

Research Article

Assessment of Antibiotics Resistance from Isolates Responsible for UTI in Four Regional Referral Hospitals in Tanzania

Adelard Bartholomew Mtenga^{1,*} , Adam Fimbo¹ , Danstan Hipolite¹ ,
Revocatus Makonope¹ , Saxon Mwambene¹ , Yonah Hebron¹ ,
Kissa Mwamwitwa¹ , Raphael Zozimus Sangeda² 

¹Tanzania Medicines and Medical Devices Authority, Dar es Salaam, Tanzania

²Department of Pharmaceutical Microbiology, Muhimbili University of Health and Allied Sciences, Dar es Salaam, Tanzania

Abstract

The global impact of antimicrobial resistance (AMR) includes increased morbidity and mortality rates and healthcare costs, particularly in low- and middle-income countries (LMICs), and it has dire economic and security implications. This study assessed the resistance of clinical isolates responsible for urinary tract infections (UTI) to antibacterial agents for treating UTIs in selected healthcare facilities in Tanzania. A total of 151 clinical isolates of *E. coli* and *S. aureus* isolated from urine samples in selected health facilities were analyzed for antimicrobial susceptibility to establish the presence of individual and multi-drug resistance (MDR). The results revealed that *E. Coli* displayed a significant difference in resistance ($\chi^2=12.808$, $p=0.002$) across the selected antibiotics, in which *E. coli* showed the highest resistance to amoxicillin (AML) and the least resistance to meropenem ($p < 0.005$). In contrast, *S. aureus* isolates showed a significant difference ($\chi^2=53.627$, $p\text{-value} < 0.001$) in resistance across the selected antibiotics, in which *S. aureus* showed the highest resistance to AML, peaking at more than 91%, and least resistant (4%) to nitrofurantoin (NIT) (4%). When $p\text{-value} < 0.005$, both *E. coli* and *S. aureus* demonstrated MDR against selected antibiotics in all health facilities under study, in which Morogoro Regional Referral Hospital showed the highest (65.4%) for *E. coli* and Benjamin Mkapa Hospital showed the highest (83.3%) for *S. aureus*. Similarly, Maweni Regional Referral Hospital demonstrated the lowest MDR for *E. coli* (23%) and *S. aureus* (13%). Finding suggest that some antibiotics are still in used in clinical practice despite of the evidence of emerging resistance against them hence it call for effective regular AMR surveillance and antimicrobial stewardship implementation to optimize antibiotics use in clinical practice and exclude less efficacious ones.

Keywords

Antimicrobial Resistance, Clinical Isolates, Urinary Tract Infections, *E. coli* and *S. aureus*, Tanzania Healthcare Facilities

*Corresponding author: amtengab@yahoo.com (Adelard Bartholomew Mtenga)

Received: 4 December 2024; Accepted: 18 December 2024; Published: 31 December 2024



Copyright: © The Author(s), 2024. Published by Science Publishing Group. This is an **Open Access** article, distributed under the terms of the Creative Commons Attribution 4.0 License (<http://creativecommons.org/licenses/by/4.0/>), which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited.

1. Introduction

Antibacterial agents are compounds that kill or immobilize bacteria and are used to treat and manage bacterial infections and perioperative procedures [1]. These are further categorized based on their activity against different bacterial agents as either broad-spectrum, which acts on a wide range of gram-positive and gram-negative bacteria, or narrow-spectrum, which works on either gram-positive or gram-negative bacteria [2].

Despite the importance of these antibiotics in treatment, there have been reports of a significant increase in unsuccessful treatments owing to emerging antimicrobial resistance (AMR) [3]. When AMR occurs, microorganisms persist or grow in the presence of antibiotics designed to inhibit or kill them [4]. Several related factors, including poor hygiene and sanitation, infection control, overprescription, lack of treatment completion, antibiotic overuse in farming, and lack of development of novel antibiotics, have been identified as drivers of emerging AMR [5, 6]. Additionally, substandard medicines are another major factor driving the development of antimicrobial resistance. For instance, a 2020 report by the Tanzania Medicines and Medical Devices Authority (TMDA) recorded nine substandard and falsified medicines, including antibiotics, circulating in the market [7].

The global effects of AMR include increased morbidity and mortality rates, increased healthcare costs, particularly in low- and middle-income countries (LMICs), and dire economic and security implications [8]. Numerous studies conducted in Tanzania over the past ten years reveal that AMR continues to rise despite various intervention efforts [9, 10]. To combat AMR, the World Health Organization (WHO), by partnering with different countries, including Tanzania, initiated antimicrobial stewardship programs to promote the optimal use of antimicrobials at all levels of healthcare facilities [11, 12]. Tanzania continues implementing its National Action Plan on Antimicrobial Resistance (NAP-AMR), the first release implemented for 2017-2022 and the second in 2023-2028. NAP-AMR intends to minimize AMR delinquency and contribute to AMR global data [13, 14].

The prevalence of urinary tract infections (UTI) in Tanzania has been reported to range from 16% in children to 38-41% in adults [15-17]. Typical human body flora under favorable conditions can cause infections such as UTI, abscesses, furuncles, cellulitis, traveler's diarrhea, bacteremia, pneumonia, and neonatal meningitis [18]. The occurrence of infectious diseases in Tanzania may be due to poor water and hygiene sanitation (WASH) and antibiotic misuse [14]. Among the most frequently diagnosed bacterial infections in healthcare facilities at different levels are *E. coli* and *Staphylococcus aureus* [19]. Emerging AMR among these bacteria can contribute to the spread of AMR and transmit it to other bacteria, including pathogenic ones. Thus, we selected representative microbial agents to determine changes

in AMR or susceptibility patterns of clinical isolates due to ongoing interventions against AMR.

As a medicine regulator in Tanzania, TMDA has a direct role in ensuring the quality and safety of medicines by utilizing different strategies in registration, inspection, pharmacovigilance, and post-market surveillance to support the National Action Plan on Antimicrobial Resistance (NAP-AMR) together with the National Strategic Health Plan in line with Sustainable Development Goals (SDG) 2030 [20]. Therefore, this study aimed to assess the susceptibility of *E. coli* and *S. aureus* clinical isolates to selected antibiotics according to standard treatment guidelines (STG) to establish the magnitude of their AMR in four major referral regional hospitals in Tanzania.

2. Methodology

2.1. Study Location

The study was conducted in three zones of the country involving four referral hospitals, namely Western Zone (Maweni Regional Referral Hospital), Central Zone (Benjamin Mkapa Hospital and Morogoro Regional Hospital), and Eastern Zone (Temeke Referral Hospital).

2.2. Study Design and Sampling Strategy

A cross-sectional study design and purposive sampling were used (Kothari 2004). Individual hospitals were selected based on their capacity to perform bacterial cultures, isolation, and identification. Bacterial isolates of *E. coli* and *S. aureus* responsible for UTI were collected from hospital laboratories in selected hospitals. Samples were cultured and identified to obtain isolates, which were used to assess the prevalence of antibiotic resistance recommended in Tanzania's Standard Treatment Guideline 2022.

2.3. Sample Size Determination

The sample size was computed using a calculator developed by Statistics Kingdom (https://www.statskingdom.com/sample_size_chi2.html). The sample size computation followed a 95% Confidence Interval (CI) and a marginal error of 5%. Based on this calculation, 174 isolates were expected to be collected from the selected health facilities.

2.4. Sample Collection, Inclusion and Exclusion Criteria

2.4.1. Sample Collection

Positive *E. coli* and *S. aureus* isolates responsible for UTIs

were collected from respective health facility laboratories responsible for UTI cases between February and March 2023 for subsequent analyses.

2.4.2. Inclusion and Exclusion Criteria

All positive clinical isolates responsible for UTI in the healthcare facilities laboratory were included in this study.

2.5. Sample Isolation

Isolates responsible for UTI were collected and subcultured for colony identification of *S. aureus* and *E. coli*. Isolates were grown on cystine lactose electrolyte-deficient (CLED) (Oxoid) and MacConkey Agar (MCA) (Oxoid), respectively [21]. Gram Staining was performed to identify gram-positive and gram-negative bacteria [22]. For *S. aureus*, colonies with deep yellow colonies and gram-positive bacteria were selected and submitted to biochemical tests (Shields and Tsang, 2006) [23]. A total of hundred three (103) isolates of *E. coli* were collected from different regions based on the hospital's capacity to perform bacterial culture, isolation, and identification as follows 30 (29%) from Benjamin Mkapa Hospital, 12 (12%) from Temeke Regional Referral Hospital, 29 (28%) from Morogoro Regional Referral Hospital, and 32 (31%) from Maweni Regional Referral Hospital. A total of eighty four (84) isolates of *S. aureus* were collected from the four regions. 31(37%) were recruited from Morogoro Referral Hospital, 32(38%) from Maweni Hospital, 8(10%) from Benjamin Mkapa Hospital, and 13(15%) from Temeke Hospital. All isolates were transported to the TMDA Microbiology Laboratory in double-strength tryptic soy broth (TSB) at room temperature to the TMDA Microbiology Laboratory.

2.6. Laboratory Analysis

2.6.1. Biochemical Identification of *S. aureus*

Colonies with a deep yellow color on Cystine, lactose, electrolyte-deficient (CLED) Agar and positive Gram staining were collected for biochemical identification of *S. aureus*. Catalase and coagulase were used, as described by Ali et al. (2019) [24].

2.6.2. Biochemical Identification of *E. coli*

Red colonies on MacConkey agar (Oxoid) with negative gram staining were selected and subjected to biochemical tests. Oxidase, Sulfur Indole Motility (SIM), triple sugar iron (TSI), and urease tests were performed according to the procedures described by Ali (2019 and Brink, 2010) [24].

2.6.3. Recovery of Isolates from Transport Media (TSB)

Using the Streak method, 151 out of 187 isolates were recovered on selective media (mannitol salt agar and MacConkey Agar) after 24 h of incubation at 37 °C. Fifty-five (55) isolates of *S. aureus* and 96 isolates of *E. coli* were recovered. Pure cultures were subcultured in their respective culture media and incubated at 37 °C for 18-24 hours and the pure colonies were used for subsequent experiments.

2.6.4. Antibiotics and Reference Microorganisms

The selection of antibiotics used in this study was based on the current recommendations of the Tanzania Standard Treatment Guide for treating all types of tract infections. Selected antibiotics are listed in table 1.

Table 1. List and details of Antibiotic agents used.

Class	Antibiotic	Batch No.	Date of expire
Penicillin	Amox/Clav (AMC 20/10 µg), Piperacillin – tazobactam (TZP 100/10 µg), Amoxicillin (AML 10 µg)	3294941	2024/05/30
Aminoglycosides	Gentamicin (CN 10)	3261708	2024/03/17
Quinolones	Ciprofloxacin (CIP 5 µg)	3552317	2025/09/21
Cephalosporins	Ceftriaxone + sulbactam (CSE30 µg)		
	Ceftriaxone (CRO 30 µg)	3545371	2025/09/4
Nitrofurans	Nitrofurantoin (NIT 300 µg)	3538222	2025/08/18
Carbapenem	Meropenem (MEM 10 µg)	3524471	2023/07/19

2.6.5. Standard Control Microorganism

This study used two controls reference standard microorganisms as shown in table 2.

Table 2. List of reference bacteria used.

No.	Microorganism	Ref Number	Batch	Expire date
1	<i>E. coli</i>	ATCC 25922	Lot: 362251	28/03/2023
2	<i>S. aureus</i>	ATCC 25923	Lot: 421086	27/06/2023

2.6.6. Antimicrobial Susceptibility Test

Pure overnight *E. coli* and *S. aureus* colonies were tested for antibiotic susceptibility on Muller Hilton agar using the Kirby-Bauer method [26]. The disk diffusion method was employed according to the Clinical and Laboratory Standard Institute (CLSI) guidelines (32nd edition 2022). Fresh colonies of each isolate were cultured on Nutrient Agar (NA). They were standardized in phosphate-buffered saline (PBS) to make a suspension equivalent to 0.5 McFarland, approximately 1.0×10^8 CFU/mL. Suspensions were uniformly spread on Mueller-Hinton agar (MHA) using a sterile swab. Antibiotic discs (Oxoid) containing ceftriaxone, amoxicillin/clavulanic acid, amoxicillin, meropenem, gentamycin, nitrofurantoin, ciprofloxacin, piperacin/tazobactam, or ceftriaxone/sulbactam were used. Nine discs were used for each isolate, and no more than five discs were placed on a single MHA plate at a distance of 24 mm [27]. The plates were incubated aerobic at $35 \text{ }^\circ\text{C} \pm 2 \text{ }^\circ\text{C}$ for 16–18 h. A calibrated Vernier caliper was used to measure the zone of inhibition, and the results were interpreted as resistance (R), intermediate (I), or susceptible (S) according to the CLSI guideline 2022. *Staphylococcus aureus* ATCC 25923 and *Escherichia coli* ATCC 25922 were the controls.

2.7. Data Analysis

Data were captured in Microsoft Excel and analyzed using the Statistical Package for Social Sciences (SPSS) for Windows version 23. Categorical variables were summarized as proportions, and comparisons between groups were estimated using Pearson's chi-square test. Fisher's exact test was used when the total score was less than 20 ($n \leq 20$) or when the total score was less than five (5). Intermediate-sensitive isolates were considered fully sensitive during analysis. Odds ratios with their respective 95% confidence intervals (CI) were calculated to measure the strength of associations. A two-sided p-value of less than 0.05 was considered statistically significant.

3. Results

3.1. Sample Size and Response Rate

Determination of The sample size per hospital was determined by Mugenda and Mugenda (2003), who considered that a response rate of 50% is appropriate for analysis and reporting, a rate of 60% is reasonable, and a rate of 70% or more is excellent. A total of 151 clinical isolates were collected to register a response rate of 87.8% (Table 3). Response rates were considered excellent and accurate for statistical analysis.

Table 3. Hospital facilities response rate.

Hospital facility	Target	Actual	Response Rate (%)
BMH	43	35	81.4
MRH	43	53	123.3
MHK	43	45	104.7
TRH	43	18	41.9
Total	172	151	87.8

3.2. Sample Collection

A total of 151 clinical isolates were collected from four

health facilities. The isolates comprised 96 (63.6%) *E. coli* and 55 (36.4%) *S. aureus*. Morogoro Regional Referral Hospital showed the highest number of isolates ($n = 53$), while Temeke ($n = 18$) had the lowest number of isolates (Figure 1)

and (Table 4).

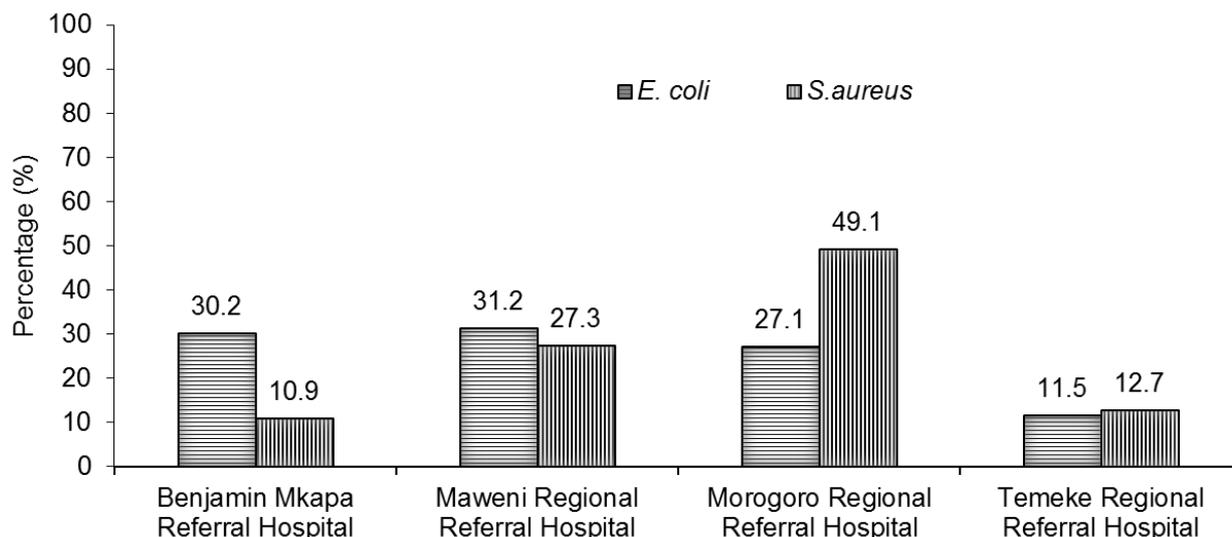


Figure 1. Bacterial isolates from four health facilities (n= 151).

Table 4. Distribution of collected bacterial isolates in relation to health facilities.

Health Facility	Isolates for <i>E. coli</i> n (%)	Isolates for <i>S. aureus</i> n (%)	Total isolates collected n (%)
BMH	29 (30.2)	6 (10.9)	35 (100)
MHK	30 (31.2)	15 (27.3)	45 (100)
MRH	26 (27.1)	27 (49.1)	53 (100)
TRH	11 (11.5)	7 (12.7)	18 (100)
Total	96 (63.6)	55 (36.4)	151 (100)

Key: BMH = Benjamin Mkapa Referral Hospital, MHK = Maweni Regional Referral Hospital, MRH = Morogoro Regional Referral Hospital, TRH = Temeke Regional Referral Hospital.

3.3. Isolation and Purification

The collected clinical isolates were identified and purified using biochemical tests; only pure isolates were used in subsequent studies.

3.4. Control Data for Standard Microorganism (Positive Control)

The results of quality control for susceptibility testing of reference microorganisms against selected antibiotics was performed using standard microorganism *Escherichia coli* ATCC 95222 and *Staphylococcus aureus* ATCC 25923 against all antibiotics in this study indicated susceptibility of

the control organism.

3.5. Prevalence of Antimicrobial Resistance

3.5.1. Prevalence of Antimicrobial Resistance Among *E. coli* Isolates

Findings revealed that *E. coli* showed varying resistance levels to all antibiotics. *E. coli* showed the highest resistance to amoxicillin (AML), peaking at 93%, whereas the lowest resistance, 1% was observed in meropenem (MEM), (Figure 2) where antimicrobial resistance patterns of *E. coli* isolates against the nine (9) antibiotics is illustrated.

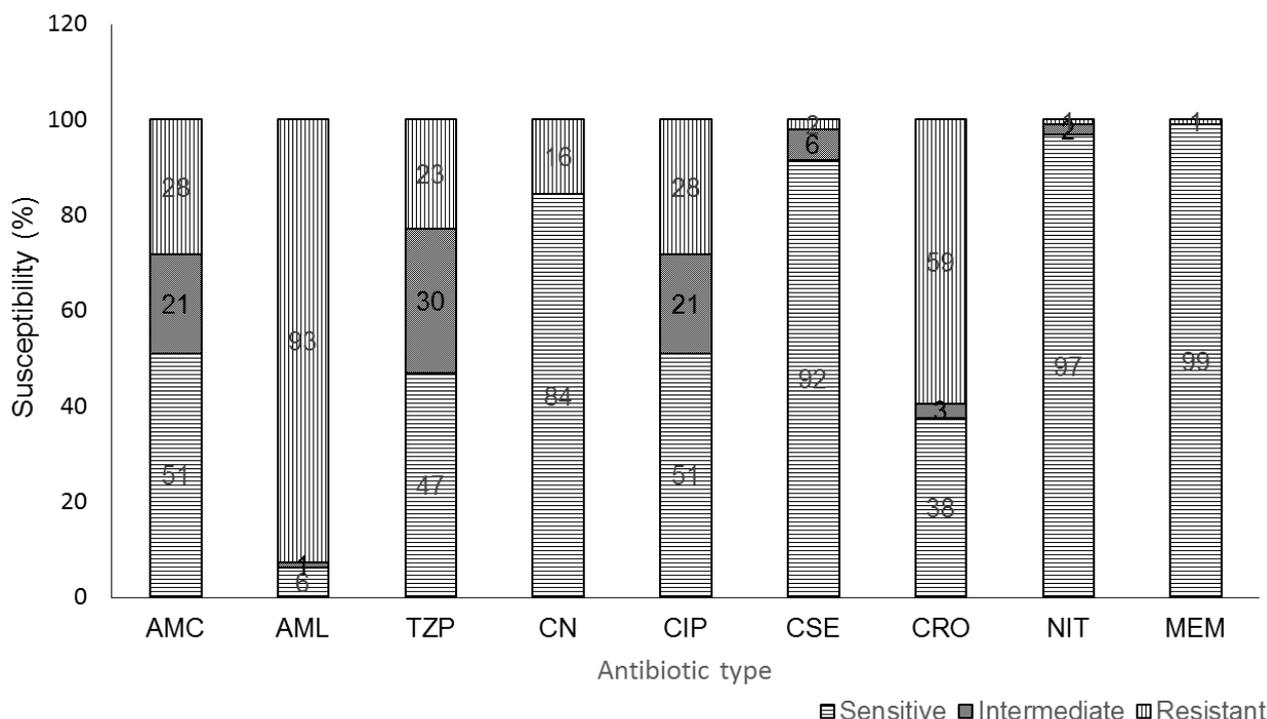


Figure 2. Overall resistance pattern of E. coli against nine antimicrobial agents (n=96).

3.5.2. Antimicrobial Resistance Pattern of E. coli

This study found a significant difference ($X^2=12.808$; p-value=0.002) in resistance across the selected antibiotics observed in which E. coli showed the highest resistance to amoxicillin (AML) and least resistance to meropenem where p-value<0.005 (Table 5).

Table 5. Antimicrobial resistance pattern of E. coli isolates across nine antibiotics.

Antibiotic	Sensitive n (%)	Intermediate n (%)	Resistant n (%)	X ²	p-value
Amoxicillin clavulanic acid	49 (51.0)	20 (10.8)	27 (28.2)		
Amoxicillin	6 (6.3)	1 (1.0)	89 (92.7)		
Piperacillin tazobactam	45 (46.9)	29 (30.2)	22 (22.9)		
Gentamycin	81 (84.4)	0(0)	15 (15.6)		
Ciprofloxacin	49 (51.0)	20 (20.8)	27 (28.2)	12.808	0.002
Ceftriaxone sulbactam	88 (91.7)	6 (6.3)	2 (2.1)		
Ceftriaxone	36 (37.5)	3 (3.1)	57 (59.4)		
Nitrofurantoin	93 (96.9)	2 (2.1)	1 (1.0)		
Meropenem	95 (99.0)	0 (0)	(1.0)		

3.5.3. Antimicrobial Resistance Among S. aureus Isolates

Resistance to S. aureus was observed for all five (5) selected antibiotics. S. aureus demonstrated the highest resistance against amoxicillin (AML), peaking at more than 91%, followed by ciprofloxacin (CIP) at 47.3%. Nitrofurantoin (NIT) demonstrated the highest efficacy, with least resistance (4%) (Figure 3).

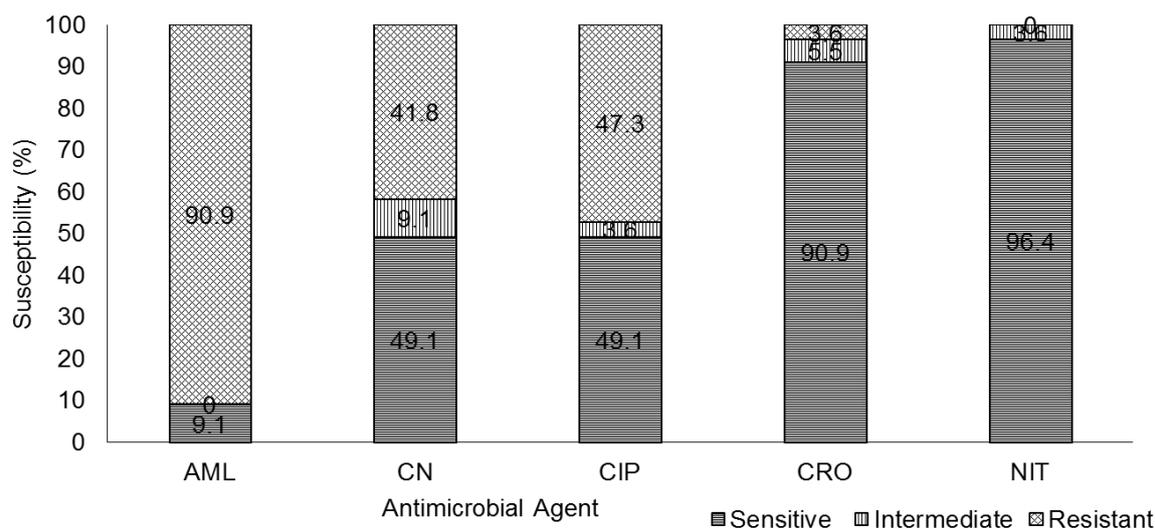


Figure 3. Overall susceptibility pattern of *S. aureus* against five antibiotics.

3.5.4. Antimicrobial Resistance Pattern of *S. aureus*

The analysis of the chi-square test indicated a significant difference ($\chi^2=53.627$, $p < 0.001$) in resistance across the selected antibiotics, in which *S. aureus* showed the highest resistance to amoxicillin (AML) and the least resistance to nitrofurantoin (NIT), where $p < 0.005$ (Table 6).

Table 6. Antimicrobial resistance pattern of *S. aureus* isolates across five antibiotics.

Antimicrobial agent	Sensitive n (%)	Intermediate n (%)	Resistant n (%)	χ^2	p-value
Amoxicillin	5(9.1)	0	50(90.9)		
Gentamycin	27(49.1)	5(9.1)	23(41.8)		
Ciprofloxacin	28(49.1)	2(3.6)	25(47.3)	53.627	<0.001
Ceftriaxone	50(90.9)	3(5.5)	2(3.6)		
Nitrofurantoin	53(96.4)	2(3.6)	0		

3.6. Multi-drug Resistance Among Isolates

Findings from this study showed that both *E. coli* and *S. aureus* isolates demonstrated multi-drug resistance (MDR) against selected antibiotics in all health facilities under study, in which isolates from Morogoro Regional Referral Hospital showed the highest (65.4%) MDR for *E. coli* and Benjamin Mkapa Hospital showed the highest (83.3%) for *S. aureus*. Similarly, Maweni Regional Referral Hospital demonstrated the lowest multi-drug resistance for *E. coli* (23%) and *S. aureus* (13%) (Figure 4).

A chi-square (χ^2) test for independence was performed to

determine whether there was any difference in MDR across the four health facilities. These findings demonstrate that the differences in MDR status across health facilities were statistically significant for *E. coli* ($\chi^2 = 10.301$; $p = 0.016$) and *S. aureus* ($\chi^2 = 11.673$; $p = 0.006$). Regarding MDR among *E. coli*, isolates Morogoro Regional Referral Hospital showed the highest (65.4%) and, Maweni Regional Referral Hospital demonstrated the lowest multi-drug resistance (23%) and Benjamin Mkapa Hospital showed the highest MDR in *S. aureus* (83.3%). Maweni Regional Referral Hospital demonstrated the lowest multi-drug resistance for *E. coli* (23%) and *S. aureus* (13%) (Table 7).

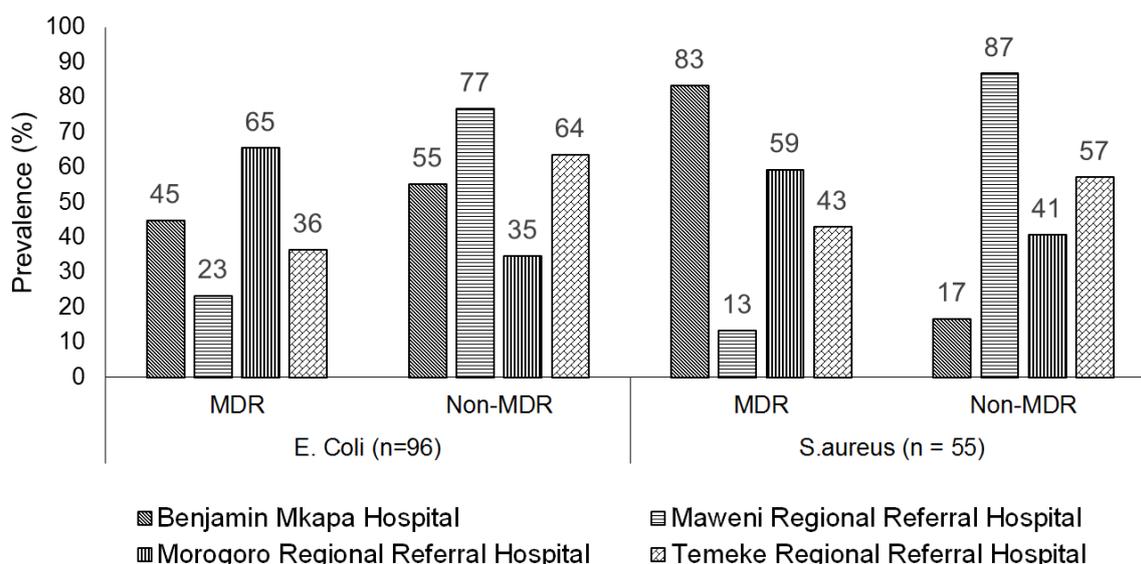


Figure 4. Prevalence of multi-drug resistance across selected health facilities.

Table 7. Multidrug-resistant patterns of isolated *E. coli* and *S. aureus* across health facilities.

Health facility	<i>E. coli</i>		X ²	P - value	<i>S. aureus</i> (%)		X ²	P- value
	MDR Status n (%)				MDR Status n (%)			
	Yes	No			Yes	No		
Benjamin Mkapa Hospital	13 (44.8)	16 (55.2)			5 (83.3)	1 (16.7)		
Maweni	7 (23.3)	23 (76.7)			2 (13.3)	13 (86.7)		
Morogoro Regional Referral Hospital	17 (65.4)	9 (34.6)	10.301	0.016	16 (59.3)	11 (40.7)	11.673	0.006
Temeke Regional Referral Hospital	4 (36.4)	7 (63.6)			3 (42.9)	4 (57.1)		
Total	41 (42.7)	55 (57.3)			26 (47.3)	29 (52.7)		

4. Discussion

This study assessed AMR among *E. coli* and *S. aureus* isolates from the urine of patients in selected health facilities against nine recommended antibiotics for treating UTI, according to the Standard Treatment Guidelines in Tanzania. Of the isolates (n=151) obtained, 96 (63.6%) and 55 (36.4%) were *E. coli* and *S. aureus*, respectively. This agrees with observations from other studies conducted in this locality and Sub-Saharan Africa, where *E. coli* was the predominant causative agent of UTIs [28]. *S. aureus* is not a commonly known causative agent of UTI. However, these bacteria cause ascending UTIs in patients with indwelling catheters or urinary tract instrumentation or patients who have recently undergone diagnostic cystoscopy and may experience transient bacteremia [29].

Of the six (6) antibiotic classes tested, the carbapenem showed the highest efficacy against *E. coli* (meropenem 99%), followed by nitrofurantoin (96.9%) and cephalosporin (ceftriaxone sulbactam 91.7%) where p-value<0.005. These findings are consistent with those reported previously [19].

In the cephalosporin class, ceftriaxone was shown to be less effective (37.5%) against *E. coli* compared to ceftriaxone sulbactam (91.7%), this difference in efficacy can be attributed to the action of sulbactam which prevents the β -lactamase activity of *E. coli*.

E. coli isolates showed the highest resistance to antibiotics belonging to the penicillin class (amoxicillin, 92.7%) and marked resistance to amoxicillin-clavulanic acid (28.2%). These findings are consistent with those of other studies [30]. The difference in resistance between amoxicillin (92.7%) and amoxicillin-clavulanic acid (28.2%), the same phenomenon has

been reported by others [31], who attributed the increased efficacy of AMC against *E. coli* to the inhibitory effect of clavulanic acid on β -lactamase in *E. coli*.

All isolates were highly susceptible to nitrofurantoin (96.4%) followed by ceftriaxone (90.9%). The high efficacy of nitrofurantoin is attributed to its multiple drug target sites that evade antimicrobial resistance. *S. aureus* showed high resistance to amoxicillin (90.9%). Followed by ciprofloxacin 47.2% and gentamycin 41.8%). Resistance to quinolone ciprofloxacin may be attributed to chromosome-mediated resistance from overprescribing quinolone antibiotics for treating bacterial infections [32].

Overall, multi-drug resistance, i.e., resistance by one species to antibiotics from at least three classes of antibiotics, was observed, *E. coli* (42.7%) and *S. aureus* (47.3%), respectively. A similar pattern was reported previously [9]. Resistance in *S. aureus* has been documented to be mediated by the synthesis of Beta-Lactamases, Penicillin Binding Proteins (PBP2A), and Mutation-Dependent Modification of PBP Proteins [33].

Resistance in *E. coli* is caused by bacterial influx pumps, beta-lactamase production, drug target modification, and antibiotic molecule modification [31]. The development of multi-drug antimicrobial resistance in both bacterial species may be driven by factors such as misuse of antibiotics in human health care due to improper prescription practices, as reported by [34] self-medication mediated by a shorter perceived distance to drug outlets and higher medical consultation fees, as documented by [34].

This study revealed *E. coli* as an MDR etiological agent for UTI, with a prevalence rate of n=41 (42.7%). Similar findings were reported [35] with a slight variation in the percentage prevalence. Antibiotic resistance in *E. coli* is caused by the bacterial influx pump, *Beta-lactamase* production, drug target modification, and antibiotic molecule modification [31].

This study revealed *S. aureus* as an MDR etiological agent for UTI, with a prevalence rate of n=26 (47.3%). This observation correlates with other findings from studies on the status of multi-drug resistance in *S. aureus* conducted in different parts of the world, with slight differences in prevalence [19, 24, 25, 36].

Escherichia coli isolates showed MDR to commonly used antibiotics among the selected, with Morogoro Regional referral hospital being the highest with 65.4%). In comparison, the least was Maweni Regional Referral Hospital, with 7% of all health facilities.

This study revealed that *S. aureus* isolates showed MDR as a commonly used antibiotic. Benjamin Mkapa Hospital had the highest rate at 83.3% and the lowest at Maweni Regional Referral Hospital at 7% in all health facility studies. This result indicates that the multi-drug resistance is vivid. This finding is consistent with those of other studies that showed a similar behavior of *S. aureus* against different classes of antibiotics.

5. Conclusion

This study found the existence of resistant and MDR isolates of *E. coli* and *S. aureus* to some antibiotics recommended in the Tanzania Standard Treatment Guidelines. Our findings call for continuous surveillance of AMR and implementation of antimicrobial stewardship at all hospital levels to identify and optimize antibiotic use against less-efficacious antibiotics still used in clinical practice. It also recommends a broader study on antibiotics currently in use to identify microorganisms that are highly resistant to and recommend their exclusion from treatment.

Abbreviations

CFU	Colony Forming Unit
AMR	Antimicrobial Resistance
LMICs	Low- and Middle-income Countries
UTI	Urinary Tract Infections
MDR	Multi-drug Resistance
AML	Amoxicillin
NIT	Nitrofurantoin
CIP	Ciprofloxacin
CN	Gentamicin
CSE	Ceftriaxone + Sulbactam
CRO	Ceftriaxone
MEM	Meropenem
TMDA	Tanzania Medicines and Medical Devices Authority
WHO	World Health Organization
NAP-AMR	National Action Plan on Antimicrobial Resistance
WASH	Water and Hygiene Sanitation
BMH	Benjamin Mkapa Hospital
MHK	Maweni Regional Referral Hospital
MRH	Morogoro Regional Referral Hospital
TRH	Temeke Regional Referral Hospital
SDG	Sustainable Development Goals
STG	Standard Treatment Guidelines
CLED	Cystine Lactose Electrolyte-deficient
MCA	MacConkey Agar
TSB	Tryptic Soy Broth
SIM	Sulfur Indole Motility
NA	Nutrient Agar
PBS	Phosphate-buffered Saline
CLSI	Clinical and Laboratory Standard Institute
ATCC	American Type Culture Collection

Author Contributions

Adelard Bartholomew Mtenga: Conceptualization, Formal Analysis, Investigation, Methodology, Supervision, Writing – original draft, Writing – review & editing

Adam Fimbo: Supervision, Writing – review & editing

Danstan Hipolite: Project administration, Writing – review & editing

Revocatus Makonope: Data curation, Investigation, Methodology

Saxon Mwambene: Data curation, Investigation, Methodology, Software

Yonah Hebron: Validation, Writing – review & editing

Kissa Mwamwitwa: Supervision, Validation, Writing – review & editing

Raphael Zozimus Sangeda: Supervision, Validation, Writing – review & editing

Funding Statement

This work did not receive any funds from funding agents and that all authors have none to declare.

Ethical Compliance

All procedures performed in studies did not involve human or animals and thus for this kind of research work ethical clearance was not required.

Data Summary

Research data for this publication is available for any kind of use, all data used to draw conclusion are provided in the manuscript as required.

Transparency Declaration

As the lead author on behalf of all authors, I confirm that the manuscript was developed based on the research data obtained from a genuine research work conducted and all information provided in this manuscript is honest, sincere and transparent account of the research to the best of our knowledge. Authors take fully responsibility of the information presented in this publication. All authors have none to declare.

Conflicts of Interest

The authors declare no conflicts of interest.

References

- [1] Denyer, S P., Hodges NA, Gorman SP, Gilmore BF. *Pharmaceutical Microbiology*. 8th ed. New Delhi: Wiley Blackwell Publishing House; 2011.
- [2] Ullah H, Ali S. Classification of Anti - Bacterial Agents and Their Functions. *Antibacterial Agents*. InTech; 2017. <https://doi.org/10.5772/intechopen.68695>
- [3] Camara N, Moremi N, Mghamba J, Eliakimu E, Shumba E, Ondo P, et al. Surveillance of antimicrobial resistance in human health in Tanzania: 2016–2021. *Afr J Lab Med*. 2023;12: 1–8. <https://doi.org/10.4102/ajlm.v12i1.2053>
- [4] Michael CA, Dominey-Howes D, Labbate M. The Antimicrobial Resistance Crisis: Causes, Consequences, and Management. *Front Public Heal*. 2014; 2. <https://doi.org/10.3389/fpubh.2014.00145>
- [5] Knobler L, Lemon SM, Najafi M. *The Resistance Phenomenon in Microbes and Infectious Disease Vectors*. Washington, D.C.: National Academies Press; 2003. <https://doi.org/10.17226/10651>
- [6] Sangeda RZ, William SM, Masatu FC, Bitegeko A, Mwalwisi YH, Nkiligi EA, et al. Antibiotic Utilisation Patterns in Tanzania: A Retrospective Longitudinal Study Comparing Pre- and Post-COVID-19 Pandemic Using Tanzania Medicines and Medical Devices Authority Data. *medRxiv*. 2023. <https://doi.org/10.1101/2023.11.27.23299060>
- [7] Tanzania Medicines and Medical Devices Authority (TMDA). *Drug Safety Bulletin 2020 1*. 2020 [cited 24 Jan 2024] pp. 1–16. Available at: <https://www.tmda.go.tz/uploads/publications/en1666874973-en1661782920-DRUG%20SAFETY%20A5%20.pdf>
- [8] Dadgostar P. Antimicrobial Resistance: Implications and Costs. *Infect Drug Resist*. 2019; Volume 12: 3903–3910. <https://doi.org/10.2147/IDR.S234610>
- [9] Ngowi BN, Sunguya B, Herman A, Chacha A, Maro E, Rugarabamu LF, et al. Prevalence of Multidrug Resistant UTI Among People Living with HIV in Northern Tanzania. *Infect Drug Resist*. 2021; Volume 14: 1623–1633. <https://doi.org/10.2147/IDR.S299776>
- [10] Silago V, Moremi N, Mtebe M, Komba E, Masoud S, Mgaya FX, et al. Multidrug-Resistant Uropathogens Causing Community Acquired Urinary Tract Infections among Patients Attending Health Facilities in Mwanza and Dar es Salaam, Tanzania. *Antibiotics*. 2022; 11: 1718. <https://doi.org/10.3390/antibiotics11121718>
- [11] Antimicrobial Stewardship—a practical guide to implementation in hospitals. *JAC-Antimicrobial Resist*. 2019; 1. <https://doi.org/10.1093/jacamr/dlz005>
- [12] Sangeda RZ, Kibona J, Munishi C, Arabi F, Manyanga VP, Mwambete KD, et al. Assessment of Implementation of Antimicrobial Resistance Surveillance and Antimicrobial Stewardship Programs in Tanzanian Health Facilities a Year After Launch of the National Action Plan. *Front Public Heal*. 2020;8: 454. <https://doi.org/10.3389/fpubh.2020.00454>
- [13] United Republic of Tanzania. *The National Action Plan on Antimicrobial Resistance 2017 - 2022*. 2017 [cited 6 Nov 2019]. Available: <https://www.afro.who.int/publications/national-action-plan-antimicrobial-resistance-2017-2022>
- [14] United Republic of Tanzania (URT). *The national action plan on antimicrobial resistance 2023-2028*. 2023. Available: <https://www.mifugouvuvu.go.tz/publications/37>

- [15] Sangeda RZ, Paul F, Mtweve DM. Prevalence of urinary tract infections and antibiogram of uropathogens isolated from children under five attending Bagamoyo District Hospital in Tanzania: A cross-sectional study. *F1000Research*. 2021; 10: 449. <https://doi.org/10.12688/f1000research.52652.1>
- [16] Mlugu EM, Mohamedi JA, Sangeda RZ, Mwambete KD. Prevalence of urinary tract infection and antimicrobial resistance patterns of uropathogens with biofilm forming capacity among outpatients in morogoro, Tanzania: a cross-sectional study. *BMC Infect Dis*. 2023; 23: 660. <https://doi.org/10.1186/s12879-023-08641-x>
- [17] Schmider J, Bühler N, Mkwatta H, Lechleiter A, Mlaganile T, Utzinger J, et al. Microbiological Characterisation of Community-Acquired Urinary Tract Infections in Bagamoyo, Tanzania: A Prospective Study. *Trop Med Infect Dis*. 2022; 7: 100. <https://doi.org/10.3390/tropicalmed7060100>
- [18] Davis CP. Normal Flora. In: Baron S editor. *MM 4th edition*. G (TX): U of TMB at G 1996. C 6. A from: <https://www.ncbi.nlm.nih.gov/books/NBK7617> Medical Microbiology. 4th edition.
- [19] Dasgupta C, Rafi MA, Salam MA. High prevalence of multidrug resistant uropathogens: A recent audit of antimicrobial susceptibility testing from a tertiary care hospital in Bangladesh. *Pakistan J Med Sci*. 2020; 36: 1–6. <https://doi.org/10.12669/pjms.36.6.2943>
- [20] Tanzania Medicines and Medical Devices Authority (TMDA). Good regulatory practices for medical products. 2023 [cited 24 Jan 2024]. Available: https://www.tmda.go.tz/uploads/publications/en1678801576-GOOD%20REGULATORY%20PRACTICE%20GUIDELIN_E_FINAL.pdf
- [21] Karah N, Rafei R, Elamin W, Ghazy A, Abbara A, Hamze M, et al. Guideline for Urine Culture and Biochemical Identification of Bacterial Urinary Pathogens in Low-Resource Settings. *Diagnostics*. 2020; 10: 832. <https://doi.org/10.3390/diagnostics10100832>
- [22] Shah HN, Gharbia SE, Collins MD. The Gram stain. *Rev Med Microbiol*. 1997; 8: 103. <https://doi.org/10.1097/00013542-199704000-00006>
- [23] Shields P, Tsang AY. Mannitol Salt Agar Plates Protocols. In: American Society for Microbiology [Internet]. 2006 [cited 24 Jan 2024] pp. 3–5. Available: <https://asm.org/ASM/media/Protocol-Images/Mannitol-Salt-Agar-Plates-Protocols.pdf?ext=.pdf>
- [24] Ali M. Prevalence of Staphylococcus Species from Clinical Samples Obtained from Some Hospitals on Kano Metropolis, Nigeria. *Am J Biomed Sci Res*. 2019; 5: 207–211. <https://doi.org/10.34297/AJBSR.2019.05.000913>
- [25] Brink B. Urease Test Protocol - Library. 2010; 1–7.
- [26] Odoch T, Wasteson Y, L'Abée-Lund T, Muwonge A, Kankya C, Nyakarahuka L, et al. Prevalence, antimicrobial susceptibility and risk factors associated with non-typhoidal Salmonella on Ugandan layer hen farms. *BMC Vet Res*. 2017; 13: 365. <https://doi.org/10.1186/s12917-017-1291-1>
- [27] Hudzicki J. Kirby-Bauer Disk Diffusion Susceptibility Test Protocol Author Information. In: American Society For Microbiology [Internet]. 2012 [cited 24 Jan 2024] pp. 1–13. Available: <https://asm.org/getattachment/2594ce26-bd44-47f6-8287-0657aa9185ad/Kirby-Bauer-Disk-Diffusion-Susceptibility-Test-Protocol-pdf.pdf>
- [28] Mwang'onde BJ, Mchami JI. The aetiology and prevalence of urinary tract infections in Sub-Saharan Africa: a Systematic Review. *J Heal Biol Sci*. 2022; 10: 1. <https://doi.org/10.12662/2317-3076jhbs.v10i1.4501.p1-7.2022>
- [29] Mlynarczyk-Bonikowska B, Kowalewski C, Krolak-Ulinska A, Marusza W. Molecular Mechanisms of Drug Resistance in Staphylococcus aureus. *Int J Mol Sci*. 2022; 23: 8088. <https://doi.org/10.3390/ijms23158088>
- [30] Fredrick F, Francis JM, Fataki M MS. Aetiology, antimicrobial susceptibility and predictors of urinary tract infection among febrile under-fives. *Afr J Microbiol Res*. 2013; 7: 1029–1034. <https://doi.org/10.5897/ajmr12.1866>
- [31] Rozwadowski M, Gawel D. Molecular Factors and Mechanisms Driving Multidrug Resistance in Uropathogenic Escherichia coli—An Update. *Genes (Basel)*. 2022; 13: 1397. <https://doi.org/10.3390/genes13081397>
- [32] Shariati A, Arshadi M, Khosrojerdi MA, Abedinzadeh M, Ganjalishahi M, Maleki A, et al. The resistance mechanisms of bacteria against ciprofloxacin and new approaches for enhancing the efficacy of this antibiotic. *Front Public Heal*. 2022; 10. <https://doi.org/10.3389/fpubh.2022.1025633>
- [33] Majumder MMI, Mahadi AR, Ahmed T, Ahmed M, Uddin MN, Alam MZ. Antibiotic resistance pattern of microorganisms causing urinary tract infection: a 10-year comparative analysis in a tertiary care hospital of Bangladesh. *Antimicrob Resist Infect Control*. 2022; 11: 156. <https://doi.org/10.1186/s13756-022-01197-6>
- [34] Mabilika RJ, Shirima G, Mpolya E. Prevalence and Predictors of Antibiotic Prescriptions at Primary Healthcare Facilities in the Dodoma Region, Central Tanzania: A Retrospective, Cross-Sectional Study. *Antibiotics*. 2022; 11: 1035. <https://doi.org/10.3390/antibiotics11081035>
- [35] Subramanya SH, Bairy I, Metok Y, Baral BP, Gautam D, Nayak N. Detection and characterization of ESBL-producing Enterobacteriaceae from the gut of subsistence farmers, their livestock, and the surrounding environment in rural Nepal. *Sci Rep*. 2021; 11: 2091. <https://doi.org/10.1038/s41598-021-81315-3>
- [36] Fredrick F, Francis JM, Fataki M, Maselle SY. Aetiology, antimicrobial susceptibility and predictors of urinary tract infection among febrile under-fives at Muhimbili National Hospital, Dar es Salaam-Tanzania. *African J Microbiol Res*. 2013; 7: 1029–1034. <https://doi.org/10.5897/AJMR12.1866>