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ANALYSIS THE STATE OF ENVIRONMENT BY BIOLOGICAL METHODS

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INTRODUCTION

The problem of environmental pollution is one of the most acute environmental problems of the whole world. Man has long considered the surrounding natural environment mainly as a source of raw materials (resources) necessary for his needs. At the same time, a large part of the resources taken from nature is returned to nature in the form of waste. Most of this waste and pollution is generated in cities. In cities, goods and people are continuously transported by railways and highways. All types of transport heavily pollute the atmosphere with exhaust gases containing substances harmful to human health.

In every modern city, a lot of industrial and household waste is generated as a result of human activities. Human activity leads to constant pollution of the natural environment: atmospheric air, natural waters and soils. The scale of pollution of soil cover and atmospheric air of urbanized territories by its emissions is about 60-71%. Among the heavy metals contained in vehicle emissions are lead and cadmium, which have a negative impact on green spaces.¹

They quickly accumulate in plants, retain toxicogenic properties for a long time, and will have an expressed negative effect and impact on vegetation growing near highways.²

Biological methods for assessing the quality of the environment allow to analyze the total effect of toxicants on biological test objects of plant and animal origin. ³ European countries have already assessed the effectiveness of complex control for both parameters (biotic and abiotic). Normative documents and standards provide for biological control in Germany, Poland, Austria, Great Britain, Hungary, Finland, etc. ⁴

¹ Зеркалов Д. В. Екологічна безпека: управління, моніторинг, контроль: навч. посібник. Київ: КНТ, Дакор, Основа, 2007. 412 с.

² Гродзинський Д. М., Шиліна Ю. В., Куцоконь Н. К. Застосування рослинних тест-систем для оцінки комбінованої дії факторів різної природи. Київ : Фітосоціоцентр, 2006. 60 с.

³ Franiel I. Fluctuating asymmetry of Betula pendula Roth. leaves – an index of environment quality. *Biodiv. Res. Conserv.* 2008. Vol. 9–10. P.7–10.

⁴ Barabash O., Lozova T., Kozlova T. Assessment of the urban environment quality in Kyiv. *Acta Carpathica*. 2017. № 28. P. 5–12.

At the end of the 20th century – the beginning of the 21st century in Europe and the USA there was a tendency to develop biological assessment methods within the ecosystem integrated approach.

Natural ecosystems are considered as a whole, taking into account abiotic and biotic factors, as well as their interrelationships. This method of assessment is approved by the Helsinki Convention. ⁵ Biological assessment primarily reflects the state of the ecosystem as a whole, and only indirectly the quality of the environment.

Plants are promising test objects for biomonitoring, due to their high sensitivity to environmental changes under the influence of anthropogenic factors. With the help of plants and their groups, it is possible to assess the effect and consequences of anthropogenic influences: disturbance of natural landscapes, pollution of air, water environment and soils; to substantiate measures for the organization of environmental monitoring.^{6,7}

The peculiarity of plants is their availability, ease of cultivation, high sensitivity to a number of pollutants, the possibility of assessing the total effect of harmful substances on the ecosystem. The studies carried out with their help are short-term, easy to perform, and do not require complex laboratory equipment.⁸

Due to genetic heterogeneity, plants show a specific reaction to the action of pollutants, in particular, certain species react to the action of only one pollutant, others to the action of two or more substances, some do not react at all, or their reaction is very weak or ambiguous.

Biotests to determine the level of environmental pollution by pollutants are reduced to assessing the degree of change in the morphological indicators of the test plant. Various approaches and scales of experimental phytotesting are known. Mainly, phytotests can be combined into three groups of methods: laboratory, vegetation and microplot.⁹

Currently, the approach using several types of plants as phytotests is quite common, which allows determining the minimally active toxic component in conditions of complex environmental pollution. Therefore, despite the rather large number of plant test systems, the question of the possibility of using

⁵ Машталер О. В. Біомоніторинг видами *Bryophyta* техногенно трансформованого середовища південного сходу України: автореф. дис. на здобуття наук. ступеня канд. біол. наук: спец. 03.00.16 "Екологія". Дніпропетровськ, 2007. 20 с.

⁶ Матейчик В. П. Методи оцінювання та способи підвищення екологічної безпеки дорожніх транспортних засобів: монографія. Київ: Нац. транспортний університет, 2006. 216 с.

⁷ Velickovi M. Developmental stability in Tilia cordata leaves. *Period biol.* 2010. Vol. 112, № 3, P. 273–281.

⁸ Лаптєв О. О. Екологічна оптимізація біогеоценотичного покриву в сучасному урболандшафті: монографія. Київ: Укр. екол. акад. наук, 1998. 208 с.

⁹ Куцоконь Н. К. Рослинні тест-системи для визначення цитотоксичності. Вісник НАН України. 2010. № 4. С. 48-52.

plants for biotesting of a certain class of substances remains open and requires the development of optimal test systems for specific pollutants. ¹⁰

All natural environments can be bioindicated with the help of plants. Indicator plants are used to assess the mechanical and acidic composition of soils, their fertility, moisture and salinity, the degree of mineralization of groundwater and the degree of atmospheric air pollution by gaseous compounds, as well as to identify the trophic properties of water bodies and the degree of their pollution by pollutants. For example, the content of lead in the soil is indicated by the species of fenugreek (Festuca ovina), broom (Agrostis tenuis), zinc – the section of violet (Viola tricolor), copper and cobalt – smolivka (Silene vulgaris), many cereals and mosses.

Sensitive indicators indicate the presence of pollutant in the air or in the soil by early morphological reactions – a change in the color of the leaves (appearance of chlorosis; yellow, brown or bronze color), various forms of necrosis, premature wilting and falling of the leaves. In perennial plants, pollutants cause a change in size, shape and number of organs, the direction of shoot growth or a change in fertility. Such reactions are usually nonspecific.

Therefore, the use of woody plants as accumulators of urbotechnogenic pollutants and bioindicators of the ecological state of the environment serves as a theoretical basis for the creation of ecologically effective cultures of phytocenoses in urboecosystems.

Exploring the bioindicative properties of woody plants, B. A. Revych believed that they manifest due to the ability to accumulate heavy metals in biomass and destroy the toxic mutagenic effects of industrial pollutants.¹¹

Biological indication is a system of assessing the state of the environment based on physiological, morphological, ecological changes of indicator plants that respond sensitively to changes in environmental factors ^{12,13,14}. Therefore, a necessary prerequisite for the indication of anthropogenic load in urbanized ecosystems is the use of woody plants as biological indicators of the quality of the environment and accumulators of negative substances.

¹⁰ Осика В. Ф. Якість вимірювань складу та властивостей об'єктів довкілля та джерел їх забруднення: монографія. Київ : Наука, 2001. 663 с.

¹¹ Парпан В. І. Деревні рослини як кумулятивні індикатори забруднення довкілля важкими металами. *Наук. зап. Тернопільського нац. педагогічного ун-ту.* 2008. № 4 (38). С. 93–97.

¹² Гойванович Н. К, Дрозд І. Ф. Вплив антропогенного навантаження на морфометричні показники рослин-індикаторів в умовах Передкарпаття України. *Розвиток природничих наук: проблеми та рішення* : матеріали Міжнародної науково-практичної конференції, 27-28 квітня 2018 р. Брно, 2018. С. 16–19.

¹³ Гойванович Н., Юзьвяк М., Біла В. Оцінка стану навколишнього середовища м. Старий Самбір за морфо-біологічними змінами *Tilia cordata. Екологічні науки*. 2022. № 3(42). С. 211–216.

¹⁴ Hoivanovych N., Pavlyshak Ya., Antonyak H. Influence of anthropogenic load in the city of Stryi on the functional state of photosynthetic apparatus of plants-indicators. *Acta Carpathica*. 2019. № 31–32. P. 52–59.

The aim of the work is an analysis of the state of the environment using biological methods using the example of the city of Staryi Sambir using indicator plants: *Tilia cordata, Acer platanoides* and *Carpinus betulus*.

1. Materials and methods

Laboratory tests were performed in the laboratory "Microbiology and Genetics" Drohobych Ivan Franko State Pedagogical University at the end of the growing season (2020-2021.). To solve the tasks was used weighing method with modification L. Dorogan with the establishment of the conversion factor. Detection of necrotic and depigmentation damage to the leaves of woody indicator plants was carried out visually. Classification of the detected lesions of the leaf was performed using the scheme proposed by R. Schubert. To determine the degree of divergence of leaf characteristics, certain foliar indicators were measured. This indicator characterizes the degree of asymmetry of the organism. For this indicator, a 5-point scale of deviation from the norm was developed, in which 1 point is a relative norm, and 5 points is a critical value with the following gradation: 1 point – up to 0.055; 2 points – 0.055–0.060; 3 points – 0.060–0.065; 4 points – 0.065–0.070; points – more than 0.070. Determination of the content of photosynthetic pigments was carried out using spectrophotometr method. ¹⁵

The calculation of emissions of harmful substances by motor vehicles was carried out using the generally accepted methodology.¹⁶ In order to determine the emissions of motor vehicles in urban conditions and further use the obtained data in the calculation of atmospheric pollution, the peculiarities of the distribution of the flow of motor vehicles in the city were studied and they were counted during the day at certain time intervals.

Assessment of the toxicity of water extracts of edafotopes in St. Sambir was carried out according to the method of A. I. Horova, in which the test object (*Allium cepa bulbs*) was placed in the test sample without prior root germination.¹⁷

Phytotoxicity was determined by measuring the length of roots (in mm) of *A. cepa* bulbs after the end of the experiment (after 4 days of growth) in the control and experimental variants, visually analyzing the turgor, color change, and shape of their root tips in the control and experimental variants.

Research area. For the convenience of conducting the work and increasing its objectivity, the studied territory was structured into separate monitoring points:

¹⁵ Осика В. Ф. Якість вимірювань складу та властивостей об'єктів довкілля та джерел їх забруднення: монографія. Київ : Наука, 2001. 663 с.

¹⁶ Купчик О. Ю. Викиди автомобільного транспорту як джерело забруднення атмосферного повітря міста Чернігова. *Молодий вчений*. 2015. № 2 (17). С. 17–20.

¹⁷ Горова А. І., Кулина С. Л. Оцінка токсичності грунтів Червоноградського гірничопромислового району за допомогою ростового тесту. Вісник Львівського університету. Серія Біологія. Львів, 2008. Вип. 48. С. 189-194.

Vidkryti Sertsia Park, Lystopadova Street, Lesya Ukrainka Street, Drohobytska Street, Lev Halytskyi Street, within which the places where 3 identical types of woody indicator plants grew were selected (see Fig. 1).



Fig. 1. Map of St. Sambir¹⁸

Vidkryti Sertsia Park is an old park that is a place for people to walk and relax. The following types of trees grow here: linden, maple, poplar, birch, spruce, etc. The park served as the background area.

Lystopadova Street is located near the railway. Noise generated by railway transport has a negative impact on people's health and local vegetation.

Lesya Ukrainka Street is located 500 m from the center, there is a local cemetery, the Starosambir district sanitation station, and a concentration of private houses and farms.

On the street Drohobytska there is a gas station 650 meters long. Green areas grow along the roadsides.

Lev Halytskyi Street is the central street that runs through the entire city. Its length is 3100 m, green areas are located on the roadsides along the entire street.

A park in the zone of suburban green spaces, which is not located in the area of concentration of enterprises and vehicle traffic St. Sambir, was chosen as the background area.

A set of biomorphometric methods and a qualitative assessment of pathological macroscopic morphological changes of the vegetative organs of

¹⁸ Карта міста Старий Самбір. URL : <u>https://www.google.com.ua/map/place</u>. (дата звернення: 01.07.2020).

the analyzed plants were used to study the reaction of woody plants to the stress factors of the habitat and to determine the degree of sensitivity of individual species to the complex of factors of urbotechnogenic nature.

Based on the analysis of literary sources, we chose the following plants: linden (*Tilia cordata*), maple (*Acer platanoides*), hornbeam (*Carpinus betulus*).

2. Results and disscusion

Morphometric changes in the leaves of woody plants in the conditions of the urban ecosystem of Staryi Sambir

Anthropogenic load of any intensity causes morphological and biochemical changes in woody plants, and their fixation and assessment allows to obtain reliable information about the ecological state of places where indicator plants grow, allows to analyze the state of pollution of a certain urboecosystem ^{19,20}. According to A. Bessonova's research, the main morphometric changes include the area of the leaf surface and the level of necrotic leaf lesions.

The study of the area of leaf blades of indicator plants: linden (*Tilia cordata*), maple (*Acer platanoides*), hornbeam (*Carpinus betulus*) was carried out during 2020-2021, at the end of the growing season. A direct relationship was established between the change in the morphometric parameters of tree species and the physical-geographical location and distance to the sources of environmental pollution within the urban ecosystem of Staryi Sambir. The results are presented in the table 1.

The maximum increase in the area of leaf blades was noted for Lystopadova Street and Drohobytska Street. Significant deviations of this indicator in the specified districts of St. Sambir can be caused by the overall effect of a man-made factor, which is especially pronounced in the area of Lystopadova Street, probably due to transport pollution of the environment.

Diagrams 1–3 present the area of leaf blades for each type of indicator. It should be noted that a species specific reaction is observed. The area of the photosynthetic surface increased the most for the linden (*Tilia cordata*) on the street. Drohobytska – more than 3 times.

¹⁹ Екологічні паспорти Львівської області за 2018-2021 р. URL: https://deplv.gov.ua/ekologichnyj-pasport/ (дата звернення: 05.09.2021)

²⁰ Регіональна доповідь про стан навколишнього природного середовища Львівської області (2018-2020 рр.). Міністерство захисту довкілля та природних ресурсів України. URL: https://menr.gov.ua/ (дата звернення: 04.06.2021)

Table 1

The area of leaf blades of woody plants in different areas
Staryi Sambir, 2020–2021

	Study ana	The true of tree	Leaf ar	rea, cm ²
n/p	Study area	The type of tree	2020	2021
		Tilia cordata	25,40±5,51*	43,3±3,6**
1	Vidkryti Sertsia park	Carpinus betulus	26,62±5,34*	21,88±2,60*
		Acer platanoides	49,18±6,54*	27,5±8,3**
		Tilia cordata	53,14±5,34*	48,8±4,95*
2	Lev Halytskyi street	Carpinus betulus	41,32±5,31*	24,9±3,06*
		Acer platanoides	66,27±8,6	52,3±2,24*
		Tilia cordata	51,88±8,7**	45,4±5,2*
3	Lesya Ukrainka Street	Carpinus betulus	27,47±5,47*	24,4±3,8*
		Acer platanoides	72,83±5,79*	49,7±8,2*
		Tilia cordata	57,12±4,75*	49,7±8,2**
4	Lystopadova Street	Carpinus betulus	36,44±29,8*	28,2±0,98*
		Acer platanoides	72,16±9,33*	43,7±6,1*
		Tilia cordata	57,85±5,82*	49,9±6,4*
5	Drohobytska Street	Carpinus betulus	31,04±2,8**	30,7±2,96*
		Acer platanoides	74,19±8,3**	54,4±4,2*

Note. In this and subsequent tables: *, **, *** – probability of differences between the control and experimental groups of animals (*– p<0.05; ** – p<0.01; *** – p<0.001).

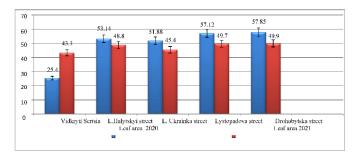
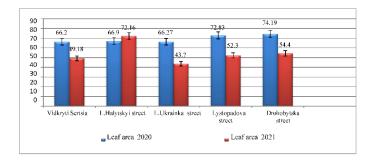
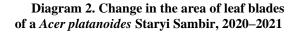


Diagram 1. Change in the area of the leaf blades of the heart-leaved linden (*Tilia cordata*) Staryi Sambir, 2020–2021





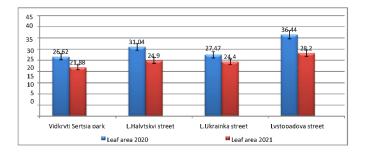


Diagram 3. Change in the area of leaf blades of *Carpinus betulus* in Staryi Sambir in 2020–2021

The increase in the area of the leaf blades of the studied species of woody plants compared to the background area can be placed in the following sequence:



Scheme 1. A series of increases in the assimilation organs of plants

According to Yu. G. Prysedskyi, the increase of assimilation organs in the studied plants can be considered as an adaptive reaction that ensures an

increase in the activity of gas exchange processes in conditions of environmental pollution. $^{\rm 21}$

The next stage of our research was the analysis of the integral degree of asymmetry of leaf blades of woody plants, because this feature is an important criterion for the sustainable development of flora in general, and is used as an important population feature in ecological studies. The results of measurements are given in table 2.

The greatest values of the degree of divergence of leaf traits of woody plants are characteristic of individuals that grew within local urban ecotopes with the highest level of anthropogenic pressure Lev Halytskyi and Drohobytska Streets, where the largest traffic is concentrated.

Table 2

	Relative mean difference between traits							
		2020		2021				
Study area	Tilia cordata	Acer Platano id es	Carpinus betulus	Tilia cordata	Acer platano ides	Carpinus betulus		
Vidkryti Sertsia Park	0,054	0,021	0,048	0,044	0,049	0,06		
Lev Halytskyi street	0,077	0,058	0,06	0,081	0,07	0,10		
Lesya Ukrainka street	0,057	0,059	0,043	0,043	0,08	0,06		
Lystopadova Street	0,056	0,071	0,06	0,10	0,06	0,08		
Drohobytska Street	0,092	0,04	0,09	0,08	0,08	0,11		

The degree of asymmetry of the leaves of woody plants in St. Sambir 2020–2021

Table 2 shows the minimum and maximum values of the divergence of traits and the integral index of leaf asymmetry of woody plants during 2020–2021. The analysis of research results shows that the maximum degree of divergence of traits for the *Tilia cordata* of Lystopadova Street (2021) is 0.1, which corresponds to the critical value of environmental pollution according to the Zakharov scale (5 points), for the sharp-leaf maple – for Lesya Ukrainka and Drohobytska streets (2021) – 0.08, which corresponds to the critical value of environmental pollution according to the *Carpinus betulus* (2021) – 0.11, which corresponds to the critical value of environmental pollution according to the Zakharov scale (5 points), for the *Carpinus betulus* (2021) – 0.11, which corresponds to the critical value of environmental pollution according to the Zakharov scale (5 points), for the *Carpinus betulus* (2021) – 0.11, which corresponds to the critical value of environmental pollution according to the Zakharov scale (5 points), for the *Carpinus betulus* (2021) – 0.11, which corresponds to the critical value of environmental pollution according to the Zakharov scale (5 points), for the *Carpinus betulus* (2021) – 0.11, which corresponds to the critical value of environmental pollution according to the Zakharov scale (5 points), for the *Carpinus betulus* (2021) – 0.11, which corresponds to the critical value of environmental pollution according to the Zakharov scale (5 points), for the *Carpinus betulus* (2021) – 0.11, which corresponds to the critical value of environmental pollution according to the Zakharov scale

²¹ Приседський Ю. Г. Адаптація рослин до антропогенних чинників : підруч. для студ. вищ. навч. закл. Вінниця: ТОВ "Ніланд-ЛТД", 2017. 98 с.

(5 points). In general, the most variable characteristics include the width of half of the leaf and the angle between the main vein and the second vein from the base of the second order. The smallest degree of divergence of woody plant traits is characteristic of the background plots.

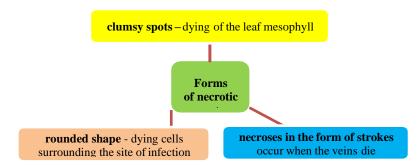
The dynamics of the divergence of traits and the increase in the level of asymmetry is species– specific, the most vulnerable species is the *Tilia* cordata, and the most resistant is the *Carpinus betulus*. A negative trend is observed, the degree of asymmetry increased almost 2 times during the observation period. If in 2020, the environment could be rated as 2–3 points for the state of tree species, which corresponds to the average value, then in 2021, the state of the environment is assessed as critical for almost all plants.

Thus, under the action of urbotechnogenic factors of the studied area, an increase in leaf asymmetry was observed in woody plants, which corresponds to the level of anthropogenic pressure at the monitoring points.

Study of the degree of damage to the leaf blades of tree species in Staryi Sambir

Anthropopression can cause the appearance of necrosis and various injuries on the leaves of plants that grow in a polluted environment. In general, violations of the structure of tissues and cells of leaves are the first signs that signal damage to tree species by various toxicants. According to Dmytryk P. M., "necrosis is dead, dry areas of the plant that are clearly separated from healthy t issues." Depending on the nature of the damage to the plant organism, the following forms of necrotic spots are distinguished.²²

Determining the percentage of damage to the leaf blade by necrosis is an important feature for assessing the state of the atmospheric air in the studied areas



Scheme 2. Forms of necrotic spots

²² Приседський Ю. Г. Адаптація рослин до антропогенних чинників: підруч. для студ. вищ. навч. закл. Вінниця: ТОВ "Ніланд-ЛТД", 2017. 98 с.

That is why the next stage of our research was to determine the percentage of the necrotic surface of the leaves of the indicator plants of the studied areas of the city of St. Sambir The obtained data are given and presented in the form of diagrams 4–6.

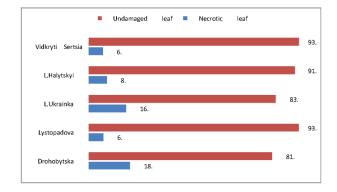


Diagram 4. The degree of necrotic lesions of the leaf blade of *Tilia cordata* in certain areas of St. Sambir 2020–2021

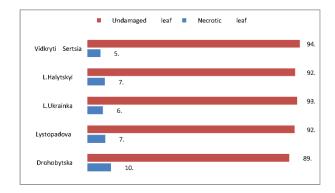


Diagram 5. The degree of necrotic lesion of the leaf blade of the *Acer platanoides* in certain areas of St. Sambir 2020–2021

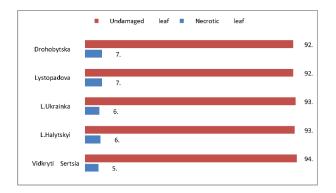


Diagram 6. The degree of necrotic lesion of the leaf blade of *Carpinus betulus* in certain districts of Staryi Sambir in 2020–2021

During 2020–2021, the maximum value of necrotic lesions of the leaf blade is typical for woody plants on the streets: L. Halytskyi (6.80-8.19%). Lystopadova (6.53–7.88%) and Drohobytska (7.4–18.4%), and the average – st. L. Ukrainka. In particular, the highest percentage of necrotic lesions on the surface of the leaves is characteristic of the Tilia cordata: S posh.=18.40% (Drohobytska St.), which is 2.9 times more compared to the control zone (S posh. = 6.38%, Vidkryti Sertsia park); the maximum value of necrotic lesions of the leaves of the Acer platanoides is S posh. = 10.30% (Drohobytska st.), which is 2 times higher compared to the control zone (S posh. = 5.63%, Vidkryti Sertsia park). The lowest values of necrotic lesions are characteristic of Carpinus betulus: S posh. = 6.3% (L. Ukrainka st.), S posh. = 6.8% (L. Halytskyi str.). The obtained results indicate that Carpinus betulus is the most resistant to anthropopression, and *Tilia cordata* is the most vulnerable species.²³ The increase in the necrotic surface is a consequence of the total load on the city's ecosystem. The highest level of necrotization was established on for all types Drohobytska street of indicator plants.

Pollutants cause the appearance of various forms of deformation damage in plants. In particular, they often lead to the deformation of leaf blades. The results are shown in table 3.

As can be seen in Table 6, the qualitative manifestations of necrotic and dechromation lesions of leaf blades are species-specific for each type of tree. The most characteristic types of necrotic lesions for *Carpinus betulus* are interveinal, *Tilia cordata* – point, *Acer platanoides* – spotted.

²³ Осика В. Ф. Якість вимірювань складу та властивостей об'єктів довкілля та джерел їх забруднення: монографія. Київ : Наука, 2001. 663 с.

Table 3

		Type of leaf blade damage						
Study area	The type of tree	Dotty	Blotchy	Inter veinal	Marginal	Apical	Type "fish Skeleton"	
	Tilia cordata	+	-	-	-	-	-	
Vidkryti Sertsia	Carpinus betulus	-	-	+	-	-	-	
park	Acer platanoides	+	+	-	-	-	-	
	Tilia cordata	+	-	+	-	-	-	
L. Halytskyi	Carpinus betulus	-	-	-	-	-	-	
street	Acer platanoides	-	-	-	+	-	-	
L.	Tilia cordata	+	-	-	-	-	-	
L. Ukrainka	Carpinus betulus	+	-	+	-	-	-	
street	Acer platanoides	-	+	-	+	-	-	
	Tilia cordata	+	-	-	-	-	+	
Lystopado	Carpinus betulus	+	-	-	-	-	-	
va street	Acer platanoides	+	+	-	-	-	-	
	Tilia cordata	+	-	+	-	-	-	
Drohobyts	Carpinus betulus	+	-	-	-	+	-	
ka street	Acer platanoides	-	+	-	+	-	-	

Forms of necrotic lesions of leaf blades of studied plants in the districts of Staryi Sambir in 2020–2021

The influence of environmental conditions in Staryi Sambir on the content of photosynthetic pigments in the leaves of indicator plants

Photosynthesis plays an important role in the life of plant organisms, as it enables the accumulation of organic substances from H_2O and CO_2 under the influence of sunlight. Accumulating toxic metals in the leaves, they lead to disruption of photosynthesis processes.

M. P. Merzlyak and N. V. Kapelyush proved that the combined effect of anthropogenic pressure is the cause of a sharp decrease in the content of chlorophyll and, as a result, slows down its biosynthesis.²⁴ Preliminary results

²⁴ Приседський Ю. Г. Адаптація рослин до антропогенних чинників : підруч. для студ. вищ. навч. закл. Вінниця: ТОВ "Ніланд-ЛТД", 2017. 98 с.

indicate the launch of gradational phenomena in plants (increase of assimilation organs, intensification of asymmetry, increase in the level of necrotization), the study of the state of the pigment composition of *Tilia cordata*, the *Acer platanoides*, and the *Carpinus betulus* in the conditions of pollution of urboecosystems will allow a detailed analysis of the physiological processes occurring in plants under by the action of harmful substances.²⁵

The research was conducted during 2020-2021, and the data was analyzed by years. The obtained calculations of the concentration of pigments in the leaves of the studied plants are given in the annex. Diagrams 7 - 9 show the concentration of chlorophyll a, b and carotenoids in the leaves of indicator plants.

The concentration of chlorophyll *a* in the leaves of the *Tilia cordata* ranges from 6.1 to 15.8 mg/l, however, the chlorophyll index in st. L. Halytskyi, L. Ukrainka and does not differ from the background plot. In the leaves of the *Acer platanoides*, the index of chlorophyll *a* varies within the range of 4.97-9.02 mg/l, and in the *Carpinus betulus* – 6.1-12.5 mg/l. In general, there is a tendency to decrease the concentration of chlorophyll *a* in the leaves of all indicator plants, the lowest values were recorded on the street. Drohobytska.

The assessment of the effect of anthropogenic pollution on the concentration of chlorophyll b in the leaves of indicator plants showed that the amount of this pigment is slightly reduced, this is best observed on the example of *Carpinus betulus*.

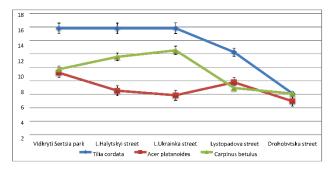


Diagram 7. The concentration of chlorophyll *a* in the leaves of woody plants in Staryi Sambir 2020

²⁵ Гойванович Н., Юзьвяк М., Біла В. Оцінка стану навколишнього середовища м. Старий Самбір за морфо-біологічними змінами *Tilia cordata. Екологічні науки.* 2022. № 3(42). С. 211–216.

The concentration of carotenoids in the leaves of indicator plants almost does not change depending on the place of growth. This may indicate the stability of this pigment before anthropogenic stress.

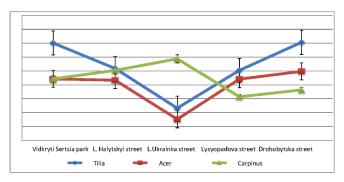


Diagram 8. Chlorophyll *b* concentration in leaves of woody plants in St. Sambir 2020

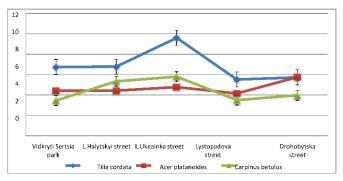


Diagram 9. Concentration of carotenoids in leaves of woody plants Staryi Sambir 2020

Next, the content of pigments in the studied material was determined, taking into account the volume of the extract and the weight. The results are presented in table 4.

Table 4

	of indicator plants in Staryl Sambir 2020								
n/p	Study area	Indicator type	Chlorophyll a	Chlorophyll b	Carotenoids				
	Widlemsti	Tilia cordata	1,975±0,131*	0,65±0,043**	$0,84{\pm}0,056{**}$				
1	Vidkryti Sertsia park	Acer platanoides	1,15±0,076**	0,55±0,037*	0,55±0,037**				
	Settsia park	Carpinus betulus	1,22±0,081**	0,55±0,022*	0,55±0,037*				
	I II-l-st-l-s-	Tilia cordata	2,78±0,011*	0,59±0,039**	1,05±0,042**				
2	L. Halytskyi	Acer platanoides	0,82±0,033**	0,54±0,036**	0,43±0,017*				
	street	Carpinus betulus	1,45±0,058*	0,63±0,042**	0,67±0,027*				
	I Illensieles	Tilia cordata	4,35±0,29**	0,29±0,012*	1,20±0,08**				
3	L. Ukrainka street	Acer platanoides	0,73±0,048**	0,19±0,01*	0,35±0,023**				
	succi	Carpinus betulus	1,57±0,10*	0,73±0,029**	0,73±0,049**				
	Lustanadava	Tilia cordata	1,53±0,061**	0,63±0,025**	0,69±0,028*				
4	Lystopadova street	Acer platanoides	$0,97{\pm}0,039*$	0,61±0,024**	0,52±0,021*				
	sueet	Carpinus betulus	0,87±0,035**	0,39±0,016*	0,41±0,016*				
	Duchchritelie	Tilia cordata	7,64±0,31**	0,88±0,035*	$0,72\pm0,029**$				
5	Drohobytska street	Acer platanoides	0,62±0,025*	0,39±0,016**	0,34±0,014*				
	sucei	Carpinus betulus	0,76±0,03*	0,45±0,018**	0,37±0,015**				

The content of pigments in the raw mass of the leaves of indicator plants in Starvi Sambir 2020

Note. In this and subsequent tables: *, **, *** – probability of differences between the control and experimental groups of animals (*– p<0.05; ** – p<0.01; *** – p<0.001).

The obtained results show that the content of pigments in the raw mass of the leaves of the indicator plants decreases.

To determine the dynamics of the concentration of photosynthetic pigments in the leaves of indicator plants in St. Sambir, their concentration was re-determined at the end of the growing season in 2021 and diagrams 10–12.

The concentration of chlorophyll *a* in the leaves of the *Tilia cordata* varies between 25.34-33.52 mg/l, but the chlorophyll index according to L. Halytskyi st., L. Ukrainka st. does not differ from the background area. In the leaves of the *Acer platanoides*, the index of chlorophyll *a* varies within the range of 10.3-17.62 mg/l, and in the *Carpinus betulus* – 20.5–27.4 mg/l. In general, there is a tendency to decrease the concentration of chlorophyll *a* in the leaves of all indicator plants, the worst indicators were recorded on the street. Drohobytska.

Based on the obtained results, we can see that the concentration of carotenoids in the leaves of the *Tilia cordata*, the *Carpinus betulus*, and the *Acer platanoides* is increasing compared to the values in 2020. This probably indicates a change in the state of the environment.

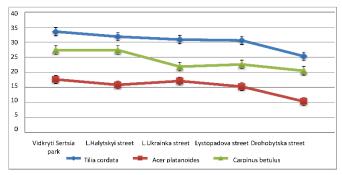


Diagram 10. Chlorophyll *a* concentration in leaves of woody plants St. Sambir in 2021

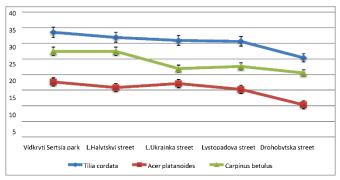


Diagram 11. Chlorophyll *b* concentration in leaves of woody plants St. Sambir in 2021

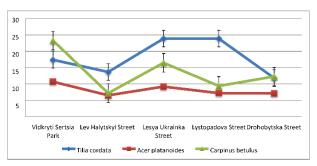


Diagram 12. Concentration of carotenoids in leaves of woody plants Staryi Sambir 2021

Based on the obtained results, we can see that the concentration of carotenoids in the leaves of the *Tilia cordata*, the *Acer platanoides*, and the *Carpinus betulus* is increasing compared to the values in 2021. This probably indicates a change in the state of the environment.

Table 5

n/p	Study area	Indicator type	Chlorophyll a		Carotenoids
шp	Study alea	mulcator type			
	Vidlemeti	Tilia cordata	3,86±0,26**	1,71±0,11*	1,4±0,09**
1	Vidkryti Sertsia park	Acer platanoides	1,98±0,13*	0,89±0,059**	$0,65\pm0,043*$
	Settsia park	Carpinus betulus	1,9±0,13**	2,9±0,12**	1,2±0,08**
	I Holytelari	Tilia cordata	3,99±0,16*	1,64±0,066*	1,52±0,06*
2	L. Halytskyi street	Acer platanoides	1,98±0,13**	$0,82{\pm}0,055*$	0,61±0,04*
	sueet	Carpinus betulus	2,74±0,11**	$0,9{\pm}0,06{**}$	0,9±0,06*
	L. Ukrainka	Tilia cordata	3,82±0,25*	2,99±0,12*	1,4±0,06**
3	L. UKrallika street	Acer platanoides	2,14±0,086*	1,15±0,05**	0,93±0,06*
	sueet	Carpinus betulus	2,56±0,10**	1,54±0,10**	1,04±0,04**
	Lustanadava	Tilia cordata	3,17±0,21**	$1,48\pm0,06*$	1,15±0,05*
4	Lystopadova	Acer platanoides	2,20±0,088*	1,3±0,09**	9,84±0,4**
	street	Carpinus betulus	2,83±0,19**	1,16±0,05**	0,99±0,07*
	Duch chartelie	Tilia cordata	4,19±0,17*	2,18±0,087*	1,44±0,096**
5	Drohobytska street	Acer platanoides	1,9±0,13**	0,91±0,060**	0,81±0,054**
	sueet	Carpinus betulus	3,21±0,13*	2,06±0,082*	1,22±0,049*

The content of pigments in the raw mass of the leaves of indicator plants in Starvi Sambir 2021

Note. In this and subsequent tables: *, **, *** – probability of differences between the control and experimental groups of animals (*– p<0.05; ** – p<0.01; *** – p<0.001).

The indicators of the content of chlorophyll a pigment in the raw mass of the selected plants in the autumn period of 2021 are greater bigger than in the autumn period of 2020, which is due to certain adaptive adjustments and the reduction of anthropopression on the environment in connection with the quarantine associated with the coronavirus pandemic. The content of chlorophyll b pigments and carotenoids in the raw mass of indicator plants of St. Sambir is growing slightly compared to 2020.

Therefore, the results of measurements of the pigment composition of the leaves of indicator woody plants of the city of Staryi Sambir in 2020-2021 show that certain adaptations of the photosynthetic apparatus are characteristic of tree species growing in areas of increased anthropogenic pressure: a decrease in the content of chlorophyll *a* and content of chlorophyll *b*.

Therefore, the assessment of the state of the environment of Staryi Sambir based on the morpho-physiological parameters of the indicator plants (*Tilia cordata*, *Acer platanoides*, *Carpinus betulus*) indicates its significant pollution. According to the results of the research, changes in morphometric parameters, an increase in the integral degree of asymmetry, an increase in the level of neuroticism and a violation of the photosynthetic function due to a decrease in the concentration of chlorophyll *a* and *b* were found in plants. This trend is most clearly observed on L. Halytskyi and Drohobytska streets.

Assessment of emissions of motor vehicles in Staryi Sambir

Today, one of the biggest environmental problems is air pollution caused by vehicle emissions. The impact of motor vehicles on the environment is discussed in the works of Mateichyk V. P. ²⁶ Kupchyk O. Yu. in his studies proved the dependence of the increase in the harmfulness of motor vehicle emissions on certain of their characteristics, namely: "the largest amount of them enters the atmosphere during starting and stopping, as well as during idling operation of the motor vehicle. As a result, the maximum content of toxic substances is observed at intersections and near traffic lights". ²⁷

At the beginning of the research, we studied how the environment of Staryi Sambir affects the physiological and morphological indicators of woody plants. The next step is the search for sources of environmental pollution and the correlation of these data with the macroscopic effects of indicator plants.

In order to determine the congestion of Staryi Sambir with cars, we chose the following streets: the street that runs along the park, L. Halytskyi str., L. Ukrainka str., Lystopadova str., Drohobytska str.

Using the method of counting cars of various types, the localization of a large number of vehicles was determined 3 times for 20 minutes at the observation points. According to the received data, it can be said that the most cars moved along the streets Drohobytska, L. Halytskyi and Lystopadova (Table 6).

Table 6

	Type of motor vehicle							
Street name	Car	Light cargo	Medium cargo (mini bus)	Heavy cargo	Bus	Σ		
Vidkryti Sertsia park	15	9	7	-	-	33		
L. Halytskyi street	55	25	15	11	17	274		
L. Ukrainka street	23	11	9	6	I	59		
Lystopadova street	35	13	11	9	5	142		
Drohobytska street	70	26	20	13	19	365		

The number of motor vehicles in Staryi Sambir

²⁶ Матейчик В. П. Методи оцінювання та способи підвищення екологічної безпеки дорожніх транспортних засобів: монографія. Київ: Національний транспортний університет, 2006. 216 с.

²⁷ Купчик О. Ю. Викиди автомобільного транспорту як джерело забруднення атмосферного повітря міста Чернігова. *Молодий вчений*. 2015. № 2 (17). С. 17–20.

According to observational data, the number of trucks in Staryi Sambir is characteristally small, and passenger cars are the most common. The largest number of motor vehicles was noted Drohobytska street and can be traced throughout the day, this is due to the fact that it connects the city with: Lviv, Uzhgorod and the customs border "Smilnytsia-Kroscienko". The smallest amount of traffic is noted for the street located near the park.

Diagram 13 shows hours of the largest number of motor vehicle on the streets of Staryi Sambir.

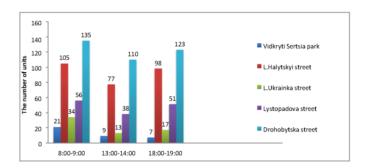


Diagram 13. Hours of the largest number of motor vehicles on the streets of St. Sambir

According to the obtained results, three main categories of streets with the most polluted atmospheric environment in St. Sambir (table 7).

Table 7

Street nome	Type of pollutant, g/km						
Street name	CO	NO ₂	СН	SO ₂	Formaldehyde	Σ	
Vidkryti Sertsia park	1,5	0,03	0,065	0,005	0,00015	1,6	
L. Halytskyi street	2	0,06	0,03	0,0006	0,00006	2,09	
L. Ukrainka street	1,5	0,146	0,14	0,0024	0,00025	1,7	
Lystopadova street	1,6	0,07	0,17	0,004	0,00032	1,8	
Drohobytska street	2,9	0,09	0,19	0,024	0,00029	3,2	

Calculation of emissions from moving vehicles in Staryi Sambir

The calculation of emissions of motor vehicles in Staryi Sambir indicate that the content of CO in exhaust emissions ranges from 1.5 to 2.9 g/km, and exceeds the norm by 1.5 times; the NO₂ content ranges from 0.03 to 0.146 g/km, and exceeds the norm by 2 times; the content of CH ranges from 0.03 to 0.19 g/km, and exceeds the norm by 2.6 times; the SO2 content ranges from 0.0006 to 0.005 g/km; the formaldehyde content ranges from 0.00015 to 0.00032 g/km.

So, according to the scale of pollution of the territory by motor vehicle emissions (Table 7), the streets that belong to the polluted zone include: Drohobytska str. and L. Halytskyi str. Streets of the moderately clean zone: L. Ukrainka, Lystopadova and streets of the ecologically clean zone – the street near the park. The problem of environmental pollution by motor vehicles is relevant because harmful emissions settle and accumulate on growing plants, which impairs their main role, namely maintaining the gaseous composition of the atmosphere.

There is a clear interdependence between the disturbance of morphophysiological processes in the leaves of indicator plants and the pollution of the environment by vehicles. The most intense changes in woody plants were found on L.Halytskyi str. and Drohobytska str., which coincides with the highest level of motor vehicle emissions.

Study of phytotoxicity of edaphotopes of monitoring points Staryi Sambir

One of the sources of environmental pollution are edaphotopes, which accumulate a large amount of pollutants of various origins, the presence of which, in turn, affects the state of the city's tree vegetation. In order to assess the impact of anthropogenic load on the environment, samples of edaphotopes were selected from our monitoring points, followed by determination of their general toxicity and phytotoxicity in the *Allium* test. The latter is a widely recognized bioassay method ²⁸, recommended by the International Commission on Protection against Mutagenic and Carcinogenic Compounds, which analyzes the growth and mitotic activity of the roots of *A. cepa* bulbs.

The general toxicity of the edaphotopes of the monitoring points in the *Allium* test was assessed by visual analysis of such macroparameters of bulb roots as growth, turgor, color, shape (swelling and bending) (see Fig. 2, Table 8).



Fig. 2. Photographs of the roots of A. cepa bulbs (for 4 days) during their cultivation on water extractors of edaphotops of monitoring points in St. Sambir: Vikryti Sertsia Park;
L. Halytskyi St.; L. Ukrainka St.; Lystopadova St.; Drohobytska St.

²⁸ Куцоконь Н. К. Рослинні тест-системи для визначення цитотоксичності. Вісник НАН України. 2010. № 4. С. 48-52.

Table 8

extractors of edulotopes of monitoring points in St. Sambir								
Research	Root length,	Indicators	Turges-	The shape	The color of			
options	mm	*t; p, T%	cene ce	of the roots	the roots			
Vidkryti Setrtsia park	45,3±3,99		Normal	Elongated and regular	Whitish with light tips			
L. Halytskyi street	8,62±1,69	$n = 4; t = 2,53; p \le 0,05; T = 35,7\%$	Normal	Elongated and regular	Dark with dark tips			
L. Ukrainka street	24±2,3	n =8; t=2,3; p≤0,05; T=6%	Normal	Elongated and regular	Whitish with dark tips			
Lystopadova street	19,3±2,1	$\begin{array}{c} n = 5; \ t = 2,97; \\ p \leq 0,05; \\ T = 34,5\% \end{array}$	Normal	Elongated and regular	Whitish with dark tips			
Drohobytska street	13,68±1.6	$\begin{array}{c} n=\!$	Normal	Elongated and regular	Dark with dark tips			
*t – coefficie	*t – coefficient of Stiudent; *p – reliability of the difference; *T% – phytotoxicity index; n– is the multiplicity index							

Morphometric indicators of *Allium cepa* roots, grown on water extractors of edafotopes of monitoring points in St. Sambir

As can be seen from Table 10, the turgescence of *A. cepa* roots in all experimental variants is normal, the shape is elongated and regular. However, the tips of the roots are dark (see Fig. 3; Table 10), and in some experimental variants – completely dark. This may indicate the presence of sufficiently high concentrations of pollutants in the studied edaphotopes.

The growth activity of the meristem of the roots of *Allium cepa* bulbs for two streets is lower, in particular, for L. Halytskyi street -5.3 times, and Listopadova street - by 2.3 times, but these indicators do not reliably differ from the control value, which is typical for the park. For the streets: L. Halytskyi, Lystopadova and Drohobytska, the growth activity is lower compared to the control by 5.3; 2.3 and 3.3 times, respectively

Phytotoxicity of aqueous extracts of studied edaphotopes and their toxic effects were evaluated in the *Allium* test according to the phytotoxicity index (T%).

The obtained indices of phytotoxicity of aqueous extracts of edafotopes testify (see Table 8) to an average ($\geq 20\%$) level of their toxicity). In particular, for L. Halytskyi str. T=35.7%, Lystopadova str. T=34.5% and Drohobytska str. T=30.5% is average (T=20.1 – 40%) and levels. Such indicators of phytotoxicity testify to the presence of toxic substances in the edaphotops of the monitoring points of Staryi Sambir in small quantities. Edaphotops. L. Ukrainka str. have the lowest phytotoxicity index (6%), which corresponds to a weak level.

A general analysis of all results shows that the assessment of the state of the environment is an integral study. The established morphological and physiological disorders of the indicator plants (*Tilia cordata*, *Acer platanoides*, and *Carpinus betulus*) depend on the state of the environment in Staryi Sambir. Expressed macroscopic effects are interdependent with emissions from moving vehicles and soil pollution. In the conditions of Staryi Sambir, there is considerable pressure on the environment.

CONCLUSIONS

1. A popular method of determining the state of the environment is bioindication. A comprehensive study of the impact of anthropogenic load on the morphological and physiological parameters of indicator plants (*Tilia* cordata, Acer platanoides, and Carpinus betulus) was carried out in the conditions of Staryi Sambir.

2. It was established that in the conditions of St. Sambir is an increase in the area of leaf blades in the studied plants. According to the results of the study, the growth of the assimilation organ can be reproduced in the following sequential series: Vidkryti Sertsia Park \rightarrow Lev Halytskyi st. \rightarrow L. Ukrainka st. \rightarrow Lystopadova st. \rightarrow Drohobytska st.

3. The analysis of the degree of asymmetry of the leaves of indicator plants showed that the greatest values of the degree of divergence of the leaves of woody plants are characteristic of individuals that grew within the local urban ecotopes with the highest level of anthropogenic pressure – Lev Halytskyi and Drohobytska Streets. The signs most prone to asymmetry were found – the width of half of the leaf and the angle between the main vein and the second vein from the base of the second order.

4. Necrotic changes of leaf blades in different districts of St. Sambir, the maximum value of the necrotic lesion of the leaf blade is typical for woody plants on the streets: L. Halytskyi (6.80–8.19%), Lystopadova (6.53–7.88%) and Drohobytska (7.4–18.4%), and the average -L. Ukrainka st. The most characteristic types of necrotic lesions for *Carpinus betulus* are interveinal, *Tilia cordata* – point, and *Acer platanoides* – spotted. The increase in the necrotic surface is a consequence of the total load on the city's ecosystem. The highest level of necrotization was established on Drohobytska street for all types of indicator plants. The obtained results indicate that *Carpinus betulus* is the most vulnerable species.

5. The analysis of the effect of technogenetic pollution on the content of chlorophylls and carotenoids in the leaves of indicator plants of Staryi Sambir in 2020–2021 showed that tree species growing in areas with increased anthropogenic pressure are characterized by certain adaptations of the photosynthetic apparatus: a decrease in the content of chlorophyll a and chlorophyll b.

6. The assessment of the state of the environment of Staryi Sambir based on the morpho-physiological indicators of plants shows its significant pollution, changes in morphometric indicators, an increase in the integral degree of asymmetry, an increase in the level of neurotization and a violation of the photosynthetic function due to a decrease in the concentration of chlorophyll a and b were found. This trend is most clearly observed on L. Halytskyi and Drohobytska streets.

7. An assessment of the congestion of the streets of St. Sambir by road transport and calculation of emissions from moving vehicles. According to the scale of pollution of the territory by vehicle emissions, the polluted zone belongs to Drohobytska and L. Halytskyi streets, a moderately clean zone are L. Ukrainka str., Lystopadova street, and an ecologically clean zone is a street near the park. The problem of environmental pollution by motor vehicles is urgent, since harmful emissions settle and accumulate on growing vegetation, and this worsens their main role is maintaining the gaseous composition of the atmosphere.

8. The general toxicity and phytotoxicity of the edaphotopes of the monitoring points of Staryi Sambir were determined. The obtained indices of phytotoxicity of aqueous extracts of edafotopes indicate an average ($\geq 20\%$) level of their toxicity. Such indicators of phytotoxicity testify to the presence of significant amounts of toxic substances in the edaphotopes of monitoring points in Staryi Sambir.

9. A comprehensive assessment of the state of the environment includes a study of sources of pollution (atmosphere, edaphotopes) and an assessment of their total impact on the morpho-physiological indicators of tree-indicator plants. The results of our research show that the streets with the most intense traffic and the toxicity of edaphotopes cause pronounced effects in plant tissues at the morphological and physiological levels. Significant anthropogenic pressure on the environment can be traced in Staryi Sambir.

SUMMARY

Determination of biologically significant anthropogenic loads based on the reactions of living organisms and their groups to them is related to bioindication. The significance of vegetation cover as an indicator of the state of the ecosystem is that it reacts very sensitively to changes in environmental factors. Ecosystems of the city are most affected by human economic activity. Therefore, it is important to monitor the state of the environment and timely analyze the pollution of the city territory. To some extent, these questions can be solved by bioindicative assessment.

Under the influence of environmental pollution, ecological and physiological signs change: pigmentation, color of plants, they are caused by an excess of toxic salts in the soil or a lack of nutrients. Bioindication has certain advantages as a method of obtaining direct information about changes in the state of biota under specific conditions of pollution, but it must be combined with chemical and geophysical experiments to obtain not only qualitative, but also quantitative information.

Therefore, in connection with the need for global monitoring, the use of the indicative capabilities of biological objects is gaining more and more importance. Indicator plants are used both to detect individual pollutants and to monitor the general state of the environment.

Biological methods for assessing the quality of the environment allow to analyze the total effect of toxicants on biological test objects of plant and animal origin. At the same time, this approach does not allow determining the causes of toxicity, but only its level. For a reliable assessment of the state of the environment, it is necessary to evenly distribute it to non-specific areas that will become monitoring points. If it is necessary to assess the state of the atmospheric air in the city, the monitoring points should be located close to the possible source of pollution. Bioindication involves assessing the state of the environment according to certain ecological and morphological parameters of indicators: quantity, structural features, growth rate, turdescence, root growth tropism, cytotoxic effects.

The assessment of the state of the environment of Staryi Sambir based on the morpho-physiological indicators of plants shows its significant pollution, changes in morphometric indicators, an increase in the integral degree of asymmetry, an increase in the level of neurotization and a violation of the photosynthetic function due to a decrease in the concentration of chlorophyll a and b were found. This trend is most clearly observed on L. Halytskyi and Drohobytska streets. According to the scale of pollution of the territory by vehicle emissions, the polluted zone belongs to Drohobytska and L. Halytskyi streets.

A comprehensive assessment of the state of the environment includes a study of sources of pollution (atmosphere, edaphotopes) and an assessment of their total impact on the morpho-physiological indicators of tree-indicator plants. The results of our research show that the streets with the most intense traffic and the toxicity of edaphotopes cause pronounced effects in plant tissues at the morphological and physiological levels. Significant anthropogenic pressure on the environment can be traced in Staryi Sambir.

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