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ENVIRONMENTAL POLLUTION IN SOIL, PLANT LEAVES, AND ROOTS USING BIOINDICATION IN HAZELNUT FIELDS

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SUMMARY

This research sought to investigate the pollution by heavy metals in soil and plants (hazelnut trees and grass plants) in natural biotopes of the Oguz Region, Azerbaijan. Besides the influence of pollution, the study considered variations in fluorescent parameters. The main task was to identify the most sensitive tree species to pollution indicators. An integrated approach to the analysis of pollution from transport gases and solid industrial waste seemed crucial and relevant for the selected hazelnut fields. The decreased ratio of Chl a+b/carotenoids in both hazelnut cultivars (Oily Hazelnut and Ata Baba) authenticates that carotenoids perform a protective function from the natural and anthropogenic factors. More sensitive cultivars of hazelnuts (Oily Hazelnut and Ata Baba) used for research mainly served landscaping purposes (as informal hedges, windbreaks, and erosion control). The study advised planting these cultivars away from roads and even in the second row after bushes as more resistant species to pollution. This is because with increased pollution from exhaust gases (CO_2, CO, and HC), trees become weaker than those away from the roads.

Keywords: Heavy metals, chlorophyll, bioindicators, soil samples, carotenoids, environmental pollution

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Key findings: The results revealed the tree species, particularly cultivars Oily Hazelnut and Ata Baba, proved sensitive to environmental pollution in Azerbaijan due to heavy metals and air pollutants. These tree species entailed studies for morphological, physiological, and biochemical variations under polluted conditions and succeeded in their identification as useful bioindicators for environmental monitoring.

INTRODUCTION

Preserving biological diversity in the context of increasing environmental pollution and technogenesis requires developing some indicators beneficial in assessing the state of ecosystems and landscapes with the influence of anthropogenic environmental factors (Bunyatova *et al.*, 2025). Various crop plants and trees absorb these heavy metals through the root system and accumulate them in their tissues, leading to physiological and biochemical stresses, such as oxidative damage, altered pigment composition (carotenoids, phycobilins), and impaired metabolic pathways (Breure and Zechmeister, 2003).

In plants, these physiological and biochemical stresses result in weakened growth and development, toxicity symptoms in leaves and roots, and, eventually, reduced productivity. Grass and herbaceous plants, along with tree species, serve as effective bioindicators by reflecting pollution levels through measurable variations in morphology, pigment content, and biochemical markers (Blanco, 2023; Bunyatova *et al.*, 2025). The most relevant is the development of biological indicators in the process of bioindication studies to assess the state of the environment, with certain properties and characteristics of plants used (Hasanova *et al.*, 2025).

Continuous monitoring of hazelnut plant characteristics helps us to quantitatively assess variations in the environment with the influence of anthropogenic load in the existing area. The development of a biological indicators' system (highly resistant microorganisms) is a pressing task in urban eco-diagnostics, requiring immediate attention (Breure and Zechmeister, 2003). Bioindication indicators can work as an effective basis for assessing the state of the natural environment, which, in the future, will also become reliable bioindicators of sustainable development of nature management. Consequently, it complements the traditional method of environmental assessment of the territory using the maximum permissible concentrations (Clements, 1920; FAO and UNEP, 2019; Hasanova *et al.*, 2025).

Bioindication allows scientists to assess the levels of chemical substances in the plant leaves, as well as their direct negative effects on life processes of plants. In soils, the heavy metal accumulation and pollution can also have a robust impact on plant life processes. Under favorable hydrometeorological conditions, a considerable enhancement resulted in the transfer of more microelements from the soil cover to the aboveground parts of crop plants and trees (Kazeev and Kolesnikov, 2012; Hasanova *et al.*, 2021; Ismayil *et al.*, 2025).

Phytoindication is one of the areas of biomonitoring, which can determine the ecologically significant loads based on the reactions of crop plants grown in that specific environment. Soil is one of the basic components of a phytocenosis, playing an important environment-forming role in these vital processes that take place in crop plants in interaction with the environment. The growth and development of the flora are directly dependent on soil conditions, which, in turn, will transform with the phytocenosis living on it (Kazeev and Kolesnikov, 2012; Ismayilova *et al.*, 2025).

The study of chlorophyll fluorescence parameters in tree leaves opens up the possibility of rapid assessment of variations in the physiological state of plant stands with the influence of anthropogenic factors in large cities. Incorporating this technology into monitoring systems will allow us to predict the growth conditions of both plantings as a whole and as an individual tree. The plant's photosynthetic activities based on chlorophyll fluorescence can serve as a reliable indicator of the functional state of plants and adequately reflect the level of stress in their environment (Kabata-Pendias, 2011; Kazeev and Kolesnikov, 2012). Therefore, the presented study sought to investigate the pollution by heavy metals in soil and plants (hazelnut trees and grass plants) in natural biotopes and the variations in fluorescent parameters with the influence of pollution in the Oguz Region, Azerbaijan.

MATERIALS AND METHODS

Field work

Objects of the study were soil, trees, and grasses grown in an area with the highest atmospheric pollution from vehicle exhaust gases. The detailed field studies began from 2022 to 2024 (Macnunlu *et al.*, 2025; Mirzezadeh *et al.*, 2025). The assessment of phytocenoses employed test plots, soil, and ecological conditions—based on excavations in a place with identical soil test descriptions (Mammadova *et al.*, 2024). The layout and descriptions of soil sections followed the FAO methodology directly in the field and in the laboratory at the Institute of Soil Science and Agrochemistry, Baku, Azerbaijan.

The secondary objects of the study were the leaves of hazelnut trees and dominant grass species in the Oguz Region, Sheki-Zagatala economic region, Azerbaijan. Standard methods served for the selection, transportation, storage, and preparation of plant samples. Leaf sample collection used the average sample method—at the height of 1.5–2.0 m from the ground surface for trees on the outer side of the crown along the circumference (out sizes) (Nasirova *et al.*, 2022; Mammadova *et al.*, 2025). The collection of samples occurred during the main vegetation period (July-September) to determine the accumulative capacity of plant leaves in urban conditions. Up to the possible extent, choosing the plants of the same age for sampling ensued.

Bioindicators

For biochemical analysis, the plant leaves used had no petioles. In the agro-laboratory, the plant leaf samples incurred thorough washing with distilled water to remove dust and soil particles before being brought to an air-dry state. Wet decomposition (chemical preparation) of samples proceeded to determine the concentration of biochemical elements in plant leaves (Rascher *et al.,* 2000). Biochemical elements with concentrations below the detection limit, as well as those with content not meeting the control criteria according to internal and external standards and interlaboratory comparison, did not pass screening. In plant leaves, the ash content determination used the generally accepted method.

Statistical analysis

The recorded data in field and laboratory compilation and analysis used the STATISTICA 8.0 and Microsoft Excel programs. The biochemical element accumulations in tree leaves and plant roots under urban conditions underwent assessment using the concentration coefficient (Cc). It is the ratio of the element content in plant leaves and roots to its concentration in plant leaves grown in background areas (Shukurov *et al.*, 2025b). The studies took place at different times on fluorimeters, recording the parameters of both delayed fluorescence (relative delayed fluorescence parameter, zero fluorescence–F_0) and fast fluorescence (relative electron transport, quantum yield–Fv/Fm). The delayed fluorescence parameters' recording continued on the Foton-10 device, a fluorimeter. The data from the delayed fluorescence measurements came from the summer of 2022.

RESULTS AND DISCUSSION

In the grass cover, the predominant representative species were *Aegopodium podagraria, Mercurialis sp., Urtica dioica*, *Asperula sp., Fragaria vesca Linnaeus,* *Gramineae,* and *Glechoma hederacea*. No detection of groundwater was evident in the soil profile, noting a better groundwater drainage system, and the groundwaters were not open. Local and violent boiling from hydrochloric acid was notable at 115 cm (Shukurov *et al.*, 2025a).

The initial fluorescence values (F_0) obtained on Photon-10 appear in Figure 1a-c. When an inhibitor acts upon the chlorophyll or any pollutant (heavy metals, harmful gases, industrial waste, vehicle emissions, and pesticides), this indicator considerably decreases to values of 0.2–0.1, and that corresponds to a strong suppression of photosynthesis, practically to a minimal level. For this study, the selected sampling areas are available in Figure 2. The luminescence measurement of each cuvette was for two preset light and time modes.

When the integrity of the chloroplast membranes reached a compromise with a decrease in photosynthetic electron transport due to the impossibility of developing a proton gradient on the thylakoid membrane, this reduced the amplitude of the fast components significantly. In the low-light mode, slow components of the attenuation of delayed fluorescence mainly represented the luminescence; however, its intensity depends on the oxidation-reduction state of the primary acceptor of the photosystem of two chloroplasts (Steven, 2017). As a result, when suppression of photosynthetic electron transport transpires, the two parameters' ratio of delayed fluorescence decreases several times with an unfavorable effect on the tested plants.

Considering the direct connection between the quantitative distribution of heavy metal in soils and plant roots (hazelnut roots accumulate various heavy metals: iron-Fe, manganese-Mn, zinc-Zn, copper-Cu, nickel-Ni, chromium-Cr, lead-Pb, and cadmium-Cd), the possibility of using bioindicators established for plant roots to assess the ecological state of territories in the impact zone of solid municipal waste landfills became a consideration (Tomasev, 2005). As an evaluation criterion, using the concentration coefficients (Cc) of these metals in roots of indicator plants (*Urtica urens* L., *Artemisia absinthium* L., and *Tanacetum vulgare* L.) entailed calculation relative to the content of these metals in plant roots in the surveyed background areas.

The distribution pattern analysis of Cc values showed the soils of the surveyed areas were generally within the range of 1–5. Except for several zones of increased local pollution, the Cc values ranged from 5 to 10 and even above 10. For plants, the Cc values sustained more rankings, showing the areas with Cc values of 1–5 located closer to the body of the polygon. It had the form of spots (Figure 3), the elongation of which made it possible to judge the dominant directions of metal flow distribution.

Since determining the degree of metal accumulation in plants was by their bioavailability mainly in ionic form, the obtained results suggested that plants react to fresh flows of heavy metal ions in the environment. Bioindication using the Cc indicator in plant roots seems more informative and has certain advantages for assessing the effect of solid municipal waste landfills on the current ecological state (2025) of the environment. Bioindication can be a more cost-effective and time-efficient method for monitoring than extensive chemical analyses of soil and water, which can be complex and require different techniques for different compounds. By identifying critical knowledge gaps, particularly in the areas of field effectiveness, economic modeling, and regulatory integration, this article contributes to a more realistic and practical understanding of bioindication (Verdiyeva *et al.*, 2025).

On the location of halos, the obtained results identified the main routes of pollution migration from the territory of the storage zone. These also determined the gaps in the system of protective structures of the solid household waste landfill. However, such types of findings could not aid the formulation by using traditional methods of environmental assessment (Kazeev and Kolesnikov, 2012). In these circumstances, a project succeeded in its development and implementation to restore the integrity of the system of engineering

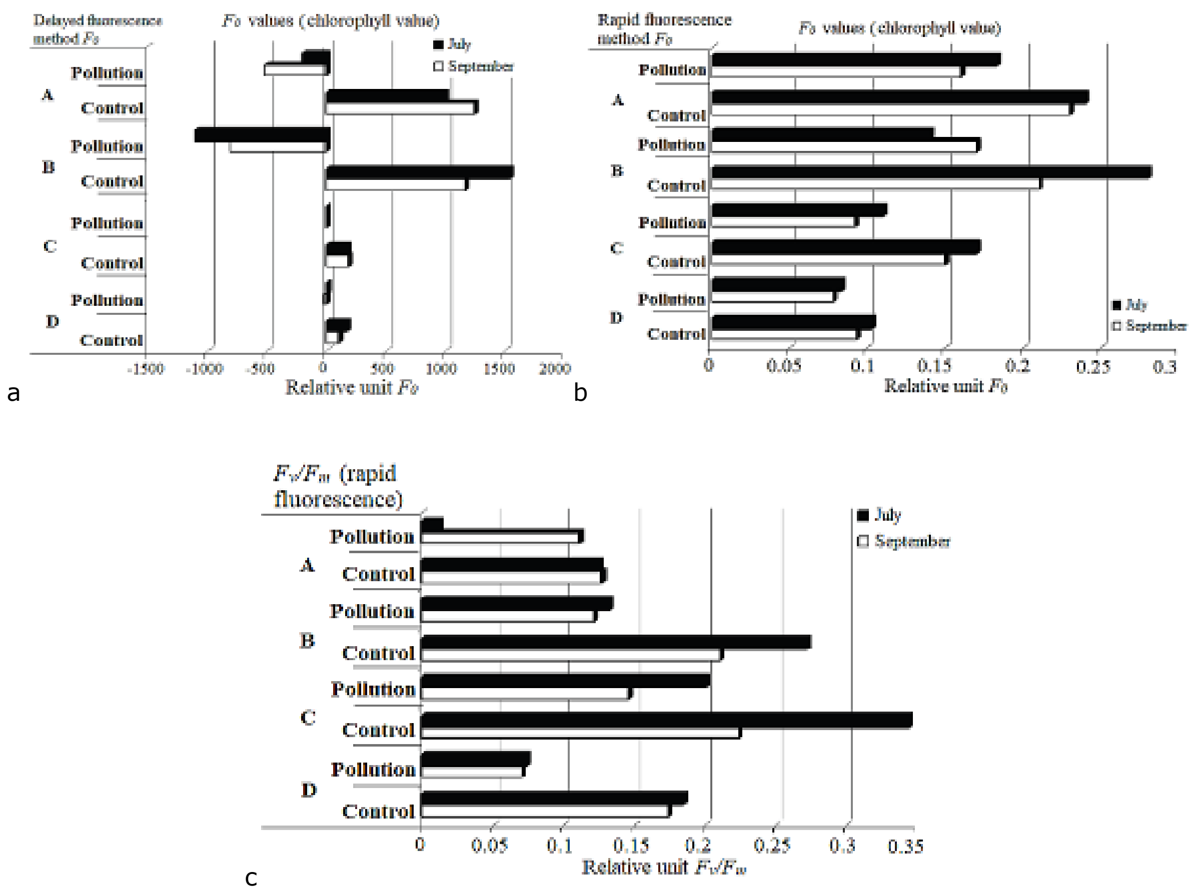

Figure 1. The chlorophyll value for soil species (A - mountain forest brown; B - forest gray-brown) and tree species (C - Oily Hazelnut leaves; D – Ata Baba hazelnut leaves) in the control area and in the contaminated area (a) on 'Photon - 10', (b) on 'RAM - 210', and (c) Quantum yield parameter characterizing rapid fluorescence for soils and hazelnut trees.

Figure 2. Testing in Oguz Region, Azerbaijan during 2024 (40.9470910, 47.6338680).

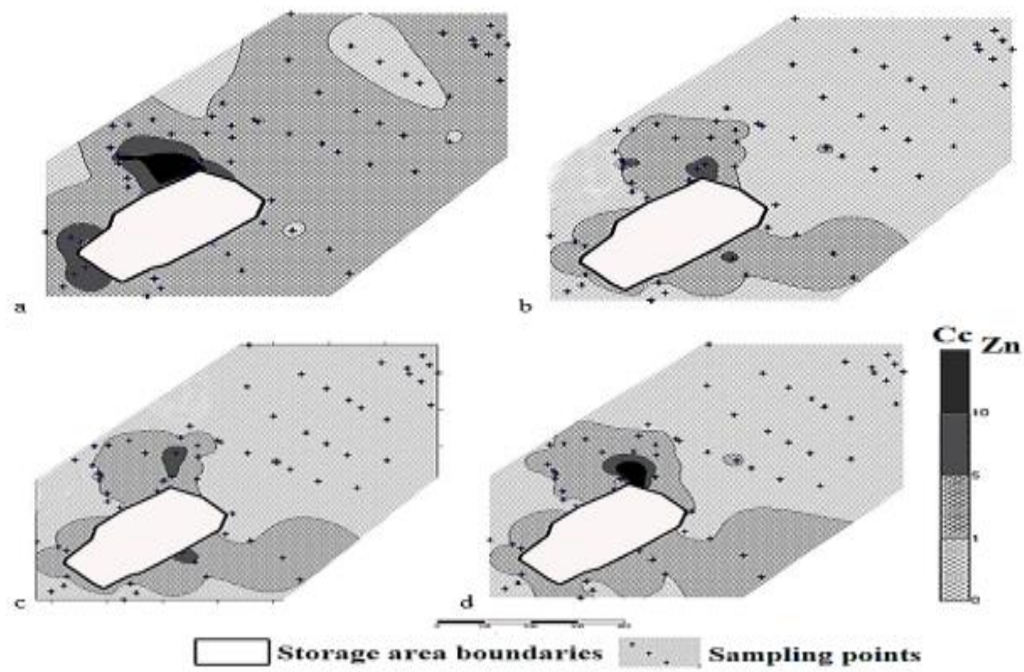

Figure 3. Distribution scheme of Cc (Zn) in soils and plants at the testing site (Oguz Region [Filfilli village], Azerbaijan), a) Soil, b) *Urtica urens* L., c) *Artemisia absinthium* L., and d) *Tanacetum vulgare* L.

protective structures of the storage zone. Moreover, it sought to assess the parameters of self-cleaning and self-restoration in the ecosystem.

Analysis of plant roots for heavy metal concentrations revealed a direct connection between the elevated levels of Fe, Zn, Cu, Pb, Cr, and Ni concentrations in the soils adjacent to the polygons. Likewise, in the levels of corresponding heavy metals in plants growing on them, primarily in their roots. In the overwhelming majority of tests, in plant roots, the metal content decreased in the series Fe>Mn>Zn>Cu>Pb>Cr>Ni, which corresponded to general patterns of quantitative distribution of heavy metals in soils. In this context, the plants reached an arrangement in the order, i.e., *Urtica urens* L. >*Artemisia absinthium* L. >*Tanacetum vulgare* L., according to Fe and Mn accumulation levels in roots. Meanwhile, according to Zn, Cu, Cr, and Ni, the plants were in the order of *Artemisia absinthium* L. >*Urtica urens* L. >*Tanacetum vulgare* L. (Hasanova *et al.*, 2025).

One should note that in the heavily contaminated area with lead (2139 mg/kg in soil), the Pb content in roots of *Artemisia absinthium* L. is 477 mg/kg, and in *Urtica urens* L., it is 261 mg/kg of dry biomass. This revealed a high degree of these plants' involvement in the Pb cycle in the environment. For the significance of the plant as an accumulative bioindicator, the important quantitative criterion is the coefficient of biological accumulation of metals (CBA) in the soil/root system, calculated as follows:

$$CBA = A/B$$

Where A is the metal content in the root biomass (mg/kg) and B is the metal content in the soil (mg/kg).

The presented results further exhibited that in most of the surveyed areas, the values of the CBA in plant roots of *Urtica urens* L., *Artemisia absinthium* L., and *Tanacetum vulgare* L., etc., remained within relatively narrow limits (0.2–0.5 for Fe, 0.1–0.4 for Mn, 0.4–0.9 for Zn, 0.3–0.8 for Cu, 0.6–2.6 for Cr,

Table 1. Pigment content in hazelnut leaves of the third year of life (mg/g fresh weight).

Indicators	Control	Filfilli village (Oguz) (41°07′01″ N; 47°35′26″ E)	Padar village (Oguz) (0°57′42″ N; 47°30′54″ E)
Oily Hazelnut variety			
Chl a	0.561±0.11	0.710±0.05	0.591±0.10
Chl b	0.229±0.04	0.246±0.02	0.241±0.04
Carotenoids	0.252±0.05	0.358±0.02	0.258±0.04
Chl a / Chl b	2.46±0.27	2.89±0.13	2.47±0.30
Chl (a + b) / carotenoids	3.14±0.12	2.92±0.06	3.23±0.24
Hazelnut variety Ata Baba			
Chl a	0.569±0.11	0.740±0.05	0.637±0.04
Chl b	0.236±0.04	0.260±0.02	0.207±0.02
Carotenoids	0.253±0.05	0.337±0.02	0.361±0.02
Chl a / Chl b	2.41±0.26	2.85±0.13	3.08±0.12
Chl (a + b) / carotenoids	3.19±0.10	2.96±0.10	2.34±0.07

0.6–1.6 for Ni, and 0.4–1.2 for Pb). This further provides the ground to consider these plants as objects that meet the goals of accumulative bioindication in contaminated areas (Mammadova *et al.*, 2025).

The ratio of Chl a+b/carotenoid*s* usually increases with the influence of anthropogenic load. In this case, the decreased ratio of Chl a+b/carotenoid*s* in both hazelnut cultivars (Oily Hazelnut and Ata Baba) authenticates that carotenoids perform a protective function, both from natural and anthropogenic factors, regardless of the adaptability of the tree to the growing conditions (Table 1) (Mammadova *et al.*, 2025). The same pattern applies to the total content of all pigments. It should decrease under the influence of environmental pollution. The two hazelnut cultivars (Oily Hazelnut and Ata Baba) have a similar mechanism for adapting the photosynthetic apparatus to anthropogenic loads (Mirzezadeh *et al.*, 2025). However, all that occurred due to a considerable increase in chlorophyll a and carotenoids.

Notably, the introduced species (different varieties of the European hazelnut [*Corylus avellana* L.])displayed a better adaptation and most likely has greater sensitivity and resistance to various anthropogenic loads. Its indicators illustrated different ranges of environmental pollution. This recognized the species with such properties served as objects for studying the content of pigment composition. The pigments' content varies dynamically due to the effect of natural and anthropogenic factors and depending on the species specificity in relation to the total pigment content (chlorophylls, anthocyanins, carotenoids). Additionally, the specific reaction of pigments to the environmental factors' influence, seasonal dynamics (summer-autumn), timing and duration of vegetation, age of leaves (hazelnut), features and arrangement of leaves on the tiers of the tree, and the plant age itself. Pigment content changes with the seasons due to shifts in light, temperature, and other factors. For example, carotenoids can act as photoprotectors (shields) in summer and additional light collectors in winter. The timing and duration of a plant's vegetation period, along with the age of the plant itself, affect pigment content (Makki *et al.*, 2025; Bunyatova *et al.*, 2025).

Based on the soil samples' analysis results, establishing more than 2-fold excesses of background contents emerged for elements such as Cu, Zn, As, Cd, and Pb. According to the value of the radial differentiation coefficients, natural sources (parent rocks) of Cr, Cu, and Zn anomalies reached their establishment; for As, Cd, and Pb, their anthropogenic origin was evident in the study area. The total pollution indicator value, as calculated by Cc (≥ 1.5), the elements As, Cd, Pb, Zn, and Cu were within the permissible pollution category. With the help of pigment composition, it is possible to assess the state of the environment. The pigment values as a bioindicator can be effective for conducting geosystem (landscape) and ecological-geographical analysis (Hasanova *et al.*, 2025). These values will further allow us to assess the environmental situation of the territory both within the framework of environmental monitoring and the development of various measures to improve the environment and sustainable urban development. Sustainable urban (large cities) development requires integrating these environmental improvements with economic and social goals through strategies like improving green spaces, managing waste, reducing pollution, and implementing smart land use planning and green transportation. A holistic approach is essential that considers cross-sectoral impacts and integrates diverse knowledge for effective and socially acceptable solutions (Makki *et al.*, 2025).

CONCLUSIONS

Bioindication studies have a prognostic value and allow for more or less adequate assessment of the degree of anthropogenic impact on ecosystems. However, these assessments entailed theoretical development and substantiation, which can be carried out at the population and ecosystem levels. The most sensitive period in a plant's life to pollution showed an association with the end of the dormant period in July-September, when depletion of the plant's sugar reserves occurs. With increased pollution from exhaust gases, trees become weaker than those far from the roads.

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