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Particulate matter deposition on roadside plants and the importance of leaf trait combinations



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ABSTRACT

Road and vehicle use in urban environments are key contributors to urban air pollution and increase concentrations of carbon monoxide, polyaromatic hydrocarbons and particulate matter (particles <100 μm diameter). Plants, which can intercept these pollutants, are increasingly recognised as practical mitigation methods to reduce ambient pollution, especially adjacent roadsides. We quantified particulate matter loads in 16 common native species along Sydney roadsides and **linked findings to leaf traits**. For each species, we tagged individuals within the first 2 m of road edges and recorded leaf area, shape and arrangement, also noting the presence of leaf hairs. We then quantified particulate matter loads deposited in each sample over three months and, for two morphologically distinct species, *Acacia parramattensis* and *A. longifolia*, the composition and concentration of metals in deposited particulate matter. We found particulate deposition varied according to species and leaf shapes but not sample months and, those species with leaf hairs accumulated significantly more particulate matter. Furthermore, we found metals associated with vehicle use including **copper, chromium and manganese** in collected particulate matter. Ultimately, our results highlight the **importance leaf trait combinations** can have in affecting particulate matter deposition.

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1. Introduction

Air pollution is a pervasive and increasing threat to both human and ecosystem health that requires practical remediation methods. One specific air pollutant of particular concern is particulate matter (PM) or particles with aerodynamic diameters in the range of 0.001–100 μm . These particles can contain toxic compounds including polyaromatic hydrocarbons and heavy metals (Ram et al., 2015) which, if inhaled can lead to respiratory and cardiac diseases (Polichetti et al., 2009). In urban areas a major source of PM is vehicle use (Vu et al., 2015). Whilst we can reduce PM by restricting the number of vehicles or limiting construction and industrial processing, an additional way to reduce PM involves using vegetation (Hirabayashi and Nowak, 2016; Nowak et al., 2014). Model estimates of PM deposition on vegetation in urban areas indicate plants offer a significant sink for PM and a route by which pollutants, including heavy metals, can be removed from the atmosphere (Räsänen et al., 2014; Popek et al., 2013; Qiu et al., 2009; Escobedo and Nowak, 2009). Quantifying the amount of PM that deposits and accumulates on different plant species is the first step towards

improving these model estimates and understanding the implications of PM accumulation on vegetation.

The surfaces and waxy epicuticular layers of leaves are the primary receptors of PM. A plant's capacity to capture PM is affected by several factors including the microstructure of the leaf's surface, the macrostructure of vegetation and environmental variables like wind and temperature (Mo et al., 2015; Chen et al., 2016). Microstructural features like rough surfaces, pubescence, thick waxy epicuticles and low stomatal densities along with macrostructural features like increased plant height, whorled leaf arrangements and larger leaf areas are all individual traits that enhance PM accumulation (Mo et al., 2015; Nowak et al., 2006; Popek et al., 2013; Chaturvedi et al., 2013; Prusty et al., 2005). To maximise the amount of PM captured and thereby the improvement to air quality that urban plantings have, it is necessary to understand which species and micro- and macrostructural traits are most effective in removing PM. Whilst the importance of individual traits on PM deposition is well known, we are yet to appreciate how different combinations of these traits interact to influence PM accumulation.

Depending on the amount and elemental composition of PM deposited on a leaf's surface and/or epicuticular wax layer and the plant species considered, PM can cause physiological and morphological impacts including increased cell alkalinity, photosynthetic

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inhibition, leaf senescence, stomatal damage, and reduced growth and yield (Rai, 2016; Daresta et al., 2015). For this reason, it is critical to quantify PM deposition in multiple plant species, especially species that are commonly planted along roadsides and in cities where ambient PM levels are greatest (Vu et al., 2015).

We quantified PM deposition on leaf surfaces in 16 native Australian species along roadsides in the Greater Sydney Region. Specifically, we determined the concentrations of metals in the PM on leaves and quantified differences in PM deposition among species, relating this to several micro- and macro-structural traits. We hypothesised that species with leaf hairs (e.g. *Westringia fruticosa*) would report greater PM deposition values because these traits typically enhance deposition.

2. Materials and methods

2.1. Plant material and study sites

We quantified particulate matter deposition on 16 native species at eight sites along moderately to highly trafficked roads in the Greater Sydney Region (Table 1). We chose sites that were greater than 10 m wide, were flat or had little slope and were adjacent roads with speed limits between 60 and 70 km/h.

2.2. Sample collection

To quantify variation in the PM depositing on plant species over time, we sampled once a month for three months between October 2012 and December 2012. In each case, sampling occurred in the last week of each month. Due to differences in plant abundance between sites, numbers of replicate plants ranged between two and six individuals. We sampled plants along ≤ 50 m strips of roadside, within ~ 2 m of the road edge. For those plants with few individuals per site, we sampled all individuals. For abundant species, we sampled replicates according to an n^{th} nearest sampling protocol, where n is a random number (Barbour et al., 1987). Once sample plants were initially located, we tagged and sampled the same individual for the next three months. For each sample, we collected 2–5 terminal shoots of similar length (< 20 cm) at breast height (approximately 1.5 m). We chose this sampling height due to height dependent differences in particulate deposition (Mitchell et al., 2010). Samples were randomly selected and either directed towards the road or not. For each terminal shoots, we picked 30 young and undamaged leaves, between the 2nd and 8th node for PM analysis. We found 30 leaves to be the minimum number required to quantify PM deposition. Upon collection, we placed 30 leaves in plastic bags with strips of absorbent towel, labeled each bag and stored in a refrigerator until analysis.

2.3. Quantitative analysis of particulate matter

We first dried all filter papers for 60 min at 100°C in a drying oven stored them in a desiccating chamber to stabilise the humidity and after 10 min weighed papers.

To quantify PM for each sample, we placed leaves in a glass container with 200 mL of reverse osmosis water and agitated for 60 s; this represents the PM washed off during rainfall and not PM captured in the epicuticular wax layer (Dzierzanowski et al., 2011). We then filtered the water using a sieve with mesh diameter $100\ \mu\text{m}$. We next filtered the solution using a 15 mm Buchner funnel connected to a vacuum pump, first on pre-weighed filter paper, Whatman grade 541 (retention $22\ \mu\text{m}$) and next on Whatman grade GF/A (retention $1.6\ \mu\text{m}$). We, therefore, collected two size fractions of PM: (1) 'large': $22\ \mu\text{m}$ – $100\ \mu\text{m}$, and (2) 'small': $1.6\ \mu\text{m}$ – $22\ \mu\text{m}$. We quantified the large fraction for all species but could only quantify the small fraction for *Acacia parramattensis*,

Acacia longifolia and *Banksia integrifolia* (small fraction results not reported due to low replicate number). We then dried and re-weighed papers following the same procedure as in pre-weighing, to calculate PM mass in each fraction of every sample.

To measure the total area of each leaf we used image analysis program imageJ (Rasband, W.S., ImageJ, U. S. National Institutes of Health, Bethesda, Maryland, USA, <http://imagej.nih.gov/ij/>, 1997–2012) following Osunkoya et al. (2010). Although PM was washed from both abaxial and adaxial leaf surfaces, we expressed the amount obtained per unit area of leaf. In addition to leaf area, we recorded habit (tree or shrub), leaf shape (linear, lanecolate, obovate, needle like or elliptic), leaf arrangements (opposite, alternate or whorled) and the presence of leaf hairs for each plant species sampled (Table 1).

2.4. Metal element analysis by ICP-AES

To determine the metal species in the PM collected, we first dissolved filter papers by gently refluxing with nitric and hydrochloric acids. We then analysed trace metal species using inductively coupled plasma-atomic emission spectroscopy (ICP-AES) following the US Environmental Protection Agency (USEPA) method 200.8 (USEPA, 1994). We tested for aluminium (Al) and iron (Fe), in addition to metal species more commonly associated with traffic, including: lead (Pb), zinc (Zn), cadmium (Cd), chromium (Cr), nickel (Ni), manganese (Mn) and copper (Cu) Hergren and Goonetilleke (2006). Issues with detectability meant we could only quantify Cu, Cr, Mn, Al and Fe in each sample. In each case, we ran metal analysis on leaf material collected from two morphologically distinct species: *A. parramattensis* and *A. longifolia* at three sites.

2.5. Statistical analysis

As data were not distributed normally, we used the non-parametric equivalent of a 1-way ANOVA, known as Kruskal-Wallis test (Field, 2009) to determine if the dependent variable, PM amount on leaves differed between each of five independent variables including (1) species, (2) leaf shapes, (3) leaf arrangements, (4) plant habits and (5) sampling sites. In cases where we found significant differences, we used Dunn's non-parametric pairwise comparisons to determine which levels of the independent variable were significantly different from each other. We used Mann-Whitney U tests to compare the PM amount between leaves with or without hairs. For each analyses, we used the average PM amount recorded over the three-month sampling period. To determine if PM deposition varied over the sampling period we used Friedman's analysis of variance (ANOVA), a non-parametric version of repeated measures ANOVA (Field, 2009).

To compare differences in the concentrations of metals (Cu, Cr, Mn, Al and Fe) quantified in the PM collected from *A. parramattensis* and *A. longifolia* leaves, we conducted separate 1-way ANOVAs. We applied a Bonferroni correction to account for increased type 1 error (adjusted significance level $P < 0.01$). We used multiple ANOVAs rather than a multivariate ANOVA owing to multicollinearity between several metals (Quinn and Keough, 2002). In each case data conformed to assumptions of normality and homogeneity of variances.

3. Results

3.1. PM deposition: species and leaf traits

PM loads differed significantly among species ($H_{15} = 82.68$, $P < 0.001$; Fig. 1), leaf shapes ($H_4 = 28.08$, $P < 0.001$; Fig. 2) and plants with or without leaf hairs ($U = 4295$, $z = 2.14$, $P = 0.03$; Fig. 3). Pairwise comparisons revealed *Westringia fruticosa* had significantly

Table 1
Plant species, leaf characteristics and number of sampling sites for sampled flora.

Species	Family	Habit	Height (m) ^a	Leaf arrange-ment	Leaf shape	Leaf hairs ^b	Petiole length ^c	Sample Sites ^d
<i>Acacia linifolia</i>	Fabaceae	Shrub	1.5–4	Alternate	Linear	–	s	4
<i>Allocasuarina littoralis</i>	Cauearinaceae	Tree	5–15	Alternate	Needle like	–	n/a	4
<i>Acacia longifolia</i>	Fabaceae	Shrub	8	Opposite	Leaflets linear	+	s	5
<i>Acacia parramattensis</i>	Fabaceae	Shrub	2–15	Alternate	Leaflets linear	–	l	5
<i>Banksia integrifolia</i>	Proteaceae	Tree	5–25	Whorled	Elliptic	+	l	4
<i>Banksia spinulosa</i>	Proteaceae	Shrub	≤3	Alternate	Linear	–	l	4
<i>Callistemon rigidus</i>	Myrtaceae	Shrub	2–3	Alternate	Linear	+	s	6
<i>Dodonaea triquetra</i>	Sapindaceae	Shrub	3	Alternate	Elliptic	–	l	4
<i>Elaeocarpus reticulatus</i>	Elaeocarpaceae	Tree	3–15	Whorled	Elliptic	–	s	2
Unidentified <i>Eucalypt</i> spp.	Myrtaceae	Tree	≥15	Alternate	Elliptic	–	l	4
<i>Hakea salicifolia</i>	Proteaceae	Tree	5–8	Alternate	Lanceolate	–	s	4
<i>Hakea sericea</i>	Proteaceae	Shrub	1–3	Alternate	Needle like	–	s	4
<i>Melaleuca styphelioides</i>	Myrtaceae	Tree	20	Alternate	Obovate	–	s	2
<i>Persoonia levis</i>	Proteaceae	Tree	5	Alternate	Obovate	–	l	4
<i>Syzygium australe</i>	Myrtaceae	Shrub/tree	35	Opposite	Elliptic	–	s	4
<i>Westringia fruticosa</i>	Lamiaceae	Shrub	1.5	Whorled	Lanceolate	+	s	6

^a Natural variation in height. Note, all species sampled were at least 1.5 m tall.

^b + and – indicate the presence and absence of hairs on leaves respectively.

^c Petioles were grouped into two categories – s: small (<0.5 cm) or l: large (>1 cm).

^d Number of sites where species was present and sampled.

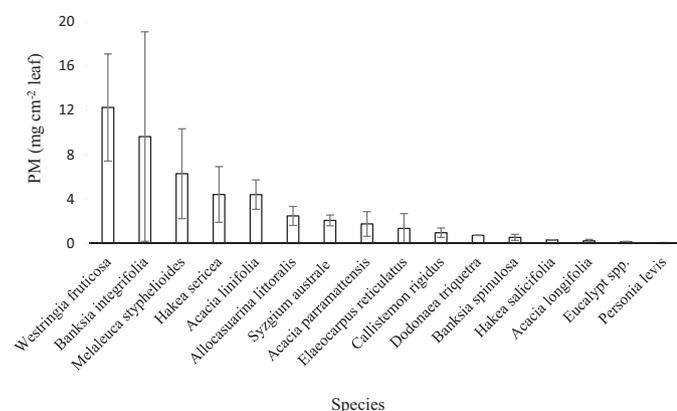


Fig. 1. Amount of PM (mean + S.E.) collected on leaves of 16 different species and expressed in mg cm⁻² of leaf. A minimum of five replicate plants per species were sampled.

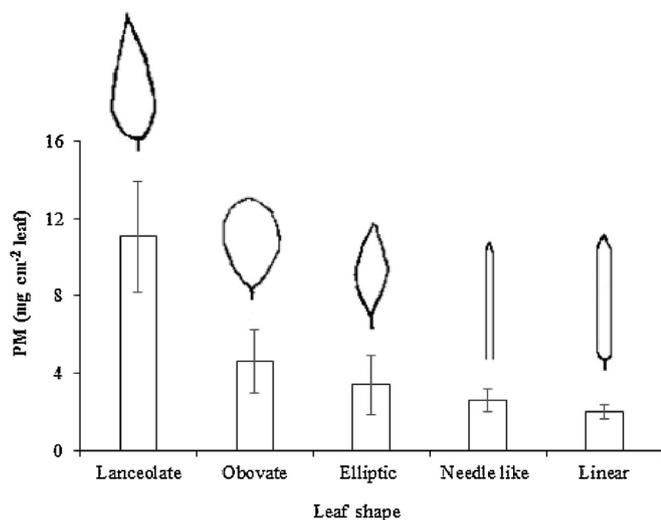


Fig. 2. Amount of PM (mean + S.E.) collected on leaves with different shapes and expressed in mg cm⁻² of leaf. For each leaf shape a minimum of two replicate plant species and 10 plants per species was sampled. Letters indicate a significant difference at $P < 0.05$.

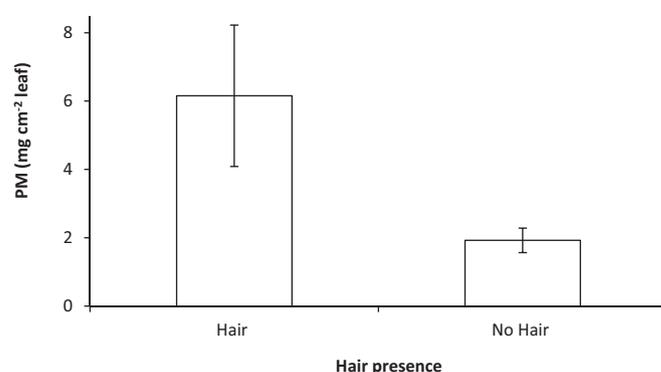


Fig. 3. Amount of particulate matter (PM: mean + S.E.) collected from plants with or without leaf hairs and expressed in mg cm⁻² of leaf. PM was quantified on the leaves of four species with leaf hairs ($n = 85$) and twelve species without leaf hairs ($n = 131$). * indicates significant difference between plant species at $P < 0.01$ significance level.

more PM than *A. longifolia*, *Banksia spinulosa*, *Callistemon rigidus*, *Elaeocarpus reticulatus* and *Persoonia levis* (adjusted $P < 0.05$ in all cases). Furthermore, *A. longifolia* had significantly less PM than *Allocasuarina littoralis*, *Acacia linifolia* and *Melaleuca styphelioides*. Additionally, unidentified *Eucalypt* spp. had significantly less PM than *A. littoralis*, *A. linifolia*, *M. styphelioides* and *W. fruticosa* and, *P. levis* had significantly less PM than *M. styphelioides* (adjusted $P < 0.05$ in all cases). Pair-wise comparisons revealed lanceolate shaped leaves retained significantly more PM than linear, elliptic and needle like leaf shapes (adjusted $P < 0.01$ in all cases). Leaves with hairs also had significantly greater PM loads than leaves without hairs ($U = 4295$, $z = 2.14$, $P = 0.03$).

PM amount (mean + S.E.) did not differ between whorled ($6.33 + 3.95$ mg cm⁻²), opposite ($0.56 + 0.32$ mg cm⁻²) or alternate ($1.78 + 0.28$ mg cm⁻²) leaf arrangements ($H_2 = 3.91$, $P = 0.14$); nor between trees ($3.00 + 0.68$ mg cm⁻²) and shrubs ($3.88 + 1.15$ mg cm⁻²) ($U = 3998$, $z = -0.55$, $P = 0.58$) or, over the three sampling months ($H_{22} = 1.45$, $P = 0.49$).

3.2. Metal species in PM

Acacia longifolia and *A. parramattensis* did not differ in the concentrations of Cu ($F_{1,7} = 0.53$, $P = 0.49$), Mn ($F_{1,8} = 2.83$, $P = 0.13$), Al ($F_{1,5} = 0.24$, $P = 0.64$) or Fe ($F_{1,8} = 3.01$, $P = 0.12$) found in their PM (Table 2); however, Cr concentrations differed significantly

Table 2
Concentration, expressed in mg kg⁻¹ PM, for 5 metal species found in PM samples collected from *Acacia parramattensis* and *Acacia longifolia* leaves. PM was collected from 6 replicate plants per species and 30 replicate leaves per plant. * indicates significant difference between plant species at $P < 0.01$ significance level.

Species	Cu	Cr*	Mn	Al	Fe
<i>A. parramattensis</i>	1.21 ± 0.53	0.51 ± 0.11	0.75 ± 0.34	249.62 ± 138.24	63.43 ± 32.10
<i>A. longifolia</i>	4.49 ± 3.82	0.09 ± 0.04	0.22 ± 0.08	300.84 ± 124.5	16.98 ± 7.25

between species ($F_{1,7} = 16.64$, $P = 0.05$). *Acacia parramattensis* had greater concentrations of Cr than *A. longifolia* (Table 2).

4. Discussion

By determining PM load on 16 native species we have provided the first estimates of PM accumulation on Australian flora. We found leaf shape, species and the presence of leaf hairs, but not habit or leaf arrangement to be important factors influencing the amount of PM on leaves. Combinations, rather than single traits may, therefore, be critical in influencing PM deposition and accumulation.

Our results show that lanceolate shaped leaves, which are broadest below the middle, accumulate more PM than obovate and elliptic shaped leaves which are narrowest below the middle. Leaves with narrow leaf bases have higher surface specific drag and flutter more erratically than leaves with broader bases (Vogel, 1989). By fluttering more erratically, we suspect leaves decrease the chance of PM deposition and increase the chance of PM dislodging. We predict additional traits which promote leaf movement and thereby the chance of removing PM include larger leaf surface areas and increased petiole length (Vogel, 1989). In support of this prediction, we found that species with larger leaf areas including *P. levis* and longer petioles including *Dodonaea triquetra* accumulated less PM on their leaves than species with smaller leaf areas and shorter petioles including *W. fruticosa* and *M. styphelloides*. Our results, therefore, provide support for the hypothesis that certain leaf traits affecting leaf movement including leaf area, shape and petiole length are also important factors influencing PM deposition on leaves. The relationship between a tree or shrub's leaf area (leaf area index) and PM deposition is of particular interest here because this factor is already known to positively correlate with deposition load (Liu et al., 2015). The possibility that large leaf areas both increase and decrease PM deposition by providing greater surface area for PM attachment but also increasing leaf movement which causes PM dislodgement is an interesting topic that warrants future research.

Our results also show that PM accumulation is greater on leaves with hair. Leaf hairs not only increase the surface area that can intercept PM but, may also make it harder for PM to dislodge when leaves are moving (Neinhuis and Barthlott, 1998; Qiu et al., 2009; Prusty et al., 2005). Additionally, by creating surface polarity, the hydrophobicity of certain leaf hairs may help to attract charged particles including certain metal species commonly found in PM Fernández et al., 2014.

Interestingly, *A. longifolia*, despite having leaf hairs still accumulated significantly less PM than *A. littoralis*, *A. linifolia* and *M. styphelloides*, species without leaf hairs. Given *A. longifolia* have waxy epicuticles, the hydrophobicity of wax particles on *A. longifolia* leaf surfaces may have decreased the PM collecting ability of these leaves (Faini et al., 1999). In support of this, two additional species that accumulated less PM than *A. longifolia* were *Eucalyptus* sp. and *P. levis*, species which also have thick waxy epicuticular layers. This finding highlights the importance that trait combinations, in this case hair presence and the wax epicuticular layer have on PM deposition.

Several of our findings further suggest certain traits may override the positive effect leaf hairs and lanceolate shaped leaves

have on PM deposition. *Hakea salicifolia*, for example, despite having lanceolate shaped leaves recorded the fourth lowest PM load. The presence or combination of additional traits including alternate leaf arrangement, absence of leaf hairs and short petioles may be responsible for this result however, it would be worth further investigating the effect of additional traits like surface roughness and stomatal density, factors known to significantly influence PM deposition (Mo et al., 2015). The large difference in PM deposition between *P. levis* and *M. styphelloides* also suggests a possible overriding effect of specific traits on trait combinations. Both *P. levis* and *M. styphelloides* have alternate leaf arrangements, obovate leaf shapes and no leaf hairs but *P. levis* accumulated the least and *M. styphelloides* the third greatest amount of PM on their leaves amongst all species sampled. In this case, because *P. levis* and *M. styphelloides* differ in leaf size and petiole length it is possible these traits led to the observed differences in PM deposition. Ultimately, these findings make it difficult to draw conclusions about trait combinations that maximise PM deposition.

Contrary to patterns found in Southeast Asian plant species, where whorled leaf arrangements accumulate significantly more PM than non-whorled arrangements (Chaturvedi et al., 2013; Prusty et al., 2005), we found no significant effect of leaf arrangement on PM load. Possible mechanisms to account for this result are (1) large variation in the PM amount quantified on plants with whorled leaf arrangements especially *B. integrifolia* and, (2) the overriding effects of other traits including leaf shape and hair presence.

Although the amount of PM (mg cm⁻²) in the two *Acacia* species we sampled were comparable to PM levels found in other species including *Syzygium cumini* (4 mg cm⁻²), *Tectona grandis* (7 mg cm⁻²), *Anthocephalus cadamba* (7 mg cm⁻²) (Chaturvedi et al., 2013), the concentrations of heavy metals associated with PM from vehicle exhaust and tire wear particles (Cu, Cr and Mn) were below levels reported for other species adjacent roads (e.g. *Bougainvillea spectabilis* Cu = 4310 mg kg⁻¹) (Zheng et al., 2013; Qiu et al., 2009). One possible reason for this is Australia's substantially lower PM emissions (4-year average: PM_{2.5}: 7.62 ± 4.36 μg m⁻³, PM₁₀: 10.49 ± 7.47 μg m⁻³) and heavy metal composition of PM compared to China (4-year average: PM_{2.5}: 42.14 ± 31.46 μg m⁻³, PM₁₀: 109.29 ± 56.47 μg m⁻³) and India (4-year average: PM_{2.5}: 44.69 ± 17.68 μg m⁻³, PM₁₀: 82.83 ± 33.01 μg m⁻³), where these studies were published (Hopke et al., 2008). The presence of heavy metals especially Cu, Cr and Mn suggests the main source of PM within our study sites is diffuse emission sources like vehicle exhaust and tire wear particles and not point emission sources like industry (Wei and Yang, 2010). We found large concentrations of Al and Fe, compared to the other metals quantified. These metals are typically associated with soil particles adjacent roadsides, including feldspar Hergren and Goonetilleke (2006). It is possible that passing vehicles increased the ambient concentration of soil particles at our sampling sites, leading to increased deposition of Al and Fe on leaf surfaces.

5. Conclusion

Our findings show Australian vegetation accumulates PM and, based on the metals present, that this PM is derived from vehicle exhaust, tire wear particles and re-suspended roadside soil. This

is particularly relevant to urban areas where road and vehicle use is a significant contributor to decreasing air quality. Traits including leaf shape and leaf hairs are important factors influencing PM deposition, however, their effectiveness may be undermined by the presence of other traits. We provide baseline data for future study including the implications of deposited PM on vegetation and the relative importance of leaf and whole plant traits in influencing the PM deposited.

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Author's contributions

Study conception and design: R.L, C.M. and D.H., acquisition of data, analysis and interpretation of data: R.L., manuscript draft and revision: R.L., D.H. and C.M. All authors approve the publication and we have no competing interests.

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