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THE PRESENCE OF HEAVY METALS IN THE WHEAT STEM AND INFLUENCE ON THE GROWTH AND HEIGHT OF THE STEM

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Abstract: One of the ways in which heavy metals get into food products is through polluted plants and animals used for their production. Thus, if plants are grown on soil with a high content of heavy metals then a part of these metals will reach in their organism. Current agricultural practices include the widespread use of chemical pesticides and mineral fertilizers known for their ability to cause negative impacts on human health and degrade the natural environment. Plants play an important role in the circulation of heavy metals in nature. Heavy metals for the most part they enter the food chain through plants. The experiment was set up in vessels where two different concentrations of heavy metal mixture 100 ppm and 250 ppm were added under controlled conditions. Higher concentrations of heavy metal mixture had negative effect on wheat stem hight and plant growth was reduced. Higher concentrations of heavy metals significantly reduced plant growth in both cultivars. The aim of the research was to determine the content and influence of heavy metals on tree development in the varieties Pobeda and Ljiljana. The vegetation trial was set up in three repetitions, with a total of 36 pots in which two varieties of wheat, Pobeda and Ljiljana, were sown. Before sowing, the vessels were filled with 2 kg of Novobalt dry extract, which was subsequently contaminated with a mixture of chemical compounds of heavy metals in the form of a solution of the following compounds, namely: zinc in the form of zinc-acetate-$Zn(CH_3COO)_2$ x $2H_2O$, lead in the form of lead-acetate $C_4H_6O_4Pb$ x $3H_2O$, chromium in the form of chromium-trioxide-CrO_3, copper in the form of copper-sulfate-CuSO4 and cadmium in the form of cadmium-nitrate-$Cd(NO_3)2$ x $4H_2O$.

Keywords: Atomic absorption spectrophotometry, Heavy metals, Wheat steam, Triticum sp, Plant war

INTRODUCTION

In soil and sediment, heavy metals exist in various forms, such as: dissolved ions (e.g., Cu^{2+}, Cd^{2+}, $CrO4^{2-}$, $Cr_2O_7^{2-}$ and MnO_4^{2-}), organic complexes (e.g., Cu^{2+}, Pb^{2+}, and Hg^{2+} bindingto dissolved organic matter), exchangeable ions (e.g., Cu^{2+}, Zn^{2+}, Cd^{2+}, Ni^{2+}, and Pb^{2+}) adsorbed onto soil solid particles, and those (co-) precipitated as part of soil particles (e.g., $Cd_3(PO_4)^2$, ZnS, $PbCO_3$, and $HgSO_4$). These three forms maintain thermodynamic equilibrium in activities and mutual concentration with insoluble precipitate as the dominant species (Liu et al.,2019).

Copper is one of the essential heavy metals that is not toxic in its nature metallic form, but its salt can be toxic. They are particularly toxic sulfate salts. Copper sulfate is a crystalline salt that has a metallic taste and a blue color. In a larger dose of 0.5 g, it acts as an irritant poison and causes intestinal and stomach problems (Ali et al., 2023).

Chromium is an important micronutrient that is involved in carbohydrate metabolism lipids and proteins. It is an important metal in the production of pigments, coatings metals, woodworking and leather tanning, and is used in metallurgy and production of refractory materials, among various other applications. The most important forms of chromium oxidation are trivalent and hexavalent chromium. Trivalent chromium is largely non-toxic, and hexavalent chromium is toxic as well as carcinogenic to living organisms. Industrial waste containing chromium is known as the main source of pollution and can have a serious impact on soil and water on human health (Shin et al., 2023).

Cadmium and lead are heavy metals that, when taken into the body in the amounts more than the maximum permitted, cause various organ damage and malignant changes. They have a negative effect on blood flow, central nervous system, cardiovascular and immune system, and the kidneys. Heavy metals are a natural part of the Earth's crust. So contamination with cadmium and lead can come from an environmental source, during packaging or food processing, but also from the packaging in which it is packed (Lalić, 2024).

Differences in stress-induced effects of elevated concentrations of toxic metals indicate differenttolerances of plants to metals. Based on the results of depth variation of heavy metal concentration,it is concluded that heavy metal concentrations present in solid waste samples depend on the site conditions, as well as on the conditions prevailing at the sampling sites. Based on the average concentration, the concentration of heavy metals in the collected solid waste sample was determined in the following order: Mn > Cu > Pb > Cd (Zhao et al., 2017).

Contamination alongside roads was evident through significant negative correlations between metal concentrations in soil and plants samples and distance from the road edge. Only cadmium concentrations in soil and plant samples were not associated with roadside pollution. Pollution of plants and soil alongside roads did not extend muchbeyond 20 meters from the road (Kanmani and Gandhimathi, 2013).

Research initiated in the 1960s has shown that soil in urban and industrial areas contains extremely high concentrations of heavy metals. Plants are most sensitive to industrial pollution, which is whythey are successfully used as indicators to assess environmental pollution and establish adequate biomonitoring. The aim of this study is to determine the content of heavy metals in the leaves of Plantain (Plantago major L.) in the Tuzla area, as well as to assess the influence of thermal power plants and industrial facilities on surrounding areas. It was found that the content of heavy metalsin Plantain leaves often did not decrease with increasing distance from the dominant anthropogenicsources of heavy metal emissions (Bektić et al., 2020).

The bioconcentration factor (FB) is the concentration of heavy metals that are present in plant biomass divided by the concentration of the corresponding weight metal in the soil on which the appropriate plants are grown (Maiti et al., 2022).

Studies have shown that it is not total concentration but the reactive fraction of heavy metals inthe soil that dictates their toxicity to plants, microbes, and human beings. Exchangeable forms of heavy metals that are soluble in water are much more reactive and biologically available than precipitated species (Kim et al., 2015). However, the distribution of heavy metal species is influenced by numerous environmental factors, especially pH, redox potential, clay content, Fe/Mnoxide content, organic matter content, and the presence of other cations and anions in the solid phase (Pietrzykowski et al., 2014). To accurately assess the bioavailability of heavy metals in the soil, factors of influence must be taken into account.

MATERIALS AND METHODS

The study on the influence of heavy metal mixture concentration on the dynamics of heavy metal accumulation in wheat plants and plant growth was conducted through experiments in pots in the greenhouse of the Faculty of Agriculture in Zemun, where controlled conditions of temperature and humidity were maintained. The vegetative trial was arranged in three repetitions, with a total of 36 pots in which two varieties of wheat, *Pobeda* and *Ljiljana*, were sown. Before sowing, the pots were filled with 2 kg of dry extract of Novobalt, which was subsequently contaminated with a mixture of heavy metal chemical compounds in the form of solutions of the following compounds: zinc in the form of zinc acetate-$Zn(CH_3COO)_2$ x $2H_2O$, lead in the form of lead acetate $C_4H_6O_4Pb$ x $3H_2O$, chromium in the form of chromium trioxide-CrO_3, copper in the formof copper sulfate-$CuSO_4$, and cadmium in the form of cadmium nitrate-$Cd(NO_3)_2$ x $4H_2O$.

The following concentrations of solutions were applied: 0 ppm (control), 100 ppm, 250 ppm. In addition to the heavy metal content in wheat plants, the height of the plants from the soil level was first measured. Afterward, the heavy metal content in wheat plants was determined. The *Pobeda* and *Ljiljana* wheat varieties were chosen because they are predominantly grown in the southern Banat region. Twelve seeds were sown in each pot at a depth of 5 cm. Samples from the trial in pots were analyzed during the tillering and heading phases, where four plants were taken from each pot. After sampling, the roots were manually separated from the wheat stem. Subsequently, the plant mass-stems were washed with distilled water and kept for several hours in 0.1 M HCl to remove soil and mineral oxides from the surface. Then, the plant mass was ground and dried in a dryer at 80˚C. A 1 g sample was taken and poured with 20 ml of 60% HNO_3. Mild boiling was performed for 2 hours. After cooling, 3 ml of H_2O_2 was added, followed by boiling for 15 minutes. The peroxide procedure was repeated. After cooling, 2 ml of $HClO_4$ was added, and mild evaporation was performed until dense white fumes of perchloric acid appeared (Jones and Case, 1990). After cooling, 5 ml of 5M HCl was added, and then the samples were quantitatively transferred into normal 50 ml vessels. The vessels were filled up to the final volumewith distilled water. The solution was filtered through quantitative filter paper. Readings were performed using atomic absorption spectrophotometry (Varian Specter AA 220FS instrument) in acetylene/air flame. The analysis of the data obtained was conducted using the statistical softwarepackages STATISTICA 8 for Windows and SPSS Statistics 17.0.

RESULTS AND DISCUSSION

Plant Height

The variety *Pobeda* had a slightly lower average height (38.70 cm), while the variety *Ljiljana* had a higher average height (44.50 cm). The average height of the tree per sample for the variety *Pobeda* varied from 34.00 to 42.60, and for the variety *Ljiljana* from 35.50 to 50.00 (Table 1, 2).

Table 1. Plant Height, *Pobeda* Variety (cm)

Treatment	Zn, Pb, Cr, Cu, Cd	LSD 5%	LSD 1%
Control	42.60	7.9889	14.6647
100 ppm	39.50	11.4729	21.06
250 ppm	34.00	1.8371	3.3723
Average	38.70	-	-

Table 2. Plant Height, *Ljiljana* Variety (cm)

Treatment	Zn, Pb, Cr, Cu, Cd	LSD 5%	LSD 1%
Control	50.00	1.8371	3.3723
100 ppm	48.00	1.8371	26.7668
250 ppm	35.50	5.1143	9.3881
Average	44.50	-	-

During metal mixture contamination at a concentration of 100 ppm, the *Ljiljana* variety had a plant height of 48.00 cm, while the *Pobeda* variety had a slightly lower height of 39.50 cm. At a concentration of 250 ppm, the plant height for the *Ljiljana* variety was 35.50 cm, while for the *Pobeda* variety, it was lower at 34.00 cm.

The process of photosynthesis is most sensitive to the toxic effect of lead. Long-term exposure causes inhibition of chlorophyll biosynthesis (Ernst et al., 2000; He et al., 2014). Studies have shown that lead negatively affects photosynthesis, transpiration, and stomatal conductivity when its concentration in the soil exceeds 300 ppm (Fu and Wang, 2015). Differences in the effects caused by stress due to elevated concentrations of toxic metals indicate varying plant tolerance tometals (Zhao et al., 2017).

At the cellular level, the consequences of prolonged exposure to high metal concentrations can be membrane disintegration, ion loss, lipid peroxidation, DNA/RNA degradation and finally cell death. In order for plants to develop and grow normally, they must maintain concentrations of essential elements within optimal values- a state of homeostasis (Stojanović, 2017).

Accumulation of lead in the bodies of plants and animals occurs in areas where the air, water and soil are contaminated with this chemical element. The insecticide lead arsenate, which is applied in orchards to control insects, also contributes to soil lead contamination (Premović et al., 2024).

Under the influence of higher concentrations of lead in plants, main processes, such as germination, growth and development, photosynthetic process, wather intake, mineral diet and enzymatic activity (Agami and Mohamed, 2013; He et al., 2016; Sharma and Dubey, 2005) are inhibited.

Cadmium is considered one of the most toxic elements. Although it cannot be found in nature in free form, it is still present in nature in zinc ores, but it also comes from anthropogenic sources. It is thought to be radioactive in small amounts. All cadmium compounds are poisonous, and cadmium oxide is the most poisonous. Apart from the mentioned compound, there are other known compounds of cadmium, such as: cadmium sulphide, i.e. griconite, which is the most well-known cadmium mineral, cadmium chloride, cadmium hydroxide and cadmium telluride (Turek, 2024).

Lead is present in many vegetable and animal foods in a concentration of 0.5 to 1.0 mg/kg. Lead is predominantly present in higher concentrations in fish (up to 2.0 mg/kg), molluscs and crustaceans (up to 10 mg/kg). An increase in the content of lead is mainly detected in foods that are packed in tin packaging with a lid with solder, which contains a certain content of lead. The quality of the soldering is sometimes poor, which can lead to the disintegration of those parts of the packaging, that is, to the contamination of food with lead. In very rare cases (up to 2%), it happens that in canned food stored in a container (especially during long-term storage), up to 3 mg/kg of lead accumulates, which in this case can represent a significant danger to human health. It is known that lead slows down the cognitive and intellectual development of children, and that in adults it causes an increase in blood pressure and the appearance of cardiovascular diseases. Changes in the nervous system are manifested by headache, dizziness, increased fatigue, irritability, sleep disorders, memory disorders, disorders in muscle activity and function, increased sweating, etc. Lead introduced into the human body through contaminated food can replace calcium in bones and thus become a source of permanent poisoning (Haydarov et al., 2023).

The Content of Heavy Metals in the Stem of Wheat

The content of heavy metals in the *Pobeda* and *Ljiljana* varieties varies significantly (Table, 3, 4). According to their average content in wheat plants, the heavy metals investigated in thePobeda variety can be ranked in the following order:

- zinc > cadmium > copper > lead > chromium

The lead content of the *Pobeda* variety in the wheat stem is on average 3.77 mg kg^{-1} and varies from 2.46 mg kg^{-1} (control) to 5.45 mg kg^{-1} (variant with 250 ppm). The cadmium content of the *Pobeda* variety in the wheat stem is on average 6.07 mg kg^{-1} and varies from 0.65 mg kg^{-1} (control) to 9.12 mg kg^{-1} (variant with 250 ppm).

According to their average content in the wheat stem of the *Ljiljana* variety, the heavy metals investigated can be arranged in the following order:

- zinc > cadmium > copper > lead > chromium

The lead content of the *Ljiljana* variety in the wheat stalk is on average 3.08 mg kg^{-1} and varies from 2.14 mg kg^{-1} (control) to 4.33 mg kg^{-1} (variant with 250 ppm). The cadmium content of the *Ljiljana* variety in the wheat stem is on average 10.39 mg kg^{-1} and varies from 0.39 mg kg^{-1} (control) to 17.31 mg kg^{-1} (variant with 250 ppm).

Table 3. Content of Heavy Metals in Wheat Plants, Pobeda Variety (mg kg^{-1})

Treatment	Zn	Pb	Cr	Cu	Cd	Ftest	LSD 5%	LSD 1%
Control	19.39	2.46	1.23	4.49	0.65	38832.27**	0.1197	0.1655
100 ppm	39.46	3.40	0.99	5.26	8.45	1094.38**	0.4569	0.6318
250 ppm	49.10	5.45	1.75	5.54	9.12	49105.52**	0.2678	0.3703
Average	35.98	3.77	1.32	5.10	6.07	-	-	-

Table 4. Content of Heavy Metals in Wheat Plants, *Liljana Variety* (mg kg^{-1})

Treatment	Zn	Pb	Cr	Cu	Cd	Ftest	LSD 5%	LSD 1%
Control	29.99	2.14	0.58	4.65	0.39	38832.27**	0.1197	0.1655
100 ppm	77.67	2.76	0.58	5.36	13.46	62999.44**	0.3917	0.5417
250 ppm	79.13	4.33	1.23	5.41	17.31	58501.43**	0.4087	0.5652
Average	62.26	3.08	0.80	5.14	10.39	-	-	-

CONCLUSION

The vegetative trial in pots was conducted with different concentrations of heavy metal mixtures.Increasing the concentration of metal mixtures had a negative impact on plant height. Higher concentrations of heavy metals significantly reduced plant growth in both varieties. The results of the statistical analysis (analysis of variance) show that the experimental factor - the concentration of heavy metal mixtures for soil contamination- had a very significant impact on the content of heavy metals in wheat plant parts, as evidenced by the calculated F-values.

Based on the results of the research on uptake of heavy metals, it can be recommended to grow the Pobeda variety, considering that it has a significantly lower uptake of heavy metals - Zn, Cu and Cd.

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