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Particulate Matter Accumulation and Leaf Traits of Ten Woody Species Growing with Different Air Pollution Conditions in Cheongju City, South Korea

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Abstract: Particulate matter (PM) is the most dangerous form of air pollution and is known to cause severe health problems to humans. Plants as biological filters can reduce PM in urban areas by accumulating PM on the surface and epicuticular wax of leaves. The present study determined the amount of PM (large PM (10–100 µm) and coarse PM (2.5–10 µm)) collected on 10 plant species from two sites (urban forest and roadside) of Cheongju City, South Korea. Selected leaf traits (chlorophyll a (Chl a), chlorophyll b (Chl b), total chlorophyll (TChl), carotenoid, relative leaf water content (RWC), specific leaf area (SLA), and pH of these plant species at the two sites were concurrently determined to find about the correlation between the leaf straits and PM accumulation on leaf. Study results showed that the amount of accumulated large PM (10–100 µm) and coarse PM (2.5–10 µm) were different depending on the plant species and the collection site. Plants from the roadside tended to have higher amounts of PM accumulation as compared to the same plant species from the urban forest. In addition, the amount of PM accumulated on the leaf surface was higher than that of the epicuticular wax. PM accumulation on the leaf surface was positively correlated with the amount of PM accumulated on the epicuticular wax. Among the 10 plant species selected, *P. strobus*, *P. densiflora*, *M. denudata*, and *S. vulgaris* were the most effective plant for PM accumulation, while *M. glyptostroboides* was the least effective plant ones. Chl a, Chl b, TChl, and carotenoid contents were higher in plants collected along the roadside than in those collected from the urban forest, whereas RWC was higher in plants from the urban forest. No distinct tendency was noted regarding the pH. Coarse PM (2.5–10 µm) was negatively correlated with leaf traits of plants along the roadside. The tolerance of plants to pollution might be due to an increase in chlorophyll content. Features of the leaf were also essential in increasing PM accumulation on the leaf surface.

Keywords: air quality; epicuticular wax; leaf trait; PM 2.5; PM 10

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1. Introduction

Air pollution is a global problem adversely affecting human health, animals, and vegetation. Natural events (volcano eruptions, forest fire) and anthropogenic activities, such as the use of polluting fuels for cooking, heating, and lighting are the main causes of air pollution [1]. Particulate matter (PM), consisting of complex solid and liquid particles suspended in the air, is one of the most dangerous forms of air pollution. In addition, human activities and natural events, the reaction of nitrogen oxide (from vehicle and industrial emission) with other air pollutants is one of the sources of PM [2]. According to their diameter, these PM pollutants are commonly classified into three categories, namely, PM 10 (diameter less or equal to 10 µm), PM 2.5 or fine particles (diameter less or equal to 2.5 µm), and ultrafine particles (diameter less than 0.1 µm [3]. PM 10 and PM 2.5, which include many small diameter particles, are very dangerous because these can easily penetrate the respiratory system and cause respiratory and cardiovascular morbidity, and even mortality [4]. Therefore, reducing air pollution has become an urgent task worldwide.

Nowadays, heavy traffic in urban areas has significantly increased the concentrations of pollutants in the atmosphere [5]. Plants play an important role in mitigating air pollution in urban areas [6–10]. The phytoremediation method involves the use of plants to improve the quality of soil, air, and water [11]. Using plants to improve air quality was reported to be the best strategic solution that uses plants as natural biological filters or sink to mitigate PM in the air [12]. PM can accumulate on the leaf surface or epicuticular wax [13]. However, the amount of PM captured by plant leaves differs depending on the plant species as well as environmental conditions [14–17]. Firstly, the amount of PM accumulated on the leaf depends on the leaf characteristics [18]. The micromorphology of leaves, such as roughness, stomata density, leaf shape, and leaf growth expansion can act as combination factors to increase the amount of PM accumulated on leaves [19,20]. For example, plants with rough leaf surfaces can capture more PM than those with smoother leaf surfaces [21,22]. Plants with leaf hair show more effective PM accumulation than those without leaf hair. In addition, the amount of PM accumulation on leaf depends on other factors, such as the levels of pollution and the environmental conditions, namely, wind, rainfall, and temperature [23–25]. In a highly polluted environment, the amount of PM accumulated by a plant would be higher than that of a plant in a less polluted environment [26]. Moreover, PM can be washed off from leaf because of strong wind and rainfall. However, the wash-off rate of different plants would differ [27].

Conversely, air pollution also can impact plants. The morphological, biochemical, physiological, and genetic traits of plants can be affected by PM in the atmosphere [28]. PM on leaf surface can reduce plant growth, flowering, leaf area, and the number of leaves. Rai [29] showed that PM can also destroy the structure of epicuticular wax. Modification of the biochemical and physiological traits of plants is due to changes of the leaf traits, such as chlorophyll content, carotenoid content, leaf extract pH, relative leaf water content (RWC), and specific leaf area (SLA) [24,30]. A high amount of PM on leaves can block stomatal pores, reducing stomatal conductance, and can also reduce light absorption, impacting the photosynthesis rate [13,31]. However, changes of leaf traits are different for various plant species. Leaf traits can be increased or decreased at a polluted site depending on the response of a plant species to air pollution and environmental conditions [13,23,31–33].

The purpose of this study was to determine the ability of PM to accumulate on leaves of plant species under different air pollution levels areas. Understanding the effect of air pollution on leaf traits is as important as determining the capacity of the plant species to reduce PM. Therefore, determining the correlation between PM accumulation on leaves and leaf traits has a prominent role in choosing which plant species to plant to reduce air pollution in urban areas. In this study, we determined PM accumulation capacities of leaf surface and epicuticular wax layer of 10 common plant species at two sites (roadside and urban forest) in Cheongju City, South Korea, with different levels of pollution. In addition, responses of these plants to air pollution were determined by measuring leaf traits, such as chlorophyll a (Chl a), chlorophyll b (Chl b), total chlorophyll (TChl), carotenoid, relative leaf water content (RWC), specific leaf area (SLA), and pH. We hypothesized that (1) PM accumulation on plants along the roadside was higher than plants in the urban forest because of the difference in air pollution between these two sites, (2) PM has a negative effect on leaf traits.

2. Materials and Methods

2.1. Study Area

Cheongju is the biggest city in Chungcheongbuk-do province in South Korea. Plant samples were collected at two sites of Chungcheongbuk-do, Cheongju City, South Korea (36°38′0″ N, 127°29′0″ E). One site was Chungcheongbuk-do Forest Environment Research Institute (urban forest) where 1593 plant species were planted on 25 ha. This site is located about 16 km from the city center and is not affected by traffic or industrial pollution. The other study site was a crossroad in the city center near the Cheongju Express Bus Terminal (roadside). This area is affected by the high traffic this city and the traffic of the Express Bus Terminal (Figure 1).

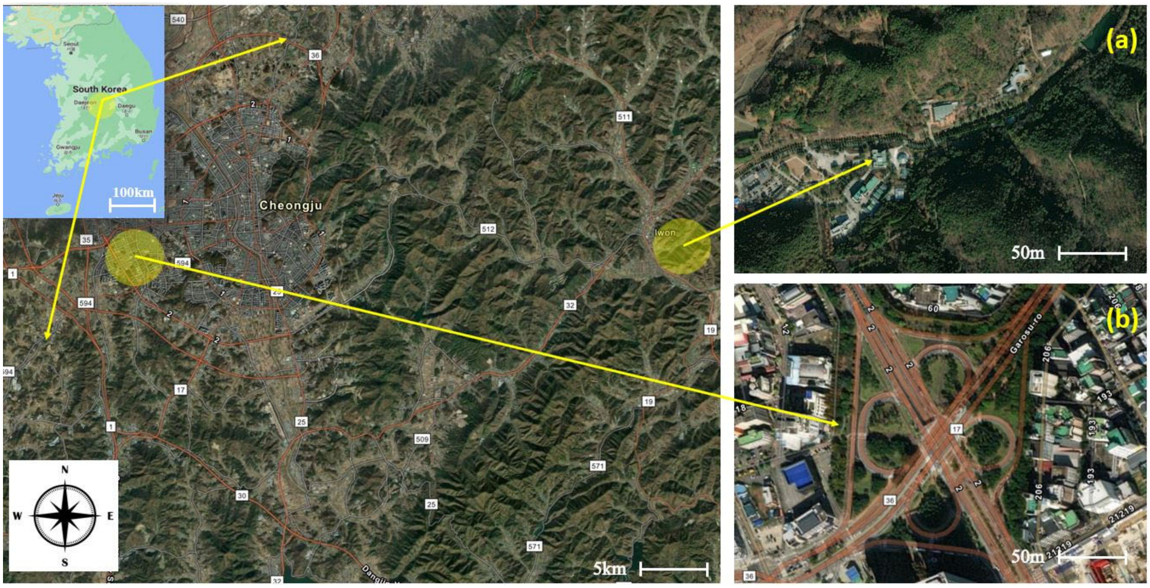

Figure 1. The location map of the two sampling sites. (**a**) Urban forest (Chungcheongbuk-do Forest Environment Research Institute) and (**b**) Roadside (Crossroad near Chenogju Express Bus Terminal).

2.2. Sample Collection

Ten common plant species in South Korea were collected for this study (Table 1). Leaf samples were collected on the same day in October 2020. Leaves in good condition (i.e., not affected by disease or pests) were selected. For each plant species, the leaves of five different trees were collected at 1.0–1.5 m above the ground (n = 5 replication). Leaf samples were cut to make up a leaf area of about 300 to 400 cm^2 and were placed into a paper bag. Subsequently, all samples were immediately conveyed to the laboratory for analysis. All samples were collected after 10 days of no rain.

Table 1. Scientific name and leaf shape of 10 plant species sampled in this study.

Plant Species	Family	Foliage	Leaf Shape	Habit
Rhododendron yedoense f. *poukhanense* (H.Lév.) M. Sugim. ex T.Yamaz.	Ericaceae	Deciduous broad-leaved	Oval	Shrub
Zelkova serrata (Thunb.) Makino	Ulmaceae	Deciduous broad-leaved	Ovate	Tree
Acer palmatum Thunb.	Aceraceae	Deciduous broad-leaved	Palmate	Tree
Magnolia denudata Desr.	Magnoliaceae	Deciduous broad-leaved	Obovate	Tree
Syringa vulgaris L.	Oleaceae	Deciduous broad-leaved	Cordate	Shrub
Metasequoia glyptostroboides Hu & W.C. Cheng	Cupressaceae	Deciduous conifer	Opposite	Tree
Juniperus chinensis L.	Cupressaceae	Evergreen needle-leaved	Scale	Tree
Taxus cuspidata Siebold & Zucc.	Taxaceae	Evergreen needle-leaved	Lanceolate	Tree
Pinus densiflora Siebold & Zucc.	Pinaceae	Evergreen needle-leaved	Needle-like	Tree
Pinus strobus L.	Pinaceae	Evergreen needle-leaved	Needle-like	Tree

2.3. Analysis of Accumulation of Surface PM, In-Wax PM, and Epicuticular Wax on Leaves

The wash-off method was used to determine the amount of leaf surface PM (sPM) and in-wax PM (wPM) [34]. For each species, leaves with an area of about 300 cm^2 were rinsed with distilled water to collect PM on the leaf surface (sPM), then with chloroform to collect PM on the epicuticular wax (wPM). The leaves were placed in individual glass beakers and washed with 250 mL distilled water for 60 s. These beakers were placed in ultrasonic cleaners (WUC-A22H, Daihan Scientific, Wonju, Korea) for 6 min to ensure that particles were separated from the leaf surface. The solution was passed through a metal sieve with a diameter of 100 μm mesh to remove particles with size over 100 μm. Subsequently, a glass filter funnel set connected to a vacuum pump (2546c-10, Welch, Concord, MA, USA) was used to filter the solution with pre-weighed type 91 and type 42 filter papers (Whatman, UK) with pore size 10 μm and 2.5 μm, respectively. After passing through type 91 filter, the collected solution was filtered with type 42 filter immediately. These collected PM in the two types of filter papers were divided into two sizes: large PM (10–100 μm) and coarse PM (2.5–10 μm), respectively. These paper filters were weighted before and after filtration with the same method. All samples were then placed in an auto desiccator cabinet (SLDeBG1K, SciLab, Seoul, Korea) for 48 h to remove humidity and were later weighted on a microbalance (EX125D, Ohaus, Parsippany, NJ, USA). The same method was used to determine the amount of PM accumulation on wax of plant. However, the chloroform was used instead of water. Leaf samples were then washed with 100 mL chloroform for 60 s and placed in an ultrasonic cleaner for 2 min. After that, the two type 91 and type 42 filter papers were used to collect PM. Collected PM was divided into two types: large PM (10–100 μm) and coarse PM (2.5–10 μm). The amount of epicuticular wax that dissolved in chloroform was measured in a pre-weighed beaker at the filtration end and was determined after chloroform evaporation. Before washing with chloroform, the total leaf area of the leaf samples was determined with an area meter (LI-3100C, LI-COR, Lincoln, NE, USA). In this study, the amounts of PM and epicuticular wax were expressed in microgram per square centimeter. PM of each leaf sample was photographed using a mini-SEM (Scanning Electron Microscope, SNE-4500M Plus A, SEC, Suwon, Korea) according to Noh [35].

2.4. Leaves Traits

2.4.1. Leaf Extract pH (pH)

The leaf extract pH was measured based on the method described by [30]. Briefly, 1.0 g of fresh leaf sample and 10 mL of distilled water were homogenized and centrifuged at 2700 rpm for 3 min. Afterward, a pH meter (HI 8424, Hana Instruments, Woonsocket, RI, USA) was used to measure the pH of the collected homogenate.

2.4.2. Relative Leaf Water Content (RWC)

After sampling, leaf samples were immediately washed to remove particles from the leaf surface. For each species, leaves were weighed to determine their fresh weight. These leaves were weighed again after soaking in distilled water at 4 °C in a dark place for 24 h to obtain their turgid weight. Finally, leaf samples were dried in an oven at 80 °C for 24 h and weighted to obtain their dry weight. Following the method of [36], the RWC was determined using Equation (1):

$$\text{RWC (\%)} = [(\text{FW} - \text{DW})/(\text{TW} - \text{DW})] \times 100 \tag{1}$$

where, FW = fresh weight, TW = turgid weight, and DW = dry weight.

2.4.3. Chlorophyll and Carotenoid Contents

Using the Lichtenthaler [37] method, 0.05 g of each leaf sample was crushed in liquid nitrogen with a mortar and a pestle. After adding 10 mL of 100% acetone into the crushed leaf sample, the liquid was collected and centrifuged (Cef-6, Daihan Scientific, Korea) at 2700 rpm for 10 min. Then, 10 mL of the supernatant was used to determine the absorbance at 470 nm, 616.6 nm, and 644.8 nm using a spectrophotometer (UV-1800, Simazuda, Japan). Chlorophyll a (Chl a), chlorophyll b (Chl b), total chlorophyll (TChl), and carotenoid contents were calculated using Equation (2):

$$\begin{aligned} \text{Chlorophyll a} &= (11.24 \times A_{616.6}) - (2.04 \times A_{644.8}) \\ \text{Chlorophyll b} &= (20.13 \times A\ 6_{44.8}) - (4.19 \times A_{616.6}) \\ \text{Total chlorophyll} &= (7.05 \times A_{616.6}) + (18.09 \times A_{644.8}) \end{aligned} \tag{2}$$

$$\text{Carotenoids} = (1000 \times A_{470}) - (1.90 \times \text{Chl a} - 63.14 \times \text{Chl b})/214$$

where $A_{616.6}$, $A_{644.8}$, and A_{470} are absorbance values at corresponding wavelengths.

2.4.4. Specific Leaf Area (SLA)

According to Chaturvedi et al. [38], the SLA present the ratio of leaf area to dry weight of leaf. An leaf area meter (LI-3100C, LI-COR Biosciences, Lincoln, NE, USA) was used to determine the leaf area of each sample for each plant species. Then, the leaf samples were dried in an oven at 80 °C for 24 h and were weighed to record the dry weight. After that, SLA was determined using Equation (3):

$$\text{SLA (cm}^{-2}\cdot\text{g}^{-1}) = \text{Leaf area/dry weight} \quad (3)$$

3. Statistical Analysis

All data were analyzed using SAS software 9.4 version (SAS Institute, Cary, NC, USA) for two-way analysis of variance (ANOVA) with Duncan's multiple range test (DMRT). The significance level was set at 5%. Two-way ANOVA was also used to identify the influence of two different factors (sites and plant species) on leaf PM accumulation and biological characteristics of plants. Pearson's correlation analysis was used to identify the relationship between the amount of PM accumulation on leaf and the plant's biochemical characteristics. The presented data are given as means with standard error (±SE).

4. Results and Discussion

4.1. PM Accumulation in Plant Species

In this study, we found that the amount of PM accumulation on the leaf of plant species was different depending on the plant species and the leaf sample collection site. In the urban forest area, the amount of large PM (10–100 μm) accumulation on leaf surface ranged from 5.03 to 43.12 μg·cm^{-2}. The highest accumulation of large PM (10–100 μm) on leaf surface was on *P. strobus*, followed by *P. densiflora*, *M. denudata*, and *S. vulgaris* with an amount of large PM (10–100 μm) accumulation of 36.52, 22.26, and 17.41 μg·cm^{-2} respectively, whereas *M. glyptostroboides* showed the lowest accumulation of large PM (10–100 μm) on leaf surface. The amount of coarse PM (2.5–10 μm) accumulation on leaf surface ranged from 1.94 to 9.61 μg·cm^{-2}. *A. palmatum* showed the highest coarse PM (2.5–10 μm) accumulation while *T. cuspidata* showed the lowest coarse PM (2.5–10 μm) accumulation. The amount of large PM (10–100 μm) on leaf surface accumulation was higher than that of coarse PM (2.5–10 μm) for all 10 plant species (from 1.30 to 6.92 times).

The amount of large PM (10–100 μm) accumulation in wax of the selected plant species ranged from 1.88 to 23.01 μg·cm^{-2}. Plant species with the highest and lowest accumulation of large PM (10–100 μm) in wax layer were *P. strobus* and *M. glyptostroboides*, respectively. *P. strobus* also showed the highest accumulation of large PM (10–100 μm) while *A. palmatum* showed the lowest value. Furthermore, among the 10 plant species, the amount of coarse PM (2.5–10 μm) in wax layer accumulation on *S. vulgaris*, *M. denudata*, *R. yedoense*, *J. chinensis*, and *M. glyptostroboides* was higher than that of large PM (10–100 μm) accumulation. The total PM accumulation of the 10 plant species at the urban forest ranged from 11.84 to 89.33 μg·cm^{-2}, with the highest and lowest PM accumulation on *P. trobus* and *M. glyptostroboides*, respectively (Table 2).

Table 2. Amount of particulate matter on leaf surface and epicuticular wax of 10 plant species from two sites.

		sPM (Mean ± SE)		wPM (Mean ± SE)		Total PM (μg·cm^{-2})	Epicuticular Wax (μg·cm^{-2})
		10–100 (μg·cm^{-2})	**2.5–10 (μg·cm^{-2})**	**10–100 (μg·cm^{-2})**	**2.5–10 (μg·cm^{-2})**		
S. vulgaris	Urban forest	17.41 ± 13.05	4.87 ± 1.21	8.47 ± 4.48	9.27 ± 4.76	40.01 ± 13.63	161.08 ± 39.73
	Roadside	23.20 ± 6.52	26.41 ± 5.57	6.04 ± 2.30	13.60 ± 8.06	69.25 ± 11.50	154.53 ± 30.30
M. denudata	Urban forest	22.26 ± 3.86	9.14 ± 2.02	2.94 ± 1.26	3.27 ± 1.96	37.61 ± 4.47	20.59 ± 4.14
	Roadside	96.10 ± 37.37	8.07 ± 2.72	2.67 ± 1.67	2.60 ± 1.77	109.43 ± 35.49	38.92 ± 34.69
A. palmatum	Urban forest	12.45 ± 2.19	9.61 ± 3.07	3.73 ± 1.15	0.83 ± 0.26	26.62 ± 2.82	18.13 ± 2.25
	Roadside	23.23 ± 10.89	4.34 ± 1.15	14.67 ± 6.37	9.82 ± 4.35	52.06 ± 16.10	34.00 ± 9.02
Z. serrata	Urban forest	11.16 ± 3.24	6.82 ± 1.00	5.91 ± 2.62	3.08 ± 1.48	26.97 ± 4.33	60.02 ± 15.64
	Roadside	27.16 ± 5.76	5.85 ± 0.68	2.88 ± 1.14	4.56 ± 2.52	40.44 ± 7.53	30.18 ± 7.36
R.yedoense	Urban forest	8.66 ± 5.00	3.94 ± 2.52	2.37 ± 0.94	3.30 ± 1.72	18.27 ± 8.04	58.85 ± 4.32
	Roadside	21.56 ± 14.81	7.36 ± 5.22	4.60 ± 3.15	8.89 ± 6.93	42.40 ± 17.63	90.52 ± 25.80
J. chinensis	Urban forest	5.41 ± 2.00	3.61 ± 1.58	3.23 ± 2.17	4.57 ± 1.58	16.82 ± 4.70	368.71 ± 69.46
	Roadside	22.57 ± 8.33	23.37 ± 9.45	7.13 ± 4.40	4.60 ± 3.02	57.68 ± 9.29	578.64 ± 34.73
M. glyptostroboides	Urban forest	5.03 ± 0.76	2.19 ± 2.14	1.88 ± 0.90	2.74 ± 0.73	11.84 ± 2.87	223.92 ± 60.21
	Roadside	14.36 ± 4.33	10.89 ± 1.49	2.26 ± 1.54	3.77 ± 3.00	31.28 ± 7.51	166.72 ± 40.32
T. cuspidata	Urban forest	10.59 ± 3.26	1.94 ± 0.51	4.10 ± 0.99	2.47 ± 0.79	19.10 ± 2.29	77.65 ± 4.54
	Roadside	18.33 ± 2.99	21.85 ± 4.92	6.30 ± 4.22	0.77 ± 0.63	47.25 ± 5.09	102.59 ± 35.07
P. densiflora	Urban forest	36.52 ± 10.31	5.28 ± 1.95	12.64 ± 1.87	7.08 ± 2.37	61.51 ± 13.22	723.04 ± 59.87
	Roadside	60.78 ± 11.63	12.66 ± 7.13	17.03 ± 8.51	8.15 ± 4.55	98.62 ± 20.29	883.46 ± 125.51
P. strobus	Urban forest	43.12 ± 16.64	6.82 ± 3.85	23.01 ± 5.10	16.37 ± 6.93	89.33 ± 28.13	1127.78 ± 242.82
	Roadside	73.75 ± 25.22	29.61 ± 9.51	49.97 ± 37.25	12.40 ± 5.56	165.73 ± 35.91	1055.31 ± 248.51

ANOVA †	**Df**	**Error**	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***
Species	9	80	23.19	<0.0001	10.2	<0.0001	13.11	<0.0001	10.77	<0.0001	40.33	<0.0001	176.36	<0.0001
Site	1	80	65.79	<0.0001	124.14	<0.0001	6.3	0.0141	4.36	0.04	129.19	<0.0001	2.77	0.1002
Species × site	9	80	6.14	<0.0001	15.17	<0.0001	2.39	0.0186	2.37	0.0198	4.37	0.0001	2.57	0.0119

† ANOVA was used to analyze the significance of PM difference between different sites and plant species. 10–100 μm: particulate matter accumulated on leaf surface with diameter ranging from 10 to 100 μm; 2.5–10 μm: particulate matter accumulated on leaf surface with diameter ranging from 2.5 to 10 μm; wPM: particulate matter accumulated on the epicuticular wax; sPM: particulate matter accumulated on the leaf surface. Total PM: sPM + wPM.

For samples collected along the roadside, the amount of large PM (10–100 µm) accumulation on leaf surface ranged from 14.36 to 96.10 µg·cm^{-2}. *M. denudata* showed the highest accumulation of large PM (10–100 µm) followed by *P. strobus* and *P. densiflora*. The plant species that showed the lowest accumulation of large PM (10–100 µm) was *M. glyptostroboides*. The amount of coarse PM (2.5–10 µm) accumulation ranged from 4.34 to 29.61 µg·cm^{-2}, with the highest coarse PM (2.5–10 µm) accumulation on *P. strobus* and the lowest accumulation on *A. palmatum*. We also found that the amount of large PM (10–100 µm) accumulation on the leaf surface of *S. vulgaris*, *J. chinensis*, and *T. cuspidata* was lower than the amount of coarse PM (2.5–10 µm) accumulation. On the epicuticular wax, the amount of large PM (10–100 µm) accumulation ranged from 2.26 to 49.97 µg·cm^{-2}, while the highest PM accumulation plant species was *P. strobus* followed by *P. densiflora* and *A. palmatum*. The lowest PM accumulation plant species was *M. glyptostroboides*. Conversely, the amount of coarse PM (2.5–10 µm) accumulation ranged from 0.77 to 13.60 µg·cm^{-2} with the highest and lowest PM accumulation plant species being *S. vulgaris* and *T. cuspidata*, respectively. The total PM accumulation of the 10 plants at the urban forest ranged from 31.28 to 165.73 µg·cm^{-2}. *P. strobus* and *M. glyptostroboides* were still the highest and lowest total PM accumulation plant species (Figure 2).

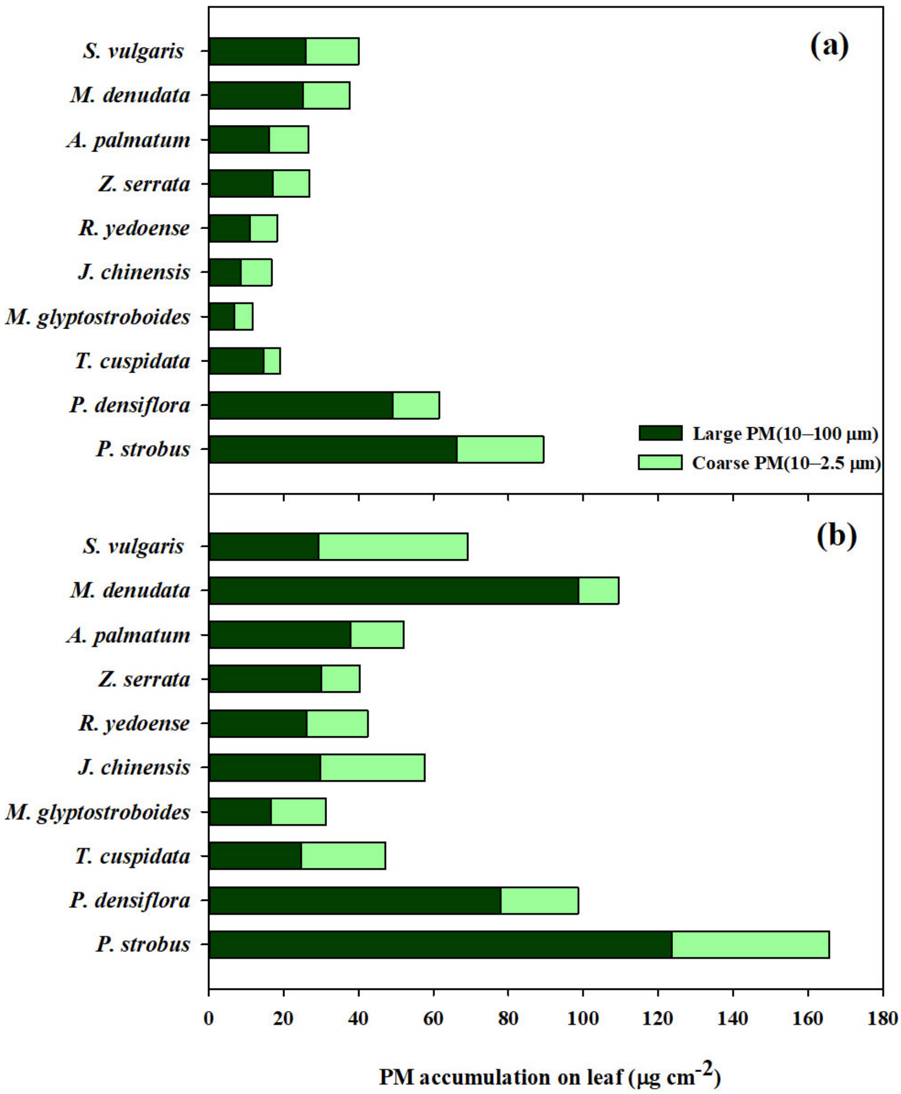

Figure 2. Particulate matter accumulation on leaves of 10 plant species in (**a**) urban forest and (**b**) roadside.

When comparing the two sites, the amount of total PM accumulation on the leaf of the selected plants species along the roadside was higher than the amount of total PM accumulation of the same plant species at the urban forest (from 1.5 to 3.4 times). Plant species with the highest difference in the ratio of PM accumulation between the roadside and the urban forest were *J. chinensis* and *M. denudata* (about 3.4 and 2.9 times, respectively). Among the 10 plant species, *P. strobus*, *M. denudata*, *P. densiflora*, and *S. vulgaris* had the highest PM accumulation while *M. glyptostroboides* had the lowest PM accumulation.

Additionally, the amount of wax on the leaves of the selected plant species differed with respect to the different sites and the different plant species. At both sites, we found that *P. strobus* showed the highest amount of wax followed by *P. densiflora*, *J. chinensis*, and *M. glyptostroboides* while the plant species with the lowest amount of wax were *A. palmatum* and *Z. serrata* at the urban forest and the roadside, respectively. The amount of wax of needle leaves was higher than that for broad leaves at both sites, except for *T. cuspidata*. Among the five species with broad leaves, the wax of *S. vulgaris*, at the urban forest and roadside (161.08 and 154.53 $\mu g \cdot cm^{-2}$, respectively), was highest.

The amount of PM accumulation on the leaves of plants differed with respect to the plant species. Many studies showed that the difference in the structure on the leaf between plant species was one of the main reasons that led to the difference in the effective accumulation of PM on the leaf [5,23,26,39]. A similar result was found in our study. Additionally, we found that the amount of total PM accumulated on plants growing along the roadside was higher than that of plants growing in the urban forest. The different concentrations of air pollution between the roadside and the urban forest could be a reason that influenced the amount of PM accumulation on the leaves of the same plant species between the two sites. Several studies have shown that the capacity PM accumulation of plants depends on PM level in the air and that higher PM level in the air could increase the amount of PM accumulated on the leaf [26,40]. However, we also found that the average total PM accumulated on plant species with needle leaves was higher than that of plant species with broad leaves at both sites. He et al. [41] and Leonard et al. [42] have also indicated that needle leaves accumulated PM more than broad leaves. Plants with needle leaves are less affected by wind that removes PM from the leaf surface due to their smaller leaf area and higher concentration of leaves than plants with broad leaves [19,42,43]. Additionally, the high roughness and high stomatal density of needle leaves enhance the adsorption of PM on leaf surface as compared to broad leaves [44]. In the urban forest, *P. strobus* had the highest PM accumulated on both the leaf surface and epicuticular wax, while *M. glyptostroboides* showed lowest PM accumulation on the leaf surface. Deep grooves observed on the leaf surface of *P. strobus* (Figure 3s,t) could have helped PM accumulation. Furthermore, *P. strobus* had the highest amount of wax that also significantly contributed to increasing PM accumulation. Many studies showed that the grooves on leaves help contain large particles, causing an increase in the PM mass accumulated on the leaf [45]. The amount of epicuticular wax of *P. strobus* has an essential role in increasing the capacity of PM accumulation. The PM accumulated on the epicuticular wax of *P. strobus* makes up a large part of the total PM accumulated. The critical role of quantity and structure of epicuticular wax in PM accumulation has been reported previously [32,46]. Additionally, *M. glyptostroboides* is a plant with needle leaves and has smooth and thin leaves. This might be the reason why PM accumulated on leaves of *M. glyptostroboides* could be easily removed by wind or washed off by rain [47]. Along the roadside, *M. denudata* was the most excellent plant species for PM accumulation on the leaf surface. Based on SEM images, we found that *M. denudata* had large PM accumulation on the leaf surface (Figure 3c,d). Kwak et al. [39] have shown that *M. denudata* has high stomatal density and leaf hairs on both the adaxial and the abaxial leaf surfaces. In this study, we did not assess the leaf hairs of plant, but we guessed that the presence of leaf hair could help increase the amount of PM accumulation. This might be the reason for the significant increase in the amount of PM accumulated on leaves of *M. denudata*. Leaf hairs can increase the area of the leaf to accumulate more PM and impede the removal of PM from the leaf surface by wind as suggested by Leonard et al. [42]. Additionally, we found grooves on the leaf surface of *S. vulgaris* and *T. cuspidata* and large size particles on the leaf surface of these plant species. In addition, we found that the amount of wax had an important role in increasing amount of PM accumulation. In the present study, we found that the feature of leaf was a key factor impacting the capacity for PM accumulation.

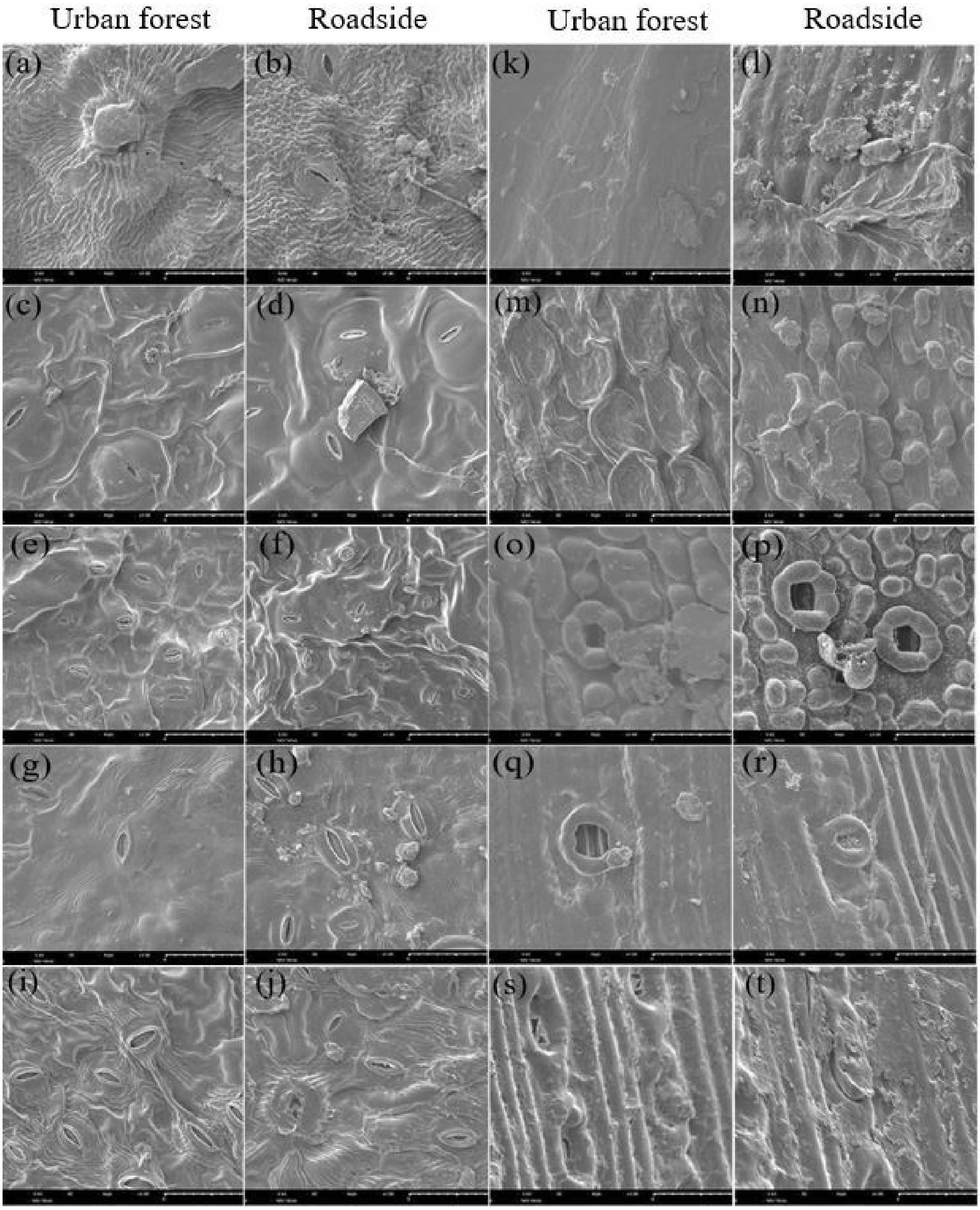

Figure 3. SEM (Scanning Electron Microscope) micrographs of 10 plant species (**a**,**b**) *S. vulgaris*, (**c**,**d**) *M. denudata*, (**e**,**f**) *A. palmatum*, (**g**,**h**) *Z. serrata*, (**i**,**j**) *R. yedoense*, (**k**,**l**) *J. chinensis*, (**m**,**n**) *M. glyptosboises*, (**o**,**p**) *T. cuspidata*, (**q**,**r**) *P. densiflora*, and (**s**,**t**) *P. trobus* in urban forest and roadside. The scale bar is 50 μm.

4.2. Leaf Traits

Leaf traits are factors that affect the amount of PM accumulation in plants. However, PM can also impact leaf traits and cause decreased growth in plants. In this study, Chl a, Chl b, TChl content, and carotenoid contents were higher of plants from roadside site than plants growing in the urban forest for all plant species excepted for *S. vulgaris* and *P. strobus*. Relative leaf water content (RWC) of plants growing along the roadside were higher than those of plants growing in the urban forest. However, the RWC of *M. denudata* did not follow this trend. The pH of plants with needle leaves growing along the roadside tended to be higher as compared to the pH of broad leaves as (Table 3). Additionally, the amount of wax of the 10 plant species was different between species and sampling sites. Among the 10 plant species, the quantity of wax of *S. vulgaris*, *Z. serrata*, *M. glyptostroboides*, and *P. strobus* were higher for plants from the urban forest than that of plants from the roadside (Table 2).

In this study, aside from a positive correlation between large PM (10–100 μm) and epicuticular wax quantity and a negative correlation between PM 10 and SLA at both study sites, we did not find any other correlations between large PM (10–100 μm) and the other leaf traits. The large PM (10–100 μm) accumulation on a leaf can lead to increasing shape area, caused reducing light absorption of the leaf [24,29]. While coarse PM (2.5–10 μm) had a positive correlation with epicuticular wax quantity at both study sites, it had negative correlations with all leaf traits (Chl a, Chl b, TChl, carotenoid, and SLA) of plants from the roadside. Thus, some leaf traits, notably SLA and epicuticular wax, are correlated with the PM accumulation capability in these plants (Table 4). Thus, the feature of leaf structures is one of the determining factors influencing the capability of PM accumulation on plants (Table 4). Conversely, PM could also affect leaves by changing leaf traits, such as leaf pigment (chlorophyll content and carotenoid), RWC, SLA, and leaf extract pH [20,22]. Under the impact of PM, plants have to respond to air pollution to stay alive. Some plants are sensitive to air pollution while others are tolerant. Plant species have different responses to air pollution [30]. The effect of PM on leaf traits, such as SLA, RWC, and chlorophyll content has been reported previously [38].

Chlorophyll content is a crucial factor because a reduction of chlorophyll content causes decreased photosynthesis, which is a critical factor for plant growth. Environmental conditions and air pollution can affect chlorophyll content. Depending on the tolerance of plants and chemical components of pollutants, the influence of pollution on a plant's chlorophyll content is different [48]. As a result, PM accumulation on the leaf surface could reduce the chlorophyll content in leaves [23,26,48,49]. A pollutant covering the leaf surface can impact light absorption by the plant or block the stomatal pores and reduce photosynthetic gas exchange of the plant, leading to reduced chlorophyll content [24]. Moreover, chlorophyll content is also strongly dependent on pH. Pollution on leaves can reduce their extract pH, leading to decreased chlorophyll content [30]. In this study, chlorophyll and carotenoid contents were increased in all plant species growing along the roadside except for *S. vulgaris* and *P. strobus*. The same result has been reported previously [50]. We found that coarse PM (2.5–10 μm) was weakly correlated with chlorophyll and carotenoid contents of plants at the roadside. Such weak correlations might not affect plants too much. The increase of chlorophyll content in plants growing along the roadside might have increased the tolerance of plants to pollution. The plant species showing the highest increase of chlorophyll content was *R. yedoense*. The chlorophyll content of *R. yedoense* was increased 2-fold, whereas the chlorophyll content of *S. vulgaris* was reduced by half compared to that of the same plant species growing in the urban forest. These results suggest that the decreasing chlorophyll content of *S. vulgaris* might be due to the large amount of coarse PM (2.5–10 μm) accumulation on leaves of this plant (Figure 3). The PM accumulation on leaf surface can caused decreasing light absorption. Coarse PM (2.5–10 μm) with small size can easily penetrate and block the stomatal pores, thus influencing chloroplast and resulting in reduced chlorophyll content [51].

Table 3. Average leaf trait parameter values of 10 plant species at two different sites.

		SLA ($cm^{-2} \cdot g^{-1}$)	Chl a ($mg \cdot g^{-1}$ FW)	Chl b ($mg \cdot g^{-1}$ FW)	TChl ($mg \cdot g^{-1}$ FW)	Carotenoid ($mg \cdot g^{-1}$ FW)	pH	RWC (%)
S. vulgaris	Urban forest	90.39 ± 24.02	0.105 ± 0.03	0.043 ± 0.01	0.148 ± 0.04	10.17 ± 2.79	5.72 ± 0.16	73.99 ± 3.16
	Roadside	97.16 ± 37.64	0.047 ± 0.01	0.021 ± 0.00	0.068 ± 0.02	4.37 ± 1.18	5.39 ± 0.20	87.05 ± 2.81
M. denudata	Urban forest	203.34 ± 92.08	0.057 ± 0.01	0.027 ± 0.00	0.083 ± 0.02	5.58 ± 1.27	6.09 ± 0.09	78.79 ± 2.30
	Roadside	183.27 ± 51.58	0.098 ± 0.03	0.041 ± 0.01	0.139 ± 0.04	9.13 ± 2.09	6.02 ± 0.14	77.90 ± 7.48
A. palmatum	Urban forest	289.523 ± 119.18	0.068 ± 0.02	0.032 ± 0.01	0.101 ± 0.02	6.75 ± 1.70	4.50 ± 0.42	92.79 ± 3.52
	Roadside	246.55 ± 77.05	0.118 ± 0.04	0.052 ± 0.02	0.170 ± 0.06	11.09 ± 3.72	5.65 ± 0.15	97.75 ± 3.69
Z. serrata	Urban forest	300.10 ± 111.24	0.072 ± 0.03	0.035 ± 0.01	0.107 ± 0.04	7.61 ± 3.23	5.92 ± 0.09	67.65 ± 5.72
	Roadside	76.74 ± 32.43	0.089 ± 0.02	0.036 ± 0.01	0.125 ± 0.02	8.00 ± 1.69	5.71 ± 0.11	69.91 ± 3.61
R. yedoense	Urban forest	115.93 ± 44.30	0.061 ± 0.02	0.029 ± 0.01	0.090 ± 0.02	5.73 ± 1.77	5.59 ± 0.33	72.97 ± 3.81
	Roadside	198.35 ± 77.19	0.146 ± 0.01	0.060 ± 0.00	0.206 ± 0.02	13.84 ± 0.94	5.58 ± 0.12	86.72 ± 3.78
J. chinensis	Urban forest	54.12 ± 22.96	0.050 ± 0.02	0.020 ± 0.01	0.070 ± 0.03	4.10 ± 1.78	5.15 ± 0.18	71.07 ± 1.93
	Roadside	51.49 ± 18.18	0.057 ± 0.01	0.023 ± 0.00	0.080 ± 0.02	4.81 ± 0.92	5.48 ± 0.25	83.52 ± 8.63
M. glyptostroboides	Urban forest	226.94 ± 105.01	0.122 ± 0.02	0.049 ± 0.01	0.171 ± 0.03	11.19 ± 2.09	5.63 ± 0.05	72.71 ± 6.69
	Roadside	297.92 ± 135.54	0.149 ± 0.04	0.058 ± 0.02	0.207 ± 0.06	13.30 ± 3.77	5.45 ± 0.12	73.39 ± 9.57
T. cuspidata	Urban forest	110.51 ± 32.16	0.063 ± 0.02	0.026 ± 0.01	0.089 ± 0.03	5.45 ± 1.80	5.25 ± 0.10	80.24 ± 4.78
	Roadside	92.72 ± 27.21	0.063 ± 0.02	0.027 ± 0.01	0.090 ± 0.03	5.70 ± 1.63	5.34 ± 0.16	84.85 ± 2.45
P. densiflora	Urban forest	33.24 ± 10.61	0.059 ± 0.01	0.026 ± 0.00	0.085 ± 0.02	5.08 ± 1.05	4.80 ± 0.04	81.91 ± 2.91
	Roadside	39.45 ± 7.25	0.082 ± 0.01	0.036 ± 0.01	0.188 ± 0.02	7.31 ± 0.94	5.09 ± 0.13	87.68 ± 2.25
P. strobus	Urban forest	16.01 ± 8.45	0.100 ± 0.01	0.044 ± 0.01	0.144 ± 0.02	8.27 ± 1.13	4.99 ± 0.04	72.70 ± 6.75
	Roadside	8.58 ± 4.79	0.100 ± 0.02	0.042 ± 0.01	0.142 ± 0.03	9.29 ± 1.90	5.11 ± 0.15	75.52 ± 1.56

ANOVA †	**DF**	**Error**	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***	**F**	***p***
Species	9	80	19.18	<0.0001	11.46	<0.0001	11.28	<0.0001	11.77	<0.0001	11.85	<0.0001	40.09	<0.0001	21.97	<0.0001
Site	1	80	65.79	<0.0001	124.14	<0.0001	6.3	0.0141	4.36	0.04	129.19	<0.0001	11.48	0.0011	2.77	0.1002
Species × site	9	80	6.14	<0.0001	15.17	<0.0001	2.39	0.0186	2.37	0.0198	4.37	0.0001	14.28	<.0001	2.57	0.0119

† ANOVA was used to analyze the significance of difference in leaf traits between different sites and plant species. SLA: specific leaf area; Chl a: chlorophyll a concentration; Chl b: chlorophyll b concentration; TChl: Total chlorophyll concentration; Carotenoid: carotenoid concentration; pH: leaf extract pH; RWC: relative leaf water content.

Table 4. Pearson correlation analysis of PM accumulation and leaf traits of 10 plant species.

		SLA	Chl a	Chl b	TChl	Carotenoid	pH	RWC	Epicuticular Wax
Urban forest	Total large PM	−0.442 **	0.078	0.067	0.093	0.005	−0.260	0.071	0.810 ***
	Total coarse PM	−0.258	0.166	0.161	0.205	0.152	−0.149	0.035	0.545 ***
Roadside	Total large PM	−0.317 *	0.010	0.107	0.057	0.041	−0.086	−0.137	0.564 ***
	Total coarse PM	−0.434 **	−0.391 *	−0.314 *	−0.384 **	−0.386 **	−0.510 ***	0.085	0.479 ***

ns, *, **, and ***: non-significant, significant at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively. Total large PM: sPM (10–100 µm) + wPM (10–100 µm); Total coarse PM: sPM (2.5–10 µm) + wPM (2.5–10 µm); SLA: specific leaf area; Chl a: chlorophyll a concentration; Chl b: chlorophyll b concentration; TChl: Total chlorophyll concentration; Carotenoid: carotenoid concentration; pH: leaf extract pH; RWC: relative leaf water content.

In this study, PM accumulated on the leaf surface had a significant correlation with epicuticular wax quantity. Epicuticular wax is an important element of plants. The epicuticular wax can prevent pollution from penetrating plant cells and losing leaf water. In this study, we found that the quantity of epicuticular wax was different depending on the plant species. This is because the amount and structure of epicuticular wax depend on genetic factors, environmental (rain, temperature, and humidity) factors, and air pollution. In the present study, the amount of epicuticular wax was higher in six plant species growing along the roadside than in plants growing in the urban forest, and the species that did not follow this trend were *S. vulgaris*, *Z. serrata*, *M. glyptostroboides*, and *P. trobus*. It was also higher in plants with needle leaves than in those with broad leaves. Similar results have been reported previously [25]. However, the epicuticular wax structure can be destroyed by air pollution or acid rain, leading to erosion of the epicuticular wax [51,52]. In the industrial and traffic regions, the epicuticular wax could be eroded because of PM on leaf [53]. It might be the reason for a large reduction in the amount of epicuticular wax on *M. glyptostroboides* and some other plant species growing along the roadside.

RWC was used as an indicator to evaluate water status of the plants. RWC is intimately associated with growth, photosynthesis, and stomatal conductance [50]. RWC not only maintains physiological balance, but also influences water and nutrients of a plant. Increasing air pollution level can increase cell permeability, resulting in the loss of water and dissolved nutrients [30,50,54]. RWC content tends to decrease in plants growing at sites with higher pollution [23]. In this study, we did not find any significant correlation between PM and RWC. However, RWC was increased in all plant species from the roadside except for *M. denudata*. We suppose that the RWC of *M. denudata* growing along the roadside was decreased due to a large amount of large PM (10–100 µm) on its leaf surface. Conversely, the increase of RWC content can contribute to a tolerance against pollution for plants growing along the roadside [33].

SLA is leaf area divided by leaf biomass. It is used to determine the leaf thickness and density. Plants with thick leaves can be more effective in absorbing light. SLA depends on the degree of shading and PM accumulation on the leaf surface that can increase shadow on the leaf. However, SLA fluctuates depending on plant species and is related to a plant's protective or adaptive mechanism [33,55,56]. In this study, 4 (*P. densiflora*, *M. glyptostroboides*, *R. yedoense*, and *S. vulgaris*) out of 10 plant species had higher SLA at the roadside. It was found that large PM (10–100 µm) had negative correlation with SLA at both sites. At polluted sites, coarse PM (2.5–10 µm) also had negative correlation with SLA. These findings are consistent with research results of [57].

The other important trait of a plant is its leaf extract pH (pH), which is a sensitive indicator of air pollution. A low pH makes a plant more sensitive to air pollution while a high pH could increase the conversion of hexose sugar to ascorbic acid, thus increasing the plant's tolerance to air pollution [58]. In this study, the pH of plants growing in the urban forest ranged from 4.50 to 6.09, while the pH of plants growing along the roadside ranged from 5.09 to 6.02. Some pH values of plants were significantly different between the two sites or between species (Table 3). The leaf extract pH of the plant can be affected by air pollutions, such as SO_2 and NO_2 [59]. The pH values of some plant species growing along the roadside were higher than those growing in the urban forest for the same plant species (*A. palmatum*, *J. chinensis*, *T. cuspidata*, *P. densiflora*, and *P. strobus*), while pH values of a few other plant species at the roadside were lower (*S. vulgaris*, *M. denudata*, *Z. serrata*, *R. yedoense*, and *M. glyptostroboides*). These differences in pH values were due to the difference in tolerance of the plants to air pollutants. In this study, we found that the amount of coarse PM (2.5–10 μm) had negative correlation with pH only in the roadside. In addition to influence of PM, the leaf extract pH of plant can be influenced by other factors, such as soil pH. We need more study to determine the influence of the individual factors on the pH value.

5. Conclusions

The ability of PM accumulation of 10 different plant species and the influence of PM on leaf traits was analyzed. In this study, we found that PM accumulation was different depending on plant species and collection site. The amount of PM accumulation on the leaf surface and the amount of PM accumulation in wax were also different. The total PM accumulation of roadside plants was higher than that of urban forest plants. Additionally, needle leaves showed higher PM accumulation than broad leaves. Among the 10 plant species, *P. strobus* was the most effective plant species for PM accumulation at both sites. The micromorphology of leaves affected PM accumulation of plants. In some plant species, we found that leaves with specific features, such as grooves or thick wax layers, could increase the PM accumulation capacity. The epicuticular wax quantity of plants was positively correlated with the amount of PM accumulation on the leaf, and the amount of PM accumulation in wax contributed to the total PM accumulation on the leaf. Coarse PM (2.5–10 μm) affected all the leaf traits of plants growing along the roadside, while large PM (10–100 μm) affected SLA and the amount of epicuticular wax. The tolerance of plants to air pollution might be due to increased chlorophyll content, carotenoid, pH, and SLA for plants growing along the roadside. Epicuticular wax was positively correlated with large PM (10–100 μm) and coarse PM (2.5–10 μm). Among the 10 plant species, on the basis of the present work evidence, *P. strobus* can be a great choice to improve air quality in urban sites. Assessing the correlation between PM and other leaf trait is considered a future study. This study showed the ability of PM accumulation of different plant species, and the influence of PM concentration level on the amount of PM accumulation on the leaves. Additionally, the influence of PM on leaf traits was also clarified in this study. However, the PM concentration levels of the two sites need to be analyzed, and the influence of different factors on leaf traits also needs analysis to determine more exactly the influence of PM on leaf traits.

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