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ORIGINAL RESEARCH

Antimicrobial Resistance Patterns of *Staphylococcus aureus* and *Enterococcus* Species at the Ethiopian Public Health Institute, Ethiopia: A Five-Year Retrospective Analysis

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Purpose: The study aimed to investigate the antimicrobial resistance patterns of *Staphylococcus aureus* and *Enterococcus* species isolated from clinical specimens over a period of five years, including resistance to methicillin and vancomycin.

Patients and Methods: Bacterial identification and antimicrobial susceptibility testing reports from 2017 to 2021 at the Ethiopian Public Health Institute were used for this retrospective study. The organisms were identified using either BD Phoenix M50, Vitek 2 compact, or conventional biochemical methods, whichever was available at the time of testing. The antimicrobial susceptibility profiles of the isolates were determined using either Kirby-Bauer disc diffusion, BD phoenix M50, or Vitek 2 compact. WHONET software was used to analyze the antimicrobial resistance patterns of both organisms. The p-values of ≤0.05 were considered statistically significant.

Results: During the study period, a total of 315 *Staphylococcus aureus* and 92 *Enterococcus* species were isolated. Out of 315 *Staphylococcus aureus* isolates, 27% and 5.1% were methicillin and vancomycin resistant, respectively. *Staphylococcus aureus* showed very high resistance to Penicillin G (86.7%). Out of 92 *Enterococcus* species recovered, 8.7% were vancomycin-resistant. *Enterococcus* species showed very high resistance to Penicillin G (71.4%) and tetracyclines (83.3%). Methicillin-resistant *Staphylococcus aureus* shows 100% resistance to penicillin followed by ciprofloxacin (50%), erythromycin (45.6%), and tetracycline (44.2%) and lower resistance to vancomycin (18.8%). All vancomycin-resistant isolates of both organisms were fully resistant (100%) to all antibiotics tested, except for linezolid and daptomycin, to which they were susceptible.

Conclusion: This study found a high prevalence of methicillin and vancomycin-resistant *Staphylococcus aureus* and vancomycin-resistant *Enterococcus* species between 2017 and 2021. However, there were no statistically significant changes in the prevalence of these organisms during the study period. This suggests that larger and more representative nationwide data is needed to show trends of these pathogens.

Keywords: antimicrobial resistance, *Staphylococcus aureus*, *Enterococcus* species, Ethiopia

Introduction

Staphylococcus aureus and *Enterococcus* species are both types of Gram-positive bacteria that can cause infections in humans.[1,2] They have been identified by the World Health Organization (WHO) as emerging causes of nosocomial infections in recent decades, posing a significant threat to public health.[3] *Staphylococcus aureus* is a Gram-positive, catalase, and coagulase-positive facultative anaerobic bacteria that colonizes up to 30% of the human population.[4] However, certain groups, such as healthcare workers, hospitalized patients, intravenous drug users, and individuals with compromised immune systems, have higher rates of colonization (up to 80%).[2] *Staphylococcus aureus* causes infection only if it manages to enter the bloodstream or internal tissues.[5] It is a major cause of healthcare-associated infections,

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including bloodstream infections, surgical site infections, and pneumonia. Additionally, it can also cause community-acquired infections, such as skin and soft tissue infections and sepsis.[2,6,7]

Enterococci are a type of Gram-positive, facultative anaerobic bacteria that form short and medium chains of cocci that are commonly found in the gastrointestinal tract of humans and animals.[1] *Enterococcus faecalis* and *Enterococcus faecium* are the most common species that cause infections in humans, and they are often associated with healthcare-associated infections, such as urinary tract infections, endocarditis, bloodstream infections, and surgical site infections.[1,8,9] *Enterococci* are intrinsically resistant to cephalosporins, clindamycin, aminoglycosides, and trimethoprim-sulfamethoxazole.[10] In addition, there have been reports of *Staphylococcus aureus* strains that are resistant to both methicillin (MRSA) and vancomycin (VRSA), as well as *Enterococcus* species that are resistant to vancomycin (VRE). This can make treatment more difficult and raise the likelihood of mortality.[7,9,11] Vancomycin is primarily used to treat infections caused by MRSA and those who are allergic to semisynthetic penicillin and cephalosporins.[12]

In recent years, the prevalence of antimicrobial resistance among these organisms has increased significantly, highlighting the need for ongoing surveillance of their resistance patterns.[13–17] A systematic analysis conducted in 2019 estimated *Staphylococcus aureus* is among the six leading pathogens for death-associated resistance. Particularly, Methicillin-resistant *Staphylococcus aureus* caused more than 100,000 deaths attributable to antimicrobial resistance. The study also reported that the *Enterococcus* species was responsible for between 100,000 and 250,000 deaths associated with antimicrobial resistance.[18] Some studies have been conducted in Ethiopia to assess the antimicrobial resistance patterns of *Staphylococcus aureus* and *Enterococcus* species, but these studies are limited in scope, as they only analyzed a smaller number of isolates from specific geographic locations within the country. Therefore, this study aimed to determine the five-year antimicrobial resistance patterns of *Staphylococcus aureus* and *Enterococcus* species isolated from clinical specimens referred from various health facilities to the Ethiopian Public Health Institute. The findings can provide valuable insights into these bacteria's distribution and resistance patterns in the country. Moreover, this study's results can guide the appropriate use of antimicrobial agents in the treatment of infections caused by these bacterial pathogens, leading to better patient outcomes and reduced healthcare expenses.

Materials and Methods

Study Design and Data Extraction

In this retrospective study, we analyzed the antimicrobial resistance profiles of *Staphylococcus aureus* and *Enterococcus* species isolated from clinical specimens referred to the National Clinical Bacteriology and Mycology Reference Laboratory, Ethiopian Public Health Institute between January 1, 2017, and December 31, 2021. The laboratory was accredited by the Ethiopian Accreditation Service in 2017 for meeting the requirements of the International Organization for Standardization (ISO) 15,189: 2012. All *Staphylococcus aureus* and *Enterococcus* species isolated from all specimens with complete information were included. Identification and antimicrobial susceptibility testing results of both *Staphylococcus aureus* and *Enterococcus* species data were extracted from the bacteriology result logbook, BD Phoenix M50, and Vitek 2 compact instruments. Data from BD Phoenix M50 and Vitek 2 compact instruments were converted to a WHONET SQLite file using the WHONET backlink for analyzing the antimicrobial susceptibility pattern of *Staphylococcus aureus* and *Enterococcus* species.

Specimen Processing, Bacterial Isolation, and Identification

Specimens were sent from different healthcare facilities across the country. The specimens obtained encompassed various types of specimens, including urine, blood, wound, eye, body fluids, and other miscellaneous specimens. Only those samples that met the laboratory's acceptance criteria were accepted and then inoculated into suitable culture media, after which they were incubated at the proper temperature and duration. *Staphylococcus aureus* and *Enterococcus* species were identified using one of the three methods: VITEK® 2 Compact (bioMérieux, France), BD phoenix M50 (Becton, Dickinson, USA), or conventional biochemical tests. Conventional biochemical tests involved macroscopic colony characterization (color, size, shape, texture, hemolysis), Gram staining, and biochemical tests (catalase test, coagulase test, L-Pyrrolidonyl Arylamidase (PYR), bile esculin agar, 6.5% sodium chloride tolerance test).

Antimicrobial Susceptibility Testing

The antimicrobial susceptibility of *Staphylococcus aureus* and *Enterococcus* species to antimicrobial agents was tested using either the BD Phoenix M50 or Vitek 2 compact systems, or the Kirby-Bauer disk diffusion method, whichever was available at the time of testing. The results were interpreted using the latest Clinical and Laboratory Standards Institute (CLSI) M100 guideline.[10] The antimicrobial disks were obtained from the following manufacturers: (Oxoid Ltd., Basingstoke, Hampshire, England), (Liofilchem, Roseto degli Abruzzi, Italy), (Abtek, Liverpool, United Kingdom), and (Hardy Diagnostics, Santa Maria, California, and Springboro, Ohio, United States).

For *Staphylococcus aureus*, the antimicrobial agents tested and reported were as follows: Penicillin G (10 units), cefoxitin (30µg), gentamycin (10µg), ciprofloxacin (5µg), trimethoprim/sulfamethoxazole (1.25/23.75µg), clindamycin (2µg), erythromycin (30µg), linezolid (30µg), chloramphenicol (30µg), tetracycline (30µg) and Nitrofurantoin (300µg). For *Enterococcus* species, the following antimicrobial agents were tested and reported: Penicillin G (10 units), ampicillin (10µg), ciprofloxacin (5µg), erythromycin (30µg), linezolid (30µg), chloramphenicol (30µg), tetracycline (30µg), nitrofurantoin (300µg), and vancomycin (30µg).

Additionally, daptomycin and vancomycin susceptibility in *Staphylococcus aureus* and daptomycin susceptibility in *Enterococcus* were tested using the VITEK® 2 Compact, BD phoenix M50 systems, and vancomycin Etest (bioMérieux, France). This was necessary because disk diffusion testing of these antibiotics for these organisms is not reliable. To screen for oxacillin or methicillin-resistant *Staphylococcus aureus* (MRSA), a cefoxitin disk was used as a surrogate agent for oxacillin. A zone diameter of ≥22 mm was considered to be susceptible to oxacillin, while a zone diameter of ≤21 mm was considered to be resistant to oxacillin. Nitrocefin and zone edge tests were used to screen for β-lactamases in *Staphylococcus aureus* strains with a penicillin zone diameter of ≥29mm before reporting them as susceptible. Sharp zone edge (cliff) was considered β-lactamase positive indicating resistance to penicillin, and fuzzy zone edge (beach) was considered β-lactamase negative indicating susceptibility to penicillin.

Quality Assurance

The quality of the culture media used was checked as per CLSI M22-A3 guidelines,[19] laboratories standard operating procedures, and manufacturers' recommendations for each culture media. American-type culture collection (ATCC) strains, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, and *Staphylococcus aureus* ATCC 25923 were used to check the quality of all the antimicrobial disks used as per clinical and laboratory standards institute (CLSI) M100 Table 4A 1–2. A 15-day replicate (3x5) plan was used to check the quality of newly bought antimicrobial agents using the ATCC strains mentioned above. Once satisfactory results were obtained from daily quality controls, the quality control testing frequency was switched to weekly testing on Tuesdays of each week.

Data Analysis

The data was regularly entered into the WHONET software. The data completeness was checked using the WHONET standard report feature, and data with incomplete information were removed from the dataset. The overall antimicrobial susceptibility profile of *Staphylococcus aureus* and *Enterococcus* species to the abovementioned antimicrobial agent was analyzed to calculate the proportion of isolates resistant to different antimicrobial agents using resistance, intermediate, and susceptible (RIS) analysis type from WHONET software. Additionally, oxacillin and vancomycin-resistant *Staphylococcus aureus* and vancomycin-resistant *Enterococcus* species were selected to analyze their susceptibility profile to other antimicrobial agents. The distribution of the bacterial isolates by health facility and ward was also analyzed.

Ethical Considerations

The institutional review board (IRB) of the Ethiopian Public Health Institute (EPHI) approved this study and waived the need for informed consent because the study was retrospective and did not involve any intervention or interaction with patients. The study was assigned the IRB approval number EPHI-IRB-413-2021. Patient confidentiality was maintained by using de-identified data in the analysis. Hence, the study was conducted following the Declaration of Helsinki.

Results

Between 2017 and 2021, a total of 7110 clinical specimens were received at the National Clinical Bacteriology and Mycology Reference Laboratory of the Ethiopian Public Health Institute, after excluding 89 records due to missing information. Of these specimens, 315 *Staphylococcus aureus* and 92 *Enterococcus* species isolates were recovered. Of the 315 *Staphylococcus aureus* isolates, 27% were methicillin-resistant and 5.1% were vancomycin-resistant. Additionally, out of 92 *Enterococcus* species, 8.7% were vancomycin-resistant (Figure 1).

Socio-Demographic Characteristics of the Patients

Table 1 summarizes the demographic characteristics of patients and the number of *Staphylococcus aureus* and *Enterococcus* species isolates obtained based on age, sex, healthcare facilities, different wards, and specimen source. Out of 407 isolates 246 and 161 were recovered from specimens obtained from males and females, respectively. The age group of 21 to 30 had 94 isolates recovered from their specimens. The majority of isolates were isolated from specimens referred from AaBET Hospital (187, representing 46% of the total). The specimens collected from patients admitted to orthopedics (102) and surgery (93) wards had the highest number of isolates, with 170 of them being *Staphylococcus aureus*, which represents about 87% of the total. Wound specimens had the highest number of isolates (259), with 248 of them being *Staphylococcus aureus*, representing 96% of the total.

Antimicrobial Susceptibility Profile

The antimicrobial susceptibility profile of *Staphylococcus aureus* and *Enterococcus* species is shown in Table 2. Out of 315 *Staphylococcus aureus* isolates, 85 (representing 27% of the total) and 16 (representing 5.1% of the total) were methicillin and vancomycin-resistant, respectively. *Staphylococcus aureus* showed very high resistance to Penicillin

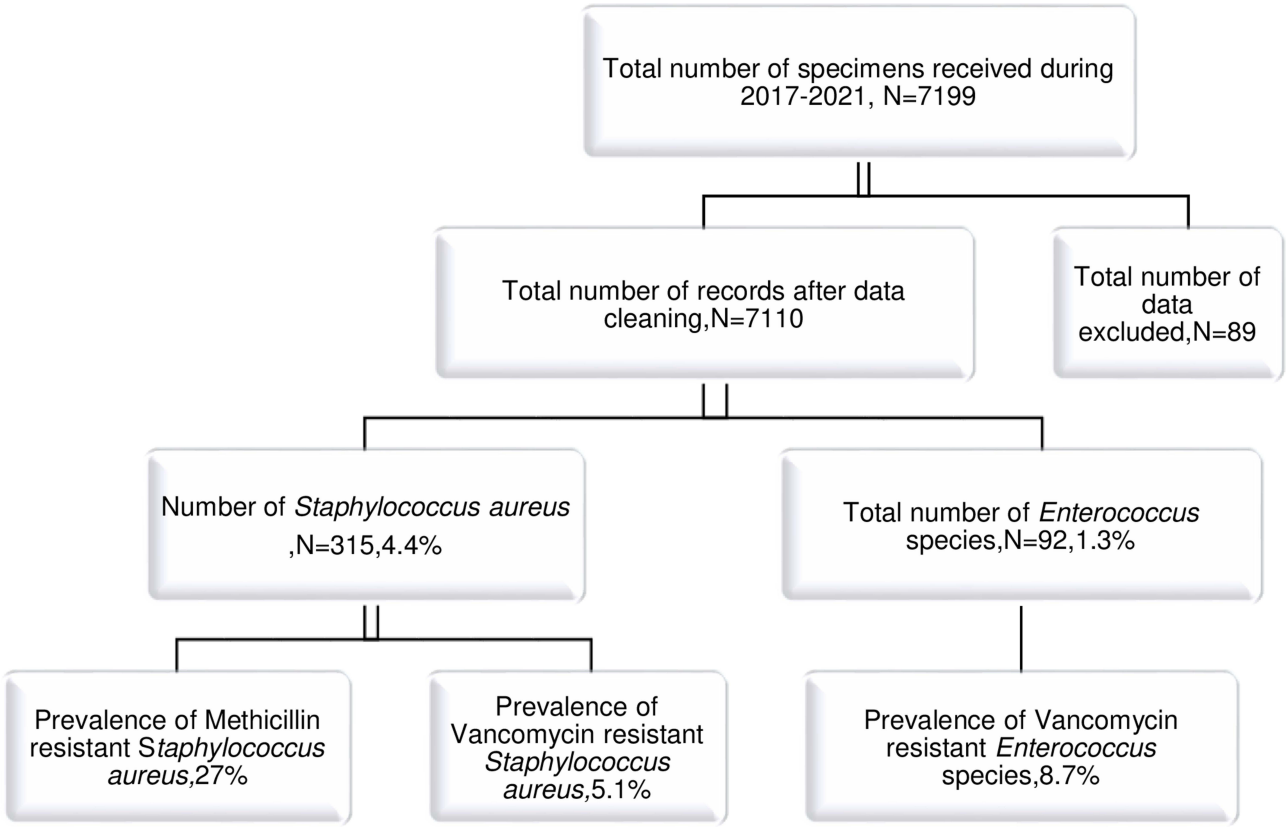

Figure 1 Flowchart of the data cleaning process.
Abbreviation: N, number.

Table 1 Socio-Demographic Characteristics of Patients from Which Specimens Were Obtained from 2017–2021

	Variables	*S. aureus*	*Enterococcus* Species	Number of MRSA	Number of VRSA	Number of VRE
Sex	Male	194	52	55	8	4
	Female	121	40	30	8	4
Age	<1 year	25	7	8	1	5
	1 to 10	44	16	14	1	1
	11 to 20	47	13	12	2	0
	21 to 30	79	15	17	5	1
	31 to 40	39	19	16	2	0
	41 to 50	29	6	5	4	0
	51 to 60	27	10	8	0	0
	Above 61	25	6	5	2	1
Health facilities	St Paul Hospital	18	9	7	1	1
	AaBET Hospital	162	25	40	7	1
	Alert Hospital	12	8	6	1	0
	Federal police Hospital	12	10	3	1	1
	Minilik II Referral Hospital	14	12	7	2	0
	Ras Desta Hospital	27	16	7	2	3
	Private clinics	6	3	1	0	2
	Other hospitals	64	9	14	2	0
Department	Emergency	37	17	7	1	2
	Intensive care unit	19	11	4	3	1
	Medical	31	9	15	0	3
	Orthopedics	91	11	29	7	1
	Outpatient	22	12	6	2	0
	Pediatrics	36	18	14	0	0
	Surgery	79	14	10	3	1
Specimens	Urine	6	39	4	1	2
	Blood	23	39	8	2	6
	Wound	248	11	66	10	0
	Eye and eye	25	0	5	3	0
	Body fluids	4	1	1	0	0
	Other	9	2	1	0	0

Abbreviations: MRSA, methicillin-resistant *Staphylococcus aureus*; VRSA, vancomycin-resistant *Staphylococcus aureus*; VRE, vancomycin-resistant *Enterococcus* species.

Table 2 Antimicrobial Susceptibility Profile of *Staphylococcus aureus* and *Enterococcus* Species

Antibiotic Name	*Staphylococcus aureus*				*Enterococcus* Species			
	%R	%I	%S	%R 95% C.I.	%R	%I	%S	%R 95% C.I.
Penicillin G	86.7	0.4	12.8	81.4–90.7	71.4	0.0	28.6	47.7–87.8
Oxacillin	27.0	0.4	72.7	20.1–33.1	–	–	–	–
Ampicillin	–	–	–	–	66.7	0.0	33.3	52.4–78.5
Gentamycin	10.3	2.8	86.9	5.5–18.0	–	–	–	–
Ciprofloxacin	13.3	1.8	84.9	8.7–19.6	71.9	12.5	15.6	53.0–85.6
Trimethoprim/Sulfamethoxazole	18.6	2.7	78.7	14.2–24.0	–	–	–	–
Clindamycin	10.6	0.8	88.6	7.1–15.4	–	–	–	–
Daptomycin	0	0.0	100.0	1.8–34.5	0.0	0.0	100.0	0.0–30.1
Erythromycin	30.2	0.0	69.8	18.7–44.5	54.5	27.3	18.2	24.6–81.9
Linezolid	0.0	0.0	100.0	0.1–11.6	0.0	0.0	100.0	1.8–34.5
Vancomycin	5.1	0.0	96.2	0.0–7.3	8.7	0.0	91.3	0.0–7.3
Chloramphenicol	44.4	11.1	44.4	15.3–77.3	25.0	12.5	62.5	10.6–47.1
Tetracycline	35.9	4.2	59.9	28.8–43.8	83.3	4.2	12.5	61.8–94.5
Nitrofurantoin	–	–	–	–	8.0	0.0	92.0	1.4–27.5

Abbreviations: %R, percent resistant; %I, percent intermediate; %S, percent susceptible; C.I., confidence interval; -, not tested.

G (86.7%), tetracyclines (35.9%), and chloramphenicol (44.4%). The resistance rates to gentamicin and clindamycin were low at 10.3% and 10.6%, respectively. Of the 92 *Enterococcus* species, 8 (representing 8.7% of the total) were vancomycin-resistant. These isolates showed very high resistance to Penicillin G (71.4%), tetracyclines (83.3%), ciprofloxacin (71.9%), and erythromycin (54.5%), and lower resistance to nitrofurantoin (8%), and chloramphenicol (25%). All isolates of *Staphylococcus aureus* and *Enterococcus* species were susceptible to daptomycin and linezolid.

Antimicrobial Susceptibility Profile of MRSA, VRSA, and VRE

The antimicrobial susceptibility profile of methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant *Staphylococcus aureus* (VRSA) and vancomycin-resistant *Enterococcus* species (VRE) is shown in Table 3. All of the 85 *Staphylococcus* aureus isolates that were resistant to methicillin also demonstrated resistance to penicillin. However, about 50%, 45.6%, and 44.2% of these isolates were found to be resistant to ciprofloxacin, erythromycin, and tetracycline, respectively. In contrast, the resistance level to vancomycin was lower, at 18.8%. Both vancomycin-resistant *Staphylococcus aureus* and vancomycin-resistant *Enterococcus* species demonstrated complete resistance to all tested antibiotics, but they exhibited no resistance to linezolid and daptomycin.

Figure 2 shows five trends in the prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant *Staphylococcus aureus* (VRSA), and vancomycin-resistant *Enterococcus*. The prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) was 25.3% in 2017. It increased to 29% and 28.7% in 2018 and 2019, respectively. However, it decreased to 16.7% in 2020 and then increased again to 27.8% in 2021 (p-value 0.59673). The prevalence of vancomycin-resistant *Staphylococcus aureus* (VRSA) was 4% in 2017. It increased slightly to 5.4%, 6.9%, and 4.2% in 2018, 2019, and 2020, respectively. However, it decreased to 2.8% in 2021 (p-value 0.5895). The prevalence of vancomycin-resistant *Enterococcus* species was 7.1% in 2017. It increased to 9.4%, 8.3%, and 12.5% in 2018, 2019, and 2020, respectively. It then slightly decreased back to 7.1% in 2021 (p-value 0.73063).

Table 3 Antimicrobial Susceptibility Profile of MRSA, VRSA, and VRE

Antibiotic Name	VRSA			MRSA			VRE		
	%R	%I	%S	%R	%I	%S	%R	%I	%S
Penicillin G	100.0	0.0	0.0	100.0	0.0	0.0	100.0	0.0	0.0
Oxacillin	100.0	0.0	0.0	100.0	0.0	0.0	–	–	–
Gentamicin	100.0	0.0	0.0	36.4	13.6	50.0	–	–	–
Ciprofloxacin	100.0	0.0	0.0	50.0	2.5	47.5	100.0	0.0	0.0
Trimethoprim/Sulfamethoxazole	100.0	0.0	0.0	31.3	4.7	64.1	–	–	–
Clindamycin	100.0	0.0	0.0	28.8	1.9	69.2	–	–	–
Daptomycin	0.0	0.0	100.0	0.0	0.0	100.0	0.0	0.0	100.0
Erythromycin	100.0	0.0	0.0	45.6	17.5	36.8	100.0	0.0	0.0
Linezolid	0.0	0.0	100.0	0.0	0.0	100.0	0.0	0.0	100.0
Vancomycin	100.0	0.0	0.0	18.8	0.0	81.2	100.0	0.0	0.0
Chloramphenicol	100.0	0.0	0.0	0.0	33.3	66.7	100.0	0.0	0.0
Tetracycline	100.0	0.0	0.0	44.2	9.3	46.5	100.0	0.0	0.0
Ampicillin	–	–	–	–	–	–	100.0	0.0	0.0
Nitrofurantoin	–	–	–	–	–	–	100.0	0.0	0.0

Abbreviations: %R, percent resistant; %I, percent intermediate; %S, percent susceptible; MRSA, methicillin-resistant *Staphylococcus aureus*; VRSA, vancomycin-resistant *Staphylococcus aureus*; VRE, vancomycin-resistant *Enterococcus* species; -, not tested.

Discussion

In this retrospective study, we analyzed the five-year antimicrobial resistance patterns of *Staphylococcus aureus* and *Enterococcus* species isolated from clinical specimens with a special focus on methicillin and vancomycin resistance. In the present study, the highest proportion of *Staphylococcus aureus*, accounting for 78.7%, was obtained from wound samples. Among these isolates, 26.6% were identified as MRSA (Methicillin-Resistant *Staphylococcus aureus*), while 4.0% were identified as VRSA (Vancomycin-Resistant *Staphylococcus aureus*). These findings are consistent with the results reported in previous studies conducted at Asmara, Eritrea,[20] Jimma University, Ethiopia,[21] Yekatit 12 Hospital Medical College, Ethiopia,[22] and Karl Referral Hospital, Ethiopia.[23] The majority (84.8%) of *Enterococcus* isolates were obtained from blood and urine samples, of which 10.3% were identified as VRE (Vancomycin-Resistant *Enterococcus*). These results align with a study conducted at the University of Gondar[24] and Felege Hiwot Hospital, Ethiopia,[25] which reported similar findings.

In this study, the prevalence of methicillin-resistant *Staphylococcus aureus* was 27%, which is almost similar to the findings from Referral Hospital, Northeast Ethiopia (28.3%),[26] a meta-analysis conducted in Ethiopia (32.5%).[15] However, the prevalence was higher than that reported in other studies, such as Mettu Karl Referral Hospital, Ethiopia (18.8%),[23] Yekatit 12 Hospital, Addis Ababa, Ethiopia (17.5%),[22] Mekelle, Northern Ethiopia (2.4%),[27] and Debre Markos Referral Hospital, Ethiopia (13.22%).[28] Conversely, the prevalence was lower than that reported in other studies, such as Tikur Anbessa Specialized Hospital, Addis Ababa, Ethiopia (35.6%),[29] Debre Markos Referral Hospital, Ethiopia (49.7%),[30] Arba Minch Hospital, South Ethiopia (82.3%),[31] Jimma University Specialized Hospital, Ethiopia (76.7%),[21] A Multicenter Study in Asmara, Eritrea (72%),[20] and a systematic review and meta-analysis in low- and lower-middle-income countries (48.4%),[32] and northwest, Iran (53.7%),[33] a meta-analysis, Ethiopia (47%).[33] These observed differences in MRSA prevalence could be attributed to factors such as differences in study design, geographic location, sample sizes, patient populations, laboratory methods, and use of antibiotics.

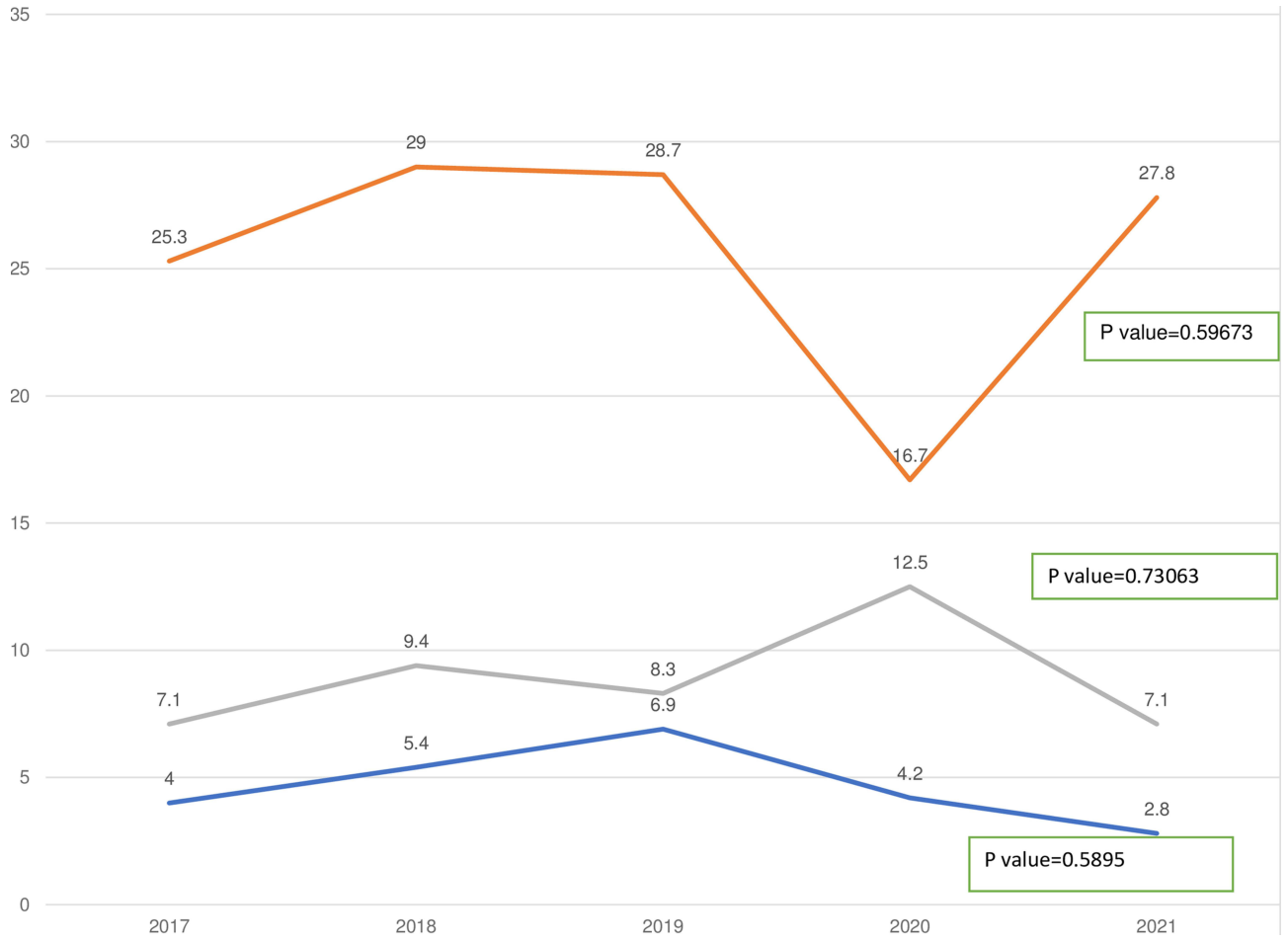

Figure 2 Five-year trends of methicillin-resistant *Staphylococcus aureus*, vancomycin-resistant *Staphylococcus aureus*, and vancomycin-resistant *Enterococcus* species.
Notes: Vancomycin-resistant *Staphylococcus aureus*. Methicillin-resistant *Staphylococcus aureus*. Vancomycin-resistant *Enterococcus species*.

In the current study, the prevalence of vancomycin resistant *Staphylococcus aureus* was 5.1%, which is in agreement with study from Debre Markos Referral Hospital, Ethiopia (4.1%), and[28] the pooled global prevalence of 6% reported between 2000 and 2019,[14] a meta-analysis, Ethiopia (5.3%),[15] but lower than the prevalence reported in another meta-analysis from Ethiopia (11%).[16] However, the prevalence was higher than the pooled global prevalence of VRSA 1.5%,[34] and the pooled prevalence in low- and middle-income countries (0.6%).[32] The different results of the studies could be due to differences in how the studies were designed, the types of samples that were used, the locations where the studies were conducted, and the policies regarding antibiotic prescription.

The prevalence of vancomycin-resistant *Enterococcus* species in this study was 8.7%. This is similar to the prevalence rates found in Asia (8.1%),[35] Addis Ababa, Ethiopia (6.7%),[36] and the SENTRY antimicrobial surveillance program (8%).[37] However, it was lower than the studies from Nigeria (26.5%),[38] Iran (14%),[39] Egypt (26%),[40] and Ethiopia (14.8%).[41] Our finding was also much lower than the prevalence rates reported from Bahir Dar, Ethiopia (34.61%),[25] and Gondar, Ethiopia (41.7%).[24] The variation in the prevalence rates could be due to a number of factors, such as the study design, the types of samples collected, the timing of the study, differences in antibiotic prescription policies, differences in geographical locations, and other potential factors. Vancomycin is a powerful antibiotic that is used to treat infections caused by antibiotic-resistant *Staphylococcus aureus* and *Enterococcus* species with better clinical outcomes. It is important to use vancomycin wisely, as it is one of the last lines of defense against these infections. It serves as a crucial treatment option in the management of such infections, offering a reliable treatment alternative when other antibiotics fail.[42–44]

In the current study, methicillin-resistant *Staphylococcus aureus* (MRSA) showed no resistance to linezolid and daptomycin, but 18.8% resistance to vancomycin. Most studies reported zero resistance rates of MRSA to vancomycin, linezolid, and daptomycin. For instance, a study from India reported 0% resistance rates of MRSA to vancomycin,[45] and North India, reported 0% resistance rates of MRSA to vancomycin and linezolid,[46] Eastern India, reported 0% resistance of MRSA to vancomycin, daptomycin and linezolid,[47] pooled result from Ethiopia, reported 5.3% rates of resistance to vancomycin,[15] and Nairobi, Kenya, less than 5% resistance rates of MRSA to linezolid and vancomycin.[48]

Vancomycin has long been considered the last-resort treatment for MRSA infections and resistance to vancomycin is mediated by a vanA gene, which is transferred from vancomycin-resistant *Enterococcus*.[34] There are multiple factors that can be directly or indirectly associated with the increasing burden of infections caused by MRSA, VRSA, and VRE strains and one of them is Excessive use of vancomycin resulted in the emergence of vancomycin-resistant *Staphylococcus aureus* (VRSA). These strains, in turn, lead to escalated expenses within healthcare facilities, prolonged hospital stays, heightened morbidity rates, and increased mortality rates.[14,34]

In the current study, all 16 vancomycin-resistant *Staphylococcus aureus* (VRSA) strains were susceptible to daptomycin and linezolid. This is consistent with the findings of other studies, including a review of VRSA cases that reported that VRSA were susceptible to daptomycin and linezolid,[11] results of the Network on Antimicrobial Resistance in *S. aureus* (NARSA) Program which reported that >90% of 13 VRSA isolates were susceptible to daptomycin and linezolid,[49] and a study from the United States that found that 7 out of 7 VRSA isolates were susceptible to linezolid and 6 out of 7 were susceptible to daptomycin.[50]

Similarly, all 8 vancomycin-resistant *Enterococcus* species (VRE) were susceptible to daptomycin and linezolid, which agrees with findings of the study from Ethiopia, which reported very low resistance of VRE to daptomycin and linezolid with a pooled estimate of 3.2% and 9.9%, respectively,[41] and a study from Egypt between 2010 and 2022 which reported VRE resistance rate of 5.54% to linezolid.[40] This is because vancomycin-resistant *Staphylococcus aureus* (VRSA) and vancomycin-resistant *Enterococci* (VRE) strains are resistant to vancomycin due to the acquisition of the vanA gene from vancomycin-resistant *Enterococci* (VRE). This gene encodes a modified peptidoglycan precursor that has a lower affinity for vancomycin, making the bacteria resistant to this particular antibiotic.[11,35] Linezolid and daptomycin, on the other hand, have different mechanisms of action and are not affected by the modified peptidoglycan precursor. Linezolid and daptomycin target different components of the bacterial cell, hence they are effective against VRSA and VRE strains.[9,33,49]

The present study showed that from 2017 to 2021, the prevalence of MRSA fluctuated, which contradicts the findings of a study from the United States from 2010 to 2014 that reported decreasing rates of MRSA infections among hospitalized patients.[51] It also contradicts the findings of a nationwide surveillance study (2008–2021) that reported decreasing MRSA infections.[52] The study also found that the prevalence of VRSA was fluctuated, which contradicts the findings of a global pooled result of a systematic review and meta-analysis between 1997 and 2019 that reported an increase in the prevalence of VRSA from 1.2% before 2010 to 2.4% after 2010.[34] Likewise, the study also found that the prevalence of VRE was fluctuated, which contradicts the findings of a three-year prospective study from India that reported increasing trends of VRE[53] and the pooled results from the European Region Surveillance Network that reported decreasing trends of VRE.[54] The varying trends observed in methicillin and vancomycin-resistant *Staphylococcus aureus*, and vancomycin *Enterococcus* species may be explained by the low number of each strains isolated between 2017 and 2021 at the Ethiopian Public Health Institute.

Limitations

This study has the following limitations that should be considered when interpreting the results. The first is, the study was conducted at a single institution. The second is, the study was retrospective, which means that the data were collected from records and may not reflect the current situation. The third is, we did not include information on the clinical outcomes of patients infected with these organisms. The fourth is, we did not detect resistance genes using molecular methods. The fifth is, we did not include information on the risk factors associated with antimicrobial resistance due to the retrospective nature of the work.

Conclusion

Between 2017 and 2021, 315 *Staphylococcus aureus* and 92 *Enterococcus* species were isolated, and there was a high prevalence of methicillin and vancomycin-resistant *Staphylococcus aureus* (MRSA and VRSA) strains, with resistance rates of 27% and 5.1%, respectively. *Staphylococcus aureus* showed a very high resistance to Penicillin G (86.7%). Methicillin-resistant *Staphylococcus aureus* showed 18.8% resistance to vancomycin. Vancomycin-resistant *Enterococcus* species isolates were also prevalent, with a resistance rate of 8.7%. There were no statistically significant changes in MRSA, VRSA, and VRE during the study period, which suggests that larger and more representative nationwide data is needed to show trends of these pathogens. Additionally, all *Staphylococcus aureus* and *Enterococcus* species isolates were susceptible to linezolid and daptomycin. The high prevalence of these bacteria in commonly used antibiotics necessitates the importance of strengthening the antimicrobial surveillance system, antibiotic stewardship, and infection prevention and control practices at the health facility.

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Disclosure

The authors report no conflicts of interest in this work.

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