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Diurnal distribution of airborne bacteria and fungi in the atmosphere of Helwan area, Egypt

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ABSTRACT

Airborne bacterial and fungal composition in an industrial town of Helwan, Egypt, was studied using a slit impactor sampler during the period from March 2006 to February 2007. Airborne bacterial concentrations were usually higher than fungi. Bacteria and fungi had similar diurnal variation patterns. Airborne microorganisms reached their concentration peaks in the evening and gradually decreased during the night time. The hourly concentration peaks of the bacteria and fungi appeared at 20:00 h. A significant difference ($P \leq 0.05$) was found between the hourly mean concentrations of airborne fungi in winter compared to other seasons. Fungi concentrations were significantly higher ($P \leq 0.05$) on working weekdays than weekends. *Aspergillus*, *Penicillium*, *Alternaria* and *Cladosporium* were the most predominant airborne fungal genera. *Aspergillus* showed double peak patterns whereas *Penicillium*, *Alternaria* and *Cladosporium* showed one peak pattern. The diurnal variations of the bacteria and fungi could be divided into four periods: 1) the morning maximum concentration (6:00 h–10:00 h), 2) midday to afternoon pattern (10:00 h–16:00 h), 3) the evening concentration peak (18:00 h–20:00 h) and 4) the gradual decrease of night time concentration (22:00 h–24:00 h). Geographical location, human activity, growth cycle of organisms and meteorological factors were the main criteria controlling the temporal variations of the air microorganisms in the Wadi Hof area.

1. Introduction

Airborne microorganisms originate from different natural sources such as soil, animals, and humans (Pósfai et al., 2003; Mouli et al., 2005; Fang et al., 2007). Man-made activities such as sewage treatment plants, animal rendering, fermentation processes and agricultural activities emit microorganisms into the air (Recer et al., 2001; Adhikari et al., 2004; Gillum and Levetin, 2008). Airborne microorganisms have been implicated in spoilage of food (Tournas and Katsoudas, 2005), damage of books and archival materials (Aira et al., 2007), biodeterioration of stones (Mohammadi and Krumbein, 2008) and spread of plant and animal diseases (Rossi et al., 2005). Exposure to outdoor air microorganisms has been associated with allergic respiratory symptoms, asthma exacerbation, asthma related death and infection (Dales et al., 2004; Peternel et al., 2004).

Airborne microorganisms have been shown to vary throughout the day and season depending on various environmental factors such as: type of vegetation (Pepeljnjak and Šegvić, 2003), air pollution (Lin and Li, 2000), human activities (Mitakakis et al., 2005), meteorological and seasonal climatic factors (Rossi et al., 2005 and Klarić and Pepeljnjak, 2006) and periodicity of the emission sources (Cariñanos et al., 1999). Members of dry air spora are found in the greatest abundance in the atmosphere with condition of low humidity and high wind speed (Levetin, 1995). The wet air spora require moisture for release and thus increase in number after precipitation events (Horner et al., 1992).

In Egypt, little attention has been paid to outdoor airborne microorganisms. Airborne fungal concentrations have been monitored in some Egyptian cities including Ismailia (Abdul Wahid et al., 1996), Cairo (Abdel Hameed et al., 1999), Menofia (Abdel Hameed and Khoder, 2001), Western Desert (Ismail et al., 2002), Giza (Abdel Hameed et al., 2007) and New Damietta (EL-Morsy, 2006). Diurnal distribution of airborne fungi was only studied in Upper Egypt, at Assiut (Abu-El-Souod, 1974) and Qena (Abdel-Fattah et al., 1981).

This study aims to assess composition and diurnal distributions of airborne fungi and bacteria on working days and weekends in an industrial district town of Helwan. Moreover the diurnal air distributions of the most common allergenic fungal genera *Alternaria*, *Aspergillus*, *Cladosporium* and *Penicillium* will be determined.

2. Materials and methods

2.1. Area description

Air sampling was performed at Wadi Hof site, 29°82′N, 31°33′E, about 4 km north of Helwan district. Helwan is considered one of the major industrial urban areas in Cairo City. It is located at 29°51′N,

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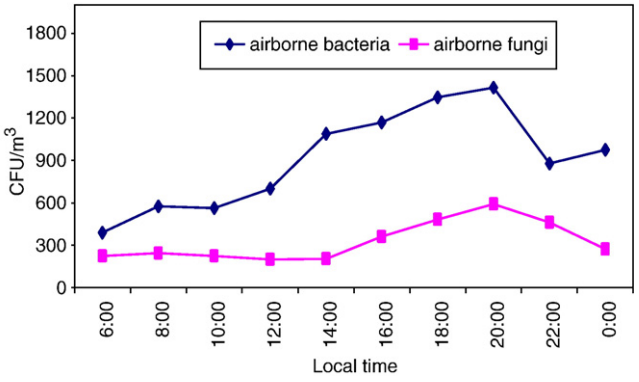

Fig. 1. The mean diurnal concentrations of airborne bacteria and fungi.

31°20′E, about 25 km south of Cairo's city center near the eastern bank of the River Nile with approximately 500,000 inhabitants. Cement, automobiles, iron, steel, textiles, and wood furniture are the main industry products in Helwan area. A variety of vegetation is present in the area but there is no predominant ground cover.

2.2. Air sampling and isolation methods

Airborne bacteria and fungi samples were collected during the period from March 2006 to February 2007, at Wadi Hof, Helwan district. Sampling was conducted from the roof of a ~15 meter height building. Slit sampler (*Model П 818N°5587*, САЕПАНО, ВСССР) was used to collect airborne microorganisms. Airborne particles impact onto a rotating agar plate by passing through a slit at a flow rate of 20 l/min for 2–4 min. Air samples were collected on Mondays and Fridays (weekends), 2 times per month, from 6:00 h to 24:00 h, at two hour intervals. Two consecutive samples were collected during every

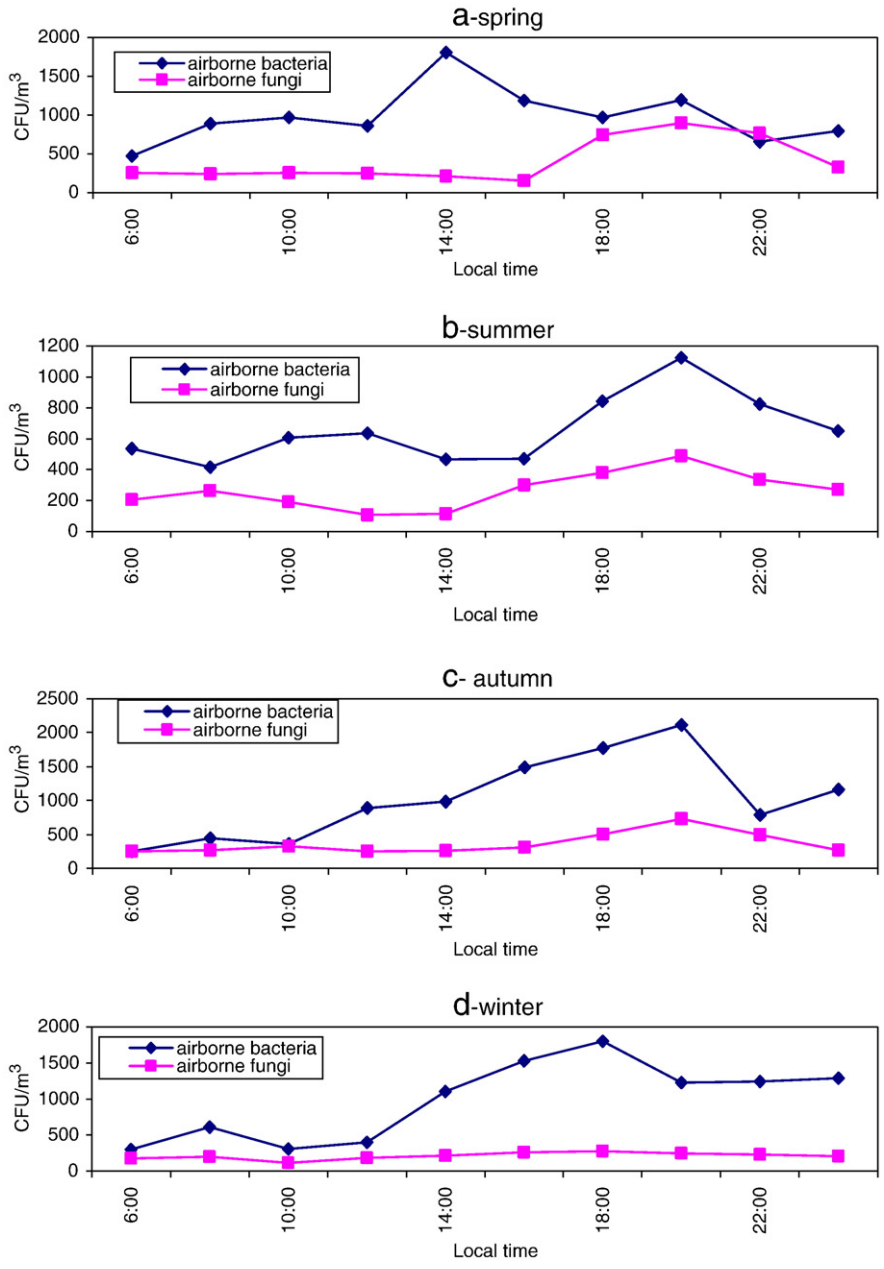

Fig. 2. Seasonal diurnal variations of airborne bacteria and fungi.

sampling event (a total of 4 sample runs were collected every month). Petri plates, (in replicates) containing nutrient agar and Rosebengal streptomycin agar media were used to collect airborne bacteria and fungi, respectively. The slit sampler was sterilized by 70% ethanol between each sampling run. Collected samples were maintained in an ice box during sampling and transportation.

Bacterial plates were incubated at 25 °C for 48 h; whereas fungal plates were incubated at 28 °C for 5–7 days and examined daily. The growing colonies were counted and the mean count from replicate plates was calculated. The airborne bacterial and fungal concentrations are calculated and expressed as colony forming unit per cubic meter of air (CFU/m^3).

Identification of all fungal isolates was done using macroscopic and microscopic features following to Raper and Fennell (1965), Ellis (1971), Singh et al. (1991) and Barnett and Hunter (1999).

2.3. Meteorological parameters

Temperature and relative humidity were measured with a Psychrometer (SATO, PC-5000 TRH-II sampler) during each sampling event. Wind speed records were obtained from the Egyptian Meteorological Authority. During this study, temperature ranged between 12 and 43 °C, relative humidity ranged between 23 and 69.5% and wind speed records varied between 0.5 and 7.2 m/s.

2.4. Statistical analysis

The Mann–Whitney *U* test was used to ascertain the significance of differences between the hourly mean concentrations of airborne microorganisms and on working weekdays and weekends. A probability of less or equal to $P \leq 0.05$ was considered significant.

3. Results

3.1. The diurnal distributions of airborne bacteria and fungi

Fig. 1 shows the diurnal distributions of airborne bacteria and fungi. The hourly concentrations of bacteria exceeded fungi. The concentration peaks of airborne bacteria (1.414×10^3 CFU/m^3) and fungi (5.90×10^2 CFU/m^3) were achieved at 20:00 h. Seasonal diurnal distributions of the bacteria and fungi are shown in Fig. 2(a–d). Their mean concentration peaks were recorded at 20:00 h in spring, summer and autumn. In contrast, the mean concentration peaks of airborne bacteria (1.801×10^3 CFU/m^3) and fungi (2.81×10^2 CFU/m^3) were observed at 18:00 h in winter, 2 h earlier than other seasons. Statistically, Mann–Whitney *U* test showed significant differences between diurnal distributions of bacteria in spring and summer seasons ($P = 0.0315$). Significant differences were found between diurnal distributions of fungi in spring and winter ($P = 0.0262$) and in autumn and winter ($P = 0.0002$).

3.2. Diurnal distributions of airborne bacteria and fungi on working days and weekends

Airborne bacteria and fungi showed similar distribution patterns on working days and weekends (Fig. 3a and b). A non-significant difference was found between mean concentrations of airborne bacteria on working days and weekends ($P = 0.5147$). The concentration peaks of bacteria were achieved at 18:00 h and 20:00 h on working days and weekends, respectively. The concentration peaks of the fungi on working days and weekends were detected at 20:00 h. A significant difference was found between the concentrations of airborne fungi ($P = 0.0446$) on working and weekends, where higher airborne microbial concentrations shifted toward working days.

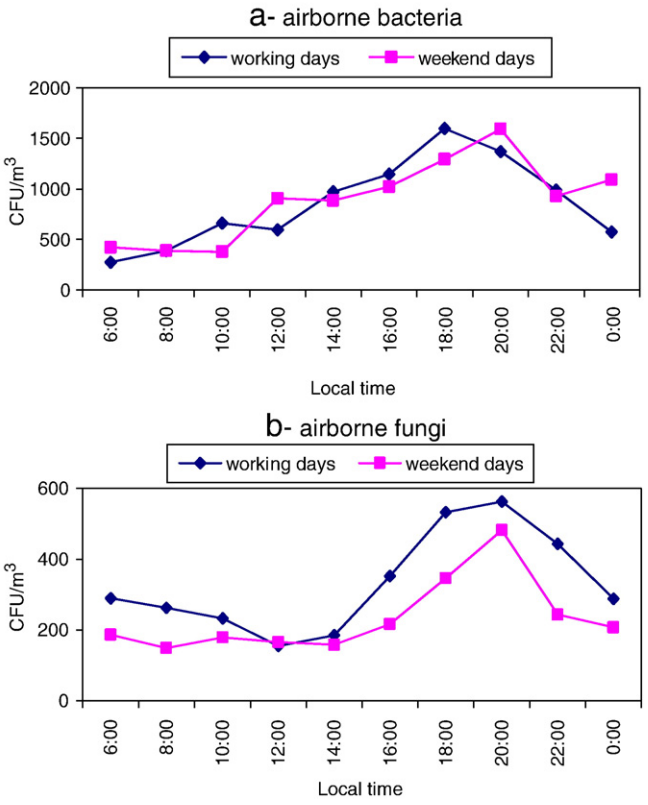

Fig. 3. Diurnal variations of airborne bacteria (a) and fungi (b) on working days and weekend days.

Table 1
The types, number of isolates and percentages of airborne fungal genera identified in Wadi Hof.

Fungal genera	Number of isolates	%
Total *Aspergillus*	3116	30.84
Aspergillus flavus	525	5.2
Aspergillus niger	1296	12.82
Aspergillus fumigatus	4	00.04
Aspergillus ochraceous	43	00.43
Other *Aspergillus* spp.	1248	12.35
Acremonium	2	0.02
Alternaria	913	09.03
Aureobasidium	124	01.227
Basidiomycetes	9	00.09
Bipolaris	12	00.118
Botrytis	1	0.001
Botryotricum	1	0.001
Chlamydomycetes	10	00.1
Cladosporium	2575	25.51
Curvularia	2	00.02
Drechslera	5	00.05
Emericella	21	00.2
Epicoccum	4	00.04
Eurotium	6	00.06
Fusarium	84	0.831
Geotrichum	3	0.029
Monilia	5	00.05
Mucor	12	0.118
Mycelia sterile	811	08.03
Nigrospora	3	00.03
Oidiodendron	386	03.82
Penicillium	1733	17.15
Rhizopus	98	0.969
Stachybotrys	9	00.09
Scopulariopsis	14	0.138
Torula	3	0.029
Ulocladium	14	0.138
Yeast	128	01.27
Total isolates	10,104	100

3.3. Diurnal distribution of the most predominant airborne fungi

The identified airborne fungi are shown in Table 1. A total of 10,104 fungal isolates belonging to 29 fungal genera were identified. *Aspergillus* (30.84%), *Alternaria* (9.03%), *Cladosporium* (25.51%) and *Penicillium* (17.15%) were the most common airborne fungi. The diurnal distributions of *Alternaria*, *Aspergillus*, *Cladosporium*, and *Penicillium* are shown in Fig. 4(a–b). The concentration peak of *Alternaria* (8.7×10 CFU/m^3) was achieved at 16:00 h whereas the minimum concentration (6.38 CFU/m^3) was detected at 10:00 h. *Aspergillus* showed double peak patterns at 10:00 h (1.15×10^2 CFU/m^3) and at 20:00 h (1.44×10^2 CFU/m^3). The concentration peaks of *Penicillium* (9×10 CFU/m^3) and *Cladosporium* (1.96×10^2 CFU/m^3) were found at 20:00 h. On the other hand, the minimum concentrations of *Penicillium* (2.5×10 CFU/m^3) and *Cladosporium* (3.1×10 CFU/m^3) were found at 14:00 h. *Aspergillus* drought appeared 2 h earlier than *Penicillium* and *Cladosporium*. Significant differences were found between diurnal concentrations of *Aspergillus* with *Penicillium* ($P<0.0001$) and *Alternaria* ($P<0.0001$) and between *Alternaria* with *Penicillium* ($P=0.0376$) and *Cladosporium* ($P=0.0216$).

4. Discussion

The hourly distribution of biological particles in the atmosphere of a city is analyzed to know at what time of the day the concentration is maximum. This knowledge can be useful for all people who suffer from respiratory diseases and help plan their outdoor activities. In the present study the diurnal variations of airborne bacteria and fungi could be divided into four periods: 1) the morning concentration peak, 6:00 h–10:00 h, 2) midday to afternoon pattern, 10:00 h–16:00 h, 3) the evening concentration peak achieved between 18:00 h and 20:00 h, and 4) the nighttime decreased concentration, 22:00 h–00:00 h. The morning concentration peak may be attributed to the heat of the sun. At sunrise, wind speed and temperature concurrently increase and relative humidity decreases which leads to evaporation of water molecules binding the microbial particles with different surfaces. High wind speed releases microorganisms into the air. In early morning, human activities and traffic flow disturb and resuspend soil particles into air that may increase airborne microbial concentrations. At midday and afternoon higher temperature, intensive solar radiation and lower relative humidity collectively decrease airborne microbial concentrations. Changed water content occurs for all bioaerosol particles and represents the most fundamental potential stress on air microorganisms. Moreover, radiation effect is exacerbated by dehydration (Cox and Wathes, 1995). The decrease in temperature, solar radiation and increase of human activities are the main causes for the evening peak. In addition, increased humidity in the evening helps physical repair of damaged air microorganisms. However, the absence of human activities leads to decrease in

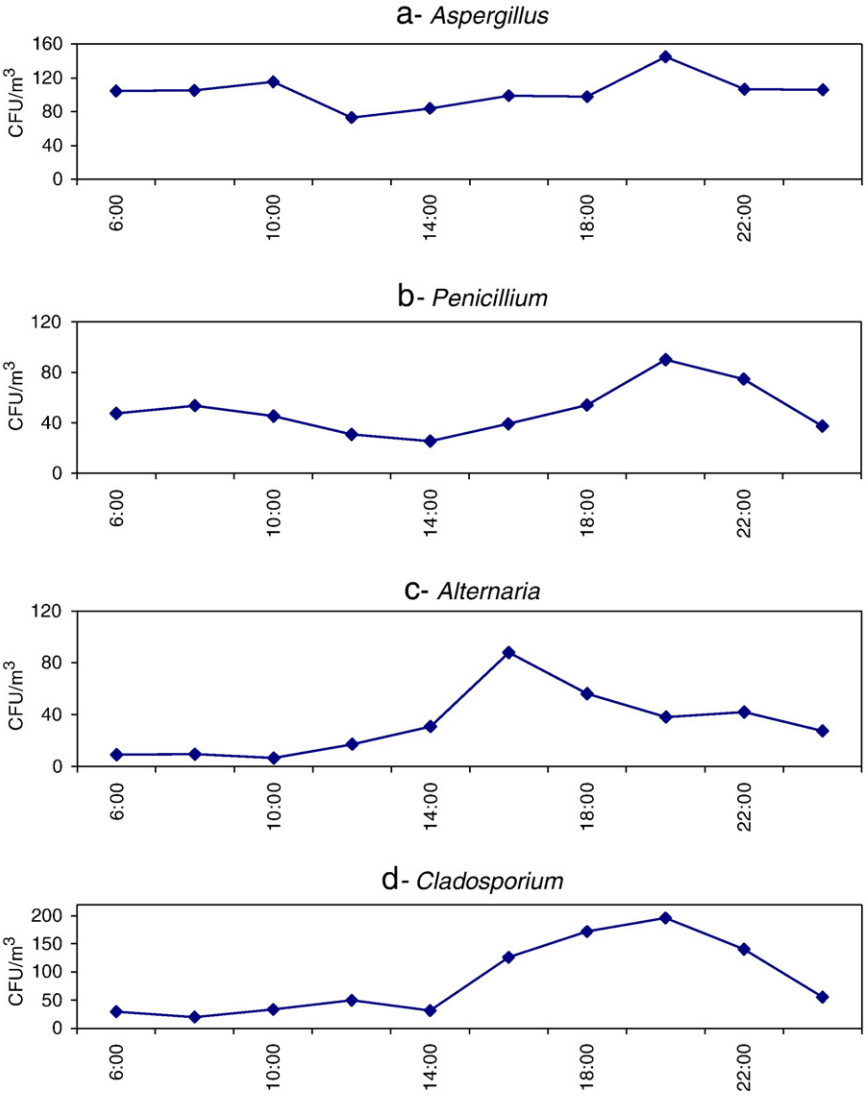

Fig. 4. Diurnal variations of the most common airborne fungi.

airborne microbial concentrations at night time. Stable conditions at night cause fast settlement of air microorganisms which re-suspend again into the air with the sun heat.

The results in the present study agree with Lighthart and Shaffer (1995) who found sunrise peak concentration of bacteria between 06:00 and 08:00 h, which is concurrent with increased winds near the ground level. Fang et al. (2007) observed higher airborne bacterial concentrations at 09:00 and 17:00 h than at 13:00 h due to heavy human activities and traffic flow in the morning. The stability of airborne bacterial patterns between 10:00 h and 16:00 h could be explained by the fact that as the day proceeds, airborne bacteria generated by the flux from vegetation, soil and anthropogenic sources are trapped below the inversion layer and accumulated with time during the day. Concentration gradients were absent and no upward flux occurred due to atmospheric stagnation and presence of thermal inversion in midday period (Lighthart and Shaffer, 1995). Moreover, Lindemann and Upper (1985) found the evening peak at 20:00 h.

Diurnal variations of airborne fungi depend on locality, human activity and weather conditions. Diurnal distributions of airborne fungal spores are associated with the sun heat effect in the morning and reduced relative humidity (Royes, 1987). Nussbaum (1990) reported that morning was the time of day where maturation and dispersion occurred for many fungi. He also attributed fewer airborne fungal spores during midday and afternoon to the effect of higher temperature, and solar radiation. Jones and Harrison (2004) attributed the rising of airborne fungal concentration in the morning to the increasing of wind speed. Fang et al. (2005) explained the morning maximum peak of fungi to the effect of sunlight which causes spore release. In contrast, extreme midday conditions reduce concentrations of biological materials (Jones and Harrison, 2004). Levetin (1995) concluded that hazard of desiccation was the greatest during midday. Concentrations of airborne fungi were lower during daytime than at nighttime due to the effect of solar radiation whereas dark and high relative humidity enhances fungal viability (Fengxiang et al., 1991). Burch and Levetin (2002) recorded two airborne fungal spore peaks, at 08:00 h and at 18:00 h, the authors explained the evening peak as a result of increasing wind speed.

The winter's concentration peak appeared 2 h earlier than other seasons. This is attributed to driving mainly within the city and returning back home soon before the sunset. During other seasons, concentration peaks were delayed until 20:00 h. This may be attributed to the lifestyle of people, higher number of daylight hours and longer staying outdoors.

In this study airborne microorganisms had similar diurnal rhythms on working days and weekends. No significant difference was found between diurnal distributions of airborne bacteria on working days and weekends, probably because of the same lifestyle of people. In contrast, there was significant difference between airborne fungi ($P = 0.0446$) on working days and weekends, because fungal diurnal variations are mainly related to weather conditions throughout the day. The slight differences observed throughout the day may be related to changes in meteorological factors including wind direction and accidental human activity near the sampling location. Fang et al. (2007) reported that atmospheric microbial pollutants are potentially correlated to population density and activity.

Airborne fungi are among the most common organisms in nature and they are considered to be correlated with adverse health effects of humans and plants (Shelton et al., 2002). In this study *Aspergillus*, *Alternaria*, *Cladosporium* and *Penicillium* were the predominant fungal genera in the atmosphere and their concentrations appeared to differ from place to place and even from hour to hour because of prevailing local environment, fungal genera and human activity. *Aspergillus* had double peak patterns, the concentration peaks of *Penicillium* and *Cladosporium* were achieved in the evening at 20:00 h whereas *Alternaria* had a midday peak pattern. These findings agree with Sreeramulu and Ramalingam (1966) who found two daytime maxima of *Aspergillus*, *Helminthosporium* and *Tetrapolea*. Lin and Li (2000) attributed insignificant diurnal pattern for *Aspergillus* and *Penicillium* to their passive launching mechanism of dry small conidia that can be liberated even by slight air currents or vibration. Ho et al. (2005) found the highest concentration of *Aspergillus*/*Penicillium* around midnight. Fang et al. (2005) found the lowest concentration of *Penicillium* at 13:00 h. Gillum and Levetin (2008) observed the highest concentration of *Penicillium*/*Aspergillus* spores at 10:00 h.

In this study, *Cladosporium* peak shifted toward the evening (20:00 h) and gradually increased in the afternoon period between 16:00 h and 20:00 h with the lowest concentration at 14:00 h. A significant concentration peak of *Alternaria* was recorded at 16:00 h. *Cladosporium* and *Alternaria* are usually found in higher concentrations during warmer periods of the day due to their dry-weather spores. The late afternoon peak coincided with the results obtained by Mediavilla (1995) for *Cladosporium* spores. Padys et al. (1969) found a single *Cladosporium* peak throughout the day occurring at different hours depending on the climatic features of the area. *Cladosporium* showed midday pattern (10:00 h–16:00 h) and double peak pattern (08:00 h–10:00 h, 14:00 h–18:00 h), (Lacey, 1981). Nayak et al. (1998) observed the *Cladosporium* peak during morning hours rather than at noon and in the evening. Fang et al. (2007) detected the highest concentrations of *Cladosporium* at 17:00 h. Troutt and Levetin (2001) found the peak concentration of *Cladosporium* at 14:00 h and the lowest at 6:00 h.

Alternaria is a member of dry air spora which favors high temperature and low relative humidity at midday. The presence of melanin pigment protects conidia from the impact effect of sunlight and UV radiation (Gregory, 1973). Members of dry-air spores are found in the greatest abundance in the atmosphere characterized by low humidity during warmer afternoon hours (Levetin, 1995). Dry spore types are released into the air during the day whereas wet spores are released at night (Sarica et al., 2005). The present results agree with Corden and Millington (2001) who found a maximum peak of *Alternaria* between 14:00 h and 22:00 h and more than half the daily *Alternaria* catch was caught between 18:00 h and 24:00 h. Sarica et al. (2007) found higher concentrations of *Alternaria* in the evening time. Generally concentration peaks of airborne fungi have varied among various studies all over the world due to differences in human activities, climatic conditions, vegetation and topographical factors.

5. Conclusion

Airborne bacterial and fungal concentrations varied from hour to hour throughout the day. The rhythmic distributions in microbial liberation depend on geographical characters, human activities, meteorological factors and growth cycle of organisms. The concentration peak in winter season appeared 2 h earlier than other seasons. Airborne microorganisms had the same diurnal rhythms on working days and weekends. *Aspergillus*, *Alternaria*, *Cladosporium* and *Penicillium* were the dominant fungal genera in the atmosphere. *Aspergillus* showed double peak patterns at 10:00 h and 20:00 h. The concentration peaks of total airborne fungi, bacteria, *Penicillium* and *Cladosporium* appeared at 20:00 h. This time is considered dangerous for people who are suffering from respiratory diseases whereas midday is dangerous for persons who are sensitive to *Alternaria*.

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