Comparative evaluation of the efficiency of enhancing phytoextraction with heavy metals in Phaseolus vulgaris, Pisum sativum, Sinapis alba using a disubstituted potassium salt of oxyethylene diphosphonic acid

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Abstract. It is possible to increase the efficiency of phytoextraction by introducing additives that convert less bioavailable compounds of heavy metals into bioavailable fractions in the soil, as well as to increase the rate of transfer of metals into the plant body. The article presents the results of a study of various chemical additives that affect the development and growth of plants when the soil is contaminated with heavy metals (Ni, Cu, Cd). It can be concluded that the disubstituted potassium salt of hydroxyethylidene diphosphonic acid is a growth stimulant for some plant species, and an increase in growth directly depends on an increase in the concentration of the additive introduced. The work was supported by the Russian Foundation for basic research (RFBR) under the project 18-29-25071.

1. Introduction

Constant continuous industrialization, agricultural practices and other anthropogenic human activities lead to pollution of the environment with heavy metals. Heavy metals have a very serious toxic effect on animals, plants and public health, so the remediation of contaminated areas is critical [1]. Proceeding from the fact that the protective capabilities of soils are very limited, and the period of self-cleaning from pollutants is many tens and even hundreds of years, at present, a huge influence is given to the development of new, innovative methods aimed at controlling and eliminating pollution, as well as for its prevention [2].

One of the most common and hazardous chemical contamination is heavy metal contamination. Heavy metals and their compounds accumulate in the soil, especially in the humus horizons, and are extremely slowly removed when they are consumed by plants, during leaching and soil erosion (deflation).
Currently, a number of methods have been developed and tested to cleanse soils from heavy metals, but despite this, rather large areas still remain contaminated, since the environmental and economic costs of cleaning these areas using existing technologies are too high and not always payback [3].

Among the various methods used, phytoremediation is considered one of the most effective, innovative, environmentally friendly and inexpensive ways to restore soil from heavy metal pollution. Phytoremediation - the use of plants to neutralize and remove heavy metals (accumulation of pollutants in the plant's body) - is a promising strategy for cleaning up contaminated areas [3].

The efficiency of phytoremediation can be increased by introducing various components that stimulate the development and growth of plants. In this work, a disubstituted potassium salt of oxyethinidenediphosphonic acid (hereinafter referred to as K2HEDP) was used as such an additive. Chelates of microelements have high biological activity - microelements in the form of complex salts with complexing agents of organic acids: oxyethinidenediphosphonic acid (hereinafter - HEDP). To accelerate the transfer of heavy metals from soil to plants and to increase absorption, chelating agents are introduced into the contaminated environment [5, 6].

The aim of this work is to study the possibility of using the potassium complex of oxyethylene diphosphonic acid to enhance phytoextraction processes in soil contaminated with polymetals.

2. Research methodology

Research objects: seedlings of the following plants: Phaseolus vulgaris, Pisum sativum, Sinapis alba.

The Levoberezhny solid municipal waste landfill was chosen as an object for creating a model of soil contaminated with heavy metals. Soil samples to determine the content of the main pollutants were taken in June 2019.

Model experiments were carried out in accordance with ISO 22030:2005 Soil quality. Biological methods. Chronic toxicity in higher plants. Using standard laboratory equipment, scales with an accuracy of ± 0.1 mg, universal soil (pH 5.8-6.2), we used a set of plastic growing pots for planting seeds with a volume of 1 liter.

When modeling pollution with heavy metals, weighed portions of reagents containing heavy metals were added to the growing pots in an amount corresponding to 5 times of the maximum permissible concentration (hereinafter - MPC) in terms of the content in the soil (\(\text{Ni(NO}_3\text{)}_2 \times 6\text{H}_2\text{O} - 38.2 \text{ mg per 1 pot; } \text{Cu(NO}_3\text{)}_2 \times 3\text{H}_2\text{O} - 23.18 \text{ mg / kg; } \text{Cd(NO}_3\text{)}_2 \times 4\text{H}_2\text{O} - 15.65 \text{ mg per pot}\), respectively [14]. For comparison (as a control), pots were prepared with soil contaminated with heavy metals, in which the introduction of additives was not provided. Also, to create a more realistic model of the Levoberezhny municipal solid waste landfill, the alkalinity of the soil was increased by adding 40 g of \(\text{K}_2\text{CO}_3\) to each kilogram of soil (pH = 8).

To improve the structure of the soil and increase the germination of seeds, potassium humate was introduced into the soil in an amount of 0.1% of the dry soil mass. Humic substances are the most common natural complex ones.

All pots, including the control sample, were planted with seeds of Phaseolus vulgaris (14 pcs), Pisum sativum (12 pcs), Sinapis alba (30 pcs) in each pot. The seed planting process is shown in Figure 1.

To study the effect of the active component on the growth and development of plants, as well as on the degree of enhancement of phytoextraction by heavy metals of Phaseolus vulgaris, Pisum sativum, Sinapis alba, an aqueous solution of K2HEDP at concentrations of 5 and 10 mmol / l, respectively, was introduced into individual growing pots.

A sample of an aqueous solution of a disubstituted potassium salt of oxyethylene diphosphonic acid with a mass content of the target component of 28.3 % was developed and provided by the laboratory for the technology of complex ones and complex compounds of the Kurchatov Institute Research Center - IREA (Institute of Chemical Reagents and Highly Pure Chemical Substances).
All variants of pots with heavy metals with additives and with heavy metals without additives were presented in triplicate.

![Pisum sativum](image1) ![Phaseolus vulgaris](image2)

**Figure 1.** Planting the seeds

### 3. Research results

A week after planting the seeds, the number of germinated shoots was counted. The data are presented in table 1.

| Plant          | 1 pot         | 2 pot         | 3 pot         |
|---------------|---------------|---------------|---------------|
| *Phaseolus vulgaris* | 2 out of 14   | 5 out of 14   | 1 out of 14   |
| *Pisum sativum*    | 5 out of 12   | 5 out of 12   | 3 out of 12   |
| *Sinapis alba*     | 2 out of 30   | 5 out of 30   | 9 out of 30   |
| *Phaseolus vulgaris* | 6 out of 14   | 9 out of 14   | 7 out of 14   |
| *Pisum sativum*    | 4 out of 12   | 5 out of 12   | 7 out of 12   |
| *Sinapis alba*     | 5 out of 30   | 22 out of 30  | 7 out of 30   |
| *Phaseolus vulgaris* | 5 out of 14   | 5 out of 14   | 6 out of 14   |
| *Pisum sativum*    | 2 out of 12   | 2 out of 12   | 1 out of 12   |
| *Sinapis alba*     | 0 out of 30   | 5 out of 30   | 23 out of 30  |

In most of the pots, the plants were green, with some leaves showing white blotches. Some of the shoots were wilted, pale with noticeable white patches, and some of the leaves were yellowed. Plant growth was measured every three days from the time of seed growth. The data are presented in **Figures 2-4**.
Based on this graph, it can be concluded that the application of disubstituted potassium salt of hydroxyethylidene diphosphonic acid has a positive effect on the growth of Sinapis alba (in comparison with the control samples, the growth of sprouts increased 1.5 times when $K_2$HEDP was added at a concentration of 5 mmol/l and 2 times when $K_2$HEDP was added at a concentration of 10 mmol/l, respectively).

![Graph of growth of Sinapis alba shoots](image1)

**Figure 2.** Growth graph of Sinapis alba shoots

Based on this graph, the following conclusions can be drawn. When $K_2$HEDP was added at a concentration of 5 mmol / l, the growth was almost equal to that of the control sample. The introduction of $K_2$HEDP at a concentration of 10 mmol/l gave the opposite effect, the growth decreased about 4 times in comparison with the control sample.

![Graph of growth of shoots of Pisum sativum](image2)

**Figure 3.** Graph of growth of shoots of Pisum sativum
Figure 4. Graph of growth of shoots of Phaseolus vulgaris

Based on this graph, it can be concluded that the introduction of a disubstituted potassium salt of hydroxyethylidene diphosphonic acid on the growth of Phaseolus vulgaris (in comparison with the control samples, the growth of sprouts increased 1.5 times with the introduction of K$_2$HEDP at a concentration of 10 mmol/l and 2.2 times with introduction of K$_2$HEDP at a concentration of 5 mmol/l, respectively).

It can be concluded that the disubstituted potassium salt of hydroxyethylidene diphosphonic acid is a growth stimulant for some plant species, and the increase in growth directly depends on an increase in the concentration of the added additive.

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