International Journal of Engineering & Technology, 7 (4.36) (2018) 222-230

International Journal of Engineering & Technology

Website: www.sciencepubco.com/index.php/IJET

Research paper

Scientific Substantiation of the Technology of Finely-Dispersed Metallurgical Sludge Bioconversion

Polischuk S.D., Nazarova A.A., Churilov D.G., Rembalovich G.K., Kostenko M.Yu., Arapov I.S.

Ryazan State Agrotechnological University Named after P.A. Kostychev,390044, Russian Federation, Ryazan, Kostychev Str., 1

Abstract.

In the course of human activities large quantities of industrial wastes, unused by-products, remnants of raw materials, materials that completely or partially change their properties, accumulate in nature. On the other hand, one of the important problems is the remediation and increase in the yield of agricultural land, especially in central Russia. Numerous data on the use of sludge sediments in the United States and Europe in growing agricultural crops have given grounds for assuming that such wastes can be used in Russian agriculture. The aim of the research work is experimental confirmation in field conditions the effectiveness of using metallurgical sludge accumulations for the remediation of soil resources in agriculture and increasing the yield of agricultural crops, especially in central Russia. The conducted research allowed to make a conclusion about the effectiveness of metallurgical sludge as a potential component of agricultural fertilizers. As test crops, technical plants,sunflower and rape, were selected as objects suitable for the bioconversion of metallurgical sludge. Crops treatment with 10 % solution of sludge turned to be the most effective means. It resulted in the increase of the sunflower field germination by 7 %, plant weight by 27.7-33.4 %, plant height by 33.9 % and leaf area by 11.6 % compared to the control. Parameters of the vegetative and generative functions of rapeseed plants changed as follows: field germination increased by 4 %, plant height by 13.8-33.6 %, plant weight by 20.0-25.5 % and photosynthesis productivity by 33.7 %. Thus, metallurgical sludge can be considered as an effective stimulator of vegetative and generative functions of agricultural crops in relation to these soil and climatic conditions. The obtained biochemical and electron microscopic data convincingly indicate the possibility of including the components of the sludge in the physiological processes of plants, including as trace elements, which is the reason for the observed stimulation of plants growth and development.

Keywords: *finely-dispersed metallurgical sludge, sunflower, rape, heavy metals, bioconversion*

1.Introduction.

The utilization of blast-furnace waste as well as wastes received from the purification of municipal wastewater and agricultural production is one of the main environmental problems. The lack of recycling technologies for such wastes leads to a permanent increase in the areas of ash dumps and an increase in pressuring the ecological state of the adjacent areas due to natural weathering, leaching of heavy metals and their transition into the atmosphere, soil and groundwater. Sludge is a finely-dispersed waste product, which includes a number of metals, such as nickel, aluminum, iron, copper, and others, which can accumulate in the cells and tissues of plants. These elements in small quantities are involved in the life processes of plants, i.e. they are trace elements. Trace elements due to their catalytic action allow plants to use main nutrient elements, the sun energy and water, more efficiently, which in turn has a positive effect on the productivity of plants and the quality of the crop. Foreign sources provide examples of the use of metallurgical sludge in agriculture, forestry and biotechnology [1, 2, 3]. The positive effects of metals and their compounds on plants are described, especially in the form of finely-dispersed particles, employees of Ryazan State Agrotechnological University Named after P.A. Kostychev work with. It is assumed that finely-dispersed metal-containing particles having prolonged action and gradually oxidizing in the soil, create unfavorable conditions for pathogenic microorganisms and are used by plants in the process of growth and development as trace elements [4-7], increasing the yield [8, 9, 10].

Heavy metal content is one of the most important criteria for environmental assessment of sediments. The concentration of the most dangerous eight elements Ni, Al, Fe, Zn, Cu, Cd, Pb and Cr is taken into account.

The maximum allowable concentration (MAC) of elements in the environment of human settlements during disposal as fertilizer varies in countries and continents.

There are several approaches to assess the influence of heavy metals (HM) in the agrocenoses, especially when introducing waste into the soil: 1) to assess the HM effect on the content of total forms in sediments and soil; 2) to assess the HM effect on the content of mobile element forms, which are extracted by ammonium acetate buffer from the sediment and from the soil; 3) to assess the effect of sediment judging by the content of HM in plants.

Analysis of available scientific data, on the one hand, indicates a high degree of novelty and relevance of the proposed direction - research on the interaction of finely-dispersed particles of anthropogenic origin with plant cells to develop the technology of using sludge organic-mineral accumulations to stimulate the growth of agricultural crops[11,12]. And on the other hand, there are difficulties in this direction associated with the probability of heavy metals bioaccumulation, including those in the form of highly active particles and compounds in plant tissues, that requires special attention to the issues of environmental friendliness and safety of the developed approaches. The low-cost and environmentally friendly technology based on the use of trace elements contained in metallurgical sludge and easily assimilated

nitrogenous compounds, taking into account the huge accumulated stocks of such wastes, created on the basis of these studies, will be of paramount social importance and will simultaneously increase the efficiency of agriculture [13].
The aim of the research work is developing scientific substantiation and experimental confirmation in field conditions of the effectiveness of using metallurgical sludge accumulations for the remediation of soil resources in agriculture and for increasing the yield of agricultural crops, especially in central Russia. These two factors ensure the undoubted relevance of the work.

2.Research design and methods

2.1. Development of laboratory process procedures for the process of finely-dispersed metallurgical sludge bioconversion.

For research, a technique was developed for dispersing the metallurgical sludge with ultrasound. In the framework of ultrasonic homogenization the following equipment was used: thermostats, cooled and heated with a range of temperature control in the working chamber from 0° C to 40° C and permissible temperature fluctuations ± 2° C; Bandelin SONOPULS HD 2200 ultrasonic homogenizer; scales for weighing the mass according to GOST 29329; drying oven with a range of temperature control in the working chamber from 50° C to 150° C and permissible temperature fluctuations ± 2° C; thermometers with a scale from 0° C to 40° C according to GOST 28498. Dispersion of sludge particles using ultrasound is an integral part of the method of preparing liquid fertilizers based on sludge. It is shown that the Bandelin SONOPULS HD 2200 ultrasonic homogenizer should be used during the dispersion operation. The maximum homogenizer power is not less than 200 watts and the working frequency is 22 kHz. The homogenization of the sludge particles in liquid (water-based) media is optimally carried out at a medium temperature of 45-55° C. Sludges are fine homogeneous powders of black or gray color, not containing large inclusions. The chemical composition of the sludge must meet the following requirements for the content of elements: zinc from 5 to 25 % of the mass, copper from 0.1 to 0.5 %, cadmium from 0.05 to 0.10 %, arsenic from 0.1 to 0.3 % and lead from 0.1 to 1 % of the mass. The average particle size determined by laser diffraction is no more than 10 microns.
Studies on the bioconversion of metallurgical sludge with the use of agricultural objects were carried out on the basis of the Center for Nanotechnologies and Nanomaterials for the Agro industrial Complex at Ryazan State Agrotechnological University (RSATU) in 2013-2017. Field experiments were carried out at the agrotechnological station of RSATU (Stenkino village, Ryazan oblast). The soil was light gray forest. The agricultural technology was carried out in accordance with regional recommendations. The experiment was small-plot and single-factor. The sown area of the plot was 56 m^2, the harvesting area was 30 m^2.
Metallurgical sludge before sowing was applied to the soil using an OP-24 trailed type sprayer with a 24.6 m wheel-bearing rod and a capacity of 2000, 2500 (according to TU - 4734-002-46558598-2003) intended for applying liquid complex fertilizers.
It was found that the stability and homogeneous distribution of highly dispersed sludge particles is one of the main methods for ensuring the quality of the product.
During the growing season, the following parameters were determined:
1) Phenological observations of the plants growth and development were carried out in all variants of the experiment. The day when at least 10–15 % of plants entered this phase was considered as the phase beginning, and the full onset of the phase was when at least 75 % plants entered the phase.
2) Determining field germination in 7 days after sowing.
3) Considering the density of standing plants in the phase of 4-5 leaves and before harvesting.
4) Determining the leaf area using the method of plots or templates [14]. The formula was as it follows:
$B(1m^2)= A/adm^2$, where
B is the leaf area from 1 m^2; A is the mass of leaves from 1 m^2; a is the mass of leaves from 1 dm^2.
5) Taking vegetative and reproductive parts of plants, as well as the root system for biochemical studies in all phases of vegetation.
6) The yield of green biomass and seeds was determined by harvesting plants from a registration plot and weighing. Counting the number of anthodia, seeds and pods on a single plant was also carried out during harvesting.
7) The grain yield was determined by collecting and threshing pods, ears and anthodia in terms of conditioned humidity and 100 % purity.
88) The moisture content of the grain at the time of harvesting was determined using laboratory tests. The ripeness of the seeds was determined during harvest by the organoleptic method.
9) The yield of dry matter of leafy mass and grain was determined after studying the moisture content of the above listed objects.
10) Dispersion analysis of experimental data was performed according to B.A. Dospekhov [15].
In laboratory setting the work was carried out to determine the effective concentrations of finely-dispersed metallurgical wastes in the habitat of agricultural crops, and a research design was defined:
1) Control - without sludge.
2) Metallurgical sludge solution with a substance concentration of 0.1 %.
3) Metallurgical sludge solution with a substance concentration of 1.0 %.
4) Metallurgical sludge solution with a substance concentration of 10.0 %.
Seeds were germinated under the conditions stipulated by GOST 12038-84 using a heated thermostat TCO-1M with a temperature range from 0° C to 60° C and admissible temperature fluctuations of ± 1° C. Determining the optimal concentrations of finely-dispersed sludge and changes in the activity of plant enzymes under its influence were carried out on the 10th day of germination in laboratory setting and in the phase of formation of the 3rd leaf in the field.
To determine the activity of nitrate reductase, catalase, and peroxidase in plant tissues, the following method was used: photometric kinetic test using a KFK-3-01-ZOMZ photoelectric colorimeter [16, 17]. Sunflower seeds [18, 19] and rape seeds [20-22] were selected as test objects for the research.

3.Results and discussion

3.1. Conducting laboratory studies on the bioconversion of finely-dispersed metallurgical sludge using agricultural crops.

A key aspect of the interaction of metal-containing particles with plant tissues is their influence on biochemical processes in plant cells. The reason for this is the key importance of trace elements, especially such as iron, zinc, copper and cobalt in the formation of enzymes and cell metabolism.
One of the most important enzymes is nitrate reductase. It is an enzyme for reducing nitrates to nitrites and its activity limits the supply of nitrogen to nitrates in plants, and the productivity of nitrogen in soil and mineral fertilizers largely depends on nitrate reductase functioning [23, 24].
Studying the activity of this enzyme is of particular interest for our research, since sludge contains trace elements and macroelements, in particular nitrogen. In addition, the study of the coordination of nitrate absorption and recovery at the level of a whole plant is necessary to determine the causes of excessive accumulation of nitrate nitrogen in the biomass of agricultural crops, since increasing its concentration above a certain limit causes

pathological phenomena in humans and animals consuming plant products. For the plant itself, the accumulation of nitrate in the tissues is not toxic, therefore, it is considered as a protective reaction that contributes to the protection of plants from the accumulation of toxic intermediate products of the assimilation of nitrate nitrogen. It is assumed that biological mechanisms of inhibiting the activity of nitrate reductase, which catalyzes the primary reduction of nitrate, play a significant role in this plant defense reaction [25, 26]. It is shown that both aboveground and underground plant organs take part in nitrates assimilation. The enzyme activity in the aerial parts (leaves and stems) is higher than that in roots. In early phases of development, it is the leaves that make up the bulk of the plants. In this regard, the proportion of reduced nitrates in them is greatest.

The results are presented in tables 1 and 2.

Table 1 - Nitrate reductase (NR) activity in sunflower roots and sprouts under the influence of metallurgical sludge during incubation of the material homogenate at 37° C in the presence of sodium nitrate at pH 7.4

Variant	Nitrate reductase activity, μmol / hr × g of tissue			
	Roots		Leaves and stems	
	Absolute value	% to control	Absolute value	% to control
Control	202.19±39.54	-	240.21±41.62	-
Sludge 1.0%	230.63±14.95*	+14.06	301.94±42.73*	+25.69
Sludge 10%	265.51±28.43	+31.31	312.15±46.12	+29.95

Note: * – $P \leq 0.05$

Table 2 - Nitrate reductase (NR) activity in rape roots and sprouts under the influence of metallurgical sludge during incubation of the material homogenate at 37° C in the presence of sodium nitrate at pH 7.4

Variant	Nitrate reductase activity, μmol / hr × g of tissue			
	Roots		Leaves and stems	
	Absolute value	% to control	Absolute value	% to control
Control	186.14±36.32	-	207.78±23.61	-
Sludge 1.0%	223.63±26.51*	+21.45	248.58±36.21*	+19.64
Sludge 10%	242.04±41.64	+30.03	269.95*±41.07	+29.92

Note: * – $P \leq 0.05$

Thus, considering the effect of metallurgical sludge on the growth and assimilation of nitrates by plants, it should be emphasized that in the field and laboratory setting it acts in combination in the same way. Nitrate reductase activity is increasing, that reflects the potential of plants to assimilate oxidized nitrogen, although the correlation between the rate of nitrates reduction and intensity of their use is not always observed, since the availability of a substrate or a shortage of reducing agents may be a limiting factor.

Catalase belongs to the class of oxidoreductases and is involved in the catalytic decomposition of hydrogen peroxide to water and oxygen. This enzyme is widely distributed in tissues. Catalase is the basic primary antioxidant of the defense system, sharing this function with GSH-PX. Both enzymes detoxify the active oxygen radical. In addition to differences in their substrate specificity, these two enzymes differ in affinity for the substrate.With a low content of H_2O_2, organic peroxides are predominantly catalyzed by peroxidase. However, at high concentrations of H_2O_2 catalases work. The level of activity varies not only in different tissues, but also within the cell itself. Catalase is a tetrameric heme-containing protein that is formed in the cytosol as monomers that do not contain heme [27].

Enzyme peroxidase is one of the links in the chain of electron transfer in the mitochondrial alternative respiratory chain. For peroxidase, its participation in redox reactions during photosynthesis has been proven, and detection in mitochondria indicates that it is involved in energy metabolism of cells; in formation of auxin and ethylene; reduction of nitrites, nitrates (in nitrogen metabolism), respiratory processes, and is involved in regulating development, organogenesis, etc. [28 - 30].

The formation of reactive oxygen intermediates on the cell surface (“oxidative burst”) is one of the earliest responses to the stressful effects of abiotic, biotic and anthropogenic nature, and represents a change of balance between the formation of reactive oxygen intermediates and the antioxidant protection activity in favor of the first. Therefore, peroxidase metabolism is very important. Enzyme peroxidase reacts differently to the presence of metallurgical sludge which different concentrations influence the development of sunflower seeds.

As a result of studying the peroxidase and catalase activity of sunflower at different concentrations of metallurgical sludge, it was established that the peroxidase activity in roots and aerial parts of sunflower was significantly reduced relative to the control. Sunflower seedlings had a minimal peroxidase activity with a sludge content of 10 %: it was 20 % in roots and 15 % in aerial parts.

At the same time, a significant increase in catalase activity was observed in roots and aerial parts of sunflower. The activity of this enzyme increased by 10 % (1 % sludge) and 15 % (10 % sludge) in roots and by 8 % and 16 % in aerial parts of sunflower compared to the control, that is, there was a positive correlation between catalase and peroxidase activity.

Consequently, the use of the studied metallurgical sludge contributes to an increase in catalase activity and a decrease in peroxidase activity, as indicators of an increase in plant resistance during the development period, and at the development stage of the 3rd true leaf, growth processes are enhanced.

With increasing sludge content, an increase in peroxidase activity in rapeseed roots is observed from 24 % (1 % sludge) to 29 % (with a sludge content of 10 %).

Peroxidase activity in sprouts is also higher than that of the control for all sludge concentrations, and higher than in roots, increasing from 18 % (1 % sludge) to 22 % (10 % sludge).

At the same time, the influence of metallurgical sludge causes a decrease in catalase activity in rape aerial part and roots. Catalase activity in rape roots varies from 11 % below the control (sludge 1 %) to 21 % (sludge 10 %). Catalase activity in the aerial part is lower than the control by 24 % (1 % sludge) and by 17 % (10 % sludge).

Catalase in addition to the main function, the decomposition of hydrogen peroxide, has some peroxidase activity. Its biological role is not only to protect against peroxide formed during metabolism, but also to provide plants with oxygen. There is a seeming antagonism between these enzymes, however, acting simultaneously, both enzymes do not violate their specific functions and in the presence of sludge the development of rape is not inhibited.

Thus, the activity of nitrate reductase, peroxidase and catalase enzymes was evaluated when determining the effect of metallurgical sludge on plant objects by biochemical parameters in field tests during the development phase of the 3rd true leaf. It has been established that enzymes nitrate reductase, catalase and peroxidase react differently to the presence of metallurgical sludge in the soil of the experimental plot, depending on the concentration. The activity of enzymes differs according to the place of determination: roots or sprouts with the same concentration of sludge, that is, it is biologically active.

3.2. Conducting field studies with dosed introduction of metal-containing wastes into the soil.

The results of the research on the effect of sludge on sunflower plants are presented in Figures 1-3 and Tables 3-5.

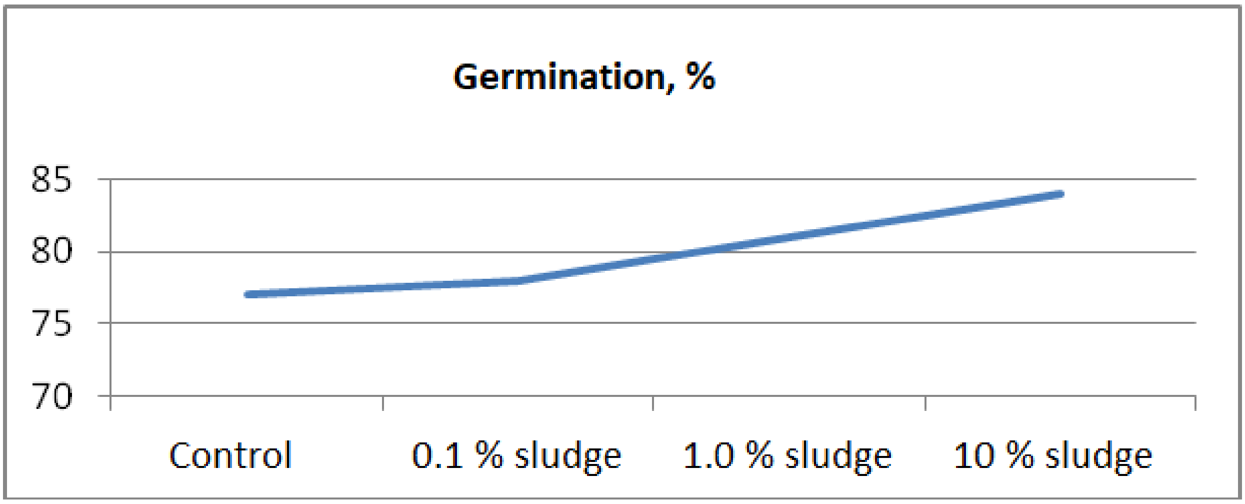

Figure1 - Field germination of sunflower when interacting with sludge from metallurgical production

An increase in the concentration of metallurgical sludge in the soil of the experimental plot contributed to an increase in field germination of sunflower seeds in all variants: by 1 % with 0.1 % sludge, by 4 % with 1.0 % sludge and by 7 % with 10 % sludge as compared to control seeds.

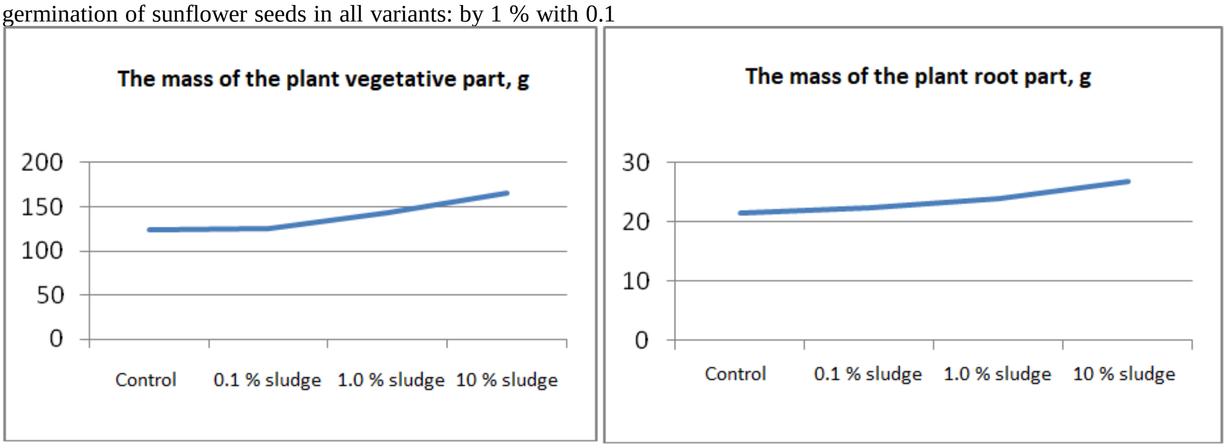

Figure2 - The mass of sunflower plants in the interaction with the metallurgical sludge

The mass of the vegetative part of sunflower plants when interacting with sludge at low concentrations was almost at the level of control values, but at high doses of sludge, the mass of the vegetative part of sunflower significantly exceeded the control, maximum by 33.4 % when 10.0 % sludge.

The weight parameters of the root system of a sunflower plant with an increase in the concentration of metallurgical sludge in all variants exceeded the control. The best result was observed when the concentration of sludge is 10 % (+ 24.1 %).

Table3 – Sunflower plant height when interacting with metallurgical sludge

Variants	Sunflower (phase of 9th – 10th real leaf)	
	Plant height, cm	Ratio to control, %
Control	46.5±1.2	–
Sludge0.1%	48.8±0.5	+4.9
Sludge 1.0%	54.6±1.8	+17.4
Sludge 10%	62.3±4.6	+33.9

* Note P≥0.95

The sludge from metallurgical production largely stimulated the growth of sunflower plants (Figure 3 and Table 3). The height of plants at experimental plots exceeded the control at high concentrations by 17.4 - 33.9 %.

Figure 3. Sunflower plants after field tests (01.07.2016)

Table4 – Leaf surface area of sunflower plants when interacting with metallurgical sludge

Variants	Sunflower (phase of 15^{th} – 16^{th} real leaf)	
	Leaves area per 1 plant, m^2	Ratio to control, %
Control	0.2543±0.0153	–
Sludge0.1%	0.2638±0.0187	+3.7%
Sludge 1.0%	0.2795±0.0215	+9.9%
Sludge 10%	0.2837±0.0203	+11.6%

* NoteP≤ 0.05

Analysis of the leaf surface area of sunflower plants showed that the introduction of metallurgical sludge into the soil before sowing at a concentration of 0.1 % contributes to an increase in the leaf surface area by 3.7 %, at a concentration of 1.0 % by 9.9 % and at a concentration of 10.0 % by 11.6 % as compared with the control value.

Figure4 - Sunflowerplantsfromthecontrol and experimental plots (31.07.2016)

Table5 –The yield of sunflower green mass when interacting with metallurgical sludge

Variants	Sunflower	
	Mass of a plant with ananthodium, kg	Ratio to control, %
Control	2.311±0.015	–
Sludge0.1%	2.408±0.023	+4.2
Sludge 1.0%	2.513*±0.044	+8.7
Sludge 10%	2.950±0.038	+27.7

* Note$P \leq 0.05$

The average weight of 1 sunflower plant, and as a result, the weight of plants per unit area of agricultural land, varied significantly depending on the variant. So, the introduction of 0.1 % metallurgical sludge into the soil increased the mass of 1 plant by 4.2 %, the increase for the variant with 1.0 % sludge was 8.7 % and that for 10 % sludge was 27.7 % relative to the control. Thus, the maximum sludge concentration of 10 % showed the best effect on the yield of the green mass of sunflower in the given conditions.

The results of the influence of various concentrations of metallurgical sludge on the mass of rapeseed plants, photosynthesis productivity and yield are presented in Tables 6-8 and Figure 5.

The effect of metallurgical sludge on the mass of rapeseed plants was reflected in a significant increase of these parameters in all variants, especially at high concentrations (1.0 % and 10.0 % sludge), increasing the mass of the plant by an average of 16-20 % as compared to the control.

Table6 – The mass of the vegetative and root parts of rape plants when interacting with metallurgical sludge

Variants	Rapeseed (phase of 5^{th} – 6^{th} real leaf)			
	Mass of the vegetative part, g	Ratio to control, %	Mass of the root part, g	Ratio to control, %
Control	8.73±0.53	–	0.76±0.004	–
Sludge0.1%	9.20±1.02*	+5.7	0.78±0.001	+2.6
Sludge 1.0%	10.10±1.06	+16.1	0.81±0.009*	+6.5
Sludge 10%	10.40±0.92	+19.5	0.85±0.012	+11.8

* Note$P \leq 0.05$

The concentrations of sludge from metallurgical production used in the experiment stimulated the growth of rapeseed to varying degrees. With 0.1 % sludge the plant height increased by 3 %, 1.0 % sludge caused 9.6 % increase and 10 % sludge resulted in the increase by 13.8 % relative to the control.

During the growing season of rapeseed, the photosynthesis productivity of plants was determined.

Table7 – The productivity of photosynthesis of rape plants

Variants	g/m^{2}per day	Ratio to control, %
Control	9.8±0.12	–
Sludge0.1%	10.4±0.15*	+6.1
Sludge 1.0%	11.5±0.04	+17.3
Sludge 10%	13.1±0.10	+33.7

* Note$P \leq 0.05$

When increasing sludge concentration, productivity increased, reaching the maximum value with 10 % sludge - 13.1 g / m2 per day, which exceeds the control by 33.7 %.

Increasing the concentration of sludge from 0.1 % to 10 % contributed to an increase in the height of rapeseed plants (Figure 5) by 3.2 - 34.6 %, as well as the weight of the vegetative part of the plant by 6.4 - 25.5 % as compared to the control.

Figure5 – Rapeseed ripening phase (control and sludge 10 %)

Rapeseed yields are directly dependent on the number and weight of the pods containing oilseeds, which are presented in Table 8.

Table 8 – The yield of rapeseed pods when interacting with metallurgical sludge

Variants	Stage of maturity			
	Number of pods from 1 plant, pcs.	Ratio to control, %	Mass of 10 pods, g	Ratio to control, %
Control	45±2	–	2.9±0.1	–
Sludge0.1%	44±3	–	3.0±0.4	+3.4%
Sludge 1.0%	51±4	+13.3%	3.8±0.3	+31.0%
Sludge 10%	58±6	+28.9%	4.1±0.2	+41.4%

* NoteP≤0.95

Both the number of pods per 1 plant and the weight of 10 pods directly depended on the concentration of metallurgical sludge. Increasing the concentration of sludge contributed to the increase of these parameters and the maximum results were observed in the variant with 10 % sludge. So the number of pods per 1 plant increased by 28.9 % and the weight of 10 pods increased by 41.4 % relative to the control.

Depending on the concentration of the preparation, the crop yield varied from 0.155 kg/m2 (0.1 %) to 0.217 kg/m2 (1 %), with control parameters being 0.161 kg/m2 (10 %). Maximum yield increases were caused by the preparation in a concentration of 10 %.

Considering the presence of heavy metals in metallurgical sludge, their content in the soil and grown agricultural crops was analyzed (Tables 9, 10).

The content of chemical elements at different stages of plant development was determined by atomic absorption spectrometry (KVANT – Z. ETA spectrometer).

Table9- The content of heavy metals in soil (mg/kg) when using finely-dispersed sludge for rape

Metals	Control	1%	10%	MAC
Before sowing				
Cu	43.4	44.0	45.1	165
Zn	68.5	69.0	69.9	165
Pb	19.3	19.6	21.3	96
Cd	1.17	1.23	1.52	9
Cr	96.7	96.8	97.4	100
Ni	51.2	52.2	53.8	105
Mo	3.10	3.12	3.68	
After harvesting				
Cu	45.6	49.4	45.1	165
Zn	74.2	78.6	79.9	165
Pb	19.8	19.8	21.3	96
Cd	1.21	1.22	1.52	9
Cr	98.1	101.6	107.4	100
Ni	51.7	53.8	53.8	105
Mo	3.11	3.14	3.68	

The content of heavy metals in the soil does not exceed the maximum allowable concentrations, including after harvesting. When released into the soil, metals are intensively absorbed by plants, which is shown by the results of the analysis of spring rape samples influenced by mineral fertilizers.

To understand the dynamics of accumulating the components of metallurgical sludge, electron microscopic studies were carried out with an elemental analysis of samples of various parts of mature rapeseed, sunflower and other crops. Particular attention was paid to accumulation of sludge in tissues of economically valuable plant organs, as a part of the mechanism of the biological effect of this waste on plants. Table 10 presents data on the content of fixed elements in economically significant parts of plants.

Table10 – Content ofNi, Al, Fe, ZnandCu (mg/kg) in tissues of experimental plants

Elements	Rapeseeds			Семена подсолнечника			ПДК*
	Control	0.1%	10%	Control	0.1%	10%	
Nickel	2.2±0. 1	2.5±0.1	3.8±0.1	2.8±0.1	3±0.1	4±0.2	-
Aluminum	5±0. 1	7±0.2	6.4±0.1	2±0.1	2.2±0.1	2.9±0.1	-
Ferrum	0.6±0.02	1±0.1	1±0.1	2±0.1	2.4±0.1	2.3±0.1	-
Zinc	42±1	46±1	45±2	39±2	44±02	46±2	50
Cuprum	2.4±0.2	3.2±0.1	2.8±0.1	-	-	-	10

*SanPiN #42-123-4089-86

Plants grown with the introduction of the lowest and highest doses of metallurgical sludge were selected for the study. The availability of test results of these dosages allows to judge about the tendency of accumulation of sludge elements in cells and to predict the features of their accumulation when using other concentrations. A similar approach based on predicting the properties of toxicants in unexplored concentrations when there are data on two far-ranging concentration values is widely used in toxicology, for example, in the course of probit analysis.

According to the results of elemental analysis, the following metals were found in plant tissues: Ni, Al, Fe, Zn and Cu. Some insignificant amounts of Cd, Pb and Cr were also found.

4. Discussion

Analyzing the data of laboratory and field experiments, it can be noted that in most cases in seeds of plants grown using sludge, there is a slight increase in the content of heavy metals, directly proportional to the increase in the rate of sludge introduction. It is known that some metals are antagonists in biochemical processes of plants, therefore, the amount of zinc and chromium naturally decreases due to high iron content in the sludge. The content of regulated heavy metals according to SanPiN No. 42-123-4089-86 does not exceed the MAC.

Sludge increases the activity of enzyme nitrate reductase involved in protein metabolism (up to 31.31 % in roots and up to 29.95 % in the vegetative part above the control), as well as the activity of redox enzyme catalase up to 15-16 % higher than the control in roots and leaves. The increase of biochemical processes in the plant is consistent with the results of field tests. Sludge at a concentration of 10 % contributed to an increase in mass and plant growth up to 33-34 % relative to the control, increased leaf area (up to 11.6 %), photosynthesis productivity (+ 33.7 %) and, as a result, the yield of sunflower seeds (+ 27.7 %) and rapeseed (41.4 %). Slurry wastes of metallurgical production showed their biological activity for industrial crops and the safety of use in selected concentrations.

5. Conclusions

1) Sludge in most of the used concentrations (application rates) in field experiments significantly increases the growth and development rates, as well as the yield of sunflower (by 8-28 %) and rapeseed (by 15-44 %), while the MAC of heavy metals loams.in plants and soil was not fixed.

2) Depending on the type of plants and soil type, various sludge application rates were most effective: 50-100 kg/ha on gray forest soils and 500-2000 kg/ha on poorer heavy

3)In aerial parts (leaves and stems) of sunflower at a sludge concentration of 1 %, the activity of enzyme nitrate reductase is higher compared to that in roots. When introducing a 10 % solution of metallurgical sludge into the soil, the content of nitrate reductase in roots increases in the range of 20 % - 30 % above the control, as in the above-ground parts. In rapeseed, the activity of enzyme nitrate reductase is the same in the aerial parts and roots and above the control, as in sunflower, i.e. nitrate reductase activity is increasing, reflecting the plant's potential to absorb oxidized nitrogen.

4) The peroxide changes in experimental plants occur as it follows: the peroxidase activity in roots of experimental rape plants increases by 25 % when sludge concentration of 1.0 % and decreases to 10 % below the control in sunflower. In aerial parts of plants, peroxidase activity in rapeseed increases by 18 % and decreases in sunflower to 10 % below the control.

With an increase in the concentration of sludge to 10 %, the peroxidase activity in rapeseed roots increases to 28 % above the control and also decreases in sunflower to 20 %. In the above-ground part of plants, it decreases to 15 % in sunflower and rises accordingly to 22 % in rapeseed relative to the control.

5) Catalase belongs to the group of enzymes with high speed work, and plays a certain role in the processes of adaptation of the organism to stress. A significant increase in catalase activity was observed in roots and aerial parts of sunflower. This enzyme activity increased by 10 % (1 % sludge) and by 15 % (10 % sludge) in roots and by 8 % and 16 % in aerial parts of sunflower as compared with the control, that is, there was a positive correlation between catalase and peroxidase activity.

Catalase activity in rapeseed roots varied from 11 % below the control (1 % sludge) to -21 % (10 % sludge). Catalase activity in the aerial part remained below the control by 24 % (1 % sludge) and by 17 % (10 % sludge).

There is a seeming antagonism between enzymes peroxidase and catalase, however, acting simultaneously, both enzymes do not violate their specific functions and in the presence of sludge inhibiting the development of rape does not occur.

6)The obtained results indicate the prospects of the chosen direction, which can reduce the impact on the environment of one of the most powerful environmental threats - huge accumulations of finely-dispersed metallurgical technogenic waste. Metallurgical sludge in certain concentrations can be considered as a regulator of plant growth and development and an environmentally friendly tool. The ability of the sludge to activate physiological and biochemical processes during the period of seed germination revealed in the process of research is of great practical interest. Changes in plant cells, both in organelles and in the cytoplasm, undergo the most significant changes in enzymes, in particular nitrate reductase, peroxidase and catalase, whose activity and spectrum change under the influence of metallurgical sludge. The use of sludge makes possible to increase growth rates, and, consequently, the yield.

7)Preliminary calculations showed that the use of metallurgical sludge instead of fertilizers for enriching the soil with certain substances has an impressive economic efficiency. The costs of using sludge will be several times lower than in a case with classical fertilizers. Thus, this approach is a promising direction in the agro-complex, primarily in regions with a developed metallurgical industry.

References

[1] LuC.M., ZhangC.Y., WenJ.Q., WuG.R.andTaoM.X. 2002. Research of the effect of nanometer materials on germination and growth enhancement of Glycine max and its mechanism. Soybean Sci 21:168–172.

[2] Hong F.S., Yang F., Liu C., Gao Q., Wan Z., Gu F., Wu C., Ma Z., Zhou J. and Yang P. 2005. Influence of nano-TiO_2 on the chloroplast aging of spinach under light. Biol Trace Elem Res 104:249–260., Zheng L., Hong F., Lu S., Liu C. 2005. Effect of nano-TiO_2 on strength of naturally aged seeds and growth of spinach. BiolTraceElemRes 104:83–91.

[3] Kondakov S.E., Kuznetsov D.V., Churilov D.G. et al. Determining the optimal concentrations of sludge from metallurgical production according to vital and morphological parameters. Modern problems of science and education, 2013, no. 5

[4] Samoylova M.V., Churilov D.G., Nazarova A.A., Polischuk S.D.andByshov N.V. Biologically Active Nanomaterials in Potato Growing. Nano Hybrids and Composites. Vol. 13, 2017. pp. 91-95, DOI 10.4028 /www.scientific.net /NHC.13.91

[5] Eskov E.K. and Churilov G.I. The influence of corn seed treatment with ultrafine iron powder on the development of plants and accumulation of chemical elements in them. Agroecology, 2011, no. 10.

[6] Zheglova T.V., Vorontsova S.V., Nazarova A.A. and Churilov G.I. The effect of iron in the ultradisperse state on the chemical composition of plants and corn seeds. Innovative fundamental and applied research in the field of chemistry for agricultural production: (collection). Proceedings of the III International Internet Conference. Orel, Publishing house Orel SAU, 2010, pp. 16-20.

[7] Khodakovskaya M., Dervishi E., Mahmood M., Xu Y., Li Zh., Watanabe F. and Biris. A. S. ACS NANO: Carbon Nanotubes Are Able to Penetrate Plant Seed Coat and Dramatically Affect Seed Germination and Plant Growth, pp 3221–3227.

[8] Lee Ch.W., Mahendra Sh., Zodrow K., Li D., Tsai Y., Braam J. and Alvarez P. Developmental phytotoxicity of metal oxide nanoparticles to Arabidopsis thaliana. Environmental Toxicology and Chemistry, Vol. 29, No. 3, pp. 669–675, 2010

[9] Lee W.M., An Y.J., Yoon H. and Kweon H.S. 2008. Toxicity and bioavailability of copper nanoparticles to the terrestrial plants Mung Bean (Phaseolusradiatus) and Wheat (Triticumaestivum): Plant agar test for water-insoluble nanoparticles. EnvironToxicolChem 27:1915– 1921.

[10] Nazarova A.A., PolischukS.D. and Churilova V.V. Physiological, biochemical and productive parameters of malting barley using biologically active nanomaterials. Sugar, 2017, no. 1, pp. 22-25.

[11] Kuzmin N.A., Korenev G.V. and Shevchenko V.E. Theoretical and practical fundamentals of crop production. Proc. allowance, Voronezh, 1998, 200 p.

[12] Vition P.G. The use of organo-mineral fertilizers for growing crops and zoomelioration of pedobionts. Modern Science Success. 2017. vol. 1. Issue 9. Pp. 138-142.

[13] Fedulova A.D., Merzlaya G.E., Postnikov D.A., Postnikova K.V. Ecological assessment mineral, organic and organo-mineralnoy of systems of fertilizer in the after-effect at cultivation of oats. Modern Science Success. 2017. Vo2. 1. Issue 10. Pp.140-143.
[14] Ashirbekov M.Zh., Batkaev Zhan Agroecological aspects of using organo-mineral fertilizers in cotton-agricultural cultivation in the south of kazakhstan. Modern Science Success 2017, vol. 1, no. 9, pp. 77-82.
[15] 15.Dospekhov B.A. Methodology of field experience (with the basics of statistical processing of research results). 5thed., M., Agropromizdat, 1985, 351 p.
[16] 16.Methods of biochemical studies of plants. Ed. by A.I. Ermakova, L., Agropromizdat, 1987, 430 p.
[17] 17.Rogozhin V.V., Verkhoturov V.V. and Kurilyuk T.T. Antioxidant system in germination of wheat seeds. News of the Academy of Science, Biological Series, 2001, no. 2, pp. 165-173.
[18] 18.Andryukhov V.G. and Ivanov H.H. Sunflower in the Central Black Earth Zone. Voronezh, 1970, 95 p.
[19] 19.Pustovoit G.V. Sunflower classification. Sunflower. Ed by B.C. Pustovoit. M., Kolos, 1975, pp. 14-21.
[20] 20.Guschina V.A., Agankin, E.V. and Zheryakev N.D. Multi-purpose rapeseed. Beekeeping, 2007, no. 10, pp. 32-33.
[21] 21.Savelyev G.S. Biological motor fuel for diesel engines based on rapeseed oil. Tractors and agricultural machines, 2005, no. 10, pp. 11-16.
[22] 22.Pokrovskaya S.F. On the production of biofuels in the European Union. Agro-industrial production: experience, problems and development trends. Ser. Technological problems of the AIC, M., VNIIESH, 2007, Issue 3, pp. 11-28.
[23] 23.Kuznetsov V.V., Ovcharenko G.A., Borisova N.N. et al. Nitratreductase as a heat shock target. Reports of the USSR Academy of Science, 1991, vol. 321, no. 3, pp. 635 -638.
[24] 24.Lipe S.G. The role of inorganic nitrogen ions in the processes of plant adaptation. PlantPhysiology, 1997, vol. 44, no. 4, pp. 487-498.
[25] 25.Beevers L. and Hageman R.H. Nitrate reduction in higher plants. Annu. Rev. Plant Physiol., 1969, vol. 20, pp. 495-522.
[26] 26.Campbell W.H. Nitrate reductase biochemistry comes of age. Plant Physiol., 1996, vol. 111, pp. 355-361.
[27] 27.Campbell W.H. Nitrate reductase structure, function and regulation: bridging the gap between biochemistry and physiology. Annu. Rev. PlantPhysiol. Mol. Biol., 1999. vol. 50, pp. 277-303.
[28] 28.Averyanov A.A. Active forms of oxygen and plant immunity. Success of modern biology, 1991, vol. 111, no. 5, pp. 722-737.
[29] 29.Andreeva V.A. Peroxidase enzyme: participation in the plant defense mechanism. M., Science, 1988, pp. 7–24.
[30] 30.Salonen E., Lin S., Reid M. L., Allegood M., Xi Wang, Rao A.M., Vattulainen I. and Pu Chun Ke Real-Time Translocation of Fullerene Reveals Cell Contraction, Small. vol. 4, no. 11, 2008.