

Antibiogram Profiles of Bacteria Isolated from Different Body Site Infections Among Patients Admitted to GAMBY Teaching General Hospital, Northwest Ethiopia

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Background: Infections with multi-drug resistant (MDR) bacteria are serious threats to many low-income countries associated with overuse and misuse of antibiotics. This study determined the antibiogram profiles of bacteria isolated from different body site infections among patients admitted to GAMBY Teaching General Hospital, Bahir Dar, Northwest Ethiopia.

Methods: A hospital-based cross-sectional study was done between November 2015 and May 2018. Various clinical specimens were sampled from patients and analyzed for aerobic bacterial isolation and Kirby–Bauer disk diffusion susceptibility testing. Chi-square test was calculated to see association among variables, and P -value <0.05 was taken as a cutoff value for statistical significance.

Results: From the 716 clinical specimens processed, 134 (18.7%) were culture-positive for aerobic bacterial pathogens. Culture-confirmed positivity was higher in ear discharge (27.3%) and urine (26.3%) samples. The prevalence of infection was significantly highest among females ($P = 0.001$). *Escherichia coli* 63 (47.4%) and 10 (7.4%) of *Klebsiella* spp. from Gram-negative bacteria were the predominant bacterial isolates, while *Staphylococcus saprophyticus* 17 (12.6%) and *S. aureus* 14 (10.4%) were from Gram-positive bacteria. Overall, 61.8% of the isolates were found to be MDR. The proportion of MDR among *Klebsiella* spp., *S. aureus* and *E. coli* isolates was 90.9%, 60.9% and 50%, respectively. Gram-positive bacteria demonstrated 20%, 48.6% and 100% of resistance against norfloxacin, ciprofloxacin and clindamycin, respectively. Gram-negative bacteria also revealed from 20% resistance for the antibiotic nitrofurantoin and 100% of resistance for ampicillin and penicillin.

Conclusion: Infections with bacterial isolates resistant to the majority of antibiotics are a major issue in the study area. Most of the identified bacteria were resistant to the routinely used antibiotics, and MDR isolates are alarmingly high. Therefore, clinicians should practice rational choice of antibiotics and treatment should be guided by antimicrobial susceptibility testing.

Keywords: bacterial isolates, antibiogram, clinical specimen, GAMBY, Bahir Dar, Ethiopia

Introduction

Antimicrobial resistance (AMR) reduces the treatment options and is becoming a global issue. Hence, it negatively impacts the health of a community, healthcare costs and gross domestic products. Globally, the burdens of infections with resistant microbes are increasing.¹

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Based on the 2014 World Health Organization (WHO) report, AMR is one of the major global threats in the spectrum of infectious diseases. Worldwide evidence has revealed the declining of the effectiveness of antibiotics in the stock and the rising of bacterial resistance to all first-line and last-resort antibiotics. Thus, the impact of antibiotic resistance is clinical, economic and societal.^{2,3} Hospital patients acquire the infection during their admission and inpatient ward and intensive care unit stay.⁴

The impact of AMR is significant; as new bacterial strains develop, it will also have an effect by reducing consumer income, employment savings, healthcare delivery and gross domestic products.⁵

Worldwide, infections with drug-resistant pathogens significantly affect not only the public health but also the economic stability of societies. At least 25% of the 60 million year-based deaths in the world is due to microbial diseases. Despite significant advances in infection control practices, clinical infections with drug-resistant pathogens remain significant causes of morbidity and mortality among hospitalized patients and in the community settings, affecting developed countries, middle-income countries and sub-Saharan Africa.^{6,7}

Escherichia coli (*E. coli*), *Staphylococcus aureus* (*S. aureus*), *Klebsiella pneumoniae* (*K. pneumoniae*), *Pseudomonas aeruginosa* (*P. aeruginosa*), *Streptococcus pneumoniae* (*S. pneumoniae*) and *Salmonella* spp. are the commonly isolated organisms from infections in the clinical and community settings.⁸ They are also the current most serious antibiotic-resistant organisms.⁹

As a result of the high burden of infections, irrational use of antibiotics, over-the-counter availability of drugs and limitations in the availability of antimicrobial susceptibility testing, drug resistance is a major challenge in low-income countries.^{10–12}

Early detection and response at local and regional levels is one of the major strategies recommended by Ethiopian Ministry of Health and US CDC to mitigate the spreading of AMR. However, there is scarcity of comprehensive data on the antibiogram of bacterial pathogens isolated from different body sites of infections from adequate study participants in sub-Saharan Africa including Ethiopia. Furthermore, there are no documented data on the bacterial isolates and antibiotic resistance profiles in GAMBY Teaching General Hospital. Patient flow in GAMBY Teaching General Hospital is increasing from year to year, and culture and drug susceptibility tests were started in 2015. Therefore, this study isolated

aerobic bacteria and determined their antibiotic resistance profiles from different body site infections that occurred among patients at GAMBY Teaching General Hospital, Northwest Ethiopia.

Materials and Methods

Study Design, Period and Setting

A prospective cross-sectional study was done between November 2015 and May 2018 in GAMBY Teaching General Hospital, located in Bahir Dar, Northwest Ethiopia. The hospital delivers medical services to all regular and referral cases from the region. It is equipped with a laboratory for clinical chemistry, microbiology and pathology test analysis. It has an average flow of 300 daily new patients each year.^{13,14} All bacterial infection presumptive patients admitted in GAMBY Teaching General Hospital were the study population.

Variables of the Study

Culture-confirmed bacterial infection was the dependent variable, while sex, age, residence, type of clinical samples and antibiotic use were the independent variables.

Specimen Collection and Processing

Clinical specimens such as swab from wound, urine, ear discharge, blood, stool, urethral or cervical discharge, nasal or throat swab, semen and CSF were sampled following standard operating procedures. Depending on the source of specimen, each sample was plated onto blood agar, mannitol salt agar, MacConkey agar, xylose lysine deoxycholate agar, chocolate agar and Thayer–Martin agar (Oxoid, UK) and incubated aerobically for 24 hrs at 37°C. Bacterial isolates were identified by standard phenotypic microbiological methods.^{15,16}

Antimicrobial Susceptibility Testing

Susceptibility of bacterial isolates to different antibiotics was analyzed by Kirby–Bauer disk diffusion susceptibility testing on Muller Hinton Agar (Oxoid, UK). All the identified bacterial isolates were checked for susceptibility to penicillin (10 µg), oxacillin (1 µg), amoxicillin (10 µg), tetracycline (30 µg), nitrofurantoin (300 µg), erythromycin (15 µg), chloramphenicol (30 µg), gentamicin (10 µg), ciprofloxacin (5 µg), cotrimoxazole (25 µg), vancomycin (30 µg), norfloxacin (10 µg), clindamycin (2 µg) and cefotaxime (30 µg) (Oxoid, UK).

The choice of those antibiotic agents is based on the commonly available drugs and drugs which are frequently prescribed by physicians following the suggestion by

Magiorakos et al.²³ Resistance data were interpreted according to zone sizes from the Clinical and Laboratory Standards Institute (CLSI) guideline.^{17–19}

Quality Control

The created data were checked for completeness, clarity, accuracy and consistency before data entry. Standard bacteriological procedures were followed to generate correct laboratory test results. American Type Culture Collection (ATCC) standard reference strains (*E. coli* ATCC-25922 and *S. aureus* ATCC-25923) were used to verify the performance of the culture media.¹⁹

Data Analysis

Data were entered and cleaned in EPI-info version 7 and analyzed using IBM SPSS statistics for Windows, version 23 (IBM Corp, Armonk, NY, USA). Basic descriptive statistics such as frequency distribution were calculated. Chi-square tests were computed to see potential associations between variables. *P*-value <0.05 was considered a statistically significant association.

Ethical Approval and Consent to Participate

This study was conducted in accordance with the Declaration of Helsinki. Ethical approval was obtained from GAMBY Medical and Business College Research Review Committee. Written informed consent was taken from each respondent and from caregivers for children

before they were requested to give data and sample. All information obtained from this study was kept confidential and utilized only for the study. Positive findings were reported to the attending physicians for the proper management of patients.

Results

Socio-Demographic Characteristics

A total of 716 patient specimens were collected and processed from nine body sites. Of this, urine samples numbered 414 (57.8%), blood 71 (9.9%), stool 62 (8.7%), vaginal and cervical discharge 47 (6.6%), cerebrospinal fluids 35 (5%), pus discharge 24 (3.4%), urethral discharge 18 (2.5%), semen 15 (2.1%), eye discharge 13 (1.7%), ear discharge 11 (1.5%) and other body fluids 6 (0.8%). Among the total samples, 403 (56.4%) were taken from female patients. The mean \pm SD age of the patients was 30.5 \pm 8.6 years. Two hundred forty-six (34.4%) patients were in the age group of 15–29 years. The majority, 398 (55.6%), of patients were urban residents (Table 1).

Bacterial Profile

Overall, 134 (18.7%) of the specimens collected were culture-positive and 144 bacteria were isolated. From the total 134 positive patients, the proportion of bacterial isolates was significantly higher in females, 102 (76.1%), than males, 32 (23.9%) (*P*=0.03), Table 1.

Of the 144 bacteria isolated, 134 (93.0%) were single isolates while 10 (6.9%) were mixed isolates. *E. coli*,

Table 1 Demographic Characteristic and Culture-Positive Status of Patients at GAMBY Teaching General Hospital

Variables	Category	Total N (%)	Culture Result		Isolates Gram-Reaction		P-value
			Negative N (%)	Positive N (%)	Gram-Negative N (%)	Gram-Positive N (%)	
Sex	Female	403 (56.3)	301 (74.6)	102 (76.1)	60 (58.8)	42 (41.2)	0.0001
	Male	313 (43.7)	281 (89.7)	32 (23.9)	24 (75)	8 (25)	
Age (years)	0–14	125 (17.5)	110 (88)	15 (12)	10 (66.6)	5 (33.3)	0.03
	15–29	246 (34.4)	194 (78.9)	52 (21.1)	34 (65.4)	18 (34.6)	
	30–44	217 (30.3)	172 (81.1)	45 (18.9)	25 (55.6)	20 (44.4)	
	45–59	83 (11.6)	71 (85.5)	12 (14.5)	10(83.3)	4 (16.7)	
	\geq 60	45 (6.3)	37 (82.2)	8 (17.8)	5 (62.5)	3 (37.5)	
Residence	Urban	398 (55.6)	322 (80.9)	76 (19.1)	49 (64.5)	27 (35.5)	0.77
	Rural	318 (44.4)	260 (81.4)	58 (18.6)	35 (60.3)	23 (39.6)	
Antibiotic use	No	121 (16.9)	105 (86.8)	16 (13.2)	11 (68.8)	5 (31.2)	0.08
	Yes	595 (83.1)	477 (80.2)	118 (19.8)	73 (61.8)	45 (38.2)	
Total		716 (100)	582 (81.3)	134 (18.7)	84 (62.7)	50 (37.3)	

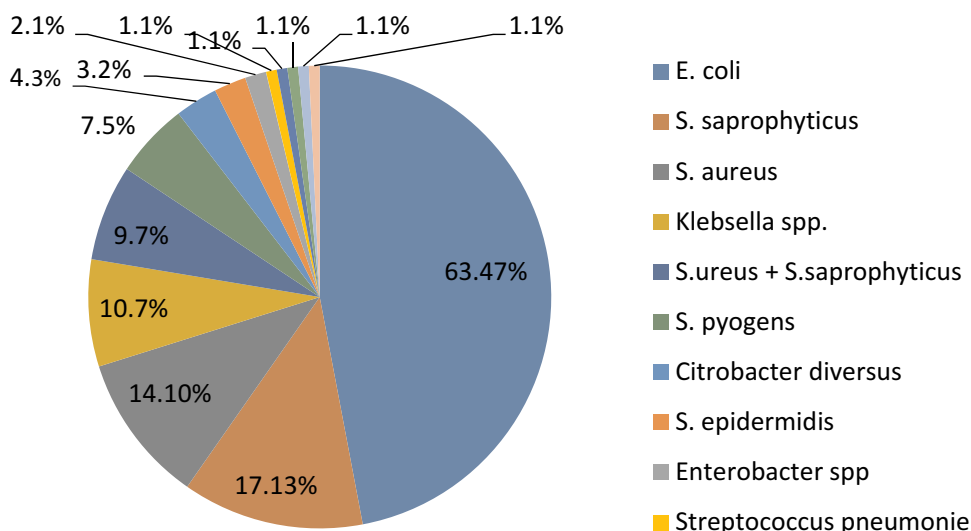


Figure 1 Bacteria isolated from patients at GAMBY Teaching General Hospital (November 2015–2018).

S. saprophyticus and *S. aureus* were the most frequently isolated bacteria from the clinical specimens (Figure 1).

Antimicrobial Resistance Profiles of Bacterial Isolates

Gram-Positive Bacteria

Five Gram-positive bacteria were isolated from clinical specimens. Overall, 133 (60.2%) Gram-positive bacteria were resistant to the tested antibiotics. Resistance rates of 75.6%, 78.4%, 75% and 62.5% were recorded for cotrimoxazole, tetracycline, erythromycin and gentamicin, respectively. In case of *S. aureus* isolates, resistance rates of 85.7% for tetracycline, 80% for each of erythromycin and

ciprofloxacin and an overall rate of 67% were recorded. *S. pyogenes* isolates showed an overall resistance rate of 79%. Gram-negative bacteria and Gram-positive bacteria accounted for 82 (56.9%) and 62 (43.1%) of the isolates, respectively, Table 2.

Gram-Negative Bacteria

Generally, 286 (54.2%) Gram-negative bacteria were resistant to the tested antibiotics. Resistance rates of 100%, 96.8%, 92.2%, 80.8% and 87.3% were recorded against ampicillin, amoxicillin, amoxicillin+clavulanic acid, cotrimoxazole and tetracycline, respectively.

E. coli isolates showed an overall resistance rate of 50%. These isolates exhibited 100% resistance to both amoxicillin

Table 2 Antimicrobial Resistance Profiles of Gram-Positive Bacterium Species Among Patients at GAMBY Teaching General Hospital

Antimicrobials	<i>S. saprophyticus</i>		<i>S. aureus</i>		<i>S. pneumoniae</i>		<i>S. pyogenes</i>		<i>S. epidermidis</i>	
	# T	R	# T	R	# T	R	# T	R	# T	R
Cefoxitin	1	0	4	2	1	1	1	1	0	0
Clindamycin	1	1	1	1	0	0	1	1	1	1
Ciprofloxacin	22	8	10	8	1	0	2	1	2	1
Chloramphenicol	0	0	3	2	0	0	0	0	0	0
Cotrimoxazole	19	17	21	13	0	0	3	3	2	1
Erythromycin	16	12	10	8	1	0	4	3	1	1
Gentamycin	3	2	3	2	2	1	0	0	0	0
Nitrofurantoin	16	0	4	0	0	0	0	0	0	0
Norfloxacin	6	1	4	1	0	0	0	0	0	0
Penicillin	1	1	5	4	1	0	0	0	1	1
Tetracycline	17	13	14	12	1	0	3	2	2	2
Oxacillin	3	1	5	3	1	0	0	0	1	0

Abbreviations: #T, number of isolates tested against each antimicrobial agent; R, isolates resistance to antimicrobial agents.

Table 3 Antimicrobial Resistance Profiles of Gram-Negative Bacterium Species at GAMBY Teaching General Hospital

Antimicrobials	<i>E. coli</i>		<i>Klebsiella</i> spp.		<i>Citrobacter diversus</i>		<i>Salmonella</i> spp.		<i>Proteus</i> spp.		<i>N. gonorrhoeae</i>	
	# T	R	# T	R	# T	R	# T	R	# T	R	# T	R
Amoxicillin	23	23	4	3	2	2	1	1	1	1	0	0
Ampicillin	22	22	4	4	2	2	1	1	0	0	0	0
Amoxiclav	38	36	8	7	4	3	1	1	0	0	0	0
Ceftriaxone	3	2	2	1	1	1	1	0	1	0	0	0
Ceftazidime	30	11	6	5	0	0	0	0	1	1	0	0
Cefepime	3	3	1	1		0	0	0	0	0	0	0
Cefixime	4	3	0	0	0	0	0	0	0	0	1	0
Clindamycin	2	0	1	0	0	0	0	0	0	0	0	0
Ciprofloxacin	61	15	10	5	5	1	1	1	1	1	1	1
Chloramphen	2	1	1	0	0	0	1	1	0	0	0	0
Cotrimoxazol	51	41	10	8	5	3	1	1	1	1	1	1
Erythromycin	1	1	0	1	0	0	1	1	0	0	0	0
Gentamycin	18	2	3	2	2	2	0	0	1	1	0	0
Nitrofurantoin	41	1	6	3	3	0	1	0	0	0	0	0
Norfloxacin	48	1	8	4	4	0	1	0	0	0	0	0
Penicillin	0	0	1	1	0	0	1	1	0	0	1	1
Tetracycline	48	40	7	7	5	5	1	1	1	1	1	1

Abbreviations: # T, number of isolates tested against each antimicrobial agent; R, isolates resistance to antimicrobial agents.

and ampicillin, 94.7% to amoxicillin+clavulanic acid, 83.3% to tetracycline and 80.4% to cotrimoxazole. *Klebsiella* spp. showed an overall resistance of 72.2%. *Salmonella* spp. and *N. gonorrhoeae* isolates demonstrated an overall resistance of 83.3% (Table 3).

Multi-Drug Resistance Profiles of the Isolates

From the total isolates, 8 (5.5%) were susceptible to all the tested antibiotics. The majority (94.4%) of isolates were

resistant to one or more antimicrobials tested. In general, 89 (61.8%) of bacterial species were MDR. The rate of MDR was 90.9%, 60.9% and 50%, among *Klebsiella* spp., *E. coli* and *S. aureus*, respectively (Table 4).

Discussion

The prevalence of culture-confirmed bacterial infection was significantly higher in females than males in the present study. The finding is in agreement with a report from Brazil.²⁰ This may be due to more specimens in this study

Table 4 Multi-Drug Resistance Profiles Among Bacterial Isolates at GAMBY Teaching General Hospital

Bacterial Isolates	R0	R1	R2	R3	R4	R5	R6	R7	Overall MDR
<i>E. coli</i> (n=64)	4	10	11	10	15	8	3	3	39
<i>S. saprophyticus</i> (n=26)	2	3	5	6	5	2	2	1	16
<i>S. aureus</i> (n=24)	0	4	8	5	3	1	2	1	12
<i>Klebsiella</i> spp. (n=11)	0	1	0	2	3	2	3	0	10
<i>S. pyogenes</i> (n=5)	1	0	2	1	0	0	0	1	2
<i>C. diversus</i> (n=5)	0	1	0	1	1	1	1	0	4
<i>S. epidermidis</i> (n=3)	0	0	0	1	1	0	1	0	3
<i>Enterobacter</i> spp. (n=2)	1	0	0	0	1	0	0	0	1
<i>S. pneumoniae</i> (n=1)	0	1	0	0	0	0	0	0	0
<i>Salmonella</i> spp. (n=1)	0	0	0	1	0	0	0	0	1
<i>Proteus</i> spp. (n=1)	0	0	0	0	0	1	0	0	1
<i>N. gonorrhoeae</i> (n=1)	0	1	0	0	0	0	0	0	0
Total (n=144)	8	21	26	27	29	15	12	6	89

Abbreviations: R0, susceptible to all antimicrobials tested; R1, R2, R3, R4, R5, R6 and R7, resistance to one, two, three, four, five, six and seven antibiotics taken from different classes, respectively.

being from urine, and females are more prone to get UTI. In this study Gram-negative bacteria were the predominant isolates. This was consistent with previous reports from other part of Ethiopia,²¹ India²² and China,²³ where Gram-negatives were more frequently found compared to Gram-positive isolates. The prevailing isolation of Gram-negative bacteria might be due to their simple nutritional requirements, frequent existence in the clinical setting and their ability to resist many antibiotics and detergents in the hospital area.

In this study, *E. coli* was the most predominant isolate. This is similar to studies from Burkina Faso²⁴ and Kigali, Rwanda²⁵ and studies from other parts of Ethiopia.^{26,27} This could be due to the abundance of *E. coli* in urinary tract infection and its virulence factors and interaction with the host. *S. saprophyticus* was the second most isolated pathogen, which was comparable with studies conducted in Nigeria²⁸ and Benin.²⁹ The frequent colonization of the rectum and genitourinary tract might be the source of infection for *S. saprophyticus* in the present study. Furthermore, *S. aureus* and *K. pneumoniae* were the other frequently isolated organisms in the present study. The finding was analogous to similar studies in Rwanda,²⁵ Nigeria²⁸ and other parts of Ethiopia.³⁰ The predominance of the above isolates could be associated with their frequent existence in the hospital settings, which is a major reason for the high chance of different body sites of infections and cross-contamination among inpatients. Moreover, *S. aureus*, *S. saprophyticus*, *E. coli* and *Klebsiella* spp. are normal residents of the skin and gut of individuals that can be displaced from the normal habitat to other sterile sites and openings on the skin and soft tissues, which results in dissemination and serious infections.

Gram-negative bacterial isolates of the present study demonstrated a high percentage of resistance to ampicillin (100%), amoxicillin+clavulanic acid (92.2%), amoxicillin (90.6%), tetracycline (84%), cotrimoxazole (78.8%) and erythromycin (77.1%). This might be linked with the widespread use of these antibiotics in the study area. These antibiotics are most frequently used in the community as they are cheap and easily available. Similarly, Gram-negative isolates in Ghana showed 94.4% resistance to ampicillin, and in India high rates of resistance were found against ampicillin and cotrimoxazole. However, the rate was relatively higher than that of the study conducted in Debre Markos, Ethiopia,²¹ where bacterial isolates showed resistance to penicillin (71%), ampicillin (71%),

amoxicillin (62.9%), cotrimoxazole (58.1%) and tetracycline (64.6%).³¹ Variation in the resistance percentages might reflect differences in sample size, study setting and prescription pattern, antibiotic therapy and epidemiology of causative organisms among different places.

E. coli and *Klebsiella* spp. in the present study were resistant to amoxicillin, tetracycline, ampicillin, amoxicillin+clavulanic acid, ceftriaxone, cotrimoxazole and ceftazidime. This showed that different body site infections with bacteria resistant to third-generation cephalosporin and other commonly used antibiotics is a major worry in the study area.

S. aureus and *S. saprophyticus* demonstrated the highest percentage of resistance to ciprofloxacin, erythromycin and tetracycline in the current study. The finding was similar to studies from different countries such as Nepal,³¹ Nigeria,²⁸ and Shiraz, Iran.¹⁶

Most (94.4%) of the isolates were resistant to one or more antibiotics tested in the present study. This was similar to a study done in Nepal (92.1%)³¹ and Debre Markos, Ethiopia (91.4%).²¹ The majority (61.8%) of the bacterial isolates of the present study were MDR. This was consistent with studies from other parts of Ethiopia^{21,27} and Indonesia.³² However, a lower MDR rate was reported in Birgunj, Nepal and Karachi, Pakistan and Khusibu, Nepal.³¹

In this study *Klebsiella* spp. showed MDR of 90.9%. This was consistent with findings from Indonesia (86.0%),³² Southern Ethiopia (92.9%)³³ and Karnataka, India (94.13%).²² However, it was higher than a report from Tanzania (63%).³⁴ It was also found that 50% of *S. aureus* was MDR. This was comparable to the study from Dessie, Ethiopia,³⁵ but it was lower than the study done in Kabul, Afghanistan (91.4%).³⁶ Similarly, 60.9% of *E. coli* was MDR in the present study. This was consistent with the study in Dessie, Ethiopia.³⁵ This revealed that antibiotic resistance especially MDR is a rising threat in Ethiopia. This might be linked with misuse, overuse and inappropriate antibacterial agents. Moreover, GAMBY Teaching General Hospital receives referrals from many districts and distant rural villages. These patients took different antibiotic treatments from the general practitioners and nurses or from over-the-counter, usually with inappropriate doses, before coming to the hospital. Therefore, the finding sounds the alarm for the implementation of a nationwide antimicrobial surveillance and in-vitro susceptibility testing at all levels of private and government hospitals with strict adherence to antibiotic

policy to inhibit the spread of drug-resistant microbes in the country.

Limitation of the Study

This study may not represent the general population of Bahir Dar area since it was an institutional study. Only those commonly used antibiotics in the institution were tested. Hence, it might not include all antibiotics used in clinical practice in the study area.

Conclusion

E. coli, *S. saprophyticus*, *S. aureus* and *Klebsiella* spp. were the predominant isolates from different body sites infections. Most of the isolates were resistant to ampicillin, amoxicillin+clavulanic acid, amoxicillin, tetracycline and cotrimoxazole antimicrobials. At the same time, the rates of multiple drug-resistant isolates are alarmingly high. Therefore, it is recommended to have strict antibiotics utilization policies within the hospital and to support clinicians on rational choice of antibiotics therapy and regularly update the list and reliable sources of drugs.

Abbreviations

AMR, antimicrobial resistance; ATCC, American Type Culture Collection; BSI, bloodstream infection; GGH, GAMBY General Hospital; HAI, health care associated infections; HMIS, Hospital Management Information System; MRSA, methicillin-resistant *Staphylococcus aureus*; spp., species; UTI, urinary tract infection; VRSA, vancomycin-resistant *Staphylococcus aureus*.

Data Sharing Statement

All data relevant to this study are available on the manuscript.

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The authors are grateful to GAMBY Medical and Business College for their permission to conduct the study in the hospital and support for culture media, biochemical tests and antibiotic discs.

Author Contributions

All authors made a significant contribution to the work reported, whether that was in the conception, study design, execution, acquisition of data, analysis and interpretation or in all of these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; agreed on the journal to which the

article was submitted; and agree to be accountable for all aspects of the work.

Disclosure

The authors declare that they have no competing interests in this work.

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