UDC: 581.526.425:581.33(497.11 Beograd)

Original scientific paper

Morphological Properties of Pollen as Bioindicators of Deciduous Woody Species in Belgrade Parks (Serbia)

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Abstract: As deciduous woody species make a large and significant portion of park plant communities, they are suitable for the programs of the urban environment biomonitoring. The study of the size and shape of pollen included 58 trees of the following 12 species: *Aesculus hippocastanum* L., *Betula alba* L., *Ginkgo biloba* L., *Paulownia tomentosa* Steud., *Platanus x acerifolia* (Aiton) Willd, three species of *Quercus* sp. (*Q. robur* L., *Q. cerris* L. *Q. rubra* L.) and four species of *Tilia* sp. (*T. argentea* Desf., *T. x euchlora*, *T. grandifolia* Ehrh., *T. parvifolia* Ehrh.). Pollen was collected in five parks in the area of Belgrade, three of which (Pioneer Park, Academic Park and Banovo Brdo Park) are located in the central city zone, while the park of the Palace of Serbia (SIV Building) and Topčider Park are in the wider urban zone. The pollen of birch, chestnut and pedunculate oak had smaller values of pollen length in the central city zone, due to the increased human activity, compared to the wider city zone. Ginkgo, chestnut and pedunculate oak didn`t have significantly different values of pollen width between the urban zones. The pollen of ginkgo, pedunculate oak and London plane had smaller mean values of the index of pollen shape in the central city zone. A reversed pattern of values, i.e. higher values in the central city zone, was obtained for the length of the ginkgo pollen and the width of the London plane pollen. The index of pollen shape in chestnut didn't show any statistically significant differences between the city zones. The results of the analysis of variance confirmed statistically significant differences between the city zones for the length and width of pollen, and the index of pollen shape. The share of phenotypic variability caused by the differences in the environmental conditions between the zones (differences between the parks) was statistically significant for the analyzed pollen properties. Based on the preliminary results, it can be concluded that the increased environmental pollution in the central city zone leads to a reduction in the size of pollen. Therefore, future monitoring projects can use this property as an indicator of the change in the morphology of pollen under the influence of stress induced by conditions of urban environment.

Keywords: pollen, size, index of pollen shape, bioindicators, urban parks.

1. Introduction

Air is an indicator of the state of the environment because it directly affects its preservation and survival. Apart from global climate change, the environment is heavily exposed to numerous harmful effects that alter the composition of air and in the interaction with climate affect the environment (Bytnerowicz et al. 2007; Ghorani-Azam et al. 2016). The main sources of air pollution, especially in the urban environment, are the products of transportation, industry and domestic heating. The most common harmful substances in the air are gases, such as carbon monoxide (CO), sulfur dioxide (SO_2), nitrogen dioxide (NO_2) and suspended microparticles of soot, as well as specific chemical elements (Pb, Cd, Mn, As, Ni, Cr, Zn and other heavy metals and organic compounds resulting from a wide range of different activities). Their effects and consequences are manifested in the overall living world, especially in urban areas. Plant species in the urban environment remove harmful pollutants from the air by sedimenting them on their surface or depositing them in their tissues (Nowak et al. 2014). The role woody species is particularly important. During their long life cycle, they accumulate significant amounts of harmful substances that cause numerous structural and physiological alterations and disturb their normal functioning. The changes that occur are indicative of the harmful effects of the entire complex of environmental stress factors, both biotic and abiotic (e.g. pathogens, climate, air pollutants, radiation) (Onete et al. 2010; Samecka-Cymerman et al. 2011; El-Khatib et al. 2016). In this sense, parks can be considered to be a defensive system of urban sites.

Pollen properties, particularly the morphological ones (size, shape, structure of the outer shell - exine), are mainly species-specific and they are often used in the systematics of plants (Jia et al., 2014). There are significant differences in the size, composition and structure of pollen between broadleaf and conifer species. The pollen of most conifer species is significantly larger and has air bubbles that facilitate its dispersal, compared to the smaller pollen of broadleaved species without air bubbles. The pollen of *Quercus* sp. is oblate and belongs to the group of medium-size pollen. It is larger than *Castanea sativa* Mill. and *Populus nigra* L. pollen. Furthermore, the pollen of different species within the same genus is different in size: the pollen of *Quercus robur* L. is larger than the pollen of *Q. pubescens* Willd., but it is smaller than the pollen of *Q. frainetto* Ten., *Q. petraea* (Matt.) Liebl., *Q. macedonica* A.DC. and *Q. cerris* L. (Erdtman, 1952). The size of pollen grains of the genus *Aesculus* sp. was different in this order (from lager to smaller): *A. flava* > *A. carnea* > *A. hippocastanum* > *A. parviflora* (Ćalić et al. 2009). It should be taken into account that literature sources often lack sufficiently precise data on the methodology of pollen morphology research, which makes the comparison more difficult (Kedves et al., 2002).

Most woody species are characterized by the periodicity of flowering, i.e . they do not flower every year, and the intensity of flowering varies greatly from year to year (temporal variability) and from tree to tree within the same species (interindividual variability) (Jato et al., 2002**;** Batos, 2013).

Recent decades have seen an increasing interest in the research of global climate change and increasing levels of air pollution (air pollutants, chemical pollutants, radiation, antioxidant enzyme content, etc.) and their impact on pollen properties (Kormutak et al. 1994; Knight et al. 2005; Rezanejad, 2009, 2012). The resulting changes, either morphological or physiological, are used as bioindicators of the state of the environment (Pidek et al. 2010). Increased pollution leads to the reduced size and germination rate of pollen, which is why some authors consider that pollen is a good indicator of the state of the environment (Pukacki and Chalupka, 2003; Azzazy, 2016; Kaur and Nagpal, 2017). Based on the findings that in the conditions of increased temperatures and the resultung increased plant desiccation intensities, plants produce pollen of larger dimensions, Ejsmond et al. (2011, 2015) suggest that the pollen size and shape should be used in the models of paleoecological reconstruction of climate change. Furthermore, the research on pollen properties can be applied in aerobiology, allergology, paleobotany, palynology, etc. (Garcia-Mozo et al. 2000; Lindbladh et al. 2002; Yu-Sheng Liu et al. 2007).

One of the starting hypothesis of the study was that, besides the effects of the biology of species and genotype, the location of the selected parks (wide or central city zone), i.e., their proximity to roads and the presence of pollutants, has significant effects on the development and physiological status of pollen. The aim of the research was to study the effects of environmental factors in the urban areas on the morphological properties of pollen of deciduous woody species, i.e., the use of pollen properties as bioindicators of the state of the environment.

2. Material and Methods

The research was conducted in the parks of the city of Belgrade (N 44° 49′, E 20° 27′). Pollen was collected in five selected parks during 2015 and 2016. It was harvested from 12 broadleaved species, i.e. 58 trees: *Aesculus hippocastanum* L. (10 trees), *Betula alba* L. (8), *Ginkgo biloba* L. (3), *Paulownia tomentosa* Steud. (2), *Platanus x acerifolia* L. (11), *Quercus cerris* L. (2), *Quercus robur* L. (6), *Quercus rubra* L. (2), *Tilia argentea* Desf. (5), *Tilia* x *euchlora* (2), *Tilia grandifolia* Ehrh. (3), and *Tilia parvifolia* Ehrh. (4). The criterion used in the selection of parks was the degree of pollution, that is, their location in the city and proximity to the main roads. Three parks are located in the zone of increased pollution, two of which are located in the central city zone (Pioneer Park – 3.0 ha and Academic Park – 1.5 ha) and one in the wider city area but in the immediate vicinity of the main road (Banovo Brdo Park - 10.9 ha), and two in the zone of small pollution, i.e., in the wider urban zone (the park around the Palace of Serbia or SIV Building – 20.0 ha and Topčider Park – 13.0 ha). The criteria for the selection of trees and species included their higher share in the parks, good vitality and physiological maturity, as well as the role of species in the greening of parks. The number of trees of a particular species varied between the study parks and was limited by the periodicity and quantity of pollen in the years of pollen sampling.

The analysis of air pollution in the city of Belgrade and its individual sites (parks), was based on data taken from the Annual Reports of the Secretariat for Environmental Protection, City of Belgrade (*www.eko.bg.gov.rs*), compiled in accordance with the Regulation on monitoring conditions and air quality requirements, "Official Gazette of RS", no. 11/2010 and 75/2010.

Pollen was sampled during the period of full flowering (April - June, depending on species) from the branches in the lower third of the outer portion of the crown (3 - 5 - 8 m), in all four aspects. It was done in the morning on a day with no wind and rain. Depending on the maturity of flowers, pollen was collected either directly from the flowers/catkins in situ or from the flowers/catkins that matured in the laboratory by the method of "aquatic cultures" (Kirby and Stanley, 1976). The anther ruptured and the pollen was released within 24 - 48 h. In both cases, the collected pollen was first separated from rough impurity and then screened through a series of sieves with the end diameter of 0.2 mm. Having been dried in a pollen dryer at +30 °C/48 h, pollen was stored in a desiccator with $CaCl_2$ in a refrigerator at +5(±1) °C and then it was ready to be used in the analysis (Figure 1a-f).

The size of dry pollen was measured using a digital microscope system (Leica Galen III) with a camera (CCD Camera Topica TP/5001) with 10(40)x zoom lens and 10x ocular zoom in the Image Tool. The length (E = equatorial axis) and the width (P = polar axis) were measured on a sample of 100 pollen grains per tree in 3 visible fields (200 measurements x 58 trees = 11600 measurements). The obtained values were corrected by the corresponding coefficient and expressed in μm (1×10^{-6} m). They were further used to determine the pollen shape index expressed in % as the ratio of the polar to the equatorial axis P/E (Panahi et al. 2012). Since the statistical processing included only the species that were present in at least two parks, *Tilia* x *euchlora*, *Paulownia tomentosa*, *Quercus cerris* and *Quercus rubra* were excluded from the study. The share of phenotypic variability between the urban zones and the parks within the zones, as well as the effects of species and trees as sources of genetic variability, were estimated using the appropriate model of the analysis of variance (ANOVA). City zones, parks and species were the fixed factors in the hierarchical model of the analysis of variance, while the trees had a random effect. The comparison of the mean values of the analyzed properties and the statistical significance of their differences were obtained using the Scheffe test.

Figure 1. The process of pollen collection, preparation and analysis. a) Direct collection of pollen in situ (*Ginkgo biloba* L.), b) Pollen collection by the method of "aquatic cultures" (*Ginkgo biloba* L.), c) Pollen collection procedure (*Paulownia tomentosa* Steud.), d) Pollen collection procedure (*Quercus rubra* L.), e) Cleaning and screening of pollen (*Betula alba* L.), f) Microscopic analysis of pollen.

3. Results

3.1. Air pollution

The analyzed parks in the area of Belgrade are located at a relatively small distance. Thus, local climate changes which occur in specific areas during the year do not have any significant effect on the climate change annual cycle (Figure 2).

However, the degree of air pollution can vary significantly between different parts of the city. The selected parks were zoned based on their location and degree of pollution. The following spatial zoning of the city was used: Zone I - central city zone: Pioneer Park, Academic

Park; - zone II - wider city zone: Banovo Brdo Park, the park around the Palace of Serbia (SIV Building), Topčider Park. According to the degree of pollution, the following city zones can be distinguished: highly polluted: Bulevar Kralja Aleksandra Street/ Takovska Street (The House of the National Assembly of the Republic of Serbia); - polluted: Požeška Street/ Kirovljeva Street (Banovo Brdo); - moderately polluted: Bulevar Mihajla Pupina Street (SIV) and - slightly polluted: Topčiderska Zvezda (www.envpl.ipb.ac.rs).

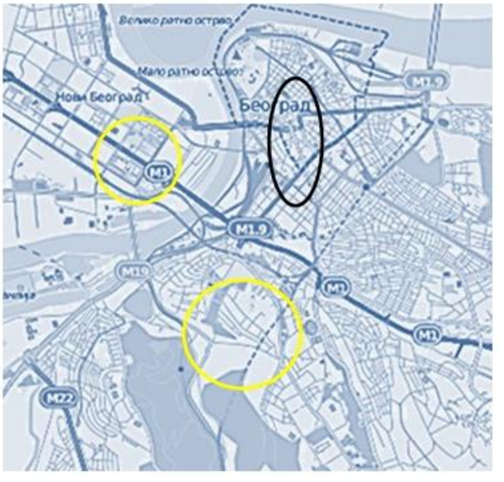

Figure 2. The location of the studied parks within the selected zones in the city of Belgrade - the central city zone of the city (framed in black): Pioneer Park, Academic Park; the wider city zone (framed in yellow): Banovo Brdo Park, The Palace of Serbia (SIV Building) Park, Topčider Park.

According to available data on the concentrations of major air pollutants (*www.eko.bg.gov.rs*) for the period preceding the research (2008-2013), the area of the central city zone (I), in which Pioneer Park is located, is most polluted. We assumed that Academic Park, also located in the central city area, has similar conditions. The concentration of the main pollutants SO_2 and NO_2 in Pioneer Park was above the limit in all study years, i.e., the air was in the category of "polluted" and "highly polluted" (except for SO_2 in 2012 and 2013).

Table 1. Air Quality Index - SAQI_11 (24 h) (www.eko.bg.gov.rs).

Polluting matter	LV* (μg/m^3)	TV** (μg/m^3)	Excellent	Good	Acceptable	Polluted	Highly polluted
CO	5000	10000	0.0-2500	2501-3500	3501-5000	5001-10000	>10000
SO_2	125		0.0-50.0	50.1-75.0	75.1-125.0	125.1-187.5	>187.5
NO_2	85	125	0.0-42.5	42.6-60.0	60.1-85.0	85.1-125.1	>125.0
< PM_{10}	50	75	0.0-25.0	25.1-35.0	35.1-50.0	50.1-75.0	>75.0
O_3-8 h max	120		0.0-60.0	60.1-85.0	85.1-120.0	120.1-180.0	>180.0
Sooth	50		0.0-25.0	25.1-35.0	35.1-50.0	50.1-75.0	>75.0

LV* – limit value; TV** – tolerable value

Other investigated parks are located in the wider urban area (II), but they differ in the degree of pollution. Banovo Brdo Park is in the zone of `polluted` air, which is a direct consequence of the high frequency of traffic in the main Požeška Street which borders the park.

This park had the concentration of SO_2 above the limit only in 2008, while in the subsequent years it was significantly lower and the air was categorized either as "excellent" or as "good". On the other hand, NO_2 concentrations were above the limit value (except in 2008 and 2011) and the air was in the category of "polluted" and "highly polluted".

Table 2. Mean annual concentrations of major pollutants in the area of Belgrade, for the period preceding the research (www.eko.bg.gov.rs).

Location of the measuring station	Pollutant	2008	2009	2010	2011	2012	2013
Bulevar Kneza Miloša Street	CO (mg/m^3)	4.32	6.27	6.97	7.12	6.19	3.96
Bulevar Kralja Aleksandra Street,	SO_2 (μg/m^3)	126	146	236	144.8	65.0	33
The House of the National	NO_2 (μg/m^3)	96	144	142	103.7	157.1	123
Assembly of the Republic of Serbia	VOC* (mg/m^3)	4.57	5.18	6.62	6.45	5.24	3.12
(**Pioneer Park**)	Pb (μg/m^3)	1.16	0.83	0.96	0.56	0.38	0.34
	CO (mg/m^3)	3.32	3.06	3.58	3.40	2.49	2.01
	SO_2 (μg/m^3)	152	53	70	41.3	23.9	20
Požeška Street, Kirovljeva Street (**Banovo Brdo Park**)	NO_2 (μg/m^3)	68	112	128	75.3	143.6	92
	VOC (mg/m^3)	3.68	2.80	3.99	3.48	2.47	2.00
	Pb (μg/m^3)	1.14	0.68	0.78	0.45	0.30	0.30
	CO (mg/m^3)					3.31	2.32
Milentija Popovića Street,	SO_2 (μg/m^3)					28.4	21
Hayatt Hotel	NO_2 (μg/m^3)					146.4	85
(**The Palace of Serbia – SIV Park**)	VOC (mg/m^3)					2.91	2.13
	Pb (μg/m^3)					0.31	0.30

*VOC (volatile organic compounds)

The park around the Palace of Serbia (SIV Building) is located in the "moderately polluted" zone, but according to data on the concentratios of pollutants, it has conditions similar to the above-mentioned park (Banovo Brdo Park). Regarding the content of SO_2, SIV Park has "excellent" air and regarding the content of NO_2, which was above the limit, the air can be categorized as "highly polluted" (2012) and "polluted" (2013). Topčider Park is said to be in the `slightly polluted` zone, although there are no concrete data on the concentrations of certain pollutants. The park is located in the immediate vicinity of the `Košutnjak` park-forest, and thus it can be assumed that it is in the less polluted zone (Tables 1, 2).

3.2. The analysis of pollen

According to the results of the morphological analysis of pollen, the analyzed species vary considerably. According to Erdtman's classification of the size and shape of pollen (1952) and based on the results of our research, the group of small-pollen species includes birch (22.8 μm) and London plane (22.6/15.2 μm), whose pollen is the smallest, and paulownia whose pollen is at the border between the small and medium pollen (27.6/13.6 μm). Chestnut (30.0/14.5 μm), ginkgo (34.1/15.3 μm), linden (37.3 μm) and oak (38.8/22.0 μm) have medium pollen. Of all the analyzed species, Turkey oak has the largest pollen (44.1/28.0 μm), which, according to the above classification, also belongs to the category of medium pollen.

Birch and linden have the most spheroidal (round) pollen with the index of the pollen grain size of 100.0%. The group of species with oblate pollen grains includes London plane (67.6%). Oaks also have oblate pollen (57.0%). Peroblate pollen is typical of ginkgo (45.1%), chestnut (48.6%) and paulownia (49.4%) (Figure 4).

Pollen color also varies between species. Most of the analyzed woody species have pollen in the shades of yellow. Chestnut and paulownia have significantly different pollen colours. They are reddish and gray/white respectively (Figure 3a-i).

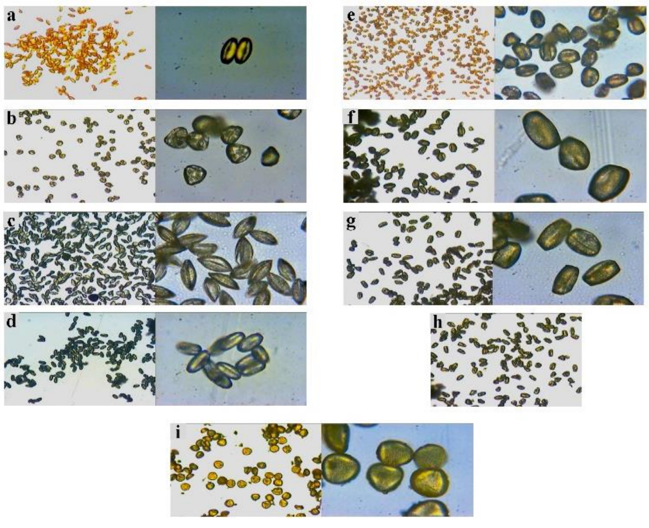

Figure 3. Pollen of the studied species: a) *Aesculus hippocastanum* L., b) *Betula alba* L, c) *Ginkgo biloba* L. d) *Paulownia tomentosa Steud.*, E) *Platanus x acerifolia* (Aiton) Willd, f) *Quercus cerris* L., g) *Quercus robur* L., h) *Quercus rubra* L. i) *Tilia argentea* Desf. The first image is pollen with 100x zoom and the second is 400x zoomed.

3.3. Analyzed species and parks - zonal (spatial) variability

The pollen of *Aesculus hippocastanum* is 30.0/14.5 μm/48.6% (length/width/shape) at the level of all analyzed trees and parks and it is peroblate (Figure 4). The size of the pollen varies considerably between the analyzed zones ($p<0.0001$) and parks within the zones ($p<0.05$), except for the index of pollen grain shape (Table 3; Figure 5). The trees in Banovo Brdo Park have the smallest pollen (29.0/13.9 μm/48.3%), and the trees in Topčider Park the largest (30.8/14.9 μm/48.6%).

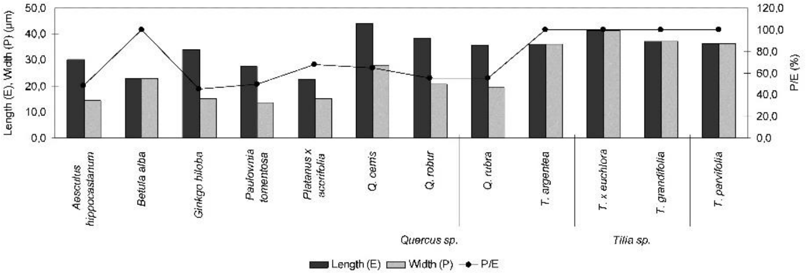

Figure 4. Morphological properties of the pollen of analyzed species.

Betula alba pollen is 22.8 μm (length=width) and it is spheroidal (P/E=100.0%) (Figure 4). The pollen size varies considerably between the zones and parks ($p<0.0001$; Table 3; Figure 5).

The smallest pollen grains of birch trees were found in SIV Park (33.3/15.4 μm/46.7%) and the largest in Pioneer Park (34.9/15.0 μm/43.3%).

Ginkgo biloba pollen is 34.1/15.3 μm/45.1% and it is peroblate (Figure 4). There are significant differences between the studied zones (except for the pollen width; p< 0.01), while there is no difference between the parks within a zone (Table 3; Figure 5). The smallest pollen was recorded in SIV Park trees (33.3/15.4 μm/46.7%) and the largest in Pionir Park trees (34.9/15.0 μm/43.3%).

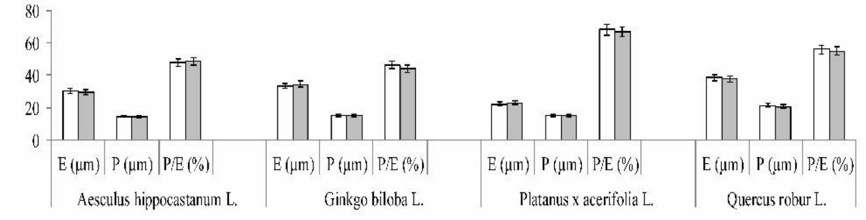

Figure 5. Differences in the values of the analyzed properties of pollen grain: length (E – equatorial axis, μm), the width (P – polar axis, μm) and the pollen shape index (P/E, %) for species *Aesculus hippocastanum* L., *Ginkgo biloba* L., *Platanus x acerifolia* L. and *Quercus robur* L. in the city zones (the wider city zone - white bars and the central city zone of the city – gray bars).

Table 3. Results of ANOVA analysis for each of the analyzed species with zone and parks as sources of phenotypic variability.

Source of variation		Lenght (E) (μm)		Width (P) (μm)		Index of pollen shape P/E (%)	
	df	MS	F	MS	F	MS	F
Aesculus hippocastanum L.							
Zone	1	385.22	50.59****	44.65	12.18****	87.07	1.73
Park(Zone)	3	27.45	3.61*	20.47	5.58****	71.36	1.42
Error	995	761		3.67		50.22	
Betula alba L.							
Zone	1	490.39	54.59****				
Park(Zone)	2	117.77	13.11****				
Error	796	8.98					
Ginkgo biloba L.							
Zone	1	99.79	6.66***	2.68	0.55	373.19	7.93**
Park(Zone)	1	23.74	1.58	6.09	1.26	194.04	1.26
Error	297	14.98		4.85		51.62	
Platanus x *acerifolia* (Aiton) Willd.							
Zone	1	181.06	25.63****	10.27	1.85	791.28	6.49*
Park(Zone)	2	66.63	9.43****	184.33	33.25****	2384.63	19.55***
Error	1096	7.06		5.54		121.97	
Quercus robur L.							
Zone	1	111.86	10.5**	152.01	13.93****	339.23	3.92*
Park(Zone)	1	0.004	0	108.06	9.84***	847.89	9.97***
Error	597	1065		10.96		86.63	

* P< 0.05; ** P< 0.01; *** P< 0.001; **** P< 0.0001

Paulownia tomentosa pollen is 27.6/13.6 μm/49.4% and it is peroblate (Figure 4). The analyzed trees do not differ significantly in terms of the size of pollen (28.3/13.6 μm/48.3% *vs.* 26.9/13.5 μm/49.4%).

Platanus x acerifolia pollen is 22.6/15.2 μm/67.6% and it is oblate (Figure 4). The studied zones show differences in the length of pollen and in the index of pollen shape (p< 0.0110), while there are significant differences between the parks in all the properties analyzed (p< 0.0001;

Tabela 3; Slika 5). The smallest pollen was found in Topčider Park trees (21.9/15.2 μm/70.1%), and the largest in Academic Park trees (23.4/16.1 μm/69.3%).

The pollen of *Quercus* sp. averages 38.0/22.0 μm/57% and it is oblate (Figure 4). There are significant differences within some species. *Q. rubra* (35.5/19.5 μm/53.3%) has the smallest and most oblate pollen. The pollen of *Q. robur* (38.1/20.8 μm/55.0%) is slightly larger and the pollen of *Q. cerris* is significantly larger and less oblate (44.1/28.0 μm/64.4%) than the above-mentioned species. The size of pedunculate oak pollen varies between the zones (for all properties analyzed, $p>0.05$) and between the parks within a zone (except for the length of pollen) ($p>0.001$; Table 3; Figure 5). Pedunculate oak trees have the smallest pollen in Banovo Brdo Park (37.8/21.1 μm/56.1%) and in Academic Park (37.8/19.8 μm/52.8%), and the largest in Topčider Park (38.8/21.6 μm/56.2%).

The pollen of *Tilia* sp. is 37.3 μm/100.0% (length=width) and it is spheroidal (Figure 4). *T. x euchlora* has the largest pollen (41.4 μm) of all linden species. *T. grandifolia* (37.3 μm), *T. parvifolia* (36.3 μm) and *T. argentea* (36.1 μm) have pollen of similar size, the largest of which is the pollen of *T. grandifolia*. Pollen size has statistically significant differences between parks only for *T. parvifolia* ($p<0.0001$).

The share of phenotypic variability conditioned by environmental differences (park effect), as well as the effect of species and trees were statistically significant for all studied pollen properties (all $p<0.05$) (Table 4).

3.4. Interindividual variability

The size and shape of pollen were in most cases different between individual trees of one species, which indicates individual or intraspecific variability. The pattern of the differences in the size and shape of pollen was specific for each individual (tree) studied within a species, which confirms the interindividual (genetic) variability of the analyzed pollen properties within a species ($p<0.05$; Table 4).

Table 4. Results of ANOVA analysis for morphological properties of pollen grains (Length (E), Width (P) and Index of pollen shape (P/E)).

Source of variability	Length (E) (μm)			Width (P) (μm)			Index of pollen Shape (P/E) (%)	
	df	MS	F	df	MS	F	MS	F
Park	4	76.13	6.68 ****	4	51.91	31.34****	23.71	6.24***
Species (Park)	24	634.12	55.65 ****	13	80.50	48.54****	79.25	20.86****
Tree (Park species)	29	11.40	21.15 ****	18	1.66	9.65****	3.80	11.78***
Error	5742	0.54		3564	1.66		0.17	

4. Discussion

In the last decades, Belgrade has been a city characterized by rapid urbanization (industry development, increasing population, higher traffic density), which has resulted in a change in the state of the environment (increased levels of harmful gases and various pollutants). For instance, the mean annual PM_{10}, $PM_{2.5}$ mass concentrations (μg m^{-3}) were significantly higher for Belgrade-2002 (77; 61, respectively) than for some other European cities such as Milan-1998. (103-winter, 68-summer; 66-winter, 43-summer, respectively), Madrid-1999-2000. (48; 34, respectively), Berlin-1998. (38, 30, respectively), Roma-1999. (60) (Tasić et al., 2006). Based on the data of regular air monitoring in Belgrade, zones of low and high pollution can be identified. A significant increase in the concentration of SO_2, NO_2, CO, O_3 has been found especially in the central zone of the city (Tasić, 2016). Woody species, which account for a significant portion of park plant communities, are prone to numerous structural and physiological changes that can be used as bioindicators of the state of the environment. Apart from the structural changes in the plant tissue of leaves and bark (Aribal et al. 2016; El-Khatib et al. 2016; Chrabaszcz and Mroz, 2017), the changes in pollen, both in the morphology and in the protein content, also contribute to the understanding of the impact of pollution on the urban environment (Bianchimano et al. 2014).

Based on the conducted research, it can be concluded that increased pollution in the urban environment, induced primarily by human activity, affects the analyzed properties of pollen. The distinguished zones of low or high air pollution had different patterns of response to morphological properties of pollen. A decrease in the size of pollen grains was recorded in the areas of increased air pollution and vice versa. It was confirmed that future monitoring projects can use pollen size as an indicator of the change in the pollen morphology under the influence of human-induced stress. Furthermore, significant differences were confirmed not only between the analyzed species (interspecific variability) but also between individual trees within the same species (interindividual variability). Given that trees in one locality grow in similar environmental conditions, the recorded differences can be attributed to individual variability within a species, that is, to the genetic potential of the responses of woody forms of park communities to the impact of environmental pollution induced by human activity.

5. Conclusions

As woody deciduous species represent a large and significant portion of park plant communities, they can be used in biomonitoring programs of the urban environment in which human activity contributes to the increase in pollution and, consequently, affects the growth of plants. Studies of the impact of human-induced stress on the morphology of pollen are necessary in order to estimate the reproductive potential of plants in urban areas. The share of phenotypic variability determined by environmental differences within the zones (differences between parks) is statistically significant for the analyzed pollen properties (except for all ginkgo pollen properties, pedunculate oak pollen grain length and chestnut pollen shape). This research makes a good basis for the monitoring of park woody species with the aim of achieving their long-term survival in the urban environment. Based on preliminary results, it can be concluded that increased pollution of the environment in the central city zone contributes to the reduction in pollen size, and future monitoring projects can use this feature as a bioindicator of the change in the pollen morphology under the influence of human-induced stress.

Acknowledgments

This paper was realized as part of the project "Studying climate change and its influence on the environment: impacts, adaptation and mitigation" (43007) financed by the Ministry of Education, Science and Technological Development of the Republic of Serbia within the framework of integrated and interdisciplinary research for the period 2011-2018, and project "Morphological - anatomical and physiological changes on woody species in Belgrade parks as an indicator of the state of the environment" (2014-2016), fInanced by the Secretariat for Environmental Protection, Belgrade.

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