular basement membrane after granular deposits removal.
Disclosure: Fibrillary glomerulopathy in a patients with multiple myeloma.

Su435 ANTIPHOSPHOLIPID ANTIBODIES (aPL) AND aPL NEPHROPATHY IN PATIENTS WITH SYSTEMIC LUPUS ERYTHEMATOSUS

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Introduction and Aims: Antiphospholipid antibodies (aPL) are associated with the presence of specific histological lesions in renal biopsies, named antiphospholipid syndrome (APS) nephropathy.
To determine the association between the positive aPL and APS nephropathy in patients with systemic lupus erythematosus (SLE).
Methods: Patients were included if the following criteria were present: a diagnosis of SLE nephritis (ACR criteria) requiring renal biopsy, a positive aPL sample at the time of the biopsy, and at least two positive determinations being. Persistent positivity was defined as more than two-thirds of the aPL determinations positive during follow-up. The associations between type of aPL and renal histological lesions biopsies were analyzed.
Results: Thirty-seven biopsies from 36 SLE patients were included (30 female [83,3%]). The mean age at time of biopsy was $30,9\pm10,3$ years (range, 15-63). An average of $12,2\pm6,9$ (range, 2-27) aPL samples were collected during a period of $110,4\pm61,4$ months (range, 3-211). The follow-up period (time between the renal biopsy and the last assessment of aPL) was $71,8\pm60,0$ months (range, 0-208). There were $3,3\pm2,6$ (range, 1-11) positive records of aPL/patient. Twenty-four (64.9%) patients were positive for lupus anticoagulant (LA), 29 (78.4%) for IgG-type anticardiolipin antibodies (aCL-IgG), and 18 (48.6%) for IgM-type anticardiolipin antibodies (aCL IgM). Three (8.1%) patients had persistent LA, 5 (13.5%) aCL IgG, and one (2.7%) aCL IgM.
Nine (24,3%) renal biopsies met diagnostic criteria for APS nephropathy. The following histological lesions were found: 3 (8.1%) cases of thrombotic microangiopathy (TMA), 4 (10.8%) of fibrous intimal hyperplasia (FIH), and 3 (8.1%) cases of focal cortical atrophy (FCA). We found association between persistent positive LA and APS nephropathy ($p<0.02$), but not with the persistence of aCL-IgG ($p=0,18$) or aCL-IgM ($p=0,48$). The relative risk of developing APS nephropathy in SLE patients with persistently positive LA was 5.6 (95% IC 2.7-11.7).
Conclusions: Development of APS nephropathy accounted in more than fifth of the SLE nephritis with positive aPL sample at the time of renal biopsy and this was associated with the persistence of positivity LA.

Haemodialysis 2

Su436 NATRIURETIC PEPTIDES AND FLUID STATUS IN HEMODIAYSIS (HD): READY TO USE THEM IN THE CLINICAL PRACTICE?

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Introduction and Aims: Natriuretic peptides (NP) are described as biochemical markers of the fluid status. To assess their usefulness on a clinical ground in the chronic HD patient, we evaluated the behaviour of two peptides, adrenomedullin and atrial natriuretic peptide (ADM, ANP) in 50 randomly selected hemodialysis patients (28 conventional HD, 23 hemodiafiltration – HDF).
Methods: Due to the short half-life of the peptides, we measured the plasma levels of their prercusors (medium region pro-ADM – molecular weight 2,460 d – and pro-ANP – molecular weight 4,000 d – respectively; immunofluorescence assay, BRAHMS Kriptor) pre- and post a mid-week HD session. Dry body weight had been previously clinically determined. The interdialytic body weight increase was taken as a measure for fluid overload, while the ultrafiltration (UF) volume was the measure of fluid removal. Any relationship between the peptide levels or their changes, and fluid overload, fluid removal and blood pressure values was the object of the study.
Results: The basal levels of both the peptides (proADM 3.6±1.8 mmol/L; proANP 836.1±438 pmol/L) did not correlate either each other (R=0.06) or with pre-HD fluid overload (ADM: r=0.11 and ANP: r=0.03) or blood pressure.
Both decreased significantly during HD (proADM: from 3.6±1.8 to 1.8±1.4 mmol/L, p<0.0001; proANP: from 836.1±438 to 599.8 +456 pmol/L, p< 0.0001) but their pre-to-post HD changes (%) did not correlate with the ultrafiltration volume (UF).
Moreover, the % change of both the peptides was significantly different in HD (low flux polysulphone membranes) compared with HDF (high flux polysulfone, polyacrylonitrile) in spite of similar UF. However, the pre-dialysis levels was comparable.

Results

	HD	HDF	p
Pre-dialysis ADM (μmol/L)	3.6±1.5	3.7 ± 2.1	NS
Pre-to-post dialysis ADM (%)	-37.4 ± 20.7	-64.7 ± 14.8	<0.0001
Pre-dialysis ANP (pmol/L)	890.1 ± 495	770.4 ± 357	NS
Pre-to-post dialysis ANP (%)	-15.8 ± 24.6	-49.13 ± 16	<0.0001
UF (L)	1.83 ± 0.9	1.9 ± 1.3	NS

The correlations between pre or post-dialysis peptide levels and pre or post-dialysis blood pressure were not significant.
Conclusions: To conclude, these data do not support the hypothesis of an association between the fluid status *per se* and NP levels. Other determinants (cardiac function, other?) probably come into play modifying this relationship. In spite of a greater reduction ratio of the NP levels with the convective-diffusive techniques, due to the medium-range molecular weight, pre-dialysis values are not lower than in HD, also suggesting a multiple control of their concentration. The use of the NP precursors in the day-by-day clinical setting still needs further studies.

Su437 HIGH CUT-OFF RENAL REPLACEMENT THERAPY FOR EXTRACORPOREAL ELIMINATION OF MYOGLOBIN IN SEVERE RHABDOMYOLYSIS AND ACUTE KIDNEY INJURY

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Introduction and Aims: Rhabdomyolysis is associated with the release of myoglobin into the circulation. Myoglobin promotes acute kidney injury (AKI) by cellular toxicity, altered renal hemo-dynamics and haeme pigment tubular obstruction. In rhabdomyolysis patients, dialysis-dependent AKI doubles mortality. Myoglobin removal by standard blood purification techniques has limited efficacy and prognostic impact. We describe the use of high cut-off (HCO) protein permeable filters as a novel concept for rapid extracorporeal elimination of myoglobin in rhabdomyolysis.
Methods: With an in vivo molecular cut-off at 45 kD, HCO filters are effective in removing myoglobin (17.8 kD). Elimination kinetics across standard and HCO filters were compared intraindividually using continuous or intermittent renal replacement therapy (CVVHD, SLEDD, hemodialysis (HD)) in patients with severe rhabdomyolysis and AKI. Elimination rates and clearances were calculated according to standard formulae.
Results: Median myoglobin clearance, normalized for m^2 of membrane surface area on CVVHD was 1.2 [interquartile range 0.8 – 1.6] ml/min. Using standard high-flux hemodialysis, median myoglobin clearance was 2.4 [1.7 – 4.2] ml/min. Up to 20-fold higher clearances were obtained using HCO filters in both continuous (SLEDD: 20.3 [18.8 – 22.7] ml/min) and intermittent (HD: 40.0 [38.7 – 42.9] ml/min) dialysis techniques. Using full size (2.1 m^2) HCO filters, median absolute myoglobin clearances in excess of 70 ml/min were achieved, resulting in rapid and highly effective reduction of plasma myoglobin concentration. Albumin losses across HCO filters require substitution over time.
Conclusions: As a novel treatment option, HCO renal replacement therapy allows the safe and highly efficient elimination of myoglobin in severe rhabdomyolysis and acute kidney injury. Limiting exposition of the kidney to myoglobin may aid renal recovery in these patients.

Su438 SUPPRESSION OF PLATELET ACTIVATION ON THE SURFACE OF DIALYSIS MEMBRANE RELEASING NITRIC OXIDE FROM THE DIALYSATE

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Introduction and Aims: Vascular diseases, including arteriosclerosis and coronary artery disease, are accelerated in patients undergoing hemodialysis. Increased oxidative stress and platelet reactivity are the most probable causes of initiation and progression of these diseases. In the blood vessels, nitric oxide (NO) generated by the nitric oxide synthase on the endothelial cells suppresses the aggregation and activation of platelets and the migration and adhesion of monocytes. Therefore, the biocompatibility of dialysis membranes can be improved with the use of a dialysis membrane with an NO-releasing surface. The aim of the present study was to examine whether dialysis membrane surface releasing NO from the dialysate can suppress platelet activation.
Methods: Porcine whole blood with sodium citrate (10 mM) was circulated for 4 hrs through either a polysulfone dialyzer (TS-1.6UL, Toray Medical, Japan) or polymethylmethacrylate dialyzer (BQ-1.6PQ, Toray Medical, Japan). Before circulation the dialyzer was primed with saline containing nafamostat mesilate. After the blood was circulated in the blood circuit and dialyzer, NO was added using an NO donor (sodium nitropruside) infused into the re-circulating dialysate. During circulation of the blood, samples were collected every 30 minutes for determination of the changes in the platelet activation status and platelet counts. Platelet activation was evaluated by platelet aggregate formation in response to ADP or collagen detected by screen filtration pressure using WBA Karuna (IMI Co. Japan).
Results: Platelet aggregation in response to ADP or collagen in the blood coming in contact with an NO-releasing membrane surface was significantly decreased as compared to that in blood coming in contact with a non-NO-releasing surface at 30 min after the start of the experiments (n=4, p<0.01 for PS, n=3; p<0.05 for PMMA, in response to both ADP and collagen). Suppression of platelet activation on the NO-releasing membrane surface was maintained until the end of the experiments (240 min). Decrease in the platelet count observed in both dialyzers without NO release was also significantly suppressed in the blood in contact with the NO-releasing surface. These results indicate that the NO-releasing surface suppressed platelet activation and the subsequent consumption of platelets occurring as a result of the formation of aggregates and/or adhesion of platelets on the dialysis membrane.

Conclusions: NO-releasing dialysis membrane surface was capable of suppressing platelet activation which was induced by its contact with the dialysis membrane.

Su439 DIALYTIC PERFORMANCE EVALUATION OF A NEW HIGH FLUX MEMBRANE IN On Line HDF: EXPERIENCE OF A FRENCH DIALYSIS CENTER

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Introduction and Aims: The importance of high flux dialysis as the appropriate way of dialysis is continuously increasing. Results from MPO study clearly showed the superiority of high flux dialysis for diabetic type II and malnourished patients in regards to survival. Increasing removal of middle molecules (MM) like béta2microglobulin (Beta2M) or others uremic toxins of higher molecular weight might be a major reason for this significant influence on patient's benefit compared to low flux dialysis. Using On Line HDF, dialyzers with a high ultrafiltration coefficient (KUF) offer a challenging system to support effective removal of MM while preventing the loss of albumin during dialysis. Xevonta Hi23 is a new dialyzer using a newly developed polysulfone-based membrane (amembris) with unique hydraulic permeability (KUF = 124).
Methods: Nine stable dialysis patients, 6 females and 3 males were included in the study during 6 months. Patients were treated for 4 weeks with Fresenius FX 1000, followed by ASAHI Rexeed 21 for 4 weeks, XEVONTA HI 23 for 8 weeks, Gambro POLYFLUX 21 for 4 weeks and ASAHI REXEED 25 for the last 4 weeks. The dialysis was performed by a 24 liters of post-dilution On Line HDF (Qb = 400 ml/mn and Qd = 850 ml/mn). Removal of béta2microglobuline and albumin was measured using spent dialysate collected thought a reversed injection pump during the whole dialysis session.
Results: With regards to small molecule clearance and removal no dialyser showed significant advantages. Urea Sp KT/V was around 2±0.2 and did not differ significantly. *In vivo* Urea, Creatinine and Phosphate clearances were stable over time of study, showing, for each membrane, a average high Urea clearance of 340 ml/mn, Creatinine clearance of 270 ml/mn and Phosphate clearance of 290 ml/mn.
Albumin and béta2M were collected as described and absolute mass transfer was determined. Beta2M RR following Bergström formula was not significantly different between FX 1000 (81.68±4.6%), XEVONTA HI 23 (81.56±3.8%), REXEED 21 (82.6±3.8%), REXEED 25 (84.6±2.68%). Only POLYFLUX 21 showed a lower RR of 77.06±5.5% under study conditions.
Albumin loss was siginificantly elevated for REXEED 21 (4.82±2.5 G/session) and REXEED25 (5.41±3.8 G/session) compared to FX 1000 (1.2±0.4 G/session), POLYFLUX 21 (1.21±0.6 G/session) and XEVONTA HI 23 (2.3±0.8 G/session).
Conclusions: The Urea spKT/V observed in our study is a strong argument to use On Line HDF in order to increase the urea RR, whatever the membrane used. On the other hand, our study showed that Beta2M RR (and MM reduction rate) has to be compared to the dialysis albumin loss level. The right balance between Beta2M-removal and albumin retention is displayed by dialyzers such as XEVONTA HI 23 or Fresenius FX 1000. High efficiency of MM reduction rate as not to be accompanied by a high albumin loss rate.
Disclosure: Our clinical research programm was sponsorised by BBraun.

Su440 DETERMINATION OF DIALYSIS DOSE: A CLINICAL COMPARISON OF METHODS

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Introduction and Aims: Guidelines recommend regular measurements of the delivered HD dose Kt/V using a validated method. As alternatives to the conventional method automatic on-line measurements are available now. In a large population covering different centres and countries the conventional method and two on-line methods were simultaneously applied and compared.
Methods: Comparison of dialysis dose determination was investigated in a multi-centre trial: the conventional method with urea concentrations from pre- and post-dialytic blood samples and Daugirdas formula (Kt/V_{Dau}), the automatic on-line clearance monitor OCM measurement with the urea distribution volume V based on the anthropometric estimate of Watson (Kt/V_{OCM}), and the automatic on-line clearance measurement with the urea distribution volume V measured with bioimpedance spectroscopy by the Body Composition Monitor BCM (Kt/V_{BCM}).
Results: In 18 German and Czech dialysis centres 1606 patients on HD or on-line-HDF were screened whether eligible for the study, 1089 patients were enrolled, and 1076 patients with full data set were analyzed.
In the analysis cohort dialysis dose was measured as $Kt/V_{Dau} = 1.74\pm0.45$, $Kt/V_{OCM} = 1.47\pm0.34$, and $Kt/V_{BCM} = 1.65\pm0.42$.
Bland-Altman plots on difference reveal increasing variability with increasing Kt/V, prefering ratios for comparison of methods instead of differences. On average, Kt/V_{OCM} resulted in 16% lower values compared to Kt/V_{Dau}, whereas Kt/V_{BCM} was 5% lower than Kt/V_{Dau}.
For Kt/V_{Dau} occasionally outliers were observed due to extremely low post-dialytic urea concentrations (probably artefacts), whereas both OCM based dose measurements Kt/V_{OCM} and Kt/V_{BCM} during the same treatment delivered plausible values. Correlation between Kt/V_{Dau} and Kt/V_{BCM} was 0.74, and increased to 0.81 without outliers.
Conclusions: Due to the automatic procedure the on-line clearance measurement with Watson estimate of urea distribution volume was easiest to use, but the difference with the conventional method was larger; the automatic on-line clearance measurement with the urea distribution value measured by BCM had a higher correlation to the conventional method.
The conventional method based on blood sampling was occasionally prone to errors, whereas the automated dialysis dose determination with OCM and urea distribution volume measured by BCM delivered plausible values.
Disclosure: The following authors are employees of Fresenius Medical Care (FME): P.Taborsky, J.Vlasak, P.Machek, M.Sagova, P.Moucka, I.Rychlik, P.Vyskocil, J.Possnickerova, R.Wojke.

Su441 CLINICAL APPLICATION OF REAL-TIME Kt/V DETERMINATION BY ULTRAVIOLET ABSORBANCE (Adimea®) IN SINGLE-NEEDLE-CROSS-OVER (SNCO) DIALYSIS AND HEMODIAFILTRATION MODES (HDF-ONLINE)

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Introduction and Aims: Kt/V determination by Adimea® has been established in regular dialysis (Castellarnau et al., EDTA 2009). The aim of this study was to compare the Kt/V-values determined by blood sampling according to Daugirdas with the Kt/V-values determined by the Adimea® device in single-needle-cross-over and hemodiafiltration-online dialysis.

Methods: For SNCO, two studies (”control” and ”routine”) were performed to compare the Kt/V determination either with Adimea® and Daugirdas. In the “control” study, 9 uremic patients (6 female and 3 male) on thrice weekly hemodialysis were included in the study. The blood was sampled every 20min during the dialysis treatment. The blood based Kt/V was calculated by fitting the timely exponential decay of blood urea concentration. In the “control” study, 11 uremic patients (8 female and 3 male) on thrice weekly hemodialysis were included. The blood was sampled pre and post dialysis following the methodology described in the KDOQI guidelines.
For HDF-online, a “control” study was performed to compare Adimea®-Kt/V to blood-Kt/V calculated by 20min blood sampling and exponential fitting of the decay of blood urea concentration.
In all studies, the UV-absorbance at 280 nm of the spent dialysate was measured every three minutes with a UV-spectrophotometer (Option Adimea®, B. Braun) coupled with the water system of the dialysis machine (Dialog+, B. Braun). The Daugirdas equations were applied to calculate the blood-Kt/V parameters. A total of n=222 (“control”: n=83, “control”: n=139) measurements were performed for SNCO and n=80 measurements were performed in the HDF-online “control” study.
Results: In the SNCO “control” study, the mean blood-spKt/V value was 1,28±0,21 and the mean Adimea®-spKt/V value was 1,25±0,18. The mean difference between both spKt/V values was -1,24%±10,5%. The Pearsons correlation coefficient between blood and Adimea®-spKt/V values was 0,77. In the SNCO “routine” study, the Pearsons correlation coefficient between blood and Adimea®-spKt/V values was 0,85. A Bland-Altman plot indicated an even distribution of measurement values. In the HDF-online “control” study, the mean blood-spKt/V value was 1,69±0,24 and the mean Adimea®-spKt/V value was 1,69±0,24. The mean difference between both spKt/V values was 0,21%±9,5%. The Pearsons correlation coefficient between blood and Adimea®-spKt/V values was 0,81.
Conclusions: The results show both a close correlation and accordance between the blood-spKt/V and the Adimea®-spKt/V values. Adimea® can therefore be used for determination of dialysis dose not only in regular HD but also in other dialysis modes, such as SNCO and HDF-online.

Su442 A COMPARISON BETWEEN THREE METODS TO DETERMINE KT/V: KINETIC MODELLING AND ON-LINE MEASUREMENT SYSTEMS

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Introduction and Aims: Kt/V is accepted as dialysis dose applied to a patient during a dialysis session. It is also used as a parameter for the dialysis quality and in some countries the reimbursement is connected to the height of Kt/V. This requires an exact and improvable determination of Kt/V.
The aim of the study is to compare the precision of Kt/V-determination of an improved kinetic modelling program (quantiplan), an online clearance measurement system (OCM) and a photometric Kt/V-determination system (Adimea).
Methods: Quantiplan calculates Kt/V with a 2-pool modelling. It was selected because of its plausibility control for Kt/V and its calculation of the amount of urea removal in dialysis.
The precision of the Kinetic Modelling calculations can be checked by comparing the calculated urea removal and the measured urea removal in dialysate. Urea was measured in the dialysate of a closed system (Genius), which collects dialysate in a vessel.
The plausibility control calculates the maximum of Kt/V, which can be reached respecting the boundary conditions (blood- and dialysate-flow, dialyser-clearance and the patients hematocrit). Kt/V-values which overstep these maximum Kt/V were sorted out.
Measurements using all 3 systems in parallel were technically impossible. Thus two groups of patients had to be investigated. In the first group (30 patients) quantiplan results were compared with OCM values and in the second group (58 patients) with Adimea values in parallel measurements.
Adimea measures the change of light intensity when the light passes dialysate. The kinetic of this intensity change correlates with the kinetic of urea removal. Based on this correlation Kt/V is calculated.
In a defined interval, OCM measures the difference of sodium concentration at the dialysate entry and exit of the dialyser after a determined sodium bolus administration. Based on this difference, OCM calculates the urea clearance of the dialyser.
The result of Adimea is Kt/V, the result of OCM is the urea dialyser clearance. Therefore, using OCM, the Kt/V has to be calculated with a tool after entering the data for V and t.
Results: The measured and calculated urea removal by quantiplan correlates with a coefficient of r=0,95. This allows to use quantiplan as reference in the comparison of the Kt/V-determinations.
In the first group a correlation was found r=0,71 where as in the second group the coefficient for correlation was 0,90. The Kt/V values of quantiplan and Adimea were not significant different (p=0,77) where as the Kt/V values of quantiplan and OCM were significant different (p=0,0002). The Kt/V values determined with OCM were always lower than the Kt/V values determined by Adimea and quantiplan. The difference was in the range of 20% (21%).
Conclusions: The Kt/V determination of quantiplan and Adimea are very close together. The correctness of these determinations is proven by a good correlation between the measured and calculated amount of urea removal with quantiplan (r=0,95).
Despite the methodical difference between quantiplan and Adimea, Adimea is connected to a machine, but quantiplan can be used independently.

Su443 LONGTERM DAILY OZONIZATION (DE-NOVO) OF AN OLDER STANDARD PVC WATER LOOP REDUCED SIGNIFICANTLY PERMEATE ENDOTOXIN LEVELS AND BIOFILM CONTAMINATION AND ACHIEVED FAVORABLE NUTRITION&INFLAMMATION MARKERS IN PREVALENT HD-PATIENTS

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Introduction and Aims: For improvements in longterm oucome of patients on maintenance hemodialysis (HD) it requires optimal permeate water quality to avoid inflammation, infections and malnutrition. However, frequent (≤weekly) prophylactic water loop disinfection is often limited by costs and unfeasable heat approaches (>80°C) for most current standard PVC water loops. Thus, cold high-level disinfection with ozone may be beneficial but longterm microbiological and clinical efficacy is unknown.
Methods: During 3-months (=Mo) run-in and 33-Mo ozone study period we tested 1-2x/Mo permeate (loop start + end) for colony forming units (=CFU) of bacteria (1 mL: water bacteria on R2A + mycobacteria on Middlebrook, pseudomonas on Cetrimid + gram-neg. bacteria on MacConkey agars) and endotoxin levels (sensitive limulus assay: ≥0.005 EU/ml). (De-novo) central PVC water loop ozonization was performed initialy weekly (9 Mo) and then daily (24 Mo) by dissolved low-dose ozone levels of 20-50 ppb (0-3 pm) in an 8-year old permeate PVC loop usually in line with current EU limits (CFU< 100/mL: R2A agar, 7 days at 23°C). In addition, we analyzed ozone (daily) impact on biofilm contamination in new installed standard dialysis inflow-stubs after use for 3-5 Mo and routinely (each quarter) measured serum markers for malnutrition and inflammation (albumin=ALB – subnormal range: <35 g/L, CRP -normal range: <5mg/dl) in 53 prevalent HD patients persistently treated in the studied (out-patient) dialysis unit (all on high-flux HD, 24-Mo mortality rate: 9%; 6 of 53).
Results: Before ozone median permeate endotoxin level was 0.03 EU/mL (R=Range: 0.01-0.31) and we detected in 12 probes 8x water bacteria (median: 16/mL, range: 0-330), 4x mycobacteria, 4x pseudomonas, 5x gram-neg. bacteria. Final median endotoxin levels (0.005 EU/ml, <0.005 – 0.069; p<0.001) and water bacteria CFU rates (4/mL, 0-12; p<0.02) declined significantly and all other pathogens were no longer detectable after start of ozone. Daily central loop ozonization reduced significantly biofilm CFU load in standard dialysis inflow-stubs after 3-5 Mo (initial: 1.2×10^4 per cm^2 vs. final: 2.4×10^2 per cm^2; p<0.01) and we observed persistently favorable levels of ALB and CRP (ALB: initial: 41.7 g/L, 33-49; final: 41.5 g/L; 28-48 and CRP: initial: 0.6 mg/dl, 0.1-3.6; final: 0.9 mg/dl, 0.1-4.2) and a mortality rate of 9% in the prevalent patients.
Conclusions: 1. Long-term frequent ozone reduces significantly permeate endotoxin levels, water bacteria (<10% of current EU limits) and usually

high biofilm contamination in new dialysis inflow-stubes after use for 3-5 Mo. 2. Stable favorable ALB and CRP levels and a rather low mortality suggest clinical benefit. 3. Frequent cold disinfection with (low-dose) ozone appears to be a reliable and cost-effective option for older PVC water loops where heat-based approaches are unfeasable.

Su444 CLINICAL EVALUATION OF POLYETHERSULFONE HIGHFLUX HEMODIALYSIS MEMBRANE COMPARED TO OTHER MEMBRANES

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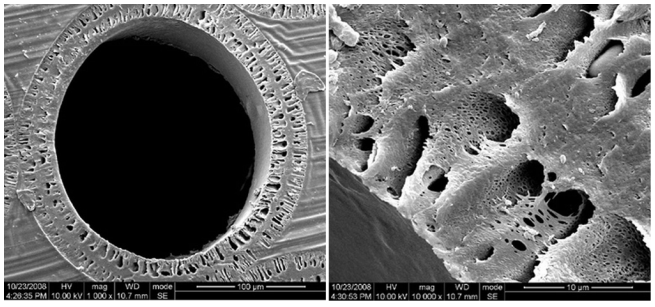
Introduction and Aims: Clinical evaluation of polyethersulfone(PES) highflux hemodialysis membrane in compare with polysulfone(PSF) and polyamide(PA) membranes.
Methods: There were three groups uremic patients who had received hemodialysis at least three months. One group received hemodialysis for 4 hours with PES highflux dialyzers, the second group with PSF highflux dialyzers and the third with PA highflux dialyzers.To evaluate and calculate the solute clearance, blood compatibility and observe the hollow membrane structure with scanning electron microscopy(SEM).
Results: The pictures show the SEM images of PES hollow fiber membrane adsorbing proteins from plasma when they contacted with blood, and this phenomenon was so-called 'membrane fouling' which can induce flux decrease. However, the PSF and PA membranes adsorbed much more and observed more 'membrane fouling'. After the hemodialysis, the clearance and the reduction ratio of small molecules (urea, creatinine, phosphate) were similar to PES, PSF and PA($P>0.05$). However the remove and reduction ratio of middle molecules b_2 microglobulin were higher in PES than that in PSF and PA. The reduction ratio of b_2 microglobulin by using PES was 29.52±13.32 mg/L, 28.71±11.97 mg/L of PSF and 27.43±12.97 of PA,but there was no *statistical significance* among three groups ($P>0.05$). There was no *statistical significance* in the Hb and album level which reflected the safety of the dialyzers of the three groups ($P>0.05$).



Conclusions: The PES hollow fiber membrane hemodialyzer was effective and safe in the therapy for uremic patients. The data indicated that the performance of PES, PSF and PA hemodialyzers in the clinical setting were comparable.May be the PES hemodialyzer was better than the others. The results indicated that PES hollow fiber membrane had a potential widely use for hemodialysis.

Su445 ASSESSING BODY COMPOSITION IN HEMODIALYSIS PATIENTS WITH A MULTIFREQUENCY BIO-IMPEDANCE DEVICE: A MULTICENTRIC EVALUATION OF REPRODUCIBILITY

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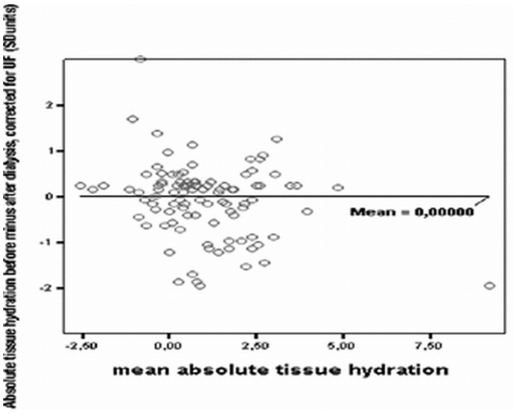
Introduction and Aims: Evaluation of hydration status is important to assure adequate dialysis. MultiFrequency Bio-Impedance (MFBI) has been tested against golden standards, and single point BFMI derived parameters are associated with mortality. However, data on reproducibility of measurements in routine everyday clinical practice are lacking.
Methods: All HD patients from 4 different HD centres were included, unless they had exclusion criteria for BFMI. Measurements were done with the BCM® device (Fresenius Medical Care, Bad Hombourg, Germany) on a regular midweek dialysis day, once immediately before and once at least 30 minutes after the HD session by the attending dialysis nurses. We assessed reproducibility with different calculations. First, lean tissue mass (LTM) was compared pre and post LTM using Bland Altman analysis (BA). Second, BA was used to compare 1) change in total body water (TBW); 2) change in absolute tissue hydration (ATH); 3) change in body weight (BW), all corrected for ultrafiltration (UF) during dialysis.
Results: In total, 104 patients (58 males) aged 67.8±14.0 years, with a pre-dialysis TBW, intracellular water (ICW), extracellular water (ECW) of 32.7±7.4, 16.2±4.2, 16.7±4.2 l and a LTM 30.9±11.0kg were included. Absolute TH was 1.8±1.6 pre and 0.1±1.8 post dialysis respectively, after an UF of 2.0±1.1l. LTM was 30.9±11.0 pre and 30.2±10.6kg post dialysis (p=NS, Spearman correlation 0.96).



BA analysis demonstrated a high level of agreement. Also for TBW, ATH and BW before and after dialysis, corrected for UF, there was a high level of agreement on BA (in declining order).
Conclusions: Evaluation of body composition as assessed by the BCM® device appears to deliver reproducible results, even when performed in routine daily practice in a multicentric trial. BCM can as such be a useful tool to detect changes in body composition and hydration status over time, and thus help to maintain ideal weight.
Disclosure: Dr Van Biesen has received travel grants and speakers fees from Fresenius Medical, Baxter and Gambro on different occasions. The BCM device was provided by FMC Belgium.

Su446 IMPACT OF ULTRAFILTRATION THERAPY ON HYPONATREMIA IN PATIENTS WITH HEART FAILURE

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Introduction and Aims: Hyponatremia is frequently encountered in patients with heart failure (HF), and its association with adverse outcomes is well-established in this population. Current therapeutic options for HF-associated hyponatremia are limited, and ultrafiltration has been suggested as a therapy in this setting. With the advent of new ultrafiltration devices for management of treatment of HF, it is increasingly used in these patients. We evaluated the impact of ultrafiltration therapy on hyponatremia in this setting.
Methods: All ambulatory patients with refractory HF who were referred to the "aquapheresis clinic" (outpatient chronic extracorporeal ultrafiltration therapy for diuretic-resistant HF) were included. Patients with less than three sessions of ultrafiltration therapy were excluded. For each session of ultrafiltration, relevant clinical data including weight, blood pressure, and ultrafiltration volume were recorded. Laboratory data prior to initiation of therapy and at one week were recorded and compared.

Results: Between March 2006 and December 2008, 16 patients (11 men and 5 women) underwent a total of 279 sessions of ultrafiltration therapy (a mean of 18 sessions per patient). The mean ejection fraction was 38%, with a baseline estimated GFR of 52.2 ml/min. The mean baseline serum sodium level was 137.4 mEq/l. Four patients (25%) presented with hyponatremia (defined as serum sodium level of <136 mEq/l) with a mean serum sodium concentration of 134.2 mEq/l. After one week of ultrafiltration therapy, there was no significant change in the serum sodium level of the overall population (137.1 mEq/l) or the hyponatremic subgroup (135.2 mEq/l). No significant change in other electrolytes was observed.
Conclusions: Isolated ultrafiltration therapy does not correct serum sodium levels in patients with HF-associated hyponatremia and cannot be recommended as a therapeutic option in this setting. This is consistent with its mechanism of action based on extraction of isotonic ultrafiltrate from serum.

Su447 DOES INDIVIDUALIZED SODIUM CONCENTRATION FIT ALL HEMODIALYSIS PATIENTS?

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Introduction and Aims: Most dialysis centers usually adopt a single default dialysate sodium prescription. The patients on chronic maintenance hemodialysis (HD) had relatively constant pre-HD serum sodium levels and tended to maintain their own set-points, but the levels varied considerably from patient to patient. Therefore, a single dialysate sodium prescription may not fit all patients. It was recently reported that individualization of dialysate sodium concentration based on pre-HD serum sodium levels decreased thirst, interdialytic weight gain, and blood pressure in HD patients. However, the design of previous study was to decrease dialysate sodium concentration in the non-hypotension prone HD patients. In the present study, we classified patients into three groups according to pre-HD serum sodium levels in relation to the standard dialysate sodium concentration and investigated the clinical impact of an individualized dialysate sodium prescription in each group.
Methods: Twenty-six stable chronic maintenance HD patients were enrolled in this study. During the standard period (3 weeks), all the subjects underwent nine consecutive HD sessions with the dialysate sodium concentration of 136 (10 pateints) or 138 mEq/L (16 patients). During the individualized period (3 weeks), the patients underwent nine HD sessions with the individualized dialysate sodium concentration set to match the patient's average midweek pre-HD serum sodium concentration during the standard period. The patients were divided into three groups: average midweek pre-HD serum sodium concentration was lower than (group 1, n=7), equal to (group 2, n=6), and higher than (group 3, n=13) the standard dialysate sodium concentration. The dialysis prescription, dry weight, diet and hypertensive medications were not modified throughout the period of study. Interdialytic weight gain, thirst score, hypotensive episodes, and pre-HD blood pressure were compared.
Results: Pre-HD serum sodium concentration did not change during the entire study period in all groups. In group 1, pre-HD diastolic blood pressures decreased (70.811.0 vs. 66.98.4 mmHg; P<0.05) and intradialytic hypotensive episodes and nursing interventions increased significantly (p<0.05) after dialysate sodium individualization. In group 2, no significant differences were observed in all parameters studied. In group 3, interdialytic weight gain increased significantly during the individualized period (2.0 0.5 kg vs. 2.3 0.4 kg; P<0.05). Thirst scores in 7 patients whose dialysate sodium was increased by 4 mEq/L increased (6.4 1.5 vs. 7.6 1.5; p<0.05).
Conclusions: Individualization of dialysate sodium concentration based on patient's serum sodium has little benefit for the chronic stable HD patients with their baseline dialysate sodium concentration of 136-138 mEq/L. An individualized dialysate sodium prescription does not fit all HD patients ant it may be considered only in the selected patients.

Su448 BIOLOGICAL VARIATION OF β_2-MICROGLOBULIN IN HAEMODIAFILTRATION PATIENTS

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Introduction and Aims: Critical evaluation of the significance of changes in results on serial analysis can be performed only by consideration of its biological and analytical variation in different settings. Knowledge of these are prerequisites for the introduction of any new tests, as is the case actually with the use of β_2-microglobulin (β_2m) as marker for middle molecules in convective therapies. Biological variation (BV) is calculated as CV_I (within subject variation) and CV_A (analytical imprecision). We study the BV of β_2m, urea and C-reactive protein (CRP) in uremic patients treated with high efficiency postdilutional haemodiafiltration (HDF). CV_I in general population: urea 12,3%, β_2m 5.9%, CRP 42,2%.
Methods: 16 patients with HDF treatment, 8M/8F. Mean age 52,7 years and 42,4 months of prevalence. BV analysis of midweek preHDF samples, each 4 weeks, of urea and β_2m (217 samples) and CRP (173 samples) during a 12 months period. β_2m MEIA Abbott. BV study by Fraser-Harris method. Extreme measures excluded by Reed criteria: difference between extreme mesure and next lower is >1/3 of the range of values. Systematic error (SE): (mean-value of control) × 100/value of control. Total error (TE): SE+CV_A. CV_A and CV_I = Standard deviation (DS) * 100/mean of individual means. Analytical imprecision must be half the within subject variability ($CV_A < 0.50CV_I$) (desirable imprecision), being the optimal imprecision CV_A <0,25 CV_I. Statistic test ANOVA.
Results: Means±SD and ranges (R). Urea 106,30±26,85 (R 52-196) mg/dl. β_2m 19,21±6,08 (R 8,1-35.9) mg/L. PCR 9,7±8,7 (R2,9-56,4)mg/L. Median β_2m 19,59 mg/L. SE urea 0,71%, SE β_2m 2,66%, SE PCR 10,9%. CV_A urea 1,9%, CV_A β_2m 6,57%, CV_A PCR 12,3%. TE urea 2,61%, TE β_2m 8,81%, TE PCR 13,2%. CV_I urea 25.14%, CV_I β_2m 31,35%, CV_I PCR 89,42%. CV_Aurea= 0,13CV_Iurea, CV_A β_{2m}=0,20CV_I β_{2m}, CV_APCR=0,13CV_IPCR. Without significant differences between urea and β_2m within subject variations (ANOVA).
Conclusions: The analytical procedures of β_2m in patients treated with haemodiafiltration show a biological variation (within subject variation of 31%) without significant differences with the biological variation of urea (within subject variation 25%).

Su449 CLINICAL PRACTICE OF LIPID APHERESIS FOR ISOLATED LP(a)-HYPERLIPOPROTEINEMIA WITH PROGRESSIVE ATHEROSCLEROTIC DISEASE IN GERMANY

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Introduction and Aims: Lipid apheresis (LA) for isolated Lp(a)-hyperlipoproteinemia (iLp(a)-HLP) with progressive atherosclerotic disease has been approved for regular reimbursement in Germany by the federal joint committee (g-BA) in 2008. G-BA recognized that for high-risk patients with life-threatening disease LA must be considered as the only therapeutic option**. However, due to shortage of clinical data g-BA in parallel demanded to prove efficacy by conducting a prospective controlled clinical trial. Aim of this working group has been to document in a complementary fashion patients treated since 2008 being not enrolled in the controlled clinical trial.
Methods: LA had been approved for included patients by committees of regional physicians councils: (1) progressive atherosclerotic disease clinically or by imaging techniques, (2) standard care of all co-existing cardiovascular risk factors failed to prevent progression, (3) Lp(a) > 60 mg/dl, LDL-cholesterol within normal range. After informed consent patients entered a retrospective-prospective documentation of their clinical course including quality of life (SF36), starting 2 years before commencing, and continuing annually during chronic LA.

Results: The first 88 consecutive patients with iLp(a)-HLP exhibited the following baseline characteristics before commencing LA (mean values): age 56 yrs, Lp(a) 109 mg/dl, LDL-C 101 mg/dl. In 92% of patients coronary artery disease was the major manifestation. Mean annual cardiovascular event rate was 0.52 during the 2 yrs before LA (0.59 in yr in -1, 0.44 in yr -2). Accounting progression documented by imaging techniques as equal event the mean rate was 0.75 (0.91 in yr -1, 0.58 in yr -2).
Conclusions: In cooperation of apheresis centers and committees of regional physicians councils critical approval of patients with iLp(a)-HLP and progressive vascular disease for LA has been established. Continuing this documentation and determination of event rates during chronic LA will provide results on efficacy of LA for iLp(a)-HLP and allow to better define this patient population.
*The working group comprises Martin Haesner, Martin Tepel, Berlin; Gerhard Riechers, Braunschweig; Wilfried Dschietzig, Claudia Ernst, Renate Jacob, Cottbus; Harald Kaul, Deggendorf; Ulrich Julius, Sergey Tselmin, Dresden; Beate Jaeger, Ralf Spitthoever, Johan Knee, Essen; Paul Breitenberger, Germering; Michael Koziolek, Gerhard-Anton Mueller, Goettingen; Eberhard Roeseler, Sabine Wehner, Hannover; Franz Heigl, Ines Schulz-Merkel, Kempten; Christina Saehn, Krefeld; Johannes Bunia, Iserlohn; Michael Wintergalen, Olpe; Josef Leebmann, Passau; Carsten Schuerfeld, Saarlouis; Albrecht Wagner, Trier.
** Jaeger et al. Nat Clin Pract Card Med 2009; 6: 229-39.
Disclosure: Reinhard Klingel received research grants from Asahi Kaesi Kuraray Medical, Japan and Diamed, Germany.

Su450 THE HEPARIN-COATED AN69 ST DIALYSIS FILTER DOES NOT REDUCE CLOTTING DURING HEMODIALYSIS WHEN COMPARED TO A CONVENTIONAL POLYSULFONE FILTER

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Introduction and Aims: We investigated whether the heparin-coated AN69 ST hemodialysis (HD) filter induced less clotting than a conventional polysulfone filter (Fx8).
Methods: Detailed analyses of clotting were scheduled in six subsequent dialysis sessions in 11 patients with alternate AN69 ST and Fx8 filters. All received priming with unfractionated heparin (UFH) and free UFH was removed by saline flushing. To obtain some degree of clotting half the normal dalteparin dose was given. Clotting was evaluated in the venous air trap each hour and graded on a 6-point scale. Clotting was also assessed by repeated measures of the coagulation marker prothrombin fragment 1+2 (PF1+2). The platelet activation marker β-thromboglobulin (β-TG) and anti-FXa activity were also repeatedly measured.
Results: Forty-five HD sessions in 10 patients were evaluated in statistical analyses; 23 with the AN69 ST filter and 22 with the Fx8 filter. One patient treated with enalapril had two repeated adverse reactions to the AN69 ST filter and was excluded from the analyses.

Effect of the AN69 ST filter and dalteparin-dose/kg on mean clot score in the air trap

	B	SE	p
AN69 ST filter	-0.047	0.189	0.8
Dalteparin dose, IU/kg	-0.015	0.014	0.29

B = linear regression coefficient, SE = standard error. The effect of the AN69 ST filter (versus the Fx8 filter) and dalteparin-dose/kg body weight on mean clot score in the air trap estimated by a linear regression model with repeated measurements.

Effect of the AN69 ST filter, anti-FXa activity and time on PF1+2

	B	SE	p
AN69 ST filter	0.177	0.084	0.038
anti-FXa activity, IE/mL	-2.455	0.683	0.0006
4 hours of HD (versus 3 hours of HD)	0.421	0.101	<0.0001

B = linear regression coefficient, SE = standard error. The effect of the AN69 ST filter (versus the Fx8 filter), anti-FXa activity and time on PF1+2 (logtransformed) estimated by a linear regression model with repeated measurements.

Multiple linear regression analyses with repeated measurements showed that use of AN69 ST adjusted for dalteparin dose/kg did not decrease mean clot in the bubble trap; estimate (B)=-0.047, p=0.8. AN69 ST increased the clotting marker PF1+2; B=0.177, p=0.04, when adjusted for anti-FXa activity and hours of HD. Compared to the Fx8 filter the AN69 ST filter significantly decreased β-TG during HD adjusted for anti-FXa activity and hours of HD; B=-0.974, p<0.001.
Conclusions: The heparin-coated AN69 ST filter did not induce less clotting in the extracorporeal circuit than the Fx8 filter. One patient treated with enalapril had a hypersensitivity reaction to the heparin-coated AN69 ST filter.
Disclosure: Research grants were received from Vingmed AS, Norway, who also granted the Fx8 dialysers. The AN69 ST dialysers were granted from Gambro, Norway.

Su451 LONG-TERM LIPID APHERESIS IN PATIENTS WITH SEVERE HYPERTRIGLYCERIDAEMIA AND PROGRESSIVE ATHEROSCLEROSIS

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Introduction and Aims: Hypertriglyceridaemia (HTG) is discussed to be an independent risk factor for atherosclerosis. Dietary management combined with lipid-lowering drugs and appropriate treatment of diabetes mellitus are the standard for severe HTG. Extracorporeal elimination of lipoproteins and triglycerides (TG) has been found to be effective mainly for the treatment of acute HTG-induced pancreatitis, a potentially life threatening complication. For drug and diet-refractory HTG-patients with severe progressive atherosclerosis at increased risk of coronary and/or peripheral complications, chronic lipid apheresis may be indicated to decrease excessively elevated serum triglycerides. In these rare cases, TG may be the pathophysiologically relevant risk factor for progression of atherosclerosis. Lipid apheresis, commonly used for the treatment of familial hypercholesterolemia, provides a more selective elimination of atherogenic lipoproteins than plasma exchange with no need for plasma substitution with the related risk of allergic reactions. Aim of this retrospective investigation was to evaluate efficacy and safety of lipid apheresis in high-risk patients with severe HTG.
Methods: We report on three patients (age 41, 55 and 69 years, 1 woman, 2 men) with severe progressive atherosclerosis involving coronary and peripheral arteries. Two of them had diabetes mellitus type 2, none had a history of prancreatitis. All patients had severe combined hyperlipidaemia with HTG. Appropriate diet and drug therapy, including antidiabetic drugs, were insufficient to prevent progression of atherosclerosis. The patients received chronic outpatient lipid-apheresis once to twice a week with lipidfiltration (double filtration plasmapheresis) using the Octo Nova system (SW version 4.30.2/5.10.6). Plasma was separated from cellular blood components with a polyethylene plasma separator and warmed by the heating system Octo Therm before filtration to optimize sieving characteristics.
Results: Mean concentration of TG before the first lipid-apheresis was 3287 mg/dl (s=952), total cholesterol level was in mean 409 mg/dl (s=90). Patients received long-term chronic lipidfiltration with 1-2 treatments per week over 11 months to 4 years. Extracorporeal elimination of lipoproteins was useful in rapidly lowering elevated serum TG. The mean reduction rate of TG during lipidfiltration was 58% (s=9.1), of total cholesterol 48% (s=12). Lipidfiltration was safe and well-tolerated. All three patients had no coronary event since initiation of lipid-apheresis. The progression of peripheral occlusive disease could be prevented, in one case amputation had been already considered.
Conclusions: Long-term outpatient lipid apheresis was safe and effective to correct the atherogenic lipid profile and resulted in stabilization of patients with severe HTG at risk for acute cardiovascular events and progression of arterial occlusive disease who had not responded to previous therapies and whose symptoms were refractory to conventional dietary and drug therapy.

Su452 HIGHER HD/HDF EFFICACY WITH CITRIC ACID ACIDIFIED-DIALYSATE- WHY?

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Introduction and Aims: Higher efficiency has been claimed in bicarbonate haemodialysis (HD, BHD) with citric acid (CA)-acidified dialysate as compared to conventional BHD with acetic acid (AA)-containing solution but the reason for this has not been properly elucidated so far. This work investigated impact of the acidifying agent on parameters, which might explain this – coagulation status, dialyser fibre bundle volume (FBV) behaviour and transient leukopenia during the first 20 minutes into HD as a marker of acute phase reactions.
Methods: Kt/V was evaluated in a group of patients (pats) on HD and on-line HDF during 1 week-interval (3 procedures) with CA- and AA-containing dialysate. Heparin elimination constant (K_e) was calculated from exponential decrease of Activated Clottting Time (ACT, $ACT(t) \sim ACT(0)*\exp(-t/K_e)$) during all evaluated procedures to assess differences in coagulation status. FBV was measured in 3 pats on on-line HDF during 3 procedures with CA- and 3 procedures with AA-containing solution at 45 minutes and towards the end to see whether increased efficacy can be attributed to lowered clotting tendency and thus better preservation of functional membrane surface area over the entire procedure. To assess possible differences in blood-membrane interaction during early stage of BHD procedure, paired measurement of leukocyte count drop in 20th minute was performed in 11 patients dialysed once with AA- and once with CA-containing dialysate.
Results: Increased efficiency was found both in BHD and HDF with CA-containing dialysate as compared to procedures with conventional AA-based solutions (on average, 7% increase in Kt/V). The heparin elimination constant K_e was on average by 10% smaller (i.e. indicating lowered clotting tendency) in HD with CA-containing dialysate. Surprising results were obtained from FBV measurements: The FBV values were consistently lowered in all proce-dures with AA-containing dialysate at the first measurement, suggesting transient plaguing of the membrane, while there was no difference towards the end of the procedure regardless of the acidifier used. In correspondence with this finding, higher leukocyte count drop was seen with AA-containing dialysate (on average 26,6±12,9% against 21,6±10,5% with CA-containing dialysate), suggesting more pronounced transient membrane plaguing and thus lower efficacy during the first part of dialysis.
Conclusions: Although prolonged ACT times were found both in BHD and HDF with CA-containing dialysate, increased dialysis efficacy does not appear to be caused (mainly or solely) by less coagulation in the dialyser with CA-containing dialysate because no differences were seen in FBV at the end of dialysis. Lower FBV during the first hour as well as higher leukocyte count drop with AA-containing dialysate rather suggest that higher efficacy of procedures with CA-containing dialysate is achieved mainly during the first hour because of less transient plaguing of the membrane.

Su453 HIGH SURVIVAL RATE AND COMPREHENSIVE REHABILITATION IN AN OPTIMAL DIALYSIS SETTING

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Introduction and Aims: Dialysis technology, quality and outcomes have been under continuous pursuit of excellence. We report clinical data from an independently owned and operated satellite dialysis center in which several different approaches toward an ideal hemodialysis treatment have been successfully addressed in the last 5 years.
Methods: Thirty-two consecutive unselected incident hemodialysis patients (*17 males, 15 females, mean age 57.9±21,0 [10-93] yrs, 72% arteriovenous fistula rate at dialysis initiation, 34.3% prevalent diabetes mellitus*), all originated from our multidisciplinary pre-dialysis care, were followed for a mean time of 28.1±12.6 (3-48) months in an *exclusive* short daily in-center hemodialysis program (*1.5-2.5h, 6 to 7 times a week, 300 ml/min blood flow, 700 ml/min dialysate flow*), that includes round-trip patient transportation, 24-stations comfortable environment, 5-treatment shifts options per day, ultrapure dialysate (*heat disinfection water treatment, PEX looping and individual ultrafilter*), single-use high-flux dialyzer, online blood pressure/clearance monitoring, exercise program and high staff/patient ratio (*1 present nephrologist, 1 dietitian, 1 psychologist, 2 nurses and 4 technicians for each group of 12 patients*). Data are expressed either as mean ± SD or percentage.
Results: Analysis from the most recent data shows 78.5% employment rate of working-age patients, 2.1% missing sessions, 4.3 days/patient-year hospitalization rate, 21.8% on anti-hypertensive medications, UF 1164±642 ml/session, URR 0.50±0.07, ESA requirements 5840±4394 UI/week, hemoglobin 11.9±1.8 g/dl, calcium 9.2±1,0 mg/dl, phosphate 4.7±1.4 mg/dl, PTH 205±210 pg/ml, albumin 4.0±0.6 g/dl, β2-microglobulin 19.7±8.4 mg/l and CRP 7.7±6.4 mg/l. Six patients underwent successful renal transplantation and one 74-year-old patient has died of lung cancer after 38 months on dialysis.
Conclusions: Full rehabilitation and high life expectancy of our patients undergoing chronic hemodialysis was achieved/accompannied by a comprehensive pre-dialysis care, convenient transportation and flexible dialysis schedule, quotidian solute and volume removal, all leading to satisfactory clinical, psycossocial, nutritional, inflammatory and mineral metabolism markers.

Su454 EVALUATION OF ITCHING OF THE PATIENTS WITH SEVERE PRURITUS SWITCHED FROM HIGHLY PERMEABLE POLYSULFONE DIALYZER TO POLYMETHYLMETHACRYLATE DIALYZER WITH A ONE-YEAR FOLLOW-UP

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Introduction and Aims: Patients undergoing hemodialysis suffer from uremic pruritus, a frequent and tormenting problem that interferes with their quality of life. The pathophysiological mechanisms of pruritus are largely unknown. Some studies have shown that a increase in the dialysis dose improves uremic pruritus. However, even under a high dose dialysis, pruritus among hemodialysis patients is still prevalent. In the present study, the patients with uremic pruritus undergoing high dose hemodialysis with polysulfone (PS) dialyzer switched to polymethylmethacrylate (PMMA) dialyzer. After the switch, the strength and duration of itching of the patients were evaluated.
Methods: Eleven stable patients with severe pruritus undergoing high dose hemodialysis using a highly permeable PS dialyzer (APS-SA, Asahi Kasei Kuraray Medical, Japan) for over 1 year were selected and were subsequently switched to a PMMA dialyzer (BG-PQ, Toray Medical, Japan). The strength and duration of itching were evaluated by using a visual analogue scale (VAS) at 1, 2, 3, 6, 9, 12 months after the switch to PMMA. Pre-dialysis values of urea, β2-microglobulin, albumin, total protein were also evaluated. Adsorbed proteins eluted by 40% acetic acid were also evaluated by SDS-PAGE.
Results: The self-assessed VAS itching strength scores significantly decreased from 48 (before the switch) to 35 after 1 month ($p<0.05$), 32 after 2 months ($p<0.01$), 21 after 3 months ($p<0.001$), 18 after 6 months, 9 after 9 months and 4 after 12 months. The itching duration scores also significantly decreased from 32 (before the switch) to 12 after 6 months ($p<0.05$), 7 after 9 months ($p<0.05$), and 4 after 12 months ($p<0.05$). After the switch, pre-dialysis values of urea did not change but that of β2-microglobulin increased whereas those of albumin and total protein decreased. The removal of β2-microglobulin by PMMA dialyzer was inferior to that by PS dialyzer, while the removal by PMMA dialyzer of larger molecules which molecular weight is near albumin was superior to that by PS dialyzer. PMMA membrane adsorbed larger amount of proteins which molecular weight ranged 10-90 kDa.
Conclusions: The strength and duration of itching of the patients significantly decreased when the patients were switched from highly permeable

PS dialyzer to PMMA dialyzer. PMMA dialyzer can efficiently remove the larger molecules which are provably associated with uremic pruritus.

Su455 ESTIMATION OF β2-MICROGLOBULIN DIALYZER CLEARANCES USING A TWO-COMPARTMENT MODEL FOR CONVENTIONAL AND SHORT HEMODIALYSIS (HD) TREATMENTS

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Introduction and Aims: The European Renal Best Practice Advisory Board recently recommended that all HD patients be treated by high-flux dialyzers. This decision was based primarily on the results of the Membrane Permeability Outcome study that showed reduced accumulation of serum β2-microglobulin (β2M) levels in patients treated by high-flux as opposed to low-flux dialyzers (Locatelli et al, 2009). The growing role for β2M as a middle molecule marker to assess HD adequacy suggests improved methods to quantify its removal are needed.
Methods: We compared β2M dialyzer clearances during conventional (241±27 min) and short (116±14 min) HD treatments using high flux dialyzers in a crossover study on 22 maintenance HD patients (16 male, 6 female; 61±18 (SD) years of age). Two patients in each treatment modality were excluded from these analyses either because of missing or erroneous data. Intradialytic blood samples were obtained at regular time intervals, and the pre- and post-dialysis blood samples were input in the new parameter estimation method based on a two-compartment, variable-volume model (Ward et al, 2006). Assuming fixed rates of β2M generation and inter-compartmental transport, dialyzer clearances were estimated and compared with clearances obtained from a single-compartment model (Leypoldt et al, 1997) and the standard (cross-dialyzer) plasma clearance equation.
Results: Comparing the single- and two-compartment models, β2M clearances estimated using the two-compartment model were closer to those calculated using the standard plasma clearance equation (38±21and 42±23 vs. 32±17 ml/min in conventional HD; 37±23 and 51±32 vs. 41±26 ml/min in short HD). This improvement results from the incorporation of an additional compartment to account for the slow transport of β2M from interstitial to plasma compartment. Furthermore, the clearances during conventional and short treatments were comparable with all three methods.

A

Conventional Treatment (4-hr)	Cross-Dialyzer Clearance	Single-Compartment Clearance	Two-Compartment Clearance
Mean	32	42	38
SD	17	23	21
SEM	4	5	5

B

Short Treatment (2-hr)	Cross-Dialyzer Clearance	Single-Compartment Clearance	Two-Compartment Clearance
Mean	41	51	37
SD	26	32	23
SEM	6	7	5

Table 1: Comparison of measured and estimated β2M dialyzer clearances based on pre and post-dialysis blood samples for conventional (top) and short (bottom) HD treatments. SD: standard deviation, SEM: standard error of the mean defined as SD/√(Mean). All dialyzer clearances are in ml/min. N=20.

Conclusions: We conclude that the use of complex, multi-compartmental models improve β2M clearance estimations during both conventional and short HD treatments. For many applications, however the previously defined single-compartment model likely provides sufficient accuracy.
Disclosure: The authors of this abstract are employed by Baxter Healthcare Corporation.

Su456 DETERMINATION OF POTASSIUM (K), CALCIUM (Ca), PHOSPHATE (PO_4) AND MAGNESIUM (Mg) LEVELS IN SERUM AND PLASMA OF UREMIC PATIENTS UNDERGOING HEMODIALYSIS

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Introduction and Aims: It has been referred that at the time of blood clot formation, degranulation of cells may release several ions, leading to falsely elevated serum values. On the other hand, Ca^{++} is an essential cofactor for interactions in the coagulation. The aim of our study was to determine plasma and serum levels of potassium (K), calcium (Ca), phosphate (PO_4) and magnesium (Mg) in uremic patients undergoing hemodialysis (HD).
Methods: We investigated the difference of plasma and serum levels of K, Ca, PO_4, and Mg pre- and post-dialysis in 25 hemodialysis patients without thrombocytosis, leukocytosis and erythrocytosis. Plasma concentrations were measured from sample of 3ml blood collected in a Vacutainer tube containing 170IU of sodium heparine and the serum ones from blood in the same tube without heparine. We performed measurements before and immediately after the session.
Results: The data of this study are summarized in table I.
Table I. Mean value ± SD of the measured ions in serum and plasma of hemodialysed patients and statistical evaluation.

Mean value ± SD of the measured ions in serum and plasma of hemodialysed patients and statistical evaluation

	K+ (mEq/L)	Mg++ (mg/dl)	PO4 (mg/dl)	Ca++ (mg/dl)
Pre-HD				
Serum	5.02±0.68	2.82±0.32	5.17±1.41	9.06±0.69
Plasma	4.83±0.69	2.77±0.23	5.02±1.36	9.39±0.71
P-value	P<0.05	P<0.05	P<0.05	P<0.05
Post-HD:				
Serum	3.19±0.43	2.50±1.84	1.76±0.7	9.05±1.31
Plasma	3.01±0.48	2.53±1.88	1.63±0.7	9.32±1.27
P-value	P<0.05	P=NS	P<0.05	P<0.05

According to our finding, serum concentrations of K and PO_4 were significantly higher than plasma concentrations before and after the HD session. On the contrary, plasma Ca concentration was significantly higher than serum Ca concentration before and after HD session. Serum Mg concentration were higher than plasma before HD without significant changes between serum and plasma post-HD.
Conclusions: According to our results the difference between serum and plasma concentrations of K, Mg, and PO_4 (mainly intracellular anions) and of Ca (mainly extracellular ion) which participates in coagulation, opens the question if the plasma values are more accurate. Probably this observation is more useful in patients with pseudohyperkalemia, where marked elevation of serum potassium levels is not accompanied by clinical evidence. It seems that the serum levels of the above ions are not the proper indicator of their excact concentration in the blood. This observation is more useful in uremic patients with abnormal high blood cells.

Su457 REFINING THE ASSESSMENT OF DIALYSIS DOSE IN CRRT

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Introduction and Aims: To introduce a new expression of dialysis dose that accounts for such factors as pre-dilution, off-dialysis time, and volume changes, that are usually neglected.
Methods: Eighteen patients on CRRT at two different ICUs were studied. The usual dialysis dose expression was computed as total effluent (TE, ml)/ Body wt (BW, kg)/total dialysis time (TDT, h). Urea clearance (K),

assumed to coincide with effluent, was corrected for pre-dilution (when present) and the time-averaged K (TAK) was computed dividing the sum of all K x t products by the whole treatment period (WTP, min), from the start of the first session to the end of the last one, including all off-dialysis times. Volume change (ΔV) from start to end of WTP was computed as the sum of daily fluid balances. The baseline urea volume (V0) was assumed to be 65% of BW (Marshall, Nephrology, 2006), and mean V (Vm) was computed as V0 + ΔV/2. TAK was normalised to Vm and multiplied times a typical V of 40 L.

Results: We studied 83 CRRT sessions in 18 patients (11 Males). The more relevant results (M±SD) were: age 63.7±12 years, BW 83±23 kg, sessions per patient 6±2.3, blood flow rate 139±17 ml/min, dialysate flow rate 1260±386 ml/h, replacement fluid rate 738±626 ml/h, studied period (WTP) 9.4±6.7 days, total effective dialysis time (TDT) 6.8±5.9 days, TDT/WTP 0.75±0.22, mean effluent (TE/TDT) 39.5±8.7 ml/min, TAK 27±10.4 ml/min, usual dose expression (TE/TDT/BW), 29.8±8.0 ml/kg/h; new index (TAK/Vm x 40 L), 20.9±10.3 ml/min. The difference between paired mean effluent and TAK was 12.5±10.5 ml/min (p<.0.000).

Conclusions: The usual dialysis dose expression overestimated the delivered dose (TAK) by 31±24%, essentially because dialysis time was 75±22% of total time. Moreover, pre-dilution reduced K by 15±2%, in 9 patients. The new index, TAK/Vm x 40 L, being a proxy of the equivalent renal clearance (EKRjc), allows an easy comparison with other dialysis modalities. For instance, based on the equation EKRjc=10 Kt/V + 1 (Casino and Lopez, NDT, 1996), one can realise that the observed mean "EKRjc" of 20.9 ml/min should correspond to a Kt/V of about 2.0 on three times per week Haemodialysis.

Su458 BETA 2 MICROGLOBULIN, KAPPA AND LAMBDA LIGHT CHAINS DEPURATION BY HEMODIALYSIS

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Introduction and Aims: Beta 2 microglobulin (β2M) is a recognized surrogate marker of middle-molecule uraemic toxins and a key component in dialysis-associated amyloidosis. Recently, it was associated with the clinical outcome in dialysed patients. Free imunoglobulin light chains (κ, λ) are also markers of middle molecule uremic toxins, less investigated than β2M, but seems to interfere with essential neutrophils functions and to participate in the disturbed immune function of CKD patients.

To compare beta 2 microglobulin, κ and λ light chains depuration by hemodialysis.

Methods: 87 prevalent HD patients [1% diabetics, 59% male; median age 61(53-72) years; median HD vintage 7(4-10) years; 23% HDF; 77% high flux HD; median CRP 6mg/L].

Investigated parameters (pre-, postdialysis) were: urea, β2 microglobulin (β2M), κ and λ light chains (LC), serum albumin; body weight, total body water (TBW), extracellular body water (ECW – measured with BCM, Fresenius Medical Care) and ultrafiltration volume. Solute concentration were normalized to pre- and postdialysis albumin levels.

Solute depuration was quantified either by reduction ratio or quantitatively (no rebound, monocompartment model, total body water in case of urea, and extracellular water, in case of β2M and κ, λ light chains).

Results: Dialysis reduced all investigated parameters levels, but the slope was different in case of λ light chains from κ and β2M. Estimated by reduction ratio, β2M depuration (68.2%) is not different from urea (70.9%), but significantly differs from κ and λ light chains (49.2% and 25.5%). When examined quantitatively, κ and λ light chains are excreted in significantly larger amonunts than β2M (1100 and 803, vs. 212mg/session; $p<0.05$). β2M and urea had similar percentual depuration (68.2% vs. 70.9%), while κ and λ light chains was lesser (49.2% and 25.5%; $p<0.05$). Albumin-normalized pre- and post-dialysis β2M and κ levels were closely correlated (r^2 0.91 and 0.23;$p<0.05$), which was not true in case of λ light chains (r^2 0.001 and 0.08; $p>0.5$), suggesting analogous distibution volumes and similar equilibration constants for β2M and κ, but not for λ. A model including the percentual reduction in body weight and pre-dialysis β2M concentration predicted in 17% of cases the reduction ratio. A significant contribution made the percentual reduction in body weight, suggesting a dominant role for ultrafiltration in the definition of β2M reduction ratio.

Conclusions: Beta 2 microglobulin, κ and λ light chains are effectively depurated by dialysis. While β2M seems to have a similar distribution volume and excretion by dialysis as urea, κ and λ light chains perform differently. A monocompartment model is more fitted to β2 microglobulin than to κ and λ light chains dialysis excretion. More studies are necessary to validate κ and λ light chains as surrogate markers of midlle molecules excretion.

Su459 A VERSATILE MATHEMATICAL MODEL FOR ACCURATE PREDICTION OF INTRADIALYTIC IONIZED CALCIUM CONCENTRATION DURING REGIONAL CITRATE ANTICOAGULATION

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Introduction and Aims: Regional citrate anticoagulation (RCA) in hemodialysis (HD) offers several advantages over heparin anticoagulation, but frequent systemic ionized calcium (iCa) measurements are required for fear of potentially life-threatening iCa derangements. We aimed to develop a versatile model for accurate prediction of systemic iCa levels at any time point during RCA dialysis.

Methods: We previously developed a model of calcium and citrate kinetics during RCA in HD based on physicochemical, biochemical, and physiologic principles [Thijssen et al., Blood Purification, Jan 2010]. It was found that the difference between model-predicted systemic iCa and measured iCa during HD follows a linear relationship with a slope related to levels of parathyroid hormone (PTH). Based on these observations, a novel model ("hybrid model") was developed. The hybrid model takes the contribution of PTH to the dynamics of systemic iCa into account. The hybrid model was validated in 8 HD subjects (17 RCA treatments) using citrate infusion into the arterial line, calcium- and citrate-containing dialysate and no venous calcium substitution. Systemic iCa was measured multiple times throughout each treatment for comparison with model predictions.

Results: Across all intradialytic time points, the average difference between predicted and measured systemic iCa (predicted minus measured) was 0.00196 mmol/L (95% CI, -0.0038 to +0.0077 mmol/L). The minimum and maximum differences were -0.12 and +0.08, respectively. There was no systematic trend in this difference over the course of the treatments (Fig. 1).

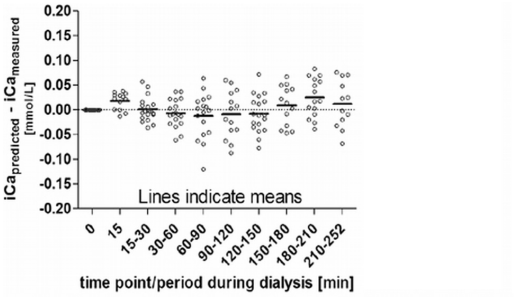

Figure 1. Difference between model-predicted and measured systemic ionized calcium (iCa), plotted against groups of elapsed dialysis time.

Conclusions: The presented hybrid model exhibits excellent agreement between predicted and measured systemic ionized calcium concentration during RCA in HD. While previous iterations of the model showed an end-dialysis prediction error of up to 0.3 mmol/L, the current model predicts systemic iCa within en error margin of only about 0.1 mmol/L. Furthermore, this level of accuracy remains stable throughout the entire HD treatment. Of note, the RCA treatments delivered were by no means uniform in nature, but instead comprised different pre-filter iCa targets, different treatment times, and even citrate infusion rate profiling. This further underlines the robustness of the underlying mathematical model. Accurate prediction of systemic iCa is a key requirement if frequent iCa measurements during RCA dialysis are to be eliminated.

Su460 EVALUATION OF FILTER PATENCY, ACID-BASE AND ELECTROLYTE BALANCE OF A PRISMAFLEX-BASED REGIONAL CITRATE ANTICOAGULATION PROTOCOL FOR CONTINUOUS RENAL REPLACEMENT THERAPY

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Introduction and Aims: Regional citrate anticoagulation has been shown to be an excellent alternative to heparin anticoagulation for patients requiring continuous renal replacement therapy (CRRT). Potential risks are disturbances of acid-base, calcium and magnesium homeostasis. Here, we evaluate a new citrate protocol commercially available by the Gambro Hospal GmbH (Planegg-Martinried, Germany) and involving the actual Prismaflex dialysis device and an isotonic citrate solution (Prismocitrate).
Methods: The Prismaflex citrate protocol was provided by Gambro Hospal GmbH. On start, this protocol was based on a Prisma Flex ST100 set (AN 69® ST membrane) mounted on the actual Prismaflex dialysis device, a blood flow 120 ml/min, 1.8 L/h Prismocitrate (10 mmol/l citrate/2 mmol/l citric acid) in pre-dilution mode and a 0.8 L/h dialysate flow (Prism0Cal). In parallel, separate i.v. infusions of potassium, calcium and magnesium were initiated according to the manufacture protocol. Blood pH, base excess and ionized calcium levels were measured before and at least every 6 h after start of CRRT, magnesium levels before and every 24 h, respectively. Scheduled hemofilter run time was 72 hours.
Results: A series of 25 CCRT treatments in 16 patients were included. Of the 25 hemofilters started, only five reached the scheduled hemofilter run time of 72 h (20%). Twenty treatments stopped prematurely (10 because of filter clotting, 10 because of Prismaflex hardware or software problems). None of the hemofilters had to be stopped because of non-CCRT related reason (recovery of renal function, death of the patient, change to intermitted dialysis, etc.).
In the patient's blood measurement at and after 12 h of CCRT 70.7% of the bicarbonate concentrations were lower than normal range (21-28 mm/l); the base excess calculations were below in 91.9% of the readings (normal range -2–3 mmol/l), respectively. During the course of CCRT treatment, mean bicarbonate decreased from 23 to 20 mmol/l and mean BE from -2 mmol/l to -5 mmol/l, respectively. In 6 treatments, substitution of bicarbonate was necessary. Regarding the electrolytes potentially bound by citrate, we found that 65.3% of ionized calcium measurements were outside the normal range (1.1–1.3 mmol/L) under treatment, while 51.4% of the magnesium concentrations were above the normal range (>1,06 mmol/L).
Conclusions: The present study shows that the evaluated Prismaflex/Prismocitrate-based regional citrate anticoagulation protocol yields not satisfying filter running times. In addition, the control of blood acid base status, calcium and magnesium homeostasis is of poor quality.
Disclosure: T.S. and H.P. have received lecture fees and grant support from Gambro Hospal GmbH and Fresenius Medical Care GmbH, Germany.

Su461 EVALUATION OF EFFECTIVENESS AND SAFETY OF ACETATE FREE DIALYSATE – PRELIMINARY STUDY

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Introduction and Aims: A hemodialysate acidified with citric acid instead of the less physiologic acetic acid has been in use, principally in acute dialysis for several years. The small amount of citrate in dialysate has been suggested to have some anticoagulant properties. The purpose of this study was to evaluate the efficacy and safety of acetate-free hemodialysis(AFHD) in chronic hemodialysis patients.
Methods: This prospective study involved 6 chronic hemodialysis patients. AFHD was performed during 21 hemodialysis procedures using citrate dialysate (2.0mEq/L citrate, ARTSTON, KRD, Korea). During the AFHD, routine heparin anticoagulation was not performed.
Results: All the dialyses with acetate-free dialysate were uneventful, and no unusual events occurred. The average age of the patients was 56.2±11.8 years, the M:F ratio was 3:3 and the average number of the treatment was 3.5±1.5 dialyses/patient. The average number of saline washing during AFHD was 1.4±0.8(0-3). Over 85% of the AFHD were successfully performed with minimal blood clotting in the dialyzer. There was no difference between predialysis BP and postdialysis BP. Postdialysis serum total calcium level was significantly lower than predialysis level and postdialysis ionized calcium level was significantly lower than predialysis level. Postdialysis hypocalcemia always asymptomatic and the patients spontaneously recovered in the interdialytic phase. There were 3 episodes(14.3%) of asymptomatic intradialytic hypotension and 6 episodes(28.6%) of asymptomatic intradialytic hypertension during the study.
Conclusions: In conclusion, acetate-free dialysate is safe and can be used without associated technical and clinical problems. Also citrate acidified dialysate can protect against intradialyzer clotting without use of other anticoagulant.

Su462 RISK OF HYPOGLYCEMIA DURING HEMODIALYSIS IN DIABETIC PATIENTS IS RELATED TO LOWER PRE-DIALYSIS GLYCEMIA

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Introduction and Aims: Asymptomatic hypoglycemia (HG) during hemodialysis (HD) has been reported in chronic renal failure (CRF) patients, either when using a dialysate without glucose or with glucose at 55mg/dL. During a recent trial, we performed a complementary study to verify the relationship between predialysis serum glucose levels and intradialytic HG episodes in diabetic CRF patients.
Methods: We randomly selected 20 diabetic individuals among the 94 CRF patients of our dialysis unit and submitted them to 3 HD sessions with bicarbonate dialysis solutions with different glucose concentrations at a 7-day interval: with a glucose-free (GluZERO), with 55 mg/dl (Glu55), and with 90 mg/dl (Glu90). In each phase, plasma glucose levels were measured at five standardized moments during the session (at 30, 60, 150 and 240 min) and immediately pre-dialysis. We considered <70 mg/dL as hypoglycemia.
Results: Five patients in GluZERO and 3 patients in Glu55 presented 7 and 5 HG episodes respectively, none during Glu 90. Mean intradialytic glycemia was lower than preHD glycemia in each of the 3 phases, this being more pronounced in GluZERO. For GluZERO, mean preHD glycemia was lower in those who presented HG episodes during dialysis than in those who did not (p=0.005; *t* test), while this was just a tendeny in Glu55 (p=0.082; *t* test), probably due to the small sample.
In those with preHD glycemia under 140 mg/dL, intradialytic glycemia levels were significantly lower only in phase GluZERO(p=0.0015; *t* test).

Table 2. PreHD and intradialytic glycemias (mean ± SD, mg/dL) inl patients with preHD glycemia <140 mg/dL in each phase (*t test)

	Glu ZERO	Glu 55	Glu 90
Patients (n)	5	6	5
PreHD glycemia (n)	122.4±14.1 (5)	107.0±18.3 (6)	114.4±16.1 (5)
Intradialytic glycemia (n)	79.4±12.3 (20)	103.7±44.4 (24)	127.2±49.4 (20)
*p**	0.0015	0.7816	0.343

Abstract Su462 – Table 1. Glycemias (preHD and intradialytic (mean ± SD,mg/dL) in those with and without HG during each phase

	Glu ZERO			Glu 55			Glu 90
Patients	All (n=20)	With HG (n=5)	Without HG (n=15)	All (n=20)	With HG (n=2)	Without HG (n=18)	itshape All (n=20)
PreHD glycemia	243.3±101.8	140.4±50.7*	277.7±91.0*	215.5±108.3	89.5±10.6**	229.7±105.0**	207.8±113.7
Intradialytic glycemia	151.9±70.2	78.0±16.2	176.4±63.8	165.2±72.2	77.0±41.9	175.0±68.2	157.6±75.4
p	0.0009	0.053	0.0008	0.0141	0.698	0.0484	0.019

*p=0.005/**p=0.082 (t test-Welch correction applied when necessary).

Conclusions: Patients with lower, though normal, preHD glycemia seems to present higher risk of developing intradialytic HG when using a dialysis solution without glucose and probably when using a solution with low concentration of glucose, too. These findings allow to suppose that diabetic ESRD patients who are in best glycemic control present higher risk of HG episodes during hemodialysis senssions with a glucose-free (or a glucose-poor) dialysis solution.

Su463 A CROSSOVER COMPARISON OF THE EFFECT OF THE ACETATE-FREE AND ACETATE-CONTAINING BICARBONATE DIALYSATE FOR HEMODIALYSIS PATIENTS

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Introduction and Aims: This study was designed to use acetate-free bicarbonate dialysate ("CARBOSTAR®", manufactured by AJINOMOTO CO., INC., hereinafter called "CAB") and conventional acetate-containing bicarbonate dialysate ("Kindaly 2E", manufactured by FUSO Pharmaceutical Industries, Ltd., hereinafter called "K2E") in a cross-over study and to analyze changes in test data and clinical symptoms.
Methods: The study involved 48 randomly selected patients on maintenance hemodialysis (23 males and 25 females). Their age was 64.4±11.7 years, and the history of dialysis was 7.0±6.8 years. For each patient, two dialysates (CAB and K2E) were used in cross-over study (each term for 6 months). Blood was sampled one month before and at the start of use of the first dialysate (CAB) and 3 months, 6 months (at the start of use of K2E), 9 months and 12 months later. Each blood sample was subjected to measurement of nutritional parameters (TP, ALB), indicators of atherosclerosis (pentosidine, PAI-1, lipoprotein homocysteine), inflammatory markers (H-CRP, IL-6), bone turnover markers (i-PTH, ALP, osteocalcin), clotting factor (fibrinogen), indicators of anemia (Ht, HB, Fe, TSAT, ferritin, TIBC) and serum electrolytes (iCa, corrected Ca, iP, Ca×P, β2–MG, hANP, BNP). In addition, changes in Kt/V, CTR and DW were recorded. As clinical symptoms, the frequency of each event (leg raising, use of saline replacement, use of hypertonic fluid, suspension of dialysis, blood pressure drop, leg cramp and discomfort) was counted. The study period was 16 months, beginning 4 months before the start of use of CAB and ending upon completion of the use of Kidaly. The setting for dialysis (dialyzer, dialysis time, blood flow rate, dialysate flow rate) was kept unchanged, as a rule.
Results: During the use of CAB, nutritional parameters tended to increase and lipoprotein level tended to decrease. Of the bone turnover markers, ALP and osteocalcin tended to increase and intact-PTH level rose in patients having low level of this parameter at the baseline. Both inflammatory parameters (H-CRP and IL-6) tended to increase. Of the indicators of anemia, ESA dose level showed no change but Ht and Ht improved. In evaluation of clinical symptoms, the frequency of leg raising, use of saline replacement and use of hypertonic fluid was lower during the use of CAB. The frequency of blood pressure fall and discomfort decreased while muscle cramp became more frequent during the use of CAB.
Conclusions: Acetate-free bicarbonate dialysate CAB was shown to be more useful than K2E in stabilizing the condition of patients during dialysis and improving the QOL. Acetate-free bicarbonate dialysate is expected in the long run to suppress induction of inflammatory cytokines, possibly leading to prevention and alleviation of MIA syndrome. Furthermore, in view of the acetate-free feature and the high bicarbonate level, this dialysate is promising as a means of symptom-free dialysis treatment.

Su464 SAFETY AND EFFICACY OF A PROTOCOL USING A HIGH CONCENTRATED CITRATE SOLUTION FOR ANTICOAGULATION DURING CONTINUOUS RENAL REPLACEMENT THERAPY

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Introduction and Aims: Regional citrate anticoagulation is being used more and more frequently during renal replacement therapy. Because citrate infusion is considered to induce metabolic and electrolyte disarrangements, protocols for citrate anticoagulation have been developed. Here we evaluate our protocol for citrate anticoagulation during continuous renal replacement therapy (CRRT) using a high concentrated citrate solution to minimize fluid load.
Methods: We studied 29 patients with acute kidney injury on the ICU, who required CRRT. All patients were at high risk of bleeding. For anticoagulation during CRRT patients were receiving 35 ml/h of a high concentrated citrate solution (1.26 M) manufactured by our pharmacy. We used a highflux dialyzer (F60S®, Fresenius) and a dialysate (HDE2® from BBRAUN) with a low concentration of $NaHCO_3$ (13.2 mmol/l) and NaCl (132 mmol/l). The blood flow rate was set to 100 ml/min. None of the patients received heparin.
Results: The average CRRT duration was 4.8±0.4 days (values are mean ±SEM). The filter lifetime was 39.1±1.8 h. The dialysate flow rate was 1.31±0.78 l/h and the transmembrane pressure ranged between 18.2 and 26.5 mmHg. The calcium concentration directly after the filter ranged between 0.20 and 0.25 mmol/l. The calcium substitution ranged between 0.52 and 1.10mmol/h. The electrolytes sodium, potassium and calcium remained in the normal range throughout the whole treatment. pH increased from slightly acidic to slightly alkaline values (7.36±0.02 to 7.43±0.01).
Conclusions: The use of high concentrated citrate solution for anticoagulation during continuous renal replacement therapy (CRRT) is not accompanied by relevant calcium, sodium and potassium or metabolic changes even in the setting of high risk ICU patients. Thus, anticoagulation with high concentrated citrate solution is safe, easy and effective.
Disclosure: The study was supported by a grant from BBraun Avitum AG.

Su465 nPCR DETERMINED ACCORDING EBPG IS HIGHLY OVERESTIMATED

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Introduction and Aims: According to the European Best Practice Guidelines (EBPG), PCR may be estimated with the use of formal single pool variable volume urea kinetic model (spvvUKM) or simplified derived equations; nPCR is then calculated as nPCR= PCR/AefBW*. To be adequate nPCR should be at least 1.1 g protein/kg ideal body weight(kgIBW)/day. Ionic dialysance (ID) constitutes an adequate estimation of dialyzer urea clearance corrected for total recirculation and its use in spvvUKM (UKM_{ID}) may be preferable to the use of "in vitro" dialyzer urea clearance.
Methods: In 81 patients, nPCR values obtained according to EBPG (nPCREBG) and to UKM_{ID} were compared with values obtained by DDQ method ($nPCR_{DDQ}$) actually considered the "gold standard method".
Results: nPCREBG resulted 1.19±0.34 g/kgIBW/day; nPCRID resulted 0.98±0.26g/kg IBW/day; $nPCR_{DDQ}$ resulted 0.93±0.25 g/kgIBW/day.
Comparison between nPCREBG ^{and} $nPCR_{DDQ}$ gives the following results: mean difference = -0.26±0.05; CI 95% from -0.35 to -0.17; p=0.000.
Comparison between $nPCR_{ID}$ ^{and} $nPCR_{DDQ}$ gives the following results: mean difference = -0.05±0.04; CI 95% from -0.13 to 0.03; p=0.214.
Finally comparison between nPCREBG ^{and} $nPCR_{ID}$ gives the following results: mean difference 0.21±0.05; CI 95% from 0.12 to 0.30; p=0.000).
Mean ID resulted 176±23 ml/min significantly lower than in vitro urea clearance (K) calculated according to mass transfer area coefficient (KoA), effective blood water flow (Qe) and adjusted for cardiopulmonary recirculation (difference -36±4 ml/min; CI 95% from -43 to -29; p=0.000).

Conclusions: nPCR determined according EBPG is highly overestimated To correctly estimate nPCR it is essential the correct estimation of urea distribution volume (V). When using the spVV urea kinetic model, V is directly related to the K value and there are some reasons for "in vitro" K overestimate "in vivo" K. Nowaday the availability of monitors allowing to to determine ID consents the correct determination of V and so of nPCR. In our patients, according to nPCREBG we could have considered as malnourished only 35 patients out of 82 while 55 were actually malnourished.
***Appendix:** AefBW = efBW+(SBW-efBW)0.25 where: AefBW = adjusted oedema free body weight; efBW = final body weight; SBW = standard body weight.
SBW = (h-100) - (h-150)/4 (male); SBW = (h-100) - (h-150)/2 (female).

Su466 THE INFLUENCE OF SUPERFLUX HEMODIALYSIS ON THE UREMIC TOXICITY PROFILE: A COMPARISON WITH POST-DILUTION HEMODIAFILTRATION

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Introduction and Aims: Both with on-line hemodiafiltration (OHDF), as well as with superflux hemodialysis (SHD) long term reductions in relevant larger molecular weight toxins have been achieved. However, the efficacy of both techniques in this respect have not yet been compared. If superflux membranes would have comparable effects on larger molecular weight uremic toxins as compared to OHDF, this might have highly significant impact for the prescription of dialysis therapies, as SHD is a technically less demanding technique compared to OHDF. The primary hypothesis of the study is that SHD and post-dilution OHDF are equivalent in terms of clearance and reduction in plasma levels of larger uremic toxins.
Methods: In this single-centre study, treatments were compared in a randomised cross-over design. Five patients were included. After a run-in period of 1 months in which all patients were continued to be treated with LHD, patients were randomised to treatment with SHD for a period of 8 weeks, followed by 8 weeks treatment with OHDF (filtration volume: 25% of blood flow), or to treatment with OHDF, followed by SHD. Uremic toxins (β2 microglobulin) inflammatory parameters and albumin (CRP and albumin), immune function (leucocytes and platelets), and other laboratory parameters (total and LDL cholesterol) were assessed. Comparison was made with paired T-test. Data were given in mean (SD).
Results: In these 5 patients no significant differences for albumin and β2 microglobulin at either one of the time points between OHDF and SHD were observed. Only ultrafiltrate albumin tended to be higher with SHD as compared to OHD (Table 1). Other parameters showed no significant differences.

Table 1. Larger molecular weight uremic toxins

	sAlb T2	sAlb T4	sAlb T6	sAlb T8	sβ2m T8	ufAlb T8	ufβ2m T8
SHD	33.9 (5,1)	33.6 (3,4)	29.5 (3,1)	32.5 (3,7)	23.4 (1,3)	87.3 (18,0)	1.5 (0.3)
OHDF	33.2 (1,7)	34.8 (2,1)	35.3 (2,8)	33.2 (4,0)	27.0 (12,8)	28.7 (8,5)	1.5 (0,8)

s = serum albumin, uf = ultrafiltrate.

Conclusions: SHD showed to be as effective as OHDF with regard to clearance of larger molecular weight uremic toxins. Only ultrafiltrate albumin tended to be higher with SHD as compared to OHDF. More patients are needed to draw definitive conclusions from this study.

Su467 DURABILITY ABILITY OF ENDOTOXIN RETENTIVE FILTER IN CENTRAL DIALYSIS FLUID DELIVERY SYSTEMS

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Introduction and Aims: Most Japanese facilities use central dialysis fluid delivery systems (CDDS). In addition, since high-flux dialysis membrane has been frequently used in Japan, the standard recommends that ultrapure dialysis fluid is used for all dialysis modalities at all dialysis facilities. So the endotoxin retentive filter (ETRF), as well as the reverse osmosis, is an indispensable device for the purification of dialysis fluid in this CDDS process. However, there are no clear safety standards or time-use specifications for the product. Concerning the timing for the replacement of ETRF, the endotoxin (ET) challenge test (durability test) was reviewed at many facilities in order to acknowledge present conditions and to study the safety and durability in clinical use.
Methods: The subject ETRFs were collected from ten dialysis facilities. Targeted ETRFs were Cuttol EF02(Nikkiso), JP-80K(JMS), TEF-1.0(Toray) and CF-609(Nipro). To confirm safety in the period of using of ETRF, the challenge test of ET for the used ETRFs should be recommended. The method of testing is to: Adjust the ET concentration level before loading it into the ETRF to be approximately 1.0 to 100EU/ml by using either untreated or incubated raw water taken from facilities. In the inhibition test, the test medium was infused at a rate of 500ml/min under a single pass (dead-end method). To evaluate the inhibition rate, 10ml samples were simultaneously taken into ET measuring tubes at two spots on both pre- and post-ETRF. Retentive capacity is estimated with the logarithm reduction value (LRV) of ET.
Results: The results showed that there are variations in the ET-retention capabilities of ETRF by different manufacturers. In addition, the LRV of some ETRF deteriorated as the duration of use increased. Between the first and second sides, the LRV on the second side tended to be higher. Most ETRF maintained LRV level 3 after three months of use, and LRV level 2 after six months of use.
Conclusions: It appears that the ET-retention capabilities of used ETRF vary depending on the method of usage and sterilization performed at each facility, however, we would like to request the manufacturers to produce ETRFs in which the retention capability remains stable for at least six month. Moreover, the retention capabilities should be able to maintain LRV level 3 concerning ET even after use. Lastly, we expect a statement from the manufacturers on the issue of recommended handling procedures for ETRF in order to secure LRV level 3 after six months of use.

Su468 OSMOLAL GAP IN HEMODIALYSED UREMIC PATIENTS

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Introduction and Aims: Osmolality is an expression of the number of particles in a given weight of solvent (mOsm/kg$H_2$0). Measured osmolality is determined by osmometer and calculated asmolality is estimated by 2xNa + UN/2.8 + glucose/18. The difference between measured and calculated osmolality is the osmolal gap. The purpose of the present study is to determine the measured and the calculated osmolality and the osmolal gap in hemodialysed uremic patients, pre- and post-HD.
Methods: To this aim in 24 non-diabetic uremic patients under regular hemodialysis (HD), blood samples pre- and post- HD were collected and serum osmolality measured (osmometer) and calculated (2xNa + UN/2.8 + glucose/18) and osmolal gap (measured – calculated osmolality) were determined. Also, the same parameters were determined in 22 healthy subjects (control).
Results: The findings of this study are summarized in table I. According to

Abstract Su468 – Table 1. Mean value ± SD of the determined parameters in patients and controls and statistical evaluation

Determined Parameters	Patients, Pre-HD Pre-HD n=24		Controls n=22		Patients, Post HD, n=24
Measured Osmolality (mOsm/kg/H2O)	313.33±2.88	←p<0.001→	293.72±2.37	←p<0.001→	302.08±2.43
Calculated Osmolality	302.33±4.03	←p<0.001→	290.54±3.11	←p<0.001→	294.7±2.96
Osmolal Gap	11±2.08	←p<0.001→	3.18±1.46	←p<0.001→	7.29±1.94
			←p<0.001→		

our findings the measured osmolality in patients are significantly higher pre- and post-HD in comparison to that of controls but post-HD is significant lower than pre-HD. Also, calculated osmolality is significant higher pre- and post-HD in comparison to that of controls but the value post- HD is significant lower than the pre- HD. The osmolal gap is significant higher pre- and post- HD in comparison to that of controls and the value post-HD is significantly lower from that of pre-HD.
Conclusions: Uremic hemodialysed patients present high measured and calculated osmolality pre-HD and remain high post-HD in comparison to that of controls in spite to the significant decrease post-HD in comparison that of pre- HD. Also, the osmolal gap is high pre-HD and in spite the significant decrease, remains high post-HD in comparison to that of controls. The high osmolal gap indicates indirectly the presence of unidentified endogenous osmoles in the serum of uremic patients which partly are improved by HD. The significant decrease of osmolal gap and osmolality by HD has an increased risk of dialysis disequilibrium syndrome.

Su469 ANTICOAGULATION IN HEMODIALYSIS IN SPAIN. A SURVEY BASED STUDY

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Introduction and Aims: Hemodialysis (HD) requires anticoagulation to avoid clotting of dialyzer and extracorporeal circuit. Low molecular weight heparin (LMWH) is reported to have several potential advantages over unfractionated heparin (UFH). However, there aren't still homogeneous and universally accepted criteria regarding the type of heparin, the mode of administration and the methods to the dosage control. The aims of study was to meet the anticoagulation methods used in the HD's regular practice of chronic renal disease patients in Spain, on what criteria depend those methods and which of them are used in the medication dosage.
Methods: Transversal design study based on a survey distributed in HD units in Spain, both public and private. This survey was answered by 87 units, in which a total of 6,093 adult patients have dialysis. The study asked about the use of UFH and/or LMWH and the criteria for its indication, the type of LMWH, the mode of administration, methods of dosage control, besides the use of UFH in the saline to rinse extracorporeal circuit.
Results: 8% of the tardet units just used UFH, 22% only used LMWH and the rest (70%) used both of them. 47% of the total of patients (6,093) used UFH, 49% used LMWH and 4% had dialysis without heparin. There are not any differences between public and private units. The type of LMWH (n=2.982) was: Enoxaparin 48.9%, Bemiparin 26.3%, Tinzaparin 9.8%, Nadroparin 9.7% and Dalteparin 3.5%. The most common indications for LMWH's used were medical criteria in 83.3% of the units, and in 29.5% the way of administration. Rinse of extracorporeal circuit with heparin was employed in 86,7% of the units when the HD's anticoagulation is done with UFH, and in 71,3% when it is done with LMWH. The most common doses of heparin in the rinse of extracorporeal circuit were 5,000 U (60%), although they oscillate between 1,000 and 10,000 U. The UFH is administrated in a continuous infusion in 35.8% of the units, in intermitent doses in 59.7% and in both ways in 4.5%. On the other hand, the LMWH is given only once at the HD beginning in 94.9% of the units. In the majority of the units the medication dosage is adjusted according to the coagulation of the circuit (88%), the bleeding of the vascular access after the disconnection (75%) and the weight of the patient (57.6%). Just 20% of them use the total time of coagulation or the partial time of thromboplastin to the UFH's dosage, and in 5.9% the anti-Xa factor to the LMWH's dosage.
Conclusions: LMWH and UFH's distribution in Spain is near 50% with each one, but there is a great disparity regarding the criteria and the mode of administration. In the majority of times, the medication dosage is fixed by trial and error, and the use of the laboratory's parameters is very infrequent.

Su470 EFFECT OF HIGH-FLUX HEMODIALYSIS USING NEPHRAL ST DIALYZERS ON BETA-2-MICROGLOBULIN (BETA-2-M) AND HS-CRP LEVELS IN CHRONIC DIALYSIS PATIENTS

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Introduction and Aims: Accumulation of beta-2-M takes part in the development of dialysis related amyloidosis and it's plasma concentration is considered as a general mortality predictor in chronic dialysis patients. Prognostic CRP value pertaining to sudden death has been reported in the general population and is suggested that CRP increase is a possible predictor of death in dialysis patients. The aim of the study was to evaluate the effect of hemodialysis using Nephral ST dialyzers on the clinical state and selected biochemical parameters.
Methods: The study included 40 chronic patients hemodialyzed during a time period of 7-31 years (mean 17,5) using polysulphone and hemophane low-flux dialyzers. Patients were dialyzed using Nephral ST dialyzers for a period of 6 months. Beta-2-M and hs-CRP concentrations were measured, as well as an evaluation of joint-bone pain intensification was performed based upon the VAS questionnaire (visual analogue scale).
Results: Results: 39 patients completed the study, one patient underwent renal transplant. The mean serum beta-2-M level during the period of treatment with low-flux dialyzers was 53,46 mg/l, while after 6 months of utilization of high-flux Nephral ST dialyzers beta-2-M decreased to 38,03 mg/l (a decrease of 28,86%). The mean serum hs-CRP level during treatment with low-flux dialyzers was 5,14 mg/l and after 6 months of utilization of high-flux Nephral ST dialyzers decreased to 4,15 mg/l (a decrease of 19,26%). Reduced symptoms of joint-bone pain were observed as noted in the VAS index.
Conclusions: High-flux dialysis with Nephral ST dialyzers reduces the inflammatory state and causes a significant decrease in beta-2-M concentration which may contribute to inhibition of dialysis related amyloidosis and decrease mortality in chronic dialysis patients.

Vascular access 2

Su471 10 YEAR SURVIVAL OF DISTAL AND PROXIMAL UPPER ARM ARTERIOVENOUS FISTULAS

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Introduction and Aims: The native upper arm arteriovenous fistula (AVF) is considered the vascular access of choice for patients with end-stage renal disease (ESRD). The radiocephalic fistula at the wrist (Brescia-Cimino) and the anastomosis between brachial artery and cephalic vein along with their variations, offer long-term access to hemodialysis patients. Most published data on AVF survival are restricted to 1 and 3 year follow up.
The aim of this study was to evaluate the 10 year survival rate of upper arm AVF and to identify risk factors for access failure, in a large series of AVF performed at a single Nephrology Center.
Methods: A retrospective analysis of the medical records of 189 patients started on dialysis between January 1991 and December 2000 was performed. Preoperative careful clinical evaluation and vascular mapping was done routinely, while in selected patients, additional imaging (venography) was performed to exclude central vein stenosis. Patient records were reviewed to determine demographic and clinical information, including age, gender, diabetes, hypertension and cardiac failure. Primary access survival was plotted using Kaplan-Meier survival techniques, with patient follow up censored for primary failure, death with a functioning AV access, change of treatment modality and transfer to another dialysis unit. Access survival was compared between proximal and distal AVFs by the log rank test.
Results: One hundred thirty patients (77 males, 53 females), with an average age of 55 years old (19 – 85) were included in the study. Overall, 156 AVF were performed (68 distal and 88 proximal), while 108 (65.8%) were initially created and 56 (34.2%) were AVF in patients with previous failed fistulas. Overall median AVF survival was 47.9 months, while survival rates were 82%, 67% and 66% at 12, 24 and 36 months for distal and 88%, 79%, 64%

for proximal AVF respectively. The additional 5 and 10 year survival was 30% and 12% for distal and 27% and 15% for proximal AVF respectively. There was no statistical significant difference in survival rates between initially and subsequently created AVF (ρ=0,188). In addition, no difference was found in the long-term survival between proximal and distal AVF (p=0,145). Among the examined patients' characteristics only age at the time of AVF construction was negatively related to access survival (p=0,002).
Conclusions: The native upper arm AVF's are associated with high long-term patency rates. Careful preoperative clinical evaluation and vascular mapping contribute to the high long-term AVF survival rate.

Su472 VASCULAR ACCESS AND MORBIDITY AND MORTALITY IN HEMODIALYSIS: THE RISCAVID STUDY

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Introduction and Aims: An adequate vascular access (VA) plays a pivotal role in the performance of dialytic treatment. Arteriovenous fistula (AVF) is the gold standard VA for haemodialysis, and the primary use of AVF is recommended by the Guidelines of all Societies of Nephrology. Some Authors reported differences in morbidity and mortality of dialytic patients with different VA, but these studies have been till now inconclusive. In this study, we have investigated the relation among different VA and mortality and morbidity in an omogeneous population of 755 HD patients enrolled in the RISCAVID Study (Cardiovascular Risk in Dialytic Patients from North-West of Tuscany) from 15 different dialytic centres.
Methods: The M:F ratio was 1.5:1, the mean age was 6514 years, and mean dialytic age was 7077 months. At the beginning of the observation (June 2004) the AVs were so divided: 82% AVF, 10% synthetic vascular graft (SVG) and 8% CVC. After two years the normalised Relative Risk (RR) for cardiovascular (CD), not cardiovascular death (NCD), VA thrombosis, and not lethal cardiovascular events in the three different groups of patients was calculated.
Results: The CVC patients were older (p=0,009) and had a higher dialytic age (p=0,02). Diabetes (p=0,048), infectious comorbidity (p=0,018) and CV disease (p=0,004) were more frequent in CVC patients. The CVC as vascular access was associated with lower albumin levels (p=0,0001), lower Kt/V (p=0,0001), lower Hb levels (p=0,013) and higher level of CRP (p=0,04).
The 3 years unadjusted survival was 72% for AVF/SVG vs 52% for CVC (p= 0,004). After adjusting for Age, Dialytic vintage ad main comorbidity CVC patients showed a 1,48 RR for all cause mortality (p=0,048). The frequency of thrombosis of the AV results to the low limits in comparison to how much generally reported in literature (0.04 events/pts year) with a RR = 2.4 for the SVG. The same resulted significantly higher in a Centre in comparison to all the others (RR max = 2.32 and RR min = 0.19). Finally, mean hospitalisation was significantly higher for CVC (12.716 days/year) and secondary AVF (11.221 days/year) when compared with first AVF (5.6 9 days/year) and SVG (9.621 days/year) (p<0,05).
Conclusions: Our study confirmed the greatest safety of the AVF and SVG in terms of cardiovascular and not cardiovascular mortality, and in the development of sepsis (it is important to notice that the results of NCD were not significantly influenced by the higher prevalence of malignancy in this group). Data regarding mean hospitalisation suggested a role for the type of VA as a predictor of global health of dialityc patients. Finally, the differences in RR for VA thrombosis among the Centres suggest the pivotal role of nurses training for preventing this complication.

Su473 ASSOCIATION OF VASCULAR ENDOTHELIAL GROWTH FACTOR AND THROMBOSIS OF NATIVE ARTERIOVENOUS FISTULA IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Venous thrombosis as a result of neointimal hyperplasia is believed to be the major cause of arteriovenous fistula (AVF) dysfunction in hemodialysis patients. The thrombotic lesions are characterized by increased expression of various growth factors including vascular endothelial growth factor (VEGF). The actual role of VEGF is still not clear. Evidences from experimental studies suggested both detrimental and beneficial effects of VEGF in the atherogenic process. In this present study, we evaluated the relationship between serum VEGF and AVF thrombosis in chronic hemodialysis patients.
Methods: Ninety-nine prevalent adult end-stage renal disease patients undergoing chronic hemodialysis via native AVF were enrolled. The patients who received intervention for AVF or had systemic infection disease within 1 month before entry were excluded. The episodes of non-infectious AVF thrombosis which results in percutaneous angioplasty, thrombectomy, or recreation of AVF were retrospectively recorded for 2 years. A mid-week pre-dialysis fasting blood sampling was done for the measurements of VEGF, high sensitivity C-reactive protein (hs-CRP), interleukin-6, tumor necrosis factor-α, transforming growth factor-β, hematocrit, albumin, cholesterol, triglyceride, high-density lipoprotein cholesterol (HDL), low-density lipoprotein cholesterol, total cholesterol to HDL ratio (TC/HDL), blood urea nitrogen, creatinine, and calcium-phosphate product. Besides them, gender, age, body mass index, diabetes, smoking habit, blood pressure and current use of lipid-lowering agents were all considered in the statistical analysis.
Results: Of the 99 patients, 29 (29.3%) experienced one or more AVF thrombosis episodes in the 2-year retrospective record. In univariate analysis, only hs-CRP $\geq$ 0.75 mg/dl (odds ratio 2.347, 95% CI= 1.03-5.81, p=0.045) and VEGF $\geq$ 200 pg/ml (odds ratio 2.903, 95% CI= 1.10-7.69, p=0.032) showed significant correlations to the presence of AVF thrombosis. In the multivariate logistic regression model, after controlling age, diabetes, TC/HDL and hs-CRP, VEGF $\geq$ 200 pg/ml still showed significant correlation with the presence of AVF thrombosis, odds ratio 3.220, 95% CI= 1.02-10.20, p=0.047.
Conclusions: Our study demonstrates a close association between VEGF and the thrombosis of native AVF in chronic hemodialysis patients. The role of VEGF seems to be more important than that of inflammatory cytokines and CRP.

Su474 DIALYSIS ACCESS IN THE US – IS HIGHER FLOW THE DETRIMENTAL FACTOR?

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Introduction and Aims: Vascular access is the Achilles' heel for hemodialysis patient care. In the United States (US) prevalence of autogenous accesses has increased in the wake of Fistula First from 24% in 2000 to 52% in 2008, however, there is much debate about the quality of these accesses. European studies of success of fistula creation and frequency of use often find much different results than their US counterparts. A defining character of dialysis accesses in European studies is that the majority of them is located in the forearm, with potential for dual outflow through upper arm cephalic and basilic veins and subsequent low access pressures.
We aimed to determine the frequency of the location of dialysis accesses in a US dialysis population and measure access flows which in setting of stenosis would translate to higher intra-access pressures.
Methods: All patients of our institutions outpatient dialysis unit where characterized regarding type (autogenous versus prosthetic/biograft) and location (forearm versus upper arm). All patients referred to our interventional nephrology service with access dysfunction underwent access flow measurements before and after angioplasty using a Transonic flowmeter.
Results: At the time of evaluation 86 of 102 patients were using a graft or fistula for dialysis. Of 22 prosthetic or biograft accesses 8 were in the forearm with arterial anastomoses at the elbow in 7 and one at the wrist. 14 accesses were in the upper arm with all but two arterial anastomoses at the elbow. Of the 64 autogenous accesses 20 were in the forearm and 44 in the upper arm.
During the study period from May 2009 to January 2010 101 patients underwent angioplasty with measurements of pre- and post-angioplasty dialysis access flow. Mean post-angioplasty blood flows were 1260ml/min in the forearm prosthetic accesses (n=7) and 1430ml/min in the upper arm prosthetic accesses (n=10), 1260ml/min in forearm biograft accesses

(n=10), and 1260ml/min in upper arm mixed-autogenous-interposition biograft accesses (n=7). Forearm autogenous access mean blood flow was 845ml/min (n=11) and upper arm autogenous access blood flow was 1580 ml/min (n=56).
Conclusions: Upper arm accesses are more prevalent than forearm accesses. In particular, upper arm autogenous accesses outnumber forearm fistulas 2:1. Access flow is highest in upper arm autogenous accesses, over 85% higher than average forearm autogenous access flow. This increase in flow has consequences for intra-access pressure dynamics if stenoses develop and development of stenoses themselves may be affected.

Su475 ARTERIOVENOUS FISTULA (AVF) CALCIFICATION AND ASSOCIATED FACTORS: THE ROLE OF FIBROBLAST GROWTH FACTOR (FGF)-23?

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Introduction and Aims: (FGF)-23 is a protein with phosphaturic activity and considered to be the most important factor for regulation of phosphorus homeostasis in chronic kidney disease.Disturbed calcium and phosphorus homeostasis contribute to vascular calcification.The aim of this study is to identify the associated factors for AVF calcification and the importance of FGF-23 in AVF calcification.
Methods: We evaluated 46 chronic haemodialysis (HD) patients from one centre. AVF calcification was meassured by spiral computed tomography(CT), lesions were scored according to 80 Hounsfield Units at arterial and venous sites. Blood was drawn before a midweek HD session for c terminal FGF-23(cFGF-23) at the time of CT. Patient's characteristics, medical history, medications, laboratory tests during 3 months prior to CT and FGF-23 levels were compared to calcification scores.
Results: Among 46 patients 58,7% were male.The mean age was 52,1±15,5.The mean HD duratian was 113,7±81,3 (6- 302) months.Primary renal disease was 28,3% HT, 17,2% glomerulonefritis, 10,9% DM. Patients with a HD Schedule of 3x4 hr/wk were 93,5% (2x4,5:6,5%). 1,25 mmol/L calcium dialysate was used in all patients. The mean Kt/V was 1,52±0,2. Patients using calcium containing phosphate binders were 97,8%. Among these 11,1% were using CaCo3 alone, % 88,9 were using Ca acetate.Two patients used sevelamer for a short period during six months. Patients using calcitriol were 67,4%. The mean AVF age was 72,6±56,8(5-273) months.We found calcification in 58,7% of AVF. Patients with evidence of any calcification on CT had longer HD duration (132,7±82,6 vs 86,6±73,1; p: 0,024) and higher AVF age compared to patients without calcification (84±63,6 vs 56,4±41,8; p: 0,071). The median FGF-23 levels were 1663,5 RU/mL (175-5521). FGF-23 values were analyzed according to quartiles(Q1 to Q4). Patients with residual urine volume of >400 cc were higher in Q1 compered to other quartiles (p: 0,005).The frequency of females in Q4 were higher (72,7%; p: 0,052). There was no differance between quartiles according to vascular calcification scores,iPTH serum levels, cumulative calcitriol and calcium containing phosphate binder doses during 6 months period.The patients in Q4 had higher serum levels of Ca (9,5±1 vs 8,5±0,4 gr/dl; p: 0,046), P (5,1±0,6 vs 4,3±0,7; p: 0,026) and CaxP product (49,8±10,4 vs 36,9±7,5; p: 0,006) compered to Q1.
Conclusions: This study demonstrated no correlation between FGF-23 levels and AVF calcification maybe due to cross-sectional nature of the study or small number of patients.Only associated factors with AVF calcification were dialysis duration and AVF age. We detected AVF calcification in most of our patients, despite relatively well controled Ca and P homeostasis.

Su476 DISTAL ARTERIAL FLOW IN PATIENTS WITH ARTERIOVENOUS FISTULA FOR HEMODIALYSIS

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Introduction and Aims: The objective of this study was to document changes in the distal circulation after creation of a proximal arteriovenous fistula and to correlate these findings with the patient's clinical condition.
Methods: We prospectively examined 36 patients scheduled for upper extremity shunt creation. We used color and spectral Doppler sonography to examine flow in the radial and ulnar arteries, noting flow direction, Doppler waveform and peak systolic velocity. After the shunt procedure, we repeated the measurements and correlated them statistically with hand symptomatology.
Results: The mean peak systolic velocities in the radial and ulnar arteries were 54 and 62 cm/sec, respectively, before surgery, and decreased to 15 cm/sec after surgery in the radial artery and 40 cm/sec in the ulnar artery. The mean percentage of decrease in peak systolic velocity was 72% in the radial artery and 35% in the ulnar artery. Sixteen patients showed reversed flow. Twelve (33%) of 36 patients were symptomatic. No statistical correlation was found between change in peak systolic velocity values before and after surgery and the presence of hand symptoms. No correlation was found between flow reversal and symptoms. The most consistent factor associated with symptoms was diabetes, but only 54% of the diabetic patients were symptomatic.
Conclusions: The difference in the peak systolic velocities in the radial and ulnar arteries after fistula creation does not correlate with symptoms. The hand can tolerate a significant decrease in the peak systolic velocity and even flow reversal without symptomatology.

Su477 RECONSTRUCTION OF VASCULAR ACCESS IN DIALYSIS USING AUTOGENOUS-VENOUS-GRAFT IMPLANTATION

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Introduction and Aims: The vascular access (VA) for hemodialysis (HD) patients can occasionally develop total occlusion of the antebrachial proximal cephalic vein, especially in patients associated with antebrachial radialis arteriovenous fistula(AVF). It is important to maintain a functional anastomosis and to simplify the cannulation for VA survival and comfortable dialysis. Autogenous-venous-graft implantation (AVGI) was designed to practice reconstruction of VA. VA failure occurs when to antebrachial proximal cephalic venous occlusion causes elevation of intravenous blood pressure in patients with adequate AVF function without stenosis of the anastomotic area. The venous outflows flow backward to the distal veins such as the dorsal venous network of the hand, dorsal metacarpal veins and basilic vein. These veins are expanded and then cause venous hypertension associated with edema and pain. These veins provide auto-grafts of the correct size. The stenotic veins with total venous occlusion are excised and replaced by the auto-graft using venous-venous anastomosis. The prognosis of VA cases demonstrates the efficacy of this technique.
Methods: This study enrolled HD patients who underwent reconstruction using AVGI because of VA failure including hardness of cannulation, edema due to venous hypertension or aneurismal formation, and could be followed from 2003 to 2009. Each patient underwent angiography to confirm the stenosis. Thereafter, percutaneous transluminal angioplasty(PTA)was performed immediately. The first patency was defined as the interval from the reconstruction to the first PTA or from a PTA to the next PTA; the second patency was the interval from reconstruction or the last PTA to the re-reconstruction due to VA failure. Cumulative patency was calculated by the Kaplan-Meier method.
Results: Eight patients were included, and 5 of them were male. The mean age was 72.4 years of age. The cause of end-stage renal disease was chronic glomerulonephritis in 5, nephrosclerosis in 2, and post renal failure in 1. The follow-up was a mean of 23.3 months (range, 3-74 months). Seven patients underwent angiography because of VA trouble or the possibility of venous stenosis. Stenosis of the venous-venous anastomosis was found in seven of eight patients. These lesions were treated with PTA an average of 2.4 times per patient (range, 1-7). The mean interval from reconstruction to the first PTA was 129 days (range, 40-213 days). There was no incidence of VA failure of the need to use either a temporary vascular catheter or to perform a re-reconstruction. The median cumulative first patency was 154 days.
Conclusions: The mortality rate is higher for those with arteriovenous graft in comparison to those with autogenous AVF. Basilic vein AVF is

not comfortable at cannulation. Indeed, AVGI has a high frequency of stenosis at the venous-venous anastomosis but the prognosis is good, if PTA is performed appropriately. This technique is extremely effective for the treatment of a total occlusion of the antebrachial proximal cephalic vein at the radialis AVF in HD patients.

Su478 THE FACTORS AFFECTING THE EFFICIENT AUTOGENIC ARTERIO-VENOUS FISTULA CREATION; THE RESULTS OF ONE YEAR PROSPECTIVE OBSERVATION

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Introduction and Aims: The creation of well functioning autogenic arterio-venous fistula (AVF) reduces the morbidity and mortality in hemodialysis programs. In the present study the factors affecting the establishment of the effective vascular access were analyzed in one year prospective observation.
Methods: Among 213 patients encompassed in HD program 189 (89%), 123 M and 66 F, 64.4±13.4 years old, were treated with use of native AVF and underwent one-year prospective observation. The remained 24 patients were dialyzed with the catheter use. The regular physical examination of vascular access was performed quarterly in all patients. Doppler ultrasound was done in selected cases. Based on the investigation results and the way of AVF puncturing (single or double needle) the patients were divided into DN I (double needles without stenosis), SN I (single without stenosis), DN II (double needle, presence of stenosis), SN II (single needle, presence of stenosis) and the catheter group.
Results: The following types of AVF were employed: distal RCF in 122 (64.5%), proximal RCF in 43 (22.7%), UBF in 1 (0.5%), Gracz fistula in 14 (7.4%), BCF in 6 (3.2%) and BBF in 4 (2.1%) patients. By physical exam stenoses were identified in 70 patients (37%). The sensitivity, specificity and accuracy of physical assessment compared to ultrasound was 93.3, 86.7 and 90%, respectively.
DN I, SN I, SN II, DN II and catheter group encompassed 91 (48%), 28 (15%), 53 (28%), 17 (9%) and 24 (11.3%) patients, respectively. No effect of the patients age and the cause of ESRD on the possibility of AVF creation, and it's quality was found. Higher percentage of single-needle fistula was observed in women, 40.9% vs. 14.6% in men, $p<0.001$. Coronary heart disease was noticed more rarely in DN I compared to others, 35.2% vs. 68.6%, p=0.029. Peripheral arterial occlusive disease was observed more often in SN II and catheter groups, 43.9% vs. 22.7% in remained, p=0.049. The highest incidence of congestive heart failure was found in catheter patients, 70.8% vs. 41.8% in others, p=0.011. The percentage of non-smokers was the lowest in patients with stenosed AVF (SN II and DN II), 24.3% vs. 49.6% in remained, p=0.007.
During one year follow-up 10 patients underwent the successful stenosis repair. Thrombosis occurred in 23 patients (12%). Vascular access failure appeared more frequently in the stenosis groups (SN II and DN II) relating to patients without AVF malfunction (SN I and DN I), 25.7% vs. 3.9%, $p<0.001$.
Conclusions: 1. The creation of the effective AVF is possible in the vast majority of the current HD population despite the significant burden of vascular comorbidities.
2. The occurrence of comorbidities (congestive cardiac failure, coronary heart disease, peripheral arterial occlusive disease) and smoking exert the negative impact on the vascular access quality.

Su479 DISTAL OR PROXIMAL FISTULA APPROACH? IN MEDIO STAT VIRTUS

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Introduction and Aims: American and European guidelines recommend the distal radial-cephalic fistula (dRCF) as the first and best hemodialysis (HD) access in end stage renal disease (ESRD) patients. However this kind of arteriovenous fistula (AVF) shows a limited primary unassisted patency and frequently needs surgical revisions and/or angiographic procedures. If dRCF is not feasible, guidelines suggest a proximal brachiocephalic(AVF). The middle-arm fistula (MAF) has been suggested as possible alternative approach to dRCF. Aim of the study was to evaluate the MAF primary unassisted patency rate, the most frequent causes of MAF failure and the possible related factors.
Methods: Data on patients with a MAF placed from January 1991 until June 2008 were retrospectively collected. The cumulative probability of MAF failure overall and by the main subgroups was estimated according to Kaplan-Meier with Greenwood standard error (SE), and comparison of failure between different subgroups was performed using log-rank test in univariate analyses. The Cox regression model was used to investigate factors that independently affected the overall hazard of failure and cause-specific hazard of thrombosis.
Results: At the end of follow-up 14.9% of MAF failed (12.9% thrombosis, 2.0% stenoses), while 42.2% of MAF were still working with patients alive. Cumulative probability of MAF unassisted primary patency after 4 years from the creation was 77%. Univariate analyses highlighted that female (p=0.019), underweight patients (p=0.020) and MAF implantation after HD starting had a higher risk of MAF failure for any cause than male, normal and overweight patients and MAF implanted before HD starting ($p<0.001$). Results of the Cox multivariate analysis for overall MAF failure confirmed that only MAF implantation before HD starting is a protective factor against any failure (p=0.002), while female gender (p=0.027) was associated to an increase of the thrombosis hazard (HR = 1.87, CI 95%: 1.07-3.25).
Conclusions: Our data demonstrate that MAF has a good unassisted primary patency and suggest that this kind of AVF could be a valuable alternative surgical approach to dRCF in ESRD patients.

Su480 ARTERIOVENOUS FISTULAS FOR HEMODIALYSIS IN DIFFICULT PATIENTS – CONTRIBUTING FACTORS FOR PRIMARY PATENCY

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Introduction and Aims: The arteriovenous fistula (AVF) is the preferred access for hemodialysis since it is associated with the lowest morbidity and mortality. Diabetic and elderly patients have higher incidence of peripheral vascular disease and other co-morbidities that may negatively affect the success of an AV fistula. Accurate preoperative vascular mapping with doppler ultrasound (DU) may contribute to increase AVF success.
Methods: The outcome of vascular fistulas created in our department between 01/01/2008 and 31/12/2009 was retrospectively reviewed. We elected 288 patients (F=110; M=178) with mean age of 65±15 years. Ninety-one (31,6%) were diabetic, 155 (53,8%) were in pre-dialysis stage, 102 (35,4%) were treated with antiaggregant and 148 (51,4%) had failure of at least one previous vascular access.
Results: These 288 were submitted to 379 interventions: in 112 (29,6%) a radial-cephalic fistula was created and in 216 (57,1%) an brachial-cephalic or brachial-basilic was made; 20 were submitted to vein transposition and 4 to surgical correction of fistula. Preoperative evaluation included DU in 198 (52,3%) cases. The global primary failure of AVF was 28% (54,5% in radial-cephalic; 17,3% in arm fistula and 5% in vein transposition). Infection complicated 14 (3,7%) interventions.
No statistical difference in primary patency was found related to age, sex, diabetes or antiaggregation. Primary failure was more frequent in radial-cephalic fistulas (50% vs. 18,5%; $P < 0,001$), in pre-dialysis patients (34,5% vs. 21,8%; P=0,003) and in patients with no preoperative vascular mapping with DU (34,4% vs. 22,2%; P=0,009).
Conclusions: Preoperative UD evaluation may contribute to increase AVF success in difficult patients.

Su481 FISTULA SURVEILLANCE WITH BLOOD TEMPERATURE MONITOR. NEW ASPECTS

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Introduction and Aims: Complications associated with vascular access in hemodialysis is a major source of morbidity in end-stage renal disease patients. Measurement of recirculation (Rn), forced recirculation with reverse blood lines (Rf) and calculating the vascular access flow (Qa) from these parameters using thermodilution technique proves to be a possible solution. Based on the National Kidney Foundation Kidney Disease Outcomes Quality Initiative (NKF K/DOQI 2006) Rn using a non-urea-based dilutional method is accepted, Qa measurement is the preferred method. The critical value of Qa suggested by NKF K/DOQI 2006 is < 500-600 ml/min in native fistulae, while the cut off value suggested by European Best Practice Guidelines on haemodialysis (EBPG 2007) is < 300 ml/min in forearm fistulas. The aim of our study is:
1. Establish the the optimal cut off value of Qa in the different locations of native fistulas.
2. Estimate the value of Rn, Rf, and low difference (< 10) between Rn an Rf (delta R).
3. Looking for additional parameters to detect fistula stenosis.
Methods: We performed 250 measurements with Fresenius 4008 blood temperature monitor in 113 hemodialysis patients bearing native arteriovenous fistulas at the beginning of dialysis sessions. Rn was measured with conventional, Rf with reverse bloodlines. Qa was calculated with the software developed by Fresenius. We evaluated the parameters using Receiver Operating Characteristic curve (ROC) analysis calculating the area under the curve (AUC) and estimated the optimal cut-off of Qa and Rf for detecting fistula stenosis. We supposed also vascular access failure at insufficient blood flow in reverse line position (RLI). The results were confirmed by colour duplex ultrasonography and/or angiography or in positive cases at clinically developed fistula failure. Stenosis >= 50% was corrected by percutaneous transluminal angioplasty (68 cases).
Results: 1. The best value of AUC was 0,866 in Qa of all cases; 0,848 in Rf, 0,808 in delta R < 10 and only 0,652 in Rn.
2. Using the ROC analysis the optimal cut off value of Qa was 500 ml at wrist and 600-700 ml/l at forearm and elbow fistulas, of Rf > 33%.
3. At Qa < 500 ml at wrist, <700 ml at forearm and elbow fistulas, or delta R < 10 or Rf > 33% or RLI the sensitivity: 0,92; specificity: 0,50; positive predictive value: 0,60; negative predictive value: 0,89.
Conclusions: The best parameter is Qa, but Rf, delta R and RLI are also useful to detect stenosis.
The predictive value of Rn is very low.
The optimal cut-off of Qa at the distal part of the upper limb is higher than the previously suggested value.

Su482 THE EFFECT OF A REMOVAL OPERATION USING THROMBASTER II ON THREE CASES OF RECURRENT VASCULAR ACCESS TROUBLE IN HEMODIALYSIS DUE TO INTIMAL HYPERTROPHY

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Introduction and Aims: Nowadays, The Conventional Percutaneous angioplasty (Conventional PTA) for vascular access trouble in hemodialysis is sufficiently effective on account of its continued development and improvements in technique. The Conventional PTA can contribute greatly to maintainance of a vascular access. However, resistance to the benefits of the Conventional PTA still remains in the form of recurrent vascular access trouble due to intimal hypertrophy.
The purpose of this report is to highlight the effectiveness of Thrombaster II, which is routinely utilized as a device in thrombectomy, on a recurrent vascular access sterosis due to intimal hypertrophy that was resisting treatment by Conventional PTA.
Results: The first case was a 65 year-old male who had a severe stenosis at the venous anastomosis of the arteriovenous graft (AVG). I performed Conventional PTA. However, The stenosis restituted a month after conducting the PTA. I then performed a second Conventional PTA procedure, but this time using Thrombaster II as an intima removal device. The lumen diameter has been maintained now for six months.
The second case was a 80 year-old male who had a vascular access occlusion of the AVG due to an arterial anastomosis stenosis. The occlusion had a relapse one week after conducting Conventional PTA. The lumen diameter has been maintained so far after conducting PTA with Thrombaster II.
The last case was a 66 year-old female who had a run-off vein occlusion of the arteriovenous fistura (AVF). The occlusion had a relapse two weeks after conducting Conventional PTA. I then performed a PTA with The Peripheral Cutting Balloon (PCB). PCB had a considerable effect, but the lumen diameter had gradually narrowed after conducting this procedure. Next I used Thrombaster II. The Thrombaster II had a sufficient effect. The lumen diameter has been retained and the vascular wall has thinned.
Conclusions: In conclusion, I hope I have demonstrated that Thrombaster II could have great potential as an intimal removal device.

Su483 THE CREATION OF NATIVE FISTULA AS FIRST VASCULAR ACCESS IN ELDERLY PATIENTS: AN ITALIAN EXPERIENCE

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Introduction and Aims: In the last decade, the number of elderly patients (>65 years) who started hemodialysis treatment increased. Elderly patients present many comorbidities that can make difficult to create a vascular access using native vessels. The type of vascular access (fistula-AVF, graft-AVG, permanent catheter-pCVC) plays an important role in the results of dialysis treatment. Many complications can affect the vascular access and interfere with the morbidity and mortality of patients. Due to lower rate of infections and thrombosis compared to AVG or pCVC, even in elderly patients AVF should be the goal.
Methods: We analyzed our patients who started hemodialysis in 2 centers in a northern Italy area between 1st January 2006 to 31st December 2008 (Trento and Brescia). Hemodialysis population was about 600 patients, in an area of 1.100.000 people. Patients were divided in two group: older or younger than 65 years. The analysis between the group was performed using the following items: demographic characteristic (age, gender, risk factors, site and type of vascular access. Moreover we evaluated the survival rate of patient and patency of vascular access.
Results: In a 3-year period 336 patients started dialysis treatment in Trento and Brescia. Patients were divided in two group: 208 were older than or 65 years and 128 were younger than 65 years. During the study we analysed the kind of the first vascular access and vascular access survival.

Type of vascular access

	< 65 years (young)	> 65 years (elderly)
distal AVF (%)	89 (69%)	102 (49%)
prossimal AVF (%)	25 (20%)	55 (27%)
AVG	6 (5%)	9 (4%)
p CVC	8 (6%)	42 (20%)

Kind of vascular access in two patients group

During the study period, in the total population 264 patients (78.6%) maintain the first vascular access, 72 (21.4%) required one or more surgical procedures in order to obtain/maintain a functioning vascular access.
The rate of primary patency of AVF was similar in the two groups at the end of follow-up; a statistically significant difference was observed in AVG patency: 100% in young and 44% in elderly patients (p=0.018). Using Cox analysis for vascular access survival, we did not observe any statistically significant difference between the two group for all the risk factors tested.
Conclusions: According to NFK DOQI Clinical Practice Guidelines a native fistula is the preferred types of vascular access. Our data suggest that this kind of vascular access can be created in more than 70% of elderly patients.

Su484 VASCULAR ACCESS ANEURYSM/PSEUDOANEURYSM IN PATIENTS ON REGULAR HEMODIALYSIS: PREVALENCE, RISK FACTORS AND OUTCOME

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Introduction and Aims: The prevalence of vascular access aneurysm (VAA) and pseudoaneurysm (VAPA) differs between clinical reports, as well as its clinical consequence. Most remain asymptomatic but the others require surgical correction or even closure of AVF.
The aim of this single-centre study is to evaluate the prevalence, risk factors and outcome of VAA and VAPA in patients on regular hemodialysis.
Methods: We examined vascular access in 197 patients on regular hemodialysis: 155 with AVF, 27 with AVG and 7 with permanent vascular catheter (excluded from further analysis).
Patients were dialyzed 12 hours per week.
Patients with VAA/VAPA were classified into 3 groups according to the scoring system (length, cm+ width, cm): Group I (score 0.5cm-4.5 cm), Group II (score 5.0cm-9.5 cm) and Group III (score > 10 cm). VAA/VAPA were correlated with patients' characteristics, duration of dialysis and duration of current VA, vascular calcification data (revealed by X-ray and ultrasound) and iPTH.
Results: Aneurismal/pseudoaneurismal changes of vascular access were detected in 97 (52%) out of 190 patients. Majority of patients had one (30.9%) and two (51.5%) aneurysms; other had three (12.3%) and even four aneurysms (5.3%). The width of aneurysms was 1 cm (49.3%), 2 cm (41.5%) and 3 cm (9.2%). The length of aneurysms was less than 3 cm (35.1%), 3-5 cm (41.4%), 5-10 cm (22.1%) and >10 cm (1.4%). There was no significant difference between patient's age; however, patients with VAA/VAPA were significantly longer on hemodialysis (9.5+5.2 years vs. 5.7+4.6 years) and they had significantly longer duration of actual vascular access (8.3+4.8 years vs. 3.8+3.4 years). Patients w/o VAA/VAPA had significantly higher: overall vascular calcification score (4.8+3.5 vs. 6.2 +3.3), VA calcification score (1.03+1.04 vs. 1.26+0.99) and pulse wave velocity (9.7+2.5 vs. 10.7+2.4). Plasma iPTH value was higher in patients with VAA/VAPA (537+611pg vs. 414+471pg). Patient with highest score were older, longer on HD, with older VA, with higher iPTH value.
12 were operated (5.4%). No accidents were detected.
Conclusions: In our group of patients, there is high percent of aneurismal complications of VA. Aneurisms are more frequent in patients with longer dialysis vintage, with longer use of actual vascular access and in those wit sHPT. VAA/VAPA are usually asymptomatic and rarely require surgical treatment. Additional effort is needed to avoid dilated regions of fistulas and to prevent further progression toward aneurysms/pseudoaneurysms.

Su485 EFFECTIVENESS OF A SHOWER CLEANSING TECHNIQUE WITHOUT ANTISEPTIC AGENTS AND HISTOLOGICAL FINDINGS AT EXIT SITES OF TUNNELED CUFFED VENOUS CATHETERS

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Introduction and Aims: Exit site infection of the tunneled cuffed venous catheter (TCC) is a serious complication, and appropriate care is important in preventing it. In general, the recommended care consists mainly of disinfection around the exit sites. However, we have developed and reported a new care method designed to prevent infection by normalizing skin conditions at the exit site: the exit site is directly cleaned using a shower without the use of antiseptic agents. The aim of this study was to evaluate this care method and to determine the histological findings for the skin at the exit site of the TCC.
Methods: The subjects were 60 hemodialysis patients (male/female, 45/15; mean age, 67.2±11.8 years; diabetes mellitus/non-diabetes mellitus, 31/29; mean duration of catheter placement, 54.9±42.0 days) who had begun use of a TCC and gave informed consent to participate in this study at Sagamihara Kyodo Hospital and Hashimoto-Minami Internal Medicine Clinic between January 2008 and December 2009. Our method involves washing the exit site of TCC with tap water immediately after catheter insertion. The exit site is washed with a tap water shower if any soiling is present. Moisture is wiped away with gauze or similar material, and the TCC is fixed with appropriate tape. No antiseptic agents are applied. Any non-sterile gauze and tape can be used. While applying the shower at home, the exit site is not to be covered with water-proof film. This cleaning can be performed at home routinely. Direct showering removes any discharge or stain. Moisture is then wiped away with a clean laundered towel, and finally the TCC is fixed in the same fashion as at the time of dialysis. Histological examination of the skin at the exit site of the TCC at the time of removal was performed in 10 patients who gave informed consent for it.
Results: Exit site infection occurred in 3 patients (5.0%). Neither tunnel infection nor catheter-related bacteremia was observed during this investigation. Histological examination revealed that epidermis at the exit site exhibited neither erosion nor ulcer, and that the normal structure of the dermis was well maintained without inflammatory cell infiltration. The epidermis surrounding the catheter insertion site exhibited downward growth to the deeper dermis, with maintenance of normal skin structure. The cuff was tightly attached to the adjacent dermal connective tissue with marked foreign body reaction and fibrosis.
Conclusions: This method of washing, using a water shower without antiseptic agents, is effective in managing TCC exit sites. The skin at exit sites retained its normal histological structure, confirming the efficacy and safety of this catheter care method.

Su486 VASCULAR ACCESS LIFE AND CORONARY HEART DISEASE: ANY LINKAGE?

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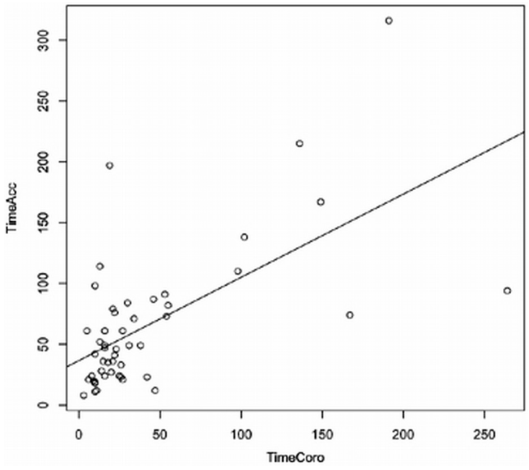
Introduction and Aims: Vascular access life represents and important issue in clinical practice, when a great effort is made in terms of time and costs.
Coronary artery disease is a frequent finding in dialysis patients, due to a lot of comorbidities.
The aim of this study was to explore a possible link between these vascular pathologies by finding any causative determinant.
Methods: Data from 50 hemodialysis patients (31 male, 19 female) were collected in a database related to coronary imaging and vascular access life time. Mean dialysis starting age was 59.8 years, 25% of patients were affected by diabetes. Mean since starting dialysis to coronary imaging was 41.22 months. Mean access life span was 64.8 months. We collected the number of coronary imaging examinations and 34 patients underwent 1 procedure, 12 patients underwent 12 procedures, and last 4 patients underwent 3 procedures. At the end of follow up time of 29 years 26 patients were still alive, while 24 patients were dead. Vascular access outcome resulted in 37 still functioning vascular access, and 13 closed access. Coronaropathy was found in 41 patients, in 12 patients only one coronary was implicated, while in 29 patients both coronaries were affected. PTH mean value at the time of imaging was 161.9 pg/ml. Descriptive statistical analysis and survival analysis by cox regression were performed with SPSS statistical software.

Abstract Su484 – Table 1

	Age (years)	HD vintage (years)	VA duration (years)	Adragao score+VA Ca-score	VA Ca-score	PWV	iPTH pg/ml
Group I 0.5-4.5 cm, N=24	61.3±10.4	8.7±5.1	7.7±4.6	4.4±3.5	1.00±1.06	9.68±2.76	247±278
Group II 5.0-9.5 cm, N=33	62.0±10.8	8.7±4.9	7.3±4.2	5.7±3.6	1.20±1.06	10.13±3.06	523±457
Group III >10 cm, N=40	62.7±9.2	10.6±5.3	9.3±5.1	4.4±3.3	0.91±1.02	9.43±1.80	530±700

Results: A strong correlation was found betweeen vascular access life time and the time to coronary imaging performing linear regression analysis (p< 1,268 e-06).



Cox analysis found that the life time of vascular access in dialysis patients, in a model that considered also patient survival, could possibly depend on number of coronary affected (p<0.08) and PTH values detected during imaging (p<0.048)

Conclusions: Dialysis patients are at risk of coronary heart disease and a low life span of vascular access with a worsening of dialysis treatment and patient life quality. Calcifications from uncontrolled secondary hyperparathyroidism could be a linkage factor between theese vascular diseases, pointing to a needing of a better comprehension of mechanisms that may affect all vascular district. These data could evidence a systemic disease threatening our patient's life.

Su487 THE SURVIVAL RATE OF NATIVE ARTERIOVENOUS FISTULA (AVF) AS VASCULAR ACCESS AND RISK FACTORS ON THE SURVIVAL

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Introduction and Aims: The maintenance of vascular access in hemodialysis (HD) patients is causing serious problems not only in terms of the quality of life of patients but also in the medical economy. In this regard, we investigated the cumulative patency rate of native arteriovenous fistula (AVF) as vascular access.

Methods: The cumulative patency rate of AVF was compared among the patients who have AVF ("AVF group"; 616 limbs). More particularly, the patency rates of AVF were investigated by different factors including primary diseases, age at the time of surgical operation, gender, condition and site. Furthermore, the influences of factors including primary disease, gender, age at the time of operation (/1 year old), and dialysis period (/1 year) on the patency rate were investigated using Cox's proportional hazard model.

Results: The patency rates of AVF in 1, 2, 3, 5 and 10 year were 85.6, 78.9, 75, 67.5 and 52.2%, respectively. According to the investigation by factors, the rates were significantly higher in males as to the gender factor. The investigation of the influences of risk factors on the patency rate showed that the significant and independent risk factors in all cases were "female" and "short dialysis duration (/year)."

Conclusions: The AVF is considered as the vascular access with the highest patency rate in the chronic HD patients. However, the gender factor influences this rate. That is, "female" has a higher risk in comparison with "male."

Su488 ACCURACY OF USING ONLINE CLEARANCE MONITOR TO MEASURE VASCULAR ACCESS FLOW IN HIGH-EFFICIENCY ON-LINE HEMODIAFILTRATION

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Introduction and Aims: The good vascular access flow is fundamental in attaining the high quality of hemodiafiltration (HDF). Measurement of the vascular access flow rate (Qa) is widely accepted as the best method for surveillance and predicting failure of the access. Among current practical methods, the ultrasound dilution technique (UDT) is standard, but this requires a costly device available in liminted hemodialysis (HD) centers. The two conductivity clearance values in normal and reversed positions of the blood lines were applied to determine the Qa and reported the accuracy in diffusive hemodialysis. However, this affordable technique has never been tested for high-efficiency HDF.

Methods: The present study was conducted to compare the values of Qa measured by Online Clearance Monitoring (OCM-Qa) in Fresenius 4008H hemodiafiltration machine with those determined by the standard UDT (UDT-Qa) in the high-efficiency pre-dilution on-line HDF (pre ol HDF).

Results: The values of Qa were determined in each of 17 patients who were treated with pre ol HDF by OCM technique and then, in the same session, by UDT using Transonic HD03 device. The values of standard UDT-Qa and OCM-Qa were 670.28±294.94 and 678.38±278.47 mL/min (NS). There was a significant correlation between the two techniques (r = 0.96, p<0.01). A Bland-Altman plot comparing the OCM-Qa and UDT-Qa was displayed in figure 1.

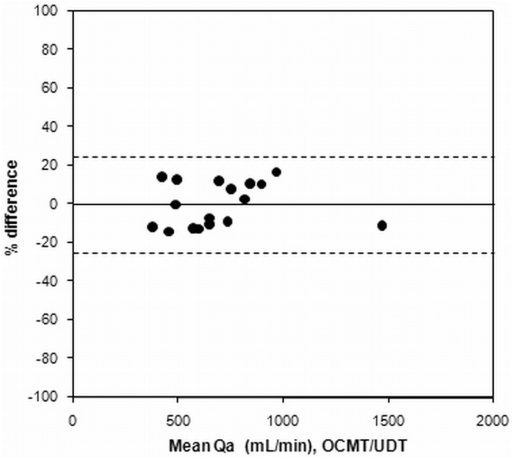

Figure 1. Bland-Altman OCMT/UDT. Qa = vascular access flow rate; OCMT = online clearance monitor technique; UDT = transonic HD03 ultrasound dilution technique.

Conclusions: The vascular access flow determined by OCM which is integrated mostly in current hemodiafiltration machine is highly accurate, easy to perform, and economical and can be used for vascular access surveillance in high-efficiency ol HDF.

Peritoneal dialysis 2

Su489 EFFECT OF ICODEXTRIN USE AT STARTING PD ON PERITONEAL PERMEABILITY

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Introduction and Aims: Peritoneal permeability differs between patients at starting peritoneal dialysis (PD) and it can increase along with time on the technique.

The aim of this study was to evaluate if the use of one exchange a day of

icodextrin from the time of DP initiation affects the evolution of peritoneal permeability.
Methods: 56 incident PD patients (mean age: 48,3±14,0; 62,5% males; 17,9%diabetics) that used one exchange a day with icodextrin from the time of starting PD. We performed a peritoneal transport kinetic study at the time of starting PD and then every 6 months during two years. We calculated the peritoneal mass transfer area coefficient of creatinine (Cr- MTAC) and urea (U-MTAC) as well as the D/P creatinine (D/P Cr). As a control group we used the results of Cr-MTAC of 249 patients that had used glucose as the only osmotic agent from the time of starting PD.
Results: The peritoneal transport, calculated using Cr-MTAC, U-MTAC and D/P Cr, diminished at 12 months (11,7±5,7 vs. 8,1±3,1; 23,5±7,3 vs. 18,9±3,8; 0,72±0,09 vs. 0,67±0,08; respectively), staying stable afterwards. We found that high transporters (HA) patients showed a higher decrease of Cr-MTAC along the first year of treatment. The diminution of Cr-MTAC after 12 months using icodextrin was significantly higher ($p<0,001$) that the one observed in the control group (10,5±5,3 vs. 10,1±4,6). High transport patients showed a higher decrease of Cr-MTAC along the first year of treatment that the others.
Conclusions: Icodextrin use at starting PD might help to correct the high transport status observed in some patients. The peritoneal transport kinetic studies performed at 6 and 12 months after starting PD are more representative of the long term peritoneal transport characteristics of the patients than those performed at starting PD.

Su490 CHANGES IN BODY FAT MASS IN PATIENTS AFTER STARTING PERITONEAL DIALYSIS

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Introduction and Aims: Peritoneal dialysis (PD) is characterized by the gain in fat mass. Visceral fat mass is associated with metabolic syndrome and atherosclerosis rather than subcutaneous fat mass. In addition, the change in visceral fat mass is more reliable predictor of survival in PD patients. In this study, we prospectively examined serial changes in fat composition and nutritional status, and analyzed factors associated with the gain in fat mass undergoing PD patients.
Methods: Body composition was assessed by bioelectric impedance analysis (BIA) and computed tomogram (CT). Nutrition status was assessed by subjective global assessment (SGA), means of protein equivalent of nitrogen appearance (nPNA), serum albumin, C-reactive protein (CRP) and lipid profile. All measurements except BIA were performed on the seventh day, 6 months and 12 months after the start of PD.
Results: Sixty patients (30 men), with a mean age 55.0±12.48 years, were enrolled. An increase in body weight continued during 12 months, but visceral and subcutaneous fat mass increased in first 6 months and decrease in second 6 months. While serum hematocrit and albumin decreased in first 6 months, they did not change in second 6 months. Serum creatinine, total cholesterol and triglyceride increased similar to the weight pattern. While nPNA decreased in 12 months, KT/V, SGA, and CRP didn't change. The patients who had more visceral fat mass at the start of PD had less gain of visceral fat mass in early 6 months (r = -0.821, p = 0.002). The patients who had more subcutaneous fat mass at the start of PD had less gain of subcutaneous fat mass in early 6 months (r = -0.709, p = 0.015). The change of weight was not associated with the change of visceral or subcutaneous fat in the first 6 months.
Conclusions: Patients starting PD have the increase of weight which includes visceral and subcutaneous fat in first 6 months. Patients with high baseline fat mass had less of an increase in fat mass than those with low baseline fat mass regardless visceral or subcutaneous fat mass.

Su491 POOR FINGER DEXTERITY BUT NOT ELDERLY AGE IS ASSOCIATED WITH INCREASE RISK OF PERITONITIS IN PATIENTS ON PERITONEAL DIALYSIS

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Introduction and Aims: More and more elderly patients requiring renal replacement therapy and whether they could self perform peritoneal dialysis (PD) exchange is the major concern. It is because PD related peritonitis is still the major cause for technique failure and mortality. Despite different educational programs, repeated training and technique assessment, the peritonitis rate has not been much reduced. Impairment of hand function (IHF) is plausible the cause for poor exchange technique. Herein, we prospective study the IHF and other factors, and its association with peritonitis risk in patients on long-term PD treatment.
Methods: All prevalent and incident PD patients were recruited except patients required helper to perform dialysis exchange. Single occupational therapist performed all hand function assessment and was blinded to patients' clinical information. IHF was defined by the power grip strength, tripod pinch, lateral pinch and finger dexterity (unimanual and bimanual). Nerve conduction test was performed to look for underlying peripheral neuropathy and carpal tunnel syndrome. Patients after assessment were follow-up for 2 years and censored for onset of peritonitis, transplant, switched to hemodialysis or death. Cox regression model and Kaplan Meier analysis were used to analyse the risk factors for peritonitis and peritonitis free survival, respectively.
Results: Total 152 (female=76) patients with age 57.6±12.5 yrs were recruited. There were 48 (32%) and 16 (11%) patients had DM and CVA, respectively. 81 (53.3%) and 72 (47.4%) patients were found to have PN and CTS, respectively. The average follow up time was 14.6±7.5 month. Throughout this period, 52 (34.2%) patients developed peritonitis. The peritonitis free survival at 12 month and 24 months were 74% and 61%, respectively. By multivariate analysis with the Cox regression model, only patients with CVA associated with a higher risk for peritonitis (RR 2.15, 95% confidence interval [CI], 0.95 to 4.92, p=0.06). Patients with age greater than 65 yrs (N= 51) did not have significantly increased risk (95% CI, 0.41 to 1.53, p=0.49). Whereas better finger dexterity conferred 12% protection (95% [CI], 0.79 to 0.99, p=0.03) from developing peritonitis. The interaction between the finger dexterity and CVA was not statistically significant (P=0.32). In Kaplan Meier analysis, patient with higher finger dexterity were also found to have higher peritonitis free survival (p=0.03). Other hand function parameters did not have effect on peritonitis risk.
Conclusions: PD patients with IHF and history of CVA may associate with a higher risk of peritonitis. Elderly age with good hand function seems not having increased risk at all. Hand function assessment is better to be performed before and throughout the PD treatment so that peritonitis risk could be better surveillance.

Su492 INFLUENCE OF ICODEXTRIN ON METABOLIC SYNDROME AND OTHER CARDIOVASCULAR RISKS ON PERITONEAL DIALYSIS

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Introduction and Aims: Metabolic syndrome (MeS) and each of its components are well-known risk factors for cardiovascular disease and diabetes mellitus. Some data suggest that the prevalence of metabolic syndrome is higher in patients undergoing peritoneal dialysis (PD). The purpose of this study was to analyze the changes of MeS incidence and the effects of icodextrin-based solutions (ICO) over glucose-based solutions (GLU) and metabolic managements of non-diabetic PD patients in continuous ambulatory peritoneal dialysis (CAPD).
Methods: We enrolled 87 non-diabetic PD patients between January 2009 and January 2010 (39 male 44,8%, 48 female 55,2% age 48,36±14,50) attending our PD center without current infections or chronic inflammatory diseases. We have performed a retrospective case-control study on the metabolic effects of ICO and GLU solutions over one year period. Forty-nine (56,3%) patients used ICO and 38 (43,7%) patients used GLU without

ICO for PD. Daily schedule of ICO group was 3x2 L GLU (2,27%) and 1x2 L ICO. Control group received 4x2 L GLU solutions (3*2,27% and 3,86% or 1,36%). Plasma lipids (total cholesterol, HDL-cholesterol and triglyceride), blood glucose, complete blood cell count, anthropometry, blood pressure, fasting glucose and insulin were measured and insulin sensitivity index by the homeostasis model assessment (HOMA-IR) were compared for two groups. MeS was defined in accordance with the National Cholesterol Education Program (Adult Treatment Panel III) criteria at the end of the 12 months.

Results: Groups were similar at baseline in all measured variables and demographic features. Overall prevalence of MeS was 42% (n=40). In these patients with MeS, 32 (80%) patients used GLU and 8 (20%) patients used ICO solution for PD. Laboratory and metabolic parameters are compared in table.

Parameter (serum)	ICO	GLU	P
Na (meq/L)	141±4	135±5	<0.001
Total cholesterol (mg/dl)	172±43	193±47	<0.005
LDL (IU/L)	108±27	137±35	<0.001
ALT (IU/L)	15±8	24±28	<0.05
TSH (IU/L)	2,3±1,7	1,6±1,0	=0.027
Insulin (IU/L)	4,8±2,8	10,5±5,3	=0.04
HOMA-IR	2,2±1,2	3,0±1,6	<0.01

Conclusions: ICO solution improved metabolic features of patients. PD with GLU solution results in MeS in CAPD. Increased ALT might be an indirect finding of fatty liver and deserves further studies.

Su493 EVALUATION OF CLINICAL OUTCOME IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: The purpose of this study was to evaluate the clinical outcome and to identify predictors of mortality in our peritoneal dialysis(PD) patients.

Methods: Three hundred twenty-two patients (pts) (171F, mean follow-up 36,7±26,9 months, mean age 44,8±15,9yrs) were started PD treatment between 2001-2010. PD treatment were withdrawn in 221 of them (group 1: drop-out) (114F, mean age 46,4±16,7yrs, mean follow-up 28,2±20 months). Remaining 101 pts (group 2: treatment) (57F, mean age 41,2±13,4yrs, mean follow-up 52,5±30,7 months) were still under follow-up. Thirty one pts had hemodialysis(HD) therapy before PD in group 1. Twenty-three pts had HD history in group 2.

Results: There was no statistically significant difference between both of groups at the begining and the last visit of PD, in regard to Kt/V urea, creatinine clearance, serum creatinine and Hb.

Table 1. Blood pressures, infectious and biochemical datas

	Group 1	Group2	P
PTH (start of PD) (pg/dl)	318±347	332±283	NS
Albumin (start of PD) (g/dl)	3,5±0,6	3,7±0,5	0,002
PTH (the last) (pg/dl)	386±393	537±465	0,006
Albumine (the last) (g/dl)	3,4±0,6	3,6±0,4	NS
Peritonitis incidence (month)	19,3±16	38,2±28,3	<0.001
Exit site inf.(month)	25,2±19	40,9±30,6	<0.001
Duration of HD (month)	45,3±41,3	16,8±21,5	0.006
CTI (The last)	0,48±0,05	0,46±0,05	0,002
SBP (mmHg) (the last)	109±27	116±27	0,03
DBP (mmHg) (the last)	68±16	73±17	0,02
Urine (ml/day) (the last)	125±251	166±305	NS
Ultrafiltration (ml/day) (the last)	1092±514	1183±576	NS

The most frequent etiologies were chronic glomerulonephritis(27,7%), diabetic nephropathy(22,1%) and unknown(35,5%). The causes of drop-out were death (37,7%), transfer to HD(37,2%), transplantation(18,3%) and lost to follow-up(6,8%). The most frequent causes death were peritonitis and/or sepsis (43,1%), cardiac events (37,5%) and unknown causes(12,5%). Whereas transfer to HD were due to peritonitis and/or sepsis(62%) and to insufficient PD(28,2%).

We found positive correlation between mortality and age, presence of HD history, mandatory PD due to vascular access problems, PD done by another person, pretreatment and last CTI (for all parameters p: <0.001; r: 0.51, 0.38, 0.48, 0.35, 0.35 and 0.29, respectively). Negative correlation was found between mortality and pretreatment urine volume, pretreatment PTH, pretreatment albumin, peritonitis, exit site infection, and Kt/Vurea (p: 0.04, 0.01, <0.001, 0.01, 0.02, 0.04; r: -0.16, -0.18, -0.29, -0.17, -0.16, -0.21, respectively). We found negative correlation between mortality and albumin, PTH, systolic and diastolic BP in the last follow-up (p: <0.001, 0.002, 0.02, 0.02 and r:-0.36, -0.24, -0.17, -0.18 respectively).

Conclusions: In conclucion: Peritonitis and cardiovascular events were the most important causes of death in PD patients. History of hemodialysis, presence of urine and infectious complications affect mortality. The presence of hypoparathyroidemia, hypoalbuminemia and development of hypotension were associated with mortality.

Su494 BENFOTIAMINE PROTECTS HUMAN PERITONEAL MESOTHELIAL CELLS AGAINST ADVANCED GLYCATION END-PRODUCT MEDIATED DAMAGE IN PERITONEAL DIALYSIS

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Introduction and Aims: Morbidity and mortality of peritoneal dialysis patients are significantly influenced by the integrity of the peritoneal membrane. High glucose and advanced glycation end-products (AGE) were suggested as contributing factors for local peritoneal damage. Previous studies demonstrated the ability of the lipid-soluble thiamine derivative benfotiamine to decrease high glucose induced tissue damage by an activation of transketolase and antioxidative effects.

Methods: Human peritoneal mesothelial cells (HMPC) were isolated from human omenta and incubated with low (1.5%) and high (3.9%) glucose containing peritoneal dialysis fluids (PDF) either +/- benfotiamine substitution for 48 h.

Expression of transketolase was assessed using immunofluorescence technique. AGE mediated damage was analyzed by immunofluorescence and western blot technique regarding the expression of the receptor for AGE (RAGE), epithelial to mesenchymal transition (EMT), as well as the organization of tight junctions and the cytoskeletal organization. For quantitation a semiquantitative score was used. Finally, the inflammatory status was analyzed by ELISA technique. Data are shown as mean ± standard deviation.

Results: After incubation with PDF, HPMC revealed a decreased level of transketolase, higher expression of RAGE and vimentin as a marker of EMT. Moreover there was a higher percentage of HPMC exhibiting a reorganized actin cytoskeleton.

When cells were incubated with benfotiamine and PDF, transketolase expression was higher in the low glucose group in comparison to the low glucose PDF group without benfotiamine incubation (PDF 1.5: 1.30±0.15 vs. PDF 1.5+Benfotiamine: 1.61±0.22, p<0.05).

RAGE expression was significantly lower when incubated with benfotiamine (PDF 1.5: 1.65±0.11 vs. PDF 1.5+Benfotiamine: 1.37±0.14, p<0.001; PDF 3.9: 1.71±0.16 vs. PDF 3.9+Benfotiamine: 1.49±0.14, p<0.001), EMT, shown by vimentin expression, was drastically reduced (PDF 1.5: 1.78±0.37 vs. PDF 1.5+Benfotiamine: 1.60±0.40, p<0.05; PDF 3.9: 2.15±0.57 vs. PDF 3.9+Benfotiamine: 1.69±0.64, p<0.05). Moreover it was noted that benfotiamine could not stabilize tight junction protein zonula occludens-1, whereas the actin cytoskeleton was reorganized in a less percentage of cells when HPMC were incubated with benfotiamine (PDF 1.5 (%): 26.3±13.1 vs. PDF 1.5+Benfotiamine (%): 17.5±7.54, p<0.01; PDF 3.9 (%): 28.5±9.75 vs. PDF 3.9+Benfotiamine (%): 24.8±11.8). Finally, interleukin-6 release was reduced by incubation with benfotiamine (PDF 3.9 [ng/ml]: 6.14±2.91 vs. PDF 3.9+Benfotiamine [ng/ml]: 3.57±2.19).

Conclusions: Our findings suggest that benfotiamine mediates activation of transketolase and thereby provides specific protective effects to HMPC that affect RAGE expression, EMT, cytoskeletal organization as well as the inflammatory status during AGE mediated damage in PD.

Disclosure: This study was supported by a grant from Wörwag Pharma.

Su495 INCREMENTAL PERITONEAL DIALYSIS FAVOURABLY COMPARES WITH HEMODIALYSIS PRIOR TO RENAL TRANPLANTATION

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Introduction and Aims: The feasibility of incremental peritoneal dialysis (PD) as a first choice renal replacement therapy (RRT) and the good clinical outcome it offers to well-motivated stage 5 CKD patients has been repeatedly reported. This strategy involves timely start of PD with a low dose, gradually increased afterwards to compensate ongoing individual residual renal function (RRF) loss to meet total (peritoneal plus residual renal) recommended small solutes clearances adequacy targets. To date its value as a bridge to renal transplantation has not been specifically addressed.
Methods: Stage 5 CKD patients with at least 1 year predialysis follow up, starting RRT with incremental PD or hemodialysis (HD) under our care and subsequently receiving their first renal transplant (Tx) were included in this observational cohort study. Age, gender, underlying nephropathy, residual renal function (RRF) and RRF loss rate before dialysis, comorbidities, RRT schedules and adequacy, dialysis-related morbidity, Tx waiting time, RRF at Tx, incidence of delayed graft function (DGF), in-hospital stay for Tx, serum creatinine at discharge and one year later were registered. Data were collected and summarized as means ±SD and proportions; continous variables were compared with the Student-t test for independent samples and categorical ones with the chi-square test between patients on incremental PD or HD before Tx.
Results: Seventeen patients on incremental PD and 24 on HD received a renal Tx after 28±13 and 32±17 months of RRT, respectively, p=0,2; all grafts were from deceased donors, except two from living related donors in 2 patients on HD. Age (37±13, median 39, range 19-68 years vs 43±14, median 41, range 20-71), underlying nephropathy and RRF loss rate in predialysis (-0,97±0,34 vs -1±0,48 ml/min/month), RRF at the start of RRT (6,97±1,1 vs 6,81±1,5 ml/min) and comorbidities did not differ significantly. DGF with need of dialysis after Tx occurred in 12 patients (25%), 1 on incremental PD and 11 on HD (χ^2=8,57 p=0,005, RR 1,73 95% CI: 1,18-2,55, p=0,005). In hospital stay lasted 19±4 vs 22±6 days, p=0,08. Serum creatinine at discharge and 1 year later were significantly higher in patients who had been on HD (2,1±0,9 vs 1,3±0,3 mg/dl and 1,96±0,9 vs 1,14±0,3 respectively, both p=0,001). At last follow up, 3 HD treated patients had returned to dialysis because of graft failure after 15, 28 and 43 months, respectively.
Conclusions: In patients receiving their first renal Tx, previous incremental PD was associated with low morbidity, excellent preservation of RRF and significantly better immediate and 1-year graft function than that observed in patients previously treated with HD. According to these results, we believe that the incremental PD option should be offered to every suitable stage 5 CKD patient who appears to be a good candidate for renal Tx. We are committed to improve the early outcome of Tx in our patients on HD.

Su496 IMPORTANCE OF RESIDUAL RENAL VOLUME IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Residual renal function(RRF) at the initiation of peritoneal dialysis(PD)therapy can predict patient outcome. This study was performed to compare the impact of baseline RRF on patient and technique and patient survival in PD patients.
Methods: Two hundred two PD patients have been evaluated retrospectively depending on urine output at the begining of the PD therapy. Anuric patients(urine-output<100ml/24hours) and non-anuric patients(urine output≥100ml/24hours) were compared according to demographic, clinical and biochemical parameters, peritonitis incidence and mortality rates.
Results: Fifty-eight patients were anuric(38F, mean age 42,8±14,9 yrs, mean follow-up period 44,2±35 months) at the beginning of dialysis. Fourty-two of these patients received CAPD and 16 APD. Twenty-seven of 58 anuric patients had HD therapy before PD. One hundred fourty-four were non-anuric(68F,mean age 43,7±14,5 yrs, mean follow-up period 39,6±26,1 months, mean urine volume 592±442 ml/day). In this group 103 patients received CAPD and 41APD. Twenty-three patients had received HD therapy. Sixty-five patients(45CAPD and 20 APD) became anuric in the following 22,5±19,6 months. No differences were noted regarding age, follow-up periods, presence of diabetes, UF volumes, albumin, Hb, P, CaxP, PTH and KtV levels. Significant differences were noted regarding systolic and diastolic blood pressure at the beginning, and gender, prior hemodialysis duration, CRP, Ca, ferritin levels between the two groups. Peritonitis rate was one episode per 28.2 vs. 30 patient-months in anuric and non-anuric groups. However there was no significant difference regarding peritonitis rate between the two groups.

Table 1. Comparison of anuric and non-anuric peritoneal dialysis patients

	Anuric patients (n=58)	Non-anuric patients (n=144)	p
Mean follow up (month)	44,2±35,0	39,6±26,1	NS
HD duration (month)	19,9±35,9	1,3±19,9	<0.05
UF volume (ml)	1053±408	1000±459	NS
Albumin (g/dl)	3,69±0.58	3,67±0,63	NS
CRP	21,5±31,9	18,7±30,2	<0.05
Hb (g/dl)	11,4±2,5	10,8±1,7	NS
Ca (mg/dl)	9,1±0,7	8,9±0,9	<0.05
P (mg/dl)	4,5±1,5	4,7±1,5	NS
PTH (pg/ml)	343±363	447±419	NS
Ferritin (ng/ml)	535±529	298±356	<0.05
Systolic BP (mmHg)	105,7±27,3	120,6±25,8	<0.05
Diastolic BP (mmHg)	67,9±16,1	78,5±15,9	<0.05
Peritonitis rate	1/28,2	1/30	NS

The two group had similar technical survival rate (87,9% and 86,1% anuric and non-anuric patients retrospectively) (p: 0.75). Although there was statistically significant difference in the mortality rate based on residual renal function (74,1% and 92,4% anuric and non-anuric patients retrospectively) (p: 0.002).
Conclusions: In conclusion;RRF is important for patient survival in PD patients.The presence of RRF had positive impact on PD patient mortality. It seems that patients who had tight blood pressure control become more anuric. Even though residual renal function is known to improve quality of life and complication rates in PD patients, no significant differences were found regarding peritonitis rates between anuric and non-anuric patients in our study.

Su497 PODOPLANIN POSITIVE CELLS ARE A HALLMARK OF ENCAPSULATING PERITONEAL SCLEROSIS

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Introduction and Aims: Encapsulating peritoneal sclerosis (EPS) is an uncommon but potentially life threatening complication of peritoneal dialysis (PD). The key factors for the diagnosis of EPS are clinical symptoms. Unfortunately, the morphological picture is not specific, and histological markers differentiating between simple sclerosis and EPS are not available at the moment.
Aim of the study was the description of a specific morphological pattern in EPS for differentiating between simple sclerosis and EPS.
Methods: Peritoneal biopsies were formalin-fixed, and paraffin-embedded (n= 69). The study population consisted of biopsies of the visceral peritoneum from patients on peritoneal dialysis (n= 16), and patients on peritoneal dialysis with clinical signs of EPS (n=18). The control biopsies were taken either at the time of hernia repair (n=15), or at the time of appendectomy (n=20).

Immunohistochemistry was performed to localize podoplanin. Additionally, markers of endothelial cells (anti CD31), mesothelial cells (anti calretinin), myofibroblasts (smooth muscle actin), proliferating cells (anti KI-67) and double immunofluorescence for D2-40 and smooth muscle actin were performed on selected biopsies.
Results: Defining the pattern as an area of podoplanin positive cells embedded in a fibrotic matrix it was present in 15 out of 18 biopsies in patients with EPS, in 3 out of 16 patients on PD without signs of EPS, but in none of the 35 controls.
In this retrospectively collected biopsies the staining pattern was significantly associated with EPS (p<0,0001). It resulted in a sensitivity of 0.83 (95% confidence interval 0,59 to 0,96) and a specificity of 0.94 (95% confidence interval 0,84 to 0.99). The positive predictive value was 0,83 (95% confidence interval 0,5857 to 0,9642) and the negative predictive value 0,94 (95% confidence interval 0,8377 to 0,9877). The likelihood ratio was 14,2.
Conclusions: In summary we describe a new marker involved in patients with EPS. Podoplanin might turn out to be useful in routine practice as well as a driving force of this devastating condition. We formulated several new hypothesis which can now be tested in vitro and in vivo.

Su498 EFFECTS OF BicaVera® (Bv) DIALYSATE FOR PERITONEAL DIALYSIS ON THE EPITHELIAL-TO-MESENCHYMAL TRANSITION (EMT) OF THE MESOTHELIAL CELL (MC) *EX VIVO*

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Introduction and Aims: Bv has less impact on EMT of MC *in vitro*. We have hypothesized that Bv may also reduce the manifestations of EMT of MC from peritoneal effluents grown *ex vivo*.
Methods: After being randomly selected, patients were treated with conventional PD fluid (C; n=20) or BicaVera® (BV; n=11; both solutions from Fresenius Medical Care on CAPD. C vs. BV follow up: baseline 20/11, 6 months 20/11, 12 months 18/11, 18 months 11/11, 24 months 3/5. Small solute and water transport capacities were determined during a 4-hour dwell (4.25% glucose dialysate). MCs from overnight effluent were seeded and grown *ex vivo* until confluence was reached. VEGF, IL-8 and TGF-β levels in the supernatant (pg/mg cell protein) and fibronectin and procollagen levels (ng/mg cell protein) in cell extracts were measured by ELISA. ICAM-1 expression was used as a marker for mesothelial nature of cells. EMT of MCs was defined by non- epitheloid morphology and higher content of fibronectin.
Results: BV was associated with increased small solute transport (p<0.0001) and lower initial UF (p<0.0001). Furthermore, Bv use was associated with a trend to better preservation of residual renal function (only the first semester). The frequency of non- epitheloid phenotype at long-term (18-24 month) was significantly lower in BV (p<0.0001). Non-epitheloid phenotype was associated with higher VEGF (p<0.04) and fibronectin (p<0.001) levels. C but not BV showed a progressive increase of VEGF (p<0,001) and IL-8 (p<0.0001) over time. MC-EMT parameter (phenotype) and IL-8 levels were favorably different for BV fluid relative to C at medium-term. There was no difference in procollagen, fibronectin and TGF-β levels between groups.
Conclusions: BicaVera® showed better biocompatibility than high-GDPs fluid *ex vivo* in terms of EMT of MCs, consistent with data obtained previously *in vitro*. Further studies including more patients are required to confirm these results.

Su499 EFFECT OF L-CARNITINE ADMINISTRATION ON SERUM ALBUMIN LEVEL IN CAPD PATIENTS IN THE ALZAHRA MEDICAL CENTER OF ISFAHAN

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Introduction and Aims: Malnutrition is a frequent complication among patients with chronic dialysis; on the other hand inflammation in patients with chronic dialysis caused synthesis and release of CRP, hemocystein, and decrease albumin that may be considered an independent predictor for cardiovascular disease, the most important factors for mortality and morbidity in dialysis patients. Recent studies have suggested that L-carnitine have useful effect on serum albumin and inflammatory factors, they showed increased serum albumin in the dialysis patients. The aim of our study is effect of oral L-carnitine on serum albumin, CRP, hemocystein, lipids profile and hemoglobin in CAPD patients.
Methods: This was a double blind – placebo controlled randomized clinical trial, that was done in the alzahra medical center of Isfahan.
Forty eight patients on chronic CAPD were randomly assigned to receive oral L-carnitine 750 mg/day or placebo for 9 months. Serum albumin was measured at baseline and after 3, 6, 9 months; CRP, hemocystein, lipids profile, hemoglobin were measured at baseline and after 9 months in two groups.
Results: Mean serum albumin in the drug group showed significant increase after 9 months (3.3±0.51 to 3.7±0.53, p value→ 0.003), while decrease in placebo group. Rate of positive CRP and hemocystein level tests significantly was decreased in drug group; also HDL-Cholestrol increased significantly in drug group. In addition, during the study 50% of the patient's drug group showed hemoglobin level increases of more than 1 gr/dl.
Conclusions: It seems that oral L-carnitine administration have positive effect on serum albumin, inflammatory factors, lipids profile and hemoglobin level in CAPD patients.

Su500 PHOSPHATE REMOVAL IN PERITONEAL DIALYSIS REGIMENS: AN UNDERESTIMATED ADEQUACY PARAMETER

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Introduction and Aims: Phosphate (Ph) control has shown a major impact on morbidity and mortality of dialysis patients, while KT/V urea, within the ranges usually achieved, often fail to independently impact on patient survival. However, phosphate handling by peritoneal dialysis (PD) is scarcely addressed in clinical investigation and patient management.
The aim of the study was to investigate peritoneal Ph clearances and its relationship with standard small solute and ultrafiltration adequacy targets, and with automated PD (APD) regimens.
Methods: 43 prevalent adult PD patients were cross-sectionally evaluated: aged 54±16 years old, on PD treatment for 35±27 (range 2-120) months, 26% on PD due to vascular access failure, 44% male, 14% diabetic, 30% anuric, 28% with Davies comorbidity score ≥ 2, 53.5% on APD.
Adequacy parameters including fluid removal, KT/V urea, creatinine clearance and normalized protein catabolic rate (nPCR) were calculated by using Adequest® software. Peritoneal phosphate clearances were simultaneously

calculated from 24 hour urine and peritoneal effluent samples. Patients were categorized according to serum Ph cut-off of 5.5 mg/dl.

Results: Whole population serum phosphate was 4.9±1.3 mg/dL; 60.5% used sevelamer; 30.2% used cinacalcet. Their adequacy targets were: net ultrafiltration (UF)/day 814±337 mL, week KT/Vurea 2.4±1.1, week creatinine clearance L/1.73 m^2 73±25.8, 4 h PET D/P creatinine 0.70±0.10; serum albumin g/dL 3.7±0.46, nPCR g/kg/day 1.1±0.38.

Total daily phosphate removal (urine plus peritoneal) averaged 461±288 mg (range 166-1514) while peritoneal dialysis phosphate removal averaged 260±96 mg (range 77-675) and was correlated with peritoneal creatinine clearance (R=0.41, P =0.011) but not with peritoneal KT/V urea nor with UF.

Estimated percentage of phosphate removal by diffusion was 95±2.6% and by convection was only 4.7±2.6%.

Thirty-tree percent of patients were hyperphosphatemic (27% in spite of adequate (>1.7) KT/V urea): statistically significant differences in categories and dose of phosphate quelants, body surface area (BSA), serum albumin, nPCR, residual renal function(RRF), and standard adequacy targets were not found between hyper and normophosphatemic groups. However hyperphosphatemic patients presented significantly lower number of PD cycles on APD (median 5 vs. 7, P = 0.008), lower cycle dwell time (median 56 vs. 60 min, P= 0.037) and lower phosphate peritoneal clearance (median 33.9 vs. 42.6 L/week, P = 0.031).

Conclusions: Hyperphosphatemia remains a clinically relevant complication in spite of achieved standard adequacy parameters. Peritoneal transport of phosphate is mainly diffusive and can be optimized by increasing the number of APD cycles and cycle dwell time. Phosphate peritoneal clearance should be taken into account as a potentially modifiable parameter of phosphate control in PD regimens and an additional adequacy target.

Su501 MARKERS OF MUSCULAR NECROSIS IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Asymptomatic elevated creatinephosphokinase (CK) levels occur in about 10 to 50% of hemodialysis patients. Possible causes for this elevation are the presence of subclinical myopathies, namely uremic myopathy, left ventricular hypertrophy and pharmacological toxicity by statins. Similarly, myoglobin levels are commonly elevated in hemodialysis patients, causing difficulty in the diagnosis of myocardial ischemia. The elevation of muscular necrosis markers in peritoneal dialysis (PD) patients is less well described. The objective of this work was to evaluate the presence of elevated levels of muscular necrosis markers in our population of PD patients, its clinical significance and relation with the type of PD technique, PD delivered dose, type of peritoneal transport, residual renal function and the use of statins.

Methods: The authors did a retrospective transversal analysis of demographic data, levels of CK, myoglobin and aldolase, D/P Creat, Kt/V, residual renal function and PD technique (automatic vs continous ambulatory) in a population of 54 PD patients. Statistical analysis was performed using t-test and one way-ANOVA for categorical variables and Spearman and Pearson correlations for continuous variables. Statistical significance was considered for $p<0,05$.

Results: Of the 54 patients analysed, 30 were male and the mean age was 52,4 years (21 to 98 years). They were on PD for 18,5 months in average. 19 patients were on automatic PD (APD) and 35 on continuous ambulatory PD (CAPD). 64,8% of the patients were on statins. Only 3 of the patients had complaints of muscle weakness. CK levels were found to be elevated in 33%, aldolase in 20,4% and myoglobin in 85,2% of patients. We found a positive and direct correlation between CK levels and age (p=0,04); no correlation was found between gender and enzyme levels. There was no correlation between the use of statins and the magnitude of muscle markers elevation. We found that APD patients had lower levels of CK (p=0,05) and aldolase (p=0,01) than CAPD patients. No correlation was found between type of peritoneal transport and residual renal function and the levels of muscle enzymes. We found an inverse correlation between myoglobin levels and Kt/V (p=0,02); a similar correlation was not found for the other enzymes.

Conclusions: In conclusion, the elevation of muscle enzyme levels is common in PD patients, is mostly asymptomatic and does not seem to be related to the use of statins. A possible relation of dialysis dose and enzyme levels was demonstrated. Given that a relation with classical mechanisms involved in muscle markers elevation was not found in this set of patients, we hypothesize that this chronic asymptomatic elevation of muscular necrosis markers in PD patients could in be in relation with subclinical uremic myopathy.

Su502 SELF LOCATING PERITONEAL DIALYSIS CATHETERS – 1 YEARS EXPERIENCE

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Introduction and Aims: In September 2008 we changed from inserting coiled Tenckoff catheters (TC) to the Gambro self -locating catheter (SLC) in an effort to overcome the problem of catheter dislocation.

The aim was to establish whether the SLC was superior to the TC with respect to dislocations, malfunction, peritonitis rates and catheter survival as has been previously documented[1]. To demonstrate that dialysis adequacy is not inferior with the SLC compared with the TC[2].

Methods: A retrospective review of all SLC insertions from September 2008 to September 2009, and all TC insertions in the year prior to this. Fisher's exact test or the unpaired t test, were used to establish the statistical significance of each outcome.

Results: In the first year of inserting the SLC 56 were placed, 41 have 6 months follow up. In the year prior to this 50 TC were inserted; 5 were removed prior to 6 month follow up for reasons other than failure of dialysis or infection (e.g. transplantation) these were excluded from analysis.[table1First adequacy assessment data from time of catheter insertion was present for 30 SLC and 31 TC. The duration from insertion to assessment varied from 1 to 10 months.

Table 1. 6 month outcomes

	SLC (n=41)	TC (n=45)	P value
Catheter survival	34 (83%)	33 (73%)	0.3
Peritonitis rates	7 (17.1%)	8 (17.8%)	1.0
Radiology manipulations	1 (2.4%)	8 (17.8%)	0.03
Surgical repositioning	4 (9.8%)	3 (6.7%)	0.7

Table 2. Dialysis adequacy and modality

	SLC (n=30)	TC (n=31)	P value
Average KT/V	2.59	2.42	1.0
Creatinine Clearance (l/wk/1.73m^2)	109.6	91.8	1.0
APD	8 (27%)	5 (16%)	0.36
CAPD	22 (73%)	26 (84%)	0.36

Conclusions: The SLC group had a significantly lower number of radiological manipulations compared with the TC group due to fewer catheter dislocations. On all other outcomes there was no significant difference after 6 months follow up.

References:

1. Di Paolo N. et al. The self locating catheter: clinical experience and follow-up. Peritoneal Dialysis International 2004; 24:359-364
2. Di Paolo N. et al. A new self-locating peritoneal catheter. Peritoneal Dialysis International 1996; 16:623-627

Su503 DEVELOPMENT OF A EUROPEAN ENCAPSULATING PERITONEAL SCLEROSIS REGISTRY

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Introduction and Aims: Encapsulating Peritoneal Sclerosis (EPS) is a rare but serious complication of peritoneal dialysis (PD). The causes are uncertain but risk factors include PD duration and stopping PD (transfer to

haemodialysis or transplantation). The overall incidence in Europe is still ill-defined and there are concerns over different used diagnostic criteria for EPS. Because numbers in individual centres are limited, research in single centres is unlikely to assist in improving understanding in this rare condition. This project aimed to develop a European Registry for EPS to act as a focus for greater understanding of epidemiology, pathophysiology and in the future for potential interventions.
Methods: The Registry database was built collaboratively by nephrologists and hosted in an academic institution with longstanding experience in this area. The diagnosis of EPS was defined by existing ISPD criteria, but also linked to a probability ranking to allow review of likelihood by expert physicians. The online database captures demographic data, CKD and RRT history, details of PD therapy (type, solutions, etc), membrane transport and adequacy, peritonitis history, treatment and outcomes from EPS itself. In addition, a future catalogue function allows identification of radiological images, photographs, histology and whether PD fluid, genetic or biopsy material is stored for each case.
Results: The online database has been used for 8 months now in a single European country and 29 historical or new suspected EPS cases have been identified, demonstrating that such an approach is feasible. It is now planned to widen the capture of cases by linking with existing country based EPS and PD Registries. It will also be possible to access and include individual cases at www.epsregistry.eu.
Conclusions: EPS is an important complication of PD therapy and collaborative approaches across Europe are essential to improve both clinical knowledge around diagnosis and investigations as well as research into causes and treatment. The European EPS Registry has been established as an online database and catalogue of images/biological material to aid physicians and researchers to gain greater understanding by working collaboratively across countries.

Su504 BASELINE PERITONEAL ALBUMIN LOSS IS NOT A SURVIVAL FACTOR IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Baseline fast peritoneal membrane status has been associated with local and systematic inflammation markers. However data that correlate this specific status with mortality are contradictory. In some studies peritoneal clearance of protein-calculated from dialysate protein losses–has been correlated with small size molecule transport and survival. Our aim was to test the hypothesis that peritoneal loss of albumin is a risk factor of death in PD patients.
Methods: Single-centre, prospective cohort study which included incident PD patients during the last 15 years. Demographic and laboratory data were collected at the initiation of PD. Baseline SPA (standard peritoneal permeability analysis) was performed within the 6 first months of PD therapy. Mass transfer area coefficient (MTAC) of creatinine and peritoneal clearances of albumin (Cl_{alb}), b_2 microglobulin (Cl_{b2m}), a_2 macroglobulin (Cl_{a2m}) and immunoglobulin G (Cl_{IgG}) were calculated. The total amount of albumin loss in the dialysate during the 4-hour SPA was calculated. Overall mortality was studied with an intention-to-treat analysis; the event was death.
Results: 258 patients were included (median age 53 years (range 18-83)-56% male). The mean amount of peritoneal albumin loss was 0.83±0.02g/L/4hours and was correlated with MTAC of creatinine (r=0.4, p<0.001) and clearances of serum proteins (Cl_{b2m}, Cl_{a2m} and Cl_{IgG}). Albumin loss was not correlated with baseline serum CRP. Fast transporters (patients with MTAC of creatinine >13.16 ml/min/1.73m^2) were compared to the other groups. They had lower serum albumin (31.4 vs 34.6g/dl, t=4.2, p<0.001), lower transcapillary ultrafiltation rate (2.9 vs 3.3ml/min, t=1.9, p=0.05), higher Cl_{alb} (130 vs 85µl/min, t=8.4, p<0.001) and higher peritoneal albumin losses (1.07 vs 0.75 g/L, t=6.2, p<0.001).
At the end of follow-up, 130 deaths were recorded. The median survival time was 69 months. Patients with baseline serum CRP>10mg/L had a greater risk of death (logrank test, p=0.01). In Cox regression analysis age (HR 1.06; 95% CI 1.04-1.09), baseline serum albumin (HR 0.9; 95% CI 0.84-0.97) and MTAC of creatinine (HR 0.88; 95% CI 0.78-0.99) were significant risk factors of mortality. However, clearance of albumin and peritoneal albumin loss were not significant predictors of death (HR 0.45; 95%CI 0.11-1.85), p=0.27).
Conclusions: In this cohort, survival is related to systematic inflammation markers, like baseline serum albumin and CRP. On the other hand, peritoneal loss of albumin is not a survival determinant.

Su505 THE RISK OF ENCAPSULATING PERITONEAL SCLEROSIS AFTER THE DEVELOPMENT OF ULTRAFILTRATION FAILURE

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Introduction and Aims: Encapsulating peritoneal sclerosis (EPS) is a severe complication of PD. Almost all EPS patients have ultrafiltration failure (UFF). However, not all patients with UFF develop EPS. The aim of this study was to investigate the risk of EPS after the development of UFF.
Methods: We included all patients who developed UFF after at least 2 years of PD. UFF in the first 2 years of PD is associated with comorbidity, mesothelial cell mass and high lymphatic absorption rate. UFF after 2 years of PD is associated with a decreased osmotic conductance to glucose and associated with EPS. UFF was defined as a net UF of less than 400mL/4H with a 3.86% glucose dwell.
Results: Between July 1995 and December 2008, 417 adult patients were treated with PD in our center. 224 of these patients had PD for more than two years. 64 of them patients developed UFF (29%) and 10 of the UFF patients developed EPS (15%). EPS patients stayed on PD for a longer period, after the development of UFF and had a longer total PD duration ($p \leq 0.05$). No differences were present for age at the start of PD, time of UFF onset, comorbidity, outcome after three years of PD with UFF and causes of death between EPS patients and controls. The interval between the diagnosis of UFF and the development of EPS or discontinuation of PD is given in the Table. 50% of the patients who stopped PD after more than three years of UFF developed EPS.

Years of PD with UFF until the discontinuation of PD	0-1	1-2	2-3	>3
EPS	4	1	4	2
Controls	27	19	6	2
EPS as proportion of totals with UFF	13%	5%	40%	50%

Conclusions: From our results it appears that discontinuation of PD should be considered within 2 years after the development of UFF.

Su506 CHANGES IN LEPTIN AND ADIPONECTINE IN PATIENTS AFTER STARTING PERITONEAL DIALYSIS

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Introduction and Aims: Leptin and adiponectin are adipokine with respective pro-atherogenic and anti-atherogenic properties. Peritoneal dialysis (PD) is characterized by the gain in fat mass. In this study, we prospectively elucidated serial changes in leptin, adiponectin and fat composition in patients undergoing PD.
Methods: Visceral and subcutaneous fat mass on umbilical level were measured by computed tomogram (CT). Nutrition status was assessed by means of protein equivalent of nitrogen appearance (nPNA), serum albumin, C-reactive protein (CRP) and lipid profile. All measurements were performed on the seventh day, 6 months and 12 months after the start of PD.
Results: Thirty six patients (17 men), with a mean age 54.9±12.5 years, were enrolled. Plasma leptin and adiponectin level on baseline were 247.2±66.9 pg/mL and 266.2±29.8 ng/mL, respectively. Leptin (383.3±116.6 vs. 95.1±29.6 pg/mL, p=0.029) and adiponectin (330.9±40.9 vs. 193.9±37.3 ng/mL, p=0.019) in women was greater than in men on baseline. Leptin in elder (age >65 years) was greater than younger (482.1±205.5 vs. 168.9±52.4 pg/mL, p=0.041). But, diabetes mellitus didn't affect these levels. Initial leptin was associated with albumin ($r = 0.480$, $p = 0.027$), sub-

cutaneous fat (r=0.772, p=0.000), BMI (r=0.571, p=0.007), and adiponectin (r=-0.436, p=0.030) after the adjustment for sex and age. Initial adiponectin was associated with albumin (r =-0.618, p=0.002), and subcutaneous fat (r=-0.535, p=0.010) after the adjustment for sex and age. While body weight and visceral fat mass didn't increase significantly, subcutaneous fat mass increased continuously in 12 months. Serum albumin, triglyceride and CRP didn't change. KT/V and n PNA didn't change. Although there is no statistical significance, leptin increased and adiponectin decreased in 12 months.

Conclusions: Patients starting PD have the gain in subcutaneous fat mass in 12 months. Leptin and adiponectin may be positively and negatively associated with gain of subcutaneous fat mass in patients undergoing PD.

Su507 PREVENTION OF PERITONITIS IN NEWLY PLACED PERITONEAL DIALYSIS CATHETERS: EFFICACY OF PARENTERAL VERSUS ORAL PROPHYLAXIS WITH CEFUROXIME AXETIL

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Introduction and Aims: Antibiotic prophylaxis has been employed to reduce the risk of peritonitis in newly placed peritoneal dialysis (PD) catheters. The development of bacterial resistance and increased cost are raising serious concerns and modification of the antibiotic regimens is usually required. In this study we aim that compare the efficacy of parenteral versus oral prophylactic cefuroxime axetil for preventing peritonitis after newly placed PD catheters.

Methods: Between January 2008- December 2009, 37 patients (F/M: 17/20; mean age: 44.4±14.1 years) undergoing 40 percutaneous PD catheter placement procedures were included to the study, retrospectively. Twenty-five patients (parenteral group) were administered a single intravenous (IV) 750 mg dose of Cefuroxime axetil, approximately 30 minutes before placement. Fifteen patients (oral group) received 500 mg dose of oral cefuroxime axetil 2 hours before procedure and the patients were continued that twice daily for three days. Patients were evaluated for peritonitis over the following 30 days. The cost for both oral and parenteral forms of cefuroxime axetil was calculated.

Results: Two groups were similar regarding age and sex characteristics. Three patients (20%) in oral group and one patient (4%) in parenteral group developed peritonitis (p>0.05). The cost of parenteral prophylaxis is 4.7 Euros, whereas the cost of oral form is 11.6 Euros. According to basic analysis, oral prophylaxis is more than twice as expensive as the parenteral administration.

Conclusions: For patients undergoing percutanaeous PD catheter insertion; single dose of intravenous cefuroxime axetil is safe, significantly less costly and equally effective in preventing early peritonitis as the same agent given orally for three days.

Su508 IMPROVEMENT OF NYHA-CLASS, BNP AND CARDIOPULMONARY CAPACITY IN PATIENTS WITH SEVERE CHRONIC HEART FAILURE TREATED WITH NOCTURNAL INTERMITTENT PERITONEAL DIALYSIS (NIPD)

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Introduction and Aims: Chronic heart failure (CHF) is one of the most common causes of death in the western countries. The cardiorenal syndrome is a vitious circle of worsening of cardiac and renal function in patients with advanced CHF leading to volume overload and diuretic resistance. The recurrent cardiac and renal failure accounts for 80% of hospitalization of CHF-patients. Nocturnal intermittent peritoneal dialysis (NIPD) is a therapeutic option for therapy-refractory CHF allowing ultrafiltration without compromizing hemodynamic effects. We aimed to assess the effects of NIPD on cardiopulmonary capacity in our CHF-patients using standard methods and a new inert-gas rebreathing (Innocor®) technique.

Methods: In 4 male patients (60±6 years) with drug-refractory CHF due to ischemic cardiomyopathy and cardiorenal syndrome we studied body weight, total body water (TBW), NYHA-class, B-type natriuretic peptide (BNP), ejection fraction (EF, echocardiography) and cardiopulmonary capacity parameters (cardiac index (CI) and oxygen uptake (VO_2) via inert-gas-rebreathing (Innocor®) and 6-minutes-walking distance before NIPD-start and after 6 weeks and 6 month (n=3) on NIPD (data are mean±SD).

Results: Before NIPD-start patients presented with NYHA-class IV and have been hospitalized every 6-8 weeks due to cardiac (and renal) failure (body weight 85±9 kg, TBW 43±8 l, BNP 1386±932 pg/ml, EF 20±3%, CI 1.5±0.7 ml/min/m^2, VO_2 2.1±0.6 ml/min/kg and 6-minutes-walking-distance 65±125 m).

For the short term, after 6 weeks on NIPD body weight (-12±5 kg) and TBW (-8±4 l) declined though doses of diuretics were reduced, NYHA-class improved to NYHA III and BNP declined (-717±386 pg/ml). EF and CI increased in 3 of 4 patients while VO_2 (+0.7±0.6 ml/min/kg) and 6-minutes-walking-distance (+240±128 m) increased in all 4 patients.

For the long term, after 6 month on NIPD all patients were assigned to NYHA-class III and presented with sustained declines in body weight (-11±7 kg), TBW (-13±3 l) and BNP (-1079±198 pg/ml). EF and CI increased in 2 of 3 patients while VO_2 (+1.7±1.2 ml/min/kg) and 6-minutes-walking-distance (+310±90 m) increased in all 3 patients.

There was no further hospitalization due to CHF since NIPD-start in all 4 patients, 2 patients even qualified for heart transplantation.

Conclusions: NIPD is a therapeutic option for drug-refractory CHF resulting in an improvement of NYHA-class, BNP and various parameters of cardiopulmonary capacity, which can be monitored by the new non-invasive inert-gas-rebreathing (Innocor®) technique.

Su509 EFFECTS OF CONVENTIONAL VERSUS BIOCOMPATIBLE DIALYSIS SOLUTIONS ON PERITONEAL AND SYSTEMIC INFLAMMATION, MALNUTRITION AND ATHEROSCLEROSIS IN CAPD PATIENTS

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Introduction and Aims: Chronic inflammation, malnutrition and atherosclerosis (MIA syndrome) is an important predictor of high mortality in CAPD patients. We aimed to evaluate the effects of PD solutions (standard *vs* more biocompatible) on some parameters of MIA syndrome in patients undergoing CAPD.

Methods: After 3±2 years of starting PD, 42 stable randomin selected CAPD patients participated in this cross-sectional study. We excluded patients who had severe anemia (Hb <10 g/l), history of or current systemic inflammatory disease or immunomodulatory therapy, peritonitis or any inflammatory conditions for at least 3 months before the onset of the study, malignant disease and acute exacerbation of heart failure. Twenty one (50%) patients continued being treated with standard PD solutions (*ANDY-disc*: CAPDP-1); while the other 21 (50% of patients) continued being treated with more biocompatible PD solutions (*Gambrosol Trio:* CAPDP-2). All patients underwent echocardiography and B-mode ultrasonography of common carotid arteries together with assessments of nutrition status, residual renal function, peritoneal solute transport and biochemical parameters of systemic and local inflammation.

Results: There were no significant differences between the groups concerning the age, gender, underlying disease, history of peritonitis, dialysis vintage and r-GFR. Patients from group CAPDP-2 had significantly lower serum level of hs-CRP (3.66±2.66 mg/l *vs* 6.28±4.52 mg/l; p=0.003); significantly better nutritional status [confirmed by Mid-arm circumference (p=0.015), bone-free arm muscle area (p=0.002) and Subjective global as-

sessment (14.28% of patients in CAPDP-2 *vs* 71% of patients in CAPDP-1 were malnourished; p=0.000)] and less presence of cardiovascular morbidity [presence of LVH (p=0.039), IMT (p=0.005), degree of carotid narrowing (p=0.000) and calcified plaques of CCA (p=0.003)]. No significant difference between the groups were observed in serum and effluent levels of inflammatory cytokines (IL-1, IL-6 and TNFα), Ca 125 effluents level, ultrafiltration volume and membrane transport characteristics.
Conclusions: Biocompatible CAPD solutions have benefit systemic effects (chronic inflammation, malnutrition and atherosclerosis), but our study didn't show statistically significant benefit influence of these solutions on local effects (peritoneal viability).

Su510 ARE THERE CLINICAL DIFFERENCES WITH THE USE OF THREE TYPES OF PATTERNS OF BIOCOMPATIBLE SOLUTIONS?

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Introduction and Aims: At present there are different biocompatible peritoneal dialysis solutions. In vitro studies show that are better than conventional peritoneal dialysis solutions. Other studies show that not all biocompatible solutions have the same characteristics, for example in the presence of of glucose degradation products. In clinical practice is not easy to decide the type of peritoneal dialysis solution based only in vitro studies. The aim of this study is to evaluate clinical differences between peritoneal dialysis three patterns with different peritoneal dialysis solutions.
Methods: 3 groups of 10 patients each followed for 18 months on CAPD program, with 4 bags of 2 liters a day.

Table 1. CAPD regimen

Group 1	1 bag Nutrineal, 1 bag of icodextrin, 2 bags of physioneal 1.36%
Group 2	3 bags Bicavera 1.5%, 1 bag Bicavera 2.3%
Group 3	3 bags of GambrosolTrio 1.5%, 1 bag GambrosolTrio 2.5%

There were no differences between demographic characteristics and the causes of kidney failure in populations of the three groups. We collected data on 3,6,9,12,15 and 18 months of the following parameters: C-reactive protein, weight, bicarbonate levels, Kt/V, glycated hemoglobin, peritonitis rate, residual kidney function, ultrafiltration.
Results: Not detected any significant difference in any of the parameters evaluated during the 18 month follow-up: CRP (mg/dL), bicarbonate (mmol/L), Kt/V, glycated hemoglobin (%), RRF mL/', number peritonitis, ultrafiltration (ml/24 day) and changes in weight (kg), with three programmed pattern.
Conclusions: With the results obtained in following 18 months are no differences in the parameters evaluated in three models using patterns biocompatible solutions.
The choice of a pattern or another, should consider other aspects such as the adequacy of conectology sistem with patient clinical characteristics or storage space.

Su511 A DAMAGED ENDOTHELIAL GLYCOCALYX IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Endothelial dysfunction and accelerated vascular disease are common features in patients with chronic renal failure (CRF). The glycocalyx is a negatively charged mesh lining the inner wall of blood vessels. It is a main regulator of vascular homeostasis. No data is available on the state of endothelial glycocalyx in CRF. In the present study we investigated whether the microvascular glycocalyx is damaged in patients with end-stage renal disease (ESRD) treated with peritoneal dialysis (PD), as compared to healthy controls.
Methods: Investigations were carried out in 8 patients with ESRD undergoing PD (male/female 7/1; median age 37.7 (18.25-53.3) years; median time on dialysis 45.7 (7.4-145.6) months and 12 healthy age and sex matched controls with normal kidney function. Exclusion criteria: diabetes mellitus, use of antioxidants, use of statins 6 weeks prior to the measurement, use of angiotensin converting enzyme inhibitors or angiotensin II receptor blockers on the day of the measurements. The status of endothelial glycocalyx in individual blood vessels was assessed by measuring the dynamic range of red blood cell (RBC) width in the sublingual microvasculature, using Sidestream DarkField (SDF) imaging with a MicroScan videomicroscope.
Results: Compared to healthy controls, PD patients appeared to have a 0.13±0.6μm (p=0.03) increase in the dynamic range of RBC width indicating the loss of glycocalyx barrier properties. This correlated with total time on renal replacement therapy (r=0.81, p=0.02)
Conclusions: Patients with ESRD on PD have a loss of glycocalyx barrier properties as compared to age and sex matched healthy controls. Impaired glycocalyx barrier properties may be an early indicator of pathogenic activation of vascular endothelium as a marker of increased cardiovascular risk.

Su512 OPEN SURGERY VERSUS LAPAROSCOPIC PLACEMENT TECHNIQUE OF PERITONEAL DIALYSIS CATHETERS – A SINGLE CENTER EXPERIENCE

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Introduction and Aims: The success of PD is determined by the success of the peritoneal dialysis catheters (PDC) placement. The ideal method for PDC placement remains debatable.
Our retrospective study compares early and late PDC – related complications and outcomes in pts. in whom PDC placement was performed by the open surgery technique (OST) and by the laparoscopic technique (LT).
Methods: A total of 62 procedures were carried out in 57 pts. from July 1996 to November 2009. Follow-up ranged from 3 to 95 months (mean 23,6±21,7 months). The procedures were undertaken by three surgeons. 34 PDC (25 straight Tenckhoff and 9 Swan-Neck double cuff catheters) were placed with the LT and 28 PDC (20 straight Tenckhoff and 8 Swan-Neck double cuff catheters) with OST. All PDC placement procedures were performed under general anesthesia.
Patient characteristics, operation-related data, procedural complications and clinical outcome were compared by using the statistical software SPSS.
Results: The median operating time was not different in the two groups (OST and LT). The median time to PD initiation was not different in the two groups, nor was any significant differences found in the infectious complications or need for secondary surgical revisions between the two groups. LT tended to have a higher incidence of pericannular bleeding (18,9% with LT versus 6,3% with OST), higher incidence of port site hernias at late follow-up (8,9% with LT versus 4,6% with OST) and a lower rate of early catheter migration (4,2% with LT versus 7,5% with OST). 10 pts. (17,5%) required catheter revision (6 pts with OST) and only 5 pts. (8,8%) required catheter removal due to technical problems (3 pts with OST). Rate and cause of overall mortality or catheter dropout did not statistically differ between the two groups. During the follow up period 12 pts. (21,05%) died, 12 pts (21,05%) were transferred to HD, 6 pts (10,5%) were transplanted and 27 pts (47,4%) are still on PD.
Conclusions: We can conclude that both PDC placement techniques (OST and LT) have specific advantages and disadvantages. They are complementary and both are safe and effective procedures for PDC placement. It's very important those tecniques to be done by dedicated and well educated surgeon.

Su513 EVALUATION OF ARTERIAL STIFFNESS AND TISSUE ADVANCED GLYCATION END PRODUCTS BY SKIN AUTOFLUORESCENCE IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Mortality is highly increased in patients with renal failure, particularly in patients with end-stage renal disease. The leading cause of death in patients with ESRD are cardiovascular complications. Advanced glycation end products are a new, non-traditional cardiovascular risk factor, implicated in the pathogenesis of age-related and chronic diseases like diabetes mellitus or chronic renal failure. Elevated cardiovascular mortality has also been shown to be associated with increased arterial stiffness. However, the contribution of tissue accumulation of advanced glycation end products (AGEs) to the increase of arterial stiffness is unclear. Our aim was to evaluate the accumulation of advanced glycation end-products in peritoneal dialysis patients and a possible correlation between AGEs and arterial stiffness in this population.
Methods: We measured the skin levels of advanced glycation end products in 75 ESRD patients from "C.I.Parhon" Hospital – Fresenius Nephrocare Dialysis Center, treated by peritoneal dialysis (33.3% male, mean age 56 y.o.). The mean duration in dialysis was 43 months. The AGE accumulation was assessed using a previously validated skin-autofluorescence reader (AGE-Reader® - Diagnoptics). In only 55 patients we evaluated both AGEs and arterial stiffness. Arterial stiffness was evaluated by pulse wave velocity (PWV) and augmentation index (AiX), determined with At-Core Sfigmocore® (Sidney – Australia). Statistical analysis was done using SPSS Statistics® 17.0. The univariate correlation between skin autofluorescence and other variables was assesses by Pearson correlation coefficient, when variables were normally distributed. Otherwise, Spearman correlation coefficient was used.
Results: Our analysis confirmed a significant correlation ($p<0.01$) between the levels of skin advanced glycation end products and the age of the patients, that supports similar data found in other study populations (hemodialysis, diabetes). The mean value of skin autoflourescence was 3.55 ± 0.7 (n = 75). We also found statistical significant correlation between AGE level and duration in dialysis ($p<0.05$). We found no significant correlation between advanced glycation end products and arterial stiffness in our study group of peritoneal dialysis patients.
Conclusions: Previous studies have suggested that AGEs accumulation plays an important role in the mechanisms that increase arterial stiffness. However, few studies have examined the association between AGEs and PWV, an established standard for measuring arterial stiffness. Previously publications have shown that stiffness is positively and independently associated with AGEs in ESRD patients treated by hemodialisys. Our study found no such correlation in patients on peritoneal dialysis. Related to duration in dialysis (correlated with AGEs), we confirm the results of a recent studies that concluded that skin AF correlated with patient age in both HD and PD but with dialysis vintage only in PD patients.

Su514 EFFECTS OF AAD(1.1% AMINOACID BASED PERITONEAL DIALYSIS SOLUTION) USE ON NUTRITIONAL MARKERS ARE ASSOCIATED WITH CHANGES OF BODY FLUID COMPOSITIONS

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Introduction and Aims: CAPD patients are frequently associated with protein-calory malnutrition due to continuous protein loss during dialysis procedure and is an important prognostic factors of patients outcome. Aminoacid based PD solution(AAD) has been used for better nitrogen balance and improving nutritional markers of CAPD patients. But its effcet on body fluid composition and various nutritional markers such as serum albumin level has been showed various contradictory results. Nutritional markers are influenced by various clinical parameters such as patient's ECF volume status. AAD may causes different ultrafiltration profile from conventional glucose based solution(GD) and causes changes of patient's volume status and various nutritional markers.
Methods: 35 CAPD patients (>6months duration of CAPD, 4 times exchanges/day) were prospectively randomized to 17 AAD group(Nutrineal, one time use/day + three times of glucose based PD solution) and 18 GD group (keep their gfour lucose based PD solution). After three month treatment follow up, we evaluate effects of AAD on nutritional markers including serum albimin and various body composition profile by using multi-frequency bioimpedance analyzer(BIA).
Results: After 3 months treatment follow up, AAD group showed favored several markrs include albumin level(3.54 ± 0.11 vs 3.74 ± 0.11, $p=0.02$), BUN(53.96 ± 3.89 vs 75.71 ± 3.88, $p=0.00$) and nPCR(1.59 ± 0.07 vs 1.98 ± 0.08, $p=0.00$). AAD group showed marginally increased drainage volume(8.77 ± 0.76L vs 9.12 ± 0.83L, $p=0.09$) and was associated with decreased ECF volume(12.45 ± 0.54L vs 12.10 ± 0.57L, $p=0.06$). Body composition analysis also showed marginally increased body weight (63.44 ± 2.5kg vs 64.24 ± 2.6kg, $p=0.09$) and fat mass(14.19 ± 1.39kg vs 15.13 ± 1.56kg, $p=0.06$) but ICF volume was not changed(22.2 ± 0.9L vs 22.3 ± 0.9L, $p>0.05$) in AAD treatment group. Although serum albumin level was increased, total plasma albumin amount, correction with ECF volume(albumin level X ECF volume) makes it no difference(43.45 ± 2.13 vs 44.80 ± 2.28, $p=0.14$). Furthermore Δalbumin vs ΔECF showed negative correlation pattern($r=-0.46$, $p=0.07$) that means serum albumin change was influenced by ECF volume change.
Conclusions: AAD treatment improved markers of better nutritional status including serum albumin, fat mass, body weight, BUN and nPCR. However the change in serum albumin level was influenced by patient's ECF volume status, which can partially explain contradictory effect of aminoacid based PD solution on serum albumin level and nutritional markers.

Su515 VARIABILITY OF EFFLUENT CANCER ANTIGEN 125 AND INTERLEUKIN-6 DETERMINATION IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Cancer antigen (CA) 125 is a glycoprotein that provides data on the state of the peritoneal membrane in peritoneal dialysis (PD). Interleukin-6 (IL-6) acts as a mediator in acute phase responses. The study evaluated the usefulness of CA125 and IL-6 in random effluent samples, by assessing their variability in individual patients during clinical practice at the outpatient department.
Methods: This longitudinal prospective study was conducted from 2007 till 2009. Participants included 52 stable PD patients aged 18 years or older. Effluent samples were obtained during regular outpatient visits, and appearance rates (AR) were calculated. Inter- and intra-individual variability were determined by the coefficient of variation (CV). A linear mixed model was used to analyze time-courses. Furthermore, the release patterns of these effluent markers were studied during a 4-hour dwell.
Results: CA125-AR of short-term patients ($\leq$24 months) ranged from 39.2-766.7 U/min, and IL-6-AR from 15.5-220.0 pg/min. Long-term patients ($\geq$25 months) had a CA125-AR of 7.3-1534.0 U/min, and IL-6-AR of 6.9-956.4 pg/min. Overall CV_{intra} was 15.3% in CA125-AR, and 28.0% in IL-6-AR. Intermediate sampling during a 4-hour dwell showed a linear increase of CA125 and IL-6 effluent concentrations. The trend of CA125-AR was different ($p=0.001$) between short and long-term patients. Relationships were found between CA125 ($r=-0.38$, $p<0.001$) or IL-6 ($r=0.23$, $p=0.04$) with PD duration.
Conclusions: The clinical relevance of effluent CA125 determinations from an unstandardized dwell during every outpatient visit is reasonable, as judged from the CV_{intra}. High CV_{intra} indicates that a single IL-6 measurement, as a predictor of outcome, should be interpreted cautiously.

Su516 PERITONEAL DIALYSIS ASSOCIATED PERITONITIS EXPERIENCE OF A PROGRAM WITH NEGATIVE SELECTION

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Introduction and Aims: Peritoneal Dialysis Program Barros Luco Hospital started in July 2007 for governmental initiative to expand therapeutic options for renal replacement in patients with restrictive criteria (exhaustion of vascular access, rural carrier or status of HBV). These patients represent a negative selection for a program of this nature, because comorbidity and risk in this group are higher when compared with open programs.Peritonitis associated with peritoneal dialysis is the mayor complication of this technique, conditioning therapy, morbidity and mortality. We intend to establish firstly a antibiotic therapy considering local bacteriology and antibiotic resistance. Also, we assets the reported peritonitis rate obtained and then we compare it with accepted international and nationally rates. We analyze the actual peritonitis rate related to the educational level of the patients.
Methods: We studied a cohort of 87 patients admitted to the program between July 2007 and July 2009, whom present perotinitis clinical evidence. There is a sampling protocol-standardized peritoneal fluid in conjunction with bacteriology laboratory of our center. Cell count is performed by sterile technique and cultivation in tubes of 50 ml, then centrifugation and culture proceed in automated blood culture vial. We analyze the peritonitis rates obtained from the first and second year of the program, bacteriological isolation and resistance to antibiotics, demographics and the Charlson Index.
Results: A 58% of our patients are between 50 to 69 years, of which 65% are women and 90% is in form APD. Of these patients, 45% have a primary education incomplete. Regarding comorbidity,70% have a Charlson index>5. The peritonitis rate calculated in both the first and second year of the program is of 0.3 patient/year with an episode every 34 months. There was no relation between the educational level and the peritonitis episodes. In the isolated microorganism, the most frequent is the Staphylococcus aureus (SA) with 33%, followed by Klebsiella pneumoniae (KP), Staphiloccocus coagulase negative (SCN) and Streptococcus viridans with 10% each.A 4.7% were Candida albicans peritonitis (1 patient).Some 28% of these cultures are reported negative. The sensitivity of isolated microorganism reported 89% of SA meticilin sensible, KP 100% to aminoglycosides and carbapenems, 75% meticilin sensible SCN.
Conclusions: This population with high comorbidity and negative selection criteria for incorporation into peritoneal dialysis program, entering 50% in emergency dialysis, peritonitis shows acceptable rates to the current international standard, thus demonstrating that this technique is suitable even for this type of population is not optimal for a peritoneal dialysis program. The bacteriology found by us in this research allow us an initial antibiotic treatment: cloxacillin or first-generation cephalosporins or carbapenems associated with aminoglycoside and then we adjust the treatment according to the agent that was isolated. The proportion of patients with negative culture must be reduced by analyzing the technique of sampling and growing also by taking the samples previously antibiotics use.

Su517 PREDICTORS OF PATIENTS' AND TECHNIQUE SURVIVAL ON CONTINUOUS AMBULATORY PERITONEAL DIALYSIS BY BIOINCOMPATIBLE AND BIOCOMPATIBLE SOLUTIONS

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Introduction and Aims: We aimed to evaluate the effects of the CAPD solutions (bioincompatible v.s. biocompatible) on patients' and techique survival.
Methods: This restrospective study included 42 CAPD patients (26 men) during the last ten years. We excluded patients who had severe anemia (Hb <10 g/l) and malignant disease. Twenty one (50%) patients continued being treated with the standard PD solutions (A.N.D.Y. Disc-CAPDP-1); while the other 21 (50%) continued being treated with biocompatible PD solutions (Gambrosol trio- CAPDP-2). All patients were analyzed for a presence of vascular calcification, nutrition status, and parameters of inflammation after 2.59 ± 1.67 years of started CAPD. These variables considered in the analysis of risk factors.
Results: Patients from CAPDP-2 group had lower level of hs-CRP (p=0.003); better nutritional status [Mid-arm circumference (p=0.015), bone-free arm muscle area (p=0.002) and Subjective global assessment (p=0.000)]; and lower vascular calcifications [IMT (p=0.005), degree of carotid narrowing (p=0.000) and calcified plaques of CCA (p=0.003)]. Kaplan-Meier analysis confirmed better survival of patients from group CAPDP-2 than from group CAPDP-1 (1-, 5-, and 10-year patients survival rate was: 100%, 62.7%, and 0.5% for CAPDP-1; and 100%, 85.2%, and 55.5% for CAPDP-2; p=0.0345). The 1-,5-, and 10-year technique survival rate was: 100%, 71.9%, and 35.9% for CAPDP-1; and 100%, 84.4%, and 76% for CAPDP-2 (p=0.0719). Duration of dialysis, serum triglyceride and Cardiovascular Morbidity Score (quantitative scoring system consisting of: EF$<$ 50%; IMT $>$ 1mm; carotid narrowing degree $>$ 50%, presence of carotid plaques in both common carotide, ischaemic heart disease, cerebrovascular event and peripheral vascular disease) were independent predictors of overall patient survival. Duration of dialysis was only independent predictor of overall technique survival.
Conclusions: Although patients treated with biocompatible solutions have shown statistically significant better survival, we can't certainly conclude that biocompatibility of CAPD solutions is a risk factor for patients' and technique survival. Namely, multivariate analysis confirmed that duration of dialysis, serum triglyceride and Cardiovascular Morbidity Score significantly predicted overall CAPD patients' survival, while only duration of dialysis was found to be independent predictor of overall techique survival.

Su518 LISTERIA MONOCYTOGENES – A RARE CAUSE OF PERITONITIS IN CONTINUOUS AMBULATORY PERITONEAL DIALYSIS (CAPD)

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Introduction and Aims: Peritonitis is a common complication of continuous ambulatory peritoneal dialysis (CAPD) and is often caused by gram-positive organisms, e.g. staphylococcus or enterococci species. Vancomycin is the antibiotic treatment of choice in such gram-positive peritonitis.
The gram-positive coccoid rods Listeria monocytogenes may be mistaken for enterococci, but are resistant to vancomycin and other empiric first line antibiotics in CAPD-peritonitis like cephalosporines.
Only a few case-reports in the 1990s describe the diagnostic and therapeutic difficulties with the uncommonly occuring peritonitis due to Listeria monocytogenes.
Methods: We report a case of a CAPD-associated peritonitis due to Listeria monocytogenes successfully treated by intraperitoneal application of ampicillin.
Results: A 41 year old female with chronic kidney disease (CKD5D) due to reflux-nephropathy starts CAPD in January 2009 after failure of living donation kidney transplantat. Thus, she continued to receive immunosuppressive therapy with tacrolimus and prednisone.
In July 2009 she presented with all signs of a CAPD-associated peritonitis. CAPD effluent contained gram-positive organisms, from CAPD effluent culture Listeria monocytogenes was grown and found to be resistant to vancomycin, but sensitive to ampicillin.
Ampicillin 500 mg (in 2000 ml 1,36% Glucose-containing dialysis solution) was administered intraperitoneal for 3 hours twice a day for 8 days. Effluent culture was negative after 3 days of antibiotic treatment.
Conclusions: Listeria monocytogenes may rarely cause CAPD-peritonitis and should be considered especially in immunosuppressed patients. Difficulties result from the fact that Listeria monocytogenes may be mistaken for enterococci, but is resistant to empiric first line antibiotics like vancomycin and cepholosporines. In our case we successfully treated Listeria monocytogenes peritonitis with intraperitoneal application of ampicillin.

Cardiovascular disease 2

Su519 INFLAMMATION, LIPIDS AND PROGRESSION OF ATHEROSCLEROSIS IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Peritoneal dialysis (PD) patients present accelerated atherosclerosis and cardiovascular disease (CVD), as well as increased levels of inflammation. The aim of the study was to evaluate the relative contribution of traditional risk factors and inflammation, as expressed by the serum levels of CRP, interleukin-6 (IL-6), tumor necrosis factor-α (TNF-α), intercellular adhesion molecule-1 (ICAM-1) and vascular adhesion molecule-1 (VCAM-1), in the progression of atherosclerosis in these patients.
Methods: The study included 44 stable PD patients (18 men, mean age 65.0±14,3 years, 13 diabetics, average time on PD 47.7±36.9 months). As surrogate ultrasonographic markers of atherosclerotic vascular lesions we used intima-media thickness and plaque score in the carotid (cIMT and cPS respectively) and femoral (fIMT and fPS respectively) arteries. Ultrasonographic measurements were repeated after a mean period of 12.5±1.80 (8 -21) months and the rates of progression (ΔIMT, ΔPS) per year were calculated. We recorded established as well as non traditional risk factors for atherosclerosis, including sex, diabetes, smoking, history of CVD, lipid profile, and the mean levels of haemoglobin, albumin, calcium, phosphorus, and iPTH. Serum levels of the aforementioned cytokines and adhesion molecules were measured by ELISA. Correlations between the above epidemiological and biochemical parameters and the ultrasonographic variables of atherosclerosis progression (ΔIMT, ΔPS) in both arteries were examined by bivariate analysis and they were further tested in multivariate stepwise regression models.
Results: In peritoneal dialysis patients, IL-6 levels was the only significant correlate of cIMT progression rate (ΔcIMT) both in bivariate and multivariate analyses (st.beta=0.471 p=0.001). Serum albumin was the only independent predictor of ΔcPS with a strong negative correlation (st.beta=-0.328, p=0.032). In bivariate analysis ΔfIMT showed a significant negative correlation with BMI (st.beta=-0.313, p=0.041) and serum levels of triglycerides (st.beta=-0.429, p=0.004) and total cholesterol (st.beta=-0.414, p=0.006), and a positive correlation with CRP (st.beta=0.409, p=0.007). In multivariate analysis, ΔfIMT was independently correlated only with serum triglycerides (st.beta=-0.380, p=0.009) and CRP values (st.beta=0.304, p=0.034), however the latter (CRP) was excluded from the final model after adjusting for sex, smoking, diabetes and history of CVD. Finally, no significant correlation was observed between fPS progression rate and any of the study variables.
Conclusions: Inflammation and malnutrition both correlate strongly with atherosclerotic vascular lesion progression in peritoneal dialysis patients. Lower lipid levels can be an important predictor of accelerated atherosclerosis in these patients possibly reflecting the presence of MIA (Malnutrition Inflammation Atherosclerosis) syndrome.

Su520 ACUTE CORONARY SYNDROME OCCURRENCE IN PATIENTS WITH END-STAGE RENAL DISEASE IS NOT ASSOCIATED WITH INCREASED CHOLESTEROL CONTENT OF ERYTHROCYTE MEMBRANES

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Introduction and Aims: Hemodialysis (HD) patients are characterized by aggressive atherosclerotic disease which cannot be fully predicted by traditional risk factors. Data from recent literature suggest that in these patients acute coronary syndromes are not associated with factors that determine plaque vulnerability but rather with novel mechanisms such as vascular stiffness, increased coagulability and increased oxidative stress. A new hypothesis for clinical instability in Coronary Artery Disease (CAD) has been proposed according which erythrocytes play a key role in atherosclerotic plaque progression and rupture. Erythrocytes are capable of increasing the size of plaque's lipid core via increased cholesterol content of their membranes. Clinical studies have reported increased cholesterol content of erythrocyte membranes (CEM) in patients with acute coronary syndromes compared to patients with stable angina. In the present study, the levels of CEM in HD patients with acute coronary syndrome, were assessed.
Methods: We recruited 68 HD patients (45 men, mean age 65±13 years) of which 16 had CAD (8 patients with acute coronary syndrome and 8 patients with stable angina). 61 (39 men, mean age 61±7 years) individuals with normal coronary arteries on angiography and normal renal function were used as a control group. Erythrocyte membranes were separated from the hemolysate by centrifugation at 15,000 rpm for 15 min. Membrane lipids were extracted by Folch's method. CEM was measured with enzymatic colorimetric method whilst membrane protein content was assessed by Bradford's method. Values are expressed as medians with inter-quartile range (IQ range).
Results: HD patients had significantly ($p<0.001$) lower CEM levels (58.1 ug/mg, IQ 24.3 ug/mg) compared to control group (76.6 ug/ml, IQ 19.7 ug/mg). CEM levels were negatively associated with creatinine levels (Spearman's rho -0.525, $p<0.001$) in whole study population as well as in patients with ESRD (Spearman's rho -0.192, p=0.047). No difference in CEM levels (p=0.971) was found between patients with (60.6 ug/mg, IQ 20.5 ug/mg) and without (56.8 ug/mg, IQ 25.3 ug/mg) Coronary Artery Disease. Similarly, no difference was observed in CEM levels (p=0.442) between patients with acute coronary syndrome (63.5 ug/mg IQ 9.3 ug/mg) and stable angina (48.8 ug/mg IQ 26.5 ug/mg).
Conclusions: CEM levels in HD patients were lower compared to healthy individuals, while, no differences were observed in CEM levels among patients with or without CAD or patients with acute coronary syndrome and stable angina. The clinical occurrence of acute coronary syndrome in HD patients do not rely on lipid core expansion via erythrocyte membranes since their CEM levels are lower compared to healthy individuals. In those patients the increased incidence of acute cardiovascular events should be explained by other pathophysiologic mechanisms.

Su521 CHRONIC HEART FAILURE IN PATIENTS WITH END STAGE CHRONIC KIDNEY DISEASE IN PROGRAM HEMODIALYSIS THERAPY

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Introduction and Aims: Cardiovascular disease (CVD) is the major cause of death in patients wih end-stage kidney disease (ESKD). The aim of the research was to study the clinical manifestations of chronic heart failure (CHF) in patients with end stage chronic kidney disease before and during program hemodialysis (PH) to assess the functional state of the heart. The detection of echocardiographic abnormalities associated with subclinical cardiac dysfunction is considered to be important.
Methods: The study involved 86 patients (47 men and 39 women) with ECKD. In all patients, clinical symptoms of CHF were evaluated. Patients underwent echocardiography and Doppler echocardiography before and during PH therapy. Systolic function was estimated by ejection fraction (EF), fractional shortening at endocardial level (endoFS) and at midwall (mwFS). EF < 50%, endoFS < 28%, and mwFS < 14% were considered indicative of LV systolic dysfunction. The maximum velocity of early diastolic filling (E), the maximum filling rate in atrial systole (A), the ratio of these velocities (E/A), isovolemic relaxation time (IVRT), deceleration time (DT) of early diastolic filling flow of the left ventricle were determined. The propagation velocity of early diastolic flow of the left ventricle (Vp) was determined with the method of color M-model Doppler echocardiography.
Results: Clinical signs of heart failure – dyspnoea – was detected in 58.1% of cases, fatigue – in 74.4%, palpitation – in 62.8%, ortopnoe – in 17.4%, edema – in 46,5%, gallop rhythm – in 7.0%, congestion in lungs – in 5.8% of cases. Concentric left ventricular hypertrophy (LVH) was diagnosed in 55 (63.95%)

patients, eccentric left ventricular hypertrophy – in 13 (15.1%) cases. Reduced ejection fraction, endFS, mwFS was detected as indicator of LV systolic dysfunction in 36 (41.9%) patients, diastolic dysfunction mainly by type of slow relaxation was detected in 66 (76.7%) cases. In patients who were on PH from 3 months to a year, the frequency of diastolic dysfunction decreased to 53.5%; in the treatment by PH which lasted 5 years, it rose to 63.95%.
Conclusions: CHF is frequently observed in patients with ECKD. Prior to PH, systolic heart function was preserved in most patients, whereas in many cases, echo-and Doppler echocardiography revealed a diastolic dysfunction of the heart. In the process of PH therapy, in the early stages of treatment indicators of the functional state of heart improved thanks to correction of metabolic disorders, nutritional status, the overload of extracellular fluid, anemia, and reduction of blood pressure in hipertension. In the later periods of treatment (after 5 years of treatment on PH) indicators of CHF gradually worsened. The results dictate the need for a regular echocardiography and Doppler echocardiography for an early detection of systolic and diastolic dysfunction for diagnosis of chronic heart failure and correction of the revealed desorders.

Su522 REACTIVE OXYGEN METABOLITES (ROMs) ARE ASSOCIATED WITH CARDIOVASCULAR DISEASE AND INCREASED MORTALITY RISK IN CHRONIC HEMODIALYSIS PATIENTS

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Introduction and Aims: The relationship between oxidative stress and cardiovascular disease (CVD) in hemodialysis (HD) patients has not firmly established and little is known about the influence of oxidative stress on survival. We measured reactive oxygen metabolites (ROMs) and correlated with prevalent CVD and all-cause and CVD-related mortality.
Methods: Sixty four prevalent HD patients of Catholic University (Rome) in March 2004 were studied (exclusion criteria: sepsis,inflammatory bowel disease, autoimmune disorders, hepatitis, liver failure, hyperthyroidism, cancer). ROMs were measured by the d-ROMs test (Diacron srl, Italy). Reference values: normal <300 CARR U, >=300 mild-severe oxidative stress >320. Patients were followed for 72 months or until death. Incident CVD were registered.
Results: ROMs levels were higher in patients with CVD (317±63.8) than in those without (242.7±49.1; p<0.0001). Multiple logistic regression analysis showed that the association between ROMs levels and CVD was independent.

Odds ratios (95% CI) for the presence of CVD at the time of inclusion

Indipendent variables	Odds ratio	95% CI	P value
Age	1.13	1.00-1.29	0.04
Dialytic age	0.99	0.97-1.01	0.80
BMI	1.00	0.75-1.33	0.99
Total cholesterol	1.03	0.98-1.98	0.15
Triglycerides	1.00	0.98-1.01	0.66
HDL	1.04	0.86-1.25	0.66
Calcium	0.33	0.06-1.81	0.20
Phosphate	1.13	0.54-2.35	0.73
Hb	0.57	0.16-2.00	0.38
Troponin	3.20	1.20-8.20	0.01
PTH	1.00	0.99-1.00	0.32
ROMs	1.02	1.00-1.05	0.03
REd cells GPX	1.00	0.99-1.00	0.71
Plama GPX	0.99	0.97-1.01	0.41

During follow-up 22 patients died, 17 from CVD-related causes. ROMs levels in surviving patients (250.2±52.2) were lower than in patients who died (320.6±67.7) (P<0.0001). Kaplan-Meier analysis showed that patients with ROMs levels <300 have lower mortality by all- (log-rank c^2 8.42, P=0.0037; HR 0.31, 95% CI 0.08-0.62) and CVD-related causes (log-rank c^2 4,91,P= 0.02; HR 0.35, 95% CI, 0.09-0.86). With Cox proportional hazard model, we corrected these differences for potential confounding factors.
Conclusions: ROMs in chronic HD patients are associated with CVD and are predictive of all-cause and CVD-related mortality.

Crude and adjusted all-cause and CVD-related mortality according to ROMS level groups

Model	Covariates	All-cause Mortality HR (95% CI)	P value	CVD-related Mortality HR (95% CI)	P value
1	Crude	3.53 (1.52-8.17)	0.003	2.79 (1.08-7.24)	0.034
2	1 + Red cell GPX	3.10 (1.31-7.29)	0.009	2.89 (1.09-7.66)	0.033
3	2 + troponin	3.02 (1.26-7.20)	0.010	2.84 (1.06-7.60)	0.038
4	3 + phosphate	3.57 (1.44-8.83)	0.006	3.39 (1.23-9.35)	0.018

Su523 ASSESSMENT OF VOLUME EXPANSION IN CHRONIC HAEMODIALYSIS PATIENTS BY CLINICAL AND BIOELECTRICAL CRITERIA

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Introduction and Aims: Chronic expansion of extracellular volume significantly contributes to high cardiovascular morbidity and mortality in chronic haemodialysis patients. The quantitative assessment and the proper adjustment of volume excess remains a challenging task for clinicians treating such patients. The aim of our study was to compare volume expansion assessed by clinical judgment with data obtained by bioimpedance analysis.
Methods: Fluid volumes were measured in supine body position before haemodialysis by whole-body bioimpedance spectroscopy (frequency range 5 to 1000 kHz). Patients with cardiac pacemakers, cardiac stents, or defibrillating devices were excluded from the study as were patients with local edema and amputated limbs. The Body Composition Monitor (BCM, Fresenius Medical Care, Bad Homburg, Germany) was used to obtain impedance data from tetrapolar measurements taken between the wrist and the ipsilateral ankle and to derive extracellular volume (V_{EC}) as well as estimated volume excess (V_X). In addition, dry weight was assessed by systematic clinical criteria (N_{SCORE}). The actual ultrafiltration volume (V_{UF}) was determined by the clinical prescription to reach the patient's target weight. Complementary, serum levels of NT-pro-BNP were evaluated before and after haemodialysis (C_{BNPpre}, $C_{BNPpost}$).
Results: 28 stable patients on chronic haemodialysis (11 women, mean age 51.3±13.3 years, mean body mass index 26.8±6.0 kg/m^2, mean arterial pressure 102.6±15.8 mmHg) were studied. At a mean V_{EC} of 17.91±3.45 L the mean volume excess V_X was 2.08±1.49 L. In comparison, the V_{UF} was 2.41±1.03 L. In spite of similarity in average values there was no correlation between V_X and V_{UF} (r = -0.145, p = 0.461). A positive correlation between C_{BNPpre} and the relative estimated volume excess (= V_X/V_{EC}) was found (r = 0.581, p = 0.001) and there was a positive correlation between $C_{BNPpost}$ and N_{SCORE} (r = 0.391, p = 0.040).
Conclusions: The strong positive correlation between C_{BNPpre} and the estimated volume excess obtained by bioimpedance analysis as well as the positive correlation between $C_{BNPpost}$ and the clinically evaluated dry weight indicate that both methods measure aspects of volume expansion. The absent correlation between V_X and V_{UF} could be explained by differences in the individual cardiac situation as well as by a striking discrepancy in the estimation of volume excess in patients with high body mass index for whom the BCM is not validated.
Disclosure: The Body Composition Monitor for the study was provided by Fresenius Medical Care Austria. W. Ribitsch receives a research grant from Baxter Austria.

Su524 LEFT VENTRICULAR HYPERTROPHY IMPROVEMENT AFTER SHORT TERM THERAPY WITH PARICALCITOL IN CHRONIC HEMODIALYSIS PATIENTS

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Introduction and Aims: Left ventricular hypertrophy (LVH) and left ventricular systolic function have both been shown to be independent risk factors for cardiovascular mortality in ESRD patients. Animal-experimental models have shown that, vitamin D deficiency promotes LVH and cardiovascular mortality.

Aim of this study was to evaluate the effect of paricalcitol, a vitamin D receptor activator, on cardiac geometry and function 3 months after treatment.
Methods: 14 stabilized chronic hemodialysis patients (M/F: 10/4, mean age 56.9±11.7years and mean dialysis duration 65,4±48,4 months) with secondary hyperparathyroidism (iPTH: 798+430 pg/ml) underwent echocardiographic examination before and 3 months after treatment with paricalcitol. Left ventricular mass index (LVMI g/m^2) and ejection fraction (EF %) were calculated. NT-Brain natriuretic peptid (BNP), Calcium, Phospat and iPTH levels were measured at the beginning and at the end of the study.
Results: LVMI decreased significantly and EF improves significantly at the end of the study (176,35±83,20 g/m^2 vs 166,58±74,45 g/m^2 and 61,23±9,14% vs 65,4±10,58%, p<0.05 respectively). BNP levels decreased also significantly (1188,2±737,3 pg/ml vs 785,5±786,1 pg/ml, p<0.05). iPTH levels fell significantly after paricalcitol therapy (798,4±430.5 pg/ml vs 413,3±244,1 pg/ml, p<0.05). All other biochemical parameter showed no significantly changes.
Conclusions: Paricalcitol improves left ventricular geometry and function leading so to a better cardiovascular survival in chronic hemodialysis patients. Direct effect on the myocard cell and indirect, due to effective iPTH suppression seems to play a pivotal role on the action of Paricalcitol on the myocard.

Su525 LYSOPHOSPHATIDYLCHOLINE, OXIDIZED LDL, AND CARDIOVASCULAR DISEASE IN PATIENTS WITH END STAGE RENAL DISEASE: ANALYSIS AT 5 YEARS OF FOLLOW-UP

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Introduction and Aims: Unlike general population, there is less of an association between dyslipidemia and the presence of cardiovascular disease (CVD) in patients with end-stage renal disease (ESRD). Although oxidized low-density lipoprotein (LDL) and lysophosphatidylcholine (LPC) has been proposed as an important determinant of the atherosclerosis, the long-term relative contribution of oxidized LDL and LPC to CVD has not been evaluated in maintaining hemodialysis patients. The objective of this study was to analyze whether or not oxidized LDL and LPC are the risk factors of CVD during long-term follow-up.
Methods: Oxidized LDL and LPC levels were evaluated in three groups (69 hemodialysis, 51 diabetic controls and 33 healthy controls). And a prospective observational study of 69 stable, chronic hemodialysis patients was conducted during 5 year periods from August 2004 to July 2009. The endpoint was the fatal and non-fatal cardiovascular events requiring admission. Analysis was performed using Cox regression analysis.
Results: Oxidized LDL and LPC levels were lower in hemodialysis patients than in diabetic and healthy controls. In a prospective follow-up study, there were 18 cardiovascular events (26.1%) including 6 deaths among the hemodialysis patients. Serum LPC levels were lower in patients with CVD than in patients without CVD, but oxidized LDL were not significant different between groups with CVD and without CVD. In adjusted Cox analysis, previous CVD (hazard ratio (HR) = 5.68, 95% confidence interval (CI): 1.94, 16.63, p=0.002) and low LPC level (≤ 255 μmol/L) (HR = 3.45, 95% CI: 1.04, 11.42, P = 0.04) had significant independent risks for development of CVD.
Conclusions: This study demonstrates that low LPC, but not oxidized LDL, is the principal risk factors for CVD in a population of hemodialysis patients who were follow-up for 5 years.

Su526 LOW TRIIODOTHYRONINE IN UREMIC PATIENTS: A LINK BETWEEN ATHEROSCLEROSIS AND INFLAMMATION?

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Introduction and Aims: Low levels of active form of thyroid hormone, free triiodothyronine, fT3, is frequently encountered in patients with chronic diseases. The relationship between thyroid hormone derangements and atherosclerotic cardiovascular disease is not fully understood in uremic patients. Therefore, we investigated whether low fT3 is associated with atreosclerotic risk factors and inflammation in chronic hemodialysis (HD) patients.
Methods: We studied 54 HD patients [M/F: 29/25; age (mean ± SE): 45.2±1.7] and 34 controls [M/F: 17/17; age (mean ± SE): 44.1±2.6]. None of the subjects had any substantial confounding medical conditions (including thyroid-related medication, diabetes mellitus, clinical cardiovascular isease or active infection). We determined fT3 levels and risk factors for cardiovascular disease including lipids, lipoprotein(a) [(Lp(a)], fibrinogen, homocysteine (Hcy), C-reactive protein (CRP) and inflammatory cytokines including interleukin-6 (IL-6), interleukin-8 (IL-8) and tumor necrosis factor-alpha (TNF-α).
Results: Gender and age was not different between the groups. HD patients, compared to controls, had higher levels (mean ± SE) of Lp (a) (mg/dL) (52.6±7.5 vs 27.7±6.5, P=0.019), CRP (mg/dL) 0.75±0.18 vs 0.45±0.16, P= 0.02), fibrinogen (mg/dL) (495±25 vs. 344±11, P=0.000), IL-6 (pg/mL)15.12±4 vs 5.18±2, P=0.006), IL-8 (6.9±3 vs 1.13±3.6, P=0.001), TNF-α (37±2.3 vs 12±1.48, P=0.00) but lower fT3 (pg/mL) 2.31±0.04 vs 2.62±0.07, P=0.000). fT4 and TSH levels were comparable between the groups. In uremic patients fT3 showed negative correlations with IL-8 (r=0.28, P=0.042) and Lp (a) (r=0.37, P=0.019). In patients, fT4 levels showed negative correlations fibrinogen (r=045, P=0.001) and TNF-α (r=0.28, P=0.036).
Conclusions: Our results suggest that low fT3 levels are associated with atherosclerotic lipoprotein-a abnormalities and increased interleukin-8 levels in chronic HD patients. The possibility of whether derangements in thyroid hormones contribute to atherosclerosis and inflammation in chronic uremic patients merits further investigation.

Su527 ECHOCARDIOGRAPHIC PREDICTORS OF PROGRESSION OF LEFT VENTRICULAR DYSFUNCTION (LVD) IN PATIENTS ON INTERMITTENT HEMODIALYSIS (HD)

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Introduction and Aims: In the course of HD treatment prevalence of LVD increases and its severity enhances. Our aim was to show changes in echocardiographic parameters which could be early symptoms of development or deterioration of LVD in HD patients.
Methods: The study, designed for 6 months, was started in 56 patients, of whom one gave up and 7 died before the end of the study. The remaining stable 48 patients (27 males, age 63.1±15.1 years, 42.1±38.6 months on HD, BMI 26.8±5.4 kg/m^2, residual urine output 200, 0 – 2000 ml/day) were included into analysis. Echocardiography (2D, PW, CW, TD) was performed in these patients before HD session (hypervolemic state, high preload) and after HD session (normovolemia, preload reduced) at the beginning of the study (stage I) and after 6 months of HD treatment (stage II) using device Pro-Sound 4000 (Aloka, Japan). Systolic LVD was diagnosed when LVEF was < 50%, diastolic LVD – according to guidelines of European Study Group on Diastolic Heart Failure with the use of tissue doppler imaging to distinguish true normal from pseudonormal filling pattern. Evaluation of prevalence of LVD based on post-HD echocardiography when preload was reduced due to proper ultrafiltration. Echocardiographic parameters obtained before and after HD session in stage I and II were respectively compared. Adequacy of each HD session was monitored using online Kt/V; routine laboratory parameters (blood count, hsCRP, Ca, P, Ca x P, intact PTH, uric acid) were evaluated in stage I and II.
Results: In both stages, systolic LVD was observed in 6.3% and diastolic LVD – in 91.7% of patients. In stage I mild diastolic LVD was diagnosed in 60.4%, moderate in 29.2%, severe in 2.1% of patients. After 6 months of HD treatment (on line Kt/V for this period 1.37±0.12; ultrafiltration volume 2115±1010 ml in stage I and 2275±1014 ml in stage II, NS; stable routine laboratory parameters) new cases of diastolic LVD were not shown,

but mild diastolic LVD was diagnosed in 54.2% of patients, moderate in 33.3%, severe in 4.2%. Although this progression in diastolic LVD was not significant, the left atrium (LA) diameter (44.0; 39.0 – 46.0 mm vs 42.0; 36.5 – 44.0 mm, p=0.001), LA area (20.0; 17.0 – 23.5 vs 19.3; 16.3 – 21.9 cm^2, p=0.017), right atrium area (15.5; 13.0 – 19.2 vs 15.0; 13.0 – 17.5 cm^2, p=0.017) and protodiastolic E wave to end-diastolic A wave ratio (1.10; 0.81 – 1.40 vs 0.94; 0.76 – 1.34, p=0.047) were greater before HD, and the isovolumetric relaxation time was longer after HD (98.0; 80.0 – 112.0 vs 90.5; 80.0 – 100.5 ms, p=0.013) in stage II compared to stage I.

Conclusions: Systolic and diastolic LVD does not deteriorate significantly during the period of 6 months of adequate HD treatment in stable HD patients, but increasing atrial diameters under hypervolemic state and changes in PW echocardiography may be the early predictive symptoms of progression of LVD.

Su528 EFFECT OF BERAPROST SODIUM (PGI_2 ANALOGUE) ON PERIPHERAL ARTERIAL DISEASE (PAD) IN PATIENTS ON HEMODIALYSIS: RESULT FROM A MULTICENTER RANDOMIZED PROSPECTIVE INTERVENTIONAL STUDY

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Introduction and Aims: Peripheral arterial disease (PAD) is one of the most serious complications and has much impact on mortality in patients on hemodialysis (HD) therapy. TASC II recommends cilostazol as an evidence-based effective drug for PAD. However, cilostazol may increase heart rate and is thought to have a high risk to induce cardiovascular events in PAD patients with concomitant severe ischemic heart disease such as HD patients. Beraprost sodium (prostaglandin I_2 analogue) has no such side effect on heart rate and seems to be safe even in patients with overt or silent cardiovascular disease. We compared the effects of beraprost sodium to antiplatelet drugs including cilostazol and sarpogrelate in PAD patients on HD by multicenter randomized prospective interventional study.

Methods: Sixty nine PAD patients on HD therapy were randomly divided into 2 groups; i.e., beraprost sodium group (Group A: N=35) and antiplatelet drug group (cilostazol or sarpogrelate, Group B: N=34), and were followed for 24 weeks. Effects of these treatments were evaluated by skin perfusion pressure (SPP), KDQOL score, and cardiovascular events and/or adverse events.

Results: Age, sex, HD duration, and PAD staging (according to Fontaine severity classification) were not different between 2 groups at study entry. Levels of SPP at 24 weeks showed significant improvement in both groups from their basal levels (Group A: 34.4±14.0 → 48.0±27.4 mmHg, p=0.02, Group B: 35.5±9.4 → 47.8±14.8 mmHg, p=0.004, mean±SD). There was no difference between 2 groups in SPP levels at basal and 24 weeks. KDQOL score showed significant improvement in emotional role score and social functioning score only in Group A. Although heart rate was not changed in Group A, 9.3/minute increase of heart rate was shown in cilostazol treated patients. There was no difference between 2 groups in cardiovascular events and adverse events during the follow up period.

Conclusions: This study clearly provided the evidence that beraprost is an effective drug for PAD in HD patients. Beraprost did not increase heart rate, and had an equivalent effect as cilostazol on PAD in HD patients.

Su529 N-TERMINAL PRO-B-TYPE NATRIURETIC PEPTIDE (NT-proBNP) IN THE PREDICTION OF LEFT VENTRICULAR (LV) FILLING PRESSURE IN PATIENTS ON HEMODIALYSIS (HD)

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Introduction and Aims: Recently, the E/Em ratio of peak transmitral E flow velocity to early (Em) diastolic mitral annular velocity has been proposed as a novel index to assess LV filling pressure. In this study, the ability of NT-proBNP to predict the E/Em ratio was assessed in the HD setting.

Methods: Twenty-four HD patients with preserved LV ejection fraction >50% participated in the study. In each patient, among others, the following parameters were measured before and after HD: E/A ratio of maximal early (E) and late (A) transmitral diastolic flow velocities, E/Em ratio, cardiac output (CO), blood pressure and NT-proBNP. One half of the patients (n=12) were treated with low flux (LFD) and the other half with high flux (HFD) dialyzers. We sought to assess the relationships between a) NT-proBNP and the measured parameters before and after HD and b) intradialytic changes in NT-proBNP and the corresponding changes in the other measured parameters.

Results:

Correlations (r) between NT-proBNP and the other measured parameters before HD (A) and after HD (B)

	E/A	E/Em	CO	SBP	Age	FVR%	TOD
A	0.539**	0.65**	0.551**	0.521**	0.454*	0.552**	-215
B	0.330	0.643**	0.415*	0.409*	0.425*	0.566**	-0.450*

SBP = systolic blood pressure; FVR% = fluid volume removed as a percentage of post-HD weight; TOD=type of dialyzer (1 = LFD and 2 = HFD); *p<0.05; **p<0.01.

NT-proBNP levels decreased after HD (from 11456±10959 to 10363±10069 pg/ml). Pre- and post-HD NT-proBNP levels correlated significantly (r=0.904; p<0.001). Multivariate analysis indicated that elevated levels of NT-proBNP after HD were associated independently with an increased E/Em, FVR%, age and the use of LFD (R2=0.726; p<0.001). NT-proBNP increased by11% in the patients treated with LFD by the end of HD, whereas it decreased by 22% in the patients treated with HFD. Multivariate analysis identified TOD (r= -0.622) and the intradialytic changes in E/Em (r=0.497) as the only independent predictors of the intradialytic changes in NT-proBNP (R2=0.494; p<0.001). A pre-HD NT-proBNP cut-off value of 5941 pg/ml and a post-HD one of 6200 pg/ml had the same sensitivity of 80% and specificity of 71% (area under curve=0.857; p<0.01 for both cutoff values) for predicting a post-HD E/Em ratio >15, a value considered as the optimal cutoff to predict an elevated LV diastolic pressure (LVDP) >15 mm Hg (Circulation. 2000;102:1788-1794).

Conclusions: Among analyzed parameters, E/Em showed the strongest correlation with NT-proBNP and intradialytic changes in E/Em were highly predictive of the corresponding changes in NT-proBNP, findings underscoring the impact of LV end-diastolic wall stress/volume overload on plasma NT-proBNP. Values of NT-proBNP above 6000 pg/ml, irrespective of TOD and sampling time, can identify HD patients with elevated LVDP that would benefit from a more aggressive clinical evaluation and therapeutic intervention.

Su530 GLOBAL AND SEGMENTAL SYSTOLIC/DIASTOLIC FUNCTION OF LEFT VENTRICLE MYOCARDIUM IN PATIENTS ON MAINTENANCE HAEMODIALYSIS

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Introduction and Aims: Left ventricular dysfunction is known to be associated with unfavorable prognosis in the patients on hemodialysis (HD). The goal of the study was to evaluate whether the global systolic/diastolic longitudinal function of left ventricle (LV) is the result of accumulation of local (segmental) alterations of myocardium kinetics.

Methods: LV ejection fraction by Simpson method ($EF_{Simpson}$), transmitral early (E), late (A) diastolic velocity, E/A and isovolumic relaxation time (IVRT) were determined by conventional pulse-Doppler echocardiography. Two-dimensional pulsed Doppler tissue imaging (DTI) of the LV was used to assess early (E'), late (A') diastolic myocardial velocities, E'/A' ratio, and peak myocardial velocity during systole (PSV') at the level of lateral margin of the mitral annulus as measures of LV global diastolic and systolic function. The same parameters by DTI (indicated as Es,As,Es/As,PSVs) were obtain from each of 12 segments of LV (basal and medial segments of septal, antero-septal, anterior, lateral, posterior and inferior parts of LV wall) to evaluate local (segmental) dysfunction. 58 HD patients (age 51±13

yrs, HD vintage 64±50 month) and 20 age- and gender matched control subjects were entered the study.
Results: Mean number of segments with systolic dysfunction (PSVs< lower limit of 95% CI of the respective segment PSVs in control group) and diastolic dysfunction (defined as Es/As<1) was 1,7 (95% CI 1,3-2,2) and 8,0 (95% CI 6,9 – 9,1), respectively. PSV' (but not $EF_{Simpson}$) correlates significantly to the number of segments with decrease of PSVs ($R_{Spearmen}$=-0,31, p=0,016) and mean PSVs at all basal segments of LV($R_{Spearmen}$=0,59, p<0,001).The number of segments with diastolic dysfunction had inter-relations with E/A ($R_{Spearmen}$=-0,46, p=0,002) and IVRT ($R_{Spearmen}$=-0,46, p=0,002) by conventional echocardiography as well as with E' ($R_{Spearmen}$=-0,48, p=0,001), A' ($R_{Spearmen}$=0,54, p<0,001) and E'/A' ($R_{Spearmen}$=-0,64, p<0,0001). In analyzing the interplay between systolic and diastolic LV functions, strong negative correlations were also found between PSV' and the number of segments with diastolic dysfunction ($R_{Spearmen}$=-0,36, p=0,008) and E/E' as indicator of global longitudinal diastolic LV function ($R_{Spearmen}$=-0,48, p=0,002)
Conclusions: It can be considered that global longitudinal LV function derives from the summation of local (segmental) disorders of myocardial velocity, and that systolic and diastolic functions of LV as determined by DTI are closely interrelated.

Su531 INFLAMMATION AND FETUIN-A AS PREDICTORS OF ALL-CAUSE AND CARDIOVASCULAR MORTALITY IN CHRONIC HAEMODIALYSIS PATIENTS

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Introduction and Aims: Recent evidence suggest that fetuin-A is a negative acute phase protein and is associated with vascular calcifications and mortality in haemodialysis (HD) patients. The aim of this prospective study was to evaluate the relative impact of inflammatory markers and fetuin-A on total and cardiovascular (CVD) mortality in stable chronic HD patients.
Methods: Eighty-five patients (45 male, mean age 58 years, mean HD duration 61 months) consecutively entered the study. Serum hsCRP was measured by immunochemistry and fetuin-A and hsIL-6 levels were determined by ELISA. All patients were followed for 5 years and all deaths were recorded. Survival was analyzed using Kaplan-Meier and Cox regression analyses.
Results: At 3 years, 25 deaths were recorded, most from cardiovascular (CVD) causes (17/25, 68%). Kaplan-Meier survival curves for all-cause and CVD mortality differed significantly between patients with fetuin-A< and ≥median (0.456 g/dl) (p=0.029 and p=0.013 respectively). In addition, compared with patients with hsCRP and IL-6<median (5.773 mg/L and 5.730 pg/ml respectively), patients with hsCRP and IL-6≥median had higher all-cause (p=0.008 and p=0.035 respectively) and CVD mortality (p=0.043 and p=0.05 respectively). In univariate Cox regression analysis all-cause mortality was correlated with age (p<0.001), albumin (p=0.006), fibrinogen (p=0.002), fetuin-A (p=0.039), hsCRP (p<0.001) and IL-6 (p<0.001). CVD mortality was correlated with age (p=0.008), diabetes (p=0.006), fibrinogen (p=0.022), fetuin-A (p=0.047), hsCRP (p<0.001) and IL-6 (p<0.001). Multivariate Cox regression analysis revealed that fetuin-A and hsCRP were independent predictors of all-cause (p=0.007 and p=0.015 respectively) and CVD mortality (p=0.012 and p=0.002 respectively). During the 5 year follow-up, 32 patients died (19/32 from CVD causes 59.4%). Kaplan-Meier survival curves for both all-cause and CVD mortality did not differ between patients with fetuin-A< and ≥median (p=0.23 and p=0.06 respectively). However, compared with patients with hsCRP and IL-6<median, patients with hsCRP and IL-6≥median had significantly higher all-cause (both p<0.001) and CVD mortality (p<0.001 and p=0.001 respectively). In univariate Cox regression analyses all-cause mortality was correlated with age (p<0.001), diabetes mellitus (p=0.04), serum albumin (p=0.005), fibrinogen (p=0.001), P (p=0.031), Ca x P product (p=0.027), hsCRP (p<0.001) and hsIL-6 levels (p<0.001). CVD mortality was correlated with age (p=0.002), diabetes (p=0.01), fibrinogen (p=0.02), hsCRP (p<0.001) and IL-6 (p=0.001). In multivariate Cox regression analyses hsCRP retained an independent effect both on all-cause and CVD mortality (p=0.006 and p=0.002 respectively).
Conclusions: In HD patients, fetuin-A is an independent predictor of short-term mortality. However, hsCRP has a stronger predictive value than IL-6 and fetuin-A for both short and long-term total and cardiovascular mortality and provides additional information for risk stratification in this high-risk population.

Su532 STUDY OF MICROCIRCULATORY CHANGES AND SKELETAL MUSCLE OXYGENATION MEASURED AT REST BY NIRS IN PATIENTS UNDERGOING HAEMODIALYSIS

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Introduction and Aims: Haemodialysis (HD) has direct and indirect effects on skin and muscle microcirculatory oxygenation which are severe enough to worsen tolerance in physical exercise and muscle asthenia to HD patients thus compromising patient's quality of life and increasing the risk of mortality. To investigate in vivo whether haemodialysis (HD) induces major changes in skeletal muscle oxygenation and blood flow in HD patients, using NIRS (near-infrared red spectroscopy).
Methods: We have included 29 patients undergoing haemodialysis (HD) in our Renal Unit in 2 months. Near-infrared red spectroscopy (NIRS), quantitative measurements of oxygenated haemoglobin (HbO_2) and deoxygenated forms (HHb) were obtained via a calf before and once hourly during 4 hours of dialysis tissue oxygen saturation (StO_2%) was continuously monitored before, during and following 3-min occlusion of the brachial artery via a pneumatic cuff, before and once hourly during 4 hours of dialysis. The same evaluation has been performed in a group of 22 matched patients who were not on HD.
Results: The total haemoglobin concentration HbT (74,15±10,87vs 56,7±12,67 μmol/lt) and HHb(32,7±9,85 vs 25,8±7,57 μmol/lt p=0,004) increased significantly during dialysis. Oxygen consumption rate (StO_2%/min) significantly improved after 4 hours of HD (11.9±10.4 vs 29.5±17.3, p=0.037). There was also a significant improvement in % oxygen consumption rate in the group of HD patients when compared with the 22 patients who were not on HD (56,6%±13,1% vs 25,7%±12,2%, p=0.005).
Conclusions: Our NIRS findings suggest that HD can have a beneficial effect on in the skeletal muscle oxygenation and peripheral microcirculation in patients undergoing haemodialysis (HD) at rest.

Su533 25-HYDROXYVITAMIN D3 LEVELS ARE PREDICTORS OF MORBIDITY AND MORTALITY IN HAEMODIALYSIS PATIENTS

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Introduction and Aims: Low serum levels of vitamin D seem to have a role in the development of cardiovascular (CV) disease, the main cause of mortality in dialysis patients.
The aim of this prospective study was to evaluate the relationship between 25-hydroxyvitamin D3 [25(OH)D3] and 1,25-dihydroxyvitamin D3 [1,25(OH)2D3] levels and the prognosis of chronic haemodialysis (HD) patients.
Methods: With this purpose, we measured [25(OH)D3] and [1,25(OH)2D3] serum levels twice (end of Winter and of Summer) and correlated them with hospitalizations and overall and CV mortality for the following 2-year period.
We studied 223 HD patients with mean age (± SD) of 62.7±15.3 years, 48% female, 27% diabetics, with mean HD time of 42.9±39.3 months. Univariate and multivariate analysis were performed and a p<0.05 was considered significant.
Results: During the 24 months of the study, 38% of the patients were hospitalized at least once and 21% of the patients died (mainly from CV causes). [25(OH)D3] levels were significantly lower in the patients that died

from all causes (16.2±8.6 vs. 23.2±12.7 ng/mL, p<0.001) and in patients that died from CV causes (16.4±9.2 vs. 22.4±12.4 ng/mL, p=0.004). [25(OH)D3] levels were also lower in patients hospitalized during the study (18.3±9.7 vs. 23.7±13.4ng/mL, p=0.001). [1,25(OH)2D3] levels were similar in all groups.
In multivariate analysis, lower levels of [25(OH)D3] were predictors of hospitalization (p=0.01), death from all causes (p=0.002) and death from CV causes (p=0.03). Patients with [25(OH)D3] deficiency (< 15 ng/mL) had a significantly lower survival at the end of the 2-year studied period (p<0.001).
Conclusions: In conclusion, [25(OH)D3] serum levels seem to be a good marker of morbidity (according to hospitalizations) and mortality (overall and CV) in HD patients.

Su534 SHORT TERM THERAPY WITH PARICALCITOL IMPROVES ENDOTHELIAL DYSFUNCTION IN CHRONIC HEMODIALYSIS PATIENTS

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Introduction and Aims: Endothelial dysfunction promotes the development of atherosclerosis and cardiovascular events in chronic hemodialysis patients.It is characterized by a reduction of nitric oxide (NO) and an increase of endothelin (ET-1), a strong vasoconstriction substance.Vitamin D receptor activators (VDRA) have shown to improve cardiovascular mortality in hemodialysis patients
Aim of the study was to evaluate the effect of paricalcitol on endothelial dysfunction in chronic hemodialysis patients.
Methods: 26 stabilized, chronic hemodialysis patients (56,4±13,5 years and mean dialysis duration 71,9±66, 7 months) were studied for 3 months. Patients suffered from secondary hyperparathyroidism (iPTH >798±430 pg/ml) and hadn't received a VDRA over a month. Paricalcitol was given i.v. according the dosage, Paricalcitol mcg = iPTH/80/HD.iPTH, Calcium, Phosphate, NO, ET-1 were measured at the beginning of the study and after 3 months.
Results: ET-1 levels decreased 10,3% and NO levels increased 15,9% after 3 months with Paricalcitol therapy. Changes were statistically significant (20,2±8,1 μmol/l vs18,1±8,7 μmol/l and 15,9±7,2 μmol/l vs 17±7 μmol/l, p<0.05,respectively).iPTH levels decreased significantly at the end of the study (798±430 pg/ml vs 348±333 pg/ml,p<0.01).The correlations between iPTH, NO and ET-1 was r=0.45 and r=0.32, respectively.All other biochemical parameters showed no significantly changes.
Conclusions: Paricalcitol has a positive effect on endothelial dysfunction and this could be very important for cardiovascular survival in chronic hemodialysis patients. Whether a direct effect, via pleiotropic action, or/and an indirect effect, via iPTH reduction, of Paricalcitol therapy it is, needs further investigation.

Su535 THE CIRCULATING INACTIVE FORM OF MATRIX Gla PROTEIN IS A SURROGATE MARKER FOR VASCULAR CALCIFICATION IN CHRONIC KIDNEY DISEASE: A PRELIMINARY REPORT

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Introduction and Aims: Vitamin K-dependent matrix Gla protein (MGP) acts as a calcification inhibitor *in vitro* and*in vivo*. The present study was performed in order to (i) determine plasma levels of the inactive, dephosphorylated, uncarboxylated MGP (dp-ucMGP) in a cohort of patients at different stages of chronic kidney disease (CKD) and (ii) evaluate the association between dp-ucMGP levels on one hand and aortic calcification and mortality on the other.
Methods: 107 patients (67±13 years; 60% males; 32% at CKD stages 2-3, 31% at stages 4-5, 37% at stage 5D) were assayed for dp-ucMGP and underwent multi-slice spiral computed tomography scans in order to quantify aortic calcification at baseline. They were prospectively monitored for mortality.
Results: Plasma dp-ucMGP levels augmented progressively with CKD stage, with a significant difference from CKD stage 4. CKD stage (p<0.0001), hemoglobin (p<0.0001), age (p=0.002) and coumarin use (p=0.006) were independently associated with plasma dp-ucMGP levels. Furthermore, plasma dp-ucMGP and age were positively and independently associated with the aortic calcification score. During follow-up (802±311 days), 34 patients died (20 from cardiovascular events). In a crude analysis, [plasma dp-ucMGP] > 921 pM was associated with overall mortality (p=0.006); this association was lost after adjusting for both age and the calculated propensity score.
Conclusions: Plasma dp-ucMGP increased progressively in a CKD setting and was associated with the severity of aortic calcification. Plasma dp-ucMGP could thus be a surrogate marker for vascular calcification in CKD.

Su536 SERUM LEVEL OF FETUIN-A IN EXTREMELY LONG TERM HEMODIALYSIS PATIENTS

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Introduction and Aims: The prevalence and extent of vascular calcification is a strong predictor of cardiovascular disease and of all-cause death in chronic kidney disease patients. Human fetuin-A has been identified as a potent circulating inhibitor of the calcification process. On the other hand, with the advances in the techniques of hemodialysis, the number of long-term survivors of end-stage kidney disease on hemodialysis has been increasing and some patients underwent hemodialysis therapy for extremely long term (more than 30 years). We conducted this study to examine the serum levels of fetuin A in extremely long-term hemodialysis survivors.
Methods: A total of 131 patients who had been under hemodialysis therapy for less than 30 years and 24 patients for more than 30 years were enrolled in this study. Also, 31 healthy age- and gender-matched control subjects were enrolled. Serum levels of albumin (Alb), calcium (Ca), phosphorus (P), fetuin-A, osteoprotegerin (OPG) and bio-parathyroid hormone were measured. Since the presence of abdominal aortic calcification (ACI) is significantly associated with both all-cause and cardiovascular mortality in hemodialysis patients, the abdominal aorta was studied in 10 sequential 8-mm slices above the bifurcation of the abdominal aorta on plain abdominal CT. The ACI was calculated as the proportion of the aortic circumference in each slice that was covered by calcification.
Results: The serum level of fetuin A was significantly lower in the hemodialysis patients (0.276±0.060 g/L) than in healthy individuals (0.340±0.054 g/L, p<0.001). The serum level of OPG was significantly higher in the hemodialysis patients (375.0±233.7 pg/mL) than in the healthy individuals (87.4±24.2 pg/mL, p<0.001). The ACI was higher in the hemodialysis patients (42.1±20.8%) than in the healthy individuals (1.5±2.4%, p<0.001). The ACI was positively correlated with the age (r=0.281), duration of hemodialysis therapy (r=0.191), serum OPG (r=0.301), serum P (r=0.217) and serum calcium-phosphorus product (Ca – P)/Fetuin A (r=0.452) in the hemodilaysis patients. The ACI was negatively correlated with the serum albumin (r=-0.181) and serum fetuin A (r=-0.386) in the hemodialysis patients. The serum level of fetuin A was significantly higher in the hemodialysis patients for more than 30 years (0.373±0.086 g/L) than in those for less than 30 years (0.279±0.063 g/L, p<0.001). And the ACI was not higher in the HD patients for more than 30 years, compared with those for less than 30 years (48.7±33.4 vs 42.2±22.7%), indicating that high fetuin-A slowed an increase of aortic calcification for the long-term.

Conclusions: We demonstrated that the ACI was negatively correlated with serum fetuin A (r=-0.386). Extremely long-term hemodialysis survivors (more than 30 years) had high serum levels of fetuin A, which may be advantageous for long-term survival.

Su537 IL-6 IS A STRONG AND INDEPENDENT PREDICTOR OF THE EXTENT AND SEVERITY OF ATHEROSCLEROTIC VASCULAR LESIONS IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Peritoneal dialysis (PD) patients present accelerated atherosclerosis and cardiovascular disease (CVD), as well as increased levels of inflammation. The aim of the study was to evaluate the correlation of traditional risk factors and inflammation, as expressed by the serum levels of CRP and fibrinogen, interleukin-6 (IL-6), tumor necrosis factor-α (TNF-α), intercellular adhesion molecule-1 (ICAM-1) and vascular adhesion molecule-1 (VCAM-1), with the extent and severity of atherosclerotic vascular lesions in these patients.
Methods: The study included 67 stable PD patients (31 men, mean age 64.6±15.2 years, average time on PD 45.5±36.6 months, 34.3% diabetics, 65.7% with history of CVD). As surrogate ultrasonographic markers of atherosclerotic vascular lesions we used intima-media thickness and plaque score in the carotid (cIMT and cPS respectively) and femoral (fIMT and fPS respectively) arteries. We recorded established as well as non traditional risk factors for atherosclerosis, including sex, diabetes, smoking, history of CVD, lipid profile, systolic and diastolic blood pressure, and the mean levels of haemoglobin, albumin, calcium, phosphorus, and iPTH. Serum levels of the aforementioned cytokines and adhesion molecules were measured by ELISA. Correlations between the above epidemiological and biochemical parameters and the ultrasonographic variables of atherosclerosis in both arteries were examined by bivariate analysis and they were further tested in multivariate stepwise regression models.
Results: cIMT was correlated with age ($p<0.000$), TNF-α ($p=0.001$), IL-6 ($p=0.000$) and ICAM-1 levels ($p=0.000$). In multivariate analyses, the parameters that retained an independent effect on cIMT values were age ($p<0.000$), history of CVD ($p=0.036$), HDL-cholesterol ($p=0.030$), ICAM-1 ($p=0.015$) and IL-6 levels ($p=0.004$). cPS was correlated with age ($p<0.000$) and ICAM-1 ($p=0.035$), IL-6 ($p<0.000$) and TNF-α ($p=0.003$). Age ($p = 0.004$) and IL-6 ($p=0.003$) retained in multivariate analyses their independent effect on cPS values. fIMT was correlated with age ($p<0.000$), IL-6 ($p=0.001$) and TNF-α levels ($p=0.005$). Multivariate analyses showed that age ($p<0.000$), BMI ($p=0.023$), smoking ($p=0.023$) and IL-6 levels ($p=0.004$) were independent predictors of fIMT values. fPS was correlated with age ($p<0.000$) and IL-6 ($p=0.003$). The above variables, as well as smoking, were also correlated with fPS in multivariate analysis ($p=0.001$, $p=0.046$ and $p=0.015$, respectively). IL-6 was the only variable apart from age that was independently correlated with all four examined ultrasonographic variables.
Conclusions: In PD patients, IL-6 is a strong and independent predictor of early as well as advanced atherosclerosis, indifferently of the arterial site examined and possibly a useful marker of cardiovascular risk.

Su538 2-YEARS SURVIVAL AND SIDE EFFECTS OF Peg-IFN-α-2a TREATMENT FOR HCV HEPATITIS IN HEMODIALYSIS PATIENTS

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Introduction and Aims: HCV infection induces inflammatory state in hemodialysis patients that precipitates worse survival. Peg-IFN-α-2a abolishes HCV infection but possibly could worse anemia. We investigated benefit of IFN-α-2a(Pegasis) based on 2-years survival and side effects monitoring in 20M and10F (32-61 years old), who became HCV(genotype 1)RNA+ after 87±65 mo of regular hemodialysis procedure.
Methods: Peg-IFN-α-2a in average weekly dose of 90 mcgr for 12 months was initiated in 15 patients, well marched by age, gender, hemodialysis duration, and co-morbidities of CVS disease and DM with control 15 patients with the same HCV infection and without Peg-IFN-α-2a treatment. Side effects according to official FDA guide and DrugLib.com data for Peg-IFN-α-2a (Pegasis), together with gender, age, dialysis duration, underlay kidney disease, co-morbidities like CVS diseases, DM and other infections, biochemical parameters for dialysis adequacy, number of HCV-PCR copies at 0 and 18th mo, blood Hb. Le, Tr number and epoetin-β(Recormon) monthly dose were analyzed 2-years survival factors by Cox analysis.
Results: Possible adverse effects of Pegasis: arthralgia (at 5th mo $p=0.033$) was less frequent, while they commonly felt fatigue (at 6th mo $p=0.007$), body pain (at 10th mo $p=0.044$), having neutropenia (at 8th mo $p=0.026$) and thrombocytopenia in relation to control group without Peg-IFN-α-2a. Peg-IFN-α-2a cessation was evidenced in 2 patients, at 3rd and 5th mo because of anemia, and in a patient struggled by cardiac infarction at 4thmo. Greater 2-years survival was found in patients on Pegasis, by Kaplan-Mayer curve (log rank $p=0.025$). The risk factors for worse 2-years survival were older age (b+)$p=0.027$, weight decrease at 1st mo (b+)$p=0.016$; myalgia at 1st mo (b+)$p=0.013$; memory impairment at 1st mo (b+)$p=0.004$; flu-like syndrome rigors at 4th mo (b+)$p=0.027$; dizziness at 5th mo (b+)$p=0.003$; back pain at 6th mo (b+)$p=0.020$; headache at 7th mo (b+)$p=0.013$; dyspnea at 2nd mo (b+)$p=0.036$; cough at 2nd mo (b+) $p=0.036$; exertional dyspnea at 5th mo (b+)$p=0.018$; and at 11th mo (b+)$p=0.016$; blood urea at 1st mo (b+)$p=0.019$, while urgent hemodialysis at 12th mo (b+)$p=0.002$; severe infection at 2nd mo (b+)$p=0.058$; blood hemoglobin at 7th mo (b-)$p=0.020$; leukocyte number at 12th mo (b+)$p=0.001$. However, the major cause of lethal outcome was cardiac failure (b+)$p=0.001$, and all 5 death events occurred in the control group of patients without Pegasis treatment (Chi-square $p=0.014$). Hemoglobin blood levels were similar between groups, applying greater epoetin-β(Recormon) doses for 50% more in Peg-IFN-α-2a group ($p=0.025$).
Conclusions: Patients with HCV infection had greater 2-yeras survival after 12 months of Peg-IFN-α-2a (Pegasis) treatment; but this is not directly because of reduction in HCV-PCR copies; the major cause was more rare cardiac failure than in the control group of HCV infection without Peg-IFN-α-2a treatment. Hematological disturbances were more frequent and serious during Pegasis treatment and epoetin-β(Recormon) dose was needed to be increased by 50% for preventing anemia.

Su539 INTRACRANIAL ARTERY CALCIFICATION IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Intracranial artery calcification is an independent risk factor for ischemic stroke, and while it is frequently observed on computed tomographic images of the brain in hemodialysis patients, its distribution has not been well studied. We investigated the frequency of calcification of each of the intracranial arteries in the hemodialysis patients.
Methods: Fifty patients on maintenance hemodialysis out of 125 outpatients at our hospital who underwent brain CT from August 2003 to December 2008 were enrolled in this study. Brain CT was performed at a slice thickness of 10mm above the tentorium cerebelli and of 5mm below the tentorium cerebelli. Patients were divided into two groups according to the presence/absence of calcification of an intracranial artery, and the age, duration of hemodialysis, distribution of the causes of the end-stage kidney disease, and serum level of calcium-phosphate(Ca-P) product were compared between the two groups. We also divided the patients with intracranial artery calcification into two groups according to the duration of maintenance hemodialysis and compared the frequency of intracranial calcification of each of the intracranial arteries between the two groups.
Results: Intracranial artery calcification was found in 36 of the 50 hemodialysis patients. Among the 36 patients with intracranial artery calcification, the

prevalence of calcification of each of the arteries was as follows: vertebral artery, 58.3%; internal carotid artery, 61.1%; basilar artery, 41.7%; anterior cerebral artery, 16.7%; middle cerebral artery, 30.6%; posterior cerebral artery, 8.3%.
The mean age tended to be higher, the mean duration of hemodialysis tended to be longer, and the mean Ca-P product tended to be higher in the patients with intracranial artery calcification than in those without intracranial artery calcification (56.6±13.3 vs. 53.6±15.7 years, 18.8±10.3 vs. 12.9±8.6 years, and 48.7±16.4 vs. 45.5±21.4, respectively). There was no difference in the distribution of the primary causes of end-stage kidney disease between the patients with and without intracranial artery calcification.
The most frequently involved site of intracranial artery calcification in the patients with a hemodialysis duration of less than 20 years was the vertebral artery(62.3%), while that in the patients with a hemodialysis duration of more than 20 years was the internal carotid artery(70.0%). However, the difference in the frequencies was not statistically significant.
Conclusions: The most frequently involved site of calcification of the intracranial arteries among the hemodialysis patients was the internal carotid artery. The prevalences of calcification of the other intracranial arteries, particularly of the basilar artery, were relatively high in the hemodilaysis patients. The frequency of calcification of each of the intracranial arteries did not differ significantly between the patients with a hemodialysis duration of more than 20 years and less than 20 years.

Su540 BIOELECTRICAL IMPEDANCE: A TOOL FOR MALNUTRITION, INFLAMMATION AND CARDIOVASCULAR RISK ASSESSMENT IN HAEMODIALYSIS PATIENTS

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Introduction and Aims: Malnutrition and overhydration are associated with high mortality in haemodialysis (HD) patients. Cardiovascular (CV) disease remains the main cause of death in these patients.
The aim of this cross-sectional study was to evaluate the relationship between body composition, assessed by bioimpedance spectroscopy (BIS), nutritional, inflammatory and CV risk markers, including brain natriuretic peptide (BNP), pulse pressure (PP), left ventricular mass index (LVMI) and vascular calcifications (VC).
Methods: We assessed 75 chronic HD patients, mean age of 63.1±15.6 years, 49% female, 29% diabetics, with mean HD time of 32.2±27.1 months. Uni and multivariate analysis was performed and a $p<0.05$ was considered significant.
Results: Malnutrition [inversely evaluated by intracellular water (ICW)/body weight (BW) ratio] was positively correlated with age (r= 0.57; $p<0.001$), diabetes (r= 0.45; p=0.004), ferritin (r= 0.37; p=0.01), C-reactive protein (CRP) (r= 0.41; p=0.008), PP (r= 0.46; p=0.001) and VC (r= 0.41; p=0.008) and negatively correlated with albumin (r= -0.44; p=0.006) and 25-hydroxyvitamin D3 [25(OH)D3] (r= -0.42; p=0.007).
Overhydration [directly evaluated by extracellular water (ECW)/BW ratio] was positively correlated with CRP (r= 0.34; p=0.009), BNP (r= 0.39; p= 0.003), PP (r= 0.47; p=0.001), LVMI (r= 0.47; p=0.001) and VC (r= 0.41; p=0.007) and negatively correlated with [25(OH)D3] (r= -0.40; p=0.006).
In multivariate analysis, malnutrition was associated with high CRP levels (p=0.001) and higher PP (> 70 mmHg) (p=0.01) and VC score (> 3) (p= 0.002). Overhydration was associated with high BNP (> 800 pg/mL) (p=0.006) and CRP levels and higher PP (> 70 mmHg) (p=0.001), LVMI (> 140 g/m^2) (p=0.004) and VC score (> 3) (p=0.002). Overhydration was also associated with low [25(OH)D3] levels (p=0.003).
Conclusions: According to this data, ICW/BW and ECW/BW ratios, assessed by BIS, reveal themselves good nutritional and inflammatory markers for HD patients and are also, significantly associated with CV risk markers.

Su541 EXCESSIVE FALL OF BLOOD PRESSURE DURING MAINTENANCE HEMODIALYSIS IN PATIENTS WITH CHRONIC RENAL FAILURE IS INDUCED BY VASCULAR MALFUNCTION AND IMBALANCE OF AUTONOMIC NERVOUS ACTIVITY

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Introduction and Aims: Acute hypotension occurring during maintenance hemodialysis (HD) is not only a critical complication, but also an independent risk factor for mortality in patients with chronic renal failure (CRF). The present study was designed to clarify the mechanisms underlying excessive fall of blood pressure during HD seen in some patients with CRF.
Methods: Fifty-six CRF patients undergoing maintenance HD thrice a week were divided into two groups according to the degree of fall of the systolic blood pressure (SBP) during HD; the hypotension group (SBP fall during HD $\geq$ 30 mmHg) and the non-hypotension group (SBP fall during HD <30 mmHg). The brachial-ankle pulse wave velocity (ba-PWV), serum high-sensitivity C-reactive protein (hs-CRP), reactive oxygen species (ROS) generation, and serum malondialdehyde-modified LDL (MDA-LDL) were measured before the HD. The high-(HF) and low-frequency components (LF) of the heart rate variability spectrum and entropy were analyzed by the maximal entropy method.
Results: The ba-PWV, hs-CRP, ROS generation, and MDA-LDL were significantly higher in the hypotension group than in the non-hypotension group. HF, LF/HF, and entropy increased significantly in the non-hypotension group, while entropy decreased significantly in the hypotension group, during HD as compared with the baseline. In addition, the LF/HF and entropy during HD were significantly lower in the hypotension group than in the non-hypotension group.
Conclusions: These findings suggest that the major factors causing excessive fall of blood pressure during HD in patients with CRF might be vascular malfunction and imbalance of autonomic nervous activity.

Su542 QTc INTERVAL AND QTc DISPERSION IN HEMODIALYSIS AND CKD PATIENTS, AND THE EFFECTS OF HD

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Introduction and Aims: Responsible mechanism for sudden cardiac death risk increases is not completely known,both arrhythmia and high incidence of ventricular early pulses were shown, the rapid change emerging in intracellular and extracellular electrolytes during dialysis, is asserted to cause those arrhythmias. *Aim:* Comparison of QTc interval and QTc dispersion among the patients possessing HD and chronic kidney disease and investigation of the influence of electrolyte variation emerging during HD over QTc interval and QTc dispersion.
Methods: Twenty five HD patients and 20CKD patiets and 10 healthy persons were enrolled. ECG recorded in 25 patients before/after a HD sesion and in 20 CRF subject. ECG were recorded before and 30 min after HD serum concentration electrolytes were monitored before and 30 min after HD. Three successive QT measurements were performed in each of the 12 leads and the mean value was calculated. QTc interval was corrected for heart rate and QT dispersion was calculated.
Results: The HD and CKD patients had an abnormally prolonged QTc interval, QTc dispersion compared to normal subjects (446±53/42±15, 408±40/39±15, 377±20/24±5 ms, $p<0.001$). When the patients having CKD and HD patients were compared, QTc interval pre and post-HD was prolonged compared to the patients with CKD($p<0.001$). PreHD; only plasma Ca^{+2} levels of patients with HD was low, postHD only K^{+1} levels was found lower.Pre and post HD in terms of QTc dispersion, there was no statistically significant variation between the patients with HD (42±15/45±18 ms) and CKD(39±15ms) ($p>0.05$).
QTc interval increased significantly postHD to pre-HD($p<0.05$). Post-HD phase, Ca was higher,Mg and K were lower than in preHD phase. An

abnormally prolonged QT c was recorded in 56% cases preHD and in 72% cases postHD. However this effect was not homogeneous. The only 16 subject had an increase in QTc duration after a dialysis session while in 5 decrease in QTc duration was recorded.The increase in QTc interval postHD correlated with Mg homeostasis. Patients with greater increase in OTc after dialysis had higher basaline plasma Mg levels, also a larger decrease in Mg postHD correlated with higher increases in QTc interval.
QTc dispersion were not increase post-HD to pre-HD($p<0.05$). Post-HD, 12 subjects had increase in QTc dispersion, 11 subject decrease in QTc dispersion was recorded.The increas in QTc dispersion post-HD correlated with Mg. Decrease in Mg post-HD correlated with higher increases in QTc interval.
Conclusions: 1. In the HD and CKD patients are an abnormally prolonged QTc interval, QTc dispersion compared to normal subjects. QTc interval is higher in HD patients compared with CKD subject, however was not QTc dispersion
2. HD increases the QTc interval in ESRD patients, mainly related to rapid changes in electrolyte plasma concentrations. However, the impact on QTc dispersion is not important. That circumstance is not homogeneous
3. Post-HD patients; among those prolonged QTc dispersion and QT interval only serum Mg level are significantly decreased, serum K and Ca level not found. Therefore, further studies with different concentration of Mg containing dialysate should be made.

Su543 ENERGY EXPENDITURE AND NUTRITIONAL STATUS IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Assessment of physical activity level and of energy expenditure is important in the clinical and nutritional care of dialysis patients. The SenseWear™ Armband (SWA) is a novel multisensory device that collects a variety of physiologic data related to physical activity; so duration and intensity of physical activity (expressed as METs =Metabolic Equivalent Task) and energy expenditure are estimated.
The aim of our study was to assess the spontaneous physical activity in stable chronic haemodialysis (HD) patients and the relation to nutritional status and dietary nutrient intake.
Methods: Forty-seven stable patients (31 m, 16 f; aged 59±13 yrs) on maintenance hemodialysis (HD) treatment, who had no major skeletal, muscular or neurological disabilities entered the study. Thirty-three normal subjects, comparable for age, sex and body mass index served as controls. The level of spontaneous physical activity and estimated daily energy expenditure was assessed by SWA; biochemistry and anthropometry data were recorded, bioelectric impedance vector analysis (BIVA) was performed after HD to determine phase angle; energy and nutrients intake assessment was obtained by a three-day food recall.
Results: In respect to controls, HD patients showed lower mean daily METs value (1.3±0.3 vs 1.5±0.2, $p<0.01$), a lower time spent on activities >3METs (89±85 vs 143±104 min/day, $p<0.05$), lower number of steps per day (5584±3734 vs 11735±5130, $p<0.001$), resulting a lower estimated energy expenditure (2190±629 vs 2462±443 kcal/day, $p<0.05$).
Thirty out of the 47 HD patients studied (64%) had a mean daily value <1.4 METS and hence defined as sedentary: they differed from the active patients for higher age (63±12 vs 54±12 yrs, $p<0.01$), lower energy intake (26.1±6.4 vs 32.4±11.3 kcal/day, $p<0.05$) and lower phase angle (5.5±1.0 vs 6.3±0.9, $p<0.05$).
SWA based estimation of daily energy expenditure was negatively related to age ($r= -0.31$, $p<0.05$) whereas positive relations were observed with BMI ($r= 0.51$, $p<0.001$), phase angle ($r= 0.40$, $p<0.01$), serum phosphate ($r= 0.49$, $p<0.001$) and albumin ($r= 0.41$, $p<0.01$).
The mean daily METs values were strongly related to energy ($r=0.47$, $p<0.001$)and protein intake ($r=0.33$, $p<0.05$) and to phase angle ($r=0.38$, $p<0.01$); similar relationships were found for the time spent on activities >3 METs. Multiple regression analysis showed that energy intake and dietary protein intake were independently related to the intensity of physical activity.
Conclusions: Our findings indicate that poor physical activity is highly prevalent in dialysis patients even when free from severe co morbid conditions or disabilities. The level and intensity of physical activity is related to body composition and to dietary nutrient intake. It confirms the strong interrelationship between exercise and nutrition, which in turn are associated with survival, rehabilitation and quality of life of dialysis patients. Physical activity programs are needed, together with a proper nutritional approach, in hemodialysis patients.

Su544 RELATIONSHIP OF THE PULSE WAVE VELOCITY WITH OSTEOPROTEGERIN AND SOLUBLE RECEPTOR ACTIVATOR FOR NF-κB LIGAND IN MAINTENANCE HEMODIALYSIS PATIENTS

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Introduction and Aims: Aortic stiffness increases end-stage renal disease all-cause and cardiovascular mortality risks. We investigated arterial stiffness through the pulse wave velocity (PWV) and the roles of osteoprotegerin (OPG) and soluble receptor activator for NF-κB ligand (sRANKL) in hemodialysis patients.
Methods: In 33 hemodialysis patients, serum OPG/sRANKL levels and arterial stiffness were measured through ELISA and Pulse Pen tonometry, respectively. Multivariate linear regression was used to assess the OPG/sRANKL–PWV relationship, with significance defined as $P < 0.05$.
Results: Compared to controls, hemodialysis patients had higher carotid-femoral (9.48±1.80 vs. 8.58±1.29 m/s, t=2.073) and carotid-radial (13.42±3.26 vs. 10.07±1.76 m/s, t=4.836) PWV values. Carotid-radial PWV positively correlated with sRANKL (r=0.349) and diastolic blood pressure (r=0.389). Serum sRANKL and phosphorus levels (or age) were independent determinants of carotid-radial (or carotid-femoral) PWV.
Conclusions: Arterial stiffness is increased in hemodialysis patients. Serum sRANKL is an independent risk factor of increased peripheral muscular-type arterial stiffness.

Su545 ANKLE BRACHIAL INDEX, MORTALITY AND VASCULAR CALCIFICATIONS IN HEMODIALYSIS PATIENTS

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Introduction and Aims: The association between hyperphosphatemia and hypercalcemia with amputations, recently described in dialysis patients (pts), may be explained by the contribution of arterial calcifications. The ankle brachial index (ABI) is a non-invasive method to evaluate peripheral artery disease (PAD). ABI<0.9 diagnoses PAD; ABI>1.3 is a false-negative caused by non-compressible arteries in lower extremities.The aim of this study was to evaluate the association between ABI with mortality and with vascular calcifications, in hemodialysis (HD) pts.
Methods: We prospectively studied 219 HD pts (60% male; 20% diabetic). At baseline, ABI was evaluated by a doppler device. Vascular calcifications were evaluated by 2 methods: the abdominal aorta calcification score (AACS) in a lateral plain X-ray of the abdominal aorta from L1 to L4 and the simple vascular calcification score (SVCS) in plain X-ray of pelvis and hands. In pelvis plain X-ray we have also identified calcifications in main arteries (PMCS) and in peripheral arteries (PPCS). The total peripheral calcification score (TPCS) was the sum of PPCS with hands vascular calcification score (HVCS). The cut-off values for the different vascular calcification scores in relation with ABI were determined by ROC curve analysis. Biochemical parameters were time averaged for the 6 months preceding ABI evaluation.
Results: An ABI <0.9 at least in one extremity, an ABI >1.3 or a normal ABI were found, respectively in 100 (45%), in 43 (20%) and in 76 (35%) pts. An ABI <0.9 in both extremities was found in 61 (28%) pts. AACS >6 and SVCS >3 were found, respectively, in 98 (45%) and 95 (43%) pts. After a follow-up of 29±7 months, 50 (23%) pts died. Adjusting for age and HD duration, an ABI<0.9 and an ABI>1.3 were associated with

mortality (HR= 3.62; p<0.001 and HR=2.78; p<0.036, respectively). The adjusted risk for having an ABI<0.9 was 2.35 (p=0.004) for AACS>6; 2.49 (p=0.003) for SVCS>3 and 3.15 (p<0.001) for PMCS>3. The adjusted risk for having an ABI>1.3 was 3.23 (p<0.002) for HVCS >2; 3.65 (p<0.01) for PPCS>2; and 3.13 (p<0.001) for TPCS>2.

Conclusions: Both low and high ABI were independent predictors of mortality in this group of patients. Vascular calcifications were associated with higher risk of PAD. Vascular calcifications in main arteries (abdominal aorta, iliac and femoral arteries) were associated with an ABI<0.9. Vascular calcifications in peripheral arteries (pelvis and hands) were associated with an ABI>1.3. The hypothesis that the correction of factors associated with development of vascular calcifications might have an impact on PAD outcomes needs to be evaluated.

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Su546 CHANGES IN HDL-CHOLESTEROL WITH CHRONIC KIDNEY DISEASE AND RENAL REPLACEMENT THERAPY: A CROSS SECTIONAL ANALYSIS

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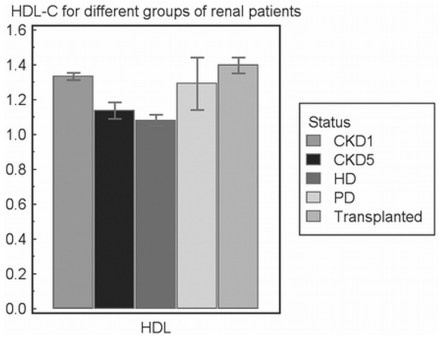
Introduction and Aims: The development of kidney failure is associated with increased risk of cardiovascular morbidity and mortality. This is multifactorial and may include the consequences of cardiovascular disease in this population as well as dyslipidemia. Worsening renal function is associated with dyslipdemia including lowering of HDL-cholesterol, believed to be protective in the development of cardiovascular disease. In this cross sectional study, we analyze the difference in HDL-cholesterol in patients with mild (eGFR>60) chronic kidney disease (CKD), stage 5 CKD, hemodialysis, peritoneal dialysis and transplant.

Methods: We retrospectively collected data from patients who attend the Northern Ireland renal service (gender, age, dialysis status, HDL, total cholesterol). Data were then analysed using Medcalc (Medcalc Software, Mariakerke, Belgium) statistical software.

Results: Data was collected from a total of 4452 patients (53±19 years, 2081 females, 2371 males). 2607 patients had an eGFR>60 and represent the control population, 381 had CKD 5, 717 were on Haemodialysis (HD), 76 on Peritoneal Dialysis (PD) and 670 were transplanted. Total cholesterol was significantly higher in the control population, compared to the other groups, and lowest in the study population on HD and with CKD-5 (Table 1). HDL-cholesterol was significantly lower in patients with CKD 5 and on HD compared with controls, PD and transplant patients (Figure 1).

Table 1

Mean ± Standard Deviation	Control	CKD 5	HD	PD	Transplanted
Total cholesterol (mmols/l)	4.8±1.1	3.9±1.1	3.7±1.0	4.4±1.3	4.5±1.0
HDL-cholesterol (mmols/l)	1.3±0.4	1.1±0.4	1.1±0.4	1.3±0.6	1.4±0.4



Conclusions: The low HDL-cholesterol in patients with CKD5 and on haemodialysis may be a direct contributor, and a difficult to modify risk factor, towards the high incidence of cardiovascular morbidity in this population. It may also partially explain the ineffectiveness of statin therapy in patients on HD, as HDL-cholesterol is only minimally impacted by this treatment. Furthermore, HDL-cholesterol profile in our study is more favourable in both the transplanted population as well as in those on PD. This supports the benefits of transplant as the gold standard renal replacement therapy (RRT) modality, and suggests that dyslipidemia that occurs with CKD is normalized upon restoration of renal function. PD may also have advantages, compared to HD in this respect, as a first modality for those entering RRT.

Su547 IMPACT OF TROPONIN I LEVEL AND LEFT VENTRICULAR HYPERTHROPY ON LONG TERM RISK OF MORTALITY IN PATIENTS ON HEMODIALYSIS

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Introduction and Aims: Some studies reveal that the patients who are suffering from chronic renal failure have elevated serum levels of cardiac markers and enzymes. But the impact of the elevation in the cardiac markers and enzymes on increased risk of all cause mortality in end stage renal disease patients on hemodialysis is unclear. We elevated the relationship between elevation in the cardiac troponin I and mortality in the patients undergoing hemodialysis. We also investigated the association between mortality risk and left ventricular mass index.

Methods: This study was carried out on 85 patients who were on hemodialysis. Serum levels of troponin Echocardiography was performed and left ventricular mass index was calculated using the Penn – cube formula. Left ventricular hyper trophy (LVH) was defined and as LVMI > 125 g/m^2. The patients were followed for 5years until date of death. We evaluated the relationship between LVH, troponin I and mortality in the patients. For data analysis we used SPSS software.

Results: Over a 5 year period, 43.5% deceased, 41.2% survived and 15.3% were transplanted. Mean age was 58.4% yrs; the youngest patient was 16 yrs and the eldest 87 yrs old. The mean level of troponin I in alive patients was 0.77±0.58 SD and 0.92±0.65 SD in dead patients. There was not significantly relation between troponin I and mortality of patients (P= 0.3). Mean left ventricular mass index in live patients was 236±67.9 SD and 296.7±98.6 SD in dead patients. There was significantly relationship between left ventricular mass index and mortality (P= 0).

Conclusions: We conclude that left ventricular hypertrophy has important role in increased risk of mortality in ESRD patients on hemoduialysis. But our study reveals that elevation of troponin I don't associated with mortality risk in this patients.

Su548 COMPARISON OF HEART RATE VARIABILITY BETWEEN END STAGE RENAL DISEASE PATIENTS ON HEMODIALYSIS AND HYPERTENSIVES IN KOREA

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Introduction and Aims: Heart rate variability (HRV) can be used to assess the effects of drug and other interventions including exercise, respiration, metabolic change and psychological or physical stress on cardiac autonomic tone. HRV is regulated by the balance of sympathetic and parasympathetic tones. Few studies about HRV of end stage renal disease (ESRD) patients were performed in Korea. So, authors investigate the autonomic nerve system activity by HRV in patients with hemodialysis due to ESRD.

Methods: We compared the pattern of cardiac sympathetic and parasympathetic activity through the time- and frequency-domain analysis of HRV with 24-hour Holter monitoring between 30 ESRD patients and 64 hypertensive control subjects. Patients have been received hemodialysis therapy at the Bongseng hospital between January 2007 and June 2008.

Results: The mean age of patients and controls were 51.17±11.91 and 55.02±13.72 years-old respectively.

In patient group, all time- and frequency-domain HRV measures including standard deviation of all normal sinus R-R intervals over 24hours (SDNN), HRV index, very low-frequency (VLF), normalized unit of low-frequency (LFnorm) and ratio of low-frequency power to high-frequency power (LF/HF) were reduced, and normalized unit of high frequency (HFnorm) was increased compared with control group.

Clinical characteristics of patients with ESRD on and hypertensive patients

	Patients	Control	P value
Case number	30	44	
Sex			0.51
Male, n (%)	17 (56.67%)	30 (46.88%)	
Female, n (%)	13 (43.33%)	34 (53.12%)	
Age, years	51.17±11.91	55.02±13.72	0.19
Hb, g/dL	10.43±0.84	13.14±1.72	0.00
Cr, mg/dL	8.84±3.66	1.04±0.22	0.00
Medication			0.54
ACE inhibitor and ARB, n (%)	21 (70.00%)	51 (79.70%)	
Beta-blockers, n (%)	15 (50.00%)	23 (35.94%)	
CCB, n (%)	20 (66.67%)	43 (67.20%)	

Data represent mean ±SD or n (%).

The comparison of time & frequency domain HRV measures between groups

	Patient	Control	P value
Mean NN (msec)	852.75±123.37	850.69±152.64	0.95
SDNN (msec)	92.53±42.93	121.48±41.47	0.00
rMSSD (msec)	57.04±60.11	43.87±46.22	0.25
SDNNi (msec)	40.12±35.35	43.12±30.25	0.67
pNN50 (%)	11.96±20.68	11.11±18.26	0.84
HRV index	11.72±5.60	16.67±5.31	0.00
VLF, msec^2	153.41±177.52	804.41±1540.41	0.02
LF, msec^2	303.69±812.14	550.26±1070.66	0.27
LF norm, nu	33.35±20.00	51.18±21.21	0.00
HF, msec^2	649.82±1528.79	712.60±2044.65	0.88
HF norm, nu	40.24±15.15	31.60±14.92	0.01
VHF, msec^2	323.26±828.00	314.36±1066.35	0.97
LF/HF	1.14±1.07	2.56±2.53	0.01

Conclusions: Autonomic tones in ESRD patients on hemodiaysis are decreased compared with those in patients with hypertension. And parasympathetic tones in ESRD patients on hemodyalysis have the preponderance over sympathetic tones.

Su549 RELATIONSHIP OF OPG/sRANKL LEVELS AND PULSE WAVE VELOCITY IN MAINTENANCE HEMODIALYSIS PATIENTS

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Introduction and Aims: Aortic pulse wave velocity (PWV), a marker of aortic stiffness has been prospectively related to all-cause and cardiovascular mortality in end-stage renal disease (ESRD) populations. Oteoprotegerin(OPG)/receptor activator for NF-κB ligand (RANKL) plays an important role in vascular disease. This study investigates the changes of arterial stiffness and relationship of OPG/sRANKL in hemodialysis patients.
Methods: Thirty-three stable MHD patients from Peking University People's Hospital were enrolled in this study (Table1). Serum OPG and sRANKL levels of 33 patients were tested by ELISA. Arterial stiffness was measured using the validated Pulse Pen tonometer. The relationship between OPG/sRANKL and PWV was assessed by multivariate linear regression.
Results: We found significant differences in PWVcr and PWVcf between the hemodialysis patients and healthy subjects [(9.48±1.80)m/s vs(8.58±1.29)m/s, t=2.073, P=0.043 and(13.42±3.26)m/s vs (10.07±1.76)m/s, t=4.836, P=0.000) respectively. Positive correlation was observed between diastolic blood pressure(DBP), sRANKL and PWVcr (r=0.389,0.349, P=0.025,0.040 respectively. Control for age, positive correlation was observed between sRANKL and PWVcr (r=0.381, P=0.029). Positive correlation was observed between PWVcf and age (r=0.466, P=0.008). In order to identify possible determinants of PWV, a multivariate regression analysis was performed using the factors including age, OPG, sRANKL, OPG/sRANKL, SBP, DBP, TG, P, TCa and Ca xP as independent variables, and PWVcr and PWVcf as the dependent variable respectively. The results showed that sRANKL and phosphate were independent determinants of PWVcr (R=0.651, R^2=0.424, Adjusted R^2=0.355, and age was independent determinants of PWVcf (R=0.466, R^2=0.217, Adjusted R^2=0.355 (Table 2).
Conclusions: Arterial stiffness is increased in HD patients. Serum sRANKL levels are independent risk factors of increasedperipheral muscular type arterial stiffness.

Su550 A PIVOTAL LINK BETWEEN HEPATOCYTE GROWTH FACTOR AND CARDIOVASCULAR DAMAGE IN END-STAGE RENAL DISEASE

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Introduction and Aims: Hepatocyte growth factor (HGF) is a pleiotropic cytokine with cardioprotective properties. It is known that its serum concentration is remarkably raised in dialysis patients. However, little is known about its role for a cardiovascular system in end-stage renal disease. In this study we assessed the determinants of serum HGF levels before and after hemodialysis and clarified its relation to cardiovascular markers in dialysis patients.
Methods: In 89 patients receiving hemodialysis, we measured serum HGF, cardiac troponin T (TnT) (positive ≥ 0.03ng/mL) and other routine biochemical parameters. We also measured left ventricular mass index (LVMI) using echocardiography and aortic pulse wave velocity (PWV). Serum HGF levels were checked both before and after dialysis.
Results: Serum HGF concentrations increased significantly after dialysis (0.214 (0.135-0.335) vs. 0.365 (0.203-0.647) ng/mL, p<0.0001). Serum HGF levels showed positive correlation with duration of dialysis (r^2=0.16, p<0.01), serum transaminase levels (r^2=0.12, p<0.01), heparin dosage (r^2=0.13, p<0.001) and aortic PWV (r^2=0.06, p=0.05), and negative correlation with serum albumin levels (r^2=0.09, p<0.05). Serum HGF levels were significantly higher in TnT positive than negative patients (predialysis 0.256 (0.153-0.438) vs. 0.166 (0.086-0.253) ng/mL, post-dialysis 0.506 (0.306-1.356) vs. 0.294 (0.173-0.46) ng/mL, p<0.05, p<0.001, respectively). There was no relation between HGF levels and LVMI. The significant difference in serum HGF levels between TnT positive and negative patients was observed in both lower and higher LVMI group. Using the multiple stepwise regression analysis, we found that duration of dialysis, positive TnT and aortic PWV were independent determinants of high HGF levels before dialysis (p<0.01, p=0.01, p<0.01, respectively), and heparin dosage, positive TnT, aortic PWV were those after dialysis (p<0.0001, p<0.01, p<0.05, respectively).
Conclusions: Although the determinant factors of high serum HGF concentrations are partially different between before and after dialysis, these are associated with positive TnT and high aortic PWV in dialysis patients, even after adjusting for LVMI. These findings suggest that HGF increase represents cardiovascular damages in end-stage renal disease.

Su551 THE EFFECT OF VOLUME STATUS ON BLOOD PRESSURE AND LEFT VENTRICULAR MASS IN CHRONIC HEMODIALYSIS PATIENTS

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Introduction and Aims: In this study we evaluated the effect of volume status on blood pressure(BP) and left ventricular muscle mass(LVM), that may be associated with them in hemodialysis(HD) patients
Methods: Patients on HD therapy at least 3 months, were included to the study. Demographic, laboratory parameters, mean interdialitic weight gains(IDWG) (kg/day), mean percentage of ultrafiltration(UF), intradialytic complications in the last month were recorded. Predialysis and postdialysis BP datas were recorded last month. Echocardiography and 24 hour ABPM were performed at the same time on the day after midweek dialysis session.
Results: Seventy four pts (36F, mean age 53.5±15.3yrs, mean duration of dialysis 41.5±41 months) were divided into two groups according to volemia status. Group 1 (n: 38), consisted of normovolemic pts (14F, mean age 50±16.7yrs, mean duration of dialysis 47.7±47.7 months) and Group 2 (n: 36), consisted of hypervolemic pts (15F, mean age 57.3±12.7yrs, mean duration of dialysis 34.9±32 months).
There were not statistically significant difference according to the duration of HD, IDWG, UF(%) and intradialitic hypotension and cramp complications between groups (p<0.05).

While 11 (28.9%) patients were found dipper in group 1, eight (22.2%) patients were found dipper in group 2 (p: 0.50). LVH had found in 33 pts (91.7%) of group 2. Whereas 21 pts(55.3%) had LVH in group 1. Significant differences were noted regarding valvular damage and LVH between the two groups (p: 0.002 and <0.001 respectively).We found positive correlation between LVMI and cardiothorasic index(CTI), IDWG, UF(%), predialysis and postdialysis BP, day and night BP in 24 hours ABPM. Negative correlation was found between LVMI and Kt/Vurea and albumin.

Table 1. 24 hour ABPM and echocardiographic datas

	Group 1	Group 2	p
SBP-day (mmHg)	125±21	142±26	0.003
Dipper	4.8±7.5	5.7±7.4	0.50
DBP-day (mmHg)	78±14	85±14	0.035
SBP-night (mmHg)	119±23	134±27	0.014
DBP-night (mmHg)	72±15	78±15	0.08
Interventricular septum thickness	1.18±0.29	1.3±0.2	0.062
LV end-diastolic diameter	4.6±0.47	4.9±0.5	0.022
LV posterior wall thickness	1.06±0.23	1.17±0.15	0.021
LVMI	131±44.9	168±42.7	0.001
EF	64±5.5	60±7.9	0.042

Table 2. Associated parameters with LVMI

	r	p
CTI	0.308	0.008
Predialysis SBP	0.461	<0.001
Predialysis DBP	0.407	<0.001
Postdialysis SBP	0.471	<0.001
Postdialysis DBP	0.408	<0.001
IDWG	0.253	0.03
UF (%)	0.279	0.016
SBP-day	0.443	<0.001
DBP-day	0.294	0.01
SBP-night	0.443	<0.001
DBP-night	0.298	0.01
Albumin	-0.236	0.04
Kt/V urea	-0.298	0.05

Conclusions: In conclucion; increased BP, IDWG and increased UF were independent predictors for developing LVH. Increasing volume and IDWG can lead to increase BP and LVMI.

Su552 ARTERIAL STIFFNESS AND QT DISPERSION IN HEMODIALYSIS PATIENTS

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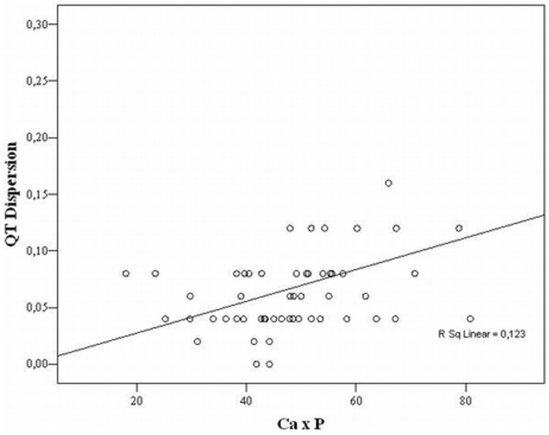
Introduction and Aims: Increased arterial stiffness and QT dispersion (QTd) are associated with cardiovascular mortality in the general population. However, the relation between these two predictors of cardiovascular risk is not clear in hemodialysis patients. The purpose of this study is to evaluate arterial stiffness and QTd, and their relation with clinical and laboratory parameters in dialysis patients.
Methods: Seventy-seven patients (47 male, mean age: 56±16 years) were included in the study. Two markers of aortic stiffness –aortic pulse wave velocity (PWV) and augmentation index (AIx) were determined by applantation tonometry. QTd and corrected QTd (QTcd) values were calculated by electrocardiograms. Serum levels of biochemical markers including serum glucose, BUN, creatinine, electrolytes, uric acid, calcium (Ca), phosphorus (P), total protein, albumin, CRP, PTH and total, LDL and HDL cholesterol levels were measured. Associations among these variables were analyzed.
Results: Mean aortic and brachial AIx in our study population were 39.4±13.6% and 14.1±31.5%, respectively. Mean aortic PWV was 10.7±2.7 m/sec. The mean QTd and QTcd values were 0.07±0.05 ms and 0.07±0.05 ms, respectively. In the univariate correlation analysis, PWV was positively correlated with age (r=0.265, p=0.022), systolic (r=0.302, p=0.008) and diastolic blood pressure (r=0.209, p=0.037) levels. There was also a significant correlation between AIx and QTd (r=0.339, p=0.043). Among biochemical markers, QTd was significantly correlated with serum P (r=0.340, p=0.012) and CaxP (r=0.379, p=0.005) levels.



However, there was no correlation between QTd and PWV.
Conclusions: The effect of arterial wave reflections might be significantly dependent on patient age in dialysis patients. Increased serum calcium and phosphate levels which were associated with QT dispersion may be involved in the pathogenesis of cardiac arrhythmia and sudden death in dialysis population.

Su553 VASCULAR CALCIFICATION, ARTERIAL STIFFNESS & BMD IN CHRONIC KIDNEY DISEASE

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Introduction and Aims: Vascular calcification with arterial stiffness as well as bone mineral density was compared in hemodialysis, peritoneal dialysis patients, and pre-dialysis patients.
Methods: The calcification level of the aorta was scored, the arterial stiffness level was examined by pulse wave velocity (PWV), bone mineral density was measured by the use of DEXA, and analyzed.
Results: PWV was significantly higher in the calcification group. The systolic blood pressure and the PWV value of the HD group was r=0.566 (p<0.001), the PD group was r=0.711 (p<0.001), and the pre-dialysis patients group was r=0.461 (p=0.001), and in all groups, a high correlation was shown. In the association of the PWV value with BMD and T score, in the PD patient group, with spine BMD, it was r=-0.351 (p<0.05), femur BMD was r=-0.510 (p<0.01), and femur T score was r=-0.527 (p=0.001). In the multivariate analysis of the PWV value, in the HD group, age and systolic blood pressure were significant and in the PD group, calcification score femur BMD, femur T score, and CRP were significant factors. In the pre-dialysis patients group, only femur T score was detected to be a significant factor for PWV.
Conclusions: In hemodialysis patients, age and systolic blood pressure, and in peritoneal dialysis patients, vascular calcification and the BMD level were analyzed to be significant factors mediating effects on arterial stiffness.

Su554 FRACTIONAL EXTRACELLULAR WATER VOLUME MEASURED BY BIOIMPEDANCE SPECTROSCOPY: IS IT CLINICALLY RELEVANT IN PATIENTS WITH END-STAGE RENAL DISEASE?

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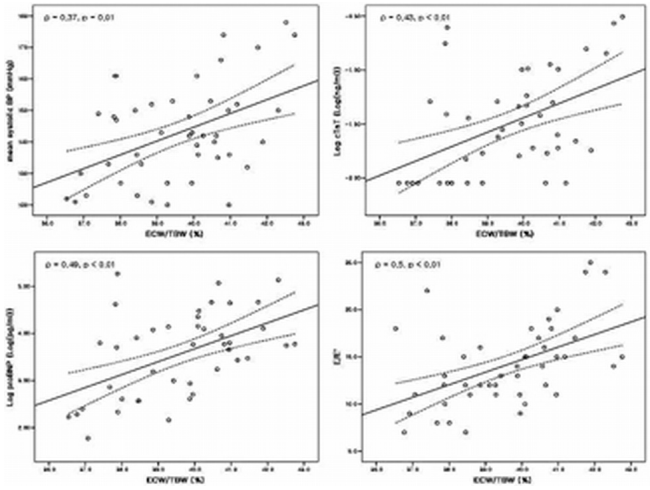
Introduction and Aims: Multi-frequency bioimpedance spectroscopy (BIS) can estimate extracellular water (ECW) and total body water (TBW) respectively. The ratio of ECW to TBW (ECW/TBW) can be used for

determination of volume overload in patients with end-stage renal disease (ESRD).

Methods: We evaluated whether ECW/TBW is actually related to blood pressure (BP), cardiac troponin T (cTnT), pro-B-type natriuretic peptide (proBNP), the mitral inflow to annular velocity ratio (E/E') and left ventricular mass index (LVMI) in patients commencing renal replacement therapy.

Results: Forty-five patients were included (male 22, age 50.1±12.5 years). Mean ECW/TBW was 39.6% (SD 1.6), which was higher than 33.1% (SD 1.4) in healthy controls (n = 181, male 118, age 49.1±14.2 years) ($p<0.01$). ECW/TBW was positively correlated with mean systolic BP during 24 hours ($\rho = 0.37$, $p = 0.01$), cTnT ($\rho = 0.43$, $p < 0.01$), proBNP ($\rho = 0.49$, $p < 0.01$) and E/E' ($\rho = 0.5$, $p < 0.01$).



However, there was no relationship between ECW/TBW and LVMI ($\rho = 0.16$, $p = 0.30$).

Conclusions: ECW/TBW measured by BIS is well correlated with systolic BP, cTnT and proBNP which are considered as biomarkers of cardiovascular outcome and E/E' reflecting left ventricular filling pressure. These results suggest that ECW/TBW by BIS is clinically relevant to estimate volume status and is helpful to control volume overload adequately in patients with ESRD.

Su555 CHARACTERIZATION OF HEART FAILURE PATIENTS WITH PRESERVED LEFT VENTRICULAR EJECTION FRACTION

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Introduction and Aims: Heart failure with preserved left ventricular ejection fraction (LVEF) has not been fully explored. This study was conducted retrospectively to evaluate clinical onset characteristics between chronic kidney disease (CKD) stage5 heart failure patients with reduced LVEF and CKD stage 5 heart failure patients with preserved LVEF.

Methods: Predialysis patients who presented heart failure were divided into two groups: patients with EF 45% and higher (n=19), and patients with less than 45% (n=6). Patients on dialysis treatment, or with cardiovascular disease were excluded. Clinical parameters and echocardiographic parameters were compared statistically. CKD stage 5 patients without heart failure (n=17) were enrolled as a control group.

Results: Patients with reduced LVEF (EF39.1±4.8%) were observed in 24% and patients with preserved LVEF (EF63.9±10.5%) were observed in 76%, which has no significant difference with control patients (EF68.0±7.9%). The pulse rate was significantly increased in patients with reduced LVEF (97.8±20.0) compared to preserved LVEF patients (81.2±16.3, $p<0.05$) and to control patients (74.0±9.0, $p<0.001$). Although there were no significant differences in systolic blood pressure or diastolic blood pressure between these three groups, pulse pressure was significantly decreased in patients with reduced LVEF compared to preserved LVEF patients (57.2±22.3 vs. 82.6±21.6 mmHg, $p<0.05$). No significant differences were seen in levels of phosphorous or calcium phosphorus products between these three groups. Left ventricular diastolic diameter (LVDd) and left ventricular endodiastolic volume (LVEDV) was little in patients with preserved LVEF compared with reduced LVEF patients. Furthermore, relative wall thickness/LVDd ratio was low in patients with reduced LVEF compared to preserved LVEF patients (0.28±0.04 vs. 0.39±0.09, $p<0.05$). LV mass index was increased in patients with preserved LVEF by 20%, while the increment was blunted in patients with reduced LVEF by 10%.

Conclusions: Preserved LVEF heart failure was more common in CKD stage 5 patients with heart failure. Our results suggest that pulse rate and pulse pressure are important markers to distinguish two different types of heart failure. Echocardiographic differences between these subtypes of heart failure may potentially characterize clinical features for diastolic heart failure.

Su556 LONG TERM HOMOCYSTEINE-LOWERING I.V. FOLIC ACID TREATMENT DECREASES PROGRESSION OF THE VASCULAR DAMAGE IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Plasma total homocysteine (t-Hcy) has been proposed as a risk factor for atherosclerosis in the general population. Its levels are elevated in at least 85% of hemodialysis (HD) patients (pts), and that could partly explain the high rate of cardiovascular events in these pts. The association between plasma t-Hcy concentration and carotid atherosclerosis has not been thoroughly studied in this populations, as well as the impact of long term t-Hcy lowering treatment on the vascular damage progression. We designed this study to evaluate the association between t-Hcy and atherosclerosis, evaluated by high resolution B-mode ultrasound to mesure the intima-media thickness (IMT) of the carotid arteries.

Methods: Forty-one pts out of 106 HD pts were studied during 24 months. All pts at baseline, and after 12 and 24 months of treatment with i.v. folic acid 50 mg/week, i.v. vitamin B6 250 mg/week and vitamin B12 p.o. underwent evaluation of: carotid IMT (CIMT; mm), and serum levels of sCa, sPO4, PTH, cholesterol, HDL, LDL, triglycerides, folic acid (n.v. 3.7-16.9 ng/ml), vitamin B12, homocysteine (n.v. 5-15 μMol/L), fibrinogen, C-reactive protein (CRP; n.v. 0-5 mg/L), and albumin.

Results: CIMT was significantly correlated with plasma t-Hcy ($r=.277$, $P<.01$).

At the end of the follow up a significant increase of folic acid and vitamin B12 levels and a significant reduction of plasma t-Hcy levels were recorded. Moreover CIMT values decreased, although not significantly, and in particular it did not increased. The main results are reported in the table.

	Baseline	12 months	24 months
Phosphorus, mg/dl	5.8±1.3	5.5±1.2	5.6±1.0
PTH, pg/ml	468±442	304±214†	385±350
HDL, mg/dl	41±13	45±14	42±13
LDL, mg/dl	96±35	71±34	65±28
Homocysteine, μMol/L	48.1±41.9	26.2±24.2†	23.4±7.5†
Folic acid, ng/ml	14.5±12.7	26.2±24.2	29.9±17.8†
Vitamin B12, pg/ml	642±962	1215±1135	1215±1135
CRP, mg/L	4.43±5.11	4.39±5.90	3.80±3.21
CIMT, mm	1.49±0.77	1.35±0.76	1.41±0.67

vs baseline: *P<0.001; †P<0.05.

Conclusions: These results show i.v. treatment with folic acid and piridossine reduced significantly plasma t-Hcy levels and the progression of the vascular damage. Our clinical data indirectly suggest a regular and timely tHcy-lowering treatment, in this high-risk populations for cardiovascular disease, might be of the benefit on the prevention of cardiovascular events.

Su557 ELEVATED SERUM MAGNESIUM LEVELS ARE NOT ASSOCIATED WITH QT INTERVAL PROLONGATION OR INCREASED INCIDENCE OF CARDIAC DYSRHYTHMIAS IN CHRONIC HAEMODIALYSIS PATIENTS

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Introduction and Aims: Sudden cardiac death is the most common cause of death in haemodialysis populations. The aetiology is complex, but in part may relate to the profound electrolyte abnormalities characteristic of these patients. As part of a dose-forcing study to assess the tolerability of fermagate (a novel inorganic polymer phosphate binder) we assessed the effect of magnesium released from this agent on QT interval and cardiac arrhythmias.
Methods: We studied 31 established haemodialysis patients. Patients were washed off their previous phosphate binding medication and then subjected to an open label escalation of fermagate dose. Dose was titrated at 2-weekly intervals to a maximum of 9g per day. Serum magnesium, phosphate and other electrolytes were measured before and after haemodialysis treatment. 12-Lead ECGs were performed in triplicate, pre-dialysis and post-dialysis at each study visit. ECGs were centrally read and the QTc interval calculated using Bazett's correction, Fridericia's correction and subject specific assessment. These assessments were supplemented by 48 hour Holter recordings made on the same visits as for the12-lead ECGs excepting follow-up.
Results: Twelve of 31 patients reached the maximum dose of fermagate. Ninety four percent of patients tolerated 3 g per day (the previously identified mean effective dose used to control serum phosphate <1.78 mmol/l). The most common adverse events were gastrointestinal, with no evidence of dose dependency of these or cardiac disorders. By end of dose titration, the greatest rise of mean pre dialysis serum magnesium and fall of mean serum phosphate was 0.64 mmol/l $\pm$ 0.41 (9.0 g per day) and 0.55 mmol/l $\pm$ 0.44 (4.5 g per day) respectively. Serum magnesium levels were effectively normalised by dialysis. There were no effects of serum magnesium levels on QT interval, before or after dialysis at any stage (mean change in pre-dialysis QTcB interval from baseline ranged from -0.32 to +3.28 ms). No treatment-emergent AV-block/prolongation of PR interval was noted, and there was no effect on any parameters derived from the Holter records.
Conclusions: Episodic pre dialysis elevated serum magnesium levels are not associated with an increase in either QT interval or the incidence of cardiac arrhythmias.

Su558 THE IMPACT OF DIALYSIS MODALITY ON MATRIX METALLOPROTEINASES (MMP-9,2) AND THEIR TISSUE INHIBITORS (TIMP-1,2) IN CHILDREN AND YOUNG ADULTS ON CHRONIC DIALYSIS

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Introduction and Aims: Among the patients with end stage renal failure, cardiovascular complications are the first cause of morbidity and mortality. Therefore, there is constant need for new markers helpful in assessing the atherosclerotic risk in that group. The MMP/TIMP system, due to its proteolytic activity, plays a role in matrix accumulation, endothelial destruction and vascular remodeling. Specific inhibitors of MMPs, TIMP-1 and TIMP-2, modify the activity of metalloproteinases, thus influencing the MMP/TIMP balance. In particular, the anti-atherogenic activity of MMP-9 and TIMP-2, as well as pro-atherogenic of MMP-2, have been observed in animal models, but the data concerning dialyzed patients are scarce, especially concerning children. The aim of the study was to assess the serum concentrations of MMP-9, MMP-2, their specific inhibitors TIMP-1 and TIMP-2, and to evaluate correlations with classical markers of atherosclerosis in children and young adults on renal replacement therapy.
Methods: 22 children on APD (median age 10 years), 17 patients on HD (median age 14 years) and 24 age-matched controls were examined. Serum concentrations of MMP-9, MMP-2, TIMP-1 and TIMP-2 were assessed by ELISA. Serum CHOL, HDL-CHOL, LDL-CHOL, TGL and hsCRP were also evaluated.
Results: Median values of MMP-9, MMP-2, TIMP-1 and TIMP-2 were significantly elevated in all dialyzed children vs. controls ($p<0.000001$) and were higher in HD than APD ($p<0.0001$). The MMP-9/TIMP-1 ratio was elevated in patients on dialysis, whereas the MMP-2/TIMP-2 ratio was decreased in all dialyzed children. In both cases the values were higher in APD than in HD ($p<0.0001$). Several correlations were notified between: MMP-9 and TGL in HD (R=-0.55, p=0.02), TIMP-1 and HDL-CHOL in HD (R=0.51, p=0.03), TIMP-1 and hsCRP in APD (R=0.49, p=0.04), MMP-2/TIMP-2 ratio and LDL (R=-0.54, p=0.03).
Conclusions: Increased concentrations of examined parameters indicate the dysfunction of MMP/TIMP system in children on renal replacement therapy, whereas differences between dialysis modalities speak in favor of APD as a method less atherogenic than HD. Correlations between analyzed parameters and classical factors influencing atherogenesis may indicate their role as new markers of atherosclerosis in the examined population.

Su559 SIX MONTHS PROGRAMS OF ADAPTED EXERCISE TRAINING IN HEMODIALYSIS PATIENTS: EFFECTS ON PHYSICAL PERFORMANCE

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Introduction and Aims: Sedentary lifestyle is frequent in the hemodialysis population, whereas regular physical exercise may reduce several cardiovascular risk factors and increase rehabilitation and quality of life. So implementation of physical activity programs are needed.
The aim of this intervention study was to evaluate the impact of 6 months programs of adapted physical training in a cohort of clinically stable hemodialysis patients.
Methods: Hemodialysis patients free from severe comorbidities or irreversible muscular or neurological disabilities, were invited to participate a training program adapted to physical capacity of the patient: 22 pts (16m, 6f, aged 62$\pm$15 yrs) entered the study; 4 of them were unable to walk and formed the "not walking group" (NWG).
Before and after a 12 months run in period (T-12, T0), after 3 (T3) and 6 (T6) months of training, the patients underwent the 6 minutes walking test (WT), and maximal constant treadmill test (TM) at speed 3 km/h and 10% grade; patients were also given a pedometer (P).
All the patients trained during the hemodialysis session (thrice weekly) with exercises of coordination, flexibility and muscular strengthening for 30 min within the first two hours of the hemodialysis session. This was the only training program for the NWG patients. The other 18 patients underwent additional training: 9 pts an advised domiciliary training "home-exercise" based on walking and monitored by pedometer (advised walking group *AWG*) in 9 pts; the advised domiciliary training plus an additional supervised ambulatory training 2 times a week by walking on a treadmill and a workout with upper limb ergometer crank (Supervised Walking Group *SWG)* in 9 pts.
Results: Results In NWG, 3 of the 4 pts regained the ability to walk and to perform the walking test at T6 (213$\pm$128 m). In both AWG e SWG no significant changes occurred during the run-in period, whereas training induced significant improvements in walking capacity and treadmill

Table 1. Data of TM and P tests, before and during the training programs in AWG and SWG hemodialysis patients

	T -12	T 0	T 3	T 6
AWG, TM, m	242$\pm$208	232$\pm$204	377$\pm$272**	615$\pm$413**
AWG, P, steps/day	2908$\pm$1622	2446$\pm$1642	3609$\pm$1978*	4700$\pm$2366*
SWG, TM, m	248$\pm$197	248$\pm$187	424$\pm$272**	890$\pm$364***§
SWG, P, steps/day	5000$\pm$2904	4353$\pm$2639	5446$\pm$2539	5130$\pm$2255

*p<0.05, **p<0.01, ***p<0.001 vs baseline §p<005 vs AWG TM

performances: at T6 there was almost a doubling of performances which were greater in SWG than in AWG (Table). No unwanted side effect occurred during the study period.
Conclusions: The results demonstrate that adapted exercise programs can significantly increase physical performances in hemodialysis patients. Collaboration and education of patients and staff are crucial to obtain adherence and successful effects. Beneficial consequences are expected in terms of rehabilitation, quality of life, cost savings and survival.

Su560 THE PREDICTIVE VALUE OF CARDIAC TROPONIN T IN HEMODIALYSIS PATIENTS: A TWO YEAR PROSPECTIVE STUDY

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Introduction and Aims: Cardiovascular events still represent the main cause of death in hemodialysis patients (HDpts). Cardiac troponin T (cTpT), a subunit of the cardiac actin-myosin complex, is released into the circulation even in silent myocardial necrosis. High levels of cTnT are associated with an increased cardiovascular mortality in HDpts.
The aim of our study was to evaluate the prognostic value of cTpT in our hemodialysis population.
Methods: Eighty four (47 males) HDpts with a mean age of 65.24±10.67 years, on hemodialysis for 69.17±54.73 months were included in the study. Twenty four (28.4%) patients were diabetics and 26 (31%) had a history of ischemic heart disease (IHD). The patients were followed prospectively for 24 months after the determination of cTpT. A cut-off of ≥0.1 ng/ml was used in assesing the prognostic significance of cTpT. The outcome after 24 months was chosen as the end-point. Also at the begining of the study patients clinical and laboratory data were collected (Table 1).

Table 1

n	cTnT ng/ml	Alb g/L	Hb g/L	CRP mg/L	URR %	V.S. (0-3)* n
84	0.062±0.062	3.69±0.32	11.95±1.54	2,69±8,9	64.42±10.2	34=1, 8=2, 42=0

*V.S. = vascular score: scoring for peripheral vascular disease of amputation, cerebrovascular disease and history of ischemic heart disease.

cTnT was correlated with clinical and laboratory parameters and outcome.
Results: Nineteen patients (22,6%) had an cTpT ≥0.1 ng/ml. The patients with cTpT ≥0.1 ng/ml had a lower serum albumin level (3.52± 0.31 vs 3.74±0.3 g/L, p=0.007), an higher incidence of previous IHD (73.7% vs 18.5%, p=0.0004) and a higher vascular score (1.11±0.49 vs 0.03±0.5, p=0.0004). In the multivariate analysis only two variables showed a correlation with cTpT ≥0.1 ng/ml, namely serum albumin (p=0.0006) and vascular score (p=0.0003).
During the study 19 patients (22,6%) died (11 males), 10 from cardiovascular causes. Twelve (63,1%) had a cTpT ≥0.1 ng/ml. 63,2% (12/19) of the patients with an high cTpT died versus 10,8% (7/65) of the patients with a low cTpT (p=0,0002). The Kaplan Meier survival curve showed a significant difference in survival time between the patients with cTpT ≥0.1 ng/ml ant the patients with cTpT <0,1 ng/ml (p=0,003). Survival was also analyzed with a Cox's proportional hazards model. After adjustment for age, sex, hemoglobin and albumin level, URR, history of IHD and vascular score survival time was significantly influenced only by the level of cTpT ≥0.1 ng/ml (p=0.01).
Conclusions: cTnT is a significant independent predictor of outcome in in our hemodialysis population and it seems that it could be used in cardiovascular risk stratification and in selecting patients that would benefit from a more invasive cardiologic investigation and intervention.

Su561 THE EFFECT OF OMEGA-3 FATTY ACID SUPPLEMENTATION ON TOTAL HOMOCYSTEINE LEVEL IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Cardiovascular disease (CVD) is a major cause of morbidity and mortality in patients on dialysis. High level of the total homocysteine (tHcy) is a strong predictor of cardiovascular events in this patients. There are some different study with controversial results about the beneficial effect of Omega-3 (fish oil) supplementation on total homocysteine (tHcy) level in dialysis patients. This study was designed to further evaluation of the effect of Omega-3 fatty acid on total homocysteine (tHcy) level in homodialysis patints.
Methods: 100 volunteer hemodialysis patients were matched (age, sex, duration on dialysis) as two group; case and control. Case group received Omega3 as a capsules 3g/day and control group were on placebo for two months. Routine laboratory tests, lipids and total homocysteine (tHcy) were measured before and 2 months after taking omega 3. Statistical analysis was done by means of the statistical package SPSS 16. Values are presented as mean and standard deviation, and 95% confidence interval. The difference between the means from the two groups was checked by Independent T test. P value less than 0.05 was considered statistically significant.
Results: from 100 patients, 88 cases completed the trial. There was no statistically significant difference regarding serum lipids, iron status and Calcium phosphorus balance between two groups. But statistically significant reduction was observed in tHcy level in omega 3 treated patients at the end of study(P=0.03). Hemoglobin level was also increased significantly in omega 3 supplemented patients.
Conclusions: Omega -3 (fish oil) supplementation by reduction of tHcy may decrease the risk of cardiovascular morbidity and mortality.

Anaemia and ESA 2

Su562 IS NT-PROBNP A PREDICTOR OF CARDIOVASCULAR EFFECTS OF ANAEMIA TREATMENT WITH ERYTHROPOIETIN IN PREDIALYSIS CHRONIC KIDNEY DISEASE PATIENTS?

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Introduction and Aims: NT-proBNP is a recognized marker of left ventricular hypertrophy (LVH) in chronic kidney disease(CKD) patients. Erythropoietin(EPO) therapy in anemic CKD patients is associated with a reduction in left ventricular mass(LVM). It is unkown the effect of this therapy on NT-proBNP levels and if this peptide could be a potential marker of LVH regression in CKD patients treated with EPO.
Methods: Seventy-two predialysis CKD patients(stage 4 or 5) and hemoglobin levels less than 11 g/dL were treated during 6 months with EPO beta to achieve hemoglobin levels between 12-13 g/dL. The effect of correction of anaemia on LVM and regression of LVH on echocardiography was evaluated. Patients whose LVM index decreased by > 10% were considered to be "responders" and those whose LVM index increased or decreased by < 10% were considered to be "nonresponders". Levels of NT-proBNP and inflammatory markers(CRP and IL-6) were measured at baseline and at the end of the study.
Results: Forty-nine patients completed the study. Mean hemoglobin levels increased from 9.9±0.6 to 12.8±1.5 g/dl, p<0.0001, LVM index decreased from 69.2±17.7 to 64.1±19.6 g/m^2.7, p=0.01, whereas Lg Nt-proBNP,

CRP and IL-6 levels remainded unchanged. Patients with LVH showed higher levels of Lg NT-proBNP than patients without LVH(3.04± 0.5 vs 2.55±0.39, p=0.007, respectively) and there was an independent association between LVM index and Lg Nt-proBNP levels at baseline(r=0.45, p=0.002) and after 6 months of EPO therapy(r=0.38, p=0.01).
Twenty-five patients were "responders" and 24 "nonresponders" [mean reduction in LVM index(%) -21.9± 9.1 vs +9.1±18.3 respectively, p<0.0001]. Baseline characteristics of two groups were similar. At 6 months, "nonresponders" had higher systolic blood pressure (154.4±14.5 vs141.5±20.3 mmHg, p=0.01) and pulse pressure(74.3±14.1 vs 62.5±16.3 mmHg, p=0.01), but similar hemoglobin, Lg NT-proBNP, CRP and IL-6 levels. After adjusting for several factors, only systolic blood pressure at the end of the study independently predicted LVH regression (OR 0.937, 95% CI 0.886-0.991, p=0.02).
Conclusions: NT-proBNP levels are closely related to LVH in predialysis CKD patients. EPO therapy is associated with a reduction in LVM index, but had no effect on NT-proBNP levels. Systolic blood pressure, but not NT-proBNP, is an important determinant of change in LVM index induced by EPO treatment in these patients, and may contribute to the increased risk of progression of LVH.

Su563 HEPCIDIN SERUM LEVELS AND ITS RELATIONSHIP WITH HAEMATOLOGICAL DATA, IRON STATUS, INFLAMMATORY MARKERS AND rhEPO DOSES IN HAEMODIALYSIS PATIENTS

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Introduction and Aims: Recently, a complex regulatory network that governs iron traffic emerged, and point to hepcidin as a major evolutionary conserved regulator of iron distribution. The synthesis of hepcidin is regulate by anaemia/hypoxia, inflammation and iron overload. We sought therefore, to study the relationship between hepcidin serum levels and, haematological data, iron status, inflammatory markers and rhEPO doses in HD patients.
Methods: We have selected 33 HD patients (15 males, 18 females; mean age 59.5±17.6 years) under rhEPO treatment. The HD patients included 16 responders and 17 non-responders to rhEPO therapy. Healthy volunteers (n=17) were used as normal controls. Haematological data, iron status [iron, ferritin, transferrin, soluble transferrin receptor (s-TfR) and prohepcidin serum levels] and inflammatory markers [C-reactive protein (CRP) and IL-6 serum levels] were performed in all patients and controls. Serum hepcidin measurements were performed by a combination of weak cation exchange chromatography and time-of-flight mass spectrometry (TOF MS).
Results: Compared to controls, HD patients presented a significantly lower erythrocyte count, haemoglobin concentration, haematocrit and transferrin levels, and a significantly higher RDW, serum ferritin, s-TfR, CRP, IL-6, prohepcidin and hepcidin levels. Among HD patients, non-responders presented lower haemoglobin, haematocrit, and a higher RDW, s-TfR and CRP levels. No statistically significant differences on transferrin saturation were found between responders and non-responders patients. Prohepcidin serum levels among non-responders were significantly lower than among responders, but were higher than those in the control group. The same trend was found on hepcidin serum levels. A statistically significant correlation was found between hepcidin serum levels and some haematological data, mean cell volume, mean cell haemoglobin and RDW, iron status markers (ferritin and transferrin) and inflammatory markers (CRP and IL- 6). We also found a positive correlation between prohepcidin and hepcidin serum levels (r=0.624, p<0.0001).
Conclusions: Our data show that the high hepcidin serum levels, found in HD patients, are dependent of the magnitude of the inflammatory process, and of rhEPO doses. A close interaction between haematological data, inflammation, iron status and hepcidin serum levels, which ultimately regulate intracellular iron availability, were also found in our HD patients. Hepcidin seems to play significant role in anaemia of HD patients.

Su564 THE IMPACT OF FERRITIN CYCLING ON STABLE Hb LEVELS IN HEMODIALYSIS PATIENTS

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Introduction and Aims: It had been reported that anemia and hemoglobin (Hb) variability were associated with mortality on patients with maintenance hemodialysis (mHD), who are treated with erythropoiesis-stimulating agents (ESA). We hypothesized that iron store in patients with mHD was an important factor, which could be associated with Hb variability.
Methods: We evaluated the relationship between ferritin (Frn) variability and Hb stability. In the present study, 246 patients with mHD were recruited and measured Hb and Frn in each month. Standard Deviation (SD) and Residual (R) SD of Hb and Frn were calculated in each patient during 12 month period. For assessing the frequency and the size of fluctuations in Hb and Frn levels, these patients were classified into 6 groups on the basis of their overall Hb and Frn patterns of fluctuation, according to FAT methods.
Results: SD value of Hb was significantly correlated with that of Frn (p=0.0001, R=2.43), and dose of ESA (p<0.00001, R=0.39). High frequent patterns of Hb fluctuation were HA-Hb (36%) and LAL-Hb (41%), while those of Frn fluctuation were HA-Frn (45%) and LAL-Frn (21%). The ratio in target Hb was significantly (p=0.0052, R=0.18) correlated with that of Frn. The ratio of the patients maintaing target Hb in target-Frn group were significantly (p<0.05) higher than that of HA-Frn group.

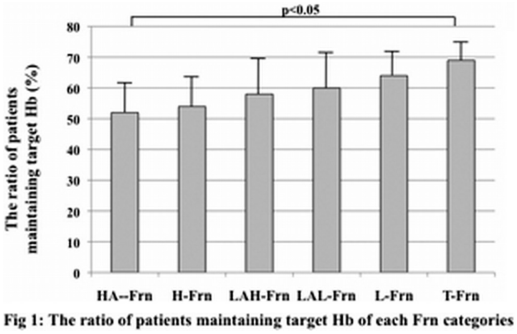

Fig 1: The ratio of patients maintaining target Hb of each Frn categories

28% patients in the target-Frn and 70% patients in Low-Frn were treated using oral iron. On the other hand, 45% patients in HA-Frn and 29% patients in High Frn were treated using intravenous iron.
Conclusions: Hb variability is associated with that of Frn in patients on mHD. Maintaining of stable iron store could be stabilized Hb levels in patients on mHD. Patients who can maintain iron store using oral iron therapy may have lower and stable ferritin levels.

Abstract Su564 – Table 1. Hb and Frn fluctuation classification

L-Hb	Consistently low Hb (<10 g/dL)	L-Frn	Consistently low Frn (<50 ng/mL)
LAL-Hb	Low amplitude fluctuation with low Hb (<10 g/dL)	LAL-Frn	Low amplitude fluctuation with low Frn (<50 ng/mL)
T-Hb	Consistently target Hb (10-11 g/dL)	T-Frn	Consistently target Frn (50-300 ng/mL)
LAH-Hb	Low amplitude fluctuation with high Hb (>11 g/dL)	LAH-Frn	Low amplitude fluctuation with high Frn (>300 ng/mL)
HA-Hb	High amplitude fluctuation Hb	HA-Frn	High amplitude fluctuation Frn
H-Hb	Consistently high Hb (>11 g/dL)	H-Frn	Consistently high Frn (>300 ng/mL)

Su565 CONVERSION OF DARBEPOETIN (DA) TO LOW DOSES OF C.E.R.A. MAINTAINS HEMOGLOBIN (Hb) LEVELS IN NON DIALYSIS CKD PATIENTS

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Introduction and Aims: In CKD, development and severity of anemia is influenced by several demographic and clinical features that also affect the treatment with erythropoietin stimulating agents (ESA) in terms of dose requirement and responsiveness to therapy. As emphasized by the results of recent trials, finding the lowest effective ESA dose is critical in the management of renal anemia. This holds particularly true in CKD where ESA are prescribed at lower doses than in dialysis patients. This study was aimed at evaluating efficacy of switching DA to C.E.R.A. at doses lower than those recommended for switch (<120μg/month).
Methods: We selected consecutive adult CKD patients treated with DA doses ≤40 μg/wk unchanged in the previous 3 months. We excluded patients with kidney transplant, iron deficiency, recent blood transfusion, bleeding, neoplasia, myocardial infarction or stroke in the last 3 months. Patients treated with DA ≤20 μg/wk were switched to C.E.R.A. 75 μg/month while those treated with DA 21-40 μg/wk were switched to C.E.R.A. 100 μg/month. After baseline, patients were evaluated monthly. Primary endpoint was the change in Hb level (target 11-13 g/dL) after 3 months and its maintenance at month 6, 9 and 12.
Results: Out of the 37 selected patients, 35 completed the study (one died at month-8 and one started HD at month-10). Mean age was 71±12 yrs and body weight was 73±17 kg; prevalence of males, diabetes and prior CV disease was 40.5%, 43.2% and 37.8%, respectively. DA dose before switching was 18±11 μg/wk with 65% of patients receiving ≤20 μg/wk. Prevalence of target and Hb values did not change at month 3 and were maintained during the study.

	Baseline	Month 3	Month 6	Month 9	Month 12
GFR mL/min/1.73m^2	31.0±10.9	31.4±12.3	28.6±11.4	29.1±11.7	28.5±10.2
Hb levels (g/dL)	11.7±1.1	11.9±1.3	11.5±1.0	12.0±1.0	12.1±0.9
Hb <11 n, (%)	5 (13.5)	6 (16.2)	9 (24.3)	6 (16.7)	4 (11.4)
Hb 11-13 n, (%)	28 (75.7)	26 (70.3)	26 (70.3)	26 (72.2)	28 (80.0)
Hb >13 n, (%)	4 (10.8)	5 (13.5)	2 (5.4)	4 (11.1)	3 (8.6)
C.E.R.A. dose (μg/month)	80±10	82±16	91±30	90±54	88±61
TSAT (%)	24.8±4.6	26.3±10.5	25.7±7.9	27.4±9.1	27.3±8.8
Ferritin (ng/mL)	173±75	165±100	169±99	192±104	179±125
Iron supplementation (%)	19 (51.4)	23 (62.2)	27 (73.0)	50 (55.6)	18 (51.4)

C.E.R.A. dose was unchanged in 350 out of 438 visits (80%), increased in 52 (12%) and reduced in 36 (8%) visits. The mean number of dose adjustment per patient was 2.38±1.72; specifically, 5 patients did not require any dose change while it was increased in 17 patients and reduced in 15. No patients required blood transfusions; three patients were hospitalized for angina, hyperglycemia and nephrotic syndrome, respectively. Blood pressure did not change; two patients had systolic >180 mmHg in a single occasion.
Conclusions: This study suggests that in CKD patients, switching from DA ≤40 μg/wk to C.E.R.A. doses (75 or 100 μg/month) lower than the recommended threshold is effective in maintaining Hb in target in absence of safety concerns and may reduce anemia-related costs.
Disclosure: Dr Minutolo received honoraria from Hoffmann-La Roche

Su566 HEMOGLOBIN STABILITY IN PATIENTS TREATED WITH EPOETIN ALFA UNDERGOING HEMODIALYSIS

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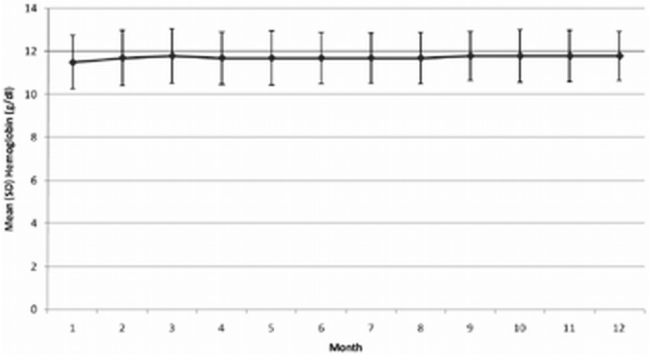
Introduction and Aims: To investigate intravenous epoetin alfa (ERYPO®) in terms of efficacy, and tolerability in patients with end stage renal failure (ESRF) undergoing hemodialysis.
Methods: Prospective, multi-center, non-interventional study. Patients undergoing hemodialysis and treatment of anemia with epoetin alfa under the discretion of the treating physician were to be included.
Results: 1630 patients from 60 centers were documented, 1480 patients with at least 1 month of documented treatment were included in the full analysis set (FAS). Mean treatment duration within the study was 320.6 (± 95.79) days. Mean dose of epoetin alfa at the beginning of the study was 7551.8 iU/w, decreasing to 7518.9 iU/w at the end of the study. The proportion of patients with stable Hb values (≥50% of Hb values between 10 and 12 g/dL) was 57.0% in the FAS population. The mean Hb value was 11.5±1.25 g/dL at month 1 and 11.8±1.14 g/dL at month 12. 7.0% of patients received blood transfusions. At study start, 91.1% of 926 FAS patients with data on iron metabolism available met the minimal criteria and 76.2% met the optimal criteria for adequate iron stores according to the European Best Practice Guideline for Anemia Management.
Figure 1: Time course of mean (SD) hemoglobin values (g/dL) – all patients.



Safety: In total, 361 of 1,630 patients (22.1%) experienced 777 AEs. For 29 AEs (3.7%) in 13 patients, a causal relationship to treatment with ERYPO® was assessed as at least possible by the investigator, most frequently being shunt occlusion, increased hematocrit and increased hemoglobin. No new safety signals have been detected.
Conclusions: With ERYPO® stable Hb values can be achieved in a majority of patients with ESRF.
Disclosure: Employment (Janssen-Cilag). This study has been conducted by Janssen-Cilag GmbH, Neuss, Germany.

Su567 INFLUENCE OF THE USE OF VITAMIN D ANALOGS IN THE ERITRHOPOIETIC RESPONSE TO C.E.R.A. IN PATIENTS NOT ON DIALYSIS PREVIOUSLY TREATED WITH EPO

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Introduction and Aims: The aim of this study is to determine the percentage of patients needing increasing doses of C.E.R.A. (Methoxi polyethylene glycol-epoetin beta) after switching from β-epoetin EPO (C.E.R.A once a month, EPO once a week), and its relation with the use of vitamin D analogs (VDA), regularly indicated for secondary hyperparathyroidism.
Methods: A retrospective and observational study conducted at our pre-dialysis unit. 40 consecutive patients were selected. 5 patients were excluded (2 not compliant with C.E.R.A. prescription, 1 died in the first month and 2 started on dialysis treatment prior to the end of the observational period). The follow-up period was 9 months. We evaluated the patients 2 times during the period of EPO use (6 and 3 months prior to the change to C.E.R.A.) and 3 months after starting treatment with C.E.R.A. Inclusion criteria: Age ≥ 18 years. GFR (Glomerular Filtration Rate) ≤ 60 ml/min/m^2. Anemia for renal reasons. All patients were on treatment with EPO at least 12 weeks previous to changing to C.E.R.A. Adequate levels of Transferrin Saturation Index (TSI) and Ferritin. Exclusion criteria: Symptomatic heart failure. Infections or acute inflammation. Bled assets or transfusion two months before the beginning C.E.R.A.
Results: 69% were men (mean age 71.0 years ±13). 40% had interstitial nephropathy, 37% nephroangiosclerosis, 17% diabetic nephropathy and the rest miscellaneous diseases. Mean ferritin level was 167mg/dl (SD=125) and mean TSI was 23% (SD=9). Average initial C.E.R.A. dose was 84 mcg (SD=34), and 3 months after change was 102 mcg (SD=51). The mean Hb levels during the observed period were: 11.6 mg/dl (SD=1.1), 11.7 mg/dl

(SD=1.2) and 11.8 mg/dl (SD=1.0) -6 -3 and + 3 months respectively. 21 (60%) patients did not require dose increase of C.E.R.A. while 14 (40%) of patients needed dose increase. The need for dose increase was the only observed difference between the groups. All other parameters were not significantly different (GFR: 22±5 versus 20±7 ml/min/1.73m^2, Ferritin: 162±130 versus 176±121 ng/ml, Transferrin 214±41 versus 229±50 mg/dl, TSI%= 21% versus 24% and iPTH (intact Parathyroid Hormone) 188±125 versus 212±176 pg/ml. 51% of patients on treatment with vitamin D analogs did not require dose increase of CERA as compared to 9% of patients not on vitamin D treatment that required the increase in dose (p=0.011).

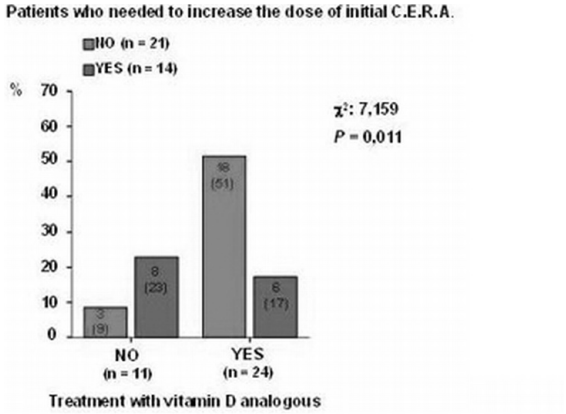

Figure 1. Patients who needed to increase the dose of inicial C.E.R.A. in relation with the use of vitamin d analogs.

Conclusions: We suggest that VDA use favors erythropoietic response to C.E.R.A. in CKD patients not on dialysis.
This erythropoietic response seems to be independent to the levels of iPTH in the patients.

Su568 GENOTOXIC ASSESSMENT AND TOXICITY EVALUATION OF HEMATIDE™ (PEGINESATIDE) IN CByB6F1 HYBRID MICE

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Introduction and Aims: Peginesatide is a synthetic, PEGylated, dimeric peptide-based erythropoiesis stimulating agent (ESA) in clinical development for the treatment of chronic kidney disease (CKD)-associated anemia. Clinical use of peginesatide will result in chronic dosing and the nonclinical data to support development should consist of carcinogenic potential. The objectives of this study were to evaluate the genotoxicity of peginesatide and to provide data for subsequent design of a pivotal 6-month rasH2 transgenic study based on a safety and PK study in CByB6F1 Hybrid Mice.
Methods: Peginesatide was assessed in a standard genotoxicity battery. Doses for a rasH2 transgenic mouse carcinogenicity assay were defined in a 28-day study in the wild type CByB6F1 parental mouse strain administered peginesatide by intravenous (IV) injection on Days 1 and 22 at 1 to 25 mg/kg.
Results: Peginesatide did not increase the number of revertants in the AMES bacterial reverse mutation assay, with or without metabolic activation and, therefore, is non-mutagenic. Peginesatide was not clastogenic in an *in vitro* chromosomal aberration assay and was negative in the murine micronucleus assay. The CByB6F1 hybrid mice findings were consistent with exaggerated pharmacology including polycythemia with associated increases in hemoglobin level and extramedullary hematopoiesis and bone marrow hypercellularity.
Conclusions: Peginesatide was not mutagenic or clastogenic in a standard genotoxicity battery. Toxicologic findings in CByB6F1 hybrid mice findings were related to the exaggerated pharmacology (polycythemia) that occurs with administration of an ESA to a normocythemic animal.
Disclosure: Dr. Woodburn is an employee and stockowner of Affymax, Inc.

Su569 ACQUIRED RENAL CYSTS PREDICT LOWER ERYTHROPOIETIN REQUIREMENTS IN DIALYSIS PATIENTS

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Introduction and Aims: Identification of predictors of hyporesponsiviness to erythropoietin-stimulating agents (ESA) in hemodialysis helps improving anemia management and reduces hemoglobin (Hb) level variability. ESA requirements show substantial inter and intra-patient variability. This study aimed to assess the associated factors with the ESA dose requirements in dialysis patients maintaining stable hemoglobin levels.
Methods: Short – acting ESA were used in anemia management. ESA dosage (defined as mean weekly ESA dose per kg body weight) was assessed in 110 stable dialysis patients maintaining Hb levels above 10.0g/dL, within 6 Hb measurements during one year. Patients on dialysis less than one year, suffering from acute infections and bleeding episodes were excluded from the study. Pertinent laboratory, demographic, dialysis and medical history data were recorded. Presence of acquired renal cysts by ultarsonography prior to the study was evaluated in all patients. Univariate and multivariate analysis were performed.
Results: 110 dialysis patients with mean age 57±13 years and dialysis vintage 117±88 month were evaluated. Presence of secondary renal cysts was found in 10 (9.1%) of patients, 7 patients (6, 4%) were receiving angiotensin converting enzyme (ACE) inhibitors as antihypertensive therapy. In the univariate analysis the male gender, lower age of participants and lower ferritin levels correlated positively with higher ESA dosage, but lost significance in the multivariate model. The use of ACE inhibitors as antihypertensive therapy was positively but insignificantly correlated with ESA dosage (r=0,169, p=0.077). The multivariate analysis showed that higher ESA dosage was associated with presence of lower urea reduction rate, (β=0.212, p=0.000), higher dosage of iron (β =0.295, p=0.001), higher phosphorous levels (β =0.216, p=0.002) and higher CRP (β =0.361, p=0.002). Presence of Adult polycystic disease and secondary renal cysts independently predicted lower ESA dose requirements (β=-0.286, p=0.000), (β=-0.340, p=0.000), respectively.
Conclusions: Dialysis adequacy, inflammatory and iron status are of extreme importance of anemia management in dialysis patients. Patients prone of secondary renal cysts formation require significantly lower doses of erythropoietin in maintenance of Hb levels.

Su570 HEMOGLOBIN VARIABILITY. TREATMENT DIFFICULTY IN ANAEMIA MANAGEMENT OF HAEMODIALYSIS PATIENTS

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Introduction and Aims: The target Hgb level according to the anaemia management guidelines is 110-125 g/l. Beside achieving this target level it is also very important to maintain the patients' Hgb values (HBV) between these limits to avoid the Hgb fluctuation. It was observed in several studies that only a small part of patients' Hgb level is stable during the months, the greater fraction of patients' Hgb values oscillate with low-frequency or high-frequency in amplitude.
Our aim was to determine the Hgb level and erytropoisesis stimulating agent (ESA) dose stability in our haemodialysis patients in a 12 months follow up term.
Methods: We performed a 12 months duration retrospective analysis in our 101 chronic haemodialysis patients' anaemia control. The patient mean age was 67 year, male: female=46:55. We were interested in whether is there any correlation concerning Hgb fluctuation and erythropoietin dose change necessity and major medical events (hospitalisation). The patients were divided in 6 groups:
1. consistently low (CL)-Hgb: <110g/l;
2. consistently high (CH)-Hgb: >125g/l;
3. low- amplitude fluctuation with cross the lower level (LAL)-Hgb: <110-<125g/l;
4. low- amplitude fluctuation with cross the upper level (LAH)-Hgb: 110->125 g/l;

5. high-amplitude fluctuation with cross the lower and upper target Hgb level (HAF)-Hgb:<110->125g/l;
6.target range (TR)group whose HBV remained in limits.
The standard deviation (SD) of HBV and ESA dose IU/kg weight/week (IU/kg/w) was determined
Results: 9% of our patients' HBV remained between the target range, 29% of them crossed the upper limit, 23% of patient's HBV fall down below the target range, and a group of 34% of patients showed large fluctuations in HBV, so they crossed both the upper and the lower limit of the recommended HBV. Only 3% belonged to the CH group and merely one patient's HBV were measured consistently below the target lower limit.
The HBV oscillation determined with standard deviation was highest in the HAF patients, and the lowest was in TR group. SD-TR=4; SD-LAL=6.6; SD-LAH=6.5; SD-HAF=12. $p<0.001$.
The mean ESA dose didn't show statistically significant difference in the groups with oscillated HBV and the group with stable HBV. TR=83IU/kg/w; LAL=122IU/kg/w; LAH= 64IU/kg/w; HAF=94IU/kg/w.
We found statisticaly significant difference in SD of ESA dose between the TR group and groups with fluctuated in HBV: LAL, LAH, HAF
TR/LAL: 9/17, p=0.02; TR/LAH: 9/15, p=0.04; TR/HAF: 9/22, p=0.0004
Our result concerning the hospitalisation:
In stable HBV patients (TR) one event was occurred per 40 patient treatment month (PTM), witch mean duration (MD) was 7 days.
In patients with oscillated HBV:
LAH: 29 month/event, MD=8 days; LAL: 18 month/event with 10 days MD;
HAF: 10 mont/event, MD=10 days
Conclusions: Conclusion: Not only important but also difficult to control the dialysis patient's anaemia such way as it is recommended in the anaemia management guidelines. Only 9% of our patients was managed to avoid crossing their HBV the target limits. The remained 91% of patients was in some degree of flux at any point in time and this fluctuation was highly associated with ESA dose change measure and necessity of hospitalization.

Su571 HIGH DOSE INFUSION OF IRON ISOMALTOSIDE 1000 (Monofer®) IN CKD AND CHF PATIENTS

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Introduction and Aims: Patients with chronic kidney disease (CKD) and iron deficiency anaemia frequently require treatment with intravenous (IV) iron preparations. Concomitant renal dysfunction is often seen in patients with chronic heart failure (CHF) and IV iron preparations have shown clinical benefit in patients with CHF. Administration of single high parenteral iron doses are particularly convenient and cost beneficial as they offer the possibility of making the patients iron replete in one single dose. This is only feasible if the iron preparation has a very low risk of free-iron related toxicity. Iron isomaltoside 1000 is a novel IV iron compound with a low risk of free-iron related toxicity. The carbohydrate isomaltoside 1000 has low immunological activity and a test dose is not necessary. The present study analyses the subgroup of patients receiving high dose infusion (defined as Total Dose Infusion (TDI) in the protocol) in 2 previously presented open-label, multicentre iron isomaltoside 1000 safety studies.
Methods: Iron isomaltoside 1000 was administered as a TDI at baseline to 40 patients with CKD and to 20 patients with CHF, both groups having iron deficiency anaemia. No initial test dose was given. The patients attended post dosing visits after 1, 2, 3, 4 and 8 weeks for safety assessments and for biochemical assessments of treatment effects. QoL was assessed on a Linear Analogue Scale Assessment in the CHF population.
Results: The mean infusion time was 59 minutes (range: 20-90 minutes) and the mean iron dose was 975.3 mg (range: 462-1800 mg) in the CKD patients and 868.3 mg (650-1000 mg) in CHF patients. 58 out of the 60 patients had a calculated iron need of less than 20 mg/kg and had their iron need covered in one dose. Only 2 CKD patients required 2 divided doses to fulfil their iron needs. One solitary treatment related adverse reaction was observed. This was a case of angina pectoris in an 80 years old male CKD patient with a medical history of angina pectoris. The reaction was classified as serious and possible related to the trial medication by the investigator, but it occurred 10-11 days after the patient had received 1400 mg iron isomaltoside 1000, and the SAE could be explained by the patient's medical history. No acute anaphylactoid/anaphylactic or delayed allergic reactions were observed. There were no clinically significant changes in routine clinical safety laboratory tests or vital signs. The mean change from baseline at week 8 was 9.8 g/L for Hb and 191.7 µg/L for s-ferritin in the CKD patients and 4.4 g/L for Hb and 216.8 µg/L for s-ferritin in the CHF patients. All QoL scores assessing "energy level", "ability to do daily activities", and "overall QoL" were significantly improved at 4 weeks from baseline.
Conclusions: Iron isomaltoside 1000 administered IV in high doses was efficacious, safe and well tolerated and allows iron repletion in one dose in both CKD and CHF patients.
Disclosure: This study has been sponsored by Pharmacosmos A/S.

Su572 THE EFFECT OF INTRAVENOUS IRON THERAPY ON OXIDATIVE STRESS PARAMETERS IN PATIENTS ON HAEMODIALYSIS

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Introduction and Aims: Iron balance is critical for adequate erythropoiesis and there remains much debate concerning the optimal timing and dosage of iron therapy for haemodialysis patients receiving recombinant human erythropoietin (rHuEpo) therapy. Accumulating evidence suggests that oxidative stress is enhanced in patients on regular hemodialysis (HD). The causes of oxidative stress in HD patients are still controversial. Beside the uraemic state and dialysis-related factors, adjuvant drug therapies such as rHuEpo and intravenous iron were involved.
In this study, we examined the influence of intravenous infusion of 100mg iron sucrose on the oxidative status (OS) of the patients on maintenance haemodialysis.
Methods: We investigated 139 patients on chronic hemodialysis who were randomized to receive either rhEPO alone (N = 72) or rhEPO in combination with intravenous iron (N = 52) and 15 healthy controls. Plasma and red blood cells (RBC) thiobarbituric acid-reactive substances (TBARS) as well as reactive carbonyl group (RCG) in plasma were used as markers of reactive species generation. The levels of total antioxidative capacity-TAOC were determined in plasma.
Results: The OS markers of HD patients did not differ, whether or not they received intravenous iron supplementation. Compared with controls, the HD patients had higher serum urea, creatinine, CRP, RBC and plasma TBARS ($p<0.001$), RCG ($p<0.001$) and lower albumin, Fe and TAOC values ($p<0.01$).
Conclusions: Our results suggest that (1) chronic HD patients appear to have simultaneously enhanced reactive species generation and antioxidative systems efficiency, and (2) intravenous iron therapy did not change their oxidative status.

Su573 MIRCERA CORRECTS ANEMIA IN KOREAN PATIENTS WITH CHRONIC KIDNEY DISEASE ON DIALYSIS: RESULTS FROM A RANDOMIZED CONTROLLED MULTICENTER STUDY

Jieun Oh[1], Kwon-Wook Joo[2], Ho-Jun Chin[2], Dong-Wan Chae[2], Sung-Gyun Kim[1], Woo-Kyung Chung[3], Wooseong Huh[4], Ha-Young Oh[4], Chul-Woo Yang[5], Sunhngwon Kim[2]. *[1]Internal Medicine, Hallym University College of Medicine, Seoul, Korea; [2]Internal Medicine, Seoul National University College of Medicine, Seoul, Korea; [3]Internal Medicine, Gachon University College of Medicine, Incheon, Korea; [4]Internal Medicine, Sungkyunkwan University School of Medicine, Seoul, Korea; [5]Internal Medicine, The Catholic University of Korea College of Medicine, Seoul, Korea*

Introduction and Aims: This study investigated the efficacy of Mircera,

a continuous erythropoietin receptor activator, for correcting anemia in patients with chronic kidney disease on dialysis. In addition, long-term safety and tolerability of Mircera was assessed for the subsequent 24 weeks of the extension period.
Methods: In this open-label, randomized, multi-center, parallel group, phase III study, eighty patients(≥ 18 yr) who were not receiving any erythropoietic stimulating agents for more than 8 weeks were randomly assigned (1:1) to either intravenous Mircera once every 2 weeks (n=39) or epoetin beta thrice-weekly (non-comparative control, n=41) during a 24-week correction period. The target hemoglobin during the correction phase was within the range of 11 to 13 g/dL and an increase in Hb from baseline ≥ 1 g/dL without RBC transfusion. Patients who achieved the target in both groups were assigned to Mircera once monthly for a 24-week extension period.
Results: The primary end point was Hb level response rate (increase in Hb level >or=1g/dl and Hb level >or=11g/dl without RBC transfusion) in the intent-to-treat population. Hb response rates were 79.5% and 87.8% for Mircera and epoetin beta, respectively, and it showed that Mircera once every 2 weeks was as effective as epoetin beta for correcting anemia. The time to response indicated that the correction of anemia was slower in Mircera group compared with epoetin beta group. Median time to response was approximately 12 weeks in Mircera and 10.3 weeks in epoetin beta. The time to response in both groups is relatively long with Korean patients. Mircera and epoetin beta were well tolerated.
Conclusions: This study demonstrates that once every 2 weeks intavenous administration of Mircera corrects anemia effectively and safely in patients with CKD on dialysis.
Disclosure: Supported by Roche

Su574 CONTINUOUS STABILIZATION OF HB VALUE IN CKD PATIENTS REQUIRING DIALYSIS: RESULTS FROM THE GERMAN OBSERVATIONAL KONTINUE TRIAL

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Introduction and Aims: The objective of this ongoing observational study (KONTINUE: Survey to assess continuous achievement of Hb target range under long-acting erythropoietic stimulation with Aranesp® in the treatment of renal anemia in dialysis patients) is to investigate the continuous maintenance of the Hb value in patients with chronic kidney disease undergoing dialysis in routine therapy.
Methods: This multicenter observational study recruited 1254 patients from 53 German sites. At baseline, ESA therapy was converted from rHuEPO to either weekly (QW) or biweekly (Q2W) therapy with darbepoetin alfa (DA), administered by either the sc or iv route. Data was collected 3 months (mo) prior to and 21 mo after conversion. This interim analysis evaluates all patient data available at the end of October 2009 in the evaluable analysis set (exclusion from EAS: DA treatment prior to BL, <2 retrospective Hb values, <6 prospective Hb values mo 4-12, no dialysis treatment before or at baseline, no ESA pretreatment) of patients who had reached the time point mo 12.
Results: A total number of 53 dialysis sites contributed 739 patients to this interim analysis. 572 of those patients (53% male, mean age 67.2 years) were included in the evaluable analysis set. The reported mean duration of underlying nephrological disease was 77.7 months. 36% of patients had diabetic nephropathy and 17% hypertensive nephrosclerosis. After conversion the mean weekly dose of DA decreased from 40.6 µg at baseline to 35.0 µg at mo 12. 86.6% of patients received 0 – 4 dose adjustments during the period from mo 4 to mo 12, where 22.6% of the patients needed no dose adjustment at all. 13.4% of patients received 5 or more dose adjustments during the time after conversion. Hb value minorly changed from 11.60 g/dl at mo -3 (95% CI: 11.49, 11.72) to 11.55 g/dl at baseline (95% CI: 11.45, 11.65) and to11.76 g/dl at mo 12 (95% CI: 11.65, 11.87). At mo 12, a total of 85% of patients has a Hb level ≥ 10 g/dl and 68% of patients had a Hb ≥ 11 g/dl. From mo 4 to 12, the mean time with Hb ≥ 11 g/dl was 76% (median 85%).
Conclusions: This interim analysis reveals that in patients with chronic kidney disease and renal anemia, who are on dialysis, therapy with darbepoetin alfa QW or Q2W, administered either by the sc or iv route, offers an effective Hb control in a heterogeneous dialysis population.
Disclosure: This study was sponsored by Amgen.

Su575 COMPLIANCE WITH GUIDELINES ON TREATMENT OF ANAEMIA IN A COHORT OF CHRONIC HAEMODIALYSIS (HD) PATIENTS

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Introduction and Aims: Recommendations and targets in the treatment of anaemia in patients with chronic kidney disease (CKD) are described in the revised European Best Practice Guidelines (EBPG) for Management of Anaemia in Patients with Chronic Kidney Disease (2004) and the KDOQI guidelines on Anaemia in CKD (2006 and 2007). The goal of the present analysis it to describe to what extent target levels as mentioned in these guidelines are reached in a cohort of chronic HD patients.
Methods: This analysis is based on baseline data from consecutive patients included in the CONvective TRAnsport STudy (CONTRAST; NCT00205556) from 26 centers (24 Dutch, 1 Norwegian and 1 Canadian). In this trial, chronic HD patients who are treated 2 or 3 times per week for at least 2 months and have a spKt/V > 1.15 were included. Levels of haemoglobin (Hb), ferritin and transferrin saturation (Tsat) and data on use of erythropoiesis stimulating agents (ESA) and iron supplementation were collected. The present analysis is not adjusted for case mix variables.
Results: 448 patients were included in the analysis (63% male, age 63.3±13.9 [mean ± SD]). 90% of all patients used ESA and 69% used intravenous iron supplementation. 83% had a Hb level ≥ 6.8 mmol/L, whereas 14% and 20% had a Hb level of > 8.6 (EBPG upper limit) and > 8.0 mmol/L (KDOQI upper limit) respectively. Only in a minority of patients, all EBPG or KDOQI treatment targets were met (table). Furthermore, a marked difference between centers in the percentage of patients reaching all treatment targets was observed: for the EBPG 2-52% (median 14%) and for KDOQI 0-29% (median 14%).

Fulfilment of EBPG and KDOQI targets

EBPG targets	% of patients meeting EBPG targets	KDOQI targets	% of patients meeting KDOQI targets
Hb 6.8-8.6 mmol/L	79	Hb 6.8-7.4 mmol/L	34
Tsat ≥ 20%	59	Tsat ≥ 20%	59
Ferritin 200-500 µg/L	38	Ferritin ≥ 200 µg/L	69
Fulfilment of all targets	19	Fulfilment of all targets	15

(Hb = haemoglobin, Tsat = transferrin saturation)

Conclusions: In a cohort of 448 chronic HD patients, 83% had a Hb of ≥ 6,8 mmol/L. Achievement of all target levels as recommended by EBPG and KDOQI guidelines on management of anaemia and iron status was relatively low (< 20%) with a wide variation between centers.

Su576 DETECTION OF COLORECTAL CANCER AND OTHER LESIONS IN PREDIALYSIS PATIENTS WITH ANAEMIA AND A POSITIVE FECAL OCCULT BLOOD TEST

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Introduction and Aims: Colorectal cancer is a major health problem; it is the second most common cancer diagnosis after breast cancer for women and the third in men following prostate and lung cancer. Screening programmes have been establish since they reduce disease-related mortality. Because chronic kidney disease may be associated with multifactorial anaemia in

predialysis patients, we hypothesized that a positive fecal occult blood test (FOBT) result should be evaluated for colonoscopy.
Aims: To determine the value of FOBT for the screening of colorectal cancer in predialysis patients.
To analyse the colonic lesions in predialysis patients who have a Hb <11 g/dl or a TSI <20% with a positive FOBT.
Methods: We prospectively identified 164 predialysis patients who had a Hb <11 g/dl or a TSI <20%. Those who had a positive FOBT were referred to a gastroenterologist to evaluate the need for a endoscopic study (gastroscopy and/or colonoscopy).
Results: Mean age was 70.8 (±11.4). There was no difference in gender. Mean MDRD was 37.6 ml/min (± 19.5). Clinically important lesions were identified in 25.6% of 138 individuals. Prevalences were: adenomas of 1 cm or greater (22.7%), carcinomas (3%), gastric ulcers (2.4%), vascular ectasias (2.4%). Colonic lesions were not related to the severity of chronic kidney disease.
Conclusions: FOBT is a useful tool for the screening of colorectal cancer in predyalysis patients with anaemia. These patients may be assymptomatic but have gastric/colonic lesions that can be treated by endoscopic studies.

Su577 EVALUATION OF ONCE-MONTHLY HEMATIDE™ (PEGINESATIDE) DOSING REQUIREMENTS AFTER HOSPITALIZATION IN HEMODIALYSIS PATIENTS

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Introduction and Aims: It has been estimated that nearly 90% of HD patients in the US do not receive treatment with ESAs during hospitalization. Nontreatment may contribute to a decline in Hb concentration, which may lead to an ESA dose increase after hospital discharge. Peginesatide is an investigational peptide-based ESA that was designed and engineered to specifically interact with the erythropoietin receptor that governs erythropoiesis. Peginesatide is in phase 3 development for the treatment of anemia associated with CKD. Peginesatide dose requirements in HD patients after hospitalization were evaluated.
Methods: These post hoc analyses included events of hospitalization pooled from 2 open-label extension studies that occurred in HD patients who received once-monthly treatment with peginesatide for up to 34 months (data available through July 17, 2009); the Safety Population included all patients. Events of hospitalization were excluded if they occurred within 2 months of another hospitalization. Peginesatide dosing requirements and Hb levels at prehospitalization (the most recent value recorded before hospitalization) and months 1 and 2 posthospitalization were evaluated for each hospitalization. These data were compared with data from an epoetin alfa study (Yaqub et al. *Am J Nephrol.* 2001).
Results: The Safety Population included 183 patients; 107 instances of hospitalization that occurred in 62 patients were included. Median duration of peginesatide exposure: 26.6 months (hospitalized patients) and 20.4 months (Safety Population). Compared with prehospitalization values, mean peginesatide dose requirements increased less after hospitalization in the current study (month 1, 9%; month 2, 10%) than they did in the Yaqub et al study with epoetin (month 1, 31%; month 2, 45%). Similarly, compared with prehospitalization values, mean Hb values changed less after hospitalization in the current study (month 1, –0.1 g/dL; month 2, 0.1 g/dL) than they did in the Yaqub et al study with epoetin (month 1, –0.7 g/dL; month 2, –0.6 g/dL). In the Safety Population, 19 patients (10%) experienced AEs that were considered possibly treatment related; those that occurred in >1 patient were hypertension (n=4), IV line clotting (n=2), and headache (n=2). Four patients (2%) experienced SAEs that were considered possibly treatment related (embolic cerebral infarction [n=1], pulmonary embolism [n=1], deep vein thrombosis [n=1], and hypertension [n=1]).
Conclusions: Dose requirements and Hb appear to change very little after hospitalization in once-monthly peginesatide-treated patients. When these results were compared with derived data from a published study with epoetin, it appears that, after hospitalization, peginesatide-treated patients may exhibit less of a decline in Hb and require less of a dose increase than do epoetin-treated patients. Reasons for these observations and their clinical significance warrant further investigation; controlled studies that compare once-monthly peginesatide with other ESAs are ongoing.
Disclosure: Dr Wiecek has received grants/research support and honoraria from Janssed-Cilag, Amgen, and Fresenius and is a consultant for Affymax Inc. and Astellas.

Su578 RELATIONSHIP BETWEEN RED BLOOD CELL LIFESPAN AND INFLAMMATORY CYTOKINE LEVELS IN CHRONIC HEMODIALYSIS PATIENTS

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Introduction and Aims: Iron and erythropoietin (EPO) deficiency are well characterized causes of anemia in CKD. Less well appreciated is the reduction of red blood cell lifespan (RBCLS) in uremic patients as an important contributor to renal anemia and possibly to EPO resistance. Our aim was to test the hypothesis that RBCLS is positively associated with pro-inflammatory cytokines, and negatively with anti-inflammatory cytokines in chronic hemodialysis (HD) patients.
Methods: Non-smoking patients without inflammatory pulmonary disease receiving maintenance HD were recruited. At two study visits per patient, spaced approximately 1 month apart, carbon monoxide (CO) concentration was measured in alveolar and environmental air samples with a 910 Buck Scientific gas chromatograph (Buck Scientific, East Norwalk, USA). RBCLS was computed from endogenous CO production (Strocchi, 1992). Pre-dialysis EDTA-plasma was aliquoted and frozen at –70°C. Plasma levels of IL-1β, IL-6, IL-10, IFN-γ and TNF-α were measured in duplicate using the xMAP technology on a Luminex100™ system (Luminex Corp., Austin, USA). Pearson correlation was analyzed, and comparisons between the study visits were performed by paired samples t test.
Results: Twenty-two patients (15 males; mean age 56.0±13.8 years; 14 Blacks, 6 Hispanic, 2 Caucasians) were enrolled. RBCLS and cytokine measurements were available for all patients on the 1st study visit and for 12 patients on the 2nd study visit. The mean RBCLS was 61.6±16.7 days (35.9 to 90.2 days). No correlation between RBCLS and cytokine levels was found for either the 1st or the 2nd visit.

Pearson correlations between RBCLS and cytokines at 1st visit

	IL-1β	IL-6	IL-10	IFN-γ	TNF-α
RBCLS	r= -0.24 r= 0.29 P=0.29	r= -0.31 P=0.16	r= -0.11 P=0.61	r= -0.10 P=0.66	r= 0.41 P=0.06

Neither RBCLS nor cytokine concentrations changed significantly between the 1st and 2nd study visit (P>0.05; paired t test). Change in RBCLS between visits did not correlate with change in cytokine levels.

Pearson correlations between ΔRBCLS and Δcytokines (N=12)

	ΔIL-1β	ΔIL-6	ΔIL-10	ΔIFN-γ	ΔTNF-α
ΔRBCLS	r= 0.17 P=0.59	r= 0.40 P=0.20	r= -0.36 P=0.25	r= 0.16 P=0.62	r= -0.34 P=0.28

Conclusions: In a group of 22 chronic HD patients, we found no correlation between RBCLS and pro-inflammatory (IL-1β, IL-6, IFN-γ and TNF-α) or anti-inflammatory (IL-10) cytokines over a wide range of RBCLS. Moreover, when studied longitudinally, changes in RBCLS were unrelated to changes in cytokine concentrations. These results indicate that shortened RBCLS in uremia is likely to be independent of inflammation. Other characteristics of the uremic milieu, such as oxidative stress or increased levels of uremic toxins, might be related to shortened RBCLS in HD patients.

Su579 ANALYSIS OF HAEMOGLOBIN (Hb) LEVELS DURING INTERCURRENT EVENTS IN PATIENTS WITH CKD RECEIVING C.E.R.A. OR OTHER ESAs

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Introduction and Aims: Clinical management of patients (pts) with chronic

kidney disease (CKD) receiving erythropoiesis-stimulating agent (ESA) therapy can often be complicated by acute events such as intercurrent infections, inflammatory or traumatic episodes. A physician's first response may be to alter the ESA dose or switch to another ESA, rather than treat the acute pathological events. We analysed the maintenance Phase III trial population who experienced acute events while receiving either methoxy polyethylene glycol-epoetin beta, the continuous erythropoiesis receptor activator (C.E.R.A.), every 2 weeks (Q2W) or once monthly (Q4W) or a comparator ESA (epoetin alfa, epoetin beta, darbepoetin alfa).
Methods: Data from 4 maintenance Phase III trials of pts who experienced acute pathological events while receiving C.E.R.A. Q2W or Q4W were compared with pooled data on pts who received a comparator ESA. For gastrointestinal (GI) bleeding, inflammation and infections, mean Hb levels after the event, change in Hb levels over time, and time to return to Hb target levels were calculated separately for the pooled population. The lowest Hb value (nadir) reported was taken within ±2 weeks of the onset of an event, and only events with an Hb nadir <10.0 g/dL were used. Pts receiving a blood transfusion during the time of the event were excluded.
Results: A total population of 1879 pts were assessed, with 1139 pts receiving C.E.R.A. and 740 pts a comparator. There were 45 GI bleeding events reported in the C.E.R.A. group vs 21 in the comparator group, with pts experiencing a mean Hb nadir of 8.14 g/dL and 8.13 g/dL, respectively. It took a median of 10 days (C.E.R.A.) and 7 days (comparator) for pts to return to an Hb level ≥10.0 g/dL, with a mean Hb >11.0 g/dL reached after 10 weeks vs 14 weeks, respectively. For infections and infestations 275 (C.E.R.A.) vs 153 events (comparator) were reported with a mean Hb nadir of 8.91 g/dL and 8.93 g/dL, respectively. Hb returned to levels ≥10.0 g/dL after a median time of 12 days (C.E.R.A.) and 14 days (comparator), with a mean Hb >11.0 g/dL achieved after 9 weeks for both groups.
Conclusions: The data show that continued treatment with C.E.R.A. Q2W or Q4W allows Hb goals to be achieved while pts receive treatment for intercurrent events. These results suggest that a prompt search for and treatment of causative factors of Hb declines in pts with CKD, rather then an immediate response to alter dose or change to another ESA, should be employed.
Disclosure: Grant/research support: Roche. Honoraria: Roche, Guerbet, Astrazeneca

Su580 HEMOGLOBIN CYCLING AND VARIABILITY IN MAINTENANCE HEMODIALYSIS PATIENTS TREATED WITY ERYTHROPOIESIS STIMULATING AGENTS: ASSOCIATIONS WITH CLINICAL CHARACTERISTICS

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Introduction and Aims: Retrospective investigations were performed to clarify the difference of hemoglobin (Hb) cycling and the relationship between Hb variability and clinical characteristics in patients undergoing maintenance hemodialysis (HD) treated with two different erythropoiesis stimulating agents (ESA).
Methods: Data were analyzed for 213 patients observed from January to December, 2009. All patients were treated with intravenous administration of either recombinant human erythropoietin (EPO: 750−9000 units/week) or darbepoetin (DPO: 10−80μg/week) in order to maintain the target Hb level between 10.0 to 12.0 g/dL. The 213 cases were divided into two groups according to the ESA: Group A (EPO: n=95, M:F=65:30, age=62.1±15.7 years old, HD duration=82.1±66.9 months) and Group B (DPO: 118, 84:34, 65.1±12.1, 65.1±49.9). The weekly average values of clinical markers such as albumin (Alb), high sensitive CRP (hsCRP), calcium (Ca), phosphorus (P) and Hb were measured at 2−day intervals just before HD, and the weekly average doses of ESAs were also examined. The frequency of Hb cycling (cycles with amplitude over 1.5 g/dL) and variability were investigated in the two groups, with variability classified according to the following categories as reported by Ebben, et al (Clin J Am Soc Nephrol 1:1205-10, 2006): consistently low (L), consistently within the target range (T), consistently high (H), low amplitude low (LAL), low amplitude high (LAH) and high amplitude (HA).
Results: 1) The target Hb levels were maintained in both A(10.9±1.3 g/dL) and B(11.0±1.4). No significant difference was found. 2) Regarding Hb cycling, a marked difference was detected between A(0.19±0.53 times) vs B(0.48±0.79), (p=0.0018). 3) According to Hb variability, the number of cases classified under six categories in the two groups were L(A=7, B=0), T(14, 4), H(0, 0), LAL(36, 27), LAH(8, 28) and HA(30, 59). 4) Comparing a combined group of L and LAL patients with another group of LAH, HA and T patients, remarkable differences were found in Hb(A: 9.9±0.6 g/dL vs 11.0±0.6, p=0.0001 and B: 9.9±0.8 vs 11.4±0.6, p=0.0001), ESA administration dose(A: 6102.9±2321.2 units/week vs 5073.9±2195.4, p=0.03 and B: 48.4±20.2 μg/week vs 27.9±15.3, p=0.0001), age(B: 69.9±11.0 years old vs 63.7±12.1, p=0.02), Alb(B: 3.6±0.4 g/dL vs 3.8±0.3, p=0.03) and hs CRP(A: 0.3±0.4 mg/dL vs 0.2±0.3, p<0.05 and B: 0.3±0.3 vs 0.2±0.2, p=0.01). Multivariate Cox's proportional hazard regression analysis showed that marked correlations in Hb(Odds ratio: 0.019, p<0.0001 in A and 0.050, p<0.0001 in B) were found in the two groups.
Conclusions: These results indicate that EPO is superior to DPO concerning Hb cycling, and quality of life in patients categrized as LAL and L is not good because of the poor therapeutic effect of these two ESAs in the treatment of renal anemia and negative clinical characteristics such as lower albumin and higher hsCRP.

Su581 ANDROGEN VERSUS ERYTHROPOIETIN FOR THE TREATMENT OF ANAEMIA OF PREDIALYSIS DIABETIC CHRONIC KIDNEY DISEASE: A PROSPECTIVE STUDY

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Introduction and Aims: Chronic kidney disease is a microvascular complication of DM. Anaemia is an important clinical manifestation to treat in chronic kidney disease.This study has designed to see the efficacy of nandrolone in comparison with recombinant human erythropoietin for management of anaemia of predialysis diabetic chronic kidney disease.
Methods: Sixty adult diabetic patients with anaemia of chronic kidney disease on conservative treatment were enrolled. Patients were studied into two groups (1 and 2) of 30 patients each. Group 1 patients received nandrolone decaonate 50 mg deep intramuscularly & Group 2, recombinant human erythropoietin 100 IU per kilogram of body weight subcutaneously once weekly. Patients of both group received oral iron supplements in order to maintain body iron stores. All the relevant haematological and renal parameters were evaluated at the end of 3rd & 6th months.
Results: There was a statistically significant rise in haemoglobin concentration, packed cell volume, in both groups. The rise in haemoglobin concentration, in Group 2 was more marked followed by Group 1, at the end of 3rd (Group 1, 9.59±1.13 vs. Group 2, 10.51±1.23; p value 0.004), and 6th months, (Group 1, 10.8±1.26 vs. Group 2, 11.36±0.97; p value 0.057). Haemoglobin increased significantly in Group 1, at the end of 3rd & 6th months (8.61±0.94 vs. 9.59±1.13; p value 0.001 and 8.61±0.94 vs. 10.8±1.26; p value 0.001) & also in Group 2 (8.31±0.91 vs. 10.51±1.23; p value 0.001 and 8.31±0.91vs. 11.36±0.97; p value 0.001). Packed cell volume was increased similarly like haemoglobin. Role of nandrolone on glycemic status was evaluated by HBA1C & reduced in both groups, but significantly (p value 0.012) at the end of 6th months, in Group1 than Group 2. Nutritional parameters of nandrolone was evaluated by albumin level, and showed significant increase (p value 0.001) in albumin level in relation with erythropoietin at the end of 3rd and 6th months.
Conclusions: Nandrolone, though not equally effective, may be considered as a valid alternative therapy for the treatment of anaemia of predialysis diabetic chronic kidney disease to that of erythropoietin in low socioeconomic population to make the treatment more cost effective.

Su582 CKD EPIDEMIOLOGY AND RENAL ANEMIA TREATMENT WITH NEORECORMON IN EVERYDAY PRACTICE IN SELECTED EUROPEAN COUNTRIES

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Introduction and Aims: Renal anemia is routinely treated with Ery-

Abstract Su582 – Table 1. eGFR values

eGFR (mean,SD)		Hungary	Latvia	Poland	Russia	p
MDRD	CKD Diagnosis	32,96±14,54	39,64±17,48	27,09±14,85	29,96±13,46	0,005
	Predialysis care	33,24±13,76	36,46±16,96	26,77±13,87	25,44±13,66	0,002
	Start of ESA therapy	29,95±12,29	37,01±16,78	23,32±10,54	22,13±12,42	0,001<
Cockroft-Gault	CKD diagnosis	40,49±17,47	45,67±17,81	31,94±18,36	34,68±17,57	0,016
	Predialysis care	40,60±16,97	43,16±16,78	29,17±14,87	33,50±18,06	0,001<
	Start of ESA therapy	36,41±15,03	41,57±15,46	26,54±10,96	30,17±17,87	0,001<

tropoiesis Stimulating Agents. However the approach toward its treatment may differ among countries. The aim of the study was to compare the retrospective data on renal anemia management followed with three years of observation in predialysis patients from Latvia, Hungary, Estonia, Slovakia, Russia and Poland.

Methods: It was a non-interventional, observational study. We present retrospective data.

Results: 285 patients(Males-103,Females-182, aged 21-90(61,7±14,8) years; 89 from Hungary,79 from Latvia,66 from Poland,27 from Russia,20 from Estonia,4 from Slovakia) were enrolled.

In Hungary,Estonia,Slovakia the most common reason for CKD was diabetic nephropathy (32,6%, 45%,50%),in Latvia and Poland pyelonephritis(29,1%;30,8%),in Russia glomeluronephritis(29,6%) and pyelonephritis (29,6%).

The mean eGFR calculated at CKD diagnosis, predialysis care and start of ESA therapy with MDRD and Cockroft-Gault formulas was higher in Latvia and lowest in Poland and Russia.

The mean haemoglobin (Hb) values at ESA treatment start were similar in all coutries (Estonia 10,1g/dl ±1,3, Hungary 10,5g/dl±0,9, Latvia 10,2±1,2, Poland 10,1 g/dl±1,0, Russia 9,6±1,2 and Slovakia 9,2±1,6).

The mean weekly dose of Neorecormon was highest in Slovakia (8000±2000 units) and Estonia (4625±3098 units),lower in Latvia (3936±2370 units), Russia (3740±2104 units) and Hungary (3441±2015,9 units) and lowest in Poland (1807±528,1 units).

The mean Hb level during predialysis care was highest in Latvia (12,2±1,1 g/dl), and Hungary (11,4±1,1 g/dl) and lowest in Russia (10,7±1,2 g/dl) and Slovakia (9,8±1,0 g/dl). In Poland it was 11,1±1,1 g/dl in Estonia 11,2±0,8 g/dl. All patients received Neorecormon subcutaneously. 70% of Estonian patients, 59,6% Hungarian patients, 41,8% Hungarian patients, 92,3% Polish patients and 22% Russian patients received NeoRecormon once a week. It was used once every two weeks in 15% of Estonian, 37,1% Hungarian, 19% Latvian, 4,6% Polish and 25,9% Russian patients.

Conclusions: There are country specific differences in the management of renal anaemia.

In Latvia the CKD diagnosis was made, predialysis care started and ESA therapy introduced at higher eGFR.

The doses of NeoRecormon are highest in Slovakia and Latvia and lowest in Poland.

Su583 DO TWO "IDENTICAL" INTRAVENOUS IRON SUCROSE PREPARATIONS HAVE THE SAME EFFICACY?

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Introduction and Aims: Patients (pts) on chronic haemodialysis (HD) with anaemia are treated in current practice with erythropoiesis stimulating agents (ESA) and intravenous (IV) iron. In our unit, over the last years, the original IV iron sucrose Venofer® (V) was used with satisfactory clinical results. In June 2009, it was decided to switch to an IV iron sucrose similar preparation, FerMylan® (FM). We observed that there was a need to re-titrate both the ESA and IV iron dose to maintain haemoglobin (Hb) values within target levels.

Methods: This retrospective analysis compared two 6 month periods(P), Period 1 (P1): Dec 2008 – May 2009 during which pts received V, and Period 2 (P2): July – Dec 2009 during which pts received FM. Pts were included in the study, if they had at least 60 dialysis sessions per P in the unit, and were treated for their anaemia with IV iron at least once. Data for June 2009 were excluded as both iron drugs were simultaneously available. Hb was assessed every two weeks; serum ferritin (SF), transferrin saturation (TSAT), C-reactive protein (CRP) and Kt/V were collected twice per P. Darbepoetin-a (DA) was injected IV once every two weeks and IV iron once a week (25-200 mg iron). Paired Student's t-tests were used for statistical analysis.

Results: The study population consisted of 75 pts (male 67%), mean age 63.4±15.2 years, mean duration on dialysis of 42 months. Diabetes was the primary renal disease in 37% of pts. The average number of dialysis sessions per pt performed during P1 was 75.1 (SD 5.1) and during P2 was 75.1 (SD 6.3) (ns). Anaemia and iron parameters are outlined in the table below:

Values	P1	P2	p-value
Hb mean (SD) g/dL	11.78 (0.99)	11.48 (0.98)	0.01
Hb median: P1 vs first 3 months P2 g/dL	11.8	11.42	0.005
Ferritin mean (SD) µg/L	570.6 (391.5)	494.5 (279.9)	0.09
TSAT mean (SD) %	49 (11.8)	24.5 (9.4)	<0.0001
CRP mean (SD) mg/L	6.40 (7.69)	8.43 (10.46)	0.13
Kt/V mean (SD)	1.41 (0.25)	1.42 (0.24)	NS
DA total dose per Period µg	73310	82300	0.04
DA µg/kg/week per pt	0.54	0.65	0.04
Iron (ampoules/Period)	880	1240.5	0.0002
Iron mean dose/pt per Period (SD) mg	1173 (873)	1654 (833)	0.0002

Clinical parameters were stable for several years prior to and during P1. Hb levels were statistically lower with FM during P2 as compared with V during P1. In order to maintain Hb target levels, a significant increase of 20% in the DA dose as well as an increase of 30% in the IV iron dose was required in P2. Nevertheless there was a decrease in TSAT and SF values during P2.

Conclusions: This retrospective evaluation suggests that V and FM are not therapeutically equivalent. A direct consequence of the switch from V to FM was the need to increase drug consumption to reach and maintain target Hb levels with a resultant increase in drug costs (+12%). These finding could have important consequences for the treatment of anaemia in HD pts. Further prospective studies are required to confirm these findings.

Su584 TIME SAVINGS ASSOCIATED WITH ONCE-MONTHLY C.E.R.A.: A TIME AND MOTION STUDY IN GERMAN AND SPANISH DIALYSIS CENTRES

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Introduction and Aims: Anaemia management in chronic kidney disease (CKD) consumes a substantial amount of time for both staff and patients (pts) in dialysis centres. Simplifying anaemia management could lessen the burden, allowing reallocation of resources. The multinational Time and Motion study presented here quantifies healthcare personnel time savings and evaluates qualitative changes in regular anaemia care procedures following introduction of C.E.R.A. Q4W maintenance therapy. First results from dialysis centers in Germany and Spain are reported.

Methods: Time spent on frequent anaemia management tasks (such as ESA preparation, distribution and injection) was recorded by trained observers using a stop watch. For each task within each centre, data samples for ESAs and C.E.R.A. were analysed and results compared after adjusting for group size. The main study end point, 'time per pt per session', was used to calculate annual time per pt and per centre. Qualitative information on expected/observed changes in practice patterns following introduction of C.E.R.A. Q4W was collected from questionnaires completed by senior healthcare personnel in each centre.

Results:

Data from German centres:

Key parameters affecting time reduction	Centre			
	1	2	3	4
Total no. pts receiving ESAs	103	130	240	150
C.E.R.A. uptake, %	24	24	25	7
Mean no. ESA administrations per pt per year (excluding pts receiving C.E.R.A.)	86	57	62	148
No. ESA administrations avoided per pt per year by switching to C.E.R.A. Q4W	74	45	50	136
Time per pt per year, min				
ESAs	76.0	127.0	55.2	172.7
C.E.R.A.	13.2	20.0	11.9	15.2
Calculated reduction in time at 100% C.E.R.A. uptake, %	83	84	79	91

Data from Spanish centres:

Key parameters affecting time reduction	Centre		
	1	2	3
Total no. pts receiving ESAs	43	100	39
C.E.R.A. uptake, %	21	26	56
Mean no. ESA administrations per pt per year (excluding pts receiving C.E.R.A.)	53	91	60
No. ESA administrations avoided per pt per year by switching to C.E.R.A. Q4W	41	79	48
Time per pt per year, min			
ESAs	55.8	38.2	50.7
C.E.R.A.	17.5	6.0	11.3
Calculated reduction in time at 100% C.E.R.A. uptake, %	69	84	78

Questionnaire findings from the participating centres reported reduction in inventory and ordering time with a switch to C.E.R.A., and confirm the reduction in preparation and injection time as shown in the time and motion analysis.

Conclusions: This study shows that 100% conversion to C.E.R.A. Q4W maintenance therapy offers annual time savings on tasks related to anaemia management of 79-91% in 4 German centres and 69-84% in 3 Spanish centres, confirming the findings of a previous study.[1] Administration of 12 injections of C.E.R.A. per pt per year results in substantial time savings, allowing scarce healthcare resources to be focused on improving patient care at a critical point in the dialysis procedure.

Reference:

1. Saueressig U et al. Blood Purif 2008; 26: 537-546.

Su585 THE SERUM LEVELS OF HEPCIDIN-25 AS THE MIRROR IMAGE OF ERYTHROPOIESIS

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Introduction and Aims: Hepcidin synthesis responds to iron overload, inflammation and inhibition of erythropoiesis in hemodialysis patients. On the other hand, it will be demonstrated that erythropoietin(EPO) down-regulates hepcidin expression in cultured hepatocytes through EPOR signaling. In this study, we investigated the early response of hepcidin -25 to EPO administration in hemodialysis patients.

Methods: This study was performed in 10 maintenance hemodialysis patients (4 men and 6 women with a mean age of 58.2±7.7 years and a mean duration of dialysis of 101±90 months) who were non-diabetic and were not receiving iron preparations. In addition to the ferritin, serum iron, TSAT, and serum haemoglobin (Hb) levels, we also measured the serum IL-6 level, and studied the serum hepcidin-25 level both prior to intravenous injection with epoetin beta (epo) and also at 3, 6, 9 and 18 hours later. The hep level was measured using a high-throughput LC-MS/MS method.

Results: The hepcidin-25 level in all subjects was 6.1±9.5 ng/mL; 4 subjects exhibited levels of 4 ng/mL or higher (14.5±10.8; the H-hep group), and 6 subjects exhibited levels below 4 ng/mL (0.6±0.6; the L-hep group). We therefore compared these 2 groups as described below. No difference was seen between the 2 groups in terms of the Hb level: it was 10.8±0.6 g/dL in the H-hep group and 9.9±1.7 g/dL in the L-hep group. The ferritin level was 74.9±60.2 ng/mL in the H-hep group, which was significantly higher than the 9.5±3.6 ng/mL in the L-hep group. The TSAT level was also significantly higher in the H-hep group than in the L-hep group: 30.2±9.4 vs. 12.2±7.5. In the H-hep group, the hepcidin-25 level increased significantly, from 14.5±10.8 before epo dosing to 23.1±17.0 at 3 hours later, 35.2±16.8 at 6 hours later and 30.7±13.7 ng/mL at 9 hours later, respectively. In the L-hep group, the level of hepcidin-25 remained at or below 1.0 ng/mL at all 4 measurement points. The weekly dose of epo was no different in the H-hep group than in the L-hep group, 4500±3240 IU vs. 5875±3563 IU, respectively, and no difference was seen between the groups in the time course of epo blood concentration. Mean serum IL-6 levels of 10 patients is 2.3±1.2 pg/mL.

Conclusions: Our data show that in the early phase of the erythropoiesis induced by EPO, hepcidin are up-regurated. These findings suggest that the serum levels of hepcidin-25, which is exclusively sensitive to iron utilization for erythropoiesis, may show the mirror image of erythropoiesis.

CKD-related bone disease 2

Su586 PTH VARIABILITY PARAMETERS FOR IDENTIFYING HIGH TURN-OVER OSTEODYSTROPHY DISEASE IN HEMODIALYSIS PATIENTS: AN OBSERVATIONAL RETROSPECTIVE COHORT STUDY

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Introduction and Aims: Abnormalities in bone morphology developed in hemodialysis (HD) are defined as Renal Osteodystrophy (ROD). Bone chemical markers are used to detect and grade bone status. Literature consider the effect of absolute levels of PTH and not the measures of their change over time, the PTH variability (PTH-var).In a retrospective observational study, we focused on the usefulness of PTH-var, in a group of HD patients, as independent predictor for High Turn Over Osteopathy (HTO) versus Low Turn Over Osteopathy (LTO).

Methods: The study was conducted on 90 HD patients classified in HTO or LTO for diagnosis based using the following criteria: PTH≥400 pg/mL associated to bone alkaline phosphatase ≥ 20ng/mL. We used a regression-based metric of PTH–Var.Every enrolled patient was observed for a variable time period basing on the number and rate of serum PTH determinations. We used a regression-based metric of PTH–Var. PTH-Var was characterized with different parameters: 1) The residual standard deviation that is the square root of the average squared vertical distance between observed PTH levels and the regression line parameter estimates from these model (PTH-Res-SD); 2) Temporal trends in PTH levels that is the slope of the regression line (PTH-Slope); 3) Absolute value in PTH levels defined as the intercept of the regression line (PTH-Intercept). We calculated the average absolute change in PTH calculated over the same exposure windows defined as the average of the absolute difference between consecutive PTH measurements (PTH-Abs-Var). Data were expressed as partial correlation coefficients (β) and p value. Receiver Operating Characteristics (ROC) analysis was employed to calculate the area under the curve (AUC) for PTH-Res-SD to find the best possible cut-off values for identifying a status of HTO.

Results: At univariate analysis PTH-Abs-Var had a significant direct correlation with PTH-Res-SD (R=0.69 p=0.0000) while no correlation was found with PTH-intercept and PTH-slope. PTH-Res-SD correlated with PTH-intercept (R=0.37 p=0.03) but not with PTH-slope. PTH-slope inversely correlated with PTH-intercept (R=-0.44 p=0.01). PTH-intercept had a strong direct link with mean PTH (R=0.61 p=0.0002). Univariate analysis demonstrated a correlation of HTO with PTH-Res-SD (beta=0.02 p=0.006), with PTH-intercept (beta=0.03 p=0.04), with z-score (beta=2.62 p=0.008), with phosphorus (beta=1.33 p=0.01), with CaxP (beta=0.15 p=0.01). At multivariate analysis only PTH-Res-SD maintained a significative predictive power on HTO diagnosis (beta=0.03 p=0.04). Receiver Operating Characteristics (ROC) analysis fixed the best possible cut-off of PTH-Res-SD at 150.4 pg/mL for identifying a status of HTO (AUC 0.91 (IC 95% 0.73-0.97)).

Conclusions: In our analysis, parameters of PTH Variability demonstrated as good independent predictive factors for High Turn-over Osteodystrophy and they had a greater sensibility respect to the use of single or/and mean PTH measurement in ROD classification.

Su587 THE SIZE OF OSTEOCYTES BEFORE AND AFTER PARATHYROIDECTOMY IN PATIENTS WITH SECONDARY HYPERPARATHYROIDISM

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Introduction and Aims: It has been reported that the lacunar walls are classified into the three groups, namely, lacunae with a predominantly eroded surface (ES.Lc), Lc with a predominantly quiescent surface (QS.Lc), and Lc with a predominantly osteoid surface (OS.Lc), indicating that the osteocytes have the important roles, including bone resorption and formation. The osteoid osteocytes differentiate into the mature osteocytes with OS.Lc and the size of the osteocyte is reduced during this differentiation. OS.Lc is mineralized and QS.Lc is also mineralized directly through the osteocyte-canalicular systm. The number of the osteocytes are also reduced after parathyroidectomy for secondary hyperparathyroidism (JASN abstract) 2008). We measured the volume of the osteocytes in ES.Lc, QS.Lc, and OS.Lc in patients with secondary hyperparathyroidis. And changes of the osteocyte volume in ES.Lc, QS.Lc, and OS.Lc were also investigated after parathyroidectomy to see the state of the disappearance of the osteocytes.
Methods: Thirty seven hemodialysis (HD) patients suffering from secondary hyperparathyroidism (age; 53.5±8.1 years, duration of HD; 17.1±7.3 years, intact PTH; 1153.8±457.8 pg/ml) underwent the iliac bone biopsies. The mean osteocyte volume in ES.Lc (ES.Ot.V/N.ES.Lc;μm^2), QS.Lc (QS.Ot.V/N.QS.Lc;μm^2), and OS.Lc (OS.Ot.V/N.OS.Lc;μm^2) were measured in cancellous bone. And changes of the osteocyte volume in each lacunae were investigated by the second bone biopsy specimens obtained at 3 or 4 weeks after parathyroidectomy in 16 patients.
Results: OS.Ot.V/N.OS.Lc was 9.9±5.6 μm^2, ES.Ot.V/N.ES.Lc was 7.7±3.1 μm^2, and QS.Ot.V/N.QS.Lc was 6.7±2.8 μm^2, respectively. OS.Ot.V/N.OS.Lc was significantly greater than QS.Ot.V/N.QS.Lc (P = 0.025). ES.Ot.V/N.ES.Lc was not different from QS.Ot.V/N.QS.Lc in these patients. OS.Ot.V/N.OS.Lc, ES.Ot.V/N.ES.Lc and QS.Ot.V/N.QS.Lc were not changed after parathyroidectomy.
Conclusions: It seems that the osteocyte volume in OS.Lc is great as these osteocytes appear immediately after the differentiation from the osteoid osteocytes to the mature osteocytes. Thereafter, osteocyte volume is unchanged during the life cycle of the osteocytes. It also seems that the osteocyte number was reduced after parathyroidectomy due to the abrupt death (apoptosis or necrosis) of the osteocytes, but not to the gradual involution of these cells.

Su588 RELATIONSHIP BETWEEN SERUM ADIPONECTIN AND BONE MINERAL DENSITY IN MALE HEMODIALYSIS PATIENTS

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Introduction and Aims: Adiponectin is a recently described adipocyte-produced hormone that correlates negatively with adiposity. Adiponectin and its receptor have been found to be produced by human bone-forming cells, suggesting that adiponection may be hormone linking bone and fat metabolism. It is reported that serum adiponectin level was correlated with bone mineral density (BMD) in general population. However, little is known about the association between adiponectin and bone metabolism in patients with chronic kidney disease. In this study, we investigated the relationship between serum adiponectin level and BMD in male hemodialysis (HD) patients.
Methods: 119 patients on maintenance HD (age 61 + 11 years; HD duration 6.6 + 3.0 years; 44% diabetics) were examined. Bone mineral density (BMD) of 1/3 distal radius and ultra-distal radius were measured by dual X-ray absorptiometry. The former represents cortical bone rich site and the latter cancellous bone rich site. Blood sample was collected immediately before starting the HD session for the measurement of adiponectin, intact parathyroid hormone (PTH), and cross-linked N-telopeptide of type I collagen (NTX).
Results: Although there was no significant correlation between serum adiponectin level and intact PTH level, there was significant and positive correlation between serum adiponectin level and NTX level (r = 0.314, p < 0.001). BMD of the 1/3 distal radius was significantly correlated in a negative manner with age (r = – 0.245, p=0.007) and serum adiponectin levels (r = -0.236, p=0.009) and in a positive manner with body weight (r = 0.226, p=0.003). Similarly, BMD of the ultra-distal radius was significantly correlated in a negative manner with age (r = -0.323, p<0.001), serum intact PTH (r = -0.187, p=0.046), and adiponectin level (r = – 0.310, p<0.001) and in a positive manner with body weight (r = 0.384, p<0.001). In multiple regression analysis, serum adiponectin level was a variables significantly and independently associated with BMD of the ultra-distal radius, in addition to other significant and independent variables such as age, body weight, and intact PTH level (R^2 = 0.297, p<0.001), whereas serum adiponectin level was not a variable significantly associated with BMD of the 1/3 distal radius.
Conclusions: These results demonstrated that adiponectin is associated with bone metabolism of cancellous bone rich site rather than cortical bone rich site of the radius in male HD patients, suggesting that adiponectin is a possible factor contributing to the development of chronic kidney disease – mineral and bone disorder.

Su589 1-84 PTH/7-84 PTH RATIO IS NOT A GOOD MARKER OF BONE STATUS IN HEMODIALYSED PATIENTS

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Introduction and Aims: Chronic kidney disease-mineral and bone disorder (CKD-MBD) is one of the main complications in patients with chronic kidney disease, especially in hemodialysed ones. A lot of studies are performed to find the best diagnostic and noninvasive marker. Bone biopsy is still a gold standard, but it is invasive method connected with serious complications. Previous data show that 1-84 PTH/7-84 PTH (CAP/CIP) ratio can be useful to interpretation the bone status in HD patients.
The aim of the present study was to evaluate the correlation between total PTH (tPTH), 1-84 PTH (CAP), 7-84 PTH (CIP), bAP and β-CTX serum levels and CAP/CIP ratio.
Methods: The study group consisted of 80 hemodialysis patients (men – 61%, women – 39%), mean age 61±13 years (range19-85), mean duration of HD treatment 43±41 months (0,5-194).
Total PTH and whole PTH (CAP) were measured by immunoradiometric assay (Duo PTH, Scantibodies Lab., USA). CIP (7,84 PTH) was calculated by subtraction of CAP from tPTH and then the ratio CAP/CIP was calculated. bPA was measured by Ostase BAP OCTEIA (IDS LTD, UK) and β-CTX by Serum CrossLaps ELISA (NBD, Denmark). Blood samples were collected immediately before the hemodialysis session, ultra-filtrated and deep frozen at -70°C before the measurements.
Results: Mean serum levels of investigated parameters were: tPTH – 432,28 pg/ml, CAP – 257,45 pg/ml, CIP 174,82 pg/ml, bAP – 26,05 ng/ml, β-CTX – 2,94 ng/ml and CAP/CIP ratio – 1,53. 44% had CAP/CIP ratio below 1,4 – characteristic for adynamic bone disease. There was high positive correlation between tPTH, CAP and CIP (r>0,90) and good positive correlation between tPTH and both bAP and β-CTX (r=0,60). No correlation between all parameters and CAP/CIP ratio (r<0,15) was observed.
Conclusions: Our findings show that 1-84 PTH/7-84 PTH ratio is not useful marker of bone status in hemodialysed patients. The levels of total PTH and

specific bone markers like bAP and β-CTX should be measured to estimate bone turnover but bone biopsy is still the only method to confirm clinical observations.

Disclosure: Research was sponsored from money on science in years 2007-2009.

Su590 THE EFFECTS OF PHOSPHATE BINDERS IN A NEW RAT MODEL OF ADYNAMIC BONE DISEASE

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Introduction and Aims: Although the prevalence of adynamic bone disease (ABD) is increasing, there are very few experimental models of this disease, most being models of ABD with metabolic syndrome. In addition, the effects of different phosphate (P) binders on ABD have not been evaluated.

Methods: We established a model of ABD induced by 5/6 nephrectomy + parathyroidectomy in rats fed a 1.2%-P, grain-based diet. Animals were divided into 4 groups: Ca (3% calcium carbonate-treated); Sev (3% sevelamer carbonate-treated); CKD (untreated); and control (normal diet/sham-operated/untreated). After 8 wks of treatment, we performed biochemical and histomorphometric analyses, as well as von Kossa staining of aortic sections.

Results: All study group rats showed higher serum creatinine and P and lower serum Ca than did control rats. Despite similar serum P in all study groups, fractional excretion of P (FeP) was significantly lower in the treated groups. Calciuria was highest in the Ca group. None of the rats developed vascular calcification. Histomorphometry revealed ABD in all study groups, as confirmed by lower bone formation rates, as well as decreases in osteoid volume, osteoblast surface and osteoclast surface and absence of fibrosis. We found no differences among the study groups in terms of bone histomorphometric parameters, except for greater eroded surface in the Ca group.

Biochemical and histomorphometric data

Parameter	Ca (n=7)	Sev (n=5)	CKD (n=7)	Control (n=5)
Creatinine (mg/dl)	1.4±0.5	1.5±0.4	1.4±0.2	0.6±0.1[a]
iCa (mmol/L)	0.5±0.1	0.5±0.3	0.6±0.2	1.1±0.0[a]
P (mg/dl)	11.5±2.2	10.6±3.5	11.3±3.0	5.2±0.2[a]
FeP (%)	1.8±1.7[a]	23.1±12.1[a]	53.8±23.3[a]	6.1±3.0[a]
uCa (mg/24h)	8.5±3.6[c,d]	4.3±2.3	2.0±1.3	0.8±0.7[b,c]
BV/TV (%)	29.1±6.4	25.1±7.5	24.0±6.1	22.3±5.7
OV/BV (%)	0.1±0.2	0.3±03	0.1±0.1	0.8±0.5[b,d]
OS/BS (%)	2.7±2.3	5.7±6.3	2.1±0.8	17.6±8.3[a]
ES/BS (%)	10.4±2.8[c,d]	6.9±1.3	5.7±2.9	19.1±3.0[a]
Ob.S/BS (%)	2.4±2.0	4.1±4.3	1.7±0.7	14.7±7.2[a]
Oc.S/BS (%)	2.4±1.1	0.9±0.4	1.0±0.8	4.3±1.8[a]
BFR/BS (μ^3/μ^2/day)	0.01±0.01	0.03±0.03	0.02±0.01	0.08±0.04[a]

iCa: ionized calcium; uCa: urinary calcium; BV/TV: trabecular bone volume; OV/BV: osteoid volume; OS/BS: osteoid surface; ES/BS: eroded surface; Ob.S/BS: osteoblast surface; Oc.S/BS: osteoclast surface; BFR/BS: bone formation rate; [a]$p<0.05$ vs. all; [b]$p<0.05$ vs. Ca; [c]$p<0.05$ vs. Sev; [d]$p<0.05$ vs. CKD

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Conclusions: Although not decreasing serum P significantly, P binders decreased FeP. However, there was no improvement in bone histomorphometric parameters in this study. This new experimental model could be useful for the evaluation of ABD in future studies.

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Su591 SERUM 25-HYDROXYVITAMIN D (25OHD) LEVELS IN HEMODIALYSIS PATIENTS: AN OBSERVATIONAL STUDY

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Introduction and Aims: K/DOQI guidelines recommend supplementation for 25OHD deficiency/insufficiency in patients with chronic kidney disease (CKD) stages 3 and 4. Low serum 25OHD levels are also usually encountered in patients with CKD stage 5 undergoing dialysis. In hemodialysis (HD) patients these levels could be related to significant morbidity and mortality. The aim of this study was to evaluate the prevalence of low serum 25OHD in HD patients and study the factors related.

Methods: Serum 25OHD levels, were measured in winter by radioimmunoassay (RIA), in 34 stable and without active liver disease HD patients, male/female: 25/9, median age: 64 (23-90) years, median time on HD: 94 (5-287) months, dialyzed thrice weekly for 4-5 hours per session. They all were under sevelamer hydrochloride (SH) phosphate binder since 5-75 months, they had not received vitamin D compounds or calcimimetics in the 3 months period before biochemical evaluation and 6/34 patients were parathyreoidectomised (PTX). Serum iPTH, alkaline phosphatase (ALP), corrected calcium (Ca), phosphate (P), CaxP product, albumin (Alb) and C-reactive protein (CRP) levels were also measured.

Results: Mean serum 25OHD levels were 15,4±10,9 ng/ml. Only two patients (5,9%) had 25OHD values within normal limits (>30 ng/ml). VD insufficiency (25OHD levels between 20 and 30 ng/ml) was found in 10/34 patients (29,4%) (Group A). VD deficiency (25OHD values <20 ng/ml) was found in 22/34 patients (64,7%) (Group B). Very low 25OHD values (<10 ng/ml) were observed in 13/22 patients of Group B (59,1%) and these patients were in HD for a longer period of time compared to the rest of the Group B (129±94 vs 72±62 months, P=0,004). Between groups A and B, apart from the significant difference in serum 25OHD levels (24,86±3,2 vs 8,96±6,1 ng/ml, P<0,001), no other differences were observed in serum iPTH, ALP, Ca, P, CaxP product, Alb and CRP. Levels of 25OHD were significantly lower in patients with iPTH<100 pg/ml (8,1±7 vs 17,7±10,9 ng/ml, P=0,03) (as they included patients with PTX and/or for longer period on HD), in patients with Ca<9 mg/dl (11,6±10,4 vs 19,3±10,2 ng/ml, P=0,04) and in those receiving SH for a period longer than 50 months (10,2±7,3 vs 19,6±11,5 ng/ml, P=0,009). Multiple regression analysis in all patients revealed significant correlations of 25OHD levels with serum phosphate (r=0,489, P=0,008) and CaxP product values (r=0,478, P=0,01) and correlated negatively with age (r = -0,386, P=0,04).

Conclusions: The majority of our HD patients had low serum 25OHD levels and was more frequently deficient. Vitamin D levels, were inversely related to age, phosphate levels, duration of HD and SH treatment.

Su592 A CASE REPORT OF SHRINKING MAN SYNDROME IN A PATIENT ON CHRONIC HEMODIALYSIS

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Introduction and Aims: We show the case of a 39-year-old male patient who had hemodialysis for 10 years and concomitant progressive bone pain for 2 years. And this patient's body height has decreased by 24cm, from 176cm to 152cm. In this case, we underwent the patient total parathyroidectomy under cervical plexus anesthesia and topical anesthesia. We show that Shrinking Man Syndrome is a serious complication and is rarely seen in long-life Hemodialysis patients.

Methods: ECG showed junctional escape; III^0 atrioventricular block; and abnormal ST-T changes. X-ray examination: Bony plate thickening resulting in typical salt-and-pepper changes in plain skull film; Type B ultrasonography of parathyroid glands showed enlargement of right superior and right inferior parathyroid glands complicated by multiple sand-like changes. ^{99m}Tc-MIBI SPECT/CT dual-phase tomography showed enlargement of bilateral parathyroid glands. Admitting diagnoses: 1. CKD V, 2. Secondary hyperparathyroidism, 3. Renal bone disease, 4. Renal anemia, 5 III^0 atrioventricular block, 6 Shrinking man syndrome. In December 2007, the patient underwent total parathyroidectomy and parathyroid autotransplantation under cervical plexus anesthesia and topical anesthesia.

Results: In the operation, four swelling parathyroid glands were excised. After the operation, symptoms such as bone pain and short breath of the patient were soon alleviated, the parathyroid hormone in blood returned to normal, and alkaline phosphatase and blood phosphorus also decreased obviously.

Blood-biochemistry examination (Alkaline Phosphatase, Blood Calcium, Blood Phosphorus, Urea Nitrogen, Creatinine, Uric Acid, Hemoglobin) at discharge after 2 weeks compared with the one before operation and now is attached on Table 1.

Follow-up of 2 years, December, 2007 to December 2009, the patient's quality of life have been improving and no shrinking at all. Now he can take care of himself very well.

Table 1

Date/Tset	iPTH (pg/ml)	ALP (IU/L)	Ca (mmol/L)	P (mmol/L)	Urea (mmol/L)	Cr (umol/L)	UA (umol/L)
2007.11	3124	1296	2.41	1.66	28.50	748	387
2007.12	24.3	1248	2.60	0.81	35.83	724	335
2008.7	9.3	155	1.35	1.26	35.64 L	938	443
2009.8	9.61	133	2.43	0.98	35.32	854	391

Figure 1. The man at the right side is the patient, before and after his illness.

Conclusions: Shrinking man syndrom is a serious complication, and parathyroidectomy and parathyroid autotransplantation undergoing is the best way to stop body shriking. We report this case as an uncommon entity in dialysis patients and because we have kept many valuable pictures.

Su593 IMPACT OF TREATMENT OF SECONDARY HYPERPARATHYROIDISM (SHP) IN SERUM MAGNESIUM LEVELS (sMg)

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Introduction and Aims: Magnesium is a cation with similar homeostatic mechanisms to calcium.Changes in serum Calcium and Phosphorus on patients treated for SHP with Cinacalcet and paricalcitol are well known. We hypothesize that these treatments may influence serum Magnesium levels (sMg), which would have impact at the time of initiating treatment with magnesium-phosphate binders in HD patients.

Objectives:

1. To study the differences in sMg depending on the situation of SHP
2. Determine the influence of treatment for SHP with Cinacalcet or paricalcitol in SMg levels.
3. Influence of treatment with 25 OH vitamin D in the sMg levels.

Methods: We studied 59 patients on HD (23 w and 36 m) with a mean age of 70.3. SHP treatment with paricalcitol was held in 15 cases, and 5 cases were treated with cinacalcet. At the time of collection, 18 patients were treated with oral calcidiol (0,266 mg/week). The paricalcitol was used in intravenous pulses with a mean dose of 3.2 ugr/week, while Cinacalcet was used in mean daily dose of 60 mg/day.

Results: No significant differences were found in sMg between patients treated or not with paricalcitol (2.15±033 vs 2.02±0.24 mg/dl), calcidiol (2.11±0.27 vs 2.04±0.30 mg/dl) and cinacalcet (2.3±0.25 vs. 2.04±0.27 mg/dl).

When sMg was analyzed by quartiles, we observed that higher magnesium quartile corresponded to higher PTH levels than those with lower magnesium (0-1.9 mg/dl), but it was non significant: 255.84±175.92 pg/ml, (1.91-2.1): 205.83±151.96, (2.11-2.3): 380.2±164.5, (2.31-4): 415.32±188.89. n.s.

Conclusions: Patients with higher sMg had superior PTH levels, although results were not significant, probably due to small sample size.

No influence of Paricacitol or Cinacalcet treatment on sMg was observed. However there was a trend for higher sMg in patients treated with cinacalcet, which should be further explored in prospective studies.

Su594 CAN CHANGES IN PARATHYROID ULTRASOUND PREDICT CLINICAL OUTCOME OF PATIENTS ON DIALYSIS WITH SECONDARY HYPERPARATHYROIDISM?

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Introduction and Aims: The ultrasonographic finding of increased parathyroid indicates poor response to treatment in patients with hyperparathyroidism secondary to renal insufficiency (RI).

Methods: Eighty-five patients with levels of PTH higher than800pg/mL, from January 2005 to January 2006 were submitted to US and followed until January 2009. We evaluated laboratory parameters, clinical and demographic data and occurrence of death, vascular events and bone disease.

Results: Fifty-three patients (62.4%) had parathyroid nodules and higher levels of PTH, Ca and P. There was no association between nodule occurrence AND morbidity or mortality. Patients who underwent parathyroidectomy (n = 15) showed significant improvement in phosphorus levels as well as Ca x P product (P = 0.03 and 0.006 respectively). They also had lower mortality (32% vs 68%, p=0.01) and lower incidence of cardiovascular or cerebrovascular events (27% vs 73%., P = 0.02). Calcium x Phosphorus product above 55mg ^{2}/dL (RR 1.48 [1.06, 2.08], p=0.03), presence of vascular calcification (1.33 [1.01, 1.76], p=0.015) previous occurrence of vascular events (RR 2.25 [1.27, 3.98], p<0.001) were risk factors for mortality in this population.

Conclusions: The presence of parathyroid nodules in ultrasound (US) is associated with worse metabolic profile in patients with severe hyperparathyroidism, but cannot predict clinical outcome of these patients. Parathyroidectomy is associated with lower cardiovascular morbidity and mortality in patients with nodules. Further studies are needed to define the usefulness of US in the evaluation of hyperparathyroidism secondary to IR.

Su595 ORAL PARICALCITOL VERSUS CINACALCET AS MAINTENANCE TREATMENT AFTER SUCCESSFUL MEDICAL THERAPY OF SECONDARY HYPERPARATHYROIDISM (SHRT) IN HAEMODIALYSIS(HD) PATIENTS

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Introduction and Aims: Treatment of SHPT with IV pariIcalcitol (especially in combination with cinacalcet) is effective, leading to discontinuation due to iPTH suppression. In the present study we compared two different strategies in order to prevent the rebound of iPTH levels after the discontinuation of initial treatment.

Methods: We investigated n=50 patients (pts) on HD who had experienced a significant drop of iPTH (498.8±184 vs. 143.2±62 pg/ml) under treatment with IV pairIcalcitol (in 30 cases in combination with oral cinacalcet.) They were randomised to monotherapy in 2 groups with oral paricalcitol (n=25) or cinacalcet (n=25) and were followed up for 12 months.

Results: The dose of oral paricalcitol was 1.87±0.4 mg thrice/week whereas that of cinacalcet was, 44.5±23 mg/day. In both groups iPTH rose significantly: *Paricalcitol group:* 144.5±69 vs. 318.6±202 pg/ml, *Cinacalcet group:* 141.9±54 vs. 358.4±189 pg/ml (between the groups: p= ns, both in the beginning and the end). In the *Paricalcitol group*, treatment was interupted in 15 pts: 10 pts were switched back to iv paricalcitol due to iPTH rebound (114.0±60 vs. 524.8±142 pg/ml p<0,01), in 4 pts treatment was terminated due to further iPTH suppression (181.5±41 vs. 81.5±23 pg/ml p=0,033) and in 1 patient due to extreme hyperphoshatemia. There was no significant change in the mean Ca and P levels (p=ns), Furthermore, there was a tendency towards a rise in the phosphate binders (sevelamer: 3.04±2.9 vs. 3.11±2.8 tab/day, p= ns and lanthanum carbonate: 1.24±2.0 vs. 1.45±2.1 tab/day, p=0.054) In the *Cinacalcet group*, in 13 pts, IV paricalcitol was administered again due to a significant iPTH rise (129.5±47 vs. 485.5±98 pg/ml p >0,01) and in 4 pts treatment was discontinued due to iPTH suppression (144.8±54 vs. 74.3±19 pg/m p

>0,01 l) Ca and P levels were reduced (Ca: 9.0±0.5 vs. 8.76±0.6 mg/dl, p=0.066; P: 5.25±0.7 vs. 4.4±0.8, p<0.001). There was a decline in the P binding agents (HCL sevelamer: 3.97±4.0 vs. 3.62±3.6 tab/day, p=ns and lanthanum carbonate: 1.28±1.7 vs. 0.95±1.5 tab/day, p=0.188). At the end of follow up period treatment was considered successful in 10 pts (40%) in the *Paricalcitol group:* (iPTH: 145.0±67 vs. 221.3±63 p= ns) and in 8 pts (32%) in the *Cinacalcet group:* (iPTH: 163.3±66 vs. 284.7±152 p=ns.
Conclusions: There was no significant difference in the efficacy of either oral paricalcitol or cinacalcet as maintenance therapy of SHPT in chronic dialysis patients. In both treatments rebound or extreme suppression of iPTH was common.

Su596 BONE MINERAL DENSITY IN CKD STAGE 3-5 – USEFUL FOR THERAPEUTIC DECISIONS?

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Introduction and Aims: Data about an association between bone mineral density (BMD) and histomorphometrical markers of bone turnover are limited. Since BMD is increasingly performed in CKD, nephrologists are faced with a therapeutic dilemma: especially if BMD is significantly decreased, bisphosphonates seem to be a therapeutic option despite off-label use in advanced kidney disease.
Methods: Osteitis fibrosa (OF), the most frequent form of ROD was histomorphometrically diagnosed in 44 patients with CKD 3-5 (mean age 51.5±14.7). Possible relationships between BMD (measured by dual energy X-ray absorptiometry at the lumbar spine, femoral neck and trochanter), bone related markers (i.e. alkaline phosphatase, osteocalcin, intact PTH, 25(OH)D, 1,25(OH)2D, pyridinium crosslinks, vitamin K, albumin, total homocysteine, BMI) and histomorphometric findings were determined.
Results: Probably related to a mean iPTH of 247±184 ng/l (within the K/DOQI-range), there was a positive correlation between iPTH and BMD at the trochanter neck (R=0.354, P=0.047). In addition, only BMI (R=0.662, P=0.004), pyridinium crosslinks (R=-0.574, P=0.008 for pyridinoline, Pyd and R=-0.556, P=0.011 for deoxypyridinoline, Dpyd) and hemoglobine (R=0.680, P=0.002) showed an association with BMD (femoral neck). Alkaline phosphatase was related to proven histological bone apposition- (R=0.404, P=0.018 for osteoblast surface) and degradation markers (R=0.568, P<0.001 for osteoclast surface) confirming an increase in bone turnover due to OF. The mineralizing surface was positively related to vitamin D (R=0.347, P=0.033 for 25(OH)D, R=0.399, P=0.035 for 1,25(OH)2D). Pyd was related to the eroded surface (R=0.570, P=0.04) as a bone resorption marker. Vitamin K was negatively correlated with resorption markers (R=-0.357, P=0.024 for eroded surface, R=-0.533, P<0.001 for osteoclast surface). There was no correlation of BMD with any histomorphometric parameter.
Conclusions: In OF, mostly associated with a decrease in BMD, reduced BMD alone should not lead to a therapeutic decision, i.e. for the use of bisphosphonates. Different laboratory markers (i.e. AP, vitamin D, iPTH) may help to define the individual situation. For indifferent cases, bone biopsy should be performed.

Su597 EIGHTEEN-YEAR EXPERIENCE IN SUBTOTAL PARATHYROIDECTOMY OF HEMODIALYSIS PATIENTS

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Introduction and Aims: Secondary hyperparathyroidism (SHP) is a frequent complication of long-term dialysis patients and surgical parathyroidectomy remains necessary in patients resistant to medical therapy. The aim of our study was to analyze the single center results in subtotal parathyroidectomy that is a diagnostic procedure, indications for parathyroidectomy and success of surgical treatment.
Methods: A total of 170 patients (90 men), aged 20-69 (44.94±11.01) years, with various renal diseases, who have been regularly hemodialyzed between 2-23 years, have undergone parathyroidectomy at our Clinical Center during the last 18 years.
Results: The patients had plasma intact parathyroid chormone (iPTH) levels 3.4-98.5 times higher than the maximal normal limit, high values of phosphatemia and alkaline phosphatase, and calcemia on the upper normal level. Radiographic changes characteristic for SHP were seen in almost all patients before parathyroidectomy and the most common were subperiosteal resorptions (57%), bone cysts (51%), and extraskeletal calcifications (51.8%). Enlarged parathyroid glands were seen by ultrasound in 64.7% of patients. Pruritus was manifested in 46% of patients, bone pain in 73.5%, joint pain in 54.7%, and myopathy in 24.7% while other symptoms and signs were present in lower proportion.
Significant drops of phosphate and calcium levels were recorded in all but four patients on the very first postoperative day. Regular peroral and parenteral supplementation of the calcium and vitamin D metabolites were used, but calcemia was not restored to normal until the end of the postoperative third week. Serum alkaline phosphatase became increased after the surgery till day ten and thereafter decreased. Plasma iPTH levels, controlled on the 21st postoperative day, were close to the lower normal limit in all but four (2.35%) patients with persistent SHP, who required reoperation. Pathohistological analysis showed that 55% of patients had nodular hyperplasia, 36.5% diffuse hyperplasia, 2.9% adenoma, and 1.2% cancer of the parathyroid gland. There was a significant correlation between the weight of parathyreoid glands and: serum calcium (r=0,340; p=0,000), phosphate (r=0,256; p=0,001) and alkaline phosphatase concentration (r=0,163; p=0,043), but not with iPTH.
Conclusions: In conclusion, subtotal parathyroidectomy was proved as successful (97.65%) and safe treatment of patients with SHP resistant to medical therapy. There was a significant correlation between the weight of parathyroid glands and serum calcium, phosphate and alkaline phosphatase but not iPTH.

Su598 ABDOMINAL AORTA CALCIFICATIONS AND HEALTH RELATED QUALITY OF LIFE IN HEMODIALYSIS PATIENTS

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Introduction and Aims: Abdominal aorta calcifications (AAC) have been related to hemodialysis (HD) patients' survival. Low scoring in quality of life questionnaires was also found to be related to patients' survival. Aim of this study was to evaluate correlation of AAC with quality of life, with demographics and serologic indices.
Methods: Seventy three HD patients were studied; mean aged 62.2±13.1 years, on HD for a median time of 43.3 months. They answered a validated translation of the Kidney disease quality of life – short form (KDQOL-SF) questionnaire, which has been used previously in ESRD patients. Lateral lumbar radiography of the abdominal aorta was used to determine the overall AAC score, which is related to the severity of calcific deposits at lumbar vertebral segments L1–L4. The reliability of the method was tested by double reading of radiographs (cc 0.9). Correlation was estimated with the help of Pearson and Spearman correlation coefficient (cc).
Results: The median score of AAC was 8 (percentiles 25%-75%= 1-17). Mean scores in three major KDQOL-SF categories were: in physical component summary 47.1±22.9, in mental component summary 53.6±25.1 and in kidney disease component summary 54.2±15.8. AAC score was significantly correlated to age (cc=0,538 p<0,001), presence of diabetes mellitus (cc=0,345 p=0,008), presence of coronary disease (cc=0,680 p<0,001) while there was no significant correlation with any of three major KDQOL-SF categories, HD duration, mean values of last year's PTH, calcium, phosphate and CaxP product. Analysing subgroups of KDQOL-SF, AAC was correlated to work status (cc=-0,406 p=0,001), sexual function (cc=-0,268 p=0,038) and physical functioning (cc=-0,346 p=0,007).
Conclusions: AAC score was not significantly correlated to three major KDQOL-SF categories, while there was correlation with subgroups of KDQOL-SF, like work status, sexual function and physical functioning.

Su599 CINACALCET ON HYPERCALCEMIA AND BONE MINERAL DENSITY IN RENAL TRANSPLANTED PATIENTS WITH SECONDARY HYPERPARATHYROIDISM

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Introduction and Aims: Persistent secondary hyperparathyroidism (SHP) is the most frequent cause of hypercalcemia observed in approximately 15% of renal transplanted (RT) patients 1 year after surgery. Persistent SHP with hypercalcemia is an important factor of bone loss after renal transplantation. This study prospectively evaluates the effects of cinacalcet therapy on serum calcium (SCa) and parathyroid hormone (PTH) blood levels, and basically on bone mineral density (BMD) in RT patients with persistent hyperparathyroidism.
Methods: 15 RT patients (9 women, 6 man) with allograft function more than 6 months were included based on total SCa more than 10.5 mg/dL and intact parathyroid hormone (iPTH) concentration more than 65 pg/mL. After inclusion, patients started on a single daily oral dose of 30 mg of cinacalcet. At inclusion and every study visit blood levels of creatinine, Ca, P, alkaline phosphatase, iPTH 1,25- dihydroxyvitamin D3, and 25-hydroxyvitamin D3 were assessed. Baseline and at the end of study radial BMD were measured. Study follow-up was 12 months.
Results: During the study period, SCa decreased from 11.72±0.39 to 10.03±0.54 mg/dL ($P<0.001$). iPTH decreased from 308.85±120.12 to 214.66±53.75 mg/dL ($P<0.05$). The mean serum creatinine decreased from 1.58±0.34 to 1.25±0.27 mg/dL ($P=0.03$) and the mean radial BMD increased from 0.881±0.155 to 0.965±0.123 gr/cm^2 ($P<0.05$). There were no significant changes in the other parameters assessed. One patient was excluded for gastrointestinal intolerance.
Conclusions: In RT patients with hypercalcemia secondary to persistent SHP, cinacalcet corrects hypercalcemia and PTH, simultaneously improving BMD.
Disclosure: Thank you for including my work in the materials of the EDTA-ERA for publication upon acceptance. Dra María Esther Raola

Su600 DIETARY INTERVENTION FOCUSED ON PHOSPHATE INTAKE IN HEMODIALYSIS PATIENTS WITH HYPERPHOSPHOREMIA

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Introduction and Aims: Elevated serum phosphorus has been identified as a cardiovascular risk factor in hemodialysis patients. Despite all the oral phosphate binders currently available, dietary restriction of this mineral remains a cornerstone for the prevention and treatment of hyperphosphatemia. This study aimed to assess the effectiveness of dietary intervention to reduce phosphorus intake and to improve calcium-phosphorus metabolism in hemodialysis patients.
Methods: A six-month, two-group experimental study. Patients were included if their previous 3-month average serum phosphorus was higher than 5,5 mg/dl. Patients were allocated to intensive dietary intervention or usual dietary recommendations. The clinical end-points were the multivariate-adjusted change in serum phosphorus and the number of patients who achieved serum phosphorus levels < 5,5 mg/dl and a calcium x phosphorus (CaxP) product < 55 mg^2/dl^2.
Results: 91 dialysis patients were selected for participation; 80 completed the study, 41 in the experimental group and 39 in the control group. After 6 months, phosphorus intake (702±168 vs. 872±242 mg/24 hours; p=0,002) and phosphorus/protein ratio (14,1±1,7 vs. 12,5±1,6; p=0,035) were lower in the experimental group than in the control group, with no difference in protein-caloric intake between the two groups. Serum phosphorus decreased 1,67 mg/dl in the experimental group and 0,58 mg/dl in the control group (multivariate-adjusted difference 0,93 mg/dl; 95% confidence interval 0,34-1,52; p=0,003). Serum phosphorus < 5,5 mg/dl was attained more frequently (51 vs. 18%; p=0,002) in the experimental group. The CaxP product < 55 mg^2/dl^2 was also attained more frequently in the experimental group (71 vs. 36%; p=0,002).
Conclusions: Current dialysis and phosphorus binders prescription are no sufficient to assure normal serum phosphorus. Intensive dietary intervention focusing on phosphorus intake may be useful to reduce phosphorus retention and to improve calcium-phosphorus metabolism in hemodialysis patients.

Su601 FLUCTUATIONS IN SERUM CALCIUM, PHOSPHATE AND PTH LEVELS AFTER PARICALCITOL OR CINACALCET TREATMENT

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Introduction and Aims: Paricalcitol (a VDR activator) and cinacalcet (a calcimimetic) are both used to treat secondary hyperparathyroidism in chronic kidney disease patients undergoing dialysis, but these two drugs affect serum calcium (Ca) and phosphate (Pi) differently. Moreover, in clinical settings serum Ca, Pi and PTH are monitored routinely but not daily or hourly. It is not well understood whether and how these drugs affect fluctuations in serum Ca, Pi and PTH levels between dosing.
Methods: The 5/6 nephrectomy (NX) was performed on male, Sprague–Dawley rats with a standard two-step surgical ablation procedure. For cinacalcet, rats at week 6 after surgery were treated with vehicle (20% Hydroxypropyl-β-Cyclodextrin, 1.65 ml/kg, p.o. by gavage, daily) or cinacalcet at 10 mg/kg, p.o., daily for 12 days. For paricalcitol, rats at week 6 after surgery were treated with vehicle (5% ethanol + 95% propylene glycol, 0.4 ml/kg, i.p, 3x/week) or paricalcitol at 0.16 μg/kg, i.p., 3x/week for 12 days. At 0, 1, 4, 8, 16, 24 hr after the last dosing, blood was collected from different groups of rats and Ca, P and PTH levels in the serum were measured. In addition, the heart and left ventricle were collected and weighed.
Results: Serum creatinine and blood urea nitrogen (BUN) levels were significantly elevated in all NX rats compared to Sham rats at 6 weeks after surgery (before drug treatment) thus confirming experimental uremia. Treatment with cinacalcet at 10 mg or paricalcitol at 0.16 μg/kg for 12 days did not have an effect on serum creatinine or BUN. Cinacalcet at 10 mg/kg caused a significant decrease (∼11%) in serum Ca levels at 1 hr after the last dosing. The hypocalcemic state continued for at least 16 hrs and went back to normal (i.e. the level in the Sham or NX group) 24 hr after dosing. On the other hand, paricalcitol at 0.16 μg/kg had no significant effect on serum Ca levels; at 24 hr after dosing, the serum Ca level trended high, but did not reach a statistical significance. Cinacalcet at 10 mg/kg caused a significant increase in serum Pi levels (∼24%), which was observed at 1 hr after dosing, and remained elevated even at 24 hr after dosing. As a comparison, paricalcitol at 0.16 μg/kg had no significant effect on serum Pi except at the 24 hr time point after dosing. Cinacalcet at 10 mg/kg caused a ∼60% decrease in serum PTH levels at 1 hr after dosing; the PTH level continued to decrease following time and then at 24 hr bounced back to ∼50% of the level before dosing. As a comparison, paricalcitol at 0.16 μg/kg effectively suppressed PTH to a similar level at all time points. There was an increase in the left ventricular weight and heart weight in the uremic rats. Cinacalcet treatment had no significant effect, but paricalcitol reduced both the left ventricular weight and heart weight.
Conclusions: These results show that, although cinacalcet and paricalcitol both reduce PTH, cinacalcet treatment results in more fluctuations in serum Ca and PTH levels, and a prolonged elevation in serum Pi. Paricalcitol reduces left ventricular hypertrophy in the uremic rats, while cinacalcet has no significant effect.

Su602 FACTORS INVOLVED IN CHANGES OF SERUM MAGNESIUM (sMg) IN HEMODIALYSIS (HD) PATIENTS

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Introduction and Aims: Magnesium is one of the cations that are elevated in HD patients. It has some similar homeostatic mechanisms to calcium and is influenced by different treatments involved in the CKD-BMD.
Objectives: 1. To determine Mg levels in HD patients depending on the technique used for HD. 2. Influence of treatment with phosphate binders in serum Magnesium levels. 3. Importance of Diabetes Mellitus in serum Magnesium levels. 4. Correlation of biochemical parameters (Ca, P, 25 OH vit.D, PTH) with changes in the CKD-BMD
Methods: 59 HD patients (23 w and 36 m) with a mean age of 70.3 years, 13 patients in on line HD (olHD) and 46 patients in high flow conventional HD (CHD). sMg in the dyalisate was 0.75 mmol/L. 27 of these patients were diabetic.
Results: Mean sMg was 2.066±0.038 mg/dl. sMg levels did not correlate with serum calcium levels (r 0,075), p 0.482. sMg level was lower in CHD than in olHD; 2.02±0.25 vs 2.2±0.304, p <.032. There was no correlation with the use of phosphate binders, or with their doses. sMg was not correlated with PTH levels or with 25 OH vit D levels. However the calcidiol was correlated with Calcium and Phosphorus, p <.05, which means that magnesium has not the same behavior to similar cations.
Contrary to previously reported there are no differences between Mg levels in diabetics and non-diabetic patients; 2.107±0.3 vs 2.023±0.24 (ns). It's also observed that when magnesium is divided in quartiles, higher levels of sMg, increase levels of calcidiol (ng/ml), but that was not significant: Mg (0-1.9): 22.53±8.2 (1.91-2.1): 21.93±19.43, (2.11-2.3): 27.53±14.87 and (2.31-4): 26.1±5.5.
Conclusions: 1. Magnesium behaves differently to calcium, so there is no correlation with PTH, 25 OH vitamin D, calcium and phosphate.
2. Magnesium in diabetic patients is not different from non-diabetic patients.
3.In on line HD sMg is higher than in CHD.
4. Calcidiol levels are higher in patients with higher sMg but this is non-significant.

Su603 SECONDARY HYPERPARATHYROIDISM (SHPT) TREATMENT BY CALCIMIMETICS IN PATIENTS ON HAEMODIALYSIS (HD) OR PERITONEAL DIALYSIS (PD)

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Introduction and Aims: SHPT aggravates morbidity and mortality of patients on dialysis (ESRD). Calcimimetics suppress parathyroid hormone (PTH) secretion and reduce serum calcium.
Calcimimetic effectiveness was evaluated on patients with ESRD, under HD or PD.
Methods: Calcimimetic (Mimpara) was administered in gradually adapted doses of 30-90mg/24h to 16 patients with PTH>300pg/ml, 9 males/1female, median age: 58 (51-77) on HD (A group) and 5 males/1female, median age: 67,5 (33-75) on PD (B group). Calcium (Ca), phosphorus (P), alkaline phosphatase (ALP), albumin (ALB), PTH, CRP and CaxP product were monthly measured for two semesters, one before and one during treatment. Comparisons of biochemical results were performed between the two semester's means and finals (T6) as well as at the start and the end of treatment (T0/T6).
Results: The two groups, initially, were significantly different (see Table 1). Semestrial mean before significantly decreased during treatment in A (Ca 9,55±0,33→8,9±0,4 mg/dl, ALP 232,7±72,6→206±53 IU/L, PTH 402±180→302±98,6 pg/ml), while in B only ALP decreased significantly (293,9±42,5→199±33 IU/L).
Both groups showed significant differences between the start and the end of treatment results (see Table 2). By the end of the treatment period, A and B differed significantly only in ALB (3,52±0.18→3,1±0,4 gr/dl).

Table 1

	A	B	p
Months on dialysis	86 (20-288)	46,5 (2-83)	<0,05
ALB (gr/dl)	3,71±0,17	3,23±0,53	<0,005
CRP (mg/dl)	5,45±0,75	14,9±12,5	0,01
ALP (IU/l)	232,7±72,6	293,9±42,41	<0,05

Table 2

		T0	T6	p
A	Ca (mg/dl)	9,6±0,5	9±0,65	<0,05
	PTH (pg/ml)	544±219	216,5±72	<0,005
B	Ca (mg/dl)	9,5±0,55	8,9±0,8	<0,05
	PTH (pg/ml)	412±162	204±100	<0,05

Conclusions: Calcimimetics led to an improvement of sHPT in patients on ESRD on HD or PD with a parallel decrease in serum Ca levels on a different ratio according to the therapeutic method used.

Su604 RELATIONSHIP OF SERUM 25-HYDROXYVITAMIN D (25OH-VD) LEVELS WITH BONE METABOLISM, INFLAMMATION STATUS AND CARDIAC FUNCTION IN HEMODIALYSIS PATIENTS

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Introduction and Aims: K/DOQI guidelines recommend repletion of 25OH-VD in chronic kidney disease stages 3 and 4. Low serum 25OH-VD levels are also observed in hemodialysis (HD) patients, but their significance has not been adequately studied.
Methods: Serum 25OH-VD levels measured in winter, were evaluated in 34 stable HD patients without active liver disease [M/F: 25/9, median age: 64 (23-90) years, median time on HD: 94 (5-287) months], dialyzed thrice weekly for 4-5 hours per session. They all were treated with sevelamer hydrochloride (SH) and had not received VD compounds or calcimimetics in the previous 3 months. Serum albumin (ALB), C-reactive protein (CRP), ferritin (FERR), iPTH, alkaline phosphatase activity (ALP), phosphate (P), corrected for albumin Ca (Ca) and Ca x P product values were recorded. Left ventricle (LV) function was evaluated by echocardiography and femoral neck bone mineral density was measured by Dual energy X-ray absorptiometry (DEXA).
Results: Mean serum 25OH-VD levels were 15,4±10,9 ng/ml. Only two patients (5,9%) had 25OH-VD levels within normal limits (>30 ng/ml). VD insufficiency (serum 25OH-VD levels between 20 and 30 ng/ml) was found in 10/34 patients (29,4%) (group A). VD deficiency (serum 25OHD levels <20 ng/ml) was found in 22/34 patients (64,7%) (group B). In 13/22 patients with VD deficiency (59,1%) very low 25OH-VD values <10 ng/ml were observed, and these patients had more time on HD compared to the remaining group B patients (129±94 vs 72±62 months, p=0,004).
Compared with group A, group B patients with the lower serum 25OH-VD levels (8,9±6,1 vs 27,3±6,6 ng/ml, p<0,001), had lower ALB (3,6±0,4 vs 4,1±0,4 g/dl, p=0,01), higher FERR (490,9±242,1 vs 312,5±227 u/l, p=0,04) and lower LV ejection fraction (48,7±12,5% vs 56,6±3,3%, p=0,04). Significantly lower serum 25OH-VD levels were observed in patients with iPTH<100 pg/ml (8,1±7 vs 17,7±10,9 ng/ml, p=0,03), with Ca<9 mg/dl (11,6±10,4 vs 19,3±10,2 ng/ml, p=0,04), with sevelamer intake for more than 50 months (10,2±7,3 vs 19,6±11,5 ng/ml, p=0,009) and with a greater (>35 mm) LV end systolic diameter (11,5±8,6 vs 19,9±11,6 ng/ml, p=0,02). Multiple regression analysis in all patients revealed direct correlations of 25OH-VD with P (r=0,444, p=0,008), CaxP (r=0,431, p=0,01) and negative correlations with age (r= -0,351, p=0,04) and LV end systolic diameter (r= -0,381, p=0,03). The other biochemical parameters as well as T-score in DEXA (-2,1±1,8 SD) were not found to be related with serum 25OH-VD levels.
Conclusions: In conclusion, the majority of our HD patients had low serum 25OH-VD levels inversely related to age and duration of HD and SH treatment. 25OH-VD deficiency in HD seems to be associated with a rather

suppressed bone metabolism, LV systolic dysfunction and inflammation status. Further studies are needed to investigate possible modifications of those factors after 25OH-VD administration in HD patients.

Su605 BONE FRACTURES IN HAEMODIALYSIS PATIENTS

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Introduction and Aims: Renal osteodystrophy is a multifactorial disorder of bone metabolism. One of the most important clinical consequences of renal osteodystrophy is bone fractures. Several studies have shown that there is increased incidence and prevalence of bone fractures, particularly hip fractures in hemodialysis patients. The aim of this multicentric, retrospective study was to detect the prevalence of bone fractures in hemodialysis patients.
Methods: A total of nine dialysis centers with 767 hemodialysis patients were included in the study. Demographic data (age, sex, hemodialysis duration), laboratory data (PTH, Ca, P, total alkaline phosphatase), and bone fracture data (hip, forearm, upper arm, rib, vertebrae, lower leg, femur, hand) were collected from medical records as well as therapy with analogs of vitamin D.
Results: In 31 patients a total of 36 fractures were recorded, i.e. the prevalence of bone fractures was 4.7%. The mean age of patients with fractures was 77.6 (range 40-85) years and hemodialysis duration was 63.3 (range 5-265) months. There were 9 male and 22 female patients. Only six patients were diabetics. Of all patients suffering fractures, there were 14 with a hip fracture (39%), 8 with a forearm fracture (22%), 5 with an upper arm fracture (14%), 4 with a femur fracture (11%), 2 with a lower leg fracture (5%), one with a rib, vertebrae or hand fracture (9%). In patients less than 40 years of age no bone fractures were observed. Eight patients (26%) aged between 41 and 60 had bone fractures as well as 23 patients (74%) older than 60 years. Bone fractures were observed in 10 patients (28%) with PTH $<$ 180 pg/ml bone fractures and in 12 patients (33%) with PTH between 181 and 300 pg/ml; in 14 (39%) with PTH $>$ 300 pg/ml. Patients with hip fractures had the highest level of PTH, 541.4 (range 23-1790) pg/ml. The lowest PTH was in patients with forearm fractures 198.5 (range 57.3-310.9) pg/ml. Sixteen patients were on therapy with calcitriol or paricalcitrol at least six months before fractures.
Conclusions: The prevalence of bone fractures in our group of hemodialysis patients is high, particularly among female patients. The higher age and hemodialysis duration is undoubtedly a risk factor for bone fractures. Although there is a slightly higher incidence of fractures in patients with higher PTH (particularly hip fracture), there are also patients with bone fractures and a very low level of PTH. This is just one more proof that there are multifactorial risk factors for bone fractures in hemodialysis patients. More data from prospective studies is needed to detect the incidence, prevalence and risk factors for bone fractures in hemodialysis patients.

Su606 PARATHYROID HORMONE LEVELS AND MOBILIZATION OF CIRCULATING BONE MARROW-DERIVED CELLS IN END-STAGE RENAL DISEASE PATIENTS

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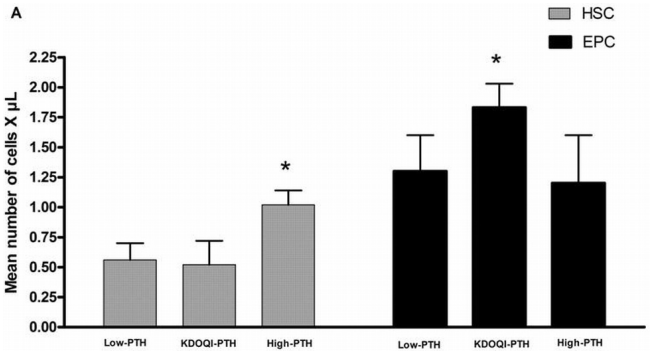
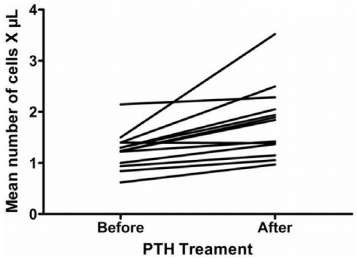
Introduction and Aims: Recently literature showed that elevated PTH serum levels result in increased mobilization of endothelial progenitor cells (EPCs) from bone marrow (Am J Physiol Endocrinol Metab. 2007) and that PTH application after myocardial infarction increases their migration to the ischemic heart, which may attenuate ischemic cardiomyopathy (Cardiovasc Res. 2008). These findings are apparently in contrast with common clinical state in ESRD characterized by increased serum parathyroid hormone (PTH) levels, a markedly decreased number of EPCs sustaining endothelial dysfunction, an important determinant involved in their increased mortality.In the present study we investigated, in a cohort of uremic patients, the effect of different levels of PTH on mobilization of progenitor cells populations.
Methods: 80 patients were enrolled. Following K-DOQI guidelines patients were divided in three groups for PTH levels: Low-PTH, K-DOQI-PTH and High-PTH group. Patients with high PTH were treated differently to achieve KDOQI targets: 5 received intravenous calcitriol and P binders,3 intravenous paracalcitriol, 10 cinacalcet. We quantified, by the combination of surface markers (CD45+,CD34+,CD31+,c-Kit+), the number of hematopoietic and endothelial progenitor cells. ROC analyses were performed in order to define the ability to identify the presence of an optimal PTH status among all patients.
Results: High-PTH group demonstrated a significant higher level of CD45+/CD34+/c-Kit+ respect to Low ($p<0.01$) and K-DOQI-PTH ($p<0.05$). CD45+/CD34+/CD31+ levels resulted significantly increased in the K-DOQI-PTH group compared to those observed in the Low- ($p=0.04$) and High-PTH group ($p=0.04$).



After 4 months we demonstrated an increase in EPCs number in 13 patients with hyperparathyroidism that achieved PTH targets after pharmacologically treatment.



Conclusions: Our data confirm the effect of PTH on bone marrow-derived progenitors cells emphasizing that, in our cohort, an intermediary PTH level, achieved following specific guidelines, results in an equilibrate balance between different subsets of progenitor cells.

Pathophysiology of kidney disease 2

Su607 METABOLIC SYNDROME (MS) IN NONDIABETIC PATIENTS (NONDM) IN HEMODIALYSIS (HD) AND ON TRANSPLANT (RT) WAITING LIST: SIMEDIT STUDY. CATALONIA-SPAIN

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Introduction and Aims: 1.-To determine, following APT III criteria, the

prevalence of MS in NONDM patients in HD and on a RT waiting list. 2.-To determine the most prevalent criteria for MS in this population. 3.- To analyze the characteristics of patients with and without MS. 4.-To determine the relationship between MS and cardiovascular disease.

Methods: *Population and methods:* Epidemiological, observational, multicenter study in NONDM patients from Catalonia in HD and on a RT waiting list (n = 353) (41,8% all NONDM patients included of Catalonia) and was sponsored by Catalonia Society of Nephrology. According to ATP III criteria, patients had MS when at least three of the following five factors were present: waist circumference (WC) (>102 cms men and >88 cms women), hypertriglyceridemia (TG-↑), low HDL-cholesterol (cHDL-↓), hypertension (HT) and hyperglycemia (HG) (fasting plasma glucose levels > 110 and <126 mg/dl). We also collected information on patients' clinical history of stroke and cardiovascular disease. The statistically significance was $p<0.05$.

Results: MS was present in 22.1% of the population. There were more men that women. With regard to the factors analyzed: 61.7% had hypertension, 36.3% had higher WC, 35.6% had TG-↑, 30.4% had cHDL-↓ and 5.6% had HG. We did not find a relationship between cerebrovascular disease and having MS. 59.7% of patients with MS had cardiovascular disease vs. 42.6% of patients without MS ($p<0.01$). There were no statistically significant differences in HbA1c between the two groups.The characteristics of patients with and without MS are shown in this table.

Characteristics of patients with and without MS. SIMEDIT Study

	MS present, n=78 (22.1%)	No MS, n=275 (77.9%)	p-value
Age (years)	54±12.3	52.2±12.2	ns
Sex (% men/women)	29.6/11.6	70.4/88.4	<0.001
BMI (kg/m^2)	27.6±5.4	24.2±3.7	<0.001
Normal weight* (%)	29.9	57.6	<0.001
Waist circunference (cm)	102.6±13.4	90.1±11.8	<0.001
Systolic BP (mmHg)	142.8±16.5	132.1±21.7	<0.001
Diastolic BP (mmHg)	79.9±11.7	76.3±12.9	<0.05
c-HDL (mg/dl)	35.1±7.6	47.7±13.8	<0.001
TG (mg/dl)	217±103.6	123.2±61.4	<0.001
Glycemia (mg/dl)	94.1±20.4	83.1±11.4	<0.001
HbA1c (%)	4.9±0.5	4.7±0.6	ns

*Normal weight (BMI 18.5-25), BP: blood pressure, c-HDL: HDL cholesterol, TG: triglycerides.

Conclusions: 22.1% of NONDM patients in HD and on a RT waiting list have MS. The presence of MS is not associated with age, but is more prevalent in men. Approximately one third of the patients with MS were of normal weight (BMI 18.5-25). Patients with MS have a higher percentage of cardiovascular disease.

Su608 DETECTION OF PULMONARY CONGESTION BY CHEST ULTRASOUND (US) IN DIALYSIS PATIENTS

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Introduction and Aims: Early detection of pulmonary congestion is a fundamental goal for the prevention of congestive heart failure in high risk patients.

Methods: We undertook an inclusive survey by a validated ultrasound (US) technique in a hemodialysis (HD) center to estimate the prevalence of pulmonary congestion and its reversibility after dialysis in a population of 75 HD patients.

Results: Chest US examinations were successfully completed in all patients (n=75). Before dialysis, 47 patients (63%) exhibited moderate to severe lung congestion. This alteration was commonly observed in patients with heart failure but also in the majority of asymptomatic (32/56, 57%) and normohydrated (19/38, 50%) patients. Lung water excess was unrelated with hydration status but it was strongly associated with New York Heart Association class ($P<0.0001$), left ventricular ejection fraction (LVEF) ($r=-0.55$, $P<0.001$), E/E' ratio ($r=0.48$, $P<0.001$), left atrial volume (LAV) ($r=0.39$, $P=0.001$) and pulmonary pressure ($r=0.36$, $P=0.002$). Lung water reduced after dialysis but 23 patients (31%) had still pulmonary congestion of moderate to severe degree. Lung water after dialysis maintained a strong association with LVEF ($r=-0.59$, $P<0.001$), LAV ($r=0.30$, $P=0.01$) and pulmonary pressure ($r=0.32$, $P=0.006$) denoting the critical role of cardiac performance in the control of this water compartment in ESRD. In a multiple regression model including age, smoking, fractional urea clearance, systolic pressure, heart rate and NYHA classification as well as LV mass index, LVEF, LAV, pulmonary pressure and E/E' ratio, only LVEF maintained an independent link with lung water excess ($\beta=-0.61$, $P<0.001$). Repeatability studies of the chest US technique (Bland-Altman Plots) showed good inter-observers and inter-US probes reproducibility.

Conclusions: Pulmonary congestion is highly prevalent in symptomatic (NYHA class III-IV) and asymptomatic dialysis patients. Chest ultrasound is a reliable technique which detects pulmonary congestion at a pre-clinical stage in ESRD. This method may be applied for early detection of pulmonary congestion in the high risk dialysis population.

Su609 PARATHYROID SURGERY IN 818 PATIENTS WITH RENAL HYPERPARATHYROIDISM

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Introduction and Aims: Renal hyperparathyroidism (rHPT) is a common and serious complication in patients with chronic kidney disease. Parathyroidectomy (PTX) remains the only option in patients with otherwise uncontrollable rHPT to improve osteodystrophy, musculopathy and vascular calcifications. The aim of this study was to evaluate clinical and surgical findings and outcome of patients with severe rHPT who underwent PTX.

Methods: Out of a prospective database of 872 patients with rHPT who underwent parathyroid surgery between 1976 and 2009, 756 patients on permanent dialysis (sHPT) and 62 patients after successful kidney transplantation (tHPT) were identified. Data were analyzed regarding perioperative biochemical changes, surgical procedures and postoperative outcome.

Results: 597/756 patients with sHPT underwent initial surgery. Total PTX with autotransplantation (AT, group A) was performed in 502, total PTX without AT in 27 (group B) and subtotal PTX in 68 patients (group C). After surgery, mean calcium dropped from 2.8 to 1.9mmol/l in group A, from 2.7 to 2.1mmol/l in group B and from 2.7 to 1.9mmol/l in group C. PTH levels dropped from 1376pg/ml to 29.7pg/ml in group A, from 1127pg/ml to 19.1pg/ml in group B and from 1403pg/ml to 104.1pg/ml in group C. Mean follow-up was 15 months (range 1-247). Persistent sHPT (PTH >5fold of the upper normal value) occurred in 7/597 (1.2%) and occurred in patients after incomplete initial surgery only. Postoperative permanent recurrent laryngeal nerve palsy (PRLNP) occured in 1.2% (7/597).

159/756 patients with sHPT underwent reoperative surgery. 66 received an AT-resection (D) and 93 a neck reexploration (E). After reoperation, mean calcium dropped from 2.7 to 2.0mmol/l (D) and from 2.7 to 2.3mmol/l (E). PTH dropped from 1269 to 129pg/ml in group D and from 1647 to 200pg/ml in group E. Mean follow-up was 55 months (range 1-161). Persistent sHPT occurred in 12/159 patients (7.6%). Postoperative PRLNP occurred in 8.6% (8/93) of patients after neck reexploration.

All 62 patients who underwent PTX after successful kidney transplantation (tHPT) included into this study underwent initial surgery. Total PTX with AT (group F) was performed in 37, total PTX without AT (group G) in 4 and subtotal PTX (group H) in 12 patients. In 9 patients less than 4 glands (group I) were identified. After surgery, mean calcium dropped from 2.92 to 2.06mmol/l in group F, from 2.85 to 2.1mmol/l in group G, from 2.88 to 2.16mmol/l in group H and from 2.86 to 2.13mmol/l in group I. PTH levels dropped from 544pg/ml to 24pg/ml in group F, from 670pg/ml to 67pg/ml in group G, from 617pg/ml to 15pg/ml in group H and from 440pg/ml to 81pg/ml in group I. Mean follow-up was 69 months (range 2-237). PRLNP occurred in 1/62 patients (1.6%).

Conclusions: Despite recent therapeutic advances such as the implementation of calcimimetics, severe rHPT may be refractory to medical treatment and require PTX. Total PTX with and without AT and subtotal PTX were able to effectively lower calcium and PTH levels. However, all efforts should be undertaken to avoid incomplete initial surgeries as reoperations are associated with higher complication- and lesser successrates.

Su610 THE ROLE OF PREGABALIN IN THE TREATMENT OF UREMIC PRURITUS

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Introduction and Aims: We evaluated the effect of pregabalin in the treatment of uremic pruritus that is not due to secondary hyperparathyroidism or inadequate hemodialysis and is refractory to conventional treatment.
Methods: Sixteen hemodialysis patients suffering from uremic pruritus resistant to conventional treatment were included in our study. Based on biochemical parameters of parathyroid function patients with secondary hyperparathyroidism were excluded. All patients re-ceived pregabalin (Lyrica, Pfizer) 25 mg/d orally at night for 1 month. Hematocrit, Ca, PO4, Ca x PO4 product, PTH, Kt/V, eosinophil counts and IgE were recorded before and after the end of the study. The effectiveness of pregabalin on uremic pruritus was evaluated by using Visual Analogue Scale at the beginning and the end of the ob-servation period. Visual Analogue Scale consisted of a 10 cm horizontal line scored from 0 (no itch) to 10 (worst imaginable itch).
Results: Four patients discontinued treatment due to side effects (3 dizziness and somnolence, 1 blurred vision and hand tremor) and therefore were excluded from the study. The remaining patients' mean age was 61.2±12.8 years and the time on hemodialysis 38±39.1 months. There was no statistically significant difference between the hemato-logical and biochemical parameters before and after the end of the study period.

Parameters recorded pre and post treatment

	Pre	Post	P
Hct (%)	37.5±5.9	36.6±4.9	N.S.
Eos (mm^{-3})	459±417.6	525±332.2	N.S.
Ca (mg/dl)	9.4±2.1	8.9±1.5	N.S.
PO4 (mg/dl)	5.5±2.1	5.3±2.3	N.S.
Ca x PO4 (mg^2/dl^2)	50.9±17.7	49.2±14.4	N.S.
PTH (pg/ml)	281±336	244±221	N.S.
IgE IU/ml	37.3±41.8	39.2±31.6	N.S.
Kt/V	1.16±0.2	1.13±0.3	N.S.

Contrarily there was a statistically significant improvement of pruritus' Visual Analogue Scale score after the one month treatment period (from 7.44±2.01 to 1.7±1.31, $p<0.00001$).
Conclusions: Pregabalin appears to be an effective alternative in the treatment of uremic pruritus that is not due to secondary hyperparathyroidism or inadequate hemodialysis and is refractory to conventional agents.

Su611 COLCHICINE TOXICITY IN ESRD PATIENTS: HYPE OR REALITY?

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Introduction and Aims: Colchicine is a micrutubule inhibitor primarily used for treatment of gouty arthritis, familial mediterranean fever (FMF) and Behcet's syndrome. Therapeutic index of colchicine is narrow. 20% of ingested colchicine is excreted via kidneys. Despite numerous colchicine intoxication cases, including fatalities, have been reported, there is no consensus in the literature in terms of recommended dose ranges in patients with chronic kidney disease (CKD). While some recommend two 0.5 mg tablets per week, others advise not to use colchicine in dialysis patients at all.
The aim of the present study was to evaluate tolerability and safety profile of colchicine in hemodialysis patients.
Methods: 20 patients (13 males and 7 females) who were undergoing maintenance hemodialysis at three private hemodialysis centers in Konya, Turkey and receiving colchicine on a chronic basis were included in the study. Daily colchicine doses, duration of and reason for colchicine use and other demographic and clinical characteristics of the patients were collected from patient charts. Interviews with patients especially focusing on syptoms of possible colchicine related toxicities were carried out. A detailed physical exam was also performed. Complete blood counts, liver function tests, creatine kinase (CK), myoglobine and other labaratory parameters were studied to detect adverse effects of colchicine.
Results: Basic demographic and laboratory characteristics of the study population are depicted in the table 1. All patients were receiving colchicine regularly: 14 for FMF, 2 patients for gouty artritis and 4 for secondary amyloidosis. 2 patients reported intermittant diarrhea. 4 patients complained of mild global alopesia. 6 patients reported generalized muscle pain. 3 patients had intermittant nausea and vomiting. None of the patients attributed their symptoms to colchicine use. CK, ALT and white blood cell counts of all patients were normal.

Laboratory and demographic charactertics patients receiving colchicine

Parameter	Value
Age	44,05±10,3
Colchicine dose	0,98±0,34
Duration	99,85±87,81
WBC	8762,5±5655,85
ALT	15,85±7,65
Creatine kinase	56,7±39,53
Myoglobine	179±110,34

WBC: Wight Blood Cell, ALT: Alanine aminotransferase.

Most Patients who reported diarrhea had amyloidosis due to FMF. Thus, their diarrhea may be due to gastrointestinal involvement of amyloidosis.
Conclusions: Results of this study showed that colchicine was well tolerated in hemodialysis patients at doses up to 1.5 mg per day.

Su612 EFFECT OF NITRIC OXIDE ON GASTRO-ESOPHAGEAL MOTILITY IN CHRONIC RENAL FAILURE PATIENTS

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Introduction: Gastrointestinal motor abnormalities may account for dyspeptic symptoms of chronic uremia patients. The gastrointestinal disorders possibly affect function and structure of the gastrointestinal tract and negatively impact on the nutritional status of the patients causing malnutrition which is a major problem with ESRD. NO is a proven gut neurotransmitters and their receptors were identified in different parts of the GIT, especially the esophagus and stomach. In CRF, previous studies on NO and VIP showed different results whether decreased, increased or even not altered, and the role of NO and VIP on gastrointestinal tract in CRF was not evaluated.
Patients and methods: The study was carried out on 60 patients with chronic renal disease divided into 3 groups group one includes 20 patients with CRF with creatinine clearance between 20-40 ml/min. group two includes 20 patients with ESRD (creatinine clearance <10 ml/min) just before the start of hemodialysis therapy. Group three includes 20 patients on regular hemodialysis (HD). Control group (group fours) includes 20 healthy subjects. For all groups full history and clinical examination, routine laboratory investigations, creatinine clearance, serum Nitrate levels an index of in vivo nitric oxide generation, esophageal manometry and electrogastrogram were done.
Results: There was a significant difference with serum nitrate between diseased CRF and healthy controls and in between patients groups. In the comparison between diseased CRF and healthy controls regarding dominant frequency, there is a highly significant difference, but no significant difference was found in between patients groups. In the patients with CRF, regarding diagnosis based on esophageal manometry and EGG: 16 patients was normal by oesophageal manometry (26.7%), 21 patients with NSMD (35%), 5 with GERD (8.3%), 8 with DOS (13.3%) and 10 with esophageal aperistalsis (16.7%), this showed a highly significant difference in comparison to the control group. 34 patients was normogastric by EGG (56.7%), 19 was bradygastric (31.7%) and 7 was tachygastric (11.7%), and also a highly significant difference in comparison to the control group. But in between the 3 CRF studied groups, no significant difference was found as regard diagnosis based on both esophageal manometry and EGG. A highly significant, weak negative linear correlation between NO and DF.There is significant association between the manometric based diagnosis, in CRF patients (3 groups), and NO.
Conclusion: Serum nitric oxide is disturbed in ESRD whether those patients

are on conservative medical management, or at the time of initiation of hemodialysis therapy or after maintaining regular hemodialysis therapy. The disturbed serum level of nitric oxide is associated with upper gastrointestinal dysmotility, which in turn may affect nutritional status of CRF patients and hence affects morbidity.

Su613 PHYSIOLOGIC, CARDIOVASCULAR AND NEUROENDOCRINE CHANGES IN HYPERTENSIVE PATIENS DURING THE HEMODIALYSIS SESSION

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Introduction and Aims: Patients with hypertension under dialytic treatment often present hypertensive episodes during the hemodialysis sessions, but the mechanism of this phenomenon is poor studied and understood. The objective of this study is to investigate possible physiologic, cardiovascular and neuroendocrine changes involved in the process of intradialytic hypertension.
Methods: A clinical study was conducted with end-stage renal disease (ESRD) patients under hemodialysis randomly selected from a University Hospital in Fortaleza, Brazil. Patients were selected through automated ambulatory blood pressure monitoring, and those with confirmed diagnosis of intradialytic hypertension were selected (group A). A control group consisted of patients who presented decrease in blood pressure during hemodialysis sessions (group B). Patients were monitoring every hour by blood pressure measurement and echocardiogram during hemodialysis session. Before and every hour during dialysis session blood samples were collected for brain natriuretic peptide (BNP) cathecolamines, endothelin and nitric oxide measurement.
Results: A total of 21 patients were included in the study (10 in group A and 11 in group B), with a mean age of age of 43±4.9 years, and 54.6% were female. There was an increase in the mean arterial blood pressure by 23mmHg in group A, and a decrease by 17mmHg in group B ($p<0.0001$). There was no significant change in the ejection fraction during hemodialysis in both groups. There was an increase in cardiac output during hemodialysis in both groups. There was no significant difference in cardiac frequency between the two groups. The mean values of BNP varied from 1643pg/mL before dialysis to 1574pg/mL after dialysis in group A ($p=0.94$) and from 1720pg/mL to 1382pg/mL in group B ($p=0.83$). The values of endothelin varied from 10.8pg/mL before dialysis to 25.9pg/mL after dialysis in group A ($p<0.001$) and from 11.1pg/mL to 13.3pg/mL in group B ($p=0.78$). The values of adrenalin varied from 208pg/mL to 80pg/mL in group A and from 153 to 212pg/mL in group B before and after dialysis ($p=0.20$). The values of noradrenalin varied from 337pg/mL to 194pg/mL in group A and from 488 to 319pg/mL in group B before and after dialysis ($p=0.20$). The values of dopamine before dialysis was 76pg/mL in group A and 71pg/mL in group B ($p=0.61$) and 76pg/mL in group A and 79pg/mL in group B after dialysis ($p=0.67$).
Conclusions: Patients with ESRD present different hemodynamic patterns during hemodialysis session, with a group presenting significant increase in blood pressure. The main contributing factor to this phenomenon seems to be the serum concentration of endothelin, which significantly increased during hemodialysis.
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Su614 EFFECT OF PARATHYROID HORMONE (PTH) ON GLUCOSE HOMEOSTASIS IN CHRONIC HEMODIALYSIS PATIENTS

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Introduction and Aims: Abnormal glucose homeostasis is a well known feature of uremic patients and might contribute to the increased incidence of atherosclerosis and cardiovascular morbidity in these patients. Several studies demonstrated the presence of insulin resistance and impaired glucose tolerance while others demonstrated pancreatic beta cell dysfunction in uremia. As hyperparathyroidism is a common feature in chronic hemodialysis patients and has been incriminated in many pathophysiological problems in these patients, it is of great interest to look into the relation between parathyroid hormone (PTH) and abnormal glucose homeostasis in chronic renal failure (CRF) patients treated with regular hemodialysis. The aim of the present work is to study the relation between PTH and glucose homeostasis variables, analyzing the effect of lowering PTH on them.
Methods: Forty one (28 males and 13 females; age range 19-64 years) non-diabetic chronic hemodialysis patients are studied after excluding cases with impaired glucose tolerance and those taking any medications that affect carbohydrate metabolism. In addition, 40 age- and gender-matched healthy persons are included as a control group. Informed consents are obtained from all subjects in the study. In addition to the routine biochemical and hematological investigations, serum PTH, fasting glucose, and fasting insulin are tested in all studied persons. Homeostasis model assessment of insulin resistance (HOMA-IR), as a measurement of insulin resistance, and HOMA-B, as a measurement of pancreatic beta cell function are calculated. Patients with plasma PTH level >450 pg/ml (n=17) are given 1-2 microgram(s) intravenous (I.V) one alpha-cholecalciferol thrice weekly at the end of the hemodialysis session for 4 consecutive months.
Results: There is significant increase in fasting insulin level in the studied hemodialysis patients versus control (10.5±6.7 vs. 6.6±0.9 IU/ml; $p<0.001$), associated with increased HOMA-IR (2.5±1.7 vs. 1.5±0.3; $p<0.001$). The pulsed dose of I.V. 1-alpha cholecalciferol is associated with decreased PTH level (before=1251±597; after=827±202 Pg/ml; $p=0.09$) and significantly improved pancreatic beta cell function (HOMA-B, before=106±117; after=181±48; $p=0.02$).
Conclusions: Insulin resistance is a constant feature of chronic hemodialysis patients; the associated hyperfunction of beta cell with the resultant hyperinsulinemia prevents marked hyperglycemia. Secondary hyperparathyroidism is linked negatively to beta cell function, that is improved significantly with intermittent pulsed intravenous 1-alpha cholecalciferol as an effective method of lowering the elevated serum PTH.

Su615 ONE YEAR OBSERVATION OF ON-LINE HEMODIAFILTRATION (HDF) ON SOME PATTERNS IN UREMICS

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Introduction and Aims: On-line hemodiafiltration (OL-HDF) is for physiological reasons the most advanced form of extracorporeal renal replacement therapy. This approach gives possible to improve the still poor prognosis of hemodialysis (HD). However, the possible benefits of this therapy have not been well defined. The purpose of study was to assess the influence of OL-HDF on dry weight, mean arterial pressure (MAP) and serum levels of total cholesterol (TCh) and triglycerides (Tg) in patients treated before by HD.
Methods: Thirty two uremics (25 non diabetic mellitus -NDM and 7 diabetics type-2 – DM) participated in this study. The NDM group had a mean age 64±14.56 and DM 56.67±15.95 yrs, an average time of HD was 48.42±41.64 months for both groups. Renal disease was caused in NDM by chronic glomerulonephritis (9), hypertensive nephrosclerosis (10), polycystic disease (6), amyloidosis (4). NDM had been stable for 6,5 and DM 5,5 yrs on maintenance bicarbonate HD, scheduled thrice weekly 12 hrs/week, achieved mean single pool Kt/V_{urea} about 1.0±0,2, with low-flux

membrane. Then they were switched to post-dilution OL-HDF. The OL-HDF was provided by 5008 Therapy System, using high-flux FX membrane, consisted of Qb-300 ml/min. Qd-500 ml/min, Kt/V_{urea} was 1,2±0,3, and time of OL-HDF was 240 min., with 10-17L hemofiltrate exchanges of the replacement fluid. Both groups of patients during HD have been treated with 40 mg/day of simvastatins and ACE inhibitors/angiotensin II receptor antagonists. At the start of OL-HDF the dose of simvastatins was reduced to 20 mg. Predialysis blood samples were drawn twice, at the beginning and after 12 months of study. The measurements of dry weight (kg), MAP (mmHg) were estimated by conventional method and TCh, Tg (mg/dl) by enzymatic assays. Means and standard deviations were calculated by conventional methods, and the statistical difference was determined by a paired Student's t – test.

Results: At the start of study the predialysis dry weight in NDM and DM was 67,4±15,2 and 77,7±14,7 respectively,which was significantly higher $p<0,05$ in DM. After 12 months of study dry weight in NDM was 66,5±14,7 and in DM 78,8±15 which did not change significantly in comparison to the begining. The MAP declined in NDM from 89,8±10,8 to 84,0±9,2 and in DM from 95,3±8,3 to 86,2±9,9 in studied patients. These values were significantly ($p<0,05$) different between two groups and the periods of study. The mean levels of TCh, at the start were in NDM 198,9±28,6, in DM 205,0±30,4, showed a non-significant trend to decrease throughout the study in both groups: 185,7±35,0, 183,6±34,0 respectively. The mean levels of Tg in NDM 165±78,2 declined to 157,2±90, in contrast to DM which was 259,3±161 and decreased statistically ($p<0,05$) to 182,3±73.

Conclusions: The change from HD to OL-HDF was associated with better dialysis adequacy, stable dry weight and better blood pressure control, allowing for reducton the dose of antihypertensive drugs about 40%. The levels in serum of total TCh and Tg especially in diabetics declined, even lower dose of simvastatins. These results showed beneficial effect of OL-HDF therapy, however futher studies are needed.

Su616 ANALYSIS BY FLOW CYTOMETRY OF THE EFFECT OF HAEMODIALYSIS ON OXIDATIVE STRESS IN LEUKOCYTES OF PATIENTS WITH END STAGE RENAL DISEASE

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Introduction and Aims: Oxidative stress (OS) is a situation in which there is increase in intracellular reactive oxygen species (ROS) that are potent oxidants and highly toxic because they react with cell molecules, altering their structure and activity. Cells have developed antioxidant activities responsible for restoring intracellular redox levels. Patients with end stage renal disease (ESRD) undergoing hemodialysis (HD) present increased OS which may lead to cardiovascular disease, malnutrition, poor outcome and low survival. We have applied flow cytometry (FCM) to assess OS in leukocytes from ESRD patients, as this technology allows to detect sensitively intracellular alterations in heterogeneous populations.

Aims: To compare the effect of a single HD in OS parameters in ESRD patients undergoing their first HD and in chronic HD patients (Chronic HD), which had undergone repeated HD. Biomarkers of OS were the intracellular levels of superoxide anion (SOX), peroxidative activity (POX) and glutathione (GSH).

Methods: 30 patients (19 males;11 females; age: 64.54±15) divided into two groups. "First HD": 7 patients starting HD for the first time and "Chronic HD": 23 patients receiving at least six months HD in our Nefrology Service. Peripheral blood samples were drawn before and after the first HD (First HD group) or in the second session of the week (Chronic HD group). OS was determined by FCM (Cytomics FC500 MCL, Beckman Coulter) in leukocyte subpopulations from peripheral blood by flow cytometry (FCM). Leukocyte subpopulations were identified by staining with CD45-PC7 antibody. Intracellular OS was quantified with flurochromes dihydroetidhium (HE) to detect SOX and Dihydrorhodamine 123 (DHR) to quantify POX. Chloromethil fluorescein diacetate (CMFDA) was used to detect intracellular GSH. To evaluate the sensitivity of ESRD leukocytes to exogenous oxidants, sample aliquots were treated with the SOX generator menadione (MD) as a positive control for induced OS.

Results: Before HD, ROS levels were increased in the Chronic HD group as compared with First HD group. Levels of GSH were higher in the First HD group. Following HD, SOX levels decreased in neutrophils in both groups without significant difference. GSH levels and POX activity decreased in the First HD group and increased in the Chronic HD group, with significant differences for GSH. In vitro treatment with MD increased significantly POX activity in all samples Pre- and Post-, but SOX decreased in the First HD group and increased in the Chronic HD group.

Conclusions: OS in leukocyte subpopulations is lower in the First HD group than in the Chronic HD group and cells have higher antioxidant capacity. Long term HD treatment decreased the capacity of leukocytes to restore the OS balance after a single HD. These results suggest that loss of antioxidant molecules through the HD membranes may occur in the course of HD.

Su617 DOES GLUCOSE PRESENT IN DIALYZING FLUID INFLUENCE ON CHOSEN ACUTE PHASE REACTION FACTORS, NON-ENZYMATIC ELEMENTS OF PLASMA ANTIOXIDATIVE SYSTEM AND THE LEVEL OF CELL DAMAGE DUE TO OXIDATIVE STRESS?

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Introduction and Aims: Patients suffering from chronic renal failure (CRF) are exposed to increased oxidative stress. It is generated by uremic toxins and hemodialysis process itself. As a result of the contact with dialyzing membranes and blood lines surfaces' the neutrophiles and monocytes liberate cytokines and the large amount of reactive oxygen species (ROS). It leads to chronic inflammatory state and cell damage (e.g. hemolysis). Apart from inter-cells free radical scavengers, there are also endogenic non-enzymatic plasma compounds like albumines, bilirubine and uric acid, which play the protective role as the first target against ROS.

The aim of our study was to establish the influence of glucose presence in dialyzing fluid on acute phase reactions intensity, concentration of non-enzymatic elements of plasma antioxidative system and indicators of hemolysis.

Methods: The erythrocytes and plasma were collected from 30 patients with CRF before and after the session of hemodialysis. The polysulfon unit with glucose–free [HD-G(-)] or glucose supplemented [HD-G(+)] dialysate were used. The following parameters in plasma were analysed: concentrations of albumine, uric acid, bilirubine (spectrophotometric method), haptoglobine, CRP (immunoturbidimetric method) and LDH activity (spectrophotometric method). The statistical analysis were performed using non-parametric U-Mann Whitney's range test and Wicoxon's pair test, $p<0,05$.

Results: The concentration of CRP was significantly higher in HD-G(-) group both before and after hemodialysis sessions, than in HD-G(+) ($p<0,001$). There was no significant difference in haptoglobin concentration, however its level was over the method's norm range in all examined patients. The bilirubine concentration achieved significantly higher values in HD-G(-) group before and after hemodialysis compared to HD-G(+) ($p<0,04$). The hemodialysis treatment caused a significant increase of LDH activity in all those examined ($p<0,02$).

General characteristics of individuals and examined parameters (mean ± SD)

Parameter	HD-G(+), 15 patients		HD-G(–), 15 patients	
	Before HD	After HD	Before HD	After HD
Age [years]	57±6		54±10	
Albumin [g/dL]	4.5±0.2	4.4±0.1	4.3±0.2	4.2±0.1
Total bilirubin [mg/dL]	0.26±0.12	0.24±0.10	0.29±0.12	0.30±0.15
Haptoglobin [mg/L]	1483.4±826.0	1490.0±880.0	1165.1±647.9	1200.2±978.0
CRP [mg/L]	4.44±0.41	3.63±0.46	10.83±3.99	8.93±2.73
Uric acid [mg/dL]	9.33±2.74	7.45±2.39	8.46±1.02	7.10±1.76
LDH [U/L]	363.4±67.5	422.9±87.4	250.7±93.7	306.7±121.8

Conclusions: Glucose present in dialyzing fluid decreases the intensity of acute phase reactions and cell damage, but does not influence on using the plasma antioxidative compounds.

Su618 SERUM TUMOR MARKERS IN PATIENTS ON DIALYSIS

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Introduction and Aims: Tumour markers (TM) are widely used for monitoring the progression of various tumors or to assess their response to therapy, but the specifity of some TM in hemodialysis patients is controversial.The aim of this study was to analyze the influence of renal function and hemodialysis on the serum levels of five TM; alphafetoprotein (AFP), carcino-embryonic antigen (CEA), cancer antigen-125 (CA-125), cancer antigen-19.9 (CA-19.9), and prostate specific antigen (PSA).
Methods: 78 patients on chronic hemodialysis (groupe A) and 50 healthy volunteers (group B), without clinical symptoms or signs of neoplasia were enrolled in this study. Age and sex distribution were comparable between the 2 groups. TM were measured once in the group B, and before and after dialysis session in the group A. Postdialysis values were compared with predialysis values in order to determine the influence of hemdialysis with low-flux polysulfone dialysers, on serum concentration of TM. All measures were performed via the ElectroChemiLuminescence method with ELECSYS 2010 analyzer (Roche Diagnostics GmbH, Mannheim, Germany).
Results: Predialysis levels of CEA and CA 19-9 were significantly higher in group A when compared to groups B (4,25±2,89 vs 2,41± 1,82 p=0,0001 and 19,65±25,02 vs 10,23±11,00 p= 0,014 respectively). There were no significant differences among the two groups in the serum concentrations of CA125,CA 15-3 and PSA. Postdialysis values, after being corrected for haemoconcentration, were compared with predialysis values. A significant increase was observed in in all TM levels: CEA (4,25±2,89 vs 5,03±3,30 avec p=0,0001), CA 125 (27,84±92,27 vs 31,41±95,86 avec p=0,0001), CA 15-3 (14,00±6,63 vs 16,64±7,69 avec p=0,0001), (CA 19-9 (19,65±25,02 vs 26,50±41,17 avec p=0,002), PSA (1,22±1,23 vs 1,44±1,50 avec p=0,0001).
Conclusions: Our results suggests that serum levels of CEA and CA 19-9 are not a good TM for hemodialysis patients, and serum CA125,CA 15-3 and PSA were as valuable as they were in patients with normal kidney function.
Also the interpretation of TM levels in dialysis patients must be made with caution, because hemodialysis affect serum levels of all TM.

Su619 ACUTE EFFECT OF HEMODIALYSIS ON MICROCIRCULATION ASSESSED BY NEAR-INFRARED SPECTROSCOPY

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Introduction and Aims: Patients (pts) with chronic kidney disease (CKD) on maintenance hemodialysis (HD) present with microcirculatory alterations, attributed to: a) endothelial damage caused by oxidative stress, b) defect in skeletal muscle mitochondrial metabolism, c) obstruction due to microbubbles, which originate in extracorporeal lines, d) decreased coronary flow reserve, e) vascular calcification and f) vitamin deficiency.
Purpose: The aim of this study was to assess the acute effect of HD on the microcirculation as assessed by Near Infrared Spectroscopy (NIRS) and the vascular occlusion technique and to correlate the microcirculatory alterations with dialysis parameters (filter, duration and adequacy).
Methods: Thenar tissue oxygen saturation (StO_2%) was measured noninvasively by NIRS (occlusion technique). The study was carried out in a Renal Unit. The vascular occlusion technique was performed in order to assess the oxygen consumption rate, the endothelial function and the vascular reactivity. Dialysis parameters (filter, duration and adequacy), the medical treatment, the aetiology of CKD and the comorbities were also recorded.
Results: Measurements were performed in 20 HD pts (15 men, 5 women) with age 70±12 years, kt/V: 1,2±4, Et: 38±3,3%, on HD for 4.5±3 years, before and after one HD session. HD pts improved the oxygen consumption rate (32±17%/min vs 46.68±27%/min before and after HD respectively, p=0.035) inclusive of the smokers and the diabetics. The microcirculatory parameters prior to the HD session were: StO_2 thenar: 77±4, oxygen consumption rate: 28±11 and vascular reactivity: 30±32.
Conclusions: HD improved the oxygen consumption rate of the thenar. However, the limited number of pts included does not allow us to define conclusions concerning the effect of HD on endothelial function and vascular reactivity. Further studies are requested to confirm these results.

Su620 HEMODIALYSIS TREATMENT INDUCES PERIPHERAL TOLERANCE

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Introduction and Aims: The increased susceptibility to infective events is an important cause of morbidity and mortality in the uraemic patient. This condition depends on the failure of the defensive immunitary system at multiple levels. One of the principal mechanisms responsible of the increased predisposition to infections is immunological tolerance that is controlled by sophisticated regulatory mechanisms. While central tolerance is controlled by thymus in the first years of aging, peripheral tolerance is under the control of T- lymphocytes regulatory activity. Particularly, CD4+ lymphocytes with high expression of IL-2 receptor (CD25bright+) are supposed to have a main role among those cells involved in the induction of tolerance. Moreover, the transcription factor forkhead (Foxp3) has been identified as an essential and specific marker of T-regulatory cells (T-reg). Foxp3 transcription factor plays a primary role in T-reg differentiation and function. The effect of haemodialysis (HD) on immunoregulatory activities of T-reg has not been extensively investigated; therefore the aim of our study was to evaluate the CD4+CD25+FOXP3+ T-cells in HD patients.
Methods: We evaluated seven patients under standard HD and seven healthy controls (CON). At the time of the study, all patients had been dialyzed for more than six months with new cuprophan membrane dialyzers three times a week. All patients used a bicarbonate bath. No patient had clinical or biologic signs evidence of infective or inflammatory diseases or malignancy; none was on steroid or immunosuppressive therapy. Blood sample was collected just before (B) and after (A) HD treatment, during the intermediate haemodialytic session. Peripheral blood mononuclear cells (PBMC) were isolated by gradient density centrifugation and stained with CD4-PE, CD25-FITC and Foxp3-PE antibodies, in order to obtain the percentage and the absolute number of CD4+CD25+Foxp3+ lymphocytes. Cytofluorimetric analysis was carried out using FACScan.
Results: Our results, number of cells (n) and percentage (%) have been reported as average ± SD in the table below.

(n/%)	CON	HD-B	HD-A
CD4+CD25 bright+	51±21/1.2±0.4	75±46/1.3±0.3	87±39*/1.7±0.5
Foxp3+	124±49/3.0±1.2	243±131*/4.6±1.8*	249±96*/5.2±2.8*
CD4+Foxp3+	80±36/1.9±0.7	172±100/3.4±1.1*	203±87*/4.0±1.5*

*$p<0.05$ vs CON.

Conclusions: Preliminary data of our study suggest that CD4+CD25bright+ and Foxp3+ cells with regulatory activity are significantly increased in HD patients, and this induction of promoting tolerance cells is chronic, in fact T reg cells are increased both before and after the treatment. These results suggest that haemodialytic treatment may induce cells involved in peripheral tolerance and this alteration may represent an ulterior mechanism of immunosuppression and, consequently, of infections in HD patients.

Su621 SELF-ASSESSED SLEEP QUALITY IN HEMODIALYSIS AND RENAL TRANSPLANT PATIENTS

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Introduction and Aims: Self-assessed sleep quality is poor in patients treated by chronic hemodialysis (HD) [*Int Urol Nephrol, 2005;37:159*]. The effect of renal transplantantion (Tx) is equivocal and some authors have even described worsening of self-reported sleep quality after kidney transplantation [*Nephrol Dial Transplant, 2005;20:194*]. We have therefore compared self-assessed sleep quality between HD and Tx patients.
Methods: We administered the Kidney Disease Quality of Life (KDQoL) sleep scale to 192 subjects (75 F,117 M) treated with HD, and 97 subjects (38 K, 59 M) with functioning renal graft.
Results: Self-assessed sleep quality score was significantly lower in HD patients as compared to Tx group (49.1±20.8 vs. 72.3±15.9; $p<0.0001$). In multiple regression analysis quality of sleep was related to employment status, depression and pain in HD patients but not in Tx patients.
Conclusions: Subjects with functioning renal graft have better self-assessed quality of sleep than HD patients.

Su622 DIAGNOSTIC VALUE OF NT-PRO-BNP FOR LEFT VENTRICULAR HYPERTROPHY IN HAEMODIALYSIS PATIENTS

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Introduction and Aims: Amino-terminal B-type natriuretic peptide (NT-proBNP) quantification is a useful marker of left ventricular hypertrophy (LVH), wich is associated with an increase in cardiovascular morbidity and mortality in general population. In hemodialysis patients, interpretation of NT-proBNP remains unclear, because of volume overload, reduced renal excretion and removal by dialysis procedure. The aim of this study was (i) to investigate the diagnostic value of the NT-proBNP level for detecting LVH in HD patients with conserved left ventricular ejection fraction (≥50%), (ii) to investigate NT-proBNP basal level alteration following hemodialysis.
Methods: We measured NT-proBNP levels in 30 stable patients on maintenance haemodialysis, before and after hemodialysis session with low-flux polysulfone dialyzers, all mesures were done by a third-generation assay (Elecsys Analyzer, Roche Diagnostics, Mannheim, Germany).We hypothesized that serum NT-proBNP cut-off value could serve as a biochemical marker to detect LVH in patients on haemodialysis treatment, regardless of chronic fluid overload. We assessed LV masse using trans-thoracic echocardiography, LVH was defined as an indexed left ventricular mass> 134 g/m^2 in man and 110 g/m^2 in woman. NT-proBNP cutoff values for LVH with different specificities and sensitivities were calculated by ROC curves.
Results: NT-proBNP levels increased significantly after hemodialysis sessions (5575,93±5509,53 versus 4114,3856,37± pg/ml $p<0,0001$). A significant positive correlation was found between NT-proBNP level and left ventricular mass ($r = 0.75$, $P < 0.0001$). NT-proBNP was significantly higher in patients with LVH: (5270,35±3410,23 versus 2477,27±1421,91pg/ml $P = 0,045$). In the multivariate regression analysis NT-proBNP was the only independent predictor of LVH ($r = 0.75$, $P < 0.0001$). An NT-proBNP level ≥ 4002pg/ml was the cut-off value for predicting LVH, with a positive predictive value of 78% (95% CI 0,22 – 0,39) and a negative predictive value of 77.8% (95% CI 0,77- 0,91), (area under the ROC curve 81,5% p =, $P < 0.0001$).
Conclusions: Our results suggests that NT-proBNP could be a potential marker of LVH in chronic renal failure patients on hemodialysis. Dialysis modalities like low-flux dialyzers influence NT-proBNP levels and should be taken into account in the interpretation of values.

Su623 EFFECT OF DIALYSIS WITH LOW AND HIGH (32 and 36 mmol/L) BICARBONATE CONTENT ON CONTROL OF METABOLIC ACIDOSIS

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Introduction and Aims: Acidosis in hemodialysis (HD) petients leads to development of bone disease, growth retardation, impaired myocardial contractility, hipoalbuminemia and is associated with increased mortality. It should be attempted to maintain serum bicarbonate levels at 22mmol/L in patients. We aimed to investigate effects of dialysate solution containing 32 and 36 mmol/L bicarbonate on control of metabolic asidosis in maintenance HD patients.
Methods: Patients receving HD thrice weekly at a private Fresenius Konya HD center were started to be dialysed with 1/44 concentration (36 mmol/L bicarbonate) instead of their previous usual 1/34 concentration (32 mmol/L bicarbonate) dialysate solution. 91 patients (43 males, 48 females) were included in the study. Mean age was 57±12, and mean dialysis vintage was 4 years. We comperated pre- and post-dialysis bicarbonate levels attained with use of either 32 mmol/L or 36 mmol/L bicabonate containing dialysate.
Results: Mean plasma bicarbonate level was 20.51±2.42 mmol/L with of dialysate containing 32 mmol/L bicarbonate. This value was below the recommended guideline targets. Only 90 patients (33%) achieved recommended target bicarbonate. Mean bicarbonate level was 23.59±3.33 mmol/L with use of dialysate containing 36 mmol/L bicarbonate. 19 patients (20.8%) had serum bicarbonate level more than 30 mmol/L.

Laboratory parameters with use of dialysate comprising 32 and 36 mmol/L bicarbonate

	Bicarbonate		p
	32 mmol/L (n=91)	36 mmol/L (n=91)	
Kt/V	1.44±0.25	1.49±0.28	NS
Urea (mg/dl)	141.21±35.49	162±41.01	NS
Potassium (mEq/L)	4.75±0.69	5.39±0.74	NS
Albumin (g/dl)	3.79±0.35	3.63±0.29	NS
Predialytic plasma bicarbonate (mmol/L)	20,51±2,42	23,59±3,33	<0,001
Postdialytic plasma bicarbonate (mmol/L)	26,04±2,43	28,81±2,09	<0,001
C-reactive protein	1.69±2.15	1.38±2.49	NS

NS: Not significant.

Conclusions: Pre- and post-dialysis bicarbonate levels were found to be different for each patient. Acidosis control was achieved beter with dialysis solution containing 36 mmol/L bicarbonate. However, approximately one fifth of patients developed significant alkalemia at the end of dialysis. This level of alkalemia may trigger arrhythmia generation. Thus, patients who have arrhythmia and prone to alkalosis should not be dialysed with high bicarbonate dialysate, if it is going to be used, the machine bicarbonate value should be set at between -4 or -8.

Su624 CARDIAC STRUCTURE ABNORMALITIES AND ELECTROCARDIOGRAM ASSESSMENT IN PATIENTS WITH AND WITHOUT METABOLIC SYNDROME IN HAEMODIALYSIS: A CASE-CONTROL STUDY

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Introduction and Aims: Metabolic syndrome (MS) is characterized by the presence of metabolic hydrocarbonate disorders, high blood pressure, lipid disorders and obesity. MS is also associated with several functions and structural disorders in the myocardium.
Objectives: 1.- Describe the main clinical features, biochemical values and cardiac abnormalities in MS on HD. 2.- Compare the data collected in MS with the other dialysis patients.
Methods: Analysis of clinical features, Echo -Doppler data and ECG register beetween MS and non MS haemodialysis patients. All the patients

underwent a Doppler echocardiography and ECG register. MS patients were diagnosed using NCEP-ATP III criteria. Alterated E/A wave relation defined DD. LVH criteria: 1.- Left ventricular mass index (LVMI)> 125 g/m^2 (men); > 110 g/m^2 (women); 2.- Sokolow–Lyon voltage ≥ 38 mm, 3.- Cornell voltage ≥ 28 mm (men); ≥ 20 mm (women), 4.- Cornell Product Voltage (CPV) > 2440 mm x mseg.
Results: We analyzed 55 patients on haemodialysis. MS were diagnosed in 24 patients.Demographical data showed MS patients were younger, had more abdominal perimeter and BMI. MS were more diabetic (76 vs 37%, p<0.05) and dyslipemics. A significant non favorable metabolic profile was shown in MS vs non MS. Echocardiography (MS vs non MS): LVMI 157±45.1 vs 138.9±42.3 g/m^2 (p<0,05), PWT 11.8±2.2 vs 11.2±2.3 mm, LVTD 52.1±9.8 vs 48.8±7.2 mm, LV LVTEF 56.5±15.5 vs 66.5±12.1% (p<0,05).MS had more atrial fibrilation (16 vs 8%, p<0,1) and diastolic dysfunction (42 vs 30%, P<0.1).The prevalence of LVH was higher in MS for LVMI,Cornell voltage and CPV. Sokolow–Lyon voltage did not found neither LVH in both groups. In relation to non fatal cardiovascular events, MS were totally higher, mainly due to isquemic heart disease (21 vs 6%; p<0,05) and peripheral vascular disease (26 vs 15%, p<0.1).
Conclusions: 1.- We observed a high prevalence of cardiac abnormalities in MS dialysis patients in our study. 2.- Echocardiogram data results more useful than ECG register in order to evaluate LVH. 3.- The worst functional and structural cardiac abnormalities in MS patients could explain in part, the more comorbidity and cardiovascular events in these patients.

Su625 CORRELATION BETWEEN YEARS ON HEMODIALYSIS AND LIVER FUNCTION IN PATIENTS ON MAINTENANCE DIALYSIS

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Introduction and Aims: The galactose single point (GSP) test attempts to assess functional liver mass by measuring its blood concentration 1 hour after administration. To date, there is no clinical report about how hemodialysis (HD) may affect GSP values. The purpose of this study was to investigate the influence of HD on short-term and long-term liver function by application of the GSP test.
Methods: Seventy-four patients on maintenance HD (46 males and 28 females, 55.3±14.0 years old with a mean time on HD of 4.90±3.89 years) were included in this study. The patients were on HD 3 times per week using low-flux polysulfone membrane. Patients with galactose intolerance (questionnaire) or significant liver disease (questionnaire, routine liver function test, liver sonography) were excluded from this study. The GSP test was performed after an 8-hour fast. Fourteen patients were selected for assessment of GSP test data before and after a HD session.
Results: Among the 74 HD patients, correlation studies between blood biochemistry parameters (BUN, Cr, AST, ALT, Albumin, Uric acid etc.), years on HD and GSP levels revealed that only the post-HD Cr levels and years on dialysis (figure 1) were significant correlated with the GSP level (r=0.280, p<0.05 and r=-0.240, p<0.05 respectively). Our data shows that patients with longer years on HD apparently have better liver function.
Among the 14 patients selected, the hepatic clearance of galactose was similar before and after a HD session (pre-HD 410±254 μg/ml, post-HD 424±288 μg/ml, p=0.73). In these patients, pre-HD GSP levels showed significant correlation with post-HD values (r=0.851, p<0.01).
Conclusions: Patients on maintenance hemodialysis for several years may experience improvement of their liver function. However, a single HD session does not appear to significantly influence liver function. Since the metabolism of galactose is dependent on liver blood flow and hepatic functional mass, whether the blood flow to the liver is slightly increased after HD, which may result in improved liver function as reflected in the GSP test, deserve further study.

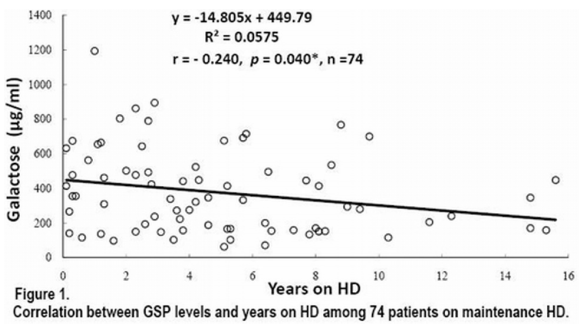

Figure 1.
Correlation between GSP levels and years on HD among 74 patients on maintenance HD.

Su626 HDL-CHOLESTEROL SUBCLASSES IN PATIENTS ON HEMODIALYSIS

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Introduction and Aims: Dyslipidemia is a common symptom in patients suffering from chronic renal failure on hemodialysis (HD)and also a factor contributing to accelerated atherogenesis. This study intends to examine the serum levels of HDL_2 vs. HDL_3-cholesterol subfraction which are considered to reflect better the protective effect of HDL compared to total HDL-C.
Methods: Thirty-three patients (age 57.8±3.0 years, average ± SEM), regularly hemodialysed (average period 95.5±15.5 months) were included (group A). A considerable percentage of them (30%) suffered from cardiovascular disease, while none of them had recent heart event, fever or known infection. Serum samples were collected prior to mid-week dialysis sessions and stored in -30°. C-LDL-Cholesstelol Apo-A1 and HDL- Chol, were measured. HDL subfractions were isolated according to a single-step precipitation method by heparin/DS/$MnCl_2$ reagent. The results were compared by Mann-Whitney test with a group of 15 healthy, matched by age and sex individuals(group B).
Results: Serum total cholesterol,LDL-C,Apo-A1 concentrations were significantly lower in group A than in B (p=0.010,0.002,0.003,respectively). HDL_2 and HDL_3 -C levels in group A (29,5±1.7 and 10.9±0.5mg/dl, respectively) were significantly lower than in group B (35.6±2.2 and 22.9±1.6 mg/dl, p=0.0407 and <0.0001 respectively).This was followed by an increased HDL_2/HDL_3 -C ratio in group A compared to B (p<0.0004).
Conclusions: It is concluded that patients on hemodialysis have lower levels of HDL_2 and HDL_3 -C subclasses compared to healthy individuals. This may reflect an alteration of serum HDL-C composition, which probably affects its antioxidant activity and constitutes a risk factor of atherogenesis in those patients.

Su627 TUBERCULOSIS IN PATIENTS ON HEMODIALYSIS IN AN ENDEMIC REGION

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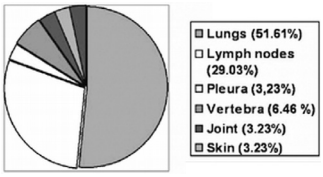
Introduction and Aims: Clinical presentation of tuberculosis is different in patients on hemodialysis than in the general population. Diagnosis is usually late due to nonspecific clinical presentations, frequent extra pulmonary involvement, negative tuberculin skin test and low probability of microbiologic evidence. This study aimed to analyze hemodialysis patients with tuberculosis in Istanbul.
Methods: Patients who were on chronic hemodialysis program in Istanbul for more than 3 months and diagnosed to have tuberculosis at least three months after the start of hemodialysis were included in the study. Their demographic and clinical data were analyzed using SPSS for Windows ver. 13.0.
Results: Of the 925 patients screened from 7 different centers, 31(3.35%) were found to have tuberculosis. The mean age was 52.3±13.5 years. Male/female ratio was 18/13. The mean duration of dialysis therapy

was 62.6±54.3 months; whereas the duration of hemodialysis up to the diagnosis of tuberculosis was 21.7±25.7 months. Extra pulmonary tuberculosis constituted 48.39% of the cases.



Eleven patients had prior history of tuberculosis. Twenty seven patients received isoniazide+rifampisin+ethambutol+pyrazinamide; one had isoniazide+rifampisin+ethambutol+a quinolone antibiotic; while in three patients drugs other than isoniazide and rifampisin were unknown. Treatment ended with cure in 18(58.05%) patients; was still ongoing in 12(38.70%) patients and one(3.25%) died of pulmonary tuberculosis.

Conclusions: With the 925 patients screened, this study is the largest scaled in our country. The lower incidence in the present study when compared with the previous studies may be related to the differences in the diagnostic criteria and the decrease in both incidence and prevalence of tuberculosis in recent years all over the world. According to the 2009 statistical reports of Ministry of Health; Istanbul is the city with the highest incidence and prevalence of tuberculosis (50.8/100000 population) but also the city with the highest rate of immigration. To minimize the effect of this factor, we included only patients who had started and continued their dialysis program in Istanbul. Demographic parameters and the primary kidney disease of the patients in our study were quite similar to the average dialysis population in Turkey. That's why we can not address a subpopulation in the hemodialysis population who has additional risk factors for tuberculosis.

It is important to prevent tuberculosis in hemodialysis patients due to difficulties in the diagnosis and frequent side effects of treatment. So we recommend routine screening of hemodialysis patients; isolate and treat effectively infected patients.

Su628 PULMONARY CONGESTION IN PERITONEAL DIALYSIS PATIENTS

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Introduction and Aims: Chest ultrasound (US) is a simple technique which provides reproducible estimates of lung water in intensive care patients and in patients with heart failure. When pulmonary congestion occurs the beam of chest US is reflected by subpleural thickened interlobular septa thus generating hyperechoic reverberation artefacts between thickened septa and the overlying pleura which are defined "lung comets" (LCs). A recent study exploring the validity of this technique in hemodialysis patients showed that LCs are strongly related with echocardiographic indicators, particularly with left ventricular ejection fraction (LVEF) [*Mallamaci F. et al, JACC imaging 2010 (in press)*].Pulmonary lung water in peritoneal dialysis (PD) patients has not been studied.

Methods: We investigated clinical and echocardiographic correlates of LCs in a series of 58 consecutive PD patients treated in 3 centers (age: 62±13 yrs). In a subgroup of 33 patients the hydratation status was also assessed by Body Impedance Analysis (BIA) and clinical evaluation.

Results: Chest US and the measurement of the number of LCs were successfully obtained in all patients. Moderate to severe pulmonary congestion (LCs ≥ 14) was found in 22 patients (38%). On univariate analysis, LCs were strongly and inversely related to LVEF (rho= – 0.54, P<0.001). In a logistic regression model, a 5 LCs excess entailed 26% increase in the odds of having a LVEF below the median value (odds ratio: 1.26, 95% CI: 1.05-1.52, P=0.015). LCs were useful to discriminate patients with LVEF below/above the median value, the area under the corresponding ROC curve (AUC) being highly significant (AUC=0.79, P<0.001). LCs were unrelated to BIA and clinical evaluation of the hydratation status (P=NS).

Conclusions: Pulmonary congestion is common in PD patients and poorly associated with hydration status. The strong association of lung comets with LV systolic function indicates that this indicator provides relevant information on cardio-pulmonary status in peritoneal dialysis patients. A clinical trial is needed to see whether chest US is useful in clinical care.

Su629 DIALYTIC BALANCE AND SERUM LEVELS OF MAGNESIUM DURING OLHDF IN PATIENTS RECEIVING Ca-ACETATE/Mg-CARBONATE AS PHOSPHATE LOWERING MEDICATION

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Introduction and Aims: Only scarce information is available about the changes in serum magnesium levels in dialysis patients during dialysis and between dialysis sessions. Therefore we performed a clinical trial to investigate these changes.

Methods: Patients, stable on three times per week online haemodiafiltration (OLHDF) (5008 online plus® machine with a FX800® dialyser; Fresenius Medical Care, Bad Homburg, Germany) with 1.25 mmol/L dialysate calcium (dCa) and 0.5 mmol/L dialysate magnesium (dMg), were recruited according to their PTH lowering medication: Group VC: oral calcitriol ≥1.0 μg/dialysis (N=6) and Group CC: cinacalcet ≥30 mg/day (N=8). All patients received Ca-acetate/Mg-carbonate (OsvaRen®) as phosphate lowering medication. Determination of dialytic balances was done during an OLHDF session with 1.25 mmol/L dCa and 1.0 mmol/L dCa subsequently. Serum samples of electrolytes and iPTH were drawn during OLHDF and during two non-dialysis days, respectively.

Results: There were no differences in serum magnesium levels or dialytic balances detected, neither between medication groups nor between OLHDF sessions. Therefore all patients were evaluated together. Interestingly we could not observe a correlation between the amount of Ca-acetate/Mg-carbonate taken and serum Mg2+ levels (r=0.213; p=0.456). Over the whole investigational period the mean fasting serum Mg2+ level was 1.19±0.23 mmol/L. Patients were divided into two equal groups according to their fasting serum Mg2+ levels. Patients with higher(1.25±0.18 mmol/L) mean fasting Mg2+ serum levels had lower iPTH serum levels than those with lower (1.02±0.12 mmol/L) serum Mg2+ levels (136.8±64.0 ng/mL vs. 219.6±82.4 ng/mL; p=0.058). The correlation between fasting serum Mg2+ and iPTH levels was inverse and borderline significant (r=-0.499; p=0.070). There was no difference in iCa, tCa, or P serum levels between these patients. The mean magnesium elimination during the OLHDF sessions was 267.33±113.91 mg. Mg2+ levels decreased during OLHDF from 1.17±0.28 mmol/L to 0.87±0.08 mmol/L (p<0.0001). During a day without OLHDF there was nearly no variation in serum Mg2+ levels (mean CV=7.3%±5.8%), as such there was no influence of food or Ca-acetate/Mg-carbonate intake. Between OLHDF treatments serum Mg2+ increased by 32% (0.90±0.10 mmol/L to 1.19±0.27 mmol/L; p<0.0001) during a 2-day interval and by 42% (0.87±0.08 mmol/L to 1.24±0.30 mmol/L; p<0.0001) during a 3-day interval.

Conclusions: There was an inverse relation between serum Mg2+ levels and serum iPTH levels which confirms the results of other studies. Furthermore, we conclude from our observations that serum Mg2+ levels are fairly stable under treatment with Ca-acetate/Mg-carbonate and 0.5 mmol/L dMg. Single meals and the concomitant intake of Ca-acetate/Mg-carbonate did not show up in changes of serum Mg2+ levels.

Disclosure: The presenting author is employee of Fresenius Medical Care.

Su630 DECREASE RATE OF THE RENAL DIAMETER AFTER INDUCTION OF CHRONIC HEMODIALYSIS

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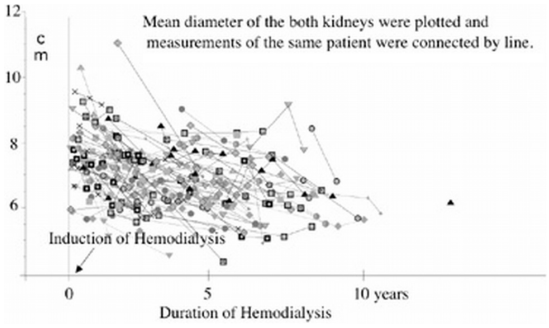
Introduction and Aims: After induction of hemodialysis(HD), it is well known that the kidney size gradually decreases. However, actual decrease

rate is poorly understood. We here present the results of ultrasonographic (US) evaluations on the alteration of renal diameter after induction of chronic HD.

Methods: Of 109 outpatient chronic HD patients who had neither severe acquired cystic disease of the kidney nor polycystic kidney disease, we performed US two or three times to measure their maximum renal diameter (mean of both kidneys), and the yearly alteration rate after induction of HD was calculated. The decrease rate was compared among the original disease, age, gender and duration of hemodialysis.

Results: The average interval of two measurements was 35.9 months and the average HD duration from the HD induction to the first measurement was 29.5 months. The measured raw data are shown in the figure.



The average decreasing rate of renal diameter in all patients was 4.34±0.4 (SE) mm/year. No difference was seen on the decrease rate in relation to gender, age and the original disease (three groups; glomerulonephritis and IgA nephropathy, diabetes, and others). However, the decrease rate was large when the first measurement was close to the induction of hemodialysis, suggesting that the alteration rate reduced according to the duration of hemodialysis (5.2±0.8 first measurement not more than 10 months after induction of HD, 1.5±1.6 first measurement 90 to 100 months after induction of HD).

Conclusions: Renal diameter decreased approximately 4.3 mm each year and the decrease rate slowed as the duration of hemodialysis increased.

Su631 EVOLUTION OF DIABETIC PATIENTS OLDER THAN 65 YEARS STARTING HEMODIALYSIS. COMPARATION WITH CONTROL GROUP

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Introduction and Aims: In the last years we have seen a significative increase of patients starting hemodialysis (HD) due to diabetic nephropathy older than 65 years. This kind of patients has more cardiovascular risk factors, drove to more morbi-mortality than other patients.

Methods: We want study the evolution, demographic and clinical characteristics of these patients (Group I) starting HD in our service, versus patients starting HD with similar age and other cause of ESRD (Group II). Between September-2005 to September-2009, 165 patients incident HD. In total 68 (41%) patients older than 65 years. Median follow up was 23 months (5-51).

Results: Main results shown in Table 1.

Cause of death in Group I: 4 due to infection disease, 3 to decline in general status and withdrawal of dialysis. Main cause of death in Group II: 6 to decline in general status and 6 for cardiological pathology. In multivariate analysis, survival was significantly influence by DHI (p 0.001, HR 1.26, 95%CI 1.114-1.401).

Conclusions: In conclusion, patients starting HD with diabetic nephropathy older than 65 years have similar survival at medium time (12 months), although less at long time (36 months), that patient in Group II. The main reason is major comorbidity, basically in relation with major percentage of ischemic heart disease and peripheral vascular disease, with more number and day's hospitalization index. Both groups have at end follow up, laboratory values in concordance with guidelines.

Abstract Su631 – Table 1. Results

Characteristics	Group I (n=20)	Group II (n=48)	p
Mean age (year)	73,5±4 (65-82)	74,5±6 (65-85)	ns
Males, %	50%	56%	ns
Body Mass Index (kgr/m^2) *	24±3	23±3	ns
Charlson Comorbidity Index	8,5±1,4	7,6±2	0,029
Ischemic heart disease (%)	35	15	0,06
Cerebrovascular disease (%)	25	33	ns
Peripheral vascular disease (%)	65	42	ns
Hypertension (%)	95	81	ns
Dyslipidemia (%)	55	40	ns
Hemoglobin >11 gr/dl * (%)	60	64.5	ns
Ferritin levels 100-500 mg/ml * (%)	55	64.5	ns
Serum calcium levels 8,4-9,5 mg/dl * (%)	65	60.4	0,002
Phosphate <5,5 mg/dl * (%)	95	92	ns
PTH levels 150-300 pg/ml * (%)	35	31.5	ns
Albumin >3,5 gr/dl * (%)	40	50	ns
Number Hospitalization Index (NHI)	0,35 (0-1,05)	0,19 (0-0,88)	0,053
Day's Hospitalization Index (DHI)	5,13 (0-23)	2,88 (0-29)	0,034
Death	7 (35%)	22 (46%)	ns
Survival at 12 months	75%	80%	ns
Survival at 36 months	26%	46%	ns
Renal transplant	2 (10%)	4 (8%)	ns

NS, no significant. NHI: number hospitalization/patients/month. DHI: day's hospitalization/patients/month. *end study level.

Su632 CENTRAL MOTOR NEURON REMAINS INTACT IN HAEMODIALYSIS PATIENTS WITH NEUROPATHY

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Introduction and Aims: Functional disturbances of Peripheral Nervous System are well known as one of frequent and sometimes severe complications in patiens with ESRD. There are some electrophysiological signs for subclinical neuropathy in about 70% of patients even at start of dialysis treatment. Most studies are aimed to prove the injury of Preipheral Motor Neuron in these cases.

In this study we used Transcranial Electromagnetic Stimulation to asses the functional state of Central Motor Neuron in a group of haemodialysis patients.

Methods: Studied subjects were divided into two groups:

Group A: 20 patients on haemodialysis treatment from 10 to 194 months.

Group B: 20 healthy subjects matched for sex and age with group A.

Presence of peripheral neuropathy was proved in Group A by Electroneurography. Nerve Conduction Velocity, Distal Latency and Amplitude of Muscle Response were abnormal for Tibial and Peroneal Nerves in all 20 patients. 8 of them showed injury of Ulnar and Median Nerves as well. Aberration from normal values was significantly more severe for nerves of lower extremities.

Subjects of Group B showed no abnormal values in Electroneurography.

First site of magnetic stimulation was pointed towards left precentral brain sulcus. Second site of stimulation was at cervical intumescence. Intensity of stimulation was adjusted to obtain evoked muscle response from Abductor Muscle of 5th finger of right hand with shortest latency and highest amplitude.

Assessed parameters were: Latency (L1) and Amplitude (A1) of evoked response at Cortex stimulation and Intensity (I1) of applied stimul. Same parameters were taken by electromagnetic stimulation of Alfa-Motor Neurons (L2, A2, I2). Central Motor Conducting Time is extracted from difference between L1 and L2.

Results:

Table 1

Groups	Transcranial stimulation			Spinal stimulation			CMCT
	L1	A1	I1	L2	A2	I2	
A	23.78	477.21	66.53	15.38	283.31	67.53	8.4
B	20.67	623.33	57.6	12.79	578.00	67.00	7.93
t	4.5640	2.3537	1.3942	3.7376	3.7161	0.1403	0.8579
p	<0.001	<0.05	>0.1	<0.005	<0.005	>0.1	>0.1

Results showed significatly longer Latency and slower Nerve Conduction Velocity of evoked responses in patients with ESRD on Haemodialysis

compared to healthy subjects for Peripheral Motor neuron. For Central Motor neuron there was no difference found.
Conclusions: Although the injury of structures of Peripheral Nervous System is relatively frequent, the results of the study prove that Nerve Fibers of Cerebrospinal Tract remain unchanged in patients with Uraemic Neuropathy on Haemodialysis Treatment. We concider further detailed study of Uraemic Neuropathy an important issue to achieve main target – to improve the quality of life of our patients.

Su633 PREVALENCE OF LUPUS ANTICOAGULANT AND VASCULAR THROMBOSIS IN PATIENTS ON DIALYSIS: PRELIMINARY RESULTS

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Introduction and Aims: Patients with end-stage renal failure (ESRF) on dialysis frequently present thrombotic events. The antiphospholipid antibodies are involved in the genesis of thrombosis, and may be present in patients with ESRF on dialysis. The thrombotic events are more frequently linked with lupus anticoagulant (LAC) than with anticardiolipine antibodies in patients with SLE or other nephropathies. Moreover, the chronic infection by hepatitis B or C virus, frequent in patients on hemodialysis (HD), has been implicated as the cause of antiphospholipid syndrome. We analyzed the prevalence of the lupus anticoagulant and the possible association with thrombotic events in patients with ESRF on dialysis, with or without B or C hepatitis virus infection.
Methods: A total of 35 patients with ESRF on hemodialysis and 16 on peritoneal dialysis (excluding patients with SLE) were evaluated. Demographic data, ESRF etiology, time on dialysis, clinical history of thrombotic complications and presence of LAC were compared between patients on HD and peritoneal dialysis (PD), as well as between patients with and without B or C hepatitis virus infection. Two blood samples were collected from each patient (in a six-week interval) for lupus anticoagulant dosage using the ACCUCLOT.dRVVT Russell's Viper method.
Results: There was a 17.7% prevalence of lupus anticoagulant in the population studied. Age and prevalence of lupus anticoagulant were greater in patients on PD than those on HD, however there was no difference in thrombosis events in the two groups. Patients on HD showed longer time on dialysis and more frequent presence of anti-HCV and HBV antigen. The comparison of patients with or without hepatitis B or C virus infection on HD did not show a difference in the presence of lupus anticoagulant or frequency of thrombosis.
Conclusions: In the population studied of ESRF patients on dialysis, the prevalence of lupus anticoagulant were high and similar to that found in the literature. The presence of lupus anticoagulant was more frequent in patients on PD and of more advanced age, compared patients on HD. However, the thrombotic events for the two groups, as well as the presence of the lupus anticoagulant between patients with and without B or C hepatitis virus infection showed no difference.

Su634 HYPERHOMOCYSTENEMIA IN HEMODIALYSIS PATIENTS: EFFECT OF C677T MTHFR

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Introduction and Aims: Hyperhomocysteinemia, an independent risk factor for cardivascular diseases, is commonly found in adult patients with end-stage renal disease. Major determinants of elevated plasma homocysteine levels in these patients include folate and vitamin B12 deficiencies, a common C677T methylenetetrahydrofolate reductase (MTHFR) genotype and renal failure. Our study aim was to evaluate plasma total homocystein (tHcy), folate and vitamin B12 levels and C677T MTHFR genotype effect in hemodialysis (HD) patients.
Methods: We recruited 98 HD patients from our dialysis unit and 105 healthy renal controls on the renal national day occasion. Fasting plasma tHcy, plasma folate and vitamin B12 were measured by immunoassay on Axsym® Abbott. The C677T MTHFR was genotyped by PCR/RFLP.
Results: tHcy was significantly higher in HD than controls (11.21±5.68μmol/L versus 9.19±4.09 μmol/L, $p<0.05$), no significant difference was observed in folate and vitamin B12 levels nor in genotype frequencies CC (62.2% vs 57.1%), CT (24.5% vs 32.4%), and TT (13.3% vs 10.5%). tHcy was significantly higher in TT genotype than CC and CT in HD and controls. TT hyperhomocysteinemia was more pronounced in HD patients (20.06±6.5 μmol/L vs 15.79±4.01 μmol/L). In another hand, tHcy was not significantly different between vitamin B supplemented and not supplemented HD patients.
Conclusions: HD patients particularly those with TT genotype have elevated tHcy. Vitamin B12 and folate supplementation seems to be inefficient. So our perspective is to adapt a supplementation protocol to the MTHFR genotype.

Transplantation – basic research 2

Su635 THE RELATION BETWEEN CTLA-4 SINGLE NUCLEOTID POLYMORPHISMS AND ACUTE REJECTION IN KIDNEY TRANSPLANTATION

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Introduction and Aims: Cytotoxic T-lymphocyte antigen 4(CTLA-4) is a cell surface protein, downregulate the T-cell response at CTLA-4/CD28/B7 pathway. Recent studies have revealed that CTLA-4 gene polymorphisms are assosiciated with autoimmune diseases, but its role in allograft celluler rejection is still unclear. The aim of this study was to investigate possible role of CTLA-4 gene single nucleotid polymorphisms (SNP); -318C/T, +49A/G,-1661A/G, CT60A/G in kidney transplant (KTx) rejection.
Methods: The study was conducted on a total of 96 living-related KTx recipients and these patients were divided into two subgroups: Group1; acute rejection group (AR,n=49) Group2; no acute rejection group (NAR, n=47). Both groups were comparable in terms of age, gender, HLA mismatch, induction and maintance immunossupressive protocols. All individiuals with a medical history of autoimmune and inflamatuary diseases were also excluded. Age and sex matched healthy control (Group 3: n=50) was also used.
Genomic DNA was isolated from peripheral blood mononuclear cells. All CTLA-4 polymorphisms were determined by restriction fragment length polymorphism(RFLP) with the use of appropriate digestion enzymes.
Results: G1 and G2 baseline characteristics were similar for age (38,1±13,8 vs 34,5±10,4 years, NS), gender (77,6% vs74,5% male patients, NS) and HLA- mismatch (2,98±1,1 vs 2,77±1,1 NS). There was no difference between induction (p=0,352) and maintaince immunsupresion as all include calcinorin inhibitors for first three months and thereafter G1 vs G2 calcinorin/mTOR inhibitors; 48%/52% vs 55%/45% (NS) between two groups.
Genotype and allele frequencies among the three mentioned groups were showed similar distributions for SNP; +49A/G,-1661A/G, CT60A/G and no statistical differencies observed. But -318C/T polymorphism, it was more than 3 times frequent in rejection group as compared with non-rejection group ($p<0,011$, OR 4,3; 95%CI) and also it was 2 times frequent than the healthy control grup ($p<0,04$, OR 2,93; 95%CI). There was significant difference between G1 and other groups.

Table 1. CTLA-4 genotype frequency distribution

	Acute rejection	Non-Rejection	Control group	p value	Odds Ratio
–318CC	35 (%71)	43 (%91)	44 (%88)	NS	
CT	14 (%29)	4 (%99)	6 (%12)	*p<0,011	4.3
TT	0	0	0	NS	
+49A/A	24 (%49)	23 (%50)	24 (%48)	NS	
A/G	20 (%40)	20 (%42)	22 (%44)	NS	
G/G	5 (%11)	4 (%8)	4 (%8)	NS	
CT60A/A	13 (%26)	17 (%36)	17 (%34)	NS	
AG	24 (%49)	19 (%40)	24 (%48)	NS	
GG	12 (%25)	11 (%24)	9 (%18)	NS	
-1661A/A	31 (%63)	35 (%75)	34 (%68)	NS	
AG	18 (%37)	11 (%23)	16 (%32)	NS	
GG	0	1 (%2)	0	NS	

Additionaly at position -318 T allele frequency was significantly higher at the rejection group compared with the non-rejection group (14% vs 5%, p<0,017, OR 3,7; 95%CI).

Conclusions: CTLA-4, single nucleotide polymorphism at -318C/T that is located within the promotor region of the gene and having T allele in this location is associated with increase risk of acute rejection.

Su636 ★ LACK OF INFLUENCE OF CTLA-4/CD28 GENE POLYMORPHISM ON KIDNEY ALLOGRAFT SURVIVAL IN CAUCASIAN RECIPIENTS

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Introduction and Aims: Participation of CTLA-4 and CD28 molecules is fundamental in cellular immune response mounting (possible induction of tolerance). It has been studied in order to explain autoimmune disease development and has been proposed as basis for possible therapies.

The aim of the study was to examine whether CTLA-4 and CD28 genes polymorphism affects outcome of kidney transplantation (KTx). We checked polymorphisms of the CTLA-4 gene (–318 C/T, +49 A/G and microsatellite polymorphism in the 3'-UTR of exon 4 (AT)n) and the CD28 gene polymorphism (IVS3 +17 T/C).

Methods: The investigation was carried out in 314 allograft recipients, with mean age of 41.9±12 years. The median time of follow-up since KTx was 97.5 months. Genotype of SNPs were determined by means of SSP-PCR and (AT)n genotype using PCR and capillary electrophoresis (ABI Prism310).

Results: In general no relationship was found between concrete allele variants and biopsy proven rejection or delayed graft function. Cox regression model of 10-years graft and patient survival did not show statistical differences between CTLA-4/CD28 genotypes (Table 1).

Lack of influence of CTLA-4 nor CD28 polymorphism on graft survival was also noticed in Kaplan-Maier analysis (Figure 1).

Table 1. Regression Cox model of 10-years graft and patient survival

Polymorphic site	Graft survival		Patient survival	
	HR	p	HR	p
CTLA4 3′UTR	1,139	0,432	0,192	0,660
CTLA4 +49	1,00	0,252	0,054	0,816
CTLA4 –318	0,998	0,604	0,141	0,706
CD28 IVS3 +17	1,275	0,208	0,075	0,783

Conclusions: In this first long-term observational study CTLA-4 and CD28 genes polymorphism did not affect long-term outcome of kidney transplantation in Caucasians.

Su637 A SELECTIVE CYCLOOXYGENASE-2 INHIBITOR, ETODOLAC, PROLONGS MICE CARDIAC ALLOGRAFT SURVIVAL

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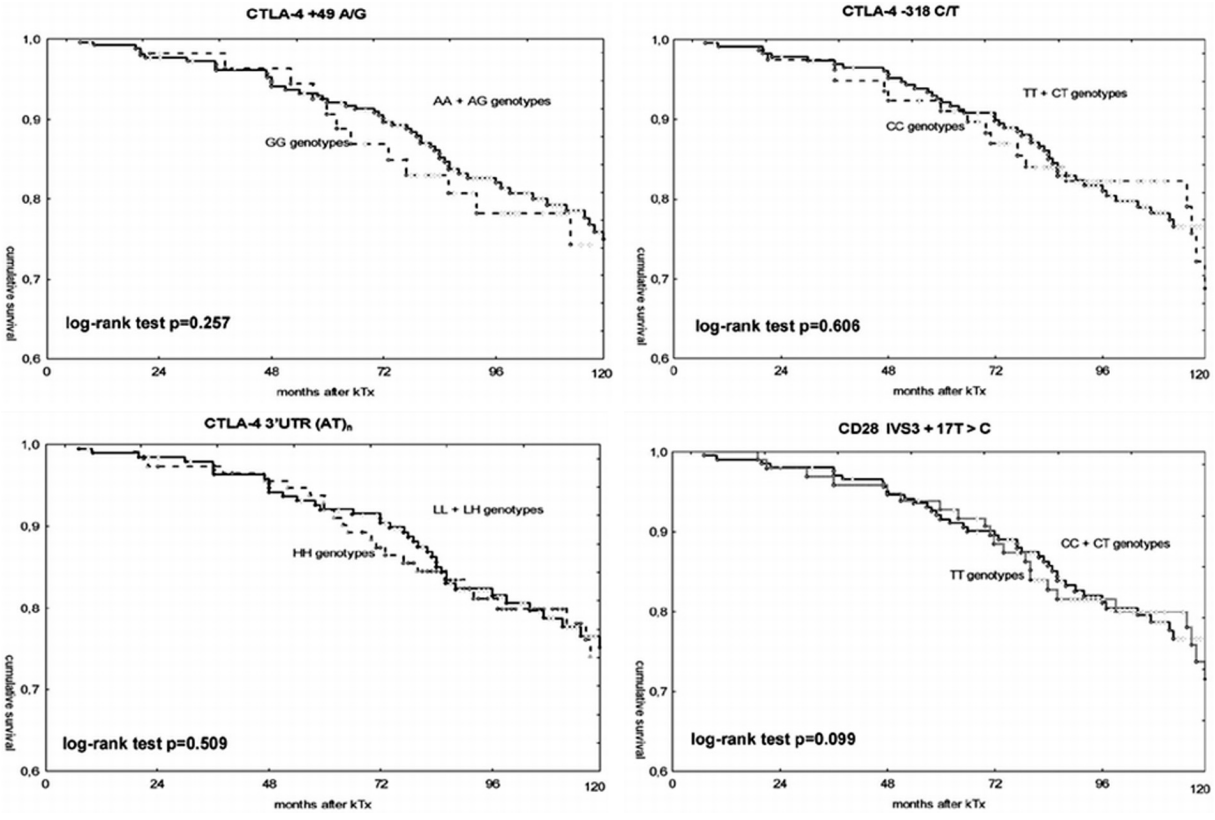
Introduction and Aims: Myocardial inflammation constitutes a major component of the pathologic changes observed during cardiac allograft rejection. Prostaglandins, along with leukotrienes and lipoxins, are lipid mediators which contribute to the vasodialation, edema, and plasma protein leakage which occur during the inflammatory response. The metabolism of arachidonic acid by either the cyclooxygenase (COX) pathway generates eicosanoids which have been implicated in the pathogenesis of a variety of human diseases, including heart disease. COX is the first enzyme in the pathway for producing PGs and thromboxane from arachidonic acid, and there are two isoforms, COX-1 and COX-2. COX-1 occurs in tissues



Abstract Su636 – Figure 1

and cells and works to protect the cell. COX-2 express momentarily and strongly on the growth factor, promotor and some endotoxins as well as being involved with inflammation, cell increase and differentiation. COX-1 is constitutively expressed in most tissues, whereas COX-2 is induced in response to pro-inflammatory cytokines and stress. The prostaglandins in particular are involved in the pathogenesis of inflammation involving cell-mediated immune responses such as those that occur in rheumatoid arthritis and allograft rejection. In this reason, the present study was designed to investigate in mice cardiac allograft model whether COX-2 is expressed and a selective COX-2 inhibitor (etodolac) contributes to myocardial inflammation during cardiac allograft rejection.
Methods: Balb/c mice (H-2d) were used as recipients, and C57BL/6 (H-2b) mice were used as donor of heart. Transplanted heart function was evaluated daily after transplantation by regular abdominal palpation of the transplanted heart beating and by laparotomy when the beating becomes weak. Rejection was defined as total cessation of cardiac muscle contraction. Recipient mice were injected subcutaneously with etodolac (10mg/kg/day). COX-2 expression was analyzed by immunohistochemistry.
Results: Cardiac isograft is definitely tolerated (>150 days), while non-treated cardiac allograft was rejected rapidly (mean 10.9±2.4). However, etodolac-treated cardiac allograft, survival was extended to 18.53±2.1 days. The necrotic area and the grade of COX-2 immunostaining were more significantly reduced in the etodolac-treated cardiac allograft than in the non-treated cardiac allograft.
Conclusions: The administration of etodolac prolongs cardiac allograft survival and reduces myocardial damage and inflammation during acute cardiac allograft rejection. These results suggest that with future study etodolac might emerge as another agent in the therapeutic armamentarium for treatment of patients undergoing heart transplantation.

Su638 THE USE OF OUR SIMPLIFIED EXTRACORPOREAL IMMUNOADSORPTION SYSTEM FOR ABO-INCOMPATIBLE KIDNEY TRANSPLANTS

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Introduction and Aims: Positive cross-match and ABO-incompatibility are the two main barriers to organ transplants because of antibody-mediated rejection (AMR) of allograft. Due to existence of selective plasma exchange (PE) and extracorporeal immunoadsorption (ECIA), kidney transplants across these immunological barriers are possible. PE plus ECIA assures high-quality blood purification with reduced risk of normal plasma constituent loss, alloimunization and/or virus transmission. The aim of this study was to investigate the efficiency of our pre-conditioning regimen in kidney transplant setting, with particular focus on the evaluation of selective PE and ECIA system efficacy based on the anti-A/B depletion, as well as overall clinical outcome for 20 major or bidirectional (major/minor) ABO-incompatible kidney transplants.
Methods: Pre-conditioning regimen used in this study was: anti-CD20 antibody, PE with ECIA and triple immunosuppressive treatment (tacrolimus/mycophenolate-mofetil/steroid). Anti-A/B reduction was obtained by PE (using Cobe-Spectra) combined with our original sterile-connected "close-circuit" antigen-specific ECIA-system. Anti-A/B removal, relevant biochemistry, hemostatic parameters and renal functions (serum creatinine – sCr) in ABO-incompatible vs. ABO-compatible (control group) kidney recipients were compared. Patients included in this study were treated in accordance with good clinical practice and guidelines for kidney transplant practice and were eligible for the use of pre-transplant protocol as certified by the Ethical Committee of MMA.
Results: The use of PE and ECIA resulted in significant ($p<0.01$) anti-A/B decrease: the degree of *in vivo* antibody depletion was 94.23±4.2% (for IgG) and 95.26±3.2% (for IgM). The mean anti-A/B titers on day 0 were: IgG = 1.27±1.03; IgM = 2.20±1.47. One HLA cross-match positive patient (beside ABO-incompatibility) subjected to double-dose anti-CD20 antibody and intensive PE-treatment had no allograft rejection. All patients underwent to renal transplants across ABO-barrier had a good graft function and normalization of sCr level from the second to five post-transplant day. The levels of sCr were 100-156 umol/L (during 12 to 45 months follow-up). One recipient (with sepsis and multi organ distress syndrome) lost kidney function in early posttransplant period. There were no confirmed event of AMR of the graft. Finally, no long-term adverse effects (e.g. no elevated risk of virus transmission), were observed during postoperative period.
Conclusions: The use of PE and our own simplified ECIA-system with anti-CD20 antibody resulted in effective, rapid and reproducible ABO-antibody depletion and beneficial short-term clinical results, as well as in radically reduced treatment-cost. However, the verification of long-term immunomodulation effects still needs to be confirmed in a larger randomized study.

Su639 ★ PRE-TRANSPLANT INTRA-GRAFT SILENCING OF CD40 SWITCHES THE REJECTION PATTERN FROM HUMORAL TO CELLULAR AND INDUCES ACCOMMODATION OF THE GRAFT

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Introduction and Aims: The humoral branch of the immune response plays an important role in acute and chronic allograft dysfunction. The CD40/CD40L costimulatory pathway is crucial in B- and T- alloresponse. Our group has developed a new siRNA molecule against CD40 that effectively inhibits its expression.
The aim of the present study was to prevent rejection and prolong survival in an acute vascular rejection model by intra-graft gene silencing with anti-CD40 siRNA (siCD40), associated or not with sub-therapeutic rapamycin. Four groups were designed: unspecific siRNA as control; sub-therapeutic rapamycin; siCD40; and combination therapy.
Survival time nearly doubled after pre-trasplant intra-graft treatment with siCD40 alone, and increased significantly when siCD40 was associated to rapamycin. The CD40 mRNA was over-expressed in control grafts but treatment with siCD40 decreased its expression, particularly when associated with rapamycin. Recipient spleen CD40+ B-lymphocytes were reduced in both siCD40-treated groups. Moreover, CD40 silencing reduced only partially donor-specific antibodies. Furthermore, CD40 knock-down reduced complement deposition and immune-inflammatory mediators, switching the rejection pattern from humoral to cellular.
Therefore, local gene silencing of CD40 is effective in the blockade of the CD40/CD40L signal, reduces the incidence of humoral vascular acute rejection, changes the type of rejection, and promotes the accommodation of some grafts.

Su640 EICOSAPENTAENOIC ACID PROTECTS AGAINST CYCLOSPORIN A SIDE EFFECTS IN OSTEOBLASTIC CELLS IN VITRO

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Introduction and Aims: Immunosuppressive agents made possible the prevention of allogenic transplant rejections as well as the therapy of autoimmune diseases. Unfortunately, several side effects are reported in subjects undergoing long term treatment with these drugs and the problem has gained importance as patients' survival has increased dramatically. Among side effects, severe clinical and experimental osteopenia has been described. Since beneficial effects of n-3 fatty acids at different organs' level were demonstrated in vivo and in vitro, in the present study we investigated the effect of Eicosapentaenoic acid (EPA) on bone metabolism during immunosuppressive treatment with Cyclosporin A (CsA) in vitro.
Methods: To this aim, pro-inflammatory cytokines expression was measured in the osteoblastlike cell line MG63 by comparative semiquantitative reverse transcriptase chain reaction (RT-PCR). Cells were treated for 48 hours with 2.5 and 5.0 μg/ml CsA, and simultaneously with 10 μM EPA. At the end of treatments, total RNA was extracted and interleukin 1 β (IL-1β), interleukin 6 (IL-6), iNOS, and COX-2 gene expression were evaluated.

Results: Analysis demonstrated that CsA induced marked up-regulation of IL-1β, IL-6, and COX2 gene expression. This effect was more evident in the case of IL-1β that was increased up to 55% by 5 μM CsA while IL-6 up-regulation reached almost 40%. COX-2 gene was slightly augmented and iNOS was not affected by CsA treatment. When the cells were incubated simultaneously with CsA and EPA the co-treatment determined a significant inhibitory effect restoring basal expression levels.
Conclusions: Our results demonstrate that EPA protects against the CsA-induced expression of cytokines known to regulate osteoclast activation and bone resorption, suggesting a beneficial effect of n-3 fatty acids on bone metabolism in the course of immunosuppressive treatment. The clinical relevance of our observation, however within the limits of an experimental model, could open new perspectives for CsA-treated patients.

Su641 FoxP3$^+$DR$^+$-Tregs ARE INVOLVED IN BOTH THE INDUCTION OF UNCONTROLLABLE PRETERM LABOR DURING PREGNANCY AND ORGAN REJECTION AFTER TRANSPLANTATION

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Introduction and Aims: Regulatory T cells (Tregs) were shown to suppress allo-immune responses after organ transplantation and are known to be of substantial importance for the induction of maternal tolerance towards the fetal antigens during pregnancy.
Methods: We used FACS-analysis to detect CD4$^+$CD127$^{low+/-}$CD25$^+$-Tregs and estimated the percentage of a distinct subset of FoxP3$^+$DR$^+$-Treg cells in non-pregnant, non-transplanted volunteers, in both healthy and preterm laboring pregnant women and in kidney-transplanted patients with stable transplant function or acute rejection. In addition, CD4$^+$CD127$^{low+/-}$CD25$^+$-Tregs were isolated by Magnetic-Associated-Cell-Sorting and their suppressive activity was tested by co-culture suppression assays.
Results: In comparison to non-pregnant, non-transplanted volunteers, the total number of CD4$^+$CD127$^{low+/-}$CD25$^+$-Tregs was significantly reduced, both during normal course of pregnancy ($p<0.05$) and after successful kidney transplantation ($p<0.001$). Their percentage of FoxP3$^+$DR$^+$-Tregs decreased during normal course of pregnancy. However, it was not diminished, neither in women with uncontrollable preterm labor nor in transplanted patients showing acute rejection. The mean fluorescence intensity of the HLA-DR expression of the FoxP3$^+$DR$^+$-Treg cell subset was significantly reduced, both in preterm-laboring women ($p<0.001$) and in kidney-transplanted patients with acute rejection ($p<0.001$). The suppressive activity of the magnetically selected CD4$^+$CD127$^{low+/-}$CD25$^+$-Tregs was significantly decreased in preterm-laboring women ($p<0.01$) and in transplanted patients with acute rejection ($p<0.05$).
Conclusions: Our results suggest that the subset of FoxP3$^+$DR$^+$-Tregs may have a potential effect on the suppressive activity of the total CD4$^+$CD127$^{low+/-}$CD25$^+$-Treg cell pool and that the immunologic mechanisms leading to uncontrollable preterm labor may be similar to those leading to acute rejection after organ transplantation.

Su642 EVOLUTION OF ALLOGRAFT FIBROSIS AND RELATED MARKERS IN KIDNEY TRANSPLANT PATIENTS UNDER TREATMENT WITH CYCLOSPORINE AND EVEROLIMUS

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Introduction and Aims: In kidney transplant, protocols to avoid CNI chronic toxicity are continuously developed in order to prevent allograft fibrosis and function loss. We analyzed the histological evolution of allograft fibrosis and the expression of related markers after conversion from cyclosporine (CsA) to everolimus performed in stable kidney transplant patients.
Methods: Seventeen patients maintained on prednisone (pred), sodium mycophenolate and cyclosporine (CsA) were considered eligible for the study. After 4.5 months of transplantation, patients were randomised to either continue CsA (n = 10) or convert from CsA to everolimus (n = 7). Kidney biopsies obtained prior and at least 6 months after randomization were analyzed for kidney fibrosis (Masson's trichrome) and collagen content (Sirius Red); immunohistochemistry was performed for TGFβ, Hif-1alpha and PDGF. Analysis was performed using image software or semiquantitative scoring system (0-12), according to the antibody stained. Evolution of the expression of the different markers was evaluated on each patient corrected by the time between biopsies; results are given as %variation/year.
Results: Baseline characteristics and time between biopsies were similar between groups. A tendency for a higher increase in fibrotic area was seen in the CSA (22.3±22.2%variation/year) compared to everolimus (1.7±25.2%variation/year, p=0.1), which was confirmed by Sirius red. TGFβ tubular expression significantly decreased in the everolimus group in compared to baseline (-50.0±33.1 variation/year, p=0.007); this was also significantly different compared to CSA (-2.94±24.4%variation/year, p=0.007). PDGF and Hif-1 alpha expressions were variable and not significantly different between the two groups, remaining stable in the observations.
Conclusions: Conversion from CSA to everolimus was associated to a significant reduction in tubular TGFβ expression. This may exert a positive impact in preventing the development of allograft fibrosis.
Disclosure: This work contains clinical information from the ZEUS study, sponsored by Novartis.

Su643 T REGULATORY ACTION ON MLR ARE ENHANCED BY CD14 POSITIVE CELLS IN KIDNEY TRANSPLANT PATIENTS

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Introduction and Aims: Following organ transplantation the deployment of immune response encompasses both up-and down-regulation reactions. Regulatory T cells play a non-dispensable role in down-modulation but it seems that they must cooperate with antigen presenting cells. We reported that fine-needle aspiration biopsy (FNAB) sample cultures from stable kidney transplants (KTx) synthesize a soluble factor endowed with both specific and unspecific down-regulating actions on donor-recipient MLR. We tested the importance of monocyte cells lineage on this effect.
Methods: We performed three FNAB on day seven post-KTx from three patients who would not develop acute rejection, treated with classical triple therapy. FNAB samples were incubated for 24h in RPMI and 10 U/ml of rIL-2. At the end cells were recovered and half of the sample was submitted to monocyte depletion by CD14 microbeads from Miltenyibiotec with autoMACS Pro Separator. This treated sample, 100uL was added to donor-recipient MLR; the other half of FNAB culture not submitted to CD14 subtraction was also added to wells with donor-recipient MLR. Another group of wells received classical non-supplemented donor recipient MLR. On the sixth day of culture thymidine was added.
Results: Cyclosporine blood levels were within the therapeutic window and creatinine was 1.4, 4.0 and 6.0 mg/dl. Results for non-supplemented MLR were 8145, 19483 and 40745 cpm. By adding whole FNAB sample cultures and by comparing with no FNAB sample supplementation, proliferation declined to 5.4%, 6.6% and 9.7%, respectively while adding CD14 depleted samples proliferation rose to 136% and declined to 10.9% and 47.6%, respectively.
Conclusions: Our results suggest that donor-recipient MLR down-modulation by FNAB sample cultures from stable KTx is enhanced by the presence of CD14 cells and/or their products. This may require either cell contact or be mediated by soluble factors. We speculate that IL-10 may be involved on the effect we observed.

Su644 REGULATORY T CELLS AND IL-10, IL-17 AND IFN-γ PRODUCING T CELLS FREQUENCIES IN KIDNEY ALLOGRAFT RECIPIENTS WITH DONOR BONE MARROW CELLS INFUSION

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Introduction and Aims: Type and dynamics of cytokines in the allograft are considered as the crucial factors in altering the tissue function and in reducing T cell responses leading to anergy, apoptosis or induction of regulatory T cells. The aim of this study was to assess the influence of donor bone marrow cells infusion (DBMI) on the frequency and function of regulatory T cells (T reg) and on the cytokine profiles in kidney allograft recipients.
Methods: 27 living unrelated renal allograft patients were included in this study as DBMI (n=14) and control (n=13) groups and followed up 2 years retrospectively. Peripheral blood mononuclear cells (PBMCs) from all patients were obtained at the end of second year post operatively and the percentages of CD25+foxP3+ and CD3+CD8+CD28- Tregs were measured using flowcytometery. Also, the frequency of IL-10, IL-17 and IFN-γ producing cells separately were determined using ELISPOT analysis using peptides corresponding to HLA-DR mismatched alleles between donor and recipients and phytohemaglutinine (PHA) as stimulators.
Results: A significant decrease in the number of IFN- γ producing cells were found in DBMI patients compared to controls (P=0.035). Also, an increase in the frequency of IL-10 producing cells (P=0.07) and a decrease in the rate of IL-17 producing cells (P=0.18) were observed. The mean number of IFN- γ/IL-10 producing cells was significantly higher in DBMI patients versus controls (P=0.02). The mean difference for the frequencies of CD4+CD25+FoxP3+Tregs and CD3+CD8+CD28- T cells between both groups were 0.5% and 4.5% respectively and higher percentages for those Tregs were shown in DBMI patients (P=0.12 and P=0.36).
Conclusions: These findings suggest that the donor bone marrow cells infusion could stimulate partially the regulatory mechanisms based on the presence of lower number of inflammatory cytokines producing cells concurrently with the higher percentage of regulatory cells in peripheral blood late after transplantation.

Su645 RESISTIVE INDEX IN RENAL TRANSPLANT RECIPIENTS: ORGAN DAMAGE OR SUBCLINICAL ATHEROSCLEROSIS?

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Introduction and Aims: Chronic kidney disease and related complications show high prevalence in renal transplant recipients. Carotid intima media thickness (IMT) appeared as an independent predictor of cardiovascular events in chronic renal failure.
Renal resistive index (RI) $\geq$ 0.80 predicts graft survival and cardiovascular mortality in renal transplant recipients. It is unclear if in these patients RI is a marker of renal or systemic vascular damage. In our hypothesis, in renal transplant recipients, an increase in RI could reflect systemic atherosclerotic damage.
Methods: 19 renal transplant recipients (mean age 50.9±12.1 years, 8.6±5.4 years since transplantation) were studied. All patients were on a calcineurin inhibitor (10 patients on cyclosporine, 5 on tacrolimus), 12 were on mychophenolate mofetil and 14 on prednisone. GFR was estimated using the abbreviated Modification of Diet in Renal Disease (MDRD) equation. IMT was measured immediately below and above carotid bifurcation on left and right arteries, then mean value was computed. RI was obtained on three proximal segmental arteries on both kidneys, and mean value was calculated.
Results: At linear regression analysis a direct correlation was observed between IMT and RI [(r= 0.32); (p<0.005)]. Both parameters directly correlated even with mean age, body mass index, low density lipoproteins and time since transplantation.
On the other hand IMT and RI inversely correlated with eGFR and high density lipoproteins.
Conclusions: Our study confirms a correlation between RI and graft function. Interesting correlation appeared between RI and atherosclerosis. Higher RI correlated with intima media thickening, resulting linked in a linear way with many risk factors, both traditional and characteristic of chronic renal failure.
So RI evaluation, in renal transplant recipients, needs care in the hypothesis that RI could depend from systemic vascular damage further that local one.

Su646 CYCLOSPORIN A-INDUCED INCREASE OF COLD SHOCK Y-BOX PROTEIN-1 IS CELL-SPECIFIC, ROS-DEPENDENT AND INVOLVES AKT/ERK-KINASE PATHWAYS

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Introduction and Aims: Cyclosporine A (CsA) effectively prevents renal transplant rejection but may result in tubulointerstitial as well as mesangial fibrosis. Fibrosis- and inflammation-related proteins like cytokine IL-2, matrix metalloproteinase 2 (MMP-2) and type I collagen (Col1A) are transcriptionally or translationally regulated by Y-box binding protein-1 (YB-1). Given the marked and very rapid increase of intracellular Y-box (YB) protein-1 after challenge of rat mesangial cells with CsA, we aimed to further study the precise mechanism and cell specificity of enhanced YB-1 protein expression.
Methods: Calcineurin inhibitor challenge was investigated by incubating different renal cells either with vehicle or with CsA (1 μM). Proteins were detected by immunoblotting using GAPDH as loading control. Involvement of reactive oxygen species (ROS) and kinases was investigated by specific inhibitors and via direct cell challenge with H2O2.
Results: A marked CsA-dependent upregulation of YB-1 was detected in rat mesangial cells. In contrast, the intracellular YB-1 protein content in human tubular cells (HK-2) was decreased following CsA incubation, whereas it remained unchanged in human embryonic kidney cells (HEK293T).
The very rapid enhancement of YB-1 protein content following CsA stimulation in rat mesangial cells might be explained by an altered degradation of YB-1 protein by the 20S proteasome since the cell-permeable proteasome inhibitor MG132 (10 μM) resulted in the induction of YB-1 protein content comparable to the one achieved by CsA administration. Furthermore, in contrast to control cells, YB-1 protein stability was enhanced following CsA-challenge. Thus, without prior CsA-challenge, cellular YB-1 protein is degradated over 8 h, that is delayed by CsA incubation. The CsA-dependent increase of YB-1 protein content was almost completely abolished following preincubation with the antioxidant N-acetylcysteine (NAC; 5 mM). In accordance with this, a concentration-dependent increase of YB-1 protein was observed after 10 min of H2O2 exposure (100 μM). To unravel the signalling pathway, we determined the impact of MAP/ERK- and PKB/Akt kinases in this event. Upon CNI-exposure upstream signalling affecting YB-1 expression involved both, ERK and Akt pathways.
Conclusions: Taken together, our results indicate a role of YB-1 in CNI nephrotoxicity, mediated by hydrogen peroxide, enhanced protein stability and posttranslational modifications.

Su647 HUMAN PERIPHERAL BLOOD CD8+CD28- T CELLS OF RENAL ALLOGRAFT RECIPIENTS DO NOT EXPRESS FOXP3 PROTEIN

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Introduction and Aims: Foxp3 molecule has been suggested as a marker of a suppressor subset of human CD8+CD28- T cells (Ts) in recent studies.

Some researchers suggest that higher level is associated with stable graft function and can allow a reduction of immunosuppressive drug doses, while others observe a correlation between the increase in CD8+CD28- cells and chronic graft rejection. The phenotype of the Ts subpopulation has not been identified explicitly. Some researchers suggest that a transcriptional factor, Foxp3, characteristic of CD4+CD25^{high} T regulatory cells (Treg), is also a phenotypic marker of the Ts subset of CD8+CD28- cells. They observed correlation between the level of Foxp3 mRNA and allograft function. The present authors suggest that the search for correlations between the Foxp3 marker and allograft function should be focused on Foxp3 protein.
The aim of this study was to evaluate whether Foxp3 protein is present in the CD8+CD28- population from the peripheral blood of renal allograft recipients and whether the level of CD8+CD28-Foxp3+ cells correlates with allograft function.
Methods: The study was performed on 30 renal allograft recipients with uneventful stable course (S) and biopsy-proven chronic rejection (CR). There were 21 patients with immunosuppression based on cyclosporine A and 9 patients with immunosuppression based on rapamycin. Peripheral blood mononuclear cells (PBMCs) were isolated from the recipients' blood samples by density gradient centrifugation using Ficoll-Isopaque. T-lymphocyte subsets were defined using the mAbs anti-human CD8, CD28, (BD Biosciences), and anti-human FOXP3 Kit (eBioscence). Flow cytometry was performed using a FACSCalibur (BD Biosciences) instrument and data were analyzed with Cell Quest software.
Results: The mean percentage of CD8+CD28- cells in the lymphocyte population was higher in the peripheral blood of the CR group than in the S group. This difference was also significant in the subgroup of patients who received immunosuppressive therapy based of CsA. There was no correlation between the percentage of CD8+CD28- cells in lymphocytes and the age of patients or the time after transplantation. Foxp3 protein expression was not observed not only in the CD8+CD28- population, but also in the whole populations of CD8+ or CD28- cells of both patient groups.
Conclusions: The expression of Foxp3 protein in CD8+CD28- cells seems to be useless as a diagnostic tool of allograft function and it probably is not a marker of the CD8+CD28- Ts subset.

Su648 INFLUENCE OF M-TOR INHIBITORS IN PATIENTS WITH PRETRANSPLANT PROTEINURIA

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Introduction and Aims: M-TOR inhibitors were accused by renal injury and worse graft outcome in some studies. However, they also were used for steroid resistant nephrotic syndrome patients. In this study, we have investigated the effect of m-TOR inhibitors on patient with proteinuria.
Methods: Between 2005-2008, 259 (F/M: 178/81) patients who were treated with m-TOR as maintenance therapy were included in this study. Treatment protocol was as follows:
–1 day: CNI+MFA
0–5 day: CNI+MFA+Prednisolone
6– 80 day: CNI+m-TORinh+ Prednisolone, (MFA stop)
81–90. day: CNI+m-TORinh+ Prednisolone +MFA (restart, ↓25% dose)
≥ 90. day: m-TORinh+MFA+ Prednisolone, (CNI stop)
Induction Therapy; 1. Basiliximab: 0. and 4.day; 20 mg IV or, 2. Daclizumab: 0. day and every 2 weeks: 1 mg/kg (5 dose total) or, 3. ATG: 8 mg/kg (single dose)
Patients were divided into three groups: Group I: Tac + SRL (F/M: 22/35); Group II: CsA + SRL (F/M: 27/72); Group III: CsA + EVE (F/M: 32/71). Three years follow-up results were analyzed.
Results: The demographics of the patients were similar. There were no differences between acute rejection rates (15, 8%- 21, 2%- 19, 6%, NS), patient survival (100%- 98%-96, 1%, NS), graft survival (98, 2%- 98%- 93, 2%, NS), respectively. As shown in Figure 1, there were also no differences in terms of proteinuria. Proteinuria was decreased during in-house period and was under critical level (800 mg/d) at the end of 7 (mean discharge time from hospital) days.

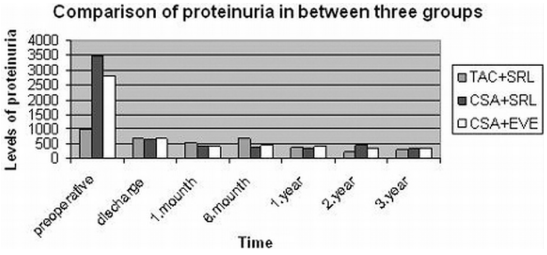

Figure 1. Comparison of proteinuria in between groups

Conclusions: Presence of pretransplant proteinuria is not a restrictive factor for choosing m-TOR for maintenance therapy for renal recipients.

Su649 THYMOGLOBULIN DOWN-REGULATES PENTRAXIN 3 SYNTHESIS THAT CORRELATES WITH ACUTE REJECTION ON KIDNEY TRANSPLANT PATIENTS

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Introduction and Aims: Long pentraxin 3 (PTX3), an acute-phase protein is an important mediator in innate immunity and inflammation. PTX3 role in kidney transplantation (KTx) awaits clarification as both up- and down-regulation of immune response may be envisaged. We present data on PTX3 synthesis by fine-needle aspiration biopsies (FNAB) cultures from KTx.
Methods: Sixty-eight KTX were divided into stable (n=55) and acute rejection cases, II (n=13). Stable cases were treated with classical triple therapy (Ia,n=21), thymoglobulin induction (Ib,n=13) and anti-IL-2 receptor antibody (Ic,n=21). Group II cases received classical triple therapy and thymoglobulin induction in 9 and 4 cases, respectively. FNAB were done on day 7 post-KTx in I and on rejection day in II. Four cases from I were also tested on day 30. FNAB samples were cultured for 48h and supernatants collected and kept frozen until analysis by ELISA from R&D. Values expressed in pg/ml, statistics by Mann-Whitney.
Results: No significant differences were observed by comparing demographic data and calcineurin inhibitors (CNI) levels between groups with the exception of a significant lower CNI values in Ic. Creatinine was equal between groups but lower on day 30.
PTX3- Ia: 1.52±0.8; Ib: 0.88±0.5; Ic: 1.6±1.22; II: 3.6±2.2. On day 30, PTX3 was 0.33±0.08. PTX3 value was significantly higher in II versus I ($p=0.0055$), in II versus Ia ($p=0.013$), in II versus Ib ($p<0.0001$), in II versus Ic ($p=0.001$). PTX3 was significantly lower on day 30 than on day 7 ($p=0.03$). Thymoglobulin induction associated with a lower PTX3 ($p=0.07$).
Conclusions: Our data suggest that PTX3 correlates with acute rejection in KTx and that thymoglobulin is endowed with the capacity to down-regulate PTX3 synthesis. Those findings corroborate reports on PTX3 role in graft-versus host disease and contradict a hypothetical positive role of immune down-regulation by sequestering cell remnants, present inside the grafts on the first days post-KTx, from antigen-presenting cells.

Su650 DIFFERENT ROLES FOR THE UP-REGULATION OF IL-22 SYNTHESIS DEPENDING ON TIMING POST-GRAFTING IN KIDNEY TRANSPLANT PATIENTS

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Introduction and Aims: IL-22 is an effector cytokine produced by Th17 cells and together with IL-17 may have essential functions in host defence and in autoimmune diseases. IL-22 role in kidney transplants (KTx) has not been defined. Previously we reported that during acute rejection IL-17 rises non-significantly but by day 30 post-KTx, unexpectedly, IL-17 rises significantly in stable cases. We present data on IL-22 synthesis by fine-needle aspiration biopsies (FNAB) cultures from KTx.
Methods: Sixty-eight KTx were divided into stable, I (n=55) and acute rejection cases, II (n=13). Stable cases were treated with classical triple therapy (Ia, n=21), thymoglobulin induction (Ib,n=13) and anti-IL-2 receptor

antibody (Ic,n=21). Group II cases received classical triple therapy and thymoglobulin induction in 9 and 4 cases, respectively. FNAB were done on day 7 post-KTx in I and on rejection day in II. Seven cases from Ia were also tested on day 30. FNAB samples were cultured for 48h and supernatants collected and kept frozen until analysis by ELISA from R&D. Values expressed in pg/ml, statistics by Mann-Whitney.
Results: No significant differences were observed by comparing demographic data and calcineurin inhibitors (CNI) levels between groups with the exception of a significant lower CNI values in Ic. Creatinine was equal between groups but lower on day 30.
IL-22- Ia: 19.1±1.20.8; Ib: 19.8±1.24; Ic: 21.3±2.0; II: 34.9±36.4. On day 30, IL-22 was 73.9±63.3. IL-22 was significantly higher in II versus I ($p=0.0023$), in II versus Ia ($p<0.001$), in II versus Ib ($p<0.001$), in II versus Ic ($p=0.01$). IL-22 was significantly higher on day 30 than on day 7 ($p=0.002$).
Conclusions: Our data suggest that IL-22 correlates with acute rejection in KTx as we had found with IL-17. Of interest, we observed a highly significant up-regulation of IL-22 synthesis on day 30 post-KTx in stable cases perfectly mirroring our previous data with IL-17. We surmise that these different roles may be akin to IL-10 behaviour in KTx.

Su651 MECHANISMS OF ACUTE TACROLIMS-INDUCED NEPHROTOXICITY AND THE RENOPROTECTIVE EFFECTS OF TELMISARTAN

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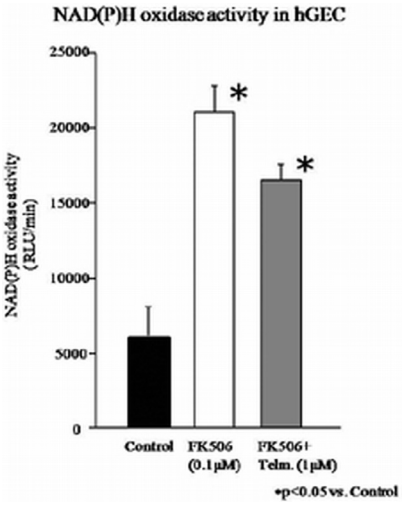
Introduction and Aims: The immunosuppressive drug tacrolimus (FK506) is used clinically to reduce rejection rate in the patients with kidney transplantation, although the nephrotoxicity induced by FK506 remains a serious problem. FK506 had been demonstrated to cause hyalinosis of afferent arterioles, constriction of arterioles, and progressive tubulointerstitial fibrosis in the chronic phase. However, the pathogenetic mechanisms of the FK506-induced nephrotoxicity especially at the early phase are not fully elucidated. We attempted to elucidate the mechanisms of acute nephrotoxicity induced by FK506 and the renoprotective effects of antiotensin II receptor blocker (ARB), telmisartan.
Methods: Male seven-week old Wistar rats were divided into three groups; vehicle group, FK506 (0.6 mg/kg/day, subcutaneously) group, FK506 and Telmisartan (1 mg/kg/day, orally) group (each n=8). Eight weeks later, we assessed kidney function and renal morphological changes including oxidative tissue injuries. Endothelial function was also evaluated in the aortic rings. Glomerular production of reactive oxygen species (ROS) was evaluated by chemiluminescence of dihydroethidium. The direct effects of FK506 on cultured human glomerular endothelial cells (hGEC) were also examined.



Co-incubation of Telmisartan inhibited NADPH oxidase activity and production of ROS.

Results: The blood pressure, heart rate and kidney weight did not differ between the groups. Blood urea nitrogen was significantly increased in FK506 group, but was attenuated in the FK506 + Telmisartan group. Increased production of ROS was recognized in the glomeruli of FK506 group. Telmisartan treatment significantly suppressed glomerular ROS production by FK506. The mRNA expressions of NADPH oxidase subunits, p47phox and p67phox were increased in the glomeruli from FK506 group, but were decreased in FK506 + Telmisartan group. FK506 directly activated NADPH oxidase activity and accelerated production of ROS in hGEC (DCFH-DA fluorescence staining, Lucigenine assay).
Conclusions: These data suggest that acute nephrotoxicity by FK506 is caused by oxidative stress mediated by NADPH oxidase activation in the endothelial cell, and that telmisartan exerts renoprotective effects through its antioxidative activity.

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Su652 CLINICAL EXPERIENCE WITH EVEROLIMUS IN 122 RECIPIENTS OF A KIDNEY OR STEM CELL TRANSPLANT

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Introduction and Aims: The mTOR inhibitor everolimus is used for immunosuppression after kidney transplantation and stem cell transplantation. Everolimus is more hydrophilic, the half-life shorter (31 vs 60 h), and the adverse events might be less frequent as compared to sirolimus [Transplant Proc. 2006;38:711-3. Transpl Int. 2009;22:586-7]. We report on our experience with everolimus and compared the therapeutic effect and adverse events between kidney and stem cell transplant patients.
Methods: Therapeutic drug monitoring was performed in 162 patients receiving everolimus in our hospital in the years 2007 to 2009. The clinical course retrospectively could be assessed in 122 patients, 76 males and 46 females. Everolimus was given to 81 patients after kidney transplantation and to 41 patients after stem cell transplantation. Everolimus co-medication was mycophenolate in 63 and glucocorticoids in 48 patients. Everolimus was taken for 1 to 51 months with a median of 7 in kidney and 5 months in stem cell patients.
Results: At start of everolimus, the kidney transplant patients were significantly older ($p<0.01$) than stem cell transplant patients (56±12 vs 47±11 y). The reason for everolimus was different in kidney transplant patients as compared to stem cell transplant patients, with calcineurin inhibitor toxicity (34 vs 5), graft versus host disease (0 vs 23), secondary malignancies mostly scin (19 vs 1) and sirolimus intolerance (15 vs 0). With a median dose of 1.5 mg/d the mean everolimus trough level was 4.9±1.9 ng/ml after kidney transplantation and 6.4±2.1 ng/ml after stem cell transplantation ($p=0.01$). The mean creatinine significantly improved ($p<0.01$) from 284±161 to 201±90 mcmol/l but deteriorated again to 266±135 mcmol/l in the kidney patients, but was throughout significantly lower with 111±83 mcmol/l in the stem cell patients. Except death (6 vs 12) and low platelets (4 vs 15), overall adverse events were significantly more frequent in kidney than in stem cell patients such as a urinary protein/creatinine ratio >1.0 g/g (16 vs 2), pneumonitis (4 vs 0), dermatitis (10 vs 0), and infectious complications (14 vs 2). A proteinuria/creatinine ratio of 4.6 and 5.7 g/g, respectively had 2 kidney patients with the nephrotic syndrome occurring 1 and 12 months after everolimus. Everolimus therapy was discontinued in 81 cases (66%) among them because of adverse events in 32 cases (40%), mainly dermatitis (n = 15), infections (n = 10), thrombotic thrombocytopenic purpura (n = 7), proteinuria (n = 12) and pneumonitis (n = 4).
Conclusions: Everolimus was a good option in kidney transplant patients with calcineurin inhibitor toxicity or secondary malignancies since serum creatinine improved significantly. However, everolimus had to be discontinued in 32/122 = 26% of all patients because of adverse events. Importantly, proteinuria and pneumonitis were observed only in kidney transplant patients but not after stem cell transplantation.
Disclosure: Our work was supported by Novartis company.

Su653 URINARY TRACT INFECTIONS IN THE RENAL TRANSPLANT POPULATION

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Introduction and Aims: Urinary tract infections (UTIs) are an important cause of morbidity and graft dysfunction in renal transplant recipients. We aimed to determine the frequency, nature and impact of UTIs in our renal transplant population.

Methods: We undertook a retrospective analysis of all urine cultures from our renal transplant patients (sent routinely at all clinic visits or when clinically indicated) from January 1st 2007 to December 31st 2008 and compared our findings to specimens from the general population over the same period.

Results: 52 out of 225 renal transplant patients had at least one episode of signficant bacteriuria (defined as $>10^5$ colony forming units). All patients were at least 3 months post transplant; 27% were less than 1 year post transplant, 63% less than 5 years and 17% greater than 10 years. The mean age was 51 years (SD 15 years). We identified 157 episodes of significant bacteriuria; all bar one were a single isolate: coliforms (76%), Pseudomonas (12%), Enterococci (6.4%), Group B Streptococci (1.9%), coagulase negative Staphylococci (1.9%), Group G Streptococci (0.6%), methicillin-resistant Staphylococcus aureus (MRSA) (0.6%). Non-coliform UTIs were significantly more common in transplant patients than in the general population (24% vs 11%, $p<0.0001$) though the only single organism significantly more frequent was Pseudomonas (12% vs 1.9%, $p<0.0001$) which may be over-represented due to recurrent infection in a small number of patients.

Compared to the general population, coliforms in the renal transplant population were significantly more resistant to amoxicillin (69% vs 50.5%, p=0.008), coamoxiclav (17% vs 7%, p=0.005) and trimethoprim (43% vs 27%, p=0.009). Trimethoprim-resistant coliforms were significantly more common in patients during the first year post transplant than thereafter (50% vs 14.7%, p=0.01).

UTI was the sole reason for hospital admission in 12 transplant patients, mean age 57 years (SD 14.5 years), with a median hospital stay of 5.5 days. UTIs were responsible for deterioration in graft function (rise in serum creatinine > 25%) in 7 patients, bacteraemia in 4 (3 coliforms, 1 MRSA) and septic arthritis in 1. No episodes of graft loss nor acute rejection were observed in these patients.

Conclusions: We demonstrate that non-coliform UTIs were significantly more common in renal transplant patients compared to the general population. There were significantly higher rates of antibiotic resistance in the coliforms isolated from transplant patients. Trimethoprim-resistant coliforms were observed more frequently in transplant patients in the first year post transplant, during which time the majority of patients take cotrimoxazole prophylaxis. UTIs were a common cause of hospital admission, graft dysfunction and morbidity. These findings should guide empirical antibiotic choice in the renal transplant population and prompt early detection and treatment of this common condition.

Su654 SUBCLINICAL INTERSTITIAL LUNG ABNORMALITIES IN STABLE RENAL ALLOGRAFT RECIPIENTS IN THE ERA OF MODERN IMMUNOSUPPRESSION

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Introduction and Aims: Interstitial lung abnormalities have been detected in up to 24% of kidney transplant patients receiving traditional immunosuppressive therapies (*eg,* cyclosporine, azathioprine); they usually occur early after transplantation. Newer immunosuppressants such as mycophenolic acid and, particularly, mTOR inhibitors (*eg*, sirolimus) may cause significant lung toxicity. However, the prevalence and severity of interstitial lung lesions in long-term, stable kidney transplant patients receiving either traditional or newer immunosuppressants is not known.

Methods: We conducted a prospective, cross-sectional study examining high-resolution lung CT scans in 63 stable kidney transplant recipients whose immunosuppressive therapy had remained unchanged for >24 months. CT findings of patients taking newer (mycophenolic acid and mTOR inhibitors) and traditional (calcineurin inhibitors and azathioprine) immunosuppressive drugs were compared.

Results: Interstitial lung alterations were found in only 3/63 patients (4.8%); the prevalence was 11.5% (3/26) and 0% (0/37) in the newer and traditional immunosuppressive therapy groups, respectively (P=0.065). The CT patterns were usual interstitial pneumonia- and nonspecific interstitial pneumonia-like. The median time-lapse between transplant and CT was 49 months in the three patients with CT alterations and 95 months in the remaining 23 patients on newer immunosuppressants. It was 75 months for all patients on newer immunosuppressive drugs and 133 months for those on traditional therapies (P=0.0015). A follow-up CT, performed in two of the three patients with interstitial abnormalities, showed that the lesions were stable in one, while they had disappeared in the other.

Conclusions: Interstitial lung abnormalities are infrequent and mild in stable kidney transplant patients treated with newer as well as traditional immunosuppressive drugs. As such abnormalities were detected in patients screened earlier after transplantation, the time-interval since transplant rather than the drug type is probably a major determinant.

Su655 RENAL FUNCTIONAL RESERVE AND SALT SENSITIVITY

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Introduction and Aims: Some individuals manifest large blood pressure changes in response to acute or chronic salt depletion or repletion, and are termed "salt sensitive" (SS) meanwhile others are called "salt resistant" (SR) because they don't show any changes to salt variations.

The renal functional reserve (RFR) was defined as the ability of the kidney to increase its level of operation under certain demands.

The purpose of our study was to evaluate RFR through the acute changes in creatinine clearance (CrCl) in response to oral protein load in SS and SR healthy people.

Methods: Data from 38 healthy people (29 women), mean age 52±10 years were included in the present study. Twenty three of them were living kidney donors (D) (15 women) mean age of 47±8 years and the other 15 people were post-donation (PD) (11 women) mean age 58±8 years. None had a history of kidney disease, diabetes, cardiovascular events or hypertension.

After two weeks on low and high-salt diet, people received a normosodic and normoproteic diet without meat for two days previous to protein load. On the third day, 24 hs urine creatinine clearance was measured (CrClb) before a supplemented soya milk protein drink containing 1 gr of protein/kg of body weight was given over a 30 minutes period. Following the milk protein drink a 2 hs urine creatinine clearance (CrClp) was performed.

Data with normal distribution were expressed as mean ± standard deviation and Student t test were used, P values < 0.05 were considered to be statistically significant.

Results: We studied 14 SS and 24 SR, SS were older than SR (55±11 vs 49±9 years p=0,07). We did not find significant differences in CrClb (94±30 vs 87±31 ml/min/1,73 m^2 p=0,56), after protein load CrClp were lower in SS than in SR without statistically significant differences (93,3±40 vs 118,5±42 ml/min/1,73 m^2 p=0,07).The percentage of variation of CrCl (ΔCrCl) in SS was significantly lower than in SR (-0,05±32 vs 42±58% p<0,02).

CrClb and CrClp found in PD group were significantly lower than in D group (72±24 vs 101±29 ml/min/1,73 m^2 p<0,004 and 88±41 vs 123±38 ml/min/1,73 m^2 p<0,01) but there were similar ΔCrCl in both groups (29±65 vs 26±46% p=0,79).

When we analyzed only D group, we found 8 SS and 15 SR without significant differences in age (49±9 vs 46±8 years p=0,45). The CrClbs were similar (100±32 vs 102±28 ml/min/1,73 m^2 p=0,80). After protein load, CrClp and ΔCrCl were significantly lower in SS than in SR (103±43 vs 134±31 ml/min/1,73 m^2 p<0,05 and 1±21 vs 39±51% p<0,05 respectively).

The PD group had 6 SS and 9 SR, SS were older than SR (64±7 vs 54±8

years p<0,03).There were not significant differences between SS and SR in CrClb (85±27 vs 63±17 ml/min/1,73 m^2 p=0,08), CrClp (80±33 vs 93±47 ml/min/1,73 m^2 p=0,58), and ΔCrCl(-1,5±45 vs 49±71% p=0,12).
Conclusions: The PD group had lower CrClb and CrClp but the same RFR than D group. We found a significantly lower increase of CrClp and ΔCrCl in SS than SR of D group that we did not find in PD group.

Su656 TEN YEARS FRACTURE RISK ASSESSMENT IN PREVALENT PATIENTS WITH KIDNEY TRANSPLANTATION

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Introduction and Aims: Several studies reported osteoporosis in renal transplant patients and in patients with kidney transplantation resulted an higher incidence of fractures than in general population. The aim of this study was the evaluation of bone loss as well of fracture risk in a population of patients with kidney transplantation of our Center.
Methods: We performed a cross-sectional observational study on 82 prevalent renal transplant patients, 52 M and 30 W, of mean age of 56±11 years with clinically stable renal graft from more than 1 year (transplant age 121±76 months). In all patients we measured lumbar spine (LS), total femur(TF) and femoral neck (FN) bone mineral density (BMD, g/cm^2) by dual energy x-ray absorptiometry (DEXA) and the ten years probability (%) of hip fracture (HF) and major osteoporotic fractures (vertebral, proximal humerus, distal femur, proximal tibia, pelvis, multiple rib: MOF) was measured by WHO fractures risk assessment tool (FRAX), including country and the main fracture risk factors (sex, BMI, previous fracture, parent fractured hip, current smoking, glucocorticoids, rheumatoid arthritis, alcohol 3 more units per day) and femoral neck BMD t score.
Results: LS, TF and FN BMD mean values±SD were 878±126, 823±134 and 688±116 g/cm^2, with t score of –1.76±1.15, -1.24±0.98 and –1.66±0.93. 43/82(52%) patients presented osteopenia (LS and/or TF and/or FN (<–1>-2.5 SD), 26/81(32%) were osteoporotic (LS and/or TF and/or FN <-2.5 SD) and only 13/81 patients (16%) were in the normal range (>-1<1 SD). 5/82 patients (6%, 4 W and 1 M) presented prevalent fractures (3 at the femoral and 2 at the vertebral level). BMI mean values were 24.8(2.5) kg/cm^2. 21/30 (70%) were postmenopausal W. All patients were on immunosuppressive treatment, in 17/82 (21%) of these non including steroid therapy. The 10 years % probability mean values of HF and MOF were 5.3± 5.1 and 12.6±7.3. 49% (41/82) of patients, 53%(23/52) of M and 43% (13/30) of F presented a >3% probability of HF and in the 24% (10/41) of these, 3 M(30%) and 7 F(70%) a > 20% probability of MOF was associated. In overall population 26/82 patients, 14/30 of F (47%) and 12/52 of M (23%) were on calcium and/or vitamin D therapy.
Conclusions: Our study confirm an high prevalence of secondary osteoporosis in kidney transplantation and in a significative percentage of renal transplant patients resulted an increase of ten years probability of hip and major osteoporotic fractures for which treatment is recommended. Calcium and vitamin D therapy alone resulted inefficient to decrease the probability of HP and MOF and less utilized particularly in males. Different more active and standardized routine treatments are needed to decrease fracture risk in renal transplant patients.

Su657 PROGNOSTIC SIGNIFICANCE OF RENAL ARTERIAL RESISTANCE INDICES FOR RENAL ALLOGRAFT SURVIVAL: THE TIME POINT MATTERS

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Introduction and Aims: The renal arterial resistance index (RI) is reported to be a significant predictive parameter for renal allograft survival. The influence of the time point after renal transplantation on the predictive value has not been sufficiently evaluated. We performed a retrospective analysis of RI and its power to predict renal allograft survival with special emphasis on the time point of RI measurement.
Methods: The present analysis is based on ultrasonographically recorded intrarenal arterial RIs, routinely obtained in our outpatient department, over a period of 13 years. Altogether 396 patients with 5717 scans were stratified retrospectively into two groups according to their RI: those with an index ≥0.75 and those with index <0.75. Cross-sectional analysis at the time periods 0-3 [n=254], 3-6 [n=147], 6-9 [n=97], 9-12 [n=99], 12-18 [n=161] and 18-30 [n=184] months after transplantation was used to detect a change of the predictive power of RI over time after transplantation.
From the 396 patients we selected a group of n=146 patients for a separate longitudinal analysis concerning the time periods 0-3 and 12-18 months. Everyone of the 146 patients had one or more ultrasonographic examinations in both time periods. Only the first RI measurement within the time period was used for analysis. The primary end point was the terminal transplant failure requiring the reinstitution of dialysis.
Results: 68 patients (17%) reached the primary endpoint at 58,23±61,8 months after transplantation. The cross-sectional analysis showed that early RIs (0-3, 3-6 and 6-9 months) after transplantation were not predictive for renal allograft survival in the log rank test (p=0.086; p=0.0584 and p=0.1406). The first significant predictive value for renal allograft survival was the RI measured between 9 and 12 months after transplantation (p<0.05). In the time periods 12-18 and 18-30 months after transplantation, the RI became a highly significant predictive value for transplant survival (p<0.01 and p<0.0001). In the longitudinal analysis only the resistance index measured between 12 and 18 months after transplantation showed a significant hazard ratio [HR=5.236, 95% confidence interval 2.299-11.926, p<0.0001] for transplant failure.
Conclusions: We confirm the prognostic relevance of the renal arterial RI for renal allograft survival. However, only an RI ≥0.75 measured more than 9 months after transplantation was a potent negative predictor, whereas RIs obtained earlier in the course after transplantation were not predictive in our population.

Su658 THE ROLE OF B-CELLS, T-CELLS AND MONOCYTES IN ACUTE REJECTION AND CHRONIC ALLOGRAFT NEPHROPATHY

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Introduction and Aims: In acute rejection infiltrates of T- and B-cells are observed. Increased monocyte infiltrates were described, when patients received a T-cell depletion therapy. The contribution of immune cells to chronic allograft nephropathy is still unknown but it is hypothesised that the antibody mediated rejection could play a role in the pathogenesis of late graft failure.
We studied cell infiltrates in kidney transplant biopsies and looked for correlations with clinical outcome.
Methods: Immunohistochemical stainings of 67 biopsies of kidney transplants with vascular rejection (vR, n=12), interstitial rejection (iR, n=17), acute tubular necrosis (ATN, n=16), and chronic allograft nephropathy (CAN, n=22) subdivided into transplant glomerulopathy (TG, n=16) and interstitial fibrosis (IF, n=6), were performed using monoclonal antibodies against CD20, CD65 and CD45RO to identify B-cells, monocytes and T-cells.
All biopsies were classified according to Banff criteria.
We determined the number of positively stained cells in glomerulus (mean cell count/glomerulus) and tubulointerstitium (cells/high power field, 400x). The results of leukocyte infiltrates were compared between the groups using the Mann Whitney Test and correlated to serum creatinine and creatinine clearance (e.g. 1-3 years after biopsy) using Kendall Tau correlation.
Results: We observed significantly increased B-cell counts in glomeruli and T-cell counts in tubulointerstitium in biopsies with acute rejection compared to CAN (B-cells: vR vs CAN: mean rank 22.75 vs 10.67, p<0.0001 and iR vs CAN: mean rank 22.76 vs 13.50, p=0.007; T-cells: vR vs CAN: mean rank 22.54 vs 13.40, p=0.008 and iR vs CAN: mean rank 26.11 vs 14.20, p=0.001).

There was no significant difference in T- or B-cell numbers between acute vascular and interstitial rejection, but an increased amount of monocytes in glomeruli was seen in vascular rejection and transplant glomerulopathy compared to interstitial rejection (vR vs iR: mean rank 20.04 vs 13.08, p=0.035; TG vs iR: mean rank 22.72 vs 12.86, p=0.004) and transplant glomerulopathy compared to interstitial fibrosis (mean rank 13.28 vs 6.75, p=0.036).
Additionally an increase of T-cells in glomeruli was observed in transplant glomerulopathy compared to interstitial rejection (mean rank 22.13 vs 14.53, p=0.029).
As expected, significantly less infiltrates of all 3 cell types were observed in biopsies with ATN.
We observed a trend to worse clinical outcome in patients with vascular rejections, who had increased B-cell infiltrates in glomeruli.
Conclusions: These results indicate a role of B- and T-cells in acute vascular and interstitial rejection and T-cell and monocyte infiltrates in glomeruli in transplant glomerulopathy. Interestingly the monocyte infiltrates in glomeruli also play a role in vascular rejection. These results and also an increase of monocytes in glomeruli in biopsies with transplant glomerulopathy compared to interstitial fibrosis indicate an immunological pathogenesis of this entity.

Su659 POLYMORPHISM IN THE FRACTALKINE RECEPTOR CX3CR1 AS A GENETIC RISK FACTOR FOR CANCER AFTER TRANSPLANTATION

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Introduction and Aims: A single nucleotide polymorphism in the fractalkin (FKN) receptor (CX3CR1) coding sequence (T280M) has been identified. The polymorphism alters ligand-receptor affinity and reduces FKN signalling. Innate immunity is critically important for tumor surveillance and regulating tumor metastasis. FKN, operating through the receptor CX3CR1, is an effective chemoattractant and adhesion receptor for NK cells and monocytes, important constituents of the innate immune response. Previous studies have shown that over-expression of CX3CL1 by tumor cells enhances antitumor responses. Cancer occurrence is increased in transplant patients. We hypothetized that T280M allele may be associated with cancer occurrence in this population. We studied the association between this polymorphism and cancer in two independent cohorts of renal transplant recipients including a total of 603 patients.
Methods: We studied the association between this polymorphism and cancer in two independent cohorts of renal transplant recipients including a total of 603 patients. The functional effect of T280M allele was analyzed by the capacity of natural killer cells to produce IFN-g. Median follow-up were 8.7 and 7.9 years for the first and second cohorts, respectively.
Results: Analysis of 603 patients identified 21 MM (3.5%), 170 TM (28.2%), and 412 TT (68.3%) carriers. The observed allele frequencies were in Hardy-Weinberg equilibrium. Cancer incidence was higher in TT patients in the two cohorts (50% vs 22%, p= [cohort 1] and 29% vs 14%, p= [cohort 2]). Patients with the MM haplotype have an independent increased risk of graft loss (HR 3,53 [1,35-9,28], p=0,010) compared to GG patients.
Conclusions: The T280M polymorphism is associated with a higher rate of cancers in renal transplant recipients. Such findings may be used to influence immunosuppressive strategies and optimize patient management.

Su660 DIABETES MELLITUS IS A RISK FACTOR FOR ZYGOMYCOSIS IN RENAL TRANSPLANT RECIPIENTS

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Introduction and Aims: Invasive fungal infection is an important cause of morbidity and mortality in renal transplant recipients (RTR). Although rare, zygomycosis carries an extremely poor prognosis when affects RTR. Diabetes mellitus, metabolic acidosis, use of steroids and iron overload are known risk factors in these patients. Diabetic patients have increased iron stores because of impaired iron utilization, which may contribute to their increased risk of zygomycosis.
Methods: We retrospectively reviewed our patient's records to find out the prevalence of the infection among them and to point out risk factors, diagnostic approaches, therapeutic options and clinical outcome.
Results: Seven patients out of 950 RTR under follow up from 1994 till 2009 were diagnosed to have invasive zygomycosis. Three had nasal sinus infection without cerebral involvement, two had pulmonary, one had hepatic and another had isolated renal graft involvement. They were all heavily immunosuppressed either through induction immunosuppression or treatment of acute rejection. Six patients were diabetic with poor control in spite of intensive insulin therapy. One of the nasal cases had a history of chronic sinusitis and another one had surgically treated renal cell carcinoma of the native kidney. The nasal sinuses cases were treated aggressively with surgical debridement and anti fungal therapy and cured. Two of them died after cure due to other reasons. The patient with liver involvement was transferred to a specialized center abroad for further management and he died after coming back with liver and renal failure. One of the pulmonary case and the patient with renal graft infection died during treatment with antifungal therapy due to associated infections.
Conclusions: Zygomycosis in diabetic renal transplant recipients is a serious life-threatening infection. High index of suspicion, early diagnosis and aggressive therapeutic management are essential for better outcome.

Su661 MATRIX-Gla-PROTEIN, FETUIN AND OSTEOPONTIN IN CALCIFICATION OF RENAL ALLOGRAFTS

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Introduction and Aims: Calcification of renal allografts is common in the first year after transplantation and is related to hyperparathyroidism. It is associated with an impaired long-term outcome (Am J Transplant 2005;5:934-41). Aim of this study is to examine factors besides hyperparathyroidism which are related to the calcification.
Methods: We analyzed protocol allograft biopsies, blood and urine samples of 30 patients with and 30 patients without allograft calcification taken at 6 weeks, 3 and 6 months after transplantation. Patients were matched for post-transplant parathormone and serum calcium levels. Demographical data, cold ischemia time, initial graft function and donor characteristics were comparable between the two groups. Analyzed factors included serum and urine electrolytes, matrix-Gla-protein, fetuin, FGF-23 and vitamin D in serum/plasma and osteopontin (OPN) in urine. Biopsies were stained for matrix-Gla-protein and Fetuin.
Results: In patients with calcification, matrix-Gla protein levels were significantly higher (25%) at 6 weeks compared to control patients without calcification, but decreased thereafter, reaching levels that were 27.5% lower than that of the controls at 6 months. Fetuin levels were higher in patients with calcification (16.2% at 6 weeks; p=0.021 and 22.9% at 3 months; p=0.06). Urinary OPN was lower compared to the control group at 6 weeks after transplantation (by 66.4%; $p<0.001$), but increased by 6.2-fold, reaching values 75.6% over the controls at 6 months after transplantation. FGF-23 and vitamin D levels were unchanged. In the biopsies with calcification, matrix Gla protein and fetuin was exclusively located in the vicinity of crystal deposits.
Conclusions: The apparent differences in serum matrix-Gla-protein and fetuin, and urinary OPN indicate that the allograft calcification is rather a systemic disease characterized by severe changes in circulating anti-calcifying factors.

Su662 PREVALENCE OF LATE POSTTRANSPLANT ANEMIA IN ADULT KIDNEY TRANSPLANT RECIPIENTS AND THE ROLE OF SIROLIMUS – A SINGLE CENTRE STUDY

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Introduction and Aims: Late posttransplant anemia (PTA) is frequent complication of kidney transplantation with ill-defined pathogenesis and prevalence. The aim of this study was to evaluate PTA prevalence and its predictors among 1523 kidney transplant recipients.
Methods: From the hospital outpatient database of all kidney transplant recipients with follow-up > 3 months, anemic patients with hemoglobin (Hb) < 11 g/dL analyzed at least twice within 2008 or those with erytropoiesis stimulating agents (ESA) therapy were identified. Control group consists of non anemic subjects who consecutively received kidney grafts to anemic ones. Detailed cross-sectional database consisting of demographical data, transplantation-related data, comorbidities, biochemical findings and therapy was constructed. Univariate analyses and multiple regression analyses were performed to find out determinants of PTA.
Results: Among 1523 patients, 147 (9.7%) anemic patients were identified. Mean Hb was 10.4±0.5 g/dL in anemic untreated patients, 10.8±1.8 g/dL in ESA-treated patients and 13.6±1.5 g/dL in the control group. Median time after transplantation was 7.4 years in the anemic group that was identical to the control group. In anemic patients compared with controls, deteriorated graft function (GFR 28.8±13.8 mL/min/1.73m^2 vs. 49.2±15.6 mL/min/1.73m^2; p=0.000), older donor age (47.0±15.3 years vs. 39.6±16.6 years; p=0.000), smaller RBC (MCV 89±7.1 fL vs. 90.6±5.6 fL; p=0.028) and lower serum albumin (37.2±4.9 g/L vs. 40.8±3.6 g/L; p=0.000) were identified in univariate analyses. Anemic patients received more frequently sirolimus (23.1% vs. 10.2%; p=0.003), whereas treatment with mycophenolate mofetil was identical in both groups. In multiple regression analyses, renal graft function (OR 9.81, 95% CI 5.65-17 for GFR < 30 mL/min/1.73m^2; OR 1.7, 95% CI 1.52-1.98 for every 6 mL/min/1.73m^2 GFR reduction; p=0.000) and sirolimus therapy (OR 3.64, 95% CI 1.61-8.23; p=0.002) only were predictive factors for PTA. The highest predictive value for sirolimus related anemia was observed in GFR 32.4-45.6 mL/min/1.73m^2.
Conclusions: In this study, kidney graft dysfunction and sirolimus therapy were identified as independent risk factors for PTA development. Moreover, sirolimus contribution to PTA was highest in CKD 3T patients.

Su663 PROGRESSION PATTERN OF CHRONIC ALLOGRAFT DYSFUNCTION AMONG LONG TERM RENAL TRANSPLANT RECIPIENTS

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Introduction and Aims: Transplantation is currently the best renal replacement modality for patients with end-stage renal disease.Graft loss due to chronic allograft dysfunction (CAD) is a major concern in renal transplant recipients. There is little data about the pattern of disease progression in this patient population. We hypothesized that K/DOQI classification of chronic kidney disease (CKD) is applicable to this patients. We apply this staging system to determine pattern of disease progression per stage in this group of pts.
Methods: We performed a retrospective study on 214 RTR with CAD among 1534 RTR at Urmia University Hospital from 1997 to 2005. Selection criteria were a functional renal allograft for at least 1 year after transplantation and a progressive decline in allograft function. The Cockcroft-Gault estimation of creatinine clearance was used to estimate kidney function. The patients have been visited at the clinic several times during the study period by nephrologists. Patients were staged by the values of creatinine clearances based on the K/DOQI classification of CKD [3]. The worst kidney CAD stage for each visit was used to define the patient's stage at the time of inclusion.
The pattern of disease progression was assessed by defining the survival rates per stage, mean waiting times of progression from one stage to the next one and death-censored graft loss by Kaplan-meier survival analysis. The log-rank tests were used to compare stage to stage progression survival and death-censored graft loss between groups
Results: Among 1534 RTR 214 (pts) fulfilled CAD criteria. Among pts with CAD 152 (71%) were male and 62 (29%) were female. Mean time to enter to CAD stage 1 was 9.8±2.4 months post transplantation. Mean of CLcr measurement during follow up period was 32.1±9.9 (range 12-56) times. At the beginning of the study 117 (54.7%) of pts belong to stage 1, 81(37.9%) to stage 2, 16(7.5%) to stage 3, and no one to stage 4 or 5. At the end of study no pts belong to stage 1, 22(10.3%) belong to stage 2, 85 (39.7%) to stage 3, 50 (23.4%) to stage 4 and 57 (26.6%) to stage 5. At the end of the study 20 (17.1%) pts from stage 1, 26 (32.1%) from stage 2 and 11(67.7%) from stage 3 progressed to stage 5. Most pts reached to stage 4 or 5 at the end of the study. There was a significant correlation (Nonparametric Kendall's Correlation) between stage of CAD at the beginning of study and the stage of CAD at the end of study ($r = 0.465$, $p<0.001$).
Conclusions: This study describes the change in GFR among transplant recipients with CAD. Because GFR decline after transplantation was relatively faster in more advanced stages of CAD, strategies to increase allograft survival by improving the baseline level of allograft function may be more effective than strategies to slow the progression of advanced stages of CAD in kidney transplant recipients.

Su664 DOPPLER ULTRASONOGRAPHY AS A PREDICTOR OF EARLY RENAL ALLOGRAFT FUNCTION

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Introduction and Aims: Despite the fact that kidney transplantation is the method of choice in renal replacement therapy problem of delayed graft function (DGF) is a major complication affecting the normal functioning of the graft at early and late periods after allotransplantation.
Methods: Sixty renal transplant patients underwent a renal transplantation in our center in 2009. Thirty patients received their grafts after deceased multiorgan donation (MOD) and thirty – after deceased kidney only donation (KOD).We studied renal blood flow using Doppler during the first and 7-10th day after the operation estimating blood circulation in interlobular blood vessels that always react to metabolic disorders early. Maximal systolic speed (cm/sec)-Vs, final diastolic (cm/sec)-Vd, resistivity index and pulsative index were assessed.
Results: Immediate graft function was observed in 24 patients (80%) who received the transplant as a result of KOD and in 19(63%) patients of MOD. Six patients (20%) with grafts from KOD and 11(37%) from MOD developed DGF.

	Kidney Donation		Graft Function	
	KOD	MOD	IGF	DGF
Vs 1	0,20±0,04	0,27±0,02	0,18±0,04	0,30±0,03
Vs 2	0,38±0,03*	0,29±0,03	0,43±0,04*	0,38±0,06
Vd 1	0,08±0,01	0,06±0,01	0,07±0,01	0,06±0,01
Vd2	0,10±0,02	0,08±0,02	0,13±0,02*	0,09±0,03
Ri 1	0,67±0,03	0,71±0,06	0,63±0,03	0,74±0,04
Ri 2	0,53±0,02*	0,68±0,04	0,43±0,02*	0,67±0,03*
Pui 1	1,02±0,30	1,39±0,28	0,99±0,27	1,39±0,63
Pui2	0,83±0,12*	1,12±0,33	0,62±0,24*	1,12±0,33

*p<0,05, Vs1, Vd1, Ri1, Pui1 – measured during 1st day after the surgery, Vs2, Vd2, Ri2, Pui2 – measured during the 7-10th day after the surgery.

We noticed an immediate and stable urine output when initial level of Ri was relatively low 0,63±0,03. A further drop in the resistive index up to 0,43±0,02 (a decrease in 1,46 times) indicated a rapid normalization of the excretory function to the beginning of the second week after the operation. In contrast, delayed graft function was observed at higher values of resistance index (0,74±0,04), while the increase in GFR has been more slowly due to continuing to 10 days of disturbances of microcirculation, as indicated by indicator Ri -0,67±0, 03 (decrease in 1,10 times).
Conclusions: This study shows the benefits of KOD vs. MOD because of ischemic damage to the tubules due to the lack of renal blood flow during

the removal of the heart and liver. Doppler study can be considered a good predictor in determining the function of kidneys at early and late periods after allotransplantation.

Su665 SINGLE CENTRE EXPERIENCE OF SCREENING FOR BK VIRUS IN RENAL TRANSPLANT RECIPIENTS

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Introduction and Aims: BK virus carrier rate in adult populations is about 60-90%. It may reactivate in renal transplant recipients (10-68%). However, of those, only 1-10% will progress to histologically proven BK virus associated nephropathy (BKVAN). Recent recommendations suggested that preemptive screening of renal transplant recipients for viruria and/or viraemia may identify at risk groups prior to the development of BKVAN. These have led to a change of practice in our unit with the introduction of monthly screening of new renal transplant recipients for the first 6 months post transplantation followed by three monthly screening for the next 18 months.

Our study aims to answer the following:

- Is random screening of new renal transplant patients picking up patients with BKVAN prior to development of clinically detectable disease?
- In patients with BKVAN was there a clinical indicator to prompt testing for the virus?
- Does screening for BK virus (BKV) alter clinical management, in the absence of biopsy proven BKVAN?

Methods: All renal transplant recipients were identified from a departmental database. We retrospectively obtained data from a computerised results server on BK virus testing and biopsy results. Further information on indication for test, treatment changes and clinical outcomes was obtained from patients' clinic letters.

Results: A total of 225 renal transplant recipients were identified. Of those, 57 had a BKV test at some point. The primary reasons for testing were rise in creatinine (27), screening (15), unknown (13), neutropenia (1) and haematuria (1).

34 patients had a urine test. Of those, 29 were negative while 5 were positive (3 of those also had subsequent viraemia). 40 patients had a blood test. 10 had viraemia while 30 were negative. In total 12 patients tested positive (7 with isolated viraemia, 2 with isolated viruria and 3 were positive for both). 9 out of those 12 patients were biopsied and 6 had BKVAN. In all 6 patients with BKVAN the reason for initial testing was a rise in serum creatinine.

All patients with biopsy proven BKVAN (n=6) had alterations in their treatment regimes. Of the 6 who either had negative biopsies for BKVAN (n=3) or were not biopsied (n=3), 3 did not require any treatment intervention while 3 had immunosuppression reduction for other reasons.

Conclusions:

- In our cohort of transplant recipients random screening for BKV did not alter clinical management.
- We identified rise in serum creatinine as a strong clinical indicator for BKVAN.
- In our study, random screening did not pick up any patients with BKVAN prior to development of graft dysfunction.
- In the absence of graft dysfunction, positivity for BKV did not alter clinical management. Patients who tested positive and were clinically stable, had several follow up urine and blood tests for BKV but no renal biopsy. This could result in unnecesary anxiety in this group of patients and raises issues of whether screening is cost effective.
- We recognize that this is a small cohort of patients. We would recommend following local guidelines for identification and management of BKV associated disease.

Su666 RELATIONSHIP BETWEEN SERUM PARAOXONASE AND HOMOCYSTEINE THIOLACTONASE ACTIVITY, ADIPOKINES AND ASYMMETRIC DIMETHYL ARGININE LEVELS IN DIABETIC, OBESE RENAL TRANSPLANT PATIENTS

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Introduction and Aims: Increased oxidative stress and inflammation are associated with atherosclerotic coronary disease in renal transplant recipients. HDL-associated paraoxonase (PON1) and thiolactonase prevent LDL-C from oxidation thus providing protection against atherosclerotic and coronary artery disease. Our aim was to investigate the correlation between serum paraoxonase and homocysteine thiolactonase (HTLase) activity, renal function, adiponectin, leptin and asymmetric dimethylarginine (ADMA) levels in kidney transplant recipients.

Methods: 79 transplanted patients (38 males, 41 females, age: 49.01±14.00 ys) were enrolled in the study. We examined fasting serum creatinine, cystatin C, homocysteine, CRP, glucose and lipid levels. PON1, HTLase activities were determined spectrophotometrically. Serum adiponectin, leptin and ADMA levels were measured by ELISA method.

Results: Our patients have hypercholesterolaemia, high LDL and ApoB levels and parallel with improved renal function, decreased cystatin C and homocysteine levels ($p<0.001$). In obese patients (BMI $>$ 30 kg/m^2, n=14) LDL ($p<0.05$) and leptin concentrations (53.64 vs. 17.09 ng/ml, $p<0.01$) were significantly higher than that in the malnourished group (n = 10). We found significant difference between serum adiponectin levels (16.61 vs. 23.69 μg/ml) and PON1 activity (99.18 vs. 79.59 U/l) in obese and malnourished renal transplant recipients. HTLase activity was 337.24 and 380.94 U/l in obese and malnourished patients, respectively (not significant). Between serum leptin concentration and PON1 activity there was a non-significant negative correlation. After transplantation a significant negative correlation was found between serum PON 1 activity and improved renal function ($p < 0.01$). Between PON1 activity and adiponectin levels there was a significant correlation (p=0.0276) and between PON1 activity and ADMA levels there was a negative, non-significant correlation (p=0.2302). Connection between ADMA and serum leptin and CRP levels there was a positive, but not significant correlation. Between HTLase and PON/HDL ratio there was a positive linear correlation in malnourished patients ($p<0.03$).

Conclusions: Dyslipidaemic, obese transplanted patients have high LDL, leptin concentrations and paraoxonase activity does not correlate significantly with leptin, ADMA and CRP levels. With improved renal function significant increase in PON1 activity can be found. HTLase positively correlated with PON/HDL ratio.

Su667 CYSTOPLASTY WITH SMALL INTESTINAL SUBMUCOSA (SIS)GRAFT. A VALID OPTION IN RENAL TRANSPLANTATION

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Introduction and Aims: Bladder augmentation is a well-established method of treating severe bladder dysfunction in patients with end stage renal disease (ESRD) who will undergo kidney transplantation. Nevertheless, cystoplasty using segments of the gastrointestinal tract is associated to surgical and medical complications such as perforation and bowel obstruction due to intestinal resection, electrolyte abnormalities, acidosis and infections. Since ileum secrets mucus, cystoplasty must be irrigated every day before transplantation. We report our experience on cystoplasty performed prior to renal transplantation in patients with a very poor bladder capacity using small intestinal submucosa graft (SIS), a collagen-based non immunogenic material harvested from the submucosa layer of porcine intestine.

Methods: 5 patients with ESRD (1 oligomeganephronia, 3 vesicoureteric reflux and 1 who underwent bilateral nephrectomy because of bilateral

renal carcinoma) and with a very poor bladder capacity due to urologic abnormalities were subjected to cystoplasty. Bladder augmentation was performed using SIS (4 layer Stratasis™). 3 renal transplants have been performed in 2 patients out of 5. One patient has been transplanted 14 months after cystoplasty, the other one received 2 kidney grafts (the first transplant complicated by a thrombosis of the renal vein was performed 4 months after cystoplasty; the second graft was transplanted 12 months later). 3 patients out of 5 are still in the waiting-list (WL).

Results: Bladder capacity after augmentation increased from a mean of 44±31.1 ml to a mean of 173±72.2 ml ($p<0.014$). The follow-up of the 2 patients who underwent a renal transplant are 14 months and 64 months, respectively. Bladder protocol biopsies performed in all patients after 3 months from cystoplasty demonstrated a complete re-epithelization of the SIS with a mild inflammation of the mucosa. 2 episodes of mild urinary tract infections (one 3 months after kidney transplant and one 2 months after cistoplasty in a patient in WL) resolved with antibiotic therapy were observed. 1 patient in WL developed macrohematuria due to bladder bleeding that resolved spontaneously. None of the transplanted patients developed electrolyte abnormalities or acidosis.

Conclusions: At our knowledge this is the first report of the use of small intestinal submucosa (4 layer Stratasis™) for bladder augmentation in patients with severe reduction of bladder capacity that underwent renal transplantation. However, many open questions need to be addressed. In particular, more studies have to be performed in order to evaluate the long-term durability of the bladder reconstruction. Nevertheless, based on our experience of 5 cases (2 patients transplanted and 3 in WL), SIS appears to be an effective reconstructive material that might be a valid option for cystoplasty in renal transplanted patients avoiding major surgical complications related to intestinal intervention.

Su668 DERMATOLOGICAL LESIONS IN KIDNEY TRANSPLANT RECIPIENTS: A RETROSPECTIVE ANALYSIS

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Introduction and Aims: Renal transplantation is the gold therapy for patients with end-stage renal failure. Chronic use of immunosuppressive drugs predispose patients to opportunistic infections and increased risk of malignancies. Reports on skin lesions in kidney transplant patients (ktx) are very few. The aim of our study was to evaluate the prevalence of skin lesions in ktx and analyze any relation between immunosuppressive drugs, demographic characteristics and cutaneous manifestations.

Methods: A retrospective analysis was performed using medical records of ktx followed up between 2000 and 2009 at the Transplant Unit of Pavia. The study included 183 patients (M 57.3%,F 42.7%), age (M±SD) 51.5±11.8 years and transplant age (M±SD) 52.3±34.9 months. Induction therapy consisted of basiliximab and steroids; maintenance therapy included associations of cyclosporine, tacrolimus, steroids, mycophenolate mofetil (MM), micophenolic acid (MA), rapamycine, everolimus. Anti-rejection therapy consisted of steroids and/or thymoglobulines. The diagnosis of cutaneous lesions have been made by a evaluation of skin, mucous membranes, nails and hair. Skin biopsies, specific cultures, seriological tests were performed according to indication. Data were compared using Fisher test and Mann Whitney test.

Results: Skin and mucosal lesions were reported in 95,7% of patients. 37,7% of pts showed viral lesions (Herpes simplex 25,6%, Herpes zooster 14,7%, Warts 8,7%), 25,6% of pts showed immunosuppression-related lesions, 20,2% bacterial lesions, 15,8% benign tumours, 14,2% mycosis, 11% precancerosis lesions, 9,2% cutaneous xerosis, 8,7% dermatitis, 8,2% malignant tumours.

A statistical correlation was found between calcineurin inhibitors and gingival hyperplasia ($p<0,001$); mTOR inhibitors and acne/folliculitis ($p<0.05$), MA and herpes simplex lesions ($p<0,05$); MM and warts ($p<0.05$). Anti-rejection therapy was related to precancerosis lesions ($p<0,001$), bacterial lesions ($p<0.05$) and gingival hyperplasia ($p<0.001$). Malignant tumours were more frequent in patients with advanced age (median age 66 vs 51.6, $p<0.001$), and surprisingly were not associated with any immunosuppressive therapy, or anti-rejection therapy. Herpes Zooster was a complication more frequent in the first two years post-transplant ($p<0.05$), immunosuppression-related lesions appear in the first year post transplant ($p<0.001$), precancerosis lesions and malignant tumours in patients after 67,5 months from transplantation.

Conclusions: Cutaneous manifestations are frequent in kidney transplanted patients. Early and continuous monitoring skin is necessary in order to make early diagnosis, to start appropriate treatment and avoid long term damages.

Su669 REASONS FOR DOSE REDUCTION OF MYCOPHENOLATE MOFETIL (MMF) AND IMPACT ON GRAFT OUTCOME IN FIRST YEAR AFTER KIDNEY TRANSPLANTATION: A SINGLE-CENTER RETROSPECTIVE ANALYSIS

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Introduction and Aims: MMF has represented a major advance in organ transplantation. Its potential side-effects, however, may ask for dose reduction implicating higher risk of rejection and graft loss. The aims of the present analysis were to describe reasons for dose reductions of MMF and to evaluate their impact on graft outcome in the first year after kidney transplantation.

Methods: We performed a retrospective analysis of single kidney transplant recipients that received MMF in their initial maintenance immunosuppressive regimen from 1996 till 2007. Data on immunosuppression, MMF dose changes, reasons for dose reduction, acute rejections, graft and patient survival up till the 400th day after transplantation were retrieved from patient files. 749 patients were included.

Results: Initial daily MMF dose was 1g (n=415) or 2g (n=302) in most patients (all bid). Other immunosuppressants were methylprednisolone (all), tacrolimus (n=620), cyclosporine (n=104), sirolimus (n=10), belatacept (n=14). In 358 patients (47.8%) MMF dose wasn't changed. In 73 (9.7%) MMF was permanently discontinued (graft loss censored). 326 (43.5%) patients had at least one dose reduction other than stopping MMF. Of all 741 reduction events (discontinuation included), 55.7% were for hematological reasons (see Figure 1). Other reasons were infection (17.7%), prescription by the trial protocol (10.9%), gastrointestinal side-effects (9.6%), correction of erroneous doses (1.3%), neoplasia (1.8%) and unidentifiable causes (10.4%) (some reasons coincided). 197 patients (26.3%) lived at least one acute rejection episode. The proportion of rejections was higher in the subgroup with MMF dose reduction and/or withdrawal than in the remainder of the patients: 34.8 vs. 19.0% (χ^2 $P<0.05$). 46 patients lost their graft or died before day 400. Their proportion was highest in the subgroup with MMF dose reduction and/or withdrawal: 8.0 vs. 4.0% (χ^2 $P<0.05$).

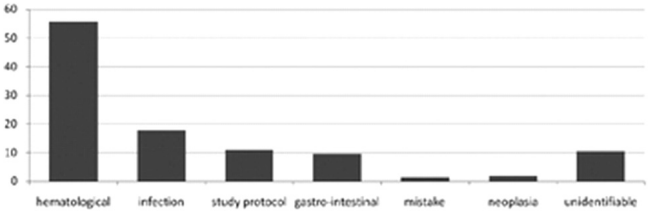

Figure 1. Reasons for MMF dose reduction expressed as percentage of total number of dose reduction events.

Conclusions: Main reasons for MMF dose reduction after renal transplantation were hematological changes and infection, while gastrointestinal side-effects were less important. MMF dose reduction was associated with higher incidence of acute rejection and graft loss during the first post-transplant year. Analysis of the interplay with other determinants of outcome needs further investigation.

Disclosure: Our department's research activities are supported by grants from Baxter, Roche, Astellas, Amgen and Genzyme.

Su670 KIDNEY TRANSPLANTATION IN PATIENTS OVER 65 YEARS – A SINGLE CENTER EXPERIENCE

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Introduction and Aims: Transplantation is the best kidney function replacement therapy regardless of patient age. Therefore, it was implemented the "old-for-old" organs allocation programme in order to maximize the grafts from donors with expanded criteria (DEC). The aim of this study was to evaluate the outcome of kidney transplants performed in recipents over 65 years, in the last ten years.
Methods: We retrospectively studied 61 patients who received their first cadaveric renal graft between August 1990 and August 2009 (44 M/17 F, a mean age of 67,3±2,1 years).
Results: 42% of the organs came from donors with expanded criteria. The cold ischemia time was 18.7±5.0 h. 31% of patients were treated with monoclonal and polyclonal antibodies and the main immunosuppressive regimen consisted in mycophenolate mofetil, cyclosporine and prednisone (46%). There was a nonfunctioning graft in 5% and delayed graft function (DGF) in 36% of patients. 28% developed acute rejection (AR). In the first 3 months, the frequency of patients who had surgical and medical complications was 21% and 57% respectively. Infection (49%) and post-transplant diabetes (20%) were the medical complications with higher incidence. After 3 months, 51% of patients required hospital admission, mostly by infectious diseases (75%). About 33% of patients lost the graft. The main causes of loss were death with functioning kidney (45%) and chronic graft nephropathy (30%). The rates of graft survival at 1 and 5 years were 76% and 58% respectively. There was a lower survival of the organ in patients with DGF (p=0.007), AR (p=0.007) and those who received a kidney of DEC (p=0.04). Eleven patients died and the causes of death were infection (73%), cardiovascular disease (18%) and cancer (9%). The rates of patient survival at 1 and 5 years were 87% and 76% respectively.
Conclusions: In our unit, the majority of patients over 65 years received a renal graft from DEC. The high prevalence of DGF and AR probably had a negative influence on graft survival. More than the cardiovascular disease, the infection was an important factor of morbidity and mortality. The rates of graft and patient survival were clearly lower than those obtained, in the center, for younger recipients.

Su671 INFLUENCE OF ANGIOTENSINOGEN GENE M235T POLYMORPHISM ON LONG-TERM OUTCOME IN RENAL TRANSPLANTATION

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Introduction and Aims: An up regulated renin-angiotensin-aldosterone system is considered to contribute to the development chronic allograft nephropathy and graft loss in renal transplantation. In the present study we evaluated the impact of angiotensinogen gene M235T polymorphism on long-term outcome after kidney transplantation.
Methods: We studied n=205 consecutive patients, who underwent renal transplantation in our center (cadaveric: n=161, living related: n=44), followed up for 5.0±2.0 years. One hundred healthy volunteers were analyzed as controls. Angiotensinogen gene M235T polymorphism was determined by PCR amplification.
Results: The genotype distribution of the investigated polymorphisms was similar in patients and control subjects (ns). Age of donor and recipient, number of HLA-mismatches, cold ischemia time did not differ between patients with different genotypes (ns). During follow up n=103 (50.2%) patients had at least one episode of acute rejection and n=28 (13.7%) experienced graft failure. No association between the M235T polymorphism and the incidence of acute rejection was detected (at least one episode of acute rejection: TT: 17.5%, MT/MM: 82.5%; no episode: CC: 17.6%, CT/TT: 82.4%, ns). Patients carrying the TT genotype however experienced graft failure more frequently (27.8%) compared to the MM/TT genotypes (10.7, x^2: 7.4, OR: 2.43, 95%CI: 1.32-4.48, p=0.007). The Kaplan Meier analysis confirmed the impact of angiotensinogen gene polymorphism (p=0.005) on graft survival. This effect was prominent after the first two years after renal transplantation. In the Cox-regression analysis carriage of the TT genotype was associated with almost triple the risk for graft loss (HR 2.87, 95%CI: 1.33-6.23, p=0.008). M235T polymorphism remained an independent risk factor for graft loss in the multivariate analysis (HR 2.99, 95%CI: 1.37-6.49, p=0.006).
Conclusions: Our results suggest that angiotensinogen gene M235T polymorphism influence the long-term graft survival in renal transplantation, possibly through up regulating the renin-angiotensin-aldosterone system and contributing to the development of chronic allograft nephropathy.

Su672 SENSITIVITY AND SPECIFICITY OF URINARY CYTOLOGY IN DETECTING BK VIRUS ASSOCIATED NEPHROPATHY IN RENAL TRANSPLANT RECIPIENTS

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Introduction and Aims: BK virus nephropathy (BKVN) is an important problem in the renal transplant patient, leading to a high pourcentage of graft failure. Monitoring of BK viral load in urine and blood has been used as a surrogate marker of BKVN. The objective of the study is to evaluate the predictive value of urinary cytology combined with direct viral DNA detection in this population.
Methods: Prospective study for a period of 3 years that used a protocol of screening for Decoy cells in the urine: monthly in the first 6 months post transplantation, then once in 3 months in the first year, then yearly. Monitoring of BK viral load in urine and blood was performed by real-time polymerase chain reaction (PCR) at the same moments. Statistical evaluation was done with the programme SPSS.
Results: We have evaluated 333 renal transplant recipients (61.5% male, 38.5% female), transplanted in a single unit in France. Mean follow-up was 10 years. Age at transplantation: 48.97±13 years. We screened 1469 urinary cytologies (UC). We obtained correspondent PCR BKV values. We found 50 cytologies showing tipical signs of BKV infection. This was more frequent in males (60%) and statistically significant (p value 0.02). The duration after transplatation was 20.8±34 months, non significant. Values of PCR BKV in the blood ≥4 log and in the urine ≥7 log were considered predictive for the BKVAN.
The positive predictive value (PPV) of Urinary cytology compared with PCR BKV in the blood ≥4 log was 6%, the negative predictive value (NPV) was 98%, sensitivity (St): 27%, specificity(Sp): 93%, false positives: 0.07% and false negatives: 73%.
Compared with a value of PCR BKV in urine ≥7 log, the urinary cytology had a PPV of 26%, NPV: 96%, FP: 6%, FN: 63%, St: 37%, Sp: 94%.
Conclusions: 1.Urinary cytology has a specificity of 93-94% and a sensitivity of 27-37% for detecting BKVN, compared with PCR BK.
2. Urinary cytolgies were statistically significant more frequently positive in male patients.
3.False negatives were frequent (63-73%), the false positives were found in renal transplant patients with urothelial tumours, urinary parasitosis and other viral infections.

Su673 OUTCOME OF KIDNEY TRANSPLANTED PATIENTS WITH EXTENDED CRITERIA DONORS

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Introduction and Aims: The shortage of organs for transplantation has led to new strategies to increase the donor pool, including the use of extended criteria donors. However, recipients transplanted with such kidneys are more likely to have lower graft and patient survival. We compared the results at our Department between patients that received kidneys from extended and standard criteria donors.
Methods: We reviewed patients transplanted from deceased donors between January 2005 and June 2009 (n=245) and compared the results between those that received kidneys from extended criteria (n=64) and standard donors (n=181). Definition of marginal donors included age over 60 years

or between 50-59 years who died of stroke or had a history of hypertension or a creatinine (Cr) greater than 1.5 mg/dl at the time of death.
Results: Mean follow-up was 29±17 months in the control group (CG) and 22±13 in the extended criteria group (ECG). The patients age at transplantation was 44±12 years in CG and 55±10 in ECG (p=NS). Time in dialysis was longer in the ECG (100±78 months *vs* 80±66, p<0.05). The prevalence of hypertension (CG-32%, ECG-27%), *diabetes* (CG-7.2%, ECG-13%) and ischemic heart disease (CG-5%, ECG-4.7%) was similar (p=NS). Donor's age in the CG was 38±12 years [8-59] and 61±7 in the ECG [51-75], p<0.05. Most of the donors (52%) in the ECG had ≥ 60 years of age. The remaining had ≥ 50 years, 52% died of cerebrovascular accident, 45% were hypertensive and 16% had Cr ≥ 1.5. HLA identities were 2±1 in both groups. The most used induction scheme was daclizumab, tacrolimus, mycophenolate mophetil and prednisolone in both groups (CG-31%, ECG-45%). The number of patients submitted to antithymocyt globulin was similar (CG-37%, ECG-36%, p=NS). Delayed graft function was not higher in ECG (45% *vs* 38% in CG, p=NS). Six patients had primary non-function in CG and one in the ECG (p=NS). Acute rejection was similar in both groups in the first 3 months (13 episodes in CG, 9 in ECG; p=NS) and thereafter (4 in CG, 1 in ECG; p=NS).
Creatinine at the 1st and 3th year post-transplantation was lower in the CG (1.3±0.8 mg/dl *vs* 1.6±0.5 in ECG in the 1st year; 1.4±0.7 *vs* 2±0.8 in the 3th year; p<0.05). Cr clearance at the 1st and 3th year was higher in the CG (66±26 ml/min *vs* 47±15 in ECG in the 1st year; 65±20 *vs* 40±17 in the 3th year; p<0.05).
Patient survival was lower in the ECG at the 1st (92% *vs* 98%; p=0.04) and 3th year (63% *vs* 95%; p<0.001). Kidney survival was similar in both groups at the 1st year (89% in ECG *vs* 95% in CG; p=NS) but lower in the ECG at the 3th year (63% *vs* 85% in CG; p=0.03). The main causes of kidney loss were dead of the receptor (5 patients in each group; p=NS) and thrombosis (CG=5 cases, ECG=2; p=NS). The main causes of death were infection (n=5) and cancer (n=3).
Conclusions: Kidney transplantation from extended criteria donors was associated with lower patient survival at the 1st and 3rd year, lower graft survival at the 3rd year and reduced kidney function. The rate of acute rejection, delayed graft function and primary non-function was similar. Marginal grafts provided acceptable survival rates.

Su674 ARFI-BASED TISSUE ELASTICITY QUANTIFICATION AND KIDNEY GRAFT DYSFUNCTION: FIRST CLINICAL EXPERIENCES

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Introduction and Aims: Acoustic Radiation Force Impulse Imaging (ARFI) is a new ultrasound-based technology (Siemens Acuson, S2000) for measuring tissue elasticity properties which is integrated into a high-end ultrasound machine.
So far no experience has been reported with the evaluation of the new method in renal transplant follow-up. The purpose of this study was to evalue changes in ARFI-measurements between stable renal allografts and biopsy-proved transplant dysfunction.
Methods: We performed 16 serial ultrasound examinations in 8 renal transplant patients in a prospective study. Patients were first examined when presenting with stable allograft function for routine transplant kidney ultrasound. A second follow-up examination was performed when patients presented with graft dysfunction for allograft biopsy and histological evaluation. All patients were examined using ARFI-quantification (15 measurements/kidney). Additionally, resistance indices (RI) were calculated based on pulsed-wave Doppler ultrasound and transplant kidney size by B-mode ultrasound images. All biopsies underwent histological examination by a reference nephro-pathologist who was unaware of the results of the sonographic studies. Pathological diagnoses were based on biopsy results.
Finally we calculated the relative changes of ARFI-quantification, resistance indices and kidney size on a percentage basis as well as absolute change of kidney size at the times of assessment and compared the results with the final pathological diagnosis.
Results: Histology revealed in five cases an acute T-cell-mediated rejection, in one case calcineurin inhibitor toxicity and in two cases acute tubular necrosis. Calcineurin inhibitor toxicity and acute tubular necrosis were subsumed under "other pathologies". Mean ARFI-values increased on average by more than 15% percent in transplants with histologically proven acute rejection, whereas no increase was observed in grafts with other pathologies. Mean RI values neither increased in the diagnostic group with acute rejection, nor in the group with other pathologies. Kidney size increased on average by 0,5 centimetres in allografts with acute rejection, whereas they decreased by only 0,17 centimetres in the group with other pathologies.
Conclusions: Our results suggest ARFI measurements as a new and promising tool for the follow-up of kidney grafts while RI-values and kidney size were of doubtful value in the evaluation of kidney allograft dysfunction as has been demonstrated in previous studies. This new parameter may also gain clinical importance as complementary tool for the diagnosis of kidney transplant rejection.

Su675 IMPACT OF HCMV INFECTION AFTER RENAL TRANSPLANTATION

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Introduction and Aims: Cytomegalovirus (HCMV) is usually associated with asymptomatic infection in immunocompetent individuals, but it is major cause of morbidity and mortality in immunocompromised patients, including transplant patients. Objective: To monitor HCMV infection in renal transplant recipients.
Methods: Methods: We collected blood in EDTA tube to perform the technique antigenemia. Polymorphonuclear cells were isolated after the merger was agreed on and 3×10^6 cells were fixed on slides for later identification of the presence of viral antigen pp65 by immunoperoxidase reaction using monoclonal antibodies. Blood samples for the detection of antigenemia were obtained whenever there was clinical suspicion of cytomegalovirus infection.
Results: Results: Were performed 903 kidney transplants at the Hospital Kidney and Hypertension in the period October 2008 to October 2009. In this period there were 2100 antigenemia tests of kidney transplant patients. These 2100 tests from 797 patients, showed 349 positive tests to HCMV (16.6%), from 214 patients. Positively ranged from 1-869 positive cells in pp65 test. Forty-nine patients were examined clinically and were divided into 2 groups: 1) Twenty-nine patients not used antilymphocyte 2) Twenty patients used antilymphocyte. Among patients without cellular allograft rejection: 48.3% showed syndrome, 34.4% showed asymptomatic and 17.3% had invasive disease (GI Tract). Among patients with cellular allograft rejection who used antilymphocytic: 30% had invasive disease (GI Tract).
Conclusions: HCMV infection remains a major cause of morbidity in kidney transplant patients, emphasizing the need for monitoring. The antigenemia for HCMV have been quite useful in preemptive treatment. The infection by HCMV is still important in this context of transplantation with high incidence (23%), with significant involvement of gastrointestinal tract (24.5%) of patients with viral replication, principally Iin patients with use of antilymphocytes.
Disclosure: FAPESP, CAPES

Su676 UROLOGICAL COMPLICATIONS AFTER KIDNEY TRANSPLANTATION. ROLE OF TAGUCHI VS. WOODRUFF URETEROVESICAL ANASTOMOSIS AND INVOLVEMENT OF THE DOUBLE J STENT

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Introduction and Aims: Urological complications in renal transplantation are an important cause of increased hospitalization and morbidity. The frecuency of these complications varies between 4 and 15% according to the center. Ischemic necrosis seems to play an important role in fistula and stenosis ureteral. The vasculature depends on fine accessory arteries situated in the lower portion of the kidney, which implies that the viability of this tissue may be compromised by ischemia – reperfusion syndrome.
Methods: This retrospective study analyzed a group of 343 transplants performed between January 1999 and July 2009, excluding those who lost the graft before two months after transplantation or those who received sirolimus or everolimus. The primary aim was to assess the risk of ureteral Fistula (F) or distal ureteral stenosis (S), depending on the urological anastomosis type, Woodruf (W) vs. Taguchi (T) and the presence or absence of Double-J stent. The secondary aims were to evaluate the role of residence time of the Double J stent as a risk factor of stenosis and other risk factors that can influence on the microvasculature of the ureter.
Results: The incidence of urinary fistula (4.4%) and ureteral stenosis (3.5%), assessing the role of the type of extravesical anastomosis used, Woodruff (W) vs Taguchi (T), and the use of Double-J Stent or not. The frequency of ureteral stenosis according to anastomosis W (without double-J), W + Double J or T + Double J were 3.7%, 1.5% and 9% respectively (p=0.031). There were differences between W + Double J vs T + Double J (p=0.015) with HR = 6.3 95% (1.3 to 30). In association with Double-J stent, the incidence of fistula in patients with Double J Stent was: W (12%) vs W + Double J (0,8%) vs T + Double J (0%) p<0.0001. HR = 15.8 IC 95% (2 to 115) in patients without Double J stent. By logistic regression, we analized other risk factors that could influence on the microvasculature of the ureter. The best predictors of ureteral stenosis were Woodruf urological anastomosis (odds ratio 0.06, 95% CI: 0.01 – 0.49) and delayed renal function (odds ratio 10.1, 95% CI: 1.4 – 72.4). The ROC curve, to evaluate the quality of the predictive equation, obtained an area of 0.786 (95% CI 0.66 – 0.91, p=0.001). As to the fistula, the best covariates were double J stent (odds ratio 0.027, 95% CI: 0.003 – 0.227) and donor age (odds ratio 1.052, 95% CI: 1.01 – 1.096). Analyzed by ROC curve, this equation had an area of 0.881 (95% CI 0.83 – 0.94, p=0.0001).
Conclusions: Double J stent protects against the development of urological fistulas. Donor age results a risk factor. Any anastomosis with Double J stent prevents the development of urological fistulas. The Woodruff anastomosis with Double J stent is the best way to prevent ureteral stenosis, being the delayed renal function an important risk factor, maybe translating an ischemic event in ureter. The length of stay of Double J stent is not a risk factor for the development of ureteral stenosis.

Su677 RELATIONSHIP BETWEEN INITIAL CYCLOSPORINE DOSE AND C2 BLOOD CONCENTRATION AND DELAYED KIDNEY GRAFT FUNCTION COMBINED WITH HIGHER INTRARENAL RESISTANCE INDICES

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Introduction and Aims: Cyclosporine A (CyA) is one of the immunosuppressive drugs used in patients after kidney transplantation (KTx), but it presents nephrotoxic potential, including pronounced renal vasoconstriction. It could be reflected by the increased values of the kidney graft resistive parameters: pulsatility index (PI) and resistance index (RI), measured by Doppler sonography. On the other hand, CyA toxicity, along with an acute rejection and an acute tubular necrosis may lead to the delayed graft function. Therefore we investigate the relationship between initial CyA dosing and blood concentration and early graft resistance indices combined with DGF occurrence in a large cohort of kidney transplant recipients.
Methods: We have analyzed 434 consecutive kidney transplant patients, who initially received CyA-based immunosuppressive regimen. CyA blood concentrations were measured, both trough level (C0) and 2 hours after drug ingestion level (C2), during the first and second week after KTx. Intrarenal PI and RI were measured by Doppler sonography at the level of the interlobar artery on 2-4 day posttransplant.
Results: Delayed graft function (DGF) was observed in 155 out of 434 patients (35.7%). Mean initial CyA dose and first C2 blood concentration were significantly lower in patients with DGF than in non-DGF group (285±161 vs. 388±181 mg, p<0.001 and 676±328 vs. 886±373 ng/ml, p<0.001, respectively). Mean PI and RI values were significantly higher in DGF- than in non-DGF group (2.08±0.84 vs. 1.52±0.47, p<0.001 and 0.84±0.10 vs. 0.75±0.10, p<0.001, respectively). A negative correlations were found between resistance indices and both initial CyA dose and first C2 blood level (for PI: R -0.223, p<0.001 and R -0.173, p=0.002; for RI: R -0.219, p<0.001 and R -0.148, P=0.01, respectively), but not with the first C0 level.
Conclusions: Low initial CyA dose and low C2 blood level early post-transplant are related to the higher intrarenal vascular resistance within the transplanted kidney and more frequent occurrence of delayed graft function.

Su678 EXPERIENCE IN THE CONVERSION FROM PROGRAF TO ADVAGRAF

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Introduction and Aims: The effectiveness and reliability of tacrolimus as an immunosuppressant for the prevention of graft rejection has been established. A new formulation of tacrolimus with delayed release has been developed(Advagraf®), and it combines the well-proven safety of Prograf® with the possibility of having only one dose per day, which improves therapeutic adherence.
The objective of our study is to assess the characteristics and the evoluion of patients with renal transplant who are under immunosuppressant treatment with Prograf® and have been switched to Advagraf®.
Methods: We carried out a retrospective analysis of 60 patients with a mean age of 50.5±13.5 years, who had received a renal transplant between 1999 and 2008, and who had switched from Prograf® to Advagraf®. The conversion rate was 1mg:1mg. The switch took place within the first year after the transplant in 30 patients, and after the first year in the other 30 patients.
All variables were monitored on the day of the conversion, after 7 days (1st visit), after one month (2nd visit) and after three months (3rd visit).
The following intra- and inter-group variables were studied: Evolution of renal function, glycemia, cholesterol, tacrolimus levels, dose changes and basal and post-conversion immunosuppression doses.
All variables are expressed as X±SD. The statistical method used was Student's t-test. All values of p<0.05 were considered statistically significant.
Results: The mean time between the transplant and the conversion was 4.52±3.32 months for patients who switched within the first year, and 50.97±30.91 months for patients who switched after the first year.
No significant differences were found with regard to levels of creatinine, glucose, cholesterol, tacrolimus or Advagraf® levels between both groups and with regard to the evolution of creatinine, glucose and cholesterol levels within each group, or in the immunosuppression levels or doses in the patients that had a renal transplant of more than one year of evolution. On the contrary, patients with a transplant of less than one year of evolution showed lower immunosuppression levels in the first visit than in the second(p<0.05). Advagraf® doses were modified in 46.7% of the patients

Main characteristics of renal transplant patients who switched from Prograf® to Advagraf®

Dose changes	P1			P2		
	1st visit	2nd visit	3rd visit	1st visit	2nd visit	3rd visit
Increased dose	8 (26.7%)	12 (40%)	2 (6.7%)	5 (16.7%)	3 (10%)	4 (13.3%)
Reduced dose	6 (20%)	3 (10%)	4 (13.3%)	4 (13.3%)	6 (20%)	1 (3.3%)
Total	14 (46.7%)	15 (50%)	6 (20%)	9 (30%)	9 (30%)	5 (16.7%)

P1: Patients who switched to Advagraf® within the first year after the transplant;
P2: Patients who switched to Advagraf® after the first year of transplant.

in the first visit, in 50% in the second visit($p<0.05$) and in 20% in the third one.
Graft and patients survival was 100%, and no acute rejection episodes or other adverse events were registered.
Conclusions: Conversion from Prograf® to Advagraf® is effective and safe. Dose changes are common after the conversion, particularly in those transplanted patients with less than one year of evolution, but these are small, and therefore require more monitoring.

Su679 BETTER RENAL ALLOGRAFT FUNCTION WITH EVEROLIMUS FACILITATED CNI REDUCTION – GRAFT TYPE, DONOR CRITERIA AND GENDER ANALYSIS

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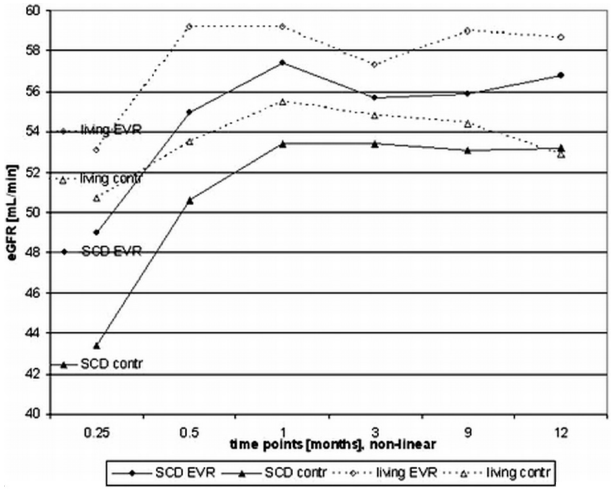
Introduction and Aims: A number of factors have been shown to influence renal allograft function (RF) and outcome. These include beside immunologic variables and procurement/shipment related factors, also donor criteria, allograft type, and presumably gender. A2309 is a randomized, prospective, multicenter trial that compares everolimus (EVR) [C0 3-8ng/mL] + reduced CsA vs mycophenolate sodium (MPA) + standard CsA.
Methods: Here, the impact of EVR facilitated CsA reduction on estimated GFR (eGFR) is assessed in (1) standard (SCD) vs extended (ECD) criteria donor, (2) deceased vs living donor allografts and (3) male vs female recipients.
Results: Immunologic variables, donor/allograft types and gender distribution between groups were comparable. The following results represent mean eGFR (MDRD) values at month 12 of the on-treatment population (n=403) (Table 1). Figure 1 displays evolution of RF over time (Fig. 1).

EVR vs MPA						
	[%]	[n]		eGFR [mL/min]		p-value
		EVR	MPA	EVR	MPA	
SCD	93	179	196	56.8	53.2	0.033*
ECD	7	15	13	50.5	47.6	0.927
deceased	47	89	99	54.6	52.3	0.601
living	53	105	110	58.7	52.9	0.027*
male	65	124	137	54.4	53.8	0.349
female	35	70	72	58.3	51.1	0.049*



Conclusions: In each of the 6 subgroup analyses EVR treated patients showed numerical higher eGFR at month 12. As expected, SCD did better than ECD, the same was observed for living vs deceased donor allografts. Interestingly, female recipients achieved a higher eGFR compared to males. For SCD, living and females the difference between EVR and MPA control reached statistical significance ($p<0.05$). In conclusion, A2309 showed that EVR allows for significant CsA reduction that translates into improved RF in distinct subpopulations.
Disclosure: Medical Scientific Advisor, Novartis Pharma AG.

Su680 LIVING-DONOR KIDNEY TRANSPLANTATION IN CROSSMATCH-POSITIVE PATIENTS WITH PERITRANSPLANT IMMUNOADSORPTION AND ANTI-CD20 THERAPY

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Introduction and Aims: Living-donor kidney transplantation in crossmatch positive patients is a challenge that requires specific measures.
Methods: Eight patients with either donor-specific antibodies against their living donor (DSA; n=1) or DSA and a positive B-cell CDC crossmatch (n=4) or DSA and a positive T- and B-cell CDC crossmatch with class I ELISA crossmatch positivity (n=3) were transplanted after successful desensitization with immunoadsorption and administration of anti-CD20 antibody immediately pretransplant. Patients in addition had either basiliximab (n=5) or thymoglobulin induction (n=3) and maintenance immunosuppression consisting of tacrolimus, enteric-coated mycophenolic sodium and steroids. Patients were followed by posttransplant antibody monitoring and protocol biopsies.
Results: Graft and patient survival rates at 1 year were 100% with a median serum creatinine of 1.72 mg/dl on day 14 and 1.68 mg/dl at year one. Seven out of 8 patients had at least one biopsy-proven acute rejection episode (borderline changes in 11 biopsies and BANFF IA rejection in 2 biopsies). Antibody-mediated rejection without graft loss was diagnosed in 3 out of 8 patients. Delayed graft function was observed in 1 patient. Infectious complications were infrequent. Notably, one allograft was lost beyond year one in a patient with systemic lupus erythematodes and antiphospholipid syndrome due to glomerular thrombi.
Conclusions: Immunoadsorption in combination with anti-CD20 therapy is highly effective for desensitization of living-donor kidney allograft recipients, even in patients with positive CDC crossmatches.

Su681 BONE HEALTH VASCULAR CALCIFICATION AND HYPERPARATHYROIDISM AFTER RENAL TRANSPLANTATION

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Introduction and Aims: The relationship between post transplant hyperparathyroidism, decreased bone mineral density, increased bone turnover and vascular calcification has not been well established.
Methods: We conducted a prospective observational study to quantify these relationships in first renal transplant recipients (eGFR <30ml/min/1.73m^2). Patients underwent DXA scan (Lunar IDXA™) of lumbosacral spine, hips and non-dominant forearm, measurement of intact PTH, 25OH Vitamin D, total and uncuffed ionized serum calcium and urine and serum markers of bone turnover. Patients had a plain radiograph of the L1 to L4 spine to quantify aortic calcification using a standardized Framingham method (Range 0-24). Results expressed as mean (sd).
90 patients were included. Patient characteristics were: Male 61%, age 47 (13) years, time on dialysis 2.6 (1.9) years. Median (Intraquartile range(IQR)) transplant duration was 2.6 (1.0-6.3) years. Mean eGFR was 53.4 (16.2) ml/min/1.73 m^2. 13% of patients were on Vitamin D or calcium supplements. 8 patients had previously experieced a cardiovascular.
Results: Mean T-score at lumbosacral spine, neck of femur and forearm was -0.26 (1.7), -0.8(1.3) and -1.2 (1.4) respectively. 19% of patients had osteoporosis. 12% of patients had experienced a post transplant fracture. The prevalence of aortic calcification was 68%, and in 8.2% it was severe (score $\geq$10). Mean uncorrected serum calcium was 2.57mmol/L (0.16). Median (IQR) iPTH was 98ng/ml (72–399). 78% had iPTH $>$65ng/ml. 8 patients had a prior parathyroidectomy with implant.
iPTH significantly and positively correlated with bone specific alkaline phosphatase (BAP) ($r=0.577$, $p<0.001$), tartrate resistant acid phosphatase 5b (TRACP5b), ($r=0.479$, $p=0.001$), urinary N-Telopeptide ($r=0.635$, $p<0.001$,

Spearman Rank) and ionised calcium (r= 0.461, p=0.001). 12% of patients had a subnormal serum phosphate and there was a significant negative correlation between iPTH and serum phosphate, (r= -0.473, p<0.001, Spearman Rank) 82% of patients were Vitamin D deficient (<50nmol/L) with a mean 25-hydroxyvitamin D of 37 (13) nmol/L.
iPTH negatively correlated with forearm T-score (r= -0.300, p=0.012).
Aortic calcification score negatively correlated with forearm T-score (r= -0.390, p=0.004) and T-score at neck of femur (r= -0.254, p=0.009). There was no significant relationship between lumbosacral spine T-score and aortic calcification score.
Conclusions: Post transplant hyperparathyroidism is common and differs in several important respects to that seen in the CKD and ESKD populations. It is associated with increased markers of bone turnover and decreased bone mineral density, especially at the radius, which consisting predominantly of cortical bone is highly susceptible to the catabolic action of PTH. Vascular calcification, which can be severe, is associated with decreased bone mineral density. Hyperparathyroidism may be an important, though neglected, mediator of long-term vascular and bone health post renal transplantation.

Su682 PHOSPHATE LEVEL, MORTALITY AND TRANSPLANT FAILURE IN RECIPIENTS OF A FIRST DECEASED DONOR RENAL TRANSPLANT

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Introduction and Aims: There is an increased prevalence of cardiovascular disease in renal transplant recipients; traditional risk factors do not explain this adequately. Phosphate has been identified as an independent predictor of mortality in both dialysis patients and renal transplant recipients. Additionally an elevated phosphate has been linked with increased risk of graft dysfunction. This retrospective study looked at renal transplant recipients and at factors which predict death and graft dysfunction.
Methods: The computerised records and case notes of all patients who received a first cadaveric transplant between 01/01/1999 and 31/12/2004 were reviewed. Demographic data including primary renal diagnosis, date of first dialysis and age at transplantation was recorded along with blood pressure, weight, renal function, bone biochemistry and haemoglobin at one year and five years from the date of transplant. Date of return to dialysis and death were also recorded.
Results: 239 patients received a first deceased donor transplant of whom 35% (n=86) were female. 2 patients were transplanted pre-emptively. At the time of transplantation the mean age was 45.3±13.4 years, 17.2% (n=41) were smokers, 10.4% (n=25) were diabetic and 17.6% (n=42) had a history of cardiovascular disease. Median time on dialysis pre transplant was 1006 days (0-5077) and median follow up time was 2749 days (379-3919). At follow up, 207 patients were alive and 32 were dead. Those who died were older at the time of transplantation (p=0.005) and had higher serum creatinine (p=0.001) and phosphate (p=0.023) at 1 year post transplantation. They were also more likely to have returned to dialysis (p=0.004). In Kaplan-Meier analysis, serum phosphate ≥1.12mmol/L at 1 year post transplant was a significant predictor of death (p=0.02) with a difference in survival of approximately 400 days. In Cox regression analysis, adjusting for serum creatinine at 1 year, diabetes, smoking status, presence of cardiovascular disease at transplantation, sex and age, serum phosphate ≥1.12mmol/L remained a significant influence on survival. Patients with diabetes and smokers were significantly more likely to have died during follow up. (Median survival 3650 days v 2798 days and 3635 days v 3339 days respectively). In 40 patients, the transplant failed. 11 of these patients died. Serum phosphate levels both at 1 and 5 years post transplant were significantly higher in the transplant failure group compared with those whose transplant continued to function (1.19±0.06 v 0.96±0.23 (p<0.001) at 1 year and 1.3±0.6 v 0.98±0.25 (p<0.001) at 5 years).
Conclusions: Serum phosphate levels at the upper end of the normal reference range are associated with increased mortality and transplant failure in renal transplant recipients. The effect of phosphate is independent of some of the other traditional cardiovascular risk factors. However, serum phosphate is modifiable with dietary and pharmacological measures and might be an important therapeutic target to help reduce mortality in the renal transplant recipients.

Su683 IS GUM OVERGROWTH A RISK FACTOR OF ATHEROSCLEROSIS IN RENAL TRANSPLANT RECIPIENTS

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Introduction and Aims: Gingival overgrowth (GO) is the main oral manifestation in transplant recipients (TR). The risk factors for GO occurrence are calcineurin inhibitors, bacterial dental plaque, gingival inflammation index, calcium channel blockers, transforming growth factor-beta, HLA-A24, IL-1A polymorphisms and down-regulation of matrix metalloproteinase levels in gingival fibroblast. Interestingly some risk factors for atherosclerosis such as bacterial dental plaque and gingival inflammation are similar with risk factors of GO. So we suggest that there may be a correlation between GO and atherosclerosis.
Methods: In this cross sectional case control study we enrolled 343 renal TRs which received their allograft between 1997-2004. Carotid intimal medial thickness (CIMT) as a marker of atherosclerosis was measured by ultrasonography in TRs by one radiologist and the CIMT 7.5 mm considered positive. All TRs were examined by one dentist and GO scoring determined based on Mc Gaw scoring system. Other demographic and clinical data obtained from medical records and the RTs and entered in the questionnaire. Data was analyzed using chi- square, T test and logistic regression.
Results: Among 343 RTs 57.7% were male, mean age was 40.54 13.08 years. CIMT 7.5 mm was found in 33.8% of RTs. GO was found in 37.6% of RTs. Mean of follow up from transplantation was 74.8867.92 months. Mean duration of dialysis was 17.3916.74 months. Mean body mass index (BMI) was 25.134.74. 74.9% of TRs have hypertension, 31.3% dyslipidemia, 8.7% history of smoking, 3.5% family history of ischemic heart disease, and 51% were diabetic. We found a strong correlation between CIMT and GO (chi- square test, P=0.0001). And also there were correlations between age (P=0.0001), dyslipidemia (P=0.0001), diabetes (P=0.002), BMI (P=006), and dialysis duration (P=0.02) with CIMT. There were correlation between age (P=0.007), using calcium channel blocker (P=0.03) and GO. By logistic regression analysis GO (P=0.0001, OR= 0.19, 95% CI: 0.1-0.35), age (P=0.0001, OR=9.28, 95% CI: 3.97-22.68), sex (P=0.019. OR=0.47, 95%CI: 0.25-0.88), dialysis duration (P=0.02, OR=0.97, 95%CI: 0.96-0.99), dyslipidemia (P=0.007, OR= 0.34, 95%CI: 0.23-0.79), diabetes (p=0.01, OR=0.27, 95%CI: 0.1-0.73) remained correlated with CIMT.
Conclusions: In this study we found a correlation between CIMT and GO. To our knowledge this is the first study. We suggest further studies to clarify this correlation which may use in clinical practice as a risk factor of atherosclerosis.

Su684 LOW THYMOGLOBULIN DOSES OR BASILIXIMAB AS INDUCTION TREATMENT IN OLD DONOR RENAL TRANSPLANTATION

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Introduction and Aims: Transplantation of kidneys from old deceased donors is followed by an increase rate of delayed graft function (DGF) and acute rejection. In these circumstances, induction treatment with antithymocyte globulin or basiliximab could delay the introduction of calcineurin inhibitors with effective prevention of rejection episodes.
Methods: We retrospectively analyzed the efficacy and safety of induction treatment with two low doses of Thymoglobulin (1.25 mg/kg) on days 0 and 2 vs. two doses of basiliximab. We compared a group of 36 patients treated with Thymoglobulin with 68 patients treated with basiliximab, and the mean follow up was 38.4 26.4 months. Immunosuppression included triple therapy with prednisone, tacrolimus and MMF.
Results: Except an older donor in the Thymoglobulin group (68.8 4.7 vs. 66.1 4.5 years, p=0.005), there were no significant differences between both groups, including the recipient age (63.5 5.3 vs. 61.4 6.6 years, p=0.11),

the incidence (33.3 vs. 42.6%, p=0.36) and duration (11.3 9.6 vs. 16 9 days, p=0.13) of DGF. The acute rejection episodes were less frequent in Thymoglobulin group (6.1 vs. 22.1%, p=0.04), observing a trend to a shorter length of stay after transplantation (12.8 6.1 vs. 15.2 5.2 days, p=0.06). We did not observe any differences in graft (p=0.35) and patient (p=0.28) survival at one and at three years, being the serum creatinine and proteinuria similar along the follow up.
There were no differences in the incidence of CMV disease (p=0.11) or admissions due to infections (p=0.36). However, we observed a higher incidence of neoplasia in the Thymoglobulin group (6 patients -20%- including 4 non-melanoma skin cancer, 1 lung and 1 prostate cancer) than in the basiliximab group (2 patients -2.9%- including one non-melanoma skin cancer, one bladder Ca, p=0.004); being the mean time to diagnosis of neoplasia since transplantation shorter in the Thymoglobulin group (14 14 vs. 38 23 months, p=0.13).
Conclusions: In conclusion, in this 'old for old' transplant population both induction treatments were equally effective preserving graft and patient survival but at the expense of an increased acute rejection incidence in the basiliximab group and a higher incidence of neoplasia in the Thymoglobulin group.

Paediatric nephrology

Su685 PLASMA LEVEL OF FIBROBLAST GROWTH FACTOR-23 C-TERMINAL (FGF-23C) AND BONE TURNOVER MARKERS IN CHILDREN WITH CHRONIC KIDNEY DISEASE

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Introduction and Aims: Fibroblast growth factor 23 (FGF-23) is a novel phosphaturic factor produced in bone. FGF-23 circulating levels is markedly elevated in patients with chronic kidney disease (CKD). The aim of the study was to investigate the serum levels of C-terminal FGF-23 (FGF-23c) and bone turnover markers in children with CKD treated and non-treated with rhGH.
Methods: Thirty two children, 7 girls, 25 boys with CKD stages 3-4 and 17 healthy children aged 10.7±5.1 years (group C) were examined. 13 children aged 12.9±2.7 years with creatinine clearance (Ccr) 35±21 mL/min/1.73m^2 were treated with rhGH (group I), 19 aged 10.2±4.8 years with Ccr 43.6±15 mLmin/1.73m^2 did not receive rhGH (group II). In all patients plasma level of FGF-23c, bone alkaline phosphatase (BAP) (by enzyme-linked immunosorbent assay) and serum level of creatinine, calcium (sCa), phosphorus (sP), alkaline phosphatase (AP), and intact PTH (PTH) were evaluated.
Results: In children from group I in comparison with children from group II and C the higher levels of plasma FGF-23c (311±141 vs 156±58 vs 115±67 RU/mL; p<0.03), AP (399±142 vs 237±76 vs 181±92 U/L; p<0.005) and BAP (79±20 vs 46±22 vs 47±26 mg/L; p < 0.005) were found. In children from group I vs group II the tendency to higher level of PTH (154±100 vs 112±114 pg/mL; p=0.053) was found. No significant differences were observed between group I and II in Ccr, sCa, sP and Ca x P.
The FGF-23c correlated positively (Spearman's rank correlation coefficient; p<0.05) with sP (R = 0.52), AP (R = 0.54), BAP (R = 0.5) and PTH (R = 0.38) and negatively with Ccr (R = – 0.53).
Conclusions: The higher plasma level of FGF 23c in children with CKD treated with recombinant human growth hormone may be result of bone turnover stimulation.

Su686 DIAGNOSTIC VALUE OF FGF-23 INTACT AND FGF-23 C-TERMINAL IN CHILDREN WITH CHRONIC KIDNEY DISEASE

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Introduction and Aims: Fibroblast growth factor 23 (FGF-23) is a novel phosphaturic factor produced in bone. FGF-23 may be involved in the pathogenesis of secondary hyperparathyroidism in chronic kidney disease (CKD). The biological activity of FGF-23 requires the C-terminal unique extended structure. Two types of enzyme-linked immunosorbent assays (ELISA) have been developed to detect the intact mature form of FGF-23 and its C-terminal portion.The aim of the study was the simultaneous investigation of plasma levels of FGF-23 C-terminal (FGF-23c) and FGF-23 intact in children with CKD treated and non-treated with recombinant human growth hormone (rhGH).
Methods: Fifty five children, aged 11.3±4.5 years; 38 with CKD stages 2-5 and 17 healthy children aged 10.7±5.1 years (group C) were examined. 14 children aged 13.2±2.8 years were treated with rhGH (group I), 24 aged 10.7±4.7 years did not receive rhGH (group II). In all children FGF-23 levels were determined in plasma using two immunoassays, termed "intact" and "C-terminal" assays. The levels of osteoprotegerin (OPG), receptor activator of nuclear factor-kB ligand (RANKL), bone alkaline phosphatase (BAP), creatinine, urea, albumin, total (TC), HDL-and LDL-cholesterol, triglycerides (TG), calcium (sCa), phosphorus (sP), alkaline phosphatase (AP), intact PTH (PTH) were also evaluated. The correlation between FGF-23 c, FGF-23 intact and other examined parameters were tested.
Results: In all examined children (n=55) the weak correlation (R = 0.29; p<0.05) between FGF-23 c and FGF-23 intact was found. In these children the correlation of FGF-23c with creatinine (R=0.59), urea (R=0.57), TG (R=0.55), AP (R=0.47) and BAP (R=0.33) and negative correlation with creatinine clearance (Clcr) (R = – 0.53) and HDL (R = -0.46) were determined. FGF-23 intact correlated with creatinine (R=0,62), urea (R=0.55), TG (R=0.43) and AP (R=0.3). In healthy children the correlation of FGF 23-c with age (R=0.58) and inverse correlation with OPG (R= – 0.53) were found. The FGF-23 intact in healthy children negatively correlated with AP (R= – 0.6) and BAP (R = – 0.65).
In children with CKD the correlation of FGF-23 intact levels with others examined parameters were not found. In these children correlations of FGF-23c levels with creatninine (R=0.46), urea (R=0.46), TG (R=0.46), AP (R=0.37) and BAP (R=0.38) and inverse correlations with Clcr (R = – 0.53) and HDL (R= – 0.63) were determined.
Conclusions: In children with CKD the FGF-23 c assay is more useful than FGF-23 intact assay.

Su687 BLOOD PRESSURE IS NOT A RELIABLE MARKER OF VOLUME STATUS IN CHILDREN WITH FIRST EPISODE OF NEPHROTIC SYNDROME

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Introduction and Aims: Edema is a common feature of nephrotic syndrome (NS) in children and majority of patients with NS are fluid overloaded. However, younger NS patients may suffer from intravascular hypovolemia despite having edema. Therefore, it may be difficult to recognize intravascular hypovolemia on physical exam. Traditionally, hypotension has been considered as a clinical marker of intravascular hypovolemia.
Aim of our study was to evaluate whether systemic blood pressure (BP) can

be used to differentiate between intravascular hypo- or hypervolemia.
Methods: All newly diagnosed patients with NS from 2004 to 2008 were enrolled. Casual BP was measured in all children at presentation of their NS and converted into Z-scores (SDS) based on age and height related reference values. Urine was tested for sodium (U_{Na}) and potassium (U_K). Intravascular volume status was calculated by the formula proposed by Donckerwolcke and Vande Wale: $U_K/(U_K + U_{Na}) > 0.6$ = hypovolemia; U_K: $(U_K + U_{Na}) < 0.6$ = normo- or hypervolemia.
Results: Thirty seven children were studied, mean (±SD) age at diagnosis was 4.74±0.28 years, 21 children were younger than 4 years and 16 children were older. Hypovolemia was diagnosed in 11 out of 37 (29%) children. In patients <4 years of age, hypovolemia was more frequent (9 out of 21; 43%; RR=1.77, 95% CI 1.075 – 2.923). However, intravascular volume status did not correlate with BP. Mean (±SD) systolic BP SDS was 1.52±0.90 in hypovolemic group and 1.35±1.28 in normo/hypervolemic group (p=0.69). Similarly, the mean diastolic BP SDS was not different between hypo-/hypervolemic groups (0.86±0.70 vs 1.20±0.96; p=0.29). Hypertension (systolic or diastolic BP>95th percentile) was diagnosed in 14 children (38%) but the presence of hypertension was not significantly different between hypovolemic (5/11) and normo/hypervolemic (9/26) groups (p=0.71, RR=0.83, 95%CI=0.45-1.53).
Conclusions: Hypertension is present in 38% children with the first episode of NS, but does not correlate with the volume status. Therefore, blood pressure is not a reliable marker of intravascular hypovolemia/hypervolemia in children with nephrotic syndrome.

Su688 PERITONEAL MEMBRANE ULTRASTRUCTURE IN CHILDREN ON PERITONEAL DIALYSIS

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Introduction and Aims: Glucose-based solutions applied in peritoneal dialysis (PD) might have a deletorious effect on peritoneal membrane, which leads to subsequent complications and ultrafiltration failure.
The study aimed at evaluation of peritoneal ultrastructure in children with chronic kidney disease.
Methods: Twelve biopsy specimens of peritoneum from 6 children on maintenance PD were evaluated. Samples were taken before, 2 and 4 years from PD beginning. In one case the sample was obtained during catheter reimplantation due to prolonged peritonitis. Peritoneal tissue biopsies were fixed in 3% glutaraldehyde solution and subsequently in 1% osmium tetraoxide for further analysis by transmission electron microscopy (TEM). Samples were dehydrated in increasing alcohol concentrations, treated with propylene oxide and embedded in Epon812 mixture. After slicing into semi and ultra thin fragments, uranyle acetate and lead citrate staining was used before TEM examination.
Results: Before PD normal mesothelial cells were present accompanied by few multiform cells. Some of cells had cytoplasm projections (blebs) on their surface. After two years of PD continuation ultrastructural changes of mesolthelial cells were observed (vacuolisation of cytoplasm, mitochondrial swelling with blunting of crest ultrstructure). Total absence of mesothelial cells was seen in some areas of the biopsies. Submesothelial connective tissue contained multiple activated fibroblasts, surrounded by the net of collagen fibres and discrete focal hyalinization areas. After four years of maintenance PD mesothelial cells were totally absent and marked fibrosis of submesothelial tissue was present. In sample taken during the inflammatory process atrophic epithelial cells were shown gradually replaced by homogeneous material of fibrinoid morphology. Photographic documentation of data is provided.
Clinical data of patients including dialysis permeability and adequacy data were analyzed.
Conclusions: Changes in peritoneal membrane are present already in children and they gradually progress with the dialysis duration.

Su689 ERYTHROPOESIS STIMULATING AGENTS IN THE TREATMENT OF ANAEMIA IN CHRONICALLY DIALYSED CHILDREN – A PROSPECTIVE MULTICENTRE ANALYSIS

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Introduction and Aims: Erythropoesis stimulating agents (ESA) are applied as a standard therapy in patients with anaemia in chronic kidney disease in children. The number of trials assessing the quality of this treatment is still not as high as necessary. The aim of our study was to describe the efficacy and details of ESA treatment in population of dialysed children in Poland.
Methods: The study has a prospective observational design and was performed in 11/12 dialysis centres for children in Poland. The analysis started on 1 Nov 2008 and lasted 6 months. We analysed basic clinical and anthropometric data, details of renal replacement therapy with its modality, efficacy (KT/V) and duration, ESA (dose, administration), iron (concentration, storage, supplementation). The target hemoglobin concentration was set to 11-12 g/dl. Detailed analysis of aneamia was performed (hemoglobin, RBC, MCV, MCHC) at 1 month interval according to the local requirements. Additional biochemical parameters were recorded (serum creatinine and urea, calcium, phosphate, parathormon).
Results: The study group comprised 112 dialysed children (DO-69,HD- 43, 54F:58M) of mean age of 146±67 months, mean body weight of 35±18 kg. The duration of dialysis treatment at the start of the study was 27±28 months. Studied patients were treated at least for 3 months with ESA.
Amongst ESA epoietin beta (epoB) was administered most frequently (80%) with mean dose of 115±70 u/kg/week. Darboepoietin (D) was given in 17% of children (dose: 0,6±0,4 mcg/kg/week), CERA and epoietin alpha were administered very rarely (3%). Eighty six patients were supplemented with iron (orally or iv) with mean serum ferritin of 303±381ug/l, TSAT- 32±43% and serum iron of 76±43ug/dl. There were no difference between epoB and D frequency between hemodialysed and peritoneal dialysis children. In HD patients most ESA preparations were given intravenously.
The mean haemoglobin concentration (calculated within 6 months of observation) was 10,9±1.2 g/dl whereas MCHC-33,8±1.7 and MCV 89,8±7,8 fl. 54/112 patients (48%) had adequate HGB concentration. There were no significant difference in anaemia treatment efficacy between HD an PD patients. The initial dose of ESA was stable: epoB rose by 1,92 u/kg/week, whereas D by -0,012 mcg/kg/week. Haemoglobin concentration rose by 0,2±1,6 g/dl in the observation period. Blood transfusions were performed in 16 patients (14%) for 34 times. We estimated the standard blood losses at 16±15 ml/month strictly related to the treatment procedures. The multivariate analysis of factors influencing haemoglobin concentration was performed.
Conclusions: The multicentre analysis of ESA treatment in Poland showed that most of the dialysed children were treated with epoietin beta, despite the treatment modality. Over eighty percent was adequately supplemented with iron. The efficacy of anaemia treatment was average with 52% udertreated subjects, despite quite high mean dose of ESA. Significant number of patients required blood transfusion.

Su690 PREMATURITY, SMALL-FOR-GESTATIONAL-AGE AND PERINATAL PARAMETERS IN CHILDREN WITH CONNATAL, HEREDITARY AND ACQUIRED CHRONIC KIDNEY DISEASE

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Introduction and Aims: Intrauterine growth retardation has been associated with a reduced nephron number,which can contribute to arterial hypertension and cardiovascular morbidity. Recent studies suggest that low birth weight also predisposes to chronic kidney disease (CKD) in later life. There is lack of data as to whether, vice versa, foetal renal impairment may lead to intrauterine growth failure and prematurity. We analysed the prevalence of prematurity, small-for-gestational-age (SGA) and anthropometrical and

clinical birth data of children with different causes and time of onset of CKD.
Methods: We analysed perinatal parameters of 435 children with CKD stages 3-5 of different aetiology and time of onset of CKD. Diseases were classified as connatal with onset of renal disease during foetal life (n=260; 60%), hereditary as genetically determined with onset after 3 months of life (n=93; 21%) and acquired CKD (n=82; 19%).
Results: The rates of prematurity and small-for-gestational-age (SGA) were elevated in children with connatal (39.3% and 29.2%), hereditary (24.7% and 22.6%) and acquired CKD (15.5% and 29.3%). Newborns with connatal CKD had a significantly lower gestational age (37.6±3.04 weeks) than those with hereditary (38.7±2.56 weeks) or acquired (39.0±2.4 weeks) CKD (p=0.001). Birth weight and length were lower in newborns with connatal than in hereditary and acquired diseases (2932±736 g vs 3156±752 g and 3240±634 g; p<0.01); (48.8±4.2 cm vs 50.0±4.2 cm and 50.8±3.8 cm; p<0.01). Head circumference was smaller in connatal than in hereditary and acquired newborn (33.3±2.4 cm vs 34.1±2.1 cm and 34.1±2.6 cm; p<0.05). APGAR scores (p<0.005) were significantly lower in newborns with connatal compared to hereditary or acquired CKD.
Conclusions: Children with connatal CKD had the highest rate of prematurity, a significantly lower birth weight, length, head circumference and APGAR scores than newborns with hereditary or acquired CKD. Irrespective of the aetiology of CKD, all children had a significantly higher rate of SGA and prematurity than the reference population. We conclude that both SGA and prematurity predispose for advanced renal disease in childhood and that foetal kidney disease impairs foetal growth.

Su691 CIRCULATING DENDRITIC CELLS IN PEDIATRIC PATIENTS WITH NEPHROTIC SYNDROME

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Introduction and Aims: Dendritic cells (DCs) represent one of the most extensively studied topics in immunology, because of their central role in the induction and regulation of adaptive immunity, and because of their therapeutic potential for manipulating immune responses.
This study aimed at evaluating circulating DC levels in pediatric patients with idiopathic nephrotic syndrome (NS) and its relation to disease activity in these patients.
Methods: Fifteen nephrotic patients in relapse (proteinuria>40mg/m^2/hour, hypoalbuminemia, and edema) before initiating steroid therapy (Group I), and another15 nephrotic patients in remission after withdrawal of steroid therapy (Group II) were compared to 15 age- and sex-matched healthy children. Besides clinical evaluation and routine laboratory investigations of nephrotic syndrome, circulating DCs were assayed by flowcytometry.
Results: Circulating DC count was lower in nephrotic patients [(48.89±13.52) and (64.64±7.69) $\times 10^6$/liter in both proteinuria & remission groups respectively] than control group (78.54±9.8 $\times 10^6$/liter) with highly significant statistical difference (p<0.001), and lower in proteinuria group than the remission group with highly significant statistical difference (p<0.001). There was a positive correlation between DC count and serum albumin (moderate association) (p=0.002) and a negative correlation between DC count and urine protein/creatinine ratio (strong association) (p=0.001).
Conclusions: Nephrotic syndrome was associated with decreased number of circulating DCs in comparison to control group and the decrease was more apparent in patients with active disease. There was a positive correlation between DC counts and total serum protein, and serum albumin, while there was a negative correlation between DC count and urine protein/creatinine ratio.

Su692 CORRELATION BETWEEN AQUAPORINS mRNA EXPRESSION AND RENAL FUNCTION IN HYDRONEPHROTIC KIDNEY IN CHILDREN

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Introduction and Aims: To investigate the relationship between mRNA expression of aquaporin (AQP)1, 2, 3, and 4 and the renal parenchyma thickness (RPT) and renal function in hydronephrotic kidney (HnK) due to pelvi-ureteral junction obstruction (PUJO) in children.
Methods: The expressions of AQP1, 2, 3 and 4 mRNA in 10 HnK of children (left kidneys, aged 62.3±18.3 months) and 6 control kidney samples from children (aged 62.7±17.1 months) with Wilm's tumor were evaluated by semi-quantitative reverse transcriptase polymerase chain reaction technique. RPT of opposite hilus renalis was measured by ultrasonography preoperatively and was verified at operation. ^{99m} Tc-labeled mercaptoacetyltriglycine (MAG3) scintigraphy was performed in all HnK children. The relationship between AQP1, 2, 3, and 4 mRNA expressions, RPT and renal function was analyzed by Pearson correlation.
Results: The AQP1, 2, 3 and 4/beta-actin ratio in the HnK and normal kidney were AQP1: 0.40±0.04 VS 0.74±0.09; AQP2: 0.48±0.06 VS 0.84±0.08; AQP3: 0.47±0.07 VS 0.82±0.08; AQP4: 0.55±0.07 VS 0.82±0.06 respectively, and the differences were significant (P<0.01). RPT measured preoperative by ultrasonography is averaged 4.89±0.93mm. Significant correlation was found between the levels of AQP1, 2, 3 and 4 mRNA and hydronephrotic RPT (r=0.743, 0.743, 0.723, 0.706, respectively; P<0.05). Expression of AQP1, 2, 3 and 4 mRNA was highly correlated with preoperative renal function (r=0.963, 0.944, 0.967, 0.960, respectively; P<0.01).
Conclusions: The xpression of AQP1, 2, 3 and 4 was correlated well with atrophy of kidney and renal function in HnK. This is helpful for further understanding the mechanism why the thinner RPT, the weaker concentration and dilution function of the kidney.

Su693 EFFICIENCY OF MYCOPHENOLATE MOFETIL IN CHILDHOOD STEROID RESISTANT NEPHROTIC SYNDROME

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Introduction and Aims: The current therapeutic approach of children with steroid-resistant nephrotic syndrome (SR NS) is still discussed. Here we present our experience on the effectiveness of Mycophenolate mofetil (MMF) in children with SR NS.
Methods: From the years 2004 to 2009 we used MMF treatment in 32 children with SR NS (age: 1-16 years). The mean duration of disease was 29±43 months. All patients were treated before by steroids and cytostatic agents, and the multi-drug resistance was found. MMF was administered at a daily dose of 500 mg/m^2/day, in two divided doses, during 12 months. Previously all patients received prednisolone (1,5-2,0 mg/kg/day) during 6 weeks, and first 6 months of MMF treatment was combined with alternate-day supporting dose of prednisolone (30-50% of initial dose with tapered on 2,5 mg every 6-8 weeks). Laboratory tests and renal biopsy were used. Follow up period after MMF was 24 months.
Results: Renal biopsy was performed in 14/43,8% cases, and demonstrated focal segmental glomerulosclerosis (9), mesangial glomerulonephritis (3), minimal change disease (1), membranous nephropathy (1). The partial remission was observed in 12/37,5% on 12 months, and remission was seen in 18/56,2% on 36 months, being 40,6% complete and 15,6% partial. The Chronic Renal Failure were developed in 11/34,4% during follow-up period (associated with genetic abnormalities or early age onset, or in adolescents). The adverse effects observed were hematological changes

(mild anemia or leucopenia, 10/31,3% and 4/12,5%, respectively), transient hepatotoxicity (7/21,9%), viral upper respiratory tract infections (5/15,6%), intestinal syndrome (2/6,3%).
Conclusions: Data from this study suggested that MMF in the dose 500-600 mg/m^2/day is an actual option as a second-line therapy in children with SR NS, resulted in remission in 56,2% cases after 36 months of follow up.

Su694 THE POSTNATAL FOLLOW-UP OF INFANTS WITH ANTENATALLY DETECTED HYDRONEPHROSIS: THE IMPORTANCE OF EARLY CIRCUMCISION

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Introduction and Aims: With the wide use of prenatal ultrasonography (USG) and increasing recognition of antenatal hydronephrosis (AH), the postnatal follow-up of these patients is of greater importance. The objective of this study was to identify the frequencies urinary tract infection (UTIs) and renal parenchymal damage as well as growth and nutrition of infants with antenatal hydronephrosis. Moreover, we aimed to determine the effect of circumcision on the frequency of UTIs in these babies.
Methods: Infants with a fetal renal pelvis diameter of 5 mm or more identified with antenatal USG were followed-up during the postnatal period. On admission, urinalysis and urine culture tests were performed and body weights and lengths were measured; tests and measurements were repeated monthly. Other investigations included USG, voiding cystourethrography and scintigraphic studies as well as MR urography and intravenous pyelography wherever indicated. All patients were evaluated in terms of UTI frequency, pelvic diameter, scarring on DMSA, differential function on DTPA. Growth (height SDS) and nutrition [relative weight (RW)] parameters were recorded as well. All male infants were recommended to be circumcised and these parameters were assessed after circumcision. Statistical evaluation was made by using khi-square test made.
Results: The study included 178 patients (234 renal units). Of these, 29 were diagnosed by VUR, 87 by obstructive uropathy and 54 by non-obstructive uropathy. The mean follow-up duration was 45±24.9 months. Of 133 males, 111 infants were circumcised. The pre-circumcision UTI frequency [(2.97±1.14/year(y)] was significantly higher than post-circumcision UTI frequency (0.25±0.67/y). Also, pre-circumcision UTI frequency (2.97±1.14/y) was significantly higher than the frequencies observed in female cases (0.85±0.91/y), in males who did not undergo circumcision (0.91±0.99/y) and in overall study group (0.73±0.79/y). In obstructive uropathy patients, the rate of renal damage was increased in uncircumcised patients (28.5% vs 35.7%) whereas no change was observed in circumcised patients (11.2% vs 12.5%). Overall growth (for height SDS, $p<0.05$) and nutrition (for RW, $p>0.05$) parameters gradually improved after circumcision.
Conclusions: In conclusion, close follow-up and early circumcision of infants with AH will prevent UTIs and renal parenchymal damage in kidneys enabling normal growth and nutrition.

Su695 POSTNATAL EVALUATION OF INFANTS WITH VESICOURETERAL REFLUX DETECTED BY ANTENATAL ULTRASONOGRAPHY

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Introduction and Aims: The aim of this study is to demonstrate the importance of the postnatal investigation and treatment in infants with Vesico ureteral reflux(VUR) diagnosed by antenatal ultrasonography(US).
Methods: Infants whose antenatal US showed a fetal renal pelvis of 5 mm or greater were investigated. All cases with VUR were evaluated in terms of the frequency of urinary tract infections (UTIs), scars on DMSA, growth [Height Z score (HZ), Weight Z score (WZ)] and nutrition status [Weight height index (WHI)]. Statistical evaluation was made by using khi-square test.
Results: Of the 277 neonates, 36 infants [56 renal units (RU)] were detected to have VUR. Of these 36 patients, 25 (69.4%) were male and 11 (30.6%) were female. Of the 56 RU, severe VUR, defined as ≥grade III, was found in 24 (45.5%). The mean duration of postnatal follow-up was 37.8±24.50 months. The annual UTI frequency was found to be 1.25±0.83 episode/year. Of these 36 infants, 22 (61.1%) were recovered from VUR with medical (8 patients, 22.2%) and surgical (14 patients, 38.9%) treatments. In 16 patients (44.4%), baseline DMSA showed parenchymal defects. Six of these 16 patients, DMSA findings were recovered on final DMSA. Although statistically insignificant, Initial growth and nutritional values (mean WHI, HZ and WZ scores: 98.19±8.81, -0.17±0.86 and 0.00±0.14, respectively) gradually improved (101.97±14.85, 0.05±1.06 and 0.06±1.071, respectively), ($P>0.05$).
Conclusions: In conclusion, postnatal early management of infants with VUR detected by antenatal US seems to prevent frequent UTIs, renal scarring and the nutritional disturbances enabling normal growth.

Su696 PROHEPCIDIN IN PEDIATRIC CHRONIC KIDNEY DISEASE: RELATION TO ERYTHROPOIETIN HYPORESPONSE

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Introduction and Aims: The introduction of erythropoiesis-stimulating agents (ESA) has led to a significant reduction in anemia and improved patients' quality of life. However, there is marked variability in the sensitivity to rHuEPO, with up to 10-fold variability in dose requirements to achieve correction of anemia. ESA resistance is associated with several conditions including iron deficiency, oxidative stress, infection, inflammation, hyperparathyroidism, aluminum toxicity and vitamin B12 and folate deficiency. Prohepcidin is a newly detected protein that plays an important role in iron metabolism and the resistance to ESA. Circulating hepcidin appears to bind to ferroportin, the ferrous iron trans-membrane transporter, on the cell surface leading to the trapping of iron at absorptive enterocytes, macrophages and hepatocytes, thus causing a decrease in available serum iron.
This study aims at investigating the relationship between prohepcidin and iron status in CKD pediatric patients, as well as its association with response to ESA therapy.
Methods: A total of 57 pediatric subjects with CKD in different stages were included in the study. The patient group was divided into group I consisting of 42 patients on regular hemodialysis (HD) [mean age 14±2.93 years], Group II included 15 patients on conservative treatment [mean age 7.47± 4.82]. Control group III included 18 healthy subjects of matched age and gender. Patients of group I were further subdivided into responders (subgroup IA) and hypo-responders (subgroup IB) to ESA therapy. Following clinical evaluation, serum prohepcidin, iron, total iron binding capacity (TIBC) and transferrin saturation (TSAT) were measured in all studied subjects.
Results: The study showed significantly lower values of serum iron, TIBC and TSAT in both patient groups compared to control while no difference could be found when comparing HD to conservative patients. Serum prohepcidin showed significantly higher values both in HD and conservative patients when compared to controls ($p<0.001$ and $p<0.05$ respectively). Moreover, significantly higher values were found in HD compared to conservative patients ($p<0.001$). Serum iron showed higher values among the responsive subgroup but with no statistical difference compared to the hyporesponsive subgroup ($p>0.05$). Both serum prohepcidin and TSAT showed significantly higher values among responsive patients in comparison to the hyporesponsive ones ($p<0.05$ and $p<0.001$ respectively). Correlation study of serum prohepcidin versus various parameters in the HD group (group I) showed a significant positive correlation with the duration of dialysis, hemoglobin, hematocrit and TSAT. While, negative correlations could be elicited with rHuEPO dose, rHuEPO/Hb ration and IV iron dose.
Conclusions: Serum prohepcidin rises progressively in pediatric patients with deterioration of their kidney function and can be used in assessment of their iron status and in monitoring their response to ESA therapy.

Su697 ADIPONECTIN, DYSLIPIDEMIA AND CARDIOVASCULAR EVENTS AMONG PEDIATRIC PATIENTS WITH CHRONIC KIDNEY DISEASE

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Introduction and Aims: Cardiovascular complications are the leading cause of mortality in patients with chronic kidney disease (CKD). This high risk results from the additive effects of traditional risk factors as hypertension, dyslipidemia and uremia-related risk factors (hyperphosphatemia, hyperhomocysteinemia and chronic inflammation). Adiponectin is a novel adipocytokine secreted exclusively by the adipocytes with a cardiovascular protective properties as it has anti-inflammatory, antiatherogenic and lipid lowering properties in the general population.
The study aimed at evaluating the adiponectin and lipid profile in pediatric patients with CKD and their possible relationships to renal replacement therapy and cardiovascular risk.
Methods: This case-control study was conducted on pediatric patients with CKD including 20 patients on regular hemodialysis (group I), 15 patients with CKD on conservative treatment (group II). They were compared to 20 apparently healthy children (group III). Lipid profile (comprising total serum cholesterol, serum triglycerides, HDL-c, LDL-c, apolipoprotein B, and calculation of coronary artery disease risk index) and fasting serum adiponectin were assayed in all subjects. For group I, adiponectin was assayed before and after one hemodialysis session. We also investigated the cardiovascular system by chest roentgenogram, electrocardiogram and echocardiography. Stress ECG and Tcm99 sestamibi myocardial perfusion imaging were done for patients who had typical anginal chest pain.
Results: We found that CKD patients had an atherogenic lipid profile which was more evident in the dialysis patients. Apolipoprotein B was higher among patients with cardiovascular morbidities. We also found that adiponectin level increased among CKD patients, and its level was higher among patients with cardiovascular morbidities.
Conclusions: High adiponectin level and atherogenic lipid profile (notably elevated apolipoprotein B) are associated with adverse cardiovascular outcomes in pediatric patients with CKD.

Su698 SKIN AUTOFLUORESCENCE (sAF) AS A NEW NONINVASIVE MARKER OF VASCULAR DAMAGE IN CHILDREN WITH CHRONIC KIDNEY DISEASE (CKD) ON CONSERVATIVE THERAPY

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Introduction and Aims: Advanced glycation endproducts (AGEs) may contribute to inflammatory processes, endothelial dysfunction and vascular damage resulting in the development and progression of atherosclerosis in CKD population. It has been shown that AF is strongly associated with arterial stiffness and mortality in hemodialyzed adults, independently of traditional risk factors. The aim of the study was to determine for the first time in children, whether sAF is increased and related to novel markers of endothelial dysfunction, being risk factors for atherosclerosis.
Methods: 36 children (17 boys and 19 girls), mean age 14,9±3,5 years with CKD (stage 2-4) were included in the study and compared to 26 healthy age-matched subjects. We evaluated sAF using AGE-Reader. Serum sE-selectin, ADMA and SDMA were measured by ELISA method.
Results: sAF, serum sE-selectin, ADMA, and SDMA were significantly higher in patients vs.controls ($p<0.001$). A significant positive correlation was shown between sAF and sE-selectin ($p<0,01$), ADMA ($p<0,03$). Additionally, sAF was strongly and negatively associated with GFR ($p<0,0003$). In the multiple regression analysis sE-selectin was an independent predictor of the sAF ($p<0,006$).
Conclusions: In CKD children the accumulation of tissue AGEs occur and is related to the progression of the disease. A significant association between sAF and investigated markers of endothelial inflammation/dysfunction supports a role for AGEs as a contributor to vascular damage and shows that sAF is a good, rapid tool for assessing the risk of atherosclerosis development in children with mild and moderate CKD.

Su699 EVALUATION OF PUBERTY AMONG ADOLESCENT PATIENTS WITH NEPHROTIC SYNDROME

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Introduction and Aims: The association between chronic renal diseases, undergrowth and pubertal delay is well known and occurs in glomerular alterations as well as in interstitial and tubular disorders. Steroid-dependent, frequent relapsers and steroid-resistant nephrotic patients all have a chronic form of the disease and need long term treatment with corticosteroids and may need additional treatment with cyclophosphamide, chlorambucil, azathioprine, high dose pulse methyl prednisolone or cyclosporine A.
This study aimed at analyzing the impact of long term nephrotic syndrome (NS) as well as the effect of different therapeutic modalities on linear growth and pubertal development in patients with childhood NS.
Methods: The study was conducted at Ain Shams Pediatric Nephrology Clinic, Children's Hospital, Ain Shams University. It comprised 30 patients between 14-18 years of age with NS of more than 2 years duration studied during remission. All patients were subjected to full history taking, thorough clinical examination, accurate determination of height. Growth and puberty were assessed by height standard deviation score (SDS) and Tanner staging respectively. Serum LH, FSH and free testosterone/estradiol (basal and after provocation) were measured only in patients with clinical delayed puberty. Lastly, X-ray on left hand and wrist was done for determination of bone age.
Results: Growth retardation (height <-2SDS) was present in 50% of patients, more in steroid-resistant NS than in steroid-dependent patients. No significant differences were observed between studied males (23) and females (17) except that females had significantly higher bone age SDS. As for puberty, 20 patients had clinical delayed puberty (66.7%). The cumulative dose of levamisole was significantly higher in patients with normal compared to those with delayed puberty. Ten patients with delayed puberty (50%) showed positive response to provocation test. Evaluation of growth and puberty together revealed that 30% of patients were stunted with delayed puberty, 36.7% were normal in height with delayed puberty, 20% were normal in puberty with short stature and only 13.3% were normally growing with normal puberty.
Conclusions: Delayed puberty is a reversible state in most of cases due to transient disturbance in hypothalamo-pituitary axis. Follow up of pubertal development is mandatory to detect patients who may need hormonal therapy. Growth retardation should be detected early to give growth hormone until patients reach target height before starting hormonal therapy for promoting puberty. It is better to keep patients of NS near puberty in full remission and avoid the use of cytotoxic drugs.

Su700 MIGRATION BACKGROUND AND PATIENT SATISFACTION IN A LARGE CITY PEDIATRIC NEPHROLOGY OUTPATIENT CLINIC

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Introduction and Aims: The socioeconomic and public health-related implications of migration are increasingly recognized as having strong effects on society as a whole and the medical profession in particular. Data on health-related issues in children and adolescents with a migration background are yet scarce but needed for future public health planning.
To obtain data on migration background, family social and financial status, education, and patient satisfaction of children and their families treated by our institution.
Methods: In a cross sectional survey of patients seen during 2008 in the Pediatric Nephrology Outpatient Department at the Charité University

Children's Hospital in Berlin, a total of 348 families answered a prepared questionaire. The final data set contained basic information, a standardized patient satisfaction score (derived from a customer satisfaction score used by industry and modified for health providers; Schmidt J et al, 1989), a subjective categorical rating of disease severity and satisfaction with treatment, and a subjective categorical external evaluation (by doctors and nurses) of patient compliance.

Results: A migration background was present in 131 patients (38%). These families were of 20 different nationalities, spoke a total of 22 different native languages and were mainly coming from Turkey, the former Yugoslavia, Russia, Poland, Vietnam, Irak, and the Lebanon.

Patient satisfaction (on a scale from 8-40) was significantly higher in families without (32.9 + 4.6) than in those with a migration backgroud (30.8 + 4.7; $p<0.0001$). In contrast, patient satisfaction was not significantly associated with income, education (school level achieved by parents), and religious background. Patient satisfaction was not significantly associated with subjective ratings of disease severity or friendliness of nurses or doctors, but showed a significant correlation with trust in doctors ($p<0.0001$). There was no apparent correlation of patient satisfaction with compliance.

Conclusions: A migration background is present in more than a third of our outpatient population; it has significant effects on patient satisfaction.