Aerosol and Air Quality Research, 12: 651–661, 2012

ISSN: 1680-8584 print / 2071-1409 online
doi: 10.4209/aaqr.2012.03.0070

Characterization of Indoor-Air Bioaerosols in Southern Taiwan

Yi-Chyun Hsu[1*], Pei-Yi Kung[2], Ting-Nien Wu[1], Yun-Hwei Shen[2]

[1] *Department of Environmental Engineering, Kun Shan University, Tainan 71003, Taiwan*
[2] *Department of Resource Engineering, National Cheng Kung University, Tainan 70101, Taiwan*

ABSTRACT

This study investigated indoor air quality (IAQ) at 39 public sites in southern Taiwan including hospitals, schools, office buildings, hypermarkets, libraries, railway stations, theaters, etc. Indoor air quality was preliminarily assessed using handy digital apparatus. Items detected include carbon dioxide (CO_2), carbon monoxide (CO), formaldehyde (HCHO), total volatile organic compounds (TVOCs), total bacteria counts (TBC), total fungi counts (TFC), PM_{10}, $PM_{2.5}$, ozone (O_3) and temperature. Based on the results of walk-through detection, the spatial distribution of indoor air contaminants was further measured over a 24 hour period using the EPA standard method. Major indoor air pollutants were found to include CO_2, TBC, and TFC. The measured CO_2 concentrations ranged between 438 and 1527 ppm, and only 38.9% of them met the Taiwan EPA suggested threshold of 600 ppm. In the schools and hospitals (Category 1), the measured TFC and TBC concentrations ranged from 62 to TNTC CFU/m^3 and from 196 to 4875 CFU/m^3, respectively. 33% TFC and 83% TBC concentrations exceeded the suggested threshold, and CO_2 concentrations were moderately correlated with TBC levels. In a case study of hospital bioaerosols, high TBC and TFC levels were effectively lowered through disinfectant housekeeping as well as ClO_2 spray. Three filamentous fungus genera were identified as *Cladosporium perangustum, Cladosporium tenuissimum*, and *Fusarium incarnatum* from outdoor samples with high TFC concentrations.

Keywords: Indoor air quality; Bioaerosol; Bacteria; Fungi.

INTRODUCTION

Life style, dietary habit and customer behaviour may directly influence in indoor air quality (IAQ) and the characteristics of indoor air pollutants. In general, people spend at least 80% of their time in various indoor spaces. Even though indoor air pollutants are present at trace concentrations, the potential threat to human health cannot be overlooked after long-term exposure in indoor environments. Poor IAQ has been shown to have adverse effects on human health (Fabian *et al*., 2005; Huboyo *et al.*, 2011). To improve IAQ and protect human health, the Taiwan EPA listed the suggested threshold levels of common indoor air pollutants in 2005. The suggested IAQ measurements include carbon dioxide (CO_2), carbon monoxide (CO), formaldehyde (HCHO), total volatile organic compounds (TVOCs), total bacteria counts (TBC), total fungi counts (TFC), PM_{10}, $PM_{2.5}$, ozone (O_3) and temperature. Regulated public sites including schools and hospitals comprise category 1, and stores, markets, office buildings, and transport stations comprise category 2 (Taiwan EPA, 2005).

Sources of indoor air pollutants may be brought about by contaminated outdoor air, air conditioning or office equipment, human activity, building components and furnishings, and other accidental events (USEPA, 1991; Lai *et al*., 2010; McNamara *et al.*, 2011). Particulate matter frequently couples with compounds of biological origins creating bioaerosol that may ranges in aerodynamic diameter from 0.01 and 100 μm (Cox and Wathes, 1995; Macher, 1999). Indoor air contains a complex mixture of bioaerosols, such as bacteria and fungi, and non-biological particles such as dust, tobacco smoke, and cooking-generated particles (Kalogerakis *et al*., 2005; Cao *et al*., 2011). The prevailing microorganisms in the air of closed spaces are bacteria, and the majority of bacteria sources are come from the presence of humans in non-industrial indoor environments (Stezenbach, 1997; Kalogerakis *et al.*, 2005; Singh *et al*., 2011). The predominant microorganism in ambient air is fungi, and the majority of fungal sources are soil, plants, and water bodies outdoors (Karbowska-Berent *et al*., 2011). Fungal spores have tiny aerodynamic diameters, high environmental durability, and are carried by air currents (Macher, 1999). The spore transport diffusion mechanism was controlled by meteorological conditions such as dew point, relative humidity (RH), temperature, and airflow turbidity. The level of airborne fungi is closely associated

* Corresponding author. Tel.: +886-6-2055952;
Fax: +886-6-2050540
E-mail address: ychsu22@yahoo.com.tw

with indoor RH, and the optimal condition for fungal growth may be above 70% RH (Burge *et al*., 1995). The presence of bioaerosols with higher concentrations was observed in the optimum temperature range of 30 to 40°C (Deguchi and Yoshizawa, 1996).

Because Taiwan is a warm humid island, bioaerosol concentrations are relatively high (Tseng *et al.*, 2011). An average indoor bacteria concentration of 1502 CFU/m^3 and indoor fungi concentration of 195 CFU/m^3 were reported for Taipei (Li, 1998). The predominant allergens in Taiwan may be caused by indoor airborne fungi that usually originate outdoors (Wu *et al*., 2000). A case study in Singapore illustrated that bioaerosols consist of 50.5% bacteria and 49.5% fungi indoors, and 20.6% bacteria and 79.4% fungi outdoors (Rajasekar and Balasubramanian, 2011). According to the hygroscopic effects of airborne fungi in Taiwan, outdoor airborne fungi (2.58 μm in summer, 2.79 μm in winter) have greater geometric mean diameters than indoor airborne fungi (1.91 μm in summer, 1.73 μm in winter). Indoor/outdoor (I/O) ratios of airborne fungal concentrations are between 0.29 and 0.58 without indoor sources during the summer, while I/O concentrations drop to 0.12 and 0.16 during the winter (Liao *et al*., 2004). Airborne bioaerosols including bacteria and fungi can be toxigenic, allergenic, and infectious (Fabian *et al.*, 2005). Human exposure to airborne fungal spores might cause adverse health effects, especially respiratory symptoms (Su *et al*., 2001). More than 80 fungal genera were reported to be linked to symptoms of respiratory tract allergies (Horner *et al.*, 1995). *Cladosporium* spp., *Alternaria* spp., *Aspergillus* spp., and *Fusarium* spp. are amongst the most common allergenic genera, and metabolites of fungi are also believed to irritate the respiratory system (Kalogerakis *et al.*, 2005). Accordingly, bioaerosol characterization has become an important issue due to its related health effects. This study was aimed at discovering common indoor air pollutants, examining the characteristics of indoor bioaerosols, and evaluating the relationship between indoor air pollutants and bioaerosols based on IAQ investigation of public sites from the southern Taiwan.

MATERIALS AND METHODS

Study Area

According to the Taiwan EPA's suggested threshold levels for common indoor air pollutants, regulated public sites are classified into two categories: category 1 includes hospitals, health care centres, schools, tutoring centres, kindergartens, and nursing centres, and category 2 includes office buildings, banks, restaurants, supermarkets, theatres, stores, and transport stations. This study selected 39 public sites in Tainan city including 8 hospitals, 7 tutoring centres, and 3 kindergartens to represent Category 1, and 5 supermarkets, 2 transport stations, 10 office buildings and 4 theatres to represent Category 2, and investigated their IAQ.

IAQ Investigation

An IAQ survey was implemented in 2 stages, and the reported investigation data was collected during 2009 and 2010. In the first stage, various handy digital apparatuses were utilized to preliminarily screen IAQ hot-spot locations in each selected public site through walk-through detection. Levels of CO_2, CO, and O_3 were quantified using a *KD Engineering AirBoxx* detector. TVOC concentration was detected using a *ToxiRAE PGM-30* PID detector, and HCHO concentration was detected using a *PPM Technology htV-M* formaldehyde meter. Levels of PM_{10} and $PM_{2.5}$ were quantified using a *Met one Instruments AEROCET 531* detector. Based on the space arrangement, 8 to 20 locations were selected for IAQ walk-through detection. A single detection was conducted for 2 min at a height of 1.2 to 1.5 m. According to the first stage IAQ survey, the concentration contours were used to examine the distributions of indoor air pollutants at each site and to identify the IAQ hot-spot locations among the investigated public sites (Wang *et al*., 2011).

The identified IAQ hot-spot location was subsequently subjected to the second stage IAQ survey. Following Taiwan EPA standard methods, the indoor air pollutants were continuously monitored for 24 hours or sampled for lab analysis. Concentrations of CO_2 and CO were measured using the non-dispersive infrared method, and O_3 was quantified using the ultra-violet absorption method. TVOCs were sampled using a canister and analysed using a HP 6890 gas chromatography equipped with a FID detector (GC/FID). The quantitative analysis of HCHO applied 2,4-dinitrophenylhydrazine (DNPH) absorption and was analysed using HPLC. Concentrations of PM_{10} and $PM_{2.5}$ were determined using the β-ray decay method. The measurements of indoor air quality by using handy digital apparatuses and the Taiwan EPA standard methods have been compared by Kung *et al*. (2011).

Bioaerosol Analysis

The numeration of TBC and TFC were sampled and analysed based on the Taiwan EPA's standard methods, NIEA E301.11C and NIEA E401.11C, respectively. Air samples were suctioned with a SKC Quick TAKE 30 impacter sampler with 400 holes 0.25 mm in diameter. The drawing air-stream had a flow rate of 28.3 L/min and was directed onto an agar surface in a standard Petri dish. Tryptic soy agar (TSA) consisting of 15 g/L tryptone, 5 g/L soytone, 5 g/L NaCl, and 15 g/L agar was used for the bacterial collection. Malt extract agar (MEA) consisting of 12.75 g/L maltose, 2.75 g/L dextrin, 2.35 g/L glycerol, 0.78 g/L peptone, and 15 g/L agar was used for fungal collection. The TSA and MEA mediums were sterilized before they were placed in an autoclave operating at 121°C and 1.2 kg/cm^2 for 20 min. The incubation periods were 48 hours at 30°C for bacterial growth and 4 days at 25°C for fungal growth. Bioaerosol concentrations as CFU/m^3 were calculated by dividing CFU counts by the sampling flow rate and sampling time using positive-hole conversion tables.

According to the appearance, colour, and distribution of fungal growth on the agar plate, flourishing colonies were selected for the identification of fungal genera by the Bioresource Collection and Research Centre. The identification procedures were followed by genus selection, separation, purification, microscopic observation, and

identification using the rDNA ITS1-5.8S-ITS2 sequence.

RESULTS AND DISCUSSION

Measurements of Indoor Air Quality

The results of the IAQ investigation at 39 public sites are summarized in Table 1. Levels of HCHO and $PM_{2.5}$ in Category 1 spaces and levels of CO, O_3, HCHO, PM_{10}, and $PM_{2.5}$ in Category 2 spaces met the Taiwan EPA's suggested threshold. As illustrated in Fig. 1, the common IAQ measurements that exceeded the Taiwan EPA's suggested threshold included CO_2, TVOC, TFC, and TBC in both Category 1 and Category 2. CO concentrations at hospitals were found to exceed the Category 1 threshold of 2 ppm. The cause of the high CO concentration may be ascribed to emissions from vehicle exhaust resulting on hospitals' central high-traffic locations. Tutoring centers and kindergartens were found to exceed the Category 1 O_3 threshold of 0.03 ppm and PM_{10} threshold of 60 ppm. The highest O_3 concentration occurred around noon (AM 11:00 to PM 2:00), and the O_3 source was most likely from outdoor air due to the absence of potentially O_3-generating electric facility indoors. The high PM_{10} levels at tutoring centers and kindergartens were also caused by outdoor air, while the high PM_{10} levels at hospitals may be associated with air conditioning, therapeutic action, and patient motion.

In general, CO_2 concentrations above 1000 ppm indicate poor ventilation of the indoor environment and can be remedied by improving ventilation (Lee and Chang, 2000). In Fig. 1, CO_2 exceeded the Taiwan EPA's suggested threshold by 61.1% in Category 1 spaces and 9.5% in Category 2 spaces. A Category 1 CO_2 threshold of 600 ppm may be too strict for most public sites. Air conditioning usually employs constant recycling based to save electricity, thus ventilation may be insufficient to lower indoor CO_2 levels when crowds gather indoors.

TVOC concentrations exceeding the suggested threshold of 3 ppm occurred in tutoring centers, office buildings, and fitness centers. The fitness centre is located in the basements of buildings, and the heating boiler is regarded as potential sources of indoor TVOCs. In tutoring centers and office buildings, high levels of TVOCs may be caused by new furniture and vehicle exhaust from outdoor air (Panagopoulos *et al.*, 2011).

TBC and TFC levels that exceed the suggested threshold are a common IAQ problem among the investigated public sites. The differences of TBC as well as TFC between Category 1 and Category 2 were examined with the aid of statistical tools, F-test and T-test. The theory test of TBC showed that the p-values are 0.193 (> 0.05) for F-test and 0.063 (> 0.05) for T-test. This result implicated that there is no significant differences on variance and mean of TBC between Category 1 and Category 2. For theory test of TFC, both p-values of F-test (9.7×10^{-6}) and T-test (0.0016) are smaller than the significant level 0.05. Thus, there exists the significant difference of TFC between Category 1 and Category 2. High TBC levels may be caused by outdoor pollutants and indoor crowds. Other factors that can also affect indoor TBC concentrations

Table 1. Results of IAQ investigations at 39 public sites.

Category		Category 1			Category 2			
Measurement	Suggested Threshold	Hospital (8)	Tutoring Center (7)	Kindergarten (3)	Hypermarket (5)	Transport Station (2)	Office Building (10)	Theater (4)
CO_2 (ppm)	600	527–**1022***	455–**1527***	438–**673***	573–**1183***	632–890	488–**1164***	618–736
	1000							
CO (ppm)	2	0.55–**2.14***	0.64–1.72	0.87–1.01	0.90–1.75	0.30–0.96	0.49–1.39	0.83–5.42
	9							
O_3 (ppm)	0.03	0.005–0.016	0.008–**0.072***	0.021–**0.039***	0.003–0.031	0.006–0.024	0.009–0.041	0.009–0.045
	0.05							
TVOC (ppm)	3	0.32–3.0	1.35–**4.0***	0.9–2.2	0.38–1.7	0.37–2.4	< 0.20–**3.6***	ND–**5.6***
HCHO (ppm)	0.1	ND–0.01	0.01–0.02	ND–0.01	ND–0.01	ND	ND–0.05	ND–0.05
PM_{10} ($\mu g/m^3$)	60	22–**90***	42–**116***	81–**97***	11–51	22–30	15–110	20–32
	150							
$PM_{2.5}$ ($\mu g/m^3$)	100	5–35	14–42	30–36	4–20	4–9	5–45	7–15
TBC (CFU/m^3)	500	958–**3025***	520–**2800***	196–**4875***	200–**3183***	733–966	178–**4125***	898–**1384***
	1000							
TFC (CFU/m^3)	1000	406–**1418***	62–**TNTC***	803–**3246***	132–**1161***	655–**1895***	107–**4500***	335–438

1. * indicates exceeding suggested threshold; 2. ND means not detectable; 3. TNTC means too numerous to count.

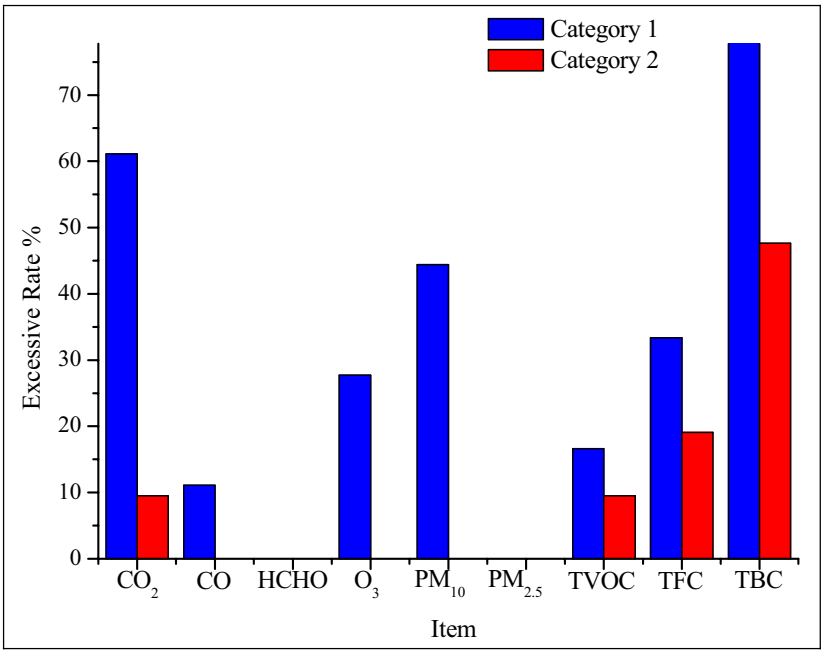


Fig. 1. Ratio excess rates of IAQ measurements at public sites of Category 1 and Category 2.

include the cleaning of air conditioning systems, disinfectant housekeeping, and control of the ventilation rate. In addition to indoor green plants, the majority of fungal sources that contribute to high indoor TFC levels may be in the surrounding outdoor area.

Correlationship between Indoor Bioaerosols and Particulate Matters

The TBC, TFC, $PM_{2.5}$ and PM_{10} concentrations were subjected to statistical linear regression analysis to examine the correlationships among them. Fig. 2 illustrated that regression coefficients were 0.12 for TBC and $PM_{2.5}$, 0.08 for TBC and PM_{10}, 0.19 for TFC and $PM_{2.5}$, and 0.27 for TFC and PM_{10}. All regression coefficients were below 0.4, indicating that indoor bioaerosols and particulate matters may have only low correlationships. Indoor air may contain bioaerosols and non-biological particles, and thus the low correlationship between bioaerosols and particulate matters implies that the percentage of bioaerosols could be low in indoor air. A comparison of the regression coefficients of TBC and particulate matters shows that TFC had higher regression coefficients and slopes of regression line for both $PM_{2.5}$ and PM_{10}. This suggests that particulate matter might contribute more to TFC than TBC. In light of the reported indoor/outdoor (I/O) $PM_{2.5}$ ratio of 0.93, indoor $PM_{2.5}$ could reasonably ascribed to outdoor sources in roadside areas (Huang *et al*., 2007). This finding is also consistent with the fact that most TFC originates outdoors (Wu *et al.*, 2000; Karbowska-Berent *et al*., 2011). In certain residences close to high traffic roads, outdoor pollutant penetration should be noted (Lai *et al.*, 2010).

Correlationship between Indoor Bioaerosols and CO_2

In order to depict the correlationships between TBC and CO_2 as well as TFC and CO_2, the results of the statistical linear regression analysis are illustrated in Fig. 3. A moderate correlationship between TBC and CO_2 was observed based on its regression coefficient of 0.44, while TFC and CO_2 had a low correlationship with a regression coefficient of 0.16. Kim *et al.* (2009) pointed out that microbial activity can be affected by CO_2 and RH, and Kalogerakis *et al.* (2005) claimed humans are the major source of bacteria in closed spaces. The literature also agrees with our result that TBC concentrations are moderately linked to CO_2 levels (Kim *et al*., 2009). Accordingly, TBC concentrations can be potentially affected by the intensity of indoor human activity and control of the ventilation rate.

Control of Indoor TBC and TFC

One hospital with an excessively high level of bioaerosols was selected for observation of the temporal transience of bioaerosols. Baseline IAQ concentrations were measured on September 2, 2009, and the concentrations of CO_2 and TBC exceeded the Taiwan EPA's suggested threshold. The ventilation period was increased and alcohol and NaOCl were used for disinfection to improve the IAQ. As shown in Table 2, IAQ measurements on January 7, 2010 showed that the CO_2 concentration dropped from 944 to 620 ppm and the TBC level dropped from 2935 to 823 CFU/m^3, which were both still higher than their respective Category 1 thresholds. Also, the measured TFC level was 1674 CFU/m^3 higher than the Category 1 threshold. After thorough disinfectant housekeeping, the TBC level decreased to 277 CFU/m^3, while the TFC level increased to 6701 CFU/m^3 based on IAQ measurements on January 25, 2010. In order to evaluate the cause of the high TFC level, indoor TFC and outdoor TFC were measured on March 5, 2010. The measured TFC concentrations were 569 CFU/m^3 in the lobby, 1422 CFU/m^3 outdoors, 1713 CFU/m^3 in the ventilation intake, and 1901 CFU/m^3 in the

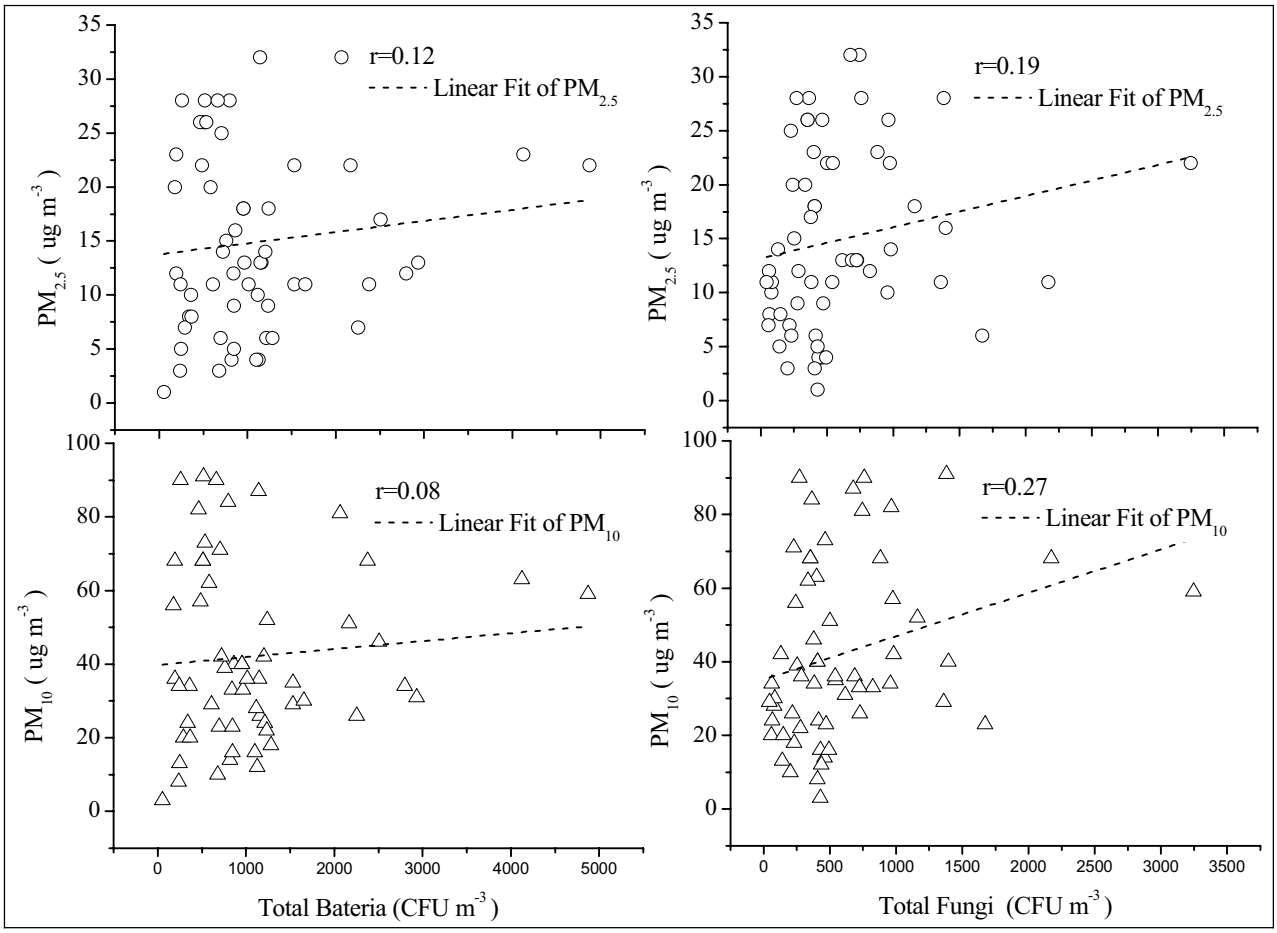


Fig. 2. Linear regression plots of bioaerosols and particulate matters.

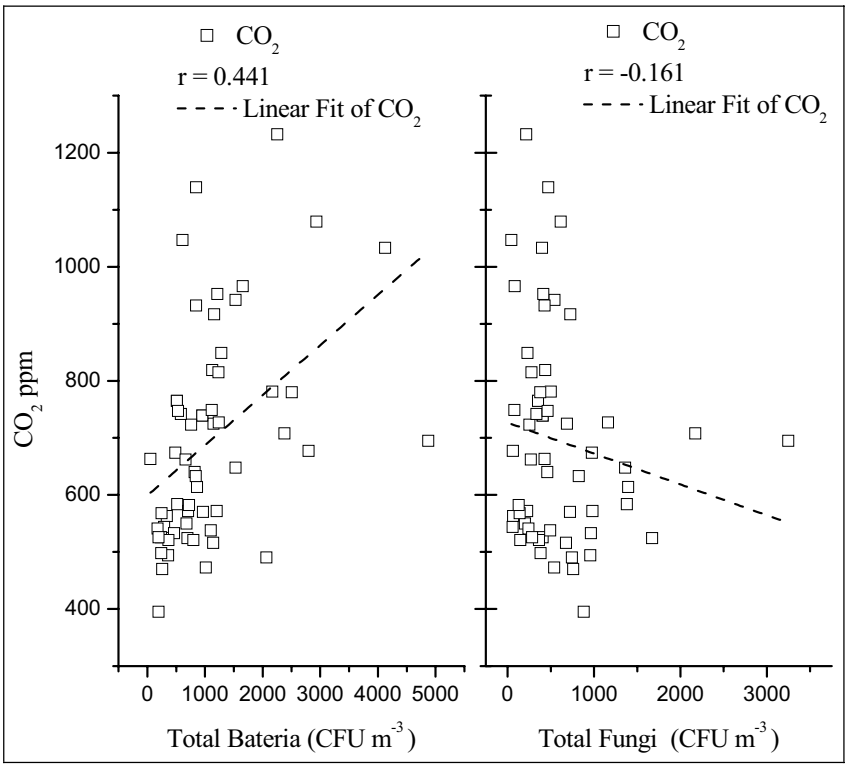


Fig. 3. Linear regression plots of bioaerosols and CO_2.

Table 2. Exceeding IAQ measurements and improving actions in a hospital.

Measurements / Date of measurements	CO_2 (ppm)	TBC (CFU/m^3)	TFC (CFU/m^3)	Improving Action
Suggested threshold	600	500	1000	
2009.09.02	944	2935	729	Before improvement
2010.01.07	620	823	1674	1. increase of ventilation period 2.using alcohol and NaOCl as disinfectant
2010.01.25	—	277	6701	
2010.03.05	—	—	569 (lobby) 1422 (outdoor air) 1713 (intake of ventilation) 1901 (ventilation exhaust)	Disinfectant housekeeping
2010.03.25	—	—	TNTC (lobby) TNTC (outdoor air) TNTC (intake of ventilation) TNTC (ventilation exhaust)	

ventilation exhaust. Thus, it was confirmed that the TFC came from outdoors. The TFC measurement was checked again on March 25, 2010, and all TFC measurements were too numerous to count (TNTC).

Increasing the ventilation period and disinfectant housekeeping can effectively control the TBC concentration, but not TFC concentration. In Table 2, the measured TFC levels in January and March were much higher than the measurement in September. The extremely high TFC levels may be ascribed to dust storms and furrowing activity on nearby farmland that increased the amount of fungal spores in the outdoor air, which then moved indoors. Consequently, the temporal transience of TFC concentration is mainly affected by the outdoor environment and seasonal climate change.

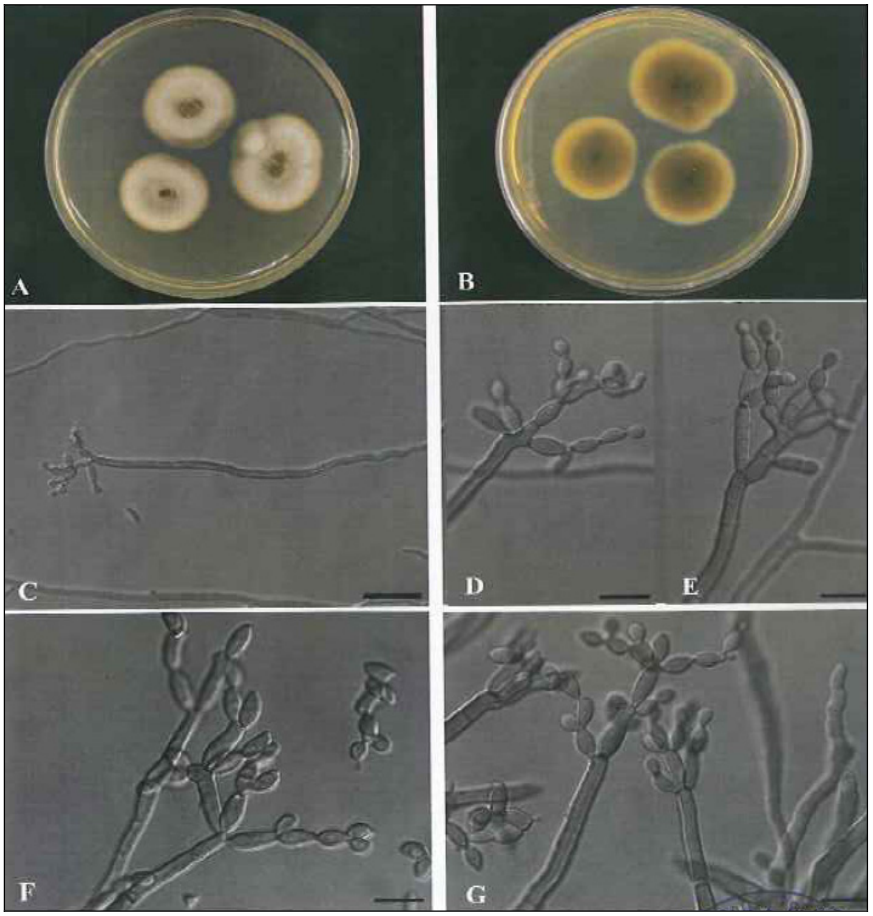
Several aerobiological technologies have been proposed to control indoor bioaerosols such as ultraviolet germicidal irradiation, photocatalytic oxidation, ionization, ozone, negative ion, etc. (Huang *et al.*, 2012). In this study, ClO_2 was used as a disinfectant to control the indoor TFC level. With ClO_2 spraying, the concentration of TFC effectively decreased from TNTC to 3071 CFU/m^3 after 0.5 hours, but jumped back to TNTC after 1 hour. The result of this disinfection illustrates that ClO_2 spraying can effectively control indoor TFC levels when done properly. The dose of disinfectant, the span of spraying, and the frequency of disinfection are all important to maintaining good IAQ conditions

Identification of Fungal Genera

MEA agar plates sampled on March 25, 2010 were selected according to their appearance, color, and the distribution of fungal growth and were sent to Bioresource Collection and Research Centre for identification of fungal genera. Three identified filamentous fungus genera included *Cladosporium perangustum* (Fig. 4), *Cladosporium tenuissimum* (Fig. 5), and *Fusarium incarnatum* (Fig. 6). Kendrick (1990) listed the big eight genera of molds as *Alternaria* spp., *Cladosporium* spp., *Curvularia* spp., *Drechslera* spp., *Epicoccum* spp., *Fusarium* spp., *Nigrospora* spp. and *Stemphylium* spp., because of their allergenicity and frequency of appearance in air. In Beijing, the dominant fungi were *Cladosporium* spp., *Alternaria* spp., *Penicillium* spp. *and Aspergillus* spp. in the outdoors (Fang *et al.*, 2005); *Fusarium* spp., *Aspergillus* spp., *Penicillium* spp. and *Basipetospora* spp. were found in west Korea during the Asian dust periods (Yeo and Kim, 2002). In Turkey, the prevailing mold genera were *Cladosporium* spp., *Aspergillus* spp., *Penicilliu,* spp. and *Rhizopus* spp. (Mentese *et al.*, 2009). In this study, *Cladosporium* and *Fusarium* were the most common allergens in Taiwan, which were usually present in soils, withered fallings, leaves' surfaces, and moldy plants. The Taiwan EPA (2012) reported the dominant fungi to be *Cladosporium* spp., *Aspergillus* spp., *Penicillium* spp., and *Curvularia* spp. in an IAQ investigation of schools and kindergartens. An I/O TFC ratio of 1.09 and I/O dominant fungal ratio of 1.0 imply that indoor fungi come from the outdoor sources (Taiwan EPA, 2012). Based on our fungal observation, the numeration of *Cladosporium* is estimated to comprise 70–80% of all fungal genera, which is higher than the reported *Cladosporium* indoor dominance of 48.8% by the Taiwan EPA (2012). The generic classifications of *Cladosporium* are *Cladosporium perangustum* and *Cladosporium tenuissimum*, which represent the dominant fungal genera in southern Taiwan. *Fusarium* is estimated to comprise 5–6% of all fungal genera that can cause plant disease and harm to 70 to 80 economic plants. *Fusarium* mostly originates outdoors, although there are some potential sources indoors as well (Taiwan EPA, 2012). In this case study, furrowing activity may have caused adsorbed microorganisms in soil and plants to be released into the outdoor air, which is our explanation of high TFC levels and identified dominant fungal genera.

CONCLUSIONS

This study investigated 39 public sites in southern Taiwan. IAQ measurements commonly exceeding the Taiwan EPA's suggested threshold included CO_2, TBC, and TFC. TBC

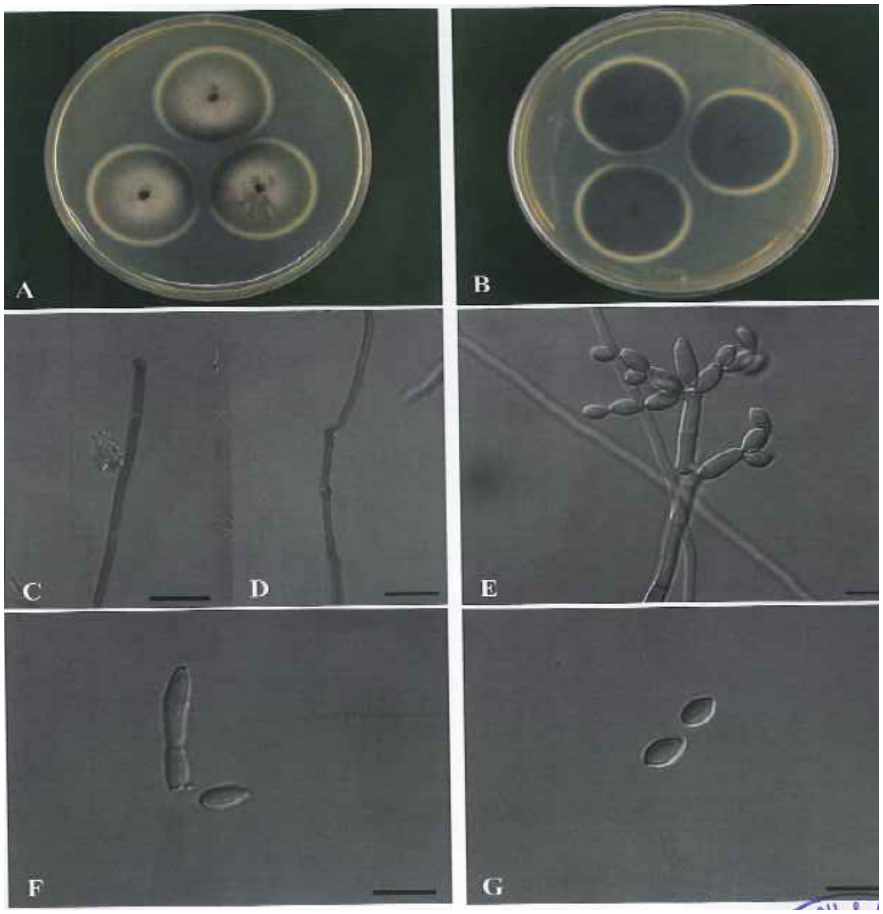


DNA gene segment of translation elongation factor 1-alpha:
TCATCGAGAAGTTCGAGAAGGTGAGCCTTACTTCACGGCAATGG
CGTCCTCTTCGCAGTGTTGTACCCCTCCGCCCCACCGCGCGCCAC
CCCACCTCGTCGCAATCTGCGATAAGATGTGGGACACGACTTGGC
CGGGCACGGACAACACTGCGATGAAGACTTCATCGCCACACGCC
ATCACCATCACAATACATTGCTAACAACCCTCCCAGGAAGCCGCT
GAACTCGGTAAGG
DNA gene segment of actin:
CGGTTTCGCCGGTGACGATGCGCCCAGAGCCGTTTTCCGTAAGTT
TGCAGACACCTGTTTCGCCGAGCTCAGAACATCCAGCTGACAGT
CTCTTAGCTTCCATTGTCGGTCGTCCCCGTCACCATGGGTATGCAC
TCCTCCCGCAAGCTCCGCCCTCGAGCCCAGATGTCTAACAGCAGC
GCAGTATCATGATCGGTATGGGCCA

Fig. 4. Morphological characteristics of *Cladosporium perangustum* (A-B: colonies on MEA agar plate incubating at 20°C for 7 days; C-G: fungal spore structure,C: bar = 25 μm, D-G: ba = 10 μm).

and TFC concentrations were weakly linked to particulate matters due to the low percentages of bioaerosols in indoor air. Particulate matters might contribute more to TFC than TBC. This is ascribed to the fact that most TFC originates outdoors. TBC concentration was moderately correlated with CO_2 level, and thus TBC concentrations can be potentially affected by the intensity of indoor human activity and ventilation rate. A case study of bioaerosols in a hospital found extremely high TFC levels that may be ascribed to the occurrence of dust storms and furrowing activities on the nearby farmland. The temporal transience of TFC concentrations is mainly affected by the outdoor environment and seasonal climate change, and ClO_2 spraying can effectively control the indoor TFC level when executed properly. The high-TFC outdoor air samples contained three filamentous fungus genera: *Cladosporium perangustum*, *Cladosporium tenuissimum* and *Fusarium incarnatum,* which are the most common allergens in Taiwan. The dominant fungal genus was *Cladosporium* (estimated 70–80%) in indoor air, which is a similar finding in other countries. Comparatively, indoor *Fusarium* (estimated 5–6%) is not so frequently found in other

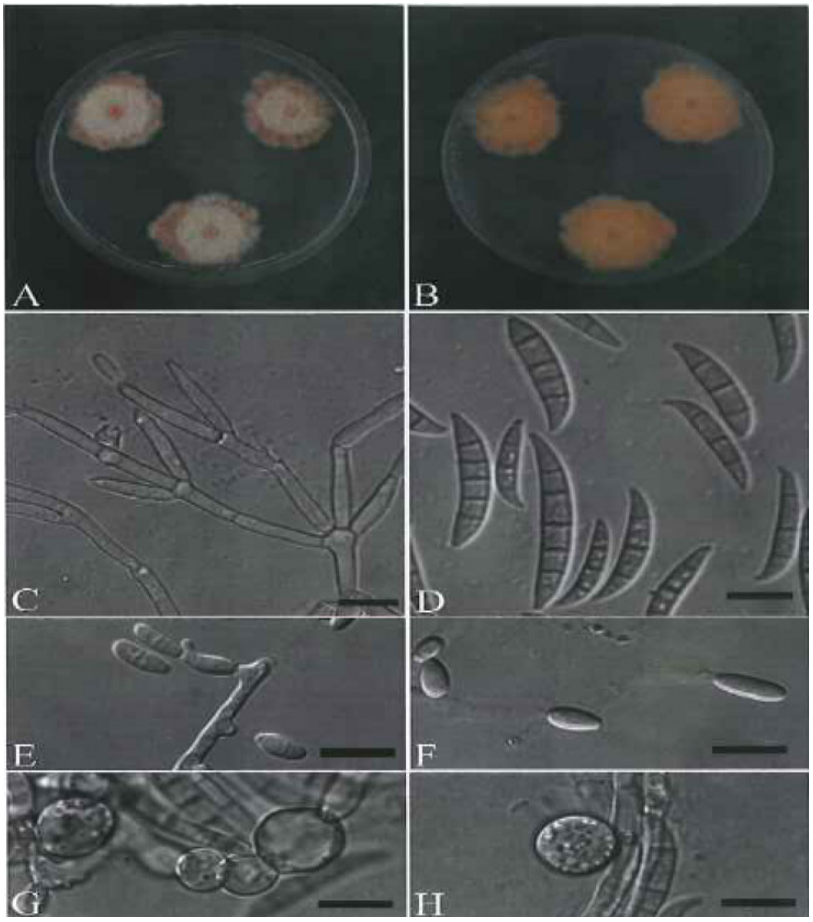
countries. An IAQ investigation including outdoor TBC and TFC measurements is suggested for examining the potential pollutant sources. The humid and warm weather in Taiwan is favorable to fungal growth and transport, and increasing ventilation rates and regular disinfectant housekeeping can effectively control concentrations of indoor bioaerosols.



DNA gene segment of rDNA ITSI-5.8S-ITS:
TCGTAACAAGGTCTCCGTAGGTGAACCTGCGGAGGGATCATTACA
AGTGACCCCGGTCTAACCACCGGGATGTTCATAACCCTTTGTTGT
CCGACTCTGTTGCCTCCGGGGCGACCCTGCCTTCGGGCGGGGGC
TCCGGGTGGACACTTCAAACTCTTGCGTAACTTTGCAGTCTGAGT
AAACTTAATTAATAAATTAAAACTTTTAACAACGGATCTCTTGGTT
CTGGCATCGATGAAGAACGCAGCGAAATGCGATAAGTAATGTGAA
TTGCAGAATTCAGTGAATCATCGAATCTTTGAACGCACATTGCGC
CCCCTGGTATTCCGGGGGGCATGCCTGTTCGAGCGTCATTTCACC
ATCTAAGCCTCGCTTGGTATTGGGCAACGCGGTCCGCCGCGTGCC
TCAAATCGACCGGCTGGGTCTTCTGTCCCCTAAGCGTTGTGGAAA
CTATTCGCTAAAGGGTGCTCGGGAGGCTACGCCGTAAAACAAAC
CCATTTCTAAGGTTGACCTCGGATCAGGTAGGGATACCCGCTGAA
CTTAAGCATATCA

DNA gene segment of actin:
TATGTGCAAGGCCGGTTTCGCCGGTGACGATGCGCCCAGAGCCG
TTTTCCGTAAGTCTGAAGACACCTGTTTCGCCCGCCCATGAATCC
CGAGCTGACACCCTTCCCAGCTTCCATTGTTGGCAGACCCCGTCA
CCATGGGTATGCATTCTCCCTGCGAACCCCTCCATCGCGCGCAGC
CAGTTCTAACCCCTCCGCAGTATCATGATCGGTATGGGCCAGAAG
CACTCGTA

Fig. 5. Morphological characteristics of *Cladosporium tenuissimum* (A-B: colonies on MEA agar plate incubating at 20°C for 7 days; C-G: fungal spore structure,C: bar = 25 μm, D-G: bar = 10 μm).



DNA gene segment of translation elongation factor 1-alpha:
AGACTCACCTTAACGTCGTCGTCATCGGCCACGTCGACTCEGGCA
AGTCGACCACTGTGAGTACTACCCACGATGATCTGCTTATCAGTA
GTCATCAACCCCGCCAGATGTGGCGGGGTAAGTTCAACTGGGAAT
ATTTGCTGACAAGATTGCATAGACCGGTCACTTGATCTACCATGTG
CGGTGGTATCGACAAGCGAACCATCGAGAAGTTCGAGAAGGTTG
GTTTCCATTTCCCTCGATCGCACGCCCTCTATCCACCGATCCATCA
CTCGAATCAGTTACGACGATTGAATATGCGCCTGTTACCCCGCTC
GAGTACAAAATCTTGCGTTTCAACCGTAATTTTTTTTGGTGGGGT
GTCAACCCCGCTACTCGAGCGACAGACGTTTGCCCTCTTCCCACA
AACTCATTTCTTGTGCATCACGTGTCCATCAGCCACTAACCACAC
GACAATAGGAAGCCGCCGAGCTCGGTAAGGGTTCTTTCAAGTAC
GCCTGGGTTCTTGACAAGCTCAAGGCTGAGCGTGAGCGTGGTAT
CACCATCGATATCGCCCTCTGGAAGTTCGAGACTCCTCGCTACTAT
GTCACCGTCATTGGTACGTTATCATCACTTACACTCAATACTTTCT
CATGCTAACATGTACTCAGACGCTCCCGGTCACCGTGATTTCATC
AAGAACATGT

Fig. 6. Morphological characteristics of Fusarium incarnatum (A-B: colonies on MEA agar plate incubating at 20°C for 7 days; C-G: fungal spore structure,C: bar = 25 μm, D-G: bar = 10 μm).

ACKNOWLEDGMENTS

This work was partially supported by funding projects 99203 and 2990529 from the Environmental Protection Bureau of the Tainan City Government, Taiwan, R.O.C. The author would like to thank the Bioresource Collection and Research Centre of the Food Industry Research and Development Institute for their assistance in identifying fungal genera.

REFERENCES

Burge, H., Chew, G., Muilenberg, M. and Gold, D. (1995). Role of Fungi in-house Dust Ecosystems. *J. Allergy*

Clin. Immunol. 95: 167.

Cao, J.J., Li, H., Chow, J.C., Watson, J.G., Lee, S., Rong, B., Dong, J.G. and Ho, K.F. (2011). Chemical Composition of Indoor and Outdoor Atmospheric Particles at Emperor Qin's Terra-cotta Museum, Xi'an, China. *Aerosol Air Qual. Res*. 11: 70–79.

Charell, P.R. and Hawley, R.E. (1981). Characteristics of Water Adsorption on Air Sampling Filters. *Am. Ind. Hyg. Assoc. J.* 42: 353–360.

Cox, C.S. and Wathes, C.M. (1995). *Bioaerosols Handbook*, Lewis Publishers, N.Y., US.

Crittenden, B.D. and Long, R. (1976). In *Carcinogenesis--A Comprehensive Survey*, Vol. 1, Freudenthal, R.I. and Jones, P.W. (Eds.), Raven Press, New York, p. 209.

Deguchi, N. and Yoshizawa, S. (1996). Study on the Pollution by Common Fungi in Houses, In *Proceedings of the 7th International Conference on Indoor Air Quality and Climate*, Institute of Public Health, Nagoya, Japan, p. 149–154.

Fabian, M.P., Miller, S.L., Reponen, T. and Hernandez, M.T. (2005). Ambient Bioaerosol Indices for Indoor Air Quality Assessments of Flood Reclamation. *J. Aerosol Sci.* 36: 763–783.

Fang, Z., Ouyang Z., Hu, L., Wang, X., Zheng, H. and Lin, X. (2005). Culyurable Airborne Fungi in Outdoor Environments in Beijing, China. *Sci. Total Environ.* 350: 47–58.

Hinds, W.C. and Bellin, P. (1988). Effect of Facial-seal Leaks on Protection Provided by Half-mask Respirator. *Appl. Ind. Hyg.* 3: 158–164.

Horner, W.E., Helbling, A., Salvaggio, J.E. and Lehrer, S.B. (1995). Fungal Allergens. *Clin. Microbiol. Rev.* 8: 161–179.

Huang, H., Cao, J.J., Lee, S.C., Zou, C.W., Chen, X.G. and Fan, S.J. (2007). Spatial Variation and Relationship of Indoor/Outdoor $PM_{2.5}$ at Residential Homes in Guangzhou City, China. *Aerosol Air Qual. Res*. 7: 518–530.

Huang, H.L., Lee, M.G. and Tai, J.H. (2012). Controlling Indoor Bioaerosols Using a Hybrid System of Ozone and Catalysts. *Aerosol Air Qual. Res*. 12: 401–411.

Huboyo, H.S., Tohno, S. and Cao, R. (2011). Indoor $PM_{2.5}$ Characteristics and CO Concentration Related to Water-based and Oil-based Cooking Emissions Using a Gas Stove. *Aerosol Air Qual. Res*. 11: 401–411.

Kalogerakis, N., Paschali, D., Lekaditis, V., Pantidou, A., Eleftheriadis, K. and Lazaridis, M. (2005). Indoor Air Quality - Bioaerosol Measurements in Domestic and Office Premises. *J. Aerosol Sci.* 36: 751–761.

Karbowska-Berent, J., Gorny, R.L., Strzelczyk, A.B. and Wlaz1o, A. (2011). Airborne and Dust Borne Microorganisms in Selected Polish Libraries and Archives. *Build. Environ.* 46: 1872–1879.

Kendrick, C. (1990). Fungal Allergens, In *Sampling and Identifying Allergenic Pollens and Moulds*, Smith, E.G. (Ed.), Blewstone Press, San Antonio, TX, US.

Kerker, M. (1969). *The Scattering of Light*, Academic Press, New York.

Kim, Y.C., Kim, H.T., Kim, D., Nakajima, J. and Higuchi, T. (2009). Distribution Characteristics of Airborne Bacteria and Fungi in the Feedstuff-manufacturing Factories. *J. Hazard. Mater.* 169: 1054–1060.

Kung, P.Y., Wang, Y.T., Chiu, J.C., Hsu, Y.C., Wu, T.N., Shen, Y.H. and Wen, S.B. (2011). Comparison of Indoor Air Quality Measurement by Using Direct Detection Apparatus and Standard Methods. *Adv. Mater. Res.* 230–232: 905–909.

Lai, S.C., Ho, K.F., Zhang, Y.Y., Lee, S.C., Huang, Y.U. and Zou, S.C. (2010). Characteristics of Residential Indoor Carbonaceous Aerosols: A Case Study in Guangzhou, Pearl River Delta Region. *Aerosol Air Qual. Res*. 10: 472–478.

Lee, S.C. and Chang, M. (2000). Indoor and Outdoor Air Quality Investigation at Schools in Hong Kong. *Chemosphere* 41: 109–113.

Li, C.S. (1998) Evaluation of Indoor Air Quality in Residencies and Office Buildings, National Science Council NSC 87-EPA-P-002-013, Taipei, Taiwan, ROC.

Liao, C.M., Luo, W.C., Chen, S.C., Chen, J.W. and Liang, H.M. (2004). Temporal/Seasonal Variations of Size-dependent Airborne Fungi Indoor/Outdoor Relationships for a Wind-induced Naturally Ventilated Airspace. *Atmos. Environ.* 38: 4415–4419.

Macher, J. (1999). *Bioaerosols: Assessment and Control*, American Conference of Governmental Industrial Hygienists (ACGIH), Cincinnati., OH, US.

McNamara, M.L., Noonan, C.W. and Ward, T.J. (2011). Correction Factor for Continuous Monitoring of Wood Smoke Fine Particulate Matter. *Aerosol Air Qual. Res*. 11: 315–322.

Mentese, S., Arisoy, M., Rad, A.Y. and Gullu, G. (2009). Bacteria and Fungi Levels in Various Indoor and Outdoor Environments in Ankara, Turkey. *Clean-Soil Air Water* 37: 487–493.

Panagopoulos, I.K., Karayannis, A.N., Kassomenos, P. and Aravossis, K. (2011). A CFD Simulation Study of VOC and Formaldehyde Indoor Air Pollution Dispersion in an Apartment as Part of an Indoor Pollution Management Plan. *Aerosol Air Qual. Res*. 11: 758–762.

Rajasekar, A. and Balasubramanian, R. (2011). Assessment of Airborne Bacteria and Fungi in Food Courts. *Build. Environ.* 46: 2081–2087.

Singh, U., Reponen, T., Cho, K.J., Grinshpun, S.A., Adhikari, A., Levin, L., Indugula, R. and Green, B.J. (2011). Airborne Endotoxin and β-D-glucan in PM_1 in Agricultural and Home Environments. *Aerosol Air Qual. Res*. 11: 376–386.

Stetzenbach, L.D. (1997). Introduction to Aerobiology, In *Manual of Environmental Microbiology*, ASM Press, Washington, DC, US. p. 619–628.

Su, H.J., Wu, P.C., Chen, H.L., Lee, F.C. and Lin, L.L. (2001). Exposure Assessment of Indoor Allergens, Endotoxin, and Airborne Fungi for Homes in Southern Taiwan. *Environ. Res.* 85: 135–144.

Taiwan Environmental Protection Administration (Taiwan EPA) (2005). Guidelines for Indoor Air Quality, Taipei, Taiwan, EPA.

Taiwan Environmental Protection Administration (Taiwan EPA) (2012). Examination of IAQ Recommendation

Levels, Investigation of Indoor Source and Establishment of Monitoring Inspection Mechanism, Project Report EPA-100-FA11-03-A018, Taipei, Taiwan.

Tseng, C.H., Wang H.C., Xiao N.Y. and Chang, Y.M. (2011). Examining the Feasibility of Prediction Models by Monitoring Data and Management Data for Bioaerosols Inside Office Buildings. *Build. Environ.* 46: 2578–2589.

US EPA (1991). Building Air Quality: A Guide for Building Owners and Facility Managers, US Government Printing Office, ISBN 0-16-035919-8 Washington, DC, US.

Wang, Y.T., Chiu, J.C., Hsu, Y.C., Wu, T.N., Shen, Y.H. and Wen, S.B. (2011). Investigation on Indoor Air Quality of Public Sites in Tainan Area. *Adv. Mater. Res.* 255–260: 1413–1417.

Wu, P.C., Su, H.J. and Lin, C.Y. (2000). Characteristics of Indoor and Outdoor Airborne Fungi at Suburban and Urban Homes in Two Seasons. *Sci. Total Environ.* 253: 111–118.

Yeo, H.G. and Kim, J.H. (2002). SPM and Fungal Spores in the Ambient Air of West Korea during the Asian Dust (Yellow Sand) Period. *Atmos. Environ.* 36: 5437–5442.

Received for review, March 1, 2012
Accepted, May 2, 2012