**Quantification of the traffic-generated particulate matter capture by plant species in a living wall and evaluation of the important leaf characteristics.**

**Abstract**

Traffic-generated particulate matter (PM) is a significant fraction of urban PM pollution and little is known about the use of living walls as a short-term strategy to reduce this pollution. The present study evaluated the potential of twenty living wall plants to reduce traffic-based PM using a living wall system located along a busy road in Stoke-on-Trent, UK. An Environmental Scanning Electron Microscope (ESEM) and ImageJ software were employed to quantify PM accumulation on leaves (PM1, PM2.5 and PM10) and their elemental composition was determined using Energy Dispersive X-ray (EDX). Inter-species variation in leaf-PM accumulation was evaluated using a Generalised Linear Mixed-effect Model (GLMM) using time as a factor; any differential PM accumulation due to specific leaf characteristics (stomatal density, hair/trichomes, ridges and grooves) was identified. The study showed a promising potential for living wall plants to remove atmospheric PM; an estimated average number of 122.08 ± 6.9 x 107 PM1, 8.24 ± 0.72 x 107 PM2.5 and 4.45 ± 0.33 x 107 PM10 were captured on 100 cm2 of the living wall used in this study. Different species captured significantly different quantities of all particle sizes; the highest amount of all particle sizes were found on the leaf-needles of *Juniperus chinensis* L., followed by smaller-leaved species. In the absence of an apparent pattern in correlation between PM accumulation and leaf surface characteristics, the study highlighted the importance of individual leaf size in PM capture irrespective of their variable micro-morphology. The elemental composition of the captured particles showed a strong correlation with traffic-based PM and a wide range of important heavy metals. We conclude that the use of living walls that consist largely of smaller-leaved species and conifers can potentially have a significant impact in ameliorating air quality by removing traffic-generated PM pollution to improve the wellbeing of urban dwellers.

Key words: Green infrastructure; Air pollution; Traffic-based pollution; Leaf characteristics

1. **Introduction**

Epidemiological studies have revealed short and long-term impacts of PM and found increased levels of, hospital admissions, morbidity and mortality around the world (Pascal *et al.*, 2014; Pope III *et al.*, 2011; Seaton *et al.*, 1995). Traffic-generated particulate matter (PM) is known to be responsible for a significant portion of urban particulate pollution (Pant and Harrison, 2013; Ranft *et al*., 2009). Globally, 25% of PM2.5 (PM with less than 2.5 µm aerodynamic diameter) and PM10 (PM with less than 10 µm aerodynamic diameter) were estimated as being generated from road traffic (Karagulian *et al*., 2015) and in Europe road traffic is known to be responsible for more than 50% of PM10 (Künzli *et al*., 2000). Some studies found direct links between traffic-based PM and detrimental health effects (Brook *et al.*, 2010; Hyun Cho *et al.*, 2005; Ranft *et al.*, 2009). Long-term exposure to these particles is known to cause premature death and various illnesses such as cardio-pulmonary diseases, lung cancer, allergies, brain damage and neuro-degenerative diseases (Brook *et al.*, 2010; Pascal *et al.*, 2014; Pope III *et al.*, 2011; Seaton *et al.*, 1995; Maher *et al.*, 2016), and have even been linked to cognitive impairments in elderly people (Ranft *et al.*, 2009). Pascal *et al.* (2014) also found a considerable rate of cardiovascular, cardiac, and ischemic mortality following short-term exposure to PM. Vehicular emission of PM0.1 in the ultrafine range was recently highlighted as being important due to its higher toxicity and larger numbers compared to other size fractions (Lin *et al.*, 2005). PM such as elemental carbon, hydrocarbons, metals, oxides of nitrogen, sulfates, ammonia (Fauser, 1999; Sharma *et al.*, 2005) from vehicle exhaust, and various non-exhaust emissions from brake wear, tyre wear, clutch wear, abrasives, and road dust, are responsible for this pollution (Mulawa et al., 1997; Slezakova et al., 2007; Thorpe and Harrison, 2008; Timmers and Achten, 2016; Wåhlin et al., 2006).

Despite several initiatives taken to mitigate this pollution, such as reducing emission levels and separating high emission zones from populous areas (Pugh *et al*., 2012), particulate levels in air still remain problematic for human health and the environment (e.g. reducing visibility, affecting urban thermal environment) (Han et al., 2012; Yan et al., 2016). Since plants are known to filter these pollutants, the ability of vegetative barriers to reduce near-road PM levels has recently been studied under different environmental and weather conditions using various techniques (Abhijith *et al*., 2017; Blanusa et al., 2015; Gautam *et al*., 2005; Maher *et al*., 2008; Mori *et al*., 2015; Šíp and Beneš, 2016; Tong *et al*., 2015; Weber *et al*., 2014; Weerakkody *et al*., 2017). The potential of vertical greenery systems is frequently cited as a possible short-term solution due to their smaller land utilization, quick installation, reduced dependency on existing soil conditions, and additional ecosystem services (Cheetham *et al*., 2012; Dover, 2015; Johnston and Newton, 2004). Previous research has mainly focused on the use of climbing species to capture PM with most not specifically relating to road traffic (Ottelé *et al*., 2010; Sternberg *et al*., 2010; Tiwary *et al*., 2008; Xia *et al*., 2011) but see Dover and Phillips (2015). Except for the work of Perini *et al*., (2017), who studied two shrub species and two climbing species in a green façade located above street level, a small-scale study on shrubs (which included few living wall species) by Shackleton *et al*. (n.d.), and work by Weerakkody *et al*. (2017), relating to PM generated by rail traffic, the value of living wall plants in the reduction of motor traffic-based PM has not been evaluated.

Living walls are vertical greenery systems holding multiple species of plants usually grown at ground level, and contrasts with façade systems using climbers; living walls require piped irrigation systems which typically also deliver nutrients (Dover, 2015). The present study evaluated the potential of different living wall plants to capture and retain near-road PM which are mainly derived from traffic pollution. Inter-species variation in capture and retention of PM under the same environmental and weather conditions were evaluated to identify the best species to use as near-road PM filters. Leaf micro-morphological variations suspected to control their ability to capture and retain PM (Dzierzanowski *et al*., 2011; Kardel *et al*., 2012; Leonard *et al*., 2016; Sæbø *et al*., 2012; Räsänen *et al*., 2013; Wang *et al*., 2011; Weerakkody *et al.*, in press) were also investigated. The elemental composition of captured particles was analysed to identify their links to traffic-generated PM and potential health effects.

**2. Material and methods**

2.1 Site description

An experimentally manipulable, freestanding, modular living wall system was employed in this research. The wall was donated, designed, and installed by Nemec Cascade Garden Ltd., Czech Republic, in June 2016. The wall was erected 6 m from the edge of Leek Road, a busy ‘A’ road adjacent to Staffordshire University, Stoke-on-Trent, UK (Fig. 1). The road has an average daily traffic flow of 20,251 vehicles (Department for Transport, 2017). Stoke-on-Trent is a city located in central England with a population density of 6,640 people/km2. The local mortality burden attributed to anthropogenic PM2.5 in Stoke-on-Trent was estimated at 5.6% in 2009 (Gowers *et al.*, 2014).

This living wall was a first-generation prototype, having two 3.98 m x 2.09 m planting areas, one on each side of the wall. The planting areas were separated from the ground by a 0.5 m tall concrete platform encased in metal. This was an artificially irrigated system equipped with water and thermal sensors for easy maintenance. As traffic pollution is of concern, only the planting area of the front side (facing the road) is described here. Ten plants per species of twenty different species (Table 1), both evergreen and deciduous species, with various leaf morphotypes (size, shape and micromorphology) were planted as ten rows and twenty columns using a Latin Square design.

2.2 Leaf sampling

Samples were only taken from the middle six rows of the wall, avoiding the upper most two rows (based on accessibility) and the bottom two rows to avoid ground-level contaminations. Prior to sampling, plants were washed using a watering hose connected to a pressure head to remove existing particles from the leaves. Plants were left undisturbed for ten weeks (23rd of June, 2016 to 1st September, 2016) allowing continuous exposure to weather changes to minimize the effect of any variability in baseline PM levels. Sampling was carried out on six days during September and October 2016 under dry weather conditions. Thirty-six leaves from *Thymus vulgaris* and 18 leaves from rest of each species were taken in total. Three leaves were randomly taken from each species on every sample day (i.e. 6 days x 3 leaves/day = 18 leaves) with the exception of *T. vulgaris* which had 6 leaves/day removed. All species were sampled on every sample day. Sampled leaves were arranged in clean plastic containers using blue tac on their petioles, avoiding cross contamination between leaves or with container surfaces. Subsequently, containers were sealed and transferred to the laboratory for analysis. Samples were analysed fresh or stored at 9 °C in a refrigerator in the same storage containers until analysed (within 2 days of sampling) (Weerakkody *et al.*, 2017).

2.3 Imaging leaves using a Scanning Electron Microscope (SEM)

Sample preparation and imaging followed the same approach as in Weerakkody *et al.* (2017). An Environmental Scanning Electron Microscope (ESEM) (Model: JSM-6610LV) was employed to image the particulates captured on leaves and the leaf micromorphology. Six leaf sections (5.0 mm x 5.0 mm), three from each adaxial and abaxial surface were excised from each of the leaves larger than 2.5 cm2 in area. Smaller leaves (apart from *T. vulgaris*) were cut into halves, one half to visualize adaxial surface, and one for the abaxial surface and mounted without further cropping. Leaves of *T. vulgaris* and leaf needles (*J. chinensis*) were scanned without cropping. *T. vulgaris* was too small to cut into halves and hence the adaxial and abaxial surfaces were scanned using adjacent leaves. Leaf sections were only excised from the leaf-blades (Weerakkody *et al.*, 2017); for the leaves mounted without cropping, three scanning areas were randomly selected from the leaf-blade following the same approach. As leaves of *J. chinensis* were needle-like, rather than having leaf blades, a different sampling approach was used: areas to be scanned were selected from the middle of the needles avoiding their tips and base. Nine micrographs from each leaf surface (both adaxial and abaxial surfaces) were taken at 450x and 1,000x using back scattered electron signals under a low vacuum to quantify the amounts of PM captured on the leaves.

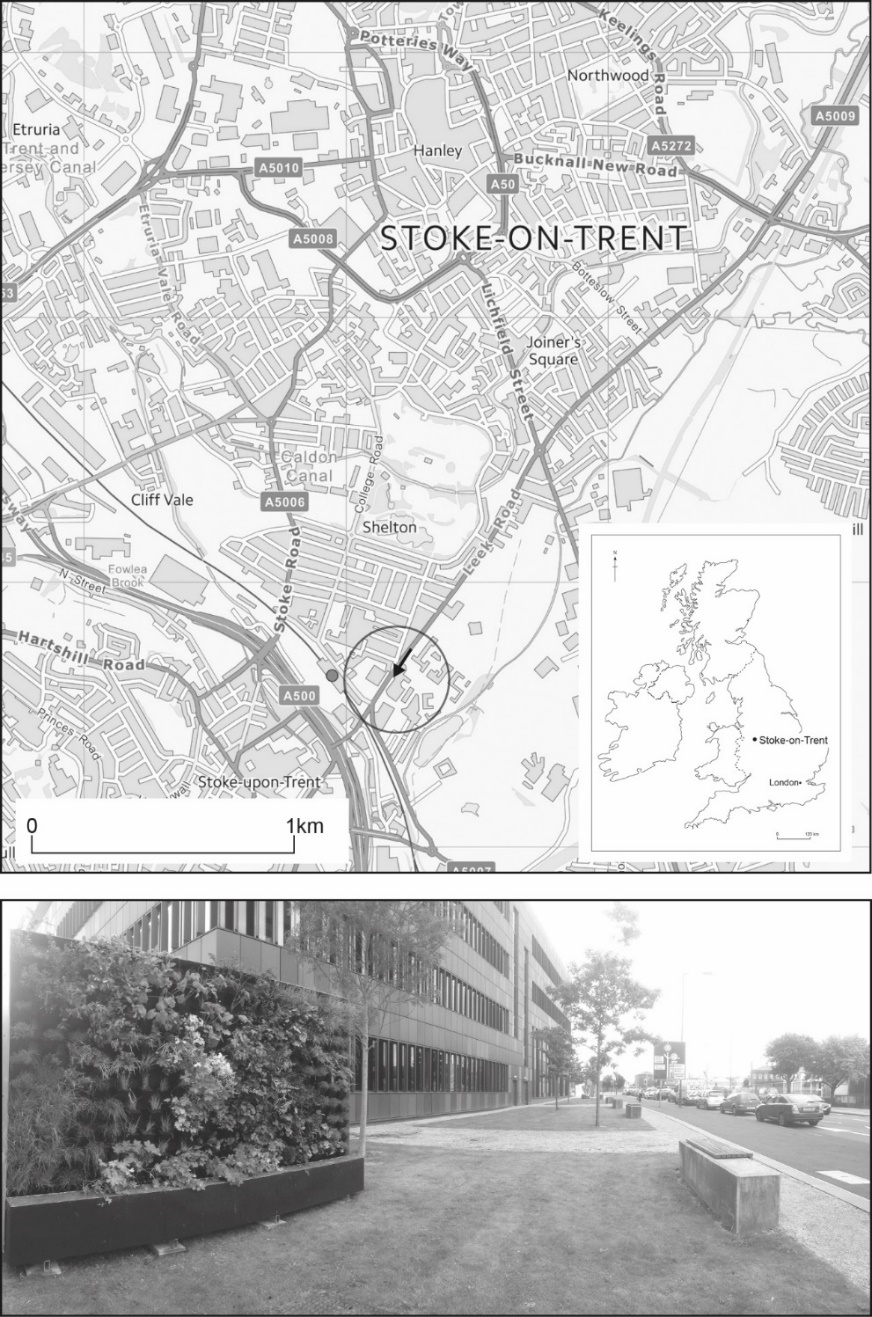


Fig. 1: The location of the study site in Stoke-on-Trent, UK (top) *Contains OS data ©Crown copyright and database right (2017),* and the location and orientation of the living wall relative to the road (bottom).

Table 1. Plant species used in the experimental living wall with a brief species description a

|  |  |  |  |
| --- | --- | --- | --- |
| **Species name** | **Species abbreviations** | **Common name** | **Species description and leaf morphology** |
| *Acorus gramineus Sol.* | A. g | Grass-leaf sweet flag | Semi-evergreen rhizomatous species having linear (“grass-like”) leaves forming tufts. Leaf surface texture was slightly rough. |
| *Berberis buxifolia* Lam. | B.b | Box-leaved barberry | Evergreen shrubs. Small, obovate, smooth and leathery leaves |
| *Bergenia cordifolia* (Haw.) Sternb. | B.c | Elephant's ears | Evergreen species. Large, ovate, glossy and smooth leaves. |
| *Berberis × media* Groot. ex Boom | B.m | Barberry 'Red Jewel' | Evergreen shrubs. Small, obovate, smooth and leathery leaves. |
| *Carex caryophyllea* Lat. | C.c | Spring sedge | Evergreen rhizomatous species. Linear, “grass like”, rough leaves. |
| *Deschampsia cespitosa* (L.) Beauv | D.c | Tufted hair grass | Evergreen bushy grass with rough, linear leaves forming dense tufts. |
| *Geranium macrorrhizum* L. | G.m | Cranesbill ‘Bevan's  Variety' | Semi-evergreen species. Large, palmately-lobed, hairy leaves. |
| *Geranium renardii* Trautv. | G.r | Renard geranium | Deciduous herbaceous plant with lobed and furrowed leaves |
| *Heuchera americana* L. | H.a | American alum root | Evergreen species. Large, glossy and palmately-lobed leaves. |
| *Heuchera villosa* Michx. var. *macrorhiza* | H.v | Hairy alumroot | Evergreen species. Large, hairy and palmately-lobed leaves. |
| *Helleborus × sternii* Turrill | H.s | Backthorn strain | Evergreen species. Large, obovate, leathery leaves with serrated margins. |
| *Veronica vernicosa* Hook.F. (*Hebe vernicosa*) | V.v | Varnished hebe | Evergreen shrub. Small, elliptic, glossy and leathery leaves. |
| *Juniperus chinensis* L. | J.c | Chinese juniper | Evergreen conifer. Small, branched leaf needles. |
| *Lonicera kamtschatica* Pojark. | L.k | Blue honeysuckle | Deciduous shrub. Large, elliptic leaves with slightly hairy/velvety texture. |
| *Persicaria amplexicaulis* ([D. Don](https://en.wikipedia.org/wiki/David_Don)) [Ronse Decraene](https://en.wikipedia.org/w/index.php?title=Ronse_Decraene&action=edit&redlink=1) | P.a | Red bistort 'Firetail' | Deciduous herbaceous species. Large, cordate and slightly wrinkled leaves. |
| *Phyllitis scolopendrium* L. | P.s | Hart's tongue fern | Evergreen fern. Large, elongated leathery fronds with irregular margins. |
| *Spiraea betulifolia* Pall. | S.be | Tor | Deciduous shrub. Medium-sized, smooth and oval leaves. |
| *Spirea japonica* L. | S.j | Japanese meadowsweet | Deciduous species. Small and ovate leaves with toothed margins. |
| *Stachys byzantina* K. Koch × *Stachys debilis* Kunth | S.by | Lamb's ear 'Silver Carpet' | Evergreen species. Densely hairy, velvety and elliptically shaped leaves. |
| *Thymus vulgaris* L. | T.v | Common thyme | Evergreen shrubs. Small, ovate leaves with slightly velvety surface texture. |

a Leaf shape descriptions based on Hickey and King (2000) and Beentje (2012)

Duplicates of each micrograph were taken with and without labels (including species name, scale, magnification and accelerating voltage) to use in the image analysis process for scale definition. As needles of *J. chinensis* were cylindrical, it was not possible to distinguish specific surfaces and hence nine micrographs were taken from a random part of the leaf surface. The same working distance, brightness, and contrast levels were used for all scans to avoid any complications in the image analysis process.

Following visualization of particulates, a further set of micrographs were taken to image micro-morphological features from ten random leaves previously scanned to quantify particles. As surface structures such as leaf hairs and trichomes are substantially larger than particulates, leaf surfaces were scanned at 90 x, 100 x and 250 x; in order to image leaf stomata, grooves and ridges, leaves were scanned at 300 x and 450 x. As these characters were unevenly distributed on leaves, focus settings were changed accordingly and ten random micrographs per character for each species were taken.

2.4. Analysing inter-species variation of PM capture

PM quantification followed the same approach described in Weerakkody et al. (2017). The micrographs taken for visualizing particulates were imported in imageJ image analysis software; those with labels were first used to define the scale, and those without labels were used to quantify PM1, PM2.5 and PM10 on each micrograph using the auto-threshold tool. As leaves have different surface areas, the amount of PM on leaves was expressed in terms of PM density, which is the number of particulates captured on a unit surface area (1 mm2). The PM density on each leaf was estimated separately for adaxial and abaxial surfaces by taking the mean PM densities on the respective micrographs. The mean PM density on each leaf was calculated by totaling the mean PM densities on adaxial and abaxial surfaces. The total PM density on leaf needles of *J. chinensis* was estimated as twice the PM density on the imaged surface to make it comparable with the other species, although we recognized this may underestimate total PM density on cylindrical leaf needles.

Inter-species variations in total PM density was identified using a mixed-Anova in a Generalized Linear Mixed-effect Model (GLMM) using R statistical software version 3.2.5 (R Development core team, 2016); as leaves were sampled on six different occasions, sample date (time) was included as a random factor. This analysis was then separately repeated for adaxial and abaxial surfaces to identify any variability between the species. Any significant difference in PM density between adaxial and abaxial surfaces of the same species of plant was identified using the Student’s t-test (R 3.2.5); as this was not applicable for needle-like leaves, *J. chinensis* was not included in this analysis.

The Leaf Area Index (LAI) was measured to estimate the total leaf area of the plants using the following formula (Weerakkody *et al.,* 2017):

LAI = Mean surface area of a leaf x Mean number of leaves per quadrat/ total surface area of the quadrat

Ten random leaves per species were used to calculate the mean leaf size and three random 10 cm x 10 cm quadrats per species were taken to estimate the mean number of leaves in a unit area of living wall surface. Individual leaf sizes were measured using ImageJ software, the area of leaf needles of *J. chinensis*  was estimated as for cylindrical shapes (Zhang *et al.*, 2015). The total PM capture by a 100 cm2 area of each species was then estimated using their mean PM densities on the leaves and LAI values (i.e. Mean PM density on the leaves x LAI of the species x 100 cm2) (Weerakkody *et al*., 2017). Inter-species variations in total PM capture levels (incorporating LAI) were identified using a mixed-Anova in GLMM (R 3.2.5) including time as a factor.

2.5 Evaluating the correlation between leaf micromorphology and PM accumulation

The number of leaf hairs/trichomes and stomata on micrographs were quantified and expressed as number of characters per unit area (1 mm2). Ridges and grooves on leaves were varied in shape, size and distribution and they were thus estimated as the percentage area covered by the character with reference to 1 mm2 area of leaf surface using ArcGIS (ArcMap 10.4 © 2015 ESRI). All epidermal protrusions of the leaves were classified as hairs/trichomes. The micrographs were loaded into ArcGIS as .jpg (image) files and an interactive supervised classification was performed by assigning twenty training zones on each character (grooves, ridges and back ground) to produce a classified image. Five hundred random points were allocated using the create accuracy assessments point tool and the number of those that fell on areas of each character was recorded. This random sampling procedure was repeated five times on each classified image and the mean number of random points that fell on each character (ridges and grooves) on each image was expressed as a percentage out of 500 total points.

The mean density of leaf hairs/trichomes, stomata, grooves and ridges were then calculated taking the mean densities of each of these characters in their respective micrographs. Ten of the leaves of each species used to assess PM levels (Section 2.3) were taken at random and used in this experiment. Any correlation between these leaf characters and their PM densities were then identified using GLMM; as these characters came from twenty species, plant species was included as a random factor in this model. As observed characters were not evenly distributed on the adaxial and abaxial surfaces, they were analysed separately.

2.6 Analyzing the elemental composition of PM captured on leaves

Elemental analysis of captured PM was conducted whilst scanning the leaves using the ESEM for their PM densities described in Section 2.3 and 2.4. Once the particles were clearly focused at 450x and 1000x magnifications at 15 Kv accelerating voltage, the areas being scanned were acquired in Integrated Calibration and Application (INCA) microanalysis software coupled with the ESEM. The elemental composition of ten random particles from each PM size fraction per species (30 particles per plant species in total) were analysed using EDX (Weerakkody *et al.*, 2017). The Point and ID analyser was used on selected particles to identify the elements and quantify their amount as a percentage of the total weight of the particle (Wt%) minimizing interference from the background leaf material.

**3. Results**

3.1 Inter-species variation in PM capture

Considering all the species of plants, on average, 122.08 ± 6.9 x 107 of PM1, 8.24 ± 0.72 x 107 of PM2.5 and 4.45 ± 0.33 x 107 of PM10 were estimated to have been captured on 100 cm2 of the living wall. However, there was a significant inter-species variation in both PM densities (p <0.0001 for all PM sizes and F = 34.69, 41.33 and 59.31 for PM1, PM2.5 and PM10 respectively) (Table 2) and in their overall ability to capture and retain PM when LAI was incorporated (Table 3 and Fig. 2) (p <0.0001 for all PM sizes and F = 62.83, 59.31 and 32.11 for PM1, PM2.5 and PM10 respectively). The separate analysis conducted on adaxial and abaxial surfaces of the leaves also revealed a significant variation in PM densities between species (on adaxial surfaces: p <0.0001 for all PM sizes and F = 16.64, 24.9 and 20.94 for PM1, PM2.5 and PM10 respectively, and on abaxial surfaces: p <0.0001 for all PM sizes and F = 16.75, 41.73 and 12.82 for PM1, PM2.5 and PM10 respectively) (Fig. 3). *J. chinensis* showed the highest PM densities on its leaves (Table 2) and the highest overall PM capture levels (incorporating LAI) for all particle size fractions (Fig. 2). Both the PM density on leaves and the overall PM capture of *J. chinensis* were significantly higher than all the other species of plants for all PM sizes (p< 0.0001). The second highest densities of PM2.5 and PM10 were found on leaves of *V. vernicosa* and the second highest density of PM1 was found on leaves of *B. buxifolia,* which was not significantly different from the third highest density of PM1 found on leaves of *V. vernicosa* (Table 2)*.* Once the LAI was taken into account (Fig. 2), the second highest ability to remove PM was shown by *V. vernicosa* for all particle sizes.Leaves of *P. amplexicaulis* showed the lowest density of PM1 and PM2.5 and *B. cordifolia* showed the lowest density of PM10 (Table 2). Once the LAI was taken into account, the lowest ability to remove PM1 and PM10 was shown by *L. kamtschatica* whilst *P. amplexicaulis* remained the lowest for capturing PM2.5 (Fig. 2).

Table 2. Mean PM densities ± 1SE on leaves of plant species in the experimental living wall system on Leek Road, Stoke-on-Trent b

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Species | Mean PM1 density on leaves (x 103) ± SE | Group assigned by Tukey’s HSD test | Mean PM2.5 density on leaves  (x 103) ± SE | Group assigned by Tukey’s HSD test | Mean PM10 density on leaves  (x 103) ± SE | Group assigned by Tukey’s HSD test |
| *A. gramineus* | 46.54 ± 2.96 | def | 1.99 ± 0.21 | ef | 1.44 ± 0.19 | de |
| *B. buxifolia* | 101.08 ± 7.34 | b | 6.25 ± 0.43 | bc | 2.44 ± 0.21 | cd |
| *B. cordifolia* | 23.14 ± 2.42 | fg | 1.04 ± 0.09 | f | 0.45 ± 0.07 | e |
| *B. ×media* | 76.86 ± 6.10 | bc | 5.34 ± 0.46 | bc | 3.10 ± 0.20 | bc |
| *C. caryophyllea* | 66.23 ± 5.75 | cd | 1.90 ± 0.19 | ef | 1.91 ± 0.19 | cde |
| *D. cespitosa* | 46.81 ± 3.79 | def | 2.08 ± 0.22 | ef | 1.79 ± 0.21 | cde |
| *G. macrorrhizum* | 51.81 ± 4.82 | cde | 2.04 ± 0.24 | ef | 0.70 ± 0.05 | e |
| *G. renardii* | 48.45 ± 5.21 | def | 2.02 ± 0.33 | ef | 1.38 ± 0.20 | de |
| *H. americana* | 39.39 ± 2.84 | efg | 1.66 ± 0.12 | ef | 1.51 ± 0.27 | de |
| *H. villosa* | 40.48 ± 2.61 | defg | 1.88 ± 0.30 | ef | 0.64 ± 0.07 | e |
| *H. × sternii* | 36.05 ± 5.62 | efg | 1.55 ± 0.10 | ef | 0.82 ± 0.09 | e |
| *V. vernicosa* | 96.35 ± 8.26 | b | 7.29 ± 1.12 | b | 4.07 ± 0.61 | b |
| *J. chinensis* | 143.90 ± 13.23 | a | 14.94 ± 1.60 | a | 6.14 ± 0.84 | a |
| *L. kamtschatica* | 18.77 ± 2.58 | g | 1.62 ± 0.17 | ef | 0.63 ± 0.03 | e |
| *P. amplexicaulis* | 16.49 ± 2.29 | g | 0.60 ± 0.04 | f | 0.60 ± 0.05 | e |
| *P. scolopendrium* | 35.04 ± 3.72 | efg | 1.38 ± 0.09 | ef | 1.65 ± 0.13 | cde |
| *S. betulifolia* | 33.80 ± 2.77 | efg | 2.51 ± 0.52 | def | 2.61 ± 0.50 | bcd |
| *S. japonica* | 52.77 ± 4.04 | cde | 5.03 ± 0.39 | bcd | 1.72 ± 0.18 | cde |
| *S. byzantina* | 51.59 ± 6.73 | cde | 2.62 ± 0.23 | def | 1.90 ± 0.24 | cde |
| *T. vulgaris* | 66.29 ± 6.05 | cd | 3.84 ± 0.16 | cde | 3.04 ± 0.25 | bc |
|  |  |  |  |  |  |  |

b PM density on leaves of species assigned with the same letter in each PM size range, were not significantly different within their respective PM size category (Tukey’s HSD post hoc test, P > 0.05).

Themean PM densities on the adaxial surfaces of the leaves were higher than those on the abaxial surfaces in all the species of plants apart from a slightly higher density of PM1 on the abaxial surface of *B. cordifolia* which was not significantly different (p =0.48) (Fig. 3). The mean PM density on the cylindrical surface of *J. chinensis* was significantly higher compared to both leaf surfaces of all the other species (Fig. 3). On the adaxial surfaces of the leaves, the second highest mean densities of all PM sizes were found on *V. vernicosa* and on the abaxial surfaces, the second highest density of PM10 was also found on *V. vernicosa.* On the abaxial surfaces, the second highest mean density of PM2.5 was found on *S. japonica* and PM1 on *B. buxifolia*. Leaves of *P. amplexicaulis* showed the lowest PM1 and PM 2.5 densities on the adaxial surfaces of the leaves whilst *B.cordifolia* showed the lowest mean density of PM10. On the abaxial surfaces, the lowest mean densities of all PM sizes were found on *P. amplexicaulis*.

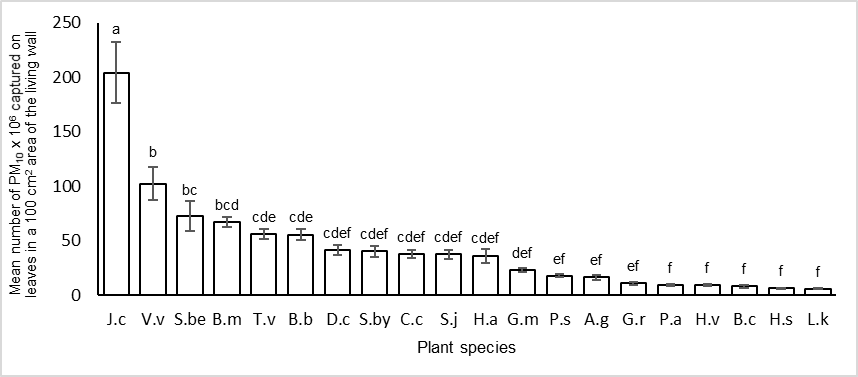
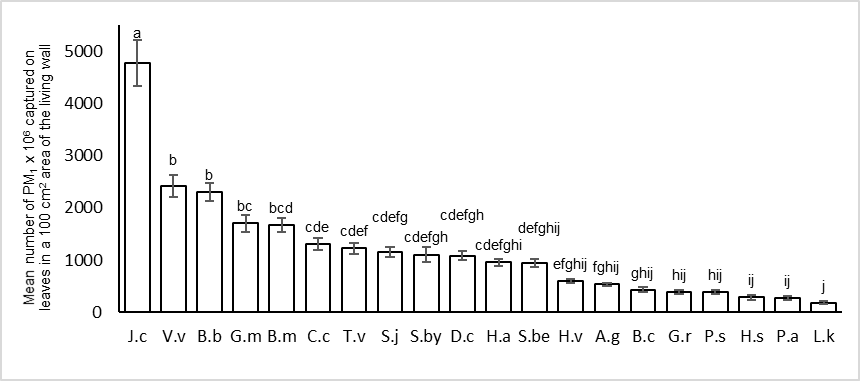
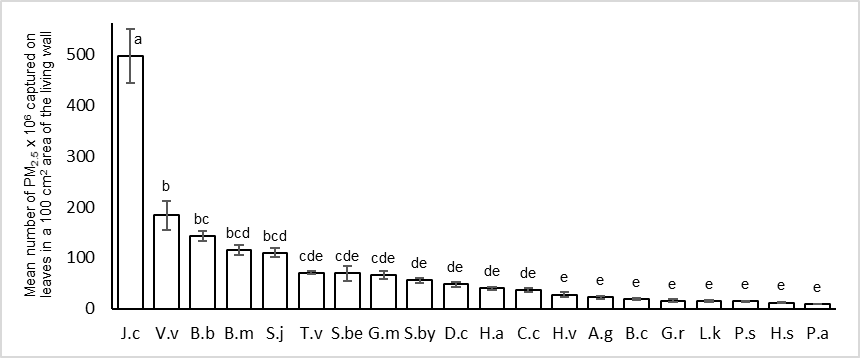
3.2 Correlation between leaf micromorphology and PM densities

The densities of micro-morphological characters varied on leaves of different species of plants and between adaxial and abaxial surfaces of the same species (Table 3 and Fig. 4). The density of PM1 did not show any significant relationship between any of the leaf characters examined (hair/trichomes, stomata, grooves and ridges) on any of the leaf surfaces. Of the characters examined, the stomatal density on the adaxial surfaces of the leaves showed a positive relationship with densities of PM2.5 (t = 38.99 and p = 0.0344) and PM10 (t = 62.72 and p <0.001). Leaf hair/trichomes showed a positive relationship with the density of PM10 (t = 3.27 and p = 0.0013). PM densities on adaxial surfaces did not show any significant relationship with ridges or grooves. Whereas on the abaxial surfaces, out of all the characters, only ridges showed a positive relationship with the density of PM2.5 (t = 2.99 and p =0.0031). The density of PM10 on abaxial surfaces was only correlated with stomatal density (t = 2.39 and p = 0.0179) but not for any of the other characters.

Deschamphia

Table 3. Mean leaf size ±1SE, LAI ±1SE and mean quantities ±1SE of micro-morphological characters of the leaves of plant species used in the experimental living wall located near Leek Road, Stoke-on-Trent

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Species | Leaf area±SE (cm2) | LAI±SE | Micromorphology on adaxial surface of the leaves ±SE | | | | Micromorphology on abaxial surface of the leaves ±SE | | | |
| Stomatal density (mm-2) | Density of Hair(mm-2) | Grooves% | Ridges % | Stomatal density (mm-2) | Density of Hair (mm-2) | Grooves% | Ridges% |
| *A. gramineus* | 5.5 ± 0.16 | 1.1 ± 0.01 | 78.8 ± 12.46 | 0 | 40.9 ± 1.0 | 43.7 ± 1.7 | 95.1 ± 4.76 | 0 | 35.8 ± 2.4 | 41.2 ± 1.8 |
| *B. buxifolia* | 1.5 ± 0.08 | 2.3 ± 0.03 | 0 | 0 | 7.6 ± 1.7 | 11.8 ± 2.5 | 207.3 ±13.17 | 0 | 10.9 ± 0.9 | 14.4 ± 1.0 |
| *B. cordifolia* | 69.8 ± 0.94 | 1.9 ± 0.12 | 0 | 0 | 0.3 ± 0.1 | 12.1 ± 0.8 | 53.7 ± 6.81 | 0 | 7.7 ± 1.0 | 11.3 ± 1.2 |
| *B. ×media* | 1.3 ± 0.08 | 2.2 ± 0.04 | 0 | 0 | 14.0 ± 0.5 | 24.9 ± 2.0 | 191.5 ± 35.72 | 0 | 22.7 ± 1.8 | 19.9 ± 2.5 |
| *C. caryophyllea* | 6.8 ± 0.13 | 2.0 ± 0.07 | 24.3 ± 6.70 | 23.0 ± 8.56 | 46.7 ± 2.8 | 50.9 ± 0.9 | 180.5 ± 48.76 | 4.2 ± 1.86 | 48.1 ± 1.7 | 48.6 ± 1.0 |
| *D. cespitosa* | 6.6 ± 0.12 | 2.3 ± 0.08 | 54.5 ± 7.62 | 75.9 ± 25.23 | 37.9 ± 1.1 | 40.5 ± 2.0 | 112.9 ± 5.37 | 34.0 ± 11.33 | 30.8 ± 2.4 | 36.2 ± 1.8 |
| *G. macrorrhizum* | 30.4 ± 2.03 | 3.3 ± 0.13 | 0 | 137.3 ± 23.39 | 31.4 ± 3.7 | 26.5 ± 2.5 | 238.7 ± 9.51 | 142.3 ± 20.48 | 26.2 ± 2.4 | 25.2 ± 2.6 |
| *G. renardii* | 7.1 ± 0.13 | 0.8 ± 0.06 | 0 | 4.6 ± 0.91 | 38.4 ± 2.9 | 40.1 ± 2.8 | 297.1 ± 73.69 | 56.7 ± 13.39 | 25.7 ± 2.8 | 25.8 ± 2.3 |
| *H. americana* | 34.7 ± 1.87 | 2.4 ± 0.10 | 0 | 5.2 ± 0.55 | 14.8 ± 2.0 | 9.9 ± 1.2 | 132.3 ± 6.25 | 27.2 ± 3.42 | 11.7 ± 1.7 | 14.4 ± 2.3 |
| *H. villosa* | 59.9 ± 1.67 | 1.5 ± 0.17 | 0 | 58.1 ± 12.52 | 21.8 ± 2.5 | 15.3 ± 2.0 | 147.6 ± 15.21 | 56.3 ± 10.51 | 16.7 ± 1.4 | 25.1 ± 2.7 |
| *H. × sternii* | 20.0 ± 0.57 | 0.8 ± 0.11 | 0 | 0 | 29.6 ± 1.9 | 37.4 ± 2.4 | 121.6 ± 7.64 | 0 | 5.9 ± 1.0 | 5.3 ± 0.7 |
| *J. chinensis* | 0.1 ± 0.001 | 3.3 ± 0.04 | 68.9 ± 15.58 | 0 | 27.3 ± 2.0 | 30.9 ± 2.6 | 73.3 ± 16.27 | 0 | 27.3 ± 2.0 | 30.9 ± 2.6 |
| *L. kamtschatica* | 12.8 ± 0.94 | 1.0 ± 0.04 | 0 | 37.2 ± 2.27 | 31.1 ± 3.0 | 26.6 ± 1.9 | 186.0 ± 14.97 | 25.2 ± 5.81 | 22.8 ± 2.1 | 28.6 ± 2.6 |
| *P. amplexicaulis* | 69.9 ± 0.25 | 1.6 ± 0.12 | 0 | 0 | 45.5 ± 1.7 | 46.6 ± 2.2 | 117.7 ± 7.32 | 10.1 ± 1.39 | 25.1 ± 2.3 | 27.8 ± 2.9 |
| *P. scolopendrium* | 19.4 ± 1.44 | 1.1 ± 0.06 | 0 | 5.3 ± 2.01 | 23.9 ± 2.2 | 24.5 ± 2.3 | 19.6 ± 3.04 | 0 | 27.6 ± 2.2 | 26.9 ± 2.0 |
| *S. betulifolia* | 3.8 ± 0.10 | 2.8 ± 0.19 | 0 | 0 | 26.2 ± 2.8 | 19.6 ± 1.8 | 286.4 ± 31.82 | 0 | 18.4 ± 1.2 | 22.5 ± 2.3 |
| *S. japonica* | 1.9 ± 0.11 | 2.2 ± 0.02 | 0 | 0 | 30.4 ± 2.1 | 27.9 ± 1.8 | 218.2±21.13 | 0 | 28.1 ± 2.6 | 31.7 ± 2.3 |
| *S. byzantina* | 14.1 ± 1.10 | 2.1 ± 0.06 | 0 | 393.9 ± 35.01 | 0 | 0 | 178.7 ± 41.28 | 323.6 ± 37.97 | 12.1 ± 1.3 | 16.2 ± 1.7 |
| *T. vulgaris* | 0.1 ± 001 | 1.8 ± 0.05 | 0 | 814.6 ± 107.2 | 28.1 ± 2.4 | 31.4 ± 1.7 | 457.3 ± 29.94 | 667.2 ± 52.05 | 27.8 ± 2.0 | 29.3 ± 1.7 |
| *V. vernicosa* | 0.1 ± 0.002 | 2.5 ± 0.02 | 89.0 ± 6.81 | 4.2 ±1.58 | 11.1 ± 1.6 | 12.6 ± 1.4 | 227.1 ± 14.14 | 0 | 14.5 ± 1.6 | 11.1 ± 1.6 |



a)

b)

c)

Fig. 2: Estimated Mean number ±1 SE (x 106) of PM captured on leaves in a 100 cm2 area of living wall (incorporating the LAI) by different species of plants on the experimental living wall on Leek Road, Stoke-on-Trent: a) PM1 b) PM2.5 and c) PM10. Data are arranged in descending order of PM accumulation for each size range, for an explanation of the species codes see further Table 1. Note different scales on the y-axes.\*Species marked with same letters were not significantly different within each particle size category (Tukey’s HSD post hoc test, P > 0.05).

3.3 Elemental composition of the captured particles

Twenty-eight different elements Ag, Al, Ba, C, O, N, Br, Ca, Cl, Cr, Co, Cu, F, Fe, K, Mg, Mn, Na, P, S, Sb, Si, Sn, Ti, Tl, V, Zn and Zr were found, in various quantities, in PM accumulated on leaves (Fig. 5). The amounts of C and O were considerably larger compared to other elements and they were thus excluded from the graphs to improve the clarity. Out of all the elements Co, Br and Tl were found only in PM1 and N, Sb and V were only found in PM10. Leaves of all the species carried PM with Ca, K, Mg and S in all particle sizes. In addition, Fe and Cl were present in PM1 captured on leaves of all the species whilst Si was found in PM10 captured on all the species. In terms of quantity, the highest Wt% was shown by Fe (13.16%) in all particle size fractions (apart from C: 48.43% and O: 32.62%).

**4. Discussion**

4.1 Inter-species variation PM capture by leaves

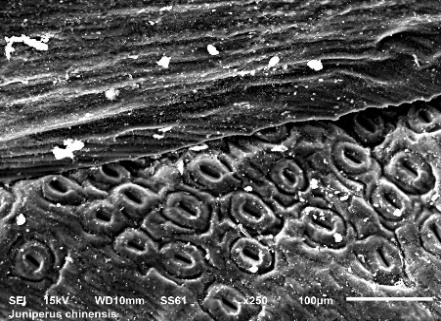
All the plant species on the living wall located in Staffordshire University along Leek Road showed considerable levels of PM capture and retention ability. In agreement with the results of Freer-Smith *et al.* (2005), Ottele *et al.* (2010) and Weerakkody *et al.* (2017), the total amount of particles captured on leaves increased with decreasing particle diameter. At the species level, although the majority of the species followed the same pattern, the amount of PM10 on *C. caryophyllea*, *P. amplexicaulis, P. scolopendrium, S. betulifolia*  were slightly higher or equal to their PM2.5 levels (Fig. 2). These varied deposition levels of particles with different aerodynamic diameter probably reflects the different aerodynamics properties of the particles and their interactions with different leaf characteristics (see Davidson and Wu, 1990, Petroff *et al.*, 2008 and Slinn, 1982; Weerakkody et al., 2017). We found a considerable variation in PM densities (i.e. excluding LAI) and in overall PM accumulation (i.e. including LAI) on different species, as has been found previously for various types of green infrastructure (Blanusa *et al*., 2015; Beckett *et al*., 2000; Freer-Smith *et al*., 2004; Song *et al*., 2015). The inter-species variation in total PM density and in density on the adaxial and abaxial surfaces of leaves reflect the influence of species-specific leaf characteristics on PM accumulation (Weerakkody et al., 2017).

b)

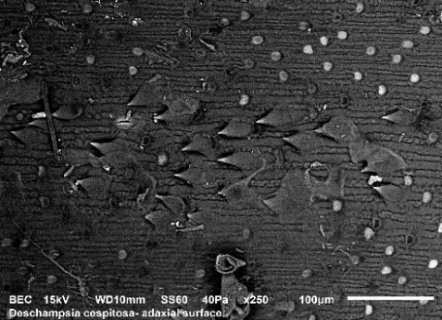
a)

c)

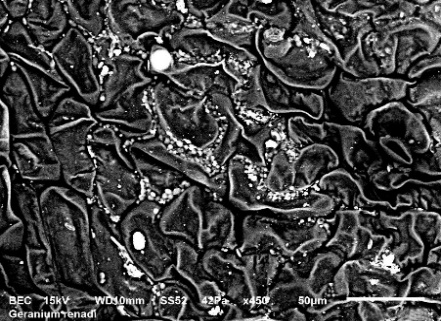
Fig. 3Mean PM density ±1 SE on 1 mm2 of the adaxial and abaxial surfaces of the leaves of plant species on the experimental living wall located on Leek Road, Stoke-on-Trent (PM densities are given in different scales): a) PM1 b) PM2.5 and c) PM10.  Plant species marked with same letters are not significantly different from each other (p > 0.05). **Upper case letters** - Variation between adaxial surfaces of the leaves; **lower case letters** - variations between abaxial surfaces of the leaves. Note different scales on the y-axes.**\*** indicates that the PM densities on adaxial and abaxial surfaces were significantly different. For explanation of the species codes see further Table 1.



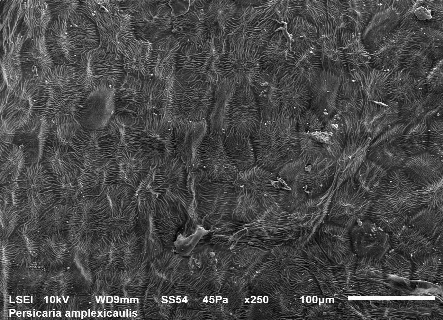
a



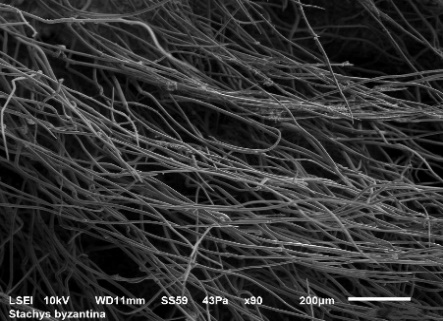
b



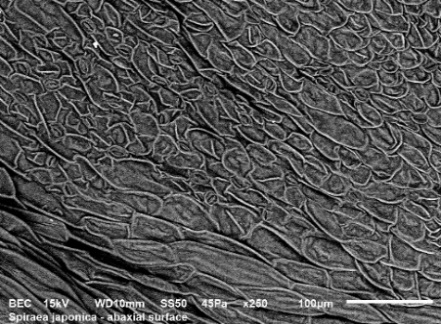
ca



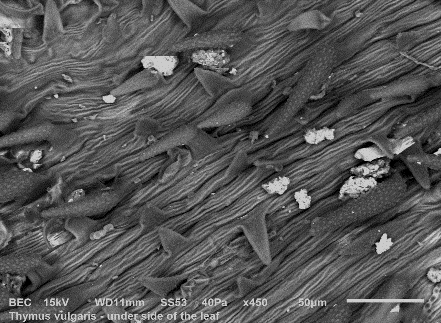
d



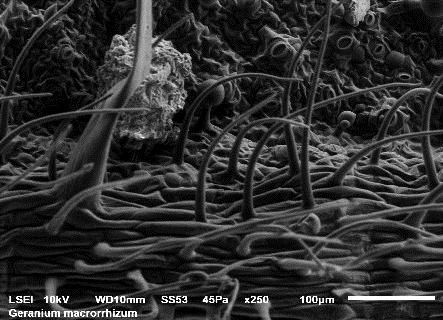
e



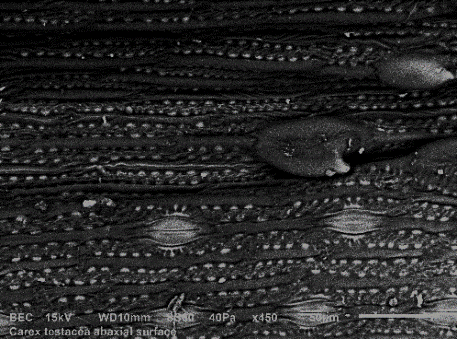
f



g



h



i

Fig. 4. ESEM micrographs showing micro-morphological characteristics on leaves of some plant species in the experimental living wall located on Leek Road, Stoke-on-Trent: a) leaf needles of *J. chinensis* (x250), adaxial surfaces of b) *D. cespitosa* (x100)c) *G. renardii* (x450) d) *P. amplexicaulis* (x250) e) *S. byzantine* (x90) and abaxial surfaces of f) *S.japonica* (x250) g) *T. vulgaris* (x450) *h) G. macrorrhizum* (x250) i) *C. caryophyllea* (x450).

Evergreen conifers have frequently been recognized for more efficient PM capture compared to other species in tree- and canopy-based studies (Beckett *et al*., 2000; Chen *et al*., 2017; Freer-Smith *et al*., 2005; Tallis *et al*., 2011); that the highest PM density found in this study was on the leaf needles of *J. chinensis* confirms the value of species with needle/needle-like leaves in living walls and probably in other vertical greenery systems. The relatively high LAI (Table 3) of conifers as demonstrated here and Gower et al. (1995) further increases their PM removal potential (Fig. 2) suggesting their greater suitability as PM filters. Larger densities of all PM sizes were evident on leaves of all the smaller-leaved species - *V. vernicosa*, *B. buxifolia*, *B. x media*, *T. vulgaris* and *S. japonica* - and confirmed their high PM filtering potential as suggested in Weerakkody *et al.* (2017) with reference PM generated from rail-traffic (Table 3). LAI values of these species were also high and enhanced their higher ability to remove PM.

a)

b)

c)

Fig. 5. Quantity of different elements as their percentage weight (Wt%) in captured PM on leaves of plant species on the living wall located on Leek Road, Stoke-on-Trent: a) PM1 b) PM2.5 c) PM10. For explanation of the species codes see further Table 1. Note different scales on the y-axes.

Supporting the findings of Beckett *et al.* (2000), Hwang *et al*. (2011) and Weerakkody *et al.* (2017), most of the wide-leaved species - *P. amplexicaulis*, *B. cordifolia*, *L. kamtschatica*, *P. scolopendrium*, *H. sternii*, *H. americana* and *H. villosa* - showed comparatively lower densities in PM capture; however, Sæbø *et al*. (2012) did not find a correlation between leaf size and PM accumulation. This study indicates that individual leaf size is an important variable causing inter-species differences in PM capture and retention as higher PM accumulation occurred on smaller leaves and lower PM accumulation on wide leaves. Freer-Smith *et al.* (2005) and Leonard *et al.* (2016) made similar conclusions about the higher PM acquisition ability of smaller leaves. The differences in opinion on the impact of leaf size evident in the literature can possibly be attributed to the influence of other variables such as leaf shape and surface micromorphology exhibited by different species of plants (Weerakkody *et al.,* 2017).

Leaf hair/trichomes were identified as potentially helping in the accumulation of particulates on leaves by several authors (Beckett *et al.*, 2000; Räsänen *et al.*, 2013; Weber *et al.*, 2014; Weerakkody *et al.*, 2017; Weerakkody et al., in press) presumably through reducing re-suspension and increasing the effective surface area (Qiu *et al.*, 2009). Suggesting a similar effect relatively high PM densities were found on hairy *S. byzantina* and *G. macrorrhizum* (Fig. 4) compared to other wide-leaved species. However, in contrast to Sæbø *et al.* (2012), there was no significant correlation between the density of leaf hair/trichomes with the density of PM captured - apart from with PM10 on adaxial surfaces. Perini *et al.* (2017) also made similar conclusions pointing out that there was no impact of leaf hairs on PM accumulation. Species-specific differences in the nature of hairs (size, length, texture, hydrophobicity and secretory/non-secretory) might have influenced their ability to help in trapping PM. Other variables such as leaf size, shape and other micro-morphological features might also have enhanced or limited these abilities (e.g. PM accumulation on hairy leaves of *H. villosa* might have been limited by a larger leaf size) (Weerakkody et al., 2017). Rough leaf surfaces with dense ridges and grooves have frequently been cited as features likely to improve PM accumulation/retention (Barima *et al.*, 2014; Kardel *et al.*, 2012; Weerakkody *et al.*, 2017; Zhang *et al*., 2017). However, in this study, the density of ridges or grooves did not show any significant correlation with PM densities apart from with PM2.5 on abaxial surfaces. The diverse morphology of these characters (Fig. 4) (e.g. curved ridges forming broader deep grooves in *G. renardii*, densely arranged ridges forming narrower grooves in *P. amplexicaulis* and parallel ridges/grooves in *D. cespitosa*) might be responsible for these results with some variants of these surface characteristics being better than others in PM capture/retention. Nevertheless, irrespective of the influence of other variables, stomatal density of the leaves was found to have a positive impact on PM accumulation by showing significant correlations with both adaxial and abaxial surface PM10 densities and PM2.5 density on adaxial surfaces.

Leonard *et al.* (2016) and Weerakkody *et al.* (2017) found relatively low PM accumulation on linear leaves. Of the four species with linear leaves used in this study, *J. chinensis* showed the highest PM accumulation whereas *D. cespitosa*, *C. caryophyllea*, and *A. gramineus* showed moderate PM densities, which were higher than most of the wide-leaved species and lower than most of the smaller-leaved species. Differences in their leaf size, micro-morphology, surface texture and flexibility probably explains this variability. The smaller, rigid, leaf needles of *J. chinensis* and the grass and ‘grass-like’ leaves of *D. cespitosa*, *C. caryophyllea*, and *A. gramineus* probably create variable drag forces with wind currents (Gemba, 2007) resulting in differential turbulence around their leaves (Davidson and Wu, 1990). The readily-bending nature of elongated flexible grass-like leaves probably creates less turbulence compared to needles, resulting in increased PM accumulation on the latter.

The higher PM accumulation on adaxial leaf surfaces, compared to abaxial surfaces, reflects previous findings of Ottelé *et al.* (2010), Ram *et al*. (2012) and Weerakkody *et al.* (2017). Leaf orientation on vertical greenery systems is such that PM exposure on abaxial surfaces, which were facing the wall, can be much lower compared to adaxial surfaces facing the road. Therefore, lower densities of all PM sizes on abaxial surfaces could mainly be attributed to their reduced exposure level. Nevertheless, significant differences in PM accumulation on adaxial and abaxial leaves became less prevalent with decreasing particle diameter such that 17 out of 19 species had significantly higher levels of PM10 on their adaxial surfaces compared with 14 out of 19 species for PM2.5 and 8 out of 19 species for PM1 (Fig. 3). Particles deposited via sedimentation under gravity are most likely to be affected by leaf orientation, and this effect is greatest for particles of larger diameter (Slinn, 1982); these factors may explain the large differences in PM10 deposition.

4.2 Elemental composition

There was a close similarity of the elemental composition of PM found on leaves taken from the Leek Road living wall when compared with the elemental composition of known traffic-generated particles (Dong *et al*., 2017; Lawrence *et al*., 2013; Lin *et al*., 2005; Sharma *et al*., 2005). This similarity indicates that, as expected, a considerable proportion of PM filtered by the plants on the living wall were likely to be generated from road traffic. Nevertheless, we did not detect Pb, or Ni in these PM, which were typically present in samples reported in the literature; this is probably due to changes in the composition of fuel additives (Lawrence *et al.*, 2013) e.g. Pb was banned in petrol in the UK in 2000 (Farmer *et al*., 2002).

In addition to the influence of surrounding plant material, higher levels of organic and elemental C in vehicle exhaust (Kleeman *et al.*, 2000; Sharma *et al.*, 2005; Zhang *et al.*, 2017) might explain elevated C masses in PM in all size ranges. High C and O levels can also indicate presence of human carcinogenic polycyclic aromatic hydrocarbons (PAHs), one of the main toxic components of PM released from road traffic (mainly from fuel exhaust (Velali *et al.*, 2016; Zhang *et al.*, 2017) and tyre wear (Boonyatumanond *et al.*, 2007). The high abundance of Fe is typical in traffic-generated PM as it can be derived from both exhaust and non-exhaust emissions. Both petrol and diesel exhaust PM are known to contain a considerable amount of Fe (Lin *et al.*, 2005; Manoli *et al.*, 2002; Wang *et al.*, 2003) and it is a predominant component in PM release from brake wear, tyre wear, road dust, and abrasives (e.g. brake linings) (Birmili *et al.* 2006; Cadle *et al.*, 1997; Cheng *et al.*, 2010; Pio *et al.*, 2013; Thorpe and Harrison, 2008). Trace quanitites of Cu, Ca, Ba, Mn, Ag, Co, Cr, Sb, Sr, Ti, V, and Zn can also be present in vehicle exhaust as functional groups of organic components (Chan, 2002; Lin *et al.*, 2005; Sharma *et al.*, 2005). In addition, Al, Ba, Ca, Co, Cr, Cu, K, Mg, Mn, Na, Sb, Sr and Zn can be attributed to non-exhaust particles released from brake and tyre wear (Pio *et al.*, 2013; Thorpe and Harrison, 2008; Weckwerth, 2001). Fine particles are mostly anthropogenic in origin and carry a variety of heavy metals which are toxic to humans and some are even carcinogens (e.g. Cr, Co, Cu, Mn) (Manalis *et al.*, 2005). Iron- rich particulates, falling in the range of PM1, are particularly hazardous due to their ability to cause oxidative brain damage potentially leading to important disease conditions such as Alzheimer’s and Parkinson’s (Maher *et al.*, 2013). In additon to PM originating from road traffic, PM10 containing Al, Ca, Na, Si, Cl, F, and N can originate from soil dust (Maher *et al.*, 2013). The presence of Tl (Thallium) has not been cited as a traffic-derived element in previous research and cement dust can be a potential source of PM1 containing Tl (Brockhaus *et al*., 1981).

4.2 Best species composition to use in living walls to filter near-road PM

Based on the outcome of this study, the use of coniferous species with leaf needles and smaller-leaved species with large LAIs can enhance the efficiency of PM filtering by living walls. Although stomatal density showed a higher impact on accumulating PM compared to other micro-morphological characters stomatal density can vary within a species depending on the environmental conditions (Beerling and Chaloner, 1993), and thus can be less useful to use as a direct indicator in species selection without a closer examination of the impact of local climate and growing conditions. Based on specific observations in the present study and previous findings, species with hairy or rough leaves can still be useful in PM capture; avoiding species with larger leaves and smaller LAI would potentially enhance living wall performance.

**Conclusion**

Leaves of plant species on a living wall located along Leek Road, Stoke-on-Trent accumulated substantial amounts of PM1, PM2.5 and PM10, demonstrating their potential to reduce PM pollutants in the atmosphere. Different plant species showed a varied potential to capture and retain particles; the highest PM levels found in leaf-needles of *J. chinenisis* were considerably higher in all particle size fractions. PM accumulation on all smaller-leaved species used in this study were considerably higher compared to most of the larger leaved species irrespective of their micro-morphological features. Accumulation of PM1 did not show any significant correlation with the leaf micro-morphological features (leaf hair/trichomes, stomatal density, surface ridges and grooves) examined. Although there was no apparent pattern valid for all particle sizes, PM2.5 and PM10 showed significant correlations with some of the micro-morphological features on at least one of the leaf surfaces (adaxial or abaxial). Of these characteristics, stomatal density showed more influence on PM2.5 and PM10 accumulation compared to other characters. Irrespective of the absence of statistical correlations when all the species were considered, higher levels of PM on leaves of *S*. byzantina and *G. macrorrhizum* suggested some positive influence of leaf hairs in PM accumulation. Morphological variation within each character probably had an impact on their influence in PM accumulation (i.e. length and shape of hair, depth and shape of ridges or grooves). Elemental composition of accumulated particles showed a strong similarity with chemical composition of traffic-generated particles.

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