

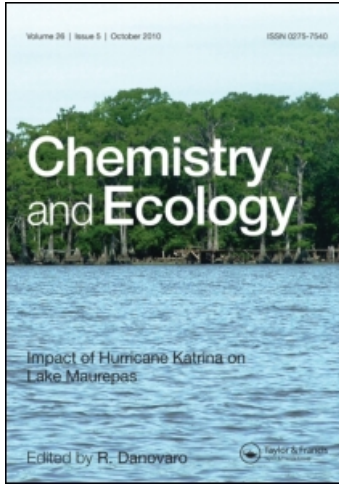
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K. Borowska^a; J. Koper^a

^a Department of Biochemistry, University of Technology and Life Science, Bydgoszcz, Poland

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The effect of long-term organic–mineral fertilisation on selenium content and chosen oxidoreductases activity under winter wheat cultivation

K. Borowska* and J. Koper

Department of Biochemistry, University of Technology and Life Science, Bydgoszcz, Poland

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The aim of the research was to determine the effect of fertilisation applying various doses of manure and nitrogen on the selenium content of soil against a background of soil catalase and dehydrogenase activity. The experiment was conducted using a crop rotation system of: potato–winter wheat + intercrop–spring barley + undersown and red clover + grasses. The soil was treated with organic fertilisation in the form of manure under potato at doses of 0, 20, 40, 60 and 80 t · ha⁻¹, and with nitrogen at doses of 0, 40, 80 and 120 kg N · ha⁻¹. A supplement of manure at 80 t · ha⁻¹ resulted in the greatest significant increase in soil total selenium content of ~100% compared with soil from control plots. We observed that interaction of the highest manure and nitrogen doses resulted in a significant decrease in the total selenium content of the soil. Selenium content in soil was statistically highly correlated with organic carbon content. Analysis of the correlation showed a significant dependence between the total selenium content in the soil and the activity of soil catalase and dehydrogenase.

Keywords: selenium; soil; catalase; dehydrogenases; organic–mineral fertilisation

1. Introduction

Selenium (Se) is considered to be an essential trace element for humans, animals and higher plants, however, at high levels, selenium can be toxic, causing deformities, decreased hatchling survival and death to aquatic wildlife, and larger animals with increased exposure [1]. The concentration of Se in plants and animals is strongly correlated with its concentration in soils. Plants and products derived from plants transfer Se taken up from the soil to humans [2]. Thus the Se cycle begins and ends with soil, and the chemical forms (dissolved in soil solution, adsorbed on oxide surfaces, fixed in the mineral lattice) and concentrations of Se in soil determine its bioavailability and thus the need for dietary supplementation [3]. Many factors, for example, soil pH, *Eh*, weathering of parent materials and microbial intervention, should be taken into account to explain the entry of Se into the biogeochemical cycles in which the soil–plant system is a key compartment [1]. Selenium occurs mainly in soil in insoluble elemental (Se⁰) and selenide (Se(II)) forms, but in oxidising environments such as aerobic soils, Se is converted to soluble

*Corresponding author. Email: kborowska56@o2.pl

selenite (Se(IV)) and selenate (Se(VI)) forms, selenate being predominant at alkaline pH. At low redox potential, selenate can be reduced to selenite, which has a much higher adsorption affinity. It is strongly retained by ligand exchange on oxide surfaces, especially at low pH, which reduces its bioavailability. Volatile Se is lost to the atmosphere from plants or through microbial activity, but Se also returns to the soil from the atmosphere with precipitation [4]. Selenium is one of the trace elements strongly affected by microbiologically mediated redox processes, which influence its solubility, and consequently its mobility, bioavailability and uptake in the soil–plant system [5]. In soil, the processes of decomposition and synthesis of mineral and organic matter occur all the time, and are monitored and activated by a variety of enzymes. All these processes make up soil metabolism, which is crucial for soil fertility maintenance and preservation [6]. Many authors have indicated a strong influence of Se on the activities of oxidoreductase enzymes, such as catalase, glutathione peroxidase and superoxide dismutase. The literature provides abundant information on the role Se plays in animals. However, there have been relatively few reports on the contribution of Se to biochemical processes in soil and plants [7]. Such processes depend on the availability of selenium. The aim of this research was to determine the effect of fertilisation with various doses of manure and nitrogen on the Se content in soil against a background of soil catalase and dehydrogenase activity.

2. Materials and methods

Soil samples were taken from a long-term experiment established in 1980 at the Experimental Station Grabow of the IUNG Pulawy on the soil classified as Albic Luvisols. The experiment was conducted with crop rotation system: potato–winter wheat + intercrop–spring barley + undersown and red clover + grasses. The soil was treated with organic fertilisation in the form of manure under potato at doses of 0, 20, 40, 60 and 80 t · ha⁻¹ and with nitrogen at doses (kg N · ha⁻¹) of: N0, 0; N1, 40; N2, 80; and N3, 120. Soil samples were taken in May 2002 from a depth of 5–15 cm under winter wheat cultivation in the 22nd year of the experiment. Total selenium content in soil was determined by the method of Watkinson [8] using a Hitachi F-2000 spectrofluorometer. Soil samples were microwave digested with concentrated nitric acid and peroxide water. The different forms of Se in the samples were reduced by boiling with 10% HCl. The Se was complexed with 2,3-diaminonaphthalene (DAN) to give the fluorescent compound, which was extracted with cyclohexane and read on a spectrofluorometer at excitation and emission wavelengths of 376 and 519 nm, respectively. Dehydrogenase activity (DHA) was assayed using the method of Casida et al. [9]. Determination of DHA in soils is based on the use of soluble tetrazolium salts as artificial electron acceptors, which are reduced to red-coloured formazans, extracted and then determined colorimetrically. Catalase activity (CAT) was determined using the method of Johnson and Temple [10]. The samples were analysed for granulometric composition according to the Bouyoucoss–Cassagrande method, organic carbon by wet oxidation with potassium dichromate, and pH in distilled water and 0.1 M KCl potentiometrically. All analyses were performed on triplicate samples. The statistical analysis was performed using Statistica software at the 0.05 level of significance.

3. Results and discussion

The general properties of the soil under study are given in Table 1. The investigated soil had a texture of light loamy sand and was a very good rye complex – IVa of soil valuation class. The pH values of the soil were in the slightly acidic range, 5.4–6.9. The application of manure resulted in the highest amounts of organic carbon in the soil, which were obtained from plots manured at

Table 1. General properties of the soil under study.

Sample symbol	Soil particle size fraction (%)		pH		C_{org} ($g \cdot kg^{-1}$)	N_{tot} ($g \cdot kg^{-1}$)
	<0.02 (mm)	<0.002 (mm)	in H_2O	in KCl		
0-N0	18	6	6.8	5.7	7.9	0.889
0-N1	17	6	6.8	5.8	8.4	0.966
0-N2	19	8	6.8	5.8	8.4	0.924
0-N3	17	7	6.7	5.7	8.3	0.966
20-N0	14	5	6.8	5.8	8.9	0.980
20-N1	16	8	6.5	5.7	8.5	0.956
20-N2	15	6	6.5	5.6	8.7	0.956
20-N3	18	7	6.5	5.4	9.3	0.956
40-N0	19	8	6.3	5.6	9.5	0.991
40-N1	17	6	6.6	5.5	9.3	1.012
40-N2	13	5	6.6	5.6	10.4	0.942
40-N3	19	6	6.6	5.5	10.3	0.987
60-N0	16	5	6.7	6.0	10.4	0.931
60-N1	16	6	6.4	5.8	10.2	0.970
60-N2	19	7	6.7	5.8	10.1	0.956
60-N3	20	6	6.7	5.9	9.8	0.921
80-N0	15	4	6.9	5.9	10.4	0.959
80-N1	16	6	6.9	5.7	10.1	0.966
80-N2	15	5	6.7	5.9	10.3	0.980
80-N3	14	5	6.8	5.9	10.4	1.005

doses of 60 and 80 $t \cdot ha^{-1}$. The Se content of control plots ranged from 0.096 to 0.112 $mg \cdot kg^{-1}$. Another increase in manure doses by 20 $t \cdot ha^{-1}$ significantly increased the total Se content in the soil by 55, 74, 84 and 101%, respectively, compared with the no-manure treatment (Figure 1). The Se content of the manure applied in the experiment was 2.28 $mg \cdot kg^{-1}$ fresh weight. Thus the increase in Se in manured soil may have been caused by the amount of this microelement in farmyard manure. According to data from the literature [11–13], Se is present in various manures in amounts varying from 0.32 to 2.4 $mg \cdot kg^{-1}$. We observed that interaction of the highest manure and nitrogen doses resulted in a significant decrease in the total Se content in soil. Supplementation

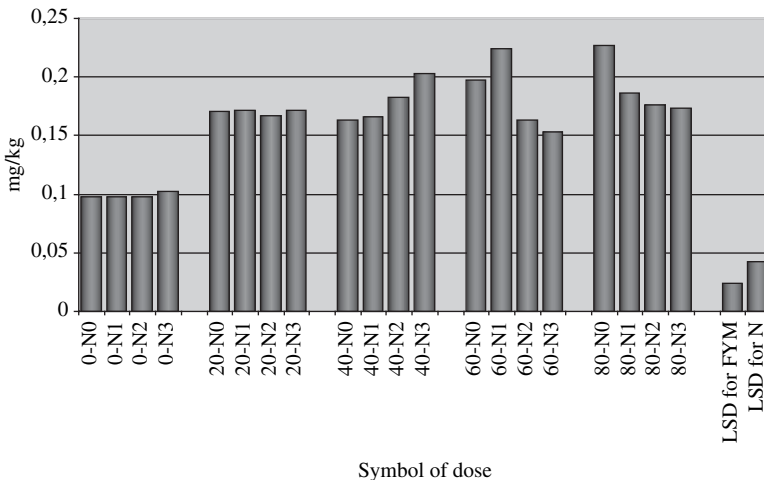


Figure 1. Total selenium content in soil.

of mineral fertilisers caused a decrease in the total Se content in soil, which may be explained as follows: increased mineral fertilisation causes greater root growth and provides plants with a larger volume of soil from which Se can be extracted, as confirmed by Blagojevic et al. [11]. Similar data were obtained by Gissel-Nielsen et al. [14], who examined the influence of N, P and S on the uptake of selenite by barley and a complex interaction between the three fertiliser anions. Nitrogen application decreased the Se concentration, which was to some extent only a dilution effect due to increased yield. The average Se content in subsurface arable soil horizons around the world is $0.33 \text{ mg} \cdot \text{kg}^{-1}$, and for Polish soils is $0.27 \text{ mg} \cdot \text{kg}^{-1}$ [1]. In Finnish soils, which also show some Se deficit, a level of $0.2\text{--}0.3 \text{ mg} \cdot \text{kg}^{-1}$ is considered too low [15]. The Se concentration in agricultural products is very low in many areas of the world, including large areas of Central and Northern Europe. This has traditionally been attributed to a poor supply of Se from the soil, and ultimately the underlying geology. For example, in German agricultural soil a mean Se concentration of $0.12 \text{ mg} \cdot \text{kg}^{-1}$ was found [2], whereas in Norwegian farmland soils, the Se concentration in general is very low ($0.3 \text{ mg} \cdot \text{kg}^{-1}$ on average) [16]. Comparison of the results offered in the literature suggests that the soils researched were poor in that element. Total Se content in soil was statistically highly correlated with organic carbon content. This agrees with our earlier findings [17] and those of other authors [1,18].

A significant increase in catalase activity of almost 40% was recorded in soil fertilised with manure at 20 and $40 \text{ t} \cdot \text{ha}^{-1}$, compared with the treatment without manure (Figure 2). Manure fertilisation at 60 or $80 \text{ t} \cdot \text{ha}^{-1}$ mostly increased catalase activity in the soil under study. The increase was 51 and 50%, respectively, compared with controls. Nitrogen fertilisation did not have a significant effect on catalase activity in the soil. Nowak et al. [14] noted that adding selenium to soil enhanced the activation of soil and plant catalase. The correlation analysis demonstrated a significant dependence between the content of total selenium (0.58^*) and organic carbon (0.56^*) in soil and the activity of soil catalase. Many authors mention that catalase activity is significantly correlated with organic carbon content and is usually higher in humus horizons of soils [10,17].

Mean dehydrogenase activity in soil from control plots was $0.034 \mu\text{g TPF} \cdot \text{g}^{-1} \cdot 24 \text{ h}^{-1}$ (Figure 3). We observed an increase in enzymatic activity with increasing doses of manure. Dehydrogenase activity in soil from a plot manured at the highest dose was approximately two times higher than dehydrogenase activity from control samples. The application of nitrogen resulted in an increase in dehydrogenase activity of 31, 25 and 9% compared with controls. We found a very significant correlation between dehydrogenase activity and organic carbon content in the soil

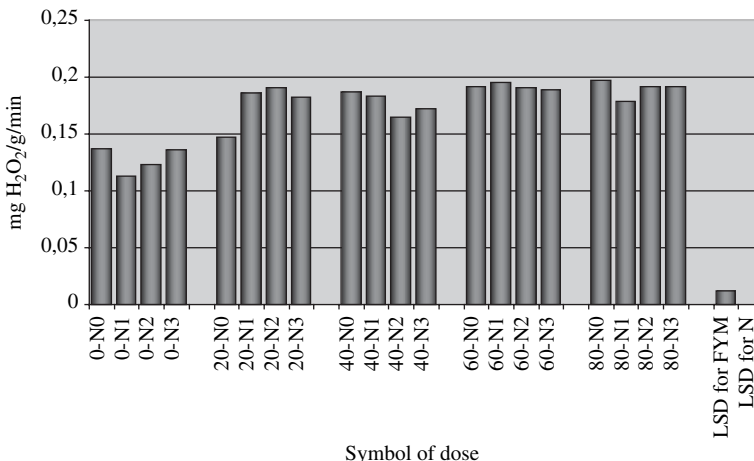


Figure 2. Catalase activity in the soil under study.

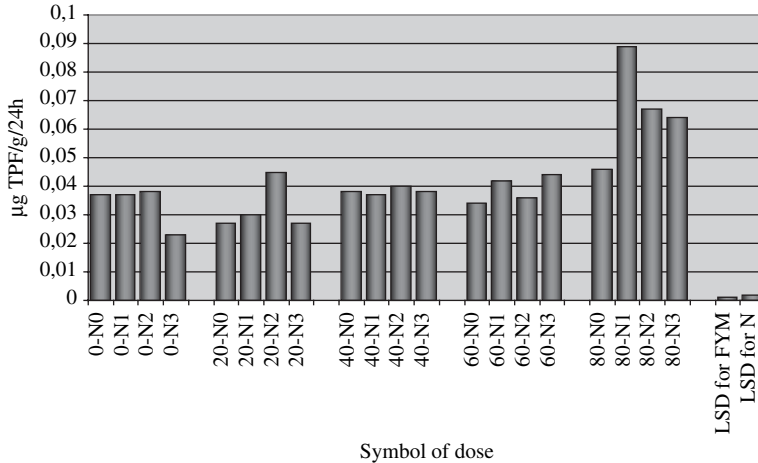


Figure 3. Dehydrogenase activity in the investigated soil.

under study. According to Hadas and Kautsky [20], mineral fertilisation increased dehydrogenase activity in mineral soil and the application of manure in organic soil also increased enzymatic activity in comparison with mineral fertilisation. In soil studied by Spychaj-Fabisiak and Smolinski [19], nitrogen fertilisation stimulated an increase in dehydrogenase activity. Moreover, the authors found that dehydrogenase activity in soil increased with increasing numbers of microorganisms and their metabolism rate, which enabled the use of organic carbon stock. According to Brzezinska et al. [21], dehydrogenase plays an essential role in the initial stages of the oxidation of soil organic matter by transferring hydrogen and electrons from substrates to acceptors. Many authors [6,7,21,22] have suggested that soil enzymes play essential roles in soil processes, such as nutrient cycling and energy transformation, by catalysing numerous chemical, physical and biological reactions, and can be used as indices of soil fertility and soil health. Any management practice that influences microbial communities in the soil may be expected to produce changes in soil enzyme activity, in agreement with our findings. Previous studies have demonstrated a significant effect of soil dehydrogenase and catalase activity, and organic matter content on the selenium status of soil.

4. Conclusions

There was a significant effect of organic and mineral fertilisation on the total Se content in the investigated soil. A manure supplement at $80 \text{ t} \cdot \text{ha}^{-1}$ resulted in the greatest significant increase in soil total Se content of $\sim 100\%$ compared with soil from control plots. The soil Se content was statistically highly correlated with the organic carbon content. We observed that interaction of the highest manure and nitrogen doses resulted in a significant decrease in total Se content in soil.

The analysis of correlation showed a significant dependence between the total selenium content in the soil and the activity of soil catalase and dehydrogenase.

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