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BIOINDICATION OF URBAN ENVIRONMENTAL STATE BASED ON LONDON PLANE TREE (*PLATANUS × ACERIFOLIA*) POLLEN ANALYSIS

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Background. The qualitative characteristics of pollen of the London plane tree, *Platanus* × *acerifolia* (Ait.) Willd., were studied for bioindication of the state of the urbanized environment based on the indicators of fertility and viability of pollen grains.

Materials and Methods. Trees of the London plane, *Platanus* × *acerifolia* (Ait.) Willd., from Kyivska Street were selected as experimental plants. In the control variant, trees from a part of the Sofiyivka Park relatively isolated from technogenic load were used. Morphometric parameters of pollen, fertility and viability of mature pollen grains were analyzed using generally accepted methods. Statistical analysis of the obtained results was performed according to Ronald Fisher (Ronald Fisher, 2017) using Microsoft Office Excel 2007.

Results. According to the mean sizes of pollen grains (31.23 and 31.18 μm), the studied trees exhibited virtually no differences regardless of the place of growth. In both variants, fertility indicators were high (98.34 % – Kyivska Street and 96.55 % – Sofiyivka Park). However, in the variants of pollen germination in five- and ten-percent sucrose solutions, pollen from Kyivska Street was inferior in viability to pollen from the park by 19.36 and 20.00 %, respectively.

Conclusion. The significant variation in the percentage of germinated pollen grains from trees growing under different air pollution conditions in Uman supports the potential use of *Platanus* × *acerifolia* pollen germination levels in 5 % and 10 % sucrose solutions as an indicator for urban air quality assessment.

Keywords: *Platanaceae* Lindl., sexual dimorphism, dioecious plants, pollen grain viability, urban green spaces, urbanized environment

INTRODUCTION

The maple-leafy plane tree, *Platanus × acerifolia* (Ait.) Willd. is one of the species in the genus *Platanus* L. of the family *Platanaceae* T.Lestib. The *Platanaceae* family consists of the only extant genus, *Platanus*, but also includes an extensive diversity in the fossil record (Danika *et al*., 2024; Yan *et al*., 2024). In 1753, Carl Linnaeus first described two species of plane trees, *P. occidentalis* L. and *P. orientalis* L. At the same time, *Platanus* plants were well known to pre-Linnaean ancient writers, and the name itself, "platanos", is the classical Greek name for the current *P. orientalis* (Nixon & Poole, 2003). Particularly in Homer's epic poem "The Iliad" (9th century BC), we read "platanistos". Theophrastus (4th-3rd centuries BC) used the name "platanos", and the name "Platanus" appears in the works of Cicero (1st century BC) already in its contemporary orthography (Tsopelas *et al*., 2017). The word "platys", which means broad or wide, is the etymological root of the Greek name "platanos", which refers to the broad palmate leaves of this tree (Tsopelas *et al*., 2017; Jones, 2025).

The ten currently accepted species of the *Platanus* genus are naturally distributed from southeastern Europe to Pakistan, Indochina, and from eastern Canada to Guatemala (Yan *et al*., 2024). All *Platanus* species can exhibit a high degree of intraspecific variation, as a result of which they can overlap with other species in terms of certain morphological characteristics (Grimm & Denk, 2008).

P. orientalis (oriental plane), *P. occidentalis* (American sycamore), and *P. × acerifolia* (a hybrid of *P. orientalis* and *P. occidentalis*) have a wider distribution range, and *P. × acerifolia* is the *Platanus* species that is used for ornamental landscaping more often than others (Hrabovyi, 2007). This species is well-known by its common names "London plane" and "hybrid plane". In an internationally recognized online taxonomic database, "Plants of the World Online" (POWO), published by the Royal Botanic Gardens, Kew, *Platanus × acerifolia* (Ait.) Willd. is included as an artificial hybrid and a synonym of *Platanus × hispanica* Mill. ex Münchh (POWO, 2025) is mentioned, too. Herein, to prevent terminological confusion, we will continue to use the common name "London plane tree" for *P. × acerifolia*, as used by previous authors (Henry & Flood, 1919; Zahorulko & Korshykov, 2020). *P. × acerifolia* was bred around 1670 in the Oxford Botanic Garden as a natural hybrid of *P. orientalis* and *P. occidentalis* (Henry & Flood, 1919).

Platanus trees became widespread in European parks in the 18th–19th centuries, when they were introduced to this territory, which is also part of their paleontological range. The most widespread of these species was *P. acerifolia*, which, thanks to its hybrid nature, proved to be the most ecologically plastic (Hrabovyi, 2007).

In the current Ukrainian territory, the introduction of plane trees began in Crimea, where seeds of one of the Platanus species were first introduced in 1786. In the Nikita Botanical Garden, a collection of plane trees was assembled in 1814–1828. Since then, plane trees have been cultivated in Crimea from local seeds. The Nikita Botanical Garden nursery produced up to 200,000 seedlings per year. *P. × acerifolia* dominates in Crimean plantations, while *P. orientalis* is rare, and *P. occidentalis* occurs in single specimens. There are more than 30 old trees of this genus growing in the old parks of Bukovina. The oldest tree in Ukraine grows in Makivsky Park (Khmelnytskyi region) and is about 150–200 years old (Zahorulko & Korshykov, 2020). *P. × acerifolia* plants are deciduous, fast-growing (particularly in the first half of their lives), and long-lived (up to 5000 years). They can reach heights of 35 to 40 meters (Gratani *et al*., 2020; Hrabovyi, 2007).

P. × acerifolia plants are also moderately frost-resistant, withstanding temperatures as low as -25 °C; they are very light-loving but can tolerate shade. *P. × acerifolia* takes root well when transplanted and grows excellently in fresh, moist, well-drained, slightly clayey, and relatively rich soils with a pH of 6.5–8.0 (Dirr, 2009; Gratani *et al*., 2020; Zahorulko & Korshykov, 2020).

Overall, plants of this species are undemanding with respect to soil conditions and can grow on almost any type of soil, even on very dry soils, provided that they are not poor. Among all representatives of the genus *Platanus*, *P. × acerifolia* is the most tolerant under cultivation conditions. It withstands air pollution, including gas contamination, smoke, and dust, as well as snow accumulation, drought, and wind, and is resistant to pests and diseases (Gratani *et al*., 2020; Hrabovyi, 2007; Kapelusch, 2009). Due to the tolerance of this species to smoke and urban pollution, it is widely cultivated in London under the name “London plane tree” (Dirr, 2009; Esper *et al*., 2023).

The crowns of *Platanus orientalis* and *Platanus acerifolia* in sanitary-hygienic plantings remove more dust from atmospheric air than the native species *Tilia cordata* and *Acer platanoides*. As part of the dust, plants deposit on the surface of their leaves a sufficient amount of various chemical elements, including toxic metals. Plants of the genus *Platanus* perform the role of sanitary-hygienic plantings, as they remove from the air a significant amount of aerosols harmful to the biosphere, acting as a barrier to their further spread, which is of great importance in preventing a number of diseases in the population (Kapelusch, 2009). There are, however, reports of increased allergenicity of *P. × acerifolia* pollen (Álvarez-López *et al*., 2022; Bedolla-Barajas *et al*., 2024; Lancia *et al*., 2024) due to its interaction with urban pollutants (Hui *et al*., 2025; Ribeiro *et al*., 2017).

In the conditions of the introduction area in the Right-Bank Forest-Steppe of Ukraine, *P. × acerifolia* reaches reproductive maturity at the age of 10–20 years. One tree can provide up to four to five kilograms of full-value seeds. *Platanus* plants can be propagated both by seeds and cuttings (Zahorulko & Korshykov, 2020).

A specific feature of *P. × acerifolia* is the gray bark of the trunk and branches, which exfoliates in large plates, leaving pale green and cream patches on the inner fresh wood of yellow color (Hrabovyi, 2007; Dirr, 2009), resulting in a highly decorative mosaic, marble-like, mottled appearance of the trunk (**Fig. 1**).

It has been suggested that this feature of *P. × acerifolia* arose through the blockage of bark pores as a protective response to environmental pollution and other ecological stresses, and the annual detachment of dead bark promotes the growth of newly formed bast (Hrabovyi, 2007). The other hypotheses regarding bark exfoliation have also been discussed. In particular, plane bark exfoliation correlates significantly with tree size and inner bark width but is strongly influenced by cold-season temperatures (Esper *et al*., 2023).

The shoots of *P. × acerifolia* are geniculate, young branches have dense gray-yellow felt-like pubescence, while old branches are bare and reddish-brown in color (Dirr, 2009; Deepdale trees, 2012; Hrabovyi, 2007).

The London plane tree palmate leaves are green and arranged alternately along the shoot; they are similar in shape to maple leaves, but have three to five lobes (rarely indistinctly 7-lobed), 12.0–25.0 cm wide (Hrabovyi, 2007; Dirr, 2009). The leaves are linearly cut or heart-shaped at the base, entire or unevenly toothed (**Fig. 2**). The green leaves of *P. × acerifolia* become brownish-green in autumn.

Fig. 1. The *P.* × *acerifolia* bark texture. Photo by Olga Opalko

Fig. 2. The *P.* × *acerifolia* leaf. Photo by Olga Opalko

In *P.* × *acerifolia* plants, the leaf lobes are broadly triangular (Grimm & Denk, 2008). They are smaller than those of the oriental plane tree (*P. orientalis*), but larger than those of the American sycamore (*P. occidentalis*). There are three (or five) main veins extending from the base of the leaf. Young leaves have felt-like pubescence on the underside, while older leaves have pubescence only along the main veins. Stipules are 1–1.5 cm long; petioles are 3–10 cm long, yellow-brown, and densely pubescent. The leaves of *P.* × *acerifolia* have candelabra trichomes, limited by large veins on the abaxial surface. Stomata are limited to small areolar areas on the abaxial part of the leaf surface (Dirr, 2009; Gratani *et al*., 2020; Hrabovyi, 2007).

The flowers of *P.* × *acerifolia* plants are small, unisexual, actinomorphic, and occur in different inflorescences on the same tree. Those wind-pollinated flowers are borne in a ball-like structure, as are the seeds that follow, and can stay on the tree for a while after the leaves have fallen. The inflorescences look like spherical heads (Hrabovyi, 2007) on long, hanging pedicels and bloom after the leaves appear (**Fig. 3**).

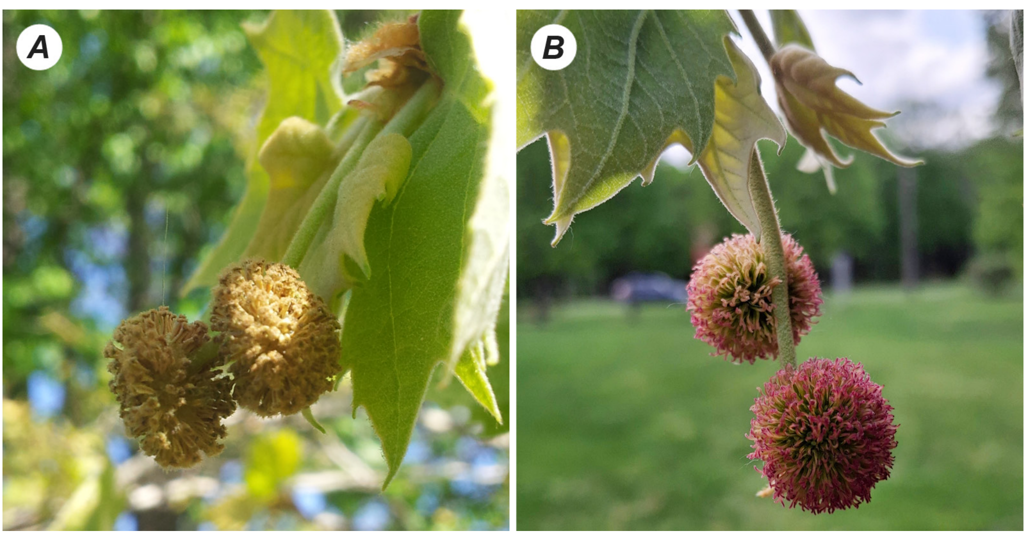

Fig. 3. The *P.* × *acerifolia* male (***A***) and female (***B***) inflorescences. Photo by Nadiia Tsybrovska

Staminate flowers of yellowish coloration have very small sepals and petals and large anthers. The anthers are leathery, pubescent (Fig. 3), bilocular, almost sessile, long, club-shaped, mostly with fleshy wedge-shaped scales. The corolla consists of 3–4 segments which, with the exception of petals in the staminate inflorescences, lack vascular bundles. The number of stamens in male flowers is usually 3–4, sometimes up to 7. The filaments are very short, almost inconspicuous. The anthers are elongated and open longitudinally. The style is expanded at the top into a stigma. (Dirr, 2009; Hrabovyi, 2007; Platan klenolystyy, 2025).

The pistillate flowers are red, numerous, and sessile, with three to five sepals, four to five petals, sometimes up to seven to eight, and often no petals at all. They are surrounded by wedge-shaped, inversely conical scales. The pistil has hook-shaped stigmas and elongated styles. The ovary usually has one ovule, sometimes two. The gynoecium is apocarpous, consisting of five to eight, less often three to four or nine, small carpels (Hrabovyi, 2007; von Balthazar & Schönenberger, 2009; Platan klenolystyy, 2025).

The London plane tree fruits appear in aggregates of hundreds in balls (multiple fruits) around two and a half centimeters across on long fruit stalks. Each tiny fruit has a tuft of hair, which is designed for anemophilous dispersal to aid dissemination. The fruit clusters contain one to three fruits, with a diameter from two and a half to three centimeters. The fruits ripen in autumn and fall off in the spring of the following year. The seeds are small, with a fine, straight embryo and endosperm (Dirr, 2009; Gratani *et al*., 2020; Hrabovyi, 2007).

Pollen quality is one of the criteria for plant reproductive capacity (Matiashuk & Tkachenko, 2024), so pollen quality characteristics can be used as bioindicators for monitoring the state of the environment (Batos *et al*., 2019; Faur *et al*., 2012). The morphoanatomical features of the embryo sac determine its protection, while pollen grains are less resistant to various stress factors. The most accessible for testing is the male gametophyte, i.e., pollen, which is produced in large quantities on a single individual, making it easy to obtain statistically reliable results. The development of reproductive structures, in particular the male gametophyte, is an important indicator of adaptation to growing conditions, as it is more sensitive to adverse environmental factors than the plant as a whole. Sterility occurs due to the inability or reduced ability of the organism to produce normal gametes, which may be caused by chemical and physical pollutants in the atmosphere (Shvets, 2011; Montano, 2020; Matiashuk & Tkachenko, 2024).

The purpose of this study was to determine whether certain characteristics of London plane tree (*P.* × *acerifolia* (Ait.) Willd.) pollen can be used to assess the state of the urban environment.

MATERIALS AND METHODS

Plant material. Over 65-year-old plane trees from Kyivska Street were chosen as the experimental plants. This street, which is a major two-way street of Uman City, runs from a bus terminal to Unity Street, leading to the Kyiv–Odesa highway. The air quality on this street is poor due to high levels of traffic exhaust and other airborne pollutants. In the control variant, trees from a relatively clean, isolated part of the Sofiyivka Park were used (**Fig. 4**).

Fig. 4. The *P.* × *acerifolia* from Kyivska Street (***A***) and from Sofiyivka Park (***B***). Photo by Olga Opalko

The morphometric characteristics of pollen, fertility, and viability of mature pollen grains were analyzed using standard methodologies (Noor *et al*., 2017; Voytsekhivs'ka *et al*., 2010; Woodhouse, 1935). Pollen was collected during the period of massive plant flowering. The pollen grains were studied at two variants: plane trees from Kyivska Street (experimental plant group) and those from the Sofiyivka Park, Quartier 13 (control plant group).

The shape and size of pollen grains were photographed and measured using a Levenhuk MED 25T microscope equipped with a 5.1MP digital camera.

Acetocarmine staining. The pollen fertility of *P.* × *acerifolia* was determined using the acetocarmine staining method. Pollen was extracted from the anthers onto a clean glass slide and immersed in a drop of acetocarmine solution to stain the grains for analysis. At least 500 pollen grains were examined in each variant.

After three to five minutes of staining, the samples were examined under a light microscope at 400× magnification in 35 fields of view for each variant. Pollen fertility was assessed by counting the number of stained pollen grains in the microscope field of view. Acetocarmine stains the nucleus of the pollen grain, which allows abnormalities to be readily detected. The granular cytoplasm and sperm cells of fertile pollen grains were stained in a dense carmine-red color. In contrast, sterile pollen grains were weakly stained with acetocarmine or showed uneven staining (Ascari *et al.*, 2020; Bisht *et al.*, 2023; Dafni, 1992; Pinillos & Cuevas, 2007).

In addition, "sterile deformed" pollen grains were counted; these are nonfunctional male gametes with structural abnormalities and irregular staining (Anamthawat-Jonsson & Karlsdottir, 2016).

The pollen viability. Pollen viability, defined as the ability of the male gametophyte to grow, was determined by germinating pollen grains in sucrose solutions with concentrations of 5, 10, 15, 20, 25, and 30 %. The number of germinated and non-germinated pollen grains was counted after 24 hours under a microscope on 15 fields of view in each variant with a 100× magnification.

Statistical Analysis. The statistical analyses of the results were performed according to Ronald Fisher (2017) and also Serhiy Skrypnyk and Viktor Rybak guidelines (Skrypnyk & Rybak, 2024) using a two-way ANOVA and were conducted using Microsoft Excel_2007.

RESULTS AND DISCUSSION

It is well known that plants growing along the sides of streets or roads are particularly vulnerable to human activities, including vehicle emissions and various types of particulate matter (Muthu *et al.*, 2021). In addition to producing particulate matter through the exhaust of cars, buses, and trucks, traffic on urban streets also lifts dust from the road surface as vehicles pass. Vehicle wear and tear, including the wear of tires, brake linings, and clutch discs, can also release particulate matter into the air along roadsides. Plants growing along streets are exposed to anthropogenic air pollution, which affects their reproductive organs, particularly pollen, through both direct and indirect impacts (Das *et al*., 2025).

Morphometric characteristics of pollen. According to the average pollen grain sizes (20.61 and 20.58 μm), the studied trees exhibited minimal difference based on the place of cultivation (**Table 1**).

Table 1. **Diameter of *P.* × *acerifolia* pollen grains by growing location (μm)**

Place of growing	Sofiyivka Park, Quartier 13	Kyivska Street, Uman	Difference, based on growing location
$\bar{X}$	20.61	20.58	0.03
max	23.12	24.00	0.88
min	18.10	18.02	0.08
SE	0.19	0.18	0.01
CV, %	6.40	6.21	0.19
s^2	1.74	1.63	0.11

The difference between the maximum and minimum pollen grain size was 19 % greater in pollen collected from the tree growing on Kyivska Street in the city of Uman. However, the coefficient of variation of pollen grain diameter in both experimental variants was less than 10 %, indicating only minor variation in this trait.

The permanent influx of airborne pollutants may interfere with early reproductive processes (Mehmood *et al*., 2024). However, the results obtained give reason to believe that air pollution from motor vehicle emissions had negligible impacts on *P.* × *acerifolia* pollen grain diameters. Differences, based on the effects of the potentially damaging airborne pollutants, were within the confidence intervals of the experimental data. The pollen grains of both locations were typically spheroidal (round). The shape of pollen grains from trees growing on Kyivska Street was almost identical to those from trees in the Sofiyivka Park, Quartier 13 (**Fig. 5**). This makes it unlikely that the shape of *P.* × *acerifolia* pollen grains will serve as a gauge of urban air quality.

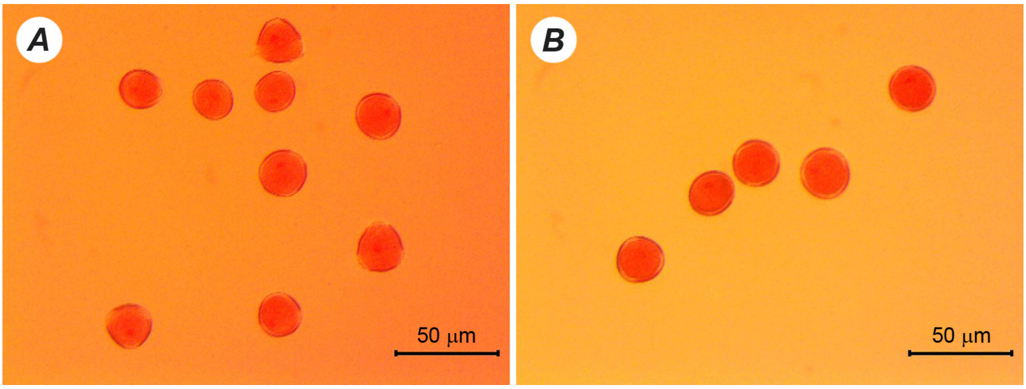

Fig. 5. Shape of *P. acerifolia* pollen grains (400× magnification): ***A*** – Sofiyivka Park, Quartier 13; ***B*** – Kyivska Street, Uman

Pollen grain fertility, and viability. The analysis of pollen from *P.* × *acerifolia* trees growing in the Sofiyivka Park, Quartier 13, and on Kyivska Street, showed high fertility (98.34 % and 96.55 %, respectively) regardless of growth location, with a few sterile (0.99 % and 2.23 %) and sterile deformed with structural abnormalities (0.67 % and 1.22 %) pollen grains in both growing locations (**Table 2**). The results of this experiment refute the hypothesis on the possibility of using the acetocarmine method by the number of stained *P.* × *acerifolia* pollen grains in the microscope field of view as an indicator of urban air quality.

Table 2. ***P.* × *acerifolia* pollen grain fertility based on growing location, %**

Pollen grain quantity, %	Place of growing		Difference, based on growing place
	Sofiyivka Park, Quartier 13	Kyivska Street, Uman	
Fertile	98.34 ± 2.11	96.55 ± 2.94	1.79
Sterile	0.99 ± 0.58	2.23 ± 1.03	1.24
Misshapen	0.67 ± 0.51	1.22 ± 0.83	0.55

The *P.* × *acerifolia* pollen grain viability analysis based on the percentage of germinated pollen in sucrose solutions confirmed the efficiency of this method. Sucrose is frequently used in pollen germination tests as an energy source for pollen, promoting germination and the growth of pollen tubes (Li *et al*., 2023). The maximum viability of *P.* × *acerifolia* pollen grains was observed in a medium with a sucrose concentration of 15 %, on which more than 50 % of pollen grains germinated in both variants of the experiment (**Fig. 6**).

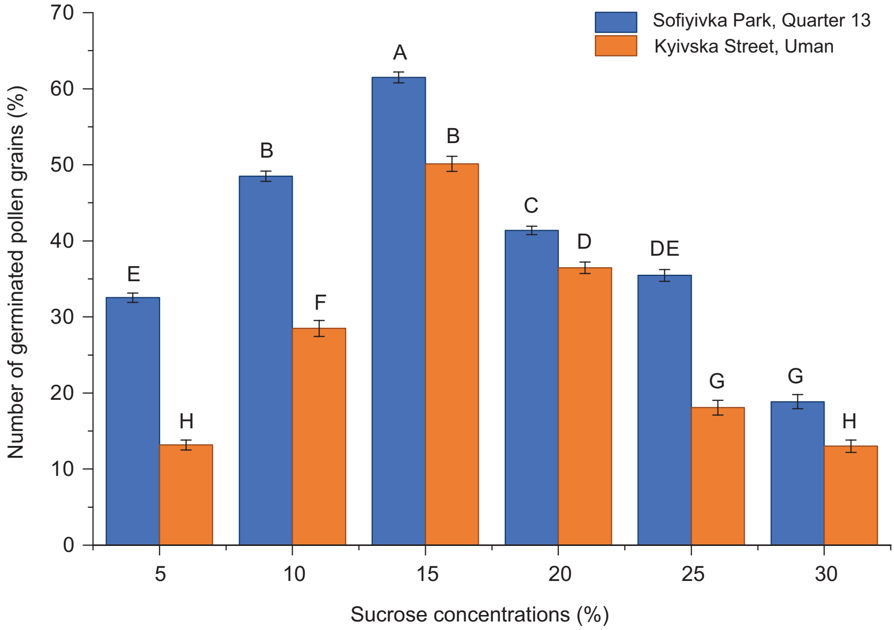

Fig. 6. Means values of *P.* × *acerifolia* pollen grain germination at different sucrose concentrations. Means that do not share a letter are significantly different (according to the results of the Tukey test)

More than 30 % of pollen grains were germinated at sucrose concentrations of 5, 10, 20, and 25 % from trees growing in Sofiyivka Park, Quartier 13, and at a sucrose concentration of 20 % from trees growing on Kyivska Street, Uman. At a sucrose concentration of 30 %, the percentage of germinated pollen was the lowest from both tree locations, at 18.86 % and 13.01 %, respectively.

A study of pollen from trees growing on Kyivska Street, Uman, revealed a 4.91 % to 20.00% reduction in its viability compared to pollen from trees growing in Sofiyivka Park, Quartier 13. The smallest difference was observed at a sucrose concentration of 20 %, at which pollen viability exceeded 30 % in both variants. The largest difference between the percentages of germinated pollen grains collected from trees from different locations was in the variants with sucrose concentrations of 5 and 10 %, at 19.36 and 20.00 %, respectively. Such a significant decrease in the percentage of germinated pollen grains from trees growing on Kyivska Street can be explained by the effects of urban air pollutants.

Our findings are generally consistent with those reported by other researchers on various plant species (Muthu *et al*., 2021; Ramírez-Aliaga *et al*., 2022; Stadnik, 2021; Ugale *et al*., 2021; Utarbayeva *et al*., 2021), including herbaceous plants (Jaconis *et al*., 2017) and woody species exposed to industrial pollution (Pukacki & Chalupka, 2003).

CONCLUSION

The comparison of some morphometric characteristics of *P.* × *acerifolia* pollen (pollen grain diameter and shape, as well as the fertility of pollen grains stained by acetocarmine), based on the growing location, indicated that when exposed to an urban street-side pollutant mixture*, P.* × *acerifolia* did not exhibit significant negative reactions.

The acetocarmine staining method is suitable for identifying aborted or sterile grains, but not for studying the actual pollen viability. Therefore, neither the diameter nor the shape of *P.* × *acerifolia* pollen grains, nor the acetocarmine staining method, can be used to monitor urban air quality.

The significant variation in the percentage of germinated pollen grains from trees growing under different air pollution conditions in Uman supports the potential use of *Platanus* × *acerifolia* pollen germination levels as an indicator of urban air quality.

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COMPLIANCE WITH ETHICAL STANDARDS

Conflict of Interest: the authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Animal Rights: this article does not include animal studies.

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

AUTHOR CONTRIBUTIONS

Conceptualization, [A.O.; O.O.]; methodology, [A.K.; N.T; O.O.]; validation, [A.O.; V.H.]; formal analysis, [A.K.; O.O.].; investigation, [A.K.; N.T.; O.O.]; resources, [V.H.]; data curation, [V.H.; O.O.]; writing – original draft preparation, [A.K.; N.T.; O.O]; writing – review and editing, [A.O.; O.O.]; visualization, [A.K.; O.O.; N.T.] supervision, [O.O.]; project administration, [V.H.]; funding acquisition, [V.H.].

All authors have read and agreed to the published version of the manuscript.

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БІОІНДИКАЦІЯ СТАНУ УРБАНІЗОВАНОГО ДОВКІЛЛЯ НА ОСНОВІ АНАЛІЗУ ПИЛКУ ПЛАТАНА КЛЕНОЛИСТОГО (*PLATANUS × ACERIFOLIA* (AIT.) WILLD.)

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Обґрунтування. Якісні характеристики пилку платана кленолистого, *Platanus* × *acerifolia* (Ait.) Willd., вивчали для біоіндикації стану урбанізованого довкілля за показниками фертильності й життєздатності пилкових зерен.

Матеріали та методи. За експериментальні рослини обрали дерева платана кленолистого з вул. Київська. У контрольному варіанті використали дерева з відносно ізольованої від техногенного навантаження частини парку "Софіївка". Морфометричні показники пилку, фертильність і життєздатність зрілих пилкових зерен аналізували загальновживаними методами. Статистичний аналіз отриманих результатів проводили за Рональдом Фішером (Ronald Fisher, 2017) з використанням Microsoft Office Excel 2007.

Результати. За середніми розмірами пилкових зерен (31,23 і 31,18 мкм) досліджені дерева майже не відрізнялися, незалежно від місця вирощування. В обох варіантах показники фертильності були високими (98,34 % – вул. Київська і 96,55 % – парк "Софіївка"). Однак у варіантах пророщування пилку в 5- й 10-відсоткових розчинах сахарози пилок з вул. Київська поступався за життєздатністю пилку з парку на 19,36 і 20,00 % відповідно.

Висновки. Оцінювання загального стану урбанізованого довкілля негативних впливів техногенного навантаження на міські зелені насадження можна виконувати за результатами пророщування пилку платана кленолистого у 5- й 10-відсоткових розчинах сахарози.

Ключові слова: *Platanaceae* Lindl., статевий диморфізм, дводомні рослини, життєздатність пилкових зерен, міські зелені насадження, урбанізоване довкілля

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