

EXPERIMENTAL VERIFICATION OF RESULTS REPRODUCIBILITY AND RESIDUAL EFFECT IN GREENHOUSE ON ARTIFICIALLY POLLUTED SOIL WITH LEAD (I)

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Abstract

Phytoextraction is a technique to remove heavy metals from soil by direct absorption into plant tissues. The implementation of phytoextraction involves growing one or more plant species that are contaminants hyperaccumulators. Specific conditions for the application of phytoextraction program for a polluted area reffers to amendments, period of vegetation, pollution degree, etc.

The paper presents experimental research in greenhouse, aimed to check the parameters determining the reproductibility of selected plant (maize) and following the conclusions of the preliminary laboratory test experiments.

Verification consisted in achieving 2 experiments with 2 degrees of artificially soil pollution with Pb: 1000 mg Pb/kg soil and concentration of EDTA with a ratio Ligand/Lead between 0 and 0.5 and 2000 mg Pb / kg soil and ratio Ligand/Lead between 0 and 0.4 and three growing cycles (Cycle 1, Cycle 2 - residual effect, Cycle 3 - residual effect).

INTRODUCTION

Metals are natural constituents of soil. Over 100 years of industrialization have led to huge changes in the overall budget of the critical chemicals in the earth's crust [1].

Soil contamination can destroy the delicate equilibria between physical, chemical and biological processes, which influences soil fertility.

The effects of soil contamination with heavy metals are already found in many zones of the entire world [2-3].

In the recent years the number of research focusing on the study of soil pollution with heavy metals has increased due to various anthropogenic sources [4-5].

Phytoremediation, a relatively new remediation technology, is mainly designed for land contamination or areas with high pollution due to urban and industrial activity. Phytoremediation by phytoextraction is an extraction and accumulation procedure

for contaminants/pollutants in plant tissues considered hyperaccumulators-including the roots and aerial part [6].

MATERIAL AND METHODS

In this stage, two experimental series were performed in greenhouse to verify the reproductibility of determined parameters on previously selected plants and the conclusions of the preliminary test experiments.

Preliminary test experiments established:

- Remaining test plant in the experiment - maize;
- Experimental scheme will include two degrees of artificial soil pollution with lead: 1000 mg Pb / kg and 2000 mg/kg;
- Experience 1 contains 7 experimental variants in three repetitions;
- EDTA concentration (expressed as EDTA:Pb ratio) in experiment with soil polluted with 1000 mg Pb / kg will be: 0, 0.1, 0.2, 0.3, 0.4, 0.5;
- Experience 2 contains 6 experimental variants in three repetitions;
- EDTA concentration (expressed as EDTA:Pb ratio) in experiment with soil polluted with 2000 mg Pb / kg will be: 0, 0.1, 0.2, 0.3, 0.4;
- Soil type: the cambic chernozem from Fundulea;
- Physical, chemical and microbiological characteristics of soil are the same in the Preliminary Test Experiments;
- Lead and ligand treatment was applied to soil before sowing.
- Quantity of soil per pot was 8 kg for both experiments in the two series.
- Pots used were MITSCHERLICH type with a capacity by 10 L.
- Pb was applied as $Pb(NO_3)_2$ - (1000 mg/kg, 2000 mg/kg) and EDTA (ethylenediaminetetraacetic acid) at the beginning of the experiment, before sowing.
- Was not applied phytosanitary treatments, neither mineral or organic fertilizers.
- Two experimental series lasted 8 weeks each.

Series II and III used the same test plant – maize, has experienced the same pots of Series I seeking residual effect of treatment.

RESULTS AND DISCUSSION

SERIES I, harvest Experience 1 and Experience 2 – Maize crop

The treatment with lead and ligand was applied to soil before sowing (Series 1). For 8 weeks there was followed the evolution of plants starting with seedling, emergence until harvest. Regarding the aspect and appearance of maize leaves was found a high influence of treatment with Pb, with EDTA or Pb + EDTA. After harvesting, maize measurements were made as parameters as plant height and weight of biological material and dosing results lead to the establishment of lead accumulated in plants. After the analysis of variance (Tukey test, Fisher test), the statistical data showed a different evolution of these parameters according to treatment.

Experience 1 - soil treated with the same lead content (1000 mg/kg) and different EDTA content (0, 0.1, 0.2, 0.3, 0.4, 0.5) - Series 1

Table 1 shows the evolution of biomass, plant height of corn and leaves the lead content of soil polluted with 1000 mg Pb/kg with increasing ligand content so that

the $\frac{\text{EDTA}}{\text{Pb}}$ ratio to reach values of 0, 0.1, 0.2 , 0.3, 0.4, 0.5. Leaves biomass

decrease distinct significantly from control to experimental variants V4, V5, V6 and V7. The increase of leaves biomass at experimental variant V2 comparatively with control is explained by higher content of nitrogen provided by lead nitrate.

Comparatively with control, the decrease is significant starting with experimental variant V7(EDTA:Pb=0,5). So, it is considered efficient the treatment, in terms of minimum biomass decrease compared with the control, the experimental variants V5 or V6. Comparatively with the experimental variant

V2 $\left[\text{Soil} (+1000 \text{ mg Pb/kg}) + \text{EDTA} \left(\frac{\text{EDTA}}{\text{Pb}} = 0 \right) \right]$ leaves biomass has a significant decrease starting with experimental variant

V5 $\left[\text{Soil} (+1000 \text{ mg Pb/kg}) + \text{EDTA} \left(\frac{\text{EDTA}}{\text{Pb}} = 0.3 \right) \right]$. This means that the ligand

effect is distinguished from V5 experimental variant, which means that treatment with high efficiency is the experimental variant V4

$\left[\text{Soil} (+1000 \text{ mg Pb/kg}) + \text{EDTA} \left(\frac{\text{EDTA}}{\text{Pb}} = 0.2 \right) \right]$.

Plant height decrease distinct significantly comparatively with control.

As regards the leaves lead content presented in Table 1 shows a distinct significantly increase in each variant comparatively with control. Significant increase of more than 58.6 cm (DL 5%) occurs in experimental variants V5, V6 and V7. The evolution of leaves biomass, plant height and leaves Pb content, can be appreciate that until V4 experimental variant

$\left[\text{Soil} (+1000 \text{ mg Pb/kg}) + \text{EDTA} \left(\frac{\text{EDTA}}{\text{Pb}} = 0.2 \right) \right]$ was not registered a significant

decrease in biomass, this ligand does not react in negative terms while the concentration of lead increased significantly. As such, are considered as efficient treatment for phytoextraction in presence of lead concentration 1000 mg Pb/kg soil, the experimental variants V4(EDTA:Pb=0,2) and V5(EDTA:Pb=0.3)

Table 1

Biological material weight at harvest (biomass), plant height and leaves content of lead in a soil polluted with Pb 1000 mg/kg soil and different content of EDTA - Series 1

Treatment	Biomass (g)	Height (cm)	Pb (mg/kg)
V1 Control Cambic chernozem from Fundulea	190.0	57.3	5.0
V2: Soil (+ 1000 mgPb/Kg) + EDTA (EDTA:Pb=0)	215.0	51.7	44.3
V3: Soil (+ 1000 mgPb/Kg) + EDTA (EDTA:Pb=0.1)	195.3	56.7	43.6
V4: Soil (+ 1000 mgPb/Kg) + EDTA (EDTA:Pb=0.2)	171.3	52.0	61.7
V5: Soil (+ 1000 mgPb/Kg) + EDTA (EDTA:Pb=0.3)	140.0	50.3	69.6
V6: Soil (+ 1000 mgPb/Kg) + EDTA (EDTA:Pb=0.4)	132.0	46.3	74.0
V7: Soil (+ 1000 mgPb/Kg) + EDTA (EDTA:Pb=0.5)	104.3	42.7	112.0
DL 5% (Tukey test)	71.3	7.2	58.6
Fisher test	**	**	**

Experience 2 - soil treated with the same lead content (2000 mg/kg) and different EDTA content of (0, 0.1, 0.2, 0.3, 0.4) - Series 1

Table 2 shows the evolution of the three parameters (biomass, height, leaves content of Pb) in a soil polluted with 2000 mg Pb/kg with increasing content of

ligand (EDTA) in the $\frac{\text{EDTA}}{\text{Pb}}$ ratio by 0, 0.1, 0.2, 0.3, 0.4.

Plants biomass and height has a distinct significantly decrease with variants and leaves lead content a significantly distinct increase. So, treatment can be efficient in the phytoextraction process in 2000 mg Pb / kg soil by application of EDTA content to produce a ration by EDTA:Pb=0.1.

Table 2

Biological material weight at harvest (biomass), plant height and leaves content of lead in a soil polluted with Pb 2000 mg/kg soil and different content of EDTA - Series 1

Treatment	Biomass (g)	Height (cm)	Pb (mg/kg)
V1 Control Cambic chernozem from Fundulea	190.0	57.3	5.0
V8: Soil (+ 2000 mgPb/Kg) + EDTA (EDTA:Pb=0)	164.3	50.0	80.8
V9: Soil (+ 2000 mgPb/Kg) + EDTA (EDTA:Pb=0.1)	147.3	49.3	85.8
V10: Soil (+ 2000 mgPb/Kg) + EDTA (EDTA:Pb=0.2)	44.3	31.7	211.0
V11: Soil (+ 2000 mgPb/Kg) + EDTA (EDTA:Pb=0.3)	40.0	23.3	439.7
V12: Soil (+ 2000 mgPb/Kg) + EDTA (EDTA:Pb=0.4)	7.3	10.0	1994.1
DL 5% (Tukey test)	47.6	8.4	120.1
Fisher test	**	**	**

Meanwhile, the leaves biomass comparatively with the control (V1) has a significantly decrease starting with experimental variant V10 $\left[\text{Soil (+ 2000 mg Pb/kg) + EDTA} \left(\frac{\text{EDTA}}{\text{Pb}} = 0.2 \right) \right]$. This means that the efficient treatment is the undecrease of leaves biomass of experimental variant V9 $\left[\text{Soil (+ 2000 mg Pb/kg) + EDTA} \left(\frac{\text{EDTA}}{\text{Pb}} = 0.1 \right) \right]$. This variation correlates very well with leaves lead content that has a significantly increase starting with experimental variant V10.

CONCLUSIONS

1. Following the evolution of the three parameters (biomass, height, Pb leaves content) in the first cycle of vegetation, it can be stated that the treatment with ligand (EDTA) on a soil polluted with 1000 mg Pb/kg stimulates the

accumulation of lead from soil in maize plants without affecting the evolution of plants and soil characteristics at an EDTA concentration to achieve a EDTA:Pb ratio between 0.2 and 0.3. Lower concentrations of ligand are not specific to the phytoextraction process on favorable conditions, while higher concentrations cause adverse effects to plants and their vegetative evolution is affected in the first cycle of vegetation.

2. Following the evolution of the three parameters (biomass, height, leaves Pb content), it can be said that treatment with ligand (EDTA) on a soil polluted with 2000 mg Pb/kg stimulates the accumulation of lead in soil without harming the maize plants plant evolution and soil characteristics at an EDTA concentration to achieve a EDTA:Pb ratio by 0.1.
3. The statistical data showed that exists differences statistically assured in terms of biological material weight at harvest (biomass) and plant height, but the biomass lead content in function with applied treatment.
4. At the same soil concentration of lead, leaves lead content increase with the applied EDTA increasing levels, which explains the ligand capability to increase the lead solubility in soil and uptake by plants.
4. Following the preliminary experiments carried out with mustard and maize, maize has met the requirements to be selected as test plant in experiments.

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