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The search for effective solutions to recover soil and vegetation cover in contaminated areas of the town of Karabash

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Abstract
The operations of non-ferrous metallurgy industries lead to the formation of persistent polymetallic contamination in the soil cover of the surrounding areas, which leads to the degradation of the soil and vegetation cover following prolonged negative exposure. Given the scale of the problem, the authors conducted a series of experimental studies to model plant growth on highly contaminated acidic soils to find a comprehensive solution for recovering the soil and vegetation cover. The experiment involved identifying the most effective soil additives for restoring the acidic soils of the town of Karabash in the Chelyabinsk Region, which are characterized by high levels of heavy metal contamination. Based on the results of theoretical studies, lime, low-moor peat, vermiculite, biochar, and melatonin were selected as soil-improving additives for the experiment. The results of laboratory studies on the soil composition and properties before and after the experiment are presented, as well as the results of visual, quantitative, and chemical assessments of plant biomass. Based on the experimental studies, the most effective soil amendments were identified, which not only reduce the impact of heavy metals on plants but also increase their biomass. The best results were obtained by neutralizing the soil with lime, improving its physical and mechanical properties through the addition of vermiculite, increasing the organic matter content using low-moor peat, and adding broad-spectrum soil-improving additives such as biochar and melatonin. The combination of soil additives made it possible to neutralize highly acidic soils to a pH of 6.6-7.5, reduce the concentration of mobile copper – the primary contaminant – by a factor of four or more, and grow sustainable root and above-ground plant parts.

Keywords
remediation; soil and vegetation cover; acidic soils; heavy metal; soil additive; lime application; biochar; melatonin; low-moor peat; vermiculite

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Introduction

Soil contamination by heavy metals (HM) poses one of the most serious global threats to the sustainability of the natural ecosystem and human wellbeing. In contrast to organic pollutants, which are susceptible to microbiological and (or) abiotic degradation, the majority of heavy metals and metalloids exhibit a high degree of persistence. These substances are seldom biodegradable or chemically decomposable within the soil environment, and they have the capacity to persist within the soil monolith, accumulating over extended periods (extending to decades or even hundreds of years). The half-life resulting from leaching, uptake by plants, erosion and deflation varies significantly between elements. For cadmium (Cd), the half-life ranges from 13 to 110 years; for zinc (Zn), from 70 to 510 years; for copper (Cu), from 310 to 1500 years; and for lead (Pb), from 740 to 5900 years [1]. The increase in heavy

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metal concentrations in soil can be attributed primarily to anthropogenic activities. The most significant impact is observed in nearby areas, particularly in proximity to major cities and industrial hubs. High concentrations of heavy metals in the soil are most frequently observed in the areas mentioned above [2, 3]. Furthermore, consideration must be given to the impact of the mining industry, which has a detrimental effect on large areas, leading to the formation of technology-influenced landscapes [4].

In areas exhibiting high environmental impact, particularly in regions containing non-ferrous metal processing facilities, polymetallic anomalies have been detected in the soil. The activity of non-ferrous metallurgical facilities has been demonstrated to result in the accumulation of Cu, Ni, Pb and Cd in the soil [5, 6]. Metal concentrations in soils in the vicinity of urban-forming enterprises exceed regulated values by a factor of 5-10 or more (Monchegorsk, Kirovgrad, Rezh, Asbest, Revda) [7, 8].

Research conducted in Russia, Canada, the USA, France, China and other countries has demonstrated that emissions into the atmosphere constitute the primary cause of heavy metal contamination in soil [9]. In areas directly impacted by metallurgical plants, the soil undergoes profound anthropogenic transformation: phytocenoses are destroyed and extensive anthropogenic wastelands are formed, with a complete absence of vegetation cover. A substantial accumulation of heavy metals, in both mobile and stable forms, has been documented in the upper layers of the soil profile. The bioavailability of metals in mobile forms is high, and they are actively absorbed by plant roots [10], resulting in heavy metal concentrations in plant biomass being many times higher than in the soil. Consequently, the accumulated environmental damage may have a long-term negative impact on the soil and vegetation cover, leading to its deterioration and eventual destruction [2]. Moreover, it has been established that areas of accumulated environmental damage can act as sources of contamination for all components of the surrounding area [11].

In the Russian Federation, there is a high density of mining and mineral processing facilities in the Middle and Southern Ural Mountains. These facilities are responsible for comprehensive pollution of the native environment [12]. The country's largest mining and metallurgical base is located in this region. In the Chelyabinsk Region, certain areas have become characterized as environmental disaster zones. The concentration of heavy metals in the soil has been recorded as being more than 25 times the maximum permissible concentration (MPC) [13].

One particularly notable example is the man-made wasteland that has formed as a result of the prolonged negative impact of the Karabashmed JSC. The company is located in the town of Karabash in the north-west of the Chelyabinsk Region. The enterprise is a town-forming specialty producer of copper, with a focus on blister and anode varieties. For a period of more than 90 years, the plant had functioned without a gas-cleaning system that was suited to the task, which resulted in the formation of accumulated environmental damage. Despite the modernization of the plant in 2004, including the installation of high-efficiency gas cleaning systems, the prolonged accumulation of pollutants has now resulted in irreversible degradation of the soil and vegetation cover.

The object of this study is the acidic, highly contaminated soil of Lysaya Mountain, which is located within the Karabash mining and industrial area. Despite a decline in atmospheric pollution levels, the soil continues to exhibit elevated concentrations of heavy metals, thereby impeding plant growth [14]. Pollutant levels have been observed to reach concentrations 400 times higher than the MPC for copper, 300 times higher for lead, and 10 times higher for nickel [15, 16]. The contamination of soil with heavy metals has resulted in the degradation of vegetation cover, which in turn has prevented erosion processes and the migration of contaminants. The topography of the area is currently characterized by the presence of deep erosion gullies. Furthermore, aerial deflation is causing changes and damage to physical properties and soil structure. It is evident that the processes under scrutiny are presently resulting in the displacement of heavy metals from the mountain slopes. These metals are being transported by wind streams in the atmosphere and accumulated in the hydrosphere through rainwater and meltwater. The long-term detrimental effects have rendered the mountain incapable of self-regeneration, even with a minimized anthropogenic impact. In order to resolve the current problem, it is necessary to implement additional effective measures to improve soil composition and properties. It is imperative to implement this in order to create a stable anti-erosion soil and vegetation cover.

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Conventional remediation methods have been demonstrated to be ineffective in the case of acidic soils that are heavily contaminated with heavy metals. The underlying reason for this phenomenon is that an acidic environment, characterized by a pH below 5.5, has been shown to enhance the solubility and mobility of metals such as Cu, Ni, Pb, and Cd. In soils with elevated levels of contamination (where concentrations exceed the maximum permissible limit by a factor of 10 or more), standard doses of soil improvers prove ineffective, and the concentration of mobile metal forms persists at high levels even after treatment. This has been shown to result in protracted remediation times and the potential for re-release of pollutants [17].

The issue of soil contamination with heavy metals is currently being addressed through the implementation of remediation measures. These remediation methods can be categorized into three primary types: physical, chemical, and biological [18, 19]. The topic is highly relevant, and a large number of studies are currently being conducted into new methods for soil remediation, especially soil contaminated with heavy metals [20]. The focus of current remediation research is on concentrations that exceed environmental quality standards by a factor of 5-10. However, in the context of long-term negative impacts on soil resulting from metallurgical plants, concentrations are capable of reaching levels that far exceed these standards, by a factor of 100 to 500. For the object of this study, concentrations of this level are valid. In addressing this challenge, the objective of this research is to create a comprehensive solution that will contribute to the remediation of a sustainable soil and vegetation cover in the contaminated area of the town of Karabash. The primary function of this layer is to prevent the development of erosion processes and the spread of pollutants. Moreover, the restoration of vegetative cover across a substantial area within the urban landscape will undoubtedly enhance the city's aesthetic appeal. In order to achieve this objective, the authors set out to achieve the following tasks: an assessment of the current state of the degraded area; a study of the theoretical aspects of soil additives which are currently in use and have future potential for recovering soils with a composition and properties relevant to those analyzed; a series of model experiments on the establishment of sustainable vegetation cover on heavily contaminated acidic soils using both individual and combined soil additives; a dynamic assessment of changes in the quality of the analyzed soils, the accumulation of pollutants by plants, and the condition of the growing plants.

Physical remediation has one major drawback – the high cost of implementation. Considering the size of the contaminated area (over 1000 ha), this method is extremely costly and undesirable. It should only be used if it is not possible to recover the vegetation cover by improving soil quality.

Phytoremediation has not been studied enough in field conditions to predict its effectiveness in removing copper from such large areas. Furthermore, many phytoremediation methods involve removing and disposing of vegetation after a short period of time, which requires significant capital investment.

Chemical remediation methods are the most effective. At present, there is a lot of research going on into the use of not only individual reagents but also complex additives and additives made from industrial waste [21, 22]. The use of soil-improving fillers, when used in a complex way, can not only lower the pH of acidic soils but also facilitate the transformation of mobile forms of heavy metals into less mobile forms, as well as the sorption of pollutants.

Given the limitations of the remediation methods reviewed, the most promising solutions to this problem are those involving integrated chemical remediation in combination with soil additives.

Materials and methods

Soil characteristics. In the course of the 2024 field season, 17 samples were collected in the Lysaya Mountain area (Karabash, Chelyabinsk Region) for the purpose of experimental studies. The samples were collected and prepared in accordance with GOST 17.4.4.02-2017[1]. Laboratory analyses revealed that the soils under investigation are characterized as very strongly acidic

[1] GOST 17.4.4.02-2017. Nature Protection. Soils. Methods for Sampling and Preparation of Soil Samples for Chemical, Bacteriological and Helminthological Analyses (put into effect as a national standard of the Russian Federation from 1 January 2019).

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with high levels of heavy metals. The results of the chemical analysis demonstrated that the concentration of HM in the mobile form exhibited variation within the following ranges, mg/kg: Cu 0.5-1400 (0.2-466 MPC), Ni 1.5-49 (0.37-12 MPC), Pb 0.1-130 (0.017-21 MPC)[2]. The samples exhibit significant variation in terms of organic matter content, with concentrations ranging from 1.6 to 17.2 %[3]. The pH of aqueous extracts[4] ranges from 4.2 to 7.0, whilst that of saline extracts[5] ranges from 3.5 to 4.9. With regard to acidity, the majority of samples are classified as being very strongly acidic ($pH_{KCl} < 4.0$). The values for hydrolytic acidity[6] exhibited a considerable variance, with a minimal measurement of less than 0.23 mgEq/100 g of soil, and a maximal measurement greater than 145 mgEq/100 g of soil.

It is evident from the data obtained from laboratory analysis of the samples that the soils are varied. The variation in chemical composition, physical, and mechanical properties is a distinguishing feature. In this regard, all samples were divided into two groups for the purposes of experimental research. This will optimize the selection of the most effective soil additives (Table 1). In accordance with the findings of previous research by the authors, the main indicators used to categorize the soil samples were pH_{KCl} and the concentration of copper as the primary contaminant in its mobile phase [15]. Following an analysis of the chemical composition and characteristics of the soils in each group, four samples from group 1 and four from group 2 were selected for the model experiment. The samples were selected from across the full range of parameters in order to assess the effect of the additives on the group as a whole.

Table 1

The characteristics of the experimental soil groups

Sample group	Number of samples in the group	pH_{KCl}	Mobile phase content of copper, mg/kg	Hydrolytic acidity, mgEq/100 g of soil
1	4	3.9-4.9	0.5-300	0.23-58
2	4	3.5-3.7	410-1400	103-132

Soil additives. A comprehensive analysis of research on the subject was carried out by the authors to select soil additives capable of improving the quality of the soils under consideration for the restoration of the soil-vegetation cover in the area in focus. Current scientific research primarily studies soil additives in the areas of chemical stabilization and phytoremediation using plant growth stimulants. Chemical stabilizers mentioned in the studies include kaolinite, attapulgite, sepiolite, montmorillonite, vermiculite, gypsum, biochar, zeolite, rice straw phosphate, hydroxyapatite, wastewater sludge, iron oxides and sulphides, wood ash, livestock waste, and peat. Biostimulants used for phytoremediation include humic substances, thiosulfates, seaweed extracts, various beneficial microorganisms (*Trichoderma pseudokoningii, Trichoderma harzianum, Burkholderia cepacia, Bacillus subtilis, Paenibacillus jamilae,* etc.), plant extracts (*Oxalis corniculate, Medicago sativa, Moringa oleifera* and *Glycyrrhiza glabra* roots, etc.), as well as melatonin. It is worth noting that the main limitation of current scientific research is its focus on soils contaminated with heavy metals concentrations of 2-50 MPC [23, 24]. However, under real-world conditions in man-made wasteland areas, including the area under study, exceedances occur significantly more frequently. Furthermore, studies into the effectiveness of soil amendments often use artificially contaminated soils with a pH > 6.0.

[2] M-MVI-80-2008. Procedure for Measuring Mass Fractions of Elements in Samples of Soil, Earth, and Bottom Sediments by Atomic Emission Spectrometry and Atomic Absorption Spectrometry. St. Petersburg: Monitoring, 2008, 27 p.

[3] GOST 23740-2016. Soils. Methods of Laboratory Determination of Organic Composition (put into effect by Order N 2096-st of the Federal Agency for Technical Regulation and Metrology, 27 December 2016).

[4] GOST 26423-85. Soils. Methods for Determination of Specific Electrical Conductivity, pH and Solid Residue of Water Extract (approved and put into effect by Resolution N 283 of the USSR State Committee for Standards, 8 February 1985).

[5] GOST 26483-85. Soils. Preparation of Salt Extract and Determination of Its pH by the CINAO Method (approved and put into effect by Resolution N 820 of the USSR State Committee for Standards, 26 March 1985).

[6] GOST 26212-91. Soils. Determination of Hydrolytic Acidity by the Kappen Method Modified by CINAO (approved and put into effect by Resolution N 2389 of the USSR Committee for Standardization and Metrology, 29 December 1991).

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The most promising soil improvers were selected from the range available for experimental studies on real samples of highly acidic, contaminated soils (Table 2).

Plants. A seed mixture containing perennial ryegrass (*Lolium perenne*) – 45 %, red fescue (*Festuca rubra*) – 30 %, meadow grass (*Poa pratensis*) – 20 % and sheep's fescue (*Festuca ovina*) – 5 % was used to conduct the growth experiment on the prepared soil samples with the previously mentioned soil additives [25]. These plants were selected for the experiment because they are used in hydroseeding mixtures for the reclamation of disturbed land and also grow in the forest-steppe zone of the Chelyabinsk Region [26]. One gram of the seed mixture was added to each soil sample in the study.

Table 2

Soil additives for the laboratory experiment

Soil additive	The impact (advantages and disadvantages)	Prospective suppliers
Lime (particle size 0-0.09 mm)	Soil neutralization. Reduction of heavy metal uptake by plants. Stimulation of activity of beneficial microorganisms (nitrogen-fixing and nitrifying bacteria). Improved development of nodule-forming bacteria. Enhanced nitrogen uptake by plants. Transformation of difficult-to-dissolve soil phosphorus compounds into assimilable forms. Adsorption of heavy metals from the soil suspension [27]. Over-application may lead to soil alkalisation and disruption of the plant nutrient balance. In the absence of sufficient organic matter, it may accelerate humus decomposition, leading to a partial loss of organic matter	The Varnenskoye deposit, Varnenskiy Crushing and Screening Complex Ltd. (Varnenskiy District, Chelyabinsk Region). Distance from the object of study – 300 km
Vermiculite (particle size 3-5 mm)	Improvement of the physical and mechanical properties of soils. Protection from the growth of fungal diseases due to its resistance to root rot [28]. Protection from overheating during hot periods due to its heat-insulating properties. Sorption of heavy metals [29]. Over moistening of soils can lead to reduced aeration and cause root rot	The Potanin deposit, Uralvermiculite Ltd. (Kyshtym, Chelyabinsk Region). Distance from the object of study – 50 km
Low-moor peat (particle size 0-15 mm, pH > 5.5)	A positive effect on the nutritional and microbiological composition of soils [30]. A slightly acidic or neutral pH (pH 5.5-7.0). Improvement in the physical and mechanical properties of soils: water permeability, air-holding capacity, density and porosity (sand binding and clay loosening). Remove various forms of heavy metals and other toxic substances accumulated in plants [31]. It is not a reliable method for neutralizing heavy metals in mobile phases. An excess leads to a disruption of the soil's aeration	The Malo-Reftinskoye mining field, Fabrika Gruntov Ltd. (Berezovsky Urban District, Sverdlovsk Region). Distance from the object of study – 230 km
Biochar (particle size 1-5 mm, made from birch wood waste)	Improvement of porosity, water permeability, aeration, and soil structure [32]. An increase in carbon stocks and CO_2 sequestration. A reduction in the availability of heavy metals. An increase in pH in acidic soils. Retention of nutrients (N, P, K, Ca, Mg, etc.) due to high cation exchange capacity and a porous surface [33]. Reduction of fertiliser leaching; increased efficiency of nitrogen and phosphorus utilisation by plants. Stimulation of microorganisms and soil biota [33]. Risk of secondary soil contamination depending on the raw materials used for biochar production	Maderum Ltd. (Plastovsky District, Chelyabinsk Region). Distance from the object of study – 170 km

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End of Table 2

Soil additives	The impact (advantages and disadvantages)	Prospective suppliers
Melatonin (suspension)	Increased plant tolerance to heavy metal contamination in soils [34]. Reduction in levels of reactive oxygen species, malondialdehyde and other markers of oxidative damage to plants caused by heavy metals. Activation of plant antioxidant systems. Increased plant biomass. Increased chlorophyll content and improved photosynthetic activity under stress [35]. Increased uptake of N, P, K, Ca and Mg in plant tissues (by up to 34-82 % depending on the element), recovery of nutrient balance. Increased populations of bacteria, fungi and actinomycetes, leading to improved biological activity and structure of the soil community. Successful combination with other soil improvers, enhancing the effectiveness of reducing the availability of heavy metals and oxidative stress [36]. High cost due to the lack of industrial-scale production as a growth stimulant. Does not have a neutralizing or sorption effect on soils.	The industrial production of the Russian Federation is not related to the mentioned activity

Design of the experiment. The studies were conducted at the Research Centre "Assessment of Anthropogenic Transformation of Ecosystems" at Empress Catherine II Saint Petersburg Mining University. The grass mixture was cultivated for 30 days in an NLO 79-03-04 LD plant growth chamber, with the results recorded periodically. The light source was a dimmable, full white spectrum LED lamp. The light intensity at a height of 0.5 m was 10,000 lux, and the photoperiod was 16/8 h (day/night). The mixture was periodically watered using low-mineralized water. A total of six sets of experiments were conducted (Table 3). For visual monitoring purposes, a control experiment on uncontaminated soil was included in each series. Each experiment was carried out on samples from both groups in two replications. The experiments were conducted in stages. In the first stage, an analysis was carried out of available and widely used additives. However, these did not achieve the target outcome of sustainable vegetation cover, although they did have a positive impact. As a result, the subsequent stage involved the application of sorbents and plant growth stimulants, which were identified as the most effective treatment.

Table 3

The structure of the model experiment

Set of experiments	Experiment number	Soil additives	
		Lime*	Other
1 (background)	1-1	No lime	No additives
2	2-1	1D	No additives
	2-2	2D	No additives
	2-3	4D	No additives
3	3-1	No lime	Peat
	3-2	No lime	Vermiculite
	3-3	No lime	Peat, vermiculite
4	4-1	1D	Peat, vermiculite
	4-2	2D	Peat, vermiculite
	4-3	4D	Peat, vermiculite
5	5-1	1D	Biochar, peat, vermiculite
	5-2	2D	Biochar, peat, vermiculite
	5-3	4D	Biochar, peat, vermiculite
6	6-1	1D	Melatonin, peat, vermiculite
	6-2	2D	Melatonin, peat, vermiculite
	6-3	4D	Melatonin, peat, vermiculite

* Dose: 1D – calculated, 2D – double, 4D – fourfold.

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Lime was applied in the calculated (single), double and fourfold doses. The increase in lime dosage compared to the standard dosage is due to the fact that lime is used for more than just neutralizing acidity during liming; it is also used to absorb heavy metals, compensate for base losses and enrich the soil with calcium [28, 37]. The lime dosage is calculated based on information about the maximum hydrolytic acidity of the group and the size of the sample,

$$P_{CaCO_3} = H \cdot 0.05hd,$$

where H – hydrolytic acidity, mgEq/100 g of soil; h – depth of the soil treatment layer, cm; d – density of the soil treatment layer, g/cm^3.

The dose for group 1 was 7.1 g and the dose for group 2 was 12.2 g.

The studies utilize a variety of soil-to-peat and soil-to-vermiculite ratio combinations, which vary widely. For example, soil is mixed with peat at ratios ranging from 50-95 wt.% soil to 5-50 wt.% of the peat, while the amount of vermiculite added varies in range of 5-20 wt.% [38-40]. Consequently, the following mass ratios were selected for the experiment based on theoretical research, experimental data and economic feasibility: soil and peat – 8:2, soil and vermiculite – 8:1, and soil, peat and vermiculite – 8:2:1.

Melatonin was applied as a 200 μmol/l suspension on the 1st and 14th days of the experiment [41]. A total dose of 10 μmol of melatonin per 1 kg of soil was applied. This amount is considered sufficient to stimulate growth and increase plant tolerance to the impact of HM [42, 43]. The proportion of biochar in the complex additive was 10 wt.%.

The analysis of the experimental results, which included an assessment of the effectiveness of soil amendments, was carried out in three areas: a visual and quantitative assessment of plant biomass; an assessment of changes in soil quality over time; and an assessment of the effectiveness of heavy metal accumulation by plants.

Visual assessment helps to identify problems associated with the impact of HM on photosynthesis (chlorosis and plant necrosis), as well as plant productivity [44]. Shoot biomass reflects the impact of soil additives on plant growth and health during the entire growing period [45]. Root biomass indicates a plant's ability to inhabit contaminated soil and uptake nutrients, as well as demonstrating its stress tolerance and the effectiveness of soil additives. Under the severe impact of heavy metals, the root system can be highly inhibited [46]. In this study, a visual assessment was carried out throughout the experiment. This included an assessment of the external appearance of the shoots, their colour and density. After 30 days, the degree of root formation was also assessed. The shoots were then separated from the soil and dried to an air-dry consistency. The roots were also dried to an air-dry state, separated from soil residues, washed, and dried again. The dried shoot and root samples were weighed using PA-214C electronic scales (OHAUS Instruments, China). The samples were then milled using an AM200S laboratory mortar grinder (ANTS Scientific Instruments, China).

Changes in soil quality were assessed based on two key parameters: pH_{KCl} and the concentration of mobile heavy metals. Hydrogen parameter of KCl was determined in accordance with GOST 26483-85[7] using a MARK-903 pH meter (VZOR Ltd., Russia). The quantitative analysis of the mobile forms of heavy metals in the soil was carried out using inductively coupled plasma atomic emission spectrometry (ICP) in accordance with the M-MVI-80-2008[8] methodology. This analysis was performed using an ICPE-9000 atomic emission spectrometer (Shimadzu, Japan).

[7] GOST 26483-85. Soils. Preparation of Salt Extract and Determination of Its pH by the CINAO Method (approved and put into effect by Resolution N 820 of the USSR State Committee for Standards, 26 March 1985).

[8] M-MVI-80-2008. Procedure for Measuring Mass Fractions of Elements in Samples of Soil, Earth, and Bottom Sediments by Atomic Emission Spectrometry and Atomic Absorption Spectrometry. St. Petersburg: Monitoring, 2008, 27 p.

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The bioavailability of heavy metals was assessed by determining the efficiency of their accumulation by plants. This indicator helps to establish the principle by which the complex additive works, either by stabilising heavy metals and preventing their migration into plant shoots, or by enhancing the plants' tolerance to the adverse effects of HM [35]. Sample preparation was carried out as follows [47]. From the soil samples (separated shoots and roots), 0.1 g samples were taken for wet ashing. The ashing process involved the use of concentrated nitric acid (2 ml) and hydrogen peroxide (2 ml) in a Multiwave 3000 microwave digestion system (Anton Paar, Austria). After cooling, the mixtures were transferred quantitatively to 20 ml polyethylene test tubes and topped up to the mark with distilled water. Each sample was prepared in two parallel versions, as well as a blank solution. The resulting solutions were analysed for total elemental content using an ICPE-9000 inductively coupled plasma spectrometer.

Results discussion

A visual and quantitative assessment of plant biomass. A visual assessment of the vegetation enabled early conclusions to be drawn regarding the effect of soil additives on plant growth and development. A visual assessment of the plants' shoots indicated the following conclusions. In experiment set 1, plants cultivated in non-additive soils exhibited characteristics such as stunted growth and weak, dull shoots. By the 16th day of the experiment, notable indications of wilting and difficulties in water uptake became visible. By the final day of the experiment, necrosis of the shoot tips and loss of turgor had been recorded. In this experiment, the negative effect of HM on the root systems of the plants was also noticed. The root system in the samples from the group 2 was practically undeveloped (Fig.1).

The liming treatment (experiment set 2) produced positive results, with root system development observed. However, signs of necrosis were also evident and a loss of turgor was noted in some samples. Furthermore, when a single dose of lime is applied, the extent of necrotic changes and shoot desiccation is reduced.

Using peat (experiment 3-1) increases the biomass of plant shoots during the beginning of the growth stage and promotes the development of the root system. However, using peat alone led to the partial rotting of the root system due to the soil sample's unsuitable physical and mechanical properties. A partially positive result was also observed when vermiculite was added to the soil (experiment 3-2). The plants exhibited comparatively high shoot turgor; however, this additive did not promote root system development, particularly in group 2, where the level stayed practically the same as in soil samples without additives. In the mixed sample (experiment 3-3), the additives helped prevent decay

Group number	Experiment 1-1	Experiment 2-1	Experiment 3-1	Experiment 3-2	Experiment 3-3	Experiment 4-1	Experiment 5-1	Experiment 6-1
1								
2								

Fig.1. Plants' appearance with different soil additives at the end of the experimental period

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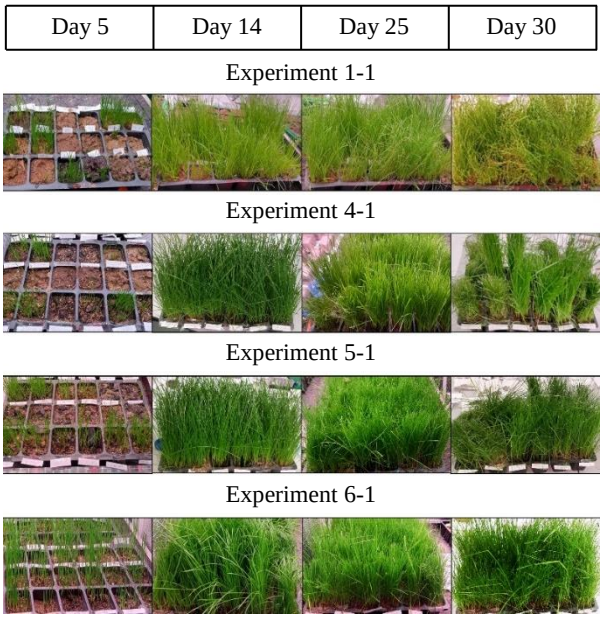


Fig.2. Growth stages of shoots with different soil additives during the entire growing period of the experiment

by improving the soil's physical and mechanical properties. However, they had practically no effect on the weak root system. The increase in shoot turgor may indicate that the original samples had a low nutrient content, causing stunted plant growth. The root system's slow response is linked to high HM concentrations, which inhibit root development even in the nutrient-rich soil. These results are insufficient for the restoration of vegetation in the area under consideration. As the study aims to create a sustainable erosion-resistant soil-vegetation cover, suppressing root development would prevent this.

The experiment set 4 was designed to facilitate an assessment of the combined effect of peat, vermiculite, and lime. Although issues with the development of root systems had been identified in a number of previous experiments, this experiment demonstrated successful root system development in all samples. The utilisation of a single dose of lime within the complex additive was found to have a positive effect, as indicated by visual assessment. In each group, an enhancement in the indicators was evident when compared with the utilisation of individual additives. Conversely, locally, when using double and fourfold doses, low turgor and necrotic changes in the plants were observed. As the lime dose was increased, there was an observable increase in the number of necrotic areas.

The application of a complex biochar-based supplement (experiment set 5) demonstrated a high level of efficacy, as evidenced by the substantial plant biomass, vivid plant colouration and high turgor observed. In this experiment, the optimal dose of lime was the single (calculated) dose. A similar outcome was observed in visual assessments when melatonin was used (experiment set 6). In experiment sets 5 and 6, the application of a fourfold dose of lime resulted in the inhibition of plant growth. The application of a double dose of lime did not yield significant improvements in contrast to the calculated dose, thus leading to the hypothesis that the application of lime in higher amounts is unnecessary.

The results of the visual assessment indicated that the most effective treatments were complex soil additives composed of peat, vermiculite and lime, with the addition of biochar or melatonin. For illustrative purposes, Fig.2 presents a visual representation of the developmental stages of shoots for experiments 1-1, 4-1, 5-1 and 6-1 over a period of 30 days.

To corroborate the results of the visual assessment and undertake a comparative analysis, a quantitative assessment of the shoots plant biomass was performed by weighing the prepared samples (Table 4). As there was no requirement for further corroboration of the outcomes of the visual assessment for the experiments utilising lime without the addition of peat and vermiculite (2-1, 2-2, 2-3), or peat and vermiculite separately (3-1, 3-2), the shoot mass of these samples was not measured.

The results presented in Table 4 confirm the conclusions from the visual assessment. The highest shoot mass values were recorded in experimental sets 4, 5 and 6. The best results were observed in each of these sets of experiments when a single dose of lime was added. Root mass was also measured in these experiments (Table 5). Experiment 3-3 was selected for comparison in group 1; in samples without soil additives, and in group 2 of experiment 3-3, the root system was insufficiently developed to allow an accurate assessment of its mass.

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Table 4

Mass of the shoots, g

Group number	Sample number	Experiment number										
		1	3-3	4-1	4-2	4-3	5-1	5-2	5-3	6-1	6-2	6-3
1	1.1	0.53	0.54	1.21	1.20	1.07	1.42	1.40	1.04	1.43	1.12	0.98
	1.2	0.43	0.56	1.18	1.12	0.98	1.23	1.15	1.02	1.42	1.38	1.03
	1.3	0.36	0.36	1.12	1.10	0.86	1.21	1.16	1.05	1.18	1.03	0.96
	1.4	0.48	0.51	1.12	1.11	0.83	1.22	1.14	1.07	1.32	1.13	0.85
	Mean value	**0.45**	**0.49**	**1.16**	**1.13**	**0.94**	**1.27**	**1.21**	**1.05**	**1.34**	**1.17**	**0.96**
2	2.1	0.42	0.47	1.08	1.01	0.98	1.11	0.98	0.95	1.31	1.3	0.92
	2.2	0.24	0.49	1.12	0.97	0.81	1.14	0.99	0.97	1.21	1.14	0.83
	2.3	0.46	0.44	1.02	1.06	0.74	1.02	1.02	0.98	1.17	1.08	0.64
	2.4	0.18	0.48	1.17	1.22	0.68	1.22	1.18	0.89	1.18	1.05	0.72
	Mean value	**0.33**	**0.47**	**1.10**	**1.07**	**0.80**	**1.12**	**1.04**	**0.95**	**1.22**	**1.14**	**0.78**

Table 5

Mass of the roots, g

Group number	Sample number	Experiment number									
		3-3	4-1	4-2	4-3	5-1	5-2	5-3	6-1	6-2	6-3
1	1.1	0.38	0.49	0.43	0.41	0.60	0.52	0.50	0.68	0.63	0.57
	1.2	0.34	0.46	0.40	0.34	0.62	0.51	0.43	0.71	0.66	0.60
	1.3	0.36	0.41	0.38	0.34	0.53	0.49	0.42	0.70	0.64	0.56
	1.4	0.27	0.37	0.24	0.20	0.46	0.38	0.35	0.57	0.56	0.49
	Mean value	**0.34**	**0.43**	**0.36**	**0.32**	**0.55**	**0.48**	**0.43**	**0.67**	**0.62**	**0.56**
2	2.1	–	0.36	0.31	0.29	0.45	0.44	0.40	0.82	0.73	0.52
	2.2	–	0.34	0.29	0.24	0.56	0.44	0.34	0.97	0.92	0.92
	2.3	–	0.31	0.29	0.25	0.46	0.34	0.31	0.79	0.71	0.59
	2.4	–	0.32	0.24	0.22	0.46	0.32	0.22	0.69	0.65	0.48
	Mean value	–	**0.33**	**0.28**	**0.25**	**0.48**	**0.39**	**0.32**	**0.82**	**0.75**	**0.63**

The results of a comprehensive evaluation of quantitative indicators for shoots and roots indicate that liming (experiments 4-1 and 4-2) positively influenced the shoot biomass of plants. However, it did not facilitate the development of a stable root system for the samples in group 2, thereby hindering the establishment of erosion-resistant vegetation cover. Moreover, the results of the root weighing experiments demonstrate that an increase in lime doses beyond the calculated amount has a detrimental effect on the root system (experiments 5-2, 5-3, 6-2, 6-3).

An assessment of changes in soil quality over time. Visual and quantitative assessments of vegetation revealed adverse effects (absence of a root system, reduced shoot turgor, signs of necrosis, etc.) in experiments 1-1, 2-1, 2-2, 2-3, 3-1 and 3-2. In consequence, samples from these sets were excluded from further quantitative analysis. At this stage, soil acidity was measured after the experiment (Table 6).

The results presented in Table 6 demonstrate that the application of peat and vermiculite (experiment 3-3) to the soils results in an increased pH level, but does not achieve neutralisation; the soils remain acidic. In turn, liming with a single calculated dose of lime is sufficient for all the soil samples under study. The application of a fourfold or, in certain cases, a double the calculated dose of lime results in the soils becoming slightly alkaline (pH > 7.2).

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Table 6

Soil pH (pH_{KCl}*) before and after growing period of the experiment

Group number	Sample number	Experiment number										
		Original**	3-3	4-1	4-2	4-3	5-1	5-2	5-3	6-1	6-2	6-3
1	1.1	4.86	5.05	7.01	7.20	7.29	7.11	7.28	7.33	7.01	7.29	7.40
	1.2	4.45	5.51	7.13	7.42	7.66	7.49	7.70	7.92	7.24	7.50	7.76
	1.3	4.48	5.50	7.24	7.33	7.43	7.28	7.59	7.65	7.19	7.32	7.48
	1.4	3.85	4.92	7.05	7.23	7.47	7.27	7.23	7.55	6.82	7.28	7.38
2	2.1	3.63	4.12	6.76	6.96	7.38	7.01	7.39	7.43	7.04	7.08	7.59
	2.2	3.50	3.99	6.98	7.05	7.21	6.95	7.34	7.44	6.9	7.11	7.36
	2.3	3.56	4.18	6.33	6.69	6.82	6.67	6.81	7.08	6.71	6.82	7.19
	2.4	3.70	4.04	6.60	6.63	6.98	6.81	6.96	7.36	6.55	6.78	7.26

* According to the method, the measurement uncertainty is ±0.05 pH.
** Original pH of the soil sample.

The assessment of heavy metal concentrations is a necessary component in the study of changes in soil properties following the application of soil additives. The application of peat and vermiculite has been demonstrated to reduce the concentration of mobile forms of copper by a factor of 2.5, nickel by a factor of 2.7 and lead by a factor of 2.1. The transformation of these elements into less mobile forms is associated with the sorption properties of the additives, a slight increase in pH, and the "dilution" of the original soil sample. It is important to note that dilution is a characteristic feature of all soil samples to which soil-improving additives have been applied. The use of lime in the additive composition has been shown reduce the content of copper by a factor of 4.7, nickel by a factor of 3.9 and lead by a factor of 2.3, due to an increase in the pH of the samples. A complex additive containing biochar has been shown reduce the concentration of copper by a factor of 5.5, nickel by a factor of 6.6 and lead by a factor of 4.6. The sorption properties of the added biochar appear to be the primary factor in the additional reduction in HM concentrations in the experiment. When melatonin was used as part of the complex additive, a reduction in the concentration of copper by a factor of 4.4, nickel by a factor of 4.1 and lead by a factor of 2.5 was observed. Melatonin has been demonstrated

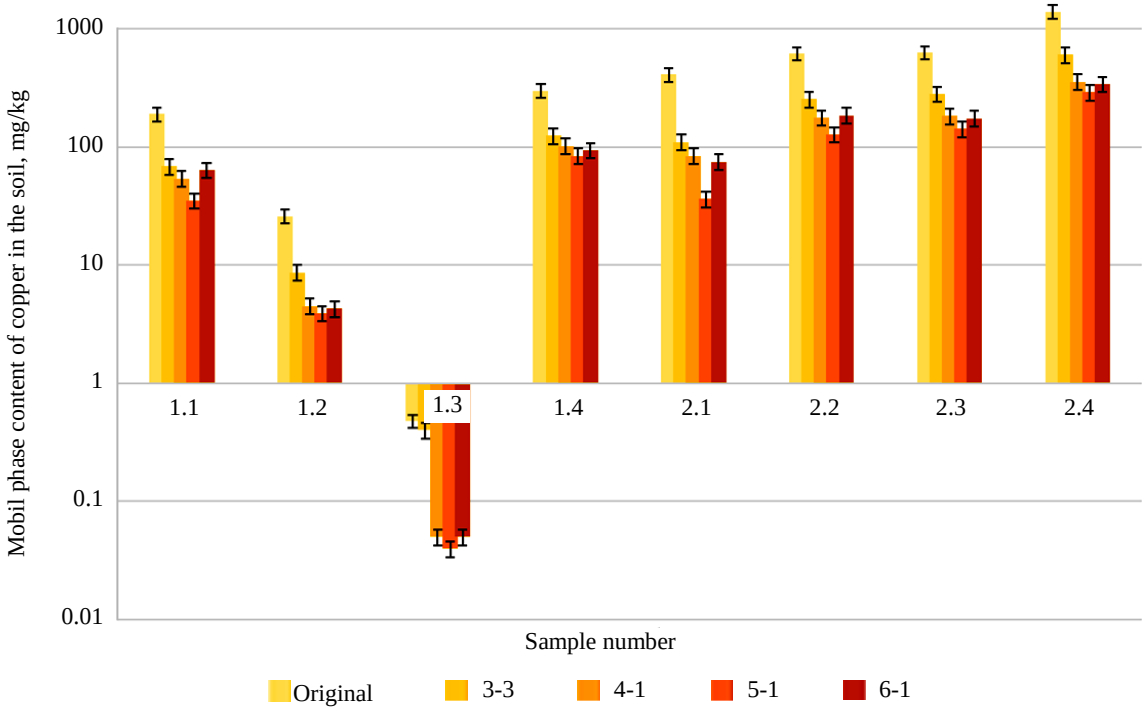


Fig.3. Content of copper in its mobile form in experiments 3-3, 4-1, 5-1 and 6-1 with different soil additives

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to be incapable of transforming HM into less mobile forms; therefore, the change in concentrations is attributed to the use of lime. The concentration changes of copper, the main pollutant, following the application of various complex soil additives are demonstrated in Fig.3. As was the case in previous stages of the experiment, the most successful results were observed in experiments involving complex soil additives containing biochar and melatonin. The most effective results in terms of reducing heavy metal concentrations were observed when using a complex additive containing biochar.

An assessment of the effectiveness of heavy metal accumulation by plants. Samples from experiment 3-3 were selected for a comparative assessment of the most prospective complex additives with regard to HM accumulation. This selection was made because there were sufficient samples to analyze the shoots of plants from both groups and the roots of group 1. In contrast, this was not possible with the samples from experiments 1-1, 2-1, 2-2, 2-3, 3-1 and 3-2. The efficiency of copper accumulation in plants as the primary contaminant in the soil samples is demonstrated in Fig.4.

The mean copper content in plant shoots under normal development is approximately 2-20 mg per 1 kg of mass [48]. The content of the samples analysed ranges from 8 to 44 mg/kg. The lowest copper content is observed in samples with added biochar, whilst the highest is found in samples with melatonin. The mean copper content in roots is generally found to be in the range of 20-200 mg/kg. The copper content of the samples achieved a maximum at 440 mg/kg, which is associated with the protective functions of plant roots. The findings indicate that the utilisation of biochar as a soil additive leads to a substantial reduction in the accumulation of copper by plants. Concurrently, melatonin has been observed to enhance the plants' tolerance to high concentrations of copper, with no apparent effect on the rate of copper uptake. This phenomenon is not exclusive to copper; it has also been observed in nickel and lead, a finding that is supported by theoretical studies.

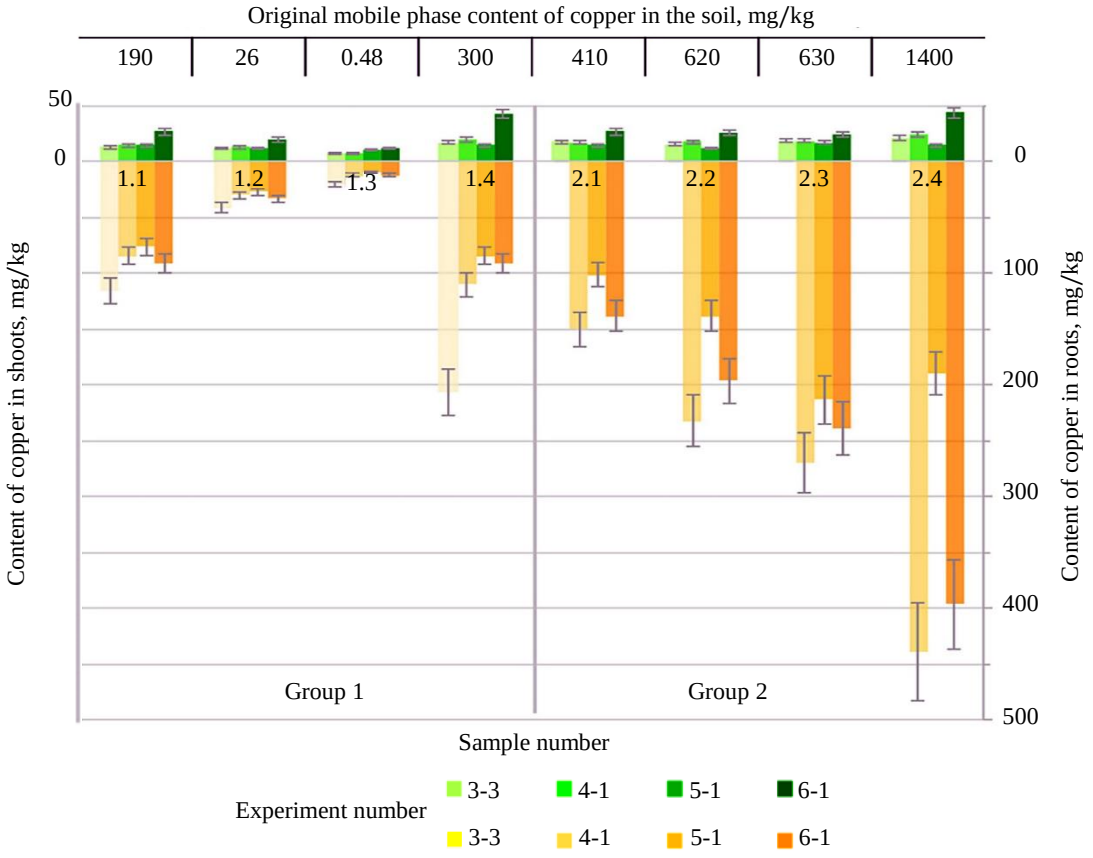


Fig.4. Content of copper in the shoots (green) and roots (yellow) of plants after the experiment

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It has been demonstrated through experimental studies that a successful method of restoring the soil and vegetation cover in the Karabash area is the application of complex soil additives comprising peat, vermiculite and lime, with the addition of biochar or melatonin. Furthermore, for plots containing soil of group 1 composition, the application of a complex amendment comprising peat, vermiculite, and lime, without the introduction of additional soil improvers, is deemed to be an effective measure. The quantity of components necessary, along with the approximate price of the additives, is outlined in Table 7.

Table 7

Characteristics of the complex soil additive

Composition	The quantity of additive in the composition, t/ha					Approximate price, rub. per t
	Lime	Peat	Vermiculite	Biochar	Melatonin	
Lime (1D), peat, vermiculite	300	1000	450	–	–	65,000
Lime (1D), peat, vermiculite, biochar				400	–	100,000
Lime (1D), peat, vermiculite, melatonin				–	0.002	> 100,000,000

Lime proved effective by reducing the mobility of elements by increasing the pH; according to the experimental results, biochar is effective as a sorbent and sorbs chemicals even before they reach the root system; in contrast, melatonin increases the plants' tolerance to HM but has almost no effect on the mobility of elements in the soil. The application of peat and vermiculite has been demonstrated to reduce pollutant concentrations through a combination of dilution, sorption, and complexation into organic complexes, in addition to the occurrence of a partial increase in pH. The results obtained from this research suggest that in order to achieve a stable, erosion-resistant soil-vegetation cover; the problem of soil contamination with heavy metals under acidic conditions must be resolved through the application of a combination of additives. The application of individual additives alone is insufficient to achieve a stable, erosion-resistant soil-vegetation cover.

Conclusion

An assessment of the effectiveness of various soil additives in solving the problem of recovering the soil-vegetation cover on acidic, heavy-metal-contaminated soils in the town of Karabash revealed the following:

- The individual use of commonly available soil-improving additives (nitrogen-free peat, lime, vermiculite) did not achieve positive results. This once again confirmed the importance of a specific approach to recovering and greening such man-made wastelands.
- A material composed of peat and vermiculite (with a soil, peat and vermiculite proportion of 8:2:1) was not able to produce stable vegetation cover in the experimental vegetation experiment, despite the fact that it improved the soil's physical-mechanical and physical-chemical properties.
- The addition of a single dose of lime to the aforementioned combination of components resulted in a reduced mobility of heavy metals (Cu, Ni, Pb) due to an increase in pH. It is evident, based on an analysis of all qualitative and quantitative indicators, that the usage of this additive had a positive effect on sample group 1. The study found that multiply increased doses of lime used as part of complex additives were not effective for any of the groups. In the majority of experiments, this led to stunting of plant growth.
- The addition of 10 wt.% of biochar to the complex additive (lime, peat, vermiculite) demonstrated an increased sorption capacity for HM, particularly group 2, which had the highest level of contamination. The experimental results demonstrated a significant decrease in metal accumulation in plants and an enhancement in visual appearance.
- The study found that the plant biomass and visual appearance were enhanced by the addition of melatonin (10 μmol/kg), which increased the plants' tolerance to HM. However, the use of melatonin as part of the complex supplement did not reduce the migration capacity of heavy metals; this is due to its mechanism of action.

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In summary, the results of the growth experiment have led to the proposal of two complex soil additives for field testing. The first additive is suitable for soils with a pH > 3.9 and a copper concentration in the mobile form of up to 100 MPC, and consists of a combination of peat, vermiculite, and lime. The second additive is suitable for soils with a pH < 3.9 and a copper concentration exceeding 100 MPC, and consists of a combination of peat, vermiculite, lime, and biochar. These solutions are environmentally and economically optimal for the recovery of the soil and vegetation cover in the town of Karabash. The findings of the study indicated that the combination of peat, vermiculite, lime and melatonin demonstrates promise for both soil groups. This strategy will not only reduce the concentration of HM in their mobile forms, but also increase the plants' tolerance to the negative impact of a contaminated soil environment.

In the next steps of the project, field tests are planned to be carried out by establishing designated testing areas in the Karabash territory.

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