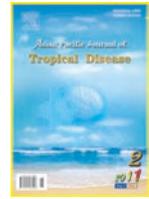




Contents lists available at ScienceDirect

Asian Pacific Journal of Tropical Disease

journal homepage: www.elsevier.com/locate/apjtd

Document heading doi:10.1016/S2222-1808(11)60053-4

Aerobacteriology of laboratories and offices: Evidence of high risk exposure to immune complex formation in Nigeria

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ARTICLE INFO

Article history:

Received 25 February 2011

Received in revised form 20 April 2011

Accepted 21 May 2011

Available online 1 June 2011

Keywords:

Aero-bacteria

Immune complexes

High risk Nigeria

Opportunistic infection

ABSTRACT

Objective: To evaluate the air bacterial load especially in high environmentally polluted areas. **Method:** The air bacterial load of 5 laboratories and 2 offices in Anambra State were sampled, using settled plate (sedimentation) method. **Results:** All the laboratories showed average of 44–55 colony forming units (CFU) within 15 minutes. Both aerobic and anaerobic bacteria were isolated. The predominant bacteria were: *Micrococcus*, *Diphtheroids*, *Staphylococcus* species, *Bacillus* species, *Corynebacterium diphtheriae* (*C. diphtheriae*), *Clostridium* species and *Propionibacterium* amongst other variety of bacteria isolated. Office with air-conditioning system gave a total of 34 CFU out of 74 CFU shown in the two 2 offices, while the office without air-conditioning system gave forty (40) colonies. **Conclusion:** It was found that the laboratories with more number of people, with frequent movement were more air loaded with bacteria than those with less number of people. All the areas sampled, contain significantly high number of colony forming units (CFU) than 3 CFU found in standard clean room $P < 0.001$. It is an evidence of high risk of persistent infections and subsequent immune complex formation.

1. Introduction

The Smith papyrus, dating back to 1600 BC, stated that the first of the incantations to the gods of sickness was devoted to cleansing the “winds of the pest of the year”. In the second century, AD Galen gave forth the ominous pronouncement, “when many sicken and die at once, we must look to a single common cause, the air we breathe. Bioaerosols are airborne particles that are living bacteria, viruses and fungi or originate from living organisms. Their presence in air is the result of dispersal from a site of colonization or growth. The health effects of bioaerosols including infectious diseases, acute toxic effects, allergies and cancer coupled with the threat of bioterrorism and SARS have led to increased awareness on the importance of bio-aerosols[1]. Some studies have shown that the atmosphere

constitutes a major source of various microorganisms which may cause serious ecological and economical problems such as plant and animal diseases, bio deterioration of food and biodegradation of industrial and research products[2]. Average human inhales about 10000 litres of air daily and with it, air-borne particles are deposited in different parts of the respiratory tract due to various physical parameters and subsequent immune complex formation. Upper respiratory tract infection (URI) causes at least one-half of all symptomatic illness in the community, exacting huge tolls that can be measured as morbidity, absenteeism from school and work, direct health care costs, and overuse of antibiotics leading to the emergence of drug-resistant bacteria.

The design of this work became manifest after a serious thought on epidemiology of air borne diseases, which gave birth to this philosophical backing, “that we can avoid food and water even when we need them but we cannot avoid air even when we don’t need it”. Therefore we

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designed this work to know the aerobacterial load of some laboratories and offices in tropical area such as Nigeria; to know the prevalent aero-bacteria and recommend ways of prevention and control of the spread. Hence, renew our fight against environmental pollutions and abuse; create awareness for us to become conscious of our environments including health centres, industries, and research institutions where microorganisms are handled, as well as review the immunological consequences of persistent exposure to infection. The importance of this work should not be over emphasized, considering the fact that human immunodeficiency virus infection has been in the increase with its characteristics of immune system suppression, more individuals are immune compromised and as such, vulnerable to air-borne opportunistic infections.

2. Materials and methods

Air in five different laboratories and two offices known to be in effective use were sampled. One of the laboratories and one of the offices have air conditioning system. The laboratories and offices include: (1) Laboratory A; (2) Laboratory B; (3) Laboratory C; (4) Laboratory D; (5) Laboratory E with air conditioner; (6) office with air conditioner; (7) office without air conditioner.

2.1. Sampling

Settle plate (Sedimentation) method was used due to its ability to retain the viability and complete size of the microorganisms and for its ability to enable short collection time[3]. Prior to sampling, environmental conditions of each laboratory and office were recorded as follows:

2.1.1. Laboratory A

The laboratory has no ultraviolet (UV) light. The number of people as at the time of sampling was four. There was limited movement of people in the laboratory. The size of the laboratory is about 4.7 m×7 m. Air current was low and there was no air conditioning system in the laboratory. Windows and doors were open. The time of sampling was 11:00 am to 11:15 am.

2.1.2. Laboratory B

The laboratory has no UV light. The population of people as at the time of the sampling was fourteen, all in their laboratory coat. There was constant movement of people in the laboratory. The size of the laboratory is about 4 m×6 m. There was no air-conditioning system in the laboratory. The windows and doors were opened for normal air flow. The benches (concrete slabs) were scrubbed but the floor was not scrubbed the morning before the air sample was taking. The time of sampling was between 2:00 pm and 2:15 pm.

2.1.3. Laboratory C

There was no UV light. Five people were in the laboratory, people were coming in and going out during the air sampling was on. There was no air conditioning system

in the laboratory, doors and windows were open for cross ventilation. General routine bacteriology tests were being carried out. The size of the laboratory is about 2 m×3.5 m (compacted laboratory). The time of the air sampling was 10:00 am to 10:15 am.

2.1.4. Laboratory D

There was no UV light except in the inoculating hood. There was no air conditioning system and the movement of people was restricted. The work being done as at the time of sampling was pouring of plate in the inoculating hood. The windows were closed except the door. The time of the air sampling was 11:30 am to 11:45 am.

2.1.5. Laboratory E

There was no UV light in the laboratory. Only two people were in the laboratory and movements were restricted. The air condition in the laboratory was put off 2 days before the day of the sampling (not deliberate) and was put on 1 hour before the air sampling was done. The doors and windows were always closed. The work being done as at the time of sampling was candling and egg inoculation. The size of the laboratory is about 3 m×4 m (a compacted laboratory). The benches and floors were not scrubbed. The time of sampling was 2:45 pm to 3:00 pm.

2.1.6. Air conditioned office

The floor was rugged. The air-conditioning system was usually on everyday and off after each day's work. The number of people in the office as at the time of the air sampling was two and there was no free movement and talking. The windows and door were continuously locked and there was no external air flow except from the air conditioning system. The size of the office was 3 m×4 m. The time of air sampling was 8:15 am to 8:30 am. The size of the laboratory is about 4 m×5 m.

2.1.7. Office without air-condition system

The office has no ultra violet light, and the floor is rugged. The population of people in the office was 15 with free movement and conversation. The size of the room was about 4 m × 7m.

2.2. Sampling procedure

Briefly, two sterile blood agar plates were exposed on benches (4 ft height from the floor) and cupboard (7 ft above the floor to aid the settlement of droplet nuclei), at a reasonable distance apart in each laboratory and offices respectively for 15 minutes. Commercially made sterile (Sterlin) swab stick was used to swab the floors and benches in various laboratories and offices. Prior to swabbing, the swab cotton buds were tested for sterility. The swabs were aseptically inoculated into sterile nutrient broth and in reinforced clostridial medium (RCM) in appropriately labelled MacCartney bottles, to enhance aerobic and anaerobic growth respectively.

2.3. Incubation

All the culture plates and broth cultures were incubated at 37 °C, one aerobically and one anaerobically (gas pak system was used for anaerobic incubation). All anaerobic plates and broth were incubated for 4 days while those for aerobic incubation were incubated for 2 days. Bacteria growing anaerobically were subcultured and incubated aerobically. Those which grew anaerobically and aerobically were grouped as facultative anaerobes and all others as strict anaerobes so that the term strict anaerobe here therefore also included microaerophilic bacteria. The nutrient broth and RCM broth cultures were subcultured onto blood agar plates. All incubations were done as previously stated.

2.4. Identification of isolates

The methods employed for the characterization of all isolates were conventional[3]. The identity of the isolates were confirmed culturally (incubation and plate reading), morphologically (wet preparations and Gram staining), antibiogram and by their various specific biochemical characteristics.

3. Results

A total number of 247 colonies were counted from all

Table 1

Percentage distribution of bacterial genus after air sampling in various laboratories.

Bacterial species	Number of isolated	% of Total isolates
<i>Staphylococcus</i>	51	20.7
<i>Micrococcus</i>	34	13.8
Diphtheroids	30	12.2
<i>Bacillus</i>	30	12.2
<i>Corynebacterium</i>	21	8.5
<i>Propionibacterium</i>	11	4.5
<i>Yersinia</i>	9	3.6
<i>Enterobacter</i>	9	3.6
<i>Pseudomonas</i>	6	2.4
<i>Streptococcus</i>	6	2.4
<i>Acinetobacter</i>	6	2.4
<i>Escherichia</i>	5	2.0
<i>Citrobacter</i>	5	2.0
<i>Branhamella</i>	4	1.6
<i>Klebsiella</i>	4	1.6
<i>Clostridium</i>	5	2.0
<i>Alcaligenes</i>	3	1.2
<i>Listeria</i>	3	1.2
<i>Norcardia</i>	3	1.2
<i>Lactobacillus</i>	2	0.8
Total	247	100%

the plates exposed in five different laboratories, while 74 colonies were counted from the plates exposed in offices.

Table 2

General distribution of bacterial (species) isolates from the air in various laboratories.

Bacterial Species	Number Isolated	Laboratory A	Laboratory B	Laboratory C	Laboratory D	Laboratory E
<i>Micrococcus spp.</i>	34	7	8	9	4	6
<i>Diphtheroids</i>	30	6	6	8	6	4
<i>S. aureus</i>	27	6	4	7	4	6
<i>Bacillus subtilis</i>	19	4	4	2	6	3
<i>S. epidermidis</i>	16	2	4	3	4	3
<i>Propionibacterium</i>	11	2	3	3	3	—
<i>C. diphtheriae</i>	14	3	4	3	2	2
<i>Bacillus cereus</i>	10	2	3	2	2	1
<i>Y. enterocolitica</i>	9	1	2	2	2	2
<i>E. aerogenes</i>	9	3	—	2	3	1
<i>S. saprophyticus</i>	8	1	2	2	1	2
<i>C. pyogenes</i>	7	2	2	—	2	1
<i>Acinetobacter</i>	6	1	—	2	—	3
<i>E. coli</i>	5	—	1	—	2	2
<i>C. freundii</i>	5	1	1	—	—	3
<i>S. pneumoniae</i>	4	—	1	1	1	1
<i>B. cartahalis</i>	4	—	2	—	—	2
<i>P. putrefaciens</i>	3	—	2	1	—	—
<i>P. specie</i>	3	1	—	—	2	—
<i>L. monocytogenes</i>	3	2	—	1	—	—
<i>Norcardia specie</i>	3	—	1	—	2	—
<i>S. pyogenes</i>	2	—	—	1	1	—
<i>K. oxytoca</i>	2	1	—	—	1	—
<i>K. pneumoniae</i>	2	—	1	1	—	—
<i>Alcaligenes faecalis</i>	3	—	1	1	1	—
<i>C. septicum</i>	2	1	—	1	—	—
<i>C. perfringens</i>	2	—	1	1	—	1
<i>Lactobacillus spp</i>	2	—	—	—	1	1
<i>C. soderlli</i>	1	—	—	1	—	—
<i>B. megaterium</i>	1	—	—	1	—	—

These show the number of settled particles that contained bacteria capable of growing on blood agar media that is the colony forming unit (CFU) for 75 minutes (247 CFU for 75 minutes and 74 CFU for 30 minutes in the laboratories and offices respectively).

Out of the 247 colonies, counted from 5 laboratories, 46 CFU (18.62%) were from laboratory A, 55 (22.27%) CFU were from laboratory C. Laboratory D gave a fraction of 50 (20.24%) colonies, while laboratory B gave 52 (21.05%) CFU while laboratory E gave 44(17.81%) CFU.

More so, 20 different bacterial genera were isolated. Among these genera are *Staphylococcus* 51 (20.65%), *Micrococcus* gave 34 (13.77%) while genus *Bacillus* gave 30 (12.15%), *Corynebacterium* 21 (8.6%), *Propionibacterium* 11 (4.5%) CFU, while *Enterobacter* and *Yersinia* have 9 (3.64%) isolates each. *Streptococcus*, *Pseudomonas* and *Acinetobacter* genera are 6 (2.43%) each. The genera *Clostridium*, *Citrobacter* and *Escherichia* were 5 (2.02%) each. Other genera include *Klebsiella*, *Branhamella* were 4 (1.62%) each. *Nocardia* and *Listeria* gave 3 (1.21%) while *Lactobacillus* was 2(0.81%) out of the total isolate (Table 1).

Out of the 247 colonies, a total of 30 different bacteria species (irrespective of the number of occurrences) were isolated. The predominant organism (bacteria species) is *Micrococcus* species, with a total number of 34 (13.77%) CFU in all laboratories. This is closely followed by Diphtheroids with a number of 30 (12.15%) CFU. *Staphylococcus aureus* gave 27 (10.93%) CFU. Other dominant bacteria are *Bacillus subtilis* 19 (7.69%), *Staphylococcus epidermidis* 16 (6.48%) while *Corynebacterium diphtheria* gave 14 (5.67%) CFU, followed by *Bacillus cereus* with a total number of 10 (4.05%) of the total isolates, and *Propionibacterium acnes* with a number of 11 (4.45%) of the total isolates. Other organisms accounted for the remaining (35.81%) (Table 2).

In the two offices sampled, a total of 74 colonies were formed. The office without air conditioning system has a total of 40 colonies (54.05%), while the office with air conditioning system gave a number of 34 (45.95%) CFU. Among the 11 different bacteria genera isolated, *Staphylococcus* and *Corynebacterium* made the highest occurrence with 16 (21.62%) isolates each, followed by *Micrococcus* with 11 (14.86%) isolates. Others are *Propionibacterium* 9 (12.16%) isolates, *Nocardia* 6 (8.11%) isolates. *Alkalegens* and *Acinetobacter* made 5 (6.76%) and 4 (5.41%) isolates respectively. *Streptococcus*, *Bacillus* and *Branhamella* made 2 (2.71%) isolates each. While *Escherichia* made 1 (1.35%) isolate.

4. Discussion

All the areas sampled, contain significantly high number of colony forming units than 3 CFU found in standard

clean room ($P < 0.001$). Bacterium are ubiquitous, they can easily mix with air particles and remain suspended on air. Breathing of air is of obligate importance to human existence, and as such, the cleanliness of the air we breathe is of paramount importance. The importance of this surveillance work should not be undermined, especially in this era when debilitating and immunologically challenging diseases ensue. The ominous pronouncement by Galen in the second century AD, “when many sicken and die at once, we must look to a single common cause, the air we breathe” is still like the word of yesterday. This is because the importance of air to life will surely out live the existence of the universe.

Isolation of 247 bacteria in 5 different laboratories, (giving an average of 49.4 CFU in each of the laboratories) is an indication that there is strong environmental treat to life in this locality. Various studies have suggested that air in the laboratories and operating rooms should not be more than 1–3 CFU/m³, using settle plate (sedimentation) method and within the same time frame as in this study[4]. Comparing this with the work of Goldner *et al*[5] who isolated 12 166 bacteria during 5 561 minutes of the air sampling, we would have equivalence of 247 bacteria isolated within 113 minutes. But in this study, we isolated 247 bacteria in 75 minutes, and with many known pathogenic organisms and potential opportunistic infectious agents on focus. The high level of bacterial suspension as detected in this study is a source of reference to the review of immunological consequences to health, and reawakening interest on the area of environmental pollution. This is in line with the work of Ekhaise *et al*[6]. Average particle in hospital, laboratory or offices, (13 μ m), settles at a rate of about one foot per minutes. Therefore using agar surface in a 100 mm petri dish, will collect particles from 1 ft³ of stagnant air in 15 minutes by sedimentation due to normal gravity. However, air flow in the vicinity may influence the delivery of particles to the agar surface[7].

Environmental problem is one major challenge facing human and animal health in tropical countries. Some factors ranging from nature such as dry weather, state of development, culture such as nomadism, personal hygiene and poor educational background may have contributed to suspension of many bacteria to the air. In this study, *Micrococcus*, *Staphylococcus*, *Corynebacterium*, Diphtheroids and *Bacillus* species are of high prevalence. This is in line with the work of Goldner *et al* and Apurva *et al*, who equally found the above 5 bacteria genus as prevalent organisms when they compared the bacterial load of UV room and non UV room and skin inhabitant bacteria as the prevailing micro-organisms in an environment respectively[5,7]. None of the laboratories has UV light, only one has air conditioning system, which had been off for two days before the day of sampling. This lack of control measures must have contributed to the large number of isolates[8]. Katara *et al*, confirmed the effectiveness of UV light in disinfecting the

air. Instead of focusing on the high prevalence ones only, we decided to show the whole isolate to serve as array of microorganisms that can be found on air in this part of the world. Wide spread of bacteria and high aero-bacterial load as detected in this study is a serious threat to human health. The findings in this study is a clear evidence of exposure to many bacteria and possibly other microbial species. This high level of bacterial inhalation would possibly increase the rate of immune complex formation. Immune complex is the binding of the antigen with its corresponding antibody by electrostatic van der Waals force, to form an interlock. Usually these complexes are eliminated by the mononuclear phagocytes system (MPS) without development of pathological changes^[9]. The formation of immune complexes (IC), due to the interaction of foreign substances (antigens) with specific antibodies, is a physiological process which constitutes an essential part of man's normal immune defence mechanisms. Soluble and insoluble immune complexes activate neutrophils by separate receptor signalling pathways. Profound changes in neutrophil responsiveness to these complexes occur after cytokine priming. It has been established that under appropriate conditions these neutrophils can actively release large quantities of reactive oxidants species (ROS) and discharge the contents of their granules extracellularly. If such large scale release of these toxic molecules occurred *in vivo*, then it is likely that local antioxidants and antiproteinases would become saturated and tissue damage would ensue^[11]. Elevated levels of ROS and down regulation of ROS scavengers and antioxidant enzymes are associated with various human diseases including various cancers^[10]. Oxygen derived species such as superoxide radical, hydrogen peroxide, singlet oxygen and hydroxyl radical are well known to be cytotoxic and have been implicated in the etiology of a wide array of human diseases, including cancer^[11].

Based on these developments, our fear remains that due to constant exposure to many infectious agents or foreign pathogens, with continuous infection and re-infection, IC accumulation may reach a plateau, and may tend to deposit on organs, constituting a great risk factor to HIV pathogenesis and to normal healthy individuals. IC induce a number of cellular functions, including the enhancement of cytokine production from monocytes, macrophages and plasmacytoid dendritic cells^[12]. Evidence has shown that production of proinflammatory cytokines such as TNF- α , interferon- γ , IL-12, IL-1, and IL-6, and nitric oxide, is a usual occurrence during infections and that the production are critical for controlling parasite growth^[13]. However, it has also been reported that excessive production of proinflammatory cytokines could lead to severe pathological conditions^[14].

Continued presence of CIC over extended periods, is a cause of consequence of some pathological condition or infection. It has been suggested that soluble immune complex diseases arising after infections may result from

the liberation of partially synthesized bacterial polypeptide or viral nucleic acid antigens. These disrupted antigens will have heterogeneous molecular weights due to antigenic material which is incomplete as a result of premature termination of synthesis. Also environmental factors play a crucial role in amount of serum levels of these substances. This makes it necessary for each region to find out its own range of normal values in apparently normal individual^[15].

Supportively, Mart^[16] reported that concentration of immune complex at any giving time in circulation depends on the rate of immune complex formation and rate of removal. The rate of immune complex formation in turn depends on the rate of antibody synthesis and rate of availability of specific antigen. The rate of immune complex removal in turn depends on the rate of removal by mononuclear phagocyte system (MPS), and on the deposition of immune complex on tissues.

In cases with inefficient clearance by the mononuclear phagocytes system (MPS) only, pathological consequences will be expected, in particular by immune complexes formed with moderate excess of antigen Tanya *et al.*, 2009^[17] have distinguished between true immune complexes and non-specifically aggregated immunoglobulin. Circulating immune complexes become fixed to basement membranes of the body, and can produce an acute inflammatory reaction by activation of complement and inflammatory cells and also interfere with the immune response.

It is important to understand that high incidence of air bacteria load should not be seen as a peculiar problem to Africa and other tropical areas. This is because the global link is more developed and wider now than before. The rate of international travels has increased. In fact it is difficult to find any country today in isolation. Based on this, doors are wide open for spread and transfer of these organisms from one country to another, from carriers to susceptible hosts and from immune consolidated individuals to immune compromised individuals especially in this era of human immunodeficiency virus, acquired immunodeficiency syndrome (HIV/AIDS) and tuberculosis. Suffice it to say that these aero-bacteria which are easily and continuously inhaled due to the need for oxygen in the air for life would definitely be a medically important source for emerging and re-emerging diseases^[18].

Little or no interest has been shown on the after effects of regular immune responses in subjects residing in those parts of the world where many infectious agents are endemic, considering that some characteristic of immune responses such as inflammation, formation and deposition of immune complexes, infiltration of phagocytic cells, allergic reactions, release of reactive oxidants, have been incriminated in patho-physiology of tissues and organs of the body^[14].

One begins to wonder about the actual healthy state of an average healthy subject in this locality, where there is high tendency of immune complex accumulation and its possible deposition on tissues, high tendency of

continuous inflammatory responses due to constant release of inflammatory cytokines, complement fixation, and constant infiltration of neutrophils and induction of neutrophil to produce reactive oxidants. All these are as a result of endemicity of infectious agents, hence constant exposure of subjects to these infectious agents^[16].

Research has shown that Immune complexes could not be detected in the Caucasian sera but were present in the Nigerian sera^[19].

We can possibly control and limit the proliferation of these microbial agents using the knowledge we have about them, and with some technical know how, we can safely guard the atmosphere of our working environment and prevent the wide spread of these microorganisms. The use of air-conditioning system and chemical disinfection have equally proved satisfactory in reducing microorganisms in the air^[20]. Other factors that could have led to the isolation of this large number of bacteria include the fact that most of these prevalent bacteria inhabit the human skin including the *Propionibacterium*^[21]. Raymond *et al* stated that the main source of air-borne aerobic bacteria is the human skin. He equally stated that a large preponderance of skin bacteria are anaerobic, proving that anaerobic non-spore forming bacteria can be found in air in large amount. Also, the majority of the anaerobic bacteria found are *Propionibacterium* species. Amongst the anaerobic non-spore forming bacteria isolated in this work, *Propionibacterium* species is the largest with 11 isolates. These skin inhabiting bacteria must have been shed off from the skin, hair and clothes worn by the laboratory occupants, as well as from the upper respiratory tracts of individuals entering and leaving the laboratory on daily bases.

In this work, the presence of the isolates, whether pathogen or non-pathogen, is considered important. While the pathogens may pose great danger to the workers, non pathogens could become opportunistic pathogens especially in debilitating subjects and subjects with HIV/AIDS, thereby encouraging emergence and re-emergence of bacteria infection. They may also continuously trigger immune response leading to chronic inflammatory activities^[22]. These bacteria may constitute laboratory nuisance causing contamination of laboratory materials, this situation would continue to influence laboratory results, cause bio-deterioration of food products, biodegradation of pharmaceuticals, industrial and laboratory research products such as drugs, vaccines and even paints^[2]. It is highly recommended in this context that protective measures against air-borne microorganisms should be given serious attention.

Conflict of interest statement

We declare that we have no conflict of interest.

References

- [1] Srikanth P, Sudharsanam S, Steinberg R. Bio-aerosols in indoor environment: composition, health effects and analysis. *Indian J Med Microbiol* 2008; **26**(4): 302-312.
- [2] Ujwala Deepake. Aero-microbiological studies of moisture affected buildings in the Indoor environment Swami Ramanand Teerth Marathwada University. *JYT* 2008; **19**(6).
- [3] Narasimhulu K, Rao PS, Vinod AV. Isolation and identification of bacterial strains and study of their resistance to heavy metals and antibiotics. *J Microbial Biochem Technol* 2010; **2**(3): 74-76.
- [4] Galson E, Goddard KR. Hospital air conditioning and sepsis control. *ASHRAE* 1968; **10**: 33-41.
- [5] Goldner JL, Mogglo M, Beissinger SF, Donald EM. Ultra violet light for the control of air-borne bacteria in operating room. *Ann NY Acad Sci* 1980; 270- 271.
- [6] Ekhaise FO, Ighosewe OU, Ajakpori OD. Hospital indoor airborne microflora in private and government owned hospitals in Benin City, Nigeria. *World J Med Sci* 2008; **3**(1): 19-23.
- [7] Apurva KP, Karuna SV. Extramural aero-bacteriological quality of hospital environment *Asian J Exp Biol Sci* 2010; **1**(1): 128-135.
- [8] Katara G, Hemvani N, Chitnis S, Chitnis V, Chitnis DS. Surface disinfection by exposure to germicidal UV light. *Indian J Med Microbiol* 2008; **26**(3) : 241-242.
- [9] Shmagel KV and chereshnev VA. Molecular bases of immune complex pathology. *Biochemistry (Moscow)* 2009; **74** (5): 469-479.
- [10] Angelopoulou R, Lavranos G, Manolakou P. ROS in the aging male: Model diseases with ROS-related pathophysiology Reproductive. *Toxicol* 2009; **28**(2):167-171.
- [11] Waris G, Haseeb A. Reactive oxygen species: role in the development of cancer and various chronic conditions. *J Carcinog* 2006; **5**: 14.
- [12] Rönnelid J, Ahlin E, Nilsson B, Nilsson-Ekdahl K, Mathsson L. Immune complex-mediated cytokine production is regulated by classical complement activation both *in-vivo* and *in-vitro*. *Adv Exp Med Biol* 2008; **632**: 187-201.
- [13] Kaufmann SHE, Sher A, Ahmed R. *Immununology of Infectious Diseases*. America: ASM Press; 2002, p. 1-495.
- [14] Ferrero-Miliani L, Nielsen OH, Andersen PS, Girardin SE. Chronic inflammation impotence of NOD2 and NALP3 in interleukin-1 beta generation 2007. *Clin Exp Immunol*. **147**(2): 227-235.
- [15] Tanyigna KB, Onah JA, Bello CSS. Serum immunoglobulin and CICs in Nigeria on Jos Plateau. *Ghana Med J* 2004; **38**(1): 28-31.
- [16] Mannik M. Pathophysiology of immune complexes. *Arthritis and Rheumatism* 2005; **25**(7): 783.
- [17] Tanya N Mayadas, George C Tsokos, Naotake Tsuboi. Mediated mechanisms of immune complex-mediated neutrophil recruitment and tissue injury. *Circulation* 2009; **120**: 2012-2024.
- [18] Gingerich BS, Ondeck DA. The infectious disease process: An overview. *Home Health Care Manag Pract* 1993; **6**(1): 8-11.
- [19] Onyewotu II. Immune complexes in healthy Nigerians. *Trans R Soc Trop Med Hyg* 1978; **22** (4): 386-388.
- [20] Perdelli F, Marina S, Anna MS, Maurizio D, Roberto L, Maria LC. A problem of hospital hygiene: The presence of aspergilli in hospital wards with different air-conditioning features. *Am J Infect Control* 2006; **34**: 264-268.
- [21] Raymond PC, Mervyn LC. Some aspects of the airborne transmission of infection. *J R Soc Interface* 2009; **6**: S697-S702.
- [22] Javier RC, Jesús MP, Olivia H Pedro L, María AM, Ignacio L. Toll-Like Receptor 4 Is Involved in Brain Damage and Inflammation After Experimental Stroke. *Circulation* 2007; **115**: 1599-1608.