

**Effect of heavy metal ions on early development of *Sinapis alba* L.****Nazariy Kozachok***

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Abstract. Soil contamination with heavy metal ions can limit land use and reduce the effectiveness of phytoremediation. *Sinapis alba* L. is considered as a promising test culture and a potential phytoremediant, but its response to various metals in the soil system needs to be clarified. The purpose of this study was to evaluate the effect of Zn^{2+} , Cu^{2+} , Pb^{2+} , and Fe^{3+} ions on the viability of seeds, the length of shoots and roots of *S. alba*. The experiment was performed in a model soil experiment with three repetitions for each variant. On Day 5, a significant decrease in viability was detected only under the action of Cu^{2+} at a concentration of 300 mg/dm^3 , where this indicator was $77.9 \pm 1.2\%$. On Day 10, the strongest effect was observed for Fe^{3+} : viability decreased to $34.9 \pm 1.2\%$, $27.7 \pm 1.2\%$ and $20.5 \pm 1.2\%$ at concentrations of 600, 800, and $1,000 \text{ mg/dm}^3$, respectively. For Cu^{2+} this indicator was $74.7-56.6\%$, $Zn^{2+} - 80.7-67.5\%$, and $Pb^{2+} - 81.9\%$ at the highest concentration. The length of shoots and roots decreased under the action of all the metals under study. Fe^{3+} caused the greatest growth suppression: the length of the shoots decreased from 54.62 to 9.94 mm, and the roots – from 35.96 to 13.09 mm. Therefore, morphometric parameters were found to be more sensitive to metal stress than to viability, and should be used for a comprehensive assessment of soil phytotoxicity. The results can be used by specialists in the field of environmental monitoring, soil science, and phytoremediation for a comprehensive assessment of the toxicity of soils contaminated with heavy metals

Keywords: phytotoxicity; biotest of phytotoxicity; grey forest soils; phytoremediation; metal-specific stress

INTRODUCTION

Soil contamination with heavy metals is an urgent problem for areas where the effects of hostilities, industrial stress, and intensive land use are combined. Ingress of Zn^{2+} , Cu^{2+} , Pb^{2+} and Fe^{3+} ions in the soil environment can disrupt seed

germination, root system development, and the development of aboveground biomass. For phytoremediation, it is important to evaluate not only the plant's ability to accumulate pollutants, but also the preservation of viability and growth in

Suggested Citation:

Kozachok, N., & Golub, N. (2026). Effect of heavy metal ions on early development of *Sinapis alba* L. *Biological Systems: Theory and Innovation*, 17(3), 84-93. doi: 10.31548/biologiya/3.2026.84.

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their presence. This is particularly important for gray forest soils, where the acidity, organic matter content, sorption capacity, and metal mobility can change the actual toxicity of the same reference concentrations.

C. Luo *et al.* (2024) summarised the effects of various hazardous substances in the soil on seed germination and showed that the effect is determined not only by the concentration, but also by the chemical form of the pollutant, the type of plant, and the duration of exposure. The researchers identified impaired water uptake, oxidative damage, and destabilisation of cellular structures as the main mechanisms of germination inhibition. However, this study was a review and examined a wide range of pollutants, so it did not provide an experimental answer regarding the comparative effects of Zn^{2+} , Cu^{2+} , Pb^{2+} , and Fe^{3+} for one test crop in the same soil environment. B. Krasnodębska-Ostręga *et al.* (2022) reviewed *Sinapis alba* as a promising plant for phytoremediation and summarised data on the accumulation of As, Tl, and platinum group elements. G.-G. Vasile *et al.* (2021) and N. Vasilache *et al.* (2023) confirmed this prospect for soils contaminated with As, Cd, Ni, and Pb, but showed a metal-specific plant response: As mostly stabilised in the roots, Cd and Ni accumulated more actively, and Pb was largely retained by the soil. The availability of metals depended on the properties of the soil and mobile fractions. However, these papers did not cover a comparative investigation of Zn^{2+} , Cu^{2+} , Pb^{2+} , and Fe^{3+} in the model grey forest soil in the early stages of development of *S. alba*.

P. Singh *et al.* (2024) examined five varieties of *Brassica juncea* in soils irrigated with wastewater and borehole water. The researchers found that contaminated irrigation reduced the morphological, physiological, and productive characteristics of plants, and the genotype determined the level of metal accumulation and the suitability of the variety for phytoremediation. These results highlighted the importance of species, genotype, and soil-water regime. However, the study concerned *B. juncea*, longer cultivation, and complex contamination, rather than an early reaction of *S. alba* on separate concentration series of metals. Y. Peng & I. Grigorieva (2024) analysed the effect of heavy metals and soil properties on plant germination in the system of *Brassica napus* and sorghum. The

researchers showed that roots may be more sensitive to soil factors than shoots, and toxicity assessment should consider the combined effects of pollution and the physicochemical characteristics of the soil. However, the multivariate model of this study did not replace the controlled comparison of specific ions within a single plant species. The paper did not consider *S. alba* and grey forest soils of northern Ukraine. A. Díaz *et al.* (2024) and D. Kramski *et al.* (2026) reported that soil additives can simultaneously alter the availability of metals and support growth of *Sinapis alba*. For As, compost increased accumulation, while the combination of dunite, compost, and nanoscale nivalent iron reduced mobilisation; for Cd, sawdust improved growth, and spruce sawdust increased the bioaccumulation rate by more than 30%.

Thus, these studies confirmed the prospects of *S. alba* for phytoremediation, but a specific combination of Zn^{2+} , Cu^{2+} , Pb^{2+} , and Fe^{3+} in the soil system, the experiment requires further investigation. Current studies confirmed the importance of soil conditions, species specificity, and simultaneous analysis of germination and growth, but do not provide an answer regarding the early metal-specific reaction of *S. alba* on Zn^{2+} , Cu^{2+} , Pb^{2+} , and Fe^{3+} in the grey forest soil. The purpose of this study was to determine how individual concentrations of these ions affect seed viability on Day 5 and Day 10 and the length of roots and shoots on Day 10, and to determine which indicators are most sensitive to metal-specific stress.

MATERIALS AND METHODS

The study was conducted under the conditions of a model soil experiment using *Sinapis alba* L. as a test crop. The substrate was soil collected from the outskirts of Kyiv, in the forest zone of Desnianskyi district. Working soil moisture was $60.0 \pm 0.5\%$, the proportion of organic matter – $3.0 \pm 0.5\%$. The studies were conducted on plant seeds and did not involve the involvement of humans, vertebrates, or invertebrates, nor the use of protected species. Therefore, bioethical approval procedures were not applied for these categories. Work with solutions of metal salts and contaminated substrate was carried out in compliance with laboratory chemical safety requirements; materials after the experiment were collected separately for disposal as metal-containing laboratory waste. The study was



conducted in accordance with the ethical standards of Convention on Biological Diversity (1992) and Convention on International Trade in Endangered Species of Wild Fauna and Flora (1973).

The experiment was performed in Petri dishes. Each Petri dish was filled with 25 cm³ of soil, after which a solution of the respective metal was added. Zn²⁺, Cu²⁺ and Pb²⁺ were introduced in the form of nitrates, Fe³⁺ – in the form of chloride. Three concentrations expressed in mg/dm³ of soil were used for each metal: Fe³⁺ – 600, 800, and 1,000 mg/dm³; Cu²⁺ – 100, 200, and 300 mg/dm³; Zn²⁺ – 200, 300, and 400 mg/dm³; Pb²⁺ – 50, 100, and 150 mg/dm³. For each variant of the experiment, 3 repetitions were performed. 30 seeds of *S. alba* were sown in each Petri dish. The control version contained the same volume of soil, water, and the corresponding amount of nitrate ions, but without the introduction of metals. Since Zn²⁺, Cu²⁺, and Pb²⁺ were introduced in the form of nitrates, and calcium nitrate was additionally introduced to equalise the effect of nitrate ions in experimental and control versions. Its amount was calculated so that the total intake of nitrates corresponded to 400 mg/dm³ of soil. After applying the solutions, all Petri dishes were supplemented with water to the same initial volume of liquid – 20 cm³. By Day 5, germination was carried out in the dark in closed Petri dishes. After Day 5, the dishes were opened and transferred to 200 lux lighting conditions. In the future, the plants were grown with sufficient watering, preventing the soil substrate from drying out. The phytotoxic effect was evaluated based on seed viability, shoot length, and root length. Viability was determined by the number of seeds that formed seedlings. The length of shoots and roots was measured on Day 10 of the experiment. These indicators were used as indicators of early response of *S. alba* to the action of Zn²⁺, Cu²⁺, Pb²⁺, and Fe³⁺ in model soil conditions.

Statistical analysis was performed separately for morphometric parameters and viability. For shoot and root lengths, ANOVA analysis of variance was used, followed by post hoc comparison between variants. The χ^2 test was used to analyse viability with subsequent post hoc correction of multiple comparisons. The viability of the control was considered to be 100%. The differences were considered statistically significant at the accepted level of significance after adjusting for multiple

comparisons. The choice of statistical approaches was determined by the nature of the data obtained. The length of shoots and roots were continuous quantitative indicators, so it was advisable to compare the average values between the experiment variants using variance analysis. Instead, the viability in each Petri dish was actually a ratio of two categories – seeds that formed seedlings and seeds that did not form viable seedlings. That is why χ^2 test was applied for this indicator. The control variant was taken as 100%, and all experimental variants were interpreted as deviations from it. This allowed assessing not the absolute similarity of the crop, but the loss of viability associated with the introduction of metal. This approach was consistent with the purpose of the study, since it was not aimed at pairwise ranking of all metals and concentrations against one another, but at determining which variants actually alter normal early development of *S. alba* compared to control.

RESULTS AND DISCUSSION

On Day 5, seed viability of *Sinapis alba* significantly differed from the control only in the variant with Cu²⁺ 300 mg/dm³ of soil and was $77.9 \pm 1.2\%$, $p = 0.002$. In other variants, viability did not significantly differ from the control on Day 5 (Table 1). On Day 10, the most pronounced decrease in viability was observed for Fe³⁺: $34.9 \pm 1.2\%$, $27.7 \pm 1.2\%$, and $20.5 \pm 1.2\%$ at 600, 800, and 1,000 mg/dm³, respectively, $p < 0.001$. For Cu²⁺, viability was significantly reduced at all concentrations and was $74.7 \pm 1.2\%$, $66.3 \pm 1.2\%$, and $56.6 \pm 1.2\%$ at 100, 200, and 300 mg/dm³, respectively. For Zn²⁺, a significant decrease was also found at all concentrations: $80.7 \pm 1.2\%$, $74.7 \pm 1.2\%$, and $67.5 \pm 1.2\%$ at 200, 300, and 400 mg/dm³, respectively. For Pb²⁺, viability was significantly reduced at just 150 mg/dm³ and was $81.9 \pm 1.2\%$, $p = 0.014$; for 50 and 100 mg/dm³ it was not different from control.

On Day 10, the average length of shoots and roots of *Sinapis alba* decreased in all variants of metal application compared to the control (Fig. 1). The strongest growth inhibition was observed for Fe³⁺: shoot length decreased from 54.62 ± 0.79 mm in the control to 16.14 ± 0.57 mm, 13.14 ± 0.74 mm, and 9.94 ± 0.69 mm at 600, 800, and 1,000 mg/dm³, respectively; root length – from 35.96 ± 0.38 mm to 18.35 ± 0.56 mm, 15.92 ± 0.52 mm, and 13.09 ± 0.41 mm. All differences in shoot and root

length were statistically significant, $p < 0.001$. For Cu^{2+} , Zn^{2+} , and Pb^{2+} there was also a decrease in the length of shoots and roots with an increase in the metal content in the soil. For Cu^{2+} , the length of shoots decreased from 35.16 ± 0.88 mm at 100 mg/dm³ up to 23.11 ± 0.74 mm at 300 mg/dm³, and the length of the roots – from 28.24 ± 0.43 mm to 20.20 ± 0.42 mm. For Zn^{2+} , the length of shoots

decreased from 44.61 ± 0.90 mm to 29.51 ± 0.69 mm, the length of roots – from 30.53 ± 0.49 mm to 24.09 ± 0.43 mm. For Pb^{2+} , the length of shoots decreased from 47.79 ± 0.82 mm to 32.62 ± 0.70 mm, the length of roots – from 32.96 ± 0.34 mm to 25.42 ± 0.39 mm. For all variants, differences from the control for shoot and root length were statistically significant, $p < 0.001$.

Table 1. Seed viability of *Sinapis alba* on Day 5 and Day 10 of the experiment

Metal	Concentration, mg/dm ³ of soil	Viability, Day 5, % $\pm$ SE	$p \chi^2$, Day 5	Viability, Day 10, % $\pm$ SE	$p \chi^2$, Day 10
Control	–	100.0	–	100.0	–
Fe^{3+}	600	96.5 ± 1.2	1.000	34.9 ± 1.2	<0.001
Fe^{3+}	800	91.9 ± 1.2	0.740	27.7 ± 1.2	<0.001
Fe^{3+}	1,000	84.9 ± 1.2	0.059	20.5 ± 1.2	<0.001
Cu^{2+}	100	88.4 ± 1.2	0.203	74.7 ± 1.2	0.001
Cu^{2+}	200	84.9 ± 1.2	0.059	66.3 ± 1.2	<0.001
Cu^{2+}	300	77.9 ± 1.2	0.002	56.6 ± 1.2	<0.001
Zn^{2+}	200	95.3 ± 1.2	1.000	80.7 ± 1.2	0.011
Zn^{2+}	300	91.9 ± 1.2	0.740	74.7 ± 1.2	0.001
Zn^{2+}	400	84.9 ± 1.2	0.059	67.5 ± 1.2	<0.001
Pb^{2+}	50	100.0 ± 1.2	1.000	92.8 ± 1.2	0.236
Pb^{2+}	100	96.5 ± 1.2	1.000	88.0 ± 1.2	0.097
Pb^{2+}	150	93.0 ± 1.2	0.820	81.9 ± 1.2	0.014

Note: values are given as the mean $\pm$ SE for three repetitions; 30 seeds were sown in each repetition. $p \chi^2$ – adjusted p-value post hoc comparison with the control; $p < 0.05$ – highlighted in red

Source: compiled by the authors

The overall picture of the results indicates that the reaction of *S. alba* had not only a concentration but also a time dependence. On Day 5, most of the variants were still indistinguishable from the viability control, but on Day 10, the differences between the metals became much more pronounced. This was especially noticeable for Fe^{3+} and Cu^{2+} , where a decrease in viability was

combined with a sharp suppression of the length of roots and shoots. For Zn^{2+} and Pb^{2+} , viability changed less, but morphometric parameters still decreased, which indicates a sublethal nature of the action. Therefore, it is impossible to fully assess the early phytotoxicity of the studied metals based on the proportion of viable seedlings alone.

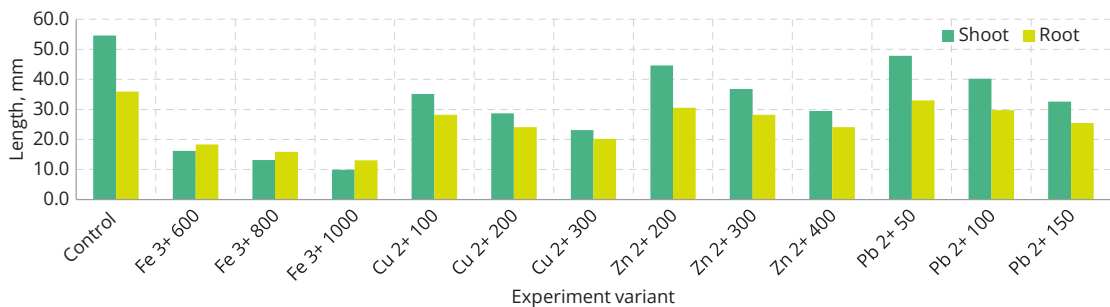


Figure 1. Length of roots and shoots of *Sinapis alba* with different content of heavy metal ions in the soil

Source: compiled by the authors

The results show that root and shoot length was a more sensitive indicator of phytotoxicity than viability. Viability decreased statistically significantly mainly under the action of Fe^{3+} and high concentrations of Cu^{2+} , while the length of roots and shoots decreased in all variants of metal application. M. Gupta *et al.* (2024) linked the Corene-centric response of *Brassicaceae* with the primary interaction of metals in the rhizosphere, apoplexy, cell wall, and root meristem. S. Bumsoms *et al.* (2021) also showed that Pb tolerance depends on metal accumulation processes and the root system response. This is consistent with the authors' data, in which morphometric parameters reacted earlier and more stable than viability.

The difference between viability and morphometric parameters is of great biological importance. The fact of the appearance of a seedling does not mean that the plant is able to develop effectively in the future in a contaminated substrate. In the first stages, the seed partially uses internal reserves, so the short-term viability index can remain relatively high even in conditions when the root meristem is already under stress. Root length in this case is a more sensitive sign of sublethal toxicity, since it reflects cell division and stretching in the growth zone, and the ability of the seedling to maintain water absorption and contact with the soil solution. The length of the shoot, in turn, shows whether early development passes into the development of biomass. That is why variants in which the viability did not differ statistically from the control, but the roots and shoots were shorter, cannot be considered completely safe for using *S. alba* as a phytoremediant.

The experimental conditions could enhance the root component of phytotoxicity. Under such conditions, the root system developed in a limited volume of substrate with high contact with the pore solution. Therefore, to interpret the results, it is important to consider the pH, organic matter content, cation exchange capacity, and availability of metals in the soil. J. Liu *et al.* (2022) noted that changes in soil properties can affect acidity, nutrient availability, structure, and microbial activity, and therefore change the plant's response to pollution.

The strongest phytotoxic effect was observed for Fe^{3+} . All its concentrations dramatically reduced viability, root length, and shoot length. In this experiment, Fe^{3+} was introduced in the FeCl_3

form and in the highest range among all metals – 600–1,000 mg/dm³ of soil. H. Li *et al.* (2022) showed that FeCl_3 can change the bioavailability of metals in the soil during remediation washing, but this procedure differs from direct application of Fe^{3+} to the substrate for germination. J. Clúa *et al.* (2024) link Fe^{3+} homeostasis regulation with processes in the root meristems. C. Xue *et al.* (2023) and F. Fodor (2024) reported that excess Fe can be associated with ROS/RNS stress, violation of K^{+} -homeostasis, damage to the root apex, and inhibition of meristem activity. Excess iron causes the accumulation of NO at the root tip; this NO can increase the leakage of potassium from the cells of the apical zone, which leads to a decrease in cell viability and stops root growth.

Delayed intensification of the Fe^{3+} effect may also be conditioned by the fact that the soil system gradually changes the availability of nutrients in the root zone. On Day 5, some of the seedlings could still support development at the expense of seed reserves, while on Day 10, the dependence on the work of the root and the availability of mineral elements increased. The high content of Fe^{3+} allows simultaneously increasing oxidative stress, local damage to meristem cells, and reducing the availability of phosphates due to their binding in a slightly acidic soil environment. This explains why Fe^{3+} -variants showed not just a decrease in the length of roots and shoots, but a sharp drop in viability at a later stage of observation.

Phosphorus was not additionally introduced in the experiment, so Fe^{3+} series may partially reflect more than just the direct toxicity of Fe^{3+} , but also changes in the availability of phosphates. In slightly acidic soil with a high concentration of Fe^{3+} , part of the available phosphorus could pass into a less accessible state, and under irrigation conditions – partially shift from the active zone of contact with the root. J. Clúa *et al.* (2024) showed that primary root suppression in phosphate deficiency is largely associated with Fe-dependent processes in the root meristem. R. Maniero *et al.* (2024) clarified that ferrit reduction in root meristems is part of the response to changes in phosphorus availability. The lack of additional introduction of phosphorus is unlikely in itself to explain such a sharp suppression of *S. alba* on Day 10. In early development, the plant can still partially use the internal reserves of seeds, while the consequences of P



deficiency are more pronounced in later stages of growth due to limited energy metabolism, biomass and root system development. N. Shen *et al.* (2021) showed that phosphate deficiency rearranged the root architecture of *Arabidopsis* by regulating Fe homeostasis and WRKY33 activity. Therefore, in this experiment, it is advisable to consider the phosphorous factor as a modifier of Fe^{3+} - toxicity, and not as an independent cause of a sharp drop in viability and growth. C. Xue *et al.* (2023), J. Clúa *et al.* (2024) and F. Fodor (2024) gave grounds to consider Fe specific action as a combination of metallic stress, violation of Fe P balance, and damage to the root meristem.

It is advisable to discard the chloride factor as the main cause of the Fe^{3+} effect. In the experiment, there was a calcium-nitrate background, since nitrates were equalised by applying $\text{Ca}(\text{NO}_3)_2$ up to a nitrate ion concentration of 400 mg/dm³ of soil; this background reduces the likelihood of a sharp pH- or salt shift as the only explanation for the results. Q. Huang *et al.* (2022) in experiments with *Brassica napus* delimited the effects of NaCl, Na_2SO_4 , KCl, and K_2SO_4 and found that the main contribution to growth suppression was made by Na^+ , and the additional effect of Cl^- and SO_4^{2-} depended on the dose and genotype. M. Rezayian & F. Zarinkamar (2023) also showed that calcium and signalling components can alter the salt response of plants. Therefore, in the presence of calcium chloride in this system, it is not the most likely factor of sharp growth inhibition; the effect of Fe variants is more logically associated with Fe^{3+} -specific toxicity and Fe P interactions.

Cu^{2+} also significantly suppressed development of *Sinapis alba*, especially at concentrations of 200 and 300 mg/dm³. G. Chen *et al.* (2022) described copper as an essential but redox active element, an excess of which causes the generation of reactive oxygen species, oxidative damage to membranes, and disruption of electron transport chains. X. Zheng *et al.* (2024) summarised the role of Cu in the plant-soil system. B. Rather *et al.* (2022) showed on mustard that Cu stress is accompanied by oxidative stress, impaired nitrogen metabolism, and reduced photosynthetic efficiency, and optimisation of nitrogen and ethylene signalling can mitigate these effects. In this experiment, the nitrate background was levelled, so the decrease in viability at high copper concentrations

and stable inhibition of root and shoot length were more likely to reflect the actual toxic effect of Cu^{2+} , not the difference in nitrate intake.

Zn^{2+} did not cause a statistically significant decrease in viability, although at concentrations of 300 and 400 mg/dm³ the p values approached 0.05. The length of roots and shoots decreased in all Zn^{2+} variants. H. Kaur & N. Garg (2021) reported that excess zinc causes erroneous incorporation of metals into metalloproteins, Fe deficiency-like signals, disruption of membrane and enzyme homeostasis, and secondary redox stress. S. Sadr *et al.* (2024) showed that Zn toxicity in rapeseed is associated with changes in cellular redox status and can be modulated by calcium and melatonin. P. Singh *et al.* (2024) demonstrated that the effectiveness of mustard phytoremediation depends on varietal characteristics and soil conditions. In grey forest soils, this additionally depends on pH, organic matter, cation exchange capacity, and Zn^{2+} mobility. This correlation of effects means that the use of white mustard for phytoremediation can be difficult on heavily contaminated Cu^{2+} and Zn^{2+} land plots due to loss of viability and biomass. However, at lower levels of contamination, plants retained sufficient development, which makes *S. alba* more suitable for moderately polluted soils, where the expected extraction of metals is not accompanied by a sharp inhibition of growth. In practice, this means that for preliminary selection of sites or metal concentrations, it is necessary to consider not only the fact of a statistically significant decrease in viability, but also the constancy of growth inhibition. If the roots and shoots are significantly shorter than the control even with relatively high viability, this option can give a weak plant cover and less biomass, and therefore – lower practical efficiency of phytoremediation in real soil conditions of long-term cultivation, especially with unstable water regime of the substrate.

Pb^{2+} did not have a statistically significant effect on viability, but it significantly reduced the length of roots and shoots. S. Busoms *et al.* (2021) showed that accumulation and tolerance to Pb in *Arabidopsis* depend on the genotype and processes associated with the root system. Z. Tan *et al.* (2022) and H. Manne *et al.* (2024) described the ability of Pb to disrupt antioxidant systems, destabilise membranes, and trigger secondary oxidative stress. At the studied concentrations



of 50-150 mg/dm³, this could manifest mainly as growth inhibition rather than as a decrease in viability. Low impact of Pb²⁺ on viability can also be associated with its binding to the soil and a corresponding decrease in the fraction available to the plant. N. Vasilache *et al.* (2023) showed that the availability of metals for *S. alba* was determined by the properties of the soil and the mobile fraction, and G.-G. Vasile *et al.* (2021) recorded a blockage of Pb in the soil. Since the availability of Pb was not measured in this experiment, this explanation should be considered as a well-founded hypothesis that requires separate testing. Y. Peng & I. Grigorieva (2024) also showed that in the soil system, the roots *Brassica* may be more sensitive to pollution factors than shoots, which supports this interpretation.

Overall, the results are better explained not by a single factor, but by a combination of metal-specific toxicity with soil system conditions. Grey forest soil with moderate organic matter without additional application of phosphorus created conditions in which root effects were most sensitive. For Fe³⁺, this implies the need for further separate investigation of the role of available phosphorus: Fe+P variants, control of P without Fe, pH measurement, mobile Fe, mobile P, and Fe-P ratios in pore solution are necessary to distinguish between direct Fe³⁺- toxicity and phosphate-modulated effect.

CONCLUSIONS

Metal ions of Pb²⁺, Zn²⁺, Cu²⁺, and Fe³⁺ had different effects on early development of *Sinapis alba*. The most sensitive indicators were the length of shoots and roots, while viability varied less steadily and depended on the metal and duration of exposure. On Day 5, a significant decrease in

viability was observed only under the action of Cu²⁺ at 300 mg/dm³ – 77.9 ± 1.2%, p = 0.002. On Day 10, the strongest and most delayed effect was found for Fe³⁺: 34.9 ± 1.2%, 27.7 ± 1.2% and 20.5 ± 1.2% at 600, 800, and 1,000 mg/dm³, respectively, p < 0.001; Cu²⁺ reduced viability to 74.7 ± 1.2%, 66.3 ± 1.2%, and 56.6 ± 1.2% at 100, 200, and 300 mg/dm³; Zn²⁺ – to 80.7 ± 1.2%, 74.7 ± 1.2%, and 67.5 ± 1.2% at 200, 300, and 400 mg/dm³; Pb²⁺ – to 81.9 ± 1.2% at 150 mg/dm³, p = 0.014. The length of roots and shoots decreased under the action of all metals under study: according to Fe³⁺, the length of shoots decreased from 54.62 ± 0.79 mm in the control to 9.94 ± 0.69 mm, and roots – from 35.96 ± 0.38 mm to 13.09 ± 0.41 mm. Cu²⁺, Zn²⁺, and Pb²⁺ also reduced this indicator. All differences in shoot and root length were statistically significant, p < 0.001. The use of *Sinapis alba* for phytoremediation of grey soils under high metal load is limited due to inhibition of early development. With moderate contamination, the crop may be suitable for further evaluation if viability and sufficient development of root and shoot biomass are maintained. Further research should focus on distinguishing the direct toxicity of Fe³⁺ and the phosphate-modulated effect by comparing variants with and without phosphorus addition, and measuring pH, mobile forms of Fe and P, and their ratios in the basal zone.

ACKNOWLEDGEMENTS

None.

FUNDING

None.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

- [1] Busoms, S., Perez-Martin, L., Llimos, M., Poschenrieder, C., & Martos, S. (2021). Genome-wide association study reveals key genes for differential lead accumulation and tolerance in natural *Arabidopsis thaliana* accessions. *Frontiers in Plant Science*, 12, article number 689316. doi: [10.3389/fpls.2021.689316](https://doi.org/10.3389/fpls.2021.689316).
- [2] Chen, G., Li, J., Han, H., Du, R., & Wang, X. (2022). Physiological and molecular mechanisms of plant responses to copper stress. *International Journal of Molecular Sciences*, 23(21), article number 12950. doi: [10.3390/ijms232112950](https://doi.org/10.3390/ijms232112950).
- [3] Clúa, J., Montpetit, J., Jimenez-Sandoval, P., Naumann, C., Santiago, J., & Poirier, Y. (2024). A CYBDOM protein impacts iron homeostasis and primary root growth under phosphate deficiency in *Arabidopsis*. *Nature Communications*, 15, article number 423. doi: [10.1038/s41467-023-43911-x](https://doi.org/10.1038/s41467-023-43911-x).

- [4] Convention on Biological Diversity. (1992, June). Retrieved from <https://www.cbd.int/doc/legal/cbd-en.pdf>.
- [5] Convention on International Trade in Endangered Species of Wild Fauna and Flora. (1973, March). Retrieved from <https://cites.org/eng/disc/text.php>.
- [6] Díaz, A.M., Forján, R., Gallego, J.R., Benavente-Hidalgo, L., Menéndez-Aguado, J.M., & Baragaño, D. (2024). Nanoscale zero-valent iron mitigates arsenic mobilization and accumulation in *Sinapis alba* grown on a metal(loid)-polluted soil treated with a dunite mining waste-compost amendment. *Plant and Soil*, 497, 241-255. doi: [10.1007/s11104-023-05879-x](https://doi.org/10.1007/s11104-023-05879-x).
- [7] Fodor, F. (2024). Iron nutrition and its biochemical interactions in plants: Iron uptake, biofortification, bacteria, and fungi in focus. *Plants*, 13(5), article number 561. doi: [10.3390/plants13050561](https://doi.org/10.3390/plants13050561).
- [8] Gupta, M., Dwivedi, V., Kumar, S., Patel, A., Niazi, P., & Yadav, V.K. (2024). Lead toxicity in plants: Mechanistic insights into toxicity, physiological responses of plants and mitigation strategies. *Plant Signaling & Behavior*, 19(1), article number 2365576. doi: [10.1080/15592324.2024.2365576](https://doi.org/10.1080/15592324.2024.2365576).
- [9] Huang, Q., Farooq, M.A., Hannan, F., Chen, W., Ayyaz, A., Zhang, K., Zhou, W., & Islam, F. (2022). Endogenous nitric oxide contributes to chloride and sulphate salinity tolerance by modulation of ion transporter expression and reestablishment of redox balance in *Brassica napus* cultivars. *Environmental and Experimental Botany*, 194, article number 104734. doi: [10.1016/j.envexpbot.2021.104734](https://doi.org/10.1016/j.envexpbot.2021.104734).
- [10] Kaur, H., & Garg, N. (2021). Zinc toxicity in plants: A review. *Planta*, 253, article number 129. doi: [10.1007/s00425-021-03642-z](https://doi.org/10.1007/s00425-021-03642-z).
- [11] Kramski, D.J., Warchoń, J., & Michalak, I. (2026). Sawdust-assisted phytoremediation: Boosting white mustard (*Sinapis alba*) growth and cadmium uptake from contaminated soil. *ACS Omega*, 11(3), 3738-3753. doi: [10.1021/acsomega.5c04779](https://doi.org/10.1021/acsomega.5c04779).
- [12] Krasnodębska-Ostręga, B., Sadowska, M., Biaduń, E., Mazur, R., & Kowalska, J. (2022). *Sinapis alba* as a useful plant in bioremediation – studies of defense mechanisms and accumulation of As, Tl and PGEs. *International Journal of Phytoremediation*, 24(14), 1475-1490. doi: [10.1080/15226514.2022.2036098](https://doi.org/10.1080/15226514.2022.2036098).
- [13] Li, H., Xiao, J., Zhao, Z., Zhong, D., Chen, J., Xiao, B., Xiao, W., Wang, W., Crittenden, J.C., & Wang, L. (2022). Reduction of cadmium bioavailability in paddy soil and its accumulation in brown rice by FeCl₃ washing combined with biochar: A field study. *Science of the Total Environment*, 851, article number 158186. doi: [10.1016/j.scitotenv.2022.158186](https://doi.org/10.1016/j.scitotenv.2022.158186).
- [14] Liu, J., Cui, J., Liu, H., Pan, Q., & He, X. (2022). Research progress of soil amelioration of acidified soil by soil amendments. *Journal of Environmental Engineering Technology*, 12(1), 173-184. doi: [10.12153/j.issn.1674-991X.20210119](https://doi.org/10.12153/j.issn.1674-991X.20210119).
- [15] Luo, C., Zhang, L., Ali, M.M., Xu, Y., & Liu, Z. (2024). Environmental risk substances in soil on seed germination: Chemical species, inhibition performance, and mechanisms. *Journal of Hazardous Materials*, 472, article number 134518. doi: [10.1016/j.jhazmat.2024.134518](https://doi.org/10.1016/j.jhazmat.2024.134518).
- [16] Maniero, R.A., et al. (2024). Ferric reduction by a CYBDOM protein counteracts increased iron availability in root meristems induced by phosphorus deficiency. *Nature Communications*, 15, article number 422. doi: [10.1038/s41467-023-43912-w](https://doi.org/10.1038/s41467-023-43912-w).
- [17] Manne, H., et al. (2024). Mitigation of lead toxicity in *Brassica juncea* L. by sulphur application – via various biochemical and transcriptomic strategies. *Journal of King Saud University – Science*, 36, article number 103175. doi: [10.1016/j.jksus.2024.103175](https://doi.org/10.1016/j.jksus.2024.103175).
- [18] Peng, Y., & Grigorieva, I.Y. (2024). Model multifactor analysis of soil heavy metal pollution on plant germination in Southeast Chengdu, China: Based on redundancy analysis, factor detector, and XGBoost-SHAP. *Science of the Total Environment*, 954, article number 176605. doi: [10.1016/j.scitotenv.2024.176605](https://doi.org/10.1016/j.scitotenv.2024.176605).
- [19] Rather, B.A., Mir, I.R., Masood, A., Anjum, N.A., & Khan, N.A. (2022). Ethylene-nitrogen synergism induces tolerance to copper stress by modulating antioxidant system and nitrogen metabolism and improves photosynthetic capacity in mustard. *Environmental Science and Pollution Research*, 29, 49029-49049. doi: [10.1007/s11356-022-19380-y](https://doi.org/10.1007/s11356-022-19380-y).

- [20] Rezayian, M., & Zarinkamar, F. (2023). Nitric oxide, calmodulin and calcium protein kinase interactions in the response of *Brassica napus* to salinity stress. *Plant Biology*, 25(3), 411-419. [doi: 10.1111/plb.13511](https://doi.org/10.1111/plb.13511).
- [21] Sadr, S., Namdjoyan, S., & Mohammadian, T. (2024). Interactive effect of exogenous melatonin and calcium for alleviation of zinc toxicity in rapeseed (*Brassica napus* L.) seedlings by modulating cellular redox status. *Journal of Soil Science and Plant Nutrition*, 24, 4017-4032. [doi: 10.1007/s42729-024-01827-5](https://doi.org/10.1007/s42729-024-01827-5).
- [22] Shen, N., Hou, S., Tu, G., Lan, W., & Jing, Y. (2021). Transcription factor WRKY33 mediates the phosphate deficiency-induced remodeling of root architecture by modulating iron homeostasis in *Arabidopsis* roots. *International Journal of Molecular Sciences*, 22(17), article number 9275. [doi: 10.3390/ijms22179275](https://doi.org/10.3390/ijms22179275).
- [23] Singh, P.K., Yadav, J.S., Kumar, I., Kumar, U., & Sharma, R.K. (2024). Screening of mustard cultivars for phytoremediation of heavy metals contamination in wastewater irrigated soil systems. *Environmental Monitoring and Assessment*, 196, article number 321. [doi: 10.1007/s10661-024-12506-4](https://doi.org/10.1007/s10661-024-12506-4).
- [24] Tan, Z., Wu, C., Xuan, Z., Cheng, Y., Xiong, R., Su, Z., & Wang, D. (2022). Lead exposure dose-dependently affects oxidative stress, AsA-GSH, photosynthesis, and mineral content in pakchoi (*Brassica chinensis* L.). *Frontiers in Plant Science*, 13, article number 1007276. [doi: 10.3389/fpls.2022.1007276](https://doi.org/10.3389/fpls.2022.1007276).
- [25] Vasilache, N., Diacu, E., Cananau, S., Tenea, A.G., & Vasile, G.G. (2023). Evaluation of the phytoremediation potential of the *Sinapis alba* plant using extractable metal concentrations. *Plants*, 12(17), article number 3123. [doi: 10.3390/plants12173123](https://doi.org/10.3390/plants12173123).
- [26] Vasile, G.-G., Tenea, A.-G., Dinu, C., Iordache, A.M.M., Gheorghe, S., Mureseanu, M., & Pascu, L.F. (2021). Bioavailability, accumulation and distribution of toxic metals (As, Cd, Ni and Pb) and their impact on *Sinapis alba* plant nutrient metabolism. *International Journal of Environmental Research and Public Health*, 18(24), article number 12947. [doi: 10.3390/ijerph182412947](https://doi.org/10.3390/ijerph182412947).
- [27] Xue, C., Li, W., Shen, R., & Lan, P. (2023). Impacts of iron on phosphate starvation-induced root hair growth in *Arabidopsis*. *Plant, Cell & Environment*, 46(1), 215-238. [doi: 10.1111/pce.14451](https://doi.org/10.1111/pce.14451).
- [28] Zheng, X., Han, G., Song, Z., Liang, B., Yang, X., Yu, C., & Guan, D.-X. (2024). Biogeochemical cycle and isotope fractionation of copper in plant-soil systems: A review. *Reviews in Environmental Science and Bio/Technology*, 23, 21-41. [doi: 10.1007/s11157-024-09681-8](https://doi.org/10.1007/s11157-024-09681-8).



Вплив іонів важких металів на ранній розвиток *Sinapis alba* L.

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Анотація. Забруднення ґрунтів іонами важких металів може обмежувати використання земель і знижувати ефективність фітореMediaції. *Sinapis alba* L. розглядають як перспективну тест-культуру та потенційний фітореMediaнт, однак її реакція на різні метали в ґрунтовій системі потребує уточнення. Метою роботи було оцінити вплив іонів Zn^{2+} , Cu^{2+} , Pb^{2+} і Fe^{3+} на життєздатність насіння, довжину пагонів і коренів *S. alba*. Дослід проводили в модельному ґрунтовому експерименті з трьома повтореннями для кожного варіанта. На 5-ту добу значуще зниження життєздатності виявлено лише за дії Cu^{2+} у концентрації 300 мг/дм³, де цей показник становив $77,9 \pm 1,2$ %. На 10-ту добу найсильніший ефект спостерігали для Fe^{3+} : життєздатність знижувалася до $34,9 \pm 1,2$ %, $27,7 \pm 1,2$ % і $20,5 \pm 1,2$ % за концентрацій 600, 800 і 1000 мг/дм³ відповідно. За дії Cu^{2+} цей показник становив 74,7-56,6 %, Zn^{2+} – 80,7-67,5 %, а Pb^{2+} – 81,9 % за найвищої концентрації. Довжина пагонів і коренів зменшувалася за дії всіх досліджених металів. Найбільше пригнічення росту спричиняв Fe^{3+} : довжина пагонів зменшувалася з 54,62 до 9,94 мм, а коренів – з 35,96 до 13,09 мм. Отже, морфометричні показники виявилися чутливішими до металевого стресу, ніж життєздатність, і мають використовуватися для комплексного оцінювання фітотоксичності ґрунтів. Результати можуть бути використані фахівцями у сфері екологічного моніторингу, ґрунтознавства та фітореMediaції для комплексної оцінки токсичності ґрунтів, забруднених важкими металами

Ключові слова: фітотоксичність; біотестування фітотоксичності; сірі лісові ґрунти; фітореMediaція; метал-специфічний стрес

