Full Length Research Paper

Microbial load of indoor airborne bacteria and fungi in a teaching hospital in Ghana

Enoch Kwaku Larrey, Jacob Nii Ayitey Laryea, Stephen Wilson Kpordze and Courage Kosi Setsoafia Saba*

Department of Biotechnology, Faculty of Agriculture, University for Development Studies, P. O. Box 1882, Tamale, Ghana.

Received 4 February, 2020; Accepted 25 February, 2020

The hospital, even though a place for the sick people, is inhabited by medical staff, administrative, security and patients who may share common microbes in the hospital environment. Some of these organisms may be pathogenic and may pose health hazard to humans through inhalation in the hospital atmosphere. Hence, the objective of this study was to investigate the microbial load of airborne microorganisms in some selected units of the Tamale Teaching Hospital and to suggest possible means to reduce the load. The microbial loads of nine units of the Tamale Teaching Hospital were estimated. Passive air sampling technique, applying open Petri-dishes containing different culture media, was employed to collect sample twice daily for the two seasons witnessed in Ghana. The concentrations of airborne microorganisms in the indoor environment of the wards ranged between 277.61 – 5395.14 CFU/m². The statistical analysis showed that the highest bacterial and fungal loads, 5395.14 CFU/m³ and 2021.87 CFU/m³, respectively were recorded in outpatient department (OPD) and the least, 492.37 CFU/m³ and 277.61 CFU/m³, respectively observed in the theatre ward. Staphylococcus aureus, Escherichia coli and Bacillus subtilis were among the bacterial isolates. The fungal isolates include: Aspergillus spp., Alternaria spp., and Fusarium spp. Generally, all units of the hospital harbored very high concentrations of bioaerosols, except the theatre ward.

Key words: Indoor air quality, Hospital air quality, microbial load, open-plate technique, Ghana.

INTRODUCTION

It is estimated that majority of the world’s population spend most of their time indoors: in houses, offices, schools and hospitals, where they encounter environmental factors which compromise their health and physical conditions (Leech et al., 2002). The World Health Organization (WHO) has described the problems of the quality of air in buildings as significant risk factors for human health in low, middle and high income countries (Gsvej, 2009). Furthermore, Fekadu and Getachewu (2015) described the healthcare settings particularly in under-developed countries as breeding grounds for the growth and survival of microorganisms due to overcrowding, improper building design and poor ventilation. These microorganisms could negatively

*Corresponding author. E-mail: csetsoafia@uds.edu.gh. Tel: +233543446929.

Author(s) agree that this article remain permanently open access under the terms of the Creative Commons Attribution License 4.0 International License.
impact human health. According to Fanger (2000), the investigation of the air quality in indoor environments is known as indoor air quality (IAQ). It has been reported that healthcare facilities should pay heed to indoor air concerns because of airborne microorganisms that have the potential to cause nosocomial infection (Ekhaise et al., 2008; Weiss et al., 2010). Nosocomial infections (hospital) during treatment, which are not the reasons for patients’ visit to the hospital. Patients who visit hospitals are prone to nosocomial infections that are driven by the degree of hygienic conditions of the hospital environment, the number as well as the kind of visitors (Adebolu and Vhriterhire, 2002). Also, a study by Fracchia et al. (2006) reported that exposure to bioaerosols that contain microorganisms and/or their by-products predisposes a person to diseases of the respiratory systems and other potentially dangerous effects like infections, hypersensitivity pneumonitis and toxic reactions. Sneezing is the most vigorous mechanism that generates millions of airborne microbes into the hospital environment (Pasquarella et al., 2000). Meanwhile, it is believed that the principal factors responsible for the build-up and spread of microorganisms in the hospital environment are the activities of people and the use of equipment (Ekhaise et al., 2008). Exposure studies have been conducted in many hospital facilities of several countries, but to the best of our knowledge, the status of airborne microbial load in health facilities in Ghana is unknown. This could be a potential source for infection of wounds, skin infections, and respiratory disease. This study aims to investigate the microbial load of airborne bacteria and fungi in the Tamale Teaching Hospital (T.T.H) and also identify the airborne bacteria and fungi in the hospital.

MATERIALS AND METHODS

Study area

This study was conducted in the T.T.H which is a regional hospital in the Northern region of Ghana. It serves as a referral hospital for the three Northern regions of Ghana and has 478 beds and over 2000 workers. It is the third largest teaching hospital in Ghana after Korle-Bu Teaching Hospital and the Okomfo Anokye Teaching Hospital.

Sample collection

The samples for the study were collected from nine different units in the hospital: the outpatient department (OPD), emergency ward (EW), children’s ward (CHW), intensive care unit (ICU), theatre ward (TW), surgical ward (SW), maternity ward (MW), administration (ADM), and the premise of the hospital (Outside). Samples were taken twice in the year with respect to the seasons in Ghana, thus wet season and dry season (harmattan). Plate exposure method, which requires that plates with specific culture media be opened for a specified duration, was used for this study (Bhatia and Vishwakarma, 2010; Ekhaise et al., 2010). This method allows bacteria or fungi in the air to settle on the respective culture media. Prepared plates of Nutrient agar, MacConkey agar (MAC) and Potato Dextrose agar (PDA) were exposed for 30 min at a height of 1 to 1.5 m from the floor to simulate human breathing zone in different wards for both morning and evening (Bhatia and Vishwakarma, 2010). Nutrient agar was used to culture bacteria for identification their loads, MacConkey agar was used to isolate gram negative airborne bacteria and PDA was also used to isolate fungi and also determine their loads. A total of two hundred sixteen (216) petri dishes were used for all of the three different media; two petri dishes were used per room for each medium. After sampling, the plates were transported to the Microbiology Laboratory of the Spanish laboratory complex in University for Development Studies - Nyankpala campus on ice chest with ice cubes for incubation. The plates were incubated for 48 h at 37 and 44.5°C in the case of analysis of bacterial samples collected with nutrient agar and MacConkey agar, respectively and for five to seven (5-7) days at 25°C in the case of fungal identification.

Bacterial count and identification

After incubation, the total number of colony forming units (CFU) for bacteria and fungi were enumerated and converted to organism colony forming units per cubic meter (CFU/m³). The bacterial isolates were identified on the basis of their morphology, microscopic appearance (using staining techniques), and biochemical reactions. The fungal isolates were identified on the basis of microscopic (employing lactophenol cotton blue staining and wet mounting techniques) and colonial growth.

Statistical analysis

Data were cleaned and entered into the computer, and analyzed using Genstat version 12.0 and where there was significant difference, XLD was used to separate the means at 5%. Descriptive statistics was used to summarize the data. Indoor air bacterial and fungal loads in the different rooms of the hospital were compared using ANOVA.

RESULTS

The results of the study on the microbial load in the T.T.H are given in Tables 1, 2, 3 and Figures 1 and 2. The results indicate that the lowest microbial load for both bacteria and fungi was observed in the theatre ward, 492.37 and 277.61 CFU/m³, respectively (Table 1). The highest concentration of bacteria and fungi was recorded in the outpatient department, 5395.14 and 2021.87 CFU/m³, respectively (Table 1). Table 2 indicates that seasonal variation has a significant effect on the microbial load in the studied units. While the two seasons observed in Ghana did witness a significant effect on the overall microbial load at TTH (Figure 2), time of air sampling on the other hand did not have a great effect (Figure 1). However, individual comparison of the time of air sampling in the various units showed a great effect on the microbial concentration.

Bacterial isolates

In this study, the bacterial isolates were Bacillus subtilis,
Table 1. General microbial load in the various units.

| Unit | Bacteria (CFU/m³) | Fungi (CFU/m³) | P-value |
|------|------------------|----------------|---------|
| ADM  | 969.03<sup>BC</sup> | 974.27<sup>AB</sup> |         |
| CHW  | 4622.54<sup>A</sup> | 1427.36<sup>AB</sup> |         |
| EW   | 4824.20<sup>A</sup> | 1704.97<sup>AB</sup> |         |
| ICU  | 2571.86<sup>ABC</sup> | 955.94<sup>AB</sup> |         |
| MW   | 4308.26<sup>AB</sup> | 929.75<sup>AB</sup> | <0.001  |
| OPD  | 5395.14<sup>A</sup> | 2021.87<sup>A</sup> |         |
| OUT  | 4787.53<sup>A</sup> | 1796.63<sup>A</sup> |         |
| SW   | 3582.79<sup>ABC</sup> | 1521.64<sup>AB</sup> |         |
| TW   | 492.37<sup>C</sup>  | 277.61<sup>B</sup>  |         |

Data are expressed in mean. Mean values followed by different superscripts in the same column are significantly different (p<0.05).

Table 2. The influence of season on the microbial load in the various units.

| Unit | Bacteria (CFU/m³) | Fungi (CFU/m³) | P-value |
|------|------------------|----------------|---------|
| ADM  | 1126.17<sup></sup> | 728.08 | 1225.69 | 0.001|
| CHW  | 4517.78<sup></sup> | 2252.34 | 1157.60 |         |
| EW   | 6110.13<sup></sup> | 1924.97 | 929.75 |         |
| ICU  | 4813.72<sup></sup> | 1592.35 | 322.14 |         |
| MW   | 4688.01<sup></sup> | 1230.93 | 628.56 |         |
| OPD  | 6081.32<sup>A</sup> | 3069.49<sup>A</sup> | 974.27<sup>B</sup> | 0.001|
| OUT  | 7555.82<sup></sup> | 2744.71 | 780.46 |         |
| SW   | 5795.85<sup>A</sup> | 2265.43 | 780.46 |         |
| TW   | 523.8<sup>C</sup>  | 361.42 | 196.43 |         |

Data are expressed in mean. Mean values in the same column followed by different superscripts are significantly different (p<0.05).

Table 3. The microbial load of both bacteria and fungi with respect to time in the units.

| Unit | Bacteria (CFU/m³) | Fungi (CFU/m³) | P-value |
|------|------------------|----------------|---------|
| ADM  | 1427.36 | 1663.07 | 288.09 |         |
| CHW  | 4130.16 | 1052.84 | 1801.87 |         |
| EW   | 4811.72 | 1592.35 | 322.14 |         |
| ICU  | 3632.55 | 1288.55 | 620.70 |         |
| MW   | 3514.70 | 934.98 | 921.89 | 0.041|
| OPD  | 6455.84 | 2422.58 | 1623.78 |         |
| OUT  | 641.11 | 1702.35 | 1890.92 |         |
| SW   | 4111.83 | 1642.11 | 1401.17 |         |
| TW   | 641.66 | 340.4 | 235.71 |         |

Bacillus cereus, Yersinia enterocolitica, Salmonella enterica, Staphylococcus aureus, Escherichia coli, Vibrio vulnificus, and Vibrio cholera.

Fungal isolates

The fungal isolates were Aspergillus species, Alternaria.
species, *Fusarium* species, *Mucor* species, *Penicillium* species, *Verticillium* species, *Abida corymifera*, *Epicoccum nigrum*, *Phema glemerata*, and *Rhizopus stolonifer*.

**DISCUSSION**

The results of this study reveal that the outpatient department had the highest microbial load for both bacteria and fungi. This could be due to the open nature of this unit which may allow bioaerosols from outdoor environment to flow in freely. This is in relation to the review of indoor bio-aerosols by Nazaroff (2016), who is of the view that, circulating bioaerosols have 100% ability to penetrate naturally ventilated buildings, implying that these bioaerosols eventually get indoors through leakages and openings of the buildings. There is no fixed universally accepted threshold for microbial load in built environments as several institutions have proposed different limits. For instance, while an expert group of WHO suggested a limit of 1000 CFU/m³ (Heseltine and Rosen, 2009), some scholars are of the view that it should not exceed 750 CFU/m³ (Rao et al., 1996). Furthermore, bioaerosols concentrations between 4500 and 10000 CFU/m³ have also been suggested for ubiquitous bacteria.
(Hameed and Habeebullah, 2013). Hence, the microbial loads in buildings can only be compared to these propositions.

Generally all the units were highly contaminated with microorganisms, with the exception of the theatre ward, the intensive care unit, and the administration block which recorded the least microbial load. A major factor that could have attributed to the high microbial load could be the fact that doors leading to the various wards were left opened for the most part of the day. The high microbial load could also be attributed to overcrowding. Being the only referral hospital serving the three northern regions of Ghana, there is so much pressure on the facility hence the carrying capacity of the hospital is far exceeded.

The results of this study affirm the reports of several studies which have reported that outdoor air microbial concentration plays significant role in raising and homogenization of the microbial load of the indoor air through the passage of bioaerosols through windows, doors and other openings into the built environment (Hyvärinen et al., 2001; Jaffal et al., 1997; Rainer et al., 2001). The result is also backed by the findings of Adams et al. (2015), who reported human occupancy as a significant factor affecting the number and community structure of bioaerosols existing in the built environment, especially in poorly ventilated or heavily occupied buildings.

The intensive care unit, theatre ward, and administration blocks which recorded the least microbial loads were enclosed areas with very high discipline as far as hygiene is concerned. These wards (intensive care unit and theatre ward) had stringent measures in place for people entering and had few patients in there. The low microbial load in these wards was again anticipated due to the high sanitary standards in these areas, compared to other hospital areas. It is worth noting that microbial rates in the theatre ward and intensive care unit were dependent on the hospital. The location of the theatre ward and intensive care unit is very important in order to reduce the microbial exchange with the other units through the air. These wards were isolated from the other wards taking the last but one and last floors of the storey building and had very good ventilation in contrast to the rest.

Ghana has two major seasons, dry and wet. The Northern part where the study was conducted has a single wet season occurring between May and November of the year. The Southern part on the other hand has two wet seasons divided into the major (March to July) and the minor (September to November) seasons (Owusu and Waylen, 2013). The results obtained in this study revealed the frequency of both airborne bacteria and fungi in the dry season to be dominant than that of the wet season. This contradicts the findings of Ekhaise and Ogboghoodo (2011), who found the bacterial load to be higher in the wet season in two major hospitals in Benin city, Nigeria. This could be as a result of geographical differences. It is a proven fact that temperature and relative humidity are two important factors for fungal spore generation, release and dispersal; especially in indoor environments. This could be attributed to the relatively high atmospheric dust particles in the atmosphere due to the north east trade winds during the dry season. Rintala et al. (2008) reported no significant difference in seasonal variation of bacterial load. The findings of this study, however, disagree with that earlier report in that seasonal variations between bacterial loads were highly significant.

The study further reveals that, generally, the microbial load was higher in the morning than in the evening. One would expect that due to the cleaning which takes place in the hospital every morning, the microbial load would be lower. The major factor which could have attributed to this is the busy nature of the hospital from morning to noon hours of the day when large number of people troop in to receive medical attention. Obbard and Fang (2003), showed that occupant density is a crucial factor affecting concentrations of airborne bacteria, their results showed that occupant density was dependent upon the time and this supports our findings.

Aspergillus species was the predominant fungi isolated from the hospital. Augustowska and Dutkiewicz (2006), brought to light that, Aspergillus species could cause invasive Aspergillosis and produce mycotoxins which are known to be carcinogens. Jain (2000) revealed that respiratory diseases, hypersensitivity reactions and allergies could also be caused by other fungal spores not only in immune suppressed patients but also in healthy individuals. The microorganisms (bacteria and fungi) isolated from the Tamale Teaching Hospital were also reported by Ekhaise et al. (2010), who reported similar isolates in their work which was conducted in the University of Benin Teaching Hospital, Nigeria.

**Conclusion**

In conclusion, the results generated in this study obviously suggest that regardless of season, the units considered in this study had very high microbial load. Thus, the Tamale Teaching Hospital should adopt effective measures to reduce the microbial loads in the wards to the barest minimum. Keeping doors closed, use of good ventilation system other than natural ventilation, regulation of visits, and sticking to the carrying capacity of the hospital could be vital to decreasing the microbial load of the Tamale Teaching Hospital.

**CONFLICT OF INTERESTS**

The authors have not declared any conflict of interests.

**ACKNOWLEDGEMENTS**

We thank the Tamale Teaching Hospital for their...
permission. We also acknowledge the help of Mr. Joseph Nzech, Mr. Isaac Asirifi, Mr. Joseph Kwodaga and Mr. Gonu Hellie

REFERENCES

Adams RI, Bhangar S, Pasut W, Arens EA, Taylor JW, Lindow SE, Nazaroff WW, Bruns TD (2015). Chamber Bioaerosol Study: Outdoor Air and Human Occupants as Sources of Indoor Airborne Microbes. PLOS ONE 10(5):e0128022.

Adebolu TT, Whitterhire KJ (2002). Survey of the microbial flora of the Ondo State Specialist Hospital Environment, Akure, Nigerian Journal of Microbiology (112):91-94.

Augustowska M, Dutkiewicz J (2006). Variability of airborne microflora in a hospital ward within a period of one year. Annals of Agricultural and Environmental Medicine 13(1):99-106.

Bhatia L, Vishwakarma R (2010). Hospital indoor airborne microflora in private and government-owned hospitals in Sagar City, India. World Journal of Medical Sciences 5(3):65-70.

Ekhaise FO, Ighosewe OU, Ajakpovi OD (2008). Hospital Indoor Airborne Microflora in Private and Government-Owned Hospitals in Benin City, Nigeria. Journal of Medical Sciences 3(1):19-23.

Ekhaise FO, Isitor EE, Idehen O, Emoghene AO (2010). Airborne microflora in the atmosphere of an hospital environment of University of Benin Teaching Hospital (UBTH), Benin City, Nigeria. World Journal of Medical Sciences 6(2):166-170.

Ekhaise AO, Fred O, Ogbogodo BI (2011). Microbiological Indoor and Outdoor Air Quality of Two Major Hospitals in Benin City, Nigeria. Sierra Leone Journal of Biomedical Research 3(3):169-174.

Fanger PO (2000). Indoor air quality in the 21st century: search for excellence. Indoor Air 10(2):68-73.

Fekadu S, Getachewu B (2015). Microbiological assessment of indoor air of teaching hospital wards: A case of Jimma University Specialized Hospital. Ethiopian Journal of Health Sciences 25(2):117-122.

Fraccia L, Pietronave S, Rinaldi M, Martinotti MG (2006). The assessment of airborne bacterial contamination in three composting plants revealed site-related biological hazard and seasonal variations. Journal of Applied Microbiology 100(5):973-984.

Gshej S (2009). Dampness and Mould. World Health Organization Regional Office for Europe. Available at: http://www.euro.who.int/__data/assets/pdf_file/0017/43325/E92645.pdf

Hameed A, Habeeballah T (2013). Air Microbial Contamination at the Holy Mosque, Makkah, Saudi Arabia. Current World Environment Journal 8(2):179-187.

Heseltine E, Rosen J (2009). WHO guidelines for indoor air quality: dampness and mould. Available at: https://www.ncbi.nlm.nih.gov/books/NBK143941/

Hyvärinen A, Vahteristo M, Meklin T, Jantunen M, Nevalainen A, Moschandreas D (2001). Temporal and Spatial Variation of Fungal Concentrations in Indoor Air. Aerosol Science and Technology 35(2):688-695.

Jaffal AA, Banat IM, El Mogheth AA, Nsanze H, Bener A, Ameen AS (1997). Residential indoor airborne microbial populations in the United Arab Emirates. Environment International 23(4):529-533.

Jain AK (2000). Survey of bio-aerosol in different indoor working environments in central India. Aerobiologia 16(2):221-225.

Leech JA, Nelson WC, Burnett RT, Aaron S, Raizenne ME (2002). It's about time: A comparison of Canadian and American time-activity patterns. Journal of Exposure Science and Environmental Epidemiology 12(6):427-432.

Nazaroff WW (2016). Indoor bioaerosol dynamics. Indoor Air 26(1):61-78.

Obbard JP, Fang LS (2003). Airborne concentrations of bacteria in a hospital environment in Singapore. Water, Air, and Soil Pollution 144(1-4):333-341.

Owusu K, Waylen PR (2013). The changing rainy season climatology of mid-Ghana. Theoretical and Applied Climatology 112(3-4):419-430.

Pasquarella C, Pitzurra O, Savino A (2000). The index of microbial air contamination. Journal of Hospital Infection 46(4):241-256.

Rainer J, Peintner U, Pöder R (2001). Biodiversity and concentration of airborne fungi in a hospital environment. Mycopathologia 149(2):87-97.

Rao CY, Burge HA, Chang JCS (1996). Review of Quantitative Standards and Guidelines for Fungi in Indoor Air. Journal of the Air and Waste Management Association 46(9):899-908.

Rintala H, Pitkaranta M, Toivola M, Paulin L, Nevalainen A (2008). Diversity and seasonal dynamics of bacterial community in indoor environment. BioMed Central Microbiology 8(1):56.

Weiss KD, Osborne SF, Callahan-Lyon P (2010). Prevention of surgical-site infections. The New England Journal of Medicine 362(16):1541-1542.