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Indoor Airborne Bacterial Concentration of a Private-Owned Hospital Laboratory in Samaru-Zaria

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Abstract
A study on indoor air bacterial concentration in a private-owned hospital (Jama'a) laboratory in Samaru-Zaria is presented. Investigations were carried out during the period of dry (8weeks) and wet (8weeks) seasons in 2007. Air samples and work surface samples were taken after the daily laboratory routine of analyses which were immediately conveyed to the department of microbiology postgraduate research laboratory in Ahmadu Bello University, Zaria for relevant microbiological analyses. In most of the samples, multiple growths of bacteria were observed with the highest bacterial airborne mean concentration: 8.0×10^3 CFU/m^2 in wet season at the 7th week of sampling and the highest surface count: 3.0×10^2 CFU/m^2 in the dry season at the 1st, 3rd and 6th weeks. The predominant bacteria isolated from investigated air and surface samples included S*taphylococcus spp*, M*icrococcus spp*, P*roteus spp,* S*treptococcus spp*, *Escherichia coli*, P*seudomonas spp* and *Bacillus spp*. Among these microbes are pathogenic and allergenic species.
Keywords: Airborne bacteria, concentration, hospital, indoor, laboratory

INTRODUCTION

Living microbes which are suspended in the air are referred to as bioaerosols (Brandl *et al,* 2008). It has been confirmed that atmospheric microorganisms are significantly less compare to those present in oceans and in soil. However, there is still large enough number that can affect the atmosphere and once suspended in the air, they have the opportunity to travel long distances by wind and precipitation thus increasing widespread of airborne diseases that can be associated with humans, animals, and plants (Amato, 2012).It has been revealed that both residential and non residential indoors have bioaerosol sources related to their environmental characteristics (Li and Hou, 2003 ; Sanchez-Monedero 2008).

According to previous studies, the survival, type and duration of microbes in the atmosphere or indoor environment is strongly determined by some physical characteristics such as relative humidity, temperature, chemical composition of the air and radiation (Verreault *et al,* 2008 ; Pepper, 2009).

Microbes are generally found in the air along with water droplets, dust particles and other matter. They followed a particular pathway which includes launching into the air after a release from humans, animals, and vegetation (Al-Dagal, 1990), transported by various means, and are finally deposited on surfaces.

The atmosphere is known for its harsh climatic condition which makes it difficult for the microbes to survive in it. However, many microbes are capable of withstanding this condition by forming endospores. *Bacillus anthracis*, a gram positive rod shaped bacteria utilizes spore formation to resist environmental stresses such as extreme temperatures, chemical contamination, and low nutrient (Gatchalian, 2010). *Aspergillus fumigatus*, a major airborne fungal pathogen is adaptable to changing environmental conditions and therefore still capable of mass infection (McCormick, 2010).

Diverse range of microbial populations are found in hospital indoor environments and they could serve as primary sources of indoor air contamination, potentially posing greater health risk to patients and workers compare to those in the outdoor environments. This is because they are confined aerosols which enables them build up to infectious level (Ekhaise *et al,* 2008). Specific activities like talking, sneezing, coughing, walking, washing and toilet flushing can generate airborne biological matter (Cox and Wathes, 1995 ; Maeir *et al*., 2002). Although indoor environments are considered to be protected, they can become contaminated with particles that present different and sometimes more serious risks when their concentrations exceed recommended maximum limits than those related to outdoor exposures (Banerjee, 2008). The recommended maximum limits are: 1000 CFU/m^3 for the total number of bio-aerosol particles set by the National Institute of Occupational Safety and Health (NIOSH); 1000 CFU/m^3 set by the American Conference of Governmental Industrial Hygienists (ACGIH) with the culturable count for total bacteria not to exceed 500 CFU/m^3 (Cox and Wathes, 1995; Jensen and Schafer, 1998).

The objective of this study was to investigate the indoor airborne bacterial concentration of a clinical laboratory in a private owned hospital located in Samaru, Zaria Kaduna State of Nigeria.

MATERIALS AND METHODS

Study area: This study was carried out in a selected laboratory of a private-owned hospital named Jama'a

located in Sarkin-pawa Street, Samaru-Zaria. Kaduna State, Nigeria. It is close to the north gate of Ahmadu Bello University, Zaria. The hospital was established about two decades ago with a clinical laboratory measuring approximately 7.3m^2 , located on the ground floor of the 2-storey building hospital. The potential sources of microorganisms included open dustbin, direct linkage to the outdoor environment through open door of entrance from the reception room and a large window, air conditioner system.

Air sampling: The samples were collected from the air and work surfaces of the laboratory without controlling any indoor environmental conditions as adopted by Yassin and Almouqatea, 2010. Air sampler MAS- 100 (Merck) corresponding to the 5th stage of Andersen's air sampler was used to collect about 100ml of air for one minute, impacted on prepared Petri dish containing nutrient agar already fixed in the air sampler. This was done in duplicate once in a week after the daily routine for a period of 8 weeks in dry season (Jan-March) and wet season (July-September) respectively. The plates were incubated at 37^{0}C for 24 hrs after which the colonies were enumerated and converted to colony forming units (cfu/ml).

Surface Sampling: Air sampling alone does not provide assurance that an area is free of biological contamination due to re-aerosolisation of the organisms from surfaces during routine activity. Surface sampling therefore is essential to identify the areas and sources of contamination in determining the effectiveness of remediation and clean-up of contaminated indoor environments (Higgins *et al.,* 2003; Anderson *et al* 1997; Buttner, 2001).

The workbench surface was sampled using swab stick moistened with distilled water to swab an area and impacted on nutrient agar surface. This was done in duplicate. The plates were incubated at 37^{0}C for 24 hrs after which the colonies were enumerated and expressed in colony forming unit. The total number of cfu is presented in Table.1

The air of the immediate outdoor environment of the laboratory were also sampled and compared with the isolated bacteria from the indoor environment in order to be able to ascertain whether some of the captured bacteria are of outdoor sources.

The colonies were characterized morphologically, microscopically, and biochemically. Some of the biochemical tests carried out included oxidase, catalase, triple sugar iron, and coagulase. (Bergey's manual, 1974; Cheesebrough, 2010).

RESULTS

The airborne bacteria present in the laboratory were quantified by conventional culture-based technique, in which colony forming units on selective media were counted and the results presented. Table 1 shows the bacterial concentration in cfu/ml per week of sampling, while Figures 1 and 2 show the graphical representation of bacterial concentration.

In dry season, the highest air sample mean count was obtained in the third week of sampling: 1.8×10^2 cfu/ml, and the lowest in the eighth week: 0.62×10^2cfu/ml. However, 8.0×10^3 at seventh week and 0.01×10^2 cfu/ml at the first week were obtained as highest and lowest air mean count respectively during the dry season.

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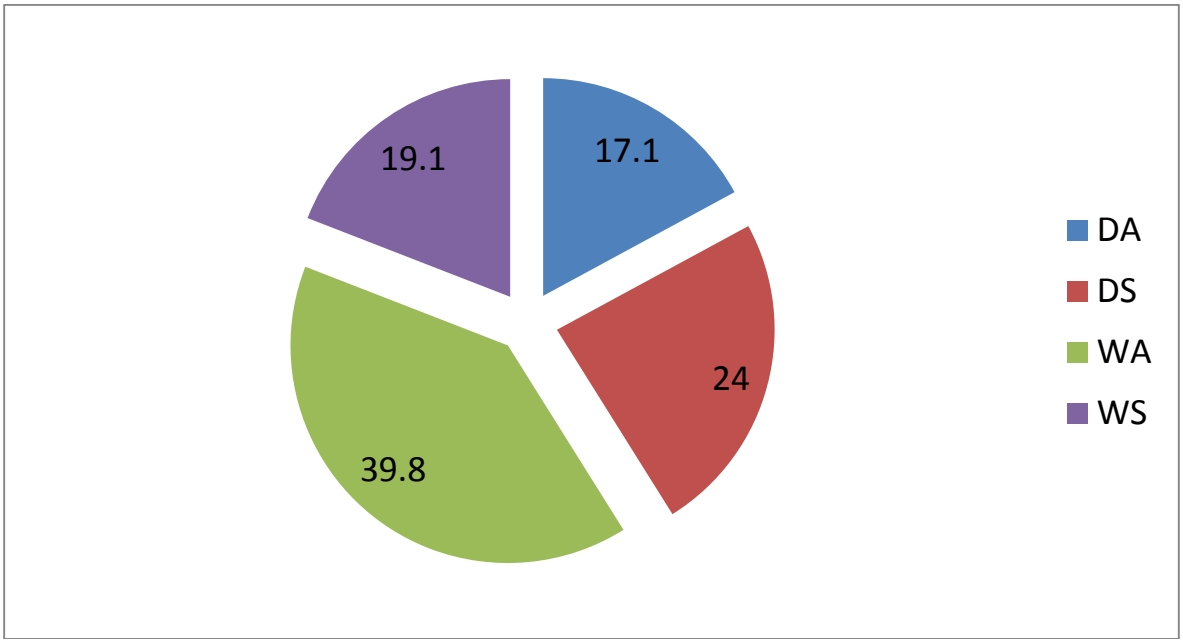
Table 1: Weekly bacterial mean count for dry and wet season (cfu/ml)

WK	DRY SEASON			WET SEASON			
	air	surface	MC/WK	air	surface	MC/WK	TMC/WK
1	1.10	3.00	4.10	0.05	1.73	1.78	5.88
2	1.60	1.90	3.50	2.90	3.00	5.90	9.40
3	1.80	3.00	4.80	1.30	1.50	2.80	7.60
4	1.10	0.15	1.25	1.10	1.50	2.60	3.85
5	1.40	0.61	2.01	0.90	1.50	2.40	4.41
6	1.60	3.00	4.60	1.30	0.10	1.40	6.00
7	0.66	1.50	2.16	8.00	1.50	9.50	11.66
8	0.62	0.73	1.35	7.50	0.25	7.75	9.10
TMC	9.88	13.89	23.77	23.05	11.08	34.13	57.90
%	(17.06)	(24.00)		(39.8)	(19.1)		

MC: mean count; **TMC**: total mean count; **WK**: week

In the surface sampling, the highest mean count 3.0×10^2 cfu/ml occurred in the first, third, and sixth week while the lowest was in the fourth week of sampling during dry season. In wet season, the highest surface counts: 5.9 x 10^2cfu/ml were in second week and the lowest occurred in eight week: 0.25 x 10^2cfu/ml of sampling.
The highest TMC for both air and surface per week occurred in the third week: 4.8 x 10^2 of sampling in the dry season and seventh week: 9.5 x 10^2cfu/ml in wet season.
Some of the bacteria identified based on microbiological and biochemical examinations included *Staphylococcus aureus*, *Micrococcus spp, Bacillus spp*, *Escherichia coli*, *Streptococcus* spp, *Proteus* spp, and *Pseudomonas* spp. others were unidentified. These are similar to those bacteria isolated in a research laboratory within Samaru in a published work of Shiaka *et al*., 2011.
Different species of Bacillus, Streptococcus, Staphylococcus, and Corynebacterium were isolated from the outdoor environment. This implies that some of the isolated bacteria such as staphylococci, streptococci, bacilli might be partly imported into the laboratory from outdoor sources via the opened window and door. More so, Staphylococci are well known as members of skin flora and could be shed from the skin of patients, visitors and the laboratory workers.
Figure 1 shows the bacterial total mean count of air and surface in both dry and wet season (%) during the period of investigation. The highest (39.80) and lowest (17.06) percentages occurred in air during wet and dry season respectively.



DA: total air bacterial count in dry season **DS**: total surface bacterial count in dry season
WA: total air bacterial count in wet season **WS**: total surface bacterial count in wet season
Fig 1: Bacterial total mean count of air and surface in dry and wet seasons (%)

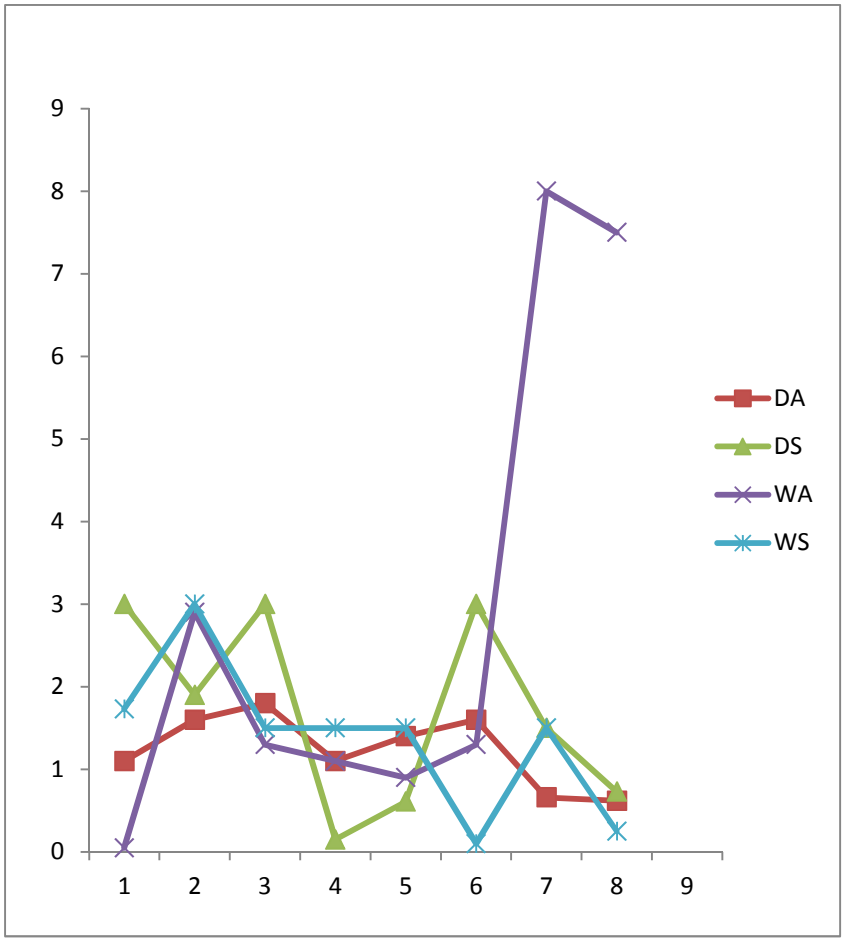


Fig 2: Bacterial concentration of dry and wet season per week

DA: air bacteria in dry season **DS**: surface bacteria in dry season **WA**: air bacteria in wet season **WS**: surface bacteria in wet season

The relationship between bacterial concentration of air in dry and wet season using paired sample t-test (95% confidence interval) was significant while the relationship between bacterial concentrations of the work surfaces in both seasons was not significant.

DISCUSSION

Several studies on microbial contaminants in different indoor environments including hospitals (Obbard and Fang, 2003, Ekaise *et a*l., 2008) have been recorded. Bacteria are known to occur in most environments and many of the species isolated in this study are harmless and frequently include members of the genera bacillus and micrococcus.

The extremely high bacterial concentration of 8.0×10^2cfu/ml and 7.5×10^2cfu/ml as shown in Figure 2 was due to mechanical ventilation (use of ceiling fan) at the time of sampling. This confirms the idea of aero microbiologists that air microbes are usually agitated when subjected to wind.

The surface concentration during the dry season was a bit high compare to the others. This might be due to inefficiency or and inappropriate use of disinfectants which could give room to re-aerosolisation of bacteria present on the same work benches.

The coagulase negative *Staphylococcus spp* isolated is a non-pathogenic group of bacteria which belong to the normal skin microflora and mucous membranes of the respiratory tract. Pseudomonas is also normal flora of the skin, as well as the respiratory, gastro-intestinal and urinary tracts. The *E.coli* is an indicator organism and its presence in the indoor laboratory under consideration reflects contamination. Bacilli were the commonest among the others. This might be due to their high capability to withstand environmental stress (Gatchalian, 2010).

Gram stain and microscopy tests had found that the gram positive rod and cocci shape bacteria were higher than the gram negative rod and cocci. This is similar to the result of Ismail and Ismail 2011 on the study of airborne bacterial contamination in indoor air environment at Universiti Malaysia Terengganu and that of Obbard and Fang: airborne concentrations of bacteria in a hospital environment in Singapore in 2003. However, the low concentration of gram negative bacteria which are pathogenic could potentially become higher, posing

health threat to most especially the laboratory workers if there is less improvement in frequency of cleanliness and hygiene in the laboratory.

The total concentrations of airborne bacteria in this study did not exceed the standard concentration approved by the American Conference Governmental of Industrial Hygienist (ACGIH) of 500 cfu/ml. This indicates that the hygienic level of the laboratory under investigation is acceptable.

Generally, the indoor bacteria concentrations of the laboratory are lower than the outdoor concentrations this is also acceptable based on the general idea of ACGIH 1989 and Macher *et al.,* 1995 on the indoor microflora concentrations of a healthy work environment. No occupational limit for bioaerosols has been promulgated by the occupational safety and health administration (OSHA). However, ACGIH (1989) states that concentrations of less than 100 cfu/ml may be unhealthy to immunosuppressed people.

A low airborne concentration of microorganisms, in and of itself does not indicate a clean and healthy environment. The airborne microbial concentration and the types could be useful in determining the degree of cleanliness of an environment as well as an index they bear in relation to human health (Bhatia, 2011). Hence adequate attention should be given to maintenance of proper hygiene in the laboratory environments since it is well known to be a reservoir of microorganisms. This would to a greater extent assist to minimize the health risk of workers among others who usually spend quality time in the laboratory. Potentially effective strategies including limiting entrance of outdoor aerosols, installing appropriate filtration devices in order to allow inlet filtered outdoor air into the indoor environment of the laboratory, keeping the relative humidity level below high levels (<60%) removing or reducing contaminant sources such as indoor organic waste, and indoor air treatment (Burge, 1990; Peccia, 2008).

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