International Journal of TROPICAL DISEASE & Health
17(4): 1-12, 2016, Article no.IJTDH.27253
ISSN: 2278–1005, NLM ID: 101632866

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Post-surgical Infection and Antibiotic Susceptibility Patterns of Bacteria Isolated from Admitted Patients with Signs of Infection at Jimma University Specialized Hospital, Jimma, Ethiopia

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Authors' contributions

This work was carried out in collaboration between all authors. Authors TS and SA did the study design and wrote the protocol. Authors TS, GB, SE and SA did the statistical analysis and literature searches while analyses of study was by authors TS, GB, SE and SA. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IJTDH/2016/27253
Editor(s):
(1) Viroj Wiwanitkit, Department of Laboratory Medicine, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand.
Reviewers:
(1) Anonymous, Dr. Pablo O. Torre Memorial Hospital, Philippines.
(2) Cucunawangsih, University of PelitaHarapan, Tangerang, Indonesia.
(3) Anonymous, Nnamdi Azikiwe University, Agulu Campus, Awka, Nigeria.
(4) M. Jayavarthinni, Indira Gandhi Medical College & Research Institute, India.
Complete Peer review History: http://sciencedomain.org/review-history/15381

Original Research Article

Received 25th May 2016
Accepted 5th July 2016
Published 13th July 2016

ABSTRACT

Introduction: Nosocomial infection (hospital acquired infections) poses a significant burden and threat for public health. Treatment of such infections is becoming difficult due to resistance of antibiotics to the bacteria that circulate in the Hospital environment.

The aim of this study was to determine the profile and drug resistance pattern of bacteria isolated from inpatients with clinical signs of infection.

Methodology: A cross sectional study was done on patients admitted to surgical and gynecological wards. All patients were followed until discharge. Patients who had shown signs of infection after 48

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hours of admission were interviewed for some socio demographic and associated factors data. Wound swab, urine and blood specimens were collected and processed to isolate and identify bacteria. The antibiotic sensitivity pattern of the isolates was done.

Results: The overall prevalence of culture positive nosocomial infection was 35%. *Staphylococcus aureus, coagulase negative staphylococci, E. coli,* and *Pseudomonas species* were the most frequent isolates. Nearly 100% of Gram positive and 92.6% of Gram negative isolates had shown multi drug resistance against two or more commonly used classes of drugs.

Conclusion: The observed multi drug resistance rate of the isolates is very worrisome and demands immediate attention.

Keywords: Nosocomial; antibiotic resistance; bacteria; Jimma University.

1. INTRODUCTION

Nosocomial or Hospital-acquired infection (HAI) continues to be a major public health concern throughout the world because of the associated morbidity, mortality and socioeconomic costs [1,2,3]. It can prolong duration of stay in hospital, increases the cost of health care and the risk of emergence of multiple antibiotic resistant microorganisms [3,4].

The most frequent nosocomial infections seen are pneumonia, intravenous device related blood stream infection (BSI), surgical site infection (SSI) and urinary tract infection (UTI) [5]. These infections are usually caused by bacteria including Coagulase-negative staphylococci (*CoNS)*, *Staphylococcus aureus*, *Enterococcus species*, *E. coli*, *Klebsiella species and Pseudomonas species* [6,7].

Nosocomial infection occurs among 7-12% of the hospitalized patients globally with more than 1.4 million people suffering from its complications. Regionally, it occurs in 5%-10% of all hospitalizations in Europe and North America. Furthermore, it occurs in more than 40% of hospitalizations in parts of Asia, Latin America and sub-Saharan Africa [8,9,10]. Treatment of nosocomial infections is becoming difficult due to the increasing trend of antibiotics resistance. This condition increases the morbidity and mortality associated with infections. It contributes substantially to the rising costs of care which is associated with prolonged hospital stays and the need for more expensive drugs. Therefore current knowledge about the etiologic agents and their antibiotic susceptibility pattern of the isolates is essential for appropriate therapy.

In Ethiopia, a report from Tikur Anbessa and Mekelle hospitals indicated how the problem is serious [4,5,11,12]. In Jimma University Specialized Hospital (JUSH) only few studies were done on patient samples in the year 2000, 2001 and 2008 [13,14,15]. These studies reported high bacterial resistance against commonly used antibiotics. However, none of these studies included surgical and gynecological wards of the hospital. Besides, most of these studies were done more than 10 years ago and could not reflect the current picture of the problem. One recent study conducted in 2011 reported high indoor air bacterial load at surgical wards and operating rooms of JUSH as compared with the standard expected indoor air bacterial load. This report suggested the need of doing a study to depict the nosocomial infection burden and the bacteria profile of isolates on patients who are admitted and have been receiving medical care in these rooms [16].

Hence, this study was done to determine the magnitude of nosocomial infection, bacteria profile and drug resistance pattern of the isolates among nosocomial suspected patients admitted to surgical and gynecological wards to generate important base line information.

2. MATERIALS AND METHODS

2.1 Study Settings and Selection of Participants

Hospital based longitudinal study was carried out from June, 2012 to February; 2013 at Jimma University Specialized Hospital (JUSH). The hospital is tertiary level hospital which is found in Jimma town, Oromia region and 352 Km away from Addis Ababa (capital city of Ethiopia) in South West direction. According to the data obtained from the hospital statistics department, JUSH has 11 wards, about 444 beds and 861 health professionals. Jimma University Specialized hospital besides its being a teaching hospital, it provides services to over 1 million populations in the region. It is the only teaching and referral hospital in South Western Ethiopia [17].

2.1.1 Inclusion criteria

All patients admitted to surgical and gynecological wards, who had elective or emergency surgery, with no clinical signs and symptoms of infection within the first 48 hours, and with negative urine culture for asymptomatic bacteriuria at admission were included. Such patients have been followed up to the date of discharge for signs of infection.

2.2 Definition of Suspected Cases

In this study, presentation of at least one of suggestive signs [fever (T°>38℃), urgency, frequency, dysuria, supra-pubic tenderness and indwelling catheter insert] was considered as a clinical sign for urinary tract infection (UTI). Likewise, presentation of at least one of [fever (T°>38℃), pain, tenderness, localized swelling, redness, heat, abscess or drainage] was considered as clinical sign for surgical site infection (SSI). Presentation of at least one of indicative signs (fever (T°>38℃), chills, hypotension (systolic pressure <90 mm) oligouria (<20 cm^3/hour)) was considered as a clinical sign for blood stream infection (BSI) [18].

2.3 Definition of Proven Nosocomial Cases

2.3.1 Confirmed blood stream infection (Septicemia)

presence of fever (T°>38℃), chills, or hypotension accompanied with a positive blood culture for isolated organism is not related to an infection at another site or for common skin contaminants isolation of the same bacteria from blood samples drawn at two different occasions [18].

2.3.2 Confirmed urinary tract infection

The presence of dysuria, urgency and frequency of urination and cloudy urine after admission or instrumentation of urinary tract followed by significant bacteriurea. In this study mid-stream urine cultures with $\geq 10^5$ colony forming units (CFU) and catheterized urine with $\geq 10^2$ colony forming units were considered as significant. On top of this indwelling catheter infection was defined by isolation of bacteria during the presence of indwelling urinary catheter or within 7 days before the onset of the symptoms [18].

2.3.3 Confirmed surgical site infection

Presence of pus or infected fluid at the site of operative incision that occurred until discharge after the operative procedure followed by isolation of bacteria from the discharge.

2.4 Sample Size

The sample size (n) was calculated using the prevalence of nosocomial infection 13% in Tikur Anbessa, Addis Ababa, Ethiopia [11]. The expected margin of error (d) taken was 0.05 with the confidence interval ($Z\alpha/_2$) of 95%. The sample size was calculated using single population proportion formula [19]. Considering 10% contingency for unknown reason, a total of 192 patients were the minimum estimated sample size for this study.

2.5 Social Demographic Data and Specimen Collection

All admitted patients during the data collection period were followed daily by the researcher and data collector nurse. Patients who fulfill the inclusion criteria during admission and developed signs of infection during follow up were consented and interviewed by pre-designed and semi-structured questioners for socio-demography and some associated factors data. Then, urine sample, wound swab and venous blood samples were collected following strictly standard operating procedure of sample collection for bacteriological investigation [20,21]. Then, immediately the samples were transported to Jimma University Medical Microbiology laboratory (located in the same compound) for further processing.

2.6 Specimen Processing and Bacterial Identification

Urine and wound swab specimens were inoculated on Blood agar, Mannitol Salt agar and MacConkey agar (Oxoid Ltd, Basingstoke Hampshire, UK) and processed using standard procedures [20,21]. Venous blood samples were drawn in duplicate on separate occasions and directly dispensed into Brain-heart infusion (BHI) broth (Oxoid Ltd, Basingstoke Hampshire, UK) and processed as described previously. Bacterial isolates were identified using colony morphology, Gram reaction and different biochemical tests [20,21].

2.7 Antibiotic Sensitivity Testing (AST)

Antibiotic sensitivity testing was done using Kirby–Bauer modified disc diffusion technique [22]. Antibiotic discs selection was made based on Clinical and Laboratory Standards Institutes recommendations [23]. Accordingly the antibiotic discs used for Gram positive bacteria were; Ampicillin (AMP, 10 µg), Chloramphenicol (C, 30 µg), Ciprofloxacin (CIP, 5 µg), Cloxacillin (OB, 5 µg), Doxycyclin (DO, 30 µg), Erythromycin (E, 5 µg), Gentamycin (CN, 10 µg), Nitrofurantoin (FM, 300 µg). Norfloxacin (NOR, 10 µg), Oxacillin (OX, 5 µg), Penicillin G (P, 10 IU), Tetracycline (TTC, 30 µg), Trimethoprim-Sulphamethoxazole (SXT, 25 µg). The disc used for Gram negative bacteria were; Amoxicillin-Clavulanic acid (AMC, 30 µg), Ampicillin (AMP, 10 µg), Ceftriaxone (CRO, 30 µg), Chloramphenicol (C, 30 µg), Ciprofloxacin (CIP, 5 µg). Gentamycin (CN, 10 µg), Nalidixic acid (NA, 30 µg), Nitrofurantoin (FM, 300 µg), Norfloxacin (NOR, 10 µg), Tetracycline (TTC, 30 µg), Trimethoprim-Sulphamethoxazole (SXT, 25 µg).

All antibiotic discs were from Oxoid Ltd, Basingstoke Hampshire, UK. Reference strains that include ATCC 25922 (*E. coli*), 25923 (*S. aureus*) and 27853 (*P. aeruginosa*) were used for quality control of reagents, culture media and antibiotic discs.

2.8 Data Management and Analysis

Data were collected and checked for completeness, edited, cleaned and entered and analyzed using SPSS windows version16. Descriptive statistics, frequency of drug resistance and MDR pattern of the isolates, frequency distribution of socio demographic and associated factors were done. Chi square test was used to see the association between some factors with culture proven infection. P-value <0.05 was considered statistically significant.

3. RESULTS

A total of 500 patients were admitted to Surgical (n=350) and Gynecological (n=150) wards. From the admitted patients only 200 patients (40%) showed clinical signs of infection (Fig. 1).

The median age of patients with clinical sign of infection was 35 years with a range of 18-75 years. The four main reasons for admission in surgical ward were; intestinal obstruction 23/151(15.2%), goiter 19/151 (12.5%), laparatomy 17/151 (11.25%) and cholecystectomy 16/151 (10.5%). Likewise the four major reasons for admission in gynecology ward were; cesarean section 12/49 (24.4%), Myoma 11/49 (22.4%) and ovarian tumor 8/49 (16.3%). The median length of hospital stay for admitted patients with clinical sign was 11 days (ranges from 3 to 45 days).

From 200 patients that fulfilled the case definition of hospital acquired infections, 70(35%) were culture positive. The most frequently encountered infection was surgical site infections (SSI) 33(47.1%) followed by UTI 21 (30%). Patients who had both blood stream infections (BSI) and SSI were 13(18.6%). The least 3 (4.3%) frequent infection was BSI.

Majority 40 (57.1%) of culture positive nosocomial patients were males and the rest (42.9%) were females. Nineteen (27.1%) patients with urinary catheter and 12 (17.1%) patients with intra-venous (IV) cannulation had developed culture positive nosocomial infection (Table 1).

In this study a total of 111 bacterial etiologic agents were isolated. Majority 67 (60.7%) of the isolates were Gram negative bacteria and the rest 44(39.3%) were Gram positive. The most predominant isolates were *Staphylococcus aureus* 22(19.6%), coagulase negative staphylococcus 21(18.7%), *Escherichia coli* 9(8.0%), and *Pseudomonas species* 8 (7.0%) (Table 2). The least frequent isolate was *S. pyrogen* 1 (2.27%) (data not shown in Table 2 due to low number of observation)

Over all 69(61.6%), 25(22.3%) and 18(11.1%) of the isolates were detected from surgical site, urinary tract and blood stream infections respectively. *Staphylococus aureus, Eschercia coli* and coagulase negative staphylococcus (*CoNS*) were the main bacteria isolated from SSI, UTI and BSI (Fig. 2).

3.1 Antibiotic Susceptibility Testing

Over all, Gram positive bacteria showed high level of resistance (> 80% resistance) to most tested antibiotics. The isolates showed 100% resistance against Penicillin (P) and Doxycycline (DO). Intermediate level of resistance (60-80%) has been observed against Ciprofloxacin (CIP), Norfloxacin (NOR), and Trimethoprim-Sulphamethoxazole (SXT) (Table 2). Gram positive bacteria isolated from urine samples showed intermediate resistance for Nitrofurantoin (Table 2). Gram negative bacterial isolate showed high level of resistance against Ampicillin (AMP) and Ceftrixaone (CRO). They also developed intermediate level of resistance for most antibiotic tested. However, low level of resistance (<60%) has also been observed against Gentamycin (CN), Ciprofloxacin (CIP) and Norfloxacin (NOR) (Table 2).

3.2 MDR Patterns

Most of the isolated bacteria showed different multi drug resistance (MDR) pattern against tested drugs. MDR is defined as bacteria, that are resistant to two or more classes of antimicrobial agents (24). Based on this definition Gram positive bacteria showed MDR pattern that ranges from 2-8 classes of antibiotics. Likewise the MDR pattern of gram negative bacteria ranges from 2-9 different classes of antibiotics (Fig. 3).

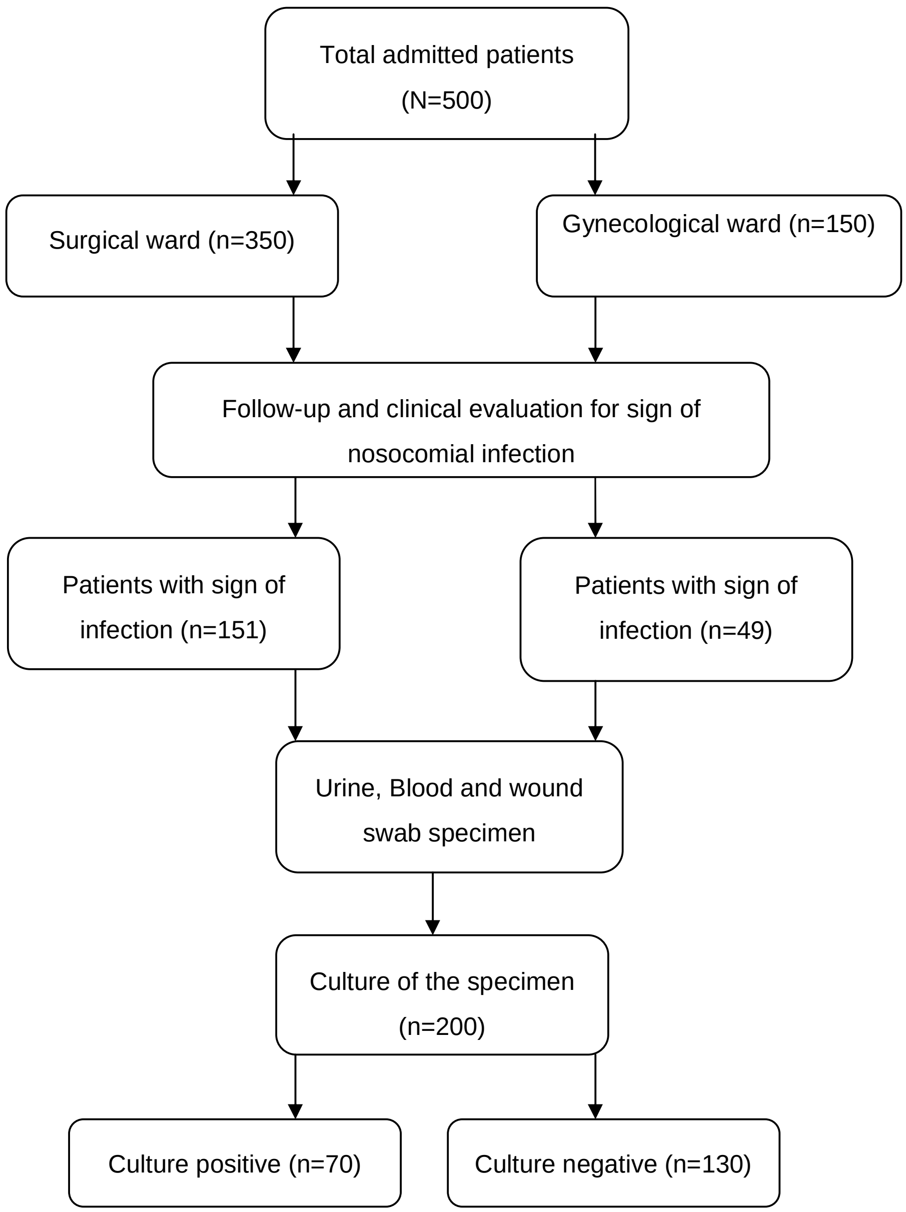

Fig. 1. Overall work flow

Table 1. Socio demographic and some associated factors distribution among participants

Associated factors	Nosocomial infection		p-value
	Yes n (%)	No n=130 (%)	
Sex			
Male	40 (57.1)	63 (48.5)	
Female	30 (42.9)	67 (51.5)	0.529
Age			
<65	61 (87.1)	126 (96.9)	
≥ 65	9 (12.6)	4 (3.1)	0.007
Wards			
Surgical	55 (78.6)	98 (75.4)	
Gynaecological	15 (21.4)	32 (24.6)	0.912
Catheterization			
Yes	19 (27.1)	5(3.9)	
No	51 (72.8)	125 (96.1)	<0.001
IV canulation			
Yes	12 (17.1)	4 (3.1)	
No	58 (82.9)	126 (96.9)	<0.001
Total (n)	70	130	

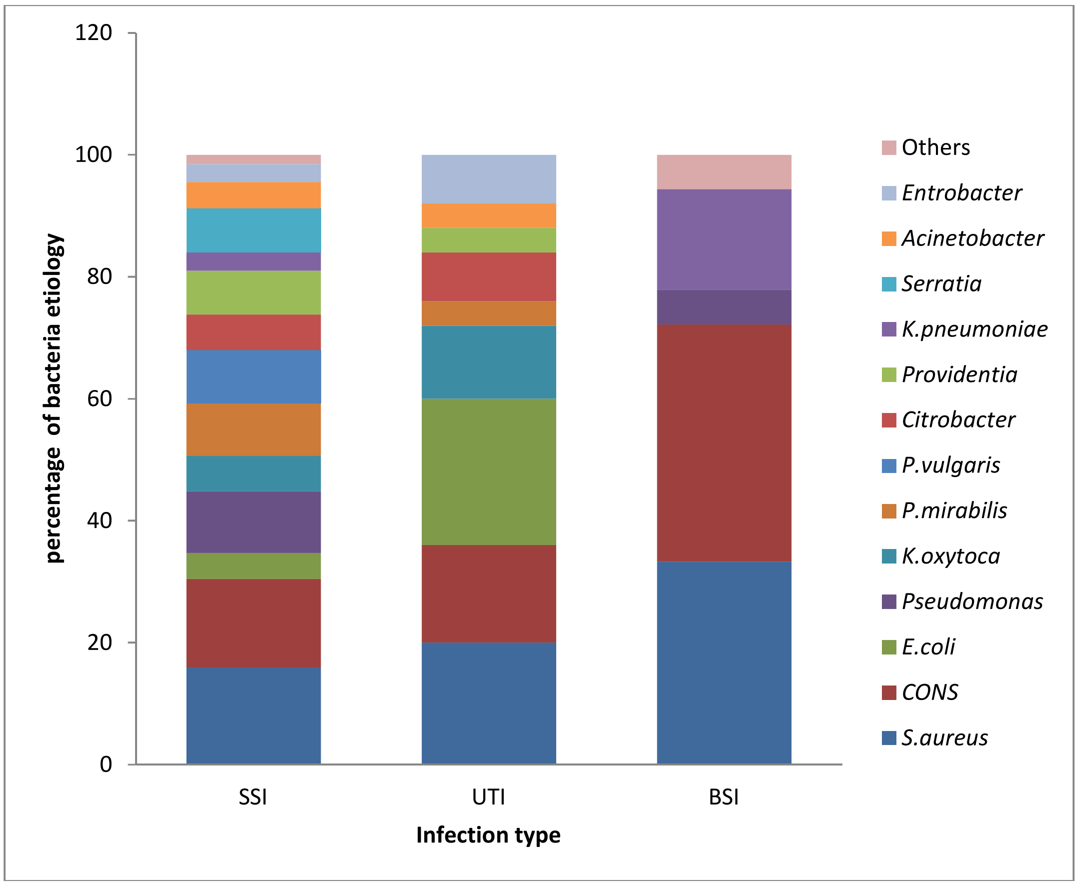

Fig. 2. Percentage component graph of isolated bacteria in relation to the type of infection
Where SSI - Surgical Site Infection; UTI - Urinary Tract Infection; BSI - Blood Stream Infection

Table 2. Antibiotic resistance patterns of bacteria isolates identified from patients admitted to surgical and gynaecological wards

	***S. aureus* n=22 %**	***CoNS* n=21 %**	***E.coli* n=9 %**	***K. pneumonia* n=5 %**	***K. oxytoca* n=7 %**	***Enterobacter* n=4 %**	***Citrobacter* n=6 %**	***Serratia* n=5 %**	***P. mirablis* n=7 %**	***P. vulgaris* n=6 %**	***Providencia* n=6 %**	***Pseudomonas* n=8 %**	***Acinetobacter* n=4 %**
Amoxicillin-Clavulanic acid	NA	NA	66.6	80	100	NA	NA	NA	71.4	83.3	NA	87.5	NA
Ampicillin	95.4	85.7	88.8	NA	100	NA	NA	NA	100	NA	NA	NA	100
Chloramphenicol	90.9	95.2	33.3	40	28.5	100	50	100	57.1	83.3	100	NA	NA
Ciprofloxacin	59.1	76.2	44.4	20	57.1	100	50	80	42.8	16.6	50	50	50
Cloxacillin	90.9	100	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Doxycycline	100	100	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Erythromycin	95.4	95.2	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Gentamycin	81.8	85.7	44.4	60	71.4	75	66.6	40	28.6	66.6	66.6	50	50
Nitrofurantoin	80	50	55.5	20	42.8	25	50	60	71.4	83.3	83.3	87.5	50
Norfloxacin	54.5	85.7	44.4	20	57.1	75	50	60	28.6	16.6	50	50	50
Oxacillin	95.4	95.2	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Penicillin G	NA	100	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Tetracycline	90.9	95.2	55.5	NA	71.4	75	50	80	NA	NA	NA	NA	100
Trimethoprim-Sulphamethoxazole	77.2	77.2	66.6	40	71.4	100	100	60	85.7	83.3	100	NA	75

NA; Not applicable.

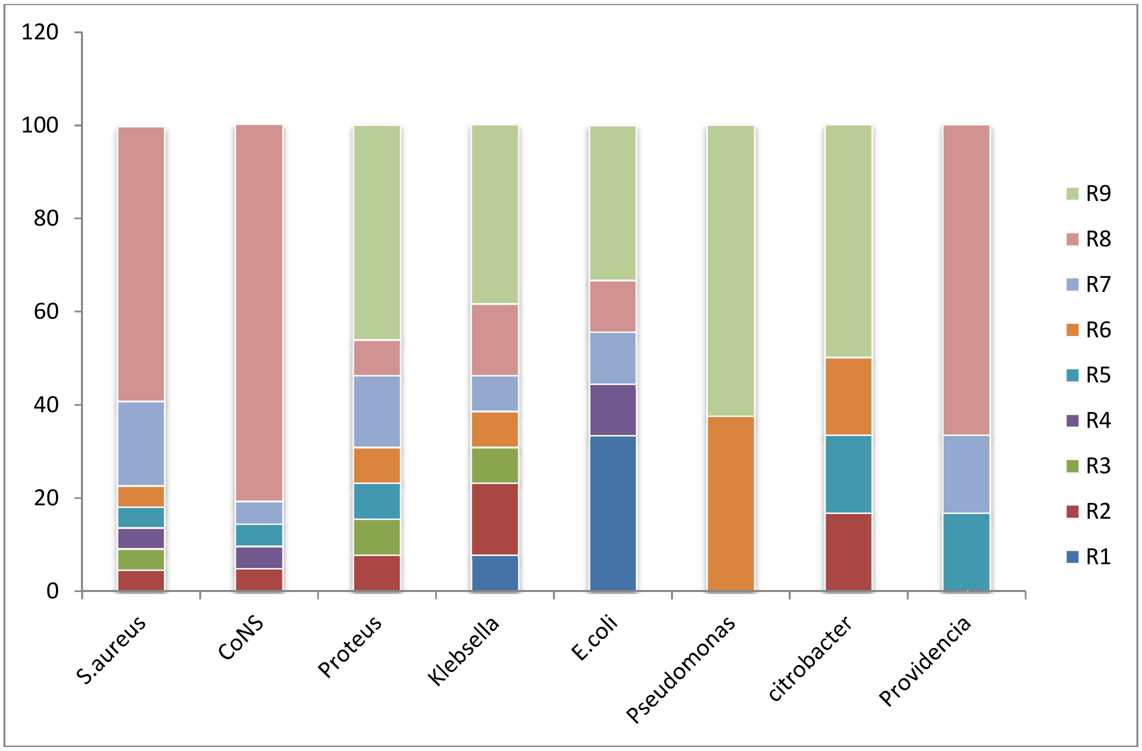

Fig. 3. Percentage component graph multi drug resistance pattern of bacteria isolated from patients admitted to surgical and gynaecological wards, JUSH, Jimma Ethiopia

Where For Gram positive bacteria: R1=single class resistance, R2=Pencilline+tetracyclines, R3=R2+Macrolids, R4=R3+Chloramphenicol, R5=R4+Cephalosporines, R6=R5+Amonoglycosides, R7=R6+Sulphonamides and R8=R7+quinolones. For Gram negative bacteria: R1=single antibiotic class resistance, R2=Penicillin +Tetracycline, R3=R2+Cephalosporines, R4=R3+Sulphonamides, R5=R4+Nitrofurans, R6=R5+Pencilline combinations, R7=R6+Chloramphenicol, R8=R7+Quinolones, R9=R8+aminoglycosides

4. DISCUSSION

The overall prevalence of culture proven nosocomial infection among patients with clinical signs was 35%. This result is higher than previous studies done in Ethiopia such as, Tikur Anbesa Hospital and Mekele Hospital with prevalence of 13% and 27.6% respectively [11,12]. It is also higher than studies conducted in other counties such as 14.3% in Greece and 13% in Tunisia [25,26].

The higher prevalence in this study might be related with high bacterial contamination of Operation Room (OR) and surgical wards. According to previous study report from this hospital, aerobic colony count of the OR room was far beyond the set 5-8cfu/hr acceptable standards for passive room [27]. Apart from this time difference of the studies could also be speculated for this huge difference of nosocomial infection rate between this study and previous report from Ethiopia, Tikur Anbesa Hospital which was done towards the end of 1980's. It is known that through time there is a possibility for microorganisms to spread in hospital environment due to resistance development against available antibiotics.

In this study surgical site infection, with or without association with BSI (65.7%) was the most commonly seen infection. This is because of an increased risk of getting an infection in the hospital by direct invading of the patient's body, which favors the entrance of bacteria in to normal sterile parts of the body. This infection can be acquired from contaminated surgical equipment, operation room, and prolonged hospitalization or from health care workers [1]. Our finding is higher as compared to local studies done in Tikur Anbessa hospital with a prevalence rate of 52.1% in 2009 and 52.4% in 2011 respectively [4,5]. However, lower than other local study done in St. Paul Hospital (2009) which had reported 79.4% prevalence [4]. Similarly it is higher than study findings reported from different countries such as from Tunisia (33%) in 2006 and Swiss (30%) in 2005 which might be related with the facility and resource difference required for surgery [26,28,29].

A total of 111 bacterial etiologic agents were identified from culture proven nosocomial patients. Of these the isolation rates of Gram negative and gram positive bacteria were 60.7% and 39.3% respectively. This is in agreement with other similar studies done in Ethiopia where more than 60% of the isolates were Gram negative [5,11,12]. Among the total 111 bacterial isolates from surgical site infection; *Staphylococcus aureus* (15.9%), CoNS (14.5%), and *Pseudomonas* (10.1%), were the dominant isolates. Similar findings have been observed in Nigeria and Swiss (29,30). Among the bacteria isolated from patients with UTI, *E. coli* (24%), *Klebsiella oxytoca* (12%) and *Citrobacter* species (8%) were dominant which is in line with many similar study findings conducted in Ethiopia, Swiss and Nigeria [5,29,31]. Among blood stream infection isolates, CoNS (38.9%) *Staphylococcus aureus* (33.3%) and *S. pyogen* (5.6%) were the important etiologic agents and which is comparable with previous studies done in Ethiopia, Nigeria, Taiwan and Tanzania [3,5,32-34].

In this study both Gram positive and Gram negative bacterial isolates showed considerable resistance to the commonly prescribed antibiotics in Ethiopia and particularly in the study area, which is comparable with other local study results done in Mekele, Addis Ababa, Bahirdar, and Jimma [5,11,12,35,36], High level of resistance (>80%) was recorded among Gram positive isolates to many tested antibiotics which is comparable with many other study findings done in different countries [36,37].

More than 95 % of the *S. aureus* are oxacilin resistant and are likely MRSA isolates. This finding is consistent with previous reports from Ethiopia where 100% and >80% of the isolates showed Methicillin resistance [11,20]. This finding is also higher than (75%) Methicillin resistance reported from Taiwan [37].

In this study Gentamycin, Ciprofloxacin and Norfloxacin were relatively effective drugs for the treatment of infections caused by Gram negative bacterial isolates, which is comparable with other local study results from Addis Ababa, Mekele and Bahirdar Felegehiwot hospital [5,12,35]. The level of resistance is higher in Gram positive as compared with that of Gram negative isolates. Possible reasons for this could be; cross resistance to cell wall synthesis inhibitors, their natural course of resistance and indiscriminate and frequent use of antibiotics in the hospital and prolonged survival of Gram positive bacteria on hospital environment than Gram negative bacteria. Our study findings are in line with other study results done in Ethiopia [31,32,35].

According to the study report from different directions of the world, currently resistance rate to the commonly prescribed antibiotics in Ethiopia and other countries is alarmingly high [38]. Due to this great emphasis, attention should be given while treating infections caused by hospital isolates. In Ethiopia it is a common practice to purchase antibiotics without prescription, which leads to misuse of antibiotics by the public. This contributes for the emergence and spread of antibiotic resistant bacterial pathogens. Other causal factors can be poor drug quality, poor hospital hygienic conditions and inadequate surveillance, all of which are crucial for good clinical practice and for rational policies against antibiotic resistance [12].

This study has few limitations and should be interpreted carefully. We were not able to include pneumonia as one of the common nosocomial infection. This is associated with the inherent limitation of sputum sample which could be contaminated by upper respiratory tract floras and technical difficulty to employ invasive sample collection procedures. In spite of this, we believe this research clearly indicated the magnitude of the problem, the bacteria profile and antibiotic susceptibility pattern of the isolates in Jimma University Specialized Hospital.

5. CONCLUSION AND RECOMMENDATION

High prevalence of bacteria nosocomial infection and drug resistance rate was seen in Surgical and gynaecological wards of Jimma University specialized Hospital. This demands an urgent attention to make a concerted effort to minimize nosocomial infections and drug resistance. Further study is recommended to identify and address the main determinant for the observed problems.

ETHICAL CONSIDERATION

Ethical approval was obtained from Jimma University, College of Public Health and Medical

Sciences Staff Research and Post Graduate coordinating office Ethical Review Committee. Participant information sheet had been explained for potential participants with clinical signs of infection. Signature on consent form was received from participants agreed to be a part of the study. Based on the laboratory results, study participants were treated by the respective Surgeon or Gynecologist on duty. Confidentiality of information was maintained by giving a particular code to each patient and no name of a patient was mentioned during reporting.

ACKNOWLEDGEMENT

The authors are grateful to Jimma University Specialized Hospital and College of Health Sciences for their genuine assistance during the course of this research.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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