Journal of Applied Sciences

ISSN 1812-5654

science alert

ANSInet
an open access publisher
http://ansinet.com

Journal of Applied Sciences 10 (11): 1011-1014, 2010
ISSN 1812-5654

Isolation of Potential Pathogenic Bacteria from the Air of Hospital-Delivery and Nursing Rooms

Sanaa O. Yagoub and Amani El Agbash
Department of Microbiology and Molecular Biology,
Faculty of Science and Technology, El Neelain University, P.O. Box 12702, Khartoum, Sudan

Abstract: This study aimed to investigate the hygienic conditions of air at delivery and nursing rooms at some hospitals located in Khartoum district Sudan. Out of seventy-nine randomly collected air samples from delivery rooms at the four largest hospitals (Khartoum Teaching Hospital, Al Ribat Hospital, Ahmed Fadul Hospital and Omdurman Maternity Hospital), 63.3% showed positive bacterial growth. Out of 60 air samples that were collected from the nursing rooms, 66.7% of them showed positive bacterial isolation. The isolated bacterial species were identified as *S. aureus*, *E. coli*, *Klebsiella* sp., *P. aeruginosa* and *Bacillus* sp. The highest percentage of bacterial isolation was recorded in Khartoum Hospital as 50.6 and 42% in delivery and nursing rooms, respectively. *Staphylococcus aureus* and *P. eruginosa* were the most dominant organisms isolated from the delivery rooms at all examined hospitals while *S. aureus.* showed the highest percentage from nursing rooms at two of the examined hospitals, *E. coli* and *Bacillus* sp. were dominant in Khartoum Hospital. The isolated bacteria showed clear resistance toward the common antiseptics used in hospitals.

Key words: Hospital pollution, nesocomial infection, microbial resistance, disinfectants

INTRODUCTION

Bacterial infections still play considerable role in diseases in Sudan. Air is not a suitable medium for growth of the pathogenic bacteria, any pathogen, that airborne must have originated from a source such as humans, animals, plants, soil, food or water (Ishida *et al*., 2006). More people are dying every year from hospital infections (Johnson, 2002). Although, many pathogens can cause hospital infection but those that are able to survive in the hospital environment for long periods and resist also disinfections are particularly important in this respect. *Streptococcus pyogenes* was perhaps the most important cause of hospital infection formerly but is hardly ever encountered-now as it is highly susceptible to antibiotics. *Staphylococcus aureus* strains, resistant to multiple antibiotics and belonging to phage type 80 colonizing hospitals and causing nosocomial infections (Newman, 2002; Crimi *et al*., 2006; Khojasteh *et al*., 2007).

Staphylococcus epidermidis and group D *Streptococci* also are sometimes responsible for hospital infection. Enteric Gram-negative *bacilli*, *E. coli*, *Klebsiella* sp., *Enterobacter* sp., *Proteus* sp. and *Neisseria* sp., have become the most important group of hospital pathogens (Krishna *et al*., 2007), multi-drug resistant *Salmonellae*, particularly *S. typhimurium,* become a prominent hospital pathogen, *Pseudomonas aeroginosa* and other *Pseudomans* sp., have always been important causes of hospital infection because of their intrinsic resistance to most antibiotics and ability to survive and even multiply at low temperatures and in disinfectant solutions (Newman, 2002; Behrends, 2003), Ohsaki *et al.* (2007) isolated *Bacillus cereus* from nosocomial patients as a causative agent for hospital outbreak.

Newman (2002) reported that nosocomial infection remains an important problem in intensive care units. Also, Ishida *et al.* (2006) reported that air borne bacteria in the environment are thought to be a cause of postoperative infection. Jimenez *et al.* (2004) evaluated the presence of bacteria in food and environment of an oncological service of national hospital, Behrends (2003) mentioned that testing of the piping of new hospital showed that the drinking water was contaminated with *Pseudomonas aeruginosa*.

The effective use of antiseptics, disinfectants and sterilization procedures is important in preventing nosocomial infections, physical agents such as moist or dry heat, play the major role in sterilization, antisepsis depends on a variety of factors, include the degree of

Corresponding Author: Sanaa O. Yagoub, Department of Microbiology and Molecular Biology,
Faculty of Science and Technology, El Neelain University, P.O. Box 12702, Khartoum, Sudan
Fax: 00249-83-776338

microbial killing required and the nature of the item or surface to be treated (Zaidi *et al*., 1995).

The objectives of this research were isolation of microorganisms from the air of delivery and nursing rooms and to study the effect of the common antiseptics used in the hospital to inhibit the growth of these isolated microorganisms.

MATERIALS AND METHODS

This study was carried in Khartoum district Sudan, from 2005-2008. The laboratory examination was done at the Department of Microbiology and Molecular Biology, Faculty of Science and Technology, El Neelain University, Sudan.

Samples collection: This study was done at Khartoum Teaching Hospital, Al Ribat Hospital, Ahmed Fadul Hospital and Omdurman Maternity Hospital. The samples technique for monitoring air borne microbial contamination is sedimentation technique using open Petri-dishes containing different media. This media were exposed at room air for 1-2 h then closing the Petri-dishes. The plates were distributed in many places of the delivery rooms and nursery at different distance from the floor.

Immediately after collection of samples, the Petri-dishes were taken to the laboratory of Microbiology, Faculty of Science and Technology El Neelain University, Khartoum, for further bacteriological analysis.

Preparation of the media, isolation and identification of bacterial species were done according to Cheesbrough (1991).

The susceptibility of the isolated organisms towards antiseptics was tested by using the cup -plate diffusion technique described by Kavanagh (1974), on Mueller-Hinton agar four cups (8 mm) were cut using a sterile cork borer and then inoculated, the four cups were filled with 0.1 mL of each antiseptic concentrations (100, 90, 80, 70, 60 and 50%) by using adjustable volume digital pipette and allowed to diffuse at romm temperature for 2 h, the plates were then incubated in upright position at 37°C for 18 h. Growth inhibition zones were measured.

RESULTS

Number and percentage of the bacteria isolated from Delivery and Nursing rooms: Out of 79 samples that collected from delivery rooms at different hospitals, 52 of them (63.3%) showed positive bacterial growth, The positive percentages of samples were 100, 84.4, 84.4 and 35% from Khartoum Hospital, Omdurman Maternity Hospital, Ahmed Fadual Hospital and El Ribhat Hospital, respectively (Table 1). The Nursing rooms revealed positive bacterial growth of 44 samples out of 66 with percentage of 66.7. Individual percentages are 40, 66.7, 80 and 100 at El Ribhat, Ahmed Fadul, Odurman Maternity and Khartoum Hospitals, respectively (Table 1).

Number and percentage of isolated bacterial species: The isolates were identified according to morphological appearance, cultural characteristic and biochemical reactions as Gram-positive bacteria which further identified as *Staphylococcus aureus* and *Bacillus* sp. The Gram-negative bacteria were identified as *E. coli, Klebsiella* sp. and *Pseudomonas aeruginosa*.

As shown in Table 2, 163 organisms were isolated from the delivery and nursing rooms of all hospitals, these were classified as *Pseudomonas aeruginosa* 47(28.8%), *Staphylococcus aureus* 43 isolates (26.4%) *Klebsiella* sp., 30 (18.4%), *E. coli* 22 (13.5%), *Bacillus* sp., 20 (12.3%), and *Rothia* sp., 1 (0.6%).

The most predominant isolated bacteria from the delivery rooms of all hospitals was *Pseudomonas aeruginosa* this was followed by *Staphylococcus aureus* at Omdurman Materinty hospital and Khartoum hospital

Table 1: Number and percentage of positive bacterial growth from delivery and nursing rooms at Khartoum state Hospitals

	Delivery room			Nursing room		
Hospitals	No. of samples	No. of positive samples	Percentage	No. of collected samples	No. of positive samples	Percentage
Ribat	20	7	35.0	25	10	40.0
Omdurman Maternity	19	16	84.4	20	16	80.0
Khartoum	20	20	100.0	12	12	100.0
Ahmed Fadul	20	9	45.0	9	6	66.7
Total	79	52	63.3	66	44	66.7

Table 2: Number and Percentage of bacteria isolated from khartoum state hospitals

Organism	No.	Percentage
Staphylococcus aureus	43	26.4
Bacillus sp.	20	12.3
E. coli	22	13.5
Klebsiella sp.	30	18.4
Pseudomonas aeruginosa	47	28.8
Rothia sp.	1	0.6
Total	163	

Table 3: Number and percentage of bacteria isolated from Delivery and Nursing rooms at different Khartoum state hospitals

	Omdurman H		Khartoum		A. Fadual		El Rhibat		Total	
Organisms	Deliv.	Nurs.	Deliv.	Nurs.	Deliv.	Nurs.	Deliv.	Nurs.	Deliv.	Nurs.
Staphylococcus aureus	10 (52.6)	10 (47.6)	3 (17.6)	3 (8.6)	3 (17.6)	-	3 (33.33)	4 (40)	19 (30.6)	17 (22.7)
Pseudomonas aeruginosa	6 (31.6)	6 (28.6)	5 (29.4)	10 (28.6)	5 (29.4)	-	3 (33.33)	3 (30)	19 (30.6)	19 (25.3)
Klebsiella sp.	3 (15.8)	4 (19)	4 (23.5)	4 (11.4)	4 (23.5)	3 (33.3)	3 (33.33)	-	14 (22.6)	11 (14.7)
Bacillus sp.	-	-	5 (29.4)	6 (17.1)	5 (29.4)	6 (66.7)	-	3(30)	10 (16.1)	15 (20)
E.coli	-	-	-	12 (34.3)	-	-	-	-	-	12(16)
Rothia sp.	-	1(4.8)	-	-	-	-	-	-	-	1(1.3)
Total	19	21	17	35	17	9	9	10	62	75

Values in brackets indicate percentage

Table 4: Effect of concentrated and diluted septol and formalin on the isolated bacterial species (Zone of inhibition by mm)

	Septol						Formalin					
Organisms	Full	10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}	Full	10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}
Staphylococcus aureus	(-)	(-)	(-)	(-)	(-)	(-)	15	13	7	(-)	(-)	(-)
Klebsiella sp.	2	(-)	(-)	(-)	(-)	(-)	14	5	(-)	(-)	(-)	(-)
E. coli	3	2	1	(-)	(-)	(-)	25	9	1	(-)	(-)	(-)
Pseudomonas aeruginosa	5	4	1	(-)	(-)	(-)	35	17	7	(-)	(-)	(-)

(-): No inhibition zone

or by *Bacillus* sp., as shown at Ahmed Fadul hospital and equal with *Staphylococcus aureus* and *Klebsiella* sp., at El Rhibhat hospital (Table 3).

Table 3 also demonstrates that in the Nursing rooms of these hospitals *Pseudomonas aeruginosa* was isolated as 19 (25.3%), *Staphylococcus aureus* as 17(22.7%), *E. coli* as 12 (16%), *Klebsiella* sp., as 11 (14.7%), *Bacillus* sp., as 15 (20%) and *Rothia* sp., as 1 (1.3%).

The most predominant organism isolated from nursing room at Khartoum hospital was *E. coli* as 12 (34.3%) followed by *Pseudomonas aeruginosa* as 10 (28.6%), at Omdurman Maternity hospital *Staphylococcus aureus* showed dominant isolation 10 (47.6%) followed by *Pseudomonas aeruginosa* as 6 isolates (28.6%) at E Rhibat Hospital isolation of *Staphylococcus aureus* was more dominant as 4 isolates (40%) followed by *Pseudomonas aeruginosa* and *Bacillus* sp., 3 (30%) for each. At Ahmed Fadual hospital the dominant isolate was *Bacillus* sp., as 6 isolates (66.7%) followed by *Klebsiella* sp., as 3 (33.3%) (Table 3).

Effect of different concentrations of septol on isolated bacteria: As shown in Table 4, *Staphylococcus aureus* showed a marked resistance towards septol antiseptic even at full concentration (100%) no inhibition zones were observed at any plates. *Klebsiella* sp., showed inhibition zone of 2 mm at full concentration of the septol otherwise no inhibition zones were detected at dilution of, 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4} and 10^{-5}. *E. coli* showed inhibition zone of 3 mm at full concentration of septol 2 mm and 1mm diameter inhibition zones were detected at 10^{-1} and 10^{-2}, respectively, at 10^{-3}, 10^{-4} and 10^{-5} no inhibition zones were detected. *Pseudomonas aeruginosa* showed inhibition zones of 5 mm at full concentration of the septol, 4 and 1 mm zones of inhibition were shown at dilution of 10^{-1} and 10^{-2} respectively, other dilutions showed absence of any inhibition zones.

Effect of different concentrations of formalin on isolated bacteria: As shown in Table 4, formalin resulted in good inhibition zones with *Staphylococcus aureus, Klebsiella* sp., *E. coli* and *Pseudomonas aeruginosa* at full, 10^{-1} and 10^{-2} except 10^{-2} with *Klebsiella* sp., other concentration showed no activity against all tested bacteria. *Pseudomonas aeruginosa* showed largest inhibition zones at all concentrations.

DISCUSSION

Child birth is one of the life's major events, The way in which it is experienced will have very significant and long term effects on the mother. It is the responsibility of all those involved in the provision of care to achieve a balance between scientific objectivity and the concern for woman wishes.

The presence study showed that the percentage of positive isolation of potential pathogenic bacteria at hospital delivery rooms and nursing rooms was 63.3 and 66.7%, respectively. Khartoum Hospital had the highest percentage, these might be due to the fact that Khartoum Hospital is the largest educational hospital with largest admission data that increase possibility of the presence of pathogenic organisms among patients and co-patients and confirm the findings of Abussaud (1991), who reported that the infection rates were higher in the large teaching hospitals, these also indicated that there is no evidence to support the claim that a hospital is the safest place for women to have normal births.

In this study, the most common isolated bacteria was *Pseudomonas aeruginosa*, the isolation of this organism which is well known as multi antibiotics resistant organism and the role of these bacteria as nosocomial organism which associated with wounds and eye infections is well documented The isolation of

Pseudomonas aerouginosa in burns units via the airborne route was reported by Govan (1992), *Pseudomonas aerouginosa* was also isolated as predominant organisms from indoor air of hospitals by Tambekar *et al.* (2007). *Staphylococcus aureus* was the one of the dominant isolated organisms, these bacteria is a common pathogenic bacterium that associated with various diseases, it is responsible for many respiratory tract, digestive system, post operative infections, urinary tract and skin disorders with multi antibiotics resistance, the presence of this organism might be due to post-sterilization, or the environment contamination. Tambekar *et al.* (2007) found that *Staphylococcus aureus*, *Staphylococcus saprophiticus* were among the predominant organisms that isolated from the air of hospitals.

The isolation of *E. coli* from both delivery and nursing rooms might be due to faecal contamination and indicated the possibility of occurrence of different diseases agents especially in immuno-compromise persons such as new babies and delivered women According to Beggs (2003), *E. coli, Klebsiella pneumonia* were the least air contaminants as the source of contamination may be water droplets and not survive for long period, otherwise in this study the *E. coli, Klebsiella pneumonia* were isolated as 13.5 and 18.4%, respectively. The isolation of *Bacillus* sp. which is spore-forming organism needed proper program for elimination.

All isolated bacteria showed resistance toward septol the disinfectant that commonly used in the most hospitals. The resistant of bacterial hospitals toward disinfectants were reported previously by Rutala *et al.* (1997), Russell (1999) and Nunez and Moretton (2007).

It could concluded that efforts are urgently needed to improve hospital hygienic environment among health care workers and mothers and it is recommended to raise the awareness and educational status of the medical workers to reduce the hazards of air-borne transmission of such potentially pathogenic microorganisms.

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