Bacterial Agents Associated With Health Care Associated Infections In Some Selected Tertiary Hospital Of Kano Metropolis, Northwest Nigeria

Alkali, B.,*[1] Agwu, E[1]., Sarkinfada, F[2]., Idris, A. M[3]. and Takalmawa, H. U[3]

[1]Department of Microbiology and Immunology, Faculty of Biomedical Science, Kampala International Hospital Bushanyi, Uganda

[2]Department of Health and Medical Sciences, Khawarizmi International College Abu Dhabi, United Arab Emirate.

[3]Department of Medical Microbiology and Parasitology, Faculty of Clinical Science, Bayero University Kano, Nigeria

Abstract: Health care acquired infections (HCAIs) are those infections that occur within 48 hours of hospital admission, 3 days of discharge or 30 days of an operation. These infections have strong effect on patients because of worsening underlying medical condition and increased mortality and morbidity. They are mostly caused by microorganisms already present in or on the patient's own body. Such organisms only cause problems when the body's defenses are weakened, or breached by surgery or other medical procedures. Such infections may also be caused by micro-organisms originating from another patient either by direct contact or through a contaminated hospital environment. This study aimed to detect the bacterial agent associated with HCAIs in some selected hospitals in Kano metropolis. A total of 401 non duplicated samples were collected from patients who spent ≥14 days and aged ≥ 18 years that were admitted in the three selected hospitals of Kano metropolis. All the samples were cultured on the appropriate culture media and subjected to standard biochemical tests according to standard bacteriological procedure. Antibiotics susceptibility testing was done using a modified form of the Kirby Bauer method. An overall prevalence of 34.4% were obtained, with higher incidence 28 (7.0%) in age group 39 – 48 years. Gram negative organisms were the most frequent organisms among which *E. coli* and *Proteus* spp have the higher percentage of 26.1% and 16.7% respectively. Most of the isolated organisms were shown to be resistant to Cotrimozaxole and Amoxicillin. From what was obtained in this study, it shows that Gram negative bacilli were frequent organisms associated with nosocomial infection in the study area. Patients with post surgical site infection has the highest percentage of nosocomial infection among which *E. coli* and *Proteus* spp are the predominant species.

Keywords: Health care acquired infections, Microorganism, Bacteria, Antibiotics, Infection.

INTRODUCTION

Healthcare-acquired infections (HCAIs), also known as nosocomial infections, are infections that patients get while receiving treatment for medical or surgical conditions. HCAIs occur in all settings of care, including hospitals, surgical centers, ambulatory clinics, and long-term care facilities such as nursing homes and rehabilitation facilities (Patient, 2018). Health care-acquired infections first appear 48 hours or more after hospitalization or within 30 days after having received health care (Revelas, 2012). These infections have a strong effect on patients because of worsening underlying medical conditions and increased mortality and morbidity.

[1*]Corresponding author: balkali2012@gmail.com; amikibiya@gmail.com +234 8035876964

Patients are more likely to be vulnerable to HCAIs infection due to their illness, their age, or the treatment for their condition. Many infections are caused by micro-organisms already present in or on the patient's own body. Such organisms only cause problems when the body's defenses are weakened, or breached by surgery or other medical procedures (NHS, 2014). Infections may also be caused by micro-organisms originating from another patient either by direct contact or through a contaminated hospital environment (NHS, 2014).

The prevalence of HCAIs is already substantial in developed countries, where it affects from 5% to 15% of hospitalized patients and as high as 50% in ICU (Vincent *et al.*, 2009). In the hospital environment, healthcare-associated pathogen scan colonizes the respiratory, urinary, and gastrointestinal tracts and wounds of the patients and those on mechanically ventilated life support.

It shows a special predilection for the ICU and causes prolonged hospital stay, challenges faced by healthcare stakeholders (Towner *et al.*, 2014). To prevent HCAIs it is necessary to identify sources and modes of transmission of infection and to implement data-driven prevention guidelines and practices.

In Nigeria, there is scarce knowledge of the risks of HCAIs. In Kano State, Nigeria, a report on HCAIs includes the agents of postoperative site infections SSIs (Nwanko *et al.*, 2010).

MATERIAL AND METHODS

Study Area

Three hospitals were selected for the study and these are; Aminu Kano Teaching Hospital (AKTH) located in Tarauni Local Government Area of Kano State, Murtala Muhammad Specialist Hospital (MMSH) and Muhammad Abdullahi Wase Specialist Hospital (MAWSH) in Nasarawa Local Government Area of Kano State. Kano state is located at latitude 10o 33N to 11o 15N and longitude 34oCE to 8o 20CE with an estimated population of over 13 million (NBS, 2018). The Hospitals are strategically located for access to both urban and rural populations from the 44 Local Government Areas of Kano State.

Study Population

The study population comprised both male and female patients who spent ≥14 days in admission in the selected hospitals and age ≥ 18 years.

Ethical approval

Ethical approval was obtained from the Kano State Hospital Management Board and the ethical committee of Aminu Kano Teaching Hospital before the commencement of the study.**Sample collection**

A total of 401 non duplicated samples were collected from the entire three study locations (AKTH = 200, MMSH = 140 and MAWSH =61). The number of samples collected from each location was according to the bed capacity of the center. Samples include wound swab, urine samples, urine catheters, and nasal intubation.

Bacterial Isolation

Wound samples and nasal intubation were cultured on blood agar (Oxoid, UK), chocolate agar (Oxoid, UK) and MacConkey agar (Tm, Media), while urine and urine catheters samples are cultured on cysteine, lactose electrolyte deficient (CLED) agar and MacConkey agar. All the plates were incubated at 37^{o}C for 18 – 24 hours.

Bacterial Identification

All incubated plates were examined for growth after 18 – 24 hour incubation at 37OC. MacConkey agar plates were screened for lactose or non-lactose fermentation, while blood and chocolate agar plates were examined for complete or partial haemolysis.

Biochemical Tests

All the isolated organisms were Gram-stained and the Biochemical test was carried out on the bases of their Gram status according to Cheesbrough (2010).

The biochemical test carried out in this study included lactose fermentation, citrate utilization, urea fermentation, indole test, methyl red, Voges Proskauer, motility test, Kovac's oxidase test and Analytical profile index 20 for Non Enterobacteriaceae (API 20NE) Gram-negative organisms and catalase, coagulase, haemolysis activities on blood agar and sugar fermentation for Gram-positive organisms (Cheesbrough, 2010).

API 20 Multi Test Systems

All the Gram-negative bacilli, oxidase negative isolates were further confirmed by API 20 NE multi-test system (Biomeniuex, France). These tests were used according to the manufacturer's protocol for the identification of non-enteric bacteria. Well of the biochemical test were inoculated with bacterial suspension made from fresh bacterial culture. The suspension matched with 0.5 McFarland turbidity standards and incubated at 37oC for 18 - 24 hours. The result was read after the addition of reagent as a 7 digit number that identifies API 20 analytical index (API 20, Biomeriux, France (2010).

Antibiotics Susceptibility Test

The antibiotics susceptibility testing of isolated bacterial organisms was carried out using the disk diffusion method according to the guideline of the Clinical Laboratory Standard Institute (CLSI, 2014). Briefly, a bacterial suspension adjusted to 0.5 MacFarland has inoculated onto Muller – Hinton agar (Lib – Biotech).

The single disk (Oxoid, UK) antibiotics used include Amoxicillin-clavulanate (10µg), Pefloxacin (20µg), Cefriaxone (30µg), Ciprofloxacin (5µg), Ceftazidime (30µg), Co-trimozaxole (30µg), Imipenem (10µg), Rocephin (10µg), Amoxicillin (10µg) and Tetracycline (30µg). All the plates were incubated at 37oC for overnight, after which the plates were held a few inches above a black, non-reflective surface illuminated with reflected light and zone of inhibition were measured.

The result was recorded and was compared with the zone diameter interpretive standard of the Clinical Laboratory Standard Institute (CLSI, 2014).

Data Analysis

Descriptive statistics were used to report prevalence rate and cumulative prevalence of HCAIs.

RESULTS

A total of 401 non duplicated clinical samples were collected from admitted patients who consented and spend ≥ 14 days and are of ≥ 18 years in all the three selected study sites. A total of 138 (34.4%) from the 401 samples collected all the three selected study sites had positive HCAIs with MMSH having the highest prevalence 41.4%, following by MAWSH 36.1% and the least were observed at AKTH 29.0%. The overall highest positivity 28 (7.0%) was obtained in the age group 39 – 48 years following by 26 (6.5%) in the age group 49 – 58 years. The least positivity was observed among the age group 18 – 28years 14 (3.5%). Of the entire subjects that participated in study 201 (50.1%) were female with 76 (19.0%) positive prevalence (Table 1).

From the 138 positive isolates obtained in this study, it was observed that E. coli was the highest isolated organism 36 (26.1%), followed by *Proteus* species 23 (16.7%), the least isolated organisms were found to be *Streptococcus* species and *Pseudomonas* species 11 (9.4%) and 13 (9.4%) respectively. Wound swab has the highest positive isolates 48 (34.8%) following by urine 43 (31.2%) and least was from nasal intubation 16 (11.6%) isolates (Table 2).

Most of the isolated organisms obtained at AKTH were showed to be resistance to Amoxicillin (10µg), Cotrimozaxole (1.25/23.75µg) and Pefloxacin (20µg) except *K. pneumonia* which shows high resistant to Tetracycline (30µg) as shown in Table 3.

Table 4 showed the antibiotics resistant profile of the isolated organisms at MAWSH. Tetracycline (30µg) shows high resistant to *E. coli, Proteus* spp, *Pseudomonas* spp, and *K. pneumonia* while Cotrimozaxole (1.25/23.75µg) showed high resistance to E. coli, *A. baumannii* and *S. aureus*. Least resistance was observed from Imipenem (10µg) to *E. coli, Proteus* spp, and *K. pneumonia*.

Antibiotics resistant profile of the isolated organisms at MMSH shows Cotrimozaxole (1.25/23.75µg) showed high resistance to *Proteus* spp, *Pseudomonas* spp, *A. baumannii* and *S. aureus*, followed Amoxicillin (10µg) to *E. coli, Proteus* spp and *Pseudomonas* spp and least resistance with Imipenem (10µg) in *E. coli, Proteus* spp and *Pseudomonas* spp (Table 5).

Table 1: Demographic Distribution of Positive Isolates Obtained from the Three Study Sites in respect to the Age group of the Subject participants

Variables	Study Site						Total Positive
	MMSH		AKTH		MAWSH		
	No. Exam	Positive n (%)	No. Exam	Positive n (%)	No. Exam	Positive n (%)	
Age (Years)							
18 – 28	25	10(7.1)	10	2(1.0)	2	2(3.3)	14 (3.5)
29 – 38	19	10(7.1)	15	9(4.5)	9	6(9.8)	25 (6.2)
39 – 48	27	12(8.6)	41	13(6.5)	7	3(4.9)	28 (7.0)
49 – 58	22	6(4.3)	62	17(8.5)	27	3(4.9)	26 (6.5)
59 – 68	30	10(7.1)	41	10(5.0)	9	3(4.9)	23 (5.7)
69 – 78	17	10(7.1)	31	7(3.5)	7	5(8.2)	22 (5.5)
Total	**140**	**58(41.4)**	**200**	**58 (29.0)**	**61**	**22(36.1)**	**138 (34.4)**
Gender							
Male	63	27(19.3)	107	28(14.0)	30	7(11.5)	62 (15.5)
Female	77	31(22.1)	93	30(15.9)	31	15(24.6)	76 (19.0)
Total	**140**	**58(41.4)**	**200**	**58(29.0)**	**61**	**22(36.1)**	**138 (34.4)**

Key: MMSH = Murtala Muhammad Specialist Hospitals, AKTH = Aminu Kano Teaching Hospitals, MAWSH = Muhammad Abdullahi Wase Specialist Hospitals, Exam = Examined

Table 2: Organisms isolated from various health care associated infections sample in three Hospital Kano State, Northwest Nigeria

Organisms	Wound Swab 48 (34.8)			Urine Sample 43 (31.2)			Urine Catheter 30 (21.7)			Nasal Intubation 17 (12.3)			Total n (%)
	AKTH n (%)	MAWSH n (%)	MMSH n (%)	AKTH n (%)	MAWSH n (%)	MMSH n (%)	AKTH n (%)	MAWSH n (%)	MMSH n (%)	AKTH n (%)	MAWSH n (%)	MMSH n (%)	
Acinetobacter spp	1(5.9)	1(10.0)	2(9.5)	2(11.8)	1(16.7)	2(10.0)	2(14.3)	1(33.3)	1(7.7)	0(0.0)	0(0.0)	1(25.0)	**14(10.2)**
Klebsialla spp	0(0.0)	1(10.0)	1(4.8)	2(11.8)	2(33.3)	6(30.0)	2(14.3)	1(33.3)	3(23.1)	3(30.0)	0(0.0)	0(0.0)	**21(15.1)**
E. coli	4(23.5)	2(20.0)	5(23.8)	9(52.)	2(33.3)	6(30.0)	5(35.7)	1(33.3)	2(15.4)	0(0.0)	0(0.0)	0(0.0)	**36(26.1)**
Proteus spp	2(11.8)	1(10.0)	2(9.5)	2(11.8)	0(0.0)	2(10.0)	4(28.6)	0(0.0)	2(15.4)	4(40.0)	2(66.7)	2(50.0)	**23(16.7)**
Pseudomonas spp	3(17.6)	2(20.0)	5(23.8)	0(0.0)	0(0.0)	2(10.0)	0(0.0)	0(0.0)	1(7.7)	0(0.0)	0(0.0)	0(0.0)	**13(9.4)**
S. aureus	4(23.5)	2(20.0)	4(19.1)	2(11.8)	1(16.7)	2(10.0)	1(7.1)	0(0.0)	2(15.4)	1(10.0)	0(0.0)	1(25.0)	**20(14.5)**
Streptococcus spp	3(17.6)	1(10.0)	2(9.5)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	2(15.4)	2(20.0)	1(33.3)	0(0.0)	**11(8.0)**
Total	**17(12.3)**	**10(7.3)**	**21(15.2)**	**17(12.3)**	**6(4.4)**	**20(14.5)**	**14(10.2)**	**3(2.2)**	**13(9.4)**	**10(7.3)**	**3(2.2)**	**4(2.9)**	**138(100.0)**

Table 3: Antimicrobial Resistance Pattern of the isolated organism in Aminu Kano Teaching Hospital

Antibiotics (µg)	Isolated Organisms						
	E. coli **n=18 n (%)**	***Proteus*** **spp n=12 n (%)**	***Pseudomonas*** **spp n=3 n (%)**	***K. pneumonie*** **n=7 n (%)**	***A. baumannii*** **n=5 n (%)**	***S. aureus*** **n=8 n (%)**	***Streptococcus*** **spp n=5 n (%)**
Amoxicillin (10)	16 (88.9)	5 (41.7)	3 (100.0)	1 (14.3)	5 (100.0)	5 (62.5)	1 (20.0)
Cotrimozaxole (1.25)	17 (94.4)	3 (25.0)	2 (66.7)	1 (14.3)	5 (100.0)	6 (75.0)	2 (40.0)
Pefloxacin (20)	3 (16.7)	1 (8.3)	2 (66.7)	1 (14.3)	5 (100.0)	6 (75.0)	1 (20.0)
Tetracycline (30)	16 (88.9)	3 (25.0)	2 (66.7)	7 (100.0)	1 (20.0)	2 (25.0)	0 (0.0)
Ciprofloxacin (5)	3 (16.7))	2 (16.7)	1 (33.3)	0 (0.0)	1 (20.0)	2 (25.0)	2 (40.0)
Ceftazidine (30)	4 (22.2)	4 (33.3)	1 (33.3)	4 (57.1)	1 (20.0)	2 (25.0)	2 (40.0)
Augmentin (10)	2 (11.1)	4 (33.3)	0 (0.0)	4 (57.1)	1 (20.0)	3 (37.5)	2 (40.0)
Recephin (10)	3 (16.7)	4 (33.3)	1 (33.3)	2 (28.6)	2 (40.0)	2 (25.0)	2 (40.0)
Ceftriaxone (30)	3 (16.7)	4 (33.3)	2 (66.7)	2 (28.6)	2 (40.0)	3 (37.5)	2 (40.0)
Imipeniem (10)	5 (27.8)	1 (8.3)	0 (0.0)	3 (42.9)	5 (100.0)	4 (50.0)	1 (20.0)
Total n=58 n (%)	**18 (31.0)**	**12 (20.7)**	**3 (5.2)**	**7 (12.1)**	**5 (8.6)**	**8 (13.8)**	**5 (8.6)**

Table 4: Antimicrobial Resistance Pattern of the isolated organism in Muhammad Abdullahi Wase Specialist Hospital

Antibiotics (µg)	Isolated Organisms						
	E. coli **n=5 n (%)**	***Proteus spp*** **n=3 n (%)**	***Pseudomonas*** **spp n=2 n (%)**	***K. pneumonie*** **n=4 n (%)**	***A. baumannii*** **n=3 n (%)**	***S. aureus*** **n=3 n (%)**	***Streptococcus*** **spp n=2 n (%)**
Amoxicillin (10)	3(60.0)	1(33.3)	2(100.0)	1(25.0)	3(100.0)	2(66.7)	0(0.0)
Cotrimozaxole (1.25)	4(80.0)	2(66.7)	1(50.0)	2(50.0)	3(100.0)	3(100.0)	1(50.0)
Pefloxacin (20)	3(60.0)	2(66.7)	1(50.0)	1(25.0)	3(100.0)	3(100.0)	0(0.0)
Tetracycline (30)	4(80.0)	3(100.0)	2(100.0)	4(100.0)	1(33.3)	2(66.7)	1(50.0)
Ciprofloxacin (5)	1(20.0)	2(66.7)	1(50.0)	1(25.0)	1(33.3)	2(66.7)	1(50.0)
Ceftazidine (30)	3(60.0)	1(33.3)	2(100.0)	2(50.0)	1(33.3)	1(33.3)	1(50.0)
Augmentin (10)	3(60.0)	0(0.0)	1(50.0)	1(25.0)	1(33.3)	3(100.0)	0(0.0)
Recephin (10)	2(40.0)	1(33.3)	0(0.0)	1(25.0)	2(66.7)	2(66.7)	1(50.0)
Ceftriaxone (30)	3(60.0)	2(66.7)	1(50.0)	3(75.0)	2(66.7)	1(33.3)	1(50.0)
Imipeniem (10)	0(0.0)	0(0.0)	1(50.0)	0(0.0)	2(66.7)	2(66.7)	0(0.0)
Total n=22 n (%)	**5 (22.7)**	**3 (13.6)**	**3 (9.1)**	**4 (18.2)**	**3 (13.6)**	**3 (13.6)**	**5 (9.1)**

Table 5: Antimicrobial Resistance Pattern of the isolated organism in Murtala Muhammad Specialist Hospital

Antibiotics (µg)	Isolated Organisms						
	E. coli **n=13 n (%)**	***Proteus spp*** **n=8 n (%)**	***Pseudomonas*** **spp n=8 n (%)**	***K. pneumonie*** **n=10 n (%)**	***A. baumannii*** **n=6 n (%)**	***S. aureus*** **n=9 n (%)**	***Streptococcus*** **spp n=4 n (%)**
Amoxicillin (10)	11(84.6)	4(50.0)	6(75.0)	1(10.0)	4(66.7)	6(66.7)	1(25.0)
Cotrimozaxole (1.25)	12(92.3)	4(50.0)	6(75.0)	4(40.0)	6(100.0)	7(77.8)	1(25.0
Pefloxacin (20)	3(23.1)	3(37.5)	5(62.5)	2(20.0)	6(100.0)	7(77.8)	3(75.0)
Tetracycline (30)	12(92.3)	1(12.5)	2(25.0)	8(80.0)	2(33.3)	2(22.2)	2(50.0)
Ciprofloxacin (5)	1(7.7)	3(37.5)	1(12.5)	2(20.0)	1(16.7)	3(33.3)	2(50.0)
Ceftazidine (30)	3(23.1)	3(37.5)	2(25.0)	7(70.0)	3(50.0)	2(22.2)	1(25.0)
Augmentin (10)	3(23.1)	2(25.0)	2(25.0)	3(30.0)	1(16.7)	3(33.3)	1(25.0)
Recephin (10)	2(15.4)	2(25.0)	2(25.0)	5(50.0)	2(33.3)	3(33.3)	2(50.0)
Ceftriaxone (30)	3(23.1)	4(50.0)	2(25.0)	3(30.0)	2(33.3)	3(33.3)	1(25.0)
Imipeniem (10)	1(7.7)	1(12.5)	0(0.0)	4(40.0)	2(33.3)	4(44.4)	1(25.0)
Total n=58 n (%)	**13(22.4)**	**8(13.8)**	**8(13.8)**	**10(17.2)**	**6(10.4)**	**9(15.5)**	**4(6.9)**

DISCUSSION

Healthcare-associated infections continue to trouble the healthcare industry. The Centers for Disease Control and Prevention estimates one in 20 patients will contract HCAIs every day, and some estimates suggest the economic burden of HCAIs reaches $35.7 billion a year. Unfortunately for hospitals, they are a great breeding ground for infections (CLIC, 2014). To prevent HCAIs it is necessary to identify sources and modes of transmission of infection and to implement data driven prevention guide-lines and practices (Chandra and Milind, 2001). In this study 401 non duplicated clinical samples were analysed of which participants of age group 49 – 58 years have the highest prevalence of 102 (25.4%) followed by age group 59 – 68 years 89 (22.1%), the least was observed in age groups 18 – 28 and 29 – 38 years with 27 (6.7%) and 44 (11.0%) respectively. One hundred and thirty eight (34.4%) samples were showed to be positive from all the samples process of which AKTH and MMSH had the higher isolated organisms of 58 (42.0%) each. The overall highest positivity 28 (7.0%) was obtained in the age group 39 – 48 years following by 26 (6.5%) in the age group 49 – 58 years. The least positivity was observed in the age group 18 – 28 and 69 – 78 years with 14 (3.5%) and 22 (5.5%) respectively. The highest positivity was found in the age group 49 - 58 years 17 (8.5%) in AKTH, 6 (9.8%) from age group 29 – 38 years from MAWSH and 12 (8.6%) from age group 39 – 48 years in subject from MMSH. Morgan and Johns (2016) report in their study that the risk of developing a healthcare-associated infection (HCAI) increases linearly with age; a 2011 prevalence study reported a 11.5% HCAI prevalence rate in patients over the age of 85, which decreases significantly with younger age 11.27% in 75–84 group, 10.64% in 65–74, and 7.37% in patients under the age of 65 (Cairns *et al.*, 2011), While surgical site infections account for the largest proportion of HCAIs in the under 65 groups, HCAIs in the elderly are primarily attributed to urinary tract infections (Smith *et al.*, 2008). In another study conducted by Hans, (2012) reported that patients have an increased susceptibility to infection because they are weakened by disease, and also often elderly. Old age weakens the immune system and the function of vital organs. Lifestyle factors such as poor quality food, lack of exercise, and tobacco and alcohol abuse also play a role (Hans, 2012).

From a total number of 138 isolated obtained in this study, Gram-negative isolates were the commonest 77.5% and Gram-positive 22.5% of the total number of the isolates. Gram-negative bacilli have been reported as the most commonly associated organisms with hospital-acquired infections (Robert *et al.*, 2005). In another study done by (Negi *et al.*, 2015), reported that organisms isolated from the wound as surgical site infections (*S. aureus, Pseudomonas* spp, and *E. coli*) were severally been implicated as the major causative agent of HCAIs. This finding corresponds with this work finding which reported the high prevalence of 34.8% was obtained from a wound sample of post-surgical infections.

Antibiotics resistance to commons used antibiotics continues been a challenge to clinicians in patient management which can contribute to prolonging hospitalize patient stays, increase in mortality rate, increases in the tendency of acquired nosocomial infection and more expenses from both patients and government in general. In our study, high resistant were observed in Amoxicillin (10µg), Cotrimozaxole (1.25/23.75µg) and Tetracyline (30µg) with the least resistant to Imipinem (10µg) and Ceftrixone (30µg). This finding corresponds with what obtained by Peter *et al.* (2017) in Uganda where 97% of the isolates were resistance to Amoxicillin (10µg). In India Kamat *et al.* reported 70% resistance of Amoxicillin (10µg) and Tetracycline (30µg) to the most Gram-negative bacilli (Kamat *et al.*, 2008). Perez *et al.* reported Imipenem (10µg) was resistant to *Acinetobacter* spp ranges between 0 – 40% in 2000 to 2004 (Perez *et al.*, 2007), which is in line with what obtained this study which *A. baumannii* has 37.7% resistance to Imipenem (10µg).

CONCLUSION

From what was obtained in this study, it shows that Gram-negative bacilli were frequent organisms associated with nosocomial infection in this study site. Patients with post-surgical site infection have the highest percentage of nosocomial infection in hospitals, where *S. aureus, E. coli*, and *Proteus* spp are the predominate species.

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