

Prevalence of Multidrug Resistant Bacteria in Postoperative Wound Infections at Tikur Anbessa Specialized Hospital, Addis Ababa, Ethiopia

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Abstract

Background: Postoperative wound infections are major problems throughout the world. Moreover, multidrug resistance (MDR) bacteria pose challenges for control. Hence, this study aimed to determine the prevalence of multidrug resistant bacteria from postoperative wound infections.

Methods: A cross-sectional study was conducted from March to August 2015. A total of 197 patients who developed postoperative wound infections after surgery were included. Wound swabs were collected using sterile cotton swabs and dipped into sterile amines transport media. All samples were cultured on blood, MacConkey and mannitol salt agar. Culture positive samples were characterized by gram stain, colony morphology and standard biochemical tests. Antimicrobial susceptibility testing was performed on Muller-Hinton agar using disk diffusion. Data was analyzed using SPSS version 20.

Results: Overall bacterial prevalence was 75.6% (n=149/197) and predominant bacterial isolates were *Staphylococcus aureus* 33.3% (n=56/168) and *Escherichia coli* 14.3% (n=24/168). Double infections were seen 11.4% (n=17/149) of which 23.5% (n=4/17) *Staphylococcus aureus* and *Pseudomonas spp.* occurred together. Highest proportion of bacteria 28.2% (n=42/149) were isolated from orthopedics department followed by general surgery 24.8% (n=37/149). Of all bacterial isolates, the multidrug resistance level was 65.5% (n=110/168). Gram positive and Gram negative bacteria showed an MDR level of 55.3% (n=42/76) and 73.9% (n=68/92) respectively. Amoxicillin (93.5%), ceftriaxone (85.3%) and penicillin (84.5%) were least effective.

Conclusion: Aseptic surgical practice and standard operating procedures for wound management should be followed to minimize postoperative wound infections. The possible choices of antibiotic options for treatment of postoperative wound infections are few hence for preventing further emergence and spread of MDR bacteria rational use of antibiotics and regular monitoring of antimicrobial resistance patterns is essential.

Keywords: Multidrug resistance; Bacteria; Postoperative wound infections; Tikur anbessa specialized hospital; Addis ababa; Ethiopia

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Citation: Asres GS, Legese MH, Woldearegay GM (2017) Prevalence of Multidrug Resistant Bacteria in Postoperative Wound Infections at Tikur Anbessa Specialized Hospital, Addis Ababa, Ethiopia. Arch Med. Vol. 9 No. 4:12

Received: August 17, 2017; **Accepted:** August 28, 2017; **Published:** August 31, 2017

Introduction

Postoperative wound infections are infections that occur within 30 days of the operation or within 1 year of operation if an implant is left in place and if the infections appear to be related

to the surgery [1]. The infection is associated with the intensity of bacterial contamination of the wound at surgery or later in wards during wound care that follows interference with the skin barrier [2]. The third most commonly reported nosocomial infection is postoperative wound infections which accounts for

approximately a quarter of all nosocomial infections [3]. It has been responsible for the increasing cost, morbidity and mortality related to surgery and continues to be a major problem even in hospitals with modern facilities and standard protocols of preoperative safety measures and antibiotic prophylaxis [1,3].

Since long ago wounds have been classified in to four categories called clean, clean-contaminated, contaminated and dirty according to the theoretical number of bacteria that contaminate wounds [4,5]. It has been reported that clean, clean-contaminated, contaminated and dirty wound categories account approximately 1.5-3.9%, 3-4%, 8.5% and 28-40% of wound infection rates respectively [5]. The source of postoperative wound infections can be either endogenously from body flora following surgical manipulation or exogenously from the hospital staffs, other patients and visitors, foods, water, fomites [6-9]. Patients who developed postoperative wound infections are 60% more likely to spend time in an intensive care unit, 5 times more likely to be readmitted and twice more likely to die than patients who don't have these infections [4].

Most common postoperative wound infection pathogens are *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella* species, *Proteus* species, *Citrobacter* species and *Coagulase negative staphylococci* (CONS) [10,11]. The role of these microbes as etiological agents in hospital wound infections is partly because of their ability to survive in the hospital environment by developing resistance to antimicrobials and disinfectants and also their ability to rapidly colonize the body surface of compromised host [9,10]. Rapidly spreading antimicrobial resistance in bacterial populations has made the management and treatment of postoperative wound infections a serious challenge in clinical and surgical practice [12]. Mainly if it is caused by multidrug resistant bacteria it worsens the condition and specifically it has become serious problem in developing countries due to crowding hospital environment, irrational prescription of antimicrobial agents and poor infection prevention program [13]. More over the battle between bacteria and their susceptibility to antibiotics is a problem among public, researchers, clinicians and drug companies who are looking for effective drugs [14].

Materials and Methods

A cross-sectional study was conducted for a period of six months (from March to August 2015) at Tikur Anbessa Specialized Hospital which is the biggest referral hospital of Ethiopia located in the capital city, Addis Ababa. All surgical patients, irrespective of age, operated during the study period that later developed symptoms of post-operative wound infections and who gave informed consent and/or assent to participate on the study were included. However, patients who developed postoperative wound infections later than 30 days after the operation and patients who had infected burn wounds were excluded. A total of 197 study participants were recruited using convenient sampling technique from various surgical wards (orthopedic, ICU, general surgery, pediatrics, obstetrics and gynecology, internal medicine and OPD) of the hospital. The sample size was calculated based on single population proportion from previous study done in Ethiopia where the prevalence was 88.1% (0.881)

[11]. Patients were identified from daily operation schedules and those patients who operated during the study period were observed daily for any signs of postoperative wound infection for a total of 30 postoperative days. From eligible participants, patient specific demographic characteristics were collected from the patient card after obtaining informed consent using data collection sheets. Information on potential predicating variables of wound infection and whether wound class was clean, clean contaminated, contaminated and dirty were recorded from the responsible surgeon. Wound swabs were collected aseptically with sterile cotton tipped swabs by the principal investigator, trained nurses and resident doctors. After cleaning skins, the sterile cotton tipped swabs were placed centrally and rolling technique was applied to collect wound samples. All wound swabs were dipped into Stuarts transport media after collection and taken to bacteriology laboratory within 30 minutes for culture and drug susceptibility testing.

Culture and identification

All wound swab specimens were inoculated on blood agar, MacConkey agar and mannitol salt agar and incubated at 37°C for 24 hours aerobically. Identification of bacterial isolates was performed using colony morphology, Gram stain and conventional biochemical tests. Gram positive bacteria were identified at species level using catalase, coagulase, latex agglutination test and Pastorex TM staph-plus (for *Staphylococcus aureus* identification). Biochemical tests used for identification of Gram negatives to species level were triple sugar iron, indole, citrate, urea, Lysine decarboxylase (LDC), motility and malonat. *Klebsiella ozaenae* were identified from *Klebsiella pneumoniae* using malonat biochemical test. *Pseudomonas aeruginosa* were differentiated from other *Pseudomonas* species using *Pseudomonas Aeruginosa* Screen 80 tablet (Rosco, DK- 2630). For all isolated pathogens antimicrobial susceptibility testing was carried out on Muller-Hinton agar using disk diffusion.

Drug susceptibility testing

The disk diffusion method was performed and after 16-18 hours of incubation at 37°C zone of inhibition was measured and interpreted as recommended by the Clinical and Laboratory Standards Institute (CLSI) [15]. Using a sterile wire loop, 3-5 pure colonies were picked from blood agar for Gram positives and MacConkey agar for Gram negatives then emulsified in nutrient broth. Standard inoculums adjusted to 0.5 McFarland standard using McFarland Densitometer was swabbed onto Muller-Hinton agar (dispensed on 100mm plate).

Accordingly detailed CLSI guidelines for each category of Gram positive and Gram negative bacteria, isolates were tested against amikacin (10 µg, Oxoid), amoxicillin (30 µg, BD), amoxicillin-clavulanic acid (30 µg, BD), ceftazidime (30 µg, BD), cefotaxime (30 µg, BD), ceftriaxone (30 µg, BD), chloramphenicol (30 µg, BD), clindamycin (2 µg, BD), gentamicin (10 µg, BD), TMP-SXT (1.25 µg+23.75 µg, BD), tetracycline (30 µg, BD), ciprofloxacin (5 µg, BD), penicillin (10 units, BBL), oxacillin (5 µg, BD) and erythromycin (15 µg, BD). Oxacillin susceptibility of *Staphylococcus aureus* and *Coagulase negative Staphylococci* was interpreted using 30 µg cefoxitin as a surrogate test. The zone of inhibition was measured to the nearest millimeter and all bacterial isolates

were classified as sensitive, intermediate and resistant according to the standardized table supplied by CLSI.

Quality Control

Standard Operating Procedures (SOP) were strictly followed verifying that media meet expiration date and quality control parameters per CLSI (15). Visual inspections of cracks in media or plastic petridishes, unequal fill, hemolysis, evidence of freezing, bubbles, and contamination was done. Quality control was performed to check the quality of medium. Each new lot was quality controlled before use by testing *Escherichia coli* ATCC 25922 and/or *Staphylococcus aureus* ATCC25923 standard control strains.

Statistical analysis and interpretation

The data was analyzed using SPSS version 20. The descriptive statistics (mean, percentages or frequency) was calculated. The bi-variant logistic regression analysis was used to see the relation between dependent variables and independent variables. Variables that showed a significant association were selected for further analysis using multiple logistic regression models with a p-value <0.05 considered statistically significant.

Data quality assurance

Socio-demographic characteristics of patients were collected using structured data collection sheets after getting informed consent. Postoperative wound swabs were collected in accordance with SOPs and brought to bacteriology laboratory within 30 minutes for bacteriological analysis. Culture results were recorded carefully before data entry and the data was double checked by a different person before analysis.

Ethics approval and consent to participate

The study was approved by “Department Research and Ethical Review Committee (DRERC)” of the Department of Medical Laboratory Science (MLS/483/15), School of Allied Health Sciences, College of Health Sciences, Addis Ababa University. Written permission letter was also obtained from the study site. The purpose and procedures of the study was explained to the study participants, participants’ parents or guardians within the study period. Those participants who gave informed consent and those children who gave assent and whose parents or guardians gave informed consent were selected and enrolled as the participants of the study. A patient result was communicated to the attending physicians.

Results

Socio-demographic characteristics

One hundred ninety seven (n=197) eligible study participants were investigated during the study period. Of these patients who developed postoperative wound infections 59.9% (n=118/197) of them were males and 40.1% (n=79/197) were females with males to females ratio of 1.49:1. The majority of patients (n=53/197; 27%) and (n=51/197; 25.9%) were between 1-10 and 21-30 years of age as shown in **Table 1** and the mean (std. deviation) ages of patients was 24.8(19.2) with age range of 0-85 years. Among

all study participants, 14.7% (n=29/197) were out patients while 85.3% (n=178/197) were inpatients. Socio-demographic characteristics of patients have shown in **Table 1**.

Prevalence of bacterial isolates from postoperative wounds

The overall prevalence of bacterial isolates from postoperative wound infections was 75.6% (n=149/197). Among all bacterial isolates, 62.4% (n=93/149) of the culture positives were from males and 37.6% (n=56/149) were from females. Gram positive and Gram negative isolates constitutes 45.2% (n=76/168) and 54.8% (n=92/168) respectively with Gram positives to Gram negatives ratio of 0.83:1. The frequent bacteria isolated from postoperative wound cultures were *Staphylococcus aureus* 33.3% (n=56/168). Other isolates were *Escherichia coli* 14.3% (n=24/168), *CONS* 11.3% (n=19/168), *Acinetobacter spp.* 10.1% (n=17/168), *Klebsiella pneumoniae* 8.9% (n=15/168, *Pseudomonas spp.* 5.3% (n=9/168) and *Pseudomonas aeruginosa* 4.8% (n=8/168) (**Figure 1**).

The spectrum of post-operative wound infections varied with the age of patients (**Table 1**). The highest proportion 28.2% (n=42/149) of post-operative wound infections were found in less than 10 years. However, there was no significant association between age of patients and culture results (OR=1.13, 95% CI=0.931-1.377, P = 0.212) (**Table 2**). In this study, 83.9%

Table 1 Socio-demographic characteristics of patients who developed postoperative wound infections at Tikur Anbessa Specialized Hospital from March to August 2015.

Variables		Bacterial culture results (n=197)		
		Positive n (%)	Negative n (%)	Total n (%)
Gender	Male	93(78.8)	25(38.8)	118(100)
	Female	56(70.9)	23(29.1)	79(100)
Age in Year	<10	42(79.3)	11(20.7)	53(100)
	Nov-20	29(82.9)	6(17.1)	35(100)
	21-30	36(70.6)	15(19.4)	51(100)
	31-40	14(77.8)	4(22.2)	18(100)
	41-50	14(73.7)	5(26.3)	19(100)
	>50	14(66.7)	7(23.3)	21(100)
Patient Type	Outpatient	24(82.8)	5(17.3)	29(100)
	Inpatient	125(74.4)	43(25.6)	168(100)
Wound status	Dirty	12(100)	0(0)	12(100)
	Contaminated	58(98.3)	1(1.7)	59(100)
	Clean contaminated	63(78.8)	17(21.2)	80(100)
	Clean	16(34.8)	30(65.2)	46(100)
Wards (inpatients)	Orthopedics	42(82.4)	9(17.6)	51(100)
	Intensive Care Unit	9(81.8)	2(18.2)	11(100)
	General surgery	37(75.5)	12(24.5)	49(100)
	Gynecology & obstetrics	13(68.4)	6(31.6)	19(100)
	Internal Medicine	4(57.1)	3(42.9)	7(100)
	Pediatrics	20(64.5)	11(35.5)	31(100)
Total		149(75.6)	48(24.4)	197(100)

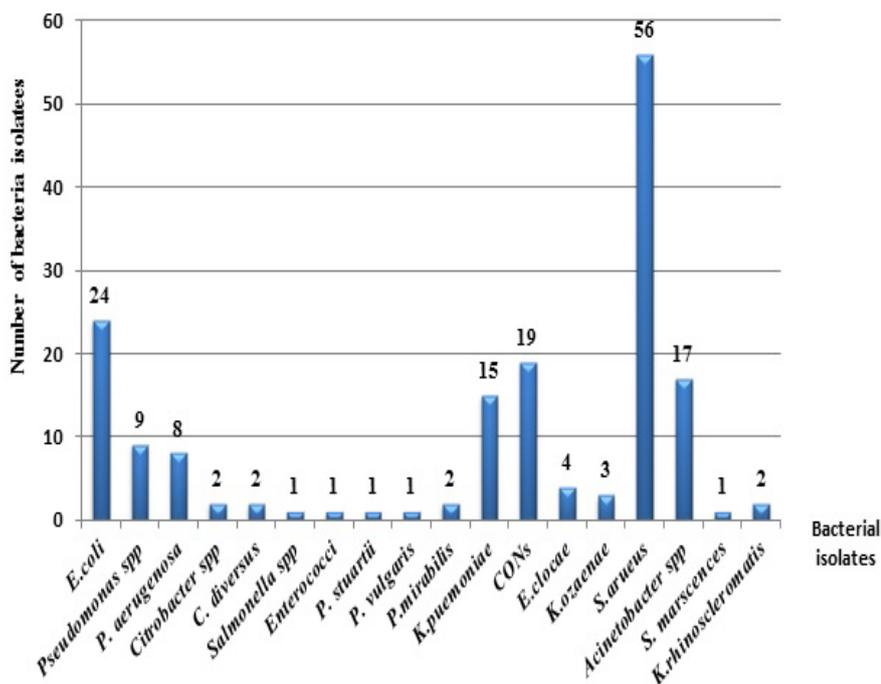


Figure 1 Frequency of bacterial isolates from postoperative wounds at Tikur Anbessa Specialized Hospital from March to August 2015.

Table 2 Association of variables with postoperative wound culture results at Tikur Anbessa Specialized Hospital from March to August 2015.

Variables	Culture results (n=197)		P-value	OR	95%CL	P-value	AOR	95% CI
	Positive n (%)	Negative n (%)						
Gender	Male	93(78.8)	25(38.8)	0.205	0.655	[0.340-1.262]		
	Female	56(70.9)	23(29.1)	1				
	Total	149(75.6)	48(24.4)					
Age in Year	<10	42(79.3)	11(20.7)	0.26	0.524	[0.170-1.612]		
	Nov-20	29(82.9)	6(17.1)	0.1711	0.414	[0.117-1.464]		
	21-30	36(70.6)	15(19.4)	0.743	0.833	[0.280-2.476]		
	31-40	14(77.8)	4(22.2)	0.445	0.571	[0.136-2.399]		
	41-50	14(73.7)	5(26.3)	0.629	0.714	[0.182-2.800]		
	<50	14(66.7)	7(23.3)	1				
	Total	149(75.6)	48(24.4)					
Patient Type	Outpatient	24(82.8)	5(17.3)	1				
	Inpatient	125(74.4)	43(25.6)	0.337	1.651	[0.593-4.597]		
	Total	149(75.6)	48(24.4)					
Wound status	Dirty	12(100)	0(0)	0.998	0	0		
	Contaminated	58(98.3)	1(1.7)	0	0.009	[0.001-0.073]	0	0.008 [0.001-0.063]
	Clean contaminated	63(78.8)	17(21.2)	0	0.144	[0.064-0.323]	0	0.133 [0.056-0.318]
	Clean	16(34.8)	30(65.2)	1			1	
	Total	149(75.6)	48(24.4)					

OR: Odds Ratio; CI: Confidence Interval; AOR: Adjusted Odds Ratio; NA: Not Applicable

(n=125/149) of bacteria were isolated from hospitalized patients while the remaining 16.1% (n=24/149) were from those who attended outpatient department; however there was no significant association between being out patient or inpatient on post-operative wound infection culture results (OR=1.653, 95%CI=0.593-4.597, P = 0.337). Among the different wards, the

highest proportion of bacteria 28.2% (n=42/149) were isolated from orthopedics followed by General surgery 24.8% (n=37/149), OPD 16.1% (n=24/149) and pediatrics 13.4% (n=20/149). However, there was no significant association between wards and culture results (OR=1.03, 95%CI=0.309-3.424, P = 0.936).

Among the different wound status of postoperative wound

infections clean contaminated wound class yields the highest culture positive results 42.3% (n=63/149) followed by contaminated 25.5% (n=38/149) and clean wounds 10.7% (n=16/149). However, there was no significant association between wound status of the patient and culture results (OR=0.109, 95% CI=0.43-0.274, P = 0.998). Dirty wounds yield 100% (n=12/12) positive culture results though it took the least proportion 6.1% (n=12/197) from all wound status. In our study, mono-microbial isolates were recovered from 88.6% (n=132/149) patients whereas 11.4% (n=17/149) had double microbial infections (Table 3). Highest double microbial infections 52.9% (n=9/17) were showed from orthopedics ward (Table 3). The most double isolated bacteria were *Staphylococcus aureus* and *Pseudomonas spp.* 3.5% (n=4/17) combination followed by *Staphylococcus aureus* and *Escherichia coli* 17.6% (n=3/17) (Table 4).

Antibiotic resistance patterns

Among the total bacterial isolates (n=168), multidrug resistance (MDR ≥ 2 different classes of drugs) was recorded in 65.5% (n=110/168) of bacterial isolates (Table 5). Antimicrobial resistance level for Gram positive isolates causing postoperative wound infections ranged from 0-100% with an MDR level of 55.3% (n=42/76). Among the Gram positive bacteria, the frequent isolate *Staphylococcus aureus* 73.7% (n=56/76) showed 44.6% (n=25/56)

an MDR level. It showed lowered resistance level for clindamycin (1.8%), chloramphenicol (7.1%) and oxacillin (10.7%) however it demonstrated high level of resistance to penicillin (80.4%) (Table 6). Coagulase negative *staphylococci* (CONS) showed an MDR level of 84.2% (n=16/19). It showed lower resistance level for sulphamethoxazole-trimethoprim (33.9%), tetracycline (28.6%) and erythromycin (16.1%) however it showed high level of resistance to penicillin (94.7%). It showed better susceptibility for ciprofloxacin (26.3%) and clindamycin (31.6%).

In the same manner to gram positive bacteria, the resistance patterns of Gram negative organisms causing postoperative wound infections were ranged from 0-100% and they showed 73.9% (n=68/92) of an MDR level. Almost all gram negative bacterial isolates showed greater than 90% level of resistance for amoxicillin and almost all isolates were sensitive for amikacin (>95%). Among the Gram negative bacteria, the frequent isolates, *Escherichia coli* 24.1% (n=24/92), showed 87.5% (n=21/24) of MDR level which demonstrated highest level of resistance to amoxicillin (90%). It showed lower resistance level for chloramphenicol (16.7%) and gentamicin (29.2%). Among the antibiotics tested, amoxicillin (93.5%), ceftriaxone (85.3%), penicillin (84.5%) and cefotaxime (82.7%) were least effective. Clindamycin (7.9%) for Gram positives and amikacin (1.1%) for Gram negative isolates were the most effective drugs.

Table 3 Rate of single and double infections from postoperative wounds among different wards at Tikur Anbessa Specialized Hospital from March to August 2015.

Wards	Culture results			
	Single infections	Double infections	No-Growth	Total
Orthopedics	33 (64.7%)	9 (17.6%)	9 (17.6%)	51 (100%)
ICU	6 (54.5%)	3 (27.3%)	2 (18.2%)	11 (100%)
General surgery	35 (71.4%)	2 (4.1%)	12 (24.5%)	49 (100%)
Gynecology & Obstetrics	13 (68.4%)	0 (0.0%)	6 (31.6%)	19 (100%)
Internal medicine	3 (42.9%)	1 (14.3%)	3 (42.9%)	7 (100%)
Pediatrics	19 (61.3%)	1 (3.2%)	11 (35.5%)	31 (100%)
Out patient	23 (79.3%)	1 (3.4%)	5 (17.2%)	29 (100.0%)
Total	132 (67.0%)	17 (8.6%)	48 (24.4%)	197 (100.0%)

Table 4 Frequency of double infections from patients who develop postoperative wound infections at Tikur Anbessa Specialized Hospital from March to August 2015.

Double infections		
Infections	Frequency	Percent %
<i>K. pneumoniae</i> and <i>Escherichia coli</i>	1	5.9
<i>Acinetobacter spp.</i> and <i>Escherichia coli</i>	1	5.9
<i>Acinetobacter spp.</i> and <i>Citrobacter diversus</i>	1	5.9
<i>Staphylococci aureus</i> and <i>Pseudomonas spp.</i>	4	23.5
<i>Staphylococci aureus</i> and <i>Acinetobacter spp.</i>	1	5.9
<i>Staphylococci aureus</i> and <i>Escherichia coli</i>	3	17.6
<i>Staphylococci aureus</i> and <i>K. pneumoniae</i>	2	11.8
<i>P. mirabilis</i> and <i>K. rihinoscleris</i>	1	5.9
<i>Escherichia coli</i> and <i>Citrobacter spp.</i>	1	5.9
<i>Escherichia coli</i> and <i>Providencia stuartii</i>	1	5.9
<i>Citrobacter spp.</i> and <i>K. rihinoscleris</i>	1	5.9
Total	17	100

Table 5 Multidrug resistance pattern of bacteria isolated from postoperative wounds at Tikur Anbessa Specialized Hospital from March to August 2015.

Organism isolated	Antimicrobial resistance level in no.(%)of bacterial isolates									
	R2	R3	R4	R5	R6	R7	R8	R9	R10	Total
Gram Positive										
<i>Staphylococci aureus</i> (n=56)	8(14.3)	6(10.7)	3(5.4)	4(7.1)	1(1.8)	1(1.8)	0(0)	2(3.6)	0(0)	25(44.6)
<i>CONS</i> (n=19)	1(5.3)	2(10.5)	2(10.5)	0(0)	1(5.3)	1(5.3)	4(21.1)	4(21.1)	1(5.3)	16(84.2)
<i>Enterococci</i> (n=1)	0	1(100)	0(0)	-	-	-	-	-	-	1
Total (n=76)	9(11.8)	9(11.8)	5(6.6)	4(5.3)	2(2.6)	2(2.6)	4(5.3)	6(7.9)	1(1.3)	42(55.3)
Gram Negative										
<i>K. ozaenae</i> (n=3)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(33.3)	1(33.3)	1(33.3)	3(100)
<i>P. mirabilis</i> (n=2)	0(0)	0(0)	0(0)	0(0)	0(0)	1(50)	1(50)	0(0)	0(0)	
<i>K. rhinoscleris</i> (n=2)	0(0)	0(0)	0(0)	0(0)	0(0)	1(50)	1(50)	0(0)	0(0)	
<i>S.marscense</i> (n=1)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(100)	0(0)	0(0)	
<i>Citrobacter spp.</i> (n=2)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(50)	0(0)	0(0)	
<i>Citrobacter diversus</i> (n=2)	0(0)	0(0)	0(0)	0(0)	1(50)	0(0)	0(0)	1(50)		2(100)
<i>Providencia stuartii</i> (n=1)	0(0)	0(0)	0(0)	1(100)	0(0)	0(0)	0(0)	0(0)	0(0)	1(100)
<i>Salmonella spp.</i> (n=1)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(100)	0(0)	0(0)	1(100)
<i>K. pneumoniae</i> (n=15)	0(0)	0(0)	2(13.3)	4(26.7)	0(0)	2(13.3)	4(26.7)	2(13.3)	0(0)	14(93.3)
<i>P. aerogens</i> (n=8)	1(12.5)	1(12.5)	0(0)	0(0)	-	-	-	-		2(25)
<i>Pseudomonas spp.</i> (n=9)	3(33.3)	3(33.3)	0(0)	0(0)	0(0)	0(0)	-	-		6(66.7)
<i>Acinetobacter spp.</i> (n=17)	1(5.9)	1(5.9)	1(5.9)	4(23.5)	0(0)	0(0)	-	-		7(41.2)
<i>Escherichia coli</i> (n=24)	1(4.2)	2(8.3)	2(8.3)	3(12.5)	2(8.3)	2(8.3)	6(25)	2(8.3)	1(4.2)	21(87.5)
<i>P. vulgaris</i> (n=1)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	1(100)	0(0)	0(0)	1(100)
<i>E. cloacae</i> (n=4)	1(25)	1(25)	1(25)	0(0)	0(0)	0(0)	0(0)	1(25)	0(0)	4(100)
Total (n=92)	7(7.6)	8(8.7)	6(6.5)	12(13.0)	3(3.3)	6(6.5)	17(18.5)	7(7.6)	2(2.2)	68(73.9)

CNS: Coagulase negative staphylococci; R2, R3, R4, R5, R6, R7, R8, R9, R 10-Resistant to two, three, four, five, six, seven, eight, nine and ten antimicrobials respectively.

Discussion

Prevalence of bacterial isolates among postoperative wound infections

The overall prevalence of bacterial isolates from postoperative wounds with clinical suspicion of wound infections was 75.6% (n=149/197) which was in line with previous studies done in Ethiopia which showed 75% and 71.1% of prevalence [16,17]. On the contrary, lower rates of isolation (11.4%, 10.9%) were reported from other localities of Ethiopia [18,19]. However, our finding was relatively lower than other studies done in Ethiopia (92%, 87.3%) [20,21] and India (96%) [22]. The possible explanation for the difference could be varying bacterial etiology and infection prevention practices in diverse geographical settings and at different sampling times [17]. The effect of antimicrobials used for surgical prophylaxis, that the infection is already resolving, antiseptics used for cleaning the wounds, bacteria fail to grow due to their fastidious nature, samples only containing already dead bacteria or wound infections caused by unidentifiable pathogens by the methods used could be main factors for the absence of bacterial growth in samples collected from surgical wounds with clinical signs of infections [19,20].

The incidence of wound infection was higher in males 62.4% (n=93/149) than in females 37.6% (n=56/149). However, there was no significant association between sex of patient and culture results (OR=0.65, 95% CI=0.340-1.262, P=0.205). This showed

agreement with studies done in different parts of Ethiopia and other countries [17,21,23]. This might be explained by the fact that traditionally, in Ethiopia, males are predominantly involved in occupations such as farming, construction works, transportation and industry works where they are at higher risk of trauma [21] and exposed to the outside environment than females [24].

The spectrum of post-operative wound infections varied with the age of patients (Table 5) where 28.2% (n=42/149) of post-operative wound infections occurred in less than 10 years in contrary to other study [24]. However, there was no statistical significant association between age of patient and culture results (OR=1.13, 95% CI=0.931-1.377, P=0.212). It showed disagreement with a study done in Ethiopia which reported that age of the patient showed statically significant association with culture results [25]. In this study 83.9% (n=125/149) of bacteria were isolated from hospitalized patients while the remaining 16.1% (n=24/149) were from those who attended outpatient departments; however there was no significant association between being out patient or inpatient on post-operative wound infection culture results (OR=1.653, 95%CI=0.593-4.597, P = 0.337). It was a similar finding with a previous study done in Ethiopia [17].

Among the different wards, the highest proportion of bacteria 28.2% (n=42/149) were isolated from orthopedics which showed similarity with a previous study [16]. However, there was no statistical significant association between different wards and

Table 6 Antimicrobial resistance levels of bacterial isolates from postoperative wounds at Tikur Anbessa Specialized Hospital from March to August 2015.

Organism isolated	Antimicrobial resistance level in no.(%) of bacterial isolates from postoperative wounds															
	AML	AMC	SXT	PEN	CLD	VA	OX	ERT	GE	CTX	CAZ	C	TE	AN	CIP	CRO
<i>S. aureus</i> (n=56)	NT	9(16.1)	19(33.9)	45(80.4)	1(1.8)	NA	6(10.7)	9(16.1)	7(12.5)	NA	NA	4(7.1)	16(28.6)	NA	7(12.5)	NT
<i>K. ozaenae</i> (n=3)	3(100)	3(100)	3(100)	NA	NA	NA	NA	NA	3(100)	3(100)	3(100)	2(66.7)	3(100)	0(0)	2(66.7)	3(100)
<i>P. mirabilis</i> (n=2)	2(100)	2(100)	2(100)	NA	NA	NA	NA	NA	2(100)	1(50)	0(0)	2(100)	2(100)	0(0)	0(0)	1(50)
<i>K. rhinoscleris</i> (n=2)	2(100)	2(100)	2(100)	NA	NA	NA	NA	NA	0(0)	2(100)	2(100)	1(50)	2(100)	0(0)	2(100)	2(100)
<i>S. marscensense</i> (n=1)	1(100)	1(100)	1(100)	NA	NA	NA	NA	NA	1(100)	1(100)	1(100)	1(100)	1(100)	0(0)	0(0)	1(100)
<i>Citrobacter spp.</i> (n=2)	1(50)	1(50)	1(50)	NA	NA	NA	NA	NA	1(50)	1(50)	1(50)	0(100)	1(50)	0(0)	1(50)	1(50)
<i>C. diversus</i> (n=2)	2(100)	2(100)	1(50)	NA	NA	NA	NA	NA	2(100)	2(100)	2(100)	1(50)	1(50)	0(0)	2(100)	2(100)
<i>Enterococci</i> (n=1)	NA	NA	NA	1(100)	NA	0(0)	NA	NA	NA	NA	NA	1(00)	1(100)	NA	NA	NA
<i>P. stuartii</i> (n=1)	1(100)	1(100)	1(100)	NA	NA	NA	NA	NA	0(0)	1(100)	0(0)	1(100)	0(0)	0(0)	0(0)	1(100)
<i>Salmonella spp.</i> (n=1)	1(100)	1(100)	1(100)	NA	NA	NA	NA	NA	1(100)	1(100)	1(100)	1(100)	1(100)	0(0)	0(0)	1(100)
<i>K. pneumoniae</i> (n=15)	14(93.3)	12(80)	13(86.7)	NA	NA	NA	NA	NA	9(100)	13(86.7)	9(60)	7(46.7)	9(60)	0(0)	4(26.7)	13(86.70)
<i>P. aerogens</i> (n=8)	8(100)	7(87.5)	7(87.5)	NA	NA	NA	NA	NA	2(25.0)	NA	1(12.5)	3(37.5)	8(100)	1(12.5)	1(12.7)	NA
<i>Pseudomonas spp.</i> (n=9)	9(100)	8(88.9)	8(88.9)	NA	NA	NA	NA	NA	7(77.8)	NA	6(66.7)	7(77.8)	9(100)	0(0)	3(33.3)	NA
<i>Acinetobacter spp.</i> (n=17)	17(100)	15(88.2)	15(88.2)	NA	NA	NA	NA	NA	12(70.6)	17(100)	16(94.1)	13(76.5)	14(82.4)	0(0)	12(70.6)	17(100)
<i>Escherichia coli</i> (n=24)	20(83.3)	14(58.3)	15(62.5)	NA	NA	NA	NA	NA	7(29.2)	15(62.5)	14(58.3)	4(16.7)	15(62.5)	0(0)	9(37.5)	17(70.8)
CONS (n=19)	NT	15(78.9)	14(73.7)	18(94.7)	6(31.6)	NA	10(52.6)	11(57.9)	12(63.2)	NA	NA	9(47.4)	12(63.2)	NA	5(26.3)	NA
<i>P. vulgaris</i> (n=1)	1(100)	1(100)	1(0)	NA	NA	NA	NA	NA	1(100)	1(100)	1(100)	0(0)	1(100)	0(0)	1(100)	1(100)
<i>E. cloacae</i> (n=4)	4(100)	2(50)	1(25)	NA	NA	NA	NA	NA	1(25)	4(100)	4(100)	1(25)	1(25)	0(0)	1(25)	4(100)
Total (n=168)	86(93.5)	96(57.5)	105(62.9)	64(84.2)	7(9.3)	0(0)	16(21.3)	20(26.7)	68(40.7)	62(82.7)	49(53.3)	57(33.9)	97(57.7)	1(1.1)	50(29.8)	64(85.3)

CNS: Coagulase Negative Staphylococci, NA: Not Applicable; NT: Not Tested, AML: Amoxicillin, AmC: Amoxicillin-Clavulanic Acid, AN: Amikacin, SXT: Sulphamethoxazole-Trimethoprim; C: Chloramphenicol, CTX: Cefotaxime, CAZ: Ceftazidime, CIP: Ciprofloxacin; OX: Oxacillin; ERY: Erythromycin; CLD: Clindamycin; VA: Vancomycin; PEN: Penicillin

culture results (OR=1.03, 95%CI=0.309-3.424, P=0.936). While General surgery 24.8% (n=37/149) and OPD 16.1% (n=24/149) took the second and third rank of bacteria proportion, pediatrics 13.4% (n=20/149), gynecology and obstetrics 8.7% (n=13/149) and internal medicine 2.7% (n=4/149) followed them. It also showed similarity with other study done in Nepal [24]. This could be due to that in orthopedic wards since patients require longer hospitalization time to recover from their bone cases, they become prone for post-operative wound infections [24].

In our study, among the different classes of postoperative wound infections clean contaminated wound showed highest culture positive results 42.3% (n=63/149). Multivariate regression analysis showed a statistical significant association between wound status of the patient and culture results (OR=0.144, 95% CI=0.0640-0.323, P=0.00). It showed agreement with other study done in Ethiopia [17]. This high rate of infections could be probably because of profound influence of endogenous contamination during the time of operation [17]. Contaminated wounds 58.9% (n=38/149) and clean wounds 10.7% (n=16/149) took the second and third culture positive ranks following to clean contaminated wounds. In the case of dirty wounds, it yields 100% (n=12/12) positive culture results though it took the least proportion from all wound status [16].

Double bacterial infections, among culture positive post-surgical wound infected patients, was seen in 11.4% (n=17/149) of patients. The rate of double pathogens in the current study was comparable with a previous study done in South West Ethiopia [21] however it was a lower finding compared to another study conducted in Southern Ethiopia (20.1%) [17]. The leading double infection was caused by *Staphylococcus aureus* in combination with *Pseudomonas* species was also found in another study from Ethiopia [25].

In our study, Gram negatives 54.8% (n=92/168) were isolated more commonly than Gram positive isolates 45.2% (n=76/168) which showed similarity with other studies [24,26]. The most frequent bacteria isolated in this study were *Staphylococcus aureus* 33.3% (n=56/168). It showed similarity with other studies done throughout the world [17,26,27]. The predominance (33.3%) of *Staphylococcus aureus* infection seen in this study could be associated with endogenous source as the organism colonizes the skin that lead access to deep sites during surgery [16]. This organism may also be transmitted from the environment, surgical instruments or contaminated hands of the health professionals [19]. A study done in Nepal found that *Pseudomonas* spp. were the most frequently isolated bacteria from postoperative wound infections [24]. The possible reason for these varied findings with our study could be due to populations; different surgical procedures as well as timing of specimen collections [24].

The second most common bacteria isolated in this study was *Escherichia coli* 14.3% (n=24/168) that showed similarity with a study done in Nepal (17.5%) [24]. Endogenous contamination from bowel flora of patients could explain this finding [19]. Other bacterial species found in this study were Coagulase negative *Staphylococci* 11.3% (n=19/168), *Acinetobacter* spp. 10.1% (n=17/168), *Klebsiella pneumoniae* 8.9% (n=15/168,

Pseudomonas spp. 5.3% (n=9/168) and *Pseudomonas aeruginosa* 4.8% (n=8/168).

Antimicrobial resistance patterns of bacterial isolates

The overall multidrug resistance level of all bacterial isolates was 65.5% (n=110/168). This finding was in line with the finding of a study done in other part of Ethiopia where the MDR level was 65.2% [25]. On the other hand, our finding was a lowered finding as compared to what has been recorded in South West Ethiopia which was 85.2% [21]. In our study, Gram positive and Gram negative bacteria showed 55.3% (n=42/76) and 73.9% (n=68/92) MDR level respectively. This high rate of antibiotic resistance might reflect inappropriate use of antibiotic, lack of laboratory diagnostic tests for appropriate antibiotic selection, unavailability of guideline for the selection of antibiotics, unskilled practitioners, expired antibiotics, self-medication, counterfeit drugs, or inadequate hospital control measures [28].

The most frequently isolated pathogens, *Staphylococcus aureus*, showed 44.6% (n=25/56) of an MDR level which disagreed with previous studies done in Ethiopia [19,20] where *Staphylococcus aureus* showed 100% MDR level. The difference could be due to the difference in prescribing this antibiotic for the treatment hospital to hospital [16]. It demonstrated high level of resistance to penicillin (80.4%) which was consistent with a study done in other part of Ethiopia [21]. It showed lower resistance level for sulphamethoxazole-trimethoprim (33.9%), tetracycline (28.6%), erythromycin (16.1%), clindamycin (1.8%), chloramphenicol (7.1%) and oxacillin (10.7%) as compared to other tested drugs.

Among the Gram negative bacteria, the predominant isolates, *Escherichia coli* 24.1% (n=24/92), showed 87.5% (n=21/24) of multidrug resistance level, It demonstrated high level of resistance to amoxicillin (90%) and a lowered resistance level for chloramphenicol (16.7%) and gentamicin (29.2%). This finding showed disagreement with a study done in Ethiopia where *Escherichia coli* isolates showed highest resistant level to ampicillin (96.6%), tetracycline (79%), chloramphenicol (65.5%), ceftriaxone (62%), sulphamethoxazole-trimethoprim (55%) and gentamicin (51.7%). The difference could be due to the difference in prescribing these antibiotics for the treatment of the bacteria from hospital to hospital [16]. In our study, amoxicillin (93.5%), ceftriaxone (85.3%), penicillin (84.2%) and cefotaxime (82.7%) were least effective drugs.

Conclusion

The overall 75.6% (n=149/197) prevalence of bacterial isolates from postoperative wound infections was high and the most frequent isolated bacteria were *Staphylococcus aureus* and *Escherichia coli*. Although complete eradication of postoperative wound infections is not possible, proper precautions should be taken to minimize the occurrence by strictly adhering aseptic surgical procedures and proper management of wounds. The choice of drugs for the treatment of bacterial isolates from postoperative wound infections was quit narrow especially for bacterial strains which were resistant to most classes of

antibiotics which have been used previously. To prevent further emergence and spread of MDR bacterial pathogens, rational use of antibiotics and regular monitoring of antimicrobial resistance patterns are essential and mandatory.

Competing of interest

The Authors declared that no competing of interests.

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Acknowledgments

We greatly appreciate Addis Ababa University for supporting this study. We are also grateful to the Department of Clinical Laboratory Sciences to give ethical clearance for this study. Our deep gratitude goes to those study participants and also parents and guardians who gave us their willingness to participate in this study by giving written consent form.