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Use of Humic Organic Matter and Rhizobacteria in the Cultivation of Winter Wheat in Areas Polluted With Heavy Metals

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Abstract. The article presents experimental results on the reduction of the content of heavy metals in the roots, stems and grains of wheat grown on soil contaminated with heavy metals belonging to "Almalik Mining Metallurgical Combine" JSC (Joint-Stock Company) under the influence of bacterial fertilizer, potassium humate and Serhosil biopreparations created on the basis of wheat rhizobacteria. In the development phases, the resistance of the crops to the harmful effects of heavy metals was increased, the metabolism in the plant, the growth and development of the plant were accelerated, and the yield of 55 centners of grain per hectare was achieved, compared to the control option, an additional yield of 7 centners of grain per hectare was achieved.
Key words: soil, heavy metals, potassium humate, rhizobacteria (PGPR), Serhosil biopreparation, Almalyk Mining Metallurgical Combine JSC, winter wheat, growth, development, root, stem, grain, productivity.

1 Introduction

Currently, heavy metal pollution is a major environmental problem, as metal ions remain in nature due to their non-degradable nature. Most heavy metals are toxic even at low concentrations and can enter the food chain, where they accumulate and harm living organisms. All metals have the potential to cause harmful effects at high concentrations, and the toxicity of each metal depends on the amount available to organisms, the absorbed dose, and the duration of exposure [1-2].

Soils contaminated with heavy metals can contaminate agricultural crops grown in them. But despite the fact that the problem of saturation of the biosphere with heavy metals has attracted public attention in the last two decades, soil pollution with heavy metals has been studied to a lesser extent. There is no maximum permissible concentration of heavy metals in the soil-plant-animal-human system. Therefore, the search for measures to protect the food chain from heavy metals is an urgent problem [3-6].

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Environmentally safe biotechnological approaches based on local strains of effective soil and rhizosphere microorganisms should be used to reduce the negative effects of heavy metals in growing crops on soils contaminated with heavy metals.

Many bacteria, actinomycetes, yeasts, single-cell microorganisms are able to accumulate heavy metals and radionuclides in amounts thousands and millions of times higher than their physiological needs.

For example, it is known that uranium or lead are accumulated by cells of bacteria belonging to the genus Micrococcus. Cadmium, nickel, cobalt, rubidium are absorbed by Bacillus and Escherichia coli generation cells. With an increase in the concentration of heavy metals, the number of beneficial soil microflora and the enzymatic activity of the soil decreases (catalase, urease, amylase, invertase, dehydrogenase, etc.). Due to their biochemical properties, silicate- and phosphate-degrading bacteria reduce the toxic effects of heavy metals and improve plant growth.

In connection with the above, we have started work on the development of biotechnological approaches for growing wheat on soils contaminated with heavy metals located in the territory of AGMK.

Microorganisms are at the origin of the trophic chain, through which metals enter higher organisms. With the help of soil microorganisms, the soil with heavy metals

Microorganisms can be used to monitor pollution levels and as soil pollution indicators.

Over the past decades, there has been a lot of conflicting information in the literature about the effects of heavy metals on soil microbiota. Therefore, determining the general patterns of soil and plant response to heavy metal pollution and developing new biotechnological approaches to improve the quality of wheat grain is the most urgent problem today.

The use of fertilizers of natural origin plays an important role in increasing the yield and quality of agricultural crops in the conditions of the harmful effects of heavy metals, because they can form complex compounds with heavy metals in low concentrations, stimulate the growth and development of plants, and increase the resistance to harmful effects.

Research shows that inoculation of plant seeds with specific PGPR species such as N_2-fixing Bacillus subtilis and phosphorus-solubilizing Bacillus megaterium instead of synthetic chemicals can serve as an effective alternative and environmentally friendly practice as they increase N and P uptake by plants. improves nutrition.

It has been studied by many scientists that the use of humus substances is more effective in reducing the harmful effects of heavy metals on plants due to the properties of forming a complex combination of organic substances [7-10].

Heavy metals enter plants mainly from the soil through the root system, to a lesser extent - from the atmosphere and through the leaves. It is known that the root system is more sensitive to the effects of heavy metals. This is because the roots are the first barrier for the transport of metals from the soil to the plant. Studying the effect of heavy metals on the "plant-microorganism" system is of not only theoretical but also practical interest. In addition, heavy metals that affect stem cells slow down their growth. It disrupts the permeability of membranes, reduces its absorption capacity [11].

Using bioremediation as a suitable method for the treatment of heavy metals from contaminated soils using additives such as organic matter and rhizobacteria [12].

The purpose of the study: to study the effectiveness of using wheat rhizobacteria, potassium humate and biopreparation Serhosil on the level of reduction of heavy metals in the organs and soil of wheat plants grown in soil contaminated with heavy metals.

2 Materials and methods

2.1. Location of sampling and physico-chemical composition of the sample.

Taking into account the above, an experiment was conducted on the basis of bacterial fertilizer, potassium humate and Serhosil complex-acting biopreparation, created from wheat rhizobacteria, which allows to increase the resistance of winter wheat to the harmful effects of heavy metals, grain quality and high yield. The experiment was carried out in 2020-2022 on the Tanya variety of winter wheat at the field experiment area owned by the owner of "Almalik Mining and Metallurgical Combine" JSC (Joint-Stock Company).

We took samples from the soil of the experimental area in accordance with GOST 17.4.4.02-84.

The object is located in the area of the pre-mountain low-high elevation zone, directly under the influence of "Nurabad" HPP, Almalyk mine and metallurgical plant and "Ohangarontsement" JSC, within a radius of 11-15 km. The physical and chemical properties of the soil were studied (Table 1).

Table 1. Physical and chemical properties of soil

№	Sampling location	pH	Humus %	Humus carbohydrate, % (C, %)	N		P		K	
					General %	Enhanced, mg/kg	General %	Enhanced, mg/kg	General %	Enhanced, mg/kg
1	Soil	8,4	1,41	0,91	0,135	12,4	0,214	17	1,13	261

Experiments were conducted on winter wheat crops according to the following schemes.

1. Control, NPK
2. Experience - NPK + potassium humate (tillage)
3. Experiment - NPK + bacterial fertilizer (wheat rhizobacteria seed treatment)
4. Experiment- NPK + potassium humate (soil treatment) + bacterial fertilizer (seed treatment) + YIELD (treatment of the plant in the budding and tuberting phases of the vegetation period)

2.2. Adding humic substances and soil.

Potassium humate was used to transform the heavy elements in the soil into a form that cannot be assimilated by plants due to sorption and formation of complex compounds. Potassium humate was obtained by oxidizing Angren charcoal with hydrogen peroxide, followed by extraction of the oxidized product with 1% KON solution. Its concentration was diluted to 0.1% (11).

Table 2. Potassium humate composition, %

Moisture	Ash	Total organic matter	Including	
			Humic acids	Fulvic acids
97,17	0,11	2,72	1,42	0,24

2.3 Treatment of wheat seeds using bacterial fertilizers

100 ml of bacterial fertilizer is mixed with 8 liters of clean water, mixed well, 250 kg of wheat seeds per hectare are treated with a spray gun, dried in the air for 1 hour and planted in the soil.

2.4 SERHOSIL treatment of wheat plant in the budding and tuberting phases of the vegetation period using biopreparation.

10 l of concentrate is added to 190 l of water and 1 hectare of plants is treated by foliar spraying.

2.5. Absorption of heavy metals

Wheat grains were dried and collected at harvest (12) and heavy metals were determined using an Inductively Coupled Plasma Atomic Emission Method and Inductively Coupled Plasma Mass Spectrometer (ICP–MS) 7500 Series in Rocks, Earth, Soil and underground sediments were determined by the methods of element composition determination.

2.6. Statistical processing.

Data obtained in MS Excel and Statistica-10.0. was processed using generally accepted mathematical statistical methods. The reliability of the differences of the mean values was evaluated by the Fisher-Student method.

3 Results and discussion

2020-2022 at the farm belonging to the Olmalik mining metallurgical combine located in the Okhangaron district of the Tashkent region. "Tanya" variety of winter wheat plant was planted with "potassium humate+bacterial fertilizer+Serhosil" complex action biofertilizers, seed germination increased and 95% of planted seeds germinated.

When treated with complex biofertilizers, wheat plant's resistance to the harmful effects of heavy metals in the development phases is increased compared to control, plant metabolism is accelerated, and favorable conditions for plant growth and development are created.

Humic substances in the soil can form complexes with metal ions and change the assimilation of nutrients by plants. They can form complex compounds in the form of metal-humate and metal-humate-mineral, in addition, due to the formation of chemical bonds with heavy metals based on the pH environment of the soil solution, they reduce the absorption of heavy metals by plants [13]. This means that humic substances in heavy metal-contaminated soils, while potentially beneficial for plant development, simultaneously reduce the ecological mobility of these pollutants. It is confirmed that the use of rhizobacteria produced by bacterial communities in the soil environment affected by the harmful effects of heavy metals is effective [14]. Inoculation of wheat seeds with RGPRs has a significant effect on reducing Pb stress [15].

According to the results of the experiment, when treated with a complex of biofertilizers. When studying the effect of positive changes in the amount of heavy metals in wheat roots, stalks and grains, the following data were obtained (Tables 4-6).

Table 3. Changes in the amount of heavy metals in the roots of winter wheat under the influence of biofertilizer complex (average, n=3)

№	Experience options	Cr	Co	Cu	Zn	As	Rb	Cd	Pb	U	V
1	Control NPK	100	14,0	140	510	31,0	97,0	0,57	61,0	2,7	93,0
2	NPK+ potassium humate	120	13,0	120	580	34,0	100	2,6	84,0	3.0	97,0
3	NPK+ bacterial fertilizer	64,0	12,0	91,0	150	31,0	88,0	0,34	60,0	2,9	87,0
4	NPK+ potassium humate + bacterial fertilizer +SERHOSIL	61,0	12,0	64,0	110	28,0	82,0	0,32	44,0	2,6	85,0

According to the obtained data, it was found that the soils of the area not treated with biofertilizers (control) were contaminated with elements of heavy metals Zn, Pb, Co, Ni, As, Cr. It was found to be 2-3 times higher than the permissible norm (REM).

It was found that the amount of toxic heavy metals in the area where the biofertilizer complex was applied was significantly reduced compared to the control: the amount of Zn was 400 mg/kg, Cd -0.25 mg/kg, Pb-17 mg/kg, Cu-76 mg/kg, Co-2 mg/kg, As- It was found that 3mg/kg, Cr -39mg/kg, V amount - 8 mg/ha decreased.

In the area where the biofertilizer complex was applied, it was found that the amount of heavy metals in the stem of the winter wheat plant was significantly reduced compared to the control: the amount of Zn was 300 mg/kg, Cd -1.4 mg/kg, Pb-3 mg/kg, Cu-10,mg/kg, Co-1, It was found that 5 mg/kg, As-2mg/kg, Cr -9mg/kg, V amount - 4 mg/ha decreased (Table 4).

Table 4. Changes in the amount of heavy metals in the stems of winter wheat under the influence of biofertilizer complex (average, n=3)

№	Experience options	Cr	Co	Cu	Zn	As	Rb	Cd	Pb	U	V
1	Control NPK	43,0	2,7	110	600	23,0	42,0	2,5	55,0	0,59	27,0
2	NPK+potassium humate	67,0	3,2	170	320	26,0	70,0	3,6	93,0	0,89	29,0
3	NPK+bacterial fertilizer	75,0	5,0	150	300	24,0	60,0	8,1	99,0	1,2	43,0
4	NPK + potassium humate + bacterial fertilizer + SERHOSIL	34,0	1,9	100	300	21,0	55,0	1,1	52,0	0,59	23,0

The results of the experiment showed that the amount of heavy metal Zn in the grain of the wheat plant treated with complex biofertilizers was 100 mg/kg, Cd -0.07 mg/kg, Pb-9 mg/kg, Cu-9mg/kg, Co-0.68 mg/kg, It was found that As-3mg/kg, Cr -10 mg/kg, V amount - 5 mg/ha decreased (table 5).

Table 5. Changes in the amount of heavy metals in grains of winter wheat under the influence of biofertilizer complex (average, n=3), mg/kg

№	Experience options	Cr	Co	Cu	Zn	As	Rb	Cd	Pb	U	V
1	Control NPK	41,0	1,2	100	370	18,0	52,0	0,17	27,0	0,29	20,0
2	NPK+potassium humate	37,0	1,3	87,0	220	14,0	38,0	0,32	19,0	0,38	21,0

3	NPK+bacterial fertilizer	33,0	0,58	100	370	15,0	60,0	0,27	23,0	0,31	17,0
4	NPK + potassium humate + bacterial fertilizer + SERHOSIL	31,0	0,52	91,0	270	15,0	30,0	0,10	18,0	0,21	15,0

Table 6. Changes in quality indicators of winter wheat plants under the influence of biofertilizer complex

№	Experience options	grain type, Gr	Gluten amount, %	IDK. ed.%	1000 pcs grain weight, Gr,	transparency, %
1	Control NPK	758	21,0	74	40	50
2	NPK+potassium humate	774	21,1	76	40,2	50,2
3	NPK+bacterial fertilizer	785	21,2	78	41	51,5
4	NPK + potassium humate + bacterial fertilizer + SERHOSIL	820	21,6	81	42,0	52,1

In the area where biofertilizers were applied, wheat yield was 55 ts/ha. It was found that control in the field was 48 t/ha and 7 t/ha additional yield was obtained.

4 Conclusion

It can be concluded from the obtained results that positive results were obtained with the complex application of biofertilizers in the cultivation of wheat plants in the conditions polluted by heavy metals.

1. It was found that 95% of wheat plants germinated seeds and reduced heavy metal elements in wheat roots, stems and grain compared to control.
2. It was determined that the quality indicators of wheat grains increased by 8.5%, transparency by 4.2%, gluten content by 2.8%, IDK by 9.4% compared to the control.
3. Productivity has increased by 7 ts/h, along with the opportunity to grow environmentally friendly products.

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