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The Biological Properties of Soil as Influenced by Chromium Contamination

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Abstract

In this experiment the effect of chromium (VI) applied at doses of 0, 40, 80 and 120 mg·kg^{-1} of soil on the growth and development of oats, and on the presence of the following bacterial groups: copiotrophic, oligotrophic, nitrogen fixing, *Azotobacter* sp. as well as actimycetes and fungi. Activity of some soil enzymes were also determined i.e.: dehydrogenases, urease, acid and alkaline phosphatases. An attempt to diminish negative impact of chromium on soil and crops was made by the addition of barley straw to the soil. The experiment was performed on loamy sand of value pH in 1M KCl of 6.6. Studies were performed in bare soil or with oats cover cv. Komes in two experimental series with and without straw amendment.

In the result of studies it was found that hexavalent chromium applied at doses of 80 and 120 mg·kg^{-1} of soil significantly inhibited growth and development of oats. Chromium adverse effects on activity of soil dehydrogenases, urease and acid and alkaline phosphatases and the number of *Azotobacter sp.* and actinomycetes, whereas stimulation of the proliferation of oligotrophic, copiotrophic, ammonifers and nitrogen fixing bacteria was observed. Soil amendment with straw resulted in enhanced activity of all studied enzymes and abundance of majority of studied microbial communities. It also diminished toxic effects of applied chromium.

Keywords: chromium (VI), microbial communities, enzymatic activity, organic substances

Introduction

One of the most common heavy metals in the environment is chromium. Transformation of chromium in the soil is a very complex process due to the variety of its forms of occurrence and factors which could modify its chemical form [1]. Chromium in soil and water usually occurs as trivalent or hexavalent ion [2]. Occurrence of different chromium species is determined by chemical and physical properties of the soil [3, 4, 5].

The chemical form of chromium determines its availability for microorganisms. The difference in toxicity between compounds of tri- and hexavalent chromium is determined by their different chemical properties [6]. Because toxicity of hexavalent chromium is from 100 to 1000 times higher then trivalent [7] it is very important to investigate the effects of hexavalent chromium on higher plants and microbial communities. This was the aim of our experiment where soil was contaminated by different doses of hexavalent chromium. An attempt was undertaken to diminish chromium's negative effects on oat yield and microbial communities made by organic matter amendment (barley straw).

Methods

Studies were made in a cold greenhouse in plastic pots (in four replicates) with 3 kg of loamy sand (chemical parameters of the soil were as follows: pH in 1M KCl - 6.6; Hh - 1.16 mmol • 100 g^{-1} of the soil; S - 14.1 mmol • 100 g^{-1} of the soil; V - 92.4%). The tested plant was oats cv. Komes (25 plants per pot). Before establishing the experiment soil was mixed with fertilizers and with chromium dichromate according to the design of the experiment. The following doses of mineral nutrients were applied (expressed as g · kg^{-1} of the soil): N - 0.15 [$CO(NH_2)_2$]; P - 0.1 [K2HPO4]; K - 0.15 [K_2HPO_4 + KCl]; Mg - 0.05 [$MgSO_4 \cdot 7 H_2O$]. To the soil $K_2Cr_2O_7$ was introduced in doses expressed as Cr · kg^{-1} of the soil as follows: 0, 40, 80 and 120. Aim different doses application of chromium was examination degree of soil contamination by this metal on biological properties. The experiment was performed in two series: soil with or without finely ground barley straw at the dose of 4 g of straw · kg^{-1} of the soil. Our earlier investigation indicated that straw can be used for heavy metal and petrol detoxication.

Studies were conducted on soil with or without oat cover. During plant growth (51 days) soil humidity was maintained at the level of 60% capillary water holding capacity. After oat harvesting at the stage of panicles emergence yield was determined and in soil from pots with or without plant cover abundance of the following microbial communities was determined: oligotrophic (Olig) and copiotrophic (Cop) bacteria on diluted and concentrated substrates with peptone and meat extract according to Onta and Hattori's method [8], actinomycetes (Act) according to Kiister and Williams's methods according to procedure presented by Parkinson et al. [9], fungi (Fun) - on glucoso-peptone agar according to Martin [10] and *Azotobacter* sp. according to Fenglerowa's method [11], nitrogen fixing microbes (Im) and ammonifiers (Am) - on Winogradski's medium [12]. The activity of the following enzymes was also determined: soil dehydrogenases (Deh) according to Lenhard's method modified by Casidy et al. [13], urease (Ure) - according to Gorin and Chine Changa [14] and acid (Pac) and alkaline (Pal) phosphatases - according to Tabatabai's and Bremener's method [15].

Results and Discussion

Our studies showed that the strength of toxic effects of hexavalent chromium on oats was related to the level of soil contamination (Fig. 1). Yield of the crop in the conditions of the lowest dose of chromium application (40 mg Cr · kg^{-1} of the soil) was close to the yield from control treatment. Studied pollutant introduced at doses of 80 or 120 mg Cr · kg^{-1} of the soil significantly inhibited growth and development of studied crops which resulted in reduction of the yield by 2.4 and 8.9 times, respectively.

Amendment of the soil with barley straw was effective in diminishing the negative influence of chromium, which was clearly seen under the conditions of high level of contamination. In the series with straw yield of oats dry matter in pots contaminated with 120 mg Cr · kg^{-1} of the soil was 7 times higher compared to the treatments without straw amendment.

From the results of our studies and available literature [3, 4, 7] it might be concluded that hexavalent chromium is toxic for plants irrespective of soil properties [16]. Adverse effects of chromium on plants is related to plant species as well as to the level of soil contamination, which is confirmed by our results [17]. Susceptibility to this element is determined by the level of its oxidation. According to Filipek-Mazur and Soja [4] and [16] chromium applied as potassium dichromate (chromium VI) is very toxic to plants whereas chromium sulphate (chromium III) manifested slight effects on plant yield. It appears that toxic effects of hexavalent chromium might be at least partly overcome by amendment with straw which affect biological, physical and chemical properties of the soil.

Oxidation and reduction of chromium in the soil are microbiological processes [18, 19, 20]. In our studies chromium introduced to the soil as potassium dichromate modified abundance of all tested microbial groups but the direction of changes was not always uniquivocal (Table 1). In light of our results it seems that microorganisms were more susceptible to chromium than oats. The most susceptible appeared to be *Azotobacter* sp., because under the condition of the application of lowest Cr dose e.i. 40 mg Cr · kg^{-1} 24 times decrease of its abundance was noted and dose of 120 mg Cr · kg^{-1} resulted in absolute disappearance of this species. Chromium application irrespective of oats growing adversely affected the number of actinomycetes, which was seen as 2.4 and

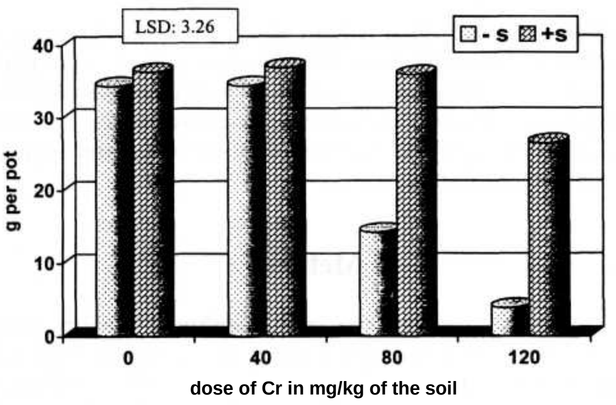

Fig 1. Yield of dry matter of oats in g·pot^{-1}
- s: soil without straw applied
+ s: soil with straw applied

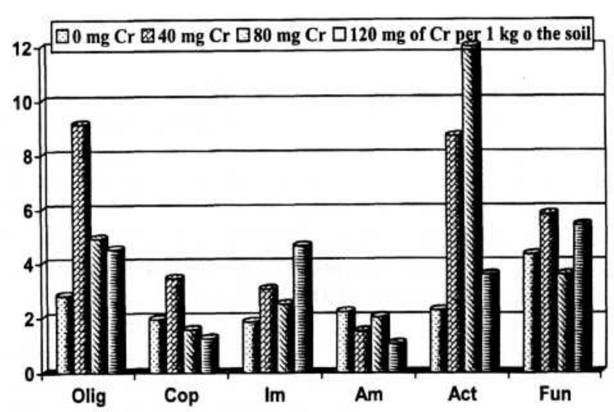

Fig. 2. Ratio of the number of microbes in straw fertilization soil to non-fertilization soil.

Table 1. Number of microorganisms (cfu) in 1 g of soil dry matter.

Cr dose (mg • kg^{-1} of soil)	Olig x 10^6	Cop x 10^6	Im x 10^6	Am x 10^6	Az	Act x 10^6	Fun x 10^3
soil not sown without straw applied							
0	2.67	3.07	4.09	4.43	5.30	3.37	3.79
40	1.74	2.48	3.44	4.30	0.00	0.67	4.07
80	1.02	3.68	3.71	4.97	0.36	0.84	4.73
120	4.57	4.18	2.38	14.48	0.00	1.15	2.97
x	2.50	3.35	3.41	7.04	1.42	1.51	3.89
soil not sown with straw applied							
0	6.94	6.69	7.31	5.67	0.00	8.26	20.47
40	13.71	13.6	15.03	13.31	0.00	16.06	26.04
80	20.48	20.51	22.74	20.96	0.00	23.86	20.04
120	17.98	14.41	22.15	15.47	0.00	18.96	14.03
x	14.78	13.8	16.81	13.85	0.00	16.78	20.15
soil sown without straw applied							
0	2.36	4.17	5.71	4.76	62.4	3.81	4.19
40	2.12	4.79	7.98	18.30	8	3.38	6.31
80	5.91	18.18	14.93	19.68	2.60	2.83	5.54
120	4.99	19.18	5.68	24.13	0.74	1.58	2.93
x	3.85	11.58	8.58	16.72	0.00	2.90	4.74
soil sown with straw applied							
0	7.19	7.38	10.73	14.66	13.2	8.06	13.96
40	21.54	11.50	19.88	20.54	6	19.09	34.23
80	13.67	13.60	23.93	28.63	4.84	20.26	16.67
120	25.27	14.81	15.21	25.38	2.96	24.3	17.87
x	16.92	11.82	17.44	22.30	0.00	3	20.68
LSD for:							
a	1.42	1.46	1.86	2.39	5.43	1.41	2.4
b	0.79	0.75	0.89	3.43	3.57	0.86	0
c	0.50	0.88	1.29	2.93	2.26	n.s.	1.71
a x b	1.57	1.50	1.78	n.s.	7.14	1.71	n.s.
a x e	1.00	1.75	2.58	n.s.	4.51	n.s.	3.43
b x c	n.s.	1.24	1.82	n.s.	3.19	n.s.	2.71
a x b x c	1.42	2.48	3.65	n.s.	6.38	n.s.	n.s.

a - chromium dose, b - straw amendment, c - plant cover of the soil, n.s. - not significant difference.

2.9-times reduction of abundance, respectively. However, in the case of oligotrophic, copiotrophic, nitrogen fixing bacteria and ammonifiers positive correlation between chromium contamination of the soil and abundance of microbes communities in the series with oats growing in pots was stated. In this series the highest chromium dose showed the highest stimulation of copiotrophic and ammonifiers reaching 4.7 and 5.1 increase of abundance, respectively. Similar relationships, excluding oligotrophic and nitrogen-fixing bacteria were observed in soil without oat cover. Chromium contamination did not affect development of soil fungi, contrary to results obtained by Babich et al. [18] and Galus [21], In an experiment performed by mentioned authors hexavalent chromium appeared to be more toxic than trivalent to the development of mycelium *of Aspergillus giganteus, Oospora sp., Rhizopus stolonifer, Penicillium vermiculatum, Trichoderma viridae* and *Aspergillus flavus.*

In our studies soil amendment with straw significantly increased abundance of all microbial groups excluding *Azotobacter* sp., and organic substance showed the most visible effects on abundance of oligotrophic bacteria, actinomycetes and fungi (Fig. 2). Growing of oats positively affected abundance of all tested microbial communities; however this increase was not always statistically significant (Fig. 3).

Enzymatic activity of the soil is determined among other factors also by microbial abundance [22], and in light of results already presented it is understandable that chromium by modifying this parameter affected the activity of dehydrogenases, urease and phosphatase (Table 2). Chromium effects on the activity of soil enzymes was related to the level of contamination. It adversely affected dehydrogenase, urease and both phosphatases. Inhibition was related to the introduction of plant cover to the pot (Fig. 4). It was more visible in bare pots and weaker on

Table 2. Enzymatic activity in 1 g of soil dry matter.

Cr dose (mg • kg^{-1} od soil)	Dehydrogenases (mm^3 H_2)	Urease µg hydrolized urea • 1 h^{-1})	Phosphatase (µmol p-nitrophenol)	
			acid	alkaline
soil not sown without straw applied				
0	2.90	26.41	1.38	1.46
40	1.34	19.97	1.04	1.40
80	1.17	17.00	0.95	1.35
120	0.25	14.67	0.73	1.32
x	1.42	19.51	1.03	1.38
soil not sown with straw applied				
0	3.09	28.63	1.97	1.71
40	1.45	21.66	1.29	1.60
80	1.30	18.96	1.10	1.54
120	0.81	16.63	1.09	1.52
x	1.66	21.47	1.36	1.59
soil sown without straw applied				
0	13.8	185.65	1.67	2.00
40	6.38	72.04	1.25	1.86
80	1.83	37.72	1.14	1.64
120	0.30	36.37	0.78	1.52
x	5.58	82.95	1.21	1.76
soil sown with straw applied				
0	14.8	212.24	1.93	2.33
40	9.68	144.93	1.51	2.36
80	6.91	140.03	1.39	2.17
120	2.45	88.60	1.30	2.05
x	8.46	146.45	1.53	2.23
LSD for*:				
a	0.05	0.10	n.s.	0.01
b	0.05	0.63	0.01	0.01
c	0.06	0.59	0.01	0.01
a x b	0.09	1.26	0.02	0.02
a x e	0.11	1.18	0.01	0.02
b x c	0.08	0.84	0.01	0.01
a x b x c	0.16	1.68	0.02	0.03

* Explanation as in Table 1.

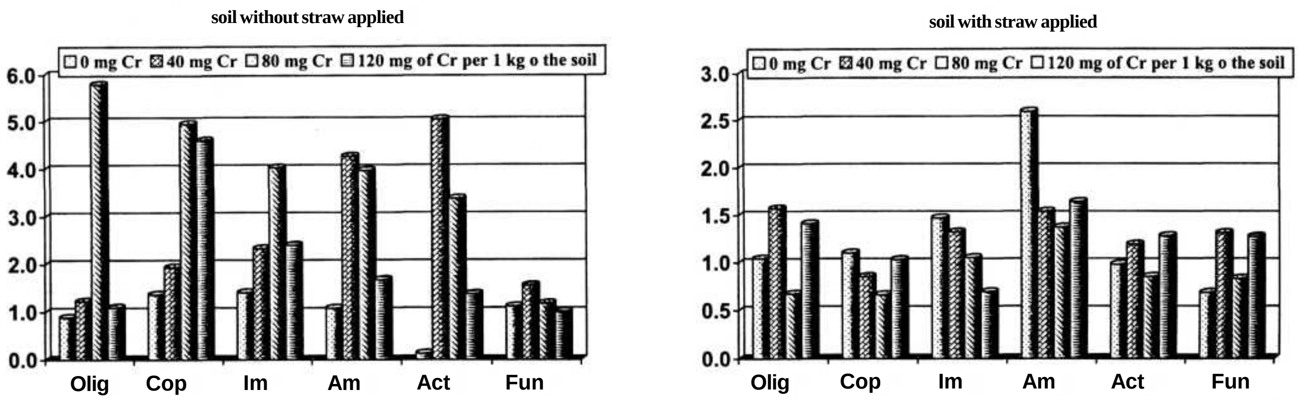

Fig. 3. Ratio of the number of microbes in sown versus unsown soil.

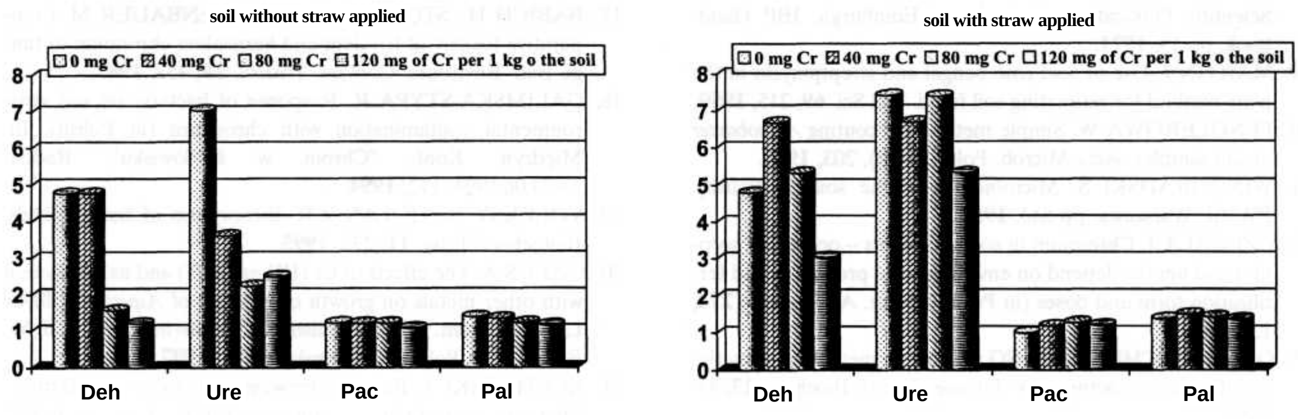

Fig. 4. Ratio of enzymatic activity in sown versus unsown soil.

soils with plant cover. It might be explained by favourable effects of root system on microbial communities abundance and indirectly on activity of their enzymes. Some effects of enzymes secreted by hair root cells should not be excluded.

Enzymatic activity of the soil was also determined by the straw amendment (Fig. 5). This is attributed to the positive effects of organic substance on oat yield and the number of microbial communities and especially to some detoxication effects of straw in terms of growth and development of the crop, microbial abundance and activity of soil enzymes. Activity of dehydrogenases in soil with plant cover with straw amendment and the highest chromium dose increased more that 8 times in relation to the series without straw. Similar relationships between the level of soil contamination by chromium and microbial communities and enzymatic activity of the soil was found in our earlier studies [23].

Decrease of the activity of soil enzymes under effects of applied chromium should be considered as very unfavourable in terms of soil fertility because soils of good quality and high content of soil organic matter show high enzymatic activity [24].

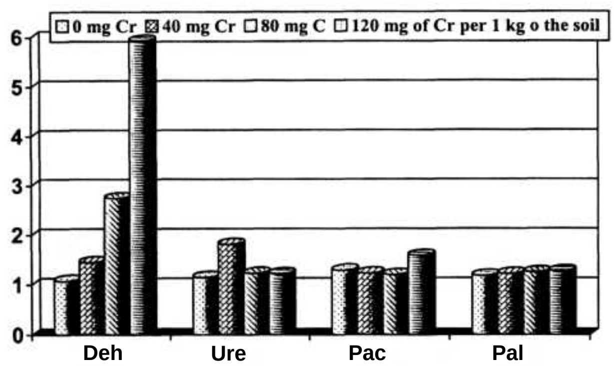

Fig. 5. Ratio of enzymatic activity in straw fertilization soil to non-fertilization soil.

Conclusions

1. Hexavalent chromium applied at doses of 80 and 120 mg Cr • kg^{-1} of soil significantly affected growth and development of oats.
2. Chromium adversely affected activity of soil dehydrogenases, urease, and alkaline phosphatase and number of *Azotobacter* sp. and actinomycetes, whereas stimulated proliferation of oligotrophic, copiotrophic, ammonifiers and nitrogen fixing bacteria was observed.
3. Straw addition to the soil intensified activity of all soil enzymes and increased the number of microbial communities. This contributed to the diminishing of toxic chromium effects.

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