

# CHANGES IN THE CONTENT OF LIPID PEROXIDE OXIDATION PRODUCTS IN LEAVES OF POPLAR SPECIES AND CULTIVARS UNDER CONDITIONS OF ENVIRONMENTAL POLLUTION WITH HEAVY METALS

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## Abstract

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Changes in the content of lipid peroxidation products (LPO) in the assimilation organs of two species and three poplar cultivars under conditions of varying degrees of environmental pollution with heavy metals were studied. The established different amounts of accumulation of the latter in plant leaves indicate the specificity of their absorption. The greatest ability to accumulate heavy metals was revealed in plants growing in a zone of severe pollution, at the Northern Iron Ore Dressing Combine experimental site (henceforth referred to as Pivnichnyi HZK or PivnHZK). In the assimilation organs of the “Lvivska” and “Hradizhka” cultivars, cadmium, one of the most dangerous elements for living organisms, accumulates on average 25.5 times more than in the leaves of plants growing on the territory of the “Arboretum of the Kyryvi Rih Botanical Garden of the National Academy of Sciences of Ukraine” (control). The obtained data are in good agreement with the enrichment factor ( $EF_p$ ) indicators of the specified element in the assimilation apparatus of poplars. A peculiarity of the stress effect of heavy metals on poplar is the absence of an increase in the formation of TBARS in the cultivars “Lvivska” and “Hradizhka” with an increase in the level of accumulation of heavy metals in the conditions of PivnHZK in comparison with plants on CHZK. Considering that TBARS is a marker of the level of stress effect on organisms, and the established fact of high accumulation of heavy metals, the cultivars “Lvivska” and “Hradizhka” can be used to create effective phytofilters with a high level of pollution by heavy metals at industrial sites.

**Key words:** poplars, heavy metals, assimilation apparatus, lipid peroxidation (LPO), thiobarbituric acid reactive substances (TBARS).

## Introduction

The increase in anthropogenic pollution, which intensified in the middle of the 20th century, poses a real danger to all living organisms (Petrov, 2023; Jonah, Archibong, 2022; Anyanwu et al., 2022). In the industrial centers of Ukraine, one of which is Kyryvi Rih, there is an increase in production volumes, as well as an increase in the density of motor vehicles, which leads to an aggravation of environmental problems in these regions (Danylchuk et al., 2024; Savosko et al., 2022; Fedorchak, 2024). To solve them, one of the main ways is to identify woody plants that are characterized by high accumulation of toxicants in combination with their physiological resistance to environmental pollution. It is such plants that are advisable to use when creating effective biological filters. Poplars, due to their many properties (rapid growth, large areas of the assimilation organ, the presence of adhesive substances on the buds and leaves, a wide distribution area, etc.), are the most successful object for clarifying the autecological peculiarities of the genus and taking them into account when creating green spaces in conditions with different levels of pollution (Hjelm et al., 2018; Traboulsi et al., 2025; Mc Carthy et al., 2017).

The peculiarities of plant autecology regarding the influence of environmental factors are determined by changes in various physiological processes that contribute to their adaptation to certain conditions of existence (Bihuňová et al., 2021). The ability of poplars to accumulate heavy metals, as a dominant environmental factor, highlighted in previous works determines the relevance of studying the course of LPO processes – one of the primary signaling systems of the body’s response (Chirkova, Yemelyanov, 2018; Baik et al., 2021). Among the known mechanisms that characterize the toxicity of heavy metals in biological systems are the formation of active oxygen and the ability to interact with cellular biomolecules.

As a result of the action of toxic compounds or other stress factors, in cellular membrane structures, the intensity of the course of LPO processes increases (Filipstsova, Yurin, 2021; Shahid et al., 2014; Jorjani, Pehlivan Karakaş, 2024). This is accompanied by inactivation of sulfhydryl groups of proteins and damage to nucleic acids (Jones, Dangel, 2006; Albert, 2013; Blavillain et al., 2011; Blokhina, Fagerstedt, 2010). The importance of studying this process lies in the fact that an increase in the intensity of LPO processes is an indicator of the first reactions of

the organism to any stress effect. The intensity of these processes can be assessed by the content of thiobarbituric acid reactive substances (TBARS) value, diene conjugates (DC), dienketones (DK), etc. Therefore, studying changes in the content of these compounds will allow us to reveal the peculiarities of the implementation at the physiological level of the adaptive potential of species and cultivars of the genus *Populus* and the formation of their resistance to the action of heavy metals. Therefore, the aim of our study was to investigate the peculiarities of the translocation of heavy metals to assimilation organs in poplar species and cultivars and the response of signaling systems at the physiological and biochemical level of plant organization.

## Material and methods

### Study area

The study was conducted at three monitoring sites. The monitoring site “Pivnichnyi HZK” (PivnHZK) is located in the Ternivskyi district of Kryvyi Rih city near the warehouse for the shipment of finished products of the “Northern Iron Ore Dressing Combine” (48°11'41.2"N 33°31'25.9"E). The topography of the area is flat. The monitoring site “Central HZK” (CHZK) is located in the Pokrovskyi district of Kryvyi Rih city near the production facilities of the ore beneficiation factory of Central Iron Ore Dressing Combine (Myrivske village, the city of Kryvyi Rih, Dnipropetrovsk region) (48°04'43.2"N, 33°22'26.8"E). The topography of the area is flat. The monitoring site “Arboretum of the Kryvyi Rih Botanical Garden of the National Academy of Sciences of Ukraine” (control) is located in the Ternivskyi district of Kryvyi Rih city (48°08'51.5"N 33°34'33.9"E) outside the sanitary protection zone of PivnHZK, at a distance more than 5 km from the source of emission of PivnHZK and at a distance of more than 20 km from the source of emission of CHZK. The topography of the area is flat.

### Objects of research

The objects of research were 3-year-old seedlings of two species and three poplar cultivars that are actively used in the national economy, as well as in landscaping. Saplings of *Populus italica* Du Roi and *P. deltoides* W. Bartram ex Marshall were local reproduction. Saplings “Hradizhka,” “Lvivska,” and “Tronco” were grown from lignified cuttings, which were obtained from the mother plant of the Lubny branch of H.M. Vysotskyi National Institute of Forestry and Agromelioration. Two-year-old seedlings were planted at three monitoring sites: in the industrial sites of PivnHZK and CHZK, which according to previous studies are areas with strong and moderate pollution, respectively, and in the Arboretum of the Kryvyi Rih Botanical Garden of the National Academy of Sciences – control (Danylchuk et al., 2023).

### Research methods

Plant material, fully developed mature leaf blades, was selected in August, after the cessation of shoot growth and the formation of apical buds, from the middle part of the crown of 10 individuals of each of the first-class cultivars by age, with the south-western exposure. The content of heavy metals in plant material was de-

termined following the guidelines for the determination of heavy metals in plant products (Danylchuk et al., 2023; Hrytsaenko et al., 2003; San PiN, 1988). The plant material was mineralized by dry ashing (at a temperature of +525 °C) and subsequent extraction with HNO<sub>3</sub>. Further determination of the concentration of heavy metals was carried out using an atomic adsorption spectrophotometer C-115PK Selmi (Ukraine) according to generally accepted methods (Kochmar, Karabyn, 2022).

To determine the content of LPO intermediates, 20% leaf homogenate (which was taken from one crown layer) was used, prepared in 0.1 M potassium phosphate buffer containing 0.25 M sucrose. LPO intermediates, extracted in isopropanol and heptane mixture (1:1) at 3000 rpm for 15 min on a Labofuge 400R HERAEUS centrifuge (Germany), were determined in the heptane fraction. The optical density of the obtained heptane extract was determined at wavelengths of 233 and 278 nm on a spectrophotometer Spekol 1500 Analytik Jena AG (Germany). The number of diene conjugates (DC) and dienketones (DK) according to the method of Stalnaya I.D. modified by Grishko, V. and Danilchuk, A. (Grishko, Danilchuk, 2004). The TBARS content was determined by the TBA reaction method (Heath, Packer, 1968; Stewart, Bewley, 1980); 2.0 ml of distilled water, 1.0 ml of trichloroacetic acid (TCA) solution, and 1.0 ml of TBA solution were added to a test tube with 0.5 ml of supernatant, which was prepared as described above. The sample was heated in a boiling water bath for 10 min, then centrifuged for 10 min at 3000 rpm in a Labofuge 400R centrifuge. The color intensity was measured at a wavelength of 540 nm on a Spekol 1500 spectrophotometer. The protein content in homogenates plant tissues was determined by the method of Greenberg and Craddock (1982) by reaction of bromophenol blue.

### Statistical analysis

Calculations of the enrichment factor values for heavy metals in the leaves (EF<sub>p</sub>) of the poplars were performed according to formula (1) (Kisku et al., 2000):

$$EF = \frac{\text{concentration of the element in the plant material under heavy metal application}}{\text{concentration of the element in the plant material in the control}} \quad (1)$$

The content of diene conjugates (uA/ml homogenate) was calculated according to formula (2) and dienketones according to formula (3):

$$DC = \frac{A_{233} \times V_H}{V_S}, \quad (2)$$

$$DK = A_{278} \times 2, \quad (3)$$

where  $V_S$  is the final volume of heptane extract,  $V_H$  is the volume of sample taken,  $A_{233}$  is the optical density of the solution at wavelength of 233 nm, and  $A_{278}$  is the optical density at 278 nm.

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.

## Results

### Research on the accumulation of heavy metals in the assimilation apparatus of poplar cultivars

According to the results of the studies, it was found that the translocation of cadmium ions, which belongs to highly toxic elements, occurs most intensively in the assimilation organs of “Hradizhka” and “Lvivska,” and the level of its accumulation in the above-mentioned cultivars at the CHZK industrial site exceeded the indicators for the conditional control by up to 15.5 times (Table 1).

Under conditions of severe pollution, similar trends are observed. High levels of cadmium translocation are characteristic of the cultivars “Hradizhka” and “Lvivska.” The value of  $EF_p$  for this metal in the leaves of the above-mentioned taxa is 26.0 and 25.1, respectively (Fig. 1).

The obtained data coincide well with the values of the absolute content of the element. Thus, in the assimilation organs of poplars “Hradizhka” and “Lvivska,” under conditions of moderate pollution, cadmium ions accumulate 4.4–19.6 times more, and under severe pollution, 4.1–24.7 times more than in “Tronco,” *Populus italica* and *P. deltoides* (Table 1).

The least cadmium accumulates in the control and heavily polluted conditions of *P. italica*, while in moderate pollution – “Tronco,” and in all variants of the experiment, the least zinc also gets to its assimilation organs. If in “Hradizhka,” “Lvivska,” *P. italica*, and *P. deltoides*, the value of the zinc enrichment

factor varies from 3.5 to 8.7, then for “Tronco,” it is on average 2.5. Comparison of the obtained data on the zinc enrichment factor for leaves showed that at the industrial site of CHZK in “Hradizhka,” “Lvivska,” *P. italica*, and *P. deltoides*, it is on average 1.8 times higher and at PivnHZK, it is 2.3 times higher than in “Tronco.”

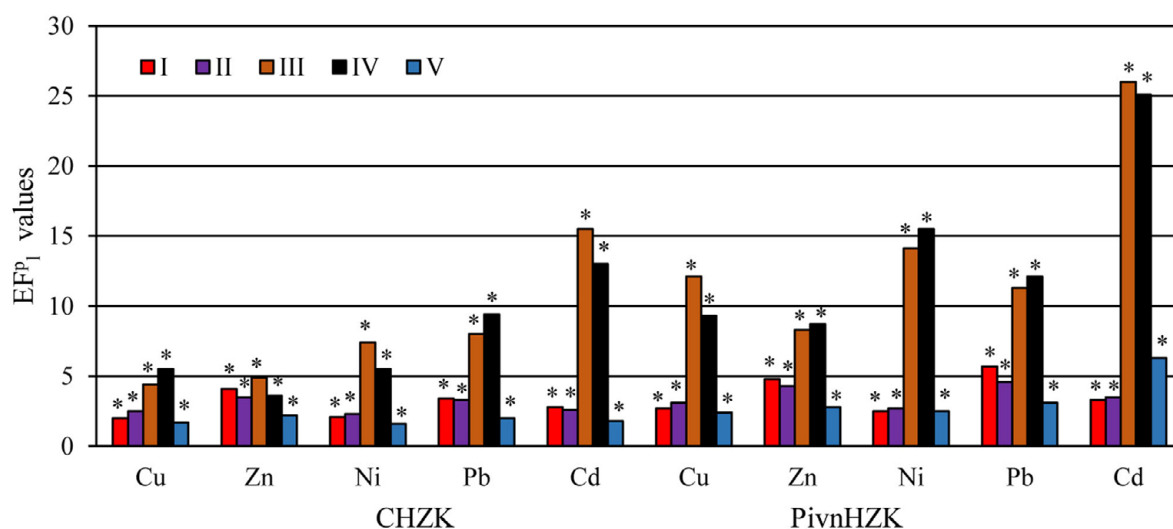
Among the less toxic elements for the environment, the processes of translocation of copper and nickel ions to the leaves of poplar cultivars are intensive. Moreover, nickel accumulated most actively under the conditions of PivnHZK in “Lvivska” and copper ions in the assimilation organs of “Hradizhka,” while for *P. italica*, *P. deltoides*, and “Tronco,” the values of enrichment factor under similar growth conditions were 3–6.5 times lower (Fig. 1). Considering that the entry of lead ions into plants also occurs with air industrial emissions, the trends of translocation of the specified toxicant among the studied taxa are preserved. Thus, the values of  $EF_p$  for the lead in the leaves of “Hradizhka” and “Lvivska” under conditions of moderate pollution were 2.4–4.7 times higher, and under conditions of strong pollution, they were 2.0–3.9 times higher than in “Tronco,” *P. italica*, and *P. deltoides* (Fig. 1).

Therefore, when determining the intensity of heavy metal translocation by poplar species and cultivars at different levels of environmental pollution, they can be divided into two groups. The first group is formed by “Hradizhka” and “Lvivska,” with a high level of metal translocation by assimilation organs, and the second one by “Tronco,” *P. italica*, and *P. deltoides* with a significantly lower level.

**Table 1.** Accumulation of some heavy metals in the assimilation organs of poplars growing in different environmental conditions, mg/kg dry matter.

| Place of sampling               | Cu            | Zn             | Ni             | Pb             | Cd             |
|---------------------------------|---------------|----------------|----------------|----------------|----------------|
|                                 | M ± m         | M ± m          | M ± m          | M ± m          | M ± m          |
| <b><i>Populus italica</i></b>   |               |                |                |                |                |
| Control                         | 14.8 ± 1.26   | 56.6 ± 1.40    | 5.6 ± 0.31     | 7.2 ± 0.45     | 4.1 ± 0.17     |
| CHZK                            | 29.6 ± 1.17*  | 232.1 ± 14.05* | 11.8 ± 0.64*   | 24.4 ± 1.58*   | 11.5 ± 0.71*   |
| PivnHZK                         | 40.0 ± 2.47*  | 271.7 ± 16.62* | 14.1 ± 0.79*   | 40.9 ± 3.79*   | 13.6 ± 0.52*   |
| <b><i>Populus deltoides</i></b> |               |                |                |                |                |
| Control                         | 15.1 ± 0.65   | 49.6 ± 2.79    | 6.4 ± 0.34     | 8.2 ± 0.43     | 6.6 ± 0.35     |
| CHZK                            | 37.7 ± 2.79*  | 173.6 ± 7.63*  | 14.6 ± 1.36*   | 27.1 ± 1.18*   | 17.1 ± 0.89*   |
| PivnHZK                         | 46.8 ± 4.83*  | 213.3 ± 11.45* | 17.1 ± 1.34*   | 37.7 ± 1.12*   | 23.0 ± 1.55*   |
| <b>“Hradizhka”</b>              |               |                |                |                |                |
| Control                         | 8.3 ± 0.73    | 77.6 ± 3.61    | 15.2 ± 0.54    | 20.6 ± 1.79    | 12.9 ± 0.70    |
| CHZK                            | 36.8 ± 1.15*  | 379.4 ± 28.45* | 111.5 ± 7.57*  | 165.7 ± 15.39* | 199.8 ± 7.81*  |
| PivnHZK                         | 101.1 ± 4.10* | 641.6 ± 39.40* | 213.5 ± 14.91* | 233.6 ± 14.55* | 335.8 ± 27.06* |
| <b>“Lvivska”</b>                |               |                |                |                |                |
| Control                         | 6.8 ± 0.25    | 91.6 ± 8.66    | 8.2 ± 0.28     | 16.1 ± 1.52    | 5.8 ± 0.18     |
| CHZK                            | 37.4 ± 2.33*  | 329.6 ± 16.09* | 44.5 ± 1.80*   | 150.8 ± 10.19* | 75.3 ± 3.91*   |
| PivnHZK                         | 63.1 ± 3.48*  | 795.6 ± 30.51* | 126.0 ± 7.94*  | 193.5 ± 5.53*  | 145.3 ± 11.46* |
| <b>“Tronco”</b>                 |               |                |                |                |                |
| Control                         | 4.4 ± 0.32    | 40.1 ± 2.18    | 4.1 ± 0.17     | 2.0 ± 0.29     | 5.7 ± 0.48     |
| CHZK                            | 7.3 ± 0.73*   | 88.7 ± 6.15*   | 6.4 ± 0.48*    | 4.0 ± 0.29*    | 10.2 ± 0.57*   |
| PivnHZK                         | 10.4 ± 0.33*  | 110.6 ± 5.57*  | 10.1 ± 0.51*   | 6.3 ± 0.57*    | 35.9 ± 3.38*   |

Notes: \*the difference is significant relative to the control,  $p \leq 0.05$ .



**Fig. 1.** The values of  $EF_p$  for poplars growing on industrial sites: I, *Populus italica*; II, *Populus deltoides*; III, “Hradizhka”; IV, “Lvivska”; and V, “Tronco.” \*The difference is significant relative to the conditional control,  $p < 0.05$ .

### Research into lipid peroxidation processes in the assimilation apparatus of poplar species and cultivars

The data obtained from the research results (Table 2) indicate that the highest level of primary and secondary LPO products in the leaves of poplar cultivars under control conditions is observed in “Tronco.” For example, the number of diene conjugates and dienketones in the leaves of “Tronco” exceeds the level in other cultivars by 1.7–2.0 times. Among poplar species, the highest level of diene conjugates formation was in *P. deltoides*, while the species did not differ statistically significantly in the number of dienketones. In the leaves of *P. italica*, 2.5 times more TBARS was formed than in *P. deltoides*, while among the cultivars, the highest level was observed in “Tronco.” The difference in the number of LPO products shown above is a species-specific response to their growth conditions.

The increased level of translocation of heavy metal ions to leaves at the monitoring sites of ore processing plants leads to a significant activation of lipid peroxidation processes compared to the control. In the leaves of *P. deltoides* and *P. italica* under CHZK and PivnHZK conditions, the number of diene conjugates increases by 10–30%, while in the cultivars “Hradizhka” and “Lvivska,” it increases by 40–60%. The greatest increase in the level of conjugated diene structures of lipid hydroperoxides was found in the leaves of the cultivar “Tronco.” In this cultivar at the CHZK industrial site, their content exceeds the level in control plants by 1.9 times and in PivnHZK conditions by 2.1 times. The results presented in Table 2 show that only in *P. deltoides* and the cultivar “Tronco,” there is a statistically significant increase in the amount of diene conjugates formed under conditions of PivnHZK in comparison with plants growing on CHZK, where the level of heavy metal contamination was lower. In plants of other species and cultivars, only a tendency to increase the amount of primary LPO products under conditions of higher levels of contamination was revealed.

Analysis of the obtained data taking into account different levels of heavy metal contamination of monitoring sites of ore dressing factories clearly indicates the formation of a greater

amount of secondary LPO metabolites. The greatest amount of dienketones is formed in the assimilation organs of “Tronco” (0.107 uA/ml) at the industrial site of PivnHZK, which is 2.2 times more than in plants of the arboretum of the botanical garden. The general trend was a 20–40% increase in dienketone levels in leaves of all poplar species and cultivars with increasing heavy metal contamination at the PivnHZK industrial site, compared to CHZK. However, only in *P. italica* and *P. deltoides* was this pattern statistically significant (Table 2).

Along with hydroperoxides, other compounds are also formed during the oxidation of fatty acids, among which researchers pay the most attention to another secondary product of hydroperoxide destruction – malondialdehyde (MDA). It should be noted that MDA is not the only TBARS. TBARS is formed not only in the process of peroxide oxidation but also under the influence of acid and heating of homogenates, which is part of the procedure for performing a color reaction with TBA. Analysis of the results presented in Table 2 shows that among the poplars studied in the conditions of the arboretum of the botanical garden, the largest amount of TBARS is produced in *P. italica* (0.249 mM/mg protein). In the leaves of “Tronco,” 2.5 times more TBARS is formed compared to other cultivars.

The impact of heavy metal pollution on the monitoring sites of ore processing plants leads to an intensification of TBARS formation. Under the conditions of the CHZK industrial site, up to 50% more TBARS is formed in the assimilation organs of the cultivars “Hradizhka” and “Lvivska” than under control conditions. In the leaves of *P. italica*, *P. deltoides*, and “Tronco,” the processes of TBARS formation occur more intensively – the number of products of the LPO formation process increases by 1.6–2 times. The calculated correlation coefficients between the increase in the level of heavy metals accumulated by plants in the leaves at the monitoring sites of ore processing plants and the formation of secondary LPO products ( $r_s = +0.76 - +0.98$ ) indicate a strong positive relationship between these indicators (Table 3).

The impact of heavy metal pollution on the monitoring sites of ore processing plants leads to an intensification of TBARS for-

**Table 2.** The content of lipid peroxidation products in the assimilation organs of poplar species and cultivars under conditions of heavy metal contamination, n=4.

| Species and cultivars                                          | Place of sampling |      |                |      |                            |      |
|----------------------------------------------------------------|-------------------|------|----------------|------|----------------------------|------|
|                                                                | Control           |      | CHZK           |      | PivnHZK                    |      |
|                                                                | M ± m             | CV,% | M ± m          | CV,% | M ± m                      | CV,% |
| <b>Diene conjugates (uA/ml homogenate)</b>                     |                   |      |                |      |                            |      |
| <i>Populus italica</i>                                         | 0.070 ± 0.009     | 23.2 | 0.084 ± 0.005  | 9.7  | 0.091 ± 0.007              | 13.5 |
| <i>Populus deltoides</i>                                       | 0.151 ± 0.004     | 4.4  | 0.172 ± 0.005* | 4.7  | 0.190 ± 0.006 <sup>v</sup> | 5.8  |
| “Tronco”                                                       | 0.087 ± 0.002     | 4.7  | 0.166 ± 0.005* | 4.9  | 0.182 ± 0.005 <sup>v</sup> | 4.5  |
| “Lvivska”                                                      | 0.052 ± 0.004     | 13.0 | 0.072 ± 0.005* | 13.0 | 0.082 ± 0.006*             | 13.0 |
| “Hradizhka”                                                    | 0.044 ± 0.004     | 15.1 | 0.061 ± 0.005* | 13.0 | 0.068 ± 0.006*             | 16.0 |
| <b>Dienketones (uA/ml homogenate)</b>                          |                   |      |                |      |                            |      |
| <i>Populus italica</i>                                         | 0.029 ± 0.003     | 20.0 | 0.042 ± 0.001* | 5.6  | 0.053 ± 0.002 <sup>v</sup> | 5.5  |
| <i>Populus deltoides</i>                                       | 0.021 ± 0.003     | 22.5 | 0.033 ± 0.001* | 5.1  | 0.037 ± 0.001 <sup>v</sup> | 4.6  |
| “Tronco”                                                       | 0.049 ± 0.001     | 4.9  | 0.097 ± 0.003* | 4.9  | 0.107 ± 0.003 <sup>v</sup> | 4.5  |
| “Lvivska”                                                      | 0.031 ± 0.001     | 4.4  | 0.046 ± 0.001* | 5.1  | 0.052 ± 0.002 <sup>v</sup> | 5.1  |
| “Hradizhka”                                                    | 0.024 ± 0.001     | 8.0  | 0.034 ± 0.001* | 4.1  | 0.038 ± 0.002*             | 6.6  |
| <b>Thiobarbituric acid reactive substances (mM/mg protein)</b> |                   |      |                |      |                            |      |
| <i>Populus italica</i>                                         | 0.249 ± 0.012     | 8.4  | 0.392 ± 0.006* | 2.8  | 0.534 ± 0.002 <sup>v</sup> | 0.7  |
| <i>Populus deltoides</i>                                       | 0.101 ± 0.003     | 4.7  | 0.172 ± 0.007* | 7.4  | 0.275 ± 0.016 <sup>v</sup> | 9.9  |
| “Tronco”                                                       | 0.165 ± 0.003     | 3.6  | 0.333 ± 0.010* | 5.4  | 0.406 ± 0.011 <sup>v</sup> | 4.7  |
| “Lvivska”                                                      | 0.066 ± 0.005     | 12.8 | 0.098 ± 0.008* | 13.3 | 0.119 ± 0.009*             | 13.3 |
| “Hradizhka”                                                    | 0.064 ± 0.006     | 16.8 | 0.093 ± 0.009* | 17.1 | 0.113 ± 0.011*             | 17.3 |

Notes: \* – differences are significant compared to control according to Student's t-test,  $p \leq 0.05$ ; v – differences are significant compared to CHZK according to Student's t-test,  $p \leq 0.05$ ; M ± m – the average value with an error; CV,% – coefficient of variation.

**Table 3.** Correlation coefficients between the content of heavy metals in the assimilation organs of poplars and TBARS in the conditions of monitoring sites of ore processing plants.

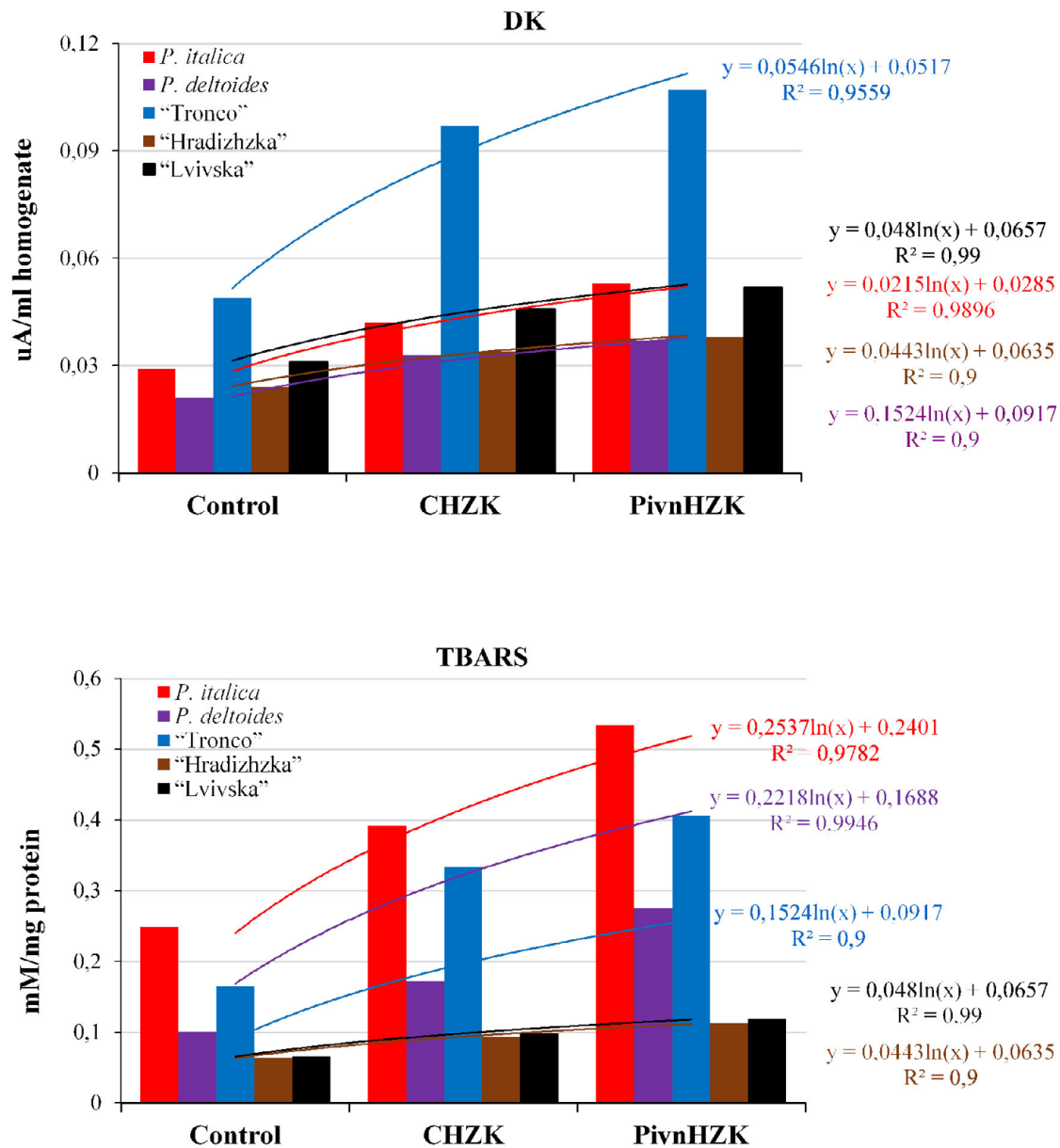
| Species and cultivars    | Elements   |            |            |            |            |
|--------------------------|------------|------------|------------|------------|------------|
|                          | Cu         | Zn         | Ni         | Pb         | Cd         |
| <i>Populus italica</i>   | +0.98±0.08 | +0.92±0.15 | +0.94±0.13 | +0.96±0.10 | +0.94±0.13 |
| <i>Populus deltoides</i> | +0.87±0.19 | +0.88±0.18 | +0.89±0.17 | +0.93±0.14 | +0.96±0.10 |
| “Tronco”                 | +0.92±0.15 | +0.94±0.13 | +0.92±0.14 | +0.94±0.13 | +0.81±0.22 |
| “Lvivska”                | +0.85±0.20 | +0.83±0.21 | +0.81±0.22 | +0.87±0.19 | +0.94±0.13 |
| “Hradizhka”              | +0.82±0.22 | +0.89±0.17 | +0.76±0.25 | +0.80±0.23 | +0.84±0.20 |

mation. Under the conditions of the CHZK industrial site, up to 50% more TBARS is formed in the assimilation organs of the cultivars “Hradizhka” and “Lvivska” than under control conditions. In the leaves of *P. italica*, *P. deltoides*, and “Tronco,” the processes of TBARS formation occur more intensively – the number of products of the LPO formation process increases by 1.6–2 times. The calculated correlation coefficients between the increase in the level of heavy metals accumulated by plants in the leaves at the monitoring sites of ore processing plants and the formation of secondary LPO products ( $r_s = +0.76 - +0.98$ ) indicate a strong positive relationship between these indicators (Table 3).

It was important for us to find out whether there is a pattern between the level of stress action, which is estimated by

the amount of formation of secondary LPO products, and the increase in the content of heavy metals in the assimilation organs of certain species or cultivars of poplars on CHZK compared to PivnHZK. The statistical significance of the difference between the content of secondary LPO products in plants on PivnHZK compared to plants growing on CHZK (Table 2) and the probability of logarithmic approximation of the content of intermediate products were assessed (Fig. 2).

The results obtained indicate that for the cultivars “Lvivska” and “Hradizhka,” there was no statistically significant increase in the content of TBARS (the main marker of the level of stress action of various environmental factors) in plants on CHZK



**Fig. 2.** Probability of logarithmic approximation of the content of secondary products of lipid peroxidation in poplar leaves at different levels of heavy metal contamination.

(with a lower level of heavy metal pollution), compared to PivnHZK (maximum level of pollution). While in the leaves of *P. italica*, *P. deltoides*, and "Tronco" under PivnHZK conditions, a statistically significant increase in TBARS is formed. A similar pattern is also observed with logarithmic approximation of the TBARS content (Fig. 2).

Comparing the studied species and cultivars with each other by the level of stress effect on plants, it can be stated that at different levels of heavy metal pollution at the monitoring sites of ore processing factories, an intensification of the formation of primary and secondary LPO products was ascertained. A peculiarity of the stress effect of heavy metals on poplar is the absence

of an increase in TBARS formation in the cultivars "Lvivska" and "Hradizhka" with an increase in the level of heavy metal accumulation under PivnHZK conditions compared to plants on CHZK.

## Discussion

Analysis of published studies shows that poplars growing on contaminated soils accumulate Cd, Cr, Pb, and Zn more intensively than control individuals from uncontaminated soils (Tözsér et al., 2023). In this respect, the results of our study coincide with the results of other researchers. At the same time, "Lvivska" and

“Hradizhka” accumulate the most toxic elements more than *P. italica*, *P. deltoides*, and “Tronco.” The obtained data allow us to divide the experimental plants into two groups. The first group is formed by “Hradizhka” and “Lvivska” (with a high level of metal translocation to the assimilation organs) and the second one by *P. italica*, *P. deltoides*, and “Tronco” (with a lower level of metal translocation to the assimilation organs).

The enhancement of lipid peroxidation processes is a universal mechanism by which the most important homeostatic physicochemical parameters of the cell are controlled, which play a significant role in the survival of organisms under the influence of extreme factors of various origins (Batra, 2021; Ren et al., 2020). The intensity of heavy metal translocation in the assimilation apparatus of poplars growing under polluted conditions affects the course of redox processes in the plant organism at the cellular level. The formed reactive oxygen species (ROS) play an important role as signaling molecules in cellular metabolism and participate in the intercellular signaling-mediated system of acquired resistance (Xie et al., 2019; Wani et al., 2021). In particular, the synthesis of  $H_2O_2$ , which occurs under stress conditions, is a signal for triggering a number of molecular, biochemical, and physiological processes in cells, including plant adaptation to them by activating antioxidant defense systems (Nowicka, 2022; Demidchik, 2015; Karpets et al., 2015).

Studies by a number of authors (Luo et al., 2016; Zhou et al., 2016; Ding et al., 2017; Flores-Cáceres et al., 2023; Romero-Puertas et al., 2018) indicate that the plant uses some mechanisms to combat Cd toxicity, including Cd exclusion, active excretion, limited Cd distribution in sensitive tissues, chelation with organic molecules, compartmentalization in the cell wall, and vacuoles. In our case, under polluted conditions, the content of LPO products in the assimilation apparatus of poplar is higher than in control conditions. That is, in cultivars growing in areas contaminated with heavy metals, LPO processes are significantly activated. At the same time, the amount of primary lipid peroxidation products in poplar in the area of severe pollution increases maximally (up to 2.1 times) in “Tronco.” The largest amount of dienketones (by 40–80%), among the studied species and cultivars of poplar, is also formed in “Tronco.” A peculiarity of the stress effect of heavy metals on poplar is the absence in the cultivars “Lvivska” and “Hradizhka” of an increase in the formation of TBARS with an increase in the level of accumulation of heavy metals in Pivn-HZK conditions in comparison with plants on CHZK. Considering that the above indicator is considered by most authors as a marker of the level of stress on organisms, and the established fact of high accumulation of heavy metals, the cultivars “Lvivska” and “Hradizhka” can be used to create effective phytofilters with high levels of heavy metal pollution at industrial sites.

## Conclusion

As a result of comprehensive studies of the accumulation of heavy metals by various species and cultivars of poplars and the course of LPO processes, the peculiarities of the translocation of copper, zinc, nickel, cadmium, and lead to the assimilation organs and the formation of primary and secondary LPO products were ascertained. The studied poplar taxa can be divided into two groups: the first includes “Lvivska” and “Hradizhka,” which are characterized by high metal-accumulating ability and

greater resistance to the effects of toxicants; the second includes *P. italica*, *P. deltoides*, and “Tronco,” which accumulate less heavy metals and are not resistant to their effects. Based on the results obtained, it can be stated that the “Lvivska” and “Hradizhka” poplar cultivars have physiological and biochemical mechanisms that cause greater accumulation of some heavy metals and have, compared to other studied poplars, a lower level of LPO processes and, accordingly, have greater potential resistance. However, it should be noted that highly hazardous heavy metals are more characterized by a “cumulative” effect than less toxic ones. This conclusion is also confirmed by higher (by 25–90%) values of the enrichment factor the metal for in the leaves of the above-mentioned poplar taxa under conditions of moderate pollution. Whereas under conditions of severe pollution, *P. italica* and *P. deltoides* cannot compete with the cultivars “Lvivska” and “Hradizhka” in terms of resistance to heavy metals and can be used to create effective phytofilters under high levels of heavy metal pollution at industrial sites.

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