African Journal of Biotechnology Vol. 7 (20), pp. 3535-3539, 20 October, 2008
Available online at http://www.academicjournals.org/AJB
ISSN 1684–5315 © 2008 Academic Journals

Full Length Research Paper

Distribution of potential nosocomial pathogens in a hospital environment

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Accepted 15 September, 2008

The distribution of probable nosocomial pathogens in a government hospital in Nigeria was investigated. Thirty swab and air samples were collected from patients, hospital personnel, formites and air in four wards namely orthopaedic (OW), paediatric (PW), surgical (SW) and medical (MW). For the patients and personnel, skin and nasal samples were taken. A total of 56 Gram positive (45) and Gram negative (11) bacteria were isolated. Gram positive cocci were the highest number of isolates of which *Staphylococcus epidermidis* (22; 39.2%) occurred the most especially from the skin in all the wards. This was followed by *Staphylococcus aureus* (16; 28.5%) and the least being *Streptococcus* spp. (5; 8.9%). Among the Gram negative bacilli, *Escherichia coli* was the highest (4; 7.1%). Others were *Klebsiella pneumonia* (3; 5.3%), *Proteus* spp. (2; 3.5%) and *Enterobacter aerogenes* (2; 3.5%). The only Gram positive bacilli isolated were *Bacillus cereus*. Orthopaedic ward (22) had the highest number of isolates followed by paediatric ward (15). Surgical and medical wards had 10 and 9 isolates, respectively. Statistical analysis of the questionnaire distributed to the patients and hospital personnel in the four wards revealed that duration of admission and length of service were significant determining factors for the carriage rate of the isolates in the individuals examined. The findings of the study showed that the hospital may be a potential reservoir of organisms likely to cause nosocomial infections.

Key words: Distribution, nosocomial pathogens, government hospital, Nigeria, gram positive bacteria, gram negative bacteria.

INTRODUCTION

Nosocomial infections (also known as hospital associated/acquired infections) are those infections that develop in a patient during his/her stay in a hospital or other type of clinical facilities which were not present at the time of admission. Hence, pathogens that cause such infections are termed nosocomial pathogens (Prescott et al., 2005). The hospital environment is a potential reservoir of infectious agents since it houses both patients with diverse pathogenic microorganisms and a large number of susceptible/immunocompromised individuals (Rhomberg et al., 2006; Zhanel et al., 2008). The nosocomial pathogens that cause infections can come either from endogenous or exogenous sources.

Endogenous sources are those that are from the patient's own microbial flora while the latter are from the surrounding hospital environment. A patient may be infected by his/her body flora following surgical manipulation, chemotherapy and diagnostic or therapeutic procedures which in most cases suppress the natural body defensive mechanisms (Pelczar et al., 1993). Animate and inanimate sources of exogenous infections include hospital staff, other patients, visitors, food, water, formites, urinary catheter, intravenous devices, respiratory equipment and other prostheses (Prescott et al., 2005).

The most important means of transmission of nosocomial infections is by contact, usually directly but sometimes indirectly by means of secretions from the body (Bergogne-Berezin and Towner, 1996). Air can also be a route of transmission of air borne-nosocomial patho-

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gens (e.g. in droplet nuclei and aerosols) that infect the respiratory tract. The faecal-oral route is a portal of entry for food-borne and water-borne infections (Pelczar et al., 1993).

Predisposing factors that make patients susceptible to these infections include concurrent infections, prosthetic devices, surgery, immunosuppressive agents, administration of broad-spectrum antibiotics, and emergence of multidrug resistant pathogens (Pelczar et al., 1993; Courvaline and Weber, 2005). Other risk factors include age of patient, duration of hospitalization, underlying diseases like diabetes, tumours or overcrowding in the hospital wards (Prescott et al., 2005)

The bacteria that commonly cause nosocomial infections include *Staphylococcus aureus*, *Streptococcus* spp., *Bacillus cereus*, *Acinetobacter* spp., coagulase negative staphylococci, enterococci, *Pseudomonas aeruginosa, Legionella* and members of the *Enterobacteriaceae* family such as *Escherichia coli*, *Proteus mirabilis*, *Salmonella* spp., *Serratia marcescens* and *Klebsiella pneumonia* (Esposito and Leone, 2007; Zhanel et al., 2008).

The most frequently reported nosocomial pathogens have been *E. coli*, *S. aureus*, enterococci and *P. aeruginosa.* Pathogenic strains of *E. coli* can cause different forms of gastrointestinal tract infections. *P. aeruginosa* is a regular cause of nosocomial pneumonia, urinary tract infections, surgical site infections and infection of severe burns. *S. aureus* is commonly associated with skin and soft tissue infections, surgical, lower respiratory tract infections and neonatal infections (McCraig et al., 2006; Pitout et al., 2005 and 2007).

The occurrence of multi-drug resistance in hospital-associated pathogens has resulted in the emergence and reemergence of difficult-to-treat nosocomial infections in patients. Examples of bacteria possessing such drug resistance are methicillin-resistant *S. aureus*, penicillin-resistant pneumococci, vancomycin-resistant enterococci, vanco-mycin resistant *S. aureus* and multi-drug resistant tuberculosis (Moran et al., 2005; Prescott et al., 2005; McCraig et al., 2006; Scheider-Linder et al., 2007).

In the developing countries, the incidence of nosocomial infections can be devastating resulting in major disease outbreaks in hospitals and other healthcare facilities. This may be attributed to poor infrastructure, over-crowding, inadequate personnel and management in most hospitals. This study was undertaken to investigate the occurrence and distribution of potential nosocomial pathogens in a government hospital in Port Harcourt, Nigeria.

MATERIALS AND METHODS

Samlpe sources

Samples used for the study were obtained from doctors, nurses, orderlies, patients, air, and formites like beds, cannula, oral thermometer, and tables. They were collected from paediatric ward (PW), medical ward (MW), orthopaedic ward (OW) and surgical ward (SW). Sterile swab sticks were aseptically used to collect nasal and skin swabs from patients, personnel and formites. A total of 30 swab samples were collected. For air samples, nutrient agar plates were exposed to the air in the wards for 10 min. Questionnaires were distributed to patients and hospital personnel in order to determine the relationship between duration of admission (for patients)/length of service (for the workers) and extent of colonization by isolated bacteria in individuals. Some of the items in the questionnaire were gender, ward, age, duration of stay (admission or service), number of patients and personnel in the ward and reason for admission. The samples were taken to the laboratory and analyzed immediately.

Isolation

The swab samples were streaked on MacConkey agar, eosin methylene blue (EMB) agar and mannitol salt agar. The plates were incubated together with the nutrient agar plates used to sample the air at 37^{o}C for 24 h. Discrete colonies were further subcultured onto nutrient agar to obtain pure cultures. The purified cultures were stored on nutrient agar slants for biochemical tests and identification.

Colonial morphology of colonies

Presumptive identification of the colonies was done by observing their individual appearance on the selective and or differential media used for isolation. Colonies with characteristic metallic sheen on EMB agar and lactose fermenters on MacConkey agar were noted. Purified colonies were further characterized using Gram stain and motility test.

Biochemical tests

The following biochemical tests were used to identify the isolates: catalase, coagulase, indole production, citrate utilization, triple iron sugar utilization and methyl red-Voges Proskauer.

Statistical analysis

Student t-Test was used to analyze the questionnaires distributed to the patients and hospital personnel.

RESULTS AND DISCUSSION

It was observed that Gram positive bacteria had higher occurrence in the 30 samples analysed than Gram negative bacteria. Out of 56 isolates identified, 45 were Gram positive, the remaining 11 Gram negative. *S. epidermidis* had a frequency of 39.2% of the total number of isolates followed by *S. aureus* with 28.5%. *Streptococcus* spp., *E. coli*, and *K. pneumoniae* had frequency of 8.9, 7.1 and 5.3%, respectively. *B. cereus*, *E. aerogenes* and *Proteus* spp. each had a frequency of 3.5%. The result presented in Table 1 shows the common organisms isolated from all the wards.

Table 1. Frequency of isolation of bacteria from the four wards.

Isolate	Occurrence	(%)
S. epidermidis	22	39.2
S. aureus	16	28.5
Streptococcus spp.	5	8.9
Escherichia coli	4	7.1
K. pneumonia	3	5.3
Proteus spp.	2	3.5
Enterobacter aerogenes	2	3.5
Bacillus cereus	2	3.5
Total	**56**	**100**

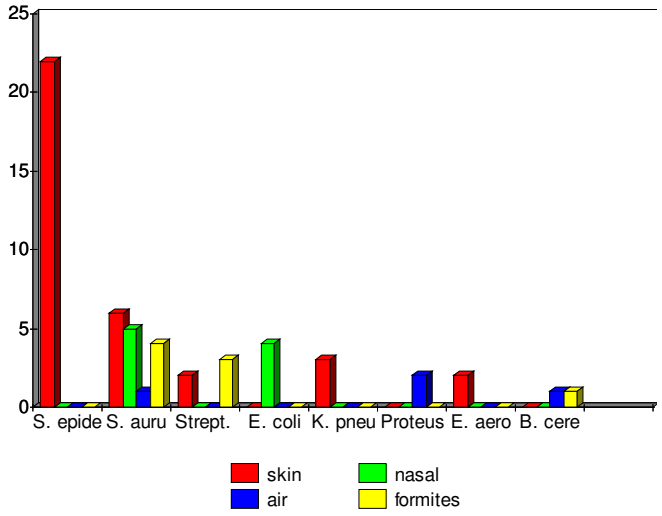

Figure 1. Distribution of bacterial isolates from the different sources of samples. S. epide = *Staphylococcus epidermidis* ; Proteus = *Proteus* spp.; S. auru = *Staphylococcus aureus* ; E. aero = *Enterobacter aerogenes*; Strept. = *Streptococcus* spp.; B. cere = *Bacillus cereus*; E. coli = *Escherichia coli*; K. pneu = *Klebsiella pneumonia.*

The ward that had the highest number of bacterial isolates was orthopaedic ward. This ward accounted for 22 out of the 56 isolates from all the wards. Paediatric ward followed closely with 15 while surgical and medical ward had 10 and 9, respectively. The most commonly isolated bacterium from the wards was *S. epidermidis* followed by *S. aureus* whereas *Proteus* spp., *E. aerogenes* and *B. cereus* were among the least isolated. The distribution pattern of the isolates for the wards is shown in Table 2.

The frequency of isolation of the different isolates from the four sample sources which were skin, nasal, air and formites is presented in Figure 1. The skin accounted for the highest number of isolates obtained. Gram positive cocci were the most isolated from the skin while Gram negative bacilli were the least. For the other sample sources, few organisms were isolated from them. The Gram positive cocci in all the samples were the dominant and most isolated organisms.

This survey was carried out to gain insight into the distribution and carriage rate of bacterial flora that could be of potential health risk in a hospital or any other healthcare facility. It was observed that more than half of all the isolates recovered were from the skin (Figure 1). The remaining came from the nose, air and formites. The bacteria that were isolated from the different sources were *S. epidermidis* (39.2%), *S. aureus* (28%), *Streptococcus* spp. (8.9%), *E. coli* (7.1%), *K. pneumonia* (5.3%), *Proteus* spp. (3.5%), *E. aerogenes* (3.5%) and *B. cereus* (3.5%). Statistical analysis using t-Test on nasal and skin isolates from the patients and hospital personnel showed that duration of admission and length of service are significant factors that determine the carriage rate of these bacteria.

S. epidermidis was the most frequently isolated from all the samples collected from the four wards. This was followed closely by *S. aureus* and *Streptococcus* spp. The reason for this may be because of the fact that staphylococci and streptococci are members of the body flora of both asymptomatic carriers and sick persons. These organisms can be spread by the hands, expelled from the respiratory tract or transmitted by animate or inanimate objects (Pelczar et al., 1993). Though strains of *S. epidermidis* are known to be non-pathogenic on the skin and nose areas, but when they harbour antimicrobial resistance they can constitute serious health hazard. Recently, methicillin-resistant *S. epidermidis* strains and other coagulase negative staphylococci have emerged as common nosocomial pathogens affecting immunocompromised patients carrying medical devices (Kainer et al., 2007). This observation that Gram positive cocci were the most isolated bacteria is consistent with the work of Fridkin et al. (2001). In the same vein, Zhanel et al. (2008) in their survey of antimicrobial-resistant pathogens in intensive care units in Canada reported that Gram positive cocci were among the most common isolates recovered from 80% of all clinical specimen collected. Strains of *S. aureus* are becoming a nightmare to the medical field as a result of their possession of antibiotic resistant genes. There are numerous reported cases of emerging nosocomial infections caused by methicillin-resistant *S. aureus* (MRSA), vancomycin-resistant *S. aureus* (VRSA) and other multi-drug (MDR) resistant strains (Courvalin and Weber, 2005; Kuehnert et al., 2005; Moran et al., 2005; Zhanel, et al., 2008).

Among the Gram negative bacteria, *E. coli* was the most common when compared to *K. pneumonia*, *Proteus* spp. and *E. aerogenes*. This finding is in consonance with the works of Lockhart et al. (2007) and Zhanel et al. (2008). All the *E. coli* strains were isolated only from the nose, *Proteus* spp. from the air, while *K. pneumonia* and *E. aerogenes* from the skin. Some strains of all the Gram negative bacilli isolated have been shown to harbour anti-

Table 2. The distribution of the isolates in each ward.

Bacterial isolate	Orthopaedic	Paediatric	Surgical	Medical
Staphylococcus epidermidis	4 (40.9%)	5 (22.7%)	4 (18.2%)	4 (18.2&)
Staphylococcus aureus	6 (37.5%)	6 (37.5%)	1 (16.3%)	3 (18.7%)
Streptococcus spp.	-	1 (20%)	3 (60%)	1 (20%)
Escherichia coli	2 (50%)	1 (25%)	1 (25%)	-
Klebsiella pneumoniae	2 (66.7%)	-	-	1 (33.3%)
Proteus spp.	1 (50%)	-	1 (50%)	-
Enterobacter aerogenes	-	2 (100%)	-	-
Bacillus cereus	2 (100%)	-	-	-
TOTAL	**22**	**15**	**10**	**9**

biotic resistance like the extended spectrum β lactamases (ESBLs) and MDR-ESBLs (Mulvey et al., 2004, 2005; Moland et al., 2006; Lewis et al., 2007; Lockhart et al., 2007, Zhanel et al., 2008). The only Gram positive bacilli encountered in this study was *B. cereus* which was isolated from the air and formites. This organism forms endospores, so it is not surprising that the aerosols formed a repository for them. Probably from the air the spores settled on the surfaces of formites in the hospital.

The orthopaedic ward had the highest number of isolates. As much as 22 out of the 56 isolates came from this ward. Paediatric ward followed with 15 while surgical and medical wards had 10 and 9, respectively. Gram positive bacteria were more isolated than the Gram negative bacteria (Table 2). Most patients in orthopaedic wards are confined to a place unlike other ambulatory patients. As such the use of medical devices to aid evacuation of waste products may be higher in orthopaedic wards. This may predispose the patients to colonization by bacteria in the environment (Pelczar et al., 1993; Prescott et al., 2005; Rhomberg et al., 2006).

In conclusion, Gram positive bacteria were more isolated from the wards especially from the patients and hospital personnel than any other sources with the skin being the most colonized by these organisms. While most of the isolates were recovered from the orthopaedic ward, other wards showed varying numbers of Gram positive and Gram negative isolates.

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