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# ACCUMULATION OF HEAVY METALS IN THE VEGETATIVE ORGANS OF POPLARS UNDER THEIR JOINT INTRODUCTION TO THE SOIL

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**Background**. Studies of plant autecology under excessive heavy metal intake require a thorough understanding of the specifics of the uptake and accumulation of these substances by plants. It is therefore important to study the redistribution of heavy metals in the soil–plant system. Poplars, due to their rapid growth, large assimilative organ surface area, etc., are a suitable object for studying their autecological properties and potential use in the creation of green spaces.

**Materials and Methods**. The objects of the study were two-year-old poplar seedlings: 'I-45/51', 'Lvivska', 'Robusta', 'Tronco', and the soils on which they grow. The plants were watered once a week for 2 months with a mixture of heavy metal compounds at 1, 5, and 10 maximum permissible concentrations (MPC). Mixtures of the following salts were used as heavy metal sources CdSO<sub>4</sub>, Ni(NO<sub>3</sub>)<sub>2</sub>, CuSO<sub>4</sub>, ZnSO<sub>4</sub>, Pb(CH<sub>3</sub>COO)<sub>2</sub>. The control plants were not exposed to heavy metals. All plants were grown under natural light and temperature. The content of mobile forms of Cu, Zn, Ni, Pb, and Cd in ammonium acetic acid extract from soil was determined and their accumulation in leaves and roots was measured by conventional methods using a C-115PK atomic absorption spectrophotometer Selmi (Ukraine). Statistical processing of the results was carried out using Microsoft Excel 2013. To assess the probability of the difference between the statistical characteristics of two alternative data sets, a univariate analysis of variance using IBM SPSS Statistics 27, ANOVA, was performed.

**Results.** The accumulation and translocation of heavy metals in the soil–plant system was studied in model experiments. The study revealed the peculiarities of heavy metal accumulation in the soils where poplars grew. It was shown that the introduction



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of heavy metal compounds leads to an increase in the content of mobile forms of the studied elements in the soil. Under the influence of 1, 5, and 10 MPC of heavy metal ions, all cultivars accumulated them more actively in the root system than in the leaves.

**Conclusions**. The studied poplars can be divided into two groups. The first one includes the cultivars 'I-45/51' and 'Lvivska' with a high level of heavy metal accumulation and the highest enrichment factor. The second group – 'Tronco' and 'Robusta' – includes those with low levels of heavy metal accumulation and low enrichment factors. The results suggest that the poplars of the second group have physiological mechanisms that determine the observed peculiarities of heavy metal translocation.

**Keywords:** cultivars of poplars, heavy metals, soil–plant system, translocation, enrichment factor

# INTRODUCTION

Recently, a number of factors, including full-scale military operations in Ukraine, have led to a rapid and sometimes irreversible deterioration of the environmental situation in the country. In the Prydniprovya Steppe, long-term extraction and processing of natural resources has created an imbalance between the growth rate of man-made environmental impacts and the inadequate level of environmental protection. This eventually turned the industrial centres of the region into an environmental disaster zone (Gryshko *et al.*, 2012; Danylchuk *et al.*, 2023; Masiuk *et al.*, 2023; Savosko *et al.*, 2022). Industrial pollutants, including heavy metals, are a new anthropogenic environmental factor that has not previously been involved in the phylogenetic adaptation of plants (Bessonova & Grytsay, 2018; Michopoulos, 2021).

The study of the peculiarities of plant autecology under excessive heavy metal loading in urbanised ecosystems is impossible without clarifying the specifics of their uptake and accumulation by plants (Barman *et al.*, 2000; Gupta *et al.*, 2008). It is therefore important to study not only the peculiarities of accumulation of different heavy metal compounds in soils, but also their redistribution in the soil—plant system (He *et al.*, 2013; Bi *et al.*, 2006). In this context, many scientists insist on considering the soil—plant system as a system whose components are interconnected by flows of chemical elements (Singh *et al.*, 2010). In this case, the fact that heavy metals accumulate in the soil is not as important as the effects they have on living organisms. Thus, without analysing the processes of heavy metal redistribution in the elements of the soil—plant system, it is impossible to determine either the functional characteristics of plant organisms under conditions of environmental pollution by heavy metals or the possibility of using the latter to develop biological systems for extracting excess elements from the soil (Agbemafle *et al.*, 2019; Pant *et al.*, 2023).

Poplars possess numerous properties, such as rapid growth, large areas of the assimilative organ, wide distribution area, etc., that make them the most suitable object for determining the autecological features of the genus, and consequently are widely used in agroforestry. These fast-growing woody species have the potential to enhance ecosystem services including the storage of carbon, and the conservation and remediation of soil and water. Industrial plantations of such trees in Germany alone amount to more than 1.6 million hectares and their wood is used in the bioenergy and woodworking industries (Mirko & Volker, 2024). Therefore, the demand for fast-growing tree

species is rising in global markets, prompting government agencies and landowners to create and search for the most cost-effective technologies for growing improved cultivars (Dhiman et al., 2024; Kort, & Schroeder, 2020). For example, in Ukraine, Western Europe, and Asia, heritability in a broad sense as well as the genotype-by-environment interactions are assessed for such traits as growth and development of physiological processes that contribute to the accumulation of biomass and the corresponding phenological stages of plant development in different climatic zones. Such research makes it possible to determine the limits of adaptation of the cultivars to certain climatic features of the regions and develop optimal schemes for their diversification in agroforestry. (Adler et al., 2021; Kutsokon et al., 2022; Langhof, & Schmiedgen, 2023; Thevs et al., 2021; Zadorozhnaya et al., 2018). Many works in this field pay great attention to the influence of environmental factors on the plant organism, on which the flow of elements largely depends (Kabata-Pendias, 2010; Mareri et al., 2022). In order to exclude the influence of other factors on the processes of accumulation and migration of heavy elements in the soil-plant system, model experiments were carried out in which different poplar cultivars growing on ordinary chernozem were exposed to certain amounts of heavy metal compounds.

# **MATERIAL AND METHODS**

The research objects were two-year-old seedlings of 4 poplar cultivars that are actively used in landscaping: 'I-45/51', 'Lvivska', 'Robusta', and 'Tronco', as well as the soils on which they grow. The seedlings were grown from lignified cuttings obtained from mother plants of the Lubny Branch of the Ukrainian Research Institute of Forestry and Forest Melioration named after G. M. Vysotsky. The two-year-old plants were planted in plastic containers with ordinary chernozem containing Cu - 6.8; Zn - 16.3; Ni - 21.2; Pb - 3.9 and Cd - 1.53 mg/kg of soil.

Research methods. The peculiarities of heavy metal accumulation in poplars were studied in vegetation experiments. For each variant of the experiments, 10 specimens of two-year-old seedlings of four poplar cultivars were planted in 5 L containers with ordinary chernozems in March and grown under natural light and temperature conditions; soil moisture was maintained at 60% throughout the experiment. In June-July, plants in the mature leaf phase were watered once a week with a solution of a mixture of heavy metal compounds in the amount of 1, 5, and 10 of maximum permissible concentrations (MPC). Mixtures of salts such as CdSO<sub>4</sub>, Ni(NO<sub>3</sub>)<sub>2</sub>, CuSO<sub>4</sub>, ZnSO<sub>4</sub>, Pb(CH<sub>3</sub>COO), were used as sources of heavy metals. The MPC for Cu was considered 3; for Zn - 23; Ni - 4; Pb - 32 and Cd - 3 mg/kg soil (Cabinet of Ministers, 2021). Two-year-old poplar seedlings that were not exposed to heavy metals were used as the control. Two weeks after the last application of the solutions of heavy metal compounds, soil and plant material was collected for further analysis. Soil samples were taken from the root layer according to conventional soil science methods (ISO, 2018). The content of the heavy metals was determined using a C-115PK atomic absorption spectrophotometer Selmi (Ukraine). The amount of mobile forms of Cu, Zn, Ni, Pb, and Cd in the ammonium-acetic acid extract (pH = 4.8) in the soil was measured using conventional methods (Kochmar & Karabyn, 2022). The content of heavy metals in plant material was determined following the guidelines for the determination of heavy metals in plant products (Danylchuk et al., 2023; Hrytsaenko et al., 2003). The plant

material was mineralised by dry ashing (at a temperature of +525 °C) and subsequent extraction with HNO<sub>3</sub>. Calculations of the enrichment factor values for active forms of heavy metals in the soil ( $\text{EF}^{\text{s}}_{a}$ ), roots ( $\text{EFP}^{\text{p}}_{r}$ ), and leaves ( $\text{EFP}^{\text{p}}_{l}$ ) of the poplars were performed according to Kisku *et al.*, (2000):

EF = concentration of the element in the soil or plant material under heavy metal application concentration of the element in the soil or plant material in the control

Statistical processing of the results. Standard statistical methods were used to process the results (Welham *et al.*, 2014). To compare groups of data, the reliability of the results was assessed using the Student's *t*-test; differences were considered significant at p <0.05 (Welham *et al.*, 2014). Statistical processing of the results was carried out using Microsoft Excel 2013. The main statistical indicators were calculated based on direct quantitative data obtained as a result of research (mean; standard error of the mean). To assess the probability of the difference between the statistical characteristics of the two alternative data sets, a univariate analysis of variance using IBM SPSS Statistics 27, ANOVA was performed. A difference with a probability of p ≥0.95, calculated using IBM SPSS Statistics 27 software with post-hoc analysis, was considered significant.

# **RESULTS AND DISCUSSION**

Due to the high buffering capacity of ordinary chernozem, a significant proportion of the pollutant elements that enter it can be converted into forms that are not readily available to plants. It is therefore advisable to determine the level of toxicity of soils contaminated with heavy metals by the amount of their mobile forms.

Our results show that the introduction of a complex of heavy metal compounds leads to an increase in the content of mobile forms of all the elements studied. Thus, at the maximum permissible concentration, a greater amount of Zn ions available to plants was observed under the 'Robusta' and 'Tronco' cultivars, amounting to 54.3 and 53.3 mg/kg soil, respectively (**Table 1**).

The content of Zn in the soil under the 'Lvivska' and 'I-45/51' cultivars was 25–40 % lower than under 'Robusta' and 'Tronco' when heavy metals were introduced at concentrations of 5 and 10 MPC. The obtained data are in good agreement with the values of the  $EF_a^s$ , which under the influence of 10 MPC were 1.5 times higher in the first group than in the second ('Robusta' and 'Tronco') (**Fig. 1**). A similar pattern was observed for Pb.

Our study has shown that under the influence of the maximum concentration of heavy metal ions the content of mobile Cu forms in the soils under the cultivars 'Tronco' and 'Robusta' was significantly higher than in the soils under 'Lvivska' and 'I-45/51', which indicates the specificity of the processes of the absorption of this element by these cultivars. Thus, in the second group of cultivars, the EFs avalue ranged from 149 to 202, while in the first group, it was up to 89 (**Fig. 1**). This indicates that the cultivars 'Tronco' and 'Robusta' are on average 2 times less active in removing Cu from the soil than 'Lvivska' and 'I-45/51'. The results obtained demonstrate similar values of EFs for Cu and Cd, which is in good agreement with the average long-term data for other plants in Palu Grand Forest Park (Palu, Indonesia). The values of the transfer factor (TF = Plant Concentration/Soil Concentration) were identical for Cd and Cu – 0.17 and 0.17 (Ramlan *et al.*, 2022).

Table 1. The content of heavy metals (ammonium acetic acid extract) in soil, mg/kg

Variant	Cu	Zn	Ni	Pb	Cd		
'I–45/51'							
Control	2.3±0.5 <sup>vvxx</sup>	6.2±0.4 <sup>vvvxxx</sup>	6.2±0.6 <sup>vvvxxx</sup>	2.1±0.2 <sup>vvvxxx</sup>	0.4±0.1 <sup>vvxxx</sup>		
1 MPC	94.1±2.6*	32.5±2.8*vvx	12.5±0.6*	46.3±2.7*vv	3.0±0.2*vvvxxx		
5 MPC	135.2±6.3*vvvxxx	121.6±6.4*vx	28.9±1.0*vvvxx	180.7±8.9*vvvxx	23.4±1.4*xx		
10 MPC	334.1±19.8*vvxx	167.7±2.7* <sup>vx</sup>	70.3±4.9* <sup>vv</sup>	309.0±12.6*vvx	45.4±4.6*xx		
		'Lviv	ʻska'				
Control	2.0±0.3 <sup>vvxx</sup>	8.3±0.6 <sup>vvvxxx</sup>	5.7±0.6 vvvxxx	2.5±0.3 <sup>vvvxxx</sup>	0.2±0.1 <sup>vvvxxx</sup>		
1 MPC	92.0±1.8*	32.1±2.0*vvx	11.3±0.5*vx	32.7±0.8*****	3.2±0.2*vvvxxx		
5 MPC	159.8±2.3*vvxx	92.2±2.1*vvvxxx	27.8±1.1*vvvxx	197.4±8.8*vvvx	24.8±1.9*xx		
10 MPC	393.2±11.6* <sup>v</sup>	163.7±6.3*vx	69.7±3.9*vv	336.3±10.8*vvx	46.4±2.0*xx		
	'Robusta'						
Control	5.9±0.5	16.0±0.5	14.2±0.6	6.0±0.5	1.0±0.1		
1 MPC	104.6±2.1*	54.3±4.4*	17.0±1.7	65.5±3.5*	6.0±0.4*		
5 MPC	234.5±10.1*	163.3±8.1*	41.9±1.1*	281.3±5.6*	31.5±2.1*		
10 MPC	514.1±30.0*	224.9±9.1*	97.5±3.0*	481.3±27.7*	60.2±0.7*		
'Tronco'							
Control	5.6±0.6	14.7±0.4	13.0±1.1	5.6±0.2	1.1±0.1		
1 MPC	108.2±6.9*	53.3±3.6*	16.4±0.6*	52.7±3.3*	6.7±0.2*		
5 MPC	227.6±13.9*	159.3±8.4*	38.0±2.0*	246.5±12.5*	37.6±1.7*		
10 MPC	492.8±29.9*	223.1±16.3*	86.1±3.0*	443.6±26.0*	72.8±4.1*		

Notes: \* – the difference is significant relative to the conditional control, p ≤0.05. Letter v indicates the significant levels of differences between variants compared with 'Robusta'; and x – with 'Tronco' (v, x: p ≤0.05; vv, xx: p ≤0.01; vvv, xxx: p ≤0.001)

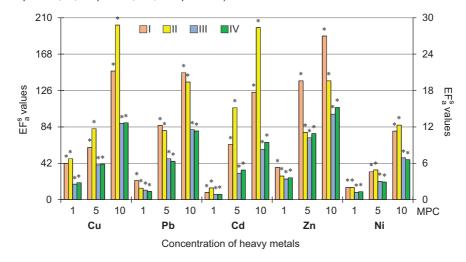


Fig. 1. The EFs a values in he studied soils under the combined effect of heavy metals: I - 'I-45/51', II - 'Lvivska', III - 'Robusta', and IV - 'Tronco'; left scale for (Cu, Pb, and Cd); right scale for (Zn and Ni); \* - the difference is significant relative to the conditional control, p ≤0.05

The data presented in **Table 1** show that under the maximum permissible concentrations of heavy metals in the soils in which 'Lvivska' and 'I-45/51' cultivars grew, there was a statistically significant increase in mobile Ni compounds by 12.2 and 11.3 times, respectively, compared to the control. A univariate analysis of variance was conducted to assess the probable difference between the statistical characteristics of the two alternative datasets (cultivar – heavy metal concentration in soil). The results demonstrated that for 'Robusta' and 'Tronco,' there is no statistically significant difference between the alternative datasets at p  $\leq$ 0.05 (**Tabl. 1**). In contrast, for 'Lvivska' and 'I-45/51', in comparison to 'Robusta' and 'Tronco', a significant difference was observed for the majority of elements at p  $\leq$ 0.001–0.008 (**Tabl. 2**).

Table 2. Results of univariate analysis of variance between the content of mobile forms of heavy metals in soils used for growing different groups of cultivars ('Lvivska', 'I-45/51 and 'Robusta', 'Tronco')

Indicators	Cu	Zn	Ni	Pb	Cd	
Control						
$R^2$	13.050	68.236	59.325	12.268	0.599	
F	17.576	112.476	35.190	40.556	29.944	
р	<0.001	<0.001	<0.001	<0.001	<0.001	
1 MPC						
R <sup>2</sup>	186.908	461.467	24.003	559.188	10.902	
F	4.048	14.171	8.306	23.852	60.568	
р	0.051	0.001	0.008	<0.001	<0.001	
5 MPC						
R <sup>2</sup>	7306.988	3403.168	143.456	6354.609	128.146	
F	28.623	24.881	24.819	24.687	13.010	
р	<0.001	<0.001	<0.001	<0.001	0.002	
10 MPC						
R <sup>2</sup>	21517.290	3408.510	540.350	20627.971	505.018	
F	12.343	11.495	12.465	15.990	16.237	
р	0.002	0.003	0.002	<0.001	<0.001	

At the same time, the  $\mathrm{EF_{a}^{s}}$  for Ni ions in the soil under the 'Tronco' and 'Robusta' cultivars was 1.7 times lower, indicating a less active absorption capacity of the latter group of plants. For Cd, this indicator was on average 2.4 times lower than in the control.

Studies on the degree of resistance and adaptive responses of plants to heavy metals and their redistribution in the soil–plant system would be impossible without clarifying the peculiarities of their further accumulation in plant organs. In poplar cultivars, a certain specificity in the uptake of metal ions by the roots has been observed. For example, Cu is most actively accumulated in the root system of cultivars 'I-45/51' and 'Lvivska'. Under the influence of a mixture of heavy metals at a concentration of 10 MPC, the underground organ of this group of poplars accumulates on average 2 times more Cu than the roots of 'Robusta' and 'Tronco' (**Table 3**).

Table 3. The content of heavy metals in the roots of poplar cultivars, mg/kg dry matter

Variant	Cu	Zn	Ni	Pb	Cd			
'I–45/51'								
Control	27.9±1.78	77.2±3.97 <sup>vvxx</sup>	28.7±1.71	30.0±1.30 <sup>vvvxxx</sup>	21.8±0.44			
1 MPC	103.2±5.20*vvvxxx	339.7±24.86*vvvxxx	81.6±5.29*	90.8±6.06*vvvxxx	56.0±2.09*vvvxxx			
5 MPC	137.4±8.04*vvvxxx	528.0±45.47*vvvxxx	125.2±5.79*vx	152.5±14.09*vvvxxx	77.5±4.79*vx			
10 MPC	204.9±6.61*vvvxxx	933.0±42.59*vvvxxx	186.9±5.93*vvxxx	203.8±10.90*vvvxxx	370.8±19.22*vvvxxx			
	'Lvivska'							
Control	24.0±1.20	69.5±4.04°	23.0±1.31	34.5±1.30 <sup>vvvxxx</sup>	13.6±1.01 <sup>vvvxxx</sup>			
1 MPC	75.4±4.77*vvxxx	251.2±18.05*vvvxxx	95.0±6.79*vvxx	146.5±7.99*****	34.4±2.01*			
5 MPC	101.4±6.50*****	431.2±14.05*vvvxxx	160.9±13.48*vvvxxx	168.0±4.68******	67.2±2.60*			
10 MPC	125.2±9.81*	786.0±44.48*vvvxxx	216.9±13.92*vvvxxx	217.3±10.11******	192.5±8.46*vvx			
		"	Robusta'					
Control	22.4±1.50	49.1±2.41	24.7±0.93	10.3±0.66	23.7±1.31			
1 MPC	45.7±1.56*	101.2±5.41*	58.3±4.06*	15.0±0.43*	35.0±2.20*			
5 MPC	57.7±3.27*	177.0±10.63*	86.6±3.51*	29.3±1.30*	55.8±5.07*			
10 MPC	106.7±7.58*	236.2±19.50*	118.3±9.82*	41.8±1.32*	99.1±7.22*			
'Tronco'								
Control	21.7±0.67	53.2±3.44	22.9±1.39	8.3±0.43	26.1±1.28			
1 MPC	37.2±0.90*	95.2±4.56*	58.9±3.53*	17.5±0.66*	32.9±2.60*			
5 MPC	47.4±2.38*	183.0±14.60*	86.3±2.77*	19.3±1.32*	56.6±2.89*			
10 MPC	96.9±8.94*	252.7±14.31*	101.5±3.94*	34.0±1.52*	130.0±5.89*			

Notes: same as in Table 1

When metals were introduced into the soil at concentrations of 1 and 5 MPC, 1.7 to 3.4 times more Cu accumulated in the roots of 'Lvivska' and 'I-45/51' than in 'Tronco' and 'Robusta'. Similar trends are characteristic of Cu accumulation by the assimilation apparatus (**Table 4**).

The results obtained are in good agreement with the high  $EF_r^p$  values for Cu in all the cultivars studied. Under 10 MPC, this indicator was 1.1–2.1 times higher in 'I-45/51' and 'Lvivska' than in 'Robusta' and 'Tronco' (**Fig. 2**).

The roots of 'Lvivska' and 'I-45/51' accumulate the most Zn and Pb, which belong to the class of highly hazardous elements (up to 933 and 217 mg/g dry matter for Zn and Pb, respectively). On the contrary, 'Tronco' and 'Robusta' contained 3.3 and 4.6 times less of these ions than the other cultivars, respectively (**Fig. 3**).

The results obtained are in good agreement with  $EF_r^p$  and  $EF_r^p$ , which were 1.9–2.7 times lower for the latter group of hybrids. In addition, a significant difference in the accumulation of Zn and Pb between 'Lvivska', 'I-45/51' and 'Robusta', 'Tronco' groups of cultivars is evidenced by the results of two-way analysis of variance, according to which the difference between the content of these elements in the roots and leaves was significantly different with a probability of p  $\leq$ 0.001 (**Table 5, 6**).

Table 4. The content of heavy metals in the leaves of poplar cultivars, mg/kg dry matter

Variant	Cu	Zn	Ni	Pb	Cd	
'I–45/51'						
Control	13.2±0.88 <sup>vvvxx</sup>	38.5±3.31 <sup>vvv</sup>	10.5±0.75	7.3±0.66 <sup>vvxxx</sup>	4.7±0.30 <sup>vvvxxx</sup>	
1 MPC	32.9±1.88*vvvxxx	92.3±4.18*vvvxxx	20.2±0.61*vvvxxx	15.8±1.15*vvxx	20.8±1.59*	
5 MPC	50.9±2.83*vvvxxx	135.9±8.41*vvvxxx	40.1±2.28*vvvxxx	22.8±1.64*vvvxxx	33.0±1.52*	
10 MPC	71.8±4.56*vvvxxx	237.1±11.43*vvvxxx	67.7±6.09*vvvxxx	38.0±1.95*vvvxxx	83.9±4.35*vvvxxx	
		'Lv	vivska'			
Control	10.8±0.40°	33.4±0.86 <sup>w</sup>	9.6±0.35	11.3±0.43 <sup>vvvxxx</sup>	3.7±0.22vvvxxx	
1 MPC	21.1±1.67*vv	76.7±5.19*vvvxxx	20.0±1.14*vvvxxx	30.0±2.84*vvvxxx	11.1±0.44*vvx	
5 MPC	33.9±1.86*vvvxxx	123.4±3.86*vvvxxx	46.9±3.08*vvvxxx	47.0±1.95*vvvxxx	23.2±0.36*xx	
10 MPC	42.5±2.01*vvvxx	257.3±13.39*vvvxxx	80.7±2.89******	69.5±2.38*vvvxxx	37.3±1.44*	
		'Ro	busta'			
Control	6.9±0.67	17.4±0.69°	8.3±0.71	3.0±0.43	10.3±0.60	
1 MPC	9.9±0.66*	25.7±1.87*	10.5±0.58*	4.8±0.25*	19.8±1.23*	
5 MPC	14.0±0.88*	33.5±1.31*	17.3±0.64*	5.3±0.43*	26.8±1.59*	
10 MPC	15.4±0.75*	55.9±3.44*	24.7±1.51*	9.5±0.66*	34.4±2.89*	
'Tronco'						
Control	8.9±0.30	29.9±1.69×	7.7±0.84	2.3±0.43	9.3±0.60	
1 MPC	13.9±1.60*	34.9±2.09	9.6±0.58	3.3±0.25	16.9±0.58*	
5 MPC	17.3±1.00*	52.1±3.97*	11.3±1.10*	3.8±0.43*	22.8±1.88*	
10 MPC	21.0±0.57*	62.6±2.60*	19.8±1.33*	6.3±0.25*	26.3±1.15*	

Notes: same as in Table 1

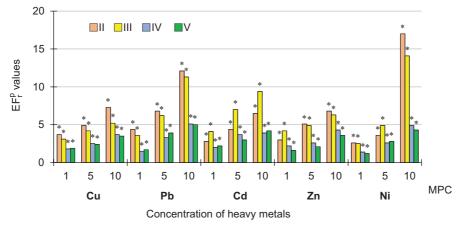


Fig. 2. The values of the EF<sup>p</sup>, for poplars: I – 'I-45/51', II – 'Lvivska', III – 'Robusta', and IV – 'Tronco'; \* – the difference is significant relative to the conditional control, p ≤0.05

When discussing the results obtained, it should be noted that the mechanisms underlying the absorption and transport of heavy metal ions include physiological processes associated with their uptake, translocation, and detoxification in tissues and cellular compartments, as well as the functioning of corresponding gene families involved in these processes, the activation of antioxidant systems and the generation of non-enzymatic antioxidants. (Sharma *et al.*, 2022).

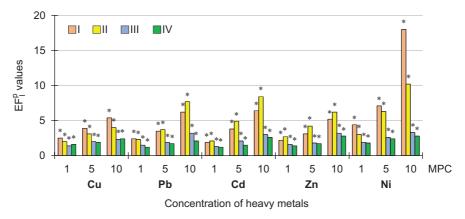


Fig. 3. The values of EF¹₁ for poplars: I – 'I-45/51', II – 'Lvivska', III – 'Robusta', and IV - 'Tronco'; \* – the difference is significant relative to the conditional control, p ≤0.05

Table 5. Results of univariate analysis of variance between the content of heavy metals in roots of cultivars of different groups ('Lvivska', 'I-45/51 and 'Robusta', 'Tronco')

Indicators	Cu	Zn	Ni	Pb	Cd		
Control							
R <sup>2</sup>	23.321	531.205	21.692	541.125	88.227		
F	4.279	14.259	3.881	180.375	25.705		
р	0.044	0.001	0.056	<0.001	<0.001		
	1 MPC						
$R^2$	2712.542	42841.688	972.092	12037.797	361.653		
F	68.170	57.471	12.586	158.620	24.093		
р	<0.001	<0.001	0.002	<0.001	<0.001		
		5 N	IPC				
$R^2$	5172.239	94473.422	3842.031	18666.125	315.097		
F	55.881	48.619	21.772	111.201	6.590		
р	<0.001	<0.001	<0.001	<0.001	0.015		
10 MPC							
$R^2$	7224.550	389165.625	9067.391	29920.547	44282.227		
F	34.736	118.539	35.473	177.258	111.872		
р	<0.001	<0.001	<0.001	<0.001	<0.001		

Table 6. Results of univariate analysis of variance between the content of heavy metals in the leaves of cultivars of different groups ('Lvivska', 'I-45/51 and 'Robusta', 'Tronco')

Indicators	Cu	Zn	Ni	Pb	Cd	
Control						
$R^2$	21.645	243.740	4.750	52.172	33.111	
F	19.732	21.576	3.369	69.563	51.269	
р	<0.001	<0.001	0.075	<0.001	<0.001	
1 MPC						
R <sup>2</sup>	307.089	3103.913	101.733	458.922	57.019	
F	44.204	79.175	58.023	64.410	16.663	
р	<0.001	<0.001	<0.001	<0.001	<0.001	
5 MPC						
R <sup>2</sup>	863.686	7793.114	893.818	1217.797	66.910	
F	87.734	100.819	73.124	236.179	10.470	
р	<0.001	<0.001	<0.001	<0.001	0.004	
10 MPC						
R <sup>2</sup>	1958.882	35553.015	2797.203	2605.922	2037.299	
F	101.178	144.275	75.417	347.456	88.478	
р	<0.001	<0.001	<0.001	<0.001	<0.001	

Of the poplars studied under model experimental conditions, 'I-45/51' was the most active in absorbing Cd, which belongs to the class of highly hazardous elements, from the soil. This cultivar accumulates 1.9–4 times more Cd in the roots and leaves than 'Tronco' and 'Robusta' under the influence of heavy metals at a concentration of 10 MPC (**Tables 2**, **3**). Under the maximum concentration of heavy metal compounds, 'Lvivska' cultivar of poplar accumulates 12–52 % more Cd in the root than 'Tronco' and 'Robusta'.

Although significant progress has been made in the last twenty years in understanding cadmium uptake and detoxification by plants, the mechanisms by which plants adapt to and tolerate cadmium toxicity remain poorly understood (Zhang *et al.*, 2024). In this regard, different levels of the *PePCR2* gene transcription induced by elevated Cd levels in *Populus euphratica* (Lv *et al.*, 2023) can be considered as a possible mechanism that leads to the differences in root uptake of cadmium and its further transport to leaves by the two groups of poplar cultivars in our experiments. Also, a different activity of the *PePCR2* gene under Cd excess affects the level of transcripts from *ABCG*, *HMA*, *YSL*, *NRAMP*, and *ZIP* families, which in turn are involved in the transport of Cu<sup>2+</sup>, Zn<sup>2+</sup>, Fe<sup>3+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup> and other divalent ions (Levenets *et al.*, 2022; Tan *et al.*, 2020).

According to the results of the model experiments, similar patterns are generally characteristic of the less hazardous element – Ni. Under the conditions of soil contamination with heavy metal compounds, a more intensive accumulation of Ni is observed in the cultivars 'Lvivska' and 'I-45/51'. Thus, the roots of these poplars contain 2–3.4 times more Ni and the leaves contain 1.5–4.8 times more of this element than 'Tronco' and 'Robusta', (**Tables 2**, **3**). These patterns are confirmed by the 1.7–2.3 times lower values of the  $\mathsf{EFP}_1$  and  $\mathsf{EFP}_2$  for 'Tronco' and 'Robusta'. Our results reveal certain mechanisms of phytoaccumulation and demonstrate the possibility of using poplar cultivars for phytoremediation, phytoextraction, and phytostabilisation of soils contaminated with heavy metals.

#### CONCLUSIONS

Based on the results of the research, poplars can be divided into two groups. The first group ('Lvivska' and 'I-45/51') includes cultivars with a high level of heavy metal accumulation and the highest  $\mathsf{EFP}_{\mathsf{l}}$  and  $\mathsf{EFP}_{\mathsf{r}}$  values. The second group ('Tronco' and 'Robusta') has a low level of heavy metal accumulation and low values of the  $\mathsf{EFP}_{\mathsf{l}}$  and  $\mathsf{EFP}_{\mathsf{r}}$ . This fact is associated with the inhibition of the translocation of elements to different plant organs, which is characteristic of the poplars of the second group. At the same time, the level of translocation of these pollutants is higher in poplar roots than in leaves. This distribution can be explained by the fact that heavy metals were applied in the form of soluble compounds and by the existence of plant defence mechanisms.

The results of this research may enable poplar cultivars to be used in two specific areas in the future: phytostabilisation and phytoextraction. In the first case, the plants reduce the availability of metals and the danger of heavy metals in the soil. When considering the possibility of using cultivars in phytostabilisation technologies, it is necessary to take into account the ability of plant roots to control the uptake and inactivation of potentially toxic elements in urban soils. This will reduce the risk of human exposure to toxic elements. Considering the fact that poplars quickly form a significant amount of biomass, the cultivars studied can be indispensable components of sustainable ecosystems with increased functional stability.

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#### COMPLIANCE WITH ETHICAL STANDARDS

**Conflict of Interest:** the authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

**Animal Rights:** this article does not include animal studies.

**Human Rights**: this article does not contain any studies with human subjects performed by the any of the authors.

# **AUTHOR CONTRIBUTIONS**

Conceptualization, [V.G.; O.D]; methodology, [O.D.; N.D.; V.G.]; validation, [L.B.; N.D.; V.G.]; formal analysis, [O.D.; L.B.]; investigation, [N.D.; L.B.]; resources, [V.G.]; data curation, [V.G.; O.D.]; writing — original draft preparation, [O.D.; V.G.; N.D.]; writing — review and editing, [L.B.; V.G.]; visualization, [N.D.; O.D.]; supervision, [O.D.]; project administration, [V.G.]; funding acquisition, [L.B.].

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# АКУМУЛЯЦІЯ ВАЖКИХ МЕТАЛІВ У ВЕГЕТАТИВНИХ ОРГАНАХ ТОПОЛЬ ЗА ЇХНЬОГО СУМІСНОГО ВНЕСЕННЯ У ҐРУНТ

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**Обґрунтування.** Дослідження особливостей аутекології рослин за надлишкового надходження важких металів неможливе без з'ясування специфіки поглинання та накопичення їх рослинами. Тому актуальним є вивчення перерозподілу важких металів у системі "ґрунт–рослина". Тополі, завдяки їхнім властивостям (швидкий ріст, велика площа органа асиміляції та ін.), є найвдалішим об'єктом для з'ясування їхніх аутекологічних особливостей і для створення зелених насаджень.

**Матеріали і методи.** Об'єктами досліджень були дворічні саджанці тополь:  $^{1-45/51'}$ ,  $^{1}$  Lvivska',  $^{1}$  Robusta',  $^{1}$  Tronco' та  $^{1}$  грунти, на яких вони ростуть. Рослини поливали один раз на тиждень протягом 2-х місяців сумішшю сполук важких металів у кількості 1, 5 та 10 гранично допустимих концентрацій (ГДК). Як джерело важких металів використовували суміші солей  $(CdSO_4, Ni(NO_3)_2, CuSO_4, ZnSO_4, Pb(CH_3COO)_2)$ . Контрольні рослини не зазнавали дії важких металів. Рослини вирощували за природного рівня освітленості й температури  $^{1}$  25–30 °C. Визначення рухомих форм Cu, Zn, Ni, Pb i Cd в амонійно-ацетатній витяжці з ґрунту та їхнє накопичення в листках та коренях проводили з використанням загальноприйнятих методів на атомно-адсорбційному спектрофотометрі C-115.

**Результати.** Досліджено накопичення і транслокацію важких металів у системі "ґрунт–рослина" за умов модельних дослідів. З'ясовано особливості нагромадження важких металів у ґрунтах, на яких зростали тополі. Внесення сполук важких металів

призводить до збільшення вмісту рухомих форм досліджених елементів у ґрунті. За дії 1, 5 і 10 ГДК іонів важких металів усі культивари більш активно акумулюють їх у кореневій системі, порівняно з листками.

Висновки. Досліджені тополі можна розподілити на дві групи. До першої групи належать 'I-45/51' і 'Lvivska' з високим рівнем акумуляції важких металів і найбільшим фактором нагромадження, до другої ('Tronco' і 'Robusta') — з низьким рівнем накопичення важких металів і малим фактором нагромадження. Тобто у тополь другої групи є фізіологічні механізми, які обумовлюють встановлені особливості їхньої транслокації.

**Ключові слова:** культивар тополі, важкі метали, "ґрунт–рослина", транслокація, фактор нагромадження