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Particulate matter accumulation – further differences between native *Prunus padus* and non-native *P. serotina*

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Abstract: Particulate matter (PM) is one of the most harmful inhaled pollutants. Where pollutants have been emitted into the atmosphere, the most effective method for cleaning the air is through phytoremediation, whereby plants act as biological filters. PM has a negative impact on plants, but knowledge of PM effects on the photosynthetic apparatus is limited. In European forests, species of the genus *Prunus* L. play a key role in the composition of the forest understory and urban as well as industrial plantings. Shrubs of the native *P. padus* L. and closely-related invasive alien *P. serotina* Ehrh. are particularly widespread. Thus, both are good model species in which to study the impact of PM pollution.
The aim of this study was to assess the accumulation of PM in the context of leaf morphology and amount of epicuticular waxes on foliage, and the efficiency of the photosynthetic apparatus of *P. padus* and *P. serotina*. The study was conducted under controlled conditions using two variants of dust, cement and roadside PM. In addition, we analyzed the absorption of dust by leaves dividing it into three fractions by size (10−100 μm, 2.5−10 μm and 0.2−2.5 μm). Results showed that both *P. padus* and *P. serotina* accumulate PM mostly on the surface of their leaves ($_S$PM), rather than in the wax layer ($_W$PM). *P. padus* accumulated higher amounts of PM than did *P. serotina*. The higher presence of PM on leaves of *P. padus* resulted in a reduction of the efficiency of the photosynthetic apparatus, manifested by lower rates of photosynthesis and chlorophyll *a* fluorescence, coinciding with an increased stomatal resistance. A strong negative correlation was found between the amount of PM accumulation and the efficiency of the photosynthetic apparatus in *P. padus*, but not in *P. serotina*.
We have concluded that alien *P. serotina* is more tolerant to the conditions of stress caused by PM pollution than is the native *P. padus*, which may partly explain its success in the invasion in Europe.

Keywords: particle deposition, PM, shrubs, black cherry, bird cherry, leaf morphology

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Introduction

Despite high expenditures on environmental protection, we continue to observe a significant deterioration of air quality in urban areas (OECD, 2012; WHO, 2016). Several studies have highlighted that one of the most dangerous components of air pollution to human health is a mixture of liquid and solid particles called particulate matter (PM; Bell et al., 2011; Jędrychowski et al., 2011; Weuve et al., 2012). These pollutants can be divided into large PM (10–100 μm), coarse PM (2.5–10 μm), fine PM (0.1–2.5 μm) and ultra-fine PM (<0.1 μm; Power, 2009; Popek et al., 2013). PM is formed by natural forces and human activity. Natural forces that cause PM include volcanic activity, forest fires and dust storms, whereas anthropogenic PM is created as a result of the incomplete combustion of fuel in car engines, road surface erosion, tyre wear and industrial activities (Farmer, 2002). PM of human origin is also frequently coated by other toxic components, such as black carbon, heavy metals, and polycyclic aromatic hydrocarbons (Jouraeva et al., 2002; Alghamdi, 2016).

Epidemiological studies investigating the impact of particulate matter on the human population have shown a negative correlation between the amount of PM in air and human health (Jędrychowski et al., 2011; Weuve, 2012; Saravia et al., 2013; Hsu et al., 2015). It has been estimated that exposure to PM causes approximately 2.1 million premature deaths annually worldwide, associated with cardiopulmonary diseases and lung cancer (Silva et al., 2013).

Technical equipment, such as filters and electrostatic precipitators, are effective in reducing dust emissions, but only for certain point sources. In contrast, plants that are widespread and widely-distributed play an important, uninterrupted function in reducing the emission of pollutants (Yang et al., 2005). Numerous studies have proven that deciduous trees and shrubs are efficient passive particulate matter collectors (Sæbø et al., 2012; Popek et al., 2013; Przybysz et al., 2014; Popek et al., 2015). Moreover research has shown that species differ in their ability to accumulate PM on the surface of their leaves (Dzierżanowski et al., 2011; Dzierżanowski & Gawroński, 2011; Nawrot et al., 2011; Sæbø et al., 2012; Popek et al., 2013; Song et al., 2015). The amount of PM gathered on leaves depends on the density of the plant, the shape of the leaves and the presence of other morphological structures, such as various types of spines, thorns and hairs (trichomes) on the leaf surface. The accumulation of PM can also be increased by the presence of epicuticular waxes, in which particular matter can be immersed (Jouraeva et al., 2002; Leonard et al., 2016).

Vegetation in urban and industrial areas significantly helps to limit the amount PM suspended in the air (Yang et al., 2005; McDonald et al., 2007; Vailshery et al., 2013). Conversely, PM also has a negative impact on plants (Armbrust, 1986; Hirano et al., 1995; Naidoo & Chirkoot, 2004; Nanos & Ilias, 2007). Particles accumulated on the surface of leaves can absorb and scatter light rays, preventing them from accessing the chloroplast freely (Hirano et al., 1995). Particles can also be located in the openings of the stomata, where they reduce the intensity of transpiration what can negatively affect on plant heat tolerance (Cape, 2009; Burkhardt & Grantz, 2017). Moreover, chemically-active particles consisting of metal oxides, calcium, cement particles and heavy metals can affect the physiological processes of plants, such as photosynthesis and respiration (Farmer, 2002; Chauhan, 2010; Abdel-Rahman & Ibrahim, 2012). In an environment with high levels of PM pollution, the amount of chlorophyll in plants cells may also be reduced (Prusty et al., 2005), and the fluorescence of chlorophyll *a* may be decreased (Naidoo & Chirkoot, 2004).

In European forests, species of the family *Rosaceae*, including the genus *Prunus* L., play a key role in the species composition of the understory (Pairon et al., 2010). Particularly widespread are shrubs, or more rarely trees, of *P. padus* L. (bird cherry) and the closely-related *P. serotina* Ehrh. (black cherry). Bird cherry is a species native to Europe, with a wide geographic range that extends from the southern to the northern limits of the continent (Łukowski et al., 2014; Houston Durrant & Caudullo, 2016). In contrast, black cherry is an alien species in Europe,originating from the north-eastern and central parts of the USA, Mexico and the north of South America (Pairon et al., 2010). Both species are found in similar natural habitats in Poland (Dyderski & Jagodziński, 2015; Łukowski et al., 2017); however, black cherry can be observed growing in soil extremely poor in minerals or in heavily anthropogenically-transformed habitats (Jagodziński et al., 2015). Moreover, these species are very common in the understory of urban forests and urban plantings (Dyderski et al., 2016), and often choose well-lit forest edges or roadsides (Mizera et al., 2016). Both species are therefore good model systems in which to study the impact of PM pollution, and the interaction of native and non-native plant species from the perspective of accumulation and response to PM pollution. Although there are a number of studies concerning the impact of dust on trees typically growing in urbanised areas, there is little research concerning forest ecosystems that are often located near large urban areas. Recent studies clearly show that Mediterranean urban forest ecosystems can sequestrate particles (Fares et al., 2016), but there is limited data concerning the impact of PM accumulating on the leaves of temperate broad-leaf forest plant species.

The aim of this study was to assess: (i) the accumulation of PM (total, surface and in-wax in three size fractions) on foliage, (ii) the amount of epicuticular waxes on the surface of leaves, and (iii) the efficiency of the photosynthetic apparatus in bird cherry (*P. padus*) and black cherry (*P. serotina*), the leaves of which were experimentally polluted with PM with different source variants (cement plant and roadside). We hypothesised that: (1) PM accumulation on leaves is higher on *P. padus* than on *P. serotina*, due to differences in leaf structure and the amount of waxes on their surface; (2) waxes play an important role in catching PM, resulting in significant fraction of PM contained in wax relative to surface PM even under controlled conditions where dust may not be rinsed off by rain; and (3) PM has a negative impact on the photosynthetic apparatus, and the effect is greater in *P. padus* than in *P. serotina*.

Materials and methods

Plant material and study area

In one vegetation season of 2015, studies were conducted on *P. padus* L. and *P. serotina* Ehrh. Seeds of both *Prunus* species of local origin (from the Palędzie Forest; 52°23'N, 16°40'E) were sown in pots (15 dm^3) in mid-April 2007, after stratification in the Laboratory of Seed Biology (Institute of Dendrology, Kórnik, Poland). The pots were filled with forest soil (collected in a mature oak/pine forest) mixed with neutralised peat (1:1) and the slow-release N-P-K fertiliser Osmocote (2 $kg \cdot m^{-3}$; Scotts Poland, Warszawa, Poland). All seedlings chosen for the experiment were the same age (8 years old), had an average height of 50 cm and were in good condition (healthy and free from pests). To protect plants from outside stress factors, especially from accumulation of PM from the air and the possibility of rinsing away dust used in the experiment via rain, they were placed in greenhouses in appropriate growing conditions. Plants were divided into three groups of four plants of each *Prunus* species and each group was placed in a separate greenhouse. Seedlings in the two first groups were dusted by equal volumes of either (i) roadside PM or (ii) cement PM, consisting of particles less than 100 μm in diameter. Roadside PM was collected from the edge of busy streets (Bolesława Krzywoustego, Matyi and Aleja Niepodległości) in Poznań, Poland (52°24'N, 16°56'E) and mixed, whereas the second variant of PM consisted of sieved Portland cement CEM I 42,5R (Cemmas A.S., Poland). Mass based ratios of particle size fractions in the applied dust were 86:12:2 for roadside PM and 93:5:2 for cement PM, using large (10–100 μm), coarse (2.5–10 μm) and fine (0.2–2.5 μm) fractions, respectively. The third group, constituting controls, was not subjected to any stress conditions. All seedlings were watered as necessary.

Analysis of accumulation of PM and epicuticular waxes on leaves

Sample collection

For each PM variant, leaves were harvested from seedlings at the end of the growing season (n = 3 PM source variants × 2 *Prunus* species × 4 seedlings). In order to obtain sufficient material to determine the fine fraction of PM and still avoid filter blockage by particles during filtration, the leaf area per sample ranged from 300 to 400 cm^2. Samples consisted of leaves gathered from different branches of crowns, making them representative of each seedling. Leaves (only leaf blades without petioles) were placed in paper bags, labelled and kept at ambient temperature until analysis.

Quantitative analysis of PM and epicuticular waxes

The amount of PM was determined in two categories: washed with water – surface PM ($_S$PM), and washed with chloroform – in–wax PM ($_W$PM). Both categories were divided in in three size fractions (10–100 μm, 2.5–10 μm, 0.2–2.5 μm) and determined by weighting. Is should be noted that the number of particles within each size fractions may vary. Each leaf was washed with water and then with chloroform. The liquids were filtered using Type 91 and Type 42 paper filters, and PTFE membrane filters (all Whatman, UK), with pore sizes of 10 μm, 2.5 μm and 0.2 μm, respectively. Before and after filtration, all filters were weighed. The quantity of waxes was weighed after evaporation of chloroform collected (after filtration) to pre-weighed beakers. The amounts of PM and waxes were then converted to $\mu g \cdot cm^{-2}$ after measuring the leaf area of samples using a scanner and WinFOLIA 2004 software (Regent Instruments Inc., Canada).

Evaluation of efficiency of photosynthetic apparatus

Measurements were carried out four times after dust application, at monthly intervals throughout the growing season (in mid-June, mid-July, mid-August and mid-September). The following parameters of efficiency of photosynthetic apparatus were measured *in vivo*.

Plant gas exchange

As part of the gas exchange measurements, photosynthetic rate and stomatal conductance were evaluated using an infrared gas analyser (LI-6400 Photosynthesis System, LI-COR Inc., Lincoln, Nebraska, USA) equipped with the 6 cm^2 leaf chamber. Measurements were conducted during cloudless weather, between 9 a.m. and 1 p.m. Leaves were randomly selected from the middle part of the annual twigs. Each leaf was enclosed in the leaf chamber and maintained until the photosynthesis rate stabilised, when a measurement was recorded. For each plant, four separate measurements were conducted on each of three leaves (biological replicates). In every month of the study period, leaves were selected for measurements from the same part of the plant (n = 3 PM source variants × 2 *Prunus* species × 4 months × 4 seedlings × 3 leaves).

Chlorophyll a fluorescence determination

Measurements of chlorophyll *a* fluorescence were performed using the portable Handy PEA (Hansatech, Pentney, UK) on leaves used for gas exchange study. Thirty minutes prior to the measurement, dark adaptationclips were placed on the leaves and the shutter was closed. Measurements of minimal (F_o) and maximal (F_v) fluorescence of chlorophyll *a* were taken using a pulse of high intensity (3000 $\mu mol \cdot m^{-2} \cdot s^{-1}$) light. Maximum quantum efficiency of photosystem II (F_v/F_m where F_v is the difference between F_m and F_o) was calculated by the instrument's software (n = 3 PM source variants × 2 *Prunus* species × 4 months × 4 seedlings × 3 leaves).

Statistical analysis

Two-way analysis of variance (ANOVA) with interaction was used to assess the statistical significance of the impact of the two *Prunus* species and PM source on the amount of PM present (total and according to size category), the amount of epicuticular waxes, the photosynthetic rate, stomatal conductance and the maximum quantum efficiency of photosystem II. In order to facilitate interpretation of the results of the above mentioned parameters of photosynthesis, only mean seasonal values are presented in the figures, without regard to the date of measurement during the growing season. In the Results section, however, we refer to the influence of this independent variable. Relationships between the amount of $_W$PM on leaves and epicuticular waxes, the photosynthetic rate and total PM amount, and the stomatal conductance and amount of fine PM 0.2−2.5 μm fraction were evaluated using Pearson's correlation coefficient. A Tukey's HSD test (P = 0.05) was then employed to assess the significance of differences between variants. The presented data are given as means with standard error (±SE). All calculations were performed using JMP Pro 12.1.0 software (SAS Institute Inc., Cary, NC, USA).

Results

For all plants tested at the end of the growing season, particulate matter of two types ($_S$PM and $_W$PM) and all three size fractions was found. *Prunus padus* accumulated approximately 14% more PM in the control group, 67% more cement PM and 45% more roadside PM than did *P. serotina* (Fig. 1). Additionally, for both *Prunus* species, more PM was deposited on the leaf surface than was immobilised in waxes (Table 1). Species and PM source have a significant effect on the amount of $_S$PM and $_W$PM on leaves. Regardless of the source of the PM, approximately 60% of total PM was $_S$PM and 40% was $_W$PM in *P. padus*, with 75% and 25%, respectively, in *P. serotina*. We also found that most PM accumulated was from the roadside PM variant (a), the next highest amount came from the cement variant (b), and the least was found in control (c; Tukey's HSD). A similar dependence of the influence of the variant of dust upon its accumulation on the leaves is evident in the comparison of each *Prunus* species (Fig. 1).

The amount of waxes on leaves differed significantly between the two species, but we found no effect of PM source (Table 1). *P. padus* had 25% more waxes than did *P. serotina*. A significant correlation was found between the amount of waxes and $_W$PM accumulation, based on data for both species (r = 0.74; P < 0.0001; Fig. 2).

Out of all the size fractions measured, regardless of species and dust source, the majority (80%) were large PM (10–100 μm), followed by 16% coarse (2.5–10 μm) and 4% fine PM (0.2–2.5 μm; Table 1). On the leaves of both *Prunus* species there was a lower amount of large PM, and more coarse and fine PM in comparison to the initial composition of the dust. *Prunus* species and PM source had a significant effect on PM accumulation on the leaves for all size

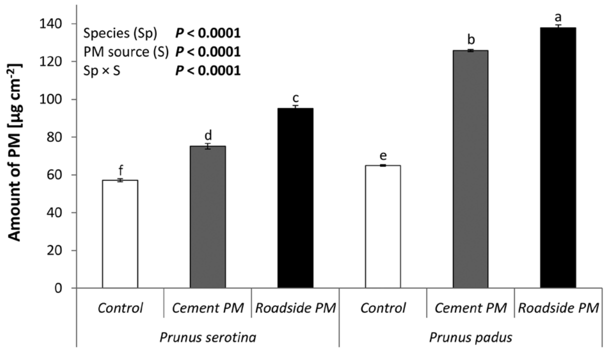

Fig. 1. Total amount of particulate matter (PM) from different sources deposited on leaves of *Prunus padus* and *P. serotina*. Analysis of variance (ANOVA) was used to assess the statistical significance and Tukey's HSD test (P = 0.05) was employed to assess the significance of differences between PM variants

Table 1. The amount of particulate matter (PM) from different sources divided into surface PM ($_{S}$PM) and in-wax PM ($_{W}$PM) and three size fractions. The amount of epicuticular waxes on leaves of *Prunus padus* and *P. serotina* is also presented. Analysis of variance (ANOVA) was used to assess the statistical significance and Tukey's HSD test (P = 0.05) was employed to assess the significance of differences between variants. *P* values <0.05 are in bold

Species	PM Source variant	$_{S}$PM [μg·cm^{-2}] (mean ±SE) Size fraction [μm] 10–100	2.5–10	0.2–2.5	Total $_{S}$PM [μg·cm^{-2}]	$_{W}$PM [μg·cm^{-2}] (mean ±SE) Size fraction [μm] 10–100	2.5–10	0.2–2.5	Total $_{W}$PM [μg·cm^{-2}]	Epicuticular waxes [μg·cm^{-2}] (mean ±SE)
Prunus padus	Control	28.42 ±1.01f	8.94 ±0.28c	1.65 ±0.11c	39.0 ±10.87f	18.44 ±0.52b	6.91 ±0.86b	0.63 ±0.08d	25.98 ±0.76b	81.22 ±3.45
	Cement	59.50 ±1.42b	12.44 ±0.26b	2.31 ±0.17b	74.25 ±1.22b	39.29 ±0.80a	10.48 ±0.53a	1.76 ±0.04a	51.54 ±1.15a	78.26 ±3.92
	Roadside	66.54 ±0.51a	15.81 ±0.20a	4.71 ±0.17a	87.06 ±0.19a	41.90 ±1.66a	7.35 ±0.17b	1.53 ±0.09ab	50.78 ±1.73a	79.39 ±3.21
Prunus serotina	Control	40.04 ±0.87e	4.17 ±0.3e	1.32 ±0.11c	45.54 ±0.91e	8.38 ±0.27d	2.66 ±0.15d	0.56 ±0.08d	11.60 ±0.23d	61.01 ±2.02
	Cement	49.72 ±0.97d	5.76 ±0.22d	1.66 ±0.12c	57.14 ±1.11d	12.20 ±0.33c	4.84 ±0.22c	0.98 ±0.23c	18.02 ±0.53c	64.62 ±2.76
	Roadside	56.10 ±0.57c	8.69 ±0.27c	2.27 ±0.09b	67.06 ±0.78c	20.48 ±0.55b	6.24 ±0.35b	1.37 ±0.06b	28.09 ±0.88b	65.73 ±3.07
ANOVA						*P* value				
Species		**0.0016**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**0.0023**	**<0.0001**	**<0.0001**
PM source variant		**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	0.8899
Species × PM source variant		**<0.0001**	**0.0005**	**<0.0001**	**<0.0001**	**<0.0001**	**0.0003**	**0.0123**	**<0.0001**	0.4954

fractions and both types of PM ($_{S}$PM and $_{W}$PM). In each of these cases, the amount of PM was greater in *P. padus* than in *P. serotina*.

The rate of photosynthesis measured on control *P. padus* seedlings was 12% and 21% higher than in the cases of plants polluted by cement dust and roadside dust, respectively (Fig. 3A). In contrast, the difference in rate of photosynthesis between *P. serotina* control and polluted plants was within 3%. Additionally, a strong negative correlation was found between photosynthesis rate and total amount of PM accumulation in *P. padus* (r = –0.92; P < 0.0001), but not in *P. serotina* (r = 0.04; P = 0.90). Reduction in

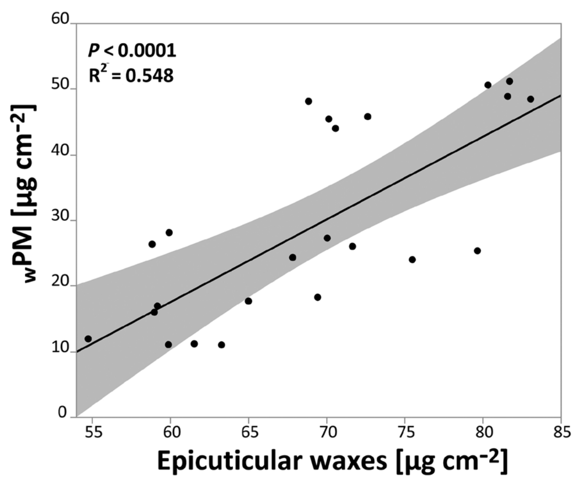

Fig. 2. Relationship between epicuticular waxes on leaves of *Prunus padus* and *P. serotina* (taken together), and in-wax PM ($_{W}$PM) accumulation. The curve shows the resulting regression. Coefficients of determinations, 95% confidence interval and associated probability are given

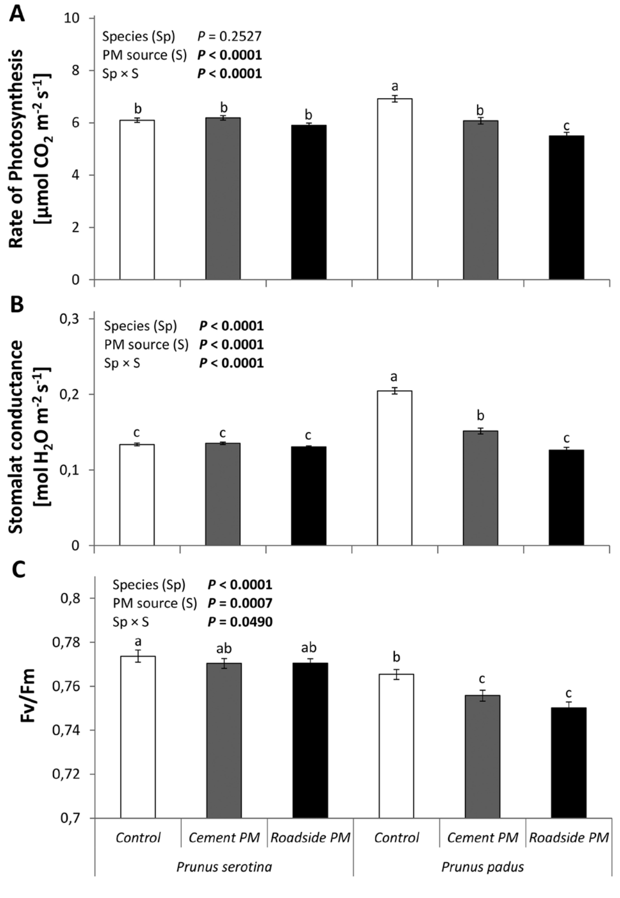

Fig. 3. Rate of photosynthesis, stomatal conductance and maximum quantum efficiency of photosystem II (Fv/Fm) of *Prunus padus* and *P. serotina* dusted by cement particulate matter (PM) and roadside PM. Analysis of variance (ANOVA) was used to assess the statistical significance and Tukey's HSD test (P = 0.05) was employed to assess the significance of differences between PM variants

stomatal conductance was associated with a greater accumulation of dust in *P. padus* (Fig. 3B). We also found a strong negative correlation between stomatal conductance and the accumulation of fine PM (0.2–2.5 μm) for *P. padus* (r = –0.76; *P* = 0.0045), but not for *P. serotina* (r = –0.18; *P* = 0.57). The results of chlorophyll *a* fluorescence showed no significant differences between dusted and control plants based on the parameter Fv/Fm for *P. serotina*, but a reduction of this parameter was recorded in polluted *P. padus* seedlings (Fig. 3C).

Discussion

We found two types of PM accumulation: on the leaf surface ($_S$PM) and in-wax layer $_W$PM), in all three size fractions of particulate matter. Even the control plants, although shielded from external contamination, accumulated a certain amount of PM, possibly originating from the exchange of air between the greenhouse and the outside environment. The results documented the differences in PM accumulation between *P. padus* and *P. serotina* (Fig. 1). Prusty et al. (2005) also observed significant variations in dust interception ability among plant species, and implied that the dust interception capacity of plants depends on their canopy shape and size, leaf phyllotaxy and leaf surface characteristics, such as hairs and cuticle. Although our studied species are very closely related, they differ in anatomy and morphology of leaves (Danielewicz & Wiatrowska, 2013). *P. serotina* has long, narrow, smooth leaves, with no appendages (Seneta & Dolatowski, 2008). Bakker et al. (1999) showed that *Plantago* species with smooth leaf surfaces accumulated smaller amounts of PM than did species with rough leaves. *P. padus* leaves are distinctly wrinkled, with numerous hollows and ridges. Moreover, both species have convex veins with trichomes on the abaxial side of the leaf blade, between which PM can be accumulated, whereas in *P. serotina*, these features may be more important because of denser trichomes along the midrib (Danielewicz & Wiatrowska, 2013). It has been reported in several studies that PM accumulation increases on the leaf margins (Mitchell et al., 2010) or along the main leaf nerves (Tomašević et al., 2005). Many morphological characteristics, such as surface roughness and presence of trichomes, have been shown to increase the amount of PM on leaves (Bakker et al., 1999; Beckett et al., 2000; Leonard et al., 2016). We speculate that in *P. serotina* PM was mainly accumulated in denser trichome layer along the midrib, but in *P. padus* due to structure of leaves, for example numerous hollows and ridges on leaf blade, while these features are not equal in terms of the level of accumulation. This aspect requires additional research, taking into account other features of leaves, important for PM accumulation, such as the length of the petioles (Prusty et al., 2005). These results are consistent with hypothesis 1, because the differences in construction of both species' leaves may potentially explain the differences between PM accumulation on the foliage of native *P. padus* and alien *P. serotina*.

We have shown that in both species, particulate matter was found both on the surface PM ($_S$PM) and in-wax PM ($_W$PM). Such a relationship was also found in studies conducted by Dzierżanowski et al. (2011) and Popek et al. (2013, 2015). It has been shown that in nature, $_S$PM can be washed off of foliage by rain or blown away by strong wind, whereas $_W$PM particles can stick to the surface or penetrate the wax layer and be immobilised (phytostabilised; Jouraeva et al., 2002). Gawrońska & Bakera (2015) suggested that a greater amount of waxes deposited on leaves can be a response of plants to conditions of stress. These authors found higher amounts of waxes on plants growing in an area with more polluted air than on those growing in a clean area. It should be mentioned that the total epicuticular wax layer is a very variable trait and may also vary with season, temperature and light (Müller & Riederer, 2005). Even within a species, the wax of plants growing in different locations can have different structural and chemical characteristics, and for this reason the amount of accumulated PM may also vary (Jeffree, 2007). These results thus support hypothesis 2, because waxes play an important role in catching PM on leaves of both species, although with varying efficiency.

Confirming findings by Sæbø et al. (2012) and Przybysz et al. (2014), also in our research the large size PM fraction (10–100 μm) comprised the largest proportion of all accumulated PM. More of this fraction was noted on leaves in roadside dust than in cement dust, and much greater accumulation was observed on *P. padus* than on *P. serotina*. Farmer (2002) showed that the large fraction is frequently found in more polluted areas near the source of emission. This PM fraction is not as dangerous to human health as is PM with a smaller diameter, but it can have a significant negative impact on plants, particularly on photosynthesis (Vardaka et al., 1995).

Smaller amount of coarse PM (2.5–10 μm) than the largest PM was present on the leaves, which is consistent with the initial composition of the applied dust. Generally, taking into account the studied variants, the obtained results are similar to those of the large PM fraction, with greater amounts noted on leaves in roadside dust than in cement dust, and with much greater accumulation observed on *P. padus* than on *P. serotina*. In the literature, studies concerning coarse PM particles are gaining increasing attention, for example due to fact that high daily mean levels of this PM fraction are associated with increased cardiovascular mortality (Villeneuve et al., 2003). The phytoremediation of this PM fraction, especially in highly-polluted areas, could contribute to an improvement in the health of the local population; therefore, it is worth supporting the presence of native *P. padus,* rather than alien *P. serotina* in urban forests and urban plantings.

Accumulation of fine PM (0.2–2.5 μm) deposited on leaves contributed the least amount of pollutants. Similarly to the case of large and coarse PM, greater amounts of this fraction were noted on leaves in roadside dust than in cement dust, and greater accumulation occurred on *P. padus* than on *P. serotina*. Removing the smallest PM from the air is the most important, because this fraction causes the greatest threat to human health (Weuve et al., 2012; Jędrychowski et al., 2015) and plants (Chen et al., 2015).

Our study showed that the photosynthetic apparatus efficiency of the examined species was significantly negatively affected by amount of PM only in the case of *P. padus*. Also Kuki et al. (2008) showed that the Brazilian pepper tree (*Schinus terebinthifolius* Radii) reacts strongly to increased amounts of PM in the air by reducing its rate of photosynthesis, but no negative effect was observed in the yellow necklace pod (*Sophora tometnosa* L.). In our study, *P. serotina* accumulated less PM on its leaves, which could protect it from the higher negative impact and could be another reason why this shrub is more tolerant to stressful conditions. Lowering the rate of photosynthesis due to the presence of PM accumulated on the surface of leaves can result from changes in the optical properties of the leaves and the decrease in radiation reaching the chlorophyll antenna, due to absorption or reflection by PM (Nanos & Ilias, 2007; Chauhan, 2010).

The same pattern seen in the rate of photosynthesis was found in measurements of stomatal conductance in these two species (Fig. 3B), similarly to results noted by Vardaka et al. (1995) on the leaves of holm oak (*Quercus coccifera* L.). Similar results were also obtained by Nanos & Ilias (2007) when studying the effect of deposition of cement dust on the leaves of the European olive (*Olea europaea* L.). The smallest particles can clog stomata, thus reducing the concentration and accessibility of CO_2 independent of the effect on light phase of photosynthesis and transpiration (Naidoo & Chirkoot, 2004; Abdel-Rahman & Ibrahim, 2012; Burkhardt & Grantz, 2017). Several studies have also observed alteration in the stomatal features of plant leaves exposed to a cement dust environment. This modification is likely an adaption in response to pollution stress that helps to restrict the entry of dust pollutants into the plants (Amulya et al., 2015; Siqueira-Silva et al., 2016). Cement dust deposition also leads to modification of stomatal frequency, which is a mechanism to counter the blockage and allow proper functioning of gaseous exchange (El–Khatib et al., 2012). Furthermore, Gostin (2009) reported a decrease in size and increase in stomatal density in leaves at polluted sites, which makes leaves more resistant to the pollutants.

The results of chlorophyll *a* fluorescence obtained in this work showed no significant differences between dusted and control plants based on the parameter Fv/Fm, which described the maximum quantum efficiency of photosystem II for *P. serotina* (Fig. 3C). Completely different response was recorded in *P. padus*, where a reduction of this parameter was recorded in polluted seedlings. These data correspond to those obtained, for example, by Heerden et al. (2007) for *Zygophyllum prismatocarpum*. In their experiment, plants responded to increased exposure to limestone dust by reducing the Fv/Fm. This could be a reason why in our study *P. padus* seedlings, which accumulated more PM, reacted by decreasing this parameter. Vardaka et al. (1995) and Naidoo & Chirkoot (2004) also reported a decrease in the parameter Fv/Fm, together with an increase in the amount of dust deposited on the leaves of the tested species (*Quercus coccifera* L. and *Avicennia marina* (Forssk.) Vierh.). These authors concluded that due to the deposition of dust on the leaves, photosystem II efficiency was reduced via impaired electron transport, which to some extent explains the previously-reported decrease in the rate of photosynthesis. With regard to hypothesis 3, we partly confirmed that PM has a negative impact on the photosynthetic apparatus, because the effect have been observed only in *P. padus*.

In summary, we have shown that native *P. padus*, as it accumulates greater amounts of PM on its leaves, experiences lowering of efficiency of its photosynthetic apparatus, which in a broader sense reduces the condition of individuals. This species is not as protected from excessive accumulation as is *P. serotina*, due to their specific morphological and anatomical leaf traits. In Europe, *P. serotina* is classified as a highly invasive species causing significant transformation of ecosystems' species composition and difficulties in forest management (Vanhellemont et al., 2009; Pairon et al., 2010). *P. serotina* has been able to invade the European continent owing to its lower demand for nutrients and water compared with bird cherry (Halarewicz & Jackowski, 2011). Our results indicate that increased PM pollution may be an additional factor that affects the observed displacement of *P. padus* species by the invasive *P. serotina* in their common habitats (Halarewicz & Żołnierz, 2014). In addition to what is presented in this paper, there are several other reasons why *P. padus* is endangered compared to the closely-related invasive alien *P. serotina*. *P. padus* is less thermophilous than *P. serotina* and is much more demanding in terms of its humidity, soil nutrient content and soil moisture requirements (Ellenberg et al., 1992); thus, it is less tolerant to environmental changes, such as extremely high temperatures and the drying out of soils in summer. Moreover, the same folivorous species occur for both species, but *P. padus* experiences a much greater loss of leaf mass as a host than does *P. serotina* (Karolewski et al., 2013). A variety of mono- and polyphagous insect pests, especially the leaf beetle *Gonioctena quinquepunctata* F. (Mąderek et al., 2015), aphid *Rhopalosiphum padi* L. (Halarewicz & Gabryś, 2012) and *Yponomeuta evonymellus* L. (Karolewski et al., 2014), have more food sources due to the presence of alien species, so their increased populations may simultaneously pose a greater risk to *P. padus* (as it is their primary host). In addition, the invasive *P. serotina* is extremely resistant (*P. padus* is as well, but to a lesser extent) to various leaf mass reducing factors, such as insects (Halarewicz & Jackowski, 2011), artificial defoliation (Karolewski et al., 2010), mechanical damage of branches or even complete removal of the aboveground part of shrubs (Annighöfer et al., 2012). Considering the high resistance of *P. serotina* to biotic and mechanical properties, PM pollution appears unable to block/slow down the invasion of this shrub species. In light of these studies, it is advisable to examine whether PM-polluted *P. padus* is more vulnerable to folivorous insects. The next step in our research is therefore to investigate the effect that PM pollution on leaves of both species has on the feeding behaviour of the main folivorous insect species that utilise these two plants.

Conclusions

1. Significantly greater amounts of particulate matter (PM) were found on the leaves of the European native *Prunus padus* than were found on the leaves of the non-native *P. serotina*.
2. Both species accumulated PM of both categories (surface and in-wax PM) and from all three size fractions on their leaves in different amounts. Surface PM from the largest size fraction (10–100 μm) was the most common type of PM to accumulate on the two *Prunus* species.
3. The amount of in-wax PM was correlated with the amount of wax deposited on leaves. Leaves of *P. padus* had 25% more waxes than those of *P. serotina*.
4. The rate of photosynthesis and stomatal conductance, as well as maximum quantum efficiency of photosystem II, were negatively affected by both roadside and cement plant PM in *P. padus*, but in *P. serotina* no negative impact was found.
5. A strong negative correlation was found between the rate of photosynthesis and the total amount of PM accumulation, as well as between stomatal conductance and the amount of fine PM (0.2–2.5 μm) accumulated on *P. padus*, but this relationship was not found in *P. serotina*.
6. We conclude that non-native *P. serotina* is more tolerant to the stressful conditions of PM contamination of air than is the native *P. padus*, which may contribute to the success of the non-native species in its invasion of various habitats in Europe.

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