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Indoor Airborne Bio Aerosols in Valiasr Hospital in Zanjan, Iran

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ABSTRACT

Background: Bio aerosols include airborne micro-organisms such as bacteria, fungi, viruses, etc and their products. Exposure to a bio aerosol is linked with a broad spectrum of health problems, including infectious diseases, acute toxic effects, allergies, and cancer. The aim of this study was to evaluate the quality and quantity of bio aerosols found in the air of different wards in Valiasr Hospital, Zanjan, in summer and fall 2012.

Methods: Air samples were collected from six wards including: operating room, infectious, ear, nose, and throat (ENT), surgery, adult intensive care unit (ICU), oncology and administrative with a single-step Anderson sampler. The type and number of colonies were determined in the laboratory, and then the bio aerosol density were calculated in terms of cfu/m^3 and compared with the recommended limits.

Results: The most common genera of isolated bacteria and fungi were *Staphylococcus* and *Penicillium*, respectively. In the infectious ward bacterial density was higher than the recommended limit of WHO (100cfu/m^3) in the visiting times (afternoon). The fungal density in the meeting time (afternoon) in the ICU, ENT, and general surgery, infectious and administrative wards, and in non-visiting times (morning) in the infectious ward was higher than the recommended limit of WHO (50 cfu/m^3).

Conclusion: From the findings of this study it can be concluded that the density of fungi and bacteria in the hospital air in some times of working period are higher than recommended levels and therefore, the condition of existing air filtration and ventilation systems should be appropriated according to the international standards of hospitals buildings.

1. Introduction

Aerosols are particles of suspended solid or liquid in a gaseous environment with a size of 0.001 to 100 microns. Bioaerosols are aerosols containing microorganisms (bacteria, fungi, viruses) or isolated compounds from microorganisms (endotoxins and metabolites, toxins and other microbial residues). The source of biological aerosol particles (cells, cell components or organic material of animal, plant and microbial sources) is sometimes up to the 50 percent of the aerosol

particles (1). Generally, particles with a diameter of 0.3 to 20 microns in medical terms have pathogenic importance, especially breathing particles which have a diameter of 1 to 10 microns, as this size of bio aerosol particle is important to control even more than in other cases (2). Exposures to bio aerosols are linked with a broad spectrum of health effects, including infectious diseases, acute toxic effects, allergies and cancer (3). Fungi are usually found in both outdoor and indoor environments and about 10 percent of people are allergic to fungi (4). Bio aerosols consist of approximately 5 to 34 percent of air pollution (1). Microorganisms are a major source of indoor air pollution. Indoor air creates risks with higher potential problems for patients than outdoor air. Because they can be confined, indoor aerosols can result in several airborne infectious diseases which are related to indoor air quality and these infections are attributed in a wide range of chemical contaminants and physical or biological products in the environment (5, 6). Bioaerosols have different shapes, sizes and categories. Human responses to bio aerosols vary from very mild to extremely severe. For people who work in hospitals and medical centers, bio aerosols are factors that can cause job loss and a reduction in work productivity and absenteeism (7). Indoor air, especially in places like hospitals, can carry a wide variety of microorganisms such as bacteria, fungi and viruses (8). Hospitals are an area where administrators, medical staff, service personnel, patients and visitors inhale bio aerosols and therefore the presence of excessive air bioaerosols in hospitals can bring about serious health threats (9). Although pollution levels, surgical procedures, wounds and cuts produce a large percentage of nosocomial infections, bacterial and fungal contamination of hospital's air also plays an important role in the spread of nosocomial infections. Approximately 10 percent of nosocomial infections in healthy adults are caused by airborne bacteria and moreover control of air borne pathogens in health care facilities is important not only for patients, but also for hospital personnel (10,11). Isolated bacteria from hospital air have been identified and these include; *Staphylococcus aureus, Klebsiella, Bacillus cereus, Bacillus subtilis, Streptococcus pyogenes* and *Sratya Marsns* and isolated fungi such as; *Aspergillus flavus, Penicillium, Fusarium, Alternaria,* and *Candida albicans*. *Staphylococcus aureus* has been cited as the most common bacteria and *Penicillium* as the most commonly isolated fungi (12). The aim of this study was to evaluate the qualitative and quantitative of bio aerosols in Valiasr Hospital, in Zanjan, Iran during summer and fall 2012.

2. Materials and Methods

This study is a descriptive - cross sectional study conducted in six wards including: operating rooms, infectious, ENT and surgery, adult ICU, oncology and administrative, using impact procedures in the Valiasr Hospital, during visiting and non-visiting times in summer and fall 2012. For sampling purposes we used a single-step Anderson sampler with flow rate of 28.3 liters per minute for 10 minutes (3, 13, and 14). The samples were collected on the bacterial culture mediums (nutrient agar) containing cycloheximide and fungi culture mediums (malt extract agar) containing chloramphenicol (14, 15, 17). They were placed under sterile conditions in special boxes and taken to the hospital. The height of the sampler was 1.20 m from the floor (breathing zone) and placed at a distance of 1 m from the side walls, and in addition the screening stations were disinfected with 70% alcohol (9, 18). In total 108 samples of bacteria and 108 samples of fungi were obtained. While sampling, the ambient humidity and temperature were measured with a $Testo_{610}$ device. Plates containing culture medium, were placed immediately after sampling beside ice pack, and returned to the laboratory. In the laboratory, plates containing nutrient agar were incubated at a temperature of 35-37°C for 24-48 h. Then after adding malt extract agar, the plates were

incubated at a temperature of 25-27℃ for 5 to 7days (3, 16, 18, 19). After incubation, the plates were taken out of the incubator and the colonies were counted. To identify the type of bacteria after gram staining and preparing microscopic slides from the mannitol salt agar medium, the tests of blood agar, MacConkey agar, TSI, and biochemical (catalase, oxidase, grown at 42℃) were conducted. In addition, for the detection of fungi we used macroscopic characteristics including color, surface preparation method with the colonies, and small colonies and culture slides. Lactophenol cotton blue solution was used for staining the fungi. According to the mycelium, the presence or absence of medial septum, and the proliferation of conidia, the presence or absence of fungi in vesicles were identified (9, 18, and 20). In order to report the number of colonies (cfu/m^3), the volume of air sample at the correct temperature and pressure was standardized (9, 13).The results were analyzed using Excel and SPSS software. WHO guidelines were used to compare the results with normal values.

3. Results

In this study, from the six hospital wards a total of 216 samples of bacterial and fungal were produced and bioaerosol concentrations by cfu/m^3 were reported. In Table 1, the density of bacteria and fungi, in the two visiting and non-visiting times are shown. According to Table 1, there is no significant difference between the density of bacteria and fungi in the two visiting and non-visiting times ($p>0.05$). In Table 2, the density of bacteria and fungi in the various ward are presented. According to Table 2, the infection ward, with an average of 95.33 cfu/m^3 and administrative unit with an average of 49.69 cfu/m^3 have the highest

Table 1: Density of bacteria and fungi in visiting and non-visiting times in Valiasr hospital, 2012.

	Bacteria			Fungi		
Time	**Average cfu/m^3**	**SD cfu/m^3**	**P value**	**Average cfu/m^3**	**SD cfu/m^3**	**P value**
visiting	66.31	52.10	0.769	48.45	66.52	0.44
Non visiting	69.25	51.50		40.57	33.95	

Table 2: Density of bacteria and fungi in different wards in Valiasr hospital, 2012.

	Bacteria			Fungi		
Ward	**Average cfu/m^3**	**SD cfu/m^3**	**P value**	**Average cfu/m^3**	**SD cfu/m^3**	**P value**
ICU	69.26	65.33	0.115	45.93	100.30	0.953
Oncology	65.14	31.14		37.47	36.73	
Operating room	64.75	48.80		39.87	27.69	
ENT and Surgery	51.65	37.87		44.10	38.28	
Infectious	95.33	63.73		51.15	40.76	
Administrative	49.69	41.59		54.61	22	

and lowest level of pollution, respectively, whereas with the fungi the greatest contamination occurred in the administrative unit and the smallest level of pollution was in

oncology. There were no significant differences found between the mean density of bacteria and fungi in the air of the hospital wards ($p>0.05$). The results of this study showed that at visiting time in the infectious unit an average of 128.53 cfu/m^3 and SD 73.28 cfu/m^3, and in the operating room an average of 33.35cfu/m3 and SD 20.01cfu/m3 were the highest and lowest levels of bacterial contamination, respectively. Also in terms of fungal infections in the visiting time in the intensive care unit there was an average of 67.29 cfu/m^3 and SD 140.77cfu/m^3 and in the operating room an average of 30.32 cfu/m^3 and SD 19.65cfu/m^3 were the highest and lowest pollution levels, respectively. Table 3 shows the percentage of detected bacteria and fungi. According to Table 3 *Staphylococcus* with an average of 55.31 percent is the most commonly isolated bacterial species and the genera Penicillium with an average of 38.44 percent is the most often isolated fungi from different wards of the hospital's air. In this study the collected bio aerosols were compared with the WHO recommendations of bacteria and fungi density (Figure 1 and 2) in comparison with the standard.

Table 3: Percentage of isolated bacteria and fungi in different wards of Valiasr Hospital in 2012.

Bacteria	Percent	Fungi	Percent
Gram positive	73.62	*Aspergillus*	32.72
Gram negative	26.38	*Penicillium*	38.44
Staphylococcus	55.31	*Cladosporium*	22.71
Streptococcus	8.61	*Alternaria*	2.62
Gram positive bacilli	7.25	*Fusarium*	0.35
Spore gram positive bacilli	2.56	*Chrysosporium*	1.07
Pseudomonas	11.41	*Rhizopus*	1.91
Acinetobacter	6.92	*Trichothecium*	0.14
Enterobacter	7.57		
Gram negative cocci	0.32		

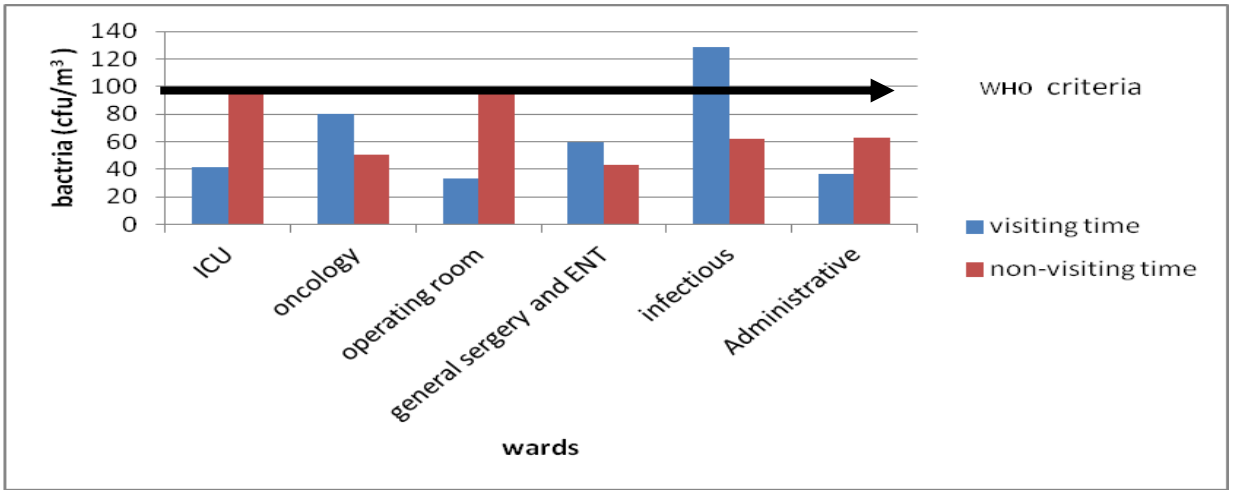

Fig. 1. Comparison of the mean density of bacteria in the air of Valiasr Hospital, with standard values - 2012.

According to the figure 1, the mean density of bacteria in the infection ward is more than the standard value, whereas in the intensive care ward (ICU) and operating room were nearly to the standard.

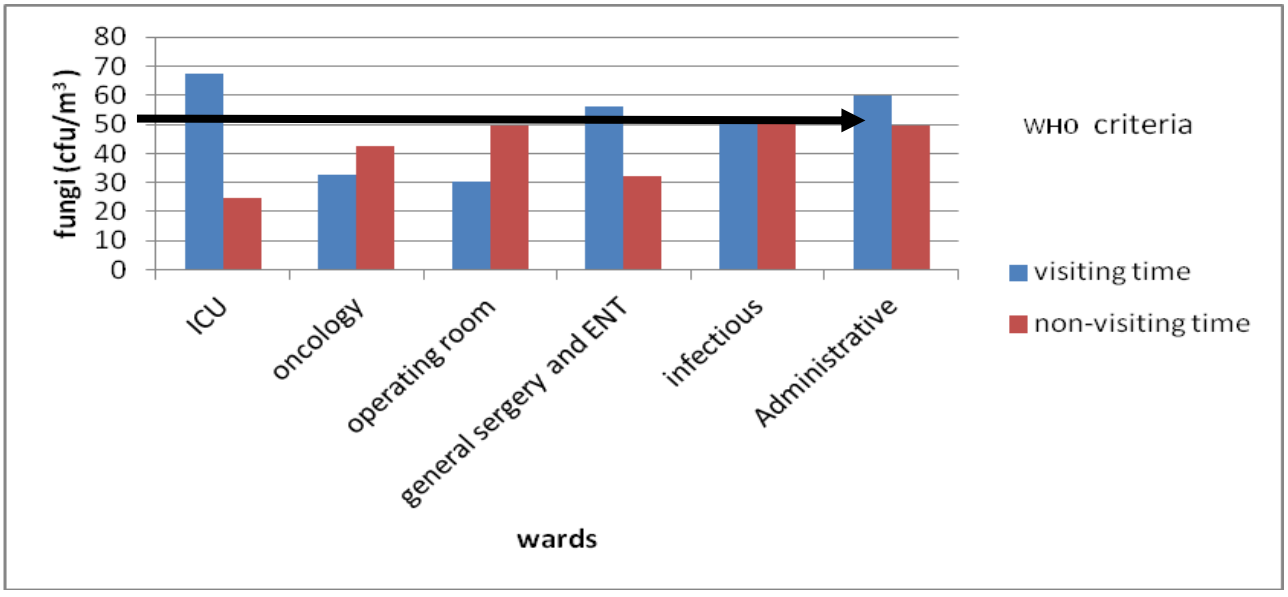

Fig. 2. Comparison of mean density of fungi in the air of Valiasr Hospital -2012.

According to figure 2, the average density of fungus in visiting time in the ICU , ENT and general surgery, Infectious, administrative and in non-visiting time in the Infectious ward is higher than the proposed standard and in the operating room and office areas they were desired.

The results of this study showed that bacterial density vary with air temperature (p=.047) and fungal concentrations with wind speed (p=0.002) significantly. Moreover, there are no significant relationships between bioaerosol concentrations (bacteria and fungi) with air pressure, humidity, or particulate matter. There was no significant correlation found between the density of bacteria and fungi (p=0.341). The statistical analysis revealed that there was a significant difference between the density of bacteria in different months (p=0.003), whereas no significant difference was observed between the density of the fungi in different months (p=0.431).

4. Discussion and conclusion

Gram positive bacteria with a mean of 49.92 cfu/m^3 (%73.62) were the most common bacteria type and Staphylococci with %55.31 was the most common bacterial species, while the Penicillium genera with an average of 17.11cfu/m^3 (%38.44) were identified as the most common fungi. In a similar study in different hospitals of Isfahan University of Medical Sciences (21) and in public and private hospitals the highest percentage of bacteria has been identified as *Staphylococci*.

In a similar study in 2010 by Kim et al. most of the identified bacteria were Staphylococcus species (up to 50 percent), while Perdile et al. and Azizifar reported that the isolated genus Penicillium were the most frequent fungi found in the hospitals' air (18, 22, 23). There is an abundance and diversity of fungi and bacteria due to different factors including: type of hospital (general, technical nature), sampling time, effects of outdoor air,

hospital air, influence of environmental factors such as temperature and wind velocity, referral of patients, types of disinfectants and efficiency, type of air condition filters used and their maintenance, restrictions on the entry and exit of visitors, observance or non-observance by health visitors and staff in various hospitals, can all combine to create different patterns.

The genus Staphylococcus rapidly develops resistance with antibiotics and because they are a part of normal flora of human skin they are easily spread in the hospitals. Genus Penicillium is one of the most common fungi that grows easily in soil and plant materials, because of the proximity of some parts of the hospital to outside doors, the movement of personnel and clients and releasing spores into the air, temperature and humidity as the favorable factors for growth, lack of proper maintenance of filters, and wet organic and inorganic substances in this material, are probably the main cause of the high amount of this material (20). The highest polluted ward in the studied hospital was the infectious ward with an average of 95.33 cfu/m^3. The results of this study are consistent with the studies of Hossein Zadeh (9) and Azizifar (18). The high bacterial pollution in infections is partly due to the hospitalization of infectious diseases, lack of air changes according to the relevant standards, excessive numbers of patients, high traffic volume of patients and their families, and lack of correctly working air condition filters in this section are other reasons. The lowest bacterial contamination level was found in the administrative unit with an average of 49.69 cfu/m^3 and that is probably because of low levels of people traffic and the type of activity that takes place in this part and also the low number of staff working in this sector. According to previous studies, there is strong evidence that the number of bacteria is associated with the number of people in each section. The high fungal density in the office areas could be due to the lack air condition, and more likely the opening and closing of doors and windows. Lower fungal density in the oncology ward is due to disinfection of this area because of its sensitivity, position, department hygiene standards, and a lack of proximity to other units along with less opening and closing of doors and windows. Operating rooms in the afternoon had the lowest levels of infection because they have the most important reasons for hygiene when compared to the other sectors. Despite the low number of active personnel in the afternoon hours, there was also less opening and closing of windows and door which is another reason for the lack of bacteria and fungi in this section. There are higher densities of fungi and bacteria in the operating room in the morning than in the afternoon, and this may be due to the existence of students and trainees (about 80-70 trainees in the morning compared with six trainees in the afternoon), Frequent lack of proper ventilation, deficient air disinfection (disinfecting the air in operating rooms is done only once a week), talking, walking, having an active operation and surgical patients.

During visiting times the bacterial density in the infectious ward is an average of 128.53 cfu/m^3 which was higher than the standard number of WHO which is equal to 100cfu/m^3 (24). During this time the fungus density in the wards of intensive care units, ENT, general surgery, infectious and administrative and in non-visiting time wards of infectious were higher than the proposed standard number of WHO which is equal to 50cfu/m^3 (24). Concentration of fungi and bacteria in the operating room were higher than 10cfu/m^3 which is recommended limit of WHO (24). In a study by Ross et al. conducted on air samples in hospitals, the fungal contamination of the hospital samples (194 cfu/m^3) were almost four times higher than WHO standards (25). From the findings of this study it can be concluded that the density of the fungi and bacteria in the hospital air in some times of working period are higher than recommended levels and therefore, the condition of existing air filtration and ventilation systems should be

appropriated according to the international standards of hospitals buildings.

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