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Original Article

Bacteriological contaminants of indoor air in a neonatal intensive care unit of a Nigerian hospital

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Abstract

Background: The indoor air contamination by microorganisms constitute source of potential infection to the occupants of the room, and is now a potential threat to the healthcare system because of the increase risk of nosocomial infection out-breaks.

Objective: The study aimed at assessing the bacteriological quality of the indoor air at the Neonatal Intensive Care Unit (NICU) of the University of Ilorin Teaching Hospital, Ilorin, Nigeria (UITH).

Materials and Methods: This was conducted over a period of 3 days in June, 2018. The passive air sampling technique was adopted using the 1/1/1 scheme. A set of Blood and McConkey agar plates were kept at 1m away from the floor and side walls of the room and exposed to the indoor air for one hour and there after they were incubated at 35^0C - 37^OC for 18-24hours. This process was performed between 10.00-11.00 am and 3.00-4.00 pm daily in the NICU which was partition into four segments. Standard microbiologic methods were employed for the identification of the bacterial isolates. The density of the bacterial contaminants was calculated in colony forming unit per cubic meter (cfu/m^3). The mean density of the bacterial contaminants for each of the four sections of the NICU, the total mean density, and the Grand mean density were also calculated. The Grand mean density was compared with the American Industrial Hygienist Association (AIHA) acceptable limit ($5x10^2$ CFU/m^3).

Results: Six bacterial contaminants were isolated namely: *Staphylococcus aureus* (44.9%), *Staphylococcus epidermidis* (14.1%), *Micrococcus* (19.2%), *Klebiella pneumonia* (10.3%), *Bacillus spp*. (6.5%), and *Staphylococcus saprophyticus* (5.1%). The highest and the lowest total mean density of the indoor air contaminants were 4.436 X 10^3 CFU/$m^3$and 2.509 X 10^3 CFU/m^3 respectively. Highest densities of bacterial air contaminants were obtained during the peak of clinical activities in the morning hours. The Grand mean density was significantly (0.006) above the acceptable AIHA standard.

Conclusion: There is need for a review of the indoor air decontamination measures in the hospital NICU towards making it safe, efficient and to reduce the risk of hospital acquired infections.

Introduction

Air is the source of oxygen, an essential requirement for human and animal survival. Pure air is a mixture of gases and it includes 78.0% nitrogen, 21.0 % oxygen, and some other gases as well as varying amounts of water vapour[1].

According to National Health and Medical Research Council (NHMRC), indoor air is defined as the one within a building occupied for at least one hour by people of varying states of health[2]. Therefore, indoor air quality is closely related to human's health and wellbeing[3].

Air pollution can occur when the environment is contaminated by any chemical, physical or biological agents that modify the natural characteristics of the atmosphere[4]. It is one of the most important problems of our age which has now reached an advanced level, constituting a potential threat to the health and wellbeing

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of the population[5]. More than 4.6 million people die every year due to air pollution most of which are indoor air pollution associated[6]. In 2002, World Health Organization stated that 2.7% of the global burden of ill-health is attributed to indoor air pollution with 80% occurring in Africa and South East Asia[7].

Biological contaminants of air include bacteria, fungi and viruses. Majority of bacteria in the hospital air are harmless saprophytes or commensals. Only about 0.01-0.1% of the air-borne bacteria are pathogenic[8]. Bacteria in air are accidental contaminants, air not being their natural habitat but medium through which they are disseminated or transmitted[9]. Dissemination of pathogenic microbes in air has significant impact on human health and particularly challenging with reference to hospital acquired infections[10].

Within hospital confines, the main contributors to indoor air bacterial flora are the ventilation and air conditioning system, high dusting, various medical devices for respiratory support, porous wet surfaces, patients and health care givers[11]. The diversity and spread of organisms is enhanced by sneezing and coughing, high door traffic, environmental disturbances and other activities[12]. When bacteria in the air are inhaled, they can cause or aggravate diseases of the respiratory system such as acute respiratory infection (ARI), chronic bronchitis, acute exacerbation of asthma and chronic obstructive pulmonary diseases (AECOPD), tuberculosis and possibly aggravate lung cancer[13]. Allergic reactions such as hypersensitivity pneumonitis, humidifier fever, and allergic rhinitis among many others may follow inhalation of bacterial contaminant of the air[14].

The indoor air of NICU is recognized as a potential reservoir of pathogenic bacteria[15]. Unfortunately, Low Birth Weight (LBW) and Extremely Low Birth Weight(WLBW) infants which are typically immunocompromised are admitted into the NICUs and therefore vulnerable to hospital acquired infections [15]. NICU therefore requires highest hygiene standards, which is directly related to the healthcare providers' attitude towards infection control and equipment maintenance culture[15].

Indoor air quality control measures and monitoring are equally important for reducing dissemination and nosocomial infection out-breaks[16-17]. Proper cleaning, disinfection, and sterilization of surfaces reduce spread of the contaminants and infection risk[18].

The common air borne microorganisms associated with hospital acquired infection in the NICU are *Streptococcus pneumoniae*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Haemophilus influenza*, and members of the enterobacteriacea[19]. Infants in the NICUs are particularly susceptible to opportunistic infections because their lungs are still in the developmental stage and because they have immature immune systems[20]. Exposure of such babies to bacterial contaminants of air poses a significant risk and may result in poor outcome of management of the diseases for winch they have been admitted.

In the NICU of UITH, the air quality is maintained by visitor's restriction to certain areas. Mothers of admitted neonates are allowed entry only to feed their wards. General cleaning is usually done in the mornings and evenings. We hypothesized that the indoor air quality of NICU is line with the acceptable standards. There has been no previous attempt to assess the bacteriological quality of the indoor air of NICU at UITH. This is an exploratory study and therefore aimed at determining the pattern and density of bacteriological contaminants in the indoor air of the NICU of UITH.

Material and Methods

Study design: This was conducted over a period of 3 days in June, 2018 at the NICU of UITH. It is a 600 bed hospital that serves as a referral center to the Middle belt zone in Nigeria. Even though there is no record of fumigation, the NICU is usually fumigated whenever there is clinical or laboratory evidence of infection out-breaks in the unit. The hospital has an infection control unit and team headed by a microbiologist. The hospital is in the process completing documentation of her infection control policy.

Air Sampling Method

Passive air sampling technique was used to access the bacteriological contaminant of air in the NICU of UITH. Briefly, this sampling technique is a sedimentation method based on the 1/1/1 principle. During sampling, agar plates were placed 1m above the floor, 1m away from the wall and for 1hour in the rooms to be sampled. Blood and McConkey agar were the media used.

Sampling was done in the mornings (10-11am) and evenings (3-4 pm) for three days, with eight plates being placed in each of the rooms in the mornings and evenings making a total of thirty-two plates per day and ninety-six plates at the end of the third day.

Bacterial Isolation

Inoculated media were then aerobically incubated at 35^0C - 37^0C for 18-24 hours after which they were examined for growth. Purity plating was done to obtain pure isolates which were then characterized using standard microbiological methods. Antimicrobial susceptibility testing was performed by the modified Kirby- Bauer disk diffusion method and interpreted according to Clinical and Laboratory Standards Institute (CLSI) [21] but its result is to be reported elsewhere.

Calculation of bacteriological contaminants density in indoor air.

The density of bacteriological indoor air contaminants was expressed as colony forming units per cubic meter of air (cfu/m3). The density of bacteriological indoor air contaminants was derived using the formula of Stryjakowska-Sekulska [22]

$CFU/m^3 = A \times 10^4 / P \times T \times 0.2$

Where **A** = Numbers of colonies on the Petri-dish

P = The surface area of the Petri plate

T = Time (duration) of the Petri dishes exposure

0.2 = Constant.

The NICU was partition into four sections at UITH (Figure 1). The mean density of bacteriological indoor air contaminants for each of the four sections was calculated by finding the average densities derived from each of the eight agar plates for morning and evenings and the 3days. The Total mean density is the sum of the mean densities for mornings and afternoons while the Grand mean density is the sum of all total mean densities. The Grand mean density was statistically compared with the American Industrial Hygienist Association (AIHA) acceptable standard ($5x10^2$ CFU/m^3). The study was approved by the Ethical Review Committee (ERC) of UITH.

Results

The highest and the lowest total mean density of the indoor air contaminants were 4.436 X 10^3 and 2.509 X 10^3 respectively. Highest densities of bacterial air contaminants were obtained during the peak of clinical activities in the morning hours. The Grand mean density ($14.238x10^3$) was significantly (0.006) above the acceptable AIHA standard ($5x10^2$ CFU/m^3)

Six bacterial indoor contaminants were identified namely: *Staphylococcus aureus* (44.9%), *Staphylococcus epidermidis* (14.1%), *Micrococcus* (19.2%), *Klebiella pneumonia* (10.3%), *Bacillus spp*. (6.5%), and *Staphylococcus saprophyticus* (5.1%). The density of *Staphylococcus aureus* ($6.92 x10^3$ CFU/m^3) and *Bacillus spp*. ($9.81x10^3$ CFU/m^3) were individually higher than the AIHA standard.

Table 1: Mean density of the contaminants for each room

Sample sites	Time of sampling	Density Day 1 Cfu/m^3	Day 2 Cfu/m^3	Day 3 Cfu/m^3	Mean density Cfu/m^3	Standard AIHA limit Cfu/m^3
Room A	**Morning**	**2.54 x10^3**	**1.95 2 x10^3**	**2.50 3 x10^3**	**2.331 x10^3**	
	Afternoon	**1.035 x10^3**	**2.46 3 x10^3**	**2.81 7 x10^3**	**2.105 x10^3**	
Total mean					**4.436 x10^3**	
Room B	**Morning**	**9.04 x10^2**	**2.75 2 x10^3**	**2.89 6 x10^3**	**2.184 x10^3**	
	Afternoon	**6.81 x10^2**	**2.55 5 x10^3**	**3.01 4 x10^3**	**2.083 x10^3**	
Total mean					**4.267 x10^3**	
Room C	**Morning**	**3.210 x10^3**	**7.73 x10^2**	**9.83 x102**	**1.655 x10^3**	
	Afternoon	**1.559 x10^3**	**1.17 9 x10^3**	**1.37 6x 103**	**1.371 x10^3**	
Total mean					**3.026 x10^3**	
Room D	**Morning**	**1.402 x10^3**	**7.60 x10^2**	**6.6 x10^1**	**7.43 x10^2**	
	Afternoon	**2.883 x10^3**	**1.17 9 x10^2**	**3.28 x10^2**	**1.766 x10^3**	
Total mean					**2.509 x10^3**	
Grand mean					**14.238 x10^3**	**5x10^2 0.006**
P-value						

Room A = In- born, Room B= out- born, Room C- Nurses station, Room D- Main entrance

Table2: Mean density of each type of organism

Organisms/ Rooms	Room A Cfu/ m^3	Room B Cfu/ m^3	Room C Cfu/m^3	Room D Cfu/m^3	Total Cfu/ m^3
S. aureus	**2.564x 10^3**	**1.729x 10^3**	**1.393x 10^3**	**1.234x 10^3**	**6.920x 10^3**
S. saprophyticus	**1.49x 10^2**	**7.7x 10^1**	**2.6x10^1**	**9x10^0**	**2.61x 10^2**
S. epidermidis	**10^3**	**10^2**		**10^2**	**10^3**
Bacillus spp.	**5.28x 10^2**	**1.57x 10^2**	**2.18x10^2**	**7.8x10^1**	**9.81x 10^3**
Micrococcus	**9.26x 10^2**	**1.215 x10^3**	**8.60x10^2**	**5.72x10^2**	**3.573x 10^3**
K. pneumonia	**1.48x 10^2**	**1.61x 10^2**	**6.48x10^2**	**1.31x10^2**	**1.088x 10^3**

Room A= In-born, Room B= out- born, Room C- Nurses station, Room D- Main entrance
S = Staphylococcus, K = Klebsiella.

Table 3: Frequency distribution of bacterial contaminants

Variables	*S. aureus* N (%)	*S. epidermidis* N (%)	*S. saprophyticus* N (%)	*Micrococcus* N (%)	*Bacillus spp.* N (%)	*K. pneumonia* N (%)
Room A	**13 (37.1)**	**2 (18.2)**	**1(25)**	**1 (6.7)**	**1 (20)**	**1 (12.5)**
Room B	**5 (14.3)**	**3 (27.3)**	**1(25)**	**2 (13.3)**	**2 (40)**	**4 (50.0)**
Room C	**10 (28.6)**	**4 (11.4)**	**1(25)**	**2 (13.3)**	**1 (20)**	**1 (12.5)**
Room D	**7(20)**	**2 (18.2)**	**1(25)**	**1 (6.7)**	**1 (20)**	**2(25)**
Total N (%)	**35 (44.9)**	**11 (14.1)**	**4(5.1)**	**15 (19.2)**	**5 (6.4)**	**8 (10.3)**

Room A= In-born, Room B= out-born, Room C- Nurses station, Room D- Main entrance (passage). S = *Staphylococcus*, K = *Klebsiella*.

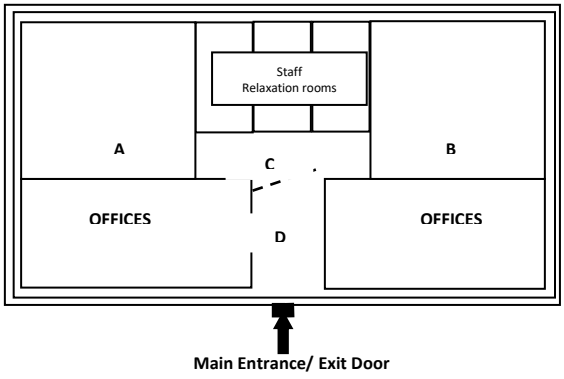

Figure 1: NICU layout at UITH
LEGEND: Room A= In-born, Room B= out-born, Room C- Nurses station, Room D- Main entrance passage/ Lobby

Discussion

The main findings of this study showed that the bacterial load was higher in morning samples than the afternoon samples and they were all above the standard acceptable limit by the AIHA of 500 cfu/m^3. There was a significant difference in the total mean densities of the contaminants

between the rooms of the NICU because the p-value is less than 0.05. All the examined rooms in this study were contaminated with bacteria and the most frequently occurring contaminant was *S. aureus.* The highest bacteria density was recorded in room A which is where the inborn patients are usually admitted. The high density of these contaminants might be potential factors for the spread of hospital acquired infection among neonates in this unit.

The relatively high level of bacterial contaminants in the morning can be attributed to high movement of medical staff due to handing over and mother of the babies in the course of treatment and breast-feeding. Another factor could be inadequate cleaning procedures and routine disinfection; because there is a possibility of increased bacterial concentration in the morning even after the normal cleaning had been done. This implies that the occurrence of these bacterial contaminations was likely due to specific activities or improper cleaning activity. This finding goes in line with the study conducted by Suzuki *et al*., which explain that bacterial load of indoor air is usually higher when the activities is high than when activities is low [23].

Findings from this study revealed that the genera of bacterial contaminants isolated from the NICU indoor environment included: *Staphylococcus aureus* (44.9%), *Bacillus spp*. (6.5%), *Staphylococcus epidermidis* (14.1%), *Staphylococcus saprophyticus* (5.1%), and *Micrococcus* (19.2%), and *Klebsiella pneumoniae* (10.3%). The bacterial contaminants isolated in this study are in conformity with those isolated by similar studies carried out by Awosika *et al*. (2012).[24]

The bacterial contaminants profile of the NICU indoor air showed that S. aureus was the most frequently isolated species among contaminants and the isolation rate was higher in room A than other rooms. This variation may be because of the difference in the presence of carriers, and in cleaning procedure that affect the load of *S. aureus* in air as it was suggested by Chikere *et al*. [25]

Although the isolates were not screened for methicillin resistance, this result was corroborated by a previous study in the same institution that found *S. aureus* is a major contaminant in the NICU. Fadeyi *et al* in 2010 found that medical staff had 52.5% of MRSA carriage rate and the NICU accounted for 50.0% of Methicillin Resistant *Staphylococcus aureus* (MRSA) carriage rate among healthcare workers in the critical care units [26]. MRSA awareness level among the Healthcare givers in this hospital is just a little above 50.0% with substantial proportion of them not realizing the medical implication of the organism [27].

Conclusion

The indoor air of NICU of UITH Ilorin is contaminated with bacteria to a level beyond what is acceptable. There is need to institute measures to maintain acceptable bacterial air contamination limit for the safety of the babies admitted to the unit and health care givers of the unit.

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