ENZYMATIC ACTIVITY OF NICKEL-CONTAMINATED SOIL

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Abstract

A pot experiment was performed to determine the effect of soil contamination with nickel, applied at a dose of 100, 200, 300 and 400 mg kg⁻¹, on the activity of dehydrogenases, urease, acid phosphatase and alkaline phosphatase. The impact of nickel on the enzymatic activity of soil was studied on samples of heavy loamy sand and light silty loam. The experiment was conducted in two series: in the first one soil was cropped to yellow lupine, and in the second one it was left uncropped. Soil samples were analyzed on day 14, 28, 42 and 56.

It was found that soil contamination with nickel reduced the activity of all the enzymes. This negative influence was most noticeable in the case of dehydrogenase. The activity of urease and alkaline phosphatase was higher in light silty loam, while the activity of dehydrogenases and acid phosphatase was higher in heavy loamy sand. The activity of dehydrogenases and urease was higher in soil cropped to yellow lupine, whereas the activity of acid phosphatase and alkaline phosphatase was higher in uncropped soil. Yellow lupine was sensitive to excessive amounts of nickel in the soil, and partly alleviated the adverse impact of this heavy metal on urease activity, but did not reduce its inhibitory effect on the other enzymes.

Key words: nickel, enzymatic activity, soil, yellow lupine.

AKTYWNOŚĆ ENZYMATYCZNA GLEBY ZANIECZYSZCZONEJ NIKLEM

Abstrakt

W doświadczeniu wazonowym określono wpływ zanieczyszczenia gleby niklem w dawkach 100, 200, 300 i 400 mg kg⁻¹ na aktywność dehydrogenaz, ureazy, fosfatazy kwaśnej oraz fosfatazy alkalicznej. Wpływ niklu na aktywność enzymatyczną badano w dwóch gatunkach gleb: piasku gliniastym mocnym oraz glinie lekkiej pylastej. Badania przeprowadzono w dwóch se-

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riach, tj. na glebie obsianej łubinem żółtym i nie obsianej. Próbki glebowe analizowano: w 14., 28., 42. i 56. dniu.

Stwierdzono, że zanieczyszczenie gleby niklem spowodowało zmniejszenie aktywności wszystkich badanych enzymów. Najbardziej niekorzystnie nikiel wpływał na aktywność dehydrogenaz. Wyższą aktywność ureazy i fosfatazy alkalicznej stwierdzono w glinie lekkiej pylastej niż w piasku gliniastym mocnym, natomiast dehydrogenaz i fosfatazy kwaśnej – w piasku gliniastym mocnym. Wyższą aktywność dehydrogenaz i ureazy odnotowano w glebie obsianej łubinem żółtym, natomiast fosfatazy kwaśnej i fosfatazy alkalicznej – w glebie nie obsianej. Łubin żółty okazał się wrażliwy na nadmiar niklu w glebie. Jego uprawa częściowo łagodziła niekorzystny wpływ niklu na ureazę, lecz nie zmniejszała hamującego oddziaływania tego pierwiastka na pozostałe enzymy.

Słowa kluczowe: nikiel, aktywność enzymatyczna, gleba, łubin żółty.

INTRODUCTION

Enzymatic activity, similarly to total microbial count, is an important indicator of soil quality since it provides information about the specific metabolic activity and functions of soil microbial communities (Kizilkaya et al. 2004, Bielińska, Tomaszewicz 2006). In general, enzymes may be divided into biotic and abiotic ones. The first group includes enzymes synthesized by plants during metabolic processes and soil organisms, while the other comprises enzymes forming complexes with minerals or organic colloids as well as those found in dead plant cells or microbial cells (Guibaud et al. 2005). The enzymes produced as a result of microbial metabolism may differ with respect to some specific properties, e.g. preferences for a given type of substrate, pH, susceptibility to inhibitors and resistance to proteolysis (Criquet et al. 2002).

Studies on soil microbial activity involve enzymes which are known to respond to stress factors. Their activity is dependent on total microbial count (Nowak et al. 2000, Antil et al. 2001). Enzymatic activity may be stimulated if the concentration of heavy metals in the soil is low, and irreversibly inhibited if their content in the soil increases several times (Klose, Tabatabai 1999). This depends, to the greatest extent, on the physicochemical properties of the soil, in particular on its pH (Vepsäläinen 2001).

The nickel content of soil has increased in recent years (Baralkiewicz, Siepak 1999). The origin of nickel is allochthonous and its presence is correlated with the degree of industrialization and motorization as well as with the use of means of agricultural production containing nickel, such as some mineral fertilizers, wastewater and crop protection chemicals (Oliveira, Pampulha 2006). According to Terelak et al. (2000), 95% of all soils used for agricultural purposes in Poland have a natural nickel content (0°) and in 4.23% of soils the concentration of this heavy metal is increased (I°). In 0.34% of soils nickel contamination is slight (II°), in

0.06% – medium (III°) and in 0.01% – severe (IV°). No arable land in Poland is characterized by very severe (V°) nickel contamination.

The aim of the present study was to determine the effect of soil contamination with nickel on the activity of such enzymes as dehydrogenases, urease, acid phosphatase and alkaline phosphatase.

MATERIAL AND METHODS

The experiment was conducted in a greenhouse. Plastic pots were filled with 3 kg of typical brown soil developed from heavy loamy sand and 3 kg of typical brown soil developed from light silty loam. The soil material was taken from the organic layer. The detailed characteristics of soils are given in Table 1.

Table 1
Tabela 1
Some physicochemical properties of soils used in the experiment
Niektóre właściwości fizykochemiczne gleb użytych w doświadczeniu

Soil species Gatunek		ometric com granulometric (mm)		pH _{KO}	Hh	S	Corg (g kg ⁻¹)
gleby	1 - 0.1	0.1 - 0.02	< 0.02	- 181		ol(+) – gleby)	(g kg ¹)
hls – pgm	66	17	17	6.90	11.25	89.30	7.50
lsl – glp	42	32	26	7.00	8.77	159.00	11.15

 $\label{eq:lossy} \begin{array}{l} hls-heavy\ loamy\ sand-pgm-piasek\ gliniasty\ mocny;\ lsl-light\ silty\ loam-glp-glina\ lekka\ pylasta;\ Hh-hydrolytic\ acidity-kwasowość\ hydrolityczna;\ S-sum\ of\ exchangeable\ basic\ cations-suma\ wymiennych\ kationów\ zasadowych;\ Corg-organic\ carbon\ content-zawartość\ wegla\ organicznego \end{array}$

Variable experimental factors were as follows:

- 1) dose of nickel in the form of $\rm NiSO_4 \cdot 7H_2O~(mg~Ni^{2+}~kg^{-1}~soil):$ 0, 100, 200, 300 and 400;
- 2) soil species: heavy loamy sand, light silty loam;
- 3) method of soil management: soil cropped to yellow lupine and uncropped soil;
- 4) time of biochemical analysis (days): 14, 28, 42, 56.

The tested crop, yellow lupine cv. Markiz (5 plants per pot), was collected at the flowering stage and dry matter yield was determined. All treatments were regularly fertilized with macro- and micronutrients, as follows (pure component per mg kg⁻¹ soil): P - 66 [KH₂PO₄], K - 125 [KH₂PO₄ + KCl], Mg - 20 [MgSO₄·7H₂O], Cu - 5 [CuSO₄·5H₂O], Zn - 5

[ZnCl $_2$], Mn - 5 [MnCl $_2\cdot 4H_2O$], Mo - 5 [Na $_2MoO_4\cdot 2H_2O$], B - 0,33 [H $_3BO_4$]. Over the entire experimental period soil moisture content was 60% of the capillary water capacity of the soil. The experiment was carried out in six replications for soil cropped with yellow lupine, and in three replications for uncropped soil.

On day 14, 28, 42 and 56 (factor 4) soil samples were assayed for the activity of dehydrogenases (Deh) – by the Lenhard method modified by Öhlinger (1996), urease (Ure) – by the method developed by Alef and Nannipieri (1998), acid phosphatase (Pac) and alkaline phosphatase (Pal) – as described by Alef et al. (1998).

The results were processed statistically by Duncan's multiple range test and a four-factorial analysis of variance. Statistical analysis was performed with the use of Statistica (Statsoft, INC. 2003).

RESULTS AND DISCUSSION

The activity of dehydrogenases (Table 2) depended on the nickel dose, soil species, method of soil management and time of analysis. It was found to be substantially higher in heavy loamy sand than in light silty loam. Until day 28 of the experiment it remained at a higher level in bare soil than in soil cropped with yellow lupine. This trend was no longer observed on day 42, and on day 56 - during yellow lupine harvest - higher activity of dehydrogenases was recorded in samples of cropped soil. Soil contamination with nickel significantly reduced the activity of dehydrogenases, which was negatively correlated with the degree of nickel contamination in both types of soil. The activity of urease (Table 3), similarly to the activity of dehydrogenases, was negatively correlated with the degree of soil contamination with nickel. Regardless of the date of analysis, the activity of this enzyme was higher in soil cropped with yellow lupine compared to bare soil, and in light silty loam compared to heavy loamy sand. Yellow lupine partly alleviated the adverse impact of nickel on urease activity, but there was no such relationship in the case of dehydrogenases, acid phosphatase (Table 4) or alkaline phosphatase (Table 5). Both acid and alkaline phosphatase were most active in soils not contaminated with nickel, and the least active - in soils containing the largest amounts of this element (400 mg kg⁻¹). The coefficients of correlation between the degree of soil contamination with nickel and enzymatic activity were -0.84 to -0.99 for acid phosphatase, and -0.89 to -0.99 for alkaline phosphatase. The activity of acid phosphatase was higher in heavy loamy sand, and the activity of alkaline phosphatase was higher in light silty loam.

Soil contamination with nickel had an adverse effect not only on enzymatic activity, but also on the yield of yellow lupine (Table 6). The val-

Table 2 Tabela 2

Effect of soil contamination with nickel on the activity of dehydrogenases in 1 kg d.m. of soil $(cm^3 H_2 \text{ kg}^{-1} d.m. d^{-1})$ Wptyw zanieczyszczenia gleby niklem na aktywność dehydrogenaz w 1 kg s.m. gleby $(cm^3 H_2 \text{ kg}^{-1} s.m. d^{-1})$

Ni dose (mo ko-1 of soil)				Time of ans Termin an	Time of analysis (days) Termin analizy (dni)			I	16	18
Dawka Ni (mg kg ⁻¹ gleby)		14	2	28	4	42	E2	56		
	IX-	IX+	IX-	Ι Χ +	IX-		IX-		IX-	[X+
			Heavy loan	ıy sand – Pi	Heavy loamy sand – Piasek gliniasty mocny	y mocny				
0	5.35	4.65	5.92	3.65	5.52	5.91	3.04	4.31	4.96	4.63
100	4.43	3.52	5.21	3.30	4.31	4.70	2.68	4.28	4.16	3.95
200	4.16	3.29	4.74	2.99	3.63	3.40	1.86	3.31	3.60	3.25
300	3.71	2.92	4.76	2.48	3.20	3.12	1.64	2.64	3.33	2.79
400	3.03	2.43	3.90	2.11	2.69	2.73	1.38	1.58	2.75	2.21
î.	-0.98	96:0-	-0.96	66:0-	-0.98	96.0-	-0.97	-0.97	-0.99	-0.99
X	4.14	3.36	4.91	2.91	3.87	3.97	2.12	3.22	3.76	3.37
			Light sil	ty loam – G	Light silty loam – Glina lekka pylasta	ylasta				
0	4.25	4.09	5.66	3.86	4.95	4.71	2.04	4.52	4.23	4.30
100	4.00	3.40	4.68	2.88	4.14	4.00	1.87	3.64	3.67	3.48
200	3.64	2.88	4.05	2.42	3.58	3.49	1.60	3.11	3.22	2.98
300	3.13	2.59	3.84	2.01	3.21	3.13	1.47	2.38	2.91	2.53
400	3.04	2.24	3.47	1.80	2.36	2.53	1.33	1.94	2.55	2.13
i.	-0.98	86:0-	-0.96	96.0-	-0.99	66:0-	-0.99	66:0-	66.0-	-0.99
18	3.61	3.04	4.34	2.59	3.65	3.57	1.66	3.12	3.32	3.08
${ m LSD}_{0.05}$ ${ m NIR}_{0.05}$	a - 0.07; b $x = 0.07$; a $x = 0.07$; a	-0.05; c-0.	$a-0.07;\ b-0.05;\ c-0.05;\ d-0.07;\ a\times b-0.11;\ a\times c-0.11;\ a\times d-0.15;\ b\times c-0.07;\ b\times d-0.09;\ c\times d-0.09;\ a\times b\times c-0.5;\ a\times b\times d-0.21;\ a\times c\times d-0.21;\ b\times c\times d-0.13;\ a\times b\times c\times d-0.30$	$a \times b - 0.11$ - 0.21; $b \times c$	L; $a \times c - 0.1$. $x d - 0.13$; a	1; a x d - 0.1	$5; b \times c - 0$ $- 0.30$.07; b x d – C	0.09; c x d –	0.09; a x b

YI-uncropped soil – gleba nieobsiana, +YI-soil cropped to yellow lupine – gleba obsiana lubinem źółtym; LSD for - NIR dla: a - soil species – gatunek gleby, b – nickel dose – dawki niklu, c – time of analysis – terminu analizy, d - method of soil management - obsiania gleby; n.s – nonsignificant – nieistotne statystycznie; n=15

Table 3 Tabela 3

Effect of soil contamination with nickel on the activity of urease in 1 kg d.m. of soil (mg N-NH $_4$ kg⁻¹d.m.) Wpływ zanieczyszczenia niklem na aktywność ureazy w 1 kg s.m. gleby (mg N-NH $_4$ kg⁻¹s.m.)

			Time of and	Time of analysis (days) - Termin analizy (dni)	– Termin a	nalizy (dni)			;	
Ni dose (mg kg ⁻¹ of soil) Dawka Ni (mg kg ⁻¹ glebv)		14	2	28	4	42	5	56	3	
	IX-		-Yl	IX+	IX-		-YI	+Y1	IX-	+Y1
			Heavy loamy sand – Piasek gliniasty mocny	sand – Pias	sek gliniasty	mocny				
0	23.65	23.90	14.48	16.88	14.51	27.00	18.15	15.56	17.70	20.84
100	20.33	26.44	10.14	15.52	11.10	25.71	9.53	12.14	12.78	19.95
200	16.04	15.51	9.28	14.19	8.09	14.62	7.55	10.11	10.24	13.61
300	12.06	11.94	8.41	10.23	7.27	13.24	6.65	7.13	8.60	10.64
400	11.69	10.85	7.59	9.79	6.84	10.32	5.15	7.10	7.82	9.52
7	86.0-	-0.91	-0.91	26:0-	-0.94	-0.95	-0.89	76.0-	-0.95	-0.97
ert arkappa ert	16.75	17.73	96.6	13.32	9.56	18.18	9.41	10.41	11.43	14.91
			Light silty	Light silty loam – Glina lekka pylasta	na lekka pyl	asta				
0	27.96	85.49	22.82	65.43	31.27	79.54	53.79	71.14	33.96	75.40
100	24.69	73.82	19.40	64.97	25.37	58.20	52.83	67.88	30.57	66.22
200	23.42	71.57	14.66	57.96	21.36	55.95	49.79	57.80	27.31	60.82
300	22.32	64.74	12.92	54.16	20.62	51.82	27.78	54.75	20.91	56.37
400	21.44	55.80	10.55	48.37	16.63	46.99	18.40	35.44	16.76	46.65
7	96.0-	-0.98	-0.99	-0.98	-0.97	-0.90	-0.93	-0.95	-0.99	-0.99
$ \mathcal{X} $	23.97	70.28	16.07	58.18	23.05	58.50	40.52	57.40	25.90	61.09
${ m LSD}_{005}$	a - 0.67; b - x + 0.000; a - 0.	a - 0.67; $b - 0.43$; $c = 0.43$; $c = 0.60$; $c = 0.95$; $c = 0.95$; $c = 0.95$; $c = 0.60$; $c = 0.60$; $c = 0.85$; $c =$	0.43; d -0.6	x = 0.0 a $x = 0.0$ x d $x = 0.0$ 3	95; $a \times c - 0$	$.95; a \times d - 1$	1.35; b x c – 0 x d – 2.70	0.60; b x d –	0.85; c x d	– 0.85; a
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 * explanations under Table 2 * objaśnienia podano pod tabelą 2

ues of all components of yield structure (mass of aerial parts, roots and nodules and the number of nodules per root) were lower in nickel-contaminated soils than in soils with a natural nickel content. The dry matter yield of yellow lupine grown on heavy loamy sand and light silty loam decreased 3.8-fold and 2-fold, respectively, following the application of the highest dose of nickel. Excessive quantities of nickel in the soil negatively affected the root growth and contributed to almost complete disappearance of root nodules. This trend was particularly noticeable when nickel was introduced into the lighter soil at a dose of 200 mg Ni²⁺ kg⁻¹ and into the heavier soil at a dose of 300 mg Ni²⁺ kg⁻¹. The current results indicate that the activity of dehydrogenases, urease, acid phosphatase and alkaline phosphatase was significantly positively correlated with yellow lupine yield (Figure 1).

Literature data (Welp 1999, Kucharski, Wyszkowska 2000, Giridhara, Siddaramappa 2002) show that the harmful effect of heavy metals, including nickel, on the biological properties of soil is dependent on their concentration in the environment. When present in low amounts, those elements may stimulate enzymatic activity of soil. However, if their threshold limit values are exceeded, they become typical inhibitors of the activity of enzymes and soil microorganisms.

In the present study all the doses of nickel, even the lowest one (100 mg Ni²⁺ kg⁻¹), reduced the enzymatic activity of both types of soil, but this activity was inhibited to a greater extent in heavy loamy sand (considered poorer) than in light silty loam. This is related primarily to a higher content of colloids in more compact soils, confirmed by their higher adsorbing capacity which plays an important role in permanent binding of metals (including nickel), thus reducing their impact on enzymes. This hypothesis is supported by the results of previous investigations (Welp 1999, Kucharski, Wyszkowska 2000, Wyszkowska et al. 2005), which demonstrated that the abundance of mineral and organic colloids in the soil could partly neutralize the negative influence of heavy metals on enzymes. A strong correlation between urease activity and the organic matter content of soil was also reported by Stepniewska and Samborska (1999). This could also explain the role of yellow lupine in alleviating the adverse effects of nickel on urease activity, observed in the present experiment. The organic compounds secreted by roots into the soil positively affect microbial communities, thus increasing the pool of enzymes. In addition, those substances may form complexes with heavy metals, including nickel (Kandeler et. al. 1999). However, this hypothesis was not confirmed in the case of dehydrogenases, acid phosphatase and alkaline phosphatase, whose activity was not significantly affected by yellow lupine grown in nickel-contaminated soil.

Environmental pollution with heavy metals has a harmful impact not only on the biological activity of soil, but also on the quality and yield of

Table 4 Tabela 4

Effect of soil contamination with nickel on the activity of acid phosphatase in 1 kg d.m. of soil (mmol PNP h^{-1}) Wpływ zanieczyszczenia gleby niklem na aktywność fosfatazy kwaśnej w 1 kg s.m. gleby (mmol PNP h^{-1})

			Time of an	Time of analysis (days)	- Termin analizy (dni)	nalizy (dni)				
Ni dose (mg kg ⁻¹ of soil)	1	14	22	28	4	42	5	56	l8	
Lawra 141 (1115 115 Broy)	-YI	+Y1	-Yl		IX-	Ι Χ +	-YI	+Y1	-YI	
			Heavy loamy sand – Piasek gliniasty mocny	sand – Pias	sek gliniasty	mocny				
0	1.96	2.20	2.20	1.71	3.07	1.99	2.48	2.15	2.43	2.01
100	1.84	1.66	2.11	1.68	2.55	1.88	2.34	1.98	2.21	1.80
200	1.65	1.61	1.79	1.53	2.44	1.66	2.06	1.78	1.99	1.65
300	1.59	1.48	1.70	1.49	2.41	1.55	1.78	1.66	1.87	1.55
400	1.50	1.31	1.68	1.33	2.26	1.37	1.83	1.56	1.82	1.39
7	-0.98	-0.93	-0.95	-0.98	-0.90	66:0-	-0.96	-0.99	-0.97	-0.99
8	1.71	1.65	1.90	1.55	2.55	1.69	2.10	1.83	2.06	1.68
			Light silty	Light silty loam – Glina lekka pylasta	na lekka pyl	asta				
0	1.95	2.05	1.79	1.83	2.55	1.86	2.15	2.05	2.11	1.95
100	1.78	2.01	1.67	1.79	2.38	1.76	2.10	1.97	1.98	1.88
200	1.73	1.88	1.72	1.61	2.28	1.67	1.98	1.70	1.93	1.72
300	1.66	1.79	1.67	1.60	2.12	1.52	1.84	1.60	1.82	1.63
400	1.51	1.71	1.62	1.32	1.95	1.34	1.73	1.49	1.70	1.47
7	-0.98	66:0-	-0.84	-0.95	66.0-	66.0-	-0.99	-0.98	-0.99	-0.99
x	1.73	1.89	1.69	1.63	2.26	1.63	1.96	1.76	1.91	1.73
$\mathrm{LSD}_{0.05}$ $\mathrm{NIR}_{0,05}$	a - 0.04; b - b x c - n.s.;	-0.02; c -0 .	.02; d – 0.03; n.s.; a x c x c	$a \times b - 0.05$ $1 - 0.10; b \times$	$c \times c - n.s.$ $c \times d - 0.07;$	$a-0.04;\ b-0.02;\ c-0.02;\ d-0.03;\ a\ x\ b-0.05;\ a\ x\ c-n.s.;\ a\ x\ d-0.07;\ b\ x\ c-0.03;\ b\ x\ d-0.05;\ c\ x\ d-0.05;\ a\ x$	7; b x c -0.0 1 -0.15	3; b x d – 0.	05; c x d – ().05; a x

 * explanations under Table 2 * objaśnienia podano pod tabelą 2

Table 5 Tabela 5

Effect of soil contamination with nickel on the activity of alkaline phosphatase in 1 kg d.m. of soil (mmol PNP h⁻¹) Wptyw zanieczyszczenia gleby niklem na aktywność fosfatazy alkalicznej w 1 kg s.m. gleby (mmol PNP h⁻¹)

,			Time of ans	Time of analysis (days) – Termin analizy (dni)	– Termin a	nalizy (dni)				
Ni dose (mg kg ⁻¹ of soil) Dawka Ni (mg kg ⁻¹ glebv)	1	14	2	28	4	42	ũ	56	3	
	-Yl	+Y1	IX-	IX+	IA-	+YI	-Yl	+Y1	-Yl	+Y1
			Heavy loamy sand – Piasek gliniasty mocny	sand – Pia	sek gliniasty	mocny				
0	2.06	1.68	2.25	1.75	2.07	1.64	2.08	2.49	2.12	1.89
100	1.96	1.29	2.04	1.38	1.63	1.44	1.57	2.44	1.80	1.64
200	1.77	1.22	1.86	1.18	1.45	1.18	1.41	1.92	1.62	1.38
300	1.67	1.15	1.82	1.16	1.40	1.08	1.27	1.67	1.54	1.27
400	1.59	1.09	1.74	1.02	1.37	1.05	1.28	1.55	1.50	1.18
7	-0.99	06.0-	96.0-	-0.94	68:0-	96:0-	06:0-	-0.97	-0.94	-0.97
x	1.81	1.29	1.94	1.30	1.58	1.28	1.52	2.01	1.71	1.47
			Light silty	Light silty loam – Glina lekka pylasta	na lekka pyl	asta				
0	10.65	3.07	4.54	3.37	96.8	2.82	4.57	4.55	5.93	3.45
100	10.40	2.62	3.97	2.55	3.20	2.78	4.38	4.49	5.49	3.11
200	10.31	2.43	3.78	2.36	3.15	2.45	4.14	3.64	5.35	2.72
300	10.16	2.31	3.50	2.25	2.93	2.50	3.55	3.37	5.04	2.61
400	10.15	2.27	3.43	2.02	2.56	1.98	3.39	3.02	4.88	2.32
7	-0.95	-0.93	-0.95	-0.92	26.0-	-0.92	-0.98	-0.97	-0.98	-0.99
\overline{x}	10.33	2.54	3.84	2.51	3.16	2.51	4.01	3.81	5.34	2.84
${ m LSD}_{0.05}$	a - 0.05; b - a x b x c - r	-0.03; c -0.03	a - 0.05; b - 0.03; c - 0.03; d - 0.05; a x b - 0.07; a x c - 0.07; a x d - 0.10; b x c - 0 a x b x c - n.s.; a x b x d - 0.14; a x c x d - 0.14; b x c x d - 0.09; a x b x c x d - 0.20	$a \times b - 0.07$ $c \times d - 0.14$:	$3 \times 3 \times$	0.7; a x d -0.9	-0.05; b - 0.03; c - 0.03; d - 0.05; a x b - 0.07; a x c - 0.07; a x d - 0.10; b x c - 0.05; b x d - 0.07; c x d - 0.07; x b x c - 0.05; a x b x d - 0.14; a x c x d - 0.14; b x c x d - 0.09; a x b x c x d - 0.20	05; b x d – C	0.07; c x d –	0.07;
60.0										

 * explanations under Table 2 * obja
śnienia podano pod tabelą 2

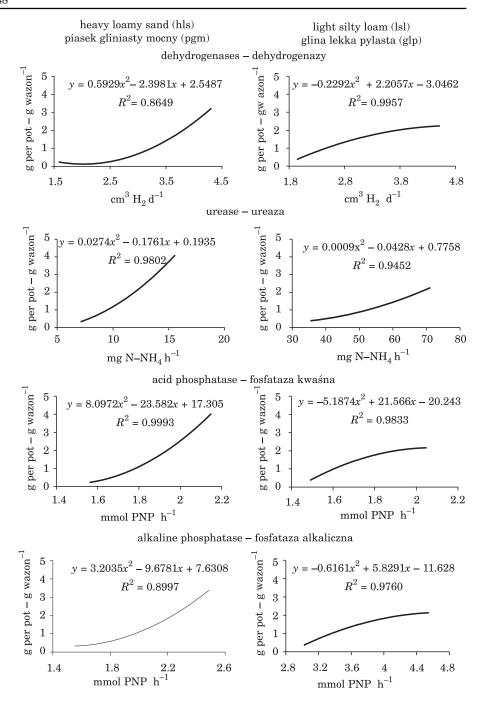


Fig. 1. Correlation between yellow lupine yield and enzymatic activity in 1 kg d.m. of soil Rys. 1. Zależność między plonem łubinu żółtego a aktywnością enzymów w 1 kg s.m. gleby

crops, and – in consequence – on the health of humans and animals (Oleszek et al. 2003). According to Pandey and Sharma (2002), excessive quantities of nickel in the soil inhibit the growth and development of plants because this metal exerts a deleterious effect on the metabolic processes taking place in the plant. The negative influence of nickel on plants was also observed by other authors (Koszelnik-Leszek 2002, Wyszkowski, Wyszkowska 2004, Wyszkowska et al. 2005). Tibazarwa et al. (2001) demonstrated that this metal may adversely affect crops even when present at substantially lower concentrations than those applied in the current experiment with yellow lupine.

Table 6
Tabela 6
Effect of soil contamination with nickel on yellow lupine yield, g d.m. pot⁻¹
Wpływ zanieczyszczenia gleby niklem na plon łubinu żółtego, g s.m. ·wazon⁻¹

Ni dose (mg kg ⁻¹ of soil) Dawka Ni (mg kg ⁻¹ gleby)		l.m. pot ⁻¹) n. wazon ⁻¹) of roots korzeni	Root nodule yield (g d.m. root ⁻¹) Plon brodawek (g s.m. korzeń ⁻¹)	Number of nodules per root Liczba brodawek na 1 korzeniu
	1	y sand – Piasek glii	iasty mocny	
0	4.04	1.19	0.18	21.33
100	2.35	0.58	0.07	6.00
200	0.95	0.26	0.00	0.00
300	0.54	0.09	0.00	0.00
400	0.19	0.04	0.00	0.00
r	-0.95	0.89	0.98	0.97
	Light silt	y loam – Glina lekk	xa pylasta	
0	2.22	0.56	0.16	16.00
100	2.02	0.46	0.10	10.33
200	1.55	0.48	0.03	7.00
300	0.87	0.28	0.00	0.00
400	0.40	0.05	0.00	0.00
r	-0.99	0.96	0.90	0.87
LSD _{0.05} NIR _{0.05}	a -0.23; b -0.15; a x b -0.33	a -0.19; b -n.s.; a x b -0.27	a -0.03; b -n.s.; a x b -n.s.	a -4.42; b -n.s.; a x b -n.s.

LSD for – NIR dla: a - soil species – gatunek gleby, b – nickel dose – dawki niklu; n.s – non-significant – nieistotne statystycznie; r – coefficient of correlation significant at – współczynnik korelacji istotny dla p = 0.05; n = 15

CONCLUSIONS

- 1. Soil contamination with nickel, applied at a dose of 100 to 400 mg Ni²⁺ kg⁻¹, disturbed its biological balance measured by enzymatic activity. The activity of dehydrogenases, urease, acid phosphatase and alkaline phosphatase was reduced along with an increase in the nickel dose.
- 2. The activity of urease and alkaline phosphatase was higher in light silty loam, while the activity of dehydrogenases and acid phosphatase was higher in heavy loamy sand.
- 3. The activity of dehydrogenases and urease was higher in soil cropped to yellow lupine, whereas the activity of acid phosphatase and alkaline phosphatase was higher in uncropped soil.
- 4. The negative effect of nickel on the activity of the tested soil enzymes was observed over the entire experimental period (56 days).
- 5. Yellow lupine partly alleviated the adverse impact of nickel on urease activity, but did not reduce the inhibitory effect of this heavy metal on the other enzymes.
- 6. Yellow lupine grown in more compact (light silty loam) and less compact (heavy loamy sand) soil was found to be sensitive to nickel contamination.

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