

# **RESEARCH ARTICLE**

# Antimicrobial Resistance Status Among Referred Patients Attending at Nova Diagnostic and Research in Mogadishu, Somalia

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### ABSTRACT

Background: Bacterial infections remain a major global health concern, contributing significantly to morbidity and mortality. While antimicrobials have been effective in treating these infections, the rising threat of antimicrobial resistance (AMR) has complicated management strategies. Methodology: This study investigates the antimicrobial resistance (AMR) patterns of bacterial isolates from clinical specimens at Nova Diagnostics and Research in Mogadishu, Somalia, over a three-year period from June 2021 to July 2024. Result: A total of 650 patient specimens were analyzed, with bacterial growth identified in 647 samples. Among these, 98.2% were Gram-negative and predominantly aerobic, while 1.4% were Gram-positive, all aerobic. The most frequently isolated pathogens were Staphylococcus spp. (43.4%), followed by Escherichia coli (17.2%), Klebsiella (13.1%), and Pseudomonas (7.2%). Antimicrobial resistance was particularly concerning among Gram-negative bacteria. Staphylococcus exhibited high resistance to erythromycin (66.6%) and tetracycline (50.7%). Escherichia coli showed 84.8% resistance to ampicillin, while Klebsiella demonstrated 90.5% resistance to ampicillin. Gram-positive bacteria commonly exhibited resistance to Erythromycin, Trimethoprim/Sulfamethoxazole, Rifampicin, and Vancomycin. Conclusion: The high prevalence of resistant strains, particularly among Gram-negative bacteria like Staphylococcus spp. and Escherichia coli, presents significant challenges to effective treatment. The widespread resistance to multiple antibiotics underscores the urgent need for enhanced infection control, improved surveillance systems and researches, and comprehensive antimicrobial stewardship programs.

Keywords: Bacterial infection; antimicrobial resistance; gram negative bacteria; bacterial isolates; disc diffusion technique

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### **1. Introduction**

Bacterial infection represents a major cause of morbidity and mortality across the world<sup>[1]</sup>. The severity of these infections ranges from mild cases to life-threatening conditions such as meningitis, sepsis, and chronic wound infections<sup>[2]</sup>. High priority pathogens including Salmonella, Pseudomonas and Enterococcus possess significant public health threat, resulting treatment failure.

An East African country, Somalia, face substantial disease burden due to bacterial infection, which affects their quality of life and economy profoundly<sup>[3]</sup>. Infections caused by bacterial species like *Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, Salmonella*, and *Pseudomonas aeruginosa* are predominant in this region, particularly among children, the elderly, and individuals with compromised immune systems<sup>[4]</sup>. However, pathogen prevalence and susceptibility profiles exhibit considerable geographic variation, as well as significant differences across different populations and environments.

While antimicrobials have been effective in treating bacterial infections, rise of antimicrobial resistance (AMR) has emerged as a global health concern. The increasing resistance to antibiotics has significantly complicated the management of bacterial infections. Several factors contribute to the high rates of AMR, including the misuse and overuse of antimicrobial agents, availability of over the counter drugs, poor sanitation, and malnutrition<sup>[5]</sup>. Furthermore, the widespread indiscriminate use of antibiotics in agriculture and livestock has exacerbated the issue, diminishing the effectiveness of these drugs in combating bacterial infections<sup>[6]</sup>.

Bacteria acquire resistance through complex mechanism such as chromosomal mutation, gene transfer, plasmid exchange, transformation and transduction. Additionally, gram negative bacteria have the ability to evolve resistance patterns within bacterial populations and across different species<sup>[7]</sup>. Reports indicate that many pathogens isolated from different specimen have extended range of resistance against multiple antimicrobial agents like Ampicillin, Cefazolin, Erythromycin, Tetracycline etc<sup>[8-9]</sup>. So there is an urgent need for enhanced surveillance, targeted research, improved infection control measures, and the implementation of effective antimicrobial stewardship programs to address this escalating issue. Given the circumstances, this study aims to look into the AMR status of the common bacterial isolates from Nova Diagnostics and Research, Mogadishu, Somalia.

### 2. Materials and Methods

This cross-sectional study was conducted at the department of Microbiology, Nova Diagnostics and Research, Mogadishu, Somalia over a three-year period from June 2021 to July 2024. The study was aimed to evaluate the antimicrobial resistance status of bacterial isolates obtained from several clinical specimens of patients attending the outpatient department. A total of 650 laboratory culture results were examined including specimens from blood, urine, skin swab, ear swab, throat swab, wound swab, high vaginal swab (HVS), pus, sputum, and cerebrospinal fluid (CSF). The susceptibility test was performed following the Clinical Laboratory Standards Institute guidelines (CLSI). Socio-demographic data were collected from laboratory records.

### 2.1. Isolation and identification of pathogenic bacteria

To isolate the concerned pathogenic microorganisms, clinical samples were collected using standard microbiological techniques. Based on the source of each specimen, samples were inoculated onto various media, including MacConkey agar, blood agar, chocolate agar, mannitol salt agar, Xylose lysine deoxycholate (XLD) agar, and Thayer-Martin agar (Oxoid, UK). The inoculated plates were incubated at

37°C for 24 hours. Colony characteristics such as color, shape, elevation, surface texture, and opacity were recorded for each medium. For initial identification, Gram staining was performed to observe the size, shape, and arrangement of the suspected isolated colonies. To identify the bacterial species, automated analysis using the VITEK® 2 COMPACT system was employed. A bacterial suspension was prepared in 0.45% saline, standardized for turbidity, and used with the appropriate identification card.

#### 2.2. Antibiotic susceptibility assay of bacterial isolates

Antimicrobial susceptibility testing was performed on Mueller–Hinton agar using the disk diffusion technique according to the Kirby–Bauer method. The antimicrobial agents tested included amoxicillin ( $10\mu g$ ), ampicillin ( $10\mu g$ ), ceftriaxone ( $30\mu g$ ), ciprofloxacin ( $5\mu g$ ), erythromycin ( $15\mu g$ ), tetracycline ( $30\mu g$ ), and gentamicin ( $10\mu g$ ).

#### 2.3. Quality control

To ensure optimal performance of the VITEK® 2 Compact system, regular calibration was performed, focusing on colorimetric accuracy and temperature control. For antimicrobial susceptibility testing (AST), antibiotic gradients and Minimum Inhibitory Concentration (MIC) values were monitored and compared against control strains in accordance with Clinical and Laboratory Standards Institute (CLSI) guidelines. Automated data validation and regular software updates kept the system current, while external proficiency testing ensured accuracy and reliability.

#### 2.4. Statistical analysis

Statistical analyses in this study were conducted with the statistical package for the social sciences (SPSS). The descriptive variables were presented as frequencies and percentages (%). P value of <0.05 was considered to statistically significant.

### 3. Results

A total of 650 specimens from the patients were collected, among them bacterial growth were identified in 647 samples. Of the participants, 348 (53.5%) were male, while 302 (46.5%) were female (Table 1). The study subjects were categorized into age groups in 10-year clusters, with the 21-30 age group comprising 22.3% of the participants and exhibiting the highest number of infections (Table 2). Among the isolated microorganisms, 638 (98.2%) were Gram-negative, with 98.5% aerobic and 4.7% anaerobic. In contrast, 1.4% was Gram-positive bacteria, all of which were aerobic. Pathogenic bacterial growth was most commonly found in skin swabs (36.6%), followed by urine (22.8%), cerebrospinal fluid (CSF) (13.8%), pus (6.8%), and stool samples (4.9%). The presence of pathogenic bacteria was notably lower in ear swabs (4.8%), high vaginal swabs (HVS) (2.9%), semen samples (2.8%), wound swabs (2.2%), and blood samples (0.3%). Additionally, only 0.2% of pathogenic bacteria were detected in throat and oral swabs (Figure 1).

### 3.1. Distribution of bacterial isolates

Among the bacterial isolates, *Staphylococcus spp.* was predominant, accounting for 43.4%, with 40.1% primarily identified in skin swabs. It was also isolated from cerebrospinal fluid (CSF) (14.5%), pus (8.5%), ear swabs (6.4%), and semen (5.7%). In urine samples, Escherichia coli were the most common isolate (47.3%), followed by *Klebsiella* (20%) and *Staphylococcus* (18.8%). Notably, 14.1% of Klebsiella was isolated from stool samples, while 9.8% of *Escherichia coli* were found in both stool and cerebrospinal fluid (CSF). Additionally, a very low number of Enterobacter, Enterococcus, Proteus, and Sphingomonas species were also detected (Table 3).

### 3.2. Antimicrobial resistance index of Gram-negative bacteria

Among Gram-negative bacteria, Staphylococcus species exhibited resistance the most to Erythromycin (66.6%) and Tetracycline (50.7%). Additionally, 84.8% of *Escherichia coli* isolates were resistant to Ampicillin, and 68.7% showed resistance to Trimethoprim/Sulfamethoxazole. *Klebsiella* species demonstrated 90.5% resistance to Ampicillin, while *Pseudomonas* species exhibited 87.2% resistance to Cefazolin (Table 4).

### 3.3. Antimicrobial resistance index of Gram-positive bacteria

Overall, Gram-positive bacterial isolates commonly exhibited resistance to Erythromycin, Gentamicin, and Tetracycline (Table 4).

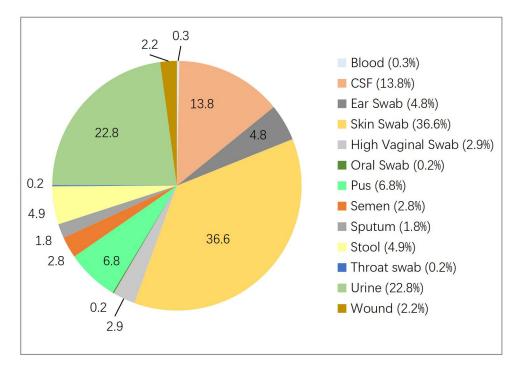


Figure 1. Proportions of sample collection sites.

Types of Isolates	Total (%)	Male (%)	Female (%)	P-Value
Acinetobacter	4 (0.6)	2 (0.3)	2 (0.3)	0.559
Burkholderia	4 (0.6)	3 (0.5)	1 (0.2)	0.004
Citrobacter	5 (0.8)	5 (0.8)	-	NA
Enterobacter	18 (2.8)	10 (1.5)	8 (1.2)	0.099
Enterococcus	10 (1.5)	4 (0.6)	6 (0.9)	0.974
Escherichia	112 (17.2)	52 (8.0)	60 (9.2)	0.715
Klebsiella	85 (13.1)	47 (7.2)	38 (5.8)	0.001
Kocuria	4 (0.6)	3 (0.5)	1 (0.2)	0.576
Proteus	30 (4.6)	17 (2.6)	13 (2.0)	0.064
Pseudomonas	47 (7.2)	24 (3.7)	23 (3.5)	0.487
Sphingomonas	13 (2.0)	6 (0.9)	7 (1.1)	0.861

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Types of Isolates	Total (%)	Male (%)	Female (%)	P-Value
Staphylococcus	282 (43.4)	159 (24.5)	123 (18.9)	0.905
Streptococcus	6 (0.9)	1 (0.2)	5 (0.8)	0.287
Others	27 (4.2)	14 (2.2)	13 (2.0)	0.717
No Growth	3 (0.5)	1 (0.2)	2 (0.3)	0.266
Total	650 (100)	348 (53.5)	302 (46.5)	0.085

Table 1. (Continued)

All results were expressed as number (%). Statistical difference was calculated by using student's unpaired 't' test. A p<0.05 was taken as level of significance.

	Age distribution										
Microorganisms	≤10	11-20	21-30	31-40	41-50	51-60	61-70	≥71			
Enterobacter (n=18)	3 (16.7)	1 (5.6)	6 (33.3)	2 (11.1)	1 (5.6)	1 (5.6)	3 (16.7)	1 (5.6)			
Enterococcus (n=10)	-	3 (30.0)	1 (10.0)	-	-	3 (30.0)	2 (20.0)	1 (10.0)			
Escherichia (n=112)	9 (8.0)	18 (16.1)	17 (15.2)	13 (11.6)	11 (9.8)	13 (11.6)	12 (10.7)	19 (17.0)			
Klebsiella (n=85)	11 (12.9)	7 (8.2)	17 (20.0)	14 (16.5)	6 (7.1)	11 (12.9)	3 (3.5)	16 (18.8)			
Proteus (n=30)	9 (30.0)	6 (20.0)	3 (10.0)	3 (10.0)	4 (13.3)	-	1 (3.3)	4 (13.3)			
Pseudomonas (n=47)	8 (17.0)	9 (19.1)	7 (14.9)	5 (10.6)	7 (14.9)	3 (6.4)	6 (12.8)	2 (4.3)			
Sphingomonas (n=13)	1 (7.7)	-	2 (15.4)	2 (15.4)	3 (23.1)	2 (15.4)	1 (7.7)	2 (15.4)			
Staphylococcas (n=282)	47 (16.7)	55 (19.5)	79 (28.0)	36 (12.8)	18 (6.4)	27 (9.6)	10 (3.5)	10 (3.5)			
Streptococcus (n=06)	-	-	3 (50.0)	1 (16.7)	-	-	2 (33.3)	-			
Others (n=44)	6 (13.6)	5 (11.4)	9 (20.5)	5 (11.4)	3 (6.8)	8 (18.2)	6 (13.6)	2 (4.5)			
No growth (n=03)	-	-	1 (33.3)	1 (33.3)	-	-	-	1 (33.3)			
Total	94 (14.5)	104 (16.0)	145 (22.3)	82 (12.6)	53 (8.2)	68 (10.5)	46 (7.1)	58 (8.9)			

Table 2. Age distribution of common bacterial isolates among the patients.

All results were expressed as number (%).

Microorganisms	Blood	CSF	Ear swab	Skin swab	HVS	Oral swab	Pus	Semen	Sputum	Stool	Throat swab	Urine	Wound
Enterobacter (n=18)	-	4 (22.2)	1 (5.6)	5 (27.8)	-	-	1 (5.6)	-	1 (5.6)	5 (27.8)	-	-	1 (5.6)
Enterococcus (n=10)	-	2 (20.0)	-	6 (60.0)	-	-	-	-	-	-	-	2 (20.0)	-
Escherichia (n=112)	-	11 (9.8)	1 (0.9)	24 (21.4)	3 (2.7)	-	7 (6.3)	1 (0.9)	-	11 (9.8)	-	53 (47.3)	1 (0.9)
Klebsiella (n=85)	-	7 (8.2)	3 (3.5)	27 (31.8)	3 (3.5)	1 (1.2)	6 (7.1)	-	7 (8.2)	12 (14.1)	-	17 (20.0)	2 (2.4)
Proteus (n=30)	-	4 (13.3)	2 (6.7)	17 (56.7)	-	-	2 (6.7)	-	-	1 (3.3)	-	2 (6.7)	2 (6.7)
Pseudomonas (n=47)	-	5 (10.6)	4 (8.4)	27 (57.4)	-	-	2 (4.3)	-	1 (2.1)	-	-	4 (8.5)	4 (8.5)
Sphingomonas (n=13)	-	5 (38.5)	-	-	1 (7.7)	-	-	1 (7.7)	1 (7.7)	-	-	5 (38.5)	-
Staphylococcus (n=282)	2 (0.7)	41 (14.5)	18 (6.4)	113 (40.1)	9 (3.2)	-	24 (8.5)	16 (5.7)	1 (0.4)	-	1 (0.4)	53 (18.8)	4 (1.4)

Table 3. Bacterial distribution across different sample collection sites.

All results were expressed as number (%).

Antibiotics	Enterobacter	Enterococcus	Escherichia	Klebsiella	Proteus	Pseudomonas	Sphingomonas	Staphylococcas	
	(n=18)	(n=10)	(n=112)	(n=85)	(n=30)	(n=47)	(n=13)	(n=282)	
Amoxicillin	1 (5.5)	-	4 (3.5)	-	1 (3.3)	1 (2.1)	-	-	
Ampicillin	-	-	95 (84.8)	77 (90.5)	17 (56.6)	-	-	1 (0.3)	
Cefazolin	17 (94.4)	-	69 (61.6)	41 (48.2)	10 (33.3)	41 (87.2)	4 (30.7)	-	
Cefepime	2 (11.1)	-	57 (50.8)	28 (32.9)	6 (20)	4 (8.5)	4 (30.7)	-	
Ceftriaxone	7 (38.8)	-	64 (57.1)	34 (40)	9 (30)	1 (2.1)	5 (38.4)	-	
Ciprofloxacin	3 (16.6)	2 (20)	55 (49.1)	17 (20)	9 (30)	10 (21.2)	5 (38.4)	49 (17.3)	
Erythromycin	-	4 (40)	-	-	-	-	-	188 (66.6)	
Gentamicin	3 (16.6)	3 (30)	26 (23.2)	13 (15.2)	8 (26.6)	3 (6.38)	3 (23)	13 (4.6)	
Levofloxacin	4 (22.2)	-	52 (46.4)	16 (18.8)	10 (33.3)	10 (21.2)	2 (15.3)	-	
Nitrofurantoin	1 (5.5)	-	10 (8.9)	15 (17.6)	27 (90)	2 (4.2)	-	-	
Piperacillin/ Tazobactum	-	-	29 (25.8)	14 (16.4)	-	6 (12.7)	3 (23)	-	
Tetracycline	-	8 (80)	-	-	-	-	-	143 (50.7)	
Frimethoprim/ Sulfamethoxazole	5 (27.7)	-	77 (68.7)	36 (42.3)	14 (46.6)	4 (8.5)	4 (30.7)	87 (30.8)	
Vancomycin	4 (22.2)	-	-	-	-	-	-	26 (9.2)	

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Table 4. Frequency of antimicrobial resistance of common bacteria for selected antimicrobial classes.

All results were expressed as number (%).

## 4. Discussion

In the recent years, the rise of bacterial diseases due to AMR is a growing concern around the world. The consequences are highly concerning especially for public health, healthcare system, and economy. Therefore, surveillance on AMR is crucial to preserve the effectiveness of current antibiotics, in order to restore a quality life.

In the present study, a total of 650 subjects were recruited to observe their AMR patterns against several antimicrobial agents. The subjects included were 53.5% male and 46.5 % female. Among the age groups, patients between 21 and 30 were found infected the most with different bacterial infections (22.3%) (Table 2).

Different pathological species were found in the patients, predominantly S. *aureus, E.coli, K. pneumonae, P. aerogenosa*, all comprising gram negative bacteria. Notably, *Staphylococcus aureus* was the most frequently isolated bacteria, accounting for 43.4% of all infections among the subjects. This finding aligns with other studies that infection with staphylococcus is very common in the East African countries<sup>[3,5,10]</sup>. Additionally, *E.coli, S. aureus, K. pneumonae*, and *P. aerogenosa* have a major contribution in hospital acquired infections, which highlights the importance of strict infection control practices <sup>[11]</sup>. The rate of infection was found more among subjects who were 40 or below, with age group 21-30 exhibiting the highest rate (28.0%). No significant difference between male and female subjects was seen (p=0.905) (Table 1). Only a few subjects with gram positive bacteria like *Enterococcus* were observed (1.5%).

In regard to sample type, skin swab was most frequently received, representing 36.6% of the total subjects (Figure 1). This suggests an increasing trend of skin infections in Somalia. Among the skin swab isolates, Staphylococcus aureus was the most commonly identified pathogen, accounting for 47.4% of cases. In addition to skin swabs, samples were also collected from urine (22.8%) and cerebrospinal fluid (CSF) (13.8%) (Figure 1). Urinary tract infections (UTIs) were prevalent, with *E.coli* and *S. aureus* being the most frequently isolated organisms, each exhibiting a prevalence of 35.8%. Among the CSF samples, S. aureus was identified in 54.4% of cases, followed by E. coli, which was present in 12.2% of the isolates. The detection of bacteria in the CSF is indicative of serious conditions such as bacterial sepsis, brain abscess, and bacterial meningitis, as described by Honda and Warren (2009)<sup>[12]</sup>.

The resistance profile of the common pathogens against multiple antimicrobial agents was assessed as the key investigatory finding. Antimicrobials such as Ampicillin, Cefazolin, Ceftrixone, Erythromycin, Tetracyclin, and Trimethoprim/Sulfamethoxazole are some of the common agents evaluated. The most resistant microorganisms against several of these agents were Escherichia and Klebsiella spp. Ampicillin demonstrated the highest level of resistance against them, being 84.8% of Escherichia spp. and 90.5% of Klebsiella spp. Followed by are Trimethoprim/Sulfamethoxazole, Ceftriaxone and Cefazolin which also exhibited significant proportions of resistance across these isolates (Table 4). Another report also showed high prevalence of multiple resistances of *Escherichia* and *Klebsiella spps*<sup>[13]</sup>. Escherichia and Klebsiella species are classified as uropathogens due to their significant role in UTI, and their resistance against multiple drugs is alarming. In this study, these pathogens were widely present in urine samples as well as skin sample, indicating immunosuppression/ healthcare associated infection.

Staphylococcus spp also exhibited very high resistance against few agents. Along with S. aureus, S. epidermidis and S. haemolyticus were also found widely resistant against these agents. Notably, resistance was specifically high against Erythromycin (66.6%) and Tetracyclin (50.7%). Erythromycin is a macrolide

antibiotic which was found resistant against some strains of *S. aureus* called Macrolide-resistant *Staphylococcus aureus* and Methicillin-resistant *Staphylococcus aureus* (MRSA)<sup>[14-15]</sup>. A similar pattern of resistance was observed in a study conducted in Somalia. However, the other study also found resistance of Staphylococcus against beta-lactams such as Ampicillin and Amoxicillin, which did not congruent with our findings<sup>[16]</sup>.

Other pathogens like Pseudomonas, Proteus and Enterobacter were also found resistant against some agents. Almost all pathogens demonstrated moderate levels of resistance against agents like Ciprofloxacin, Gentamycin, and Trimethoprim/Sulfamethoxazole, suggesting antibiotic susceptibility testing for effective treatment.

## 5. Conclusion

This study underscores the growing concern of antimicrobial resistance (AMR) and its significant impact on public health, the healthcare system, and the economy. The high prevalence of bacterial infections, primarily caused by *Staphylococcus, Escherichia coli, Klebsiella,* and *Pseudomonas*, is concerning. Significant levels of resistance were observed against antibiotics, including ampicillin, trimethoprim/sulfamethoxazole, ceftriaxone, cefazolin, erythromycin, and tetracycline. Addressing the rising AMR emphasizes the urgent need for effective surveillance and infection control measures.

# **Disclosure of conflict of interest**

No competing interests exist by the authors. This manuscript has not submitted to, nor is under review at another journal or other publishing venue.

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