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Microbial Indoor Air Quality of Public Places in a Semi-dry City in Iran

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Abstract: Majority of people spends 90% of their time in indoor; therefore, quality of indoor air is of great importance from health viewpoint. Because of exposure to bioaerosols in indoor and the proven relationship between them and a wide range of undesirable health effects, the present study is aimed to surveying microbial indoor air quality in some of public places of Gonabad City, Iran. The study was carried out in spring 2015 and bacterial and fungal bioaerosols were measured. The bioaerosol samples from each location (municipality building, students' dormitory, public library and hospital) were collected in morning and afternoon from randomly selected spots. Results of measurements of each sampling occasion were reported as mean values. Samplings were done using active Zefon A6 Inpacter sampler at 1.5 height from the floor (normal respiratory height). Totally, 192 samples (96 bacterial and 96 fungi with replication) were collected. Afterward, the bacteria and the fungi were determined based on microbiology tests. Effects of the environmental factors (temperature and relative humidity) on concentration of bioaerosols were also taken into account. concentration of the bacteria and fungi varied in the range 13-813 CFU/m^3 and 0-288 CFU/m^3 respectively. Air quality in waiting hall of the hospital was the lowest quality with mean bacterial and fungal concentrations of 296.1 and 105.4 CFU m^{-3}, respectively. In addition, air quality in the public library and municipality building was the highest quality with mean concentration of bacteria and fungi 60.6 and 37.8 CFU m^{-3}, respectively. Data analyses showed that there was a direct significant relationship between concentration of bacteria and air temperature ($p<0.01$). In addition, there was a significant relationship between concentration of bacteria and fungi ($p<0.01$). The findings also highlighted a significant relationship between concentration of fungi and relative moisture of the air ($p<0.05$). Taking into account the significant relationship between concentration of bioaerosols and background factors such as temperature and humidity using effective air conditioning system to control temperature and humidity of the air, detecting potential contamination focus points and temporary factors effective on bioaerosols in indoor appears to be effective on improving quality of indoor air. These measures, in turn, result in decrease in concentration of bioaerosols and pathological bioaerosols in particular.

Key words: Microbial quality, public places, bioaerosols, indoor air, pathological

INTRODUCTION

Air is one of the five primary vital elements of life which is usually available with relatively high rate of impurities (Raju, 1997; Barkhordari *et al*., 2011). These impurities are known as air pollutants. Air pollution refers to presence of unwanted particles and materials in the air that harm man and other living creatures' health (Alidadi, 2011; Yarmohammadi *et al*., 2016). Taking into account what we might let into our body by respiration, quality of air has drawn ever increasing attention (Kermani *et al*., 2004). Air pollutions might be in the form of gas or suspended particles which can have natural or human-caused sources (Griffin, 2006). Contaminators enter human's body, mostly, through the air (Abbaspour, 2011). A normal human being inhales 20m3 air and the microorganisms in it every 24 h. The microorganisms are omnipresent in our environment including in water, soil and air and humans and animals body. Those in the air are called airborne microorganisms or bioaerosols

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(Sekhavatjou *et al*., 2013). In other words, the term "bioaerosol" refers to the airborne particles with biological sources (Ghasemkhan, 2010). They include dead/living and pathogenic/non-pathogenic bacteria ranged from 0.001-100 μm size (Mirhoseini *et al*., 2014). Although, these agents are not considered as usual air contaminators, their concentration in indoor might reach to a level enough to cause allergic or health problems for those living in such places. The problems caused by the microorganisms in indoor have drawn a great deal of attention in the recent years. Majority of the microorganisms of the respiratory system remain in the air for short period of time as in most of the cases they are transferred to other person. Some human pathogenic factors such as staphylococcus and streptococcus survive relatively dry environment so that they might survive for a long time on fine particles (Dehghani, 2001). People tend to spend 90% of their time in indoor such as home, office, school and the like (Stryjakowska-Sekulska *et al*., 2007). Therefore, it is imperative to take quality of air in indoor into account knowing that quality of indoor air is more important than that of outdoor. People might be influenced by poor air quality of indoor at three levels of unpleasant and undesirable environment, development of serious health problems and development of chronic disease.

Bioaerosols are of the main causes of indoor air pollutions so that 5-34% of indoor air contamination is caused by airborne particles (Mirhoseini *et al*., 2014). Several studies have highlighted the undesirable effects of bioaerosols (Sharma, 2010). Indoor air might be featured with a great deal of non-pathogenic microorganisms while our main concerns are pathogens and allergen agents. The most common airborne microorganisms that cause allergic reactions are bacteria and fungi (Rostami and Tahsini, 2014). Symptoms of exposure to the bioaerosols are coughing, wheezing, rhinorrhea, itchy eyes or throat, skin rash, diarrhea, asthma and headache (Darvishzadeh *et al*., 2013).

Despite the proven facts about the health problems caused by exposure to bioaerosols, there is no standard for this type of air pollutants so that there are only recommendations about the safe concentrations of these pollutants. The main reasons for this are the wide diversity of bioaerosols, their different pathogenic potentials and difference between individual with regard to sensitivity to the pollution in different people (Ghasemkhani *et al*., 2014).

Taking into account the effects of exposure to the bioaerosols and their relationship with variety of undesirable health consequences and that few studies have been carried out on microbial quality of indoor air in Iran, the present study is an attempt to survey microbial quality of public indoor air from bacterial and fungal bioaerosols viewpoint. The study was carried out in Gonabad, Khorasan-Razavi Province, Iran.

MATERIALS AND METHODS

The study was carried out as an analytical and cross-sectional study in spring 2015. Study population included indoor air (municipality building, girl students' dormitory, waiting hall and emergency ward of 22 Bahman Hospital-Gonabad and a public library). Samples were collected with two weeks interval and twice per day (morning and afternoon) at two randomly selected spots. Results of each measurement occasion were reported as mean value. The samples were collected using zefon A6 Impactor (made by a reliable European company) at 1.5 m height (normal respiration height). It was assumed that the indoor air was homogenous. The sampling devise worked based on direct contact between air samples and growth medium on the plate. The impactor would be disinfected using 70% alcohol. Each sampling operation would take 5min with 16li/m air discharge rate (using an SKC air sampling pump). Sabrouraud dextrose agar growth medium (supplied from European producers) containing chloramphenicol antibiotic (250 mg L^{-1}) was used for fungi samples and tryptic soy agar growth medium (supplied from European producers) containing nystatin (250 mg L^{-1}) was used for the bacterial samples. The plates were placed in a cold box after sampling and then transported to the laboratory. Totally, 192 samples were collected (96 bacterial and 96 fungal samples with replication). To examine the relationship between the environmental factors and concentration of the bioaerosol, temperature and humidity were also measured. The bacterial samples were placed in an incubator for 48 h at 37°C and after counting the colonies by colony counter, the cultures were replicated. To detect the grown bacteria, gram staining and other microbiology methods were used. Fungal samples were placed in an incubator for 5-7days at 25-28°C and the samples were examined on slide and using microscope. Colony density was counted based on CFU/m^3 (colony forming unit per cubic meter). The collected data was analyzed in SPSS (19) and to this end, descriptive statistics such as mean, standard deviation, tables and figures were used. To determine the relationship between the variables Spearman test was used.

RESULTS

The results showed that waiting hall and emergency ward of the hospitals had the poorest indoor air quality with mean bacterial and fungal concentrations of 296.1 and 105.4 CFU m^{-3}, respectively. In addition, the lowest mean bacterial (60.6 CFU m^{-3}) and fungal (37.8 CFU/m^3) concentrations were obtained in the public library and the municipality buildings. Mean score, minimum and maximum concentration of bioaerosols in indoor are listed in Table 1. Figure 1 shows frequency percentage of fungal species in indoor so that highest percentage belongs to Alternaria (22.9% of the samples). According to our findings, on a descending order, the most prevalent fungi found in indoor were Alternaira, Ulocladium, Sterile hyph and yeast. Moreover, the most prevalent bacterial species, on a descending order were micrococcus (33.6%), bacillus (23.35%) and staphylococcus epidermis (19.15%). Analyses of bacterial aerosols in the indoor air indicated that 33.6 and 23.35% of species were micrococcus and bacillus, respectively (Fig. 2). Frequency of micrococcus species in the public library, the municipality building, the girls' dormitory and waiting hall/emergency ward of the hospital were 30.6%, 30.9, 34.3 and 38.6%, respectively; clearly, frequency of bioaerosols in the hospital is higher than that of other places. Temperature and humidity range were 18-33°C and 19-54%, respectively. Table 2 lists Spearman's ranked correlation coefficients of different parameters. The analysis showed that there was a direct relationship

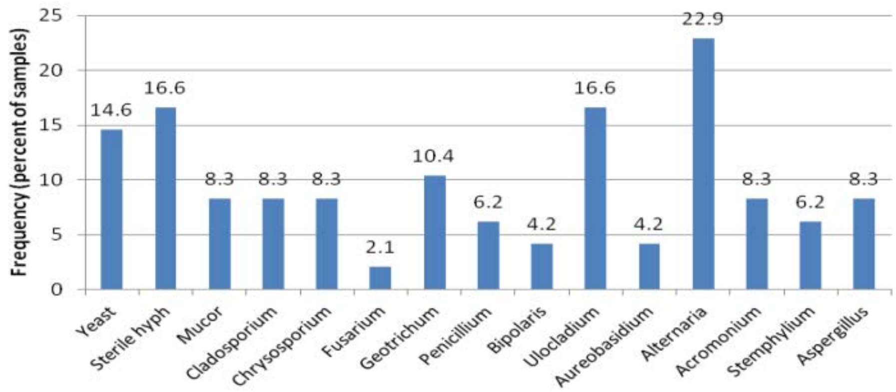

Fig. 1: Frequency percentage of main fungi in public indoor

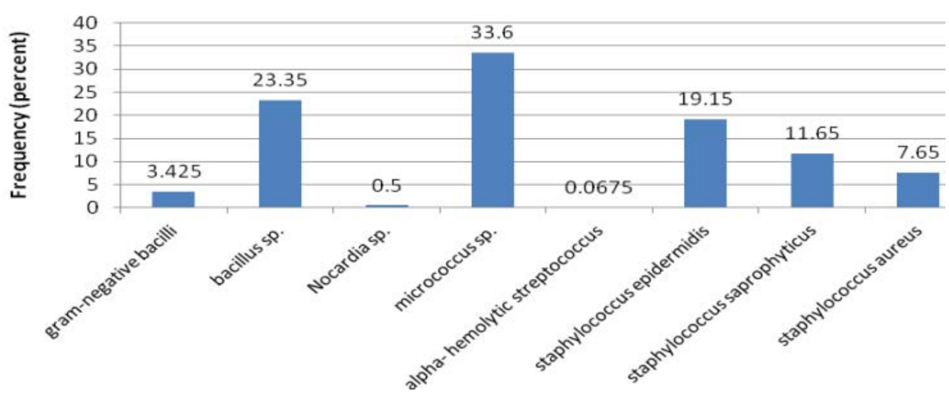

Fig. 2: Frequency percentage of main bacteria in public indoor

Table 1: Mean score, min, and max of bioaerosol concentration in different public indoor (CFU m^{-3})

Microorganism	Bacterium			Fungus		
Indoor	Mean±SD	Max	Mean	Mean±SD	Max	Min
Waiting hall/emergency ward	296.1±268.7	813	100	105./4±53.0	200	38
Municipality	268.9±226.8	813	75	37.8±46.5	167	0
Students' dormitory	200.3±132.8	488	75	54.5±77.5	288	0
Public library	60.6±47.9	163	13	41.9±33.7	125	13

Table 2: Spearman correlation coefficient between parameters

Parameter	Bacterium	Fungus	Temperature	Humidity
Bacterium	-	-	-	-
Fungus	0.484**	-	-	-
Temperature	0.428**	0.223	-	-
Humidity	-0.081	0.286*	-0.226	-

**p<0.01, <0.05

between concentration of the bacteria and temperature ($p<0.01$); a significant relationship between concentration of bacteria and fungi ($p<0.01$) and a significant relationship between concentration of fungi and humidity ($p<0.05$).

DISCUSSION

Presence of airborne microorganisms in indoor air (home and work) is a serious public health concern. Comparing with outdoor, air circulation in indoor is very limited and UV ray in indoor is not notable. In addition, temperature and humidity in indoor are almost constant and in a range that guarantees long-term survival of airborne microorganisms. Consequently, the environment in many indoor such as hospitals and office building is suitable for growth of microbes.

According to the WHO's guidelines, mean concentration of a combination of different fungus species should be less or equal with 150 CFU/m^3 (Rao *et al*., 1996). Fifteen genera of fungi in indoor air were identified and studied. As the results showed, mean concentration of fungus in indoor was less than that of the WHO's guidelines. However, maximum concentration values at waiting hall and emergency ward of the hospital, the girl students' dormitory and the municipality building was higher than the guideline's limits. Although, some fungi such as Aspergillums fumigate cause infectious diseases, most of these pathogenic airborne agents create antibody reactions by generating metabolite products such as proteins. Fungi can cause health problems by generating airborne mycotoxins (Mirhoseini *et al*., 2014). High fungi concentration at waiting hall of the hospital is expectable given the large number of people using the place. With regard to girl students' dormitory, long hours that the students would stay in their rooms and variety of activities that they might do are of the factors that increase concentration of bioaerosol fungi in the place. Moreover, higher concentration of fungi in the municipality indoor is expectable due to large green places in the yard and the constant exchange of indoor and outdoor air in the building.

The results highlighted eight genera of bacteria in indoor; according to ACGIH and AIHA standards, accepted range of microbial load in indoor for non-pathogenic bacteria is 0-500 CFU m^{-3} and this range for pathogenic bacteria is zero (Kowalski, 1998). As listed in Table 1, maximum concentration of the bacteria load in indoor of waiting hall/emergency ward of the hospital and the municipality building was higher than the standards. High bacterial concentration in the emergency ward is expectable given the large number of health personnel and referrals in the ward which is also a managerial concern. With regard to higher maximum concentration of airborne bacteria in the municipality building than the recommended maximum microbial load, large green places and the air circulation between indoor and outdoor is one factor to blame. A study in Poland showed that majority of bacteria and fungi in different indoor of a university were staphylococcus, micrococcus, serratia, aspergillus, penicillium, rhizopus, cladosporium and alternaia (Stryjakowska-Sekulska *et al*., 2007). A study in Austria measured background concentration of fungus spores inside and outside of 185 residential places and reported that aspergillus and penicillium were the main fungal species in indoor (Haas *et al*., 2014). Our study showed a significant and direct relationship between concentration of bacteria and temperature; while no such relationship was found for the fungi. A study in Brazil examined bacterial and fungal quality of public and industrial indoor equipped with air ventilation and showed that temperature and humidity had notable effect on bioaerosol concentration (Nunes *et al*., 2005). A study in girl high schools in Islamshahr-Iran on bioaerosols showed a significant correlation between number of biological aerosols in classroom space and relative humidity/temperature (Kashi and Bigloo, 2015).

A study on microbial quality of air in indoor (residential, institutional, educational) in Isfahan showed that residential indoor had higher bacterial concentration and classrooms has highest concentration of fungi; in addition, the lowest bacterial and fungal concentration was found in institutional and university classroom spaces. The study reported no significant relationship between concentration of fungi and temperature humidity, while there was a direct relationship between concentration of bacteria and temperature/humidity (Mirhoseini *et al*., 2014). As noted, results of the present study supported a significant relationship between concentration of fungi and humidity; this result is consistent with Ren et al. who studied the relationship between fungi spore in indoor and architectural specification of house (Ren *et al*., 2001).

The results showed that chrysosporium (16.6%) and stemphylium (12.5%) and yeast (12.5%) were the main fungi found in the emergency ward air. A study on types and concentrations of bioaerosols in 30 hospital wards of five hospitals in Hamedan, Iran showed that the most prevalent fungi, in a descending order, were penicillium, cladosporium, Aspergillus fumigates and Aspergillus Niger. Highest prevalence rates of the bacteria, in a descending order, were obtained by staphylococcus, negative coagulase, bacillus, micrococcus and staphylococcus aureus (Hoseinzadeh *et al*., 2012).

There is no notable pattern in frequency and diversity of bacteria and fungi species reported by different studies. There are different explanations for this, such as sampling season, air circulation between the indoor and outdoor, type of air ventilation system, temperature, humidity, number of users and type of disinfectors used in the case of health centers.

Biological quality of indoor air is a function of microbial sources in outdoor and indoor contamination sources such as human, plants, construction materials and furniture. Air temperature, relative humidity, amount of sun light received to indoor and even artificial lighting influence biological quality of indoor air. Natural sources of particles are usually considered as background and permanent pollution of the air which are hardly controllable. Quality of air in indoor is not easy to measure or control and its poor quality inflict health and wellbeing of the users (Kashi and Bigloo, 2015). Air in indoor and hospitals in particular can result in transfer of variety of microorganism and risk health of the personnel and referrals who have to inhale the bioaerosols. Therefore, it is essential to take measures like cleaning the environment, using air quality improvement facilities, using air conditioning systems (insulated system if possible is recommended) and solving architectural problems of the buildings.

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