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EFFECT OF SLURRY FERTILIZATION ON THE SELENIUM CONTENT AND CATALASE ACTIVITY IN LESSIVE SOIL

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Abstract

The aim of the present research was to determine the total selenium content in soil and plants from a microplot experiment with different nitrogen fertilization regimes, and to identify the relationships of the selenium content in soil and plants versus the soil catalase activity. The experiment was conducted in randomized blocks with three replications. The soil and plant samples were collected from a microplot experiment established at the IUNG in Pulawy. The soil was enriched with mineral nitrogen and with nitrogen supplied in slurry, both applied at doses of 100 kg N ha^{-1}. The total selenium content in soil under each of the crop rotation systems was no more than 0.2 mg kg^{-1}. Data from the references imply that the soil was deficient in selenium. The highest amount of selenium was under winter wheat and spring barley with undersown crop in crop rotation A, and in soil under maize crop rotation B. Slurry fertilization significantly stimulated the activity of catalase in soil, as compared with the control and mineral nitrogen fertilization treatments. The highest catalase activity – nearly double the control – was detected in soil under winter wheat in crop rotation A and under spring barley in crop rotation B; winter wheat, regardless of the type of crop rotation, accumulated on average 0.3 mg Se kg^{-1} d.w. in aerial parts and 0.344 mg Se kg^{-1} d.w. in roots. The highest amounts of selenium in the investigated parts of plants were reported in the control plots and in the plots with slurry fertilization. Mineral fertilization reduced selenium availability to plants. In both crop rotation systems, the highest bioaccumulation of selenium was noted in winter wheat roots from control plots, while the lowest one was detectetd in aerial parts of plants from the plots with slurry fertilization. Despite the fertilization applied, the selenium content in plant roots was higher than its content in aerial parts. The correlation analysis of the results on selenium concentration in soil and plants as well as the catalase activity of soil identified only a significant dependence between the total selenium content and catalase activity in soil from crop rotation B.

Key words: selenium, catalase activity, slurry, plants.

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WPŁYW NAWOŻENIA GNOJOWICĄ NA ZAWARTOŚĆ SELENU W GLEBIE I ROŚLINACH UPRAWIANYCH W ZMIANOWANIU

Abstrakt

Celem pracy było określenie wpływu nawożenia azotem w formie mineralnej oraz w formie gnojowicy od trzody chlewnej na całkowitą zawartość selenu w glebie oraz roślinach występujących w zmianowaniu na tle aktywności katalazy glebowej. Do badań wykorzystano próbki gleby i roślin z doświadczenia mikropoletkowego prowadzonego przez IUNG w Puławach. Zastosowano nawożenie azotem w postaci saletry amonowej w ilości 100 kg N ha^{-1} oraz w formie gnojowicy w ilości 100 kg N ha^{-1}. Doświadczenie wykonano w dwóch zmianowaniach z następującym doborem roślin: A: koniczyna – pszenica ozima – jęczmień jary z wsiewką; B: kukurydza – pszenica ozima – jęczmień jary. Zawartość selenu w glebie i roślinach oznaczono metodą Watkinsona z użyciem spektrofluorymetru F-2000 firmy Hitachi. Całkowita zawartość selenu w glebie, w obu zmianowaniach, nie przekraczała średnio 0,200 mg kg^{-1}. Nawożenie gnojowicą spowodowało istotny wzrost całkowitej zawartości selenu w glebie, w odniesieniu do jego zawartości w obiektach kontrolnych i nawożonych azotem w formie mineralnej. Najwyższą zawartość tego pierwiastka w zmianowaniu A wykazano w glebie pod uprawą jęczmienia jarego z wsiewką koniczyny, natomiast w zmianowaniu B – w glebie pod kukurydzą. Nawożenie azotem w formie gnojowicy istotnie stymulowało aktywność katalazy w badanej glebie w porównaniu z jej aktywnością w obiektach kontrolnych oraz w obiektach nawożonych azotem w formie mineralnej. W glebie ze zmianowania A najwyższą aktywność katalazy – ponad 2-krotnie wyższą – wykazano pod uprawą pszenicy ozimej, natomiast w zmianowaniu B – pod uprawą jęczmienia jarego. Pszenica ozima, niezależnie od rodzaju zmianowania, zgromadziła największą ilość selenu spośród roślin uprawianych w obu zmianowaniach: w częściach nadziemnych średnio 0,300 mg kg^{-1} s.m., natomiast w korzeniach 0,344 mg kg^{-1} s.m. Największą zawartość selenu w badanych roślinach wykazano w obiektach kontrolnych oraz w obiektach, na których stosowano azot w formie gnojowicy. Zastosowanie azotu w formie mineralnej ograniczyło pobieranie tego pierwiastka przez rośliny testowe. Wykazano, że najwyższą zdolność kumulacji selenu miały korzenie pszenicy ozimej pobrane z obiektów kontrolnych, natomiast najmniejszą – części nadziemne jęczmienia jarego z obiektów nawożonych azotem w formie gnojowicy. Niezależnie od zastosowanego nawożenia, zawartość selenu w korzeniach badanych gatunków roślin była większa od jego zawartości w częściach nadziemnych. W warunkach doświadczenia wykazano istotną korelację między aktywnością katalazy a całkowitą zawartością selenu w glebie.

Słowa kluczowe: selen, aktywność katalazy, gnojowica, rośliny.

INTRODUCTION

Selenium (Se) is an essential micronutrient for most plants, animals and people. This element builds selenoproteins, acts against oxidative stress, is involved in the production of thyroid hormones and contributes to the functioning of the immune system (Amouroux et al. 2001). The global distribution of selenium in water, air, soils and live organisms is uneven, so that some areas on the Earth are depleted of Se and pose serious health risks to both animals and humans, such as development of cancer and heart diseases. Soil-borne selenium is the primary source of human food Se. Most soils of the temperate humid climate zones and developed from sedimentary rocks contain low selenium levels, insufficient to produce food and animal feed plants with adequate Se content (Kabata-Pendias 1998). According to Winkel et al. (2012), selenium bioavailability is a function of the interaction among the prevailing geochemical parameters, i.e., pH and redox conditions, and soil properties, such as organic carbon, Fe hydroxide, and clay contents, and Se speciation. In soils, Se comes in a broad range of oxidation states: +6 in selenates, +4 in selenites, 0 in elemental Se, and -2 in inorganic and organic selenides. Selenate, which is poorly adsorbed on oxide surfaces thus being the most mobile Se form, can be expected to occur under high oxidative conditions. At low redox potential, it can be reduced to selenite, which has much higher adsorption affinity. It is strongly retained by ligand exchange on oxide surfaces, especially at low pH, which reduces its bioavailability (Hartikainen 2005, Patorczyk-Pytlik, Kulczycki 2009). The transformation of easily soluble selenates added to acidic or neutral soils into slightly soluble forms is relatively fast. In soil, processes of decomposition and synthesis of mineral and organic matter occur all the time, being monitored and activated by a variety of enzymes. Catalase (EC 1.11.1.6) is an iron porphyrin enzyme which catalyses very rapid decomposition of hydrogen peroxide to water and oxygen (Nelson, Cox 2000). The enzyme is widespread in nature, which explains its diverse activities in soil. The activity of catalase, alongside dehydrogenase, is tested to provide information on microbial activities in soil (Achuba, Peretiemo-Clarke 2008). A study of the Se content in soils and its forms can improve our understanding of Se cycling and balance in geosystems and their impact on health problems.

With the above in mind, the present investigation has been launched to study the total selenium content and catalase activity in soil fertilized with mineral nitrogen and swine slurry, and their impact on the bioavalability of this microelement to crops.

MATERIAL AND METHODS

Soil and plant samples were collected in March (winter wheat) and May (other plants) 2008, from a microplot experiment established by the Department of Plant Nutrition of the Institute of Soil Science and Cultivation in Pulawy. The soil, according to the FAO classification, was Haplic Luvisol with the texture of loamy sand and sandy loam. The experiment was conducted with two crop rotation systems: A (red clover - winter wheat - spring barley + undersown crop) and B (maize – winter wheat – spring barley). Some agrotechnical elements of plants under crop rotations are presented in Table 1. The soil was enriched with mineral nitrogen in the form of ammonium nitrate at the dose of 100 kg N ha^{-1} and in the form of pig slurry at the dose of 100 kg N ha^{-1}. The experiment was designed as split-plot trials with three replications on 1x1 m plots. Soil samples were air-dried and sieved through a 2 mm screen. The plant material was rinsed in deionized water to remove soil particles, separated into aerial biomass and roots, and dried.

Table 1

Some agrotechnical information about the plants in the crop rotations

Plant	Cultivar	Date of sampling	Development stage
Crop rotation A			
Red clover	Hruszowska	25.05.2008	BBCH 33
Winter wheat	Turnia	20.03.2008	BBCH 25
Spring barley with undersown crop	Justina	25.05.2008	BBCH 30
Crop rotation B			
Maize	Ainergy	25.05.2008	BBCH 15
Winter wheat	Turnia	20.03.2008	BBCH 25
Spring barley	Justina	25.05.2008	BBCH 30

The total selenium content in soils and plants was determined applying the Watkinson method (1966) on a Hitachi F-2000 spectrofluorometer. The samples were microwave-digested with concentrated nitric and perchloric acids. The different forms of selenium in the samples were reduced by boiling with 10% HCl. The selenium was complexed with 2,3-diaminonaphtalene (DAN) to yield the fluorescent compound, which was extracted with cyclohexane and read on a spectrofluorometer at the excitation and emission wave lengths of 376 and 519 nm, respectively. The analytical procedures gave satisfactory values for the standard reference material CRM024-050 Resource Technology Corporation (RTC), soil from Western US of a texture of loamy sand; Se 0.558 mg kg^{-1} (certified value 0.540 mg kg^{-1}). The certified reference material was included in each batch of samples for quality control. The bioaccumulation coefficient (BC) was calculated as a ratio of selenium concentration in plant aerial parts or roots to its amount in soil. Catalase activity (CAT) was measured applying the Johnson and Temple method (1964). Soil was incubated with hydrogen peroxide for 20 min at 20^{o}C. The remaining material, H_2O_2, not decomposed by catalase, was treated with *potassium* permanganate in the presence of H_2SO_4. To eliminate probable overestimation of the enzymatic activity due to chemical reduction of added H_2O_2, correction for autoclaved soil (0.1 MPa, 120°C, 30 min) was made. The results were expressed in mmol O_2 kg^{-1} h^{-1}. The soil samples were analysed for organic carbon by wet oxidation with potassium dichromate, total nitrogen following the Kjeldahl method and pH in 1M KCl potentiometrically. All the analyses were performed on triplicate samples. The multi-replication data from the analyses of soil and plant samples underwent statistical procedure consiting of variation analysis for one-factorial experiments carried out in a split-plot design. The data were analysed for treatment effect with the analysis of variance (Anova), after which the Tukey test at $p < 0.05$ was applied. The analysis was carried out using Statistica for Windows software.

RESULTS AND DISCUSSION

General properties of the soil are given in Table 2. The data on total Se concentrations of the soil (Table 3) indicate that slurry application significantly increased the total selenium content in the soil. The results show that the total selenium content did not exceed 0.2 mg kg^{-1}. Such low levels of selenium in soils suggested that plants growing on these soils were deficient in this microelement. According to Kabata-Pendias (1998), the mean total selenium content in soils worldwide is estimated at 0.44 mg kg^{-1}, while its background content in various soil groups ranges from 0.05 to 1.5 mg kg^{-1},

Table 2

Some general properties of the soil

Treatment	Crop rotation A			Crop rotation B		
	red clover	winter wheat	spring barley with undersown crop	maize	winter wheat	spring barley
Organic carbon (g kg^{-1})						
Control	7.55	6.98	6.87	6.95	6.55	6.28
$N_{MINERAL}$	6.98	6.86	6.91	6.55	6.83	6.63
N_{SLURRY}	7.68	7.55	7.86	7.45	7.59	7.56
Total nitrogen (g kg^{-1})						
Control	0.679	0.681	0.678	0.667	0.645	0.643
$N_{MINERAL}$	0.711	0.713	0.723	0.719	0.698	0.715
N_{SLURRY}	0.728	0.731	0.735	0.725	0.718	0.731
pH_{KCl}						
Control	5.5	5.4	5.4	5.3	5.4	5.3
$N_{MINERAL}$	5.3	5.3	5.2	5.2	5.2	5.2
N_{SLURRY}	5.4	5.4	5.3	5.3	5.3	5.5

being the lowest in Podzols and the highest in Histosols. Aro and Alfthang (1998) as well as Hartikainen (2005) claim that soils containing less than 0.5 mg Se kg^{-1} are likely to lead to crops and pastures having inadequate selenium concentrations (<0.05 mg kg^{-1} d.w.). In the soil from both crop rotations, the application of slurry significantly increased total selenium compared with the control and soil where mineral fertilization was applied. Sager (2007) stated that average total selenium in slurry ranged from 0.21 to 0.35 mg kg^{-1}. Thus, the increase in Se in slurry-treated soil could have been caused by the amount of this microelement in slurry. The supplementation of

Table 3

Total selenium content in the soil (mg kg^{-1})

Treatment (I factor)	Crop rotation (factor II)					
	A			B		
	cultivated plant (factor III)					
	red clover	winter wheat	spring barley with undersown crop	maize	winter wheat	spring barley
Control	0.118	0.121	0.133	0.161	0.125	0.122
$N_{MINERAL}$	0.100	0.131	0.144	0.144	0.125	0.124
N_{SLURRY}	0.124	0.152	0.186	0.177	0.153	0.152
Mean	0.114	0.135	0.154	0.161	0.134	0.133
Mean for treatment (factor I)						
	Control	0.129	$N_{MINERAL}$	0.126	N_{SLURRY}	0.157
Mean for crop rotation (factor II)						
	A		0.134	B	0.141	
Mean for plants (factor III)						
	Red clover/ maize	0.135	Winter wheat	0.134	Spring barley	0.143
$LSD_{0.05}$	I	0.004	interactions	II/I 0.003 I/II 0.005		
	II	0.002		I/III 0.005 III/I 0.003		
	III	0.002		II/III 0.005 III/II 0.002		

mineral fertilizers decreased the total selenium content in soil, which may be explained in the following way: increased mineral fertilization causes greater root growth and provides plants with a larger volume of soil from which Se can be extracted, an explanation supported by Blagojevic et. al. (1980).

Generally, the organic addittives increased enzyme activities of soil and their application to soil can improve soil structure and stimulate microbiological activity (Jezierska-Tys, Frąc 2009, Jezierska-Tys et al. 2011). Catalase activity of the soil from crop rotation B was significantly higher than in soils treated with mineral nitrogen and from control plots (Table 4). In crop rotation A, CAT activity was significantly stimulated by slurry application only in the soil under winter wheat, as compared to that of control. The correlation analysis of the results on selenium concentration in soil and plants as well as the catalase activity of soil yielded a significant relationship only between the total selenium content and catalase activity in soil from crop rotation B (0.56). Our results coincide with the literature data reviewed by Samuel (2010) and with our earlier studies (Borowska, Koper 2011).

Variation of the Se status in humans largely depends on diet. Plant foods are the major dietary sources of Se in most countries around the world, followed by meat and seafood (Winkel et al. 2012). As shown in Tables 5 and 6, winter wheat accumulated in an average 0.3 mg Se kg^{-1} d.w. in aerial organs and 0.344 mg Se kg^{-1} d.w. in roots, regardless of the crop rotation. The highest amounts of selenium in the investigated parts of plants were

Table 4

Catalase activity (CAT) in the soil (mmol O_2 kg^{-1} h^{-1})

Treatment (I factor)	Crop rotation (II factor)					
	A			B		
	cultivated plant (III factor)					
	red clover	winter wheat	spring barley with undersown crop	maize	winter wheat	spring barley
Control	74.10	163.19	57.34	52.04	75.86	160.54
$N_{MINERAL}$	62.63	157.89	60.86	53.81	70.57	149.96
N_{SLURRY}	71.45	171.13	54.69	67.92	82.92	166.72
Mean	69.39	164.07	57.63	57.92	76.45	159.07
Mean for treatment (factor I)						
	Control	97.16	$N_{MINERAL}$	92.61	N_{SLURRY}	102.46
Mean for crop rotation (factor II)						
		A	97.02	B	97.80	
Mean for plants (factor III)						
	Red clover/ maize	63.64	Winter wheat	120.25	Spring barley	108.34
$LSD_{0.05}$	I	0.066	interactions	II/I 0.034 I/II 0.068		
	II	0.020		I/III 0.069 III/I 0.040		
	III	0.023		II/III 0.033 III/II 0.032		

Table 5

Selenium content in aerial parts of plants (mg kg^{-1} d.w.)

Treatment (I factor)	Crop rotation (II factor)					
	A			B		
	cultivated plant (III factor)					
	red clover	winter wheat	spring barley with undersown crop	maize	winter wheat	spring barley
Control	0.132	0.318	0.145	0.120	0.330	0.140
$N_{MINERAL}$	0.125	0.287	0.132	0.124	0.288	0.129
N_{SLURRY}	0.135	0.265	0.128	0.194	0.324	0.118
Mean	0.131	0.290	0.135	0.146	0.314	0.129
Mean for treatment (factor I)						
	Control	0.196	$N_{MINERAL}$	0.181	N_{SLURRY}	0.195
Mean for crop rotation (factor II)						
		A	0.185	B	0.196	
Mean for plants (factor III)						
	Red clover/ maize	0.139	Winter wheat	0.301	Spring barley	0.132
$LSD_{0.05}$	I	0.013	interactions	II/I 0.003 I/II 0.013		
	II	0.002		I/III 0.013 III/I 0.004		
	III	0.003		II/III 0.004 III/II 0.003		

Table 6

Selenium content in plant roots (mg kg^{-1} d.w.)

Treatment (I factor)	Crop rotation (factor II)					
	A			B		
	cultivated plant (factor III)					
	red clover	winter wheat	spring barley with undersown crop	maize	winter wheat	spring barley
Control	0.129	0.403	0.151	0.125	0.399	0.143
$N_{MINERAL}$	0.126	0.298	0.141	0.132	0.294	0.122
N_{SLURRY}	0.141	0.328	0.132	0.263	0.344	0.123
Mean	0.132	0.343	0.141	0.173	0.346	0.129
Mean for treatment (factor I)						
	Control	0.225	$N_{MINERAL}$	0.185	N_{SLURRY}	0.221
Mean for crop rotation (factor II)						
		A	0.205	B	0.215	
Mean for plants (factor III)						
	Red clover/ maize	0.152	Winter wheat	0.344	Spring barley	0.134
LSD $_{0.05}$	I	0.003	interactions	II/I 0.002 I/II 0.004		
	II	0.001		I/III 0.004 III/I 0.002		
	III	0.001		II/III 0.002 III/II 0.002		

recorded in control plots and in the plots with slurry fertilization. These results confirm that among common cereal crops, wheat is the most efficient accumulator of Se (wheat> rice> maize> barley> oats). Wheat is also the most important Se source for humans. Thus, this cereal is an obvious target crop for agronomic biofortification to increase the dietary Se intake (Broadley et al. 2006).

According to Terry et al. (1992) and Munier-Lamy et al. (2007), the value of bioaccumulation coefficient (BC) reflects the plant's capacity for the uptake of nutrients from soil, while telling us about the amount and rate of nutrient translocation from soil solution to aerial organs of plants. The bioaccumulation coefficients (BC) of selenium demonstrated that aerial parts and roots of winter wheat and spring barley from both crop rotation systems absorbed selenium more easily from soil of the control plots or plots where mineral fertilization was applied (Figures 1 and 2). However, the BC factor calculated for maize indicated an opposite tendency, namely the highest amounts were recorded for plants from plots fertilized with slurry (Figure 2).

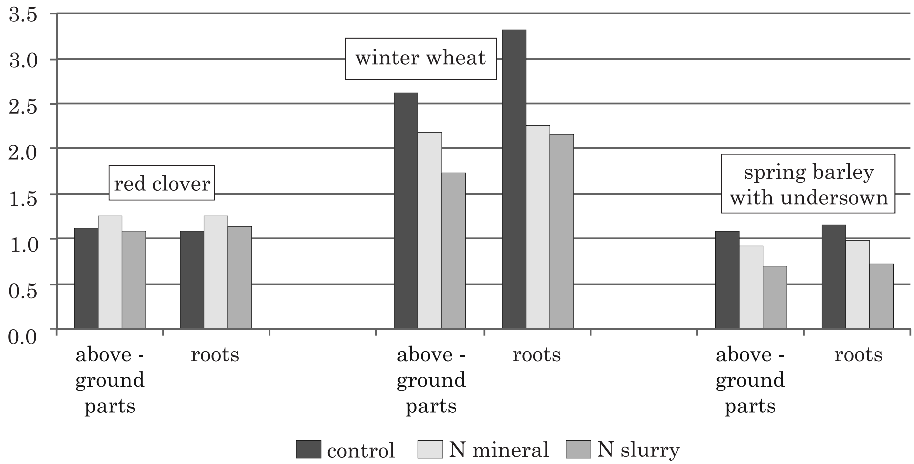

Fig. 1. Bioaccumulation coefficients (BC) for plants in crop rotation A

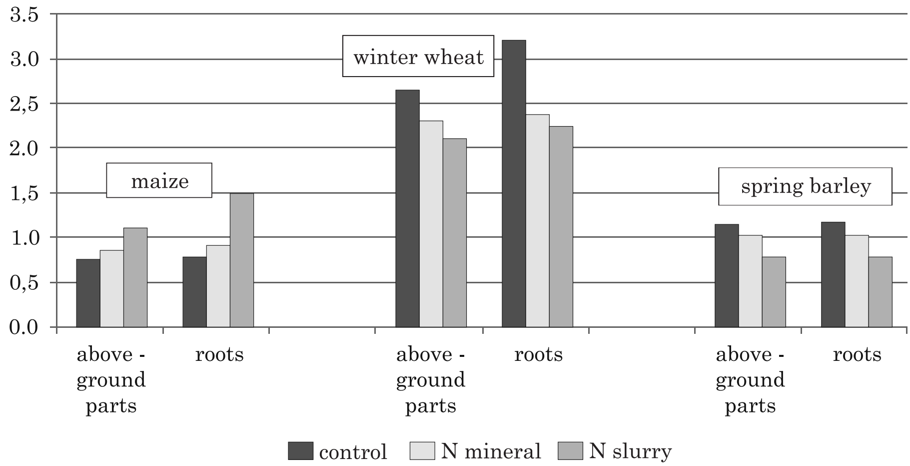

Fig. 2. Bioaccumulation coefficients (BC) for plants in crop rotation B

Winkel et al. (2012) mentioned that a large amount of organic matter can lead to reducing conditions, where both biotic and abiotic mechanisms induce the formation of elemental selenium or metal selenides. Elemental Se is not soluble in water and it is classified as unavailable to biota. The accumulation of selenium by plants may be affected by competition or interactions with sulphur fertilization, as well as by other interactions in soil during the adsorption, transport and transformation processes (Hawkesford, Zhao 2007).

CONCLUSIONS

1. The total selenium content in soil did not exceed 0.2 mg $kg^{-1.}$ In the light of the relevant literature, the analyzed soil is deficient in this microelement.

2. Slurry fertilization significantly stimulated catalase activity in the investigated soil in comparison with the control and soil from plots fertilized with mineral nitrogen.

3. Regaardless of the crop rotation system, winter wheat accumulated in 0.3 mg Se kg^{-1} d.w. in aerial organs and 0.344 mg Se kg^{-1} d.w. in roots, on aveerage. The highest amounts of selenium in the investigated plant parts were obtained from control plots and the plots with slurry fertilization. Mineral fertilization reduced selenium availability to plants. Irrespective of the applied fertilization, the selenium content in plant roots was higher than in aerial organs.

4. The correlation analysis of the results on selenium concentrations in soil and plants as well as the catalase activity in soil showed a significant dependence only between the total selenium content and catalase activity in soil from crop rotation B.

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