ORIGINAL ARTICLE—STUDENT AWARD WINNER 2015*

# T helper cell-related cytokine gene polymorphisms and vitamin D pathway gene polymorphisms as predictors of survival probability in patients on renal replacement therapy

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**KEY WORDS**

gene polymorphisms, renal replacement therapy, survival probability, T helper-cell cytokines, vitamin D

**ABSTRACT**

**INTRODUCTION** Negative outcomes in patients on renal replacement therapy (RRT) may have a source in T helper (Th)-cell imbalance or vitamin D deficiency.

**OBJECTIVES** We examined the association of genes encoding cytokines related to Th1 and Th2 cells and vitamin D pathway genes with survival probability of patients on RRT.

**PATIENTS AND METHODS** The study included 1253 patients on hemodialysis. *IL13*, *IL4R*, *IL18, IL12A*, *IL12B*, *IL28B, MCP1*, *GC*, *VDR,* and *RXRA* gene polymorphisms were tested. The Kaplan–Meier method with the log-rank test was used to estimate significance of survival probabilities.

**RESULTS** Patients carrying the *IL13* minor T allele had an increased risk of death compared with CC subjects (log-rank test, $P = 0.005$; hazard ratio [HR], 1.40; 95% confidence interval [CI], 1.11–1.76; $P = 0.005$). *IL28B* rs8099917 GG patients had higher mortality rates compared with *IL28B* GT+TT carriers (log-rank test, $P = 0.04$; HR, 1.82; 95% CI, 1.10–3.01; $P = 0.02$). *IL28B* rs12979860 TT carriers had an increased risk of death compared with CC+CT carriers (log-rank test, $P = 0.02$; HR, 1.62; 95% CI, 1.09–2.42; $P = 0.02$) only when they were negative for hepatitis B virus (HBV) or hepatitis C virus (HCV) infection. The prevalence of coronary artery disease differed significantly among patients with the *IL28B* rs12979860 TT genotype compared with CC+CT carriers (odds ratio, 1.87; 95% CI, 1.14–3.09; $P = 0.01$). There was no association between the *GC*, *VDR*, and *RXRA* nucleotide variants and survival.

**CONCLUSIONS** The *IL13* rs20541 T allele and *IL28B* rs8099917 GG genotype are negative predictors of survival in patients on RRT, while the *IL28B* rs12979860 TT genotype increases the risk of death only in patients negative for HBV or HCV infection.

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**Received:** March 25, 2015.
**Accepted:** April 25, 2015.
**Published online:** June 3, 2015.
**Conflict of interest:** none declared.
Pol Arch Med Wewn. 2015; 125 (7-8): 511-520
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* The authors won the first award of the Editor-in-Chief for the best student paper in 2015. For more information, go to www.pamw.pl.

**INTRODUCTION** Adjusted rates of all-cause mortality are 6.5- to 7.9-times greater for dialysis patients than for the general population.[1] Successful renal transplantation improves survival.[1] Cardiovascular diseases, infections, and cancers are the main diagnosed causes of death. Fatal outcomes may have a source in T helper (Th) -cell imbalance or vitamin D deficiency which are among the most prevalent abnormalities in end-stage renal disease (ESRD).

Cytokines such as interleukin (IL) 18, IL-12, and IL-28B are, among others, associated with the Th1 pathway, while IL-4, IL-13, and monocyte chemoattractant protein 1 shift the balance towards Th2. Aberrant Th-cell responses lead to diseases. Th1-cell domination was shown to be

related to type 1 and 2 diabetes,[2-4] diabetic nephropathy,[4] lupus nephritis,[5] atherosclerosis,[6] and graft-versus-host disease,[7] whereas Th2 supremacy was shown in infections,[2,8] allergy,[9] asthma,[3] and cancers.[2,10] The involvement of Th1/Th2 cells in hepatitis C virus (HCV) infection seems to depend on clinical manifestation of this infection.[11-13] Patients with ESRD have altered Th1/Th2 balance; however, it is disputable which pattern dominates in patients on hemodialysis (HD).[14-16] Vitamin D deficiency is epidemiologically linked to life-threatening diseases such as myocardial infarction (MI),[17,18] cerebral stroke,[18] cancer,[19] and bone fractures.[20] Low levels of serum vitamin D may be associated with atherosclerosis.[21] In patients on dialysis, the above conditions are more frequent than in the general population.

There is a link between vitamin D and T-cell functional balance: an active form of the vitamin, $1,25(OH)_2D$, has an inhibitory effect on the Th17 and Th1 response,[22,23] causing a shift toward the Th2 profile. In stimulated peripheral blood mononuclear cells, $1,25(OH)_2D$ caused suppression of the IL-2 production.[24] However, in peripheral blood mononuclear cells isolated from HD patients showing lower IL-2 production compared with healthy controls, vitamin D restored IL-2 production.[25] IL-2, IL-10, and IL-12B were shown to be under direct transcriptional regulation by $1,25(OH)_2D$.[26] Additionally, $1,25(OH)_2D$ increased the low percentage of IL-13-producing $CD4^+$ and $CD8^+$ T cells,[27] upregulated IL-4 receptor densities on a murine osteoblast cell line,[28] and suppressed the IL-18 mRNA expression.[29] Moreover, *VDR* rs2228570 influences the IL-12 expression.[30] However, the above effects do not always result in differences in circulating cytokines in in-vivo studies in which vitamin D is given to patients.[31] Deviations from the T-cell cytokine balance[32-34] and low plasma vitamin D concentrations[35-37] are related to cardiovascular events[34,37] as well as altered immunocompetence during infections[32,35] and vaccinations.[33,36] Serum parathyroid hormone (PTH) levels are dependent on serum vitamin D concentrations,[38] and T cells are implicated in the mechanism of the PTH action in the bone.[39]

We aimed to check a frequency distribution of polymorphic variants of the genes encoding cytokines/ILs related to Th1 and Th2 cells (*IL18* rs360719, *IL13* rs20541, *IL4R* rs1805015, *IL12A* rs568408, *IL12B* rs3212227, *IL28B* rs8099917, *IL28B* rs12979860, and *MCP1* rs1024611) as well as polymorphisms of the vitamin D pathway genes (the vitamin D-binding protein gene, also referred to as the group-specific component gene: *GC* rs2298849, *GC* rs1155563, *GC* rs7041; vitamin D receptor gene: *VDR* rs2228570, *VDR* rs1544410; and retinoid X receptor alpha gene: *RXRA* rs10 776 909, *RXRA* rs10881578, and *RXRA* rs749759) in relation to survival probability of patients with ESRD requiring renal replacement therapy (RRT).

## PATIENTS AND METHODS

### Patients and controls

The enrollment of HD patients to the study started in January 2009 and was completed in May 2014. Twenty-one dialysis centers participated in the study. Demographic data (sex, metrical age), clinical data (causes of ESRD, comorbidities, age at the start of RRT, length of RRT, frequency of parathyroidectomy and treatment with cinacalcet, response rate to hepatitis B vaccination, prevalence of HBV/HCV infections, frequency of renal transplantation, causes of death), laboratory parameters (liver enzymes, calcium–phosphorus balance parameters, HBV/HCV seromarkers), and blood samples for genotyping were collected and updated every year (the last revision was done in August – September, 2014). The adherence of dialysis physicians to collecting specific patient data varied from 100% to 65% for γ-glutamyltranspeptidase (GGT), which is not obligatorily tested in dialysis facilities.

Controls (n = 378) were recruited from blood donors and healthy volunteers unrelated to patients and to one another. They lived in the same geographical region as patients.

### Genotyping

*IL13*, *IL4R*, *IL12A*, *IL28B*, *GC* rs2298849, *GC* rs1155563, *RXRA* rs10776909, and *RXRA* rs10881578 polymorphisms were genotyped using a high-resolution melting curve analysis. Polymerase chain reaction–restriction fragment length polymorphism was used for *IL18*, *IL12B*, *MCP1*, *GC* rs7041, *VDR* rs2228570, *VDR* rs1544410, and *RXRA* rs749759 genotyping. For quality control, approximately 10% of the randomly chosen samples were regenotyped. Samples with ambiguous results were excluded from further statistical analyses.

### Statistical analysis

The results were presented as percentage for categorical variables, as mean with 1 standard deviation for normally distributed continuous variables, or as median with range for not normally distributed continuous variables as tested by the Shapiro–Wilk test.

Survival probability since the start of RRT was analyzed with respect to the tested polymorphic variants using dominant, recessive, and additive models of inheritance. The Kaplan–Meier method with the subsequent log-rank test was used to estimate significance of differences in cumulative proportion surviving curves characterizing the genotype groups in each model of inheritance. Hazard ratios (HRs) and 95% confidence intervals (CIs) were calculated using a Cox proportional hazard model to show the effect of the genotype on the risk of death (all-cause mortality). The Cox model was applied to show the significance of polymorphic variants in the prediction of survival among the collected demographic, clinical, and laboratory data. If an association between the tested polymorphic variants and survival probability was significant (*P* values for both HR and the log-rank test was less than 0.05), we tried to establish which phenotype was associated with

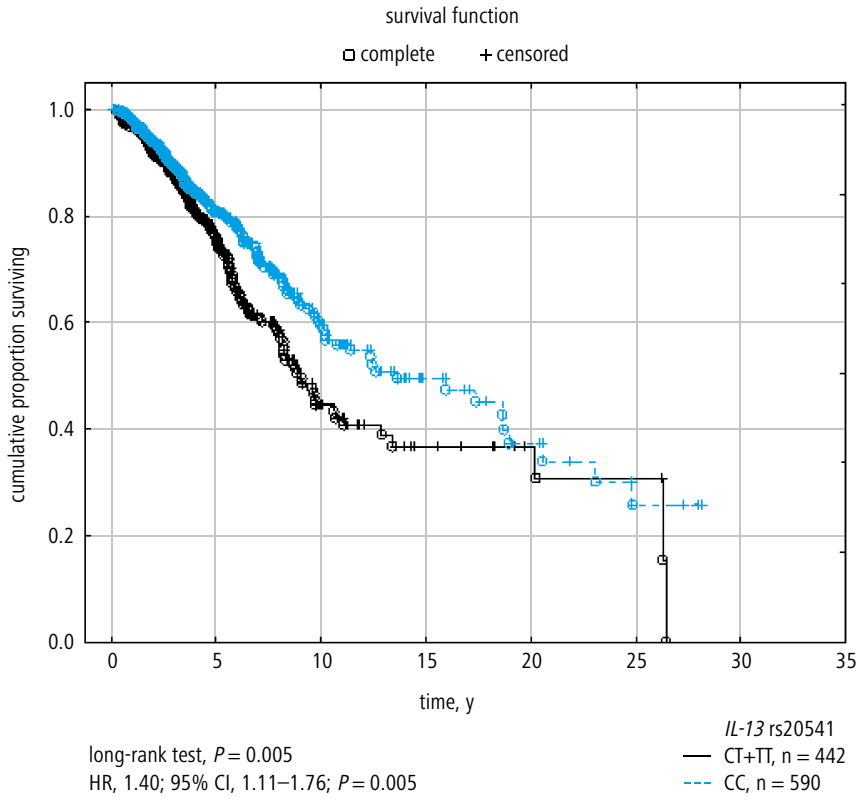



**FIGURE 1** Survival probability of hemodialysis patients with respect to *IL13* rs20541 polymorphic variants
Abbreviations: HR, hazard ratio

this specific gene calculating odds ratios (ORs) with 95% CIs. All probabilities were 2-tailed. A *P* value of less than 0.05 was considered significant. The Bonferroni correction for multiple comparisons was used, where appropriate.

The survival analysis was performed in the whole groups with determined genotypes and also in the groups divided according to demographic/clinical parameters, where reasonable.

A statistical analysis was performed using Graph-Pad InStat 3.10, 32 bit for Windows, created July 9, 2009 (GraphPad Software, Inc., San Diego, California, United States), CytelStudio version 10.0, created January 16, 2013 (CytelStudio Software Corporation, Cambridge, Massachusetts, United States), and Statistica version 10, 2011 (Stat Soft, Inc., Tulsa, Oklahoma, United States).

**Ethical approval** The study design was approved by the Institutional Review Board of the Poznan University of Medical Sciences, Poznań, Poland. Informed consent was obtained from all study participants.

## RESULTS

**Patient characteristics** The study group included 1253 patients on HD (men, 55.9%; mean age at the start of RRT, 59.0 ±16.2 years; RRT duration, 4.31 years [0.07–29.0]). The main causes of ESRD were diabetic nephropathy (n = 366), hypertensive nephropathy (n = 231), chronic glomerulonephritis (n = 179), and chronic tubulointerstitial nephritis (n = 118). Coronary artery disease (CAD) was diagnosed in 442 patients, and MI occurred in 243 patients. The response rate to hepatitis B vaccination (anti-HBs ≥10 IU/l) was 76%. Among the recruited HD patients, there were 180 individuals who had undergone renal transplantation (74 returned to HD treatment after irreversible graft failure). There were 437 deaths during the study period. Two individuals died in accidents, other causes of death were clear in 386 cases. Four main causes of death included chronic or acute cardiac diseases (45.9%), cerebral stroke (15.0%), sepsis or pneumonia (10.9%), and cancer (10.6%).

Survival probability since the start of RRT in this group was negatively associated with age at the start of RRT ($P < 0.0001$), presence of diabetic nephropathy ($P < 0.0001$), presence of CAD ($P < 0.0001$), history of MI ($P < 0.0001$), and serum GGT activity ($P = 0.02$). Chronic glomerulonephritis as the cause of ESRD ($P < 0.0001$), a history of renal transplantation ($P < 0.0001$), and development of anti-HB antibodies in response to hepatitis B vaccination or HBV infection ($P = 0.02$) increased the survival probability.

**Genotype frequencies in patients and controls** A frequency distribution of the tested single nucleotide polymorphisms (SNPs) did not differ between the experimental and control groups ($P_{trend} > 0.05$).

**Th cell-related cytokine gene polymorphisms as predictors of survival probability** The only polymorphisms of Th cell-related cytokine genes associated with survival probability of the examined patients were those of *IL13* and *IL28B*.

Patients carrying the *IL13* minor T allele (CT and TT genotypes) had an increased risk of death since the start of RRT compared with *IL13* CC patients (FIGURE 1). The *IL13* T allele was an independent predictor of death since the start of RRT (HR, 1.28; CI, 1.01–1.63; $P = 0.04$) also among other variables: older age at the start of RRT (HR, 1.03; 95% CI, 1.02–1.04; $P < 0.0001$), CAD

**TABLE 1** Demographic and clinical data of hemodialysis patients divided according to *IL13* polymorphic variants

| Parameter | *IL13* rs20541 | | | Odds ratio (95% CI) | *P* value |
|---|---|---|---|---|---|
| Demographic data obtained from 1032 patients | CC n = 590 | CT n = 379 | TT n = 63 | | |
| diabetic nephropathy | 168 (28.5) | 114 (30.1) | 21 (33.3) | TT + CT vs CC: 1.105 (0.835–1.460) | 0.5 |
| | | | | TT vs CC + CT: 1.218 (0.672–2.147) | 0.6 |
| | | | | TT vs CC: 1.256 (0.685–2.244) | 0.5 |
| hypertensive nephropathy | 100 (16.9) | 78 (20.6) | 7 (11.1) | TT + CT vs CC: 1.167 (0.836–1.626) | 0.4 |
| | | | | TT vs CT + CC: 0.555 (0.210–1.250)) | 0.2 |
| | | | | TT vs CC: 0.613 (0.229–1.402) | 0.3 |
| chronic glomerulonephritis | 88 (14.9) | 51 (13.5) | 11 (17.5) | TT + CT vs CC: 0.931 (0.643–1.341) | 0.8 |
| | | | | TT vs CC + CT: 1.263 (0.580–2.526) | 0.6 |
| | | | | TT vs CC: 1.207 (0.546–2.457) | 0.7 |
| chronic tubulointerstitial nephritis | 54 (9.2) | 38 (10.0) | 10 (15.9) | TT + CT vs CC: 1.209 (0.784–1.860) | 0.4 |
| | | | | TT vs CC + CT: 1.799 (0.788–3.726) | 0.2 |
| | | | | TT vs CC: 1.873 (0.802–3.994) | 0.2 |
| age at the start of RRT, y | 60.2 (11.8–90.8) | 61.4 (11.2–91.1) | 58.4 (14.1–82.7) | TT + CT vs CC: | 0.04[a] |
| | | | | TT vs CC + CT: | 0.5 |
| | | | | TT vs CC: | 0.08 |
| clinical data obtained from 947 patients | CC n = 548 | CT n = 341 | TT n = 58 | | |
| coronary artery disease | 196 (35.8) | 148 (43.4) | 19 (32.8) | TT + CT vs CC: 1.293 (0.983–1.699) | 0.07 |
| | | | | TT vs CC + CT: 0.772 (0.414–1.395) | 0.4 |
| | | | | TT vs CC: 0.875 (0.464–1.601) | 0.8 |
| myocardial infarction | 111 (20.3) | 86 (25.2) | 8 (13.8) | TT + CT vs CC: 1.213 (0.877–1.676) | 0.3 |
| | | | | TT vs CC + CT: 0.562 (0.226–1.222) | 0.2 |
| | | | | TT vs CC: 0.630 (0.251–1.391) | 0.3 |
| parathyroidectomy/cinacalcet | 18 (3.1) | 9 (2.4) | 2 (3.2) | TT + CT vs CC: 0.835 (0352–1.891) | 0.8 |
| | | | | TT vs CC + CT: 1.140 (0.128–4.736) | 1.0 |
| | | | | TT vs CC: 1.052 (0.115–4.580) | 1.0 |

Data are presented as number (percentage) of patients or median and range.

a not significant after the Bonferroni correction ($P > 0.017$)

Abbreviations: RRT, renal replacement therapy; others, see FIGURE 1

(HR, 1.86; 95% CI, 1.44–2.40; $P < 0.0001$), development of anti-HB antibodies in response to hepatitis B vaccination or HBV infection (HR, 0.72; 95% CI, 0.55–0.95; $P = 0.02$), chronic glomerulonephritis (HR, 0.67; 95% CI, 0.44–1.00; $P = 0.05$), and diabetic nephropathy (HR, 1.22; 95% CI, 0.93–1.59; $P = 0.2$). A $P$ value for this Cox model was less than 0.00001. No significant associations between *IL13* polymorphisms and patient data were shown (TABLE 1). The negative association of the T allele with survival probability was more pronounced in men (log-rank test, $P = 0.03$; HR, 1.40; 95% CI, 1.03–1.90; $P = 0.03$; n = 586) than in women (log-rank test, $P = 0.06$; HR, 1.41; 95% CI, 0.98–2.01; $P = 0.06$; n = 446).

In the entire study group, there were no significant associations between *IL28B* rs12979860 and survival since the start of RRT. A significant association was found in the case of *IL28B* rs8099917: GG carriers showed an increased risk of death compared with GT+TT carriers (FIGURE 2). Clinical and laboratory parameters evaluated in this study were not associated with the polymorphic variant rs8099917 of *IL28B*.

We also examined whether *IL28B* SNPs may predict survival since the start of RRT in patients with a history of HBV or HCV infection. No association was found in the group of infected patients, while it was revealed in patients without a history of those infections. HBV- or HCV-negative patients carrying the *IL28B* rs12979860 TT genotype had an increased risk of death compared with *IL28B* CC+CT carriers (FIGURE 3). HBV- or HCV-negative *IL28B* rs8099917 GG carriers showed an increased risk of death compared with GT+TT carriers (FIGURE 4) and TT carriers (FIGURE 5). *IL28B* rs12979860 was not significant in the model composed of the same variables as for *IL13* (HR, 1.41; 95% CI, 0.93–2.15; $P = 0.1$); *IL28B* rs8099917 showed borderline significance (HR, 1.90; 95% CI, 0.96–3.75; $P = 0.07$). The prevalence of CAD differed significantly among patients carrying the *IL28B* rs12979860 TT genotype compared with CC and CT carriers (OR, 1.87; 95% CI, 1.14–3.09; $P = 0.01$; significance was maintained with a Bonferroni-corrected $P$ value of 0.017) as well as with CC carriers (OR, 1.91; 95% CI, 1.16–3.15; $P = 0.01$, significance was maintained with

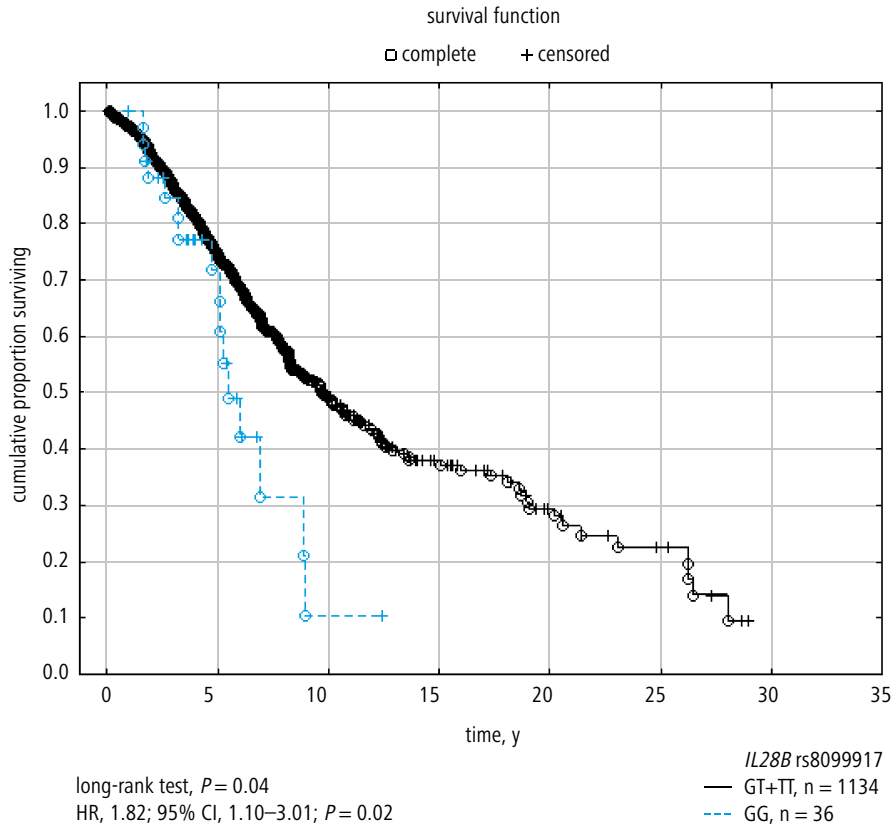



**FIGURE 2** Survival probability of hemodialysis patients with respect to *IL28B* 8099917 polymorphic variants
Abbreviations: see **FIGURE 1**

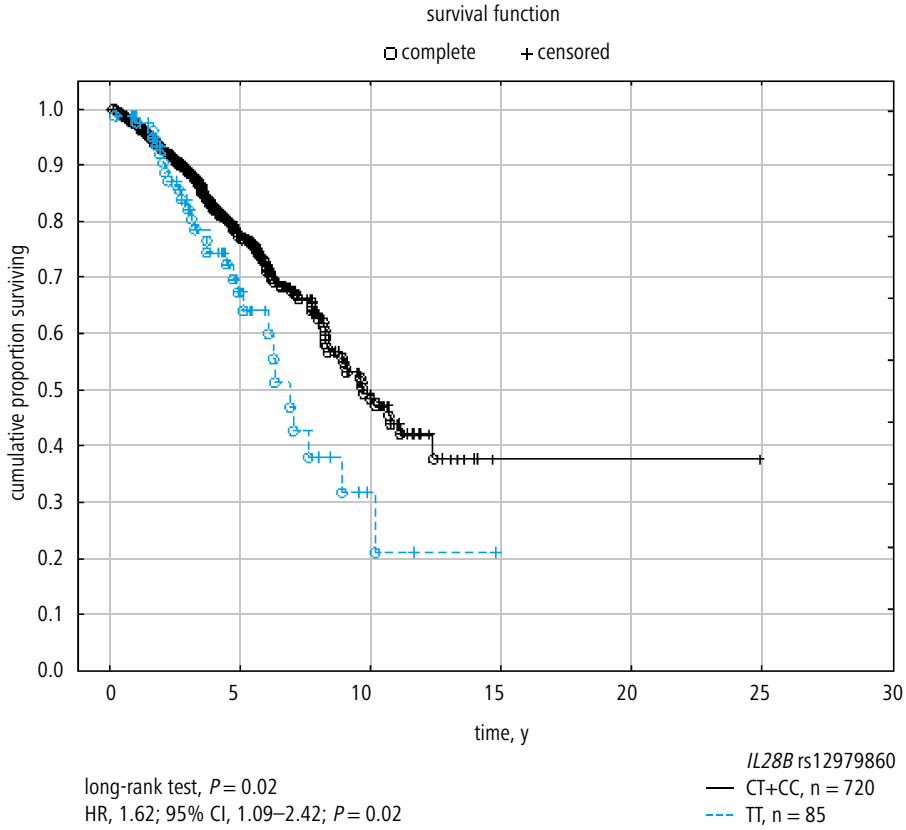



**FIGURE 3** Survival probability of HBV/HCV negative hemodialysis patients with respect to *IL28B* rs12979860 polymorphic variants
Abbreviations: see **FIGURE 1**

a Bonferroni-corrected *P* value of 0.017). Data are presented in **TABLE 2**.

**Vitamin D pathway gene polymorphisms as predictors of survival probability in patients on renal replacement therapy** Survival probability was not significantly associated with vitamin D pathway gene polymorphisms, irrespective of whether analyses were performed from the start of RRT or from patients' birth.

**DISCUSSION** Our results show that not only well-established factors such as age at the start of RRT, presence of diabetes mellitus complicated by

**FIGURE 4** Survival probability of hemodialysis patients negative for hepatitis B virus and hepatitis C virus infections with respect to *IL28B* 8099917 polymorphic variants
Abbreviations: see **FIGURE 1**

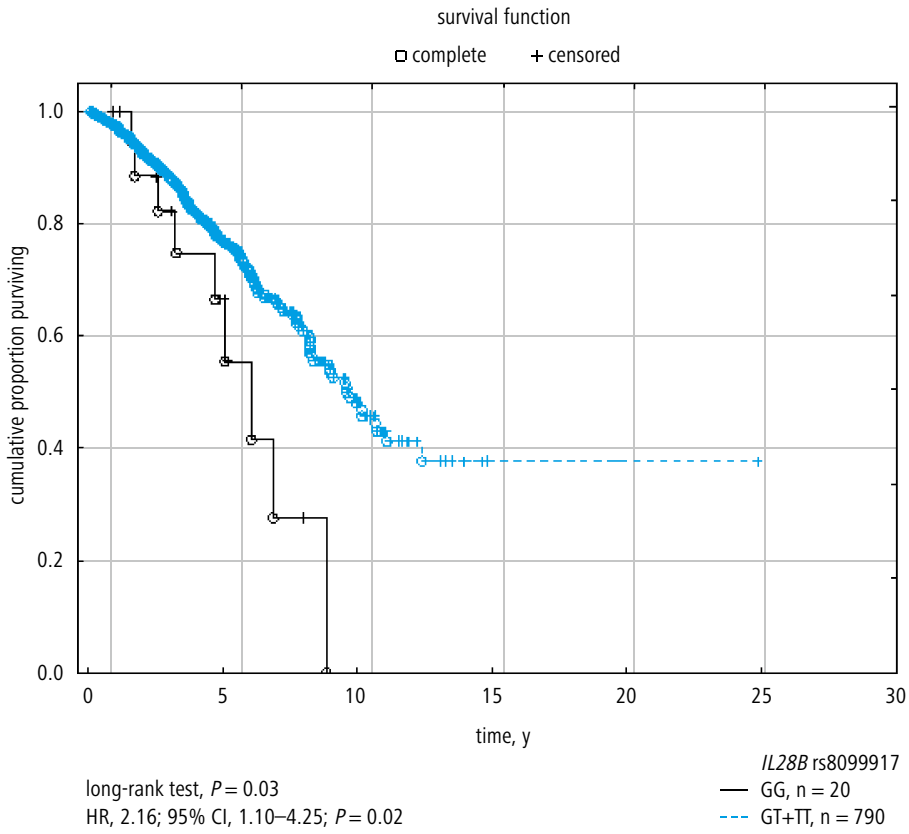
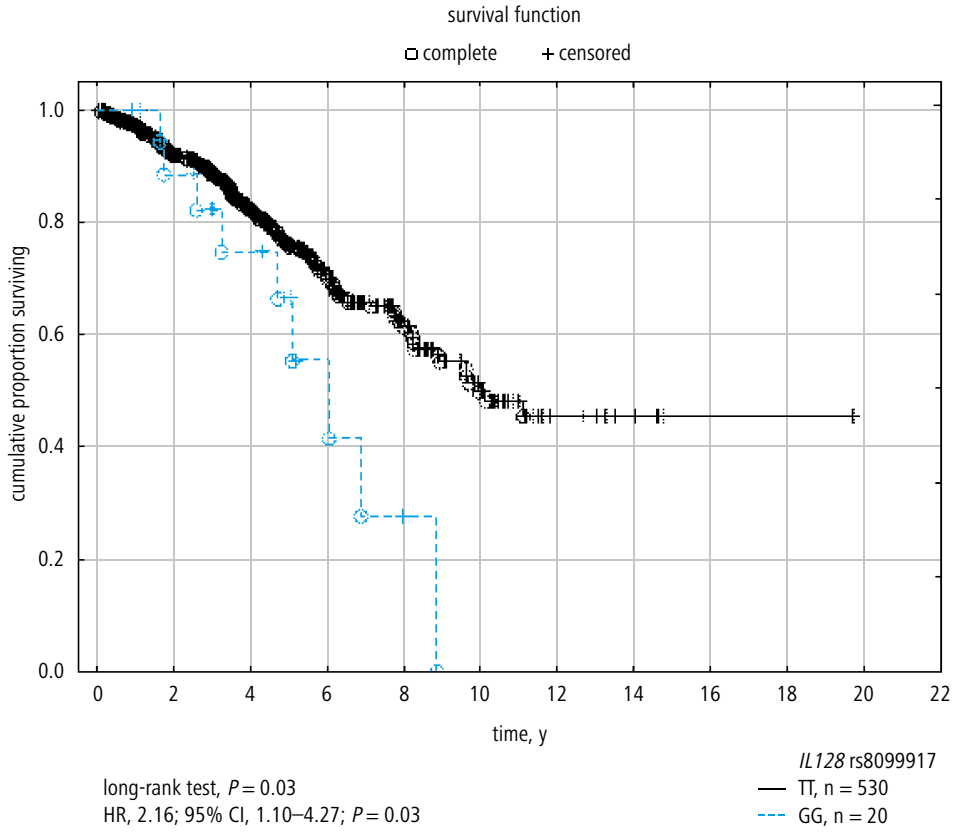



**FIGURE 5** Survival probability of hemodialysis patients negative for hepatitis B virus and hepatitis C virus infections who were *IL28B* 8099917 homozygotes
Abbreviations: see **FIGURE 1**




diabetic nephropathy, CAD or MI, but also the *IL13* rs20541 T allele, *IL28B* rs12979860 TT genotype, and the GG genotype of *IL28B* rs8099917 may negatively influence survival in HD patients.

*IL13* rs20541 (Gln144Arg) is also known as +2044 G/A (Arg130Gln). Rare *IL13* rs20541 allele homozygosity correlates with higher serum IL-13 levels than those observed in major allele homozygosity.[40] In the pilot study by Xenophontos et al.,[41] rare *IL13* rs20541 allele was associated with MI in Greek Cypriot males without impaired renal function. In our study on HD patients, the association between *IL13* rs20541 showed only borderline significance for CAD.

**TABLE 2** Demographic and clinical data of hemodialysis patients negative for hepatitis B and hepatitis C virus infections and divided according to *IL28B* rs12979860 polymorphic variants

| Parameter | *IL28B* rs12979860 | | | Odds ratio (95% CI) | *P* value |
|---|---|---|---|---|---|
| demographic data obtained from 805 patients | CC n = 344 | CT n = 376 | TT n = 85 | | |
| diabetic nephropathy | 99 (28.8) | 117 (31.1) | 23 (27.1) | TT + CT vs CC: 1.079 (0.786–1.485) | 0.7 |
| | | | | TT vs CC + CT: 0.866 (0.498–1.461) | 0.7 |
| | | | | TT vs CC: 0.918 (0.513–1.601) | 0.9 |
| hypertensive nephropathy | 70 (20.3) | 78 (20.7) | 17 (20.0) | TT + CT vs CC: 1.016 (0.709–1.461) | 1.0 |
| | | | | TT vs CC + CT: 0.966 (0.516–1.724) | 1.0 |
| | | | | TT vs CC: 0.979 (0.506–1.815) | 1.0 |
| chronic glomerulonephritis | 46 (13.4) | 39 (10.4) | 13 (15.3) | TT + CT vs CC: 0.824 (0.528–1.290) | 0.4 |
| | | | | TT vs CC + CT: 1.349 (0.656–2.587) | 0.4 |
| | | | | TT vs CC: 1.170 (0.550–2.346) | 0.8 |
| chronic tubulointerstitial nephritis | 35 (10.2) | 38 (10.1) | 12 (14.1) | TT + CT vs CC: 1.074 (0.665–1.750) | 0.9 |
| | | | | TT vs CC + CT: 1.457 (0.687–2.865) | 0.3 |
| | | | | TT vs CC: 1.451 (0.652–3.034) | 0.4 |
| age at RRT beginning, y | 65.8 (17.1–93.2) | 66.6 (20.1–95.2) | 66.9 (28.9–91.0) | TT + CT vs CC: | 0.3[a] |
| | | | | TT vs CC + CT: | 0.2[a] |
| | | | | TT vs CC: | 0.2[a] |
| clinical data obtained from 737 patients | CC n = 321 | CT n = 338 | TT n = 78 | | |
| coronary artery disease | 114 (35.5) | 123 (36.4) | 40 (51.3) | TT + CT vs CC: 1.170 (0.856–1.601) | 0.3 |
| | | | | TT vs CC + CT: 1.874 (1.136–3.091) | 0.01 |
| | | | | TT vs CC: 1.911 (1.160–3.149) | 0.01 |
| myocardial infarction | 62 (19.3) | 67 (19.8) | 17 (21.8) | TT + CT vs CC: 1.057 (0.722–1.553) | 0.8 |
| | | | | TT vs CC + CT: 1.145 (0.606–2.067) | 0.7 |
| | | | | TT vs CC: 1.164 (0.595–2.191) | 0.7 |
| parathyroidectomy/cinacalcet | 5 (1.6) | 4 (1.2) | 2 (2.6) | TT + CT vs CC: 0.925 (0.233–3.868) | 1.0 |
| | | | | TT vs CC + CT: 1.901 (0.196–9.415) | 0.7 |
| | | | | TT vs CC: 1.663 (0.155–10.38) | 0.8 |

Data are presented as number (percentage) of patients or median and range

a Mann–Whitney test

Abbreviations: see FIGURE 1 and TABLE 1

The major alleles of *IL28B* rs12979860 and rs8099917 SNPs promote spontaneous resolution of HCV infection.[42] Also a response to treatment with pegylated interferon and ribavirine is dependent on *IL28B* SNPs.[42,43] Similar results were obtained for patients with HBV infection.[44] However, *IL28B* rs12979860 and rs8099917 seem not to be related to survival probability of HBV/HCV-infected patients on HD. Interestingly, HD individuals never infected with HBV or HCV showed a higher risk of death when carrying the *IL28B* rs12979860 TT or *IL28B* rs8099917 GG genotype. To our knowledge, such associations have not been documented so far. It was shown that the CC genotype of rs12979860 protects against HCV infection.[45] A possible relationship between *IL28B* rs12979860 and carotid atherosclerosis in patients with nonalcoholic fatty liver disease was investigated but no association was found.[46] Our results demonstrated an association of *IL28B* rs12979860 TT genotype with a higher prevalence of CAD in HD patients. The *IL28B* rs12979860 TT polymorphic variant was associated with lower IL-28B production compared with the CC genotype,[47] but how this may influence the development of CAD in HD patients without HBV or HCV infection remains unknown.

Marco et al.,[48] in a study on 143 patients, showed that *VDR* rs1544410 affects survival, while *VDR* rs2228570 does not. *VDR* and *RXRA* polymorphisms were associated with susceptibility to ESRD, and the *GC* rs7041 T allele was associated with a necessity to start RRT at a younger age.[49] There were also associations between vitamin D pathway gene polymorphisms and the prevalence of CAD or MI, mineral disorders, as well as severity of secondary hyperparathyroidism among patients of this group.[50] The *VDR* rs1544410 AA genotype was shown to play a negative role (but not as an independent factor) in determining response to hepatitis B vaccination in patients from the study group.[51] Moreover, 2 of the tested SNPs of *GC* (rs7041, rs1155563) were identified as associated with 25(OH)D concentrations in a European population in a genome-wide association study.[52] Despite these

findings, suggesting a possible association of vitamin D pathway gene polymorphisms with survival probability in patients on RRT, we did not reveal associations between *GC* rs7041, rs1155563 and rs2298849, *VDR* rs2228570 and rs1544410, as well as *RXRA* rs10881578, rs10776909, and rs749759 polymorphic variants and survival either since the start of RRT or since birth.

In summary, the *IL13* rs20541 T allele and *IL28B* rs8099917 GG genotype are negative predictors of survival in patients on RRT; *IL28B* rs12979860 TT and *IL28B* 8099917 GG genotypes are negatively associated with survival in patients on RRT negative for HBV or HCV infection seromarkers. In addition, the *IL13* rs20541 T allele is an independent mortality risk factor among other clinical variables. In patients on RRT negative for HBV or HCV infection, the association of *IL28B* rs12979860 with survival may be at least partially explained by its relationship with the prevalence of CAD. Further studies are needed to establish which phenotypes of *IL13* rs20541 and *IL28B* rs8099917 are associated with prediction of survival. Vitamin D pathway gene polymorphisms examined in this study are not predictors of survival probability in patients on RRT.

**Contribution statement** AEG conceived the idea for the study and contributed to the design of the research. AEG and MS were involved in data collection and elaboration. AM was responsible for genotyping. AEG and MS were involved in writing the manuscript. All authors edited and approved the final version of the manuscript.

**Acknowledgments** We would like to thank Anna Sowińska, PhD, for her assistance in statistical analysis. We also thank the physicians of the dialysis centers for their consent in collecting the participants' data during the study period. This study was partially funded by the scientific grant of the Baxter company, allocated by the Chapter of the Polish Society of Nephrology as a blindly reviewed award-winning project, grant number 504-04-02 225 363-00 013-03 071 (granted to AEG) and partially by the Poznan University of Medical Sciences, Poznań, Poland, grant numbers 502-01-02 225 363-03 679 and 502-01-01 124 182-07 474.

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# Polimorfizmy genów cytokin związanych z limfocytami T pomocniczymi oraz genów szlaku metabolizmu witaminy D jako predyktory prawdopodobieństwa przeżycia pacjentów leczonych nerkozastępczo

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**SŁOWA KLUCZOWE**

cytokiny związane z limfocytami Th, polimorfizmy genów, prawdopodobieństwo przeżycia, terapia nerkozastępcza, witamina D

**STRESZCZENIE**

**WPROWADZENIE** Niekorzystne wyniki leczenia pacjentów wymagających terapii nerkozastępczej (*renal replacement therapy* – RRT) mogą mieć źródło w zachwianiu równowagi limfocytów T pomocniczych (*T helper* – Th) lub niedoborze witaminy D.

**CELE** Zbadano powiązania genów kodujących cytokiny związane z komórkami Th1 i Th2 oraz genów kodujących białka szlaku metabolizmu witaminy D z prawdopodobieństwem przeżycia pacjentów poddawanych RRT.

**PACJENCI I METODY** Badaniem objęto 1253 pacjentów leczonych hemodializą. Zbadano polimorfizmy genów *IL13*, *IL4R*, *IL18*, *IL12A*, *IL12B*, *IL28B*, *MCP1*, *GC*, *VDR* oraz *RXRA*. Użyto metody Kaplana–Meiera i testu logarytmicznego rang do określenia istotności prawdopodobieństwa przeżycia.

**WYNIKI** Pacjenci posiadający rzadszy allel T *IL13* mieli wyższe ryzyko zgonu w porównaniu z posiadającymi genotyp CC (test logarytmiczny rang: p = 0,005; HR = 1,40; 95% CI 1,11–1,76; p = 0,005). Pacjenci z genotypem *IL28B* rs8099917 GG mieli wyższy współczynnik umieralności w porównaniu z posiadaczami *IL28B* GT i TT (test logarytmiczny rang: p = 0,04; HR = 1,82; 95% CI 1,10–3,01; p = 0,02). Posiadacze genotypu TT *IL28B* rs12979860 mieli zwiększone ryzyko zgonu w porównaniu z nosicielami genotypów CC i CT (test logarytmiczny rang: p = 0,02; HR = 1,62; 95% CI 1,09–2,42; p = 0,02) tylko wtedy, gdy nie byli zakażeni wirusem HBV lub HCV. Częstość występowania choroby wieńcowej była istotnie większa u pacjentów z genotypem TT *IL28B* rs12979860 w porównaniu z posiadaczami CT+CC (OR = 1,87; 95% CI 1,14–3,09; p=0,01). Nie wykazano związku między badanymi polimorfizmami *GC*, *VDR* i *RXRA*, a przeżyciem.

**WNIOSKI** Allel T *IL13* rs20 541 oraz genotyp GG *IL28B* rs8099917 są negatywnymi predyktorami przeżycia pacjentów poddawanych RRT, natomiast genotyp TT *IL28B* rs12 979 860 zwiększa ryzyko zgonu tylko u pacjentów niezakażonych wirusem HBV lub HCV.

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Praca wpłynęła: 25.03.2015.
Przyjęta do druku: 25.04.2015.
Publikacja *online*: 03.06.2015.
Nie zgłoszono sprzeczności interesów.
Pol Arch Med Wewn. 2015; 125 (7-8): 511-520
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