
ENVIRONMENTAL CONDITIONS CAUSING SELENIUM DEFICIENCY IN SHEEP

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Abstract

The research was carried out on the Experimental Farm in Uhrusk, which belongs to the University of Life Sciences in Lublin. Two-year experiment were conducted in order to assess the influence of environmental conditions on the selenium content in sheep. The investigations included meadows grazed by sheep, from which samples of soils and meadow herbage were obtained for analysis of selenium content. The animal material consisted of 64 lambs and their mothers, whose blood serum was taken for determinations of the selenium content in their organism. The research results showed that the analyzed soils and meadow grasses were characterized by low selenium content. Also, the selenium content in fodder fed to the sheep and lambs was below the animals' standard demand for this element. The low selenium level found in the sheep's blood serum distinctly proved a deficiency of this element in their organisms. The results show that environmental conditions have a significant influence on the supply of selenium to adult sheep and their offspring. Selenium deficiency in particular trophic chain links can present a serious threat to the health of animals fed with homegrown fodders.

Keywords: selenium, sheep, blood serum, milk, plants, soil.

UWARUNKOWANIA ŚRODOWISKOWE NIEDOBORU SELENU U OWIEC

Abstrakt

Eksperyment przeprowadzono w Gospodarstwie Doświadczalnym w Uhrusku należącym do Uniwersytetu Przyrodniczego w Lublinie. W cyklu dwuletnim oceniano wpływ warunków środowiskowych na zawartość selenu w organizmie owiec. Z łąk przeznaczonych do wypasu owiec pozyskano próbki gleby i roślin łąkowych, a następnie poddano analizie na zawartość selenu. Materiał zwierzęcy stanowiły 64 jagnięta i ich matki, od których pobrano krew w celu pozyskania surowicy do oznaczania zawartości selenu. W badanych glebach i florze łąkowej wykazano niską zawartość selenu. Również zawartość selenu w paszy dla owiec i jagniąt była

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poniżej zapotrzebowania zwierząt na ten pierwiastek. Niska koncentracja selenu w surowicy krwi badanych zwierząt wyraźnie wskazuje na niedobór tego mikroelementu w ich organizmach. Z badań wynika, że uwarunkowania środowiskowe mają istotny wpływ na podaż selenu dla dorosłych owiec i ich potomstwa. Niedobór selenu w poszczególnych ogniwach łańcucha pokarmowego może stanowić poważne zagrożenie dla zdrowia zwierząt karmionych paszami produkowanymi lokalnie.

Słowa kluczowe: selen, owca, surowica krwi, mleko, roślina, gleba.

INTRODUCTION

Selenium is a crucial trace element, responsible for the proper functioning, growth, and development of a living organism. It is also characterized by a very narrow margin between the lowest acceptable level of intake and the levels causing toxicity (ZHANG et al. 2001). It plays a complex biological function, mainly related to its presence in enzymatic proteins. Selenium is a constituent of 35 enzymes and selenoproteins (CHEN et al. 1999). Selenium has four natural oxidation states – 2 (selenides), 0 (elemental), +4 (selenites) and +6 (selenates) (BARCELAUX 1999).

An insufficient supply of this element results in an inferior productivity of animals. Several metabolic diseases have been linked to selenium deficiency, including white muscle disease (WMD) in calves and lambs (SMART et al. 1981), hepatitis in pigs, exudative diathesis and pancreatic degeneration in poultry.

Ensuring that animals are in good health requires a good biological value of fodder, which in turn depends on the mineral composition of soils. Elemental selenium and precipitated metal selenides are not bioavailable for plant uptake. It is only the water soluble forms that plants can readily take up. Beside plants, some microbial populations can reduce selenium to volatile chemical forms (COMBS 2001, CHASTEEN, BENTLEY 2003). High soil selenium not available to plants is due to the acid soil reaction causing a lower level of water soluble and therefore phytobioavailable selenium. Plants absorb soil selenium most effectively when it is in the form of selenite or selenate and synthesized selenomethionine (Se Met) (RAYMAN 2004). Hence, causes of deficiency or relative excess of a given mineral should be sought in soil, the first link in the trophic chain composed of soil – plant – animal (HARTIKAINEN 2005). Numerous authors have proven empirically that large parts of Poland, including the Lublin District, lie on selenium-deficient soils (PIOTROWSKA 1984, BIERNACKA, MAŁUSZYŃSKI 2006, PATORCZYK-PYTLIK, KULCZYCKI 2009).

Locally produced fodder is mainly used in the nutrition of lambs, which may suffer from mineral deficits due to some local deficiency of elements in soil and, consequently, in plants (PATORCZYK-PYTLIK 2009, BOMBIK et al. 2010). Animals fed on fodder with less than 0.1 mg kg⁻¹ selenium may develop symptoms of diseases related to selenium deficiency.

The aim of the present work has been to determine selenium levels in organisms of adult sheep and their offspring.

MATERIAL AND METHODS

The investigations were carried out in two replicates completed at the turn of May and June in two consecutive calendar years (2005 and 2006). They covered meadows on two very different soil habitats: mineral lessive soil (alternate meadow) and peat-organic soil (permanent meadow). Both meadows lie in an area periodically flooded by the Bug River. They are characterized by a poor variety of plant communities. Soil samples were taken from the surface layer (0-20 cm deep) scattered at 12 randomly selected sites representative of the whole meadow area. Laboratory assays were made on averaged samples of friably dry soil. Soluble selenium forms in soils were determined in a solution obtained after six-hour extraction of 3 g of soil by 20 cm³ 0.05 M EDTA, whereas total selenium content was checked in a solution achieved after mineralization of 1 g of soil with a 3:1 mixture of oxygenating acids HNO₃ and HCl.

Meadow plants were sampled from 1 m² squares located at the same 12 sites as the soil samples. Plant samples were digested in a microwave stove Multiwave 3000 (Anton Paar, Austria). A similar resolution procedure was applied before determination of the selenium content in fodder and milk. The determination of selenium in the extract and mineralizates was carried out in an atomic absorption spectrometer SpektrAA 220Z (Varian, Australia) with electro-thermal induction and deuter Zeeman-effect background correction.

The experiment was repeated twice in two consecutive years (2005 and 2006). The evaluation of sheep's selenium status was based on analyses of this element in blood serum. The investigation involved 64 five-week-old ram lambs of the synthetic prolific-meaty line BCP and their dams. During the experiment, the lambs were nursed on their dams' milk and the adult sheep were fed on fodder produced on the experimental soils. In order to obtain serum, blood was drawn from the *vena jugularis* to heparinized test tubes (Medlab Poland) and centrifuged at 3000 rpm. Selenium in the blood serum of lambs and their dams was determined by the GFAAS method according to the modified protocol by NEVE and MOLLE (1986). Simultaneously, determinations were conducted on a reagent trial and reference material Seronorm (Nycomed and Co., Norway). The criteria of analytical reliability were as follows: LOD – 8 µg dm⁻³, LOQ – 16 µg dm⁻³.

The results were statistically processed by analysis of variance run in the statistical program Statistica (Data Analysis Software System Version 6).

RESULTS AND DISCUSSION

The tested soils differed significantly both in the total and soluble selenium content (Table 1). The highest determined total selenium concentration in organic soil was almost 10-fold higher than the total content of this element in mineral soil. The small amount of selenium detected in mineral soil ($< 0.5 \text{ mg kg}^{-1}$) is considered insufficient (RAMIREZ-BRIBIESCA et al. 2001). The concentration of easily soluble selenium forms, which are the ones mainly absorbed by plants, constituted a small proportion of this element in the tested soils: about 10% in samples of mineral soil and about 6% in organic soil. Similar results were obtained by other authors (HAMADA, HATTORI 1989). The low availability of selenium from organic soils may result from the fact that this type of soil contains a prevalent share of stable compounds with iron oxides, hydroxides and organic matter, which makes selenium less than fully available to plants despite its high total content.

Presence of stable selenium compounds is also proven by the reaction of the analyzed soils. In the two consecutive years, the soils had similar values of pH ($7.4 \text{ pH}_{\text{H}_2\text{O}}$ and 6.6_{KCl} in mineral soil and 7.3 and 6.3, respectively, in organic soil), which are characteristic of neutral soils, with selenins which are difficult to be absorbed by plants (JOHNSON 1991, WANG, CHEN 2003).

The low concentration of selenium bioavailable forms in soil significantly determined the concentration of this element in meadow flora, below the

Table 1
pH reaction; total and soluble selenium contents in two different soil systems

Year	Parameters		Soil habitat	
			I	II
2005	pH	H ₂ O	7.4	7.3
		KCl	6.6	6.3
	total Se \pm SD (mg kg ⁻¹)		0.075 ^a 0.006	0.726 ^b 0.014
	soluble Se \pm SD (mg kg ⁻¹)		0.007 ^a 0.001	0.042 ^b 0.01
2006		H ₂ O	7.2	7.3
		KCl	6.5	6.3
	total Se \pm SD (mg kg ⁻¹)		0.065 ^a 0.004	0.722 ^b 0.013
	soluble Se \pm SD (mg kg ⁻¹)		0.007 ^a 0.003	0.038 ^b 0.040

I – mineral soil – habitat of ‘alternate meadow’

II – organic soil – habitat of ‘permanent meadow’

Statistically significant differences between selenium content in soils from two soil habitats were designated with different letters: *a, b* – $p < 0.01$.

Table 2
Selenium content in grasses of two different soil habitats

Year	Se (mg kg ⁻¹ d.m.) ± SD in habitat	
	I	II
2005	0.063 ^a 0.006	0.043 ^b 0.003
2006	0.061 ^a 0.005	0.043 ^b 0.005

I – mineral soil – habitat of ‘alternate meadow’

II – organic soil – habitat of ‘permanent meadow’

Statistically significant differences between selenium contents in grasses from different soil habitats were designated with different letters, *a-b* at $P \leq 0.01$.

level covering the sheep’s daily demand for this micronutrient (0.1 mg kg⁻¹ d.m. of fodder).

Significant differences were observed in the selenium content in meadow herbage from the two soil systems (Table 2). In grasses grown on mineral soils the concentration of selenium was even 1.5-fold higher than in grasses grown on organic soils. The results can support the dependence between concentrations of particular selenium forms in different soils and Se bio-availability. Similar observations were reported by other authors (MUNIER-LAMY et al. 2007).

Additionally, fodder used in winter (oat and meadow hay) was analyzed. The tests showed that the mean selenium concentration in oat was 0.053 and 0.043 mg kg⁻¹ d.m. in the first and second year, respectively, while meadow hay contained 0.071 and 0.068 mg Se kg⁻¹ d.m. in the two years. The research results reported by other authors led to the conclusion that the sheep’s demand for selenium is covered by fodder with about 0.1 mg Se kg⁻¹ d.m. content, and lower values should be considered as insufficient, possibly leading to diseases caused by selenium deficiency (CLOETE et al. 1999, BEYTUT, KARATAS 2002). The dietary requirement of Se by both ruminants and non-ruminants is 0.1 mg kg⁻¹ d.m. in a diet, while the maximum tolerable level of Se has been increased from 2 (NRC 1980) to 5 mg kg⁻¹ d.m. (NRC 2005). In diagnosed Se deficiencies, supplementation with inorganic Se sources is used, for example: sodium selenite, sodium selenate, as well as with an organic form of Se, e.g. selenized yeast (PAPPAS et al. 2008). Selenium may be administered to livestock as *ad libitum* Se mineral supplement, fertilization, injection, oral drenching, distribution in water or ruminal pellets (SZAREK et al. 1997).

Table 3 presents selenium concentrations in blood serum and milk of adult sheep and in blood serum of lambs. The selenium level in blood serum is one of the diagnostic clues used for an assessment of the degree of Se sup-

Table 3

Selenium content in sheep blood serum ($\mu\text{g dm}^{-3}$) and sheep milk (mg dm^{-3}) as well as in lamb blood serum ($\mu\text{g dm}^{-3}$) ($n = 32$; $\bar{x} \pm \text{SD}$)

Parameters	Sheep blood serum		Sheep milk		Lamb blood serum	
	2005	2006	2005	2006	2005	2006
Mean	29.33	29.21	0.018	0.017	24.57	22.56
SD	3.79	5.01	0.002	0.003	2.735	1.90
Min	25.42	20.32	0.015	0.012	20.12	18.32
Max	39.89	44.42	0.022	0.022	30.36	27.32

ply to an organism. The mean selenium concentrations in the blood serum of adult sheep reached similar values in the consecutive years: 29.33 in 2005 and 29.21 $\mu\text{g dm}^{-3}$ in 2006. Lower values were obtained when analyzing selenium concentrations in blood serum of sheep offspring (24.57 and 22.56 $\mu\text{g dm}^{-3}$, respectively). It should be noted that the concentrations values reported in the present work are in the range of 6-30 $\mu\text{g dm}^{-3}$, which PULS (1994) considers as indicative of selenium deficiency in sheep's blood serum. Selenium concentrations in blood serum above the deficiency threshold were found only in about 16% of adult sheep and about 6% of lambs in the first year of research and in 31% of dams in the second year of research. All the lambs tested in 2006 were selenium-deficient.

Significantly higher selenium levels in sheep's blood serum were obtained by other authors, who at the same time proved a large effect of the year of research and selenium level in fodder on this element in sheep's blood serum (LIPECKA et al. 2003).

Determination of low selenium concentrations in lamb blood serum can encourage search for ways to supplement this element in lambs. Selenium supplementation could reduce the risk of subclinical symptoms of alimentary muscle dystrophy in lambs whose selenium concentration in blood fell below 26 $\mu\text{g dm}^{-3}$ (SOBIECH, KULETA 2002). Juxtaposing the above results with ours seems to suggest that the lambs tested herein may be at risk of alimentary muscle dystrophy.

The low selenium concentrations determined in lambs' blood serum probably resulted from Se low levels in their mothers' milk (0.018 in 2005 and 0.017 mg dm^{-3} in 2006). The research showed the mean selenium levels in milk below the critical value of 0.02 mg dm^{-3} suggested by UNDERWOOD, SUTTLE (1999). Other authors detected similarly low selenium concentrations in milk (BRZÓSKA et al. 2000, VALLE et al. 2003, KHAN et al. 2006). Selenium deficiency found in sheep's milk during lactation can be caused by a relatively low content of this element in fodder (URSINI et al. 1999).

Our observations are also supported by the high correlation coefficients between selenium concentrations in blood serum and milk of mother sheep and in blood serum of lambs, presented in Table 4.

Table 4

Correlations between selenium concentrations in blood serum and milk of sheep and blood serum of lambs

Selenium level	Milk of mother sheep	Blood serum of lambs
In blood serum of mother sheep	0.94*	0.77*
In milk of mother sheep	-	0.72*

* statistically significant differences ($p \leq 0.01$)

Statistically, all the correlations turned out to be significant at $p \leq 0.01$. It is possible, then, that selenium level in blood serum and milk of sheep may be a good indicator in the assessment of Se supply to sheep. The correlations discovered herein are confirmed by other authors (KAUP 1998, ROCK et al. 2001, DAVIS et al. 2006, KARIMI-POOR et al. 2011).

Moreover, the dependencies described above can help to design supplementation of sheep's diets with preparations containing selenium in order to prevent deficiencies of this element in their offspring. This concept is supported by research of LACETERA et al. (1999), MUNIZ-NAVEIRO et al. (2006), ABDEL-GHANY et al. (2008), who have proven that selenium supplementation of sheep's and cows' diets during lactation results in a large increase of this element concentration in colostrum and milk, which protects newly-born lambs and calves from the risk of diseases related to selenium deficiency. Selenium in the form of seleno-amino acids, selenomethionine and selenocysteine are absorbed through the active amino acid transport mechanism and are more bioavailable than selenite or selenate (VANDELAND et al. 1994). In monogastric animals, the relative selenium absorption is greater than in ruminants (BOPP et al. 1982) and organic forms of selenine are more easily absorbed, thus being a better source of Se in tissues and blood (PEHRSON et al. 1999). The intestinal active absorption of selenium concerns selenomethionine and selenocysteine, and varies between 90-95 % for selenite and less than 10% less for selenate (LEE et al. 1996). In ruminants, the relative absorption ranges between 29-50% for selenate (SUTTLE, JONES 1989). The lower absorption in ruminants is due to microbial reduction of selenium form in the rumen to selenides and elemental selenium, which are not bioavailable (PETER et al. 1982). Some rumen microbes reduce selenium more efficiently, while others effectively incorporate it into selenium containing amino-acids. The incorporation proteins as well as systemic absorption can be competitively inhibited by natural methionine and cysteine (SERRA et al. 1996).

Recent research shows that positive effects of Se supplementation on productivity and breeding in animals depend on many factors, such as environmental, nutritional and management factors (source of Se, time and length of the treatment, presence of interfering elements, diet feeding pattern) (PALMIERI, SZAREK 2011).

CONCLUSION

The research proves that environmental conditions have a significant influence on the supply of selenium to adult sheep and their offspring. Selenium deficiency in particular trophic chain links can pose a serious threat to the health of animals fed homegrown feeds.

Administration of selenium to animals for therapeutic or prophylactic reasons should be preceded by a thorough analysis of its content in organs and tissues. Moreover, the kind of compound, dose and duration of the supplementation should be well considered. Another crucial factor is the determination of the Se content in fodders fed to animals.

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