

Effects of increased iodine supply on the selenium status of kids

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ABSTRACT: The aim of the study was to monitor the effect of increased iodine supplementation of the clinically healthy kid organism on the selenium status. The study included 7 clinically healthy 14-day-old kids from mothers with high iodine supplementation (group E) and 7 clinically healthy kids from mothers with hypiodaemia (group C). Kids in group E were administered potassium iodide orally from 14 to 90 days of age. During the experimental period, the group E kids had a total daily iodine intake (from the feeding ration and from the *per os* potassium iodide administration) of 440–590 µg per head and day in comparison with 140–190 µg per head and day in the group C kids (only from the feeding ration; no potassium iodide administration). In kids of both groups, selenium concentration (Se), glutathione peroxidase activity (GSH-Px), concentration of the thyroid gland hormones (T₃ and T₄) and animal weights were monitored. In the group E kids, lower Se concentration (88.1 ± 10.9 µg/l; $P < 0.01$) and lower activity of GSH-Px (484.0 ± 125.4 µkat/l; $P < 0.05$) were proved at the end of the experiment (at 105 days of age of the kids) in comparison with the group C kids (131.8 ± 23.2 µg/l and 713.3 ± 153.3 µkat/l, respectively). No significant differences were found out in the T₃ or T₄ concentrations or in the weights of animals of both groups. The results indicate that increased iodine supplementation may have a negative effect on selenium metabolism and/or selenium status in kids.

Keywords: goat; trace elements; interaction; glutathione peroxidase; thyroid hormones

Essential trace elements are functional, structural and regulatory components of a number of biomolecules that play a vital role in the metabolism of living organisms. In order to synthesize them, trace elements must be available in a corresponding chemical form and concentration (Underwood and Suttle, 1999).

Biological functions of iodine are mediated chiefly by its impact on the thyroid gland. Its insufficiency is manifested by a development of goitre, abortions and stillbirths. It negatively affects offspring vitality, growth and development, adult animal health and production, and it also influences the immune system (Bires et al., 1996; Sargison et al., 1998; Slosarkova et al., 1999; Underwood and Suttle, 1999). However, other elements such as selenium, iron and zinc affect the thyroid gland metabolism

and thus also the levels of thyroidal hormones in the organism (Zimmermann and Kohrle, 2002).

Selenium affects the thyroid gland metabolism and its hormones both by deiodinases that regulate the synthesis and degradation of the biologically active thyroid gland hormone (T₃) and by selenoperoxidases that protect the thyroid gland from hydroperoxide produced during the synthesis of thyroid hormones (Hess and Zimmermann, 2004). Deiodinases are selenoproteins regulating thyroxine conversion (T₄) into the biologically active form 3,3',5-triiodothyronine (T₃) or reverse triiodothyronine (rT₃) – inactive hormone of the thyroid gland (Arthur et al., 1990; Larsen and Berry, 1995; Kohrle, 2000; Birringer et al., 2002). Selenium deficiency can cause a decrease in the concentration of T₃ and take part in the development of

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the signs related to disorders of the thyroid gland hormone metabolism. After the supplementation of animals with selenium, there appears to be an increase in the concentration of T_3 (Wichttel et al., 1994; Awadeh et al., 1998; Pavlata et al., 2004a). Other biological functions of selenium are carried out by a series of selenoproteins whose main role is participation in the removal of reactive forms of oxygen from the organism (Birringer et al., 2002).

In order to maintain the metabolic homeostasis of essential trace elements, an organism mostly utilizes regulation on the level of absorption and excretion. The regulation of intestinal element absorption concerns for instance Zn, Fe, Cu and Mn. On the other hand, the metabolic homeostasis of selenium and iodine is mostly maintained through the renal excretion of these elements and the regulation of their resorption in the intestine is minimal (Windisch, 2002).

Besides the primary deficiencies due to deficiencies of trace elements in feed also the secondary deficiencies have some impact. They are mostly caused by a negative action of an overload of other agents. Secondary iodine deficiencies are usually caused by the occurrence of goitrogenic agents in feedstuffs and water. Some of them are nitrates, nitrites, glucosinolates, humic acids, etc. (Cox-Ganser et al., 1994; Reid et al., 1994; Pisarikova et al., 1996; Kursá et al., 2000; Herzig et al., 2001; Travnicek et al., 2001; Slosarkova et al., 2002). Selenium interactions with other elements are among the factors bearing on selenium metabolism in the organism. Interactions of selenium and sulphur, and also of selenium and cadmium, arsenic, copper, cobalt, manganese, lead, iron and others were described (Shamberger, 1983). An increase in the sulphur content in feed has an impact on lowering the plasma concentration of selenium in dairy cows (Ivancic and Weiss, 2001). Negative effects of sulphur interactions on the activity of glutathione peroxidase (GSH-Px) were also reported (Murphy and Quirke, 1997). The same applies to the concentration of selenium in the liver (Van Ryssen et al., 1998). On the other hand, the protective action of selenium from toxic effects of cadmium, mercury, lead and thallium was noted (Shamberger, 1983).

Due to the current deficiencies of iodine, selenium and other trace elements in many regions of the world, including the Czech Republic, attention is paid to their sufficient supply (Herzig and Suchy, 1996; Travnicek and Kursá, 2001; Pavlata

et al., 2002, 2005) in both veterinary and human medicines.

From this perspective, there is still a lack of information on potential interactions between highly supplemented iodine and other trace elements. The aim of this study was to examine the impact of varied iodine supply on selenium metabolism in kids.

MATERIAL AND METHODS

Animals

The experiment was carried out in stables of the Clinic of Diseases of Ruminants of the University. A total of 14 clinically healthy kids of the white short-haired goat breed were involved. The experimental group (E) contained 7 kids highly supplemented with iodine and the other 7 kids were in the control (C) group – without iodine supplementation. The kids in the experimental group were from iodine-supplemented mothers (the mean iodine concentration in their urine was above 350 $\mu\text{g/l}$). The kids in the control group were from mothers not supplemented with iodine (the mean iodine concentration in their urine was below 30 $\mu\text{g/l}$). The actual experiment was performed in an age period between 14 days and 105 days. In the period between birth and weaning at the age of 75 days, the kids of the individual groups (E and C) were housed in groups, together with their dams, i.e. the kids had free access to their mothers' milk throughout this period. After weaning, the dams were moved. The feed rations of all kids consisted, at first, of mostly mother's milk complemented with hay available *ad libitum* and grain mixture whose amount gradually grew from approximately 100 g to 300 g per kid/ day. The grain mixture contained 29.7% of nitrogenous compounds, 10.8% of fibre, 4.8 MJ of net energy for fattening, 20.0 g of calcium, 13.9 g of phosphorus, 6.3 g of sodium, 5.4 g of magnesium, 152.3 mg of zinc, 104.4 mg of manganese and 25.2 mg of copper in 1 kg of dry matter. The grain mixture was enriched with selenium and vitamins A, D and E and it contained 0.3 mg of iodine/kg of dry matter and 1 mg of selenium/kg of dry matter. The animals had unlimited access to drinking water. In the period between the 14th and 90th days of age, iodine in the form of a solution of potassium iodide (0.262‰) was administered *per os* daily to the experimental group animals. The daily amount

Table 1. Iodine content in the particular components of feed rations in the kids of the experimental and control groups

Feed ration components	Iodine concentration	
	experimental (E)	control (C)
Milk ($\mu\text{g/l}$)	136.7 ± 50.4	71.5 ± 16.1
Hay ($\mu\text{g/kg}$ of dry matter)	50	50
Grain mixture ($\mu\text{g/kg}$ of dry matter)	300	300

of the administered iodine was 200 μg until 45 days of age and 300 μg until the age of 90 days, *i.e.* 1.2 or 1.5 ml of the solution, respectively. Iodine contents in the feed ration components are shown in Table 1. Its content in the milk of the individual dams was measured (Bednar et al., 1964) once a month. The content of iodine in grain mixture was determined using the method described by Bednar et al. (1964). In hay iodine was established based on its estimated value (Sommer, 1994). The maximum possible total daily iodine intakes by the kids of both groups are given in Table 2. The milk intake was determined by estimate. In the first two weeks of age, only 1 litre of milk was assumed to have been taken in. Further intake until the weaning (at the age of 75 days) was assumed to be at the volume of 1.5 l of milk. In the grain mixture, the intake was established at 100, 200 or 300 g until the age of 45, 75 and more days, respectively. The assumed hay intake was established at 200, 500 or 1 000 g until the age of 45, 75 and more days. All of the kids were repeatedly weighed before the start of the experiment and during the experiment.

Laboratory examination

Heparinised and nonheparinised blood was collected from kids of both groups from the *vena jugularis* at the age of 14 days (before the start of potassium iodide administration to the E group) for laboratory purposes (sampling No. I). The following samples were taken in the same manner at the age of 45 days, 75 days and 105 days, *i.e.* 15 days after the application of potassium iodine was discontinued (sampling Nos. II, III, IV). The selenium supply was established via the concentration of selenium and glutathione peroxidase (GSH-Px) activity in whole heparinised blood of the kids. Iodine metabolism was monitored indirectly by determining the concentrations of thyroidal hormones (T_3 and T_4) in the blood serum.

The method of selenium determination consisted in the following steps: samples of whole heparinised blood were mineralised in a closed system using a microwave (MLS-1200, Milestone, Italy) digestion technique with HNO_3 and H_2O_2 . Samples were evaporated and the mineral residue was dissolved in

Table 2. Total iodine intake (μg) per kid/day in animals of the experimental (E) and control (C) groups before the start of the experiment and during the experiment with detailed blood sampling times

Age (days)		< 14	14–45	46–75	76–90	91–105
Dietary iodine intake	E	140	245	290	140	140
	C	70	150	190	140	140
Iodine administration	E	0	200	300	300	0
	C	0	0	0	0	0
Total iodine supply	E	140	445	590	440	140
	C	70	150	190	140	140
Blood samplings*		I	II	III		IV

*always at the end of the given period

water to which 20% HCl was added. Selenium was then determined with Solar 939 AA Spectrometer (Unicam, UK) using a hydride AAS technique.

The catalytic activity of GSH-Px enzyme was determined on automatic analyser Cobas Mira (Roche, UK) according to Paglia and Valentine (1967) with the Ransel Randox set. Thyroid hormones were determined using Immulite machine by means of chemiluminescence sets by BioVendor (Total T₃ – LKT 31 and Total T₄ – LKT 41).

Statistical methods

The Microsoft Excel 2003 software was used to compute the basic statistical characteristics of the results (mean, standard deviation) of the particular groups and to carry out correlation and regression analyses of the relation between the concentration of selenium and GSH-Px activity values and the comparison between the groups (using the *F*-test for determining the variance of the individual sets and by the result using two-sided Student's *t*-test assuming equal/unequal variances) as well as the evaluation of the dynamics of changes in the individual parameters using the paired *t*-test.

RESULTS AND DISCUSSION

The experimental group kids when compared to the control group ones received approximately a double iodine amount via the milk prior to the start of the experiment and approximately a triple iodine amount during the supplementation period. Meschy (2000) recommended the minimum iodine supply in a goat diet to be 0.4 mg/kg of dry matter and, in production dairy goats, it could be higher than 0.6 mg/kg of dry matter. Iodine intake in the group E kids thus exceeded the recommended daily

dose for kids during the supplementation period (between 200–300 µg a day with respect to weight and age; it was established based on doses for adult goats and other ruminants) by some 50–100%. This increase in dosage cannot, however, be considered as toxic because iodine surplus induced disorders were noted only with manifold excess of the recommended doses and toxic doses in ruminants rank among the highest (Paulikova et al., 2002).

At the age of two weeks before the start of the potassium iodide administration to the group E kids, the concentrations of selenium and the GSH-Px activity in the whole blood of kids of both groups were very similar but relatively low (Table 3, Figures 1 and 2). Owing to the fact that ruminants display a good selenium placenta transfer and milk concentration and the fact that the supply status of their young thus corresponds to the supply in their dams (Pavlata et al., 2003, 2004b) one can state that the selenium intake in both groups was very balanced but relatively low. The values of selenium concentration in the whole blood ranging around 65 µg/l of whole blood can be classified as marginal to insufficient selenium supply because the concentration of selenium in blood should range around 100 µg/l (Pugh, 2002). As regards feeding the animals the selenium enriched grain mixtures during the experiment, both groups displayed a gradual increase in the concentration of selenium and GSH-Px activity (Table 3). In group C, however, this increase was surprisingly bigger.

Compared to the group C animals, the animals with increased iodine supplementation had a statistically lower concentration of selenium ($P < 0.01$) in the last two samples (at the age of 75 and 105 days) and also the GSH-Px activity in the last sample ($P < 0.05$) (Figures 1 and 2).

The results show a possible negative impact of higher dosage iodine supplementation on the selenium metabolism when determined on the basis of

Table 3. The dynamics of changes in the selenium concentration and GSH-Px activity (mean ± SD) in the whole blood of kids supplemented with iodine (E) and without iodine supplementation (C) with the statistical significance of the value variation of the given parameter in the course of the experiment

Age (days)		14	45	75	105
Se (µg/l)	E	65.7 ± 11.2 ^{bc}	66.1 ± 14.1 ^c	76.0 ± 11.7 ^{bf}	88.1 ± 10.9 ^{cef}
	C	65.5 ± 7.8 ^{bc}	77.3 ± 11.1 ^{de}	101.4 ± 11.9 ^{bdf}	131.8 ± 23.2 ^{cef}
GSH-Px (µkat/l)	E	247.6 ± 60.8 ^{abc}	335.8 ± 117.9 ^{ade}	408.2 ± 88.5 ^{bdf}	484.0 ± 125.4 ^{cef}
	C	235.2 ± 28.5 ^{abc}	414.0 ± 84.6 ^{ade}	523.0 ± 120.1 ^{bdf}	713.3 ± 153.3 ^{cef}

aa, bb, cc, dd, ee, ff on one table line: $P < 0.05$

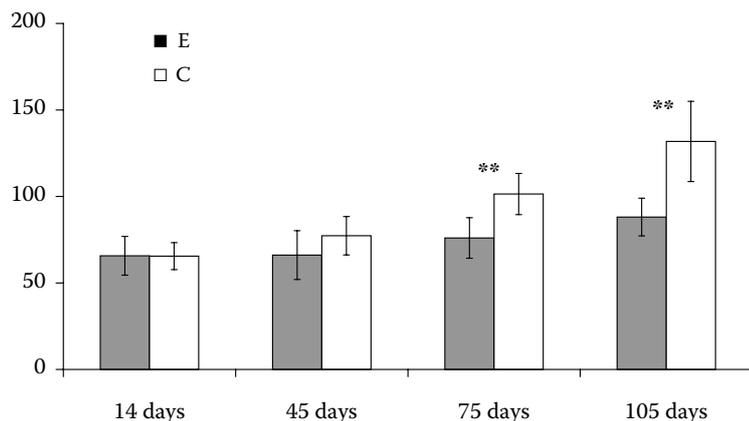


Figure 1. The concentration of selenium ($\mu\text{g/l}$) in the blood of kids of the group supplemented with iodine (E) and without iodine supplementation (C) with the statistical significance of the value variation between the groups (** $P < 0.01$)

the selenium concentration and the GSH-Px activity in whole blood. In the group without iodine supplementation, there was an increase in the concentration of selenium and in the GSH-Px activity onto a sufficiently high level but the group E displayed the values of both these parameters at a marginal supply status level. The concentration of selenium and the GSH-Px activity in the blood of the highly iodine supplemented kids reached almost 70% of the value in kids without iodine supplementation (C) at the end of the experiment.

The mutual relation between the GSH-Px activity and the concentration of selenium is documented by the results of correlation and regression analyses of the relation between the values of both parameters (Figure 3).

It is clear from the figure that the relation between both parameters is close (the correlation coefficient $r = 0.93$; $P < 0.01$) and that it is linear. It is then clear that the GSH-Px activity in the blood of kids is related to the content of selenium in their blood, and similarly like in cattle or horses, we can use it for indirect assessment of their selenium supply status (Pavlata et al., 2000; Ludvikova et al., 2005). Given

the fact that selenium is incorporated into the erythrocytic GSH-Px already during erythropoiesis, erythrocytic GSH-Px is considered a suitable indicator of biologically active selenium and also of its more permanent supply status while blood or serum levels of selenium involving enzymatic and nonenzymatic components reflect short-term dietary variations of this trace element (Hoffman et al., 1978; Gerloff, 1992).

In this context we can also clarify a somewhat later significant variance of the GSH-Px activity between the E and C groups (sampling IV at the age of 105 days) in comparison with the selenium concentration where the significant variance appears as early as since sampling III at the age of 75 days. This significantly lower GSH-Px activity in kids highly supplemented with iodine indicates a possible negative impact of the increased iodine supply on other important selenoproteins. They mediate biological functions of selenium in the organism (Birringer et al., 2002).

Despite the important difference in the iodine contents taken in by the kids of the individual groups, we did not register any significant differ-

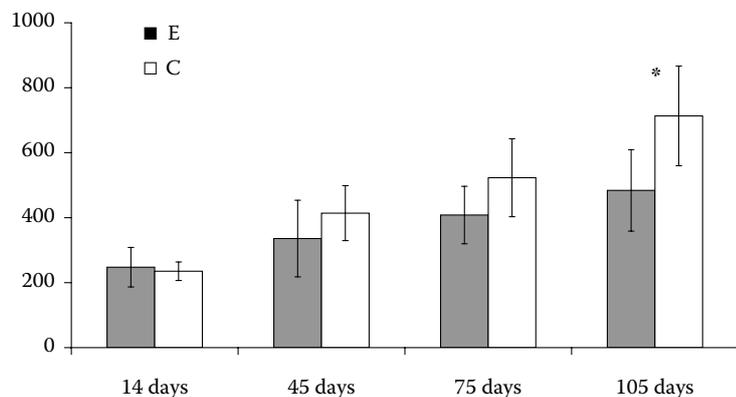


Figure 2. The GSH-Px activity ($\mu\text{kat/l}$) in the blood of kids of the group supplemented with iodine (E) and without iodine supplementation (C) with the statistical significance of the value variation between the groups (* $P < 0.05$)

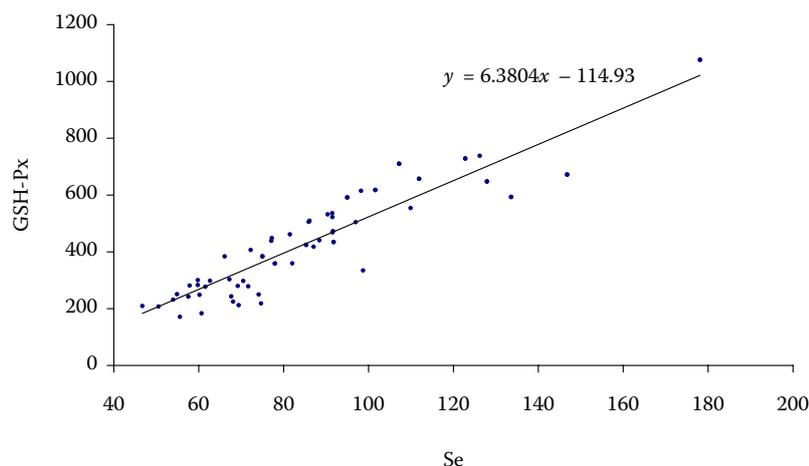


Figure 3. The relation between the values of selenium concentration ($\mu\text{g/l}$) and GSH-Px activity ($\mu\text{kat/l}$) in the whole blood of kids with presentation of the equation of regression line

ences in the concentrations of the thyroid gland hormones (Table 4).

It is clear from the table that the concentrations of T_3 and T_4 displayed relatively substantial dynamics. There was an insignificant increase in their values in the first month of monitoring that, nevertheless, gradually fell in the following period. This decrease was statistically significant.

Both animal groups reached the reference values of T_4 , which are in goats within the range of 39–103 nmol/l given the fact that the values are usually higher in the young (65–130 nmol/l) (Kraft and Durr, 2001). It can also clarify the gradual decrease in values we found during the experiment. The decrease in the T_3 and T_4 concentrations can also be related to the termination of mother’s milk feeding. One can assume that the animals acquired mother’s milk iodine in the form more ready to use and that therefore iodine utilization from other sources was not sufficient to sustain the original thyroid gland levels after weaning.

Despite the varied iodine supplementation, no differences were found in the thyroid gland hor-

mone concentrations. This can be caused by the fact that both groups had sufficient iodine supplementation and thus there were no differences in the T_3 and T_4 concentrations caused by iodine deficiency. Moreover, despite the well-known iodine influence on the thyroid gland metabolism and its hormones, close relations are not always found between the iodine supply status and the T_3 and T_4 concentrations. For example, Pechova et al. (2004) did not find a correlation between the iodine concentrations in urine and the T_3 and T_4 concentrations. Neither did Randhawa and Randhawa (2001) detect any differences in the T_3 and T_4 concentrations nor in the T_4/T_3 ratio in subclinical iodine insufficiency diagnosed on the basis of iodine concentration in blood.

The results do not prove a relation between the selenium supply status and the T_3 and T_4 concentrations either. Given the fact that deiodinases which influence the production of the active form of the thyroid gland hormone (T_3) are selenoproteins, one could assume that an increase in the selenium supply to the organism will also improve the

Table 4. The T_3 and T_4 concentrations (mean \pm SD) in the blood serum of kids of the group supplemented with iodine (E) and without iodine supplementation (C) with the statistical significance of the value variation of the given parameter in the course of the experiment

Age (days)		14	45	75	105
T_3 (nmol/l)	E	4.1 \pm 1.74 ^a	4.9 \pm 0.85 ^{bc}	3.2 \pm 0.42 ^{bd}	2.3 \pm 0.40 ^{acd}
	C	4.7 \pm 0.86 ^{ab}	4.9 \pm 0.66 ^{cd}	3.0 \pm 0.54 ^{ac}	2.5 \pm 0.78 ^{bd}
T_4 (nmol/l)	E	81.9 \pm 27.60	98.3 \pm 20.40 ^{ab}	78.5 \pm 13.40 ^a	72.5 \pm 7.53 ^b
	C	89.8 \pm 20.69 ^a	105.1 \pm 12.03 ^{bc}	83.3 \pm 13.61 ^b	71.1 \pm 21.05 ^{ac}

a:a, b:b, c:c, d:d on one table line: $P < 0.05$

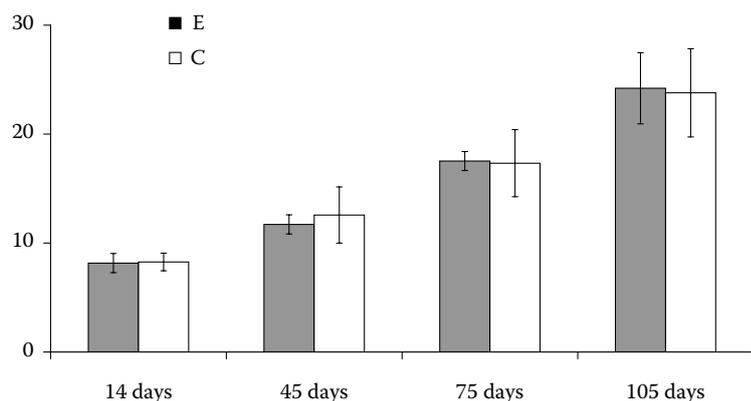


Figure 4. Mean weight of kids in both groups (E, C) during the experiment (kg)

concentration of T_3 (Pavlata et al., 2004a). Neither changes in the monitored hormone concentrations in time nor the non-existent variance between the E and C groups that had different concentrations of selenium in blood support these assumptions.

The initial mean weights of both groups were very similar, and the weight gains during the experiment were also comparable in both groups. During the whole time of the experiment, the mean weight of animals in both groups did not vary significantly (Figure 4). Neither did various levels of iodine supplementation nor significant differences in the selenium supply status have an influence on weight gains and weights of the experimental animals. Because weight gains were very similar, one can rule out that the found variance in the selenium supply status in kids was brought about by the intake of different feed volumes.

Taking into account the surprising result indicating a possible negative influence of increased iodine supplementation on selenium metabolism, one can surmise that the form, the concentration or the amount of the supplied iodine in the form of potassium iodide have had some negative impacts on absorption or further selenium metabolism in kids. The interpretation of such a finding is not, however, easy on the basis of the experiment that we carried out.

It has been noted that during passage through the intestines, neither selenium nor iodine display any significant interactions with the other diet components and that regulation impacting their intestinal absorption is minimal. This leads to the organism being passively exposed to their supply in relation to their contents in the diet. The main homeostatic regulation of the selenium and iodine metabolisms in the organism takes place through their renal excretion (Windisch, 2002; Arova et al.,

2003). In ruminants, however, this process can be to a certain extent under the influence of the changes during ruminal fermentation. In it, compounds with various degrees of solubility can develop as a result of the varied composition of ruminal microflora. Their further use in the gastrointestinal tract varies (Koenig et al., 1997; Underwood and Suttle, 1999). One can therefore speculate that a daily supply of potassium iodide could influence the composition and function of ruminal microflora in a manner that resulted in lower biological availability of selenium in the subsequent sections of the gastrointestinal tract.

The extent of the monitored parameters during the experiment does not enable to interpret the acquired data unambiguously and further experiments will be necessary to do so. This study can be considered as a pilot study that shows the potential negative influence of higher *per os* supply of potassium iodide on the selenium metabolism in ruminants. Or, as the case may be, that there are potentially important interactions between selenium and iodine whose study should attract further attention.

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