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Green wall technology for sustainably improving environmental quality:

Investigations into green wall plant health and particulate deposition

A thesis submitted by Naomi Paull to the School of Life Sciences,

University of Technology Sydney, in partial fulfilment of the requirements of PhD

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Statement of Original Authorship

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Abstract

Air pollution is of significant concern, affecting millions of people globally. Plants are effective air pollution remediators; certain species, however, may exhibit higher removal capacities. Additionally, due to the continual pollution exposure, some species may exhibit sensitivity to pollution and will thus be ineffective for use in *in situ* applications.

This thesis assessed the particulate matter removal capacity of common green wall species used in *in situ* applications over a 6 month duration. High accumulating species were then identified, and leaf traits associated with enhanced particulate matter accumulation assessed. Leaf traits were not found to be exclusively related to enhanced particulate matter deposition; with small linear leaved species exhibiting the lowest particulate accumulation. The health of the green wall species from pollution exposure was then assessed. Most species did not encounter any significant differences among their health variables between polluted test sites and control glass house conditions, indicating their suitability for use *in situ.* The particulate matter removal capacity of *in situ* Sydney green walls was then examined. To do this, air quality tests were conducted in front of green walls and matched reference walls across the test sites. There were no significant differences observed for ambient particulate matter concentrations between green wall and reference wall sites, perhaps due to the 'passive' nature of the green wall systems tested. There was also no significant difference observed between the wall types for proximal temperature conditions, but there was a significant difference for ambient noise reduction, with green walls having significantly lower noise conditions. Lastly, the pollutant removal capacity of Australian native species used in active green walls was assessed. Active native green walls were effective at reducing benzene, with similar removal efficiencies to previously tested ornamental species. They were also capable of removing particulate matter,

however at lower efficiencies than ornamental species. Native plant active green walls were inefficient for carbon dioxide removal.

The results of this thesis highlight the importance of species selection for maximum pollutant removal efficiency and the capacity for vegetation to have positive impacts on ambient conditions. The results also indicate improvements that can be made to green wall systems for a higher efficiency for *in situ* applications, including the conversion of passive systems to active systems and the inclusion of select species for increased removal efficiency and tolerance to pollution exposure.

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List of Abbreviations

Analysis of variance	ANOVA
Carbon dioxide	CO_2
Carbon monoxide	CO
Crassulacean acid metabolism	CAM
Hydrogen peroxide	H_2O_2
International agency for research on cancer	IARC
Linear mixed models	LMM
Methane	CH_4
Nitrogen dioxide	NO_2
Organisation for economic cooperation and development	OECD
Oxides of nitrogen	NO_x
Oxides of sulphur	SO_x
Ozone	O_3
Particulate matter	PM
Particulates less than 10 micrometres in size	PM_{10}
Particulates less than 2.5 micrometres	$PM_{2.5}$
Particulates less than 0.1 micrometres	$PM_{0.1}$
Photosystem one	PSI
Photosystem two	PSII
Polychlorinated biphenyl	PCB
Polychlorinated dibenzodioxins	PCDD
Polycyclic aromatic hydrocarbons	PAH
Principal components analysis	PCA
Reactive oxygen species	ROS
Relative water content	RWC
Ribulose bisphosphate	RuBP
Scanning electron microscope	SEM
Single pass removal efficiency	SPRE
Standard error of the mean	± SE or ± SEM
Statistical package for social sciences	SPSS
Sulphur dioxide	SO_2
Total suspended particulates	TSP
United States environmental protection agency	USEPA
Volatile organic compounds	VOCs
World health organization	WHO

Chapter 1 – General Introduction

1.1 Air pollution – overview

The number of people residing in urban environments is expected to increase to 66% worldwide by 2050 (United Nations, 2014), meaning a greater proportion of people will inevitably be exposed to harmful air pollution. It has been estimated that 60 – 70 % of air pollution in urban regions world-wide is due to vehicle emissions (Olukanni and Adebiyi, 2012). Pollution emitted from road traffic includes gaseous pollutants such as nitrogen oxides (NO_x), sulphur dioxide (SO_2), carbon monoxide (CO) and particulate matter ranging from PM_{10} (particulate matter ≤10 μm in aerodynamic diameter), $PM_{2.5}$ (particulate matter ≤2.5 μm) and ultra-fine particulates (UFP <100 nm) (Abhijith et al., 2017). Diesel based vehicles emit the larger proportion of atmospheric PM and NO_x, whilst gasoline and petrol driven vehicles emit mostly hydrocarbons and CO (CPCB, 2010).

Air pollution can result from both anthropogenic and natural sources, such as bush fires, volcanic eruptions and sea spray (Jyethi, 2016). In urban environments anthropogenic air pollution sources can be further defined as mobile (i.e. trucks, cars and buses) or stationary, such as industries, power plants and refineries (Kulshrestha and Saxena, 2016). Fossil fuel use from the transport and industrial sectors has led to a rise in gaseous pollutant concentrations including SO_2, NO_x, CO, carbon dioxide (CO_2) and particulate matter (PM), which are increasing with time (Agbaire and Esiefarienrhe, 2009; Rai et al., 2011). This is concerning as air pollutants are capable of travelling large distances, which can result in transboundary pollution (Oishi, 2016), meaning that harmful air pollution is almost inescapable.

Air pollution has further been associated with various forms of environmental degradation including: altered plant species composition, reduced agricultural yield and soil chemistry deviations (Thomas, 1961; Brimblecombe, 2003); long term changes in water quality such as

eutrophication and ocean acidification, changes to the Earth's radiation budget and visibility degradation in the form of hazes (Seinfeld and Pandis, 2006); and bioaccumulative, neurological, reproductive and behavioural changes in wildlife, particularly birds and fish (Welch, 1998). As well as direct effects and changes to plants, air pollution can also alter the abiotic features on which plants are dependent, including ambient humidity, soil, CO_2 and oxygen concentrations, temperature and light conditions (Stevović and Markovic, 2016). This highlights the impacts of air pollution that reach beyond human health.

1.2 Health impacts associated with air pollution exposure

Exposure to air pollution can result in varying degrees of human illness including: respiratory and reproductive disorders, hepatic and cardiovascular disorders, asthma, hay fever, and cancer (Tchounwou et al., 2012). The International Agency for Research on Cancer (IARC) has declared outdoor air pollution as a Group 1 carcinogen (IARC, 2013). Whilst short term pollution exposure can result in minor symptoms such as headaches, eye and throat irritation, nausea, allergic reactions, bronchitis and pneumonia (Jyethi, 2016); long term exposure can result in more damaging effects to the kidneys, nervous system, liver and brain as well as cardiovascular disorders and lung cancer (Kim et al., 2014).

Between 2005 and 2010, the death rate associated with outdoor air pollution exposure increased globally by 4%, by 5% in China and by a staggering 12% in India (Maurya, 2016; UNEP, 2014). The Organisation for Economic Co-operation and Development (OECD) stated that outdoor air pollution exposure is predicted to become the leading environmental cause of premature mortality globally by 2050 (OECD, 2012). In 2012 alone, it was estimated that approximately 7 million deaths were related to outdoor air pollution (WHO, 2014). Pollution exposure also has a negative impact on the economy, with a reported ~USD$ 1.7 trillion spent on health related costs associated with air pollution in 2010 (OECD, 2014).

1.3 Vegetation as an air pollution remediator

It is well known that the mitigation of air pollution is significantly aided by the presence of vegetation (Cavanagh et al., 2009). This is due to various plant processes such as detoxification, adsorption, absorption and accumulation through which the plants act as 'living filters' that do not endure acute foliar damage (Garbisu et al., 2002; Jim and Chen, 2008). One of the most widely understood concepts by which vegetation improves air quality is through generating and releasing oxygen into the atmosphere (Hill, 1971; Beckett et al., 1998). Gaseous air pollutants are reduced in the ambient environment by plants absorbing and metabolizing the pollutants into less toxic forms (Gupta and Kulshrestha, 2016). For example, SO_2 and NO_2 enter the leaf via the stomata, after which they form sulphurous and sulphuric acid and nitrous and nitric acid respectively once reacting with water; and are then further transported through the plant for use in plant metabolic processes (Nowak, 1994; Legge and Krupa, 2002; Pandey et al., 2015). Furthermore, plants are also able to adsorb PM onto their leaf surfaces, partially filtering the air by this process (Shi et al., 2017). Dry and wet deposition are the main processes in which PM is removed by vegetation (Pandey et al., 2015). However, the ability of vegetation to remove PM appears to be species specific, as different species possess varying characteristics such as epicuticular wax layers, trichomes and surface roughness (Shi et al., 2017), which appear to regulate their PM accumulation capacity.

The ability of plants to remove pollutants does not appear to be uniform across all species and plant types. For example, the intake of toxic pollutants in sensitive species usually results in visible injuries which form more quickly than in tolerant species (Sonwani et al., 2016). Pollutant sensitive species may therefore be used as bioindicator species; monitoring their stomatal pore size, foliar injury and chlorophyll content is a practical means to indicate pollutant presence (Gupta and Kulshrestha, 2016). Roadside vegetation is often used as a bioindicator and biomonitoring system for air pollution (Anyanwu and Kanu, 2006; Holt and

Miller, 2011; Esfahani et al., 2013; Miria and Khan, 2013). Tolerant species, in contrast, can in some cases be used as a pollutant removal source (Rao, 1983). In particular, air pollution can be reduced if tolerant plant species are grown in urban regions if careful species selection is used (Kaur and Nagpal, 2017).

There are, of course, existing methods for PM removal, which include: scrubbers, cyclone separators, baghouse filters, fabric filters, wet collectors, gravitational settling chambers, electrostatic precipitators and combustion, adsorption and condensation methods (Kumar and Gupta, 2016). Similarly, for removal of gaseous pollutants there are existing methods which utilise absorption, adsorption, condensation and combustion (Kumar and Gupta, 2016). These processes, however, are not always effective, parts often need frequent replacement, and all mechanical methods are expensive; potentially making plant based remediation technologies more appropriate for long term use. Phytoremediation technology has many advantages, including the capacity of plants to remove multiple pollutants simultaneously, having a relative long product lifespan, requiring only solar input as its energy source, and being recognised as a 'green', sustainable product.

1.4 Particulate matter as an air pollutant and its associated effects

Suspended PM is mainly comprised of carbonaceous matter, including both organic and inorganic components (Jyethi, 2016). PM is distinguished by its aerodynamic diameter, with ultra-fine particles having an aerodynamic diameter of $\leq$2.5 μm; fine particles being > 2.5 μm and < 10 μm and coarse particles being $\geq$ 10 μm (Ottelé et al., 2010; Jyethi, 2016). In many cases, anthropogenic PM contains heavy metals and polycyclic aromatic hydrocarbons (PAHs) which are highly toxic (Jouraeva et al., 2002; WHO, 2006; Yu et al., 2006; Uzu et al., 2010) and as a result, PM pollutants have shown to have genotoxicological impacts on both humans and plants (Rai, 2015). Regarding plant health, PM accumulation can have many negative

impacts (Nawrot et al., 2011) such as alterations to the size of stomata, the rupturing of epidermal hairs and guard cell swelling (Gupta et al., 2015a,b,c), all severely altering the foliar morphology (Gostin, 2016). Fine PM can enter the stomata, altering gaseous exchange, water retention, photosynthesis and the overall yield and growth of the plant (Tomasevic and Anicic, 2010; Rai et al., 2010).

PM deposition results in leaf blade coverage which reduces light penetration and blocks the opening of stomata, resulting in photosynthesis, respiration and growth rate reductions (Grantz et al., 2003; Dhir, 2016; Kulshrestha and Saxena, 2016). PM accumulation can also cause epicuticular wax degradation (Bermadinger et al., 1988; Sauter and Pambor, 1989) from the increased wax structure erosion rate (Huttunen, 1994); preventing transpiration, effecting photosynthesis and gas exchange (Sauter and Voß, 1986; Sauter et al., 1987); altering leaf wettability (Saneoka and Ogata, 1987) and reducing the flow of solutes within leaf cells (Bystrom et al., 1968). PM accumulation most notably reduces the amount of photosynthetically active radiation (PAR) absorbed by changing leaf optical properties, namely the surface reflectance in the visible and short-wave infrared radiation range (Eller, 1977; Hope et al., 1991; Keller and Lamprecht, 1995) as well as increasing the leaf temperature (Naidoo and Chirkoot, 2004). As a result, the reduced CO_2, light, chlorophyll content and stomatal conductance, as well as the increased stomatal resistance, negatively impacts photosynthesis (Vardaka et al., 1995; Beckett et al., 1998; Farmer, 2002; van Heerden et al., 2007; Rai et al., 2010). PM deposition into the substrate matrix can affect nutrient cycling through changes to rhizopheric fungi and bacteria (Grantz et al., 2003); with the intake of elements sourced from the deposited PM additionally reducing plant resistance to insects, frost, fungi and drought (Shanker et al., 2005).

1.5 SO_x as an air pollutant and its associated effects

Sulphur oxides (SO_x) are usually emitted from anthropogenic sources such as the combustion of sulphur-containing fuels, notably oil and coal used in electricity generation (Jyethi, 2016). Sulphur dioxide (SO_2) is the main emitted sulphur source, originating from high temperature combustion processes (Bowman, 1991), and is a key phytotoxic by-product of fossil fuel burning (Gupta, 2016). In contrast, natural sources of SO_x exist, and include volcanic eruptions and bush fire smoke (Jyethi, 2016), however these events do not occur as frequently as industrial processes. Sulphur particles are known to cause haze formation, scatter visible light, cause global cooling (IPCC, 2007) and can react in the atmosphere to cause acid rain (Jyethi, 2016). SO_2 concentrations within the air are not only source dependent, but can also be influenced by environmental factors such as humidity, temperature, atmospheric pressure and air movement (Stevović and Markovic, 2016). For example, in high humidity environments, SO_2 present in the air oxidizes and is partially converted to sulphuric or sulphurous acid (Stevović and Markovic, 2016), altering ambient concentrations.

Regarding human health impacts, SO_2 exposure can heighten pre-existing pulmonary and cardiovascular diseases in the elderly, children and asthmatics, as well as increase overall respiratory illness (Bremmer et al., 1999; Mar et al., 2000; Maynard and Ayres, 2014).

SO_2 exposure in plants has been reported to reduce crop yield and cause foliar damage (Malhotra and Hocking, 1976; Varshney et al., 1979; Winner et al., 1985). SO_2 notably has a profound impact on plant photosynthesis (Darrall, 1989; Agrawal et al., 2006; Chauhan and Joshi, 2010) due to the oxidation of chlorophyll (Shimazaki et al., 1980). Sulphur is however, important for plant metabolism as it is a key component of proteins, amino acids and some vitamins, however plant requirements for sulphur can be achieved from ambient SO_2 at low concentrations (Gupta, 2016). When SO_2 is dissolved in plant cells it forms bisulphite and

sulphite ions, and at low concentrations the plant is able to detoxify these ions, with sulphite being metabolized by the chloroplasts (Kulshrestha and Saxena, 2016). However, if the ambient SO_2 concentration reaches the plants biochemical threshold layer, the sulphite becomes toxic and cannot be detoxified before the plant suffers injury (Kulshrestha and Saxena, 2016). This results in changes to plant physiology, respiration and photosynthesis, leading to irreversible damage to pigments and tissues (Darrall, 1989; Agrawal and Verma, 1997), the acidification of the cellular pH (Liu et al., 2008, 2009), and alterations to plant growth, leading to plant death (Agrawal and Deepak, 2003; Agrawal et al., 2006; Gupta, 2016).

The presence of high levels of certain gaseous pollutants such as SO_2 usually results in stomatal closure to prevent entry into the leaf, this however restricts photosynthesis (Kulshrestha and Saxena, 2016). Conversely, in some cases SO_2 concentrations have shown to promote stomatal opening (Mansfield and Majernick, 1970), with this response appearing to differ across species (Biggs and Davis, 1980). Additionally, the duration to which the plant is exposed to SO_2 appears to be influential; with short term exposure sometimes resulting in stomatal opening, and long term exposure leading to partial closure (Abeyratne and Illeperuma, 2006). Further, pollutant combinations and leaf age also appear to influence the stomatal response to SO_2 pollution (Parshina and Rygalav, 1999), which inevitably effects plant processes such as photosynthesis.

1.6 NO_x as an air pollutant and its associated effects

The combustion of fossil fuels is a leading NO_x source, followed by biomass burning, soil respiration and lightning (Seinfeld and Pandis, 2006). In urban environments, the majority of nitrogen oxides are emitted from vehicle exhaust gases (Stević and Markovic, 2016), with NO_2 levels often increased in areas that have high traffic densities (Pleijel et al., 2004). Some sources account vehicular emissions being responsible for approximately half of all the NO_x

emissions (Seinfeld and Pandis, 2006); with power plant boilers producing ~40% (USEPA, 1999). This is concerning as NO_2 presence within the atmosphere can lead to the initiation of photochemical smog, ozone formation, acid rain (Seinfeld and Pandis, 2006) and eutrophication within water bodies (USEPA, 1998). Furthermore, human exposure to NO_x in low concentrations can cause shortness of breath, nausea, lethargy, nose, throat, lung and eye irritation; whilst high concentrations can cause fluid accumulation in the lungs and respiratory tract, decrease oxygenated body tissues and ultimately result in death (ATSDR, 2002).

Regarding plant health, toxic gases such as SO_2 and NO_x enter the leaf via the stomata and pursue the same diffusion pathway as CO_2; henceforth dissolving into the leaf cells (Kulshrestha and Saxena, 2016). Dissolved NO_x transforms into NO_2^- (nitrite ions) which are toxic at high concentrations (Kulshrestha and Saxena, 2016). NO_2 exposure thus results in the creation of acidic changes in leaf tissues which alter electron flow and photophosphorylation, resulting in a decrease in chlorophyll content (Dhir, 2016). Furthermore, NO_2 exposure can result in chloroplast membrane swelling, creating membrane injury and biochemical changes which reduces photosynthesis (Dhir, 2016). Stomatal conductance is also reduced at high concentrations of NO_2 (~100 ppb) (Dhir, 2016). Short term exposure to high concentrations, or longer term exposure to lower levels, however, can increase ribulose bisphosphate (RuBP) carboxylase activity, resulting in polyamines such as spermine and spermidine which help prevent NO_2 induced leaf damage (Dhir, 2016). Additionally, when NO_2 reacts with water within plant cells, it is converted to HNO_2 and HNO_3, which can be used in plant metabolism (Sudalai and Devaanandan, 2015; Dhir, 2016), usually for the production of organic compounds such as amino acids (Davies, 1986; Allen et al., 1988), if the NO_2 concentration is not severe. As with most air pollutants, NO_2 appears to be manageable by plants at low concentrations, but detrimental at high concentrations.

1.7 CO_2 as an air pollutant and its associated effects

The growth of urbanization has seen anthropogenic CO_2 levels increase from activities including burning fossil fuels for transportation, heating and industrial purposes (Stevović and Markovic, 2016). The global average atmospheric carbon dioxide content hit a new record high in 2018 of 407.4 ppm (Blunden and Arndt, 2019). The annual rate of increase in CO_2 levels over the last 60 years is roughly 100 times faster than previous natural increases (Blunden and Arndt, 2019), which is having drastic impacts on many environmental factors, most notably climate change.

In plants, ambient CO_2 concentrations regulate the opening and closing of stomata (Joshi and Bora, 2005; Ainsworth and Rogers, 2007). Under elevated CO_2 concentrations, plants can experience stomatal closure from changes in the turgor pressure of the guard cells (Dhir, 2016); reducing stomatal conductance (Darrall, 1989), CO_2 assimilation and fixation rates (Warren et al., 2007). However, in most cases of increased ambient CO_2, increased photosynthesis, plant productivity and growth have been recorded, with a simultaneous decrease in photorespiration (Allen, 1990). This increased photosynthesis results from the greater abundance of CO_2 available for RuBisCo activity (Dhir, 2016), and carbon assimilation (IPCC, 2007; Reddy et al., 2010). Generally, the amount of carbon fixed is greater than the amount of carbon lost under high CO_2 conditions, resulting in increased productivity and growth (Ryan, 1991).

Whilst short term increases in photosynthesis from enriched CO_2 conditions have been recorded, the carboxylation capacity of plants decreases after long term exposure, resulting in decreased photosynthetic rates (Ainsworth et al., 2004; Long et al., 2004; Aranjuelo et al., 2005, 2008). More specifically, long term exposure to high CO_2 concentrations results in a decrease in RuBisCo activity, which further reduces the energy demand per unit of fixed carbon, the overall photorespiration rate and the number of carotenoids and chlorophylls

produced (Dhir, 2016). This leads to increased respiration rates (Dhir, 2016) and as such, lowered rates of photosynthesis. Long term exposure to elevated CO_2 conditions has also been shown to alter leaf structures, such as increased leaf thickness and changes to chloroplast and cell development (Dhir, 2016).

1.8 O_3 as an air pollutant and its associated effects

Ozone is the leading cause of air pollution-associated mortality after PM exposure, and results in a global mortality burden of 0.7 million deaths / year (Anenberg et al., 2010). O_3 is a secondary pollutant, which is created from reactions between many precursor gases i.e. non-methane VOCs, CO, CH_4 and NO_x (Seinfeld and Pandis, 2012) which are produced from activities such as transport, industrial processes, biomass burning, land use changes and energy generation (Royal Society, 2008). Human induced tropospheric O_3 can affect many environmental conditions including forest productivity (Karnosky et al., 2007); ecosystem functions such as carbon storage (Sitch et al., 2007; Nikolova et al., 2010; Galant et al., 2012) and agriculture crop yield (Feng et al., 2008).

Plant exposure to O_3 can result in stomatal closure, decreasing stomatal conductance (Tiwari et al., 2006; Calatayud et al., 2007; Rai et al., 2007; Pellegrini et al., 2011a,b; 2015), and can also rupture stomatal apparatus from epidermal cell damage (Dhir, 2016). Stomatal closure is the main plant defensive mechanism against O_3 effects (Dhir, 2016). However, once O_3 enters the leaf via the stomata it is dissolved in the apoplastic fluid (Rai et al., 2016), producing reactive oxygen species (ROS) (Felzer et al., 2007), which damage nucleic acids, denature proteins, alter metabolism, stomata and chloroplasts, impedes photosynthesis (Kulshrestha and Saxena, 2016) and changes the fluidity and permeability of cells in the plasma membrane (Gupta and Kulshrestha, 2016); usually increasing the permeability of plasmalemma which results in ionic imbalances (Kaur, 2016). O_3 can produce both ROS and H_2O_2 within the leaf,

which alters cellular function resulting in cell death, reacts with cell membranes, impacts negatively on photosynthetic apparatus, alters antioxidant regulation, changes metabolic pathways and defence reactions, decreases carbon assimilation and can induce premature senescence (Booker et al., 2009; Fuhrer, 2009; Singh et al., 2014; Singh et al., 2015; Rai et al., 2016).

O_3 concentrations of 0.20 µL/L or higher are considered phytotoxic (Reich, 1983). At such high O_3 exposure, effects on photosynthetic and respiration pathways will occur (Kaur, 2016), resulting in lowered CO_2 assimilation rates (Ashmore, 2005; Fiscus et al., 2005; Rai and Agrawal, 2012; Ainsworth et al., 2012) from thylakoid structural damage, reducing photosynthetic pigments and the efficiency of solar energy capture, and by having negative effects on the electron transport system in PSI and PSII (Calatayud and Barreno, 2001; Fiscus et al., 2005), and by reducing the amount of RuBisCo (Agrawal et al., 2002). O_3 exposure can therefore result in increased dark respiration and decreased primary productivity, photosynthesis and chlorophyll content (Morgan et al., 2003; Kaur, 2016). Plant responses, however, can vary amongst species, plant age, concentration of pollutant exposure (Kaur, 2016), environmental conditions (i.e. temperature) and other types of pollution present (i.e. organic vs acid rain) (Günthardt-Goerg and Vollenweider, 2007; Vollenweider et al., 2008).

1.9 Heavy metals as air pollutants

Whilst plants require some heavy metals such as zinc, iron and copper for the biosynthesis of enzymes, growth and development (Onder and Dursun, 2006), large changes to these trace concentrations can result in substantial changes to plant production and biochemical processes (Bucher and Schenk, 2000). Processes such as gasoline and oil combustion, electroplating industries, rubber tyre wear and auto workshops, are all sources of anthropogenic heavy metals (Nadgórska-Socha et al., 2017). High levels of heavy metals can cause oxidative stress by

generating ROS within sub cellular components, or by reducing levels of enzymatic and non-enzymatic antioxidants (Benavides et al., 2005; Gupta et al., 2012). High heavy metal accumulation can result in chlorosis and leaf death (Bergman, 1983), from the metals such as copper destroying plant subcellular structures (Sresty and Madhava, 1999). Heavy metals can also decrease photosynthetic efficiency (Krupa and Baszyński, 1995; Burzynski and Klobus, 2004); excess concentrations of Pb, Cu or Cd directly inhibits photosynthetic electron transport (Krupa and Baszyński, 1995; Myśliwa-Kurdziel et al., 2002); and additionally, alters the net assimilation of CO_2 and Calvin-Benson cycle enzymes (Prasad and Strzalka, 1999; Burzynski and Klobus, 2004).

1.10 PAHs as air pollutants

PAHs such as benzo(α)pyrene are highly toxic to humans (Boström et al., 2002), and are widely distributed in the air (Bohlin et al., 2008). PAHs are organic compounds with two or more fused aromatic rings, and are emitted from incomplete combustion (Maliszewska-Kordybach, 1999). Industrial processes, vehicular exhaust, heating and power generation are all sources of PAH emissions (Mastral and Callén, 2000), with vehicular exhaust being considered one of the leading sources in urban environments (Piccardo et al., 2005). Influential traits on the formation of PAHs (i.e. whether they become particle bound or gas phase) include temperature, the presence of absorbing surfaces, and the physiochemical properties of the PAH compound (Pankow, 1987). PAHs which have 3 or 4 rings are usually associated with the vapour phase, whilst PAHs with 5 or 6 rings are usually bound to particles (Klingberg et al., 2017). PAHs often have carcinogenic and mutagenic properties, and many are very harmful to human health (Maliszewska-Kordybach, 1999; Aas et al., 2001).

PAHs can inhibit plant respiration and photosynthetic processes by altering the physiochemical properties of plant membranes (Huang et al., 1996; Duxbury et al., 1997; Tukaj and Aksmann,

2007). Specific leaf traits however, have shown to be influential in removing PAHs from the ambient environment, without causing irreversible plant damage. For example, a waxy cuticle (Simonich and Hites, 1995; Piccardo et al., 2005) as well as the presence of trichomes or leaf hairs (Howsam et al., 2000) assists in PAH capture; with larger leaf surface areas being correlated to a greater absorption of PAHs (Simonich and Hites, 1995).

1.11 Pollutant Mixtures

Pollutant mixtures have a different impact on stomatal physiology compared to single pollutants (Dhir, 2016), with plant stomatal conductance response to pollutant mixtures being considered species specific (Darrall, 1989). Additionally, stomatal resistance is thought to increase when exposed to pollutant mixtures compared to single pollutants (Noormets et al., 2001, 2010). Pollutant mixtures such as SO_2-NO_2, O_3-NO_2 and SO_2-O_3 have synergistic effects, causing severe damage to the photosynthetic mechanisms; decreasing photosynthesis, even at low concentrations (Dhir, 2016).

When exposed to ambient CO_2 conditions and O_3, there is usually an initial increase in stomatal conductance and photosynthesis but under high CO_2 conditions, the decline in chlorophyll content associated with O_3 exposure is less rapid, and photosynthesis increases with increasing CO_2 (Dhir, 2016). As such, increased ambient CO_2 plays an important role in minimizing O_3 effects on photosynthesis (Mulholland et al., 1997). It is thought that partial stomatal closure from increased ambient CO_2 conditions reduces the impact of O_3 and other pollutants, by limiting the pollutant uptake (Allen, 1990). Additionally, RuBisCo activity is increased with increased CO_2 concentrations, assisting with mitigating O_3 induced damage (Dhir, 2016). As such, since air pollution invariably consists of a variety of air pollutants with varying concentrations, it is important to consider the impact of pollutant combinations on plant health.

2.1 Green wall technology

A topical solution to urban air pollution is green infrastructure (Irga et al., 2015; Salmond et al., 2016), which includes green roofs, street trees, living walls and vegetation barriers (Abhijith et al., 2017). One major mechanism by which green infrastructure assists with air pollution mitigation is due to plants' generally 'porous' structure, which increases airborne pollution removal and deposition, as well as influencing the local pollutant dispersion patterns (Nowak et al., 2006; Escobedo and Nowak, 2009; Yin et al., 2011; Fantozzi et al., 2015; Janhäll, 2015). Similar to other forms of phytoremediation, the leaf stomata and plant surfaces act as the main site for pollutant removal via simple absorption (Escobedo and Nowak, 2009; Fantozzi et al., 2015; Salmond et al., 2016; Vesa Yli-Pelkonen et al., 2017). Green infrastructure has been recognised as a passive air pollution abatement system which requires minimal adjustments to the built environment (McNabola, 2010), making this technology both unique and desirable.

Green walls, living walls and green facades are one of the newest permutations of green infrastructure. Whilst all green wall systems commonly have plants growing on them, each system differs in its functional application. For example, direct green facades have plants growing directly attached to a wall; whereas indirect facades have plants growing on the wall via a supporting mechanism (i.e. mesh, modules, ropes or cables). 'Living walls' comparatively are a newer concept in which plants are attached to the wall along with the growing media (i.e. substrate, peat, natural fibres etc.) to support plant growth (Pérez et al., 2011, 2014; Manso and Castro-Gomes, 2015; Susorova, 2015). These various forms of green infrastructure were originally installed for aesthetics, however, currently their design is continually the subject of research so as to improve their functionality, and to maximise their contribution to the sustainable urban environment (Abhijith et al., 2017).

The presence of green infrastructure has many demonstrated benefits including: climate change mitigation (Matthews et al., 2015); noise pollution abatement (Berardi et al., 2014; Cohen et al., 2014; Salmond et al., 2016); urban heat island mitigation (Gago et al., 2013; Chen et al., 2014); increased stormwater management (Czemiel Berndtsson, 2010; Roy et al., 2012; Wurm, 2016); and a reduction in built environment energy consumption (Berardi et al., 2014; Pérez et al., 2014). As such, the implementation of green infrastructure appears to be beneficial in many regards, and not exclusive to air pollution mitigation.

2.2 The Junglefy green wall system

The current study focussed on the most commonly used green wall system in Sydney, Australia. Junglefy Pty Ltd are currently the biggest living plant wall construction company in Australia. Their green wall system (Fig. 1) is comprised of multiple small (0.25 m^2) modules which contain 16 plants per module. The modules are composed of recycled plastic and contains a coconut fibre based substrate. The module dimensions are 500 x 500 x 100 mm, with 16 circular compartments for plant insertion. Currently, several specific details of the Junglefy system, such as the composition of the substrate matrix and the nutrients used to support the microbial biomass and botanical components of the system are 'trade secrets' and are protected by a non-disclosure agreement, and will not be described here.

Figure 1: An example of a Junglefy green wall module containing plant species *C. comosum variegatum*.

Research conducted by Torpy et al. (2015) utilizing Junglefy's green wall modules, demonstrated that the modules are able to effectively remove all particulate matter size fractions including PM_{10} and $PM_{2.5}$ from chamber air. Modules were also shown to produce a 41% lower noise reverberation than that of a hard surface, indicating that reflected noise could be reduced by 4.1 dBC.

There is, however, limited knowledge of different plant species' abilities to survive in, and remove high particulate matter levels, such as those that may be experienced in highly polluted cities. The efficiency of the green wall system has currently only been tested in laboratory chamber experiments, and as such its performance in *in situ* conditions is not known.

2.3 Gaps in Knowledge

Whilst some literature exists on the particulate matter removal capacity of vegetation, it is limited to studies conducted in Europe (i.e. Weerakkody et al., 2017, 2018), and it is unknown what particulate matter reduction capabilities are likely to be displayed by plant species used in other areas, including Australia. Similarly, most plant health studies are limited to southern Asia (i.e. Prajapati and Tripathi, 2008; Govindaraju et al., 2012; Krishnaveni, 2013; Rai and Panda, 2014), with the exception of the Plants and Environmental Quality Research Group's paper examining the short term high dosage effects of diesel smoke exposure (Paull et al., 2018). Internationally there remain knowledge gaps for green wall technology plant health (Paull et al., 2018). As such, it is not known whether the species commonly used in green walls will function in Sydney's urban atmospheric conditions. Furthermore, previous work conducted on both topics have focused on a small number of species and small time periods.

In situ studies examining PM removal from green walls as well as the effect green walls have on ambient noise and temperature conditions are lacking (Pérez et al., 2014; Simunich, 2016; Abhijith et al., 2017). Most studies testing the capacity of green walls to reduce ambient

temperature, noise and PM pollution are based on computational modelling and simulation studies (e.g. Patel and Boning 2016; Alspach and Göhring 2016; Ghazalli et al., 2018). These studies cannot accurately replicate complex *in situ* conditions, such as wind patterns, humidity, varying air quality and differing building characteristics, which may be influential on the ability of green walls to improve ambient conditions. As such, this thesis provides much needed *in situ* results on the ability of green walls to reduce particulate matter concentrations, noise pollution and ambient temperature conditions across the Sydney region.

The species utilized in green walls are frequently limited to popular international ornamental species (i.e. *Chlorophytum comosum variegatum* and *Epipremnum aureum*). However, there is growing interest from industry about the suitability of Australian native species for use in green walls. Theoretically, local species should be able to withstand the harsh environmental conditions of Australia (notably extreme heat and low rainfall conditions) better than the international species; however, their pollutant removal capacity is unknown. To the author's knowledge the only previous studies examining native species pollutant removal capacity is Leonard et al. (2016), which was limited to roadside native tree species PM deposition. As such, the results presented here will be the first to determine the suitability of Australian native plant species (*Blechnum gibbum, Callistemon citrinus, Dianella caerulea, Eremophila glabra, Lomandra longifolia* and *Westringia fruticosa*) used in green walls for air pollutant removal. The natives tested are fast growing, evergreen shrub species which are appropriate for the space confined growing area and aesthetic need of the green wall modules.

Henceforth, the research presented here will be the first of its kind in regards to a long term, multi species, spatially diverse experiment examining differences between species capacity to remove suspended particulate matter, identifying potentially pollutant tolerant species determined by plant health responses, testing the effectiveness of pre-existing green walls at

particulate matter removal, and testing the value of Australian native species for botanical air pollutant biofiltration.

Each chapters experimental methodology differed in order to meet the aims of the different hypothesises. For example, Chapter 2 utilised gravimetric and microscopic assessment, whereas Chapter 3 analysed plant health metrics, Chapter 4 consisted of *in situ* air monitoring assessment and Chapter 5 used chamber trials for pollutant removal. Having said that it is important to note that Chapters 2, 3 and 4 assessed the same plant species from the same green walls across the same Sydney locations. The reasoning for this was to provide a comprehensive data set of different plant species exposed to different environmental conditions and testing. The experiments followed a logical flow and was highly industry based, which often dictates the availability of the projects. This PhD thesis was publication focused due to the high impact of publications.

Whilst each chapter of this thesis represents an important and stand-alone aspect to increasing global knowledge of green wall functions, the results of each chapter helped shape and refine the direction of the subsequent chapters. More specifically, the authors initial focus was to determine plant specific characteristics that could enhance PM removal. This then led to questioning whether the select plant species were able to withstand this PM exposure without enduring health effects. From this, we questioned what local ambient PM reductions could be achieved from *in situ* green walls. Lastly, with the previous chapters results in mind the notion of Australian native species being used for active phytoremediation was developed.

2.4 General Aim

The aims of this PhD project were to:

- Conduct experiments to determine the differences between plant species' PM deposition and to identify effective PM accumulator species suited for high pollution environments.
- Conduct experiments to determine the effect of air pollution on different plant species' health and to identify pollutant tolerant species for implementation in high pollution environments.
- Conduct *in situ* air quality tests to determine the impact of green walls on pollution mitigation, specifically airborne PM, and to determine the effect of green walls on ambient noise and temperature conditions.
- Conduct manipulative laboratory chamber studies to determine Australian native species pollutant removal capacity.

The overall outcome is to provide information to the horticultural industry and designers of the built environment indicating how they can maximize the air purifying capacity of the green wall system, whilst reducing investment and maintenance costs.

Chapter 2 – Airborne particulate matter accumulation on common green wall plants.

ABSTRACT

In order to better design greening systems for effective PM removal, it is important to understand the impact leaf traits have on PM deposition. There are, however, inconsistences amongst the leaf traits that have previously been correlated with effective PM accumulation. The aim of this chapter was to identify vegetation characteristics of green wall plants that were associated with the accumulation of PM. To determine patterns associated with different leaf morphologies, 11 common ornamental plant species were sampled across 15 sites, over a 6 month duration. PM deposition was determined gravimetrically, and its associated size fractions determined microscopically. Linear mixed models were used to identify statistical patterns relating to differences in PM deposition across plant species. The level of PM deposition and the relative frequencies of particle size fractions were found to be statistically different amongst species. Green wall plants were shown to be effective at PM accumulation, however with some differences amongst species. There was however, little evidence of specific leaf characteristics that were influential in enhanced PM removal, and thus the differences among plant species in PM removal efficacy remains unresolved.

2.1: INTRODUCTION

2.1.1 Air Pollution

Air pollution is a major risk factor to human health (Dockery and Stone, 2007) and is a widespread environmental concern (Rai, 2016). In 2012, an estimated 3.7 million premature deaths were caused globally from outdoor air pollution exposure (WHO, 2014). Airborne PM is one of the common 'criteria pollutants' (USEPA, 2004). It is a heterogeneous solid–liquid mixture, containing toxic substances that is transported in the atmosphere, sometimes over long distances (WHO, 2005; Yu et al., 2006; Oishi, 2016). Within urban regions, vehicle use is the primary PM source (Vu et al., 2015), with road traffic contributing 80% of health concerning particulate emissions into the environment (Ottelé et al., 2010). PM particles can be comprised of toxic, carcinogenic compounds which are harmful to health, including polyaromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), polychlorinated dibenzo-p-dioxins and dibenzofurans (PCDD/Fs) and heavy metals (Caricchia et al., 1999; Jouraeva et al., 2002; USEPA, 2004; Ariola et al., 2006; Yu et al., 2006; Dzierzanowski et al., 2011). Health problems caused by exposure to particulate pollution are related to the sizes of atmospheric particles (Dockery et al., 1993; Nemmar et al., 2002; EEA, 2007). Compared to large particles, small particles are more damaging to health (WHO, 2006), more stable in the air and are slower to achieve natural sedimentation on land surfaces (Lin et al., 2018).

2.1.2 PM Size Fractions and Origins

Particulate matter, especially the smaller size fractions, almost always arise from anthropogenic sources (Beckett, 1998) including: road dust, vehicle exhaust, fertiliser production, coal burning, cement and industrial processing (USEPA, 2004). $PM_{0.1}$ (aerodynamic diameter $\leq$ 0.1μm) mainly originates from transport related emissions and photochemical reactions within the atmosphere, more specifically from the condensation, nucleation or coagulation of gaseous

pollutants (SO_2, NH_3, VOCs and NO_x; USEPA, 2004). $PM_{2.5}$ (aerodynamic diameter ≤ 2.5 μm) mainly originates from industrial processes, vehicle emissions and combustion (Chow et al., 2006), whilst PM_{10} (aerodynamic diameter ≤ 10 μm), in contrast, can originate from both anthropogenic and natural sources (Chow et al., 2006). Natural emitting sources of PM can include soil and rock erosion, forest fires, volcanic eruptions and sandstorms (USEPA, 2004). Globally, traffic generated pollution has been categorised as the most toxic class of PM (WHO, 2005), with diesel exhaust being classified as a Group 1 carcinogen by the International Agency for Research on Cancer (IARC) (Silverman et al., 2012). Whilst traffic generated PM is a major source of air pollution, other anthropogenic sources exist; such as railway networks (both electric and diesel systems), with PM being generated from the friction created between overhead cables, and brake and wheel friction (Thornes et al., 2017). With the continuous rapid expansion of the already highly urbanized region of Sydney, its future projections for increasing transportation infrastructure represents an equally rapid increase in health concerning air pollution.

2.1.3 Particulate Matter Health Impacts

There is a strong relationship between increased levels of ambient PM exposure and adverse health conditions (WHO, 2013). Coarse particles (PM_{10}), fine particles ($PM_{2.5}$) and ultra-fine particles ($PM_{0.1}$ and smaller) are known for their toxicity and ease of inhalation (Solomon et al., 2012). PM exposure can cause cardiac and respiratory diseases (Polichetti et al., 2009), including asthma (Anderson et al., 2013), atherosclerosis (Araujo, 2011), lung cancer and cardiopulmonary diseases (Pope et al., 2011; Solomon et al., 2012). $PM_{2.5}$ are particularly harmful to human health because they can reach narrow spaces within the lung (Brunkeef and Holgate, 2002), causing negative health effects (Powe and Willis, 2004). Ultra-fine particles have the capacity to cross cell membranes; influencing intracellular functions, making them the most health damaging PM size fraction (Riddle et al., 2009). When entering the blood

stream, $PM_{0.1}$ can impact blood coagulability by creating systemic inflammatory changes (Seaton et al., 1995) and can further enter the brain, liver and spleen through the olfactory nerves, causing major damage to the central nervous system (Solomon et al., 2012) (resulting in disorders such as Parkinson's disease and Alzheimer's disease) if the toxicity and chemical composition of the particulates is severe (Allsop et al., 2008; Maher et al., 2013). Actions that reduce our exposure to PM are therefore paramount to ensure healthy and safe ambient environmental conditions.

2.1.4 Green walls as a PM pollution remediator

Vegetation has significant potential as a sink for PM in urban regions (Popek et al., 2013; Räsänen et al., 2014). PM is removed from the ambient air by adhesion to leaf surfaces (Ottelé et al., 2010; Sternberg et al., 2010), with additional sequestration evident from penetration of the wax layer of leaves if the PM contains organic pollutants of a lipophilic nature (Dzierzanowski et al., 2011). Vegetating urban landscapes with trees is a process constrained by many factors, including space limitations, sunlight availability, sub-surface infrastructure requirements, the size ratio between the tree and adjacent buildings and the suitability of the prevailing soil (Johnston and Newton, 2004). Green walls in comparison, do not consume additional space at the street level, instead utilizing pre-existing building surfaces, thereby increasing the particulate collection area of the building due to the large surface area presented by plants (Ottelé et al., 2010).

2.1.5 Influential factors on PM deposition

The means in which particles can be sequestered from the atmosphere include, occult (contaminated water droplets via mist and clouds), wet and dry deposition (NEGTAP, 2001). In the case of dry deposition, particulates can be removed by diffusion across surface boundary layers, such as through Brownian motion; by sedimentation caused by gravity or by

interception and impaction caused by turbulent movement (Slinn, 1982; Beckett et al., 2004; Wang et al., 2006). Aerodynamic resistance which is the resistance exerted on particles by the air; surface resistance which is the deposition surface properties; and boundary layer resistance which is the resistance offered by the laminar air layer which is adjacent to the deposition surface, however, are all means which can affect the rate at which PM deposition occurs (Davidson and Wu, 1990).

Additionally, to the aforementioned processes, the structure and traits that vegetation possess provide an additional factor to consider when assessing PM deposition. Vegetation characteristics such as leaf orientation, shape, size and surface morphology have been identified as significant factors associated with variations in PM deposition rate amongst plant species (Litschke and Kuttler 2008; Petroff et al., 2008; Chen et al., 2016; Leonard et al., 2016). Macrostructural leaf traits that have been shown to increase PM accumulation include whorled leaf arrangements and larger leaf area; whilst advantageous microstructural traits include pubescence, low stomatal densities, rough surfaces and thick waxy epicuticles (Chaturvedi et al., 2013; Popek et al., 2013; Mo et al., 2015). Additionally, the chemical composition and structure of the epicuticular wax has also been found to be influential on PM accumulation (Dzierzanowski et al., 2011; Leonard et al., 2016). Similarly, the structure of leaf hairs can also alter PM deposition, with some leaf hairs exhibiting hydrophobicity, attracting charged particles such as heavy metals found in PM (Fernàndez et al., 2014).

Nonetheless, the plant traits that have been correlated with increased PM accumulation are inconsistent amongst previous research. For example, different ideal leaf shapes for PM removal have been concluded from different studies, with Beckett et al. (2000), Dzierzanowski et al. (2011) and Wang et al. (2011) noting the positive effects of needle like leaves in comparison to broad leaved species for PM accumulation. Leonard et al. (2016), in contrast, found that lanceolate leaves demonstrated more effective PM accumulation than both needle-

like and linear leaves. Microstructural traits that have been found to be advantageous for particulate accumulation are also inconsistent throughout the literature, with some previous studies noting the importance of epicuticular wax on PM deposition (e.g. Dzierzanowski et al., 2011; Sæbo et al., 2012), whilst others have found a negative relationship between PM deposition and epicuticular wax (i.e. Liu et al., 2012). Similarly, leaf hair presence has been associated with high PM accumulation in many studies (e.g. Beckett et al., 2000; Kardel et al., 2012; Ram et al., 2014), however, Perini et al. (2017) detected a negative association between PM capture and leaf hairs. In view of these discrepancies, it remains important to determine the relationships between green wall plants, leaf traits and ambient PM accumulation; so as to maximize practical PM reduction through appropriate plant use.

2.1.6 Aim

The PM deposition capacity of plants has received insufficient research attention (Pugh et al., 2012). Whilst the influence of individual leaf traits on PM accumulation is noted in the literature, the interactions between different leaf characteristics are not yet understood (Leonard et al., 2016). Additionally, uncertainty remains surrounding the impact of individual leaf traits on PM retention due to variable conclusions from previous studies (Weerakkody et al., 2018). In order to better design greening systems for maximum PM removal, the impact leaf traits have on PM deposition must be better known. Previous research on the ability of plants to reduce ambient PM has been heavily focused on single species testing, usually using climbing plant species such as *Hedera helix* (Ottelé et al., 2010; Sternberg et al., 2010; Cheetham et al., 2012). Research on green walls and their capacity to reduce PM is limited to a few studies (Perini et al., 2017), and is not yet well understood. Given the high leaf density presented by most green wall systems, it is probable that their PM accumulation potential is substantial.

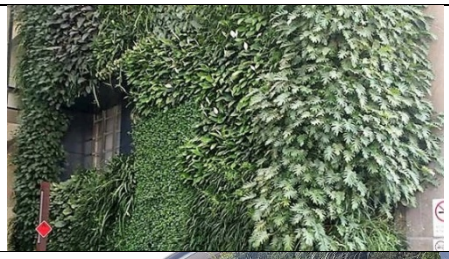
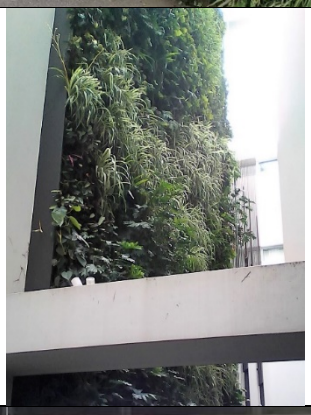
The aim of this project was to assess the effectiveness of plant species used within green walls in Sydney Australia for accumulating ambient PM, and to identify vegetation characteristics that result in maximum pollutant attenuation.

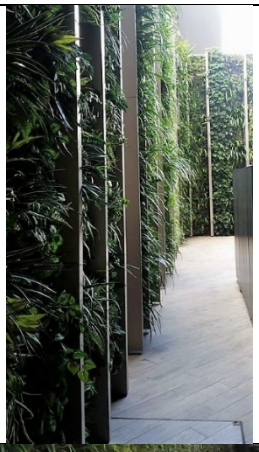
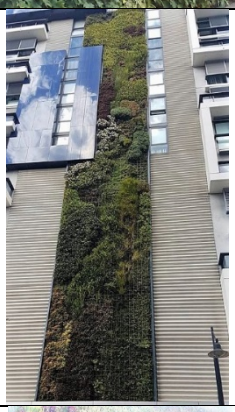
2.2: METHOD

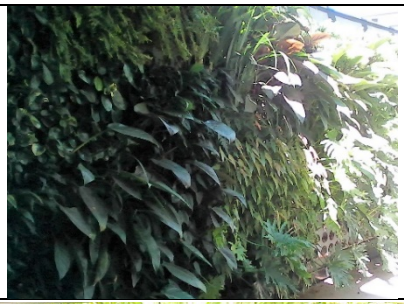
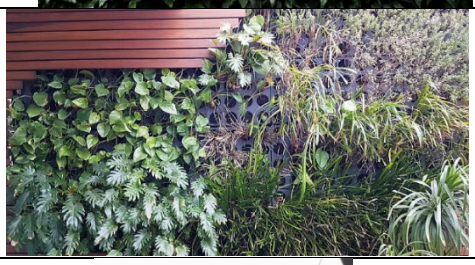
2.2.1 Sample Sites

Fifteen sites within urban Sydney, Australia were selected based on the presence of similar outdoor green walls. Green walls were of a modular design, produced by Junglefy P/L, Sydney Australia. The sites varied in location, use and pollutant conditions (Table 1). All sampled leaves were taken from pre-existing *in situ* green walls. All 15 sites had their green walls installed a minimum of 24 months prior to sampling. It was not possible to standardise the year of green wall construction in this project, as these walls had been installed in various enterprises years prior to the study. Whilst the overall atmospheric exposure time was not known, the sample size used was sufficient to randomize these effects within, but not amongst species. Leaf life expectancy is another characteristic that will differentiate between the various species in their capacity to collect PM, thus only young mature leaves were selected for sampling. Additionally, a level of standardisation was achieved through environmental effects, as rainfall volume was consistent across the sampling area and rainfall events temporally standardised PM accumulation through leaf washing effects (e.g. Prajapati and Tripathi, 2008).

Table 1: Test site location description, ranked from highest plant biomass to least.

Site Number	Site Location	Coordinates	Notes	Image
1	Ultimo	33°52'60.0"S 151°12'03.7"E	Green wall situated on tertiary education facility	
2	Gordon	33°45'33.2"S 151°09'20.0"E	High School. Green wall situated in a courtyard	
3	The Rocks, Site 1	33°51'45.6"S 151°12'20.4"E	Extensive green wall situated on expressway	
4	Mosman	33°49'41.7"S 151°14'04.1"E	Apartment complex with outdoor green wall	
5	The Rocks, Site 2	33°51'39.4"S 151°12'29.9"E	Green wall situated under rail line support structure	

6	Ashfield	33°53'25.7"S 151°07'40.3"E	Apartment complex with green wall situated in the back foyer	
7	Crows Nest	33°49'37.6"S 151°12'08.4"E	Green wall situated on the exterior of a grocery store	
8	Camperdown	33°53'05.2"S 151°10'35.9"E	Apartment complex with green wall situated on the exterior of the building	
9	Tamarama	33°53'53.5"S 151°16'23.6"E	Residential property, green wall situated in back yard	
10	Rose Bay	33°52'32.4"S 151°15'55.5"E	Residential property	

11	Lane Cove	33°48'55.8"S 151°10'10.6"E	Display home with green wall situated in the outdoor area	
12	North Bondi	33°53'16.0"S 151°16'55.0"E	Residential property	
13	Bondi	33°53'22.0"S 151°16'58.5"E	Residential property	
14	Woollahra	33°53'15.7"S 151°14'59.1"E	Residential property, green wall situated in front courtyard	
15	Summer Hill	33°53'57.0"S 151°07'54.3"E	High School, green wall situated in a courtyard	

2.2.2 Ambient PM Concentrations

Ambient PM quantification was performed at the sites to determine any relationships between ambient PM conditions and species PM accumulation. The ambient PM concentrations at each site were assessed using a DustTrack II 8532 laser densitometer (TSI, Shoreview, Minnesota). At each site, PM_{10} and $PM_{2.5}$ were sampled to obtain 3 minute time weighted averages. Air quality samples were restricted to collection between 10 a.m. and 3 p.m. to avoid spikes created by peak hour commuters (Irga et al., 2015). Samples were taken once a month at each site for the project's 6 month duration (June 2017 – November 2017).

2.2.3 PM Deposition

To determine the PM accumulation performance of different plant species, 11 species growing in the green walls amongst the test sites that had different leaf shapes, sizes and morphologies were chosen (Table 2). Not all plant species were present at each site.

Table 2: Plant species and their characteristics used for PM accumulation assessment. [1]Leaf size bins were determined as follows: small < 30 cm^2, medium 30 - 60 cm^2, large > 60 cm^2.

Scientific Name	Common Name	Leaf Shape	Leaf Size[1]	Leaf Hair Present	Leaf Arrangement
Philodendron xanadu Croat, Mayo & J. Boos	Xanadu	Lobed	Large	N	Rosettes
Peperomia obtusifolia (L.) A. Dietr	Baby Rubber Plant	Obovate	Medium	N	Alternate
Plectranthus madagascariensis George Bentham	Variegated Mintleaf	Lobed	Small	Y	Opposite
Nematanthus glabra Schrad.	Goldfish Plant	Pinnate	Small	N	Opposite
Nandina domestica Thunb.	Pink Blush	Lanceolate	Small	N	Opposite
Neomarica gracilis (Herb.) Sprague, Bull. Misc. Inform. Kew.	Walking Iris	Linear	Large	N	Rosettes
Nephrolepis exaltata bostoniensis (L.) Schott	Boston Fern	Pinnate	Small	Present on stems but not leaves	Opposite
Chlorophytum comosum variegatum (Thunb.) Jacques	Variegated Spider Plant	Linear	Medium	N	Rosettes
Chlorophytum comosum (Thunb.) Jacques	Spider Plant	Linear	Medium	N	Rosettes
Spathiphyllum wallisii Regel	Peace Lily	Lanceolate	Large	N	Rosettes
Peperomia glabella (Sw.) A. Dietr.	Small Leaf Peperomia	Obovate	Small	N	Alternate

Leaf arrangement (whorled, opposite or alternate) and leaf shape (elliptic, lanceolate, needle like, linear or obovate) were determined for the sampled species as per Leonard et al. (2016). At each site, 5 replicate leaves of each species were hand-picked and individually sealed into pre-labelled sample bags so as to minimize PM loss. This form of sample collection has been used in other studies (i.e. in Leonard et al., 2016). The position of leaf samples was randomized within the green walls at each sampling occasion and month to randomize variations in green wall characteristics at each site. Samples were taken monthly for a 6 month period from June to November, 2017.

From the 5 replicate leaves, 3 were used to determine the deposited PM mass using a dry gravimetric technique. This was carried out by weighing the intact leaves, then removing PM using a camel-hair brush and reweighing. The brush used was soft bristled and leaves were handled carefully, to avoid removing any leaf components in this process i.e. wax layers and leaf hairs (Das and Pattanayak, 1978). The leaf was then sized with a leaf area meter (Licor LI-3000-A, Nebraska, USA) to obtain an accurate area measurement. The amount of PM collected for each species was expressed per unit area of leaf, as PM was dusted from both adaxial and abaxial surfaces. Previous studies (Dzierzanowski et al., 2011; Leonard et al., 2016) similarly expressed their results. The deposited PM content was then calculated using Formula 1.

Formula 1:

$$PM\ Deposition\ \left(\frac{mg}{cm^2}\right) = \frac{(Mass\ of\ intact\ leaf - initial\ mass\ of\ leaf)}{Total\ surface\ area\ of\ leaf}$$

2.2.4 PM Size Fractions

To quantify the proportional contribution of different particle size fractions to the PM deposited on the green wall plant leaves, microscopic analysis was conducted. More specially, on the remaining 2 leaves from each species, for each site and month, a 2 cm length of adhesive tape was placed onto the middle upper surface of each leaf, pressed down, gently removed and placed onto a microscope slide. Weerakkody et al. (2017) observed that the leaf blade had less variable PM distribution compared to leaf tip, base, mid rib and edges, and as such the leaf blade only was sampled in this process. Images of the microscope slides were then taken using a Nikon Automated Upright Fluorescence Microscope at x20 magnification for 15 random surface fields on each slide. Each image was then analysed using NIS-Elements Viewer 4.20, which generated data for the diameter of each particle present on the image. From this, two PM size fraction ranges: $PM_{<5}$ and $PM_{>5}$ were categorized using MS Excel. Leaves can accumulate a range of different PM size fractions, and as such, microscopic analysis was used to determine the relative accumulation of small and large sized PM ($PM < 5$ mg cm^{-2} and $PM > 5$ mg cm^{-2}). Thus, for each species a probability density result was produced for $PM < 5$ mg cm^{-2} and $PM > 5$ mg cm^{-2}.

2.2.5 Statistical Analysis

For statistical analysis, the R programme was used, coupled with the packages car, multcomp, nlme and Hmisc. Mean values for particle counts per image for the two PM fractions ($PM_{<5}$ and $PM_{>5}$) were determined for the eleven species for each month (June–November), at each site in which they occurred, from six replicate samples per species per site. Principal components analysis (PCA) was performed using square root transformed ambient airborne PM data recorded near the green walls (two fractions, $PM_{2.5}$ and PM_{10}), with the first principal component (capturing 91% of the variance in the ambient PM data set) used as an independent

variable in subsequent analyses. The variables included in the PCA were the site-wise values for the two ambient PM concentrations (PM < 2.5, and PM > 5). This was done because despite a strong correlation between the concentrations of the two PM fractions (Pearson's correlation = 0.80, P < 0.0001), we wished to use the most accurate means of conveying the nature of the tested relationships between ambient and leaf accumulated PM. The square root transformation prior to PCA was used so that the resultant PC1 did not contain several high outlying values (present also in the PM fractions), which might skew results when regression coefficients were calculated.

To test for differences among species, and whether leaf PM accumulation was related to ambient PM concentration, linear mixed models (LMMs) were fitted, using species as a fixed categorical factor (11 levels), the ambient PM PC as a fixed continuous factor, and a species x ambient PM PC interaction term. To control for variation amongst months and variation within species amongst sites, two random factors were used; the month in which observations were recorded, and a nested species x site term. Where significant differences were found among species or for the species x ambient PM interaction, pairwise comparisons between species, or between slopes were made using a Tukey correction for multiple comparisons. Following this, the relationships between the two PM fractions were explored for each species, first using paired sample *t*-tests (repeated for all species), followed by LMM modelling of $PM_{>5}$ as a function of $PM_{<5}$, including a species x $PM_{<5}$ interaction term to test if the relationship between deposition of the PM fractions was consistent across species.

To test the effect of leaf traits on leaf PM deposition and their relationship with ambient PM, models were built with terms for the leaf traits (a fixed categorical factor with four levels), ambient PM (as a fixed continuous factor), a leaf trait term nested within species (fixed factor, included to test for differences among species with the same leaf traits), a leaf trait x ambient

PM interaction term (fixed factor) and a leaf traits/species x ambient PM (fixed factor, included to test for differences in the relationships between ambient PM among species within leaf trait groups). The same random terms used in the first two models were also used in these models. In all models accumulated leaf PM data was log transformed prior to analysis.

2.3: RESULTS & DISCUSSION

2.3.1 Differences among species and relationship with ambient PM

Significant differences were found among species for the accumulation of both $PM_{<5}$ (χ^2_{10} = 75.1, P < 0.0001; Fig. 2a) and $PM_{>5}$ (χ^2_{10} = 71.0, P < 0.0001; Fig. 2b); similar to the results of Leonard et al. (2016), Weerakkody et al. 2017 and 2018. No significant relationship was found between $PM_{<5}$ deposition and ambient PM concentration (χ^2_1 = 0.1, P = 0.7; Fig. 3a), and no significant interaction among species accumulated PM and ambient PM was detected (χ^2_{10} = 4.3, P = 0.9). For $PM_{>5}$, a significant interaction among species and ambient PM emerged (χ^2_{10} = 23.3, P < 0.01; Figs. 3b and 3c). The interaction was found to be generated by three species showing a significant association between higher accumulated PM and greater ambient PM (*C. comosum variegatum*: Variegated Spider Plant, *N. exaltata bostoniensis*: Boston Fern, and *N. glabra*: Goldfish Plant), and a further three species having a significant association between lower accumulated PM and greater ambient PM (*N. gracilis*: Walking Iris, *P. obtusifolia*: Baby Rubber Plant, and *P. xanadu*: Xanadu; Fig. 3c). This difference is likely driven by leaf trait differences, specifically between the small linear and large rosette species. More specifically, of the listed species, two small linear species (*N. exaltata bostoniensis*: Boston Fern, and *N. glabra*: Goldfish Plant) were found to have higher accumulated PM at greater ambient PM;

whilst two of the large rosette species (*N. gracilis*: Walking Iris and *P. xanadu*: Xanadu) were found to have lower accumulated PM at greater ambient PM.

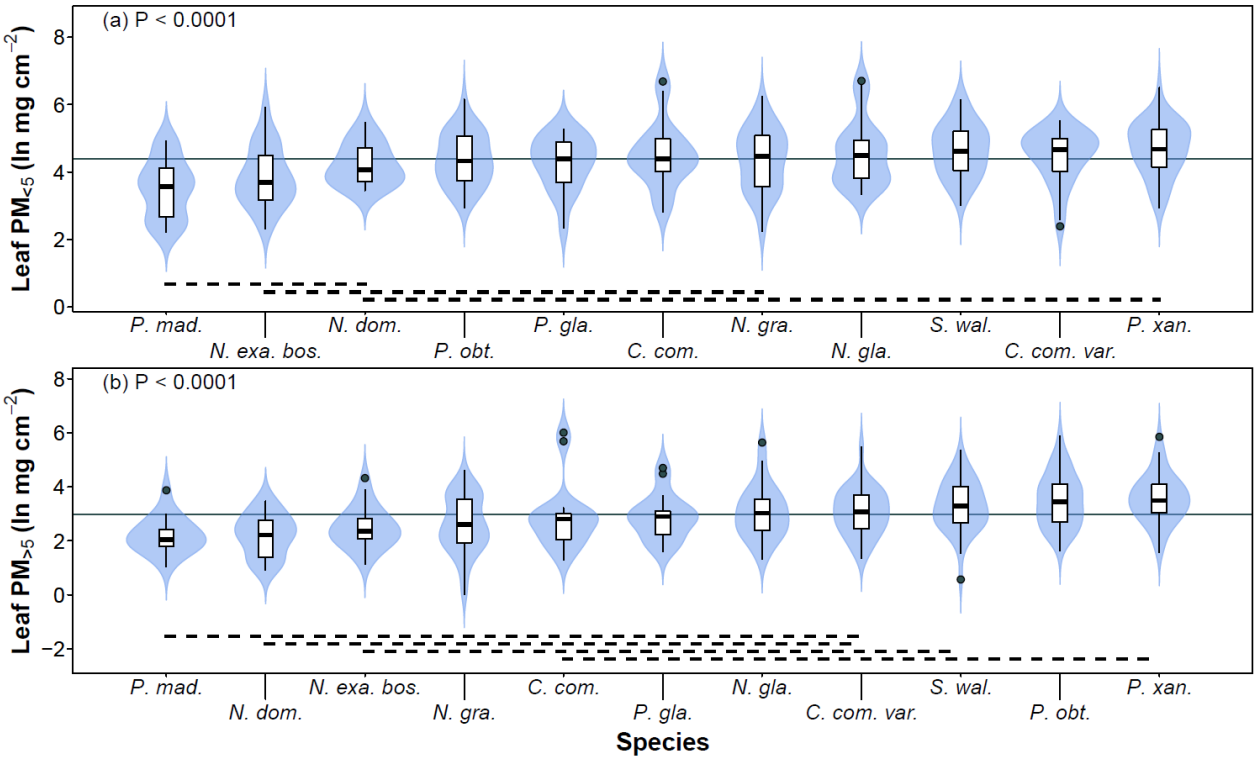


Figure 2: Plots of $PM_{<5}$ (a) and $PM_{>5}$ (b) leaf deposition in the study species, sorted from lowest mean values to highest (left to right). Units are Ln (PM accumulation in mg cm^{-2}). The shaded areas show the estimated probability density, with boxplots displayed. Broken lines below the density shapes indicate which groups of species did not differ significantly. Note: *P. xan = Philodendron xanadu; P. obt = Peperomia obtusifolia; P. mad = Plectranthus madagascariensis; N. gla = Nematanthus glabra; N. dom = Nandina domestica; N. gra = Neomarica gracilis; N. exa. bos = Nephrolepis exaltata bostoniensis; C. com. var = Chlorophytum comosum variegatum; C. com = Chlorophytum comosum; S. wal = Spathiphyllum wallisii & P. gla = Peperomia glabella.*

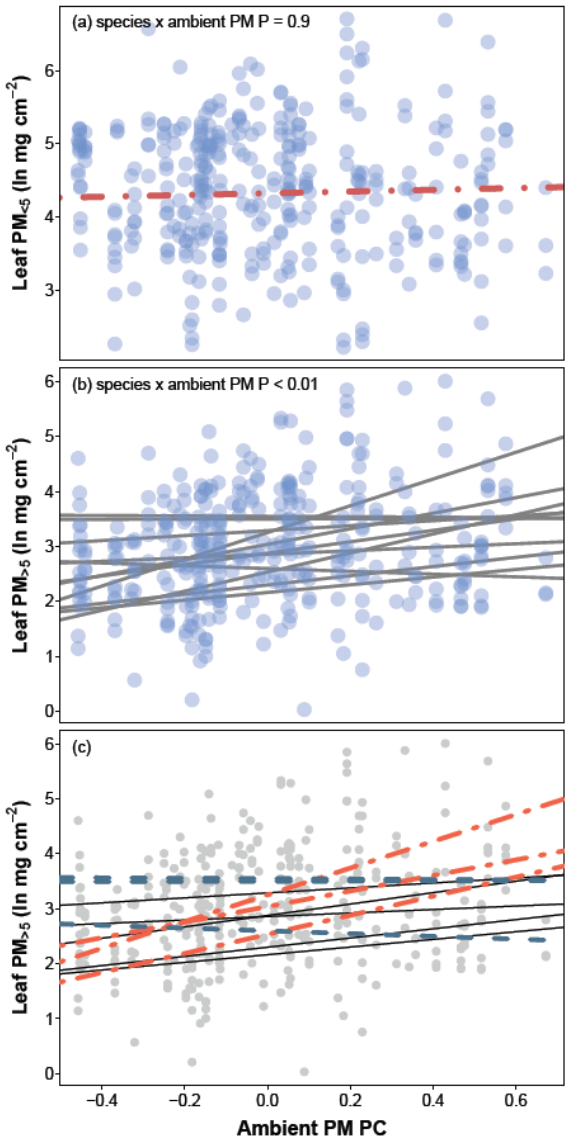


Figure 3: Relationships between accumulated leaf $PM_{<5}$ (a), and $PM_{>5}$ (b, c) deposition and the ambient PM principal component (see text for explanation). In (a), the dot-dashed line shows the relationship across species. In (b) the individual species' relationships are shown as solid lines, with (c) showing the species that contributed to the significant species x ambient PM interaction, with dashed lines for species showing a significant negative relationship, dot-dashed lines for species showing a significant positive relationship, and solid lines for species with no significant relationship.

The density of the accumulated $PM_{<5}$ fraction was found to be significantly greater than the $PM_{>5}$ fraction density across all species (Fig. 4). This trend has also been observed in previous studies (i.e. observed in: Freer-Smith et al., 2005; Ottelé et al., 2010; Perini et al., 2017; Weerakkody et al., 2017, 2018). This finding suggests that green walls may be more effective at reducing smaller PM size fractions (e.g. Weerakkody et al., 2017), or that leaves are more capable or retaining smaller PM (e.g. Przybysz et al., 2014), or simply that the ambient PM

was predominantly composed of smaller particles. The variation in PM deposition across size fractions is thought to be due to different deposition velocities resulting from the different aerodynamic behaviour displayed by different sized particles (Slinn, 1982; Weerakkody et al., 2017). For example, the increased turbulence in the boundary layer around a deposition surface has a greater effect on the turbulent transfer of smaller PM size fractions (Slinn, 1982; Petroff et al., 2008). Additionally, the effects of deposition velocities vary amongst the various processes in which dry deposition can occur i.e. interception, impaction and sedimentation under gravity (Weerakkody et al., 2017), resulting in the PM deposition differences amongst size fractions.

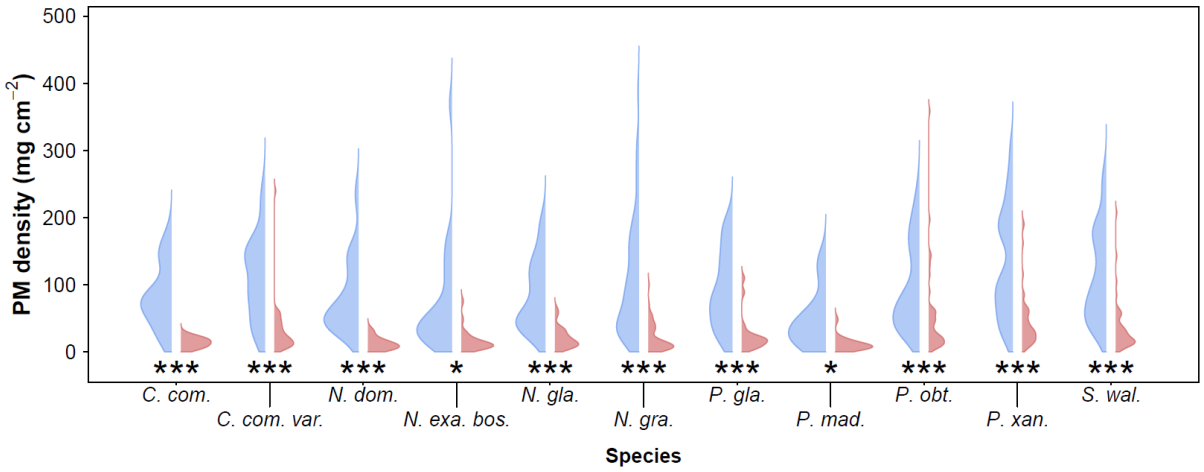


Figure 4: Probability densities of $PM_{<5}$ (left side, blue), and $PM_{>5}$ (right side, red), for each species. Asterisks beneath the density shapes indicate the significance of paired sample *t*-tests comparing densities of the two particle fraction sizes for a given species. One asterisk indicates a P value < 0.01, and three a P value < 0.0001. Note: *P. xan = Philodendron xanadu; P. obt = Peperomia obtusifolia; P. mad = Plectranthus madagascariensis; N. gla = Nematanthus glabra; N. dom = Nandina domestica; N. gra = Neomarica gracilis; N. exa. bos = Nephrolepis exaltata bostoniensis; C. com. var = Chlorophytum comosum variegatum; C. com = Chlorophytum comosum; S. wal = Spathiphyllum wallisii & P. gla = Peperomia glabella.*

N. exaltata bostoniensis (Boston Fern) accumulated less $PM_{>5}$ for a given amount of $PM_{<5}$ when compared to *C. comosum variegatum*: Variegated Spider Plant, *N. gracilis*: Walking Iris, *P. obtusifolia*: Baby Rubber Plant, *P. Xanadu*: Xanadu, and *S. wallisii*: Peace Lily (Fig. 5).

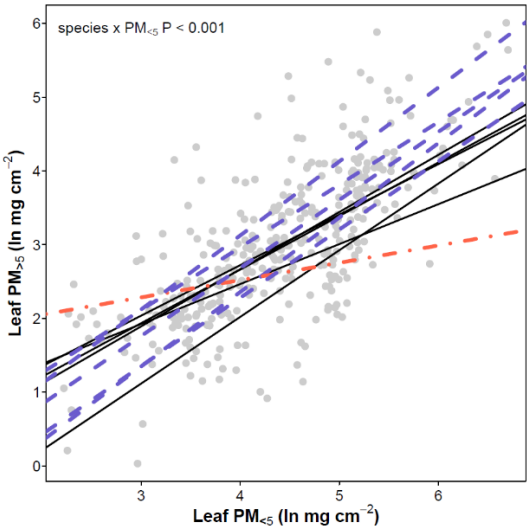


Figure 5: Scatterplot showing accumulation of the two PM fractions, by the species *C. comosum variegatum*, *N. gracilis*, *P. obtusifolia*, *P. xanadu*, and *S. wallisii* as dashed lines, and *N. exaltata bostoniensis* as the singular dot-dashed line. The remaining species, represented with solid lines, did not differ significantly from the species shown as dashed lines, or *N. exaltata bostoniensis*.

2.3.2 The effect of leaf traits on PM deposition

Significant differences were found among species within leaf trait groups for $PM_{<5}$ accumulation ($\chi^2_7 = 41.3$, $P < 0.0001$; Fig. 6a), and also for $PM_{>5}$ ($\chi^2_7 = 42.1$, $P < 0.0001$; Fig. 6b), with relatively large interspecific variation for both PM fractions amongst the small linear-leaved species. Significant differences among the leaf trait groups were found for $PM_{<5}$ ($\chi^2_3 = 33.9$, $P < 0.0001$; Fig. 7a), with the small linear-leaved species demonstrating lower accumulation of these particles compared to the medium and large rosette plant groups, particularly for *P. madagascariensis* (Variegated mintleaf), and *N. exaltata bostoniensis* (Boston Fern) (Fig. 4a). $PM_{<5}$ accumulation was the same for the medium linear and medium and large rosette groups (Fig. 7a). The deposition of $PM_{>5}$ differed significantly amongst the leaf trait groups ($\chi^2_3 = 31.4$, $P < 0.0001$; Fig. 7b). Whilst leaf deposition of $PM_{>5}$ did not

significantly differ amongst the small linear and medium rosette groups, the medium linear and large rosette groups accumulated comparatively higher quantities of $PM_{>5}$ (Fig. 7b). On the contrary, most previous research has observed higher PM deposition rates for smaller sized leaves (e.g. Freer-Smith et al., 2005; Weerakkody et al., 2017, 2018). This is thought to be due to the reduced tendency of smaller leaves to move with the wind, and thus resuspend accumulated PM (Leonard et al., 2016), combined with larger edge effects for smaller leaves, leading to a higher frequency of PM impaction (Weerakkody et al., 2018). In contrast, in the current project, the smallest leaves demonstrated the least effective PM accumulation. Weerakkody et al. (2017) did note that two of their small-leaved species showed comparatively low PM deposition, suggesting that this was a result of their lower rigidity and attendant lower capacity to withstand PM contaminated air flow, thus lowering the turbulence surrounding the leaf boundary. They concluded that small-leaved species with a complex morphology were the most efficient species for reducing ambient PM. It is therefore possible that the current observations resulted from the soft structure and simple morphology of the tested species, specifically, *N. exaltata bostoniensis*, and so were in line with the findings of Weerakkody et al. (2017).

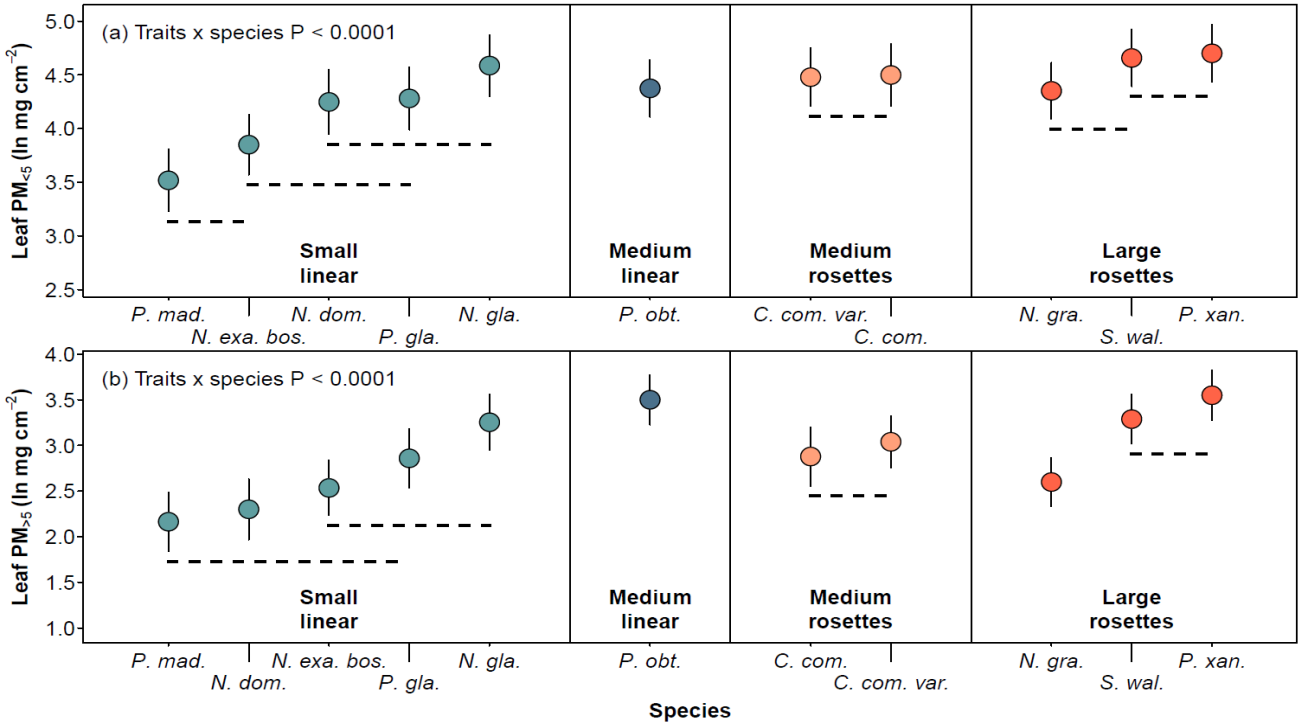


Figure 6: Plots of species mean values ± SE for $PM_{<5}$ (a) and $PM_{>5}$ (b) accumulation by leaf trait group. Broken lines beneath points indicate species that did not differ significantly within each leaf trait group. At the top of each plot, the results for the nested traits x species term from the model are presented. Note: *P. xan = Philodendron xanadu; P. obt = Peperomia obtusifolia; P. mad = Plectranthus madagascariensis; N. gla = Nematanthus glabra; N. dom = Nandina domestica; N. gra = Neomarica gracilis; N. exa. bos = Nephrolepis exaltata bostoniensis; C. com. var = Chlorophytum comosum variegatum; C. com = Chlorophytum comosum; S. wal = Spathiphyllum wallisii & P. gla = Peperomia glabella.*

Similar to the $PM_{<5}$ findings, *P. madagascariensis* and *N. exaltata bostoniensis* with the addition of *N. domestica* (Pink Blush) exhibited low levels of $PM_{>5}$ accumulation, driving an overall statistical difference between the small linear and other leaf groups for the accumulation of this particle size fraction (Fig. 6b). For both fractions, there was no significant leaf PM x ambient PM effect when comparing species within each group ($PM_{<5}$: $\chi^2_7 = 2.2$, P = 0.9; $PM_{>5}$ $\chi^2_7 = 8.3$, P = 0.3; Fig. 7d). There were no significant leaf trait x ambient PM interactions found for $PM_{<5}$ ($\chi^2_3 = 2.1$, P = 0.6; Fig. 7c), in contrast to $PM_{>5}$ ($\chi^2_3 = 15.0$, P = 0.002; Fig. 7d). This finding was driven by the small linear, and medium rosette groups showing a positive relationship between ambient PM and accumulated $PM_{>5}$, while the medium linear and large

rosette groups showed no relationships. Similarly, to the current observations, linear leaved or 'grass like' species have previously been found to have an overall low PM accumulation ability (e.g. Dochinger, 1980; Currie and Bass, 2008; Leonard et al., 2016; Weerakkody et al., 2017, 2018). This is likely due to the tendency for linear leaves to bend easily with wind flow due to their narrow bases or petioles (Weerakkody et al., 2018), thus presenting small boundary effects. Furthermore, Weerakkody et al. (2017) suggested that species that have simple leaf arrangements with larger gaps between their leaves may produce lower turbulence surrounding the foliage, resulting in less frequent impaction rates.

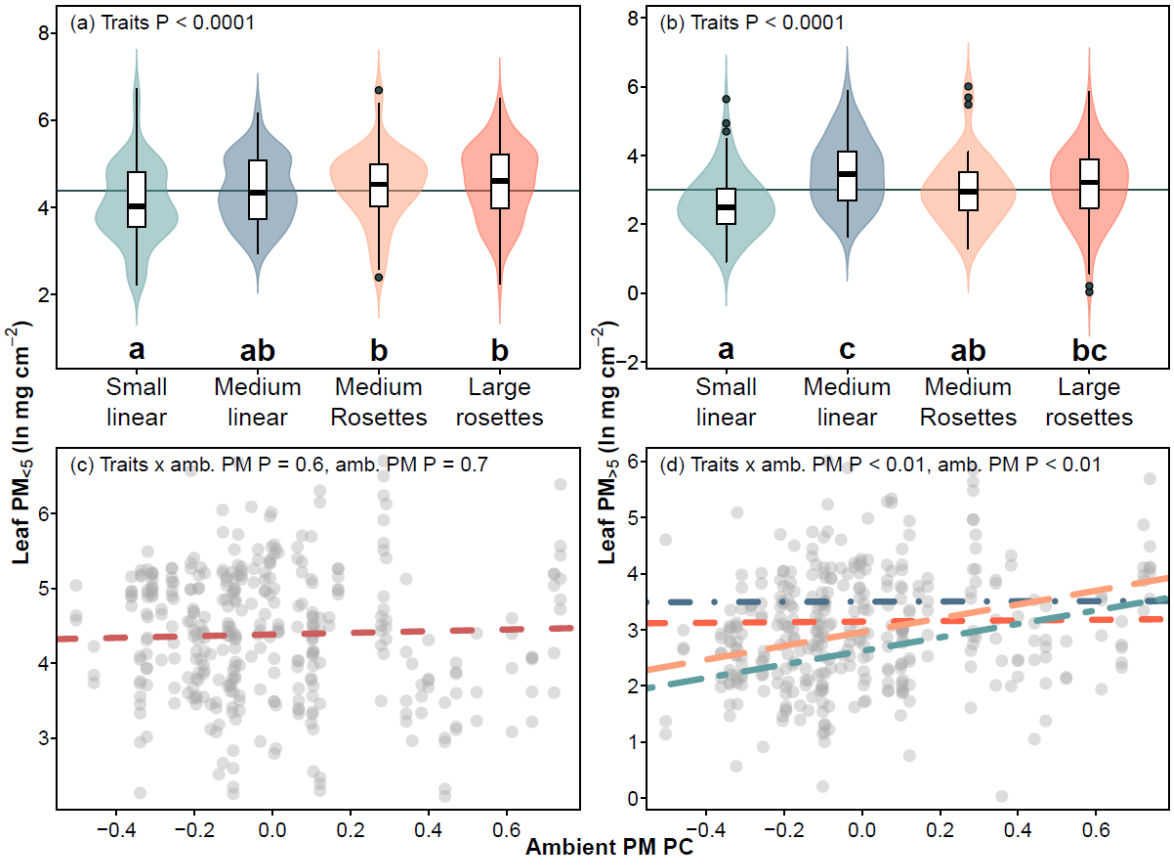


Figure 7: Plots of $PM_{<5}$ (a) and $PM_{>5}$ (b) accumulation by leaf trait groups. Shaded areas show the estimated probability densities, with boxplots displayed. The relationship between the groups and ambient PM is shown in (c) for $PM_{<5}$ and (d) for $PM_{>5}$. In (c) the mean relationship is shown (mean intercept and slope for groups). The individual relationships between the leaf groups $PM_{>5}$ deposition and ambient PM are shown in (d). The regression lines in (d) from top to bottom are as follows: medium linear, large rosettes, medium rosettes and small linear as per plots in (a) and (b).

The presence of leaf hairs (trichomes) has been shown to increase PM accumulation in multiple previous studies (Beckett et al., 2000; Sæbo et al., 2012; Räsänen et al., 2013; Ram et al., 2014; Leonard et al., 2016; Chen et al., 2017). Leaf hairs are thought to increase PM retention by preventing the resuspension of deposited PM, and by increasing the leaf surface area for the collision of particles (Prusty et al., 2005; Qiu et al., 2009). In the current project, only one of the tested species had leaf hairs, *P. madagascariensis* (Variegated Mintleaf), which showed one of the lowest PM accumulation values. Whilst this finding was not in line with the majority of previous studies, it aligned with the findings of Perini et al. (2017), who found that hairy leaves were negatively related to PM deposition.

The results from the current project highlights that many plants used in green walls are capable of accumulating airborne PM, but that this property varies amongst plant species. In particular, *P. madagascariensis* (Variegated Mintleaf) and *N. exaltata bostoniensis* (Boston Fern) were not effective PM accumulators, indicating that plants of this structural form may be ineffective for passive PM accumulation.

The quantity of PM accumulated per unit of time was estimated by dividing the total amount of PM accumulated per unit of leaf area for each species from the September 2017 samples by the number of days since the last rain event that was of a magnitude that would be expected to largely remove PM from plant leaves (Xu et al., 2017). 38.8 mL of rain fell on the 9th of June 2017 with no significant rainfall events between this date and September 2017. This date was used for the calculation.

It was found that all species demonstrated quite different PM accumulation efficiencies (Table 3). The daily accumulation values were also found to differ from the previously recorded values shown in Figure 2. This was likely due to methodological differences between the microscopic and dry gravimetric techniques. Most notably, *P. madagascariensis* was found to

have the lowest PM accumulation based on the microscopy findings (Fig. 2); gravimetric analysis contrastingly showed this species to accumulate the highest amount of PM (Table 3). Whilst the reasoning behind this is not clear it is possible that this was a result of leaf hairs being removed through the brushing process, increasing the weight of PM recorded. This reasoning is further warranted by the change in ranking for *N. exaltata bostoniensis* from within the lowest 3 PM accumulating species as per the microscopy results (Fig. 2) to within the highest 3 accumulating species as per the gravimetric data (Table 3). Whilst *N. exaltata bostoniensis* does not possess trichomes on the leaf itself, the stems of this species contain dense, small leaf hairs. It is possible that the dry gravimetric method is unsuitable for quantifying PM on species that have significant trichrome development. As is proposed in later sections of this thesis, a methods development procedure will be required to resolve issues such as this. Nonetheless, *P. glabella, C. comosum* and *N. gracilis* remained amongst their previous rankings, indicative that these species possess average PM deposition capacities amongst those tested. *P. xanadu* and *S. wallisii* remained within the higher PM performing species across both methodologies, highlighting the consistency for these species to effectively filter PM *in situ*.

Table 3: The test species PM accumulation presented in mg/cm^2/day.

Species Name	**PM Accumulation (mg/cm^2/day)**
C. comosum	1.98
C. comosum variegatum	1.79
N. domestica	1.80
N. exaltata bostoniensis	2.64
P. glabella	2.07
N. glabra	4.07
N. gracilis	0.77
P. madagascariensis	4.36
P. obtusifolia	1.86
P. Xanadu	2.17
S. wallisii	2.07

The methods used to determine the PM filtering capacity of plants have varied amongst previous investigations (i.e. Beckett et al., 2000; Freer-Smith et al., 2004; McDonald et al., 2007; Ottelé et al., 2010; Sternberg et al., 2010; Dover, 2015; Maher et al., 2013; Terzaghi et al., 2013; Lenoard et al., 2016; Zhang et al., 2017), likely causing some of the inconsistencies observed in the literature. Common methods used include Scanning Electron Microscopy (SEM) and imaging (e.g. Weerakkody et al., 2017), filtration (e.g. Leonard et al., 2016) and gravimetric assessment (e.g. Das and Pattanayak, 1978). However, with no standard, universal method of PM determination, comparisons between studies are difficult to interpret. Furthermore, all of these methods have limitations, including: the limited capacity of water to remove PM from leaf and wax structures; the limited capacity of chloroform to dissolve non-polar components of PM, and the SEM scanning area being much smaller than the leaf surface

area, requiring a large sample size of micrographs to constitute a representative sample of the overall PM deposition (Weerakkody et al., 2017). Additionally, SEM and other microscopic analyses are unable to determine the total mass of deposited PM, instead providing a particle count for each PM size fraction. Thus, the current project aimed to provide both total deposited PM mass and PM size fraction counts with the two methodologies used. Future studies should conduct a comparative assessment of previously used methodologies for the determination of leaf deposited PM, in order to determine which method is the most suitable for standardised use in future studies.

2.4: CONCLUSION & FUTURE DIRECTIONS

This chapter assessed a representative sample of common living wall plant species with a range of different morphologies for their impact on PM deposition, along with their abilities to accumulate PM from the air. All species tested accumulated airborne PM, however, the amount of deposited PM was not uniform across the species. Contradictory to previous studies, no specific leaf traits were found to strongly influence PM deposition. Whilst it is important to determine the plant characteristics that are influential on PM deposition so as to maximise the PM removal performance of green walls, it remains difficult to do so. Plant species possess many different characteristics, making it difficult to attribute their PM accumulation capacity to any specific trait. Nonetheless, species *C. comosum variegatum*, *N. glabra*, *P. xanadu*, and *S. wallisii* demonstrated consistent, high PM deposition, making these species functionally applicable for green wall use to reduce ambient PM concentrations.

Additionally, the selection of green wall species for combined pollutant removal is also unresolved. For example, Weerakkody et al. (2017) discovered that two of their lower performing PM capturing species were ferns. Ferns have however, proven to be an excellent species for VOC removal (Kim et al., 2008) and their fibrous root systems have shown to be

significant in filtering PM when used in an active green wall system (Pettit et al., 2017). Additionally, whilst the PM removal observed in the current project for the *Chlorophytum* species were not amongst the highest performing species; Spider plants have been previously shown to be capable of removing significant amounts of SO_x and formaldehyde from the air (Godish and Guindon, 1989, El-Sadek et al., 2012). This raises the question of whether species selection based on the more prominent pollutant type in a location may be the most effective decision, a concept which deserves more research focus.

Whilst green walls demonstrate potential for ambient PM pollution removal, with considerable effects on smaller PM size fractions (Weerakkody et al., 2017); morphological and physiological damage to the vegetation can occur from PM accumulation (Daresta et al., 2015; Rai, 2016). For example, stomatal blockage from PM accumulation can result in decreased gas exchange and stomatal conductance, which can alter the water regime, photosynthesis (Farmer, 1993) and plant growth as a whole (Rai et al., 2010). It is therefore important that plant health assessments be conducted to determine potential pollutant tolerant species for use in high PM pollution environments.

Chapter 3 – Pollution tolerance of green wall plants.

ABSTRACT

Air pollution exposure can impact plant physiology, morphology and biochemistry, leading to dramatic alterations to plant systems, function and growth. The use of plants for air pollution mitigation is increasing in popularity, particularly in the form of green wall systems, making the identification and classification of pollution sensitive and tolerant species essential. This chapter examined a range of *in situ* green wall species health parameters in response to ambient air pollution exposure. To do this, 11 plant species were sampled across 15 green wall sites, over a 6 month duration, and were tested for leaf chlorophyll content, pH, relative water content and carbon content. Linear mixed models were used to examine patterns in plant health traits across species, to potentially identify tolerant species that would be suitable for use in high pollution environments. Significant differences were found between species in each of the plant health parameters between the pollution exposed plants and the control, nonexposed plants. The individual species response across the health parameters however, was not consistent. As such, there is no clear distinction of the most tolerant species. As most species showed no significant health differences from pollution exposure, it is reasonable to conclude that all test species were able to withstand pollution exposure without any adverse effects.

3.1: INTRODUCTION

3.1.1 Green wall use for pollution removal

Green infrastructure including green walls and green roofs (Abhijith et al., 2017) show potential as a solution to urban air pollution (Irga et al., 2015; Salmond et al., 2016). This is due to the plants porous nature which increases airborne pollution removal and deposition as well as influencing the local dispersion patterns of the air pollutants (Nowak et al., 2006; Escobedo and Nowak, 2009; Yin et al., 2011; Fantozzi et al., 2015). Furthermore, various innate plant processes such as detoxification, adsorption, absorption and accumulation allow plants to act as 'living filters' without causing acute foliar damage (Garbisu et al., 2002; Jim and Chen, 2008). The leaf stomata and plant surfaces act as the main site for pollutant absorption (Escobedo and Nowak, 2009; Fantozzi et al., 2015; Salmond et al., 2016; Vesa Yli-Pelkonen et al., 2017), whereby gaseous air pollutants are absorbed and metabolized into less toxic forms (Gupta and Kulshrestha, 2016). Plants can also accumulate PM on their leaf surfaces, realistically filtering the air by this process (Chapter 2, Shi et al., 2017). With green walls, plants are situated vertically on pre-existing building walls, increasing the potential for pollution reduction. Green infrastructure has therefore been recognised as a passive air pollution abatement system which requires minimal adjustments to the built environment (McNabola, 2010), making implementation of this technology desirable.

3.1.2 The impact of air pollution on plant processes

Plant physiology, morphology and biochemistry are all affected by both particulate and gaseous air pollutants (Rai et al., 2010), with direct impacts on stomatal conductance, leaf cuticles, carbon allocation, respiration and photosynthetic systems (Darrall, 1989; Saxena and Kulshrestha, 2016). Previous studies have recorded air pollution effects on leaf pH (Scholz and Reck, 1977; Klumpp et al., 2000; Joshi and Bora, 2011), relative water content (Rao, 1979,

Agrawal et al., 1991; Karthiyayini et al., 2005; Joshi & Swami, 2009; Krishnaveni, 2013; Marimuthu et al., 2014) and chlorophyll content (Flowers et al., 2007). Under stress conditions, plants respond with various anatomical, physiological and morphological changes (Inamdar and Chaudhari 1984; Iqbal, 1985; Gravano et al., 2003; Dineva, 2004), and as such these 'health' parameters can be used to indicate a plants resistance or sensitivity to air pollution (Pandey et al., 2016).

Plant species are usually classified as either 'sensitive', showing visual signs of lesions and malformations from pollutant exposure, or 'accumulator' species, in which pollutants are collected on or within the plant itself (Rai, 2016). Whilst some species can flourish in polluted environments (Rai, 2016); sensitive plants experience pathological effects (Tiwari et al., 2006). The identification of sensitive and tolerant species is therefore essential, as tolerant species can be implemented in high pollution environments, whilst sensitive species could be used as a form of early detection for high ambient air pollution (Prajapati and Tripathi, 2008).

3.1.3 The impact of pollutants on chlorophyll content

Chlorophyll facilitates the conversion of gaseous precursors to chemical energy (Kaur and Nagpal, 2017) and as such, the photosynthetic apparatus is one of the most likely plant structures to be damaged by air pollution (Prusty et al., 2005; Tripathi and Gautam, 2007; Joshi and Swami, 2009; Honour et al., 2009). Air pollutant stress renders chloroplasts vulnerable to the generation of reactive oxygen species (ROS) and thus oxidative stress (Woo et al., 2007), which is cytotoxic (Pukacha and Pukacha, 2000). When ROS are produced, chloroplast membranes suffer lipid peroxidation resulting in a loss of chlorophyll (Dhir, 2016). More specifically, acidic air pollutants can cause pheophytin formation which changes the light spectrum affinity of chlorophyll (Saxena and Kulshrestha, 2016) and blocks stomata, resulting in chlorophyll degradation (Rao and Leblanc, 1966; Anthony, 2001; Joshi and Bora, 2011).

Thus, under stress conditions chlorophyll content usually decreases (Speeding and Thomas, 1973); with the resulting changes to photosynthesis having detrimental impacts on plant productivity and growth (Tiwari et al., 2006; Woo et al., 2007; Liu and Ding, 2008). Plants that are able to maintain their chlorophyll content during polluted conditions are considered tolerant (Singh and Verma, 2007).

3.1.4 The impact of pollutants on leaf extract pH

Cellular pH regulation is critical for the intracellular movement of various small molecules, vesicles and proteins (Sharma et al., 2013; Ogunkunle et al., 2015; Pandey et al., 2015; Zhang et al., 2016). When acidic, gaseous pollutants such as CO_2, NO_2 and SO_2 enter plant cell sap, they react with cellular water forming acid radicals, altering leaf pH (Kaiser et al., 1993; Swami et al., 2004; Das and Prasad, 2010), leading to ionic disturbances and thylakoid membrane swelling (Pukacki, 2000). Leaf acidification is more profound in sensitive species (Rai, 2016), as it retards the conversion of hexose to ascorbic acid (Escobedo et al., 2008; Jyothi and Jaya, 2010), lowering the plants' capacity to mitigate the effects of ROS.

3.1.5 The impact of pollutants on relative water content

Relative water content (RWC) in plants refers to the physiological balance between water uptake and release (Sen and Bhandari, 1978; Jones, 1994), which influences respiration, growth and transpiration (Dhankhar et al., 2015). Thus, high RWC assists the maintenance of plant physiological balance during air pollution stress (Singh and Verma, 2007). Leaf protoplasmic permeability is associated with RWC, hence higher RWC values are indicators of pollutant tolerance (Singh et al., 1991). A higher RWC can also dilute cell sap acidity (Palit et al., 2013) and otherwise support physiological balance during stress conditions (Dedio, 1975; Ritchie et al., 1990; Chaves et al., 2003; Geravandia et al., 2011; Sharma et al., 2013; Ogunkunle et al., 2015; Pandey et al., 2015; Zhang et al., 2016).

3.1.6 The impact of pollutants on carbon allocation

Stomatal closure due to air pollution stress decreases CO_2 availability, impeding carbon fixation (Honour et al., 2009; Saxena and Kulshrestha, 2016) and changing the carbon allocation throughout the plant (Darrall, 1989; Wolfenden and Mansfield, 1990). This further affects the dark and light reactions of photosynthesis (Farage and Long, 1999; Shavin et al., 1999; Castagna et al., 2001). Pollutant exposure injury can also inhibit or stimulate leaf respiration (Gupta, 2016). SO_2 and NO_2 exposure under ambient CO_2 concentrations prevents stomatal closure, whereas under increased CO_2, stomatal closure is promoted (Atkinson et al., 1991). Whilst the effects of respiration changes on plant carbon balance are not profound in plants which exhibit high photosynthetic rates; they can result in yield and growth changes in slower photosynthesising species (Koziol and Whatley, 2013). As such, plants carbon content provides a good indication of pollutant exposure effects on overall plant growth and health.

3.1.7 Aim

Whilst air pollutant exposure effects on plants have received significant research, most previous studies are limited to southern Asia (i.e. Prajapati and Tripathi, 2008; Govindaraju et al., 2012; Krishnaveni, 2013; Rai et al., 2013; Rai and Panda, 2014). Furthermore, most studies have focussed on roadside species as bio-indicators of air pollution, or tree and shrub selection for green belt development; and have tested a small number of species over time periods insufficient to represent seasonal changes. To the author's knowledge, no studies have examined green wall species health responses to air pollution *in situ*. A previous experiment by the authors (Paull et al., 2018) examined the health effects on 8 green wall species to intermittent, high concentration diesel exhaust over a 5 week duration. Whilst the findings provided an indication of pollution exposure health effects and tolerant species identification,

they were not representative of *in situ* conditions, where green walls are continuously exposed to pollutants of varying concentrations.

This project aimed to examine the health of common green wall species in response to pollution exposure *in situ*, and thus to identify tolerant species for use in high pollution environments.

3.2: METHOD

3.2.1 Sample Sites

The same 15 sites within the Sydney urban region containing a passive outdoor Junglefy green wall, as described in Chapter 2 Section 2.2.1 (see Table 1) were sampled for this chapter. The same 11 species described in Chapter 2 Section 2.2.3 (see Table 2) were also used for this work, and are summarised in Table 4. Each site differed in its pollution and environmental conditions, thus providing a good comparison for species differences in health parameters.

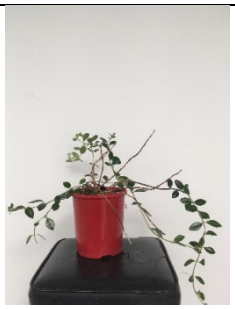
3.2.2 Sample Species

Eleven different common green wall species found within the green walls were selected for plant health testing (Table 4). Whilst not all species were present in every green wall, all species were sufficiently spatially distributed to provide a range of ambient conditions in all cases.

Procedural control plants of the same species as the test plants were obtained from the Junglefy Pty (Ingleside, Sydney) nursery and were of the stock that was equivalent to that used in the *in situ* commercial green walls. Plants were supplied in the same substrate as the *in situ* green walls. Control plants were maintained in a glasshouse lined with shade cloth, with an average temperature of 23.7 ± 3.6 °C, relative humidity of 68.1 ± 16.0 %, and a maximum mid-day light level of 90 ± 10 $\mu mol.m^{-2}.s^{-1}$ (4860 ± 54 lux). All plants were watered once weekly

(equivalent to the *in situ* samples) and their position within the greenhouse was randomized on a weekly basis.

Table 4: Test species general information.

Scientific Name	**Common Name**	**Image**	**Phylogenic Group**	**Replication Across Sites**
Philodendron xanadu	Xanadu		Eudicot	8
Peperomia obtusifolia	Baby Rubber Plant		Eudicot	7
Plectranthus madagascariensis	Variegated Mintleaf		Eudicot	3
Nematanthus glabra	Goldfish Plant		Eudicot	4

Nandina domestica	Pink Blush		Eudicot	3
Neomarica gracilis	Walking Iris		Monocot	9
Nephrolepis exaltata bostoniensis	Boston Fern		Monilophyte	4
Chlorophytum comosum variegatum	Variegated Spider Plant		Monocot	6

Chlorophytum comosum	Spider Plant		Monocot	3
Spathiphyllum wallisii	Peace Lily		Monocot	8
Peperomia glabella	Small Leaf Peperomia		Eudicot	3

3.2.3 Sampling Process

Leaf sampling was conducted from each green wall monthly for 6 consecutive months, starting in June 2017 and concluding November 2017. A minimum of 15 young mature healthy leaves of each species were collected from each location and stored in a large sample bag with moist paper towel to minimize dehydration. An additional 5 leaves were collected for RWC testing which were individually inserted into pre weighed bags without any paper towel to provide accurate respiration values. Once at the laboratory, samples were stored at 4°C and processed within 48 hours. Leaves collected for RWC were analysed as soon as possible, whilst leaves

collected for the remaining plant health tests were cleaned, weighed, frozen in liquid nitrogen and stored at -80°C until analysis.

3.2.4 Plant Health Tests

3.2.4.1 Relative Water Content Determination

RWC was determined by first weighing the fresh leaf content, then immersing the leaf into water and leaving it overnight, before dabbing dry and reweighing the next day to provide the turgid weight. The leaf was then oven dried at 80°C for 48 h and the dry weight recorded (Henson et al., 1981). From the weights obtained, the relative water content was derived using Formula 1:

Formula 1: RWC = (FW-DW) / (TW-DW) * 100

Where: RWC = Relative water content (%), FW = Fresh weight (g), DW = Dry weight (g), TW = Turgid weight (g).

3.2.4.2 Chlorophyll Content Determination

Leaf material (0.5 g) was ground in a mortar and pestle and transferred to a 15 mL centrifuge tube with 10 mL of chilled 80% acetone. The solution was then extracted for 15 minutes before centrifugation at 2,500 rpm for 3 min, followed by spectrophotometric analysis at 645 and 663 nm (Arnon, 1949). Total chlorophyll content was calculated using Formula 2:

Formula 2: Total chlorophyll content = Chlorophyll a + b mg/g

Chlorophyll a = [(12.7 * DX 663) – (2.69 * DX 645)] * [V / (1000 * W)] mg/g

Chlorophyll b = [(22.9 * DX 645) – (4.68 * DX 663)] * [V / (1000 * W)] mg/g

Where: DX = Absorbance of the extract at the stated wavelength (nm), V = Total volume of the chlorophyll solution (mL), W = Weight of the tissue extracted (g).

3.2.4.3 Leaf pH Determination

Leaf pH was measured by grinding 0.5 g of leaf sample in 50 mL of de-ionized water, followed by centrifugation at 2,500 rpm for 10 min at 21°C, and quantification with a digital pH probe (Singh and Rao, 1983).

3.2.4.4 Leaf Carbon Content Determination

Oven dried leaves (1 g; 80°C for 48 h) were dry ashed in a muffle furnace for 3 h at 550°C (Baxter et al., 2014). From these data, leaf carbon content could be determined using Formula 3:

Formula 3: LOI = [(DW-AW) * 100 / (DW)]

Where: LOI = Loss on Ignition (%), DW = Dry weight (g), AW = Ash weight (g).

3.2.5 Air Quality and Environmental Variable Determination

3.2.5.1 Green Wall Total Suspended Particulates

To determine the total suspended particulate (TSP) concentration at the green wall sites, a DustTrack II 8532 laser densitometer (TSI, Shoreview, Minnesota) was used to record 3 minute time weighted averages for TSP, monthly at the test sites.

3.2.5.2 Traffic Density

Traffic density was quantified at the closest intersection to the green walls for a 30 minute duration. Traffic was classified based on vehicle type to attain average traffic density estimates for cars, buses, trucks and motorcycles. Days of the week and times at which air quality and traffic density tests were conducted were limited to weekdays between 10 a.m. and 3 p.m. to avoid peaks caused by work and school commuters (Irga et al., 2015), and randomized amongst sites and months.

3.2.5.3 Green Wall Temperature

Temperature readings were taken using a handheld Digitech Multifunction Environment Meter, at 4 evenly distributed sections across the green wall (one towards either end of the green wall and two spaced in the middle of the green wall). This was done 0.5 m in front of the green wall and reference wall at each site, once a month for a 6 month duration. The Environment Meter was allowed to stabilize for a 60 second duration at each sample interval before recording the result. The average and standard error were then determined using the 4-point samples.

3.2.5.4 Relative Humidity

The mean monthly relative humidity at the green wall sites was recorded using the Bureau of Meteorology Data for each month of the study. The mean monthly data was recorded to provide an accurate representation of relative humidity incorporating daily temperature fluctuations.

3.2.5.5 Accumulated Leaf PM

The accumulated leaf PM values recorded in Chapter 2 were also used in the current chapter to determine if there were any correlations between PM deposition and plant health. Accumulated leaf PM was determined as previously described in Chapter 2, Section 2.2.3.

3.2.6 Statistical Analysis

The statistical approach used focused on species specific patterns so as to identify which species were more or less pollution tolerant on the basis of the plant health variables. In order to achieve this, all models incorporated interaction terms containing species as a predictor. The first suite of models tested for differences in the four health variables between control plants and the plants from the green wall sites. LMMs were fitted with a term for species (11 level categorical fixed factor), treatment (2 levels; control and green wall plants), a species x

treatment interaction term, along with a random term for site, included to control for repeated measures (13 levels, 12 for the sites, one for the control group). In the random term, control plants were all assigned to the same factor level. Where the treatment term, or the species x treatment term was significant, *post hoc* tests were carried out comparing control to green wall plants within species. For each species, ten replicate measurements were used for each site and for the control group.

The relationship among health variables for the green wall plants was then examined using bivariate LLMs, testing the association between all unique combinations of the health variables in six separate models. These models were fitted with the first plant health variable of interest as the response, and the second health variable of interest as predictor, including a term for species (11 level categorical fixed factor), and a species x second health variable interaction term, to test if relationships were consistent across species. A random intercept for site (12 levels) was again included in these models. Where the interaction term was non-significant, models were refit without the interaction term, to best model the relationships between the variables across species. Here, and in subsequent models, six replicate values per site per species were used in the modelling, one replicate for each month of the study duration. Where a significant interaction was found, 95% confidence intervals were estimated for the coefficients of the response and continuous predictor for each species, and determined to be statistically significant when the confidence interval did not contain zero.

Next, differences within species in the species health variables amongst sites was tested. In this case, linear models were employed given that the differences among sites were the primary interest. The models used terms for species, site, and a species x site interaction term. Where this interaction term was significant, pairwise tests were employed within species to determine which species generated the interaction (i.e. sites contrasted within levels of species to determine which species health variables differed among the sites where they were located).

As species health differences amongst sites was the focus of these models, and the fact that the sites featured different species, interpretation of the site term was not attempted.

Finally, to understand how plant health may be related to site conditions, relationships between green wall plant health and site conditions were tested. The four health variables were regressed against four site environmental variables (green wall TSP, mean traffic density, temperature and relative humidity), along with a leaf level indicator of site air quality (accumulated leaf PM from Chapter 2). Using LMMs, the health variables were modelled using a term for species, the continuous environmental variable of interest, and a species x environmental variable/leaf PM interaction, which was removed where non-significant to model the across-species relationship between the health variables and environmental/leaf PM conditions.

Prior to analysis, to improve variance homogeneity, leaf chlorophyll and leaf pH were log transformed, and RWC and leaf carbon content were logit transformed. Of the site environmental/leaf PM variables, green wall TSP and accumulated leaf PM were square root transformed, mean traffic density and green wall temperature were log transformed, and relative humidity logit transformed. All results reported used marginal means (Type II Wald chi squared tests for LLMs, and type II ANOVA for the linear models) to account for the unbalanced nature of the data. All analyses and graphical presentations of the data were performed in R 3.6.1 (R Core Team, 2014), using the packages lme4 (Bates et al., 2015), lmerTest (Kuznetsova et al., 2017), emmeans (Length, 2019) and car (Fox and Weisberg, 2011).

3.3: RESULTS

3.3.1 Plant health variables

There was a significant difference for species health variables between the test sites exposed to pollution and the control sites not exposed to pollution. For example, chlorophyll content was found to be significantly higher in four species (*Chlorophytum comosum variegatum*, *N. glabra*, *N. gracilis and S. wallisii*) at the green wall sites (exposed to pollution) compared to the controls, which were not exposed to pollution (species x site interaction: $\chi^2_{10} = 195.1$, $P < 0.0001$; Fig. 8a), while in *C. comosum* and its cultivar *C. comosum variegatum*, plants at the green wall were more acidic than controls (species x site interaction: $\chi^2_{10} = 41.5$, $P < 0.0001$; Fig. 8b). Three species, *N. exaltata bostoniensis*, *N. gracilis* and *S. wallisii* were found to have significantly lower RWC compared to control plants (species x site interaction: $\chi^2_{10} = 37.9$, $P < 0.0001$; Fig. 8c), with three species, *P. madagascariensis*, and the closely related *P. glabella* and *P. obtusifolia*, having lower leaf carbon content (species x site interaction: $\chi^2_{10} = 46.5$, $P < 0.0001$; Fig. 8d) than the controls.

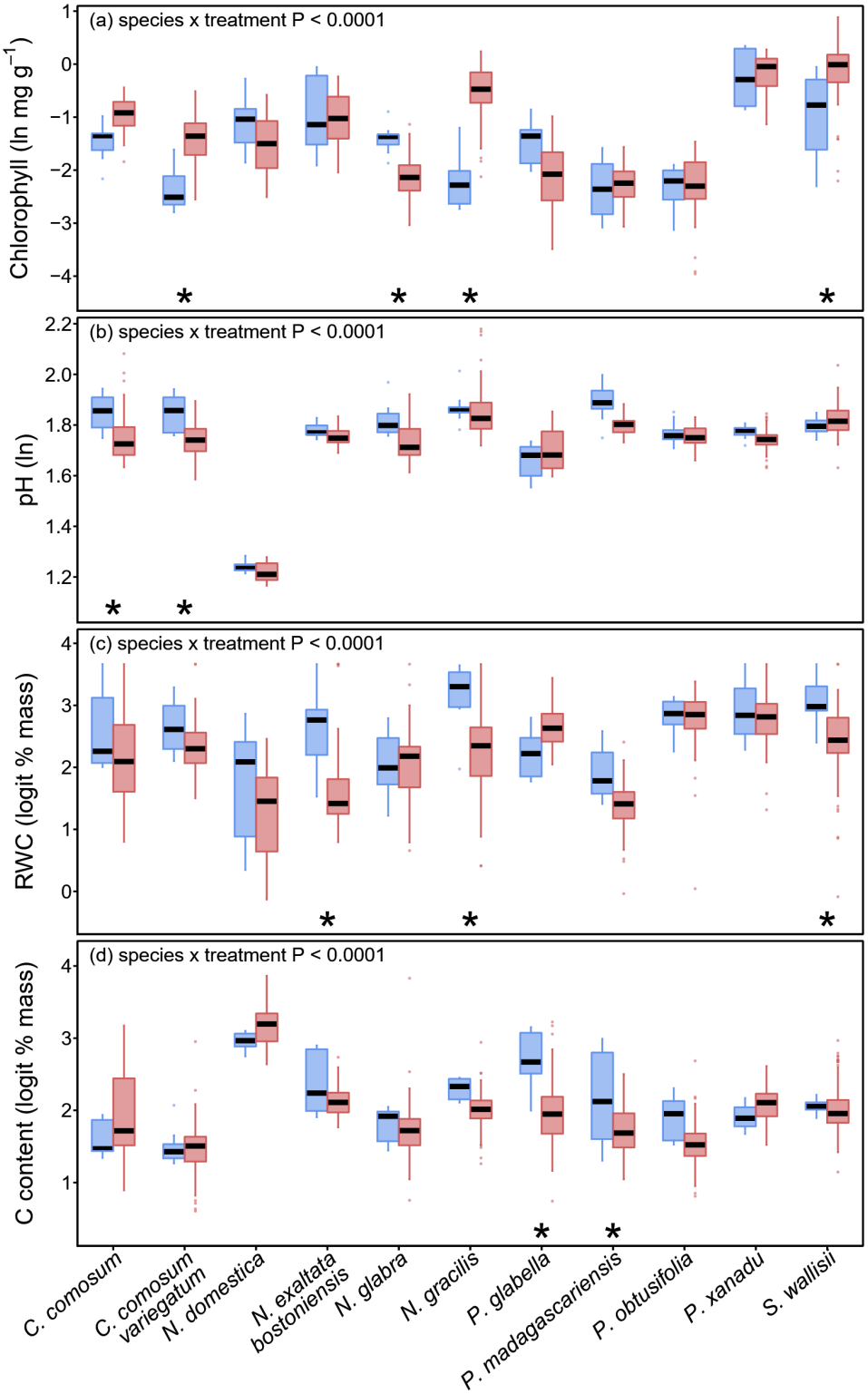


Figure 8: Boxplots of leaf chlorophyll (a), leaf pH (b), RWC (c) and leaf carbon content (d) by species. Blue boxplots indicate control plants, red boxplots the plants at the green walls. Asterisks are shown below species where a significant effect in *post hoc* tests comparing control group and green wall plants was found. The *p*-values for the species x treatment interaction term from the models is presented in each plot at top left.

A significant association between lower RWC and higher leaf carbon content was found across species (χ^2_1 = 4.8, P = 0.029; Fig. 9), and two species displayed associations between lower RWC and lower leaf chlorophyll concentration (*P. glabella* and *S. wallisii*), with no other species showing a significant relationship amongst the significant species x RWC interaction tests (χ^2_{10} = 19.8, P = 0.031; Fig. 9). No significant association was found between leaf chlorophyll concentration and pH (χ^2_1 = 3.5, P = 0.06; Fig. 9) or leaf chlorophyll and leaf carbon content (χ^2_1 = 0.7, P = 0.4; Fig. 9), pH and RWC (χ^2_1 = 0.6, P = 0.4; Fig. 9) or pH and leaf carbon content (χ^2_1 = 1.7, P = 0.2; Fig. 9). In all six models, a significant effect of species was found (full results presented in Appendix 2).

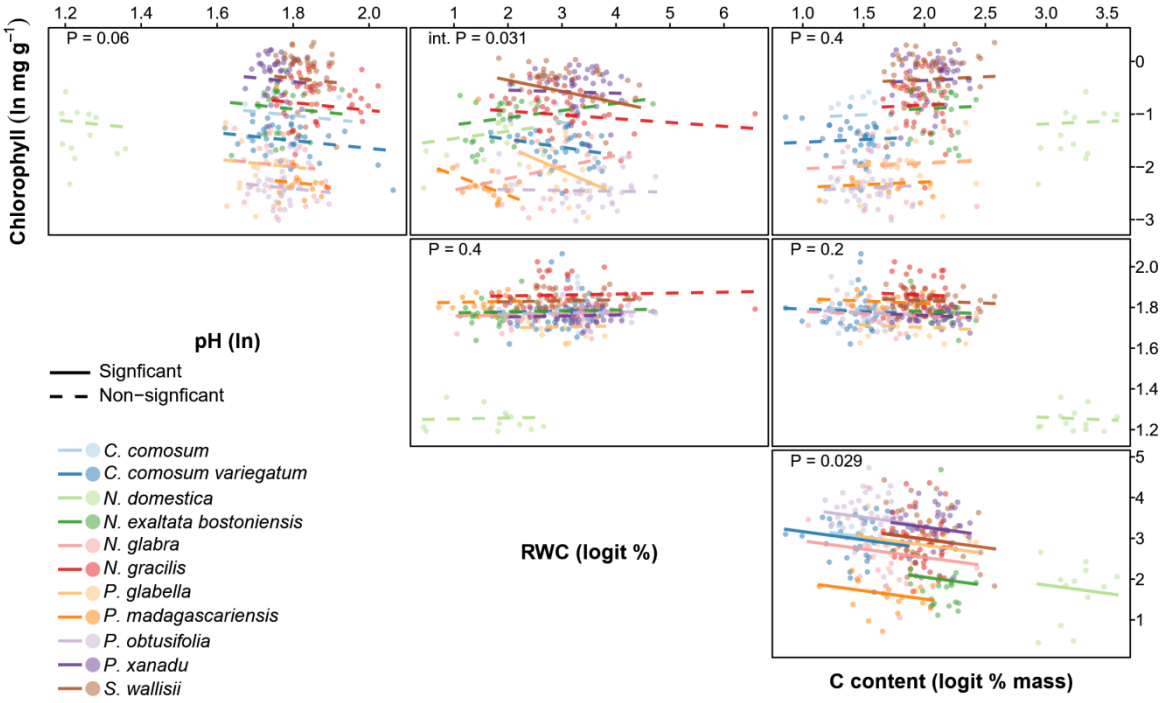


Figure 9: Scatterplots of the unique combinations of plant health variables. Lines are the fitted coefficients for individual species, coloured as per the key at lower left. Solid lines are used to indicate significant relationships, broken lines for non-significant relationships. The *p*-value for the relationship between the two variables is shown in the top left corner, with the exception of leaf chlorophyll against RWC, where the *p*-value reports the interaction term.

3.3.2 Differences amongst sites, and the effect of environmental factors on plant health

Differences among sites in leaf chlorophyll were found for five species (*N. domestica*, *N. exaltata bostoniensis*, *N. glabra*, *P. glabella* and *P. xanadu*), generating a significant species x site interaction ($F_{27,245} = 1.9$, $P = 0.006$; Fig. 10a). No significant species x site interaction was found for pH ($F_{27,245} = 0.6$, $P = 0.97$; Fig. 10b), or RWC ($F_{27,245} = 1.2$, $P = 0.2$; Fig. 10c). The species *C. comosum variegatum*, *P. xanadu* and *S. wallisii* were found to differ in carbon content across sites (species x site interaction: $F_{27,245} = 2.2$, $P = 0.0007$; Fig. 10d). In all models, the species and site terms were significant (full results presented in Appendix 3).

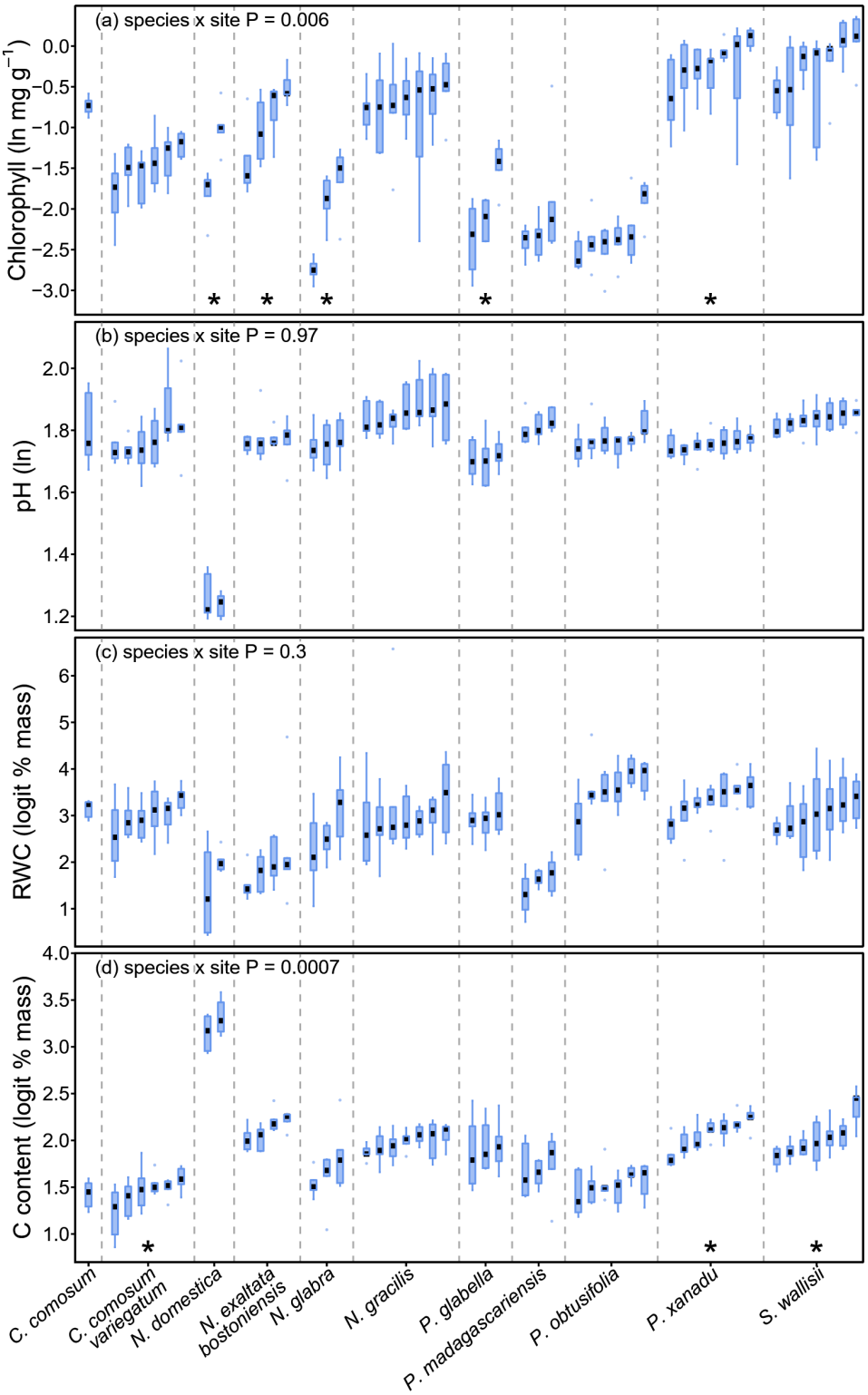


Figure 10: Grouped boxplots of plant health variables by species, showing leaf chlorophyll (a), leaf pH (b), RWC (c) and leaf carbon content (d). Each boxplot represents a site, with broken vertical lines separating species. Asterisks at the bottom of plots indicate species for which one or more significant pairwise comparisons amongst sites was found. At the top left of each plot the *p*-value for the site x species interaction term is presented.

Leaf chlorophyll was not significantly related to TSP ($\chi^2_1 = 0.2$, P = 0.7; Fig. 11a), accumulated leaf PM ($\chi^2_1 = 0.1$, P = 0.8; Fig. 11b), mean traffic ($\chi^2_1 = 0.2$, P = 0.6; Fig. 11c), green wall temperature ($\chi^2_1 = 0.5$, P = 0.5; Fig. 11d), nor humidity ($\chi^2_1 = 0.001$, P = 0.97; Fig. 11e). Higher pH was found to be related to higher TSP ($\chi^2_1 = 4.1$, P = 0.044; Fig. 11f), and lower green wall temperature ($\chi^2_1 = 10.6$, P = 0.001; Fig. 11i) across species, with no significant relationship found between accumulated leaf PM ($\chi^2_1 = 0.1$, P = 0.7; Fig. 11g), mean traffic ($\chi^2_1 = 1.9$, P = 0.2; Fig. 11. f h), or humidity ($\chi^2_1 = 1.5$, P = 0.2; Fig. 11j). Several significant associations between RWC and site conditions emerged with greater TSP ($\chi^2_1 = 21.2$, P < 0.0001; Fig. 11k) and accumulated leaf PM ($\chi^2_1 = 16.2$, P < 0.0001; Fig. 11l), both found to be significantly associated with higher RWC, and higher green wall temperature significantly related to lower RWC ($\chi^2_1 = 30.1$, P < 0.0001; Fig. 11n), with no significant relationship found between RWC and traffic ($\chi^2_1 = 0.001$, P = 0.95; Fig. 11m). Three species showed a significant association between greater humidity and higher RWC; *N. glabra*, *N. gracilis* and *S. wallisii*, with no other species showing a significant relationship, nonetheless generating a significant species x humidity interaction ($\chi^2_{10} = 25.4$, P = 0.005; Fig. 11o). Lower leaf carbon content was significantly related to lower RWC across species ($\chi^2_{10} = 5.7$, P = 0.017; Fig. 11t), while differing responses across species to green wall temperature drove a significant species x green wall temperature interaction ($\chi^2_{10} = 23.0$, P = 0.011; Fig. 11s), with two species *P. glabella* and *P. madagascarensis* showing significant positive relationships between leaf carbon content and green wall temperature. No significant relationships were found between leaf carbon content and TSP ($\chi^2_1 = 0.01$, P = 0.9; Fig. 11p), accumulated leaf PM ($\chi^2_1 = 0.02$, P = 0.9; Fig. 11q), nor traffic ($\chi^2_1 = 0.3$, P = 0.3; Fig. 11r). In all models the species term was significant (full results presented in Appendix 4).

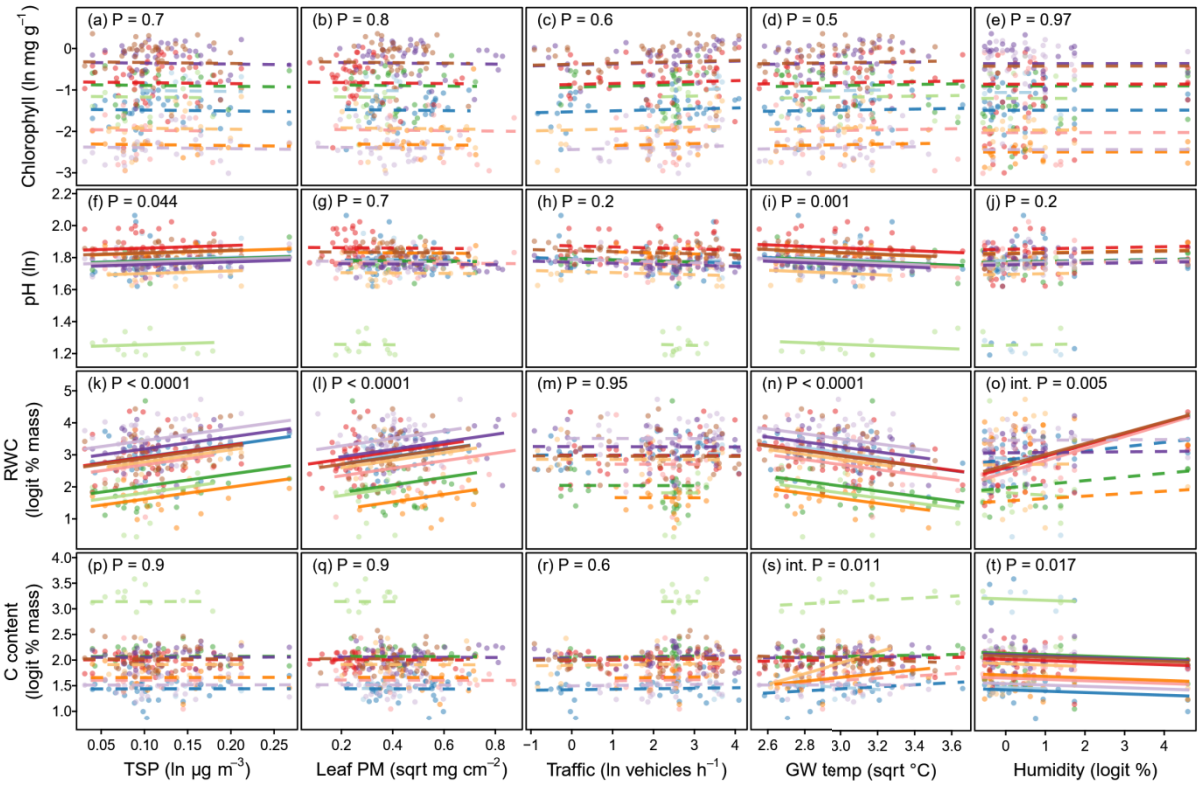


Figure 11: Plots of the plant health variables leaf chlorophyll (a–e), Leaf pH (f–j), RWC (k–o) and leaf carbon content (p–t), against the site variables: green wall TSP, accumulated leaf PM, traffic, green wall temperature, and humidity. Lines are the fitted coefficients for individual species, broken lines indicate non-significant effects, solid lines significant effects. Where the species x site variable interaction was significant (plots o and s), the interaction term *p*-value is given at top left, for all other plots, the *p*-value shown is for the relationship between the response and the site variable, from models where the non-significant interaction term has been removed. Colours used to indicate species are the same as in Fig. 9.

3.4: DISCUSSION

3.4.1 The effect of site differences on plant health

In the current project, no significant differences were observed across sites for plant leaf pH or relative water content. Differences amongst sites in leaf chlorophyll content were found for only five species (*N. domestica*, *N. exaltata bostoniensis*, *N. glabra*, *P. glabella* and *P. xanadu*), and *C. comosum variegatum*, *P. xanadu* and *S. wallisii* were found to differ in their carbon content across sites. Previous studies have indicated that site specific differences in plant health may be due to the physiological damage which differs depending on species' sensitivity and

pollutant exposure (Agbaire and Esiefarienrhe, 2009; Rai et al., 2009). However, the aforementioned species were not found to be significantly different between the control and pollution sites, which is indicative of no adverse health effects arising from pollution exposure. The tolerance of a plant to air pollution is dependent on the level and type of pollution, which is site specific (Noor et al., 2015), however as no site specific differences were observed in particulate matter concentrations, this is unlikely to be the reason for these site specific species differences, and instead is more likely to be the reason for the non-significance observed for site differences in leaf pH and relative water content. Chlorophyll content can vary due to factors other than pollution levels, such as leaf age, biotic and abiotic conditions (which can be different across locations) (Katiyar and Dubey, 2001; Rai and Panda, 2015), potentially explaining the difference observed in these species. Further, PM removal from leaf surfaces due to rainfall is thought to result in increasing chlorophyll content (Shyam et al., 2006), and environmental stressors including drought, daylight intensity, salt stress, heavy metal substrate pollution and high ambient temperatures can affect species chlorophyll content (Pandey et al., 2015; Zhang et al., 2016). It is possible that some of these variables affected the listed species' chlorophyll and carbon contents, generating the site specific differences observed.

As shown here, site specific plant health differences can exist, and as such, despite a species being classified as tolerant or sensitive within a certain geographical region, it may exhibit variant responses on a small spatial scale as a result of both different pollutant or environmental conditions (Raza et al., 1985). Thus, it may be of interest to conduct further research over a wider geographical scale to encompass a range of pollutant and environmental conditions.

3.4.2 The effect of monthly difference on plant health

There were inconsistencies in the environmental variables that were influential on plant health responses, with leaf chlorophyll content being not significantly correlated with TSP, accumulated leaf PM, traffic density, green wall temperature or humidity. Higher leaf pH was associated with higher TSP and lower green wall temperature. Higher RWC was associated with greater TSP and accumulated leaf PM, and was lower when green wall temperatures were higher. *N. glabra*, *N. gracilis* and *S. wallisii* saw higher RWC with higher humidity. *P. glabella* and *P. madagascarensis* demonstrated higher leaf carbon content at higher green wall temperatures. Ambient pollution levels vary across seasons due to both emission patterns, and environmental factors such as rain, temperature and humidity (Kaur and Nagpal, 2017), with differences in seasonal pollution levels influencing plant health responses (Abeyratne and Illeperuma, 2006). For example, if environmental factors such as temperature and light are limiting and already affecting plant photosynthesis; if a plant is then further exposed to high pollutant concentrations, photosynthesis can be significantly reduced (Gupta, 2016), which can then alter respiration rates, severely changing plant carbon balance, leading to premature senescence (Gupta, 2016). Both leaf extract pH and RWC had their lowest values in November, the hottest month, with temperature clearly impacting these two water dependent health variables. Chlorophyll and carbon content were maximal in November and October respectively, reflecting rapid Spring plant growth. Clearly not all environmental conditions affect plant health variables consistently, however overall, the test species showed little negative effect from the *in situ* conditions.

3.4.3 The impact of pollutants on chlorophyll content

In the current project a chlorophyll content range of 0.114 – 0.874 mg/g was observed, which is slightly lower than found in previous studies such as Kaur and Nagpal (2017) who noted a total chlorophyll content range of 0.444 – 1.817 mg/g. Rai and Panda (2014) noted that chlorophyll content was lower at their polluted site than at a control site; with 1.24 and 6.60 mg/g chlorophyll being their lowest and highest values for polluted and control sites respectively. However, in the current project, no significant difference in chlorophyll content was observed between the test site and controls for 7 of the species tested. This could be due to a reasonable level of innate pollution tolerance amongst the selected species negating pollutant induced stress at the test locations. It is alternatively possible that the pollutant conditions of the test sites were not as extreme as the test sites used in other studies.

Pollution exposure has been found to negatively impact plant chlorophyll content, with previous studies (i.e. Raina and Sharma, 2003; Swami et al., 2004 and Kaur and Nagpal, 2017) discovering significantly lower total chlorophyll in plants situated in high pollution study sites. This effect can result from many factors including an inhibition of chlorophyll biosynthesis (Esmat, 1993); chloroplast damage (Pandey et al., 1991) or enhanced chlorophyll degradation (Rai, 2016). Alternatively, under stress conditions plant protective processes may compensate, leading to cellular destruction such as pigment degradation (Senser et al., 1990). The degradation of chlorophyll has rippling effects throughout the plant, impacting productivity, stomatal conductance and photosynthetic rates (Tripathi and Gautam, 2007; Mir et al., 2008; Giri et al., 2013; Bora and Joshi, 2014).

Thus, whilst the trend in previous studies appears to indicate chlorophyll reductions from pollution exposure, this was not observed in the current project. *C. comosum variegatum*, *N. glabra*, *N. gracilis* and *S. wallisii* were found to have an increased chlorophyll content at the

polluted sites compared to the controls. Some studies have seen an increase in chlorophyll content from pollution exposure, such as in the Red Gum in Seyyednejad and Koochak's (2011) study, and in Mangoes in Tripathi and Gautam's (2007) study. As increased chlorophyll content in response to pollution has previously been associated with pollution tolerance (Beg et al., 1990; Shannigrahi et al., 2003; Singh and Verma, 2007; Prajapati and Tripathi, 2008; Jyothi and Jaya, 2010; Pathak et al., 2011; Sharma et al., 2013; Rai and Panda, 2014; Ogunkunle et al., 2015), it can be suggested that *C. comosum variegatum*, *N. glabra*, *N. gracilis* and *S. wallisii* are pollutant tolerant species.

3.4.4 The impact of pollutants on leaf extract pH

Leaf extract pH varied significantly amongst species, and was most notable in *N. domestica* which had the lowest pH of 3.42. However, the pH of this species was not significantly different between the test and control sites, indicating that pollution exposure did not play a role in this species' low pH. Nonetheless, the naturally low pH of this species may make it more susceptible to damage from acidic air pollutants (Escobedo et al., 2008; Jyothi and Jaya, 2010). Previous plant pollution tolerance studies recorded pH ranges of 5.46 – 6.48 (Kaur and Nagpal, 2017) 4.6 – 6.7 (Aji et al., 2015) and 5.89 – 6.37 (Tsega and Prasad, 2014). In the current project a pH range of 3.42 – 6.4 was observed. In contrast to previous literature, no significant differences were observed between the leaf extract pH at the test sites and controls for 9 of the tested species. This was probably due to either the pollution tolerance of the selected species, or the pollutant conditions in the current work not being as extreme as the test sites used in other studies. For example, in Rai and Panda (2014) the test species leaf pH levels were more acidic at their polluted site than the control site; but with their study conducted in the Indo-Burma hot spot region for air pollution.

C. comosum and *C. comosum variegatum* both demonstrated lower pH values at the test locations compared to the controls, possibly indicating pollution stress. Acidic air pollutants such as NO_x and SO_2 change the leaf pH and can induce oxidative stress, resulting in physiological and metabolic alterations (Dizengremel et al., 2008), with the decrease being greater in sensitive plants (Rai and Panda, 2014; Nadgórska-Socha et al., 2017). Lowering of cytoplasmic pH in guard cells can distort the shape of the stomata, altering the turgor systems within the stomatal complex (Kondo et al., 1980) which will affect stomatal sensitivity, decreasing photosynthesis and negatively effecting chlorophyll (Turk and Wirth, 1975). The pH of the PM can alter the leaf extract pH of vegetation, which further influences stomata sensitivity (Rai, 2016). Whilst the pH of the deposited PM was not determined in the current study, a correlation between ambient TSP and pH was observed, which could be worth examining more closely in future experiments.

As a result of this study, it is suggested that *C. comosum* and *C. comosum variegatum* are considered acidic pollution 'sensitive' species, whilst the 9 test species that did not show leaf pH reductions at the test sites could be considered to be tolerant.

3.4.5 The impact of pollutants on relative water content

A RWC range of 82.47 – 96.81 % was observed in the current work. Previous studies have observed similar ranges (Kaur and Nagpal, 2017). Rai and Panda (2014) detected lower relative water content at the polluted site compared to the control site; with 62.16% and 87.23% being their lowest and highest values for polluted and control sites respectively. However, in the current project no significant differences were observed between the leaf RWC at the test sites and controls for 8 of the tested species. Again, this could be due to the selected species having a tolerance to pollution, or generally lower pollution levels relative to past studies.

Three of the test species however, demonstrated lower RWC between test and control sites: *N. exaltata bostoniensis; N. gracilis* and *S. wallisii.* Air pollution impacts RWC by affecting the transpiration rate within the leaves (Swami et al., 2004; Joshi and Swami, 2007). High RWC values, in contrast, are indicators of pollutant tolerance (Singh et al., 1991). As such, the 8 test species which saw no RWC effects could be considered pollution tolerant for this health variable. Alternatively, *N. exaltata bostoniensis*; *N. gracilis* and *S. wallisii*, were sensitive due to their lowered relative water content values compared to the controls.

3.4.6 The impact of pollutants on carbon content

Previous research on carbon content has been heavily focused on sediment analysis (i.e. Heiri et al., 2001); and a few studies on plants used in agriculture (i.e. Rahman et al., 2013) and bio-fuels (i.e. Baxter et al., 2014). To the author's knowledge, no previous literature exists examining air pollution induced stress effects on the carbon content of leaves, making comparisons difficult.

Carbon allocation is an important plant health response and can be significantly altered by air pollution induced stress (see section 3.1.6). The results obtained for carbon content saw similar species trends as the other aforementioned health variables. More specifically, no significant differences were observed between the leaf carbon content at the test sites and controls for 8 of the tested species. As previously suggested, this could be due to the selected species having a tolerance to pollution. The carbon content of three of the test species however, was found to significantly differ between test and control sites: *P. glabella, P. madagascariensis* and *P. obtusifolia*. Interestingly, carbon content was found to be significantly correlated with leaf RWC, indicative that perhaps carbon content is a more important plant health variable than previously thought, and is thus worth including in future studies.

3.4.7 Species differences in plant health

Previous work has shown that plants vary in their responses to air pollutants (Rai and Panda, 2014; Nadgórska-Socha et al., 2017). The results obtained in the current study also detected differences amongst species for all plant health variables. In the current project, however, due to the non-significant differences in health variables observed between the test sites and controls for the majority of the trial species, it can reasonably be concluded that all of the test species are capable of withstanding the *in situ* pollution conditions encountered without enduring major pollutant induced health stress. Some plants have previously been shown to have a detoxification response to air pollutants which results in the isolation and storing of pollutants in mostly inactive tissues away from the vital leaf tissues, with this process being considered a protective mechanism (Choi et al., 2001; Gunthardt-Goerg and Vollenweider, 2007). It is plausible that protective mechanisms such as this were occurring in the test species, protecting them from enhanced pollutant stress, although pollutant storage mechanisms apart from pH changes were not tested here.

Plant performance under stress conditions may also be correlated with functional and structural leaf features (Gostin, 2016). For example, it has been shown that a plant's capacity to mitigate air pollution such as traffic exhaust varies between species due to differences in characteristics such as stomata, epicuticular wax, trichomes and cuticles (Neinhuis and Bathlatt, 1998). Furthermore, leaf shape, size and orientation are all influential factors on leaf boundary resistance which will affect the rate at which gaseous air pollutants interact with the plant (Heath et al., 2009). Species with waxy cuticles are thought to be more resistant to pollutant entry (Gupta, 2016); acidic gases, however, can penetrate the cuticular wax layer (Rai et al., 2011), causing pollutant induced harm. The tested species, however, had a broad range of different leaf traits, and no particular species stood out as being especially pollutant tolerant. Therefore, it may be possible that pollution tolerance in the test species was not related to leaf

characteristics, but rather due to an inherent, accumulated tolerance to continual pollutant exposure *in situ*.

3.5: CONCLUSION & FUTURE DIRECTIONS

Although all of the tested species were able to withstand long term pollution exposure, it is important to note that the primary pollution source in the current work was vehicular exhaust, and as such it of interest to determine whether the species would suffer more if they were situated near heavy industrial sites, or other locations with different pollutant types and concentrations. Whilst internal plant features were not examined in the current research, it is worth examining pollution induced effects in plant cell features in future studies, to better understand potential pollution effects on a cellular level. There are of course other plant variables which could be examined as indicators of plant health, such as general morphology and the structure of the epicuticular wax (Neinhuis and Barthlott, 1998). Additionally, soluble sugar content could also be used as a measure of plant density due to its role in physiological activity (Tripathi and Gautam, 2007) and protection against stress conditions (Finkelstein and Gibson, 2001). A decrease in soluble sugar content is usually a result of plant stress resulting from an increase in respiration and decrease in CO_2 fixation from chlorophyll destruction (Wilkinson and Barnes, 1973). Similarly, other plant health tests were previously conducted by the authors (see Paull et al., 2018) including stomatal conductance, substrate pH and the maximum efficiency of photosystem II (PSII) on plants exposed to concentrated diesel fumes; with substrate pH being the most influential variable on plant health. These tests were not tested in the current project due to their destructive nature. As a result, there remains an array of associated plant health parameters which may prove worthwhile testing in future studies.

The results showed that common green wall species are able to withstand continual pollutant exposure, representative of *in situ* conditions, with minimal health associated damage. Certain species, however, may be more susceptible to pollution induced damage such as *C. comosum* and *C. comosum variegatum,* as shown by their decline in leaf extract pH. Additionally, *P. madagascariensis*, *P. glabella* and *P. obtusifolia* showed pollution sensitivity from decreased carbon content and *N. exaltata bostoniensis*, *N. gracilis* and *S. wallisii* from decreased relative water content. Contrarily, *C. comosum variegatum*, *N. glabra*, *N. gracilis* and *S. wallisii* showed pollution tolerance from an increased chlorophyll content. Clearly, no exceptional species for pollution tolerance or sensitivity was identified, with some species being sensitive in one plant health category but tolerant in another. Nonetheless, the majority of the test species experienced no significant effects on plant health from pollution exposure, indicative of suitability in green wall implementation in contemporary urban environments.

Chapter 4 – The capacity of *in situ* green walls to reduce ambient particulate matter, noise pollution and temperature conditions.

ABSTRACT

Green walls have previously shown the capacity to reduce PM, noise and temperature. However, previous research has been founded on manipulative experiments and computational modelling, with minimal empirical evidence suggesting green walls are quantifiably influential on these characteristics in the ambient environment. Additionally, most studies that purport the benefits of green walls rarely take into consideration the variable environmental conditions encountered *in situ*, and how these might alter a green wall's ability to quantifiably influence the proximal environment. The aim of this chapter was to determine if green walls have a quantitative effect on the *in situ* ambient air quality in an urban environment. This was achieved by recording the ambient PM concentration, noise and temperature at 12 green wall and adjacent reference wall locations across a dense urban centre. The results indicated that the PM and temperature conditions at the green wall sites did not significantly differ to those at the reference wall sites. The noise conditions at the green wall sites, however, were significantly lower than at the reference wall locations. It is suggested that mechanically assisted, or active green wall systems may have a higher PM reduction capacity, and if so they will be more valuable for installation *in situ* compared to the standard passive systems, although this will also require further research.

4.1: INTRODUCTION

4.1.1 Urban environmental quality

The proportion of people living in dense urban areas has increased from 34% in 1960 to 54% in 2014 (Cho, 2015), with living in cities being correlated with a range of health problems (Galea et al., 2005). Diminishing air quality in dense urban environments, in particular, is an emergent health problem (Speak et al., 2012; Przybysz et al., 2014; Weber et al., 2014). It has been suggested that more than 1.78 billion people have inhaled polluted air over the last decade (WHO, 2014), with an estimated 7 million deaths from air pollution exposure in 2012 (Simunich, 2016). Air pollution can contain a combination of solid, gaseous and liquid particles, particularly from vehicle exhaust, dust and factory emissions (Simunich, 2016), with the smaller sized particles (i.e. $PM_{2.5}$) penetrating deeper into the lungs and alveolar regions, making them especially dangerous to human health (Dzierzanowski et al., 2011; Speak et al., 2012; Song et al., 2015; Wang et al., 2015). Further, as urban areas become increasingly dense, issues such as excess heat and noise are produced (Uttara et al., 2012), which negatively impacts wildlife, vegetation and human populations; altering local climate and increasing energy demands (McAlexander et al., 2015; Simunich, 2016). As such, technologies that reduce our exposure to, and mitigate the effects of, the factors associated with dense urban environments: air pollution, the urban heat island effect and noise pollution, are paramount.

4.1.2 Green wall technology

The capacity of plants and their associated growing substrates to effectively clean the air, produce cooler ambient temperatures and reduce ambient noise has been demonstrated (Scheuermann, 2016). The amount of space for green areas such as parks within cities, however, is rapidly declining (Scheuermann, 2016), commonly being replaced with buildings. It is thought that at least 80% of buildings within cities will still be in use by 2050 (Jofeh and

Li, 2016), making the implementation of green walls onto pre-existing building surfaces a space efficient initiative. Vertical greenery utilises plants which are grown in small pots, planter boxes or specially designed surfaces, and are hung vertically on walls (Ghazalli et al., 2018). Green walls can positively impact the urban environment in many ways including: mitigating air pollution (Sternberg et al., 2010; Marchi et al., 2015; Charoenkit and Yiemwattana, 2016); decreasing surface temperatures (Hasan et al., 2012; Mazzali et al., 2013; Bolton et al., 2014; Coma et al., 2017; Cuce, 2017; Vox et al., 2017) and reducing noise (Azkorra et al., 2015).

4.1.3 Green walls as PM pollution remediators

Green walls act as a particulate sink, (Smith and Staskawicz, 1977), due to the plant surfaces influencing PM diffusion and sedimentation (Beckett et al., 1998); by acting as a source of turbulence and increasing turbulent diffusion; resulting in a dilution of pollutant concentrations (Abhijith et al., 2017). Furthermore, green walls have been suggested as an appropriate tool to reduce PM without altering the air exchange (Litschike and Kuttler, 2008) by forcing polluted air to either pass through the vegetation or flow over it (Tong et al., 2016). Similar to solid barriers (i.e. low boundary walls), vegetation barriers which have low porosity and a high density have very little to no infiltration of air flow; whereas high porosity and low density vegetation barriers allow the majority of air to flow through the barrier (Bowker et al., 2007; Baldauf et al., 2008; McNabola et al., 2009; Gallagher et al., 2012; Brantley et al., 2014; Janhäll, 2015; Gromke et al., 2016; Abhijith et al., 2017) increasing their PM filtration efficiency.

Some studies have shown the positive impact of vegetation presence on ambient air pollutant removal. Irga et al. (2015) recorded lower PM concentrations in areas of Sydney which had abundant tree vegetation; Al-Dabbous and Kumar (2014) noted that roadside vegetation had a significant, wind dependent effect on nanoparticle concentrations in the UK, and García-

Gómez et al. (2015) detected significant effects of peri-urban forests in Spain on ambient air quality. There, however, still remains some uncertainty regarding the capacity of green walls to effectively remove ambient PM pollution. For example, wind strength, the presence of buffer zones, the distance from the pollution source and particle quality all effect the distribution of pollutants (Stevović and Markovic, 2016); making it difficult to draw general conclusions, as these factors are variable both temporally and spatially.

4.1.4 Green walls as noise pollution mitigators

Noise pollution has been defined as any disturbing or unwanted noise that interferes or harms humans or wildlife (Jain et al., 2016). The number of people exposed to noise pollution continues to increase in urban areas due to the expansion of transport, residential areas and infrastructure (DOH, 2018). Noise pollution is common and more frequent in dense urban environments due to peoples' inherent proximity to an array of continuous noise emitting sources **(**Brown and Bullen, 2003). Sources of noise pollution that are large contributors in dense urban environments include transport (road, rail and air), industry, construction, public works and neighbourhood related noise (DOH, 2018). Of these sources, it has been suggested that > 70% of unwanted sound in urban Australia is from road traffic (Marquez et al., 2005). Exposure to excessive outdoor noise can have negative impacts on human health and well-being (McAlexander et al., 2015); as it disrupts sleep and work productivity, limits cognitive function, contributes to mental illness and can even cause cardiovascular disease (Den Boer and Schroten, 2007). The hard surfaces of street canyons (i.e. glass, brick, concrete and asphalt) reflect sound, thereby increasing overall urban ambient noise (Thompson, 2015). Green walls however, have the ability to absorb urban noise rather than reflect it, which is the case of normal building surfaces (i.e. steel, concrete and glass; Simunich, 2016). Whilst green walls are not able to effectively reduce direct sound, they can absorb the noise that would otherwise be

reflected between buildings, as well as noise that bends around corners, essentially lowering the overall ambient noise (Wurm, 2016).

Sound can be reflected and diffracted by plant components, including trunks, twigs, branches and leaves (Van Renterghem et al., 2012). Plants can also reduce noise pollution by absorbing the sound waves. This effect is due to mechanical vibrations in plant elements caused by sound waves, leading to dissipation by converting sound energy to heat (Embleton, 1963; Martens & Michelsen, 1981; Tang et al., 1986). Additionally, the thermo-viscous boundary layer at vegetation surfaces assists with sound reduction (Azkorra et al., 2015). Regarding substrate effects on noise attenuation; the presence of soil can lead to destructive interference between the direct contribution from the source to the receiver and a ground-reflected contribution **(**Azkorra et al., 2015). This effect is referred to as the acoustical ground effect or ground dip (Azkorra et al., 2015). The presence of vegetation leads to an acoustically very soft (porous) soil, due to plant roots and a litter layer (Van Renterghem et al., 2012). This results in a distinct ground effect, producing a shift towards lower frequencies compared to sound propagation over grassland (Huisman & Attenborough, 1991). Therefore, this ground dip is effectively better at limiting typical engine noise frequencies (approximately 0.100 kHz; Van Renterghem et al., 2012).

It has been suggested that leaves have a sound absorption effect predominantly in the high frequency range (> 1 kHz), whilst the woody parts of vegetation (i.e. branches, twigs and stems) have a sound absorption effect in the mid frequency range (0.5 – 2 kHz; Martens, 1980). Traffic related noise is often strongest in the 1 kHz region, suggesting that vegetation could have the capacity to significantly reduce these levels (Klingberg et al., 2017).

Whilst the capacity of vegetation to absorb noise has been documented, it is of interest whether multiple green walls in various locations across many months will produce a uniform ambient noise reduction.

4.1.5 Green wall influence on ambient temperature conditions

Urban areas usually have much higher temperatures than rural and peri-urban areas, which is referred to as the urban heat island effect (Simunich, 2016). This is due to how urban areas are constructed, such as the heavy use of glass facades, concrete sidewalks, steel surfaces and asphalt roads which radiate heat rather than absorb it (Simunich, 2016). Increases in urban heat can result in increased air pollution levels, altered rain and wind conditions, increased energy demands, increased associated cooling costs, increased heat related illnesses and mortality rates (Simunich, 2016). More specifically high temperatures can increase air pollution levels in three main ways: by an increase in the use of cooling mechanisms, which are a primary source of pollutant emissions; the sun and heat can transform primary pollutants into secondary, more toxic pollutants; and increased heat leads to high atmospheric pressure, which creates a stagnant air layer, capturing and trapping pollutants (TEQOYA, 2020). Urban vegetation however, can be influential in reducing the ambient air temperature by evapotranspiration and shading (Bowler et al., 2010; Konarska et al., 2016). Green walls are effective at reducing the heat island effect and can remove 50% of solar radiation (Alspach & Göhring, 2016). This is due to the leaves absorbing the ambient heat energy and direct solar radiation through the process of photosynthesis (Wong et al., 2010). Additionally, a subset of plant species known as Crassulacean acid metabolism (CAM) plants photosynthesize during the night hours, reducing residual trapped heat energy at night (Winter and Holtum, 2011). Pollution, noise and heat reduction, however, can all be influenced by the geographical, morphological and climatic conditions of the area (Pauli 2016). As such, it is important to examine the effect of green walls

on temperature reductions in a range of spatial and temporal environments to uncover their true potential.

4.1.6 Aim

The number of studies on vertical greenery systems is comparatively low compared to research conducted on other types of urban greenery (Ghazalli et al., 2018). Furthermore, the majority of green wall studies are limited to Europe and Asia, and as such there is a lack of general understanding of green wall effects due to differing systems and conditions, especially different plant species and ambient environmental conditions (Pérez et al., 2014). Although some studies have highlighted the pollution reduction potential of green walls **(**Ottelé et al., 2010; Sternberg et al., 2010; Joshi and Ghosh, 2014), research regarding air pollution reduction by green walls within the built environment at a local scale is limited (Abhijith et al., 2017). Additionally, most studies conducted on PM removal by green wall systems assess removal on the leaf scale, then use modelling and simulation to generalise their findings to an *in situ* urban ambient scale (Ghazalli et al., 2018). Whilst computational modelling is a common method to determine the effect of green walls on PM removal, it usually does not account for specific environmental effects which could impact the effect of green walls on pollutant removal, such as wind speed (which can have a pollutant diluting effect), wind direction (which can have an impact on the location of pollutant emissions) and relative humidity (which can affect the size of the PM).

The experiments presented in this chapter investigated the ambient PM concentrations, temperature and noise conditions at green wall and reference wall locations across 12 Sydney test sites, over a 6 month duration. In doing so, this project aimed to determine the effectiveness of pre-existing green walls at reducing ambient PM concentrations, noise levels and temperature conditions within the Sydney region.

4.2: METHOD

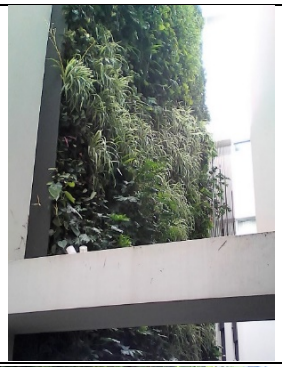
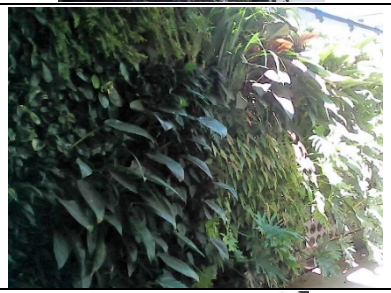
4.2.1 Sample sites

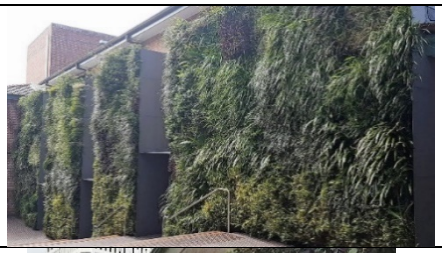
Twelve sites within the urban Sydney region were selected based on the presence of similar passive outdoor green walls. The sites varied in location, use and pollutant conditions (Table 5). Sydney was chosen as the city for this project due to the high number of Junglefy P/L modular green walls installed across the area.

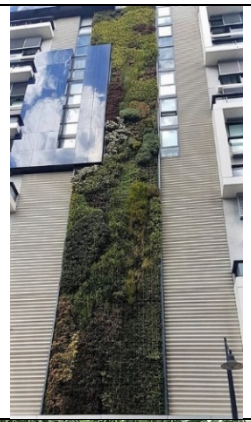
Sydney, Australia has a population of 5.2 million and lies on a coastal lowland plain between the Pacific Ocean and elevated sandstone tablelands. Sydney's climate is warm and temperate. Days on which rainfall events occur are evenly distributed throughout the year, however rainfall volume is at its highest in Autumn. Sydney city's air quality is generally good by international standards, although levels of particulate matter can exceed the national standards on occasion. Sydney has a population density of 9,186 per km^2, with land use broken into: commercial (28.9%), parkland (22.8%), residential (21.1%), industrial (14.5%), education (7.5%) and transport (3.2%). The noise pollution conditions in Sydney are concerning, with Sydney having the highest traffic related noise exposure of all the Australian capital cities (Brown and Bullen, 2003).

Table 5: Test site descriptions.

Site Number	Site Location	Notes	General Land Use	Elevation above sea level (m)	Picture	Size (m^2)	Number of Plants
1	Ashfield	Apartment complex with green wall situated in the back foyer	Residential and industry	18		27	1,296
2	Tamarama	Residential property, green wall situated in back yard	Residential and green space	33		12.5	600

3	Mosman	Apartment complex with outdoor green wall	Residential and industry	70		72	3,456
4	Lane Cove	Display home with green wall situated in an outdoor area	Industry	50		9	432
5	Woollahra	Residential property, green wall situated in front courtyard	Residential	85		6	288

6	Gordon	High School. Green wall situated in a courtyard	Residential	121		140	9,150
7	The Rocks, Site 1	Extensive green wall situated on expressway	Transport	19		142	6,891
8	The Rocks, Site 2	Green wall situated under rail line support structure	Transport	19		25	1,600
9	Summer Hill	High School, green wall situated in a courtyard	Residential	55		4.5	216

10	Camperdown	Apartment complex with green wall situated on the exterior of the building	Residential and industry	30		18	864
11	Ultimo	Green wall situated on a tertiary education facility	Industry	15		145	9,280
12	Crows Nest	Green wall situated on the exterior of a grocery store	Industry	101		25	1,200

4.2.2 Sampling Process

All air quality, traffic density, noise and temperature assessments were conducted once a month for 6 consecutive months at each site; starting June 2017 and concluding November 2017. The order in which sites were sampled was randomised amongst months to eliminate systematic temporal variation. Samples were not taken on rainy days, as rainfall removes PM from the air (Nishihara et al., 1989) and no bare soil was present within a 30 m proximity of the sampling locations so as to not artificially spike ambient PM concentrations. Average monthly weather variables were also collected for each site using Bureau of Meteorology Data to account for any weather dependent correlation with *in situ* conditions.

4.2.2.1 Air Quality Assessment

To assess PM reductions between sites with green walls and reference sites, PM air quality assessment was conducted using a DustTrack II 8532 laser densitometer (TSI, Shoreview, Minnesota) monthly at the twelve sites. At each site, 3 minute time weighted averages for each PM size fraction (PM_{10} and $PM_{2.5}$) were collected between 10 a.m. and 3 p.m. at both the green wall and a reference wall. At each site, a reference wall was selected based on the following criteria: the reference wall was exposed to the same traffic pollution source as the green wall; the reference wall had similar building characteristics as the green wall, and the reference wall was within 10 meters distance of the green wall. These criteria were used to eliminate any confounding factors effecting the variables between wall types.

4.2.2.2 Traffic Density Assessment

Traffic density was measured at each site, as it was predicted to be the predominant pollution source, and is known to be a source of varying sized PM, which was the pollutant of interest in the current project. Most sites were situated near residential properties, academic institutes or

highways; and were not near any industrial sources of pollutant emissions other than minor infrastructure work. Traffic density was quantified at the closest intersection to the green wall for a 30 minute duration. Traffic was classified based on vehicle type to attain average traffic densities for cars, buses, trucks and motorcycles. Days of the week and times at which air quality and traffic density tests were conducted were limited to weekdays between 10 a.m. and 3 p.m. to avoid peaks caused by work and school commuters (Irga et al., 2015), and randomized amongst sites and months.

4.2.2.3 Noise and Temperature Assessment

Noise and temperature readings were taken at 4-point sources across both the green wall and reference walls at each site using a Digitech Multifunction Environment Meter. The average and standard error were then determined from the point samples.

4.2.3 Statistical Analysis

Mean values were calculated for $PM_{2.5}$, PM_{10}, noise, temperature and traffic for each month of the study at each site (i.e. six values per site for each variable). For use in subsequent analysis, differences between the reference walls and the green walls (henceforth Δ values) for $PM_{2.5}$, PM_{10}, noise and temperature were calculated following the form: reference wall value – green wall value, so that higher values of Δ indicate higher concentration of PM at the reference wall relative to the green wall. Prior to analysis, Δ $PM_{2.5}$ and Δ PM_{10} were transformed to satisfy the assumptions of the models by square root transforming the absolute values of all observations, and giving a negative sign to untransformed values that were less than zero. After transformation, observations retained their original sign (i.e. positive values were not made negative, and vice versa), while decreasing the deviation of the Δ PM values.

To test if a systematic difference in PM existed between the green walls and reference walls, single tailed paired sample *t*-tests were used. Following this, the relationship between Δ $PM_{2.5}$ and Δ PM_{10} was tested using a LMM regression with Δ PM_{10} as the response, and Δ $PM_{2.5}$ as the predictor, with a random slope between Δ $PM_{2.5}$ and Δ PM_{10} and a random intercept fitted for each site. Site level differences in Δ PM were then examined, by fitting a linear model to the Δ PM data and using site as predictor (categorical fixed factor, 13 levels). Using a joint test, the coefficients produced by this model were then tested for differences from zero. Finally, to understand the relationship between PM and environmental factors, multiple regression models of green wall $PM_{2.5}$ and PM_{10}, and Δ $PM_{2.5}$ and Δ PM_{10} were built using traffic density, wind, humidity, green wall size and the number of plants used in the green walls as predictors. Here, LMMs were again employed, with a random intercept fitted for each site.

Using a similar approach to that used for PM, overall differences in noise and temperature between green and reference walls were first tested using single tailed paired *t*-tests. Site level differences from zero in Δ noise and Δ temperature were then tested by fitting a linear model to each response using site as predictor (categorical fixed factor, 13 levels). Where the joint test of these models was significant, the coefficients from the models were tested for differences from zero to determine which site or sites generated the difference.

All analyses and visualisation were performed in R 3.6.1 (R Core Team, 2014), using the packages lme4 (Bates et al., 2015), lmerTest (Kuznetsova et al., 2017), emmeans (Length, 2019) and car (Fox and Weisberg, 2011). Where LMMs were used and models did not include categorical terms, degrees-of-freedom were approximated using the Satterthwaite method for *t*-tests, while LMMs containing categorical terms used Wald Chi-square tests to generate ANOVA tables.

4.3: RESULTS & DISCUSSION

4.3.1 Differences in PM concentration between wall types

The difference between the ambient *in situ* PM concentrations at green wall and reference wall locations are presented in Figure 12. Concentrations of particulate matter measured at the green walls were found to not be significantly lower than those recorded at the paired reference walls for $PM_{2.5}$ (t_{71} = -1.10, P = 0.1; Fig. 12a), and PM_{10} (t_{71} = -0.50, P = 0.3; Fig. 12b). Additionally, no significant association between Δ $PM_{2.5}$ and Δ PM_{10} was found ($t_{10.4}$ = 1.93, P = 0.08; Fig. 12c). This result was surprising as many green wall studies have shown high pollutant removal capacities (Pugh et al., 2012; Abhijith et al., 2017). This removal capacity is thought to be due to the leaves creating turbulence with the surrounding air, forcing compaction with aerosolized PM particles, leading to deposition and eventual accumulation of PM on the leaf surfaces (Ottelé, 2011; Abhijith et al., 2017). PM accumulation by this process was apparent in the current project from visual inspection of the leaves and from the results presented in Chapter 2. As such, despite the detection of PM accumulation from the ambient air, it is clear that this level of PM removal exhibited a small overall effect on the proximal air quality conditions. Previous studies have also shown minimal effects for the role of urban vegetation on the proximal ambient air pollution levels (Setälä et al., 2013). However, even small reductions in pollutant concentration can have significant positive impacts on human health (Klingberg et al., 2017).

Green walls have shown to have similar pollution dispersion patterns to that of solid walls (Morakinyo et al., 2016; Tong et al., 2016), potentially explaining the current similar PM results between green wall and reference wall sites. Passive, solid structures such as noise barriers and low boundary walls have shown to improve the local air quality (Gallagher et al., 2015), with the reference walls in the current study potentially matching the pollutant removal

capacity of the green walls. It is likely that in the current project, the vegetation in the green walls were removing ambient PM concentrations whilst the reference walls were also simultaneously acting as a hard surface for PM impaction and deposition; with both of these wall types improving the proximal air quality to an equally small degree.

A limitation of both the current study and previous similar studies is that comparisons between identical locations, with and without a green wall were not conducted (Abhijith et al., 2017). Such comparisons would allow for a direct comparison of ambient air quality conditions pre and post green wall implementation, rather than a comparison between wall types within close proximity to one another. Due to this limitation, many previous studies have instead focussed on wind-tunnel and modelling experiments which facilitate before and after comparisons; albeit without the ability to replicate complex *in situ* weather and traffic patterns (Abhijith et al., 2017). For example, in a multi-national computational fluid dynamic modelling study conducted by Sanjuan & Bull 2016; it was determined that green walls could reduce local PM concentrations by 10 – 20 %. However, these reductions were localised to within the street canyon and overall city reductions would be much lower. Similarly, in a modelling study conducted by Nowak et al. (2006) examining 55 US cities, it was determined that urban vegetation did not have substantial effects on local air pollution concentrations. It has therefore been suggested that the effect of urban vegetation on local air pollution mitigation has been exaggerated (Pataki et al., 2011; Nowak et al., 2013, 14; Whitlow et al., 2014 a, b).

There clearly remains some uncertainty regarding the pollutant removal capacity of green walls, and as such further studies need to be conducted to identify the true role vegetation plays on local air pollution mitigation (Klingberg et al., 2017).

As the current project saw no significant difference in PM conditions between the green wall and reference wall locations, it could be suggested that passive green walls are not able to quantifiably reduce ambient PM levels to an effective rate for phytoremediation purposes in *in situ* conditions. A power analysis was conducted on the data to determine if a larger sample size was required to see a significant effect. The results of the power analysis indicated a high confidence in the non significant result, meaning a larger sample size would provide the same result. Previous controlled laboratory experiments (Torpy et al., 2016; Irga et al., 2017; Pettit et al., 2017; Irga et al., 2019) have shown that PM, VOC and CO_2 pollutant conditions can be mitigated at a greater efficiency with the conversion of passive systems to active systems. The primary difference between passive and active green wall systems is the use of assisted aeration using some form of mechanical fan, which actively forces air through the plant root and substrate membrane (Soreanu et al., 2013). This leads to an increased surface area for PM adherence, leading to PM entrapment within the substrate and plant root matrix; thus filtering the air more effectively than through the simple diffusion mechanisms of passive systems (Llewellyn and Dixon, 2011; Veillette et al., 2011; Franco et al., 2012). Therefore, it is suggested that future studies focus on the potential effect active green wall systems may have on ambient PM conditions *in situ*.

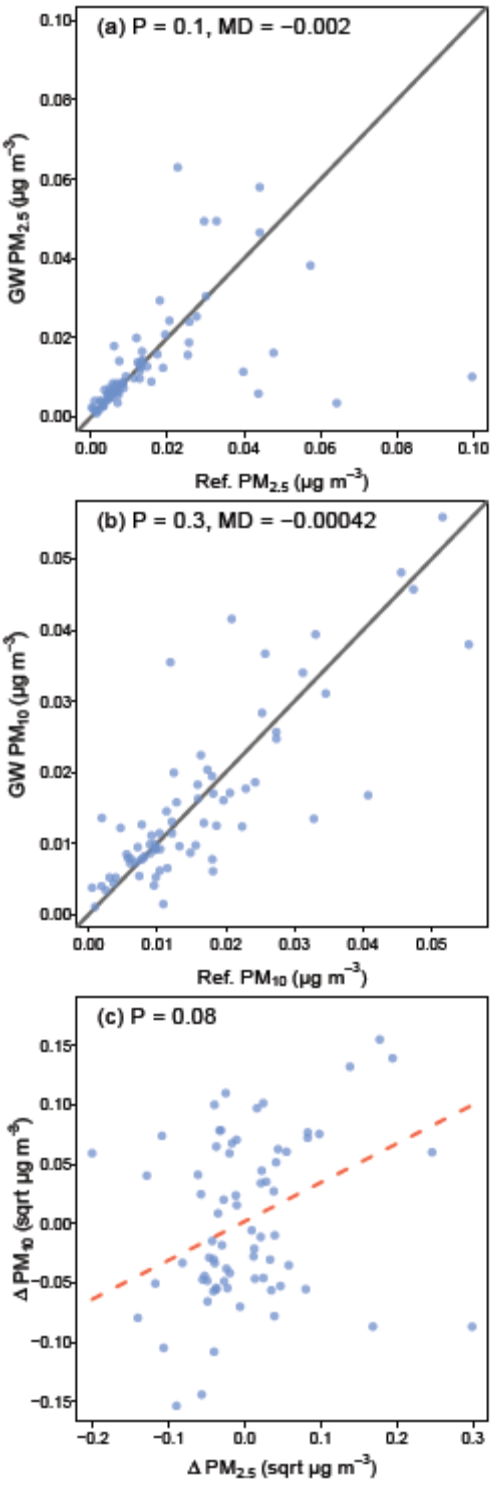


Figure 12: Scatter plots showing concentrations of $PM_{2.5}$ (a), and PM_{10} (b) at the green wall (y-axis) and reference wall (x-axis), and Δ PM (c). The black line in (a) and (b) represents a 1:1 relationship, therefore all points falling below the line indicate lower values at the green wall relative to the refence wall, and the converse for points above the line. In (a) and (b) *p*-values (paired *t*-test) and the mean difference (MD, green wall value – reference wall value) between paired samples is shown. Plot (c) shows the line of best fit as a broken red line, and the *p*-value from the LMM for the relationship between Δ PM_{10} and Δ $PM_{2.5}$.

Potential differences in site specific PM concentrations were investigated to determine if any site specific sources were influential on the non-significance observed between the PM conditions at the green and reference wall locations within each site. The results indicated that there was no significant difference from zero found in, Δ $PM_{2.5}$ across sites ($F_{12,60}$ = 0.72, P = 0.7; Fig. 13a), with similar results for Δ PM_{10} ($F_{12,60}$ = 0.52, P = 0.9; Fig. 13b). The reason for this is not clear. The non-significance observed between the sites could be due to the relatively stable pollution conditions within Sydney; with the small site specific variation between each site not leading to significant differences between PM_{10} and $PM_{2.5}$ levels across the study sites. It is probable that more noticeable pollutant differences across sites would be evident if the sites ranged across multiple cities or locations with more variable high and low pollution sources, such as power plants and industrial areas compared to residential properties.

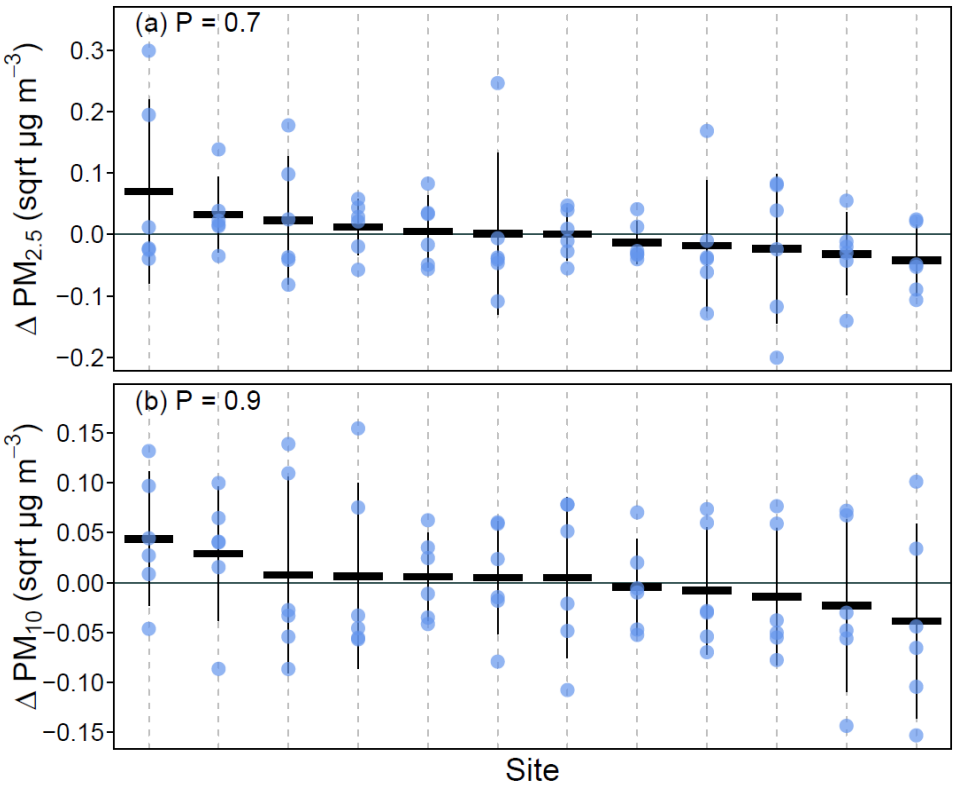


Figure 13: Plots of Δ $PM_{2.5}$ (a) and Δ PM_{10} (b). Blue points show the Δ PM values, thick black horizontal lines represent the means for the sites, and thin vertical black lines the 95% confidence intervals of the mean for the sites. The *p*-value at the top of each plot is the result of a joint test of model coefficients with the null hypothesis that the sites do not differ from zero. Sites are sorted on the x-axis by their mean value (highest to lowest) for ease in interpretation. The solid horizontal line indicates zero on the y-axis, representing equal values of PM at the green wall and reference wall.

As ambient *in situ* conditions such as traffic and weather patterns have been previously shown to have an influence on PM conditions, monthly traffic density and weather variables were recorded at each site. The results indicated that higher green wall $PM_{2.5}$ was significantly related to both greater traffic density and higher humidity (Table 6; Fig. 14a and 14b), while higher green wall PM_{10} was related only to traffic density (Table 6; Fig 14c). This result was not surprising, as traffic density is well known to influence air pollution conditions (Kim et al., 2012; Klingberg et al., 2017). A significant association between lower Δ $PM_{2.5}$ and heavier traffic, and faster maximum wind speed was found (Table 6; Fig. 14d and 14e), indicating that both these factors may affect the remediation ability of green walls. More specifically, wind speed can have a pollutant diluting effect; wind direction can have an impact on the location of pollutant emissions and relative humidity can affect the size of the PM (Sanjuan & Bull, 2016).

Table 6: Results from multiple regression LMMs of green wall $PM_{2.5}$ and PM_{10}, Δ $PM_{2.5}$, and Δ PM_{10}.

Response	Terms	Estimate	SE	df	t value	P
Green wall $PM_{2.5}$	Traffic	0.281	0.101	7.8	2.782	**0.025**
	Max wind speed	-0.139	0.077	62.2	-1.811	0.07
	Humidity	0.465	0.194	62.3	2.391	**0.02**
	Green wall size	-1.198	1.214	6.4	-0.986	0.4
	Plant number	1.173	1.125	6.4	1.042	0.3
Green wall PM_{10}	Traffic	0.23	0.081	8.6	2.847	**0.02**
	Max wind speed	-0.098	0.073	64.5	-1.334	0.2
	Humidity	0.197	0.185	64.6	1.062	0.3
	Green wall size	-0.389	0.955	7.4	-0.407	0.7
	Plant number	0.333	0.885	7.4	0.376	0.7
Δ $PM_{2.5}$	Traffic	-0.02	0.009	66	-2.299	**0.025**
	Max wind speed	0.017	0.008	66	2.076	**0.042**
	Humidity	0.002	0.021	66	0.081	0.9
	Green wall size	0.079	0.101	66	0.778	0.4
	Plant number	-0.071	0.094	66	-0.758	0.5
Δ PM_{10}	Traffic	0.004	0.007	66	0.628	0.5
	Max wind speed	-0.004	0.007	66	-0.497	0.6
	Humidity	0.02	0.018	66	1.131	0.3
	Green wall size	-0.053	0.088	66	-0.604	0.5
	Plant number	0.05	0.081	66	0.62	0.5

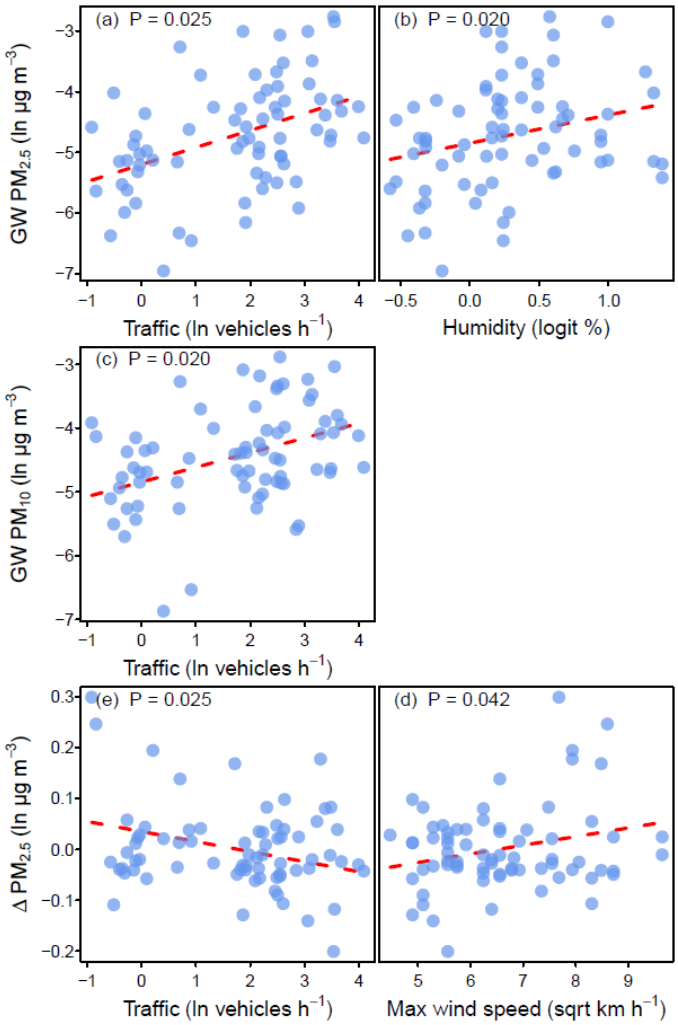


Figure 14: Scatterplots of the significant patterns emerging from the multiple regression models of green wall $PM_{2.5}$ and PM_{10}, Δ $PM_{2.5}$, and Δ PM_{10}. Broken lines are coefficients for the model term of interest, with *p*-values for the terms shown at top left of each plot. Full results are shown in Table 6.

4.3.2 Differences in noise and temperature conditions between wall types

Green walls sites were significantly quieter than their paired reference sites (t_{71} = -3.55, p = 0.0003; Fig. 15a), while no significant difference in temperature was found between sites (t_{71} = -1.34, P = 0.1; Fig. 15b). The lower noise readings at the green wall sites in the current project was probably due to the ability of plants to absorb more noise compared to hard surfaces, which reflect noise of all wavelengths (Dunnet & Kingsbury, 2008; Azkorra et al., 2015).

Previous modelling studies have also noted the noise reduction capacity of green walls, indicating that they can reduce between 2 and 5 dB (A) of single point source noises (Ismail, 2013), and up to 1.6 dB (A) of road traffic associated noises (Jeon et al., 2013). Similarly, in a study conducted by Klingberg et al. (2017) it was determined that the presence of leaves significantly reduced noise levels; noting that the noise reductions became greater as the distance between the main road and vegetation increased, as a result of an increasing amount of greenery between the road and recording device; with an overall reduction of 0.6 – 2.3 dB recorded. The results obtained in this chapter demonstrated somewhat higher noise reductions than the abovementioned studies, falling mainly in the range of 1.34 – 6.40 dB. Interestingly, one of the tested sites (Tamarama), saw an overall noise reduction of 12.13 dB between green wall and adjacent reference wall, this effect being greater than 10 times the reduction capacity of the lower noise reduction sites. This difference in noise reduction capacity is likely due to noise type and duration of noise exposure, as the predominant noise type at Tamarama was ocean related noise i.e. waves crashing compared to the predominant traffic related noise at the other sites.

Other studies have also noted the noise reduction capacity of green walls, such as the computational modelling study conducted by Patel and Boning (2016) who determined that green walls could reduce emergent and traffic noise sources by up to 10 dB (A). Whilst literature exists on green wall capacity to reduce urban noise; no two cities are the same and as such green walls may behave differently in different street layout conditions, with the effect of urban conditions on green wall performance still remaining understudied (Patel and Boning, 2016).

No significant differences were observed between the green wall and reference walls locations regarding ambient temperature. This result was surprising as plants are known to have an air cooling capacity resulting from evapotranspiration. Pérez-Urrestarazu et al. (2016) noted

temperature reductions of 0.8 – 4.8 °C from an active green wall, and most indoor green wall studies have shown a significant effect on indoor temperature (i.e. Darlington et al., 2000; Meier, 2010). The effect of outdoor temperature reductions, however, have been more variable. In a simulation study conducted by Alspach and Göhring (2016) it was found that green walls reduced the urban heat island effect most effectively in cities with a height to width ratio greater than 2, which includes dense urban cities such as Melbourne and Hong Kong (Pauli, 2016). In this case temperature reductions of up to 10 °C were modelled. It is therefore possible that the building characteristics and unorthodox grid street design of Sydney did not allow for significant temperature reductions from green wall presence in this project. Furthermore, Pérez et al. (2014) noted that most studies on green wall ambient temperature effects have been conducted in warm climates, with temperature reductions from green walls ranging from 1 – 15.18 °C; suggesting a variety of climates including cooler climates need to be tested to determine the true capacity of green walls to reduce ambient temperature. Additionally, Alspach and Göhring (2016) noted peak temperature reductions were much greater than average reductions, thus the effect was of most benefit during extreme heat waves. Similarly, in the current study, peak temperature reductions were observed, however when this was averaged across sites and months, the reductions became non-significant. These peak reductions were noticed when comparing the temperature data recorded at the 4 point sources across the wall types, during an isolated period of a day each month. It would be recommended that future studies use a recording device which can account for a longer sampling duration to account for daily temperature fluctuations or to record subsequent days of the month, to allow for a clearer interpretation of temperature reductions. Whilst the green walls within the current project were not effective at consistently reducing ambient temperatures, they still provided occasional peak reductions over the sampling period, and thus may be beneficial in Sydney's extreme heat conditions during the Summer periods.

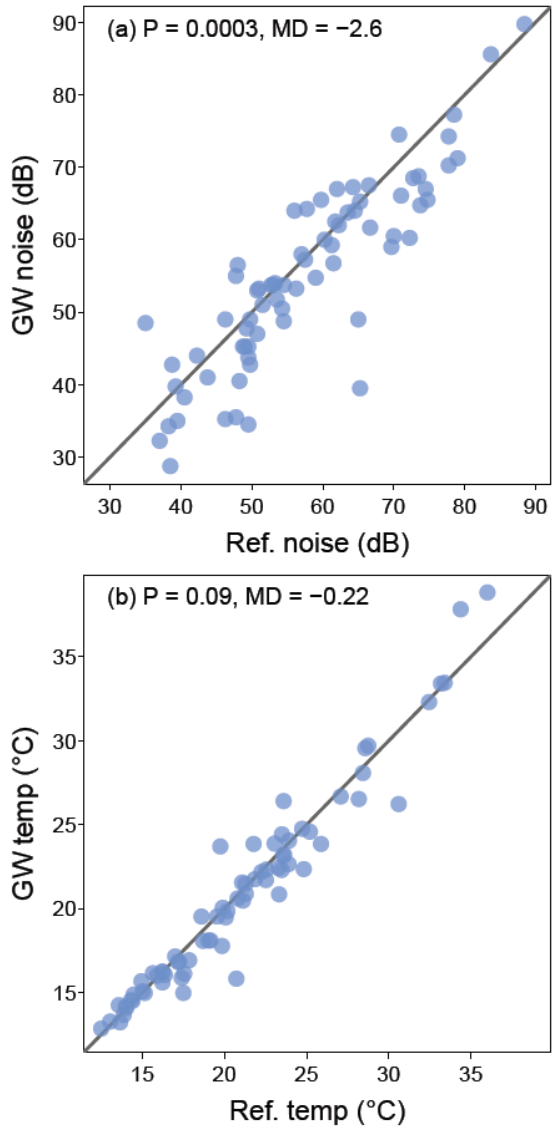


Figure 15: Scatterplots of concentrations of noise (a), and temperature (b) at the green wall (y-axis) and reference wall (x-axis). The black line represents a 1:1 relationship, therefore all points falling below the line indicate lower values at the green wall relative to the refence wall, and the converse for points above the line. In both plots *p*-values (paired *t*-test) and the mean difference (MD, green wall value – reference wall value) between paired samples in shown.

Across sites, it was found that both Δ noise ($F_{12,60} = 4.82$, $P < 0.0001$), and Δ temperature ($F_{12,60} = 3.02$, $P = 0.002$) significantly differed from zero. At site level, Δ noise was found to be significantly greater than zero at four sites (i.e. green wall noise was lower relative to the reference wall; Fig. 16a), while Δ temperature showed more equivocal results with one site exhibiting Δ temperature significantly more than zero, and one other site having Δ temperature significantly less than zero (Fig. 16b).

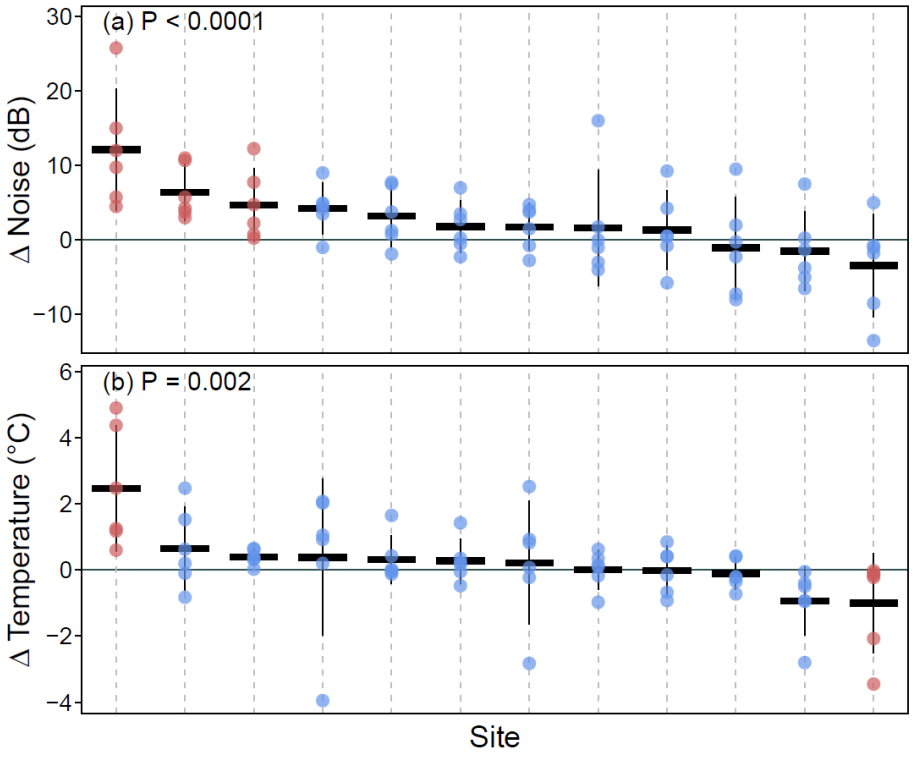


Figure 16: Plots of Δ noise (a) and Δ temperature (b). Blue points show the Δ PM values, thick black horizontal lines the means for the sites, and thin vertical black lines the 95% confidence interval of the mean for the sites. The *p*-value at the top of each plot is the result of a joint test of model coefficients with the null hypothesis that the sites do not differ from zero. Sites are sorted on the x-axis by their mean value (highest to lowest) for ease in interpretation. The solid horizontal line indicates zero on the y-axis, representing equal values of noise (a) or temperature (b) at the green wall and reference wall.

4.4: CONCLUSION & FUTURE DIRECTIONS

This chapter assessed the capacity of *in situ* passive green walls to reduce ambient PM pollution and modify ambient noise and temperature conditions. No significant differences were observed between the PM concentrations at the green wall and reference wall locations across the 12 sites; indicating that the current passive systems were not capable of reducing PM conditions to a detectable level. The green walls tested in the current project, however, had significantly lower proximal noise levels compared to the reference walls, indicating that the plants were potentially absorbing ambient noise. Furthermore, temperature conditions at the green wall and reference wall locations were not significantly different. Many studies claiming vast temperature, noise and PM reductions from green walls have been the product of computational and modelling experiments which enable before and after comparisons; however their lack of ability to accurately replicate complex *in situ* weather and traffic patterns, which ultimately play a role on ambient conditions and removal capacities, may make some of the previous observations overestimates.

Chapter 5 – Active botanical biofiltration of air pollutants using Australian native plants.

ABSTRACT

Air pollutants are of public concern due to their association with adverse health effects. Biological air filters have shown great promise for the bioremediation of air pollutants. Different plant species have previously been shown to significantly influence biofilter pollutant removal capacities, although the number of species tested to date is small. The aims of this project were to determine the active biowall PM, VOCs and CO_2 removal capacity of different Australian native species and to compare removal rates with previously tested ornamental species. The single-pass removal efficiency for PM and VOCs of native planted biofilters was determined with a flow-through chamber and CO_2 removal was tested by a static chamber pull down study. The results indicated that the native species were not effective for CO_2 removal likely due to their high light level requirements in conjunction with substrate respiration. Additionally, the native species had lower PM removal efficiencies than ornamental species, with this potentially being due to the ornamental species possessing advantageous leaf traits for increased PM accumulation. Lastly, the native species were found to have similar benzene removal efficiencies to ornamental species. As such, whilst the native species showed a capacity to phytoremediate air pollutants, ornamental species have a comparatively greater capacity to do so and are thus more appropriate for air filtration purposes in indoor applications. However, as Australian native plants have structural and metabolic adaptations that enhance their ability to tolerate harsh environments, they may find use in botanical biofilters in situations where common ornamental plants may not be suitable, especially in the outdoor environment.

5.1: INTRODUCTION

5.1.1 The use of phytoremediation for pollution removal

Phytoremediation involves the utilization of plants and their associated microbial communities to ameliorate pollution, and is generally considered to be an environmentally friendly and economical technology. The application of phytoremediation for air purification originated with investigations by Wolverton and colleagues (Wolverton et al., 1984), who demonstrated the capability of foliage plants for purifying VOC contaminated air. Subsequently, the biological activity of plants and their associated substrate microflora has been shown to be capable of reducing various air pollutants including CO_2 (Irga et al., 2013; Torpy et al., 2014; Su and Lin, 2015), PM (Gawrońska and Bakera, 2015; Pettit et al., 2019), O_3 (Abbass et al., 2017) and VOCs (Godish and Guindon, 1989; Wolverton and Wolverton, 1993; Wood et al., 2002; Orwell et al., 2004; Wood et al., 2006; Aydogan and Montoya, 2011; Irga et al., 2013; Torpy et al., 2013).

5.1.2 Relationships between plant characteristics and pollution removal

Different characteristics of plants are known to influence their suitability for air phytoremediation (Lin et al., 2017). Variation in shapes, volume of crown, leaf macro- and micromorphology, leaf size and cuticular waxes are important traits that must be considered for efficient air pollutant removal (Litschke and Kuttler, 2008; Petroff et al., 2008; Sæbø et al., 2012; Ram et al., 2014; Chen et al., 2016; Leonard et al., 2016). Whilst there are a number of studies linking air pollutant removal to effects restricted to plant behaviour (Hosker and Lindberg, 1982; Fowler, 2002; Singh and Verma, 2007; Ottelé et al., 2010; Sternberg et al., 2010), it is widely thought that the performance of botanical air filtration can largely be attributed to rhizospheric microbial activities (Wood et al., 2002; Kim et al., 2010; Pettit et al., 2017).

5.1.3 Systems for pollution removal

Conventionally, most built environment solutions to deal with air pollution involve filtration, especially for indoor air in modern buildings which are usually mechanically ventilated (Hwang and Park, 2017). The filters used within these systems have varying levels of PM removal efficiency, but are unable to remove gaseous pollutants other than by dilution with outdoor air (Tong et al., 2018). Whilst some mechanical filter systems have shown to be somewhat efficient; they have high maintenance needs, use a great amount of energy (Montgomery et al., 2012) and in many cases remain ineffective for gaseous pollutant removal. The use of plants as phytoremediators allows not only the effective simultaneous removal of multiple air pollutants, but with further development, may have the potential to be cost effective, energy efficient and suitable for long-term usage (Torpy et al., 2015).

5.1.4 Green wall use for pollution removal

Building on the 30 + years of studies investigating the use of potted plants to remove air pollutants and advancements in air phytoremediation, active green walls (also known as active plant walls, functional green walls, phytosystems and botanical biofilters) have been developed. Green walls are vertical structures in which one or several plant species are grown on a soil or a soilless support fabric or growth medium (Manso and Castro-Gomes, 2015). Apart from being aesthetically pleasing, green walls provide environmental, social and economic benefits which can be attributed to their design, plant choice, density of vegetation and location (Beecham et al., 2019). For example, green walls have shown to be capable of removing VOCs (Darlington et al., 2001; Chen et al., 2005; Wang and Zhang, 2011; Wang et al., 2014; Lee et al., 2015), PM (Irga et al., 2017; Pettit et al., 2017) and CO_2 (Torpy et al., 2017). Currently, this technology is being developed by numerous research groups and companies, as such, several active botanical biofilters have been developed. Although these systems differ in design, they all use active airflow using devices such as impellers that increase

the flow of polluted air to the active components of the systems and therefore allow large volumes of air to be processed. Many questions remain however, especially regarding pollutant removal efficiencies that may arise due to plant species-specific differences.

5.1.5 Plant use for VOC and PM removal

Plant species selection has been shown to be influential on the overall VOC, PM and CO_2 removal capacity of active green walls (Torpy et al., 2014). Regarding VOC removal, rhizospheric bacteria are the primary agents of VOC removal (Wood et al., 2002); however, plant-associated effects also play a role in VOC removal as shown by Irga et al. (2017). Certain groups of plants have shown the potential for higher VOC removal (Kim et al., 2016), however, the specific plant features that are influential remain unclear.

Pettit et al. (2017) examined the influence of plant species on active green wall PM single-pass removal efficiency (SPRE), focusing on the anatomical components of different plant species that correlated with improved SPRE. Fern species recorded the highest removal efficiencies across a range of particle size fractions. Upon assessing plant morphological data, it was found that the plant root structure most strongly influenced removal efficiency.

5.1.6 Plant use for CO_2 removal

Botanical biofilters provide promising potential for reductions in ambient CO_2, which could be of use in indoor environments, where a large proportion of the energy consumed by existing mechanical ventilation systems is used for CO_2 dilution (Redlich et al., 1997). Different plant species have strongly variant efficiencies for photosynthetic CO_2 removal, due both to their differing requirements for light, along with different intrinsic photosynthetic rates per unit of leaf area, which interacts with the average leaf area per plant that can functionally fit into vertical garden systems for different plant species (Torpy et al., 2017). For example, Torpy et al. (2017) showed that *Chlorophytum comosum* removed 13% of chamber CO_2 at a light

intensity of 50 µmol m^{-2} s^{-1} photosynthetically active photon density, whist *Epipremnum aureum* removed < 1%. At an increased light level of 100 µmol m^{-2} s^{-1}, *Chlorophytum* removed 20% of chamber CO_2, whilst *Epipremnum* removed only 8%, highlighting the importance of species selection for effective pollutant removal under specific conditions.

5.1.7 Australian native species

Currently, significant development of active green wall technology is being conducted in Australia. Australia's climate is highly variable, whilst being relatively warm and dry, which has significantly influenced Australian native plant species evolutionary traits. Australian native plants are also subject to a scarcity of essential abiotic factors including water and nutrients. Due to the very low phosphorous availability in Australian soils (Kooyman et al., 2017), many species have developed genetic adaptations to survive (Sulpice et al., 2014). Many Australian native plants have a range of water conservation traits (Wright et al., 2001) and nitrogen fixating capabilities (Sprent et al., 2017). Many Australian native species have evolved a high level of drought tolerance, through small evergreen leaves, comparatively high root biomass, high leaf mass per unit area, and stomata adapted to water use efficiency in water-limited environments, indicative of drought tolerance (Ullmann, 1989; Brodribb and Hill, 1993; Pasquet-Kok et al., 2010), all traits associated with water conservation (Schenk and Jackson, 2002; Thompson, 2005). Due to their capacity to grow in unfavourable environmental conditions, Australian native species may be suitable for green wall use elsewhere, due to reduced watering requirements and ability to survive dry spells that may occur in outdoor applications, or due to maintenance failure indoors.

5.1.8 Aim

Previous research examining plant species differences in active green wall pollutant removal have been limited to common ornamental species. As the green infrastructure industry becomes more water conscious, locally focused and 'ecofriendly', there is a growing interest in the use of Australian native species for urban greening. To date, however, the pollutant removal capacity of Australian natives has not been tested. The aim of this chapter, therefore, was to determine what capacity Australian native species have for actively removing CO_2, PM and VOC, and to compare these removal rates to previously tested ornamental species, and thus to determine whether the native species are appropriate for phytoremediation use.

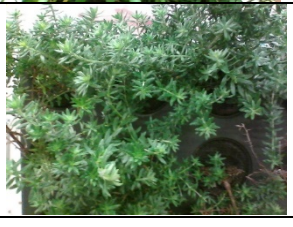
5.2: METHODS

5.2.1 Plant species

The plants used had been nursery grown for 6 months prior to testing. All tested plants were healthy upon visual inspection, as is normal practice in the horticultural industry, roughly the same size, and of the stock that is currently used in commercial green walls. Plants were supplied by Junglefy Pty (Sydney) and planted in Junglefy's green wall modules as previously described in Chapter 1, Section 2.2, and supplied with active airflow as described in the next section. There were inconsistencies both between and within species for biomass variables such as leaf area and height; however, the test plants were representative of their expected performance *in situ*; thus such inconsistencies are innate to the system, and their elimination would misrepresent their real-world performance. Plants were supplied in a substrate type and volume that were consistent across species modules and representative of *in situ* application. All plant modules were watered to field capacity and allowed to drain prior to testing. All testing was conducted between 0900 and 1700 which is when natural photosynthetic activity normally occurs. When not being tested, all modules were maintained on the University's

rooftop, with all plants exposed to the same daily environmental conditions, including ambient outdoor light conditions and watered as required (approximately once per week). The plant species chosen were common Australian native species that display growth habits indicating suitability for green wall applications (Table 7).

Table 7: The Australian native plant species used for the pollutant removal efficiency tests.

Species Name	**Common Name**	**Clade**	**Image**
Blechnum gibbum (Labill.) Mett.	Silver lady	Monilophyte	
Callistemon citrinus (Curtis) Skeels	Bottlebrush	Eudicot	
Dianella caerulea Sims	Native flax	Monocot	
Eremophila glabra (R. Br.) Ostenf.	Emu bush	Eudicot	
Lomandra longifolia Labill.	Basket grass	Monocot	
Westringia fruticosa (Willd.) Druce	Coastal rosemary	Eudicot	

5.2.2 Single-pass removal efficiency chamber

For the determination of VOC and PM removal efficiencies, a flow-through chamber was used, previously described in Irga et al. (2019). In brief, the modules were placed in a sealed Perspex chamber (0.6 m^3, 216 L). Ducting connected the chamber's front facing side to a second smaller chamber, where pollutants were generated. A 100 mm diameter 16 W fan was connected to the port on the back of the module to facilitate pollutant flow through the biofilter (see Abdo et al., 2016 for details). A 40 mm electric fan was situated within the Perspex chamber to circulate and homogenize the pollutant concentration. Attached to the back of the Perspex chamber was additional ducting leading into a 15 L Perspex chamber which housed the pollutant recording device. The filtered air was then removed through a vacuum exhaust.

The SPRE was determined by the percentage of pollutant that was removed from the air stream by the biofilter in relation to a control treatment without any biofilter present inside the chamber. Control data was also used to quantify any background pollutant removal caused by the flow-through apparatus. The following equation was thus used to determine the SPRE:

$$\text{SPRE (\%)} = [(\text{Pollutant amount}_{\text{control}} - \text{Pollutant amount}_{\text{trial}}) / \text{Pollutant amount}_{\text{control}}] \times 100$$

5.2.3 VOC trials

To determine the native species' VOC removal capacity, the same method described in Irga et al. (2019) was applied. In brief, gaseous benzene was used as the VOC in this experiment (solubility at 25 °C = ~ 1/71 g L^{-1}). 4.0 mL of liquid benzene was poured into a 10 mL sealed glass vial and allowed to stabilize for 24 h. 2.5 mL of the VOC saturated vapour from the headspace of the vial was removed with a gas chromatograph plunger syringe and injected into the pollution generation chamber such that the vacuum created by the in-duct fan passed the VOC through the system. The concentration of the benzene after passing through the biofilter

was then monitored for a 10 min period using a photo-ionization detector (PID; ppbRAE 3000, RAE Systems, San Jose, CA, USA). This process was repeated 4 times for each species.

5.2.4 PM trials

To determine the native species PM removal capacity, the same method used by Pettit et al. (2017) was applied. In brief, PM was generated by burning 4 µL of filtered retail grade diesel fuel (Shell) absorbed onto a 1 cm^2 536:2012 80 gsm square piece of paper in the pollution-generating chamber. In the pollutant-detecting chamber, a laser nephelometer (Graywolf PC-3016A, Greywolf Sensing Solutions, CT, USA) was used to record the average PM density and size distribution for a 10 min period. The average PM concentration was recorded for each of the following PM size fractions: $PM_{0.3–0.5}$, $PM_{0.5–1.0}$, $PM_{1.0–2.5}$, $PM_{2.5–5.0}$ and $PM_{5.0–10.0}$. This process was repeated 15 times for each species.

5.2.5 CO_2 chamber trials

To determine the CO_2 removal capacity of Australian native species, static chamber CO_2 monitoring was conducted, using the methodology outlined in Torpy et al. (2017). More specifically, all testing was conducted in a 216 L air-tight Perspex chamber containing a 40 mm electric fan to circulate air. Plant species were tested one at a time, with 3 independent replicates per species. The light source used was a 90 W/0.4 A red-blue plant growth-specific LED array which contained a ratio of 2:1 red to blue LEDs, with a total of 90 LEDs ('UFO' grow light, China). This type of lighting has been shown to provide an adequate spectrum of light for plant growth (Massa et al., 2008). Photosynthetic photon flux density was measured using an Apogee quantum sensor (Apogee Instruments, UT, USA). The light level used in this project ranged from 1505.5 µmol m^{-2} s^{-1} at the uppermost level of the foliage of the green wall to 111.6 µmol m^{-2} s^{-1} at the bottom of the green wall. This light was selected as it approximated the maximum light level achievable in an indoor setting. For each trial, the starting CO_2

concentration within the chamber was ~1000 ppmv, which is the ASHRAE (2011) recommended maximum for air-conditioned buildings. The CO_2 concentration was monitored using an Infra-Red Gas Analyser (IRGA; TSI IAQ-CALC, TSI Inc., MN, USA) which was sealed inside the chamber. The test was allowed to run for 40 min. Chamber leakage control treatments for the CO_2 removal trials used chambers with a starting concentration of ~1000 ppmv CO_2 with no plants present, also monitored for 40 min. The duration of 40 min was chosen based on previous studies (Torpy et al., 2014) that showed that after a 40 min duration, the rate of CO_2 removal was no longer exponential and the relative humidity increased to a point that affected CO_2 conditions.

Substrate only/no plant treatments were also tested to allow separation of the effects of substrate respiration from plant photosynthetic activity or respiration.

5.2.6 Morphological traits

Once the chamber tests had been completed, the plants were removed from the module and the substrate washed from the roots. Plant morphological characteristics were then recorded to determine if they were influential on either VOC or PM removal. Four individual plants of each species were used as replicates for each trait.

Digital callipers were used to determine the root and leaf diameters, recording 4 composite measurements per plant across the 4 replicate plants. The root and leaf fresh and dry weights were recorded using a 4 decimal place scale. Dry weights were recorded after the samples had been oven dried for 7 days at 60 °C. Root and leaf areas were determined using a leaf area machine (Licor LI-3000-A, NE, USA).

5.2.7 Statistical analysis

The data was checked for homogeneity of variance using Levene's test and checked for normality with a Kolmogorov-Smirnov test. A one-factor ANOVA and Tukey's *post hoc* test was used to determine differences in PM and VOC SPREs amongst species. A one-factor ANOVA and Tukey's *post hoc* test was used to determine differences amongst leaf and root morphologies. Pearson correlations were used to determine the strength of the association between the different plant traits and CO_2, VOC and PM removal. To compare the native species' capacities to remove CO_2, regression models were made from each chamber trial to calculate the CO_2 removed or generated after a 60 min period. The data at the 60^{th} min was used to statistically compare species CO_2 removal capacities. This was done by conducting a one-factor ANOVA and Tukey's *post hoc* test. Statistical analyses for this chapter were performed using IBM SPSS Statistics Version 21 (IBM Corp, Armonk, NY, USA).

5.3: RESULTS & DISCUSSION

5.3.1 Australian native plant species VOC removal efficiency

There were significant differences amongst the native species benzene removal efficiencies ($P = 0.000$; Fig. 17). *Dianella* had the highest SPRE of 59.04% and *Lomandra* had the lowest removal efficiency of 39.96%. The difference in removal efficiencies between these two species was surprising due to their similar morphologies, notably their similar leaf areas. The substrate only control SPRE was significantly lower than only the *Dianella* ($P = 0.000$) and *Blechnum* modules ($P = 0.004$), indicating that in most cases, soil microorganisms are probably the main agents of VOC removal. *Dianella* benzene SPRE was significantly greater ($p < 0.05$) than every species except *Blechnum*, which was the second most efficient species for benzene SPRE. The reason for these species having higher removal efficiencies may have been due to

these plants modifying the hydrophobicity/hydrophilicity of the substrate such as to alter the affinity of the VOC to the substrate binding sites (Irga et al., 2019).

In a similar experiment conducted previously (Irga et al., 2019), the VOC SPREs of 4 common ornamental species in active green walls were compared, also detecting species differences for both benzene and ethyl acetate removal efficiencies. The benzene removal efficiency range amongst species recorded by Irga et al. (2019) was relatively consistent, with < 15% variability amongst species, with SPREs ranging from 45.54 – 59.50 %. The ornamental species *Nematanthus glabra*, was found to have the highest benzene removal efficiency, likely due to its high leaf wax content. In the current project, a similar range of benzene SPREs was found, indicating that using Australian native species in active green walls results in similar benzene removal efficiencies to those possible with common ornamental species.

VOC removal appears to be mainly due to substrate bacteria metabolizing the VOCs as a source of carbon (Wood et al., 2002; Orwell et al., 2004; Irga et al., 2013). As such, differences amongst root morphological characteristics may facilitate increased microbial activity if these differences result in improved nutrient supply for soil microorganisms (Kim et al., 2018), which could in turn increase VOC removal efficiency. However, as the total residence time of the benzene within the active green wall systems was < 10 min, it was probable that insufficient time for substantial microbial metabolism occurred, and instead VOC removal was likely to primarily be a simple sorption process (Irga et al., 2019). This hypothesis is supported by the absence of significant positive correlations between benzene removal efficiency and any of the plant leaf or root traits (described in detail in following sections) in both the current project and Irga et al.'s (2019) study. Further, Irga et al. (2019) recorded negative correlations between root surface area, root mass and root diameter and benzene removal; however, these correlations were fairly weak, with $r < -0.7$ in all cases. In the current project, benzene removal was not associated with any plant traits, indicating a root-plant consistency in Australian native

plant species that is different to the patterns detected in ornamental taxa. The reason for this pattern could not be resolved, although it is possible that it results from adaptations to the environments from where the native species originate, as they are all predominantly dry shrubland species, whilst ornamental species typically originate from rainforest understorey environments.

Irga et al. (2019) proposed that green wall VOC SPRE was dependent on hydrophilic adsorbent sites in the substrate which were in turn affected by increasing root mass, surface area and diameter. In the current project however, no significant correlations were observed between any leaf or root trait and VOC SPRE. It has been proposed that leaf components allow an additional pathway for VOC removal via the stomata and cuticle (Gkorezis et al., 2016; Jindachot et al., 2018), with large leaf areas (Parseh et al., 2018) and stomatal uptake (Setsungnern et al., 2017) being characteristics influential on benzene removal. Further, the plant leaves and leaf-associated microbes have been implicated in the ability of a plant to remove VOCs (Wei et al., 2017).

It has additionally been hypothesized that different plant species can affect both physical and chemical properties of the substrate, thus altering the VOC removal by physiochemical mechanisms (Deng and Deng, 2018). As no plant belowground morphological traits influenced benzene removal in the current project, no patterns of this type could be detected in the current work, and may indicate that these mechanisms do not extend to Australian native plants. Nonetheless, *Dianella* was found to be the most appropriate species for maximum benzene removal, and could provide valuable VOC removal effects when used in indoor botanical biofiltration systems. Additionally, it would be worthwhile investigating the efficiency of native species on removing other VOCs of concern, for example methyl ethyl ketone, which was tested for ornamental species by Torpy et al. (2018), where a 57% removal efficiency was detected.

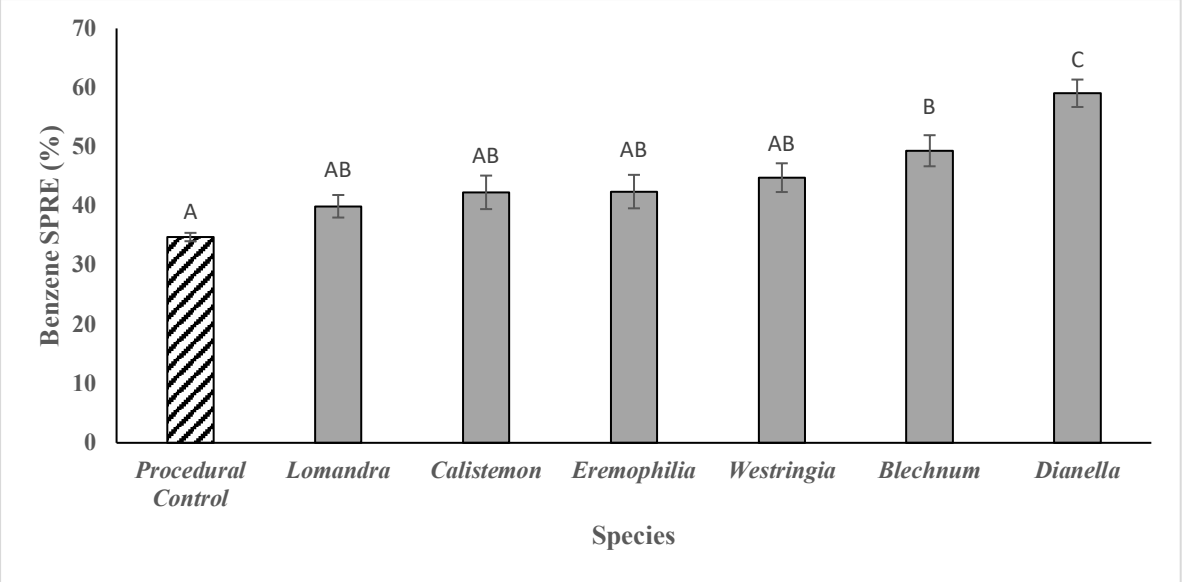


Figure 17: The different native species benzene single pass removal efficiencies; *n*=4, error bars are the standard error of the mean. Treatments with the same letter are not significantly different from each other (p>0.05 ANOVA).

5.3.2 Australian native plant species PM removal efficiency

There were no significant differences observed amongst species' SPREs for any of the PM size fractions: $PM_{1–2.5}$, $PM_{2.5–5}$ and $PM_{5–10}$ ($P > 0.05$). For $PM_{0.5–1}$, the only significant difference observed was between the *Dianella* and *Eremophila* species ($P = 0.003$). The smallest PM size fraction, $PM_{0.3–0.5}$, produced the greatest species differences, where active green walls containing *Callistemon* species were found to be significantly different to every other species ($P < 0.05$), although the direction of these differences was variable (Fig. 18). The *Dianella* and *Lomandra* species were both relatively inefficient at $PM_{0.3–0.5}$ removal, filtering significantly less PM of this size fraction than every species except one another. As was the case in the study by Pettit et al. (2017), differences across the species SPREs for PM size fractions were also detected, with SPRE generally increasing as the PM size fractions increased.

In a study conducted by Pettit et al. (2017) testing ornamental plant species using the same apparatus as the current research, considerable differences amongst different species' PM SPREs were found, with the fern, *Nephrolepis exaltata* '*Bostoniensis*' demonstrating the

highest removal efficiencies of 45.78% and 92.46% for $PM_{0.3–0.5}$ and $PM_{5–10}$ respectively. In the current project, the active green wall plant species tested had generally lower removal efficiencies across all PM size fractions than the ornamental species tested by Pettit et al. (2017). Furthermore, Lee et al. (2015) determined that their ornamental plant wall system had a 65 – 90 % PM removal efficiency, which was also considerably greater than the native species removal efficiencies detected in the current chapter.

Pettit et al. (2017) noted the influence of root structure on the PM SPRE of active green walls, proposing that different root structures modified the structure and physiochemical properties of the substrate, which increased filtration capacity. More specifically, the simple, rhizomatous root systems of ferns and herbaceous species were associated with more effective filtration characteristics, compared to woody plants which typically have complex, branching root systems (Dong et al., 2015). In the current project however, no specific root features nor leaf traits were found to be correlated with high PM SPRE. Pettit et al. (2017) suggested that species which have leaves that grow horizontally and sitting at a perpendicular angle to the green wall face allow greater foliar impaction, compared to species which have their leaves arranged at a more prominent vertical angle. Although the *Callistemon* had leaves which were arranged angled upwards, potentially increasing the PM absorption area, its performance in the current study was not different to the other species tested. Additionally, the *Callistemon* species was the only tested species which had leaf hairs, a trait shown previously to be advantageous in PM accumulation (Beckett et al., 2000; Sæbø et al., 2012; Leonard et al., 2016; Chen et al., 2017), which could be predicted to have an influence on the SPRE. Whilst these leaf structures are known to increase PM filtration efficiency, surprisingly all native species PM SPRE were similar to one another.

When grown vertically in green wall systems, the root structures of tree and other woody species may be restricted, unlike plants such as ornamental ferns that generally grow in dense colonies (Coelho et al., 2014; Large and Farrington, 2016; Ng et al., 2016). This may have increased root competition effects (Pettit et al., 2017). In the current project, however, the woody *Callistemon* and *Westringia* species displayed a similar SPRE to the *Blechnum* fern and monocot shrub species tested. It is likely that this was a result of the considerable root morphological differences between *Blechnum* and the ornamentals tested by Pettit et al. (2017). This is evidence that plant influence on biofilter pollutant removal performance should not be generalized across broad taxonomic groupings, and individual species' performance should be tested in isolation.

In conclusion, active botanical biofilters containing Australian native species were shown to be able to effectively reduce PM, with all tested species having similar SPRE values. However, the SPRE of the native species was lower than the previous recorded SPRE values of ornamental species.

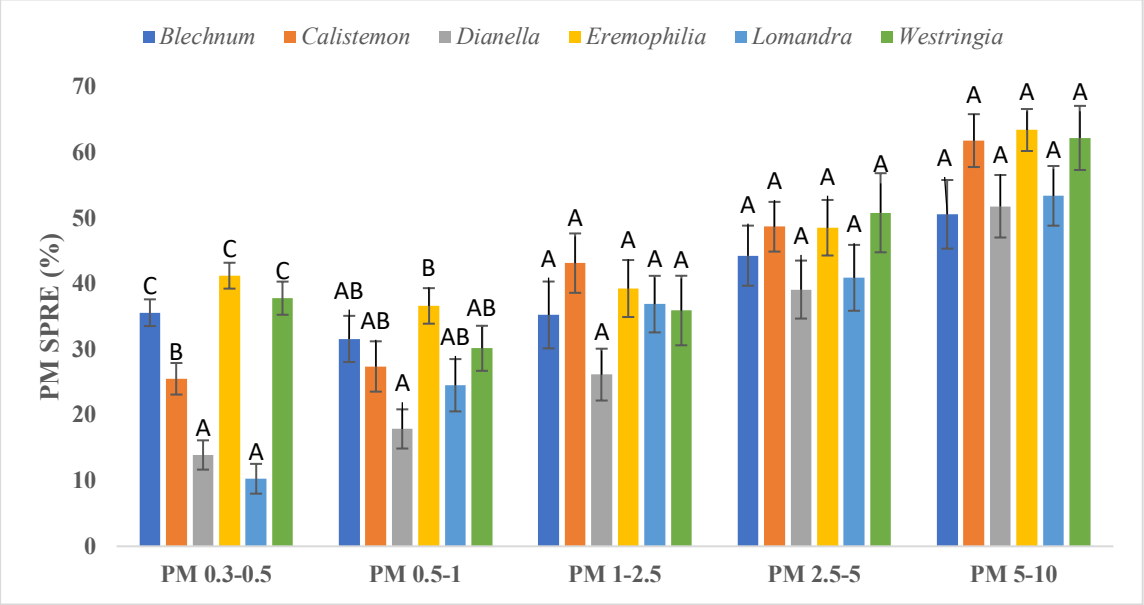


Figure 18: Australian native plant species' PM single pass removal efficiencies across different PM size fractions; n=15, error bars are the standard error of the mean. Treatments within each particle size fraction with the same letter are not significantly different from each other (p>0.05 ANOVA).

5.3.3 Australian native plant species' CO_2 removal efficiency

The final concentration of CO_2 for all species was significantly higher than the CO_2 empty chamber leakage data ($P < 0.05$), indicating that all biofilters generated CO_2 under the lighting conditions used. There were no significant differences between any species and the substrate only control treatments ($P > 0.05$), indicating that soil microorganism respiration dominated the CO_2 generation observed. Nonetheless, several significant differences were observed for the CO_2 generation rate amongst species, with *Eremophila* producing the greatest amount of CO_2 and *Blechnum* producing the least ($P = 0.005$). *Eremophila* and *Westringia* also produced significantly more CO_2 than *Callistemon* and *Blechnum* ($P = 0.032$ and $P = 0.023$, respectively). There were no other significant differences amongst species CO_2 removal efficiencies, indicating that none of the native species tested was able to remove CO_2 under the light levels used. This is evidence that these species would be of little use for the phytoremediation of this gas indoors.

Pennisi and van Iersel (2012) noted that due to the low light levels of indoor environments, an impractical number of potted plants would be needed to make a significant impact to indoor CO_2 levels. Torpy et al. (2014) nonetheless identified plant-light level combinations that could lead to some reductions in indoor CO_2, but added that for adequate CO_2 removal, plants would require higher light levels than those generally used indoors. At the light levels tested, all native species green walls increased the total CO_2 concentration in the test chambers. This was due to respiration by the microorganisms located within the substrate (Somova and Pechurkin, 2001; Torpy et al., 2017). As all plants were maintained under natural sunlight conditions prior to and also during the experiment when not being tested, it is not likely that the plants' inability to reduce CO_2 was related to photo-inhibition (Torpy et al., 2014).

The photosynthetic photon flux density supplied to plants is a key determinant of the CO_2 removal capacity of different plant species. Whilst the natives tested in the current chapter were ineffective for CO_2 removal, Torpy et al. (2017) found that active green walls containing *Chlorophytum comosum* and *Epipremnum aureum* could remove some CO_2 at higher light levels (50 μmol m^{-2} s^{-1} (2375 lux)). Similarly, in a study conducted by Gubb et al. (2019), it was determined that their ornamental species (*Spathiphyllum wallisii, Dracaena fragrans* and *Hedera helix*) could remove 1000 ppm of CO_2 at 22,200 lux. The light levels used in the current study were significantly higher than both tested light levels in Torpy et al.'s (2017) study and were predicted to be sufficient to promote net photosynthesis by the Australian native plants tested, despite these species being known to require comparatively high light levels (Borthwick et al., 1952; Toole et al., 1955; Willis and Groves, 1991; Bell, 1993). The light source in the current project ranged from 1505.5 μmol m^{-2} s^{-1} at the uppermost foliage of the green wall to 111.6 μmol m^{-2} s^{-1} at the bottom of the green wall. The light levels normally used in indoor environments range between 4 and 10 μmol m^{-2} s^{-1} (180–460 lux; Safe Work Australia, 2011), with the light levels used in this project considered a practical maximum possible in indoor environments with the use of targeted plant location or plant-specific lighting systems. As the plant species tested in the current work were unable to remove CO_2 at maximum achievable indoor photon flux densities, it is proposed that certain ornamental species will be more effective than natives for CO_2 removal in indoor applications, unless systems can be located near to a very strong natural light source.

Whilst it is likely that the native plants were photosynthesizing, this rate was clearly insufficient to offset respiratory CO_2 emissions from the plant and substrate. This phenomenon has been observed previously (i.e. Torpy et al., 2014). Whilst the height, leaf area and other biomass variables were inconsistent between and within cultivars, the individual plants used were identical to those used in their intended commercial applications and were thus accurately

representative of their performance when used *in situ*. Plant cultivar CO_2 removal on a whole plant plus substrate basis was assessed, rather than removal on a per unit leaf area basis. Whilst the latter scale has intrinsic value, it has limited relevance in practical applications, as indoor plants are not selected based on the quantitative leaf area they provide. Substrate type and volume were consistent for all species, as per commercial practice. The light level used is generally sufficient for ornamental indoor plants to photosynthesize (Torpy et al., 2014); however, it was clearly not adequate to balance the combined CO_2 emissions from the native plants and their substrates, and thus, more light than the currently used level would need to be supplied if native green wall systems were to be used for indoor CO_2 removal purpose (Fig. 19). Given the constraints of contemporary interior design practice, this would be impractical at the current time.

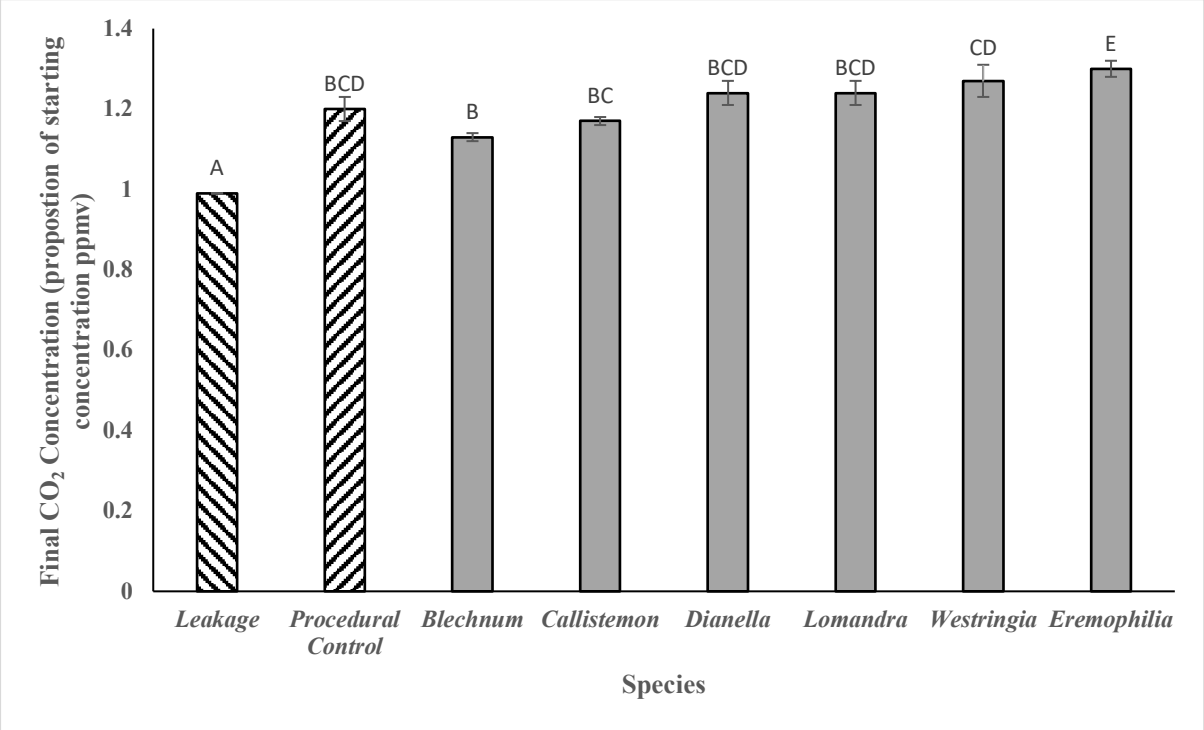


Figure 19: The projected average final CO_2 concentrations at the 60th minute across the different Australian native species, displayed as the proportion of the starting concentration of 1000 ppmv. Data are means ± the standard error of the mean, n=3. Treatments with the same letter are not significantly different from each other (p>0.05 ANOVA).

5.3.4 Plant morphological data

Leaf and root morphology were variable amongst species (Table 8), with significant differences observed amongst leaf widths, leaf areas, leaf fresh weights and leaf dry weights (all P = 0.000). There were also significant differences amongst the species' root diameters (P = 0.023). Figure 20 illustrates the differences between the species' root morphologies.

Table 8: Australian native plant species leaf and root morphological traits. All data is representative of the respective traits within a singular green wall module, which contains 16 individual plants. Data are means ± the SEM (*n* =4).

Plant Species	**Average Leaf Width (mm)**	**Leaf Area (m^2)**	**Leaf Fresh weight (g)**	**Leaf Dry Weight (g)**	**Average Root Diameter (mm)**	**Root Area (m^2)**	**Root Fresh Weight (g)**	**Root Dry Weight (g)**
Blechnum	11.2 ± 0.66	359.2 ± 0.25	443 ± 0.00	52.9 ± 0.00	1.14 ± 0.06	14.48 ± 9.23	136 ± 1.65	19.6 ± 0.23
Callistemon	7.06 ± 0.18	154.6 ± 0.12	418 ± 0.00	162 ± 0.00	1.58 ± 0.19	13.10 ± 9.89	107 ± 0.60	28.8 ± 0.15
Dianella	15.0 ± 0.46	239.3 ± 2.28	632 ± 0.07	172 ± 0.02	1.17 ± 0.07	24.01 ± 5.15	198 ± 1.09	30.6 ± 0.14
Eremophilia	10.1 ± 0.34	364.6 ± 0.12	1278 ± 0.00	301 ± 0.00	1.34 ± 0.14	6.30 ± 6.05	49.6 ± 0.46	15.6 ± 0.20
Lomandra	10.3 ± 0.30	238.4 ± 2.71	1499 ± 0.26	721 ± 0.07	1.24 ± 0.10	1.63 ± 12.5	129 ± 0.73	37.6 ± 0.26
Westringia	3.88 ± 0.10	301.7 ± 0.03	792 ± 0.00	193 ± 0.00	1.04 ± 0.08	26.04 ± 6.51	96.0 ± 1.07	21.7 ± 0.11

Figure 20: Root structures of the tested species. A: *Blechnum,* B: *Callistemon,* C: *Westringia,* D: *Lomandra,* E: *Dianella,* F: *Eremophilia*.

5.3.5 Associations between plant morphological traits and pollutant removal efficiencies

No leaf nor root morphological trait was found to be significantly correlated with VOC, CO_2 or PM removal efficiencies (all P values > 0.05; Table 9), indicating that these plant characteristics were not the cause for species differences.

Table 9: Correlation analysis across species plant morphological traits and the different removal efficiencies for the three tested pollutants. TSP = total suspended particulates.

Pollutant Type	**Statistical Result**	**Leaf width**	**Leaf area**	**Leaf fresh weight**	**Leaf dry weight**	**Root diameter**	**Root area**	**Root fresh weight**	**Root dry weight**
VOC	P	0.496	0.601	0.572	0.560	0.754	0.135	0.355	0.156
	r	0.351	-0.273	-0.294	-0.303	0.166	-0.682	-0.463	-0.659
CO_2	P	0.717	0.118	0.112	0.110	0.650	0.241	0.780	0.306
	r	0.191	0.705	0.713	0.715	-0.238	0.567	0.148	0.506
TSP	P	0.180	0.069	0.143	0.143	0.803	0.102	0.064	0.051
	r	0.630	0.778	0.673	0.672	0.132	0.727	0.787	0.809

5.4: CONCLUSION & FUTURE DIRECTIONS

It is important to note that the results obtained in the current project can only provide a surrogate indication of the removal efficiency of the plant species tested, due to the obvious differences between chamber studies and the built environment. Chamber studies cannot realistically be extrapolated to real-world building environments (Llewellyn and Dixon, 2011; Irga et al., 2013; Soreanu et al., 2013) due to the plant density per unit volume of experimental chamber atmosphere being far higher than would be possible in buildings (Torpy et al., 2015). The results obtained from laboratory chamber pull down experiments are thus not often projected into real-world situations due to the complex dynamics of indoor settings (Llewellyn and Dixon, 2011). Nonetheless, the results obtained in this chapter provide an indication on the more efficient species for different pollutant removal, which could be tested in *in situ* conditions to provide a more realistic removal capacity.

The Australian native species tested were shown to be effective at removing benzene, with similar SPRE values to ornamental species. *Dianella* was found to remove the greatest amount of benzene, although the characteristics of this species leading to its greater efficiency could not be resolved. The native species were also capable of reducing PM, however, at lower efficiencies than previously tested ornamental species. All tested native species were shown to be inefficient for the reduction of CO_2 at practical light levels, in contrast to previously tested ornamental species. As has been the case in previous work, pollutant removal characteristics were inconsistent amongst species. Whilst *Dianella* was found to be the highest performing species for benzene removal, it was the lowest performing species for PM filtration, indicating that plant species selection should focus on the dominant pollutant in any specific application. It is suggested that ornamental species remain the most appropriate choices for active biofilter phytoremediation use in indoor applications, due to their higher and more consistent removal efficiencies for the key pollutants likely to be found indoors. Australian native species may still have value, as their tolerance for harsh environmental conditions may be of use in outdoor biofiltration applications. Field trials in varied environments will be required before strong recommendations of plant species selection can be made for all conditions.

Chapter 6 – Significance of findings and conclusions

6.1 Significance of findings

Air pollution is of growing concern due to its adverse health effects. Traffic pollution and industrial activities are common sources of air pollution emissions, with these activities releasing harmful pollutants such as PM and VOCs. Green wall technology has been shown to be an effective air pollution remediator, however, are generally used in urban environments for intrinsic value, and as such more research is needed to assess their air pollution removal capacities.

This thesis represents an expansion on the proof-of-concept testing previously performed in a research collaboration between Junglefy Pty Ltd and the Plants and Environmental Quality Research group at the University of Technology Sydney, aimed at developing the green wall system to maximise its capacity to phytoremediate air pollution. The research presented in this thesis relates to: identifying the capacity for common green wall species to accumulate airborne PM, experiments for green wall species health associated from continual pollution exposure, studies to determine the effectiveness of pre-existing green walls at making pollution, noise and temperature reductions *in situ*, and finally an assessment of the appropriateness for Australian native species to be used in active green walls for air pollution mitigation.

This thesis first addressed the gap in knowledge for PM deposition on green walls used within Australian urban environments, and additionally assessed leaf traits for enhanced PM accumulation which has been previously shown to vary amongst plant species. Previous work on the PM removal capacity of vegetation has been generally limited to studies conducted overseas, mostly in Europe, with no work being done on species used within Australian green walls. Different regions of the world have different pollutant concentrations and climates,

making it unreliable to make green wall species comparisons across countries; highlighting the need for locally specific studies. Previous studies have also noted the role of leaf characteristics in enhanced PM accumulation, notably leaf hairs and waxy cuticles. Ideal leaf traits, however, remain inconsistent throughout the literature. It is currently difficult to accurately select the best species for most locations. The results of this thesis, therefore, were the first to provide knowledge on common green wall species PM accumulation within Sydney, Australia and to quantitatively analyse leaf traits as they relate to PM deposition. This thesis was also the most spatially and temporally representative study performed to date, and spanned the performance of multiple species. The majority of previous studies in comparison have examined species from only one site, for only a short amount of time, in most cases, only examining single species. This thesis therefore incorporated site and seasonal variation, which undoubtedly have an impact on species PM accumulation capacity. Furthermore, this study assessed PM deposition for 11 species, thus incorporating far greater interspecies effects than previous work on plant PM filtration. The temporal, spatial and multi-species replication of this thesis therefore provided much needed information in an important field of research.

The finding that all tested species were able to accumulate airborne PM at different rates highlights the importance of species selection for enhanced PM removal, which will become vital for future *in situ* green wall performance. Tests for influential leaf traits were found to differ from previous literature, with small leaved species accumulating the least PM, with the species containing leaf hairs (Variegated Mintleaf) being the least efficient species. This highlights the difficulty in attributing leaf traits to increased PM accumulation. With these findings, green wall systems can be better designed by identifying high accumulating species such as Xanadu for enhanced *in situ* particulate removal efficiency.

Whilst there is a broad array of existing knowledge for plant health effects from pollution exposure and stress conditions, this has commonly been documented for soil pollution, water stress and nutrient deficiencies; with these factors not being applicable to green wall systems, which are frequently maintained, use integrated irrigation systems and a nutrient rich growing substrate. Furthermore, the majority of tests conducted on plant health associated effects from air pollution exposure have focused on bio-indicator species or shrub species used in green belt development, overseas. In particular, most of these studies are limited to Southern Asia and do not assess common green wall species, nor green wall systems on a whole. If green walls are to effectively reduce ambient pollutant conditions, the health of the vegetation is among one of the most important aspects. Therefore, this thesis provided much needed information on common green wall species plant health impacts from long term *in situ* air pollution exposure. The health variables assessed; chlorophyll content, leaf extract pH, RWC and carbon content all play vital roles in overall plant health, function and productivity. Furthermore, carbon content has not previously been tested in plant health studies. The current findings indicate that it is correlated with RWC, providing evidence that it may be worth including in future studies. These results indicated that the common green wall species were able to withstand *in situ* pollution conditions and could continue to grow and thrive irrespective of the pollution levels. The results showed that some species are more susceptible to pollution damage, from decreased health variables at the polluted test locations compared to the non-pollution controls. There however, was no standout species for pollution tolerance or sensitivity, with some species being tolerant in one plant health category but sensitive in another, but overall, all species demonstrated suitability for use *in situ*.

Urban vegetation is known to be capable of reducing ambient pollutant concentrations. Trees have been found to be the most efficient form of vegetation due to their large leaf size and the turbulence created by tree crowns, however, in an ever increasing urbanized world, the space

available for planting trees is rapidly declining. Green wall technology is thus an ideal solution, as it incorporates vegetation into the urban environment without taking up any additional space at the street level. Studies have suggested that green walls provide an array of environmental benefits including air pollution mitigation, noise pollution reduction and the abatement of the urban heat island effect. However, the majority of experiments that make claims on green wall remediation capacities for ambient PM, noise and temperature conditions, have been a product of computational modelling and simulation studies. These studies cannot accurately replicate complex *in situ* conditions that include wind patterns, humidity and varying air quality, which have an effect on the ambient conditions, and the capacity for vegetation to reduce negative effects. Furthermore, street layout, building height and aspect ratios all have an influence on the capacity for green walls to mitigate urban heat and noise pollution. As such, computational studies have not been able to accurately determine ambient reductions from green wall presence, and it has thus been suggested that the reported reductions have been exaggerated. This thesis therefore filled a gap in research regarding *in situ* field trials for the removal of PM, noise and temperature conditions by green walls. The results presented in this thesis were the first to conduct *in situ* air quality, noise and temperature assessments within the Sydney region from the presence of green walls. The results indicated that there were no significant differences observed between the green wall and reference walls across the 12 sites, over the 6 month study for PM concentrations nor temperature conditions. This finding is significant as it indicates that the current systems implemented for pollution and temperature abatement are not effective and as such, modifications can be made to enhance their efficiencies *in situ.* A significant difference was however, found between wall types for noise, with the green walls having significantly lower ambient noise conditions. Notably, maximum reductions of 12.13 dB were found at one of the test sites, highlighting the significant noise reduction capacity of

green walls. Previous laboratory studies conducted using the test green walls have recorded maximum noise reductions of only 4 dB in comparison.

The findings that passive green wall systems were not highly effective for PM pollution removal, and that some plant species exhibited pollution sensitivity in certain plant health categories lead to the final project which assessed native species used in active green walls. Previous laboratory studies have indicated that active green wall systems may be more effective at pollutant removal than passive systems. Further, all previous green wall studies have only tested common, non-native species. As such, there was a gap in knowledge regarding Australian native species capacity in active green wall systems to reduce air pollutants. Australian native species should be able to withstand the harsh environmental conditions of Australian better than ornamental species, making them ideal for use *in situ*. However, irrespective of their tolerance to environmental conditions, if they are unable to remove air pollutants at an effective rate, their use *in situ* becomes invalid. Therefore, this thesis was the first to assess the air pollutant removal capacity of native species used in active green walls.

The native species' pollutant removal capacity was determined by calculating the SPRE, which is a standardized means of reporting air filter pollutant removal efficiency. Henceforth, this thesis allowed for comparisons to not only previously conducted studies on biofilters, but other air filtration devices such as those currently used in mechanical building air filtration systems. This thesis utilised a state of the art flow through chamber to determine the species SPRE values. The native species' benzene and PM SPRE, and CO_2 removal capacity were determined, and these removal abilities compared to previously tested ornamental species. The results of this thesis indicated that Australian native species had similar benzene removal efficiencies to previously tested ornamental species, with *Dianella* species having the highest removal efficiency. This result was significant as it indicated that native species are appropriate

for green wall use for VOC removal with similar removal rates to ornamental species. The native species were also able to remove PM, however, at comparably lower efficiencies than ornamental species. This finding indicated that ornamental species are more appropriate for PM removal, at least considering the small subset of native species tested. Lastly, all tested native species were not able to reduce CO_2 concentrations under reasonable lighting conditions. This finding was indicative that native species are not appropriate for indoor CO_2 phytoremediation due to their high light requirements. For now, it appears that ornamental species are more appropriate for indoor air phytoremediation, however, this thesis provided base line, proof-of-concept that native species are able to remediate PM and VOC pollution, which may become fundamental if the future of green wall implementation is aimed at increasing urban biodiversity.

In conclusion, the current findings have strengthened our understanding of the PM filtering efficiency of pre-existing green walls and species-specific differences in PM accumulation. The experiments have identified a number of specific aspects of green wall components, including certain species and the conversion of passive systems to active systems, that could be altered to obtain a higher efficiency. An extensive review of plant species' tolerance and health effects associated with *in situ* pollution concentrations was also provided. Finally, initial trials on native species used in active green walls for air pollutant removal was provided, indicating that active green walls are an effective air filtering system, with the use of native species worthwhile testing further.

6.2 Future directions

Discrepancies exist in literature regarding plant PM deposition capacities, which may be due to the differing methodologies used to determine leaf PM deposition (Weerakkody et al., 2017). For example, SEM imaging analysis, filtration and gravimetric assessment are among the methods used to determine deposited leaf PM; however, each method is with its flaws. To date, there is no consistent, accurate methodology which can provide information on both the mass of deposited PM and its associated size fraction breakdown, with both forms of information being vital to the determination of botanical PM removal efficiency. As such, it is suggested that future studies assess the accuracy of the commonly used methodologies to develop a method that can report PM accumulation on both a total accumulated mass and PM size fraction basis, to allow for a standardized method of leaf PM accumulation.

Future studies should also incorporate a larger range of plant health tests and expose the green walls to more intense pollution to better discriminate between pollutant tolerant and sensitive species. Similarly, future studies should also examine a wider range of native species, across various pollutant conditions, and the health of the native species should also be examined to determine if they will continue to grow irrespective of the pollution and environmental conditions, compared to the potential sensitivity of ornamental species.

Whilst this thesis assessed a range of different sized green walls, even at the largest green wall tested, ambient PM reductions were not observed. This suggests that perhaps ambient pollutant removal is not based on the size of a green wall, but rather is dependent on the density of green walls within an area. It is plausible that having a greater number of green walls present in an urban environment will lead to significant pollutant reductions. It has previously been estimated that an 80% vegetation coverage of urban forestry is needed to make noticeable pollutant reductions. As such, it would be worthwhile for future studies to determine what

percentage coverage of green walls within a given area is needed to cause significant pollutant reductions. The balance between the density of walls that could support green walls and their potential pollutant reductions would thus indicate the true value of green walls as pollution mitigation systems in urban environments. Furthermore, if the required coverage of green walls is not feasible, the simple conversion of passive systems to active systems is known to correlate to increased pollutant removal, although once again, the total size of active green wall required to make real differences remains unknown. Currently most outdoor green wall installations have been limited to passive systems. Passive systems of course rely on the simple diffusion of pollutants to the plant components without any form of mechanical assistance. Previous laboratory studies have however, indicated that active green walls reduce significantly greater amounts of air pollution, and as such it is plausible that significant PM reductions would be observed at the tested sites, if the passive systems were converted to active walls. As a result, future studies should involve field trials assessing the capacity of active green walls to reduce *in situ* pollutant conditions.

This thesis mainly assessed PM pollution, as it is a leading cause of global mortality, and touched on CO_2 and benzene pollution in the last chapter. Plants, however, vary in their tolerance and removal capacity when exposed to different air pollutants. As such, a wider range of pollutants should be tested in future studies to better understand plant responses. Similarly, in the final chapter benzene was used as the VOC pollutant, however, there are many different VOCs which are worthwhile testing in order to determine the capacity of green walls to phytoremediate various air pollutants. It would also be worthwhile assessing green walls capacity to reduce ozone (O_3), an important air pollutant that has not been widely tested to date.

It is plausible that significant pollutant reductions were not observed from green wall presence due to the overall low pollutant conditions within Sydney. Similarly, it is possible the green wall species did not show distinct pollution tolerance or sensitivity due to low *in situ* pollution

exposure. As a result, it would be worthwhile testing green wall pollutant removal capacity and species tolerance in known high pollution areas, such as China and India. It is likely that more prominent trends would be observed if these tests were to be conducted in regions where air pollution is a more significant issue. The results obtained in these high pollution environments would also provide benefits to the populations which are most at risk from air pollution health associated effects.

Similar to the aim of testing Australian native species which are better suited to local climatic conditions; green wall testing in other regions of the world should test species endemic to those regions, and which thrive in those particular climates. Whilst green walls throughout the world utilise common ornamental species, it is plausible that species that are native to each region could effectively reduce pollutant conditions, whilst having tolerance to the local environmental conditions. This concept should be examined in future studies to enhance the specificity, productivity and longevity of green walls.

Whilst this thesis presented results obtained over a long study period, which allowed for seasonal changes and ambient environmental condition variations to be incorporated, it would be worthwhile conducting even longer term trials in high pollution environments. This would allow for an accurate representation of the green walls' filtering capacity over time, and to see if a pollutant saturation point occurred. Furthermore, it would allow for an analysis of plant health from long term pollution exposure, providing an indication of built up tolerance over time from pollutant exposure or eventual plant death from a bioaccumulation of toxic air pollutants. This information may prove vital for an indication of ongoing maintenance requirements and the identification of self-regulating species which can thrive in high pollution environments.

As green walls are a relatively new concept which are lacking in scientific research, the systems are continually being changed and updated for better efficiency. As such it is possible that utilising different air flow rates in active systems; altering the plant species used in green walls and changing substrates could lead to enhanced pollutant removal capacity. These factors should be tested in future studies, in order to determine if any improvements can be made to the green wall systems, leading to enhanced air pollution mitigation. Just as research is lacking for green wall technology, more research is also needed for green roofs. It is possible that green roofs could have a higher pollutant removal capacity in comparison to green walls. As such, future studies should also look at the pollutant removal capacity and species pollution tolerance in green roofs and compare these findings to those from green wall studies.

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APPENDIX

Appendix 1: Results from LMM analyses of plant health variables testing the effects of species, treatment (control and green wall plants) and the species x treatment interaction.

Response	**Term**	χ^2	**DF**	**P**
Leaf chlorophyll	Species	1611.39	10	< 0.0001
	Treatment	0.96	1	0.3
	Species x treatment	195.08	10	< 0.0001
Leaf Ph	Species	2955.21	10	< 0.0001
	Treatment	0.69	1	0.4
	Species x treatment	41.51	10	< 0.0001
RWC	Species	275.56	10	< 0.0001
	Treatment	1.85	1	0.2
	Species x treatment	37.94	10	< 0.0001
Leaf carbon content	Species	2624.03	10	< 0.0001
	Treatment	1.03	1	0.3
	Species x treatment	92.82	10	< 0.0001

Appendix 2: Results from LMMs testing the relationships between all unique combinations of plant health variables, including a term for species, and a health variable x species interaction. Where non-significant, interaction terms were removed, leaving the predictor health variable and the term for species.

Response	Term	χ^2	DF	P
Leaf chlorophyll	Leaf pH	3.52	1	0.06
	Species	935.32	10	< 0.0001
	RWC	1.67	1	0.2
	Species	958.90	10	< 0.0001
	RWC x species	19.85	10	0.031
	Leaf carbon content	0.71	1	0.4
	Species	678.6	10	< 0.0001
Leaf pH	RWC	0.63	1	0.4
	Species	910.01	10	< 0.0001
	Leaf carbon content	1.69	1	0.2
	Species	641.35	10	< 0.0001
RWC	Leaf carbon content	4.77	1	0.029
	Species	164.72	10	< 0.0001

Appendix 3: Results from linear models testing whether species health variables differed among sites. Given the hypotheses of this study, the species x site interaction term is the single term of interest. Site level differences are not interpretable, given that each site featured differing suites of species.

Response	Term	SS	DF	F	P
Leaf chlorophyll	Species	135.45	10	96.93	< 0.0001
	Site	13.73	11	8.93	< 0.0001
	Species x site	7.2	27	1.91	0.006
	Residuals	34.24	245		
Leaf pH	Species	3.00	10	81.34	< 0.0001
	Site	0.10	11	2.44	0.007
	Species x site	0.05	27	0.55	0.97
	Residuals	0.90	245		
RWC	Species	52.65	10	14.7	< 0.0001
	Site	10.22	11	2.60	0.004
	Species x site	11.40	27	1.18	0.3
	Residuals	87.74	245		
Leaf carbon content	Species	32.24	10	99.5	< 0.0001
	Site	1.44	11	4.03	< 0.0001
	Species x site	1.96	27	2.24	0.0007
	Residuals	7.94	245		

Appendix 4: Results of LMMs using the plant health variables as the response, and green wall TSP, accumulated leaf PM, traffic, green wall temperature and humidity as predictors, a categorical term for species, and a predictor x species interaction term. Where non-significant, interaction terms were removed, leaving the predictor health variable and the term for species.

Response	Term	χ^2	DF	P
Leaf chlorophyll	Green wall TSP	0.16	1	0.7
	Species	920.93	10	< 0.0001
	Accumulated leaf PM	0.10	1	0.8
	Species	911.48	10	< 0.0001
	Traffic	0.21	1	0.6
	Species	921.25	10	< 0.0001
	Green wall temperature	0.45	1	0.5
	Species	921.79	10	< 0.0001
	Humidity	0.001	1	0.97
	Species	869.10	10	< 0.0001
Leaf pH	Green wall TSP	4.07	1	0.044
	species	943.11	10	< 0.0001
	Accumulated leaf PM	0.14	1	0.7
	species	934.64	10	< 0.0001
	Traffic	1.87	1	0.2
	species	931.89	10	< 0.0001
	Green wall temperature	10.63	1	0.001
	species	967.12	10	< 0.0001
	Humidity	1.52	1	0.2
	species	962.28	10	< 0.0001
RWC	Green wall TSP	21.17	1	< 0.0001
	species	182.14	10	< 0.0001
	Accumulated leaf PM	16.15	1	< 0.0001
	species	189.10	10	< 0.0001
	Traffic	0.001	1	0.95
	species	179.64	10	< 0.0001
	Green wall temperature	30.10	1	< 0.0001

	species	205.14	10	< 0.0001
	Humidity	31.41	1	< 0.0001
	species	161.76	10	< 0.0001
	Humidity x species	25.45	10	0.005
Leaf carbon content	Green wall TSP	0.01	1	0.9
	species	933.60	10	< 0.0001
	Accumulated leaf PM	0.02	1	0.9
	species	888.94	10	< 0.0001
	Traffic	0.32	1	0.6
	species	932.08	10	< 0.0001
	Green wall temperature	6.41	1	0.011
	species	994.47	10	< 0.0001
	Green wall temperature x species	23.03	10	0.011
	Humidity	5.66	1	0.017
	species	912.9	10	< 0.0001