

Quantitative and Qualitative Evaluation of Bio-Aerosols in Surgery Rooms and Emergency Department of an Educational Hospital

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Background: Bio-aerosols are a potential hazard in hospitals and are mostly produced by hospital staff, patients and visitors. Bio-aerosols are solid or liquid particles pending in the air and they consist of aerosols accompanying micro-organisms or organic compounds of micro-organisms such as endotoxin, metabolite, toxin and other parts of organism. Those are a potential hazard in hospitals and are mostly produced by hospital staff, patients and visitors.

Objectives: This study aimed to determine the types and amount of bacterial contamination in operating rooms and emergency department of an educational hospital in Zahedan, South-East of Iran.

Materials and Methods: In this study, 72 samples were collected from three operating rooms and three rooms in the emergency department of an educational hospital during 2012. On the first day of every month, a sample was taken from each room during the morning shift; active sampling was done on plates consisting of blood agar and brain-heart infusion agar (BHI) for 10 minutes in the axis of a one-story Anderson impactor (flow rate 28.1 litter per minutes) and SIBATA air pump SIP 32-L and samples were then placed in a 35°C Incubator. Bacterial colonies were counted; warm coloring and differential tests were done and the data were analyzed using Mann-Whitney U and Kruskal-Wallis tests.

Results: Seventeen types of bacteria were detected including *Staphylococcus*, *Micrococcus*, *Viridians*, *Pneumococcus*, *Escherichia coli*, *Streptococcus*, *Bacillus cereus*, *B. subtilis*, *Klebsiella*, *Pseudomonas*, *Diphtheroid*, *Citrobacter* and *Enterobacter*. Quantitative bacterial results showed that the number of observed bacteria in the emergency department with an average of 103.88 ± 33.84 cfu/m³ was more than that of the surgery rooms with an average of 63.32 ± 32.94 cfu/m³. Furthermore, the highest average number of all counted colonies (106 ± 28.45 cfu/m³) was determined in autumn. In all samples, *S. aureus* and *Micrococcus* were the most detected bacteria.

Conclusions: The World Health Organization (WHO) has suggested relatively relaxed limits of 100 cfu/m³ for bacteria and 50 cfu/m³ for fungi in the hospital air. Therefore, quantitative and qualitative outcomes of this study demonstrate that contamination level and bacterial variety in surgery rooms and emergency departments is high and effective measures must be taken to control the possible health risks.

Keywords: Operating Room; Hospital; Bioaerosol, Bacteria, Fungi, Indoor

1. Background

Aerosols are solid or liquid particles suspended in an atmosphere or gaseous medium with sizes ranging from 0.001 to 100 μ m (1, 2). Bio-aerosols are airborne particles consisting of microorganisms such as bacteria, viruses and fungi or organic compounds derived from micro-organisms such as endotoxins, metabolites, toxins and other microbial fragments; bioaerosols vary in size from 20 nm to more than 100 μ m (3). Bio-aerosols monitoring is one of the tools used by occupational health professionals to evaluate the quality of inside air in hospitals, healthcare centers, residential rooms as well as congested centers such as schools, cinemas, banks and job environments to preserve the contagion of infectious diseases and improve aspiration air. Breathing and digestive

disorders result after long-term exposure to bio-aerosols in contaminated areas (1).

Recently, there has been more concern on sampling of bio-aerosols due to their hygienic effects and them causing infectious diseases, acute poisoning, allergies, stress and cancer. Infections result from the increase and growth of microbes while allergies are the result of exposure to antigens that induce allergic reactions in the human body (4, 5). A study of inside and outside environments of residential apartments in Korea showed no significant bacterial and fungal differences between higher and lower floors with a geometric mean concentration of 10-1000 cfu/m³ for bacteria and fungi, respectively (6). The prevalence of nosocomial infections must be

one of the main concerns as one out of ten hospitalized patients have nurture nosocomial infections (7). In the United States, about 1.5 million people become infected during their hospitalizations each year (8).

A research from Madrid (Spain) compared the concentration of filamentous fungi in the air before and after deconstruction of a maternity department by controlled explosion. The amount of filamentous fungi in the atmosphere during the days after the explosion was more than that in the hospital before the explosion (9). Although the amount of microorganisms that exist in the hospital is important, the main focus should be determining the type of microorganisms due to nosocomial activities (10). This area is one of the most deprived areas in Iran sharing borders with Afghanistan and Pakistan. Low socio-economic level of most families, cultural issues and prevalent infectious diseases overload hospitals in this area, especially in the capital city of the province where patients are referred from all around the province. Furthermore, limited-controlling facilities in the hospitals of this area make it difficult to deal with the situation properly.

2. Objectives

This study aimed to determine the contamination level and types of airborne bacteria by measuring aerosols in one of the educational hospitals of Zahedan, the capital city of Sistan and Baluchestan province located South-East Iran.

3. Materials and Methods

Bacterial contamination of inside atmospheres like surgery rooms and emergency departments can be determined by active or passive sampling and by counting colony numbers per cubic meter of air or square meters of floor area. Devices used to sample airborne bacteria mainly rely on different principles namely, impaction, impingement, filtration, suction and electrostatic precipitator by cultivation and non-cultivation methods (1). Nevertheless most of the studies on bio-aerosols of inside atmospheres have been based on culturing methods (11). In this study, both active sampling and culturing were used for determining qualitative and quantitative contamination level. On the other hand, there are different kinds of active sampling (12, 13) for which a suitable sampler tool is used on the basis of availability and influence. However, Pavicic et al. have suggested that eight-step Anderson is better than two-step (14).

In this descriptive-analytical study, instruction number NIOSH 0800 was used for sampling (15). A total of 72 samples were collected from three operating rooms and three rooms in the emergency department of an educational hospital during 2012. On the first day of every month, one sample was collected from every room at the beginning of the morning shift (7-8 a.m.). Plates were located one meter above the floor and one meter far from obstacles and walls to collect the samples. Plates consisting

of blood agar, MacConkey agar and brain-heart infusion agar (BHI) (Germany's Merck company) were exposed to the air of the study area for 10 minutes by sampling axis including one-story Anderson impactor (flow rate 28.1 liter per minutes, England) and SIBATA pump (Sibata Scientific Technology Ltd, Japan). Then, the lid of the plate was closed by Para-film and was transferred to the microbiology laboratory where the plates were placed in a 35°C incubator for 24-48 hours.

Bacterial colonies were counted and examined by warm coloring and biochemical differential tests. Finally, the result was recorded in the bacterial report. Also eosin methylene-blue lactose sucrose (EMB, Merck KGaA, Darmstadt, Germany), coagulase test, Salmonella-Shigella agar (Merck KGaA, Darmstadt, Germany) and methyl red and Voges-Proskauer (IMVIC) test were performed for differential tests. Although many biological agents that may cause diverse health effects have not been identified yet and maximum limits of various bio-aerosols has not been determined individually, recommended maximum limits set by the National Institute of Occupational Safety and Health (NIOSH) and the American Conference of Governmental Industrial Hygienists (ACGIH) is 1000 cfu/m³ for the total number of bio-aerosol particles, with culturable counts for total bacteria not exceeding 500 cfu/m³ (16). The ACGIH also recommended < 100 cfu/m³ for hospitals (17). Furthermore, for hospital air, WHO suggested relatively relaxed limits of 100 cfu/m³ for bacteria and 50 cfu/m³ for fungi although many facilities fail to meet this range (18). Descriptive statistics was reported as mean \pm SD. In order to compare mean amount of bacteria in the emergency department and operating rooms, Mann-Whitney U test was used. Kruskal-Wallis test compared mean amount of bacteria for different seasons.

4. Results

In this study, 17 types of bacteria were detected including *Staphylococcus aureus*, *Micrococcus*, *Viridians*, *Pneumococcus*, *Escherichia coli*, *Streptococcus*, *Bacillus cereus*, *B. subtilis*, *Klebsiella*, *Pseudomonas*, *Diphtheroid*, *Citrobacter* and *Enterobacter*. Mean density for all bacteria was 103.88 ± 33.84 cfu/m³ in emergency rooms compared to 63.32 ± 32.94 cfu/m³ in operating rooms ($P = 0.003$). The most detected bacteria were *Micrococcus* and *S. aureus* in both emergency and operating rooms. The least detected bacteria were *Citrobacter* and *Branhamla* in emergency rooms and *Branhamla* and *Enterobacter* in operating rooms. Mean amount of *Pneumococcus*, *B. subtilis*, *S. epidermidis*, *B. cereus*, *Diphtheroid* was significantly higher in the emergency rooms than operating rooms (Table 1).

Generally, *Micrococcus* (15.5 ± 9.63), *S. aureus* (12.5 ± 7.5), *S. saprophyticus* (8.3 ± 9.68) and *S. epidermidis* (8.25 ± 6.94) were the most detected bacteria. Mean density for all bacteria during autumn was 106.9 ± 28.45 followed by 84.7 ± 36.11 in spring, 69.61 ± 43.16 in winter and 47.56 ± 22.69 in summer ($P = 0.03$). The most frequent bacteria were *S. aureus* (15.3 ± 3.74), *Micrococcus* (11.37 ± 5.85) and *S. sapro-*

phyticus (10.57 ± 15.12) in spring, *Micrococcus* (14.03 ± 4.89), *S. aureus* (8.21 ± 5.92) and *S. epidermidis* (6.83 ± 6.39) in summer, *Micrococcus* (17.84 ± 10.83), *S. epidermidis* (12.56 ± 6.65) and *S. aureus* (11.03 ± 5.33) in autumn, *Micrococcus* (16.91 ± 12.35), *S. aureus* ($14 \pm$

11.73) and *S. epidermis* (8.38 ± 7.24) in winter (Table 2). There was a significant difference between seasons in terms of *Viridans streptococci* ($P = 0.007$), *Pneumococcus* (0.003), *E. coli* (0.01), *S. epidermidis* ($P = 0.02$) and *Klebsiella* ($P = 0.004$) (Table 2).

Table 1. Mean and Standard Deviation of Bacteria Density in Operating Rooms and Emergency Department ^a

Place Type of Microbe	Emergency Rooms, cfu/m ³	Operation Rooms, cfu/m ³	P Value
Total	103.88 ± 33.84	63.32 ± 32.94	0.003
<i>Micrococcus</i>	14.85 ± 7.5	16.09 ± 11.51	0.58
<i>Streptococcus A</i>	1.26 ± 1.96	2.19 ± 2.78	0.31
<i>Viridans streptococci</i>	2.92 ± 3.33	1.72 ± 2.58	0.42
<i>Pneumococcus</i>	7.81 ± 5.26	3.81 ± 5.54	0.03
<i>Escherichia coli</i>	6.91 ± 10.84	2.00 ± 2.82	0.14
<i>Bacillus subtilis</i>	6.64 ± 5.46	1.63 ± 2.62	0.002
<i>Staphylococcus aureus</i>	14.17 ± 6.02	10.92 ± 8.56	0.19
<i>S. epidermis</i>	10.95 ± 8.24	5.72 ± 4.39	0.04
<i>S. saprophyticus</i>	11.35 ± 11.47	5.45 ± 6.88	0.12
<i>Bacillus cereus</i>	7.14 ± 6.41	1.73 ± 2.15	0.002
<i>Streptococcus A</i>	4.20 ± 3.35	2.90 ± 3.21	0.25
<i>Diphtheroid</i>	5.28 ± 4.72	2.27 ± 2.82	0.03
<i>Pseudomonas</i>	4.00 ± 4.29	3.47 ± 7.44	0.22
<i>Klebsiella</i>	4.19 ± 4.6	1.09 ± 1.57	0.06
<i>Enterobacter</i>	1.17 ± 1.59	0.27 ± 0.76	0.08
<i>Citrobacter</i>	0.77 ± 1.9	1.62 ± 3.24	0.66
<i>Branhamla</i>	0.19 ± 0.73	0.36 ± 1.09	0.59

^a Data are presented as Mean \pm SD.

Table 2. Mean and Standard Deviation of Bacteria Density for Different Seasons ^a

Season, Type of Microbe	Spring, cfu/m ³	Summer, cfu/m ³	Autumn, cfu/m ³	Winter, cfu/m ³	Total, cfu/m ³	P Value
Total	84.7 ± 36.11	47.56 ± 22.69	106.9 ± 28.45	69.61 ± 43.16	82.9 ± 38.72	0.03
<i>Micrococcus</i>	11.37 ± 5.85	14.03 ± 4.89	17.84 ± 10.83	16.91 ± 12.35	15.5 ± 9.63	0.78
<i>Streptococcus A</i>	1.37 ± 3.64	1.36 ± 1.93	1.35 ± 1.8	2.73 ± 2.19	1.74 ± 2.52	0.20
<i>Viridans Streptococci</i>	0.98 ± 2.58	0.00 ± 0.00	4.48 ± 2.86	1.88 ± 2.73	2.3 ± 2.98	0.007
<i>Pneumococcus</i>	8.03 ± 7.9	4.44 ± 3.03	8.98 ± 4.58	0.34 ± 0.96	5.74 ± 5.69	0.003
<i>Escherichia coli</i>	5.49 ± 4.62	0.68 ± 1.37	8.02 ± 12.31	0.68 ± 1.26	4.37 ± 8.05	0.01
<i>Bacillus subtilis</i>	5.29 ± 5.39	2.39 ± 2.32	5.16 ± 4.59	2.4 ± 5.76	4.05 ± 4.87	0.13
<i>Staphylococcus aureus</i>	15.3 ± 3.74	8.21 ± 5.92	11.03 ± 5.33	14 ± 11.73	12.5 ± 7.5	0.26
<i>S. epidermis</i>	2.74 ± 3.07	6.83 ± 6.39	12.56 ± 6.65	8.38 ± 7.24	8.25 ± 6.94	0.02
<i>S. saprophyticus</i>	10.57 ± 15.12	2.05 ± 2.37	9.57 ± 7.34	7.85 ± 8.8	8.3 ± 9.68	0.42
<i>Bacillus cereus</i>	5.09 ± 3.23	1.71 ± 1.31	4.78 ± 6.33	4.45 ± 7.07	4.34 ± 5.38	0.44
<i>Streptococcus A</i>	3.92 ± 3.01	2.39 ± 2.81	3.39 ± 3.96	3.92 ± 3.29	3.53 ± 3.29	0.75
<i>Diphtheroid</i>	2.35 ± 3.61	2.39 ± 2.81	6.01 ± 4.69	2.73 ± 3.51	3.72 ± 4.08	0.21
<i>Pseudomonas</i>	4.7 ± 4.02	0.68 ± 0.79	6.69 ± 8.85	0.68 ± 1.03	3.72 ± 6.03	0.05
<i>Klebsiella</i>	5.09 ± 5.29	0.00 ± 0.00	3.68 ± 3.01	0.34 ± 0.96	2.59 ± 3.68	0.004
<i>Enterobacter</i>	1.37 ± 1.93	0.34 ± 0.68	0.68 ± 1.15	0.34 ± 0.96	0.7 ± 1.29	0.55
<i>Citrobacter</i>	0.00 ± 0.00	0.00 ± 0.00	2.57 ± 3.45	1.19 ± 2.86	1.21 ± 2.67	0.07
<i>Branhamla</i>	0.97 ± 1.71	0.00 ± 0.00	0.00 ± 0.00	0.17 ± 0.48	0.28 ± 0.92	0.23

^a Data are presented as Mean \pm SD.

5. Discussion

In this study, type and density (cfu/m³) of bacteria were determined in operating rooms and emergency departments of an educational hospital. The highest density was determined for *Micrococcus* (15.5 ± 9.63), *S. aureus* (12.5 ± 7.5), *S. saprophyticus* (8.3 ± 9.68) and *S. epidermidis* (8.25 ± 6.94). Average bacterial aerosol's density in different parts of Silesian hospitals was estimated between 100-1000 cfu/m³ and the density of positive warm cocci were 110 cfu/m³ (19). In the current study, most bacteria colonies in the atmosphere of operating rooms and emergency department during all seasons were *Staphylococcus*, which is in line with the results of the study that detected bio-aerosol contamination of the inside air of Silesian hospitals; the main source of this type of bacterium is human (19). In hospitals/clinic air of Silesian hospitals the *Staphylococcus/Micrococcus* group was dominant amongst bacteria: 58-78% of the total bacteria concentration (19). Although the most detected bacteria were *Staphylococcus* in the current study, the emergency department had a greater level of contamination.

Exposure to bio-aerosols can cause breathing disorders and harmful hygienic effects like infections, hypersensitivity pneumonitis and poisonous reactions (20). A study in Hamedan (Iran) on four hospitals showed that the amount of bio-aerosol in the hospital with active air conditioning system (even if not very efficient) was less than the others. Furthermore, *Micrococcus*, *Staphylococcus*, *B. subtilis*, *Stainobacter*, *Mycobacterium*, *Diphtheroid*, *Streptococcus*, *Pseudomonas*, *Nocardia* and fungi were detected in surgery rooms (21) which is in line with the results of our study, regarding the types of microorganisms. Also, because there was no air conditioning in surgery rooms of our study hospital, an appropriate air conditioner can decrease the bacterial density of this hospital.

Botzenhart and Hoppenkamps monitored microorganisms in operating rooms equipped with either an efficient air conditioning system or a current air-flow system. An average colony count of 8 cfu/m³ was determined for an unoccupied air-conditioned operating room compared to about 70 cfu/m³ for the same room when occupied by people. Furthermore, the amount of bio-aerosol was 1 cfu/m³ in an operating room with a current air flow system (22). This can justify the high number of colonies of bio-aerosols in our study area where no ventilation was in use. On the other hand, the qualitative and quantitative bacterial results in the aforementioned study illustrated that the emergency department was more contaminated than operating rooms and also a higher mean for detected bacteria was reported during fall compared to winter (22), which is in line with our study. However, Ozdemir et al. reported particle counts as 2491 and 2308 cfu/m³ for general surgery intensive care units and in operating rooms, respectively (23).

Although the total number of bio-aerosols detected during summer was more than other seasons in the current study, a study conducted in high-rise apartment build-

ings in Korea reported more bio-aerosols during summer than winter (6). This difference could be due to climate differences. Due to the fact that the micro organisms' existence in the atmosphere of operating rooms stretches the healing time of wounds and infections, quantitative and qualitative determinants of microorganisms can be effective if controlling actions are used and managers and experts pay more attention. Hence, it is suggested that bioaerosol contamination are monitored and microorganism types are determined by hospital managers continuously.

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Author Contributions

The overall implementation of this study including the research design, data extraction and analysis, report writing and manuscript preparation were the results of joint efforts by multiple individuals who are listed as co-authors of the paper.

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