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Original Article

Bio-aerosols concentrations in different wards of Khorramabad Hospital, Iran, 2013

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ABSTRACT

Aims: This study was conducted to investigate the indoor and outdoor air quality at Ashayer Hospital in Khorramabad, Lorestan, Iran, from May-August 2013.
Materials and Methods: Air samples were collected using the ZEFON pump (ZEFON factory, USA) based on manufacturer instructions. Bacteria were isolated via differential methods. Diagnosis of fungi was performed based on amount of fungal colonies growth, shapes, colors, pigments, and microscopic procedure. The effects of various environmental factors including temperature, humidity and outdoor bioaerosol levels were also investigated.
Results: *Staphylococcus spp.* were the most predominant isolated bacteria from studied wards. Furthermore, other microorganisms including *Streptococcus spp*. *Corynebacterium spp*., and *Micrococcus spp*. were also isolated from the hospital air. Moreover, the most frequent fungus in indoor environments of hospital was *Penicillium spp*., while *Aspergillusspp*. and *Alternaria spp.* were the next frequent ones. In addition, the most frequent fungi in adjacent outdoors were *Penicillium spp*., *Aspergillus spp*., and *Cladosporium spp*., respectively. No significant difference was found between the mean concentrations of bioaerosols in inside and outside of hospital ($P > 0.05$).
Conclusion: This study suggests that the bioaerosols level in the hospitals were relatively high. Thus, hospitals should enhance practice of good sanitation protocols and infection control measures.

Key words: Aeroflora, bacteria, fungi, hospital, Iran

BACKGROUND

Microbial air contamination in hospital environments are affected by airborne microorganisms considering their existence, quantities, and kinds. There are wide varieties of factors which influence airborne counts, and therefore influence hospital infection rates.[1] The level and diversity of bio-contamination in hospital environments depend on different factors such as the number and activities of visitors, ventilation system, patients and their activities that contribute to spreading bioaerosols, design of hospital rooms, disinfestations processes and methods, outdoor air and dust, and other factors.[2-6]

Bioaerosols are airborne particles (bacteria, viruses, fungi, etc.) which enter the human body via several

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routes such as inhalation, ingestion, and skin contact.[7] Airborne particles have high significance in different parts of hospitals. Therefore, it is necessary to investigate the bioaerosols, extensively. The bioaerosols can originate from patients or various indoor hospital features and outdoor environmental sources. Compared to outdoor environments, indoor spaces can potentially place humans at greater risk, because enclosed spaces can confine aerosols and build to infection levels.[8,9] These include the patient's own normal flora, patient activities (sneezing, coughing, talking, yawning), linens, bed sheets, staff clothes, visitors, the number of patients per room, materials (such as flowers), and other factors.[10] Therefore, patients themselves can be as a source of pathogenic microbes to other people or the environment.[3] Furthermore, it is believed that outdoor air is the most important source of indoor fungi.[11]

Hospital infections are a serious problem in contemporary medicine worldwide. It is also a true indicator of the quality of the hospital and the level of air pollution.[12-14] Herein, types of airborne micro-flora and correlates of environmental factors (e.g. air humidity and temperature) and bioaerosols concentration were investigated in Ashayer Hospital, Khorramabad, Iran. The hospital opened in 1980 with 11 wards, but has since expanded to more than 20 wards and 350 beds.

MATERIALS AND METHODS

Sampling points and procedure

During 90 consecutive days, 75 bacteria samples and 75 fungal samples were gathered. The results were reported as both qualitative and quantitative form in CFU/m^3 unit. Samples were from divisions of the Intensive Care Unit (ICU), including the burn ICU, surgical ICU, general ICU, and neurology ICU, and outside air, covering all weekdays with 6-day intervals.

Outdoor air samples were matched with each ward by the date of sampling, resulting in 15 samples during 90 days. The ZEFON pump was used for air sampling based on manufacturer instructions.

The flow rate was calibrated at 28.3 l/min and samples were collected in 2 min.[15,16] Sampling locations were selected at a height of 1.5 m above ground and 1 m away from any obstacles. Active air sampling was repeated twice in each location. After which the plates were covered and transferred to the Microbiology Laboratory Unit.

Procedure of bacteria and fungi identification

Tryptic Soy Agar (TSA) with chloramphenicol and SabroDexteroz Agar (SDA) mediums were used as transmission medium for bacteria and fungi, respectively. After sampling, TSA mediums were incubated at 35˚C for 48 h. SDA mediums were placed at room temperature and dark place for 70-120 h. After which colonies were counted according to the colony forming units per cubic meter (CFU/m^3). Diagnostic tests such as Gram staining and biochemical tests including catalase, oxidase, DNAse, Bile esculin, urease, resistance to bacitracin and novobiocin (NB) disc, sugar applied to determine bacteria species and fungal. Also some of their detected through microscopic examination. Bacteria were isolated via differential methods. Diagnosis of fungi was performed based on amount of fungal colonies growth, shapes, colours, pigments and microscopic procedure. In addition, air humidity and temperature were measured to evaluate the effect of environmental factors on bioaerosols concentration.

Measuring the metrological parameters

Temperature and humidity were measured using a Taiwan-made temperature meter and TES-1360A Humidity respectively. Wind speed elements were not assessed.

RESULTS

The average number of fungi and bacteria in the air sample of various parts of the hospital are provided in Tables 1 and 2. The bacterial organisms include *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Staphylococcus saprophyticus*, *Streptococcus spp*., *Corynebacterium spp*. and *Micrococcus spp*., while the airborne fungal isolates include *Penicillium spp*., *Aspergillusspp*., *Cladosporiumspp*., *Alternariasspp*. and *Rhizopusspp*. The highest and lowest concentrations of air bacterial pollution were observed outside of the hospital (327 ± 193 CFU/m^3) and general ICU (199 ± 86 CFU/m^3), respectively. *Staphylococcus aureus* was the predominantly isolated bacteria in all of studied wards, except the general ICU, in which *Staphylococcus epidermis* was the most frequent. *Staphylococcus aureus*, *Staphylococcus epidermis*, and *Staphylococcus saprophyticus* were seen in all of studied wards [Table 2]. No statistically significant difference was found between mean concentrations of air bacterial pollution in terms of CFU/m3 between indoor and outdoor samples ($P > 0.05$). Similarly, its concentration in different wards of the hospital was the same ($P > 0.05$). The highest average number of fungi was observed in outdoor air samples (233 ± 176) and the lowest one was seen in the neurology ICU (88 ± 87).

The most frequent fungus in the indoor environments of the hospital was *Penicillium spp*. while *Aspergillus spp*. and *Alternaria spp*. were the next frequent ones. *Penicillium spp*. *Aspergillus spp*., and *Alternaria spp*. were detected in all of the studied units. In addition, *Cladosporium spp*., and *Rhizopus spp*. were the least frequent ones. *Cladosporium spp*. was isolated from indoor air of both general ICU and women burn units of hospital. Moreover, the most frequent fungi in outdoor samples were *Penicillium spp*., *Aspergillus spp*. and *Cladosporium spp*., respectively. The average

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Table 1: The average number of bacteria in the air of various parts of the hospital

Wards	CFU/m^3	Mean CFU/m^3	Staphylococcus aureus (%)	Staphylococcus epidermidis (%)	Staphylococcus saprophyticus (%)	Streptococcus (%)	Corynebacterium (%)	Micrococcus (%)	Other (%)
General ICU	71-371	199 ± 86	23	24	15	13	11	9	5
Surgery ICU	53-389	208 ± 95	22	21	17	15	0	14	11
neurology ICU	71-618	205 ± 153	28	24	21	20	0	0	7
Women burn	71-530	261 ± 132	18	26	23	19	9	0	5
Outdoor	71-707	327 ± 193	19	16	18	12	12	13	10

number of fungi in the outdoor air is higher than other parts of the hospital ($P > 0.05$). Statistical analysis did not confirm a correlation between the mean outdoor and indoor fungal concentration based on CFU/m^3 ($P > 0.05$). In addition, no statistically significant difference was found between different wards of the hospital in term of CFU/m^3 ($P > 0.05$).

Air humidity and temperature were measured to evaluate the affect of environmental factors on bioaerosols concentration. Pearson's correlation analysis indicated that the relative humidity has no significant association with the fungal levels ($P > 0.05$) air, except in the general ICU room and outdside ($P < 0.01$). Moreover, temperature had no correlation with the fungal levels ($P > 0.05$). Both humidity and temperature had no association with the bacterial levels ($P > 0.05$).

DISCUSSION

The results of the present study revealed various degrees of contamination in all of the examined environments. Therefore, air treatment systems and equipment installation should be improved.

Our statistical analysis did not approve significant difference between the mean concentrations of bioaerosols in inside and outside of the hospital ($P > 0.05$). This result is in agreement with Nourmoradi *et al*. who have found similar pattern.[17] However, Obbard *et al*. realized that there is a relationship between the outdoor and indoor bacteria concentrations.[18] Moreover, no statistically significant difference was found between the mean concentrations of fungi and bacteria in the air of various parts of hospital based on CFU/m^3 ($P > 0.05$). Therefore, the hospital's indoor air quality may be affected by the outdoor air quality (Sautour, 2009).

The highest and lowest average of bacterial levels (CFU/m^3) were detected in outdoor of hospital and general ICU, respectively. The lowest bacterial air contamination in the general ICU may be due to air purification system, while the neurology ICU had the highest air contamination. Such situations are influenced highly by various factors including outdoor-indoor air interaction in hospital, kind of admitted patients, and the efficiency of the ventilation system. *Staphylococcus aureus* was the predominantly isolated bacteria in all of studied wards except the general ICU, in which *Staphylococcus epidermis* was the most frequent. These airborne micro-flora obtained were similar to that reported by Ekhaise *et al.*, who reported bacterial isolates including *S. aureus*, *S.epidermidis*, *Escherichia coli*, *Bacillus* spp. *Proteus mirabilis* and *Streptococcus spp.*, with *S. aureus as the* most prevalent ones.[19] *Staphylococcus* and *Micrococcus* were the most prevalent bacterial species in another study.[20] Yagoub and Agbash have shown that *S. aureus* can cause opportunistic infections when host resistance is deteriorated, especially after the settlement

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Table 2: The average number of fungi in the air of various parts of the hospital

Wards	CFU/m³	Mean CFU/m³	Cladosporiumsp (%)	Penicilliumsp (%)	Aspergillussp (%)	Alternariasp (%)	Rhizopusspothers (%)	other (%)
General ICU	18-318	120 ± 90	15	34	18	22	0	11
Surgery ICU	0-389	113 ± 100	0	42	26	20	0	12
neurology ICU	0-300	88 ± 87	0	39	24	23	0	14
Women burn	0-318	104 ± 78	21	28	17	16	10	8
Outdoor	35-565	233 ± 176	19	31	22	14	7	7

of a primary infection like influenza.[21] It demonstrates as a cause of infections of the skin, deeper tissue and organs, urinary tract infections, pneumonia, postoperative infections, and food poisoning with multi-antibiotic resistance.[21,22] These microorganisms are known primary agents of nosocomial infections in hospitals.[10,23,24] Proper control measures such as improving hygiene are required to control infections by *S. aureus*in hospitals wards.[23,24] Furthermore, other microorganisms including *Streptococcus spp*., *Corynebacterium spp*., and *Micrococcus spp*. were also isolated from the air [Table 1]. The average number of fungi in the outdoor air is higher than various parts of the hospital (mean number 233 ± 176 CFU/m³), ($P > 0.05$). Considering that the number of fungi must be less than 15 CFU/m³ in the hospitals equipped with a filtration system, the observed number of fungi spores in the indoor air may be due to contact with the outdoor air.[25] In addition, the lowest average number of fungi (based on CFU/m³) was seen in neurology ICU (mean number 88 ± 87).

The frequency and diversity of fungi inside the hospital depends on different factors such as the sampling season, outdoor sources, type of patients, and ventilation system.[26] We also showed that the most frequent fungus in indoor environments of hospital was *Penicillium spp*. followed by *Aspergillus spp*. and *Alternaria spp*. Contrarily, *Cladosporium spp*. and *Rhizopus spp*. were the least frequent ones. *Cladosporium spp*. were isolated from indoor air of two general ICU and women burn units of hospital. Moreover, the most frequent fungi were *Penicillium spp*., *Aspergillus spp*. and *Cladosporium spp*. outside, respectively. *Aspergillus spp*. and *Penicillium spp*. provoke allergic disease and induce acute intoxication and many adverse effects to human health.[6] *Aspergillus spp*., *Penicillium spp*., *Acremonium spp*., *Mucor spp*., *Alternaria spp*. and *Cladosporium spp*. Airborne fungi can cause respiratory infections and allergic reactions.[9]

Fungal contamination in healthcare systems has been assessed in several studies and their frequency varies in different reports. Consistent with our findings, Perdelli and Faure have isolated the same fungi from the hospital air. They showed that the most frequent fungus in hospital ward air was *Penicillium* while *Cladosporium* and *Aspergillus* were the next frequent ones.[27,28] Ekhaise *et al*. also found that *Penicillium spp*. and *Aspergillus spp*. were the highest common fungal species in the air of government-owned hospitals in Benin City, Nigeria. Their results were also close to those of the present study. Mahdavi *et al*. reported that the most common fungus in the air of Babol Hospital in Iran was *Penicillium spp*.[29] The results of previous studies showed that fungi including *Candida albicans* and different species of *Aspergillus*, *Cladosporium*, and *Penicillium* are present in some hospital infections and were considered as the major source of hospital fungal infections.[30-32]

CONCLUSION

The findings clearly suggest that the bioaerosols level in the hospitals was relatively high. Thus, hospitals should execute more drastic sanitation protocols and infection control measures. Their air purification equipment should also be improved. Regular monitoring of hospital aero-flora is particularly recommended. High microbial concentrations in various indoor environments point out a potential health risk such as respiratory diseases. Since the isolated bacteria and fungi could be pathogenic, it is pertinent that their presence should be controlled. Efforts should be made to minimize airborne transmission of opportunistic microorganisms and their potential impact on patients.

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