# **Mapping Urban Aerosolized Fungi: Predicting Spatial and Temporal Indoor C oncentrations**

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# **A bstract**

The prediction of bioaerosols, specifically airborne fungi, can be achieved using various mapping techniques, potentially enabling the determination of ambient indoor concentrations within environments where people spend most of their time. The concentration and composition of indoor air pollutants are determined by a multitude of variables, with building ventilation type being the most predominant factor in most scenarios. A predictive statistical model-based methodology for mapping airborne fungi was developed utilizing satellite-based technology. Mapping was carried out for total aerosolized fungal spores and the diversity of aerosolized fungi in Sydney, Australia, over four seasons. Corresponding data for a range of environmental parameters known to influence airborne fungi were also used, notably green space density, land cover, altitude, meteorological variables, and other locally determined factors. Statistical models previously developed from the combined meteorological and environmental variable data were used to establish spatiotemporal models for airborne fungi across the study area for each season. Results showed that the models produced reasonable predictions of monitored

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aeromycota concentrations; although, the accuracy of these predictions for individual survey periods was variable. Using known indoor/outdoor (I/O) ratios of airborne fungi for the area, the prevalence and concentrations of indoor aeromycota were modeled for buildings with both natural and mechanical ventilation. As accurate manual assessment of the aeromycota is labor, time, and cost intensive, the current findings should assist in the prediction of fungal aerosols in both urban and indoor environments. Additionally, understanding the indoor microbiome has great importance for the health and well-being of the occupants concerned.

Keywords: airborne fungi, fungal diversity, indoor microbiome, GIS, outdoor exposure

## **Introduction**

Ambient bioaerosols include microorganisms, particles originating from plants and animals, and toxins originating from microorganisms (Calvo et al., 2018). Bioaerosols are abundant, with fungal spores being one of the most prevalent suspended particles in the ambient environment, accounting for up to 60% of ambient organic particulate matter (PM), with a diameter of  $\leq 10 \mu m$  (393.7 µin) (i.e., PM<sub>10</sub>; Bauer et al., 2008; Kallawicha et al., 2015). Exposure to high concentrations of fungal aerosols is associated with asthma, allergic rhinitis or sinusitis, and atopic dermatitis (Fisher et al., 2012). Further, the importance of fungal spores is highlighted in urban environments, where the prevalence of allergies has been estimated to be higher than in rural environments (Majkowska-Wojciechowska et al., 2007). Many studies have investigated the distributions of fungal spores and the associations between airborne fungi and air pollutants, meteorological factors, and other environmental events (Almeida et al., 2018). Although the variations of bioaerosol distributions within cities are not yet fully clear, studies have indicated that levels of other air pollutants (e.g., nitrogen dioxide) have significant within-city differences, with this trend likely extending to bioaerosols (Kallawicha et al., 2015; Maya-Manzano et al., 2017).

When inhaled, fungi contribute to adverse health effects. A large proportion of people experience allergic responses to specific fungi, and have respiratory conditions that are exacerbated by fungal exposure (Baxi et al., 2016; Carrer et al., 2001; Dutkiewicz, 1997). Further, the health effects of inhaled fungal particles can include exacerbation of asthma, allergic rhinitis or sinusitis, hypersensitivity pneumonitis, allergic respiratory ailments, and atopic dermatitis. The health effects of aerosolized fungi also vary based on the differing pathogenic properties of the fungi themselves (Dutkiewicz, 1997).

As individuals in urban societies spend most of their time indoors (Leech et al., 2002; Lyytimaki, 2012), where microorganisms too inhabit, indoor environments represent a major interface of contact between the two entities (Tong et al., 2017). Understanding the factors that shape the indoor microbiome and its distribution

patterns has great importance for occupant health and well-being. Indeed, the indoor microbiome has in recent years received much research interest (Adams et al., 2015; Crawford et al., 2009; Kemp et al., 2003; Lee et al., 2006; Li & Kendrick, 1995; Sautour et al., 2009). The concentration and composition of indoor bioaerosols may be determined by a multitude of variables, with the microbial source and building ventilation type likely to be the most predominant factors in most scenarios (Prussin & Marr, 2015). The relationship between these two variables can be complicated at times, as in many buildings the inflow of outdoor air may change temporally, as will ventilation requirements associated with thermal conditioning or other indoor environmental quality requirements (Irga et al., 2018).

Three different ventilation strategies are utilized in modern urban buildings, whether residential or commercial. Natural ventilation occurs through windows, doors, skylights, and roof ventilators, and such air is not conditioned and rarely filtered (Irga & Torpy, 2016b). Mechanical ventilation, by which the air is forcibly supplied to an indoor environment by centralized heating, ventilation, and airconditioning (HVAC) systems, usually filter the supply air to prevent the distribution of contaminants (Irga & Torpy, 2016b; Kemp et al., 2003). The third ventilation type, which is common in tropical and subtropical climates, is mixed-mode ventilation, combining natural and mechanical ventilation methods (Irga & Torpy, 2016b). Although natural ventilation has numerous benefits, the concentration of indoor airborne pollutants can be higher in naturally ventilated buildings, due to outdoor air pollution being transported indoors through openings and leaks in the building envelope (Crawford et al., 2009; Irga & Torpy, 2016b). However, it is largely unknown if these three different ventilation types affect the microbiome within their associated buildings. Thus, this paper aims to quantify the seasonal outdoor airborne fungal spores and subsequent indoor airborne fungal spores based on building ventilation type across Sydney.

## **Research Purpose and Transdisciplinary Implications**

Predictive models for airborne fungi would be of substantial value, informing building ventilation system choice, design, and operation to minimize health effects on occupants, as well as provide a greater understanding of the relationship between outdoor exposure levels and subsequent indoor exposure levels. Further, accurate empirical assessment of the aeromycota is labor, time, cost, and training intensive. Thus, mapping techniques are valuable to enable the prediction of total concentration of airborne fungi encountered at a specific space and time. Given that the types and concentrations of aeromycota in indoor air respond to variations in urban design as well as environmental and meteorological variables, transdisciplinary research that integrates mycology, biotechnology, spatial sciences

and statistics, and built environment engineering knowledge will also be necessary to produce generally effective outcomes. The application of these findings further extends across numerous disciplines that encompass human environments, as they will influence building practices and design to predictively control occupants' exposure to aeromycota. Coupling spatiotemporal fungal distribution models with building infiltration factors (i.e., the ratio of fungal spores likely to penetrate into an indoor space from the outdoor environment) may allow for predictions on the concentration of fungal propagules in any indoor setting.

The fungal aerosol distribution maps presented in this paper have been developed to facilitate the study of human exposure to potentially allergenic fungi. Combining these models with hospital admissions data for asthma would be valuable in studies examining the effects of allergenic fungi. By producing detailed maps of the locations of allergenic fungi, it may be possible to identify fungi associated with increased hospital admissions for asthma in a specific region or season (Mclnnes et al., 2017). This level of detail could help with the accurate measurement of health effects as well as monitoring for climate effects through changes in source distribution, and changes to fungal allergenicity. Similarly, aeromycota mapping may also help affected individuals selfmanage their allergy or asthma. Sensitive individuals have the potential to limit their exposure when coupled with readily available information, such as weather data, and with an understanding of their ventilation system (i.e., whether to close their windows if they are in a naturally ventilated building or turn on their air-conditioning systems in a mechanically ventilated system). Once linked with health effects, these maps allow for increased guidance, with respect to ventilation management practices, to limit exposure to the most allergenic fungi or to those who have compromised health. The application of these findings further extends into architecture, development, and urban planning, as they will influence one's decision-making to maximize both workability and livability of buildings through the minimization of aeromycota exposure.

## Materials and Methods

The current work used geographic information system (GIS) mapping techniques to predict the concentration and diversity of airborne fungal bioaerosols. Statistical models based on a previous empirical assessment of fungal bioaerosols in urban Sydney were employed therein (Irga & Torpy, 2016a).

### **Site Description**

The study was conducted in the Sydney metropolitan area (approximately 4.5 million inhabitants) of eastern Australia (33.8688°S, 151.2093°E). The city includes both constructed urban environments with a limited proportion of vegetated ground area, and natural environments occupied by diverse dry sclerophyllous vegetation. The study area has the characteristics of a warm-temperate maritime climate (Vaneckova et al., 2008). The mean annual temperature is 21.9°C (71.4°F), with the monthly averages ranging from 17.1°C (62.8°F) during winter to 25.7°C (78.3°F) during summer, while the mean annual precipitation is 1,216 mm (48 in), with a monthly average ranging from 76.0 mm (3 in) during spring to 126.0 mm (5 in) during autumn (Table 1).





Source: Bureau of Meteorology (2018a).

### **D a ta**

The current work utilizes the empirical data generated by Irga and Torpy (2016a) for model development. Briefly, field sampling was conducted to determine the diversity and relative abundance of airborne fungal genera in Sydney, with the aim of determining seasonal and meteorological relationships, while also identifying potential source relationships. The field sampling involved monitoring airborne fungal spores across 11 sites with a range of varying predictor characteristics. Samples were collected monthly at each location using a Reuter centrifugal air sampler, followed by seven days of incubation and genus-level identification microscopic morphology for each sample (Irga & Torpy, 2016a). The models predict aeromycota

concentration in colony-forming units  $(CFU/m^3)$  and diversity (number of genera encountered), and are expressed in the supplementary data published by Irga and Torpy (2016a).

Four data sets were necessary to generate the maps: percentage grass cover, wind speed, rainfall, and temperature. These variables were utilized, as they significantly correlated with aeromycota concentration (CFU/m<sup>3</sup>) and diversity (number of genera encountered), and added predictive power to the statistical model, as published by Irga and Torpy (2016a). To enable temporal comparisons, input data were limited to the middle month of each season, as those months were considered the most representative. Thus, January was selected for summer, April for autumn, July for winter, and October for spring. Additionally, data from 2014 were collected where possible, as this corresponds to the year in which the majority of samples were taken to generate the statistical model.

Further, the indoor/outdoor (I/O) ratio data from Irga and Torpy (2016b) was used to predict indoor exposure. Briefly, paired indoor and outdoor samples were taken across 11 buildings in urban Sydney (Irga & Torpy, 2016b) during the same study period as the field study previously described (Irga & Torpy, 2016a). Commercial buildings that were proximal to central Sydney with a general distribution across the central urban area were selected, while building ventilation types were classified as natural, mechanical, and mixed-type ventilation (Irga & Torpy, 2016b). These data, coupled with the predicted outdoor aeromycota levels, allowed for the development of predictive indoor exposure models.

### **Percentage Grass Cover**

Percentage grass cover was derived from the normalized difference vegetation index (NDVI), which is a numerical indicator of terrestrial-based vegetation founded on the red and near-infrared reflectance ratio gathered from satellite remote-sensing measurements (Bhandari et al., 2012). The ratio is based on the chlorophyll present in plants absorbing red light while leaf structures scatter near infrared light (Pettorelli et al., 2005). This index rose with increasing "green" coverage, and was used in the current work as a proxy for vegetation cover (Pettorelli et al., 2005). Typical NDVI values ranging from 0.2-0.5 were considered indicative of sparsely vegetated landscapes (Palanisamy & Gurugnanam, 2014), while NDVI values less than 0.2 were typically representative of non-vegetated areas such as exposed soil, rock, or sand (Bhandari et al., 2012; Carlson & Ripley, 1997; Scanlon et al., 2002; Turner et al., 2013). Conversely, NDVI values higher than 0.5 were considered to indicate densely vegetated landscapes (Bhandari et al., 2012; Scanlon et al., 2002; Turner et al., 2013). NDVI data were further processed to obtain a proxy for grass cover (see Processing and Analysis), and were obtained from the Advanced Very High-Resolution Radiometer instruments carried on the United States National Oceanic and Atmospheric Administration (NOAA)  $(-11, -14, -16,$  and  $-18$ 

satellites). These were received and processed by Australia's Bureau of Meteorology (BOM, 2018b), and were provided in 0.05° x 0.05° gridded monthly averages across Australia (BOM, 2018b).

### **Rainfall**

Mean monthly rainfall total (mm) data from 1961 to 1990 were collected from the BOM's network of weather stations, and processed by averaging the months across years. An analysis technique using an optimized Barnes successive correction was next applied to the station-weighted averages to produce the data sets (Jones et ah, 2009). Again, the data were obtained in a 0.05 x 0.05° gridded output (BOM, 2016a).

#### **Temperature**

Mean temperature (°C) data were sourced from the BOM (2016b) and obtained in the same manner as the rainfall data, in a 0.025° x 0.025° gridded output.

### **Wind Speed**

Daily near-surface wind speed was obtained from the Commonwealth Scientific and Industrial Research Organisation (CSIRO) as daily averages from a network of low-set anemometers. These were subsequently interpolated and smoothed using a trivariate thin plate spline as a function of longitude, latitude, and distance inland from the coast (CSIRO, 2018; McVicar et al., 2008). As monthly averages were required, four randomly selected days across each of the months of interest were averaged. The data were provided in a 0.01 x 0.01° gridded output (CSIRO, 2018).

#### **Processing and Analysis**

All gridded data were converted to raster images and resampled to a resolution of  $0.01^\circ$  x  $0.01^\circ$  (approximately 1 km x 1 km [0.6 mi x 0.6 mi]), if they were not already in that resolution. All raster data were vectorized to allow for processing and overlaying, and clipped to represent the area of urban Sydney that was sampled by Irga and Torpy (2016a). All data for each seasonally representative month were overlaid through spatial joining to ensure the data required for aeromycota concentration and diversity (see Equations 1 and 2, Supplementary Data) were present in each pixel.

Two of the data sets required further progressing before the equations could be applied. NDVI data were converted with the use of a scaling NDVI equation (Equation 3, Supplementary Data) to obtain an index representing grass cover. As near-surface wind speed was provided as daily averages during 2014, four randomly selected days from each of the four months were selected to calculate

monthly averages. A linear wind speed correction factor of 0.1667 was applied to the averaged data due to the original instrument used by Irga and Torpy (2016a) being limited to a maximum reading of 10 m/s (33 ft/s). Thus, a 'wind speed index' with a maximum of 10 m/s (33 ft/s) was used as a surrogate for wind speed.

After all data sets were overlaid and processed, both equations were run for each pixel to develop predictive models for all seasons, both for total aeromycota density and diversity. Further, the I/O ratios were modeled for building ventilation types classified as natural, mechanical, and mixed-type ventilation for each season.

### **Results and Discussion**

### **Comparison of Spatial and Temporal Trends in Outdoor Aeromycota for Urban Sydney**

The current study provides the most comprehensive mapping of aeromycota across urban Sydney to date, elucidating both spatial and temporal patterns. The predicted total aeromycota concentrations across urban Sydney for each season are shown in Figure 1. Concentrations ranged from 330 CFU/m<sup>3</sup> (9.3 CFU/ft<sup>3</sup>) to  $1,765$  CFU/m<sup>3</sup> (50.0 CFU/ft<sup>3</sup>). Clear spatial and seasonal differences were detected, with high concentrations occurring in summer and spring. This supports expectations, as these seasons tend to have more green vegetation and higher wind speeds that assist with aerosolization and dispersal (Lin & Li, 2000; Pyrri & Kapsanaki-Gotsi, 2017), relative to autumn and winter (see Table 1; Figure 1). Further, both seasons have higher monthly rainfall totals resulting in greater rates of removal, as rainfall washes fungal particles out of the atmosphere (Pyrri & Kapsanaki-Gotsi, 2017; Troutt & Levetin, 2001). Spatial differences across the four seasons were primarily governed by increased wind speeds in the coastal suburbs, which had higher predicted concentrations of aeromycota.

The predicted aeromycota diversity across urban Sydney for each season is shown in Figure 2. The predicted diversity of fungal genera was quite homogenous across the city for all seasons, ranging between 6 and 9 genera for each 80 L  $(2.8 \text{ ft}^3)$  sample (Figure 2). This is likely a result of the highly urbanized environment supporting a very limited fungal diversity due to the altered environmental conditions (Irga & Torpy, 2016a; Parajuli et al., 2018). Spring's higher diversity of aeromycota was matched by its high concentration of total aeromycota, which was most likely due to the low rainfall in this season and, thus, lower removal rates, in combination with higher wind speeds throughout the day assisting with aerosolization (see Table 1; Figure 2). Winter had the highest diversity, which appeared to be driven by moderate to high wind speeds aiding dispersal, along with low temperatures. Summer supported the lowest fungal diversity, resulting from high temperatures

being inversely related to genera diversity. Further, diversity tended to be higher within central Sydney, which could be due to the altered landscape creating humanmade wind tunnels (Yuan et al., 2017). Overall, all four seasons displayed a similar spatial distribution, with lower diversities in the north.

Several investigations have studied the spatial variation of airborne fungi (Arobba et ah, 2000; Frenz et ah, 1997; Gonzalo-Garijo et ah, 2006; Irga & Torpy, 2016a; Kallawicha et ah, 2015; Pyrri & Kapsanaki-Gotsi, 2017). High average temperatures and high relative humidity favor microbiological growth, therefore, acting as a source for bioaerosol proliferation (Kallawicha et al., 2015; Oliveira et al., 2005). Sunlight intensity, magnitude of air currents, wind direction, and wind speed also play major roles in bioaerosol concentration and their transportation and dispersal from one environment to another (Ghosh et al., 2015; Oliveira et al., 2005). For example, recently in Taipei, Taiwan, spatial and temporal differences in aeromycota were observed, with higher concentrations noted on a rural to urban gradient during the warmer months (Irga & Torpy, 2016a; Kallawicha et al., 2015).

Further, predictor variables for fungal exposure levels in urban environments may need to consider meteorological factors, air pollutant concentrations, land-use types, and socioeconomic factors (Irga & Torpy, 2016a; Kallawicha et ah, 2015; Maya-Manzano et al., 2017; Parajuli et al., 2018; Wollan Anders et al., 2008). Notably, Parajuli et ah (2018) investigated the land-use types surrounding Finnish homes to gauge if indoor concentrations and diversities of aeromycota were affected. They found the more the percentage cover of impervious surfaces increased within an urban environment, the more diversity of aeromycota decreased. Additionally, Kallawicha et al. (2015) identified various factors that positively correlated with the concentrations of aeromycota such as temperature, relative humidity, and rainfall, while wind speed was negatively correlated with high fungal numbers. Correlations between land uses and types varied dependent on the types of fungi detected, but both *Aspergillus* and *Penicillium* were positively correlated with commercial and residential areas (Kallawicha et al., 2015).

Wollan et ah (2008) found temperature and solar radiation had strong influences on the distribution of macrofungi across Norway, while precipitation did not produce a clear relationship, unlike the current study. Pyrri and Kapsanaki-Gotsi (2017) found air temperature, solar radiation, and wind speed had significant positive correlations with the total fungal concentration, and the prevalence of almost all genera studied (Irga & Torpy, 2016a), with temperature being the single best predictor for total aeromycota. Additionally, relative humidity and atmospheric pressure were negatively correlated with the genera examined, except for *Penicillium* (Pyrri & Kapsanaki-Gotsi, 2017). Further, Pyrri and Kapsanaki-Gotsi (2017) found meteorological factors tended to have statistically significant positive or negative correlations with total aeromycota concentrations and diversity, while air pollutants typically had insignificant or weak correlations.

It is clear that different environmental variables affect the airborne fungal community within different geographical areas. Thus, it is likely that individual predictive models will be required for different areas. Currently, the inconsistent approach taken by different studies, notably concerning the specific range of environmental variables assessed, confounds direct comparisons. Further work will be required before accurate comparisons will be possible.



Figure 1. The predicted total aeromycota densities across urban Sydney based on percentage of grass cover, wind speed, and total monthly rainfall during summer (A), autumn (B), winter (C), and spring (D), with a spatial resolution of 0.01 x 0.01 degrees. Total airborne fungi equation sourced from Irga and Torpy (2016a).

Note: British imperial conversions for these calculations are available in the supplementary data for Equation 1.

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**Number of Genera**

6.00 - 7.00 *WM* 7.01 - 8.00 ■ I 8.01 - 9.00

**Number of genera (per 80 L sample) = 0.218A - 0.207B - 0.0065C + 9.8**

**Where A = Wind speed (m/s) B = Temperature (°C) C = Total monthly rainfall (mm)**



Figure 2. The predicted aeromycota genera diversity across urban Sydney based on wind speed, temperature, and total monthly rainfall during summer (A), autumn (B), winter (C), and spring (D), with a spatial resolution of 0.01 x 0.01 degrees. Number of genera equation sourced from Irga and Torpy (2016a).

Note: British imperial conversions for these calculations are available in the supplementary data for Equation 2.

### **Predicting the Concentration of Aeromycota in Indoor Environments**

To better understand the microbial population assemblage to which humans are exposed, the changes in occurrence and distribution of aeromycota within indoor environments must be investigated. Indoor exposure is primarily caused when fungi is translocated indoors from outdoor environments through HVAC systems or natural ventilation (Li & Kendrick, 1995; Medrela-Kuder, 2003).

As the distribution of microbes in outdoor air is geographically and temporally variable, these patterns transfer to indoor environments (Irga & Torpy, 2016a; Kallawicha etal., 2015; Leeetal., 2006; Li & Kendrick, 1995; Medrela-Kuder, 2003; Oliveira et al., 2005). Building ventilation strategies have been shown to influence the input of microbial communities from outdoor sources through ventilation and other forms of infiltration into the indoor atmosphere (Kemp et al., 2003; Medrela-

Kuder, 2003; Sautour et al., 2009). The source strength of outdoor air and its relative contribution to the indoor aerobiota varies based on ventilation type: within mechanically or naturally ventilated buildings, it is dependent on the inlet filtration efficiency, the rate of ventilated airflow, and the source of ventilated air. Rooms with natural ventilation (i.e., open windows) or modest-supply mechanical ventilation systems show microbial profiles that are similar to outdoor air, with low influence from other sources (Crawford et ah, 2009; Irga & Torpy, 2016b; Li & Kendrick, 1995). Therefore, as I/O ratios of airborne fungi for buildings with both natural and mechanical ventilation have been determined (Irga & Torpy, 2016b; Kemp et ah, 2003), the species' distribution and concentrations of indoor aeromycota can be predicted. The relationship between indoor and outdoor aeromycota concentrations based on season and ventilation type for urban Sydney is displayed in Table 2. Additionally, the predicted total aeromycota concentrations across urban Sydney for each season and each ventilation type are shown in Figures 3, 4 and 5. Consequently, with knowledge of building location, season, and building ventilation type, it is possible to predict the concentration of aeromycota within an indoor space using these models.



Table 2. Indoor/Outdoor ratios of aeromycota based on season and ventilation type of building.

Source: Irga and Torpy (2016b).

The ventilation types were predicted to have a marked effect on the indoor aeromycota concentrations, with the greatest bioparticle reductions predicted with mechanical ventilation in all seasons except autumn (Table 2). Indoor concentrations in naturally ventilated buildings were predicted to follow a similar trend to the outdoor concentrations (Table 2). Summer was predicted to have the lowest similarity among building types due to increased window usage, while winter had the greatest similarity, as openings in the buildings would remain closed to prevent heat loss (Lee et al., 2006). Mixed-ventilation indoor concentrations had a less predictable trend, as the ratios were the lowest for autumn and highest for spring (Table 2).

#### Mapping Urban Aerosolized Fungi



Figure 3. The predicted total indoor aeromycota densities in buildings that have mechanical ventilation across urban Sydney during summer (A), autumn (B), winter (C), and spring (D), with a spatial resolution of 0.01  $\times$ 0.01 degrees. Total airborne fungi equation sourced from Irga and Torpy (2016a) and Indoor/Outdoor ratios sourced from Irga and Torpy (2016b). Note: British imperial conversions for these calculations are available in the supplementary data for Equation 1 and the ratios from Table 2.

After applying the mechanical ventilation I/O ratios, the lowest total indoor aeromycota concentrations generally occurred in summer, while the higher indoor concentrations were experienced in autumn and spring (Figure 3). This ventilation type resulted in the lowest indoor exposure occurring in summer, but the highest exposure for the outdoor concentrations (Figures 1 and 3). Indoor exposure for buildings with natural ventilation followed the same trend as the outdoor concentrations (i.e., the I/O ratios were close to 1), with the highest concentrations occurring in summer and then spring, and the lowest in winter and then autumn (Figures 1 and 4). Buildings with mixed ventilation were predicted to have lower indoor exposure during autumn and winter, and higher contact in spring (Figure 5), which did not correspond with the outdoor concentrations. While the seasonal trends may have differed based on ventilation type and associated ratio differences, the spatial pattern did not differ, with highest concentration distribution tending to the east of the study area, and the lowest concentrations tending toward the northwest.



Figure 4. The predicted total indoor aeromycota densities in buildings that have natural ventilation across urban Sydney during summer (A), autumn (B), winter (C), and spring (D), with a spatial resolution of 0.01  $x$ 0.01 degrees. Total airborne fungi equation sourced from Irga and Torpy (2016a) and Indoor/Outdoor ratios sourced from Irga and Torpy (2016b).

Note: British imperial conversions for these calculations are available in the supplementary data for Equation 1 and the ratios from Table 2.

The current study's findings were similar to other published literature that focused on quantifying the effect of ventilation type on indoor fungal bioaerosol concentrations and diversity, particularly when related to mechanical ventilation. For example, Kemp et al. (2003) compared two buildings in central business districts and investigated the potential reduction offered by mechanical HVAC systems. This demonstrated that HVAC systems could reduce indoor fungal counts at the supply air outlets down to 13-27% of the levels in the outdoor air. Additionally, HVAC systems were effective at removing some potentially pathogenic and allergic taxa (Kemp et al., 2003), while mechanical HVAC systems were found to reduce aeromycota concentrations and diversity at both the air outlet and within the system at the cooling coils (Kemp et al., 2003). Li and Kendrick (1995) also found that HVAC systems reduced the concentrations of most airborne fungi, thus, resulting in individuals residing in buildings with air-conditioners exhibiting significantly less severe fungal allergy symptoms.

#### Mapping Urban Aerosolized Fungi



Figure 5. The predicted total indoor aeromycota densities in buildings that have mixed ventilation across urban Sydney during summer (A), autumn (B), winter (C), and spring (D), with a spatial resolution of 0.01 x 0.01 degrees. Total airborne fungi equation sourced from Irga and Torpy (2016a) and Indoor/Outdoor ratios sourced from Irga and Torpy (2016b).

Note: British imperial conversions for these calculations are available in the supplementary data for Equation 1 and the ratios from Table 2.

Sautour et al. (2009) investigated the difference between the outdoor environment and heavily ventilated indoor environments by conducting a one-year aerosolized fungal exposure study between outdoor and indoor air inside two hematological units of a French hospital. During this study, a total of 52 weekly outdoor samples were taken along with 231 and 124 indoor samples across the two hospital units, with a mean outdoor viable aeromycota concentration of  $122.1 \text{ CFU/m}^3$ (3.5 CFU/ft<sup>3</sup>) while the indoor concentrations were  $4.1$  CFU/m<sup>3</sup> (0.12 CFU/ft<sup>3</sup>) and 3.9 CFU/ $m^3$  (0.11 CFU/ $ft^3$ ), respectively (Sautour et al., 2009). Again, this indicated the potential effectiveness of HVAC filters in reducing indoor bioaerosol levels.

## **Model Improvements**

In addition to location and building ventilation, architectural and interior building design have been implicated in influencing the bioaerosols that accumulate indoors. This is because variations in building form and interior spatial arrangements can alter how occupants utilize built spaces, and even affect the magnitude and directionality of indoor human-microbe interactions (Meadow et al., 2014). If interior building design could be incorporated into the model, its fine scale accuracy could be improved.

With the data currently available, this study's models can only predict fungal behavior on coarse temporal and spatial scales. Further sampling at finer scales in both space and time would result in models with a greater probability of accurate predictions. In turn, this would allow users to make more informed decisions regarding their health based on fungal bioparticle exposure, without a need for the currently widespread practice of manually sampling air.

## **Conclusions and Further Work**

This study demonstrates that differences in atmospheric fungal spore concentrations can be expected within an urban area. The present study is, to the knowledge of the authors, the first published urban-scale airborne fungal propagule study, which uses an integrated interdisciplinary approach to the construction of a predictive model for airborne fungi across a time scale, with added reference to indoor concentrations. The applied methodology is novel and uses widely available, validated data and land-use information, synthesized with statistical techniques. With additional empirical data and the construction of more complete predictive model functions, the flexibility of the model, coupled with the ease of availability of the predictor data, will allow for the production of high-resolution maps that can be modified according to changes of any of the predictor variables. Further value from these models is related to the capability of predicting aeromycota concentrations without the need for expensive ad hoc on-site measurements. If the sources of fungal spores in urban areas can be further characterized, the next step will be to further develop a local-scale dispersion model to develop and apply a fungal spore emission inventory, and use this as a basis for understanding and explaining air movements transporting fungal distribution on a local scale. This improved understanding can be used to develop an integrated urban-scale exposure system for exposure to allergenic fungal spores in a similar vein to those developed for chemical air pollutants and allergenic pollen.

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# **Glossary**

Aeromycota: Aeromycota is a term for aerosolized fungi.

CFU: CFU refers to colony forming units.

Genera: Genera is the plural form of genus.

GIS: GIS refers to geographical information systems, a variety of software tools that facilitate data capture, processing, management, manipulation, analysis, and visualizations.

Microbiome: Microbiome is the collection of microorganisms and microbes, such as fungi and bacteria, which inhabit an environment.

Sclerophyllous vegetation: Sclerophyllous vegetation refers to vegetation that has hard leaves, which are closer together on the stem, and that hang parallel to direct sunlight. This type of vegetation is common in Australia.

## **Supplementary Data**

Total airborne fungi (Equation 1) and number of genera encountered (Equation 2), as published in Irga and Torpy (2016a), are detailed as follows.

### **Equation 1**

*Total airborne fungi* (*CFU/m*<sup>3</sup>) = 16.6A + 138B - 1.9C + 370.1

*Total airborne fungi (CFU/fP)* = 1*6.6A +* 42.065 — 48.26C + 35.315

#### Where

A = Percentage of grass cover (per unit area)  $B =$  Wind speed  $(m/s)$  [ft/s]  $C = Total monthly rainfall (mm/in)$ 

### **Equation 2**

*Number of genera encountered (per 80 L sample)*  $= 0.218A - 0.207B - 0.0065C + 9.8$ 

*Number of genera encountered (per 2.8 ft<sup>3</sup> sample)*  $= 0.0664A - (0.115B - 32) - 0.1651C + 9.8$ 

Where

 $A = Wind speed (m/s) [ft/s]$ B = Temperature (°C/°F)  $C = Total monthly rainfall (mm/in)$ 

NDVI data were converted with the use of a scaling NDVI equation (Equation 3) to obtain an index representing grass cover.

### **Equation 3**

 $\emph{Fractional vegetation cover } = \frac{NDVI - NDVI_s}{NDVI_{\infty} - NDVI_s}$ 

Where:

*ND VI* is the vegetation index value of an individual pixel

NDVI represents the typical NDVI of bare soil areas for the study area

*NDVI* is the value of a pure green vegetation pixel for the study area

*NDVIs* and *NDVI^* are seasonal and geographical constants that were extracted from a histogram of the NDVI of the study area across the months; these were subsequently confirmed through the use of satellite imagery to ensure the NDVI constants was representative of soil and pure green vegetation. To ensure only grass cover was considered by this index, any pixel with an NDVI greater than 0.35 or less than 0.12 was excluded, based on the methods used by Bhandari et al. (2012), Scanlon et al. (2002), and Turner et al. (2013). These were also validated through the random selection of 20 pixels on a satellite image of the study area to manually confirm the presence of grass cover.

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