

# Effect of nickel contamination on soil enzymatic activities

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## ABSTRACT

The effect of soil contamination with nickel applied in the doses of 100, 200, 300 and 400 mg Ni/kg of soil on the activity of dehydrogenases, urease and acid and alkaline phosphatase was studied in a pot experiment. Heavy loamy sand and silty light loam were used in the experiment that comprised of two series: with spring barley cultivation and without plant cultivation. The enzyme activity was determined on day 14, 28, 42 and 56 of the experiment. Based on the study, it was found that soil contamination with nickel applied as  $\text{NiCl}_2 \cdot 6 \text{H}_2\text{O}$  decreased the activity of dehydrogenases, urease and acid and alkaline phosphatase. This decrease was determined by the applied dose of this metal. Nickel exhibited a stronger effect on the soil with spring barley cultivation than on the unsown soil. In the heavy loamy sand sown with spring barley, more than 50% inhibition of the activity of dehydrogenases was caused by 300 and 400 mg Ni contamination, and in the case of urease by 200, 300 and 400 mg Ni/kg of soil. In the silty light loam sown with spring barley, more than 50% decrease in the activity of dehydrogenases and alkaline phosphatase was observed under 400 mg Ni contamination. The inhibition of the other enzyme activities did not exceed 50%. Urease and alkaline phosphatase exhibited a higher activity in the heavier soil, whereas dehydrogenases and acid phosphatase exhibited a higher activity in the lighter soil. In the nickel-free soil, spring barley cultivation had a positive effect on the enzyme activity and a positive correlation between the spring barley yield and the activity of dehydrogenases, urease and acid and alkaline phosphatase was observed.

**Keywords:** soil contamination; nickel; spring barley; enzyme activity; dehydrogenases; urease; acid phosphatase; alkaline phosphatase

Environmental pollution by industrial emissions and agricultural chemicals has a negative effect on the physico-chemical properties and the biological activity of soil. Enzymatic activity influencing functional processes occurring in a given soil profile determines, among others, the soil biological activity (Koper and Piotrowska 1996, Nowak et al. 2003, Šmejkalová et al. 2003). Heavy metals, including nickel, and their compounds significantly decrease the enzymatic activity of soils (Nowak et al. 2000). This chemical element does not belong among the essential elements for metabolic processes of living organisms; however, there are plants unable to develop properly without its concentration in the soil. Pollution of the soil environment with heavy metals, its effect on the soil microorganisms (Kucharski and Wyszowska 2004) and further on their enzymatic activity depends, apart from other things, on the soil pH, the content of organic and mineral colloids and on a metal type and its chemical properties (Nowak et al. 2000, Wyszowska and Wyszowski 2003b).

Dehydrogenases are enzymes that indicate rather closely the soil biological activity. Their activity can be inhibited up to 90% by high soil contamination with heavy metals (Tresar-Cepeda et al. 1998).

According to Welp (1999), metals that inhibit the dehydrogenase activity up to 50% can be arranged in following order:  $\text{Hg}$  (2 mg) >  $\text{Cu}$  (35 mg) >  $\text{Cr}_{(\text{VI})}$  (71 mg) >  $\text{Cr}_{(\text{III})}$  (75 mg) >  $\text{Cd}$  (90 mg) >  $\text{Ni}$  (100 mg) >  $\text{Zn}$  (115 mg) >  $\text{As}$  (168 mg) >  $\text{Co}$  (582 mg) >  $\text{Pb}$  (652 mg/kg of soil).

Also urease and acid and alkaline phosphatase are sensitive to soil contamination with heavy metals (Zheng-ChungRong et al. 1999). Literature on the effect of nickel on the soil enzymatic activity is scarce (Nowak et al. 2000, Wyszowska and Wyszowski 2003a), the aim of the study was thus to determine the effect of soil contamination with this metal on the activity of dehydrogenases, urease and acid and alkaline phosphatase.

## MATERIAL AND METHODS

The experiment was conducted in a vegetation hall in plastic pots, each filled with 3 kg of soil. There were following variable factors of the experiment:

1. Two types of soil Eutric Cambisols according to WRB (1998). Characteristics of soils are given in Table 1;

Table 1. Characteristics of Eutric Cambisols soils

Kind of soil	Granulometric composition (mm)			pH <sub>KCl</sub>	C <sub>organic</sub> (g/kg)	CEC mmol(+)/kg	BS (%)
	1–0.1	0.1–0.02	< 0.02				
Heavy loamy sand (hls)	66	17	17	6.9	7.5	100.5	88.9
Silty light loam (sll)	42	32	26	7.0	11.2	167.8	94.7

hls – heavy loamy sand, sll – silty light loam, CEC – cation exchange capacity, BS – base saturation

2. Ni dose as NiCl<sub>2</sub>·6 H<sub>2</sub>O in mg/kg dry matter of soil: 0, 100, 200, 300 and 400;

3. Soil utilisation: soil sown with spring barley Rabel variety (12 plants per pot) and unsown soil.

All pots were given equal doses of macro- and microelement fertilisation in amounts calculated

as a pure component in mg/kg of soil amounting to: N – 100, P – 44, K – 83, Mg – 20, Zn – 5, Mn – 5, Mo – 5 and B – 0.33. Nitrogen was used as CO(NH<sub>2</sub>)<sub>2</sub>, phosphorus as KH<sub>2</sub>PO<sub>4</sub>, potassium as KH<sub>2</sub>PO<sub>4</sub> and KCl, magnesium as MgSO<sub>4</sub>·7 H<sub>2</sub>O, zinc as ZnCl<sub>2</sub>, manganese as MnCl<sub>2</sub>·4 H<sub>2</sub>O, molybdenum as Na<sub>2</sub>MoO<sub>4</sub>·2 H<sub>2</sub>O and boron as H<sub>3</sub>BO<sub>4</sub>.

Table 2. Effect of soil contamination with nickel on the activity of dehydrogenases in 1 kg of dry matter of soil (cm<sup>3</sup> H<sub>2</sub>/day)

Ni dose (mg/kg of soil)	Day of analysis (No. of days)								$\bar{x}$	
	14		28		42		56			
	-p	+p	-p	+p	-p	+p	-p	+p	-p	+p
Heavy loamy sand (hls)										
0	5.36	7.00	5.92	8.05	5.55	9.88	3.02	9.08	4.96	8.50
100	4.90	5.77	5.36	6.71	5.23	5.32	2.39	6.16	4.47	5.99
200	4.77	5.26	5.21	5.12	4.28	4.95	2.12	4.71	4.10	5.01
300	4.47	4.75	5.03	4.63	3.90	4.00	1.95	2.65	3.84	4.01
400	3.62	4.52	4.49	3.99	3.72	3.88	1.65	1.69	3.37	3.52
$\bar{x}$	4.62	5.46	5.20	5.70	4.54	5.61	2.23	4.86	4.15	5.41
Silty light loam (sll)										
0	4.14	5.49	5.59	6.98	5.00	9.09	2.04	9.29	4.19	7.71
100	3.84	4.74	4.86	6.02	4.22	7.88	1.78	7.97	3.68	6.65
200	3.52	4.44	4.69	5.46	4.08	6.59	1.75	6.61	3.51	5.78
300	3.33	4.44	3.94	4.26	3.58	5.14	1.51	5.27	3.09	4.78
400	3.14	4.23	3.37	3.99	3.27	3.31	1.17	3.07	2.74	3.65
$\bar{x}$	3.59	4.67	4.49	5.34	4.03	6.40	1.65	6.44	3.44	5.71

LSD a = 0.07\*\*; b = 0.11\*\*; c = 0.10\*\*; d = 0.07\*\*; a × b = 0.15\*\*; a × c = 0.14\*\*;  
a × d = 0.10\*\*; b × c = 0.21\*\*; b × d = 0.15\*\*; c × d = 0.14\*\*; a × b × c = 0.30\*\*;  
a × b × d = 0.19\*\*; a × c × d = 0.19\*\*; b × c × d = 0.30\*\*; a × b × c × d = 0.43\*\*

Soil utilisation: –p – unsown soil, +p – spring barley sown soil

LSD (least statistical difference) for: a – soil type, b – nickel dose, c – day of analysis, d – soil utilisation

n.s. – non-significant

LSD significant at: \*P < 0.05; \*\*P < 0.01

The experiment series with spring barley cultivation was carried out in 6 replications and the one without sowing was carried out in three replications (the bigger number of replications with the sown soil resulted from the necessity to minimize the variability in the soil caused by root secretions). Throughout the entire period of the experiment (56 days), a constant soil humidity at 60% of the capillary water holding capacity was maintained. Spring barley was harvested at the phase of ear formation. On day 14, 28, 42, 56 of the experiment, the soil samples were analysed for the activity of dehydrogenases according to the Lenhard method modified by Casida et al. (1964), the activity of urease – with the Gorin and Chin Chang method (1966) as well as for the activity of acid and alkaline phosphatase with the Tabatabai and Bremner method (1966). Biochemical analyses were completed in three replications. The results were elaborated statistically using Duncan's test and four-factor analysis of variance ANOVA. Statistical analysis used also Pearson's simple correlation coefficients between the variables and was completed using Statistica software (StatSoft Inc. 2001).

## RESULTS AND DISCUSSION

The enzyme activity was determined by the type of soil, its utilisation and its contamination with nickel. The activity changed throughout the experimental duration (Tables 2–6).

The spring barley cultivation increased the activity of dehydrogenases by 42% in the heavy loamy sand (contaminated with nickel) and by 46% in the silty light loam (Figure 1). The activity of this enzyme was 15% lower in the loam unsown with barley than in the sand unsown with barley and it was 10% lower in the sown loam than the in sown sand. Soil contamination with nickel from 100 to 400 mg/kg decreased the activity of dehydrogenases from 12 to 32% in the unsown heavy loamy sand and from 30 to 59% when sown with spring barley, in comparison to control. The activity of dehydrogenases in the silty light loam was from 12 to 34% (sown) and from 14 to 53% (unsown). It is clear that the mineral and organic colloids of the heavier soil had a cushioning effect on the direct inactivation of dehydrogenases. The soil enzymes occur freely or in a bound form (Pacha 1984, Gołębiowska and

Table 3. Effect of soil contamination with nickel on the activity of urease in 1 kg of dry matter of soil (mg N-NH<sub>4</sub>)

Ni dose (mg/kg of soil)	Day of analysis (No. of days)								$\bar{x}$	
	14		28		42		56			
	-p	+p	-p	+p	-p	+p	-p	+p	-p	+p
Heavy loamy sand (hls)										
0	25.12	23.28	13.62	31.16	13.46	20.86	17.89	34.65	17.52	27.49
100	22.54	18.31	11.69	17.30	11.95	17.98	10.56	16.97	14.19	17.64
200	20.41	18.26	11.15	7.79	10.64	13.48	8.58	13.89	12.70	13.36
300	17.52	17.64	9.30	6.81	9.89	8.61	8.12	10.30	11.21	10.84
400	13.72	16.62	8.99	6.66	9.27	8.51	7.25	7.74	9.81	9.88
$\bar{x}$	19.86	18.82	10.95	13.94	11.04	13.89	10.48	16.71	13.08	15.84
Silty light loam (sll)										
0	26.67	28.45	22.82	39.18	29.68	44.29	53.71	55.35	33.22	41.82
100	24.51	28.40	21.10	30.66	23.45	30.37	46.69	55.07	28.94	36.13
200	24.00	27.98	15.54	29.27	21.42	28.39	34.37	46.11	23.83	32.94
300	23.27	27.82	15.41	14.46	16.10	28.37	28.13	38.66	20.73	27.33
400	21.82	26.06	14.02	12.46	15.35	24.87	24.09	35.29	18.82	24.67
$\bar{x}$	24.05	27.74	17.78	25.21	21.20	31.26	37.40	46.10	25.11	32.58
LSD	a = 0.28**, b = 0.45**, c = 0.40**, d = 0.28**, a × b = 0.63**, a × c = 0.57**, a × d = 0.40**, b × c = 0.89**, b × d = 0.63**, c × d = 0.57**, a × b × c = 1.27**, a × b × d = 0.89**, a × c × d = 0.80**, b × c × d = 1.27**, a × b × c × d = 1.79**									

\*for explanations see Table 2

Table 4. Effect of soil contamination with nickel on the activity of acid phosphatase in 1 kg of d.m. of soil (mmol PNP/h)

Ni dose (mg/kg of soil)	Day of analysis (No. of days)								$\bar{x}$	
	14		28		42		56			
	-p	+p	-p	+p	-p	+p	-p	+p	-p	+p
Heavy loamy sand (hls)										
0	1.97	1.88	2.20	2.44	3.09	3.69	2.47	2.55	2.43	2.64
100	1.90	1.63	1.90	2.05	2.55	3.25	1.99	2.15	2.09	2.27
200	1.83	1.51	1.78	1.92	2.52	2.40	1.97	2.04	2.03	1.97
300	1.77	1.51	1.75	1.75	2.51	2.29	1.94	1.82	1.99	1.84
400	1.64	1.47	1.68	1.73	2.30	2.21	1.74	1.66	1.84	1.77
$\bar{x}$	1.82	1.60	1.86	1.98	2.59	2.77	2.02	2.04	2.07	2.10
Silty light loam (sll)										
0	1.90	1.70	2.01	2.18	2.57	3.49	2.14	2.35	2.16	2.43
100	1.81	1.66	1.76	1.90	2.37	3.05	2.01	2.16	1.99	2.19
200	1.74	1.62	1.69	1.87	2.21	2.41	2.00	2.07	1.91	1.99
300	1.68	1.63	1.67	1.85	2.17	2.31	1.92	2.03	1.86	1.96
400	1.66	1.56	1.58	1.70	1.97	2.19	1.78	1.95	1.75	1.85
$\bar{x}$	1.76	1.63	1.74	1.90	2.26	2.49	1.97	2.11	1.93	2.03
LSD	a = 0.02**; b = 0.03**; c = 0.03**; d = 0.02**; a × b = 0.05**, a × c = 0.04**; a × d = 0.03**; b × c = 0.07**; b × d = 0.05**; c × d = 0.04**; a × b × c = n.s.; a × b × d = 0.07*; a × c × d = 0.06**; b × c × d = 0.09**; a × b × c × d = n.s.									

\*for explanations see Table 2

Grzyb-Miklaszewska 1991a, b, Dick 1994). Enzyme complexes with minerals and organic colloids are more stable and more resistant to denaturation and proteolysis. In the lighter soil (sown with barley) over 50% inhibition of this enzyme occurred as the effect of both 300 and 400 mg Ni/kg, whereas in the silty light loam it occurred only as the result of the highest dose (400 mg Ni/kg). The inhibiting effect of nickel contamination intensified during the spring barley vegetation, especially in the heavy loamy sand soil which is most likely closely related with the undesirable effect of this metal on the growth and development of the crop measured by the yield (Figure 1). Nickel had a strong toxic effect on the plant cultivated in the loamy sand when applied from 200 to 400 mg/kg and in the silty light loam from 300 and 400 mg/kg. Under influence of 300 mg Ni/kg in the loamy sand, the spring barley yield decreased by 78% and in the sandy loam it decreased only by 25%; however under effect of 400 mg Ni/kg the yield decreased by 96 and 73%, respectively.

Spring barley cultivation and its development had an effect on the activity of urease (Table 3). The activity of this enzyme in the soil contaminated with

nickel increased during spring barley vegetation and was always higher in the silty light loam than in the heavy loamy sand. This is the result of the protective activity of the soil colloids towards this enzyme. They absorb urease and thus prolong its activity. According to Abmayyan (1993) there are more enzymes bound with humic acids and the fulvous than other soil components. Nevertheless, the role of mineral colloids in both enzyme activity and soil fertility should not be overseen. This role was indicated previously by Kucharski et al. (1996), whose study involved 3 soils varying in granulometric composition. Based on the results of this study, a correlation between the finest soil fraction and enzyme activity was found. Although the catalytic capacity of enzymes bound with colloids is generally lower than freely occurring or intracellular enzymes, they are more resistant to periodical changes in ecosystem conditions and they mainly determine the direction of biochemical changes in soil and may influence its fertility (Abmayyan 1993).

This could be confirmed by the intensity of the toxic effect of nickel contamination on urease in the

tested soils. In the heavy loamy sand sown with spring barley, the activity of urease was inhibited by approx. 50% (51–64%) as the result of the applied contamination from 200 to 400 mg Ni/kg, whereas in the silty light loam, none of the nickel doses applied inhibited this enzyme activity by more than 50% (14–41%). The differences between the urease activity in the experimental soils were highly significant.

The activity of acid phosphatase was to a lesser degree determined both by the spring barley cultivation and the soil contamination with nickel (Table 4). The activity of this enzyme was usually higher in the lighter soil and significantly higher in the pots sown with spring barley. However, the differences between the activity of the sown and unsown soil were not as large as in the case of dehydrogenases and urease. Increasing contamination with nickel inhibited the activity of acid phosphatase. Again, this significant inhibition was considerably lower than that of the activity of dehydrogenases and urease. Similar to the other enzymes, the inhibition of acid phosphatase was stronger in the soil sown with spring barley

and it ranged from 14 to 33% in the heavy loamy sand and from 10 to 24% in the silty light loam, compared to uncontaminated soil.

Contrary to the acid phosphatase, the activity of alkaline phosphatase (Table 5) was significantly higher (2.6-fold) in the silty light loam than in the heavy loamy sand. Also, the activity of this enzyme in the pots with nickel uncontaminated soil increased as an effect of spring barley cultivation. Nickel contamination caused a stronger enzyme activity inhibition in the sown soil. In the lighter soil, this inhibition was from 15 to 32% and in the heavier one from 13% (100 mg Ni/kg) to as much as 55% (400 mg Ni/kg). These differences were highly significant.

Even under such extreme conditions as the soil contamination with nickel, the enzyme activity closely correlated with the spring barley yield, which is shown by the regression equations included in Figure 2. The coefficients of the correlation between the analysed factors are interesting (Table 6). For all enzymes there were a significantly negative correlation with the soil contamination with nickel, and a positive correlation between

Table 5. Effect of soil contamination with nickel on the activity of alkaline phosphatase in 1 kg of d.m. of soil (mmol PNP/h)

Ni dose (mg/kg of soil)	Day of analysis (No. of days)								$\bar{x}$	
	14		28		42		56			
	-p	+p	-p	+p	-p	+p	-p	+p	-p	+p
Heavy loamy sand (hls)										
0	2.07	2.18	2.25	2.48	2.08	2.29	2.07	2.36	2.12	2.33
100	1.85	1.99	2.05	2.13	1.71	1.81	1.78	2.00	1.85	1.98
200	1.77	1.70	1.97	2.00	1.66	1.66	1.66	1.63	1.77	1.75
300	1.69	1.65	1.69	1.88	1.58	1.63	1.55	1.63	1.63	1.70
400	1.52	1.59	1.65	1.69	1.54	1.51	1.55	1.58	1.57	1.59
$\bar{x}$	1.78	1.82	1.92	2.04	1.71	1.78	1.72	1.84	1.78	1.87
Silty light loam (sll)										
0	10.38	9.51	4.48	4.91	3.99	4.25	4.56	5.03	5.85	5.93
100	9.94	8.89	4.07	4.31	3.38	3.48	4.24	4.00	5.41	5.17
200	9.85	8.66	3.66	4.12	3.14	3.21	3.87	3.79	5.13	4.95
300	9.80	2.71	3.51	3.63	3.05	3.12	2.78	2.63	4.79	3.02
400	9.64	2.47	3.27	3.14	2.33	2.61	2.60	2.50	4.46	2.68
$\bar{x}$	9.86	6.45	3.80	4.02	3.18	3.33	3.61	3.59	5.11	4.35
LSD	a = 0.08**; b = 0.13**; c = 0.11**; d = 0.08**; a × b = 0.18**, a × c = 0.16**; a × d = 0.11**, b × c = 0.25**; b × d = 0.18**; c × d = 0.16**, a × b × c = 0.35**; a × b × d = 0.25**, a × c × d = 0.22**; b × c × d = 0.35**; a × b × c × d = n.s.									

\*for explanations see Table 2

Table 6. Pearson linear correlation coefficients for the variable factors of the experiment

Variable	Ni dose	Deh	Ure	Pac	Pal
<b>Heavy loamy sand (hls)</b>					
Ni dose	1.00				
Deh	-0.63**	1.00			
Ure	-0.65**	0.70**	1.00		
Pac	-0.51**	0.44**	0.22*	1.00	
Pal	-0.82**	0.75**	0.65**	0.37**	1.00
<b>Silty light loam (sll)</b>					
Ni dose	1.00				
Deh	-0.50**	1.00			
Ure	-0.51**	0.40**	1.00		
Pac	-0.42**	0.54**	0.41**	1.00	
Pal	-0.33**	-0.01	-0.01	-0.27**	1.00

Deh – dehydrogenases, Ure – urease, Pac – acid phosphatase, Pal – alkaline phosphatase,  
*r* – correlation co-efficient significant at: \*\**P* < 0.01; \**P* < 0.05; *n* = 120

the activity of dehydrogenases and the activity of urease and acid and alkaline phosphatase in the heavy loamy sand as well as between the activity of dehydrogenases and the activity of urease and acid phosphatase in the silty sandy loam. Additionally, the correlation between the activities of phosphatases was positive, regardless of soil type.

In conclusion, nickel is a strong inhibitor of the tested soil enzymes. Soil contamination with nickel (100, 200, 300 and 400 mg Ni/kg of soil) applied as  $\text{NiCl}_2 \cdot 6 \text{H}_2\text{O}$  decreased the activity of dehydrogenases, urease and acid and alkaline phosphatase. This decrease was determined by the metal dose introduced into the soil. The nickel contamination had a stronger effect in the soil sown with spring barley than in the unsown soil. In the heavy loamy sand sown with spring barley, the activity

of dehydrogenases and urease were inhibited by 50% when the nickel contamination doses were 300 and 400 mg, and 200, 300 and 400 mg, respectively. In the silty light loam sown with spring barley, the activity of dehydrogenases and alkaline phosphatase was reduced by 50% in the case of the soil contamination of 400 mg Ni. The inhibition of the activity of the other enzymes did not exceed 50%. Greater activity of dehydrogenases, urease and acid and alkaline phosphatase was observed in the soil sown with spring barley than in the unsown soil. The activity of urease and alkaline phosphatase was higher in the heavier soil (silty light loam) than in the less compact soil (heavy loamy sand), however, the activity of dehydrogenases and acid phosphatase was higher in the heavy loamy sand.

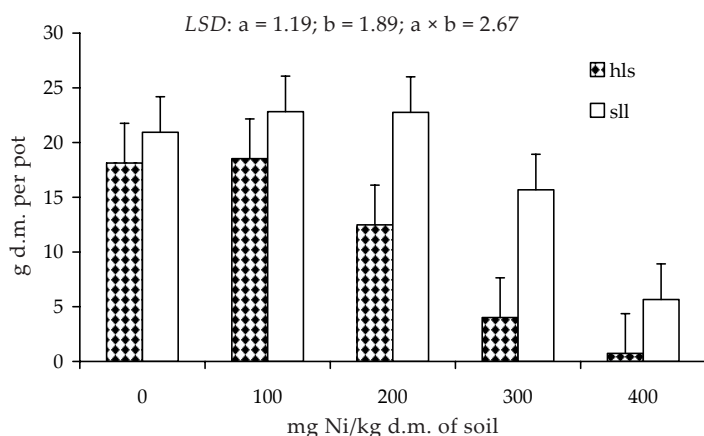


Figure 1. Effect of soil contamination with nickel on the yield of spring barley in g dry matter per pot

LSD (least statistical difference) for: a – soil type, b – nickel dose  
 LSD significant at *P* < 0.01  
 hls – heavy loamy sand, sll – silty light loam

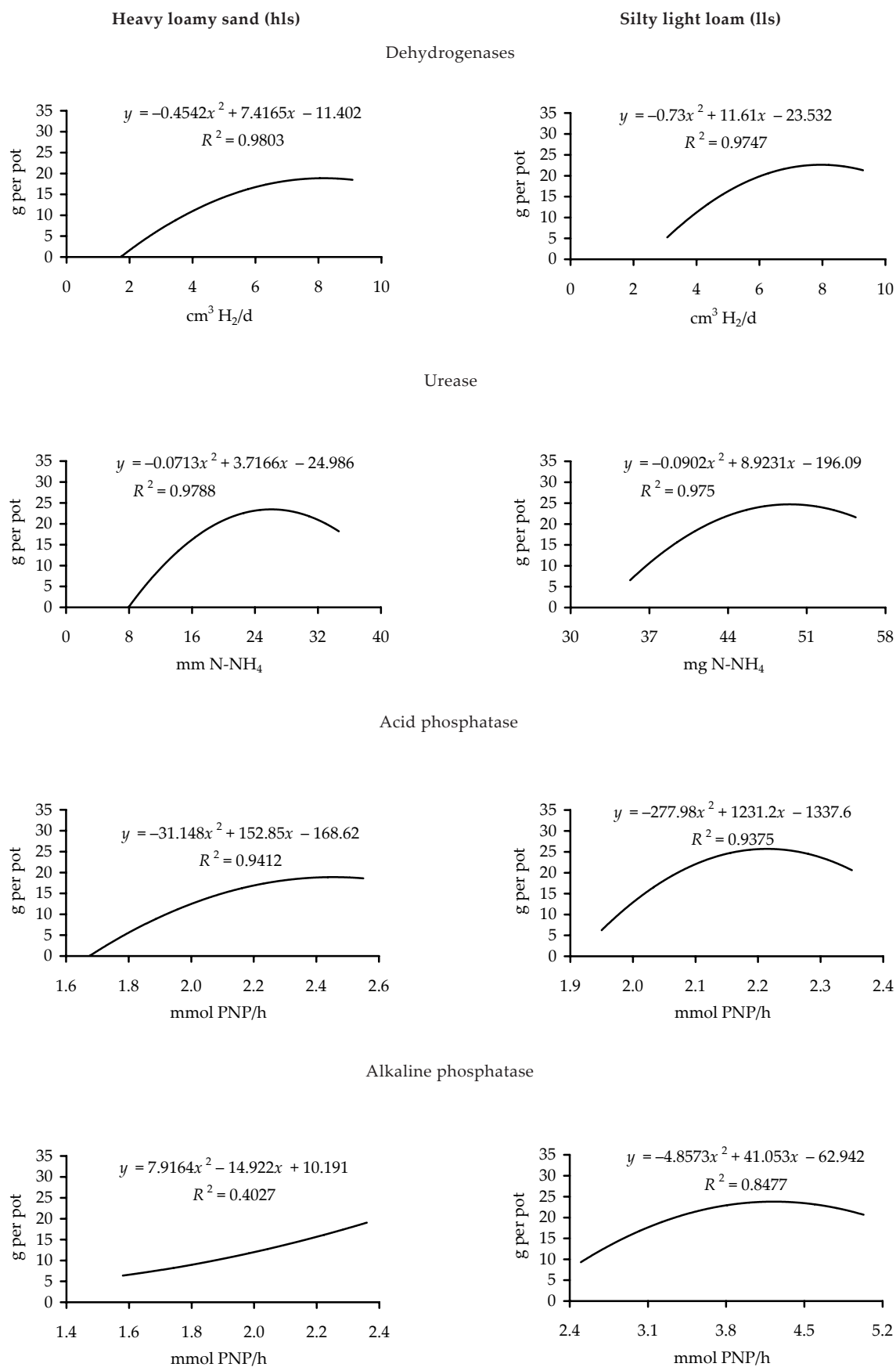


Figure 2. Correlation between spring barley yield and enzyme activity in 1 kg d.m. of soil



The same effect was reported in previous studies (Nowak et al. 2000, Wyszowska and Kucharski 2003, Wyszowska and Wyszowski 2003a). According to Nowak et al. (2000) even a small nickel dose (Ni – 25 mg/kg of soil) can affect the enzymatic activity and even 60 mg/kg of this contaminant can reduce crop yields (Koszelnik-Leszek 2002). The spring barley response to the soil contamination with nickel was determined by the soil granulometric composition. Doses of 100 mg and 200 mg Ni did not decrease the yield of spring barley cultivated in the silty sandy loam, while the dose of 200 mg Ni/kg had a clear toxic effect on this crop cultivated on the heavy loamy sand and the application of the highest dose of this metal almost entirely inhibited the growth and development of this plant. This indicates that for an analysis of the effect of a given metal on the soil metabolism and crop yielding, the soil type must be considered. Based on the completed studies, it was difficult to determine explicitly the soil contamination level at which nickel can modify the soil biological properties. When based only on model experiments without sowing the soil, the conclusions can be erroneous. In such a case, nickel inhibits an enzyme activity but it usually does not exceed 50%. On the other hand, the same level of contamination caused significantly greater inhibition of the activity in sown soils and the highest doses (200–400 mg/kg) can inhibit the activity of some enzymes over 50%. Dehydrogenases and urease were found to be particularly sensitive to soil contamination with nickel. The measurement of dehydrogenases activity seems to be a more objective indicator of soil contamination with nickel than the activity of urease because the dehydrogenases response to this xenobiotic is less dependent on the soil type. In addition, Burns (1982) and Januszek (1999) also reported on the suitability of the measurement of dehydrogenases activity for the determination of soil biological condition.

## REFERENCES

- Abraýman S.A. (1993): Variation of enzyme activity of soil under the influence of natural and antropogenic factors. *Eurasian Soil Science*, 25: 57–74.
- Burns R.G. (1982): Enzyme activity in soil: Location and a possible role in microbiological ecology. *Soil Biology and Biochemistry*, 14: 423–427.
- Casida L.E, Klein J.D., Santoro D. (1964): Soil dehydrogenases activity. *Soil Science*, 98: 371–374.
- Dick R.P. (1994): Soil enzyme activities as indicators of soil. In: Doran J.W., Coleman D.C., Bezdicek D.F., Stewart B.A. (eds.): *Defining soil quality for a sustainable environment*. Special Publ. 35, Soil Science Society Inc., Madison, Wisconsin: 107–124.
- Gołębiewska D., Grzyb-Miklaszewska J. (1991a): Kompleksy humus-enzym. I. Aktywność enzymatyczna gleb w świetle właściwości kompleksów humus-enzym. *Postępy Nauk Rolniczych*, 4/5/6: 105–115.
- Gołębiewska D., Grzyb-Miklaszewska J. (1991b): Kompleksy humus-enzym. II. Oddziaływanie kompleksów humus-enzym w układach modelowych *in vitro*. *Postępy Nauk Rolniczych*, 4/5/6: 117–127.
- Gorin G., Ching Chang Ch. (1966): A new method of assay the specific enzymic activity. IV. Urease. *Analytical Biochemistry*, 17: 49–58.
- Januszek K. (1999): Aktywność enzymatyczna wybranych gleb leśnych Polski południowej w świetle badań polowych i laboratoryjnych. *Zeszyty Naukowe AR w Krakowie*, 250: 1–130.
- Koper J., Piotrowska A. (1996): Aktywność enzymatyczna gleby płowej w zależności od uprawy roślin w zmiانowaniu i monokulturze. *Roczniki Gleboznawcze*, 47: 89–100.
- Koszelnik-Leszek A. (2002): Dynamika pobierania niklu przez dwie odmiany jęczmienia jarego. *Roczniki Gleboznawcze*, 53: 41–49.
- Kucharski J., Ciećko Z., Niewolak T., Niklewska-Larska T. (1996): Aktywność drobnoustrojów w glebach zaliczanych do różnych kompleksów przydatności rolniczej nawożonych azotem mineralnym. *Acta Academica Agriculturae et Technica Olsztyn, Agriculture*, 62: 25–35.
- Kucharski J., Wyszowska J. (2004): Inter-relationship between number of microorganisms or spring barley yield and degree of soil contamination with copper. *Plant, Soil and Environment*, 50: 243–249.
- Nowak J., Szymczak J., Słobodzian T. (2003): Próba określenia 50% progu toksyczności dawek różnych metali ciężkich dla fosfatyz glebowych. *Zeszyty Problemowe Postępów Nauk Rolniczych*, 492: 241–248.
- Nowak J., Tyrakowska-Bielec U., Szymczak J. (2000): Wpływ chlorku rtęci i niklu na zmiany aktywności fosfatyz w czarnych ziemiach. *Roczniki Gleboznawcze*, 51: 5–16.
- Pacha J. (1984): Relacje między mikroorganizmami, enzymami, materią organiczną i koloidami glebowymi oraz ekologiczne znaczenie tych procesów. *Postępy Mikrobiologii*, 23: 91–107.
- Šmejkalová M., Mikanová O., Borůvka L. (2003): Effects of heavy metal concentrations on biological activity of soil micro-organisms. *Plant, Soil and Environment*, 49: 321–326.
- StatSoft, Inc. (2001). STATISTICA (data analysis software system), version 6. [www.statsoft.com](http://www.statsoft.com).
- Tabatabai M.A., Bremner J.M. (1969): Use of p-nitrophenyl phosphate for assay of soil phosphatase activity. *Soil Biology and Biochemistry*, 1: 307–310.
- Tresar-Cepeda C., Leiros C., Gil-Sotres F., Seoane S. (1998): Toward a biochemical quality index for soil: An expression relating several biological and biochemical properties. *Biology and Fertility of Soils*, 26: 100–106.



Welp G. (1999): Inhibitory effect of the total water-soluble concentrations of nine different metals on the dehydrogenase activity of a loess soil. *Biology and Fertility of Soils*, 30: 132–139.

Word Reference Base for Soil Resources. (1998): FAO. Rome. *World Soil Resources. Reports* 84.

Wyszkowska J., Kucharski J. (2003): Właściwości biochemiczne i fizykochemiczne gleby zanieczyszczonej metalami ciężkimi. *Zeszyty Problemowe Postępów Nauk Rolniczych*, 492: 435–442.

Wyszkowska J., Wyszkowski M. (2003a). Effect of soil contamination with nickel on enzymatic activity. *Polish Journal of Natural Science*, 14: 299–307.

Wyszkowska J., Wyszkowski M. (2003b): Wpływ niklu i magnezu na namnażanie się drobnoustrojów w glebie pod uprawą łubinu żółtego. *Roczniki Gleboznawcze*, 54: 73–81.

Zheng-ChungRong, Tu-Cong, Chen-HuaiMan, Zheng C.R., Tu C., Chen H.M. (1999): Effect of combined heavy metal pollution on nitrogen mineralization potential, urease and phosphatase activities in a typic udic ferrisol. *Pedosphere*, 9: 251–258.

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## ABSTRAKT

### Vliv kontaminace niklu na půdní enzymatické aktivity

V nádobovém pokusu byl sledován vliv kontaminace půdy niklem aplikovaným v dávkách 100, 200, 300 a 400 mg Ni/kg půdy na aktivitu enzymů dehydrogenázy, ureázy a kyselá a alkalická fosfatázy. Pro pokus byly použity těžká hlinito-písčité a lehká prachově-hlinitá půda. V jedné variantě pokusu byl na půdách pěstován jarní ječmen, v paralelní variantě rostliny na pokusných půdách pěstovány nebyly. Enzymatické aktivity byly stanovovány po 14, 28, 42 a 56 dnech pokusu. Bylo zjištěno, že kontaminace půdy niklem aplikovaným jako  $\text{NiCl}_2 \cdot 6 \text{H}_2\text{O}$  snížila aktivitu dehydrogenázy, ureázy, kyselá i alkalická fosfatázy, přičemž míra poklesu aktivity souvisela s aplikovanou dávkou kovu. Nikl měl větší vliv na půdy, na nichž byl pěstován jarní ječmen, než na půdy neoseté. V těžké hlinito-písčité půdě dávky 300 a 400 mg Ni/kg půdy způsobily více než 50% inhibici dehydrogenázy a dávky 200, 300 a 400 mg Ni/kg půdy inhibovaly ve stejné míře aktivitu ureázy. V lehké prachově-hlinité půdě, na níž byl pěstován jarní ječmen, byla zjištěna více než 50% inhibice aktivity dehydrogenázy a alkalická fosfatázy při koncentraci 400 mg Ni/kg půdy. Inhibice ostatních enzymů nepřesáhla 50 %. Ureáza a alkalická fosfatáza vykazovaly vyšší aktivitu na těžší půdě, zatímco aktivita dehydrogenázy a kyselá fosfatázy byla vyšší na lehčí půdě. Pěstování jarního ječmene na niklem nekontaminovaných půdách mělo pozitivní vliv na enzymatické aktivity a zároveň byly zjištěny i pozitivní korelace mezi výnosem jarního ječmene a aktivitou dehydrogenázy, ureázy, kyselá a alkalická fosfatázy.

**Klíčová slova:** kontaminace půdy; nikl; jarní ječmen; enzymatická aktivita; dehydrogenáza; ureáza; kyselá fosfatáza; alkalická fosfatáza

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