Lucrări stiinţifice **Zootehnie si Biotehnologii**, vol. **42 (1)** (**2009**), Timisoara

STUDY ON PHYTOEXTRACTION BALANCE OF ZN, CD, PB FROM MINE-WASTE POLLUTED SOILS BY USING *MEDICAGO SATIVA* AND *TRIFOLIUM PRATENSE* SPECIES

STUDIUL BILANTULUI FITOEXTRACTIV AL ZN, CD SI PB DIN SOLURI POLUATE CU STERIL DE MINA IN CAZUL UTILIZARII SPECIILOR DE *MEDICAGO SATIVA* SI *TRIFOLIUM PRATENSE*

B. LIXANDRU[1], ANCA PRICOP[1], N. DRAGOMIR[1], SMARANDA MASU[2], C. BOGATU[2], I. PET[1]

[1]*Faculty of Animal Sciences and Biotechnologies, Timisoara, Romania,* ben_lixandru@yahoo.com
[2] *National Institute for Research and Development of Industrial Ecology (ECOIND) from Bucuresti, branch of Timisoara*

For a term of two years was studied phytoextractive potential of Zn, Cd and Pb using successive culture of alfalfa (Medicago sativa) and red clover (Trifolium pratense). In the experimental plot was incorporated a quantity of 20 kg mine waste per square meter, providing in soil 1209 mg Zn/kg d.s., 4.70 mg Cd/kg d.s. and 188.2 mg Pb/kg d.s. The metals content accumulated in plants was determined at the two moments of biomass harvesting, and through balance calculations we could establish the phytoextraction efficiency of the two forage-grasses species. The obtained results indicate that both perennial forage-legumes species have a good phytoextractive capacity and tolerance for Zn and Pb, especially Trifolium pratense specie. By using this species as phytoextractors on soil polluted with 3.76 times more Pb and 4.03 times more Zn, is provided the reduction of metallic ions concentration in soil to limits admitted by laws in a period of 3, respectively, 4 years.
Keywords: pollution, Zn, Cd, Pb, phytoextraction, alfalfa, red clover

Introduction

During the process of extraction from soil of essentials macro- and micro-nutrients, plants have developed a specific phytoextractive mechanism helping at extraction, translocation and storage of nutrients. In general, metals taking-over mechanism are selective, plants having preferences for one kind of ions. This selectivity depends on structure and properties of biological membranes that allow the recognition of transporters agent, captivation and intermediation of transport in membrane of specific metal ions. Secondly, the extractive plant must have biological capacities of toxicity resistance of tissue accumulated or storage metals.

Due to these characteristics, there is a large genetic variability of plants with variable and exiguously known phytoextractive potential. Plants capable to accumulate metals in quantities of about 100 times over the average registered in the same ecological conditions, are considerate hiperaccumulator plants.

The aim of this research was to study the phytoextractive potential of two perennial forage-legumes species, *Medicago sativa* and *Trifolium pretense*.

Materials and Methods

In two experimental plots, in surface of 3 sq m, a experimental plot with a content of 20 kg of mine-waste per sq m and control plot, was studied the phytoextractive potential of Zn, Cd and Pb using two perennial forage-legumes species, alfalfa (*Medicago sativa*) and red clover (*Trifolium pretense*). The mine-waste originated from the Copper Exploitation Factory in Moldova Nous, Caras-Severin County. The chemical composition of the mine-waste was as follows: Cu – 669 mg / kg d.s., Zn – 610 mg / kg d.s., Mn – 676 mg / kg d.s., Cd – 2.76 mg / kg d.s. and Pb – 110 mg / kg d.s. The mine-waste bedding thickness per surface unit was about 8 cm, and through agro technical processing was incorporated at a depth of about 20 cm, resulting in a final mass ratio between mine-waste and soil of 1:2,5.

The experiment lasted two years. At the end of each year, plants were removed with roots from soil for analyzes. In the first year, alfalfa was cultivated, followed by red clover in the second year. Before each sowing was determined the Zn, Cd and Pb content in the soil, determining the polluting "contribution" of the mine-waste.

The analysis of metal quantity accumulated in the plants was made through two harvests. At the last mowing, the average sample also contained samples from the radicular system. Through balance calculations we could establish the metal quantity extracted each year by the two fodder plant species; in the end we also determined the phytoextraction efficiency. The chemical analyses have been performed through spectrophotometry in the Technological Research Platform of USAMVB Timisoara.

Results and Discussion

In Table 1 are presented the results of the phytoextractive balance of Zn, Cd and Pb from the soil, through cultivation of alfalfa (*Medicago sativa*).

Table 1

Metal phytoextraction balance for the species *Medicago sativa*

Metal pollutants	Maximum admitted limit of heavy metals in soil in Romania * (mg / kg d.s.)	Control parcel					Experimental parcel				
		Amount of metals at the beginning of the experiment (mg / kg d.s.)	Extracted amount (mg / kg d.s.)			Phytoextraction efficiency (%)	Amount of metals at the beginning of the experiment (mg / kg d.s.)	Extracted amount (mg / kg d.s.)			Phytoextraction efficiency (%)
			I-st harvest	II-nd harvest	Total			I-st harvest	II-nd harvest	Total	
Zn	300	67.80	22.6	35.7	58.30	85.9	1209.0	149.60	174.50	324.10	26.80
Cd	3	0.90	0.31	Udl	0.31	34.4	4.70	0.31	0.36	0.67	14.25
Pb	50	30.00	12.5	10.7	23.2	77.3	188.20	38.20	49.60	87.80	46.65

* Order 344/2004

It can be ascertained that the soil form the control plot had a metal concentration bellow the maximum admitted limits by Romanian legislation. In this case, alfalfa phytoextractive efficiency was: 85.9% in case of Zn, 34.4% for Cd and 77.3% for Pb.

In case of the soil polluted with mine-waste the recorded values was 17.8 times higher in case of Zn, 5.2 times higher on Cd and 6.27 times higher in case of Pb. In this situation with a larger quantity of metallic ions in soil, alfalfa plants extract significant quantities of Zn and Pb. Extracted and bio accumulated cadmium quantities were lower and comparable with values obtained on control-unpolluted soil.

In both harvesting, from a soil with 17 times higher Zn quantity, alfalfa plants extract about 6 times more Zn in comparison with unpolluted soil. Even so, phytoextractive efficiency is 26.8%, being 3 times lower compared to phytoextractive efficiency of plants cultivated on soil with lower metals ions concentration.

Same situation is registered in case of ionic excess of Pb in soil. Thus, alfalfa plants extract about 3-5 times more Pb on first and second harvest, in comparison with plants cultivated on unpolluted soil. Compared to Zn, the difference is that Pb phytoextractive efficiency is barely 1.65 times lower then in case of plant cultivated on soil with lower metal ionic concentration.

Concluding, alfalfa is pretty well adapting to polluted soils with Zn and Pb ionic excess. Moreover, this specie proves to be a very good Zn extractor and then a Pb extractor.

In Table 2 are presented the results of the phytoextractive balance of Zn, Cd and Pb from the soil, through cultivation of red clover (*Trifolium pratense*).

Table 2

Metal phytoextraction balance for the species *Trifolium pratense*

Metal pollutants	Maximum admitted limit of heavy metals in soil in Romania * (mg / kg d.s.)	Control parcel					Experimental parcel				
		Amount of metals at the beginning of the experiment (mg / kg d.s.)	Extracted amount (mg / kg d.s.)			Phytoextraction efficiency (%)	Amount of metals at the beginning of the experiment (mg / kg d.s.)	Extracted amount (mg / kg d.s.)			Phytoextraction efficiency (%)
			I-st harvest	II-nd harvest	Total			I-st harvest	II-nd harvest	Total	
Zn	300	9.50	12.46	11.32	23.78	+100	884.90	82.46	114.32	196.78	22.23
Cd	3	0.59	udl	udl	udl	-	4.03	udl	udl	-	-
Pb	50	6.80	1.55	1.27	2.82	70.15	100.40	18.23	24.23	42.46	42.29

* Order 344/2004

Soil metals content, in both experimental plots, was decreased through phytoextracted quantities by using of alfalfa in antecedent experiment. Thus, in case of control plot, red clover extract the entirely quantity of Zn and over 70% of Pb quantity.

In case of soil polluted with mine-waste, red clover extract larger quantities of Zn and Pb from soil. On this experimental plot, soil had a Zn and Pb content of more then 2 times over maximum admitted limits of pollution.

Due to the quantity of phytoextracted Cd and Pb, being more then 7 times larger in comparison with unpolluted plot, in both harvesting, can be ascertain that red clover, also have a good phytoextractive potential for these metals.

Conclusions

1. On soils with Zn, Cd and Pb quantities under maximum admitted limits by ecological legislation, alfalfa and red clover register phytoextractive efficiencies of over 70% for Pb and over 80% in case of Zn.
2. In case of ions excess, although phytoextractive efficiencies are reducing to half, *Medicago sativa* extract about 5 times more Zn and 3 times more Pb, and *Trifolium pratense* extract about 8 times more Zn and over 20 times more Pb.
3. On soils polluted with 3.76 times more Pb and 4.03 times more Zn, period of phytoremediation using successively alfalfa and red clover is of 3 years in case of Pb and 4 years in case of Zn.

References

1. Chaney R.L., Angle J.S., Broadhurst C.L., Carinne Peters, Tappero R.V., Sparks D.L., 2007, Improved Understanding of Hyperaccumulation Yields Commercial Phytoextraction and Phytomining Technologies, *J. Environ. Qual*. **36**: 1429-1443

2. Cooke J.A., Johnson M.S., 2002, Ecological restoration of land with particular reference to the mining of metals and industrial minerals: A review of theory and practice. *Environ. Rev*. **10**, 41-71

3. Lasat M.M., 2002, Phytoextraction of Toxic Metals: A Review of Biological Mechanisms, *J. Environ. Qual*. **31**, 109-120

4. Lombi E., F.J. Zhao, S.J. Dunham and S.P. McGrath, 2001, Phytoremediation of Heavy Metal–Contaminated Soils - Natural Hyperaccumulation versus Chemically

5. Prasad M., Narasimha Vara, Helena Maria De Oliveira Freitas, 2003, Metal hyperaccumulation in plants - Biodiversity prospecting for phytoremediation technology, *Electronic Journal of Biotechnology*, **6** No. 3