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PLANTS AND ENVIRONMENTAL QUALITY RESEARCH GROUP

INVESTIGATIONS INTO AIR POLLUTANT CONCENTRATIONS ACROSS INNER SYDNEY AND THEIR RELATIONSHIPS WITH URBAN FORESTRY

A thesis submitted by Peter J Irga to the School of Life Sciences, University of Technology Sydney, in partial fulfilment of the requirements of the degree of Doctor of Philosophy

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Statement of Original Authorship

I certify that this thesis has not already been submitted for any other degree and is not being submitted as part of the candidature for any other degree.

I also certify that the thesis has been written by me, and that any help I have received in preparing this thesis, and all sources used, have been acknowledged in this thesis.

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Abstract

It is widely understood that the activities of plants can influence the concentration of ambient air pollutants. The research presented here assessed urban air pollution across Sydney, and examined whether higher concentrations of urban forests were associated with quantifiable effects on ambient air pollutant levels. The findings indicate that areas with higher concentrations of urban forests may lead to better air quality with respect to reduced ambient particulate matter, however, if the greenspace was composed of grass, increased fungal concentrations were observed. A further investigation was made, aimed at assessing the potential contribution of senescent leaves to the diversity of airborne fungal propagules during autumn. The fungi aerosolized from autumn leaf samples were commonly found in the autumn air samples, thus it is likely that phyllospheric fungi present on deciduating leaves contribute to the aeromycota of these urban areas. An additional investigation studied the diversity of aeromycota associated with forty urban bird roosts. Associations were established between *Rhodotorula* and Pacific black ducks, wood ducks, myna birds and miner birds. Further associations were established between *Penicillium*, *Scopulariopsis* and *Cunninghamella* and pigeons, sparrows and swallows. Indoor air quality in buildings located within the same sampling sites as used in the first study, were used to make a comparison across building ventilation types. Generalising, it was found that the indoor air quality of a typical Australian office building does not pose a health issue to occupants. As the air in naturally ventilated buildings largely resembles that of the proximal outdoor air, urban forests will influence the composition of air pollutants within these buildings, both positively and negatively. The results combined, demonstrate that urban forests does influence air pollutants substantially, either through the reduction of ambient particulate matter, or the facilitation of bioaerosols either directly or indirectly. In light of these results, I propose that the research methods developed here can be used for other field studies related to air pollutants, and that the data here not only contributes new valuable data on the distribution and behaviour of air pollutants but also identifies possible sources and preventative mechanisms.

Keywords

Aeromycota

Airborne fungi

Allergy

Asthma

Avian droppings

Bioaerosols

Environmental sources

Guano

I/O ratios

Leaf surface fungi

Mechanical ventilation

Occupational health

Offices

Particulate matter

Phylloplane

Public health

Rhodotorula

Risk assessment

Seasonal

Sydney

Street trees

Urban ecology

Workplace safety

Zoonosis

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Preface

This thesis consists of nine chapters. Chapters two to six are derived from journal articles that have been peer reviewed and published, presented in logical order. I have presented them similar to their published form; consequently, some repetition occurs in regards to themes and background, and formatting and referencing may differ slightly across chapters. To prevent unnecessary duplication, a single reference list has been provided at the end of the thesis.

This thesis is a compilation of my own work with guidance from my supervisors and additional assistance from others. I conceptualized the research, designed the experiments including choice of methods, and instrumentation, conducted all data collection and analysis, and wrote the manuscripts. My supervisors and co-authors proof-read and edited the final manuscript versions. Publication details are listed below and contributions of co-authors are detailed at the start of each chapter.

List of peer reviewed publications

The publications presented here form chapters 2-6.

Irga, P. J., M. D. Burchett and F. R. Torpy (2015). Does urban forestry have a quantitative effect on ambient air quality in an urban environment? Atmospheric Environment **120**: 173-181. https://dx.doi.org/10.1016/j.atmosenv.2015.08.050

Irga, P. J., and F. R. Torpy (2016). A survey of the aeromycota of Sydney and its correspondence with environmental conditions: grass as a component of urban forestry could be a major determinant. Aerobiologia. **32**: 171-186. https://dx.doi.org/10.1007/s10453-015-9388-0

Irga, P. J., M. D. Burchett, G. O'Reilly and F. R. Torpy (2015). Assessing the contribution of fallen autumn leaves to airborne fungi in an urban environment. Urban Ecosystems **19**(2): 885–898. https://dx.doi.org/10.1007/s11252-015-0514-0

Irga, P. J., B. Armstrong, W. L King, M. D. Burchett and F. R. Torpy (2016) Correspondence between urban bird roosts and the presence of aerosolised fungal pathogens. Article not yet assigned to an issue. pp1-11 Mycopathologia. https://dx.doi.org/10.1007/s11046-016-0013-8

Irga, P. J., and F. R. Torpy (2016). Indoor air pollutants in occupational buildings in a sub-tropical climate: Comparison among ventilation types. Building and Environment **98**:190-199. https://dx.doi.org/10.1016/j.buildenv.2016.01.012

Other peer reviewed publications

The publications presented here were published during the candidature but do not form part of this thesis

Torpy, F. R., **Irga, P. J** and Burchett M. D (2015): Reducing Indoor Air Pollutants Through Biotechnology. In Biotechnologies and Biomimetics for Civil Engineering (Pacheco Torgal F, Labrincha JA, Diamanti MV, Yu CP & Lee HK eds.). Springer International Publishing, pp. 181-210.

Torpy, F. R., **Irga, P. J** and Burchett M. D (2014): Profiling indoor plants for the amelioration of high CO_2 concentrations. Urban Forestry & Urban Greening. **13**, 227-233.

Irga, P. J, Braun JT, Douglas ANJ, Pettit T, Fujiwara S, Burchett MD and Torpy FR (2016) The distribution of green walls and green roofs throughout Australia: Do policy instruments influence the frequency of projects. Landscape and Urban Planning. *In Review.*

List of conference presentations

The presentations presented here resulted from this work, however have not otherwise been included in this thesis

Irga, P. J., and F. R. Torpy (2016). A survey of aeromycota for urban Sydney and their relationships with environmental parameters. In *Proceedings of the* Australasian Mycological Society & Fungal Network of New Zealand Joint Meeting Conference. Queenstown, New Zealand.

Irga, P. J., M. D. Burchett and F. R. Torpy (2014) Ecological determinants of the aeromycota in urban Sydney. In *Proceedings of the* 10th International Congress of Aerobiology. Campbelltown, Australia

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List of Abbreviations

Analysis of Similarity	ANOSIM
Analysis of Variance	ANOVA
Antigen-presenting cells	APCs
Canonical correspondence Analysis	CCA
Carbon dioxide	CO_2
Carbon monoxide	CO
Central Business District	CBD
Colony Forming Units	CFU
Constant air volume	CAV
Mixed model ventilation systems methods	CVS
General Linear Model	GLM
Heating, Ventilating and Air Conditioning systems	HVAC
Indoor/outdoor	I/O
Local Government Area	LGA
Major histocompatibility complex II	MHC
Natural ventilation	NV
New South Wales Environmental Protection Agency	NSW EPA
Centralised mechanical ventilation systems	MVS
New South Wales Office of Environment and Heritage	OEH
Nitric oxide	NO
Nitrogen dioxide	NO_2
Oxides of nitrogen	NOx
Non-metric multidimensional scaling	nMDS
Nota Bene	NB
Oxides of sulfur	SOx
Particulate matter	PM
Particulates less than 10 micrometres in size	PM_{10}
Particulates less than 2.5 micrometres	$PM_{2.5}$
Pathogen-associated molecular patterns	PAMPs
Polycyclic aromatic hydrocarbons	PAH
Repeated Measures General Linear Model Analysis of Variance	RM GLM ANOVA
Reuter Centrifugal air sampler	RCS

Sabouraud Dextrose Agar	SDX
Sick Building Syndrome	SBS
Standard Error of the Mean	$\pm$ SE or $\pm$ SEM
Statistical Package for the Social Sciences	SPSS
Sulfur dioxide	SO_2
Tapered Element Oscillating Microbalance	TEOM
The National Environment Protection Ambient Air quality Measure	Air - NEPM
Toll-like receptors	TLR
Total Suspended Particulate Matter	TSP
United States of America	USA
Urban Air Pollution	UAP
Urban Forest Effects Model	UFORE
Volatile Organic Compounds	VOCs
World Health Organization	WHO

Chapter 1 INTRODUCTION

1.1 Scope and Gaps in knowledge

The general aim of this thesis was to investigate air pollutant concentrations across urban Sydney, Australia and to investigate the relationship urban forests have with the distribution, composition, and concentrations of those air pollutants. The investigations presented in this thesis include a series of descriptive experiments across inner Sydney along with manipulative laboratory experiments, detailing not only the air quality of the region, but also describing the air pollutant mitigation properties of urban forests as well as its potential to directly and indirectly create air pollutants. Whilst each study focused on specific questions relating to the type and spatiotemporal patterns of air pollutants, potential relationships with the type of urban forests, biometeorological, urban forests detritus, and avian relationships, and indoor concentrations; integrated as a broader thesis, the combined results provide a novel framework for understanding the important role that urban forests has in influencing the concentrations and composition of air pollutants within urban Sydney, Australia.

Only three studies on the diversity of airborne fungi have ever been conducted in Sydney, and they either used dated techniques, or lacked spatial or temporal replication. Thus, the current work addresses a previous absence of understanding of the ecology and phenology of the aeromycota of inner Sydney. Similarly, the association between the phenology of airborne fungal behaviour had not previously been assessed with respect to the sources or substrates of the fungi. The contribution of fungi from the phylloplane of senescent leaves to the viable propagules that are present in the air has not previously been assessed. The decay of senesced leaves may be of particular significance in an inner urban or city centre environment, where deciduous street trees could be the only substantial source of biological detritus in the area, and thus the primary focus of fungal growth and dissemination. Deciduous trees are often preferred in inner urban environments; if these trees support a constrained diversity of phylloplane fungi, it is possible that the low biodiversity of street trees in Sydney may potentially yield a limited diversity of aerosolized fungi in high individual numbers. If these species are potential pathogens or allergens, such a pattern could partially explain the increased incidence of allergy that occurs in Autumn.

Habitat fragmentation in urban environments concentrates bird populations into areas of dense urban forests or into highly developed environments to which they have become adapted. Consequently, the roosts of these birds potentially create environments conducive to fungal growth and dissemination. As birds are homotherms, and are known to harbour a range of fungi that are pathogenic to humans, these roosts could act as foci of infection. Similar to the senescent leaves, airborne fungi derived from these environments have been poorly studied. Thus, the current research documenting the diversity of culturable airborne fungal propagules associated with urban bird roosts, whilst accounting for environmental variables and the substrate that the birds occupy fills a considerable gap in our understanding of urban ecology.

Lastly, information encompassing all aspects of indoor air pollutants relevant to public health for Sydney has not previously been documented, and no baseline indoor: outdoor ratios for physicochemical pollutants has been recorded for Occupational Health scientists to facilitate routine air quality investigations. This gap in knowledge is further complicated by the influence of natural, mechanical and mixed-type ventilation systems on indoor air quality. The current work provides a comprehensive temporal and spatial assessment of these variables, and should be of value in future air quality risk assessments.

1.2 Literature review

1.2.1 Air quality

Throughout history, it has been known that substances in the air can have adverse effects on human health (Kenworthy and Laube, 2002). Urban air pollution (UAP) is a world-wide health problem, causing approximately 5.5 million deaths globally each year (IHME 2015). In Australia it is estimated that UAP causes over 1,400 deaths per annum in Sydney alone (Department of Health, 2009), with national health costs estimated to be as high as 1 per cent of gross domestic product (Brindle et al 1999) or 12 billion Australian dollars a year, from lost productivity and medical expenses (Environment Australia, 2003). A considerable proportion of UAP come from fossil fuel emissions, which comprise a mixture of solid particulate matter (PM), and gases including oxides of sulfur (SO_x), oxides of nitrogen (NO_x), carbon monoxide (CO), carbon dioxide (CO_2), volatile organic compounds (VOCs), polycyclic aromatic hydrocarbons (PAHs), and ozone (Hedge, 2009). Additionally, some air pollutants are biological in nature and are referred to as bioaerosols, whist most other pollutants

1.2.3 Gaseous pollutants

1.2.3.1 Volatile Organic Compounds

Volatile organic compounds (VOCs) are defined as any gas or volatilised substance comprised of aliphatic and aromatic hydrocarbons, alcohols, aldehydes and chlorinated compounds with a vapour pressure greater than 2 mm Hg at 25 °C (Australian National Pollutant Inventory 2009). Indoors, these chemicals are emitted from furniture, carpets, construction materials, sprays, and cleaning products (Mitchell 2013). Outdoor concentrations are derived from industrial and transport-related activities (vehicle emissions, vehicle manufacturing, printing, equipment coating, electronics and furniture manufacturing).

A mixture of hundreds of VOCs can be found in indoor air. These compounds exhibit very large variations in concentration as well as physical, chemical, and biological properties (Guieysse et al 2008; Thevenet et al 2014). Differences in composition and concentrations occur due to variations in building materials, cleaning products, or type of ventilation used (Guieysse et al 2008), which interacts with outdoor VOC sources.

The concentrations may also show great temporal variations, as levels of VOCs emitted from coatings and furniture decrease over time (Zhongkai et al 2010).

1.2.3.2 Inorganic gases

The most prevalent urban inorganic gaseous compounds of concern are CO, CO_2, NO_2 and sulfur dioxide (SO_2). These compounds are mainly generated from various combustion processes such as the burning of fossil fuels, with most outdoor sources sourced from road transport, power stations, and refineries, whereas residential sources include kerosene heaters, gas-fired appliances, wood stoves and gas-fired hot water heaters, and tobacco smoking (Seow et al 2015; Soreanu et al 2014).

Not normally considered a toxic air contaminant, CO_2 can become as simple narcotic with increasing levels above the current outdoor ambient concentration. It has been associated with adverse symptoms related to the mucous membranes (dry eyes, sore throat, nose congestion, sneezing) and to the lower respiratory tract (tight chest, short breath, cough and wheezing) (Torpy et al 2015). This mainly occurs in closed-in spaces, indoors, where excess CO_2 is produced mainly by human respiration. (Erdmann and Apte 2004). At concentrations between 2,500 and 5,000 ppmv, CO_2 can cause headache, with serious health consequences arising at concentrations greater than 50,000 ppmv (Gurjar et al 2010).

The incomplete combustion of fossil fuels, can produce CO. Exposure to low concentrations of CO result in headache, drowsiness, and severe chest pains, while high concentrations leads to neurological damage and death (Ozgok-Kangal et al 2016). CO reduces the ability of the blood to carry oxygen throughout the body, as it converts haemoglobin to carboxyhaemoglobin. Ambient concentrations of CO are on the decline, with indoor concentrations considered a greater health concern (Naima et al 2012)

Concentrations of NO and NO_2 in ambient air has declined since the early 1990s, however high concentrations can still be found near busy roads. Ozone, another air pollutant of concern, is produced when NO_2 reacts with VOCs. High concentrations of NO_x cause inflammation of the airways and other respiratory effects (Naima et al 2012). Similarly, SO_2 is also a respiratory irritant, causing adverse respiratory symptoms (Powe and Willis 2004). However, most ambient SO_2 is produced through the combustion of sulfur containing fossil fuels, and thus its main source is industrial sources, which are less likely to be encountered in urban Sydney.

1.3 Particulate aerosols

Aerosol particulates include all atmospheric pollutants that are not gases (Anderson, 2009). These particulates can be of anthropogenic or environmental origins, and are composed of solid particles, suspended droplets, or a mixture of the two. The behaviour of the various forms of airborne particles is dependent on their size, chemical composition and morphology. The source of PM determines the size of particulates generated, with combustion leading to smaller particles, whilst biological sources form larger particles (Lazaridis 2011).

Airborne PM is comprised of a mixture of many different substances. Quantitatively, the major components of PM are materials of biological origin, mineral dust, sulphates, nitrates, carbonaceous material (black carbon), ammonium, nitrate and sea salt (Philip et al 2014). In urban areas, both fine and coarse particles mainly have an anthropogenic origin, largely through combustion processes, although natural sources are also important, influencing source contribution as well as temporal variability of concentrations. In an urban context, other than vehicle derived particulates, sources include wood burning, coal burning for power generation, mining, bushfires, dust storms, industrial activities such as brickworks, iron and steel making, quarrying, and smelters (NSW EPA 2009). During the cooler months, smoke from residential wood-heaters is common in Australia, and results in elevated pollution levels that may lead to health risks (Gupta et al 2007). Temperature inversions can prevent the removal of PM from the atmosphere of Sydney, thereby quickly degrading air quality (Gupta et al 2007).

Epidemiological studies mainly focus on particulate matter which is 10 μm in diameter and smaller (PM_{10}), as these particles are able to penetrate beyond the nasal passages and into the respiratory system, with the smaller the particle the further it can penetrate (Taeger 2013). Once there, these particles have the potential to cause adverse pulmonary and extra pulmonary health effects (Millar et al 2010), potentially resulting in acute exacerbations emphysema and chronic bronchitis and asthma (Guarnieri and Balmes 2014). Exposure to PM can also affect other parts of the body, specifically the cardiovascular system, with disease states including myocardial infarction, stroke, heart failure exacerbation, and arrhythmias (Brunekreef and Forsberg 2005). Additional evidence suggests that the $PM_{2.5}$ fraction (i.e. particles 2.5 μm in diameter and smaller)

are more strongly associated with these cardiovascular health effects. This health risk is higher in children and the elderly (Simoni et al 2015), and patients with pre-existing chronic cardiopulmonary diseases (Chen and Goldberg 2009).

1.3.1 Effects of meteorology and physical environment on particulate air pollution

Meteorological and topographical factors have substantial effects on the transport and deposition of air pollutants. The most important factors determining the dispersion of airborne pollutants are wind velocity, atmospheric turbulence or stability, temperature, and humidity (Seigneur and Dennis, 2011). Wind is the major determinant of the distribution and transportation of atmospheric air pollutants (Saliba et al 2010). The horizontal transportation of air pollutants is in the direction of the prevailing wind, while vertical transportation is determined by atmospheric turbulence (Fang et al 2007). Fluctuations in the wind play an important role in creating turbulence in the atmosphere and thus directly affect the level of dispersion, whilst precipitation removes many forms of pollutants from the atmosphere and deposits them on the ground, either as particle suspensions or solutions (Leppä et al 2011). Despite their relatively short lifetimes, aerosols originating from one region can travel long distances to affect the environment of downwind regions (Chin et al 2016). These trends notwithstanding, other processes like radioactive decay, chemical reactions and gravitational or boundary layer deposition of air pollutants also play important roles in the spatiotemporal dynamics of dispersion patterns (Leelőssy et al 2014).

Deposition processes, including precipitation, scavenging, and sedimentation, cause downward movement of pollutants in the atmosphere, dropping them out of suspension (Samson 1988). In terrestrial environments, vegetation is a major determinant of the deposition of air pollutants (Janhäll 2015), as it disrupts the natural flow of air through its complex geometry and associated boundary layer, and thus the transport of pollutants, encouraging deposition (Carvalho et al 2007). Plant geometry is comprised of an array of leaves, branches, and other structures, both living and dead, forming a porous barrier to air flow (Shaw 2007). Despite the visual impression of high density, the actual volume occupied by plant material makes up a relatively small fraction of the total canopy volume, usually less than 5% (Phattaralerphong et al 2006). Nonetheless, the

primary sink for the momentum of air and thus deposition, is the canopy itself, rather than the underlying ground surface (Janhäll 2015).

1.4 Measurement and Monitoring of particles

The National Ambient Air Quality Status and Trend report (OEH 2004), notes that New South Wales has only 15 $PM_{2.5}$ monitoring stations located across the state, which several ground-based air pollutant-monitoring sites are located in Sydney and its surrounding areas. These monitoring sites are operated by the NSW EPA and Office of Environment and Heritage (OEH). Each station measures PM_{10}, $PM_{2.5}$, CO, NO_2 and SO_2. The use of fixed air quality monitoring stations, can be insufficient for providing air quality information in all areas, and cannot provide adequate data to allow the spatial interpolation of air pollutant patterns over broad areas (Oaimo et al 2015). Geostatistical techniques, like kriging, can be used to enhance the interpolation of the intra-urban gradients in air pollutant load observed between these monitoring sites (Vicedo-Cabrera et al 2013). However, the accuracy of interpolation methods are restricted by the number of monitoring sites available (Oaimo et al 2015), and do not effectively represent small scale spatial variations associated with local land use patterns such as proximity to roadways and urban forests (Jerrett et al 2005).

The monitoring sites closest to central Sydney include Randwick (1 km from the perimeter of the city's Central Business District [CBD]); Rozelle (3.5 km from the perimeter of the city's CBD) and Earlwood (10 km from the CBD). The OEH air quality monitoring sites utilise a tapered element oscillating microbalance (TEOM) for particulate matter quantification as per the Australian Standard (AS 3580.9.8 - 2001), approved by the New South Wales Environmental Protection Agency (NSW EPA 2007).

Air quality in Sydney has received limited study. Gupta et al (2006) used satellite remote sensing to analyse PM mass concentrations over Sydney, and compared this data to empirical data from six ground-based air quality stations in the area. The authors noted significant diurnal variations and an increase in $PM_{2.5}$ during the spring and summer seasons, due to bushfires. They concluded that air quality throughout the year in Sydney is very good relative to other major cities in industrialised countries, except during major bushfires. However, they did note that, although Sydney levels of $PM_{2.5}$ are below both European and United States air quality limits, air quality in these locations has been improving in recent decades, while $PM_{2.5}$ levels in Sydney have remained relatively

static. Despite low ambient pollution levels in Sydney, the existing levels of air pollutants have been estimated to lead to 2% of total deaths per year (Broome et al 2015). The ambient indoor air quality across Sydney has not been well studied, and the contribution of outdoor air pollutants penetrating into indoor environments is inadequately described for this region. Further, research involving the use of satellite data to approximate PM concentrations is still subject to inaccuracies , as it relies on the accurate correlation between satellite-derived aerosol optical depth data and ground-level quantitative $PM_{2.5}$ concentrations (Gupta et al 2006). To date, most current studies have focused on developing empirical models based on ground-level $PM_{2.5}$ concentrations and satellite imagery data (Gupta et al 2006). These models are yet to be refined for many spatial regions, as the addition of ancillary information such as vertical distribution of aerosols and meteorological information from LIDAR, sun photometers, and size-resolved chemistry measurements is needed to further refine the analysis (Gupta et al 2006). However, advancement in satellite remote-sensing techniques facilitates air quality information over large spatial scales, when compared with surface instruments that only allow point observations (Gupta et al 2006).

1.5 Urban forests and its ability to mitigate air pollutants

'Urban greening' has been proposed as a means to reduce airborne pollutant levels (Chen and Jim 2008), with mounting evidence indicating that urban forestry can offer a range of benefits for urban residents that includes the mitigation of air pollution (Brack 2002; Roy et al 2012; Zheng et al 2013). Most of the related studies focus on the ability of urban forests to reduce airborne PM and NO_2 (Vos et al 2013). The capacity of urban forests, specifically trees, to reduce air pollutants is through a number of mechanisms. Particles in the airstream are most readily impacted onto moist, rough, or electrically charged surfaces. Trees can provide these surfaces intercepting and accumulating atmospheric particles. By providing a large waxy surface on which deposition can occur, and through leaf pubescence, the particles are impacted and prevented from resuspension (Beckett et al 2000). Vegetation is also able to sequester air pollutants through open stomata and either store it or process it through a series of chemical reactions (Janhäll 2015). Further, various tree configurations can alter wind profiles or create wind inversions via their geometry which assist in the deposition rate of pollutants from the air, or may act as physical barriers preventing the penetration of pollutants into specific areas (Salmond et al 2013).

Many cities have plans for increasing their urban greenspace to reduce air pollution (Andersson-Sköld et al 2015). The City of Sydney for example, has proposed to increase the city's urban canopy by 50 per cent from the current 15.5 per cent by 2030 (City of Sydney 2013). Such initiatives have been theorised to be both economically and environmentally effective, with urban forestry in Canberra, Australia estimated to have a combined energy reduction, pollution mitigation and carbon sequestration value of US$20–67 million between 2008 and 2012 (Brack 2002). While equivalent estimates for other Australian cities are unavailable, it is likely that the urban forest will have similar if not greater value for cities with higher population densities, such as Sydney.

Sydney's urban forest has not previously been investigated with respect to its ability to reduce urban air pollution. Despite the numerous known benefits urban forests has on ameliorating and mitigating air pollutants, there is a scarcity of quantitative data available for its ability to do so, with many regions' vegetation-atmosphere interactions relatively unstudied. Further, few studies from any location are available that provide experimental data on the air pollutant removal capacity of urban forestry/greenspace, as opposed to model estimates such as the Urban Forest Effects Model (UFORE) by Nowak (2006), potentially related to the challenges related to assements on an urban scale, compared to a city scale. Similarly, few reports (Tallis et al 2011, Setälä et al 2013) provide empirical evidence of the association between overall ambient particulate matter densities and urban forestry densities (Pataki et al 2011). To date, efforts to demonstrate or validate the model estimates have either found substantially reduced improvements relative to estimates from models (Tallis et al 2011) or have shown minimal positive effects (Setälä et al 2013).

1.6 Indoor air quality

Australians are known to spend up to 90 per cent of their time indoors (Molloy et al 2012), and thus indoor air quality has a major impact on the health of most Australians (Newton 2001). The accumulation of and continued exposure to indoor air pollution may result in a condition known as Sick Building Syndrome (SBS) (Hedge 2009). The term 'SBS' is used to describe situations in which building occupants experience acute or subacute health and discomfort effects that appear to be linked to the duration of time spent in a building (Engvall et al 2005; Wang et al 2008). Typical SBS symptoms include headache; eye, nose and throat irritation; dry cough; dry or itchy skin; dizziness and nausea; difficulty concentrating and fatigue. The direct cause of the symptoms is not

completely known, but symptoms are rapidly relieved after leaving the building, implicating poor indoor air quality (Fisk et al 2009; Simoni et al 2010).

1.6.1 Workplace legislation in regards to air pollutants

In Australia, ambient air pollution standards are described in the *National Environment Protection (Ambient Air Quality) Measure* (Air-NEPM 2011). These standards are similar to those from most western countries. The Air-NEPM standards cover most of the common air pollutants, including PM_{10}, $PM_{2.5}$, SO_2, NO_2, and CO. Australian threshold limits for indoor workspace CO_2 and volatile organic compounds (VOCs) are set for occupational scenarios only, by Work Safe Australia (2012).

1.7 Indoor/outdoor ratios

The rate and extent of outdoor air pollution penetration into indoor spaces can depend on many parameters, including air exchange rate, building envelope porosity, building ventilation rate, deposition rate on surfaces, particulate coagulation and chemical reactions. Calculation of the indoor/outdoor (I/O) ratio can quantify how much of the outdoor air pollutant load is infiltrating into the indoors. I/O ratios less than 1 indicate that the indoor air is better than outdoors, I/O ratios closer to 1 indicate that outdoor air pollutant concentrations are potentially infiltrating in the building, I/O ratios greater than 1 indicate and accumulation effect or that indoor sources of a pollutant contribute more to the indoor air concentrations than outdoor sources.

The increase in air-tightness of modern buildings to gain greater control over the regulation and conditioning of indoor air, have resulted in decreases in the infiltration of large, filterable particulates, thus decreasing the I/O ratio for these pollutants. However, this is not the case for many of the gaseous pollutants, which do not get filtered from the ventilated air. As such, these pollutants indoors are heavily influenced by outdoor sources, and tend to have I/O ratios approaching 1. In office workplaces, the major indoor sources of these pollutants have been regulated out (i.e. tobacco smoke, woodstoves and fireplaces, gas appliances and kerosene heaters), however if they are generated from proximal residential dwellings or have an outdoor origin they may infiltrate indoors. NO_x and fine particulate matter may infiltrate into indoor environments as they are unable to be filtered out of the incoming air, even in buildings with efficient mechanical air-conditioning systems (Kilbert 2012). Volatile organic compounds on the other hand, are released from a wide variety of indoor building

materials, including carpets, furniture, power cables, and products such as paint, cleaners, pesticides, deodorisers, and air fresheners (Cheng et al 2016).

1.7.1 Ventilation in the built environment

The concentration and composition of indoor air pollutants is the result of a number of factors, with the presence of a pollutant source and building ventilation system type and rates likely to be the predominant factors. The relationship between these factors can be complicated, as the pollutant component continually changes over time, as do the ventilation system effects due to thermal conditioning requirements that vary throughout the day and seasonally. The rate at which the volume of air in a building is replaced is called the air exchange rate (McDonald-Gibson et al 2013). Low air exchange rates can lead an accumulation of air pollutants within indoor environments, while higher air exchange rates result in lower air pollutant levels, assuming that the outdoor air is relatively clean. Appropriate use of ventilation and air exchange rates are often used to reduce the level of indoor air pollution, although other factors such as building design, material characteristics, as well as building operational factors also influence indoor air quality (Kilbert 2012). Ventilation system type, therefore, can be a major determinant of the composition and concentration of indoor air pollutants, and can provide a valuable explanatory component when assessing indoor air pollutants.

Three different ventilation system strategies are normally utilised in commercial and other non-residential buildings. The first is *Natural ventilation*, where windows, doors, skylights or roof ventilators are simply left open to the atmosphere. This method supplies an ample amount of air for buildings if there is sufficient open area and flow-through, although this may not be the case for many modern buildings. Air from natural ventilation is not conditioned, so it will permit the entry of all outside air contaminants, as well as permitting the diffusion of indoor-sourced pollutants to atmosphere. In Australia, this ventilation type is found in a restricted number of smaller commercial buildings, as well as in many schools, kindergartens and many residential buildings. The second is *Mechanical ventilation*, where indoor air is supplied, conditioned (humidity modified) and thermally regulated by a Heating, Ventilation and Air Conditioning system (HVAC). Most of these buildings have no operable windows, with all fresh air passing through the mechanical system. Central HVAC systems are generally composed of an intake for outdoor air located on the roof or side of the building, a duct bringing outdoor air to an air treatment unit and vents in room ceilings and / or floors for air

circulation The air treatment unit filters, heats or cools, and sometimes dehumidifies or humidifies the air and distributes it through a duct network to air vents throughout the occupied spaces of the building. In Australia, most buildings run at constant air volume (CAV), with 20% of the total volume of inlet air sourced from outdoors and 80% recirculated. The third system, *Mixed model ventilation,* is common in the Australian climate, combining natural and mechanical ventilation systems, usually through the use of wall mounted air conditioners in high use or sensitive spaces, but with a substantial and highly variable natural ventilation component through opening windows and doors. Due to the variability inherent in these usually retrofitted systems, no generalisations on their performance can be made.

1.8 Bioaerosols

Bioaerosols are a mixture of aerosolized particles that are either living or are from a living origin. When inhaled, bioaerosols may have serious health implications (Tham et al 2014), leading to clinical conditions ranging from temporary allergic reaction, to systemic disease (Crook and Burton 2010; Pfaller et al 2006). These particles include fungal cells and spores, intact bacterial cells and spores, protozoa and their cysts, and plant pollen grains (Douwes et al 2008; Macher et al 2012). Bioaerosols may also include cell fragments, and also particles of decayed plant and animal matter; wood and grain dusts; the droppings and dried body parts of arthropods; and particles from the detritus of larger animals, including saliva, faeces, and urine (Sturm, 2012). Other biological agents may also be present, including viral material, toxins, microbial excreta and metabolites (Dart and Thornburg, 2008). These non-living agents are considered bioaerosols due to their origin and their ability to elicit an immune response from the antigens present (Crameri et al 2014). Of the bioaerosols, fungal spores are of the most concern as they constitute the majority of the total bioaerosol load in the atmosphere and their potential to not only cause allergic reaction but can also cause infection (Green et al 2005).

1.8.1 Fungal bioaerosols

The presence of fungal bioaerosols in the atmosphere stems from the primary mechanism of fungal reproduction. Fungi have the ability to reproduce asexually, and in some cases, also sexually (Dix and Webster 1995); with the asexual forms representing quantitatively the most significant method for survival and dissemination (Staab and Wong 2009). Asexual propagules arise following mitosis of a parent nucleus, and are

termed either spores or conidia depending on their mode of production (Walker and White 2005). Conidia arise either by budding from conidiogenous somatic hyphae, or by the differentiation of preformed hyphae. Asexual spores are commonly formed within an enclosure called a sporangium, with large sporangia containing up to 100,000 spores (Biggane and Gormally 1994). Consequently, asexual fungal propagules constitute a major component of bioaerosols.

Fungal propagule aerosolisation can arise from both active and passive processes (Dix and Webster 1995). Active propagule dispersal is a result of a force or energy created by a fungus in order for the propagules to be aerosolized. This can be in the form of ballistospore discharge, spherical launching through sudden changes in cell shapes or the bursting of turgid cells full of spores (Recio et al 2012). Passive spore aerosolization involves propagules becoming disarticulated from conidiogenous structures in airstreams, or simply lifted into the air by currents. Desiccated but viable yeast cells are also sometimes distributed by this method.

Of the fungal phyla, bioaerosol production is limited to the terrestrial Ascomycota, Basidiomycota and Zygomycota. Although most fungal bioaerosols are composed of mould spores or fragmented hyphae, yeasts are also known to be capable of aerosolisation, and can sometimes constitute a major component of total bioaerosols on occasion, and cause serious diseases such as cryptococcosis (Reiss et al 2011). Most opportunistic human pathogenic fungi are environmental organisms from all three of the aforementioned phyla, whose primary route of infection is via the inhalation of airborne conidia. Common pathogenic species include *Aspergillus fumigatus*, *Histoplasma capsulatum*, *Paracoccidioides brasiliensis*, *Penicillium marneffei* and *Coccidioides immitis*. However, not all human fungal pathogens are of environmental origin; the majority of infections caused by fungi are caused by organisms that are normally associated with human hosts (Whittington et al 2014). These include *Candida*, *Malassezia* and *Pneumocystis* species and dermatophytes including *Microsporum* and *Trichophyton* (Bignell 2014).

1.8.2 Ascomycota

Asexual reproduction is the most prevalent method of reproduction for the Ascomycetes with the formation of conidia from specialised aerial hyphae (Moore and Novak-Frazer 2002). These organisms are saprophytic and parasitic, and constitute most of the species

found in air samples (Gurjar et al 2010). Ascomycota can prevail in water damaged building materials, and thus can be highly prevalent in indoor environments. Aerosolized ascomycotan conidia are typically dominated by the genera; *Aspergillus*, *Cladosporium*, *Alternaria*, *Stachybotrys*, and *Verticllium* (Dix and Webster 1995; Smith and Kagan 2009).

1.8.3 Zygomycota

The Zygomycota are characterised by having thick-walled sexual spores called zygospores and asexual spores known as sporangiospores, which develop within a thin-walled structure known as a zygosporangium. The Zygomycota tend to be found in the soil of natural habitats (Heitman, 2011), particularly in areas with high relative humidity (Ellis et al 2007). Asexual spores are dispersed by wind; from ruptured zygosporangia at tips of aerial hypha. Aerosolized zygomycotan spores may include *Rhizopus, Mucor,* and *Absidia* (Chao et al 2002; Green et al 2003).

1.8.4 Basidiomycota

Basidiomycota are unique amongst the fungi, as they mostly reproduce sexually at the tips of hyphae, through the formation of a basidium, whilst a small number of Basidiomycota reproduce exclusively asexually through conidia (Shepherd and Totterdell 1988). As with the Ascomycota, these species can thrive in water damaged building materials, however most are plant saprophytes. Common aerosolized Basidiomycota conidia include; *Wallemia* (Shelton et al 2002) and *Schizophyllum* (Mentese et al 2009). The pathogenic yeast *Cryptococcus neoformans* is also classified in this phylum, and is most commonly distributed in air as desiccated yeast cells, although conidia can also occur under certain circumstances.

1.8.5 The health effects and immunological response to aerosolized fungal spores

Indoor elevated airborne fungal spore concentrations are of great concern, having been linked to health issues such as sick building syndrome (Meyer et al 2004; Takeda et al 2009) and fungal disease (Hunter et al 1988; Staab and Wong 2009). Adverse allergic effects caused by fungi result from the production of allergens, irritants and mycotoxins, eliciting symptoms of headache, coughing and dermatitis (Mendell et al 2011). Most fungal pathogens that infiltrate our bodies cause no problems for healthy individuals because there is adequate pre-existing immunity. However, serious fungal infections

may result from exposure to certain opportunistic fungal species in severely immune-compromised individuals (Maschmeyer et al 2007).

The human immune strategies to prevent fungal infections include a general innate immunity and acquired or adaptive immunity. The majority of fungi within the human body are recognized and inactivated rapidly by cellular and non-cellular innate defence mechanisms (Gardy et al 2009). Commensal organisms also control the growth of certain potential pathogens, primarily by competitive exclusion (Heitman 2011). The first aspect of the innate immune system to prevent fungal infection is in the form of physical barriers, such as skin and mucosa, which prevent the intrusion of pathogens. If these physical barriers fail to prevent the fungal intrusion, the cellular innate immune system is initiated, with phagocytic cells (tissue macrophages, monocytes and dendritic cells) and the recruitment and activation of neutrophils to the site of infection (Figdor et al 2002, Heitman et al 2006) to facilitate the killing or containment of the fungal spore or yeast (Gardy et al 2009, Yan and Hansson 2007). The secretion of chemokines and cytokines by macrophages can also produce an effective inflammatory response (Blanco and Garcia 2008). However, inappropriate inflammatory responses (allergic reaction) can often be implicated, which do not offer protection against the offending organism (Staab and Wong 2009). This happens frequently with fungal exposure (Ospelt and Gay 2010).

On failure of the innate immune response to clear fungal infection, the adaptive immune response is activated (Palm and Medzhitov 2009), where antigen-presenting cells (APCs), macrophages or dendritic cells containing fungal antigen are carried in the lymph to lymphoid organs (Wuthrich et al 2012). In the lymph, the fungal antigen binds to B-cells and the APCs present antigen peptides on their membrane to the major histocompatibility complex II (MHC) to activate antigen-specific T-cells (Kindt et al 2007). These T cells are biased to secrete specific cytokines that can either produce a protective inflammatory activation of fungistatic and fungicidal processes by phagocytes that results in clearance of infection, or an uncontrolled allergic response and hypersensitivity responses (Ismail and McGinnis 2008; Staab and Wong 2009). Antibodies are also involved in immunity to fungi, with most people having naturally occurring antibodies to *Candida albicans*, *Cryptococcus neoformans*, *Histoplasma capsulatum*, and *Pneumocystis jirovecii*, however the protective role of these antibodies is not so well defined as cell-mediated adaptive immunity (Verma et al 2014).

1.8.6 Immunocompromised opportunistic infections.

Individuals with deficiencies in immune function may be more susceptible to fungal diseases (Reponen and Green 2012). Inhaled fungal propagules in immunocompromised hosts remain largely unchallenged by the immune defences and tissue-invasive diseases can predominate. Such infections by ubiquitous fungal organisms are termed opportunistic infections. As opportunistic infections mostly occur through inhalation, knowledge of the type of fungi in the air, and people's exposure to them, is paramount. When opportunistic infections present themselves, they can signal that the immune system of the patient is compromised (Maschmeyer et al 2007).

1.8.7 Factors that influence fungal growth and reproductive propagule release

Fungi exhibit a remarkable ability to use almost any carbon source for growth (Aringoli et al 2008). In addition to the availability of a suitable nutrient source, other factors that influence growth and spore release are moisture, temperature, pH, and oxygen availability (Fischer and Kues 2006). Climatic conditions such as temperature, sunshine, humidity, rain, snow, and wind speed may therefore be important in the distribution and deposition of outdoor airborne fungal propagules (Jones and Harrison 2004). Outdoors, it has been found that the active discharge of propagules of Ascomycota and Basidiomycota is abundant when conditions are damp, whereas the passively launched dry propagules of such genera as *Cladosporium* and *Alternaria* are abundant when the weather is dry (Morris et al 2008). However these are broad generalisations and individual species within all three phyla behave differently.

1.8.8 Fungal bioaerosols found indoors

The majority of indoor airborne fungal species are derived from outdoor sources (Hargreaves et al 2003; Torpy et al 2013), in particular from regional vegetation, which is known to strongly affect proximal airborne fungal concentrations (Morey et al 2001). However, under suitable conditions (relative humidity, temperature, and building air exchange rate) fungi may also thrive on indoor man-made constructions (Chao et al 2002). As previously mentioned (Section 1.6), due to our prolonged exposure to indoor contaminants, the indoor airborne fungal spore load and community assemblage may be more important to health than those found outside for many people.

1.8.9 Workplace exposure limits of fungal bioaerosols

There is no Australian law dealing with workplace fungal exposure, and no Australian recommendations regarding fungal concentrations present in buildings. Occupational hygienists and air quality professionals commonly adopt international guidelines from the American Conference of Industrial Hygienists (*Air Sampling Instruments for Evaluation of Atmospheric Contaminants*, 1995) and the World Health Organization (WHO, 1988). These international recommendations show large inconsistencies because of the manner in which fungal contamination can occur, as fungi possess diverse physical properties and the factors that lead to high concentrations are multifaceted. Furthermore, the recommendations may not be applicable for Australian conditions including climate, latitude and the variant ecology displayed by indigenous fungi. Unfortunately, there is a tendency to attribute certainty to such values without a full appreciation of the complexity of the factors involved, particularly in the case where multiple determinants interact (Spickett et al 2013).

An additional cause for inconsistencies in the international recommendations is the lack of uniformity in the way the regulations are presented. For example, some are based on the ratio between indoors and outdoors (Gaskin et al 2012), while others indicate that indoor levels of over a certain quantity should be investigated *per se* (Horner et al 2008). Some address total numbers (e.g. WHO 1990), while others address high numbers of specific species only (Stennett and Beggs, 2004; Pearce et al 2009). The focus on individual species may be very important for pathogenic species, especially when associated with the presence of immuno-compromised individuals (Maschmeyer et al 2007).

Further, there are differences between the sampling methods used in the various regulations. This is especially important for occupational hygienists; as large variation in results are likely, depending on the sampling method used. Some air samplers only measure total fungal spores (or biochemical surrogates such as ergosterol, mycotoxin, beta-glucans), while others measure total culturable fungi, data from which is valuable for speciation, especially for pathogenic species, however these methods underestimate the total number of spores present as they may not be able to be cultured (Lee et al 2006). Thus there is a need for research to address the multiple factors that contribute or influence fungal assemblage in urban and suburban areas of Sydney Australia, with the

goal of contributing to the background literature that one day may assist in the creations of Australian recommendations for fungal contaminants within buildings.

1.8.10 Studies on the aeromycota conducted in Australia

Previous studies have determined that the density and species composition of airborne fungi vary substantially amongst different parts of the world. It is thought that such variations occur because of local environmental variables, growth substrates, and human activities such as agriculture and the structure of urban development (Gots et al 2003). These contributing variables can differ greatly from country to country and from urban to non-urban areas. Thus, the value of each survey is specific to the area from which samples were collected, and therefore only broad generalisations can be made among studies.

No predictive modelling has been undertaken on Australian airborne fungal density, although limited studies have assessed airborne fungal diversity in most of Australia's major cities; with Brisbane (Rutherford et al 1997; Hargreaves et al 2003), Melbourne (Dharmage et al 1999; Mitakakis and Guest 2001), Adelaide (Gaskin et al 2012) and Perth (Kemp et al 2003) studied as well as rural studies in Northern NSW (Green et al 2006), and the La Trobe valley (Garrett et al 1997).

The absence of a broad ecological assessment of the aeromycota of the Sydney region is a gap in our knowledge that may have significant human health consequences. Only three studies on airborne fungal spores have been conducted in Sydney. The first study (Frey and Durie 1960), addressed the species composition of airborne moulds in Sydney. The study only sampled outdoor areas, and the sampling method was simplistic and out-dated; as the method involved leaving agar plates out in open areas – lighter fungal spores do not penetrate the agar's boundary layer are thus not detected by this method (Dix and Webster 1995).

The second study sampled total conidia and fungal fragments over a 21-day period during Spring (Green et al 2005). However, this study's focus was on the immunostaining of fungal allergens rather than identification of fungal species present. Thus the study did not address species diversity seasonality, or make comparisons between the indoor and outdoor aeromycota.

Torpy et al (2013) assessed the effect of indoor plants on indoor aeromycota levels and composition. Indoor samples from the offices of two Sydney buildings were compared to offices in the same buildings with plants present. Outdoor air samples were taken, but only in order to act as reference samples. This study's conclusions were limited to the specific locations of the buildings (southern edge of the CBD), rather than establishing a general assessment of the density and community structure of the aeromycota in Sydney.

There is little data available for Sydney, with respect to relationships between indoor and outdoor spore concentrations, which as mentioned above, may have elevated levels of pathogenic species, and have been well studied overseas (section 1.8.13). The current work represents the first research conducted in Sydney addressing the density and diversity of airborne fungal propagules found indoors.

1.8.11 Fungal modelling

To date, most studies on exposure to fungal spores have focused on the relationship between indoor and outdoor concentrations and species assemblages (Horner et al., 2008). The emphasis of these studies has been on the identification of independent variables that potentially influence the quantitative dynamics of specific fungal species; in particular phenology (Stennett and Briggs 2004), proximity to vegetation (Dharmage et al 1999), climate (Kemp et al 2003), building design (Parat et al 1997), and method of ventilation (Kemp et al 2003). Alternatively, several studies have attempted to investigate the possible point sources for pathogenic fungi, for example potted-plants (Lass-Flörl et al 2000; Hedayati et al 2004; Engelhart et al 2009). There have been few previous studies that combine all these factors, and few studies have developed statistical models for the prediction of airborne fungal spore densities. Sabariego et al. (2000) used rank correlation to investigate the effects of temperature, sunlight, relative humidity, rainfall and wind speed on atmospheric concentrations of *Alternaria*, *Cladosporium* and *Ustilago* in Granada (southern Spain). They found that the presence of *Alternaria* and *Cladosporium* were significantly correlated with temperature and hours of sunlight, while *Ustilago* was significantly correlated with increasing relative humidity and a decreasing wind speed. Chao et al (2002) used multivariate analysis to examine the characteristics of airborne fungal populations and correlations with a restricted set of environmental variables in office environments in Boston, Massachusetts. Total airborne fungal concentrations varied significantly between seasons (highest in summer, lowest in winter) and were positively correlated with

relative humidity and negatively related with CO_2 concentrations. Green et al (2003) developed a prediction model for indoor concentrations of airborne mould spores in the Greater Cincinnati area, by fitting a multiple linear regression to variables that were thought to have had a major effect on spore loads. Independent variables affecting indoor bioaerosol concentrations included indoor relative humidity, indoor temperature, outdoor airborne mould density, season, water damage, visible mould, damaged materials, building age, health of occupants, number of occupants, and indoor pets. The authors claimed that their model accurately predicted spore loads 97% of the time. Recio et al (2011) conducted a similar study, however narrowed their experimental question to only *Cladosporium* and *Alternaria* in the outdoor atmosphere of Malaga, Spain. The study used stepwise-multiple regressions, with independent variables similar to those of Green et al (2003). They determined that the variables that best explained the daily and weekly variations in concentration of *Alternaria* and *Cladosporium* were temperature.

Evident from these studies is that predictive models may be relevant exclusively to the regions in which they were developed due to the geographical independence of many environmental variables. Clearly, there is a need to evaluate predictors of fungal concentrations in indoor environments specific to regional geographical locations (Green et al 2003). It also appears that the value of models may also be constrained to the seasonal conditions within which they were developed, and thus across-season sampling is necessary to incorporate phenological variability (Chao et al 2002). Previous work indicates that there is a high probability that there will be trends and relationships with meteorological factors, but the direction and magnitude of these effects is unpredictable.

Prior to the work presented here, it was generally unknown whether there were relationships between the aeromycota and other phenomena that influence air quality. This work aimed to provide knowledge on the correspondence between airborne fungi and other components of air pollution, with the goal of establishing important background knowledge for predicting air quality warning levels.

There has been considerable research into the factors that affect urban air quality, some of which has focussed on airborne fungi. However, there is no consensus on the combined effects of urban characteristics on the aeromycota. Whilst previous work has examined a limited number of explanatory variables, or compared a small number of

buildings or sites, none has had sufficient sampling scope to allow reliable generalisations to be made.

1.8.12 Urban forests and the aeromycota

As hypothesised by Mitakakis et al (2001) and Hargreaves et al (2003), elevated aerosolised fungal propagules in urban areas could be a result of proximal vegetation. There is, however, insufficient empirical evidence to determine the strength of this relationship, and the relationship between urban vegetation and fungal diversity has not previously been considered. Furthermore, the type or types of greenspace that leads to elevated concentrations has never investigated, leading to questions about the relative contribution of grass, tree and shrub canopy coverage, or whether airborne fungi are predominantly derived from leaf litter and detritus. It is plausible that saprobic colonization and sporulation of grass endophytic fungi could become unrestrained when their host plant tissue dies (Vázquez de Aldana et al 2013). Additionally, turf grass in urban areas is regularly mown: potentially mechanically aerosolizing endophytic and epiphytic fungi (Streifel 1988) and exposing damaged plant tissues and leaking protoplasm which may encourage fungal growth. Comtois et al (1995) demonstrated that the act of mowing lawns can aerosolize fungal spores, increasing the exposure of lawn mowing workers to aeroallergens. Similarly, previous work by Mitakakis et al (2001) showed that the harvesting of crops increase aeromycota concentrations in nearby towns (Mitakakis et al 2001). Prior to the current work, it was unknown whether urban forests type would have a quantifiable influence on fungal phenology. Further, the maintenance of urban forests may potentially have an effect: trees tend to line fully paved surfaces in Sydney and these street surfaces are cleaned regularly, as opposed to grass cover, which exists in parks and may harbour greater quantities of decaying organic material; which, if the fungi are saprophytes, could proliferate and then produce aerosolized spores.

1.9 Urban forests and zoonotic pathogens

Urban development and urbanisation eliminates or fragments native forests habitats. The remaining urban forests and parkland environments tend to be highly managed and simplified ecosystems, rarely resembling the pre-existing natural habitats (Marzluff and Ewing 2001; Chance and Walsh 2006). Nevertheless, urban forests and green areas play important ecological roles, supporting biodiversity by acting as the only suitable habitat or refuge for many species within urban environments, particularly for birds (Meffert and Dziock 2013). These limited and fragmented environments favour and select for few

avian species, largely excluding bird species that are solitary, behaviourally inflexible, ground-nesting, and species that have short parental care durations (Davis et al 2013). This leads to a subset of resilient species that are able to adapt to urban environments (Garden et al 2006). The communities of urban birds are usually dominated by few species, whose numbers are often in high abundance, and concentrated into relatively small pockets of suitable habitat (Chance and Walsh 2006, Taylor et al 2013).

Birds are well known to be vectors of human fungal diseases, and for species that form colonies, can develop a suitable environment for fungal pathogens via their nutrient-rich droppings (guano), which provides an ideal medium for the growth of some fungi. Consequently, urban forests that harbours bird roosts has the potential to create a public health threat, especially if humans are in close proximity to the bird droppings (Tsiodras et al 2008).

Histoplasma capsulatum is a fungal pathogen often associated with bird guano. *H. capsulatum* is a dimorphic yeast which thrives in the highly nitrogenised environment created by bird droppings (Schwarz et al 1957). It is a primary pathogen known to cause severe systemic infections (Histoplasmosis) in both the immunocompromised but also immunocompetent people, if they are to inhale sufficient spores to facilitate an infectious dose (Kauffman, 2001). Another pathogen with a strong avian association is *Cryptococcus neoformans*, a yeast that can cause severe systemic infections (Cryptococcosis). However unlike *H. capsulatum*, *C. neoformans* is an opportunistic pathogen with 90% of infections occurring in AIDS patients (Sugar 1991). Other zoonotic pathogens of concern include *Candida* spp., and *Aspergillus* spp., both of which cause serious invasive infections when spores or cell fragments are inhaled (Hubálek 2004). There are also associations between birds and the dermatophytic fungi *Microsporum* spp. and *Trichophyton* spp. but there is little evidence to indicate whether these fungi are regularly transmitted from birds to humans.

Most studies that assess the association between fungal pathogens with birds focus their attention to directly sampling the bird (Hubalek 1994; Cafarchia et al 2006), or the guano or other substrates present at bird roosts (Wu et al 2012). In the few instances air samples have been taken, they have been limited to occupational safety risk assessment analysis scenarios (Banerjee 2009), and not part of designed scientific experimentation. Further, previous assessments have focussed on single pathogens (e.g. *Aspergillus*

fumigatus or *Cryptococcus neoformans*) and their presence or absence in a given circumstance.

Clearly, with an expanding urban landscape, it is paramount that a detailed assessment and evaluation of fungal pathogens derived from urban forests and its associated biodiversity are made. Similarly, bird species within urban forests that carry and transmit fungal pathogens in addition to the environmental conditions that facilitate biosafety risks need to be identified.

1.10 Study Sites

Sydney, Australia is a city with a population of 4.5 million and lies on a coastal lowland plain between the Pacific Ocean and elevated sandstone tablelands. The climate for Sydney has a warm climate, and has been described as sub-tropical (Crawford et al 2016). During summer, average temperatures range from 18.6°C - 25.8°C, and average humidity increases to 65%. During winter, average temperatures range from 8.8°C – 17.0°C, and average humidity decreases to 56%. Days in which rainfall events occur are evenly distributed throughout the year, however rainfall volume is maximal in Autumn (March–May). Little variation in topography exists across all sites used in these studies as the central region of Sydney has a relatively constant elevation (approx. 22 m above sea level across the sampled region).

Sydney ambient air quality is relatively good compared to many other cities, although concentrations of PM and NO_2 can exceed national standards on occasion (for the year 2015, $PM_{2.5}$ exceeded the national daily standard on only 4 days (OEH 2015). The main contributing source of Sydney's air pollution is fossil fuel combustion, specifically motor vehicle derived pollutants; however, domestic wood smoke in winter, and bush fires in summer can cause severe pollution events for a few days a year (NSW EPA 2008). Even though the ambient pollution levels in Sydney are low by world standards, the existing levels of air pollutants have been estimated to lead to significant mortalities (see section 1.21 and 1.4).

1.11 Objectives and Experimental Aims

The objective of this thesis was to investigate air pollutant concentrations across urban Sydney, Australia; specifically, to determine whether there were environmental

associations between air pollutant concentrations and urban forests and greenspaces including the detritus produced by these areas, and urban animal colonies.
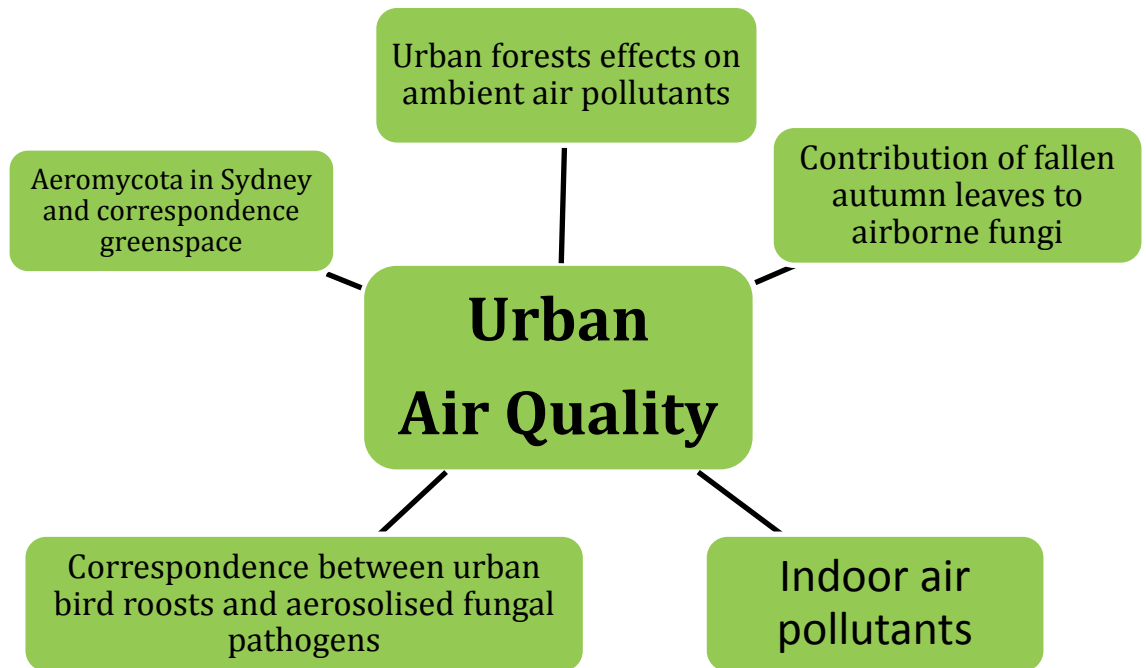
Figure 1.1 Radial cluster diagram of the 5 major chapter themes of this thesis.

The project comprised five main studies.

The first part of the project (Irga et al 2015; Chapter 2) involved a study of the correspondence between pollutant concentrations and a range of independent environmental variables in central Sydney. This component has a strong focus on urban forests and the ecosystem services relating to the urban air pollution abatement services they provide. The specific aims were to:

- To determine the quantitative seasonal periodicity of air pollutants across several sites in urban Sydney displaying varying areas of proximal greenspace or urban forest, densities of population and traffic flows, and usage profiles.
- Determine whether variations in outdoor air pollution can be predicted from the characteristics of the surrounding environment (including greenspace density).
- Identify the environmental variables most strongly associated with good and poor air quality.

The second study (Irga and Torpy 2015; Chapter 3) focused on addressing the absence of a broad ecological assessment of the factors associated with the species distribution

and density of the aeromycota of urban Sydney. The research further aimed to explore the multiple factors that contribute or influence the airborne fungal assemblages, especially those that are human pathogens, in Australia's largest city. Models were developed to predict total airborne fungal density and diversity based on a series of environmental variables that are readily available to the wider community to allow the practical use of the model. The specific aims were to:

- Determine the diversity and abundance of outdoor airborne fungal concentrations for urban Sydney.
- Quantify the seasonal trends of airborne fungi in urban Sydney.
- Investigate the spatial variability of the aeromycota across urban Sydney.
- Determine the correspondence between a range of key environmental variables and the diversity and abundance of airborne fungi.

The third study (Irga et al 2016a; Chapter 4) focused on the potential contribution of fallen leaves to the diversity of airborne fungal propagules during autumn. This investigation tested the senescent leaves of five commonly planted, exotic urban deciduous street-tree species. The leaves were subject to a manipulative experiment in which their phyllospheric fungi were identified, then allowed to become aerosolized, after which they were cultured and re-identified. The composition of the aerosolized fungi was compared with the fungi detected from direct observation of the phyllosphere. The dominant phyllosphere genera were then compared across plant types. Special attention was paid to the fungi that are known to produce allergy and other health symptoms in humans. The specific aims were to:

- Identify the phylloplane fungi on senesced autumn leaves from prevalent deciduous street tree species of Sydney.
- Assess the contribution of the phylloplane fungi to the diversity of spores that are aerosolized from those fallen leaves.
- Estimate the potential contribution of phyllospheric fungi on deciduating and decaying leaves to the aeromycota of their proximal atmospheres

The fourth study (Irga et al 2016b; Chapter 5) involves a comparison of the diversity of fungal propagules present in air samples from urban forests sites with high urban bird frequency, to sites with relatively lower bird frequency. In doing so, on the study establishes the correspondence between potential contributing environmental variables

(including bird species and habitat preferences) and the presence of fungi that could pose a human health risk. The specific aims were to:

- Compare the diversity of fungal propagules present in air samples taken from sites with high urban bird frequency, to sites with relatively lower bird frequency.
- Establish the correspondence between potential contributing bird species and environmental variables to the presence of fungal pathogens that could pose a human health risk.

It was expected *a priori* that there would be outdoor factors that could lead to changes in air quality composition, and in turn affect the types and concentrations of pollutants in indoor air, although it was unknown if, or how, airborne pollutants might respond to urban design and environmental variables. The fifth study (Irga and Torpy 2016; Chapter 6) examined indoor quality to determine the key factors associated with changes in the type and number of air pollutants detected indoors, specifically occupational buildings, and thus focused on a different range of independent variables to those tested in Chapter 2. After completion of experimental work, the data were combined with the results of the first two experiments, to determine the correspondence between indoor and outdoor air. The specific aims were to:

- Determine the relationship between the indoor air quality of office buildings at the same times and locations matched to the outdoor sites.
- Determine whether variations in indoor air quality can be predicted from the surrounding outdoor air quality, and hence from the characteristics of the outdoor environment.
- Determine and establish baseline Indoor/Outdoor ratios of air pollutants for Sydney.
- Determine whether natural, mechanical and mixed-type ventilation systems affect the indoor air quality of Sydney's buildings.
- Determine diversity and abundance of indoor airborne fungal concentrations for urban Sydney including seasonal patterns.

Following the results chapters, Chapters 7, 8 and 9 discuss the broader significance of the findings, suggest future research directions and provide a conclusion.

Chapter 2 DOES URBAN FORESTRY HAVE A QUANTITATIVE EFFECT ON AMBIENT AIR QUALITY IN AN URBAN ENVIRONMENT?

P.J. Irga, M.D. Burchett, F.R. Torpy

Statement of Joint Authorship

Peter Irga: Designed the study, collected samples, analysed and interpreted the data, and is responsible for the content and writing of the manuscript.

Fraser Torpy: Contributed to experimental design, advised on statistical analysis of data and assisted in the drafting of the manuscript.

Margaret Burchett: Offered advice and editorial comments during the drafting of the manuscript.

Keywords

2.1 Abstract

Increasing urban greenspace has been proposed as a means of reducing airborne pollutant concentrations; however limited studies provide experimental data, as opposed to model estimates, of its ability to do so. The current project examined whether higher concentrations of urban forestry might be associated with quantifiable effects on ambient air pollutant levels, whilst accounting for the predominant source of localized spatial variations in pollutant concentrations, namely vehicular traffic. Monthly air samples for one year were taken from eleven sites in central Sydney, Australia. The sample sites exhibited a range of different traffic density, population usage, and greenspace / urban forest density conditions. Carbon dioxide (CO_2), carbon monoxide (CO), total volatile organic compounds (TVOCs), nitric oxide (NO), nitrogen dioxide (NO_2), sulfur dioxide (SO_2), total suspended particulate matter (TSP), suspended particles <10 μm in diameter (PM_{10}) and particulate matter <2.5 μm ($PM_{2.5}$), were recorded, using portable devices. It was found that air samples taken from sites with less greenspace frequently had high concentrations of all fractions of aerosolized particulates than other sites, whilst sites with high proximal greenspace had lower particulates, even when vehicular traffic was taken into account. No observable trends in concentrations of NO, TVOC and SO_2 were observed, as recorded levels were generally very low across all sampled areas. The findings indicate, first, that within the urban areas of a city, localized differences in air pollutant loads occur. Secondly, we conclude that urban areas with proportionally higher concentrations of urban forestry may experience better air quality with regards to reduced ambient particulate matter; however, conclusions about other air pollutants are yet to be elucidated.

2.2 Introduction

Air pollution is ubiquitous in industrialised and densely populated regions (Begg et al 2007). Most urban air pollution comes from road traffic, and is comprised of a mixture of airborne particulate matter (PM), oxides of sulfur (SOx), oxides of nitrogen (NOx), carbon monoxide (CO), carbon dioxide (CO_2), volatile organic compounds (VOCs), polycyclic aromatic hydrocarbons (PAHs), and ozone (Thurston 2008). Outdoor air pollution kills approximately 8 million people across the world every year (WHO 2014), with a global cost of 1.7 trillion dollars (OECD 2014). Exposure to traffic-related air pollution can have infant respiratory health effects (Saravia et al 2013), and has even

been associated with autism (Volk et al 2013). In Australia it is estimated that urban air pollution causes over 1,400 deaths per annum in Sydney alone (Department of Health 2009), with national health costs estimated to be as high as 1 per cent of gross domestic product (Brindle et al 1999) or $AUD 12 billion pa, from lost productivity and medical expenses (Environment Australia 2003).

Whilst Australia has made progressive improvements to its overall air quality through regulatory measures (DEH 2004), standards set by the National Environment Protection Council are still exceeded on a few days every year. This is usually related to bushfire events, including hazard reduction burns (NSW EPA 2009), however the impact on air quality of locally sourced pollutants associated with urbanisation, climatic variability, and long periods of drought, are all contributing factors when standards are exceeded (Friend et al 2013). Clearly, governments and societies in general have a social responsibility to reduce, mitigate or ameliorate urban air pollution for the health of all organisms in urban environments, humans included.

'Urban greening' has been proposed as a means to reduce airborne pollutant levels (Chen and Jim 2008), with mounting evidence indicating that urban forestry can offer a range of 'ecosystem services' for urban residents that includes the mitigation of air pollution (Brack, 2002; Zheng et al 2013). Most of the related studies focus on the ability of urban forestry to reduce airborne PM and NO_2 (Vos et al 2013). The capacity of urban forestry, in particular trees, to reduce air pollutants is through a number of mechanisms. Trees can intercept and accumulate atmospheric particles through leaf pubescence and by providing a large waxy surface on which deposition can occur (Beckett et al 2000), and also absorb various gaseous pollutants through the stomata (Janhall 2015). Further, various tree configurations can alter wind profiles or create wind inversions via their geometry which assist in the deposition rate of pollutants from the air, or may act as physical barriers preventing the penetration of pollutants into specific areas (Salmond et al 2013; Janhäll 2015).

Many cities have plans for increasing their urban greenspace to reduce air pollution (Andersson-Skold et al 2015). The City of Sydney council is no exception, the City Council proposing to increase the city's urban canopy by 50 per cent from the current canopy cover of 15.5 per cent by 2030 (City of Sydney 2013). Such initiatives have been

shown to be both economically and environmentally effective, with urban forestry in Canberra, Australia estimated to have a combined energy reduction, pollution mitigation and carbon sequestration value of US$20–67 million between 2008 and 2012 (Brack 2002). While equivalent estimates for other Australian cities are unavailable, it is likely that the urban forest will have similar if not greater value for cities with higher population densities, such as Sydney.

Sydney's urban forestry has not previously been investigated with respect to its ability to reduce urban air pollution. Further, few studies from any location are available that provide experimental data on the air pollutant removal capacity of urban forestry/greenspace , as opposed to model estimates such as the Urban Forest Effects Model (UFORE) by Nowak (2006). Similarly, few reports provide empirical evidence of the association between overall ambient particulate matter densities and urban forestry densities (Pataki et al 2011). Efforts to demonstrate or validate the model estimates have either found substantially reduced improvements relative to estimates from models (Tallis et al 2011) or have shown no positive effects (Satala et al 2012).

The current project aimed to determine whether a discernible relationship does exist between higher densities of urban forestry and reduced local ambient air pollutant levels.

2.3 Methods

2.3.1 Study area

Sydney, Australia has a population of 4.5 million and lies on a coastal lowland plain between the Pacific Ocean and elevated sandstone tablelands. The climate for Sydney 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 (OEH 2014). The main source of Sydney's air pollution is fossil fuel combustion, specifically motor vehicle exhaust; however domestic wood smoke in winter, and bush fires in summer can cause severe pollution events for a few days a year (NSW EPA 2008).

Within the City of Sydney Local Government Area (LGA), the total tree canopy cover has been estimated at 15.5% (City of Sydney 2013), of which 6.6% of the canopy cover

is on private land, 4.9% is street trees, and 4.1% is in parks. The City's street trees are restricted in diversity, with whole lengths of streets planted with single species. *Platanus* x *acerifolia* is (London Plane Tree) is the most common Sydney street tree, comprising 9.5% of the urban forest, followed by *Melaleuca quinquenervia* (Broad-leaved Paperbark) and *Lophostemon confertus* (Brush Box), both of which comprise 8.8% of the urban forest (City of Sydney 2013).

2.3.2 Sample Sites

In collaboration with the City of Sydney Council, sites were selected so as to encompass a range of different conditions with respect to traffic density, usage, and greenspace / urban forest density (Fig. 1; Table 1). High canopy cover of > 20% of the total land area was present in the sample sites at Centennial Park, Sydney Park, Surry Hills, and Rushcutters Bay (City of Sydney 2013). These sites have high canopy coverage due to both highly planted residential areas and/or extensive public parklands. Sites with moderate canopy coverage (10% – 20% of total land area) included Chippendale, Glebe, and Prince Alfred Park. Sites with low canopy cover (0–10% of total land area), included Haymarket, Zetland and Town Hall and Pitt St sites in central Sydney; which are built up, inner city areas. Little variation exists in topography across all selected sites as the central region of Sydney has a relatively constant elevation (approx. 22 m above sea level across the sampled region).
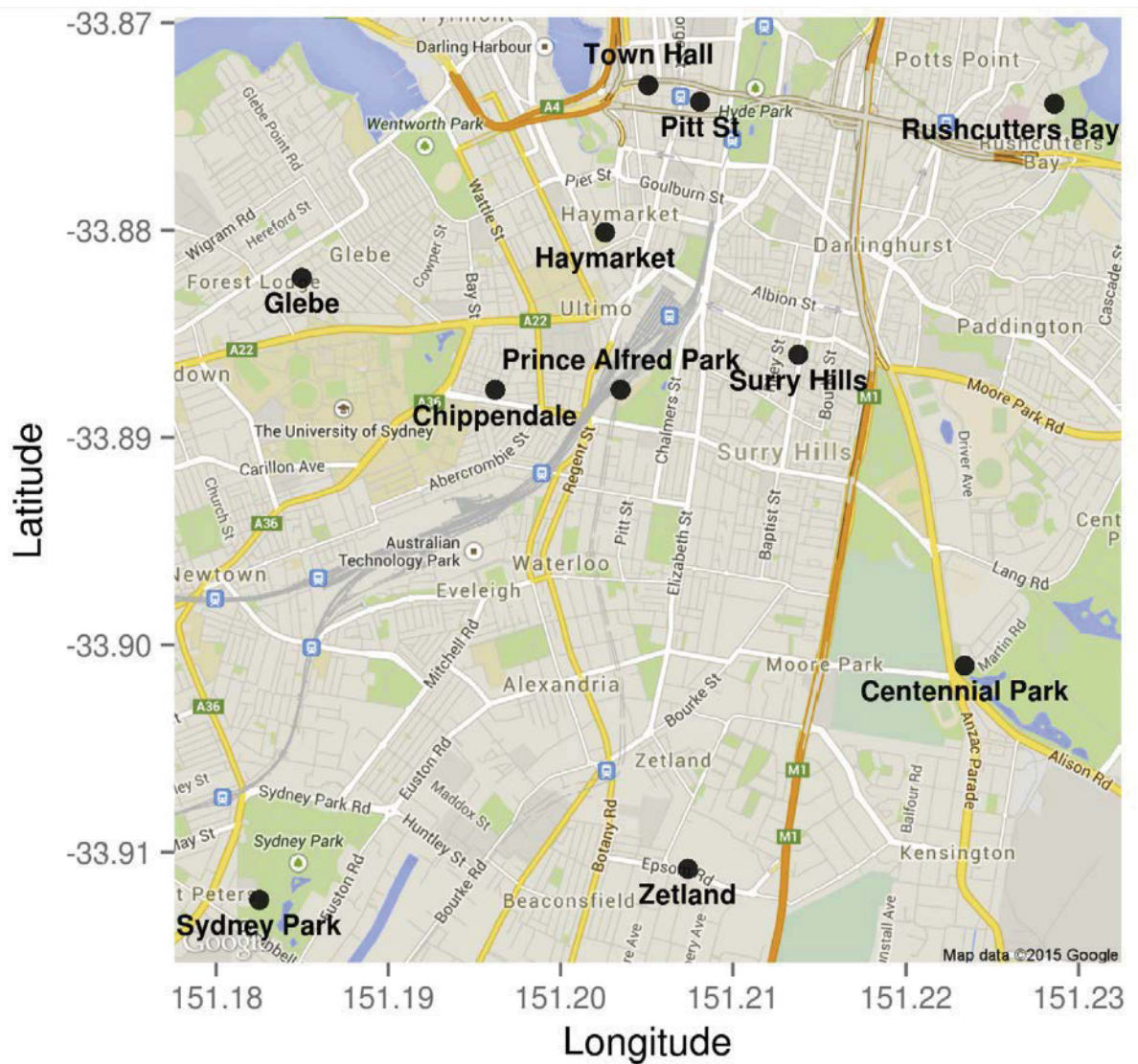
Figure 2.1 Map of central Sydney, showing the locations of the eleven sampling sites. Figure made using the packages ggplot2 and ggmaps for the program R (The R foundation, 2015), and static maps from Google Maps.

2.3.3 Traffic density and Greenspace assessment

The concentration of greenspace at the sites was estimated using satellite imagery from Google maps, within 100 m, 250 m and 500 m radii from the geographic centre of each sample site, forming areas of 3.14, 19.6 and 78.6 ha respectively. The proportions of these areas under tree cover (including shrubs >1 m in height), grass cover and total greenspace (trees + grass) were calculated using Google Maps Distance Calculator (2013). The zoom capability of the Google maps allowed accurate estimates of greenspace cover to be made. The data is shown in Table 2. Please note that the data are

cumulative, meaning that the 250 m data includes the greenspace within the 100 m radius sites etc.

Previous work that has empirically assessed the relationships between urban vegetation and air pollution has not accounted for spatial variation in the primary source of air pollutants. Motor vehicle exhaust is the main contributor to locally sourced pollutants for the sample area, and thus can be expected to be a major determinant of between-site variation in ambient air pollution. Despite all samples being conducted in the city's CBD, the traffic density was not homogenous. To estimate traffic densities at the sample sites, several traffic sampling points were selected within each site: 2 points within the 100 m radius areas, 4 between the 100 and 250 m radii and a further 7 between the 250 and 500 m radii areas. Traffic sampling points were selected based on a stratified random sampling process among high, medium and low traffic density roadways. Traffic was sampled manually by counting vehicles passing the sample point for one 3 hour period per location. Samples were taken mid-week, between 1100 and 1400 h (the same time interval during which the air quality samples were taken) to avoid the three daily peak traffic periods. Total traffic density was estimated by calculating vehicle movements per minute and multiplying by the number of streets of each roadway type within each specified radius. Whilst these traffic density estimates do not provide a quantitatively precise measure of the total road transport density at the 11 locations, this method represents a functional proxy for the overall road use in the areas surrounding the sample sites, as the data was collected during the same 'traffic behaviour' periods as the air samples, as well as allowing for the randomization of vehicle type. Traffic densities at the sites are shown in Table 3.

Table 2.1 Attributes of the sample sites

Site	Coordinates	General land use and environment	Relative greenspace	Vegetation type	Vegetation composition
Sydney Park	(−33.9108, 151.2074)	Medium density residential and parkland area	High	Parkland	Grassed lawns and a combination of the following tree species: *Casuarina* spp, *Corymbia maculata*, *Angophora costata*, *Eucalyptus sideroxylon*
Centennial Park	(−33.9010, 151.2234)	Medium density residential and parkland area	High	Parkland	Pond side vegetation and a combination of the following trees: *Ficus macrophylla*, *Araucaria cunninghamii*, *Eucalyptus saligna*, *Melaleuca quinquenervia* and *Liquidambar styraciflua*
Rushcutters Bay	(−33.8739, 151.2286)	Medium density residential harbour side area	Relatively high	Highly planted residential area	*Lophostemon confertus*, *Ficus macrophylla*, *Jacaranda* spp, *Eucalyptus sideroxylon* and *Eucalyptus saligna*
Prince Alfred Park	(−33.8884, 151.2035)	Medium density residential and parkland area	Moderately high	Parkland	*Lophostemon confertus*, *Melaleuca quinquenervia*, and *Ficus* spp.
Surry Hills	(−33.8860, 151.2138)	Medium density residential and commercial area	Moderately high	Planted residential areas	*Lophostemon confertus*, *Liquidambar styraciflua*, *Platanus* × *acerifolia*, *Poplus nigra*
Chippendale	(−33.8877, 151.1962)	Medium density residential and commercial area	Moderate	Street trees and planted residential areas	*Lophostemon confertus*, *Liquidambar styraciflua*, *Platanus* × *acerifolia*
Glebe	(−33.8823, 151.1850)	Medium density residential area	Moderate	Street trees and planted residential areas	*Callistemon* spp, *Lophostemon confertus*, *Liquidambar styraciflua*, *Platanus* × *acerifolia*, *Jacaranda* spp
Haymarket	(−33.8801, 151.2026)	High density residential and commercial area	Moderate low	Only street trees are present	*Platanus* × *acerifolia*
Zetland	(−33.9108, 151.2074)	High density residential and commercial area	Low	Only street trees are present	*Lophostemon confertus* street trees
Town Hall	(−33.8730, 151.2051)	High density commercial area	Very low	Only street trees are present	*Platanus* × *acerifolia* and *Poplus nigra*
Pitt St	(−33.8738, 151.2081)	High density commercial area	Very low	Only street trees are present	*Platanus* × *acerifolia* and *Poplus nigra*

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Table 2.2. Greenspace cover composition (area %) at the sample sites within 100 m, 250 m and 500 m radii. 'Canopy' cover was comprised of tree and shrub species. 'Combined' cover is the sum of canopy and grass cover.

Site	100 m radii			250 m radii			500 m radii		
	Canopy	Grass	Combined	Canopy	Grass	Combined	Canopy	Grass	Combined
Sydney Park	2.1	84.9	87.0	7.3	72.4	79.7	4.3	42.0	46.2
Centennial Park	35.4	37.2	72.6	13.6	54.2	67.8	13.6	58.9	72.5
Rushcutters Bay	40.5	27.1	67.5	30.5	13.8	44.3	11.8	12.3	24.2
Prince Alfred Park	20.0	7.1	27.1	16.7	8.6	25.3	8.6	5.6	14.2
Surry Hills	21.1	2.6	23.7	12.5	0.7	13.2	8.7	1.6	10.3
Chippendale	21.5	2.0	23.5	12.6	0.8	13.4	4.2	12.7	16.8
Glebe	17.9	0.3	18.2	13.9	0.4	14.3	11.1	1.9	13.0
Haymarket	11.0	0.0	11.0	7.4	0.0	7.4	8.6	1.1	9.7
Zetland	8.5	1.8	10.3	4.0	2.1	6.1	7.3	1.2	8.5
Town Hall	3.6	0.0	3.6	5.7	0.2	5.9	1.6	0.8	2.4
Pitt St	1.9	0.0	1.9	8.1	0.6	8.8	18.4	4.5	22.9

Table 2.3 Cumulative traffic movements per minute at the sample sites, within 100 m, 250 m and 500 m radii.

Site	Traffic density (movements. min^{-1})		
	100 m radii	250 m radii	500 m radii
Sydney Park	7.4	105.9	300.9
Centennial Park	1.3	145.9	215.9
Rushcutters Bay	7.3	29.2	319.2
Prince Alfred Park	42.7	217.4	517.5
Surry Hills	11.5	73.6	254.0
Chippendale	61.1	93.2	363.9
Glebe	11.6	75.2	164.5
Haymarket	13.4	82.5	331.5
Zetland	26.8	42.9	199.9
Town Hall	53.4	190.8	562.7
Pitt St	38.9	217.4	424.6

2.3.3 Air quality sampling

Monthly air samples were taken across eleven sites within the City of Sydney LGA between September 2013 and August 2014. A temporally independent design was implemented, where air samples were collected from different randomly selected points within the 100 m radius of the site centres each month, to negate temporal non-independence and the requirement for repeated measures analysis. Air samples were collected from the sites with several portable instruments. Carbon dioxide (CO_2), carbon monoxide (CO), total volatile organic compounds (TVOCs), nitric oxide (NO) and sulfur dioxide (SO_2) were measured with a *Yessair* 8-channel IAQ Monitor (Critical Environment Technologies Vantage Way Delta, Canada). Total suspended particulate matter (TSP), respirable suspended matter (PM_{10}: suspended particles <10 µm in diameter) and very fine particulate matter ($PM_{2.5}$: particles <2.5 µm in diameter) were recorded with a *DustTrack II* 8532 laser densitometer (TSI, Shoreview, Minnesota). The DustTrak has been shown to overestimate particulate matter for certain particulate materials (Kingham et al 2006), especially the $PM_{2.5}$ fraction; thus data recorded was corrected with data sourced from the NSW Office of Environment and Heritage (OEH) (explained further below). Meteorological data was obtained from the Australian Government Bureau of Meteorology 2013–2014 (rainfall, wind speed, wind direction and humidity). Nitrogen dioxide (NO_2) was recorded with a *GasAlert Extreme* T2A-7X9 (BW Technologies, Canada). Temperature, light, noise and relative humidity were

recorded using a *Digitech Multifunction Environment meter* (Digitech, China). A *Turbometer Davis anemometer* (Davis Weather Gadgets, Cannon Beach, Oregon) was used to measure wind speed.

2.3.4 Quality assurance

Air sample collection was conducted at least 30 m from roadways to allow the dispersal of pollutants sourced from the street. Rainy days were avoided, as rain has been shown to remove particulate matter from the air (Nishihara et al 1989). Furthermore, no bare soil was present within 30 m proximity of sampling, to avoid any dust contribution to PM concentrations. The order in which sites were sampled was randomised for every sampling day, to remove any systematic temporal variation within the allocated sampling time interval. Reference data from three air quality monitoring sites operated by the OEH were obtained for comparison on the days that samples were collected, for PM_{10}, $PM_{2.5}$, CO, NO_2 and SO_2. The reference sites included: Randwick (1 km from the closest sample site); Rozelle (3.5 km from the closest sample site) and Earlwood (10 km from the closest sample site). The OEH air quality monitoring sites utilise a tapered element oscillating microbalance (TEOM) for particulate matter quantification as per the Australian Standard (AS 3580.9.8 - 2001), approved by the NSW EPA (2007). The average TEOM data sourced from these monitoring sites was used to correct the over estimation of particulate matter data obtained from the *DustTrak,* as per the recommendation of Kingham et al (2006), by calculating the difference in recorded data with the mean from the three OEH sites and applying it as a correction factor for each sampling event.

2.3.5 Data analysis

All data are expressed as means ± standard error. All analyses were performed using Minitab Ver. 14 (Minitab Inc., 2003). To allow us to determine the relationship between proximal greenspace and air quality, it was necessary to account for inter-site variability related to pollutant density associated with road traffic. Thus a stepwise multiple linear regression was used to determine the traffic variable that had the strongest relationship with the air quality variables, which was traffic density within the 100 radii in all instances. The air quality variables were corrected for the effects of traffic by performing subsequent analysis on the residuals from linear regressions between the air quality variables and this traffic variable.

The presence and strength of linear associations between pollutant concentrations and environmental conditions were examined by computing Pearson correlation coefficients after checking the normal bivariate distribution of the data graphically. Stepwise multiple linear regression was then carried out to determine the relative influence of the environmental variables on the inter sample variance in all pollutant concentrations. For this analysis we treated all samples as independent (ie. samples were collected using a temporally independent design), and thus it did not allow the distinguishing of any seasonally-dependent interaction effects between air quality and environmental variables, should they be present.

2.4 Results

Site and monthly trends for PM are displayed in Figure 2.2 and 2.3. Samples taken from the sites that exhibited the lowest concentrations of greenspace, i.e. Town Hall, Pitt St and Haymarket, generally had the highest concentrations of total suspended particles; recording 34.0 ± 4.0 $\mu m/m^3$, 33.3 ± 3.3 $\mu m/m^3$ and 28.4 ± 4.9 $\mu m/m^3$ respectively. In comparison, the sites that had the most greenspace (Centennial Park and Rushcutters Bay), recorded the lowest total suspended particle concentrations relative to other sites, with 17.5 ± 2.1 $\mu m/m^3$ and 19.3 ± 4.2 $\mu m/m^3$. These levels were significantly lower than the three sites with the lowest greenspace (GLM ANOVA, both $P<0.000$ compared with Town Hall, Pitt St and Haymarket sites). This same trend was observed in the other fractions of particulate matter, with Town Hall, Pitt St and Haymarket consistently recording significantly higher concentrations of PM_{10} and $PM_{2.5}$ than sites with higher density of greenspace (GLM ANOVA, all $P<0.000$ compared with Chippendale, Glebe, Rushcutters Bay, Centennial Park).
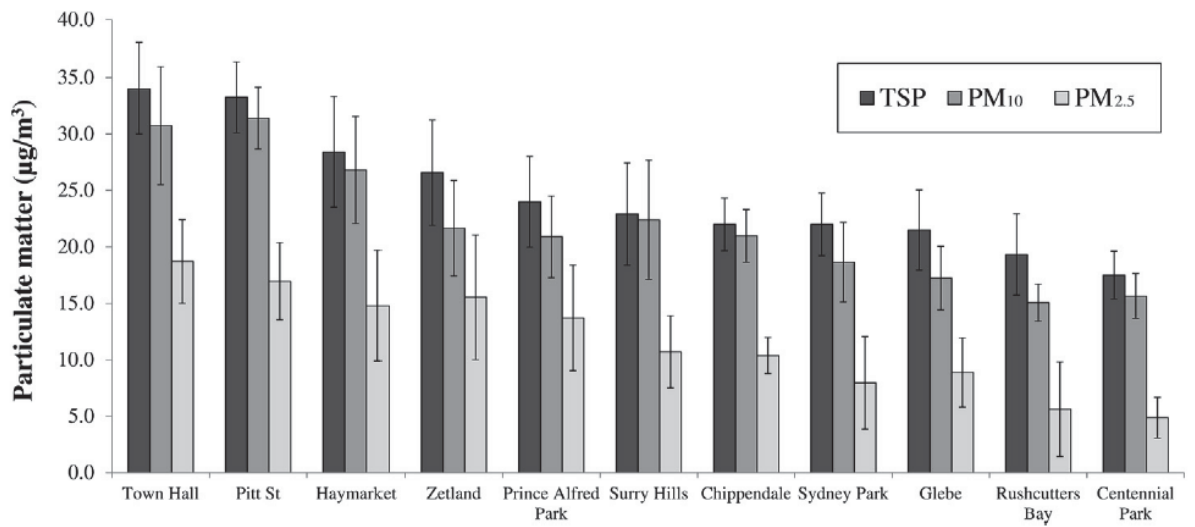
Figure 2.2. Average levels of atmospheric particulate matter fractions for each sampling site, over a 12-month period (Means ± SEM, n =12).
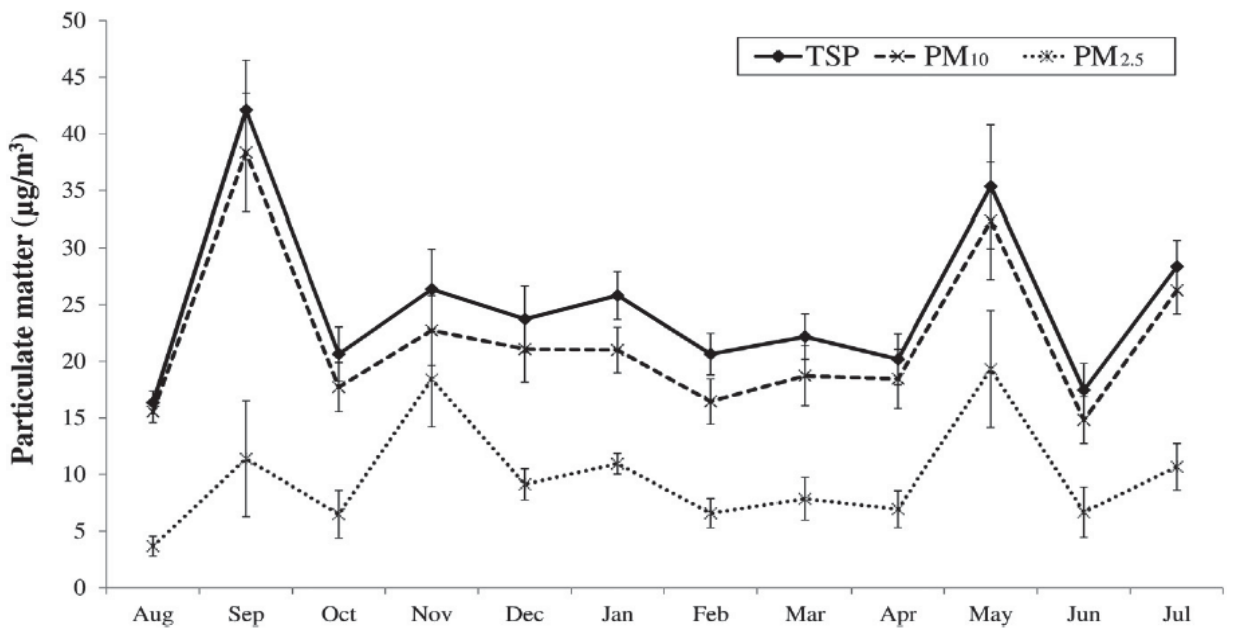
Figure 2.3. Average levels of atmospheric particulate matter fractions averaged across sites, over the 12-month sampling period (Means ± SEM, n = 11).
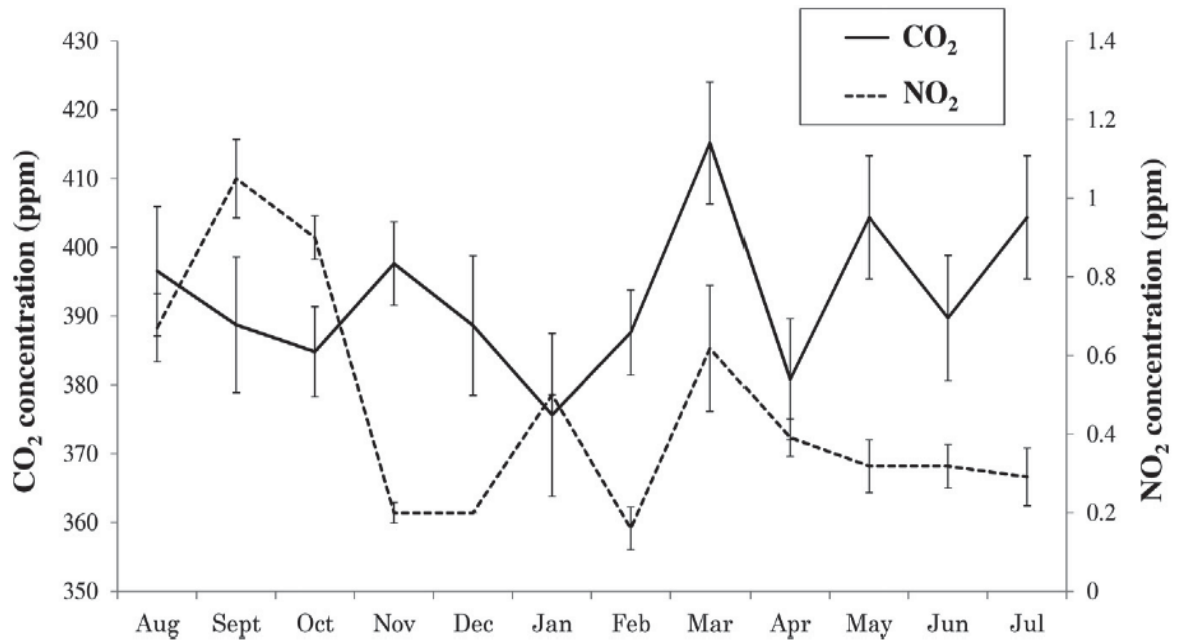
Figure 2.4. Temporal concentrations of ambient atmospheric CO_2 and NO_2 averaged across sites, over the 12-month sampling period (Means ± SEM, n = 11).
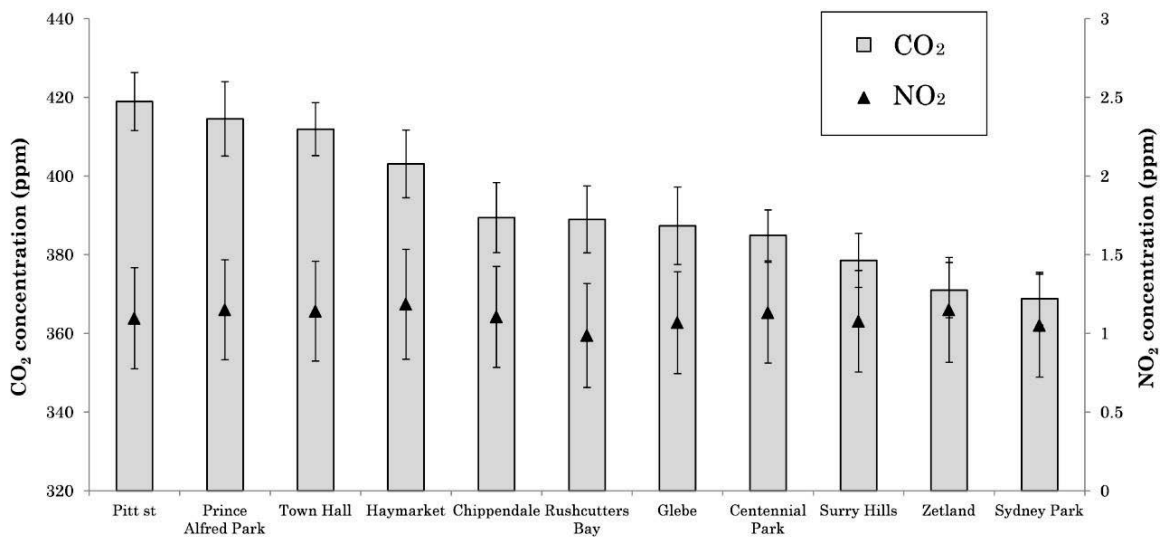
Figure 3.5. Average concentrations of atmospheric CO_2 and NO_2 for each sampling site, averaged over the 12-month period (Means ± SEM, n = 12).

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Little seasonal variation was observed in particulate concentrations other than significantly higher concentrations in TSP and PM_{10} in September and May (GLM ANOVA, both P<0.000 compared to all other months). The high concentrations observed in September are attributable to the hazard reduction burns that took place during that time (OEH 2014). A secondary peak was observed across all sited during the month of May, a probable consequence of the low precipitation rates at the time (OEH 2014). No seasonal trend was observed with $PM_{2.5}$ across the sites (GLM ANOVA, P>0.000).

Trends for CO_2 and NO_2 are displayed in Fig. 3.4 and 3.5. Some significant differences were present in CO_2 and NO_2 concentrations between months. No consistent pattern was observed across months, and amongst sites, the pattern of CO_2 concentrations (Fig. 5) was variable, the only statistically significant difference observed being Pitt St, Prince Alfred Park and Town

Hall recording significantly higher concentrations than those recorded for Sydney Park and Zetland (GLM ANOVA, P<0.05 for all differences mentioned). There was no variation in NO_2 concentrations amongst sites. No seasonal trend in CO_2 was observed, with mean concentrations ranging from 377 ppm to 414 ppm. No seasonal trends in NO_2 were observed other than significantly higher concentrations in August and September which were once again attributable to the hazard reduction burns that take place during that period (GLM ANOVA, P<0.05). The temporal and spatial variation amongst CO_2 and NO_2 samples was not of a magnitude that warranted detailed multivariate analysis.

Data for NO, TVOC, CO and SO_2 were consistently below detection limits, and were thus not analysed individually. However, these air quality variables were used for the multivariate analyses, since there is evidence that multiple air pollutants may have additive effects (e.g. Dominici et al 2002).

2.4.1 Relationships with Environmental variables

Correlations between particulate matter measurements acquired from the Bureau of Meteorology and our samples were positive and significant (r≥0.836, P=0.000), indicating that our instrument readings were closely proportional to the TEOM data, and any variation between the readings was thus likely attributable to spatial separation. To

test the potential effect of temperature, humidity and local wind direction measured at each site, these variables was used as a predictor data variables and collated and assessed univariately. Daily prevailing wind direction and all precipitation data obtained from the Bureau of Meteorology was also used as predictors, however this data represented averages from across Sydney, and was not different amongst sites.

Traffic corrected total suspended particle concentrations were significantly negatively correlated with canopy coverage within a radius of 100 m (r= –0.293, P=0.001), canopy coverage within 250 m (r = – 0.221), P=0.011), percentage total greenspace cover measured at 100 m radii (r = –0.189, P=0.03), 250 m radii (r = –0.191, P=0.028) and 500 m radii (r= –0.181, P=0.038). However, total suspended particle data was also significantly negatively correlated with monthly total rain recorded per the sampling period (r= –0.244, P=0.005), total rain recorded in the preceding week (r= –0.244, P=0.005), and significantly positively correlated with time duration since last rain event (r= 0.417, P=0.000). Traffic corrected PM_{10} data were significantly negatively correlated with canopy coverage within 100 m (r= –0.250, P=0.004), canopy coverage within 250 m (r = – 0.213, P=0.014), and percentage greenspace cover measured at 100 m radii (r = –0.179, P=0.04). However, PM_{10} data was also significantly negatively correlated with wind speed (r = –0.180, P=0.039), monthly total rain recorded during the sampling period (r= –0.226, P=0.009), total rain recorded in the preceding week (r= –0.141, P=0.004), and significantly positively correlated with time duration since last rain event (r= 0.461, P=0.000). Traffic corrected $PM_{2.5}$ data was not significantly correlated with any greenspace variable, but was significantly negatively correlated with monthly total rain recorded per the sampling period (r= –0.226, P=0.009), total rain recorded in the preceding week (r= –0.232, P=0.008), and significantly positively correlated with time duration since last rain event (r= 0.443, P=0.000).

Multiple stepwise linear regression analysis was used to determine which environmental variables were the strongest predictors of the aerosolized particulate matter. The analysis ranked the variables in order of predictive power, with backward elimination performed to check the significance of each variable. Only predictor values that contributed to over 2% of the overall explanatory power and were significant (P<0.05) were considered.

For traffic corrected TSP concentrations, the time since last rain event was the largest contributor to the overall variation in the model, explaining 17.41 % of the linear pattern in the TSP data ($R^2=17.41$). Adding canopy coverage within 100 m to the model explained an additional 9.86% of the variation, and adding canopy coverage within the 500 m radii added 2.94 % explanatory power. The three variable model thus explained 30.45% of the variability in the data set ($R^2=30.45$). When traffic corrected PM_{10} was used as the response variable, six combined predictors were detected: time since last rain event was the largest contributor to the overall variation in the model, explaining 21.24 % of the linear pattern in the PM_{10} data ($R^2=21.24$). Adding canopy coverage within 100 m to the model explained an additional 6.25% of the variation, adding wind speed explained an additional 4.80% of the variation, adding canopy coverage within 500 m radii of the site centres explained an additional 4.9% of the variation, and adding monthly total rain explained an additional 3.69 % of the variation. Combined, the six variable model thus explained 41.08% of the variability in the data set ($R^2=41.08$). For traffic corrected $PM_{2.5}$, the analysis indicated that only three predictors were worthy of consideration, as these variables were the only ones that were statistically significant and adding further predictors to the model made little contribution to its overall explanatory power. Time since last rain event was the largest contributor to the overall variation in the model, explaining 19.59 % of the linear pattern in the $PM_{2.5}$ data ($R^2=19.59$). Adding canopy coverage within 100 m to the model explained an additional 6.16 % of the variation, and adding and wind speed added 3.20% explanatory power. The three variable model thus explained 28.59% of the variability in the data set ($R^2=28.59$).

2.5 Discussion

This study provides data on a range of ambient and seasonal air pollutants for sites across central Sydney, Australia, and is the first study to use a competitive model to determine the relative importance of environmental predictors for air quality, including proximal greenspace.

Sites that frequently had high concentrations of PM; Town Hall, Pitt St and Haymarket, all had low greenspace densities, with proximal greenspace coverage within 100 m radii measuring 1.9%, 3.6% and 11.0% respectively. Conversely, sites that had the lowest concentrations of PM; Centennial Park, Rushcutters Bay and Glebe had both the highest greenspace recorded and the highest canopy coverage, with proximal tree coverage within 100 m radii measuring 35.4%, 40.5% and 17.9% respectively. Additionally, all

fractions of particulate matter were significantly negatively correlated with greenspace; thus increasing greenspace was associated with decreasing particulate matter despite the data being corrected for traffic density. The strongest associations with decreasing PM were canopy coverage within 100 m radii of the sample sites. However, associations were also found between all fractions of particulate matter and environmental elements such as wind speed, time since last rain event and quantity of rain recorded in the time proximal to the samples being taken. Clearly, these meteorological factors are also important factors in determining ambient particulate concentrations, as demonstrated previously by Cavanagh et al (2009). In the current study, when these factors were analysed for their combined effects utilising stepwise multiple linear regression, time since last rain event was the strongest predictor of the concentrations of all PM types; however in all models, canopy coverage within 100 m radii was the next strongest contributing factor. Thus, whilst meteorological factors had the strongest influence in determining particulate matter concentrations; greenspace, and especially canopy coverage proximal to the sample sites, were integral influences on reduced ambient particulate matter concentrations. This outcome is in agreement with most of the studies that have assessed the ability of urban forestry to reduce particulate concentrations, with Freiman et al (2006) finding that ambient PM concentrations were lower in neighbourhoods with dense vegetation, and Cohen et al (2014) demonstrating reduced pollutant concentrations including PM levels in an urban park compared to proximal street canyons. Similarly, Cavanagh et al (2009) observed decreasing particulate concentrations with increasing distance inside an urban forest patch in Christchurch, New Zealand. However, whilst these studies demonstrated that urban vegetation may have benefits in regards to pollution mitigation, they lacked spatial and temporal replication, with Freiman et al (2006) sampling 6 sites, Cohen et al's (2014) total study duration was 6 days at 4 sites, and Cavanagh et al (2009) only studying one forest patch with nested sites within the site. The only study with substantive replication was Setälä et al (2013), who did not detect a relationship between urban vegetation and air pollutants in Finland; subsequently concluding that the effect of greenspace was minor in northern conditions.

A recently developed tool to measure urban forest ecosystem services is the Urban FORest Effects (UFORE) model developed by the U.S Forest Service (Nowak 2006). Although the model was specifically designed for US studies, it has been widely applied

across the world, with use in Barcelona, Spain (Baró et al 2014), Shenyang, China (Li et al 2014) and Perth, Australia (Saunders et al 2011). However, as detailed by Baró et al (2014), the model has limitations, as it is based on PM deposition rates for specific plant species which limits the usefulness of the model to areas dominated by the species included in the database. Further the model has uncertainty in relation to particle re-suspension rates and fine scale spatial variability in air pollutant concentrations; thus Baró et al (2014) concluded that the model should be used for approximate estimations rather than precise quantification. Although the dry deposition velocity of PM on urban forestry was not documented in the current study, we did, however, find that areas with proportionally higher densities of urban forestry were quantifiably associated with reduced ambient particulate matter levels, thus providing empirical evidence that could verify the UFORE ecosystem service approximations.

The statistical model utilised in this experiment did not fully explain majority of the variation in the data set, indicating that there were other variables not accounted for, and that determining all, or even most of the causative factors associated with urban air pollution experiments can be challenging. Thus there are clearly manifold environmental variables that influence air quality in a city environment at any one time and in any specific location. Whilst we cannot account for majority of the temporal and spatial variation in air quality with the environmental variables chosen for analysis in this study, the identification of greenspace as an important determinant of city airborne pollutants is a significant contribution to our understanding of UAQ, and should assist in future air quality modelling exercises.

Model estimates indicate that areas with high greenspace could have lower NO_2 concentrations (Pugh et al 2012), however empirical data to demonstrate this is lacking and efforts to demonstrate this trend have failed to find effects of the magnitude detected by empirical estimates (Grundström et al 2014; Setälä et al 2013). Similarly, substantial reductions in NO_2 were not evident in the current study. Additionally, no associations with NO_2 and any measured environmental variable were found. Increases in NO_2 were seen in the Spring months, whilst all other months showed no seasonal trends. Atmospheric NO_2 is associated with combustion, and thus the increased concentrations during Spring appears to be associated with hazard reduction burning (OEH 2014).

Variation in temporal and spatial ambient atmospheric CO_2 concentrations is influenced by a range of seasonal, meteorological and land usage factors (Henninger and Kuttler 2010). In urban environments, variations in ambient atmospheric CO_2 concentrations are mainly associated with combustion processes (Idso et al 2001), leading to peak CO_2 concentrations in the centre of cities. Whilst the city centre sites had high CO_2 concentrations, this pattern was not consistent across the full sample. Restrictions in the upward movement of pollutants released at ground level has been demonstrated in areas constricted with tall buildings; therefore, accumulation of pollutants may occur in built up areas (Gratani and Varone 2005); which may also explain the high inner city CO_2 concentrations we detected on occasion. Higher wind speeds facilitate increased CO_2 advection in the atmosphere, which also could be a contributing factor to the reduced levels observed in less built up areas. Clearly, there are many challenges in relation to assessing urban CO_2 fluxes (Grimmond et al 2002), and further research on this air pollutant are needed. Whilst CO_2 levels varied considerably between months in our study (Fig. 4), there was no explainable pattern observed.

Concentrations of NO, TVOC, CO and SO_2 were frequently below the detection limits of the devices utilised in this experiment. This may be a testament to the efficacy of the governmental regulatory efforts imposed to improve Sydney's air pollutant levels (DEH State of the Air Report, 2004). If these pollutants do not exceed concentrations of concern, the ability of urban greenspace to reduce them will not be realised. Whilst it cannot be deduced from this experiment, it is possible the Sydney's urban forestry is biomitigating NO, TVOC, CO and SO_2 concentrations so as to maintain low concentrations. Yin et al (2011) demonstrated that densely vegetated areas within urban environments in a city in China had reduced atmospheric concentrations of SO_2 and NO_2. However, it remains unknown if Sydney's urban forestry is having a quantifiable influence on ambient NO, TVOC, CO and SO_2 concentrations.

We also tested the effect of wind direction on the variation in pollutant levels during the study by using wind direction as a categorical variable for univariate assessment of air pollutant levels. Whilst there were minor differences in pollutant levels when the prevailing winds varied, none of these effects approached statistical significance, and thus could not explain a meaningful proportion of the overall variability in air quality during the study.

Many tree species that comprise Sydney's urban forestry are deciduous, and thus have no leaves during winter, consequently affecting their ability to intercept and accumulate atmospheric particulates, and to absorb various gaseous pollutants. It is difficult to evaluate whether the seasonal behaviour of Sydney's urban forestry had a quantifiable influence on proximal ambient air pollutants from this study. Further, different vegetation types are known to have different deposition rates for particulates (Beckett et al 2000, Sæbø et al 2012). Thus, future work that documents the density and type of greenspace, as well as calculating approximate total leaf area based on allometric equations, whilst monitoring ambient urban air pollution could be of value.

Using a simulation model, Wania et al (2012) determined that dense tree cover in deep street canyons inhibit the upward flow of particulate matter, and thus diminish its dispersal rates. Their model indicated that particulates created from traffic are increased in street canyons with large height-to-width ratios, and that vegetation within these street canyons could compound the issue. As it was not the focus of the current study to examine such phenomena, these scenarios were avoided when sampling. Thus, future research that considers such factors as streetscape design when assessing the effects of urban vegetation on ambient air quality in urban environments would be of value.

In the current study, the volume of accumulative traffic movements was used as a measure of the primary local pollutant source. As vehicle derived exhaust pollution is known to vary between vehicle type, fuel type and vehicle age (Rhys-Tyler et al 2011), further work that takes into account the potential differences in vehicle pollution due to traffic variables such as traffic lights as well as the continuousness of traffic flow may also be of value.

2.6 Conclusion

Samples taken from low greenspace sites frequently had higher concentrations of all fractions of aerosolized particulate matter than other sites. Comparatively, sites with high proximal greenspace had lower particulates, even when pollutant sources were corrected for and factored into the analysis. Further, all fractions of particulate matter were significantly negatively correlated with greenspace, with increasing greenspace associated with decreasing particulate matter, even when meteorological and traffic density being considered. This is the first study to comprehensively demonstrate, with substantial temporal and spatial replication, that areas with proportionally higher

concentrations of urban forestry are quantifiably associated with reduced ambient particulate matter levels. Conclusions concerning other air pollutants (CO_2, NO_2, NO, TVOCs or SO_2) are yet to be elucidated.

2.7 Acknowledgments

Chapter 3 A SURVEY OF THE AEROMYCOTA OF SYDNEY AND ITS CORRESPONDENCE WITH ENVIRONMENTAL CONDITIONS: GRASS AS A COMPONENT OF URBAN FORESTRY COULD BE A MAJOR DETERMINANT

P.J. Irga, F.R. Torpy

Statement of Joint Authorship

Peter Irga: Designed the study, collected samples, analysed and interpreted the data, and is responsible for the content and writing of the manuscript.

Fraser Torpy: Contributed to experimental design, advised on statistical analysis of data and assisted in the drafting of the manuscript.

3.1 Abstract

A comprehensive survey of airborne fungi has been lacking for the Sydney region. This study determined the diversity and abundance of outdoor airborne fungal concentrations in urban Sydney. Monthly air samples were taken from 11 sites in central Sydney, and culturable fungi identified and quantified. The genus *Cladosporium* was the most frequently isolated fungal genus, with a frequency of 78% and a mean density of 335 CFU/m^3. The next most frequently encountered genus was *Alternaria*, occurring in 53% of samples with a mean of 124 CFU/m^3. Other frequently identified fungi, in decreasing occurrence, were: *Penicillium*, *Fusarium*, *Epicoccum*, *Phoma*, *Acremonium* and *Aureobasidium*. Additionally, seasonal and spatial trends of airborne fungi were assessed, with increases in total culturable fungal concentrations experienced in the summer months. The correspondence between a range of key environmental variables and the phenology of airborne fungal propagules was also examined, with temperature, wind speed and proximal greenspace having the largest influence on fungal propagule density. If the greenspace comprised of grass, stronger associations with fungal behaviour were observed.

Irga, P. J., and F. R. Torpy (2016). A survey of the aeromycota of Sydney and its correspondence with environmental conditions: grass as a component of urban forestry could be a major determinant. Aerobiologia. **32**: 171-186.
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Chapter 4 ASSESSING THE CONTRIBUTION OF FALLEN AUTUMN LEAVES TO AIRBORNE FUNGI IN AN URBAN ENVIRONMENT

P.J. Irga, M.D. Burchett, G. O'Reilly, F.R. Torpy

Statement of Joint Authorship

Peter Irga: Designed the study, collected samples, analysed and interpreted the data, and is responsible for the content and writing of the manuscript.

Fraser Torpy: Contributed to experimental design, advised on statistical analysis of data and assisted in the drafting of the manuscript.

Gabe O'Reilly: Assisted in the collection of samples and prepared a figure utilised in the manuscript.

Margaret Burchett: Offered advice and editorial comments during the drafting of the manuscript.

Keywords

4.1 Abstract

Street trees in urban areas are often deciduous and drop leaves during autumn. This investigation aimed to assess the potential contribution of senescent leaves to the diversity of airborne fungal propagules during the season of autumn. The senescent leaves of five deciduous tree species were subject to a manipulative experiment in which their phyllospheric fungi were aerosolized, and air samples taken to document the spores derived from leaf material. Aerosolized fungi were compared with the fungi detected from direct leaf of the phyllosphere. Thirty-nine genera were identified across the plant species sampled, of which twenty-eight genera were present in the corresponding air samples. Significant differences were observed amongst the fungal genera growing on the leaves of the different trees, however few differences were found in the composition of fungal spores that were aerosolized. The dominant genera that were aerosolized were: *Penicillium*, *Cladosporium*, *Alternaria*, *Chaetomium*, *Botrytis* and *Trichothecium*. As these fungal genera are commonly identified in autumn air samples in other studies, it is likely that the phyllospheric fungi present on deciduating leaves contribute to the aeromycota of urban areas.

Chapter 5 CORRESPONDENCE BETWEEN URBAN BIRD ROOSTS AND THE PRESENCE OF AEROSOLISED FUNGAL PATHOGENS

Peter J. Irga, Brigette Armstrong, William L. King, Margaret Burchett, Fraser R. Torpy

Statement of Joint Authorship

Peter Irga: Designed the study, analysed and interpreted the data, and is responsible for the content and writing of the manuscript.

Brigette Armstrong: Collected samples and assisted in the drafting of the manuscript.

William L. King: Collected samples and assisted in the drafting of the manuscript.

Margaret Burchett: Assisted in the drafting of the manuscript.

Fraser Torpy: Contributed to experimental design, advised on statistical analysis of data and assisted in the drafting of the manuscript.

Keywords

Airborne fungi; *Rhodotorula*; Avian droppings; Guano; Environmental sources; Zoonosis

5.1 Abstract

Habitat fragmentation in urban environments concentrates bird populations that have managed to adapt to these newly developed areas. Consequently, the roosts of these birds are potentially creating environments conducive to fungal growth and dissemination. Airborne fungi derived from these environments are relatively unstudied, as is the potential health risk arising from these fungi. This study documented the diversity of culturable airborne fungal propagules associated with forty urban bird roosts. Environmental variables from each site were recorded to allow us to analyse the correspondence between different bird species, the substrate they occupy and airborne fungal propagules. Associations were established between *Rhodotorula* and Pacific black ducks, wood ducks, myna birds and miner birds when in the presence of bare soil as a substrate. Further associations were established between *Penicillium*, *Scopulariopsis* and *Cunninghamella* and pigeons, sparrows and swallows living in areas with hard surfaces such as bitumen and rocks.

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Chapter 6 INDOOR AIR POLLUTANTS IN OCCUPATIONAL BUILDINGS IN A SUB-TROPICAL CLIMATE: COMPARISON AMONG VENTILATION TYPES

P.J. Irga and F.R. Torpy

Statement of Joint Authorship

Peter Irga: Designed the study, collected samples, analysed and interpreted the data, and is responsible for the content and writing of the manuscript.

Fraser Torpy: Contributed to experimental design, advised on statistical analysis of data and assisted in the drafting of the manuscript.

NB. Supplementary data displayed at end of Chapter.

6.1 Abstract

Few studies have concurrently assessed both abiotic and biotic air pollutants in the built environment in sub- tropical areas. The investigation comprised a field study of air pollutants in eleven indoor environments in Sydney throughout one year, to elucidate Indoor/Outdoor ratios of carbon dioxide, carbon monoxide, total volatile organic compounds, nitric oxide, nitrogen dioxide, sulfur dioxide, total suspended particulate matter, suspended particles <10 μm in diameter (PM_{10}) and particulate matter <2.5 μm ($PM_{2.5}$). Further, a concurrent assessment of airborne fungi was conducted along with the other air pollutants to determine their diversity and abundance for urban Sydney and to establish baseline Indoor/Outdoor ratios of airborne fungi. Building ventilation types were identified as natural, mechanical and mixed-type ventilation, to assess whether building ventilation type has an impact on prevalence and concentrations of indoor air pollutants. We found that generally the indoor air quality of a typical Australian office building is relatively good. The ventilation type of the buildings did affect indoor air quality; however not to the extent that occupant health was at risk in any case. Low concentrations of airborne fungi were encountered in samples, across all buildings and months, with naturally ventilated buildings having higher concentrations. Buildings with high airborne fungal concentrations also supported higher diversity of fungal species. Few organisms of concern to public health were identified. Significant differences were observed when comparing the structure of airborne fungal communities across building types, with buildings with centralised mechanical (air conditioning) systems harbouring different communities to the other ventilation types.

Irga, P. J., and F. R. Torpy (2016). Indoor air pollutants in occupational buildings in a sub-tropical climate: Comparison among ventilation types. Building and Environment 98:190-199.
View/Download from: Publisher's site

Chapter 7 GENERAL DISCUSSION

Air pollution in urban areas remains an increasingly important and un-resolved problem on a global scale. Although our understanding of the causes and effects of urban air pollution are increasing through time, there are still regions where well documented data on air pollution is scarce. Moreover, it is now well recognized that any level of pollution in the air is harmful for human health, increasing mortality and morbidity as well as lowering the general quality of life for populations worldwide.

The general aim of this thesis has been to investigate air pollutant concentrations across urban Sydney, Australia and to investigate the relationship urban forests have with the distribution, composition, and concentrations of those air pollutants. I conducted a series of descriptive studies and manipulative experiments across inner Sydney, which facilitated the quantitative assessment of the air pollutant mitigation properties of urban forests as well as its potential to directly and indirectly create air pollutants. Each of the five complementary investigations focused on specific questions relating to the type of air pollutants, spatiotemporal relationships with air pollutants, type of urban forests, biometeorological relationships, urban forests' detritus relationships, avian relationships, and indoor concentrations. The integration of these findings as a broader thesis provides a novel framework for understanding the important role that urban forests has in influencing the concentrations and composition of air pollutants within inner Sydney, Australia.

7.1 Spatiotemporal periodicity trends of air pollutants across urban Sydney

Prior to the current study, spatially representative data for airborne pollutant concentrations across inner urban Sydney was lacking within 15 km of the city's boundaries. Other monitoring sites do exist for Sydney, however they are located in the greater western metropolitan area of Sydney, and are not relevant for investigated area. Combined, these stations monitor PM_{10}, $PM_{2.5}$, CO, NO_2 and SO_2. The spatial coverage of these stations may be insufficient for providing air-quality information across inner Sydney, which could be subject to periodic low air quality, and cannot provide adequate data to allow the spatial interpolation of air pollutant patterns over all of urban Sydney.

Chapter 2 describes the spatiotemporal trends for a range of air pollutants across urban Sydney, Australia. Little seasonal variation was observed in particulate concentrations other than significantly higher concentrations of TSP and PM_{10} in September and May. The

differences were attributable to increased concentrations caused by prescribed hazard reduction burns that take place during that time in the year (Spring) (OEH 2014). Many rural centres across NSW experience an increase in PM levels during winter, when domestic wood burners are utilised, which add to the perennial vehicle- derived PM concentrations. For example, in our southern neighbouring state, Victoria, approximately 15% of households use firewood for primary or secondary heating (Driscoll et al 2000), significantly contributing to atmospheric particle concentrations in the airshed during winter. As evident from this study, domestic wood burners do not appear to be affecting the seasonal periodicity of PM concentrations for inner Sydney. However, samples were taken mid-week, during the day (between 1100 and 1400 h to avoid the daily peak traffic periods). Domestic wood burners would tend to be utilised at night-time, when temperatures are lower, thus it is possible that the sampling regime did not detect PM seasonality as affected by this potentially major source.

No seasonal trend in CO_2 was observed across the sampling area, with mean concentrations ranging from 377 ppm to 414 ppm. Seasonal atmospheric CO_2 fluctuations are observed all across the world, largely driven by terrestrial vegetation photosynthesis increases during Spring and Summer (Hellevang and Aagaard 2015). These fluctuations are greatest in the Northern Hemisphere, where there is greater variation in plant productivity between seasons. It is possible that the domes of high CO_2 levels that form over the cities' centre could have subsumed the effect of seasonality. Similarly, no seasonal trends in NO_2 were observed other than significantly higher concentrations in August and September which were once again attributable to the hazard reduction burns that take place during that period — wood smoke is rich in NO_x (Muala et al 2015) Amongst sites, the pattern of CO_2 concentrations was variable, the only statistically significant difference observed being Pitt St, Prince Alfred Park and Town Hall demonstrating significantly higher concentrations than those recorded for Sydney Park and Zetland. It is most likely that these differences were due to higher population density and traffic volume at the former sites. In urban environments, variations in ambient atmospheric CO_2 concentrations are mainly associated with combustion processes (Idso et al 2001), leading to peak CO_2 concentrations in the centre of cities. Whilst the city centre sites had high CO_2 concentrations, this pattern was not consistent across the full sample. Restrictions in the upward movement of pollutants released at ground level has been demonstrated in areas constricted with tall buildings; therefore, accumulation of pollutants may occur in built up areas (Gratani and Varone 2005). This may also explain the high inner

city CO_2 concentrations we detected on occasion. Higher wind speeds facilitate increased CO_2 advection in the atmosphere, which also could be a contributing factor to the reduced levels observed in less built up areas. Clearly, there are many challenges in relation to assessing urban CO_2 fluxes (Grimmond et al 2002), and further research on this air pollutant are needed.

7. 2 Sydney's urban forests ability to improve air quality

Higher concentrations of urban forests were associated with quantifiably lower concentrations of all fractions of PM; thus increasing the proportional area of greenspace was associated with decreasing particulate matter, despite the data being corrected for traffic density. The strongest association with decreasing PM was with increasing canopy coverage within 100 m radii of the sample sites. However, associations were also found between all fractions of particulate matter and environmental elements such as wind speed, time since the last rain event and the quantity of rain recorded over the period nearest to the samples being taken. These findings indicate, first, that within the urban areas of a city, localized differences in air pollutant loads occur due to meteorological conditions, as would be expected. Secondly, it appears that urban areas with proportionally higher concentrations of urban forests may experience better air quality with respect to reduced ambient particulate matter.

The statistical model developed in this experiment did not fully explain the variation in the air pollutant data sets, indicating that there were other variables not accounted for in the model, and that determining all, or even most of the causative factors associated with localized urban air pollution experiments can be challenging. There are clearly manifold environmental variables that influence air quality in a city environment at any one time, and in any specific location. Whilst the model developed here cannot account for the majority of the temporal and spatial variation in air quality with the environmental variables chosen for analysis in this study, and thus further research is needed to explore the range of additional factors that could be involved. The empirical identification of greenspace as an important determinant of city airborne pollutants is a significant contribution to our understanding of UAQ, and should assist in future air quality modelling exercises.

The results are in agreement with most of the studies that have assessed the ability of urban forests to reduce particulate concentrations, with Freiman et al (2006) finding that ambient PM concentrations were lower in neighbourhoods with dense vegetation, and Cohen et al (2014) demonstrating reduced pollutant concentrations including PM levels in an urban park

compared to proximal street canyons. However, whilst these studies demonstrated that urban vegetation may have benefits in regards to pollution mitigation, they lacked spatial or temporal replication, with Freiman et al's (2006) sampling limited to 6 sites; and the total study duration of Cohen et al (2014) being 6 days at 4 sites.

In the case of NO_2 and VOCs, contrary to the literature that utilises model estimates to demonstrate the potential of urban forests to mitigate high levels of these materials (e.g. Vos et al 2013), the results of the current study provided little evidence that urban trees remove significant amounts of these pollutants. Setälä et al (2013) documented slightly but often non-significantly lower concentrations of VOCs and NO_2 under tree canopies compared to adjacent open areas, suggesting that the role of urban forests in removing air pollutants is minor. PM reductions associated with vegetation in Setälä et al's (2013) study were more substantial, suggesting that the urban forests in both the current study and Setälä et al's work was not of sufficient density to ensure that measureable effects with VOCs and NO_2 could be detected. As it stands, current empirical evidence for ambient VOC and NO_2 reduction due to urban forests is insufficient to support this phenomenon, and the postulated role of urban vegetation in reducing local concentrations of VOCs and NO_2 may have been exaggerated (Pataki et al 2011). Future research must thus test a greater range of greenspace densities to determine whether the capacity for VOC and NO_2 mitigation is possible.

In any case, given the relationship detected between greenspace density and PM levels, I propose that if future urban greenspace developments are to have functions beyond improving the utility and attractiveness of an area (which are important functions in themselves), they must contain a sufficient density and extent of planted area using appropriate species (yet to be determined) to ensure measurable effects will occur.

7.3 Sydney's urban forests ability to compromise air quality

The benefits of urban forests notwithstanding, it is important to acknowledge that urban vegetation can also affect air quality negatively, including facilitating aerosolized pollen and fungal spores, with both allergenic and pathogenic potential and by emitting biogenic volatile organic compounds (BVOCs), which can eventually react to form ozone (Abelsohn et al 2002). As detailed above, urban forests were neither positively or negatively associated with ambient VOC concentrations in this study, thus any such effects within the study area were of a small magnitude.

Chapters 3, 4 and 5 investigated the influence of urban forests on aerosolised culturable fungal propagules, be that through density association, or indirectly through creating environments encouraging the proliferation of fungi through detritus deposition or providing a habitat for avian species that engender an environment in which fungi thrive.

When the spatiotemporal dynamics of Sydney's aeromycota was explored, the parkland sites at Centennial park and Newtown showed significantly higher mean numbers of fungal propagules indicating that greenspace, especially the grass component, could be a major determinant of the density of airborne fungi in urban areas. As hypothesised by Mitakakis et al (2001) and Hargreaves et al (2003), locally elevated mould spore densities in urban areas could be a result of proximal vegetation. In the current work, total fungal CFU, the number of fungal genera encountered, and the densities of some allergenic fungi including *Cladosporium*, *Alternaria* and *Epicoccum,* all showed positive correlations with greenspace density. One of the more noteworthy findings of this study is the finding that grass coverage consistently showed stronger associations with fungal density and diversity than with tree and shrub canopy coverage. It was previously unknown whether urban forests type would have a quantifiable influence on fungal behaviour. One potential reason why grass contributed to higher concentrations of fungal propagules might be related to saprobic colonization and sporulation of grass endophytic fungi may become unrestrained when their host plant tissue dies (Vázquez de Aldana et al 2013). Additionally, turf grass is regularly mowed in urban areas, which potentially aerosolises fungi (Streifel 1988). Additionally, the act of mowing grass which exposes plant tissues and nutrients could encourage fungal growth. Comtois et al (1995) demonstrated that the act of mowing lawns can aerosolize fungal spores, increasing the exposure of lawn mowing workers to aeroallergens. Similarly, previous work by Mitakakis et al (2001) showed the presence of agriculture and the harvesting of crops leads to increased aeromycota concentrations in nearby towns (Mitakakis et al 2001).

Increases in exposure to airborne fungal material are associated with increased allergenic symptoms, incidence of asthma and unscheduled hospital visitations (Pongracic et al 2010). In the current study, the highest concentrations of airborne fungi were observed in summer. Asthma hospitalisations tend to increase in summer periods (AIHW 2013), a pattern that is potentially related to increases in allergens at this time of year (Sharpe et al 2015), a proportion of which are thus likely to be fungal in origin. This trend is less distinct in northern Australia, where the seasonal differences are less defined (Sharpe et al 2015). Fortunately, relatively few human pathogens were identified in the survey. *Aspergillus* spp.

was present in 15.2 % of samples, however it accounted for less than 0.8 % of the total fungal propagules detected, which is comparable with the ambient background frequencies recorded throughout the year in New York (Recer et al 2001). *A. fumigatus* was found in less than 1.5% of samples taken, with a mean concentration of 0.8 CFU/m^3. Whilst the presence of this pathogenic species undoubtedly poses a risk for immunocompromised individuals no associations were made with urban greenspace, and the levels detected are considered safe for the general population.

As increases in exposure to airborne fungal material may have health implications, statistical models for the prediction of airborne fungal spore densities are invaluable. However, accurate assessment of the aeromycota is labour, time, cost, and training intensive. In the interests of practical industry application, an equation was derived in chapter 3 for the prediction of total concentration of airborne fungi encountered:

Equation:

Total airborne fungi (CFU/m^3) = 16.6a + 138b – 1.9c + 370.1

Where a = Percentage grass cover within 100 m radius, b = Wind speed (m/s), c = Total rainfall in the past month (mm).

The equation developed approximately predicts the levels of aeromycota for inner Sydney using a number of independent variables that can be quickly calculated without expensive, time-consuming methods. However, the model could be improved, with the three variable equation only explaining 54.87% of the variability in the fungal data. With so many potential influential factors determining airborne fungi concentrations, it is no surprise that research attempting to model airborne fungi do not find strong patterns. Nevertheless, health practitioners, occupational hygienists and aeromycologists may actively be able to predict airborne fungal concentrations through the utilisation of this equation.

The investigations reported in chapter 3 established that urban forests was associated with and increased density of aeromycota; however, it was clear that the relationships between the type of urban forests and the contribution to differences in fungal density in the atmosphere were not fully characterised by the data collected here. Thus in chapter 4, further experiments were developed to manipulatively investigate one component of this causal relationship, in

regards to senesced leaves acting as a substrate and reservoir for fungal growth. It is well known that fungi growing on plant material produce spores which may aerosolize and then colonise other plants (Brown and Hovmøller 2002), however the contribution of fungi from the phylloplane of senescent leaves to the spores present in the air has not previously been assessed. The current study is the first time that the contribution of fungi from the phylloplane of senescent leaves to the spores that are present in the air, and thus those that we *breathe*, has been assessed. The decay of senesced leaves may be of particular significance in an inner urban or city centre environment, where deciduous street trees could be the only substantial source of colonisable detritus in the area. The work revealed that, although differences were observed in the phyllospheric fungal genera presence and relative abundances amongst plant species, and different fungi proliferated under different humidity levels; few differences were found in the fungal spores that were aerosolized, with *Penicillium*, *Cladosporium*, *Alternaria*, *Chaetomium*, *Botrytis* and *Trichothecium* associated with all tree species. The most frequent genera detected in this study were similar to those reported in Chapter 3, during autumn in urban Sydney, with *Penicillium*, *Cladosporium* and *Alternaria* predominant. Furthermore, the community of aerosolized fungi in this study is similar in diversity and relative frequencies of species encountered in outdoor autumn samples reported by Torpy et al (2013). Few serious human pathogens were observed in concentrations so as to be of concern to health; although pathogens like *Aspergillus fumigatus* were present in a small number of samples. It is clear from this study that phyllospheric fungi present on deciduating leaves have the potential to contribute substantially to the aeromycota of urban areas during the season of Autumn, but are unlikely to be a significant source of pathogens apart from aeroallergenic fungi such as *Alternaria*.

Further exploration was undertaken to determine urban forests' potential to generate aerosolized fungal pathogens indirectly through facilitating habitat for avian zoonotic pathogens. Birds can be vectors of human fungal diseases, as well as creating a suitable environment for fungal pathogens to grow in their faecal matter, allowing the proliferation of pathogenic species, discussed in Chapter 1. This has the potential to result in a public health threat, especially if humans have increased exposure to these areas. Chapter 5 presents a comparison of the diversity of fungal propagules present in air samples taken from sites with high urban bird frequency, to sites with relatively lower bird frequency, with the aim to identify any correspondence between potential contributing environmental variables to the presence of fungal diversity that could pose a human health risk. Of the fungal species

present at sites with birds, known pathogens were identified, including *Mucor, Microsporum, Rhodotorula,* and *A. fumigatus*. However, despite rigorous exploration, no *Histoplasma capsulatum*, *Sporothrix schenckii*, *Candida albicans* or *Cryptococcus* spp. were identified in the investigation. Thus it is clear that wild birds harbour fungi and have the capacity to disseminate them throughout the environment. Many of these fungi may pose potential health risks for humans and represent a significant zoonotic concern, especially due to the vast distances birds cover on a daily basis between feeding and roosting areas. The roosts of these birds occur in areas of urban forests, which are commonly frequented by humans who are potentially at increased risk of contracting opportunistic diseases, especially if immunocompromised. It is thus a recommendation of this thesis that individuals who are at high risk of contracting mycotic infection, such as AIDS sufferers, transplant recipients and chemotherapy patients avoid areas with high bird density.

7.4 The indoor environment

Few previous studies have assessed air pollutants in the built environment in Sydney. It was hypothesised that the composition and density of the outdoor air pollutants detected in the outdoor sampling, work would lead to changes in the types and concentrations of pollutants in indoor air, although it was previously unknown if or how airborne pollutants will behave in response to variations in urban design and environmental variables. In parallel with the samples collected for Chapters 2 and 3, Chapter 6 represents a field study of air pollutants across eleven indoor environments in Sydney within the same calendar year. Analysis of this data elucidated the Indoor/Outdoor ratios of CO_2, CO, TVOCs, NO, NO_2, SO_2, TSP, PM_{10}, and $PM_{2.5}$. Additionally, an assessment of airborne fungi was conducted along with the other air pollutants, indoors and outside, to establish baseline Indoor/Outdoor ratios of airborne fungi. Building ventilation types were identified as natural, mechanical and mixed-type, to assess to what extent building ventilation systems might have an impact on prevalence and concentrations of indoor air pollutants. It was found that the indoor air quality of a typical Australian office building is unlikely to pose an immediate health issue to occupants. Further, although building ventilation types were found to influence indoor air pollutant composition, concentration ranges were not sufficient to put occupant health at risk. Naturally ventilated buildings tended to have higher I/O ratios in most instances. Significant differences were observed when comparing the composition of air pollutants across building types. Buildings with MVS tended to have lower PM and airborne fungi, while having higher CO_2. I/O ratios for PM rarely exceeded those encountered outdoors for MVS buildings, however NV and

CVS buildings I/O ratios were near parity. MVSs have the capacity to reduce PM penetration into the building as they filter the incoming air stream, while NVS and CVS air supply is through the building envelope and is not filtered. Consequently, NVS and CVS air pollutant concentrations resemble that of the incoming outdoor air supply. As discussed in Section 7.2 and 7.3, urban forests significantly affects the composition of air pollutants across inner Sydney. Thus, if NVS and CVS buildings are located in areas of high urban forest density; urban forests are likely to influence the composition of air pollutants within these buildings, both positively and negatively.

Chapter 8 RECOMMENDATIONS FOR FURTHER RESEARCH.

This section presents possible future research directions, with some additional comments on alternative methodologies which could be utilised in future work.

8.1 Greater temporal and spatial replication.

The investigations reported here has shown that areas with high densities of green plants have lower levels of ambient PM, and that this effect cannot be explained by variables other than those associated with the presence of the plants themselves. However, this research was constrained within the centre of Sydney, where greenspace is limited to a small number of extensively grassed parks and constrained areas with tree coverage. Further studies are required to expand the spatiotemporal scope of this work, along with testing areas with greater densities of tree canopy. With Australia's increasing population, carbon emissions and polluting technologies are still increasing in proportion, with a concomitant increase in poor air quality. Similarly, air pollutant concentrations are likely to increase, either through increases in load on power stations, increases in ozone and particulates due to higher temperatures and increased incidence of bushfires associated with climate change (Brown et al 2011). Urban consolidation and 'densification' (Abel 2010) is a concept proposed to contain these growing populations. Whilst higher density housing leads to less reliance on motor vehicles, the 'urban canyon' effect concentrates pollutants, and may prevent their dispersal (Salmond et al 2016). Alternately, the growth of many modern cities, including Sydney, is influenced by housing affordability, and thus the city grows at its fastest on the urban fringe. In Sydney's case, growth areas in western and south western suburbs could increase the exposure of the populace to air pollutants, as air pollution concentrates in these areas due to the topography of the Cumberland basin. As Sydney grows, there is an impending air quality crisis that is not currently being addressed, as extant air pollution laws are failing to mitigate the problem — with an estimated 1400 premature deaths from urban air pollution in Sydney alone (Department of Health 2009). While there is a general perception that plants and urban forests may be an effective mitigation method for most air pollutants, studies that provide empirical evidence for this phenomenon are scant, especially in regards to the density and type of plants involved. Further, variations in the type of urban

forests may lead to variations in the types of fungal spores released, some of which can illicit allergenic symptoms and initiate asthma attacks.

Thus, recommendations for future research derived from the main evaluation of the project findings of this work should focus on:

- Determining the influence of high densities, different structural arrangements and extent of urban vegetation on controlling a range of aspects of air pollutants across the greater Sydney basin, with emphasis on assessing the relationship between both density and type of greenspace and air pollution components.
- Calculating the net quantitative air pollutant attenuation level for Sydney's urban, peri-urban forests.
- Investigating whether urban vegetation barriers (e.g. hedges, street trees, grassed areas) can affect air pollution penetration inside buildings.
- Assessing and quantifying the emissions from a broader range of urban plants, including fungal particles, for their potential effects on human health.

8.2 Quantification and valuation of Sydney's urban forests' air pollutant abatement services – limitations and caveats

The general perception that plants and urban forests may be an effective tool in the mitigation of air pollutants requires further assessment, especially in regards to the effects of plant density and type of plant. Further, variations in the type of urban forests may lead to variations in plant sourced fungal spores that can illicit allergenic symptoms and initiate asthma attacks. In this study, only deciduous senescent leaves and bird roosts were assessed as a potential source for fungal propagules. Further research is needed to assess and quantify fungal emissions from other plant sources, be that from existing living plant biomass, or from other saprophytic material in the leaf litter (leaves from evergreen plants, bark, fallen flowers etc.). Consequently, evidence based dialogue incorporating both the advantages and the disadvantages of urban forests to urban air quality can be developed.

An increasingly utilised measure of urban forest ecosystem services is the Urban Forest Effects (UFORE) model developed by the U.S Forest Service (Nowak 2006). However, as detailed by Baró et al (2014), the model has limitations owing to the fact that the particulate matter deposition rates that form the foundation of the model have not been calculated for the

majority of the most prevalent plant species within Sydney, and the uncertainty related to both particle re-suspension rates and the fine scale spatial variability often observed in concentrations of air pollutants within urban areas. Baró et al (2014) concluded that the model should be used for approximate estimations rather than a precise quantification of ecosystem services. Although the dry deposition velocity of PM on urban forests was not documented in the current study, I did however find that areas with proportionally higher concentrations of urban forests were quantifiably associated with reduced ambient PM levels. Further work that details the deposition velocity of these particles may allow the UFORE estimates to be applied to Sydney. Additionally, the BVOCs emitted from Sydney's urban forests will differ to vegetation of other regions, and thus need to be characterised in order to be compared to estimate studies. Clearly, a similar approach to the research conducted here needs to be applied and replicated in other urban areas, to further test the identified relationship between both the density and type of greenspace and air pollution components. Similarly, the current methodology could be applied in peri and extra urban and rural areas within the wider Sydney region, facilitating modelling of the net quantitative air pollutant attenuation level for all of Sydney's urban forests.

In the current study, the volume of accumulative traffic movements was used as a measure of the primary local pollutant source. As vehicle derived air pollution varies between vehicle type, fuel type and vehicle age (Rhys-Tyler et al 2011), further work that takes into account the potential differences in vehicle pollution due to vehicle characteristics is required. Additionally, incorporation of variables such as the presence of traffic lights along with the continuity of traffic flow may also be of value, as vehicles can produce just as many if not more particulates when slowing down for traffic lights, as a result of tyre wear dust, brake pad dust, and road dust re-suspension (Grigoratos and Martini 2015).

8.3 Further characterisation of the dynamics of urban forests' ability to reduce air pollutants

The strategic use of urban vegetation plant barriers (including hedges, street trees, grassed areas) to reduce air pollution penetration inside buildings is a promising direction of research. Changes in PM_{10} concentrations have been documented in a row of roadside houses, after temporarily installing a line of young birch trees on the footpath (Maher et al 2013), with the observation that >50% reductions were recorded in PM levels inside houses screened by the temporary tree line. Electron microscopic analyses revealed that leaf-captured PM was

concentrated on trichomes on the leaf. Further manipulative experiments could be conducted, investigating air quality mitigation effects of the strategic implantation of vegetation barriers lining major traffic routes, and comparing plant barriers with conventional concrete sound barriers, that presumably do not have an equivalent PM collection efficiency.

8.4 Green infrastructure as a means of increasing urban forests

While it is desirable to implement urban forests by increasing tree canopy coverage for controlling air pollution across the city, the constraints of a highly developed area may make it difficult to plant sufficient numbers of trees in a densely populated city. Green roofs and green walls may be a solution to this dilemma (Byrne and Yang 2009; Matthews et al 2015), since they make use of walls and rooftops, the latter comprising 40–50% of the impermeable surface area in a city (Dunnett and Kingsbury, 2004). Currently, there are a limited number of studies on the air pollutant removal capacity of green roofs and green walls, which as of yet, do not provide adequate information to judge their effectiveness in air pollution control Yang et al 2008). As green roofs and green walls gain popularity, research into their ability to mitigate air pollutants will be of interest to the urban landscaper, atmospheric scientist, building occupants and the wider community.

8.5 The application of biotechnology to urban forestry to enhance air pollution mitigation

A promising avenue of research for the implementation of urban forestry into the built environment is the use of living/green wall biofilters that integrates the biofiltration and phytoremediation capacities of plants into building design, as a complement to the existing HVAC system. These systems filter air from the occupied space through the plant wall and then return clean air to the occupied space. Work developing, implementing and assessing the air pollutant removal potential of these systems may be a functional means of applying the air pollutant mitigation capacity of urban forestry to the indoor environment, whilst alleviating the space constraints of broad scale urban forests.

8.6 Alternative methodologies for determining particulate air pollutants.

Other sampling methodologies for PM estimation are available, which may increase the value of future research.

8.6.1 Gravimetric Analysis

Traditional monitoring networks for airborne particulate matter have employed filter-based samplers. While filter-based instruments have proven to be robust and accurate for the study of the detailed nature of airborne particles (Batterman et al 2012), they have several drawbacks when used in a routine monitoring network for regulatory purposes. The chief disadvantage associated with filter-based samplers for $PM_{2.5}$ is the fact that they do not provide information in real time. Often weeks months pass between the time when samples are collected and when $PM_{2.5}$ data becomes available (Chung et al 2001). The use of light scattering laser densitometry in the current project obviated the need for long sampling times, but was subject to the temporal vagaries of PM concentrations within the sampling periods. Using both methods simultaneously would provide valuable data on the comparability of the two alternative methods for sampling PM in urban conditions.

8.6.2 Tapered Element Oscillating Microbalance

Tapered Element Oscillating Microbalances (TEOM) measure suspended particulate matter concentrations by passing sample air and particles through a hollowed tapered channel where the particles are collected on a filter (Chung et al 2001). The tapered inlet tube oscillates at a frequency that is inversely proportional to the amount of sample deposited on the collection substrate (US EPA 2013). TEOMs are commonly used for routine monitoring purposes across Australia and are the standard method of measurement. This approach to air sampling has its limitations, mainly as the methodology does not account for diurnal variations of aerosol concentration due to the long sampling time required.

8.6.3 Geographical information systems

Advancement in satellite remote-sensing techniques allows the detection of air quality data over large spatial scales, when compared with surface instruments that only allow point observations. The sensor instruments provide high-quality aerosol information over the globe (Remer et al 2005). Measurement of aerosol optical thickness (AOT), a measure of columnar aerosol loading from the surface to the top of the atmosphere, can be determined with the use of these sensors. Correlations have been established with AOT and $PM_{2.5}$ mass concentration measured from ground based instruments, thus allowing estimation of $PM_{2.5}$ (Gupta and

Christopher 2009). This method thus allows the satellites to derive PM air quality information on the ground. However, ancillary information such as vertical distribution of aerosols and meteorological information is also needed to further refine the analysis (Gupta et al 2006). Further, satellite based sensors can only estimate daytime $PM_{2.5}$ on cloudless days (Christopher and Gupta 2010). Sydney experiences on average 105 cloud free days per year (BOM 2004). Incorporation of variables related to the shapes of buildings, urban vegetation and weather conditions, coupled with the data from ground based measurements, such as the data recorded in this project, would have the potential to improve our predictive capacity for particulate traffic pollutants concentration across the city.

8.6.4 Computational fluid dynamic modelling

One challenge in air pollution research is accurately mapping the movement of contaminants in regards to airflows and turbulence. Computational Fluid Dynamics (CFD) methods can account for these variables through the simulation of the movement of one substance through another, whilst accommodating the influence of complex solid structures, convection sources and meteorological changes, thus indicating the movement of pollutants from source to sink. CFD models have become an important tool in the field of air quality research, in both indoor and outdoor environments (Gao and Niu 2004). They are currently applied for investigating indoor airflow fields for building design and optimum ventilation and for pollutant dispersion in working areas for health and safety. However, few studies have combined theoretical (ie. modelling) and experimental methods to investigate air quality, in order to ground truth the model simulations. For indoor environments, pollutants are sometimes non-homogenously distributed throughout the room, thus ventilation effectiveness is the major indicator for the pollutant concentration at breathing zone. For outdoor environments, additional variables need to be taken into account, including topography, complex vegetation geometry, and boundary layer profiles. One of the biggest limitations in the utilisation of CFD models for outdoor pollutants is the use idealised buildings to model wind flow and pollution dispersion. Further development of CFD models that incorporate the use of real building and vegetation data, then validated with the sampling methodologies utilised in this study would vastly improve our knowledge on the movement of pollutants and the use of vegetation to remove them.

8.7 Alternative methodologies for the assessment of aeromycota

8.7.1 Quantification of bioaerosols

Various investigative techniques are available for aeromycological studies. However, future research should investigate a range of methods for the identification of fungal propagules concurrently with the methods utilised here, as no single method is currently available that can detect, identify and quantify all fungal species with accuracy. For example, in chapter 4, some genera were found in decaying street tree leaf samples that were not present in the corresponding air samples, including: *Mycosphaerella*, *Puccinia*, *Polythrincium*, *Ustilago*, *Pestalotiopsis* and *Geotrichum*. It would thus appear likely that some taxa eluded detection. Estimating the concentration of fungi in the atmosphere is commonly conducted using viable samplers or spore traps, as was the case here. With viable samplers, only viable propagules can be enumerated, whereas some other measures of sampling fungal spores can provide total propagule counts regardless of their viability. These may be of value in some health-related studies, as non-viable fungal materials are still capable of eliciting allergic responses.

Current viable sampling methods are based on three principles: impaction, filtration and electrostatic methods. Most instruments are based on sampling a fixed air-flow to collect a known volume of air. After collection, the samples can be analysed with microscopy, biochemical analysis or immunoassays.

8.7.2 Impactors and inertial sampling

Impactors and inertial samplers operate by drawing air through a nozzle or impeller, and forcing the jet of air to impact on a sampling surface which normally has some adhesive qualities, forcing airborne materials through the sampling surface boundary layer. This sampling surface may be a collection medium like agar, a coated microscope slide, filter or cellotape. The Reuter Centrifugal Sampler (RCS) used in this study and suction samplers such as Anderson samplers utilise this method. These samplers potentially underestimate the total airborne moulds spores as the device can only detect fungal spores that are able to be cultured on the medium used and will not detect non-viable spores, unculturable spores or spores that have specific nutrient requirements (Levetin 2004). However, these samplers are the most widely utilised in the literature and have been used extensively in previous research (eg. Hargreaves et al 2003; Bonetta et al 2010), allowing robust comparisons to be made amongst experiments that utilise common sampling methods. Results from the Anderson sampler and RCS are relatively interchangeable (An et al 2004), however the Burkard

sampler does not allow for the differentiation of many species, as spores are identified directly from the impaction surface which is error prone (many taxa produce identical spores) and requires vast operator experience (Levetin 2004).

8.7.3 Filtration samplers

Filter samplers collect particles of a size determined by the samplers' inlet characteristics and filter medium. Filter cassettes specifically developed for individual samplers are often used. Many types of filters can be used for bioaerosols, for example polycarbonate, polytetraflouroethylene (TEFLON), mixed cellulose ester (MCE), and gelatine filters. Porous membranes can be used for culturing and immunostaining. Teflon and polycarbonate filters are preferred for when the material needs to be washed from the filter to allow for culturing.

8.7.4 Electrostatic precipitation

Microorganisms in air can carry up to 10^4 elementary charges, which can be utilised for their collection by using electrostatic force (Mainelis et al 2001). Bioaerosols with sufficient charge (greater than 5 kV/cm) have collection efficiencies of 70% and 90% for indoor and outdoor samples, respectively (Xu et al 2012). This gives the potential to individually collect positive and negative charged bacteria and fungi, although it seems no commercially available bioaerosol collector/sampler has been yet designed utilising this sampling method at the time of writing.

8.7.5 Direct microscopic methods

Characterizing airborne fungi through the use of light microscopy is a technique that continues to be the primary method for the identification and quantification of fungal bioaerosols (Ellis et al 2007). This method is constrained by the viability of the fungal spores and the growth requirements of species, thus it has been postulated that only a small fraction of airborne microbes in a sample can be cultivated, resulting in numbers less than those determined by cultivation-independent methods (Douwes et al 2008). Furthermore, some organisms can only be identified to genus level, and some may remain morphologically indistinguishable under laboratory conditions as they do not produce fruiting bodies in culture — these organisms are traditionally termed *mycelia sterila* (Pitkäranta et al 2008).

The failure of the culture-dependent method used in the current study to detect non-culturable taxa was not considered a major issue, as all of the common airborne pathogens are known to grow on the culture medium used. Nonetheless, there are a range of biochemical and

molecular methods that have been developed in an attempt to alleviate problems associated with non-culturability, and while these methods do not allow for the determination of the diversity of fungal species detected, they do provide approximations on density.

8.7.6 Detection of β -N-acetylhexosaminidase (NAHA)

β -N-acetylhexosaminidase is an enzyme secreted by all members of the kingdom Eumycota. These chitin-degrading enzymes contribute to a number of morphogenic processes in filamentous fungi including spore germination, hyphal branch formation, and autolysis (Slámová et al 2010). Theoretically, the quantification of NAHA may be a very robust, alternative method for quantifying fungal spores (Allermann et al 2006).

8.7.7 Ergosterol

An often used biomarker for determining the fungal density is ergosterol, which is a biological precursor of Vitamin D2 found in cell membranes of fungi (Lau et al 2006). Ergosterol is almost exclusively found in fungi and is therefore appears to be a reasonably accurate biomarker in specific circumstances. Photochemical degradation can cause a significant decrease in the ergosterol content in living fungi, therefore it is necessary to avoid light exposure once the fungi are collected. As the molecule degrades rapidly after cell death, it has been suggesting that it could be useful as an indicator of living fungal biomass (Burshtein et al 2011). Gas Chromatography-Mass Spectrometry (GCMS) is the most efficient means for the quantification of ergosterol (Miller and Young 1997). However, the method is not without flaws; ergosterol concentrations are known to vary between cultures of the same species depending on the physiological state of the fungus, and unlike other biochemical markers and antigens derived from fungi (Lang-Yona et al 2012), ergosterol itself has not been associated with any adverse health outcomes. Furthermore, in order to get samples that provide adequate quantities of ergosterol, high volume samplers are required, with run times around 72 hours, thus making this method time constrained in some circumstances (Burshtein et al 2011; Lang-Yona et al 2012).

8.7.8 MVOCs

It has been proposed that microbial volatile organic compounds (MVOCs) derived from fungi could be used as a surrogate measurement of airborne fungi (Foruk et al 2001). These compounds usually lead to the subjective indication of fungal contamination in buildings, as they constitute the perceptible 'mouldy smell' released by secondary metabolites during

fungal growth. However, MVOCs are only present when there is active fungal growth. Consequently, the measurement of MVOC signature patterns may require high levels of fungal contamination (Horner and Miller 2003), and thus may not be an effective measure of ambient human exposure levels.

8.7.9 β glucan

Another nonspecific surrogate for airborne fungal density is the determination of β 1,3-D glucan, which, unlike ergosterol, is toxic, and thus of specific concern in indoor air. The measurement of β glucan has been shown to be a robust indicator for airborne fungi (Sander et al 2008), and has been strongly correlated with ergosterol concentration and visible mould damage (Foto et al 2005). As β glucan can elicit adverse health symptoms (Fogelmark et al 2001), it has the potential to be a more accurate measure of aerosolized fungal materials that is directly related to health, but is yet to find wide use in bioaerosol assessment.

8.7.10 Immunoassays

Fungal antigens can be used as markers of mould exposure (Prester, 2011). Increases in fungal spore germination rates have been shown to correlate with increases in airborne antigen concentrations for *Aspergillus fumigatus* antigen Asp f 1 (Sporik et al 1993), *Alternaria* allergen Alt a 1 (Mitakakis et al 2001) and other fungal aeroallergens (Green et al 2003). The measurement of fungal antigens is through the use of enzyme-linked immunosorbent assay (ELISA). Commercial ELISAs are available for specific antigens like Alt a 1 and Asp f 1 (Sporik et al 1993; Green et al 2005). Further work that explores the interaction between the treatments assessed in this work and the use of immunoassays would undeniably be of value.

8.7.11 Molecular genetic assays

Another alternative to sampling using culture based methods is through the use of quantitative polymerase chain reaction (qPCR) on air sampled material. qPCR has been proposed as a sensitive and convenient method to monitor indoor fungal concentrations (Norback and Cai 2011). For bioaerosol analysis, this method allows for the detection and enumeration of fungi independent of culturing, thereby circumventing the concerns in regards to viability of environmental microorganisms (Hospodsky et al 2010). qPCR may thus provide a more accurate estimation of the total concentrations of environmental fungi, in some cases being found to detect 1 to 2 orders of magnitude greater numbers of individual organisms than culture based methods used on the same samples (Yamamoto et al 2010).

However, non-culture based methods are prone to producing inflated estimates due to the amplification of nucleic acids from fungal cell-derived fragments present in the sample (Green et al 2005). This would likely annul the value of this method for the enumeration of aerosolized fungal cells, however this technique has become an invaluable tool for the hygiene scientist, as the viability of fungi is not essential for allergenicity, as any protein-based materials can act as allergens (Green et al 2005).

The use of qPCR for the quantitative assessment of aerosolized fungal cells suffers from several additional artefacts. The accuracy of qPCR can be influenced by a range of chemical inhibitors found in environmental samples (Haugland et al 2004). Furthermore, the detection of genomes by PCR-based techniques does not provide information about the infectivity of the pathogen, or the level of risk for the population (Girones et al 2010).

qPCR requires the optimisation of a range of conditions for the successful amplification and quantification of fungal DNA; for example the optimisation of the dilution ratio of the fungal DNA extract, which must be done in order to achieve an appropriate balance between PCR success ratios and detection ratios (Yamamoto et al 2010). There is no uniformity or standardisation in regards to this, so making comparisons across studies becomes difficult. If the standardisation of sample preparation efficiencies, requirements for sample replication, opportunities for improving precision, and approaches for estimating and meeting method detection limits can become standardised across studies, qPCR has the potential to be the most comprehensive assessment tool for aerosolized fungal cells.

9 CONCLUSIONS

The work presented here provides novel information on the relationships between a range of urban characteristics and the concentration and types of air pollutants in the air, with an emphasis on urban forests and greenspaces. The study also provides data on air pollutant concentrations, the origins and dissemination of air pollutants and some of the factors affecting our exposure to them within an urban context. Quantifiable differences in Sydney's urban air were observed at a very fine spatial scale, indicating that the air quality within this region is less homogenous than previously assumed. Most importantly, it was found that air samples taken from sites with less greenspace frequently had higher concentrations of all fractions of aerosolized particulates than other sites, whilst sites with high proximal greenspace had lower particulate levels, even when local vehicular traffic was taken into account. No observable trends in concentrations of NO, TVOC or SO_2 were observed, as recorded levels were generally very low across all sampled areas, indicating that Sydney has generally good air quality. The findings indicate, first, that within the urban areas of a city, localized differences in air pollutant loads will occur on a fine spatial scale. Secondly, it appears that urban areas with proportionally higher concentrations of urban forests may experience better air quality with regards to reduced ambient PM; however, conclusions about other abiotic air pollutants presented only weak relationships with the environmental variables I tested, probably due to their consistently low concentrations. These benefits notwithstanding, a caveat in regards to the implementation of urban forestry exists; the potential contribution urban forests to the production and dissemination of aerosolized fungal material has not previously been addressed. Thus, a comprehensive survey of the culturable aeromycota of Sydney was conducted by determining the diversity and abundance of outdoor fungal propagule concentrations for urban Sydney. Seasonal trends in airborne fungal density and diversity in urban Sydney were observed, with increases in concentrations experienced in the summer months. The environmental variables that had the largest detectable influence on the diversity and abundance of fungal propagule loads were temperature, wind speed and proximal greenspace. If the greenspace was comprised of grass, stronger associations with the aeromycota were observed, indicating that specific types of urban greenspace, whilst mitigating some air pollutants, will contribute to biological particulate air pollution. From this work, an equation was created to enable the prediction of the density of aerosolised culturable fungi, using environmental parameters that are much more convenient and easier to measure than the process of determining than the bioaerosols themselves. Aeromycota patterns were

further explored when the phyllospheric fungi present on abscised senescent autumn leaves from the five most prevalent deciduous street trees in the study area were identified. Differences were observed in phyllospheric fungal species presence and relative abundances across plant species, and certain fungi proliferated under different humidity levels; however, few differences were found among the fungal species that were aerosolized. Very few pathogenic species were observed from the sampled trees. The fungi identified were also common in the autumn air samples from the previous study, thus indicating that phyllospheric fungi present on deciduating leaves contribute to the gross aeromycota of urban areas. It is clear from this study that phyllospheric fungi on deciduating leaves have the potential to contribute greatly to the aeromycota of urban areas in seasons with substantial leaf fall. Another major potential source of airborne fungi in urban areas, which often interacts with urban forests, is urban bird colonies. The incidence of pathogenic fungi aerosolized from these sites – specifically *Rhodotorula* spp. and sporadic high concentrations of allergenic fungi such as *Aspergillus fumigatus*, were considered as a potential public health concern. Relationships were established between *Rhodotorula*, and three common urban bird species: Pacific black ducks, wood ducks, Indian mynas and Common miners. These results indicate possible health risks for sensitive individuals, and the roosts of urban bird colonies could present a risk of infection to those with a compromised immune system, or respiratory predisposing factors.

Contemporary urban populations spend the great majority of their time indoors, thus to link urban air pollution sources to potential health outcomes, indoor air pollutants were assessed. The concentrations of air pollutants across indoor office environments in Sydney were found to be below guideline maximum levels. The ventilation type of the buildings did affect indoor air quality; however not to the extent that occupant health was at risk. The findings indicate that if a building is to be constructed in a region of a city of similar structure, climate and ambient pollutant concentrations to Sydney, the use of naturally ventilated buildings and buildings with combined mechanical and natural ventilation could provide an indoor environmental quality of a sufficient standard, thus saving the major infrastructure and running costs associated with mechanical ventilating systems. However, it is advisable that a localized study should always be performed prior to implementation to ensure that unsafe conditions will not arise. Low concentrations of airborne fungi were encountered in indoor samples, across all sites and across months, with naturally ventilated buildings tending to have higher concentrations, with the aeromycota from outdoor sources identified in the

previous studies potentially penetrating into the building envelope. Buildings with high airborne fungal concentrations also supported higher diversity of fungal species. No organisms of concern to public health were identified.

From the results combined, it is clear that urban forests influence air pollutants substantially, either through the reduction of ambient particulate matter, or the facilitation of bioaerosols either directly or indirectly. The research methods developed here can be used for other field studies related to air pollutants, and that the information here not only contributes new valuable data air pollutants but also shows trends on possible sources and preventative mechanisms.

References

Abel, C. 2010. The vertical garden city: Towards new urban topology. Journal of Tall Buildings - Urban Habitat 2:20-25.

Agrios, G. N. 2005. Plant diseases caused by fungi. In: G. N. Agrios (ed.) Plant Pathology (Fifth Edition). p 385-614. Academic Press, San Diego.

AIHW. 2007. The burden of disease and injury in Australia 2003, AIHW, Canberra, Australia.

Alami, I., S. Mari, and A. Clérivet. 1998. A glycoprotein from Ceratocystis fimbriata f. sp. platani triggers phytoalexin synthesis in *Platanus × acerifolia* cell-suspension cultures. Phytochemistry 48: 771-776.

Alexopoulos, C. J., C. W. Mims, and M. Blackwell. 1996. Introductory Mycology. John Wiley & Sons, New York.

Alirol, E., L. Getaz, B. Stoll, F. Chappuis, and L. Loutan. 2011. Urbanisation and infectious diseases in a globalised world. The Lancet Infectious Diseases 11: 131-141.

Allermann, L., C. K. Wilkins, and A. M. Madsen. 2006. Inflammatory potency of dust from the indoor environment and correlation to content of NAGase and fungi. Toxicology In Vitro 20: 1522-1531.

An, H. R., G. Mainelis, and M. Yao. 2004. Evaluation of a high-volume portable bioaerosol sampler in laboratory and field environments. Indoor Air 14: 385–393.

Anderson, H. 2009. Air pollution and mortality: A history. Atmospheric Environment 43: 142-145.

Andersson-Sköld, Y., S. Thorsson, D. Rayner, F. Lindberg, S. Janhäll, A. Jonsson, U. Moback, R. Bergman, and M. Granberg. 2015. An integrated method for assessing climate-related risks and adaptation alternatives in urban areas. Climate Risk Management.

Andersson, M., S. Downs, T. Mitakakis, J. Leuppi, and G. Marks. 2003. Natural exposure to *Alternaria* spores induces allergic rhinitis symptoms in sensitized children. Pediatric Allergy and Immunology 14: 100-105.

Araujo, R., C. Pina-Vaz, and A. Rodrigues. 2010. Mould Infections: A Global Threat to Immunocompromised Patients. In: I. Ahmad, M. Owais, M. Shahid and F. Aqil (eds.) Combating Fungal Infections. p 1-19. Springer Berlin Heidelberg.

Aringoli, E.E., J.C. Basilico, R.L. Althaus, and J.C. Basilico. 2008. Multivariate analysis of fungal associations in the indoor air of Argentinean houses. International biodeterioration & biodegradation 62:281-286.

Arnold, A. E. 2007. Understanding the diversity of foliar endophytic fungi: progress, challenges, and frontiers. Fungal Biology Reviews 21: 51-66.

ASHRAE. 1992. Thermal Environmental Conditions for Human Occupancy No. 5. American Society of Heating, Refrigerating and Air-Conditioning Engineers Inc, Atlanta.

ASHRAE. 2011. GreenGuide: The design, construction, and operation of sustainable buildings, 3rd edition. http://www.ashraeregion12.org/?p=105 Accessed 15th of March 2012.

Avis, T. J. 2007. Antifungal compounds that target fungal membranes: applications in plant disease control. Canadian Journal of Plant Pathology 29: 323-329.

Banerjee, U. 2009. Opportunistic Pathogenic Yeasts. In: T. Satyanarayana and G. Kunze (eds.) Yeast Biotechnology: Diversity and Applications. p 215-236. Springer Netherlands, Dordrecht.

Baró, F., L. Chaparro, E. Gómez-Baggethun, J. Langemeyer, D.J. Nowak, and J. Terradas. 2014. Contribution of Ecosystem Services to Air Quality and Climate Change Mitigation Policies: The Case of Urban Forests in Barcelona, Spain. Ambio 43: 466-479.

Basilico, M., C. Chiericatti, E. E. Aringoli, R. L. Althaus, and J. C. Basilico. 2007. Influence of environmental factors on airborne fungi in houses of Santa Fe City, Argentina. Science of The Total Environment 376: 143-150.

Batterman, S., L. Du, G. Mentz, B. Mukherjee, E. Parker, C. Godwin, J.Y. Chin, A. O'Toole, T. Robins, Z. Rowe, and T. Lewis. 2012. Particulate matter concentrations in residences: an intervention study evaluating stand-alone filters and air conditioners. Indoor air 22: 235-252.

Beckett, P., P. Free-Smith, and G. Taylor. 2000. Effective tree species for local air-quality management. Journal of Arboriculture 26: 12-19.

Berger, C. L., W. G. Merz, and M. Silva-Hutner. 1973. Differences in the growth of *Aspergillus fumigatus* on cycloheximide media at three temperatures. Antimicrobial Agents and Chemotherapy 4: 388-391.

Biggane, R. P. J., and M. J. Gormally. 1994. The effect of dung beetle activity on the discharge of *Pilobolus* (Fungi, mucorales) sporangia in cattle, sheep and horse faeces. Entomophaga 39: 95-98.

Byrne, J., and J. Yang. 2009. Can urban greenspace combat climate change? Towards a subtropical cities research agenda. Australian Planner 46:36-43.

Bignell, E. 2014. 2 *Aspergillus fumigatus*: Saprotroph to Pathogen. In: O. Kurzai (ed.) Human Fungal Pathogens. p 19-43. Springer Berlin Heidelberg, Berlin, Heidelberg.

Blanco, J. L., and M. E. Garcia. 2008. Immune response to fungal infections. Veterinary immunology and immunopathology 125: 47-70.

Bonetta, S., S. Bonetta, S. Mosso, S. Sampo, and E. Carraro. 2010. Assessment of microbiological indoor air quality in an Italian office building equipped with an HVAC system. Environ Monit Assess 161: 473-483.

Brack, C. L. 2002. Pollution mitigation and carbon sequestration by an urban forest. Environmental Pollution 116, Supplement 1: S195-S200.

Brindle, R., N. Houghton, and G. Sheridan. 1999. Transport Generated Air Pollution and its Health Impacts. A Source Document for Local Government. In: R. R. A. 336. (ed.). ARRB Tranport Research, Vermont South, Victoria.

Broome, R.A., N. Fann, T.J.N. Cristina, C. Fulcher, H. Duc, and G.G. Morgan. 2015. The health benefits of reducing air pollution in Sydney, Australia. Environmental Research 143, Part A: 19-25.

Brown, J. K. M., and M. S. Hovmøller. 2002. Aerial Dispersal of Pathogens on the Global and Continental Scales and Its Impact on Plant Disease. Science 297: 537-541.

Brown, H., K. Proust, J. Spickett, and A. Capon. 2011. The potential role of health impact assessments in tackling the complexity for climate change adapatation for health Health promotion journal of Australia 22:S48-S53.

Brown, K.W., T. Minegishi, J.G. Allen, J.F. McCarthy, J.D. Spengler, and D.L. MacIntosh. 2014. Reducing patients' exposures to asthma and allergy triggers in their homes: an evaluation of effectiveness of grades of forced air ventilation filters. The Journal Of Asthma: Official Journal Of The Association For The Care Of Asthma 51: 585-594.

Brunekreef, B., and B. Forsberg. 2005. Epidemiological evidence of effects of coarse airborne particles on health. European Respiratory Journal 26: 309-318.

Bunse, T., and H. Merk. 1992. Mycological aspects of inhalative mould allergies. Mycoses 35: 61-66.

Bureau of Meteorology. 2014. Commonwealth of Australia. (ABN 92 637 533 532) (http://www.bom.gov.au/nsw/forecasts/sydney.shtml September 2013 and August 2014.

Burshtein, N., N. Lang-Yona, and Y. Rudich. 2011. Ergosterol, arabitol and mannitol as tracers for biogenic aerosols in the eastern Mediterranean. Atmospheric Chemistry & Physics 11: 829-839.

Burton, P. K., and C. H. Katelaris. 2014. Aerobiological survey of fungal spores in Sydney: seasonal distribution of *Didymella* and *Coprinus* spores. In: 10th International Congress of Aerobiology, Campbelltown, Australia

Bush, R. 2009. Fungal Allergy as Yet Unsolved. In: R. Pawankar, S. Holgate and L. Rosenwasser (eds.) Allergy Frontiers: Clinical Manifestations. Allergy Frontiers No. 3. p 471-485. Springer Japan.

Cafarchia, C., A. Camarda, D. Romito, M. Campolo, N.C. Quaglia, D. Tullio, and D. Otranto. 2006a. Occurrence of yeasts in cloacae of migratory birds. Mycopathologia 161: 229-234.

Cafarchia, C., D. Romito, R. Iatta, A. Camarda, M.T. Montagna, and D. Otranto. 2006b. Role of birds of prey as carriers and spreaders of Cryptococcus neoformans and other zoonotic yeasts. Medical Mycology 44: 485-492.

Crameri, R., M. Garbani, C. Rhyner, and C. Huitema. 2014. Fungi: the neglected allergenic sources. Allergy 69: 176-185.

Carvalho, J. C., M. T. Vilhena, and D. M. Moreira. 2007. Comparison between Eulerian and Lagrangian semi-analytical models to simulate the pollutant dispersion in the PBL. Applied Mathematical Modelling 31: 120-129.

Cavanagh, J., P. Zawar-Reza, and J. Wilson. 2009. Spatial attenuation of ambient particulate matter air pollution within an urbanised native forest patch. Urban Forestry & Urban Greening 8: 21-30.

Challoner, A., and L. Gill. 2014. Indoor/outdoor air pollution relationships in ten commercial buildings: $PM_{2.5}$ and NO_2. Building and Environment 80: 159-173.

Chao, H. J., J. Schwartz, D. K. Milton, and H. A. Burge. 2002. Populations and determinants of airborne fungi in large office buildings. Environmental Health Perspectives 110: 777.

Chatoutsidou, S.E., J. Ondráček, O. Tesar, K. Tørseth, V. Ždímal, and M. Lazaridis. 2015. Indoor/outdoor particulate matter number and mass concentration in modern offices. Building and Environment 92: 462-474.

Chance J. F. & Walsh J. J. 2006. Urban effects on native avifauna: a review. Landscape Urban Planning. 75: 143–154.

Chen, H., and M. S. Goldberg. 2009. The effects of outdoor air pollution on chronic illnesses. McGill Journal of Medicine : MJM 12: 58-64.

Chen, C., and B. Zhao. 2011. Review of relationship between indoor and outdoor particles: I/O ratio, infiltration factor and penetration factor. Atmospheric Environment 45: 275-288.

Chen, W., and C. Y. Jim. 2008. Assessment and valuation of the ecosystem services provided by urban forests. In: M. Carreiro, Y.-C. Song and J. Wu (eds.) Ecology, Planning, and Management of Urban Forests. p 53-83. Springer New York.

Cheng, M., I.E. Galbally, S.B. Molloy, P.W. Selleck, M.D. Keywood, S.J. Lawson, J.C. Powell, R.W. Gillett, and E. Dunne. 2016. Factors controlling volatile organic compounds in dwellings in Melbourne, Australia. Indoor Air 26: 219-230.

Chin, M., T. Diehl, H. Bian, and T. Kucsera. 2016. Aerosols in the Atmosphere: Sources, Transport, and Multi-decadal Trends. In: Air Pollution Modeling and its Application XXIV. G.D. Steyn and N. Chaumerliac, eds. Springer International Publishing, Cham. pp 3-10.

Christopher, S., and P. Gupta. 2010. Satellite Remote Sensing of Particulate Matter Air Quality: The Cloud-Cover Problem. Journal of the air & waste management association. 60:596-602.

Chung, A., D.P.Y. Chang, M.J. Kleeman, K.D. Perry, T.A. Cahill, D. Dutcher, E.M. McDougall, and K. Stroud. 2001. Comparison of Real-Time Instruments Used To Monitor Airborne Particulate Matter. Journal of the Air & Waste Management Association 51: 109-120.

City of Sydney. 2013. Urban Forest Strategy, 2013, The City Of Sydney, Sydney.

Clarke, K. 1993. Non-parametric multivariate analyses of changes in community structure. Australian Journal of Ecology 18: 117-143.

Cohen, P., O. Potchter, and I. Schnell. 2014. A methodological approach to the environmental quantitative assessment of urban parks. Applied Geography 48: 87-101.

Committee on the Assessment of Asthma and Indoor Air. 2000. Impact of ventilation and air cleaning on asthma. Clearing the Air: Asthma and Indoor Air Exposures. p 327-393. National Academies Press (US), Washington (DC).

Commonwealth Scientific and Industrial Research Organisation (CSIRO). 2010. Indoor air project Part 1: Main Report Indoor Air in Typical Australian Dwellings. A report to the Air Quality Section, Environment Standards Branch, Department of the Environment, Water, Heritage and the Arts. Commonwealth of Australia.

Cowan, M. M. 1999. Plant products as antimicrobial agents. Clinical Microbiology Review 12: 564-582.

Cragg, J., and Y. M. Clayton. 1971. Bacterial and fungal flora of seagull droppingsd in Jersey. Journal of Clinical Pathology 24: 317-319.

Crawford, J. A., P. F. Rosenbaum, S. E. Anagnost, A. Hunt, and J. L. Abraham. 2015. Indicators of airborne fungal concentrations in urban homes: Understanding the conditions that affect indoor fungal exposures. Science of The Total Environment 517: 113-124.

Crawford, J., A. Griffiths, D.D. Cohen, N.B. Jiang, and E. Stelcer. 2016. Particulate Pollution in the Sydney Region: Source Diagnostics and Synoptic Controls. Aerosol and Air Quality Research 16:1055-1066.

Crook, B., and N. C. Burton. 2010. Indoor moulds, Sick Building Syndrome and building related illness. Fungal Biology Reviews 24: 106-113.

Dart, A., and J. Thornburg. 2008. Collection efficiencies of bioaerosol impingers for virus-containing aerosols. Atmospheric Environment 42: 828-832.

Davis, R., C. Gole, and J. D. Roberts. 2013a. Impacts of urbanisation on the native avifauna of Perth, Western Australia. Urban Ecosyst 16: 427-452.

Davis, R. A., C. Gole, and J. D. Roberts. 2013b. Impacts of urbanisation on the native avifauna of Perth, Western Australia. Urban Ecosyst 16: 427-452.

Dejon, L., H. Mohammed, P. Du, C. Jacob, and A. Speicher. 2013. Synthesis of chromenoindole derivatives from *Robinia pseudoacacia*. MedChemComm 4: 1580-1583.

Denning, D. W., B. R. O'Driscoll, C. M. Hogaboam, P. Bowyer, and R. M. Niven. 2006. The link between fungi and severe asthma: a summary of the evidence. European Respiratory Journal 27: 615-626.

Department of Environment and Heritage. 2004. State of the air: National ambient air quality status and trends report 1991-2001, Canberra ACT.

Dharmage, S.C., M. Bailey, J. Raven, T. Mitakakis, F. Thien, A. Forbes, D. Guest, M.J. Abramson, and E. Walters. 1999. Prevalence and residential determinants of fungi

within homes in Melbourne, Australia. Clinical & Experimental Allergy 29: 1481-1489.

Dix, N. J., and J. Webster. 1995. Fungal Ecology. Chapman & Hall, London.

Dominici, F., L. Sheppard, and M. Clyde. 2003. Health Effects of Air Pollution: A Statistical Review. International Statistical Review 71: 243-276.

Douwes, J., W. Eduard, and P. S. Thorne. 2008. Bioaerosols. In: H. Editor-in-Chief: Kris (ed.) International Encyclopedia of Public Health. p 287-297. Academic Press, Oxford.

Douwes, J., P. Thorne, N. Pearce, and D. Heederik. 2003. Bioaerosol Health Effects and Exposure Assessment: Progress and Prospects. Annals of Occupational Hygiene 47: 187-200.

Driscoll, D., G. Milkovits, and D. Freudenberger. 2000. Impact and Use of Firewood in Australia, CSIRO.

Dunnett, NP., and Kingsbury N. 2004. Planting Green Roofs and Living Walls. Portland Oregan: Timber Press.

Ellis, D. H., S. Davis, H. Alexiou, R. Handke, and R. Bartley. 2007. Descriptions of Medical Fungi. 2nd ed. Women's and Children's Hospital, Adelaide, Australia.

Engvall, K., P. Wickman, and D. Norbäck. 2005. Sick building syndrome and perceived indoor environment in relation to energy saving by reduced ventilation flow during heating season: a 1 year intervention study in dwellings. Indoor Air 15: 120-126.

Environment Australia, E. 2003. BTEX *Personal Exposure Monitoring in Four Australian Cities,* Technical Paper No. 6. In: E. Australia (ed.), Canberra, ACT, Australia.

Erdmann, C. A., and M. G. Apte. 2004. Mucous membrane and lower respiratory building related symptoms in relation to indoor carbon dioxide concentrations in the 100-building BASE dataset. Indoor Air 14: 127-134.

Fang, G.-C., Y.-S. Wu, C.-C. Wen, W.-J. Lee, and S.-Y. Chang. 2007. Influence of meteorological parameters on particulates and atmospheric pollutants at taichung harbor sampling site. Environ Monit Assess 128: 259-275.

Figdor, C. G., Y. van Kooyk, and G. J. Adema. 2002. C-type lectin receptors on dendritic cells and Langerhans cells. Nature Reviews. Immunology 2: 77-84.

Fischer, R., and U. Kues. 2006. Asexual sporulation in mycelial fungi the mycota growth, differentiation and sexuality. Springer-Verlag, Berlin.

Fisk, W. J., A. G. Mirer, and M. J. Mendell. 2009. Quantitative relationship of sick building syndrome symptoms with ventilation rates. Indoor Air 19: 159-165.

Fogelmark, B., J. Thorn, and R. Rylander. 2001. Inhalation of (1,3)-beta-D-glucan causes airway eosinophilia. Mediators of Inflammation 10: 13-19.

Fonseca, Á., and J. Inácio. 2006. Phylloplane Yeasts. In: G. Péter and C. Rosa (eds.) Biodiversity and Ecophysiology of Yeasts. The Yeast Handbook. p 263-301. Springer Berlin Heidelberg.

Foruk, M. J., S. Uhlman, D. B. Baker, and H. Yang. 2001. Analysis of microbial contamination of a ventilation system detected by measurement of microbial volatile organic compounds. In: E. Johanning (ed.) Bioaerosols, Fungi and Mycotoxins: Health Effects, Assessment, Prevention and Control. Boyd Printing, Albany, New York.

Foto, M., L.L.P. Vrijmoed, J.D. Miller, K. Ruest, M. Lawton, and R.E. Dales. 2005. A comparison of airborne ergosterol, glucan and Air-O-Cell data in relation to physical assessments of mold damage and some other parameters. Indoor Air 15.

Freiman, M. T., N. Hirshel, and D. M. Broday. 2006. Urban-scale variability of ambient particulate matter attributes. Atmospheric Environment 40: 5670-5684.

Frey, D., and E. B. Durie. 1960. The incidence of airbourne fungus in Sydney. Mycopathologia 13: 93-99.

Friend, A. J., G. A. Ayoko, and S. Kokot. 2013. Source apportionment of airborne particulate matter: An overview of Australian and New Zealand studies. Air Quality & Climate Change 47: 13-19.

Gange, A. C., S. Dey, A. F. Currie, and B. C. Sutton. 2007. Site- and species-specific differences in endophyte occurrence in two herbaceous plants. Journal of Ecology 95: 614-622.

Garden, J., Mcalpine, C., Peterson, A., Jones, D., and Possingham, H. 2006. Review of the ecology of Australian urban fauna: A focus on spatially explicit processes. Austral Ecology 31: 126–148

Gardy, J. L., D. J. Lynn, F. S. L. Brinkman, and R. E. W. Hancock. 2009. Enabling a systems biology approach to immunology: focus on innate immunity. Trends in Immunology 30: 249-262.

Garrett, M., B. Hooper, F. Cole, and M. Hooper. 1997. Airborne fungal spores in 80 homes in the Latrobe Valley, Australia: levels, seasonality and indoor-outdoor relationship. Aerobiologia 13: 121-126.

Garrett, M. H., M. A. Hooper, B. M. Hooper, and M. J. Abramson. 1998. Respiratory Symptoms in Children and Indoor Exposure to Nitrogen Dioxide and Gas Stoves. American Journal of Respiratory and Critical Care Medicine 158: 891-895.

Gaskin, S., M. Taylor, R. Bentham, and D. Pisaniello. 2012. Understanding and managing risks associated with fungal contamination in indoor environments Final Report, The University of Adelaide, Adelaide.

Gao, N., and J. Niu. 2004. CFD study on mocro-environment around human body and personalized ventilation. . Building and Environment 39: 795–805.

Gilbert, G. S., and C. O. Webb. 2007. Phylogenetic signal in plant pathogen–host range. Proceedings of the National Academy of Sciences 104: 4979-4983.

Girones, R., M.A. Ferrús, J.L. Alonso, J. Rodriguez-Manzano, B. Calgua, A. de Abreu Corrêa, A. Hundesa, A. Carratala, and S. Bofill-Mas. 2010. Molecular detection of pathogens in water – The pros and cons of molecular techniques. Water Research 44: 4325-4339.

Gómez de Ana. S, Torres-Rodríguez, E. Ramírez, S. García, and J. Belmonte-Soler. 2006. Seasonal Distribution of *Alternaria*, *Aspergillus*, *Cladosporium* and *Penicillium* Species Isolated in Homes of Fungal Allergic Patients. The Journal of Investigational Allergology & Clinical Immunology 16: 357-363.

Gots, R. E., N. J. Layton, and S. W. Pirages. 2003. Indoor Health: Background Levels of Fungi. AIHA Journal 64: 427-438.

Gratani, L., and L. Larone. 2005. Daily and seasonal variation of CO_2 in the city of Rome in relationship with the traffic volume. Atmospheric Environment 39: 2619-2624.

Gratani, L., and L. Varone. 2014. Atmospheric carbon dioxide concentration variations in Rome: relationship with traffic level and urban park size. Urban Ecosyst 17: 501-511.

Green, B. J., T. O'Meara, J. K. Sercombe, and E. R. Tovey. 2006. Measurement of personal exposure to outdoor aeromycota in northern New South Wales Australia. Annals of Agricultural and Environmental Medicine 13: 225-234.

Green, B. J., J. K. Sercombe, and E. R. Tovey. 2005. Fungal fragments and undocumented conidia function as new aeroallergen sources. The Journal of Allergy and clinical Immunology 115: 1043-1048.

Green, C., P. Scarpino, and S. Gibbs. 2003. Assessment and modeling of indoor fungal and bacterial bioaerosol concentrations. Aerobiologia 19: 159-169.

Grigoratos, T., and G. Martini. 2015. Brake wear particle emissions: a review. Environmental Science and Pollution Research 22: 2491-2504.

Grimmond, C. S. B., T. S. King, F. D. Cropley, D. J. Nowak, and C. Souch. 2002. Local-scale fluxes of carbon dioxide in urban environments: methodological challenges and results from Chicago. Environmental Pollution 116, Supplement 1: S243-S254.

Grinn-Gofroń, A., and B. Bosiacka. 2015. Effects of meteorological factors on the composition of selected fungal spores in the air. Aerobiologia 31: 63-72.

Grundström, M., and H. Pleijel. 2014. Limited effect of urban tree vegetation on NO_2 and O_3 concentrations near a traffic route. Environmental Pollution 189: 73-76.

Guarnieri, M., and J. R. Balmes. 2014. Outdoor air pollution and asthma. Lancet 383: 1581-1592.

Gupta, P., S. A. Christopher, M. A. Box, and G. P. Box. 2007. Multi year satellite remote sensing of particulate matter air quality over Sydney, Australia. International Journal of Remote Sensing 28: 4483-4498.

Gupta, P., and S. Christopher. 2009. Particulate matter air quality assessment using integrated surface, satellite, and meteorological products: Multiple regression approach. Journal of Geophysical Research:114, D14205.

Gurjar, B., L. Molina, and C. Ojha. 2010. Air Pollution: Health and Environmental Impacts. CRC press, Florida, USA.

Haas, D., J. Habib, J. Luxner, H. Galler, G. Zarfel, R. Schlacher, H. Friedl, and F.F. Reinthaler. 2014. Comparison of background levels of culturable fungal spore concentrations in indoor and outdoor air in southeastern Austria. Atmospheric Environment 98: 640-647.

Hargreaves, M., S. Parappukkaran, L. Morawska, J. Hitchins, C. He, and D. Gilbert. 2003. A pilot investigation into associations between indoor airborne fungal and non-biological particle concentrations in residential houses in Brisbane, Australia. Science of The Total Environment 312: 89-101.

Haugland, R. A., M. Varma, J. Wymer, and S. J. Vesper. 2004. Quantitative PCR of selected *Aspergillus*, *Penicillium* and *Paecilomyces* species. Systematic and Applied Microbiology 27: 198-210.

He, C., L. Morawska, and L. Taplin. 2007. Particle Emission Characteristics of Office Printers. Environmental Science & Technology 41: 6039-6045.

Hedge, A. 2009. Indoor Work and Living Environments: Health, Safety and Peformance Indoor Air Quality, Health and Productivity. p 247-267. Science publishers Inc, Ithaca, New York.

Heitman, J. 2011. Microbial pathogens in the fungal kingdom. Fungal Biology Reviews 25: 48-60.

Heitman, J., J. E. Edwards, S. G. Filler, and A. P. Mitchell. 2006. Molecular Principles of Fungal Pathogenesis. ASM Press, Washington, D. C.

Hellevang, H., and P. Aagaard.2015. Constraints on natural global atmospheric CO2 fluxes from 1860 to 2010 using a simplified explicit forward model. Scientific Reports 5:17352.

Henninger, S., and W. Kuttler. 2010. Near surface carbon dioxide within the urban area of Essen, Germany. Physics and Chemistry of the Earth, Parts A/B/C 35: 76-84.

Hoff, G. L., and W. J. Bigler. 1981. The role of bats in the propagation and spread of histoplasmosis: a review. Journal of Wildlife Diseases 17: 191-196.

Horner, W. E., C. Barnes, R. Codina, and E. Levetin. 2008. Guide for interpreting reports from inspections/investigations of indoor mold. Journal of Allergy and Clinical Immunology 121: 592-597.e597.

Horner, W. E., and J. D. Miller. 2003. Microbial volatile organic compounds with emphasis on those arising from filamentous fungal contaminants of buildings. ASHRAE Transactions 109: 215-231.

Hospodsky, D., N. Yamamoto, and J. Peccia. 2010. Accuracy, precision, and method detection limits of quantitative PCR for airborne bacteria and fungi. Applied Environmental Microbiology 76: 7004-7012.

Hubalek, Z. 1994. Pathogenic microorganisms associated with free-living birds (a review). Acta Sc. Nat. Brno 28: 1-74.

Hubálek, Z. 2004. An annotated checklist of pathogenic microorganisms associated with migratory birds. Journal of Wildlife Diseases 40: 639-659.

Hunter, C. A., C. Grant, B. Flannigan, and A. F. Bravery. 1988. Mould in buildings: the air spora of domestic dwellings. International Biodeterioration 24: 81-101.

Idso, C. D., S. B. Idso, and R. C. Balling Jr. 2001. An intensive two-week study of an urban CO_2 dome in Phoenix, Arizona, USA. Atmospheric Environment 35: 995-1000.

Inácio, J., P. Pereira, M. Carvalho, Á. Fonseca, M.T. Amaral-Collaço, and I. Spencer-Martins. 2002. Estimation and Diversity of Phylloplane Mycobiota on Selected Plants in a Mediterranean–Type Ecosystem in Portugal. Microb Ecol 44: 344-353.

IHME. 2015. Institute for Health Metrics and Evaluation (IHME), University of Washington. Air Pollution. Available at: ⟨http://www.healthdata.org/air-pollution⟩ , (accessed 20.10.15).

Irga, P. J., F. R. Torpy, and M. D. Burchett. 2014. Ecological determinants of the aeromycota in urban Sydney. In: 10th International Congress of Aerobiology, Campbelltown, Australia

Irga, P.J., M.D. Burchett, and F.R. Torpy. 2015. Does urban forestry have a quantitative effect on ambient air quality in an urban environment? Atmospheric Environment 120:173-181.

Irga, P.J., and F. Torpy. 2015. A survey of the aeromycota of Sydney and its correspondence with environmental conditions: grass as a component of urban forestry could be a major determinant. Aerobiologia: 1-15.

Irga, P.J., M.D. Burchett, G. O'Reilly, and F.R. Torpy. 2016a. Assessing the contribution of fallen autumn leaves to airborne fungi in an urban environment. Urban Ecosyst:1-14.

Irga, P.J., B. Armstrong, W. King, M.D. Burchett, and FR Torpy (2016) Correspondence between urban bird roosts and the presence of aerosolised fungal pathogens. Mycopathologia. 1-11.

Irga, P.J., and F.R. Torpy. 2016. Indoor air pollutants in occupational buildings in a sub-tropical climate: Comparison among ventilation types. Building and Environment. 98: 190-199.

Ismail, N., and M. McGinnis. 2008. Fungal Infections. In: A. Gaspari and S. Tyring (eds.) Clinical and Basic Immunodermatology. p 373-410. Springer London.

Janhäll, S. 2015. Review on urban vegetation and particle air pollution – Deposition and dispersion. Atmospheric Environment 105: 130-137.

Janhäll, S. 2015. The effect of vegetation on air quality. Swedish National Road and Transport Research Institute (VTI). VTI report 876.ISSN:0347-6030

Jerrett, M. et al. 2004. A review and evaluation of intraurban air pollution exposure models. J Expo Anal Environ Epidemiol 15: 185-204.

Jones, A. M., and R. M. Harrison. 2004. The effects of meteorological factors on atmospheric bioaerosol concentrations—a review. Science of The Total Environment 326: 151-180.

Kauffman, C. A. 2001. Pulmonary histoplasmosis. Current Infectious Disease Reports 3: 279-285.

Kauserud, H., M. Lie, Ø. Stensrud, and M. Ohlson. 2005. Molecular characterization of airborne fungal spores in boreal forests of contrasting human disturbance. Mycologia 97: 1215-1224.

Kembel, S. W., and R. C. Mueller. 2014. Plant traits and taxonomy drive host associations in tropical phyllosphere fungal communities. Botany 92: 303-311.

Kemp, P. C., H. G. Neumeister-Kemp, B. Esposito, G. Lysek, and F. Murray. 2003. Changes in Airborne Fungi from the Outdoors to Indoor Air; Large HVAC Systems in Nonproblem Buildings in Two Different Climates. AIHA Journal 64: 269-275.

Kenworthy, J., and F. Laube. 2002. Urban transport patterns in a global sample of cities and their linkages to transport infrastructures, land use, economics and environment. World Transport Policy and Practice 8: 5-20.

Kilbert, C. J. 2012. Sustainable Construction: Green Building Design and Delivery. 3rd ed. John Wiley & Sons, Inc, Hoboken, New Jersey.

Kindt, T. J., B. A. Osborne, and R. A. Goldsby. 2007. Kuby Immunology. 6 ed. W.H Freeman Publishers.

Kingham, S., M. Durand, T. Aberkane, J. Harrison, J. Gaines Wilson, and M. Epton. 2006. Winter comparison of TEOM, MiniVol and DustTrak PM10 monitors in a woodsmoke environment. Atmospheric Environment 40: 338-347.

Kishimoto, R. A., and G. E. Baker. 1969. Pathogenic and potentially pathogenic fungi isolated from beach sands and selected soils of Oahu, Hawaii. Mycologia: 537-548.

Klich, M. A., and J. I. Pitt. 1988. A Laboratory Guide to Common Aspergillus Species and their Teleomorphs. CSIRO, Sydney australia.

Kornartit, C., R. S. Sokhi, M. A. Burton, and K. Ravindra. 2010. Activity pattern and personal exposure to nitrogen dioxide in indoor and outdoor microenvironments. Environment International 36: 36-45.

Korniłłowicz-Kowalska, T., and I. Kitowski. 2013. *Aspergillus fumigatus* and other thermophilic fungi in nests of wetland birds. Mycopathologia 175: 43-56.

Fulleringer, S.L., D. Seguin, S. Warin, A. Bezille, C. Desterque, P. Arné, R. Chermette, S. Bretagne, and J. Guillot. 2006. Evolution of the environmental contamination by thermophilic fungi in a turkey confinement house in France. Poultry Science 85: 1875-1880.

Ladds, P. 2009. Pathology of Australian Native Wildlife. CSIRO Publishing Collingwood, Victoria Australia.

Lai, H.K., L. Bayer-Oglesby, R. Colvile, T. Götschi, M.J. Jantunen, N. Künzli, E. Kulinskaya, C. Schweizer, and M.J. Nieuwenhuijsen. 2006. Determinants of indoor air concentrations of PM2.5, black smoke and NO_2 in six European cities (EXPOLIS study). Atmospheric Environment 40: 1299-1313.

Lang-Yona, N., K. Dannemiller, N. Yamamoto, N. Burshtein, J. Peccia, O. Yarden, Y. Rudich, and R. MacKenzie. 2012. Annual distribution of allergenic fungal spores in atmospheric particulate matter in the Eastern Mediterranean; a comparative study between ergosterol and quantitative PCR analysis. Atmospheric Chemistry & Physics 12: 2681-2690.

Larone, D. H. 2002. Medically Important Fungi: A Guide to Identification,. 4th ed. American Society for Microbiology Press, Washington D.C.

Lau, A., A. Y. Lee, C. K. Chan, and M. Fang. 2006. Ergosterol as a biomarker for the quantification of the fungal biomass in atmospheric aerosols. Atmospheric Environment 40: 249-259.

Lazaridis, M. 2011. Atmospheric aerosols first principles of meteorology and air pollution. environmental pollution No. 19. p 169-199. Springer Netherlands.

Łaźniewska, J., V. K. Macioszek, and A. K. Kononowicz. 2012. Plant-fungus interface: The role of surface structures in plant resistance and susceptibility to pathogenic fungi. Physiological and Molecular Plant Pathology 78: 24-30.

Lee, T. et al. 2006. Culturability and concentration of indoor and outdoor airborne fungi in six single-family homes. Atmos Environ (1994) 40: 2902-2910.

Leelőssy, Á., F. Molnár, F. Izsák, Á. Havasi, I. Lagzi, and R. Mészáros. 2014. Dispersion modeling of air pollutants in the atmosphere: a review. Central European Journal of Geosciences 6: 257-278.

Leppä, J., Anttila, T., Kerminen, V.-M., Kulmala, M., and Lehtinen, K. E. J. 2011. Atmospheric new particle formation: real and apparent growth of neutral and charged particles, Atmos. Chem. Phys 11: 4939-4955

Levetin, E. 2004. Methods for aeroallergen sampling. Current Allergy and Asthma Reports 4: 376-383.

Levetin, E., and K. Dorsey. 2006. Contribution of leaf surface fungi to the air spora. Aerobiologia 22: 3-12.

Lin, W.-H., and C.-S. Li. 2000. Associations of Fungal Aerosols, Air Pollutants, and Meteorological Factors. Aerosol Science and Technology 32: 359-368.

Liu, K. Y., and A. Baskaran. 2005. Using garden roof systems to achieve sustainable building envelopes. Construction Technology Update. National Research Council of Canada: 1-6.

Lowe, K., C. Taylor, and R. Major. 2011. Do Common Mynas significantly compete with native birds in urban environments? Journal of Ornithology 152: 909-921.

Lugarini, C., C. Goebel, L. Condas, M. Muro, M. de Farias, F. Ferreira, and M. Vainstein. 2008. *Cryptococcus neoformans* isolated from passerine and psittacine bird excreta in the state of Paraná, Brazil. Mycopathologia 166: 61-69.

Macher, J., J. Douwes, B. Prezant, and T. Reponen. 2012. Bioaerosols. In: Aerosols Handbook. CRC Press. pp 285-344.

Mainelis, G., K. Willeke, P. Baron, T. Reponen, S.A. Grinshpun, R.L. Górny, and S. Trakumas. 2001. Electrical charges on airborne microorganisms. Journal of Aerosol Science 32: 1087-1110.

Marzluff J. M. and Ewing K. 2001. Restoration of fragmented landscapes for the conservation of birds: a general frame- work and specific recommendations for urbanizing land- scapes. Restoration Ecology. 9, 280–92.

Maschmeyer, G., A. Haas, and O. A. Cornely. 2007. Invasive aspergillosis: epidemiology, diagnosis and management in immunocompromised patients. Drugs 67: 1567-1601.

Matthews, T., A.Y. Lo, and J.A. Byrne. 2015. Reconceptualizing green infrastructure for climate change adaptation: Barriers to adoption and drivers for uptake by spatial planners. Landscape and Urban Planning 138:155-163.

McDonald-Gibson, J., A. Brammer, C. Davidson, T. Folley, F. Launay, and J. Thomsen. 2013. Environmental Burden of Disease Assessment. Springer+Business Media Dordecht.

Meadow, J.F., A.E. Altrichter, S.W. Kembel, J. Kline, G. Mhuireach, M. Moriyama, D. Northcutt, T.K. O'Connor, A.M. Womack, G.Z. Brown, J.L. Green, and B.J.M. Bohannan. 2014. Indoor airborne bacterial communities are influenced by ventilation, occupancy, and outdoor air source. Indoor Air 24: 41-48.

Meffert, P. J., and F. Dziock. 2013. The influence of urbanisation on diversity and trait composition of birds. Landscape Ecology 28: 943-957.

Meklin, T., T. Reponen, C. McKinstry, S.-H. Cho, S.A. Grinshpun, A. Nevalainen, A. Vepsäläinen, R.A. Haugland, G. LeMasters, and S.J. Vesper. 2007. Comparison of

mold concentrations quantified by MSQPCR in indoor and outdoor air sampled simultaneously. The Science of the total environment 382: 130-134.

Mendell, M. J., A. G. Mirer, K. Cheung, M. Tong, and J. Douwes. 2011. Respiratory and allergic health effects of dampness, mold, and dampness related agents: A Review of the Epidemiologic Evidence. Environmental Health Perspectives 119: 748-756.

Mentese, S., M. Arisoy, A. Y. Rad, and G. Güllü. 2009. Bacteria and Fungi Levels in Various Indoor and Outdoor Environments in Ankara, Turkey. CLEAN - Soil, Air, Water 37: 487-493.

Meyer, H.W., H. Wurtz, P. Suadicani, O. Valbjorn, T. Sigsgaard, and F. Gyntelberg. 2004. Molds in floor dust and building-related symptoms in adolescent school children. Indoor Air 14: 65-72.

Millar, G., T. Abel, J. Allen, P. Barn, M. Noullett, J. Spagnol, and P.L. Jackson. 2010. Evaluating Human Exposure to Fine Particulate Matter Part I: Measurements. Geography Compass 4: 281-302.

Miller, J. D., and J. C. Young. 1997. The use of ergosterol to measure exposure to fungal propagules in indoor air. American industrial hygiene association journal 58: 39-43.

Mitakakis, T., and D. Guest. 2001. A fungal spore calendar for the atmosphere of Melbourne, Australia, for the year 1993. Aerobiologia 17: 171-176.

Mitakakis, T. Z., A. Clift, and P. A. McGee. 2001. The effect of local cropping activities and weather on the airborne concentration of allergenic *Alternaria* spores in rural Australia. Grana 40: 230-239.

Mitakakis, T. Z., E. R. Tovey, W. Xuan, and G. B. Marks. 2000. Personal exposure to allergenic pollen and mould spores in inland New South Wales, Australia. Clinical & Experimental Allergy 30: 1733-1739.

Mitchell, B. 2013. Building Materials Can Be a Major Source of Indoor Air Pollution. Occupational Health & Safety 82:62-64.

Molloy, S. B. et al. 2012. Indoor air quality in typical temperate zone Australian dwellings. Atmospheric Environment 54: 400-407.

Moore, D., and L. Novak-Frazer. 2002. Why Study the Genomes of Fungi? Essential Fungal Genetics. p 1-25. Springer New York, New York, NY.

Morawska, L., C. He, J. Hitchins, D. Gilbert, and S. Parappukkaran. 2001. The relationship between indoor and outdoor airborne particles in the residential environment. Atmospheric Environment 35: 3463-3473.

Morey, P. R., G. N. Crawford, and R. B. Rottersman. 2001. Indoor Air Quality in Nonindustrial Occupational Environments Patty's Industrial Hygiene. John Wiley & Sons, Inc.

Morris, C.E., D.C. Sands, M. Bardin, R. Jaenicke, B. Vogel, C. Leyronas, P.A. Ariya, and R. Psenner. 2008. Microbiology and atmospheric processes: an upcoming era of research on bio-meteorology. Biogeosciences Discuss. 5: 191-212.

Muala, A., G. Rankin, M. Sehlstedt, J. Unosson, J.A. Bosson, A. Behndig, J. Pourazar, R. Nyström, E. Pettersson, C. Bergvall, R. Westerholm, P.I. Jalava, M.S. Happo, O. Uski, M.-R. Hirvonen, F.J. Kelly, I.S. Mudway, A. Blomberg, C. Boman, and T. Sandström. 2015. Acute exposure to wood smoke from incomplete combustion - indications of cytotoxicity. Particle and Fibre Toxicology 12: 33.

Mycology Online, University of Adelaide. 2014. Fungal Descriptions. http://www.mycology.adelaide.edu.au/ Accessed August 2013 – August 2015.

Naima, B., H. Henrietta, H. Greg, K. Robie, K. Andrew, and M. Virginia. 2012. Essentials of Environmental Public Health ScienceA Handbook for Field Professionals. 'Oxford University Press', Oxford, UK.

Nardoni, S., F. Mancianti, M. Sgorbini, F. Taccini, and M. Corazza. 2005. Identification and seasonal distribution of airborne fungi in three horse stables in Italy. Mycopathologia 160: 29-34.

Nathanson, T. 1995. Report 93- EHD-66 Health Canada, Ottawa, Ontario, Canada.

National Environment Protection Authority. 2008. Biogenic and Geogenic Emissions: Results, Environment Protection Authority, Sydney.

New South Wales Department of Health, 2009. Air Pollution Health Risks. http;//www.health.nsw.gov.au/PublicHealth/environment/air/air_pollution.asp Accessed 21st of November 2011.

New South Wales Environment Protection Agency. 2007. Approved Methods for the Sampling and Analysis of Air Pollutants in New South Wales, NSW Environment Protection Authority (EPA), Sydney

Newton, P. 2001. Human Settlements, Australian State of the Environment Report 2001 (Theme Report). CSIRO Publishing on behalf of the Department of the Environment and Heritage, Canberra, Australia.

Nieguitsila, A., P. Arné, B. Durand, M. Deville, H. Benoît-Valiergue, R. Chermette, S. Cottenot-Latouche, and J. Guillot. 2011. Relative efficiencies of two air sampling methods and three culture conditions for the assessment of airborne culturable fungi in a poultry farmhouse in France. Environmental Research 111: 248-253.

Norback, D., and G.-H. Cai. 2011. Fungal DNA in hotel rooms in Europe and Asia—associations with latitude, precipitation, building data, room characteristics and hotel ranking Journal of Environmental Monitoring 13: 2895-2903.

OECD. 2014. The Cost of Air Pollution Health Impacts of Road Transport, OECD Publishing.

OEH. 2015. New South Wales Air Quality Statement 2014, The State of NSW and Office of Environment and Heritage, Sydney.

Office of Legislative Drafting, A.-G. s. D., Canberra. 1994. National Environment Protection (Ambient Air Quality) Measure. In: N. E. P. C. A. 1994 (ed.).

Oliveira, M., H. Ribeiro, J. L. Delgado, and I. Abreu. 2009. The effects of meteorological factors on airborne fungal spore concentration in two areas differing in urbanisation level. International Journal of Biometeorology 53: 61-73.

Osono, T. 2006. Role of phyllosphere fungi of forest trees in the development of decomposer fungal communities and decomposition processes of leaf litter. Canadian Journal of Microbiology 52: 701-716.

Osono, T., and D. Hirose. 2009. Ecology of Endophytic Fungi Associated with Leaf Litter Decomposition. In: M. Rai and P. D. Bridge (eds.) Applied Mycology. 92-109. Cambridge, CABI Publishing, Wallingford, UK.

Osono, T., and A. Mori. 2004. Distribution of phyllosphere fungi within the canopy of giant dogwood. Mycoscience 45: 161-168.

Ospelt, C., and S. Gay. 2010. TLRs and chronic inflammation. The International Journal of Biochemistry & Cell Biology 42: 495-505.

Ozgok-Kangal, K., İ. Arziman, G. Uzun, and Ş. Yildiz. Carbon monoxide poisoning in the workplace: A hidden danger. Apollo Medicine.

Paker, Y., Y. Yom-Tov, T. Alon-Mozes, and A. Barnea. 2014. The effect of plant richness and urban garden structure on bird species richness, diversity and community structure'. Landscape and Urban Planning 122: 186-195.

Palm, N. W., and R. Medzhitov. 2009. Pattern recognition receptors and control of adaptive immunity. Immunological Reviews 227: 221-233.

Parat, S., A. Perdrix, H. Fricker-Hidalgo, I. Saude, R. Grillot, and P. Baconnier. 1997. Multivariate analysis comparing microbial air content of an air-conditioned building

and a naturally ventilated building over one year. Atmospheric Environment 31: 441-449.

Park, J. S., N. Y. Jee, and J. W. Jeong. 2014. Effects of types of ventilation system on indoor particle concentrations in residential buildings. Indoor Air 24: 629-638.

Pataki, D.E., M.M. Carreiro, J. Cherrier, N.E. Grulke, V. Jennings, S. Pincetl, R.V. Pouyat, T.H. Whitlow, and W.C. Zipperer. 2011. Coupling biogeochemical cycles in urban environments: ecosystem services, green solutions, and misconceptions. Frontiers in Ecology and the Environment 9: 27-36.

Pearce, D.A., P.D. Bridge, K.A. Hughes, B. Sattler, R. Psenner, and N.J. Russell. 2009. Microorganisms in the atmosphere over Antarctica. FEMS Microbiology Ecology 69: 143-157.

Pepeljnjak, S., and M. Šegvić. 2003. Occurrence of fungi in air and on plants in vegetation of different climatic regions in Croatia. Aerobiologia 19: 11-19.

Pfaller, M. A., P. G. Pappas, and J. R. Wingard. 2006. Invasive fungal pathogens: Current epidemiological trends. Clinical Infectious Diseases 43: S3-S14.

Phattaralerphong, J., Sathornkich, and H. Sinoquet. 2006. A photographic gap fraction method for estimating leaf area of isolated trees: assessment with 3D digitized plants. Tree Physiology 26: 1123-1136.

Philip, S., R.V. Martin, A. van Donkelaar, J.W.-H. Lo, Y. Wang, D. Chen, L. Zhang, P.S. Kasibhatla, S. Wang, Q. Zhang, Z. Lu, D.G. Streets, S. Bittman, and D.J. Macdonald. 2014. Global chemical composition of ambient fine particulate matter for exposure assessment. Environmental Science & Technology 48: 13060-13068.

Pilotto, L. S., R. M. Douglas, R. G. Attewell, and S. R. Wilson. 1997. Respiratory effects associated with indoor nitrogen dioxide exposure in children. International Journal of Epidemiology 26: 788-796.

Pitkäranta, M., T. Meklin, A. Hyvärinen, L. Paulin, P. Auvinen, A. Nevalainen, and H. Rintala. 2008. Analysis of Fungal Flora in Indoor Dust by Ribosomal DNA Sequence Analysis, Quantitative PCR, and Culture. Applied and Environmental Microbiology 74: 233-244.

Powe, N.A., and K.G. 2004. Willis Mortality and morbidity benefits of air pollution (SO_2 and PM_{10}) absorption attributable to woodland in Britain. Journal of Environmental Management 70:119-128.

Prester, L. 2011. Indoor exposure to mould allergens. Archives of Industrial Hygiene and Toxicology 62: 371-380.

Pugh, T. A. M., A. R. MacKenzie, J. D. Whyatt, and C. N. Hewitt. 2012. Effectiveness of Green Infrastructure for Improvement of Air Quality in Urban Street Canyons. Environmental Science & Technology 46: 7692-7699.

Rao, C. Y., H. A. Burge, and J. C. S. Chang. 1996. Review of quantitative standards and guidelines for fungi in indoor air. Journal of the Air & Waste Management Association 46: 899-908.

Recio, M., M. Trigo, S. Docampo, M. Melgar, J. García-Sánchez, L. Bootello, and B. Cabezudo. 2012. Analysis of the predicting variables for daily and weekly fluctuations of two airborne fungal spores: *Alternaria* and *Cladosporium*. International Journal of Biometeorology 56: 983-991.

Reiss, E., H. J. Shadomy, and G. M. Lyon. 2011. Cryptococcosis fundamental medical mycology. p 303-332. John Wiley & Sons, Inc.

Reponen, T., and B. J. Green. 2012. Microbial bioaerosols in the occupational environment: Exposure, Detection, and Disease. In: E. Bingham and B. Cohrssen (eds.) Patty's Toxicology No. 6th. p 499-533. John Wiley & Sons, Inc, New York.

Rhys-Tyler, G. A., W. Legassick, and M. C. Bell. 2011. The significance of vehicle emissions standards for levels of exhaust pollution from light vehicles in an urban area. Atmospheric Environment 45: 3286-3293.

Robine, E., I. Lacaze, S. Moularat, M. Boissier, and S. Ritoux. 2006. Mesure des aérosols fongiques dans l'air : utilisation de l'ergostérol. Revue Française d'Allergologie et d'Immunologie Clinique 46: 174-179.

Roy, S., J. Byrne, and C. Pickering. 2012. A systematic quantitative review of urban tree benefits, costs, and assessment methods across cities in different climatic zones. Urban Forestry & Urban Greening 11:351-363.

Rutherford, S., J. A. K. Owen, and R. W. Simpson. 1997. Survey of airspora in Brisbane, Queensland, Australia. Grana 36: 114-121.

Sabariego, S., C. Díaz de la Guardia, and F. Alba. 2000. The effect of meteorological factors on the daily variation of airborne fungal spores in Granada (southern Spain). International Journal of Biometeorology 44: 1-5.

Sæbø, A., R. Popek, B. Nawrot, H.M. Hanslin, H. Gawronska, and S.W. Gawronski. 2012. Plant species differences in particulate matter accumulation on leaf surfaces. Science of The Total Environment 427–428: 347-354.

Safe Work Australia. 2011. Managing the Work Environment and Facilities: Code of Practice. http://www.safeworkaustralia.gov.au/ Accessed 16/01 2013.

Safe Work Australia. 2012. Guidance on the interpretation of workplace exposure standards for airborne contaminants.

Saliba, N. A., F. El Jam, G. El Tayar, W. Obeid, and M. Roumie. 2010. Origin and variability of particulate matter (PM_{10} and $PM_{2.5}$) mass concentrations over an Eastern Mediterranean city. Atmospheric Research 97: 106-114.

Salmond, J. A. et al. 2013. The influence of vegetation on the horizontal and vertical distribution of pollutants in a street canyon. Science of The Total Environment 443: 287-298.

Salmond, J. A. et al. 2016. Health and climate related ecosystem services provided by street trees in the urban environment. Environmental Health 15: 95-111.

Salonen, H., C. Duchaine, M. Mazaheri, S. Clifford, and L. Morawska. 2015. Airborne culturable fungi in naturally ventilated primary school environments in a subtropical climate. Atmospheric Environment 106: 412-418.

Samson, P. 1988. Atmospheric Transport and Dispersion of Air Pollutants Associated with Vehicular Emissions. In: Air Pollution, the Automobile, and Public Health. A. Watson, R. Bates and D. Kennedy, eds. National Academic Press, Washington DC.

Sander, I., C. Fleischer, G. Borowitzki, T. Bruning, and M. Raulf-Heimsoth. 2008. Development of a two-site enzyme immunoassay based on monoclonal antibodies to measure airborne expsure to (1,3)-beta-D-glucan. Journal of Immunological Methods 337: 55-62.

Schulz, B., and C. Boyle. 2005. The endophytic continuum. Mycological Research 109: 661-686.

Schulz, B., A.-K. Römmert, U. Dammann, H.-J. Aust, and D. Strack. 1999. The endophyte-host interaction: a balanced antagonism? Mycological Research 103: 1275-1283.

Schwarz, J., G. L. Baum, C. J. K. Wang, E. L. Bingham, and H. Rubel. 1957. Successful infection of pigeons and chickens with *Histoplasma capsulatum*. Mycopathologia et mycologia applicata 8: 189-193.

Seigneur, C., and R. Dennis. 2011. Atmospheric Modelling. In: G. Hidy, J. Brook, K. Demerjian, L. Molina, W. Pennell and R. Scheffe. (eds.) Technical challenges of multipollutant air quality management. Springer Science+Business Media B.V.

Seow, W.J., G.S. Downward, H. Wei, N. Rothman, B. Reiss, J. Xu, B.A. Bassig, J. Li, J. He, H.D. Hosgood, G. Wu, R.S. Chapman, L. Tian, F. Wei, N.E. Caporaso, R. Vermeulen, and Q. Lan. 2015. Indoor concentrations of nitrogen dioxide and sulfur dioxide from burning solid fuels for cooking and heating in Yunnan Province, China. Indoor Air. *Epub ahead of print.*

Setälä, H., V. Viippola, A.-L. Rantalainen, A. Pennanen, and V. Yli-Pelkonen. 2013. Does urban vegetation mitigate air pollution in northern conditions? Environmental Pollution 183: 104-112.

Shaw, R. H. 2007. Observation and simulation of flow in vegetation canopies. In: Y. A. Gayev and J. C. R. Hunt (eds.) Flow and Transport Processes with Complex Obstructions: Applications to Cities, Vegetative Canopies, and Industry. p 179-198. Springer Netherlands, Dordrecht.

Shelton, B. G., K. H. Kirland, W. Flanders, and G. Morris. 2002. Profiles of Airbourne Fungi in Buildings and Outdoor Environments in the Unites States. Applied and Environmental Microbiology 68: 1743-1753.

Shepherd, C. J., and C. J. Totterdell. 1988. Mushrooms and toadstools of Australia. Inkata Press, Melbourne.

Simeray, J., J.-P. Chaumont, and D. Léger. 1993. Seasonal variations in the airborne fungal spore population of the east of France (Franche - Comté). Comparison between urban and rural environment during two years. Aerobiologia 9: 201-206.

Simoni, M., I. Annesi-Maesano, T. Sigsgaard, D. Norback, G. Wieslander, W. Nystad, M. Canciani, P. Sestini, and G. Viegi. 2010. School air quality related to dry cough, rhinitis and nasal patency in children. European Respiratory Journal 35: 742-749.

Simoni, M., S. Baldacci, S. Maio, S. Cerrai, G. Sarno, and G. Viegi. 2015. Adverse effects of outdoor pollution in the elderly. Journal of Thoracic Disease 7: 34-45.

Slámová, K., P. Bojarová, L. Petrásková, and V. Křen. 2010. β-N-Acetylhexosaminidase: What's in a name…? Biotechnology Advances 28: 682-693.

Smith, C. M., and S. H. Kagan. 2009. Prevention of systemic mycoses by reducing exposure to fungal pathogens in hospitalised and ambulatory neutropenic patients. Oncology Nursing Forum 32: 565-579.

Soltani, M., M. Bayat, S. J. Hashemi, M. Zia, and N. Pestechian. 2013. Isolation of *Cryptococcus neoformans* and other opportunistic fungi from pigeon droppings. Journal of research in medical sciences: the official journal of Isfahan University of Medical Sciences 18: 56.

Soreanu, G., M. Dixon, and A. Darlington. 2013. Botanical biofiltration of indoor gaseous pollutants – A mini-review. Chemical Engineering Journal 229:585-594.

Sowiak, M., K. Bródka, A. Kozajda, A. Buczyńska, and I. Szadkowska-Stańczyk. 2012. Fungal aerosol in the process of poultry breeding--quantitative and qualitative analysis. Medycyna Pracy 63: 1-10.

Spickett, J., D. Katscherian, and P. Harris. 2013. The role of Health Impact Assessment in the setting of air quality standards: An Australian perspective. Environmental Impact Assessment Review 43:97-103.

Spitaler, R., S. Gurschler, E. Ellmerer, B. Schubert, M. Sgarbossa, and C. Zidorn. 2009. Flavonoids from *Celtis australis* (Cannabaceae). Biochemical Systematics and Ecology 37: 120-121.

Sporik, R. B., L. K. Arruda, J. Woodfolk, M. D. Chapman, and R. B. Platts-Mills. 1993. Environmental exposure to Aspergillus fumigatus allergen (Asp f 1). Cinical & Experimental Allergy 23: 326-331.

Staab, J. F., and B. Wong. 2009. Fungal Infections, Systemic. In: M. Schaechter (ed.) Eukaryotic Microbes. Academic Press, san Diego, Ca.

Stenkat, J., M. E. Krautwald-Junghanns, and V. Schmidt. 2013. Causes of Morbidity and Mortality in Free-Living Birds in an Urban Environment in Germany. EcoHealth 10: 352-365.

Stennett, P. J., and P. J. Beggs. 2004. *Alternaria* spores in the atmosphere of Sydney, Australia, and relationships with meteorological factors. International Journal of Biometeorology 49: 98-105.

Sturm, R. 2012. Modeling the deposition of bioaerosols with variable size and shape in the human respiratory tract – A review. Journal of Advanced Research 3: 295-304.

Sugar, A. M. 1991. Overview: Cryptococcosis in the patient with AIDS. Mycopathologia 114: 153-157.

Sundell, J., H. Levin, W.W. Nazaroff, W.S. Cain, W.J. Fisk, D.T. Grimsrud, F. Gyntelberg, Y. Li, A.K. Persily, A.C. Pickering, J.M. Samet, J.D. Spengler, S.T. Taylor, and C.J. Weschler. 2011. Ventilation rates and health: multidisciplinary review of the scientific literature. Indoor Air 21: 191-204.

Sweeney, K. 2013. Personal conversation, 5th of September 2013.

Takeda, M., Y. Saijo, M. Yuasa, A. Kanazawa, A. Araki, and R. Kishi. 2009. Relationship between sick building syndrome and indoor environmental factors in newly built Japanese dwellings. Int Arch Occup Environ Health 82: 583-593.

Tallis, M., G. Taylor, D. Sinnett, and P. Freer-Smith. 2011. Estimating the removal of atmospheric particulate pollution by the urban tree canopy of London, under current and future environments. Landscape and Urban Planning 103: 129-138.

Tampieri, M. P. 2006. Mycetes and urban areas. Parassitologia 48: 121-124.

Taylor, L., C. Taylor, and A. Davis. 2013. The impact of urbanisation on avian species: The inextricable link between people and birds. Urban Ecosyst 16: 481-498.

Tham, R., S.C. Dharmage, P.E. Taylor, C.H. Katelaris, D. Vicendese, M.J. Abramson, and B. Erbas. 2014. Outdoor fungi and child asthma health service attendances. Pediatric Allergy and Immunology 25: 429-449.

Thevenet, F., L. Sivachandiran, O. Guaitella, C. Barakat, and A. Rousseau. 2014. Plasma–catalyst coupling for volatile organic compound removal and indoor air treatment: a review. Journal of Physics D: Applied Physics 47:224011.

Thurston, G. D. 2008. Outdoor air pollution: sources, atmospheric transport, and human health effects. In: H. Editor-in-Chief: Kris (ed.) International encyclopedia of public health. p 700-712. Academic Press, Oxford.

Tippayawong, N., P. Khuntong, C. Nitatwichit, Y. Khunatorn, and C. Tantakitti. 2009. Indoor/outdoor relationships of size-resolved particle concentrations in naturally ventilated school environments. Building and Environment 44: 188-197.

Torpy, F., P. Irga, J. Brennan, and M. Burchett. 2013. Do indoor plants contribute to the aeromycota in city buildings? Aerobiologia 29: 321-331.

Torpy, F.R., P.J. Irga, and M.D. Burchett. 2015 Reducing Indoor Air Pollutants Through Biotechnology. In: Biotechnologies and Biomimetics for Civil Engineering. F. Pacheco Torgal, J.A. Labrincha, M.V. Diamanti, C.P. Yu and H.K. Lee, eds. Springer International Publishing. pp 181-210.

Tsiodras, S., T. Kelesidis, I. Kelesidis, U. Bauchinger, and M. E. Falagas. 2008. Human infections associated with wild birds. Journal of Infection 56: 83-98.

US EPA. 2013. Ambient air quality monitoring program. Quality Assurance Handbook for Air Pollution Measurement Systems. Office of Air Quality Planning and Standards Air Quality Assessment Division RTP, NC 2771

van Heezik, Y., A. Smyth, and R. Mathieu. 2008. Diversity of native and exotic birds across an urban gradient in a New Zealand city. Landscape and Urban Planning 87: 223-232.

Vega, F.E., A. Simpkins, M.C. Aime, F. Posada, S.W. Peterson, S.A. Rehner, F. Infante, A. Castillo, and A.E. Arnold. 2010. Fungal endophyte diversity in coffee plants from Colombia, Hawai'i, Mexico and Puerto Rico. Fungal Ecology 3: 122-138.

Verma, A., M. Wüthrich, G. Deepe, and B. Klein. 2015. Adaptive Immunity to Fungi. Cold Spring Harbor Perspectives in Medicine 5.

Vicedo-Cabrera, A., A. Biggeri, L. Grisotto, F. Barbone, and D. Catelan. 2013. A Bayesian kriging model for estimating residential exposure to air pollution of children living in a high-risk area in Italy. Geospatial Health 8:87-95.

Viegas, C., J. Malta-Vacas, R. Sabino, S. Viegas, and C. Veríssimo. 2014. Accessing indoor fungal contamination using conventional and molecular methods in Portuguese poultries. Environ Monit Assess 186: 1951-1959.

Visagie, C.M., J. Houbraken, J.C. Frisvad, S.B. Hong, C.H.W. Klaassen, G. Perrone, K.A. Seifert, J. Varga, T. Yaguchi, and R.A. Samson. 2014. Identification and nomenclature of the genus Penicillium. Studies in Mycology 78: 343-371.

Vorholt, J. A. 2012. Microbial life in the phyllosphere. Nat Rev Micro 10: 828-840.

Vos, P. E. J., B. Maiheu, J. Vankerkom, and S. Janssen. 2013. Improving local air quality in cities: To tree or not to tree? Environmental Pollution 183: 113-122.

Walker, G. M., and N. A. White. 2005. Introduction to Fungal Physiology Fungi. p 1-34. John Wiley & Sons, Ltd.

Wang-Li, L., Q. Li, and G. E. Byfield. 2013. Identification of bioaerosols released from an egg production facility in the Southeast United States. Environmental Engineering Science 30: 2-10.

Wang, B.-L., T. Takigawa, Y. Yamasaki, N. Sakano, D.-H. Wang, and K. Ogino Symptom. 2008. Symptom definitions for SBS (sick building syndrome) in residential dwellings. International Journal of Hygiene and Environmental Health 211: 114-120.

Wania, A., M. Bruse, N. Blond, and C. Weber. 2012. Analysing the influence of different street vegetation on traffic-induced particle dispersion using microscale simulations. Journal of Environmental Management 94: 91-101.

Whittington, A., N.A.R. Gow, and B. Hube From commensal to pathogen: *Candida Albicans*. In: Human Fungal Pathogens, 2nd ed. O. Kurzai, ed. Springer-Verlag, Berlin-Heidelberg. 2014.

Whipps, J. M., P. Hand, D. Pink, and G. D. Bending. 2008. Phyllosphere microbiology with special reference to diversity and plant genotype. Journal of Applied Microbiology 105: 1744-1755.

Wittmaack, K., H. Wehnes, U. Heinzmann, and R. Agerer. 2005. An overview on bioaerosols viewed by scanning electron microscopy. Science of The Total Environment 346: 244-255.

World Health Organisation (WHO). 1990. Indoor air quality: Biological Contaminants. WHO Regional Publications, Copenhagen.

Wu, Y., P.-C. Du, W.-G. Li, and J.-X. Lu. 2012. Identification and molecular analysis of pathogenic yeasts in droppings of domestic pigeons in Beijing, China. Mycopathologia 174: 203-214.

Wuthrich, M., G. S. Deepe, and B. Klein. 2012. Adaptive Immunity to Fungi. The Annual Review of Immunology 30: 115–148.

Xu, Y., C. Zheng, Z. Liu, and K. Yan. 2012. Electrostatic precipitation of airborne bio-aerosols. Journal of Electrostatics 71: 204–207.

Yamamoto, N., M. Kimura, H. Matsuki, and Y. Yanagisawa. 2010. Optimization of a real-time PCR assay to quantitate airborne fungi collected on a gelatin filter. Journal of Bioscience & Bioenginering 109: 83-88.

Yan, Z.-q., and G. K. Hansson. 2007. Innate immunity, macrophage activation, and atherosclerosis. Immunological Reviews 219: 187-203.

Yang, J., Q. Yu, and P. Gong. 2008. Quantifying air pollution removal by green roofs in Chicago. Atmospheric Environment 42:7266-7273.

Yin, S., Z. Shen, P. Zhou, X. Zou, S. Che, and W. Wang. 2011. Quantifying air pollution attenuation within urban parks: An experimental approach in Shanghai, China. Environmental Pollution 159: 2155-2163.

Yu, B.F., Z.B. Hu, M. Liu, H.L. Yang, Q.X. Kong, and Y.H. Liu. 2009. Review of research on air-conditioning systems and indoor air quality control for human health. International Journal of Refrigeration 32: 3-20.

Zak, J. C. 2002. Implications of leaf surface habitat for fungal community structure and function. In: S. E. Lindow, E. I. Hecht-Poinar and V. J. Elliott (eds.) Phyllosphere Microbiology. p 299-315. APS Press, St Paul, USA.

Zheng, D., M. J. Ducey, and L. S. Heath. 2013. Assessing net carbon sequestration on urban and community forests of northern New England, USA. Urban Forestry & Urban Greening 12: 61-68.

Zhongkai He, Wenjuan Wei, and Yinping Zhang. 2010. Dynamic—static chamber method for simultaneous measurement of the diffusion and partition coefficients of VOCs in barrier layers of building materials. Indoor and Built Environment 19: 465-475.