國立臺灣大學公共衛生學院職業醫學與工業衛生研究所

碩士論文

Graduate Institute of Occupational Medicine and Industrial Hygiene

College of Public Health

National Taiwan University

Master thesis

# 大氣中真菌孢子的分布與環境因子的相關性

# Temporal distribution of ambient fungal spores and the association with environmental parameters

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中華民國 105 年 7 月

July, 2016

doi:10.6342/NTU201603517

# 國立臺灣大學碩士學位論文
# 口試委員會審定書

大氣中真菌孢子的分布與環境因子的相關性

Temporal distribution of ambient fungal spores and the association with environmental parameters

本論文係黃聖宏君（R03841017）在國立臺灣大學職業醫學與工業衛生研究所完成之碩士學位論文，於民國 105 年 07 月 13 日承下列考試委員審查通過及口試及格，特此證明

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doi:10.6342/NTU201603517

# 誌謝

時光飛逝，研究所兩年就這樣過去了，在當中無不受到許多人的幫助，也成就了今天這本論文的誕生。首先最要感謝指導教授郭育良老師，憶起當時初進實驗室時，老師就說這裡是你最後可以犯錯的機會，更是你改進錯誤的地方，當中也許莽撞、也許迷失，或許事情並無做得盡善盡美，但感謝老師不放棄的指導，不僅給予學術上的教導，重要的是那份態度與自我要求的精神。感謝吳焜裕老師、吳章甫老師、黃耀輝老師與張靜文老師在論文上的指導與建議。感謝趙馨老師、沈偉強老師在我需要協助時，引領我去看到事情的不同面向。再來幫助我最多的就是 Bird，從一開始帶我們做實驗，每天都耗費許多精力，帶領我們判讀玻片，到後來論文及報告的部分，只要有對於真菌上的疑惑，總是能提供我很好的想法及方向。感謝秉鈺學姐、士群學長在我對於研究上有困難時，總是適時的給予幫助，以及其他在我報告時，不吝於分享意見的每一位實驗室夥伴，還有我的實驗室同學蔡宜秦，一起承擔那做實驗的辛勞。最後我要感謝家人與朋友，在背後支撐著我，在我徬徨苦悶時，能有所依靠。

研究所的結束不是個終點，兩年的淬煉使自己成長，也期許在未來的道路上，能依舊保有學生時代的熱誠。要感謝的人太多，我想就感謝天吧!



doi:10.6342/NTU201603517

## 中文摘要

**背景:** 暴露到真菌會造成不良的健康影響，像是呼吸道疾病、過敏性疾病和感染等等。真菌會存在於多項種環境介質中，且在生物氣膠中占有相當的部分。真菌孢子在生成、釋放、運輸和沉降的過程中，已經知道會受到氣候因子的影響。但我們對於台北都會區真菌孢子濃度的認知還是有限的。因此本研究目的是要去探討每日的真菌孢子濃度，並去評估它與環境因子的相關性。

**方法:** 在 2015 整年間，採用 Burkard 七天連續性孢子採樣器去監測每日的孢子濃度，採樣器架設在台北市古亭國小的頂樓，流率為每分鐘 10 公升。氣象資料從中央氣象局-台北測站取得，空氣汙染物資料由環保署空氣品質監測站-古亭測站取得。除描述性統計外，真菌孢子濃度和環境因子的相關性使用多變項線性回歸分析。

**結果:** 我們取得了 309 個樣本在本次研究中。出現頻率超過 70%的真菌孢子為 Ascospores, *Aspergillus/ Penicillium*, Basidiospores, *Cladosporium*, Smuts 及 *Arthrinium* 。大部分的真菌孢子在夏季的濃度較高，且發現到真菌孢子會與風速呈現負相關，與溫度、露點溫度、和空氣汙染物呈現正相關。

**結論:** 真菌孢子濃度會受到氣象因子及空氣汙染物所影響，且在台北有著一定的分布狀況。在夏季，較高的溫度及濕度會使得孢子的濃度提高；而當大雨或強風出現時，濃度就會下降。本篇研究提供了台北真菌孢子室外基礎濃度。未來研究可用此結果，進行更進一步的健康研究。



**關鍵字 :** 大氣、真菌孢子、環境因子、空氣污染物





doi:10.6342/NTU201603517

## Abstract

**Background:** Fungal spores are important ambient pollutants which present in all kinds of environment and it contributes as a major component of ambient bio-particle. Exposure to fungal spores is resulting to the adverse health outcomes such as respiratory diseases, allergic disease and infection. Fungal spores are mainly determined by climate factors include production, release, transport and deposition patterns. Previous study had reported the temporal distributions of ambient fungal spore in Taipei area. However, fungal spores were monitored only 7-day/month during 2005-2009. Little is known about daily concentrations of ambient fungal spores. Therefore, the aim of this study is to monitor daily concentration of fungal spore and evaluate their relationship with the environmental parameters.

**Method:** Using the Burkard seven-day volumetric spore trap to monitor the daily concentration of fungal spore during 2015 in Taipei. Sampler was set up on the rooftop of Guting elementary school. The flow rate of sampler is 10 L/min. Daily meteorological data were retrieved from Central Weather Bureau (CWB)- Taipei station. Daily air pollutants were acquired from Taiwan Environmental Protection Administration monitoring station-Guting station. Descriptive statistics of concentrations of ambient fungal spores presented the distribution and characteristic of fungi. The relationships between the concentrations of fungal spores and environmental parameters were



doi:10.6342/NTU201603517

estimated by using multiple linear regression.

**Results:** A total of 309 samples were successfully collected during 2015. The most prevalence fungal taxa were Ascospores, *Aspergillus/ Penicillium*, Basidiospores, *Cladosporium*, followed by Smuts, *Arthrinium*, presenting in more than 70% of the samples. Most concentration of fungal taxa were highest in summer. Fungal spores were negatively correlated with wind speed, and positively associated with temperature, dew point temperature and air pollutants.

**Conclusions:** We found the temporal distribution of fungal spores in Taipei. Concentration of fungal spores were affected by the meteorological parameters and air pollutants. Higher levels of fungal concentration was found in summer, likely related to higher temperature and humidity. While there were the heavy rainfall and strong wind, the concentration decreased. This study provides baseline information on concentration of ambient fungal spores in Taipei. Further study can utilize it to investigate the health outcomes associated with fungal spores.

**Keyword:** Ambient, Fungal spores, Environmental parameters



doi:10.6342/NTU201603517

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doi:10.6342/NTU201603517

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# Chapter 1 Introduction

## 1.1 Background

Fungal spores are one of the most important ambient pollutants which present in all kinds of environment and it contributes as a major component of ambient bio-particle (Kochar et al, 2014). Some fungi such as Zygomycota, Ascomycota, and Basidiomycota contain most genera of fungi that produce airborne fungal allergens (Levetin et al, 2016).Some fungi produce mycotoxin such as aflatoxins, ochratoxins . These fungi and it production are not only find in food, but also find in the air or the settled dust. Exposure to fungal spores is resulting to the adverse health outcomes such as respiratory diseases, allergic disease, infection and even cancer. Aerobiological studies can help us to make sure of the concentration of the fungal spores present in the atmosphere and give better understanding of the relationship between their concentrations and the meteorological parameters (Grinn-Gofron et al, 2015). Previous study by Chao and Lee in 2013 had reported the temporal distributions of ambient fungal spore in Taipei area. However, fungal spores were monitored only 7-day/month during 2005-2009 (Chao et al, 2013). Little is known about daily concentrations of ambient fungal spores.



doi:10.6342/NTU201603517

## 1.2 Objective

The aim of this study is to monitor daily concentration of fungal spore and evaluate their relationship with the environmental parameters.



doi:10.6342/NTU201603517

## Chapter 2 Literature review

### 2.1 Introduction to fungi

A fungus, any of about 99,000 known species of organisms of the kingdom Fungi, which includes the yeasts, rusts, smuts, mildews, molds, and mushrooms, is a eukaryote that digests food externally and absorbs nutrients directly through its cell walls (Carris et al, 2012). A typical fungus consists of a mass of branched, tubular filaments, called hyphae (singular hypha), enclosed by a rigid cell wall (Moore, 2016). Many fungi are free-living in soil or water; others form parasitic or symbiotic relationships with algae, plants or animals. Fungi are involved in a wide range of activities like some fungi are decomposers, which are responsible breaking down organic matter and releasing carbon, oxygen, nitrogen, and phosphorus into the soil and the atmosphere with bacteria.

### 2.2 Sporulation and fungal spores releasing

Following a period of growth, fungi enter a reproductive phase by forming and releasing vast quantities of spores. In both sexual and asexual reproduction, fungi produce spores, which are usually single cells produced by fragmentation of the mycelium or within specialized structures (sporangia, gametangia, sporophores, etc.) (Boundless,2016). There are two effects of meteorological variables on release processes include inert



doi:10.6342/NTU201603517

release process and active release process (Jones et al, 2004). The inert release process of material from a surface will depend on the balance of two groups of forces, i.e. bonding forces and removal forces. Forces such as the electrostatic force if the particle and surface are differently charged, or surface tension if the surface is wet, will tend to retain the particle on the surface, as will any physical attachment, it is called bonding forces. Bonding effects are most likely to be affected by the temperature and humidity of the surrounding air, and by the radiation balance of the surface. Forces, which is greater than the forces attaching the particle to the surface, might remove the particle from the surface when the movement of the surface is varying and the surface accelerates away from the particle. Such movement may occur as a result of wind, impact of raindrops or other physical disturbance. The active release process are most in Ascomycetes and Hymenomycetes (the largest order of Basidiomycetes), active release of ascospores and basidiospores takes place release frequently depends on the activity of turgid cells, which require a supply of water, and spores can be ejected for substantial distances—in the case of asci, 2–300 mm depending on species.

### 2.3 Fungal importance and disease

Fungi are essential to many household and industrial processes, making of bread, wine, beer, and certain cheeses (Moore, 2016). Studies of fungi have greatly contributed to the



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accumulation of fundamental knowledge such as molecular biology, genetic engineering, and other basic disciplines of biology. The medical relevance of fungi was discovered by Scottish bacteriologist Alexander Fleming, who published a scientific report announcing the discovery of penicillin, the first of a series of antibiotics in 1929. However, there are also adverse health effects of fungi. For example, Aspergillosis, an infection caused by *Aspergillus*, cause allergic reactions, lung infections, and infections in other organs (CDC, 2016). Candidiasis, a fungal infection caused by yeasts that belong to the genus Candida, caused health hazard depending on the area of the body that is infected such as in the mouth or throat is called "thrush", in the vagina is referred to "yeast infection", in bloodstream and spread throughout the body is called Invasive candidiasis.

**2.4 Determinants of ambient fungal spores**

Fungal spores are known to be influenced by the environment and biological factors such as geographical location, air pollution, weather conditions, human activity and local source of vegetation (Grinn-Gofron´ et al, 2015). Other study reported fungal spores are mainly determined by climate factors include production, release, transport and deposition patterns. In this study, there were a strong relationship between the lichen-forming fungal spores and rainfall events by using spearman's correlation tests (Favero-Longo et al, 2014). Study in Poland reported temperature is the environmental factor that



doi:10.6342/NTU201603517

can significantly affect the growth and development of fungi, including the abundance of their sporulation (Kasprzyk et al, 2016). The fungal spores also associated with lag day. In Sydney study reported the environmental parameters like relative humidity in the previous 1,2 and 3 days were positively correlated with the concentration of Alternaria and other parameters associated with spores on sampling day. (Stennett et al, 2004).



doi:10.6342/NTU201603517

## Chapter 3 Materials and Methods

### 3.1 Study design

We monitor daily ambient fungal spores during 2015 in Taipei, Taiwan. Investigating the composition and temporal distribution of concentrations of ambient fungal spores during the study period. We also evaluate the relationships between the concentrations of fungal spores and environmental parameters and air pollutants.

### 3.2 Ambient fungal spores sampling

Using the Burkard seven-day volumetric spore trap to monitor the daily fungal spores from January to December during 2015 in Taipei. Sampler was set up on the rooftop of Guting elementary school of Taipei city at a height 15 meters above the ground, where the Environmental Protection Administration (EPA) monitoring station is located. The flow rate of sampler is 10 L/min and the spores were impacted onto Melinex tape coated with Lubriseal grease (A.H. Thomas, Inc., Philadelphia, PA, USA) which moves past the inlet at 2 mm per hour over a 24-hour period. We calibrated the flow rate after we replaced the drum with the tape once a week. The collected 7-day tapes were cut into seven segments which spores were trapped on it through a 2 mm X 14 mm orifice and presented fungal concentrations over 24 hours on a 48 mm band. Each segment was fixed on



doi:10.6342/NTU201603517

microscopic slide and colored with glycerin jelly. We identified samples under microscopic at a 1000x magnification using longitudinal traverse method. Then, we transformed spores counts into average daily concentrations (spore $m^{-3}$). Samples identified base on fungal spore morphological characteristics. We using the fungal spore identification of American Academy of Allergy Asthma & Immunology (AAAAI) to identify 24 fungal taxa including *Alternia*, Ascospores, *Aspergillus/Penicillium*, *Arthrinium*, Basidiospores, *Botrytis*, *Cercospora*, Cladosporium, *Curvularia*, *Drechslera/Helminthosporium*, *Epicoccum*, *Fusarium*, *Nigrospora*, *Oidium/Erysiphe*, *Periconia*, *Peronospora*, *Pithomyces*, *Polythrincium*, Rusts, Smuts, *Stemphylium*, *Torula*, *Tetraploa* and *Ulocladium*. The identified fungal spores which were not on the list were categorized as other fungi. Fungal spores were broken or covered by gel or particle that difficult to identify were categorized as "unidentified" spores. Fungal spores were still undiscovered were categorized as "unknown" fungi.

### 3.3 Air pollutant and meteorological data

Daily meteorological data were retrieved from Central Weather Bureau (CWB)- Taipei station (121°30' 24.15"E, 25°02' 22.62"N). The Taipei station was the closest station to our sampling site. The data included temperature (°C, Sheathed Thermometer.), relative humidity (%, Hair hygrometer), rainfall (mm, tipping-bucket raingauge), dew point



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temperature (°C, hair hygrometer), solar radiation ($MJ/m^2$, solar-cell sunshine recorder) and average wind speed (m/s, anemometers). Daily air pollutants were acquired from Taiwan Environmental Protection Administration monitoring station-Guting station (121°31’ 46.40’’E, 25°01’ 4.19’’N). The data Included sulfur dioxide, carbon monoxide (ppm, carbon oxide analyzer, absorbing non-dispersive infrared.), ozone (ppb, ozone analyzer, ultra-violet (UV) absorption.), $PM_{2.5}$ ($\mu g/ m^3$, beta ray analyzer, differences of radiation strength on the filter paper), $PM_{10}$ ($\mu g/ m^3$) and nitrogen dioxide (ppb, nitrogen oxide analyzer, theorem of chemiluminescence.).

### 3.4 Statistic analysis

Analysis were performed by Microsoft Excel, JMP 10 and SAS version 9.4 (SAS Institute Inc., Cary, NC, USA). Descriptive statistics of concentrations of ambient fungal spores such as mean, median, standard deviation, minimum, maximum, and IQR presented the distribution of fungi.

The relationships between the concentrations of fungal spores and environmental parameters and air pollutants were used multiple regression. The day-lag before the sampling day of meteorological parameters also analysis on it. We used base-10 logarithm to transform fungal concentration to normality. Simple linear regression was used to determine which variable were significantly associated with fungal spores. Those



doi:10.6342/NTU201603517

significant variable were included for forward stepwise regression with p-value of 0.2 or smaller, and then the variables with highest p-value were excluded until all remaining variables were with p-value of 0.01 or smaller. Avoiding zero values, we add 0.5 to fungal spore counts on log transformation of fungal concentrations.



doi:10.6342/NTU201603517

## Chapter 4 Results

### 4.1 Distributions and temporal trends of ambient fungal spores

There were total 309 fungal samples in this study during 2015. The missing data included New year, Chinese new year, failure drum exchange and power problem. Table 1a showed the distribution of ambient fungal spores. The mean concentrations of total fungal spores was 4448.08 spore $m^{-3}$. The most prevalence fungal taxa were Ascospores, *Aspergillus/ Penicillium*, Basidiospores, *Cladosporium*, followed by Smuts, *Arthrinium*, presenting in more than 70% of the samples. Table 1b showed the distribution of environmental parameter. The mean temperature was 23.86 ℃.

The temporal distribution for monthly concentrations of total fungal spores during the sampling period showed in figure 1. The concentrations of total spores was highest in summer (June to August). Figure 1a-1c showed the temporal distribution for monthly concentrations of fungal categories. Most concentration of fungal taxa were highest in summer, except for a peak in *Curvularia* in September and *Botrytis* in May.

### 4.2 Determinants of ambient fungal spores

Figure 2-3 showed the monthly averages of meteorological factors in Taipei during the study period. Temperature, solar radiation and dew point temperature were highest in



doi:10.6342/NTU201603517

summer. Relative humidity was highest in March. Wind speed was highest in winter. Rainfall was highest in August. Figure 4 showed the monthly averages of air pollutants. Most air pollutants were lowest concentration in summer, except for $SO_2$.

The relationships between the concentrations of major ambient fungal spores and environmental parameters showed in Table 2. Most fungal taxa were positively associated with air pollutants, except for $O_3$. Most fungal taxa were negatively associated with relatively humidity, wind speed and rainfall, but positively with solar radiation, temperature and dew point temperature.

Table 3 showed the multiple regression for fungal spores including total spores, Ascospores, *Aspergillus/Penicillium*, *Arthrinium*, Basidiospores, *Cladosporium* and Smuts and 0- , 1- and 2-day lag environmental parameters. Most fungal taxa were positively associated with dew point temperature, but negatively with wind speed. We found 72.6% of the variability in the total spore concentrations was explained by the model. In the model, total spores were positively associated with dew point temperature, solar radiation and CO on sampling day, but negatively with wind speed. We found 68.0% of the variability in the Ascospores concentrations was explained by the model. In the model, Ascospores were positively associated with dew point temperature, but negatively associated with wind speed on sampling day. 58.2% of the variability in the *Aspergillus/Penicillium* concentrations was explained by the model. In the model,



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*Aspergillus/Penicillium* were positively associated with temperature and $PM_{2.5-10}$, but negatively with wind speed on sampling day. On two day before sampling day, *Aspergillus/Penicillium* were positively associated with dew point temperature. 30.9% of the variability of the *Arthrinium* was explained by the model. *Arthrinium* were positively associated with temperature and CO on sampling day, but negatively associated with relatively humidity. It was also negatively associated with 1-day-lag ozone. 75.2% of the variability of the Basidiospores was explained by the model. Basidiospores were positively associated with dew point temperature, $NO_2$, $O_3$ on sampling day. Basidiospores were positively associated with 2-day-lag relative humidity, but negatively with 1-day-lag wind speed. We found 33.2% of the variability of the *Cladosporium* was explained by the model. In the model, *Cladosporium* were positively associated with solar radiation and $PM_{2.5-10}$, but negatively with wind speed on sampling day. 57.9% of the variability of the Smuts was explained by the model. Smuts were positively associated temperature and $PM_{2.5-10}$, but inversely associated with wind speed on sampling day. Smuts were positively associated with 2-day-lag relatively humidity, but negatively with 2-day-lag rainfall.



doi:10.6342/NTU201603517

## Chapter 5 Discussions

### 5.1 Distributions and temporal trends of ambient fungal spores

This study using Burkard seven-day volumetric spore trap collect ambient fungal spores, and investigate the distributions and characteristic of ambient fungal spores in Taipei during 2015. We found the most prevalent fungal spores in Taipei were Ascospores, *Aspergillus/Penicillium*, Basidiospores, *Cladosporium*, presented in almost 100% of the samples. Smuts, *Arthrinium*, presented in more than 70% of the samples. The results were similar to previous study in Hualien, Taiwan. Ho et al. (2005) showed Ascospores, *Cladosporium*, *Aspergillus/Penicillium* and *Ganodema* were the most prevalent fungal categories in Hualien, Taiwan, presented in more than 60% of the samples. Wu et al. (2004) found that *Cladosporium*, Ascospores, *Periconia*, Basidiospores, *Botrytis*, Smuts, *Alternaria*, *Aspergillus/Penicillium* were the most prevalent fungal taxa in Tainan, Taiwan during the sandstorm episode days, and days before and after the episodes. Both in previous study in Taiwan, although the sampling site were different, the results were the same to us. Study using the Burkard portable air sampler in Taipei, found the most prevalent fungal taxa were *Cladosporium* and *Penicillium*, presented in more than 70% of the sample (Chao et al, 2012). Burch et al. (2002) showed the major composition of fungal taxa in Tulsa site were *Cladosporium*, Basidiospores and Ascospores. Study in



doi:10.6342/NTU201603517

Havana (Cuba) showed the results that *Cladosporium*, *Coprinus*, *Lepthosphaeria*, *Aspergillus/Penicillium* were the most prevalent fungal categories (Almaguer et al, 2014). Other study in Seoul showed that Ascomycota and Davidiella (anamorph : *Cladosporium*) were the most prevalent fungal taxa (Oh et al, 2014). Study in other country, the dominants fungal taxa were similar to our findings. There were differences of the fungal taxa compositions between our findings and the previous studies because of different study period, climate, local source of vegetation.

It is well known that weather conditions influence the daily variability as well as seasonal levels of ambient spore concentrations (Grinn-Gofroń et al, 2011). In our study, concentrations of ambient fungal spores had seasonal patterns, with higher level in summer. There were similar results in previous study in Taiwan (Ho et al, 2005; Wu et al, 2004). Because of higher temperature and humidity, concentration of fungal spores were higher in summer.

**5.2 Determinants of ambient fungal spores**

Weather conditions influence the biology of fungi such as production, release, dispersion and deposition of spores as well as the diversity and number of airborne bio-particles (Almaguer et al, 2014). A typical pattern of growth depends on response to the nutrients in the environment, modified by other environmental factors (University of



doi:10.6342/NTU201603517

Sydney, 2004; Carlile et al, 1994; Robson et al, 2007).

During sporulation, the environment plays a major role in determining whether a fungus forms sexual or asexual spores. The important factors are include water, light, nutrients, oxygen, pH and temperature (University of Sydney, 2004). The majority of fungi are mesophilic, growing usually be correlated with a limited range of temperatures between 15°C and 35°C. In our findings, there were similar trend on temperature and fungal categories. The higher temperature in summer, the higher concentration of fungal spores. Most fungal taxa were positively correlated to temperature in our study. It was similar to previous study in Taiwan (Ho et al, 2005; Wu et al, 2004) and other study in the world (Rodríguez-Rajo et al, 2005; Artac et al, 2014). In our study, most fungal taxa were positively correlated to dew point temperature. The water vapor in the air becoming saturated and condensing out is called dew point temperature. Most often humidity we meet as ‘‘relative humidity’’ given as a percent—meaning the higher the percent, the closer the temperature and dew point are (Grinn-Gofron et al, 2011). The higher relative humidity and dew point temperature, the more water vapor content in the environment to produce the spores. We found that relative humidity was little difference. It was almost 70-80% in whole year of 2015. However, the dew point temperature was highest in summer. Thus, there were more water vapor to help fungi to grow. Previous study showed total spores, Basidiospores, Ascospores, and *Alternaria* were significantly and positively



doi:10.6342/NTU201603517

correlated with dew point temperature (M. Burch et al, 2002). Study in Poland showed the *Aspergillus* and *Penicillium* were positively correlated with dew point temperature on 1-, 2-, 3-day-lag and sampling day (Grinn-Gofron et al, 2011).

In the atmospheric environment, air movement is often unpredictable in the transport and dispersal of fungal spores (Wiley et al, 2007). How well spores are dispersed and survive over horizontal distance is determined by their ability to survive in the ambient environment, such as size (Jones et al, 2004). Pasanen et al. (1991) found that spores of *Aspergillus fumigatus* and *Penicillium* were released from leaves when the air velocity was 0.5 m/s, while *Cladosporium* required an airflow of at least 1.0 m/s for spore release. Minimum wind was directly correlated with spore counts, while maximum wind was negatively correlated. High wind speed is also likely to disperse spore clouds to dilute spore concentrations (Wiley et al, 2007). In our study, most fungal taxa were negatively correlated with wind speed. It was similar to the previous study that Li found that wind speed was negatively correlated with Basidiospores (Li, 2005).

Movement due to the wind (either waving or fluttering) or being struck by a raindrop or other physical disturbance of the plant surface may result in material resting on the surface being lifted into the air (Jones et al, 2004). We found that most fungal categories were negatively correlated with rainfall. Previous study found that rainfall had significant influence on decreasing fungal spore number (Sivasakthivel et al, 2015). There was a



doi:10.6342/NTU201603517

study found intermittent rain produced a spore concentration peak for each rainfall event, but a lower total number of spores than for continuous rain (Jones et al, 2004; Gottwald et al, 1997). Although rainfall can help spores to disperse, rain also removes particles from the air by both rainout and washout effects (Burge et al, 2000).

In our finding, fungal taxa were positively correlated to radiation. There was a report found that high UV-B radiation levels of the Antarctic environment as spores have shown higher germination rates (Tosi et al, 2005). Other study reported that some fungi absorb solar radiation to enhance the growth (Dadachova et al, 2007). However, a study found that UV-B radiation reduced hyphal growth (Tosi et al, 2005), and high-intensity 405-nm light inactivate germinating spores of fungi (Murdoch et al, 2013).

Approximately 24% of the count of total atmospheric particles and 5-10% of the total suspended particulate matter were reported to be contributed by bio-aerosols (Adhikari et al, 2006). Previous study found that $PM_{10}$ were positively correlated to fungal taxa (Sousa et al, 2008). In our study, particulate matter were positively correlated with most fungal categories. PM could bind with airborne pollen and fungal spores altering their morphology and changing the dispersal pattern of bio-aerosols in ambient air by altering the particle aerodynamic properties (Adhikari et al, 2006).

Tropospheric ozone is a strongly phytotoxic oxidant, possibly altering the production content of pollen (Adhikari et al, 2006). Study reported that ozone has been shown to



doi:10.6342/NTU201603517

significantly reduce plant growth and yields through the induction of oxidative stress in plants, leading to enhancement of senescence, reduction of net photosynthesisand the premature degradation of vital leaf proteins (Tiedemann et al, 2000). Previous study written by S.I.V. Sousa et al found that most fungal categories were negatively correlated with ozone (Sousa et al, 2008). However, other study found that Cladosporium and Alternaria were positively correlated to ozone (Grinn-Gofron et al, 2011). Tiedemann and Firsching. (2000) reported that the pathogenicity of rust fungi could be increased by ozone. Therefore, besides causing respiratory health hazard, ozone may also influence the sources of ambient bioaerosols (Adhikari et al, 2006). We found that ozone was negatively correlated with fungal taxa. Sulfur dioxide is known to have antifungal activity and has been used for the control of postharvest fungal diseases (Fenn et al, 1988). We found sulfur dioxide was negatively correlated with some fungal taxa. Sulfur dioxide has been shown to inhibit growth of fungi and to inhibit germination of fungal spores (Babich et al, 1978). Effect of $SO_2$ such as formation of sulphate and toxic intermediate solution products, reductions in pH and loss of nutrients, were probably reduced the abundance of fungal species (Newsham et al, 1992). Although $SO_2$ was positively correlated with most fungal categories in spearman's rank test, we found sulfur dioxide was not the mainly parameters to affect the spores concentration in multiple regression.



doi:10.6342/NTU201603517

### 5.3 Advantages and limitations

This study collected daily ambient fungal spores in Taipei during 2015. The results could clearly understand the concentrations of fungal spores in Taipei, further know about its distribution, characteristic and relationships with environmental parameters. However, there were limitations during this sampling period. For example, first of all, there were only one instrument located 15 m above ground level. This can not interpret whole of Taipei, and the exposure of fungal spores may not realistically reflect to the human. Second, for microscopic method, we counted only 24 fungal taxa, but there were more than ten thousands of fungi in the world. Third, unidentified fungi on fungal categories, because of broken or covered by gel or particles may underestimated the concentration of fungal spores. Forth, meteorological parameters and fungal spores samples acquired from different place.



doi:10.6342/NTU201603517

## Chapter 6 Conclusions

We used Burkard seven-day volumetric spore trap to monitor daily ambient fungal spores in Taipei from January to December during 2015, and investigate the distributions and characteristics of ambient fungal spores. We found the temporal distribution of fungal spores in Taipei. Concentration of fungal spores were affected by the meteorological parameters and air pollutants. In summer, the higher temperature and humidity, the higher level in concentration. While there were the heavy rainfall and strong wind, the concentration decreased. Most air pollutants bind with fungal spores. When the concentration of pollutants were higher, it found higher concentration of fungal spores. This study is baseline of concentration of ambient fungal spores in Taipei. Further study can utilize it to investigate the health outcome.



doi:10.6342/NTU201603517

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doi:10.6342/NTU201603517

**Table 1a. Descriptive statistics for ambient fungal concentrations (#/M$^3$) in Taipei in 2015. (N=309)**

| Fungal categories | Freq(%) | Mean | Median | SD | Min | Max | IQR |
|---|---|---|---|---|---|---|---|
| **Total spores** | 100.00 | 4448.08 | 2838.89 | 4187.03 | 155.56 | 18950.56 | 5281.11 |
| **Ascospores** | 100.00 | 1850.47 | 1170.56 | 1890.24 | 31.11 | 9601.67 | 2080.56 |
| ***Aspergillus/Penicillium*** | 100.00 | 372.62 | 221.67 | 432.04 | 11.67 | 4281.67 | 423.89 |
| **Basidiospores** | 99.68 | 1500.68 | 758.33 | 1747.74 | 0.00 | 8458.33 | 2080.56 |
| ***Cladosporium*** | 99.68 | 529.19 | 326.67 | 589.07 | 0.00 | 3780.00 | 544.44 |
| **Smuts** | 95.47 | 69.18 | 46.67 | 69.62 | 0.00 | 416.11 | 81.67 |
| ***Arthrinium*** | 77.02 | 18.63 | 11.67 | 29.92 | 0.00 | 272.22 | 19.44 |
| ***Nigrospora*** | 69.26 | 10.47 | 3.89 | 14.79 | 0.00 | 116.67 | 15.56 |
| ***Periconia*** | 63.75 | 7.15 | 3.89 | 9.05 | 0.00 | 50.56 | 11.67 |
| ***Curvularia*** | 53.40 | 5.69 | 3.89 | 8.85 | 0.00 | 58.33 | 7.78 |
| ***Botrytis*** | 53.07 | 17.59 | 3.89 | 32.14 | 0.00 | 194.44 | 23.33 |
| ***Torula*** | 51.13 | 6.20 | 3.89 | 9.76 | 0.00 | 77.78 | 7.78 |
| ***Fusarium*** | 42.72 | 7.27 | 0.00 | 15.77 | 0.00 | 167.22 | 7.78 |
| ***Alternaria*** | 39.16 | 3.67 | 0.00 | 7.03 | 0.00 | 54.44 | 3.89 |
| ***Cercospora*** | 19.74 | 1.69 | 0.00 | 4.69 | 0.00 | 35.00 | 0.00 |
| ***Drechslera/Helminthosporium*** | 11.00 | 0.79 | 0.00 | 3.77 | 0.00 | 50.56 | 0.00 |
| ***Peronospora*** | 9.71 | 0.94 | 0.00 | 4.47 | 0.00 | 58.33 | 0.00 |
| **Rusts** | 8.41 | 0.58 | 0.00 | 3.46 | 0.00 | 54.44 | 0.00 |
| ***Pithomyces*** | 6.80 | 0.33 | 0.00 | 1.33 | 0.00 | 11.67 | 0.00 |
| ***Tetraploa*** | 5.18 | 0.24 | 0.00 | 1.21 | 0.00 | 15.56 | 0.00 |
| ***Oidium/Erysiphe*** | 4.53 | 0.21 | 0.00 | 1.04 | 0.00 | 7.78 | 0.00 |
| ***Epicoccum*** | 3.24 | 0.14 | 0.00 | 0.79 | 0.00 | 7.78 | 0.00 |
| ***Stemphylium*** | 3.24 | 0.15 | 0.00 | 0.87 | 0.00 | 7.78 | 0.00 |
| ***Ulocladium*** | 0.32 | 0.01 | 0.00 | 0.22 | 0.00 | 3.89 | 0.00 |
| ***Polythrincium*** | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| **Other fungi** | 80.58 | 29.42 | 19.44 | 35.00 | 0.00 | 202.22 | 38.89 |
| **Unidentified fungi** | 72.82 | 14.13 | 3.89 | 22.88 | 0.00 | 155.56 | 15.56 |
| **Unknown fungi** | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |

Freq-frequency; SD-standard deviation; Min-minimum; Max-maximum; IQR-inter-quartile range

*Frequency was the percentage of samples (total n=309) with presence of that specific fungal category.



doi:10.6342/NTU201603517

**Table 1b. Descriptive statistics for environmental parameters in Taipei in 2015. (N=309)**

| Variables | Mean | Median | SD | Min | Max | IQR |
|---|---|---|---|---|---|---|
| Temperature (°C) | 23.86 | 24.80 | 5.38 | 11.30 | 32.20 | 9.50 |
| $T_d$ (°C) | 19.15 | 20.80 | 5.18 | 5.10 | 25.40 | 8.20 |
| RH (%) | 75.76 | 76.00 | 7.57 | 51.00 | 93.00 | 12.00 |
| WS (m/s) | 2.30 | 2.10 | 1.20 | 0.50 | 9.60 | 1.80 |
| Rainfall (mm) | 7.58 | 0.00 | 25.58 | 0.00 | 306.70 | 4.00 |
| Solar radiation ($MJ/m^2$) | 12.51 | 12.80 | 7.43 | 0.00 | 27.81 | 12.88 |
| CO (ppm) | 0.53 | 0.49 | 0.17 | 0.13 | 1.21 | 0.22 |
| $NO_2$ (ppb) | 19.49 | 18.90 | 5.64 | 2.38 | 41.38 | 6.23 |
| $O_3$(ppb) | 25.19 | 25.10 | 7.80 | 6.86 | 55.13 | 9.38 |
| $PM_{2.5}$ ($\mu g/ m^3$) | 17.08 | 14.58 | 11.26 | 0.00 | 68.91 | 11.14 |
| $PM_{2.5-10}$ ($\mu g/ m^3$) | 27.61 | 27.33 | 7.57 | -5.04 | 57.04 | 8.12 |
| $SO_2$ (ppb) | 2.87 | 2.66 | 1.28 | 1.09 | 9.88 | 1.42 |

SD-standard deviation; Min-minimum; Max-maximum; IQR-inter-quartile range



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**Table 2 Spearman's correlation coefficients between the concentrations of major ambient fungal spores and environmental parameters in 2015.**

| Variables | Ascospores | *Aspergillus/Penicillium* | *Arthrinium* | Basidiospores | *Cladosporium* | Smuts | Total spores |
|---|---|---|---|---|---|---|---|
| Temperature (°C) | 0.748*** | 0.737*** | 0.339*** | 0.784*** | 0.423*** | 0.701*** | 0.789*** |
| $T_d$ (°C) | 0.789*** | 0.726*** | 0.285*** | 0.787*** | 0.371*** | 0.680*** | 0.799*** |
| RH (%) | 0.049 | -0.183** | -0.298*** | -0.066 | -0.218*** | -0.183* | -0.068 |
| WS (m/s) | -0.483*** | -0.429*** | -0.320*** | -0.523*** | -0.493*** | -0.411*** | -0.554*** |
| Rainfall (mm) | 0.142* | -0.069 | -0.254*** | -0.095 | -0.273*** | -0.133* | -0.028 |
| Solar radiation ($MJ/m^2$) | 0.430*** | 0.506*** | 0.338*** | 0.529*** | 0.462*** | 0.488*** | 0.534*** |
| CO (ppm) | 0.186** | 0.135* | 0.283*** | 0.218*** | 0.390*** | 0.157** | 0.263*** |
| $NO_2$ (ppb) | 0.090 | 0.015 | 0.145* | 0.132* | 0.268*** | 0.020 | 0.150** |
| $O_3$(ppb) | -0.251*** | -0.189*** | -0.175** | -0.222*** | -0.174** | -0.188** | -0.259*** |
| $PM_{2.5}$ (μg/ $m^3$) | -0.093 | -0.086 | 0.208*** | -0.051 | 0.330*** | 0.053 | -0.007 |
| $PM_{2.5-10}$ (μg/ $m^3$) | 0.014 | 0.201*** | 0.242*** | 0.137* | 0.218*** | 0.226*** | 0.128* |
| $SO_2$ (ppb) | 0.143* | 0.216*** | 0.253*** | 0.291*** | 0.440*** | 0.273*** | 0.277*** |

*p<0.05; **P<0.01; ***P<0.001

Note: Temp-temperature ; $T_d$-dew point temperature ; RH-relative humidity ; WS-wind speed ; $O_3$-ozone ; CO-carbon monoxide ; $SO_2$-sulfur dioxide ; $NO_2$-nitrogen dioxide



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**Table 3 Multiple regression for major fungal spores and 0- , 1- and 2-day-lag environmental parameters in 2015.**

| Fungal categories ($log_{10}$ spores/$m^3$) | β coeff. (95% C.I.) | $R^2$ |
|---|---|---|
| Total spores | | |
| 0-day-lag $T_d$*** | 0.060 (0.054, 0.067) | 0.726 |
| 0-day-lag WS*** | -0.072 (-0.104, -0.040) | |
| 0-day-lag Solar radiation*** | 0.007 (0.002, 0.011) | |
| 0-day-lag CO*** | 0.322 (0.107, 0.538) | |
| Ascospores | | |
| 0-day-lag $T_d$*** | 0.068 (0.062, 0.075) | 0.680 |
| 0-day-lag WS*** | -0.088 (-0.115, -0.060) | |
| *Aspergillus/Penicillium* | | |
| 0-day-lag Temp*** | 0.041 (0.029, 0.054) | 0.582 |
| 0-day-lag WS*** | -0.072 (-0.105, -0.039) | |
| 2-day-lag $T_d$*** | 0.023 (0.011, 0.036) | |
| 0-day-lag $PM_{2.5-10}$*** | 0.009 (0.004, 0.014) | |
| *Arthrinium* | | |
| 0-day-lag Temp*** | 0.024 (0.015, 0.033) | 0.309 |
| 0-day-lag RH*** | -0.022 (-0.029, -0.016) | |
| 0-day-lag CO*** | 0.916 (0.622, 1.210) | |
| 1-day-lag $O_3$*** | -0.015 (-0.021, -0.008) | |
| Basidiospores | | |
| 0-day-lag $T_d$*** | 0.122 (0.113, 0.131) | 0.752 |
| 1-day-lag WS*** | -0.091 (-0.129, -0.052) | |
| 2-day-lag RH*** | 0.016 (0.010, 0.021) | |
| 0-day-lag $NO_2$*** | 0.024 (0.016, 0.032) | |
| 0-day-lag $O_3$*** | 0.017 (0.011, 0.023) | |
| *Cladosporium* | | |
| 0-day-lag WS*** | -0.154 (-0.194, -0.113) | 0.332 |
| 0-day-lag Solar radiation*** | 0.020 (0.013, 0.026) | |
| 0-day-lag $PM_{2.5-10}$*** | 0.012 (0.006, 0.018) | |
| Smuts | | |
| 0-day-lag Temp*** | 0.067 (0.059, 0.078) | 0.579 |
| 0-day-lag WS*** | -0.076 (-0.112, -0.040) | |
| 2-day-lag RH*** | 0.012 (0.006, 0.018) | |



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| | | |
|---|---|---|
| 2-day-lag Rainfall*** | -0.002 (-0.004, -0.001) | |
| 0-day-lag $PM_{2.5-10}$*** | 0.011 (0.006, 0.017) | |

**P<0.01; ***P<0.001

Note: Temp-temperature ; $T_d$-dew point temperature ; RH-relative humidity ; WS-wind speed ; $O_3$-ozone ; CO-carbon monoxide；$SO_2$-sulfur dioxide；$NO_2$-nitrogen dioxide



doi:10.6342/NTU201603517

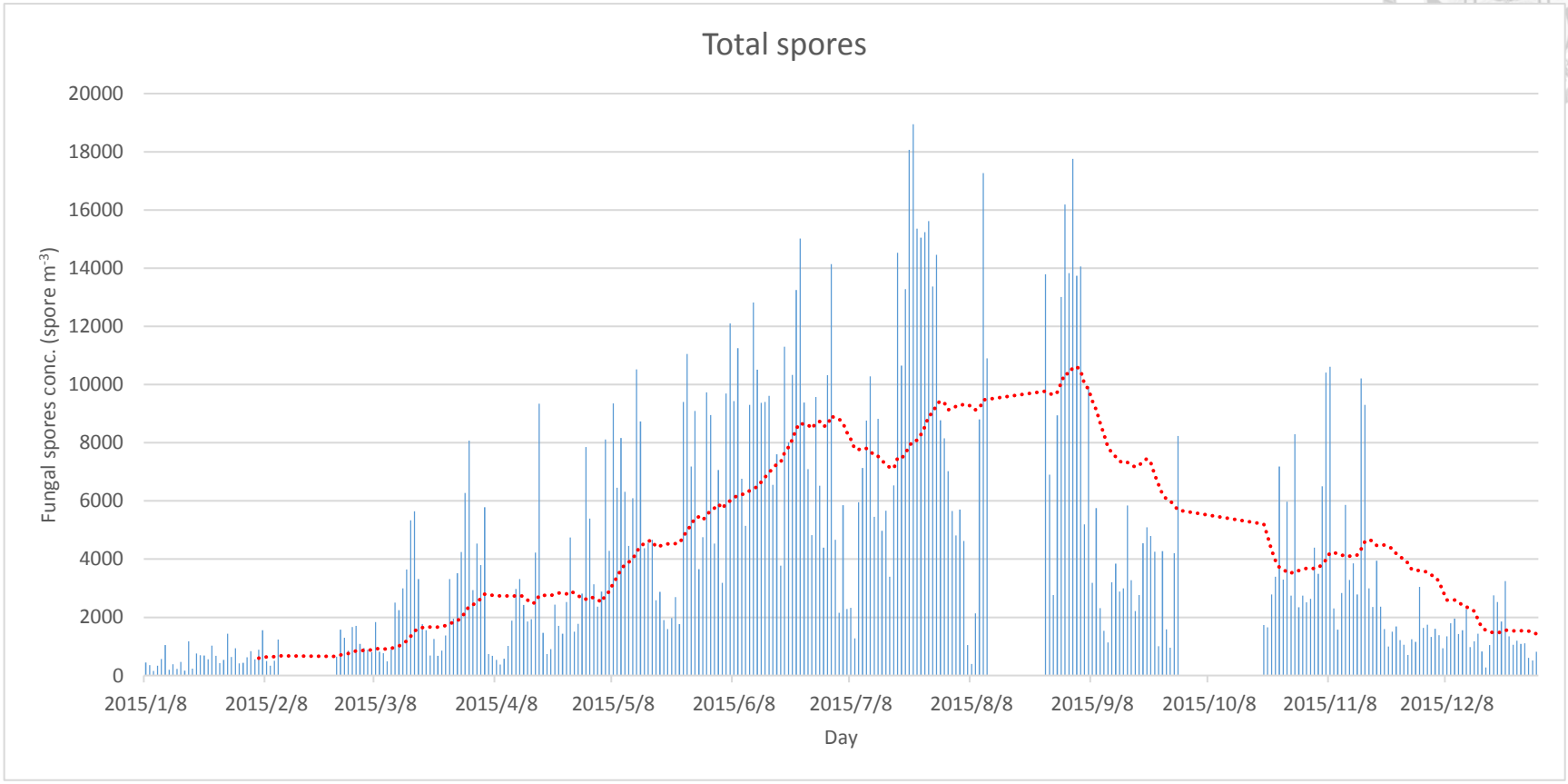



Figure 1. Temporal trend for daily concentration of total spores (spore $m^{-3}$) in Taipei during 2015.



doi:10.6342/NTU201603517

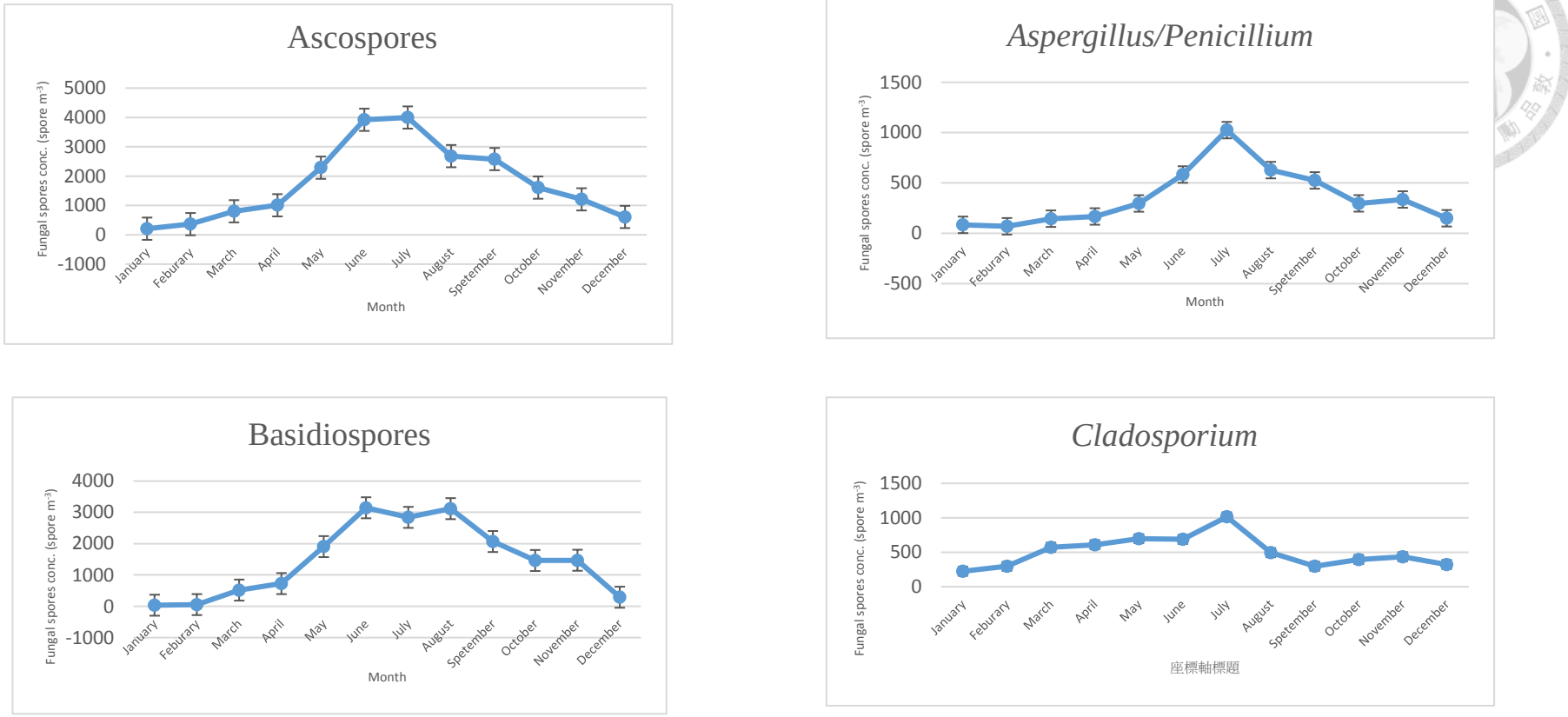



Figure 1a. Temporal trend for monthly concentration of Ascospores, *Aspergillus/Penicillium*, Basidiospores, *Cladosporium* (spore $m^{-3}$) in Taipei during 2015.



doi:10.6342/NTU201603517

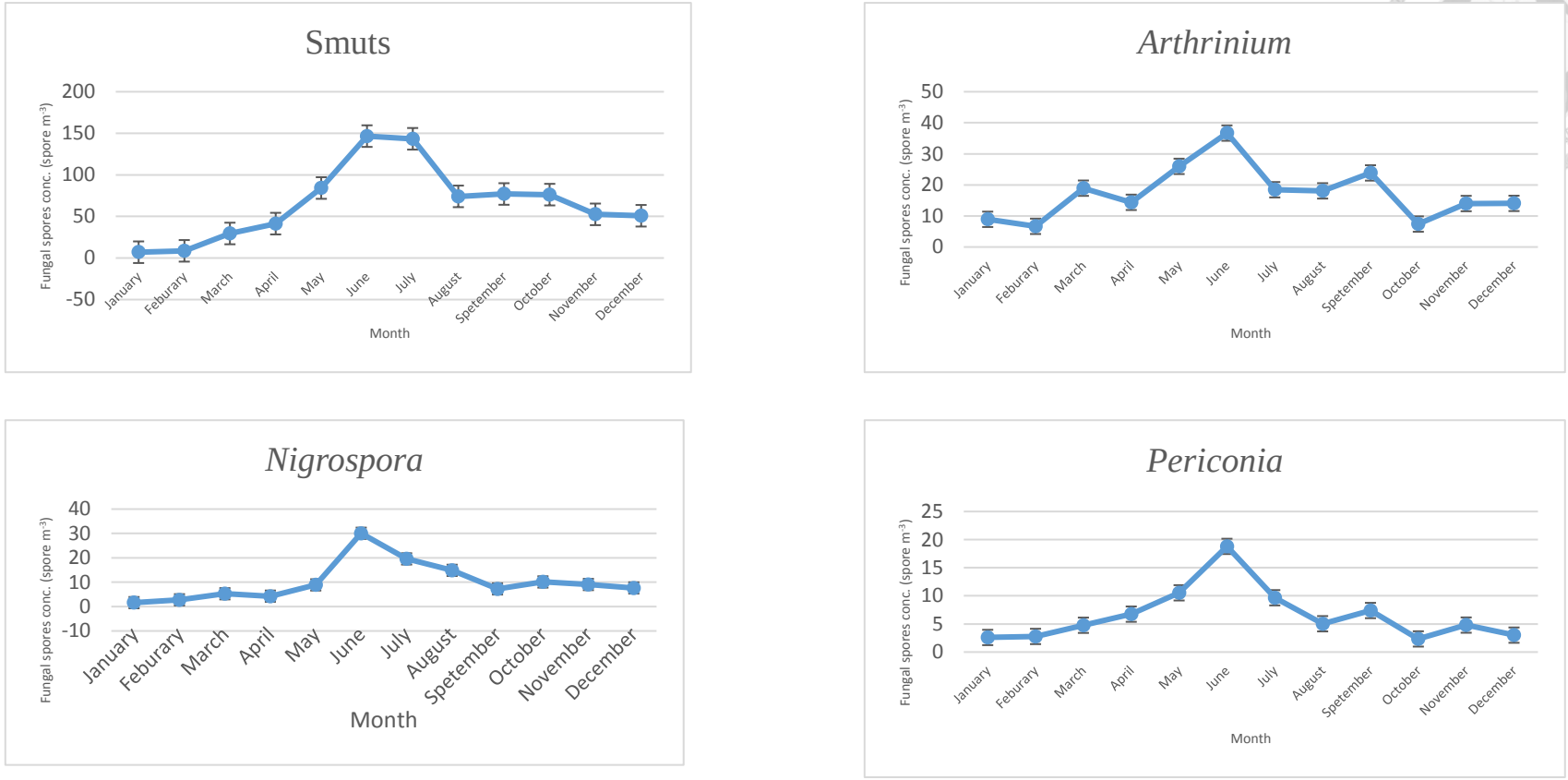
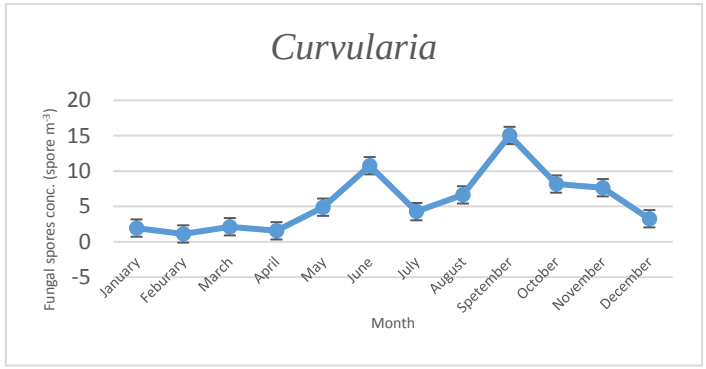
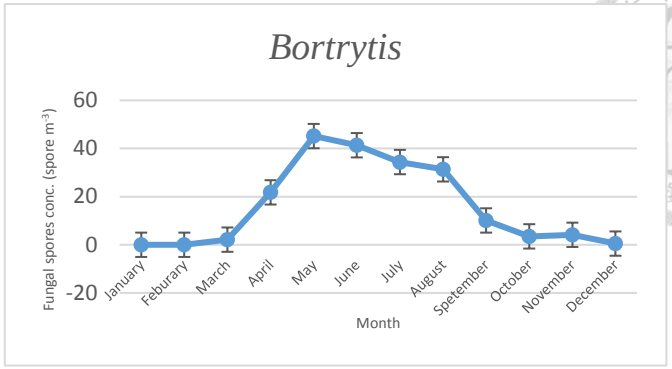



Figure 1b. Temporal trend for monthly concentration of Smuts, *Arthrinium*, *Nigrospora*, *Periconia* (spore $m^{-3}$) in Taipei during 2015.



doi:10.6342/NTU201603517



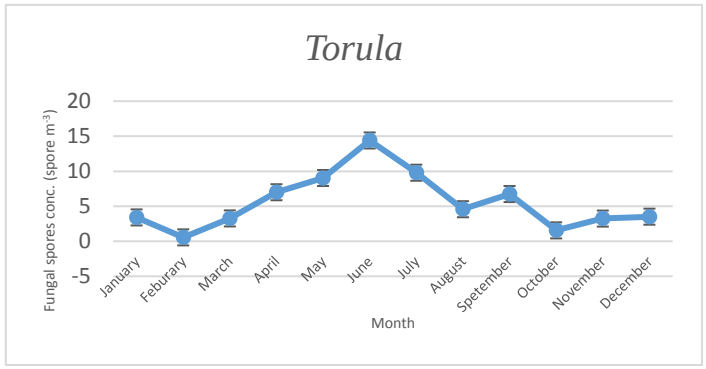




Figure 1c. Temporal trend for monthly concentration of *Curvularia*, *Bortrytis*, *Torula* (spore $m^{-3}$) in Taipei during 2015.



doi:10.6342/NTU201603517

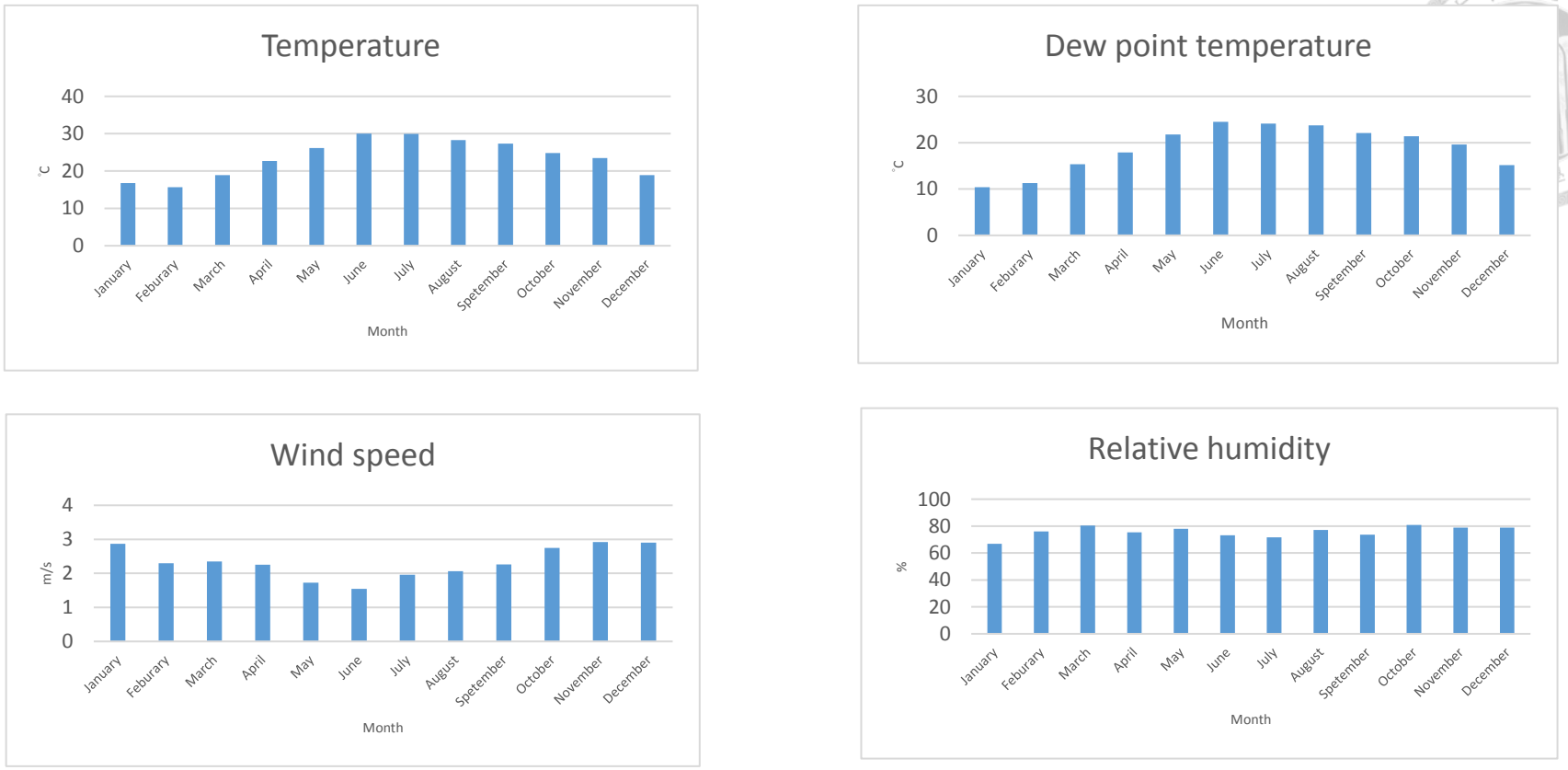
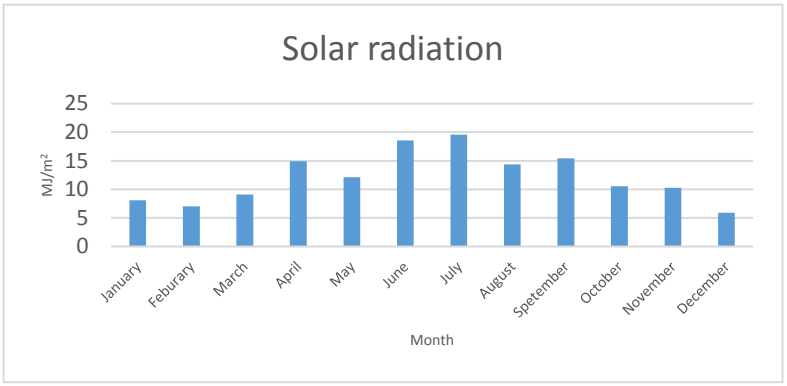



Figure 2. Monthly averages of meteorological parameters (Temperature, Dew point temperature, Wind speed, Relative humidity) in Taipei during 2015.



doi:10.6342/NTU201603517



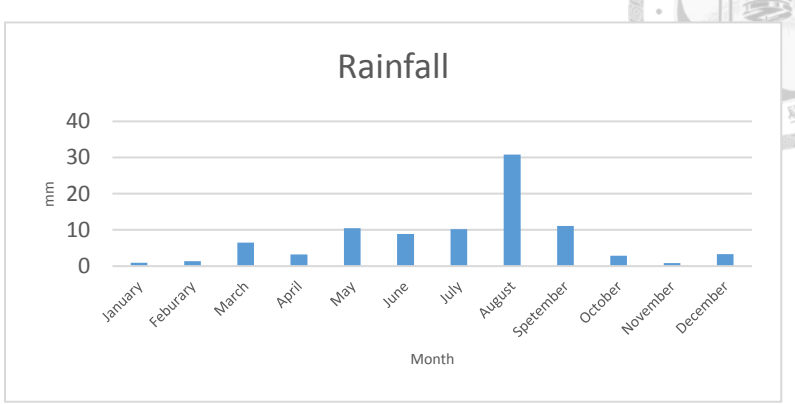




Figure 3. Monthly averages of meteorological parameters (Solar radiation, Rainfall) in Taipei during 2015.



doi:10.6342/NTU201603517

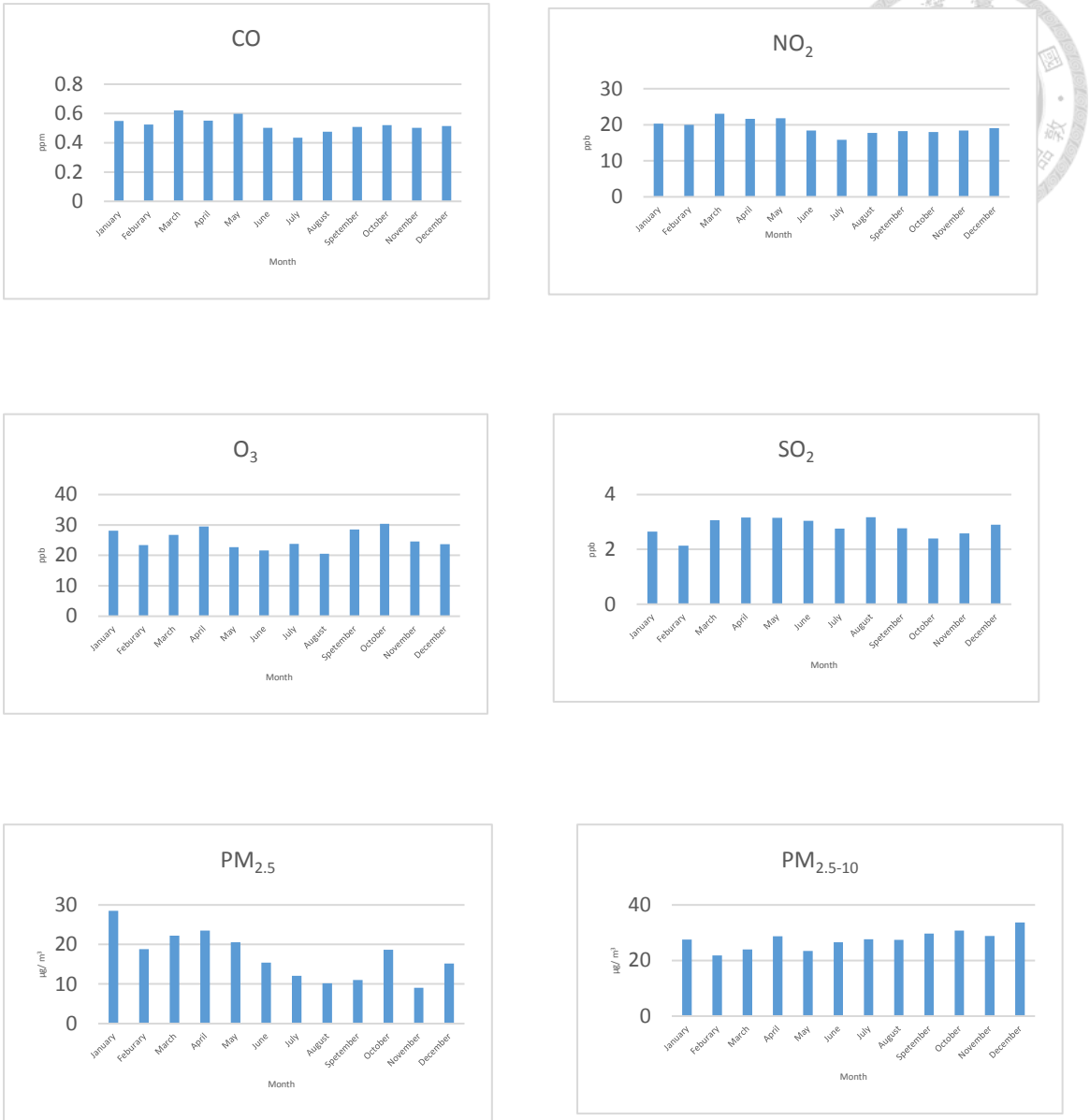



Figure 4. Monthly averages of air pollutants (CO, $NO_2$, $O_3$, $SO_2$, $PM_{2.5}$, $PM_{2.5-10}$) in Taipei during 2015.



doi:10.6342/NTU201603517