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FREQUENCY AND ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF BACTERIAL ISOLATES IN URINE AND PUS SAMPLES IN A HEALTH-CARE SETTING

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Abstract

Background: *Bacterial infections, particularly urinary tract and wound infections, are significant healthcare concerns, while increasing antimicrobial resistance has made their treatment increasingly challenging. This study aimed to determine the frequency of bacterial pathogens isolated from urine and pus samples and to evaluate their antimicrobial susceptibility patterns in a healthcare setting.*

Methods: *A cross-sectional study was conducted in the Microbiology Department of Arif Memorial Teaching Hospital over a six-month period from August 2024 to January 2025. A total of 151 clinical specimens, comprising 75 urine and 76 pus samples, were collected using convenient sampling. Samples were processed using standard microbiological procedures, including culture on MacConkey and Blood agar, Gram staining, and biochemical tests. Antimicrobial susceptibility testing was performed using the Kirby-Bauer disk diffusion method on Mueller-Hinton agar according to Clinical and Laboratory Standards Institute (CLSI) guidelines. Data were analyzed using SPSS version 25.0.*

Results: *Of the 151 samples, 110 (72.84%) were culture positive. Females constituted 60.93% of the study population, while males accounted for 39.07%, with the 31–45-year age group being the most represented (31.13%). Escherichia coli was the most frequently isolated pathogen (28.48%), followed by Staphylococcus aureus (20.53%), Pseudomonas aeruginosa (9.93%), Klebsiella pneumoniae (7.28%), and Enterobacter spp. (6.62%). High antimicrobial resistance was observed against ceftriaxone and nalidixic acid (100%), ampicillin (98.5%), penicillin (96.7%), trimethoprim-sulfamethoxazole (96.1%), and colistin*

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(94.1%). In contrast, vancomycin showed 100% sensitivity, while cefoxitin and linezolid demonstrated 96.3% and 80% sensitivity, respectively.

Conclusion: *The study demonstrates a high prevalence of bacterial infections and concerning levels of antimicrobial resistance among commonly isolated pathogens. The predominance of E. coli and S. aureus, along with high resistance to several commonly used antibiotics, emphasizes the need for routine antimicrobial susceptibility testing, continuous resistance surveillance, prudent antibiotic use, and appropriate infection-control measures to support effective treatment and improve patient outcomes.*

INTRODUCTION

Bacteria play an important role in human health but are also responsible for severe infections worldwide (1). Infectious diseases account for up to 25% of global deaths annually, in which urinary tract and wound infections are commonly observed and are mainly caused by *Escherichia coli*, *Klebsiella*, *Pseudomonas* and *Staphylococcus aureus* (2). Healthcare-associated infections (HAIs) contribute significantly to the global burden, with prevalence rates up to 25% in developing countries compared to 5–15% in developed nations (3,4). The increasing threat of antimicrobial resistance (AMR) has worsened the problem due to which Gram-negative bacteria have a fatality rate of above 45% in critically ill individuals (5), while resistant gram-positive pathogens remain equally concerning, leading the World Health Organization (WHO) to classify them into priority group (6).

Urinary tract infections (UTIs) are among the most prevalent infectious diseases, with *Escherichia coli (E. coli),* being the primary causative agent (7). Wound infections also remain a major clinical and public health concern, with common pathogens including *Staphylococcus aureus* (particularly MRSA), *Enterococcus faecium*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae* (8).

Antimicrobial susceptibility testing (AST) is important for guiding effective therapy, as it not only identifies the most appropriate antibiotic but also helps monitor developing resistance patterns and improve antibiotic stewardship strategies (9,10). However, antibiotic overuse increases resistance through processes such as decreased drug absorption, target modification, and enzyme inactivation (11,12). Antimicrobial resistance (AMR) is now recognized as a major global health threat, with estimates of nearly 10 million annual deaths by 2050, representing not only a significant clinical challenge but also an economic and public health crisis worldwide (13,14). In countries like Pakistan where resources are limited, the problem becomes even more serious due to weak surveillance, un-controlled antibiotic use, and a lack of strong healthcare facilities (15). This increase in resistance among both community- and hospital-acquired pathogens prolongs hospital stays, increases costs which results in limited therapeutic options (16). This study aims to identify the bacterial pathogens most found in urine and pus samples and to evaluate their antimicrobial resistance patterns through biochemical methods and susceptibility testing.

METHOD

This cross-sectional study was conducted at the Microbiology Department of Arif Memorial Teaching Hospital. The study was cross-sectional and was carried out over a period of 6 months, from August 2024 to January 2025. The sampling technique was Convenient Sampling. After taking consent from patients, a total of 151 clinical specimens, including urine and pus samples, were collected for bacteriological analysis. Only properly labeled and aseptically collected samples were included, while contaminated specimens were excluded. Midstream urine samples

were collected in sterile wide-mouthed containers after appropriate instructions to patients. Pus samples were obtained by sterile

swabs, depending on the site of infection. All specimens were transported immediately to the laboratory and processed within two hours of collection. Samples were inoculated on MacConkey and Blood agar plates, followed by incubation at 37 °C for 18–24 hours. Bacterial growth was identified based on colony morphology, Gram staining, and a series of standard biochemical tests, including catalase, urease and triple sugar iron reactions, as appropriate for the suspected organism. Antimicrobial susceptibility testing was performed using the Kirby-Bauer disk diffusion method on Mueller-Hinton agar following Clinical and Laboratory Standards Institute (CLSI) guidelines. A panel of commonly prescribed antibiotics was tested, including ampicillin (10 µg), amoxicillin-clavulanate (20/10 µg), ceftriaxone (30 µg), ceftazidime (30 µg), gentamicin (10 µg), amikacin (30 µg), ciprofloxacin (5 µg), levofloxacin (5 µg), imipenem (10 µg), meropenem (10 µg), vancomycin (30 µg) and linezolid (30 µg). The diameter of inhibition zones was measured and interpreted as sensitive, intermediate, or resistant according to CLSI standards. Statistical Package for Social Sciences (SPSS) version 25.0 was used for data entry and analysis. Expected quality metrics of data were mean, frequency and percentages.

RESULT

A total of 151 patients were included, with 92 (60.9%) females and 59 (39.1%) males. Participants were distributed across five age groups, with the highest representation in the 31–45 years group (31.1%) and 16–30 years (25.8%).

Out of 151 clinical samples (urine, pus, and pus-related categories), 110 (72.5%) were culture positive. *Escherichia coli* was the most common isolate (28.5%), followed by *Staphylococcus* aureus (20.5%), *Pseudomonas aeruginosa* (9.9%), *Klebsiella pneumoniae* (7.3%), and *Enterobacter spp.* (6.6%). Both Gram-positive and Gram-negative bacteria were significantly represented.

Antibiotic susceptibility testing revealed high resistance rates to several commonly used antibiotics. Nalidixic acid and ceftriaxone showed complete resistance (100%), while ampicillin, penicillin, and trimethoprim-sulfamethoxazole exhibited resistance above 96%. Aztreonam, azithromycin, and colistin had resistance rates above 85%, whereas gentamicin and ciprofloxacin showed moderate resistance (80.9% and 76.2%) respectively. In contrast, vancomycin was fully effective (100% sensitivity), cefoxitin showed 96.3% sensitivity, and linezolid 80% sensitivity.

Table 1: Age wise Distribution of Patients

Age	Frequency	Percentage
Up to 15	14	9.27%
16-30	39	25.83%
31-45	47	31.13%
46-60	29	19.21%
< 80	22	14.57%

Table 2: Gender Wise Distribution among Patients

Gender	Frequency	Percentages
Male	59	39.07%
Female	92	60.93%
Total	151	100%

Table 3: Types and Frequency of Samples submitted for culture (n=151)

Types of Samples	No. of Samples submitted	Frequency
Urine	75	49.67%

Pus (swabs, ear discharge, sputum, wound, HVS, abscesses)	76	50.3%

Table 4: Frequency of Isolated Pathogens (n=151)

Isolated Pathogens	Frequency
Escherichia Coli	28.48%
Staphylococcus Aureus	20.53%
Pseudomonas Aeruginosa	9.93%
Klebsiella Pneumonia	7.28%
Enterobacter	6.62%

Table 5: Culture Result

Culture	Frequency	Percentage
Positive	110	72.84%
Negative	41	27.15%

Table 6: Antibiotic Resistance Pattern of Drugs on Bacterial Pathogens

Sr. No	Antibiotics	Resistant (%)	Sensitive (%)
1	Aztreonam	88.20%	10.30%
2	Gentamycin	80.90%	19.10%
3	Septran	96.10%	3.90%
4	Vancomycin	0.00%	100%
5	Penicillin	96.70%	3.30%
6	Ciprofloxacin	76.20%	22.60%
7	Cefoxitin	3.70%	96.30%
8	Clindamycin	69.00%	20.70%
9	Azithromycin	86.50%	10.80%
10	Colistin	94.10%	5.90%
11	Nalidixic Acid	100%	0.00%
12	Ampicillin	98.50%	1.50%
13	Ceftriaxone	100%	0.00%
14	Ceftazidime	86.50%	13.50%
15	Linezolid	20.00%	80.00%

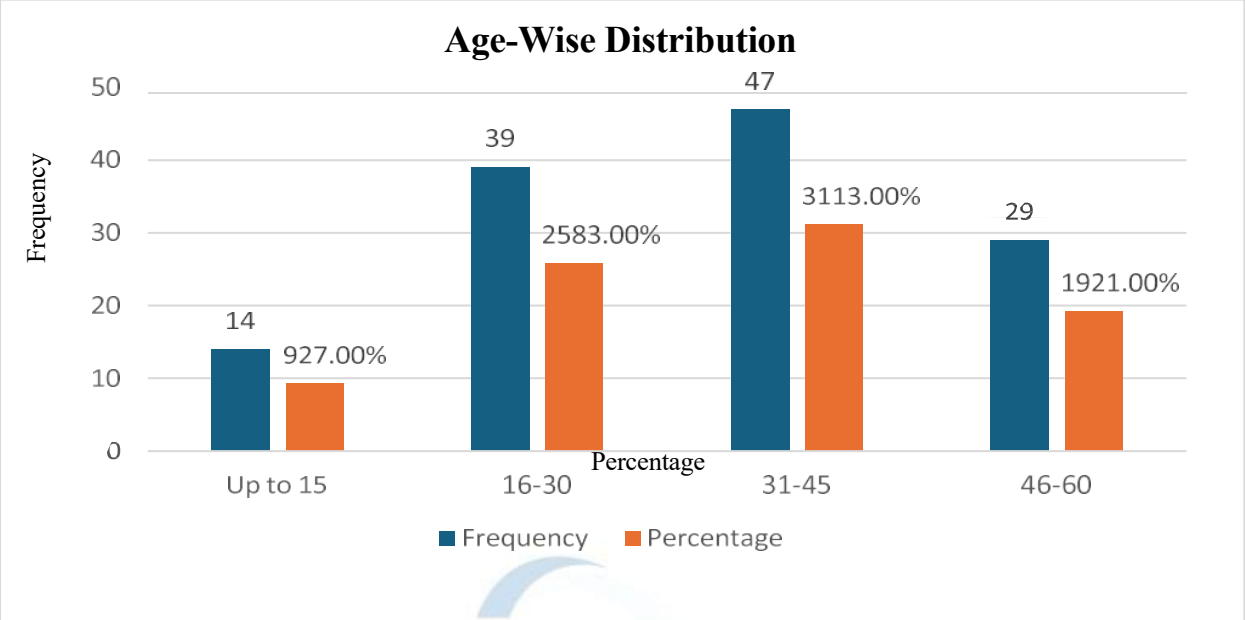


Figure 1: Bar Chart showing Age Wise Distribution

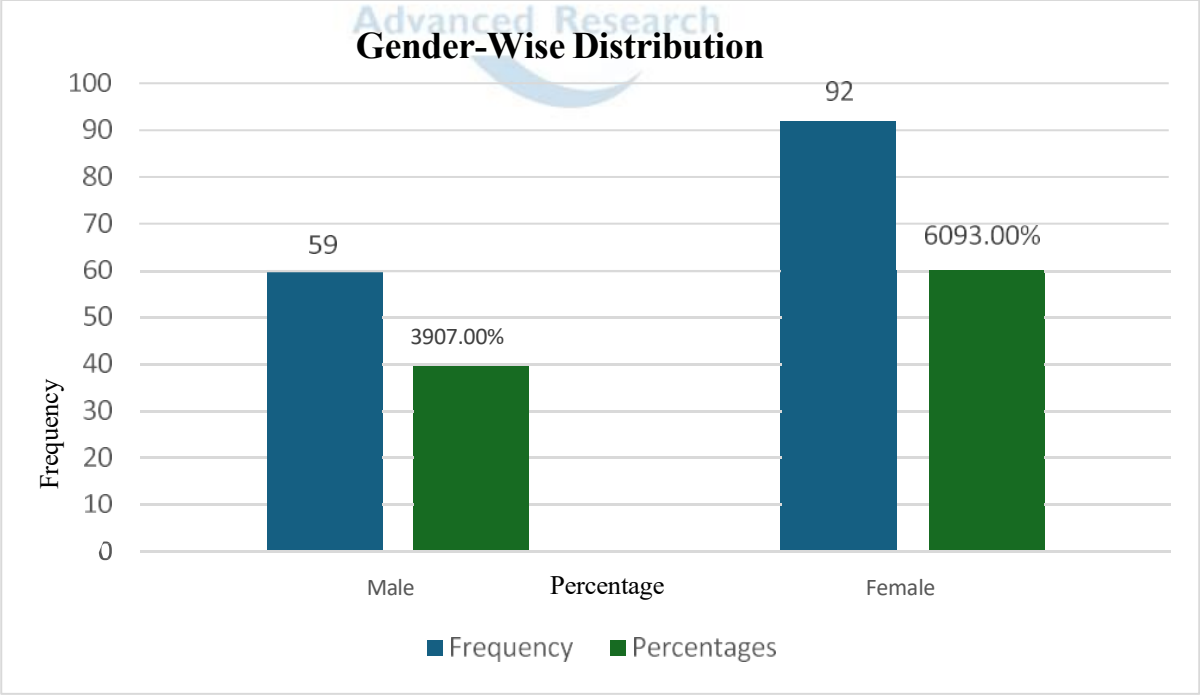


Figure 2: Bar Chart showing Gender Wise Distribution

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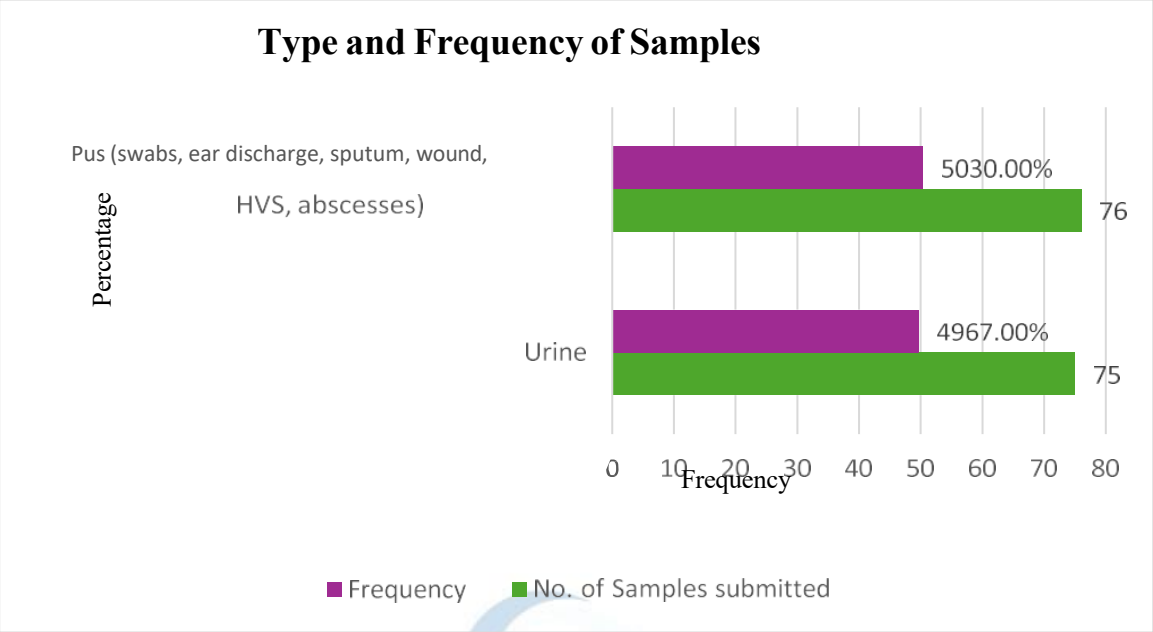


Figure 3: Bar Chart showing Types and Frequency of samples submitted for Culture

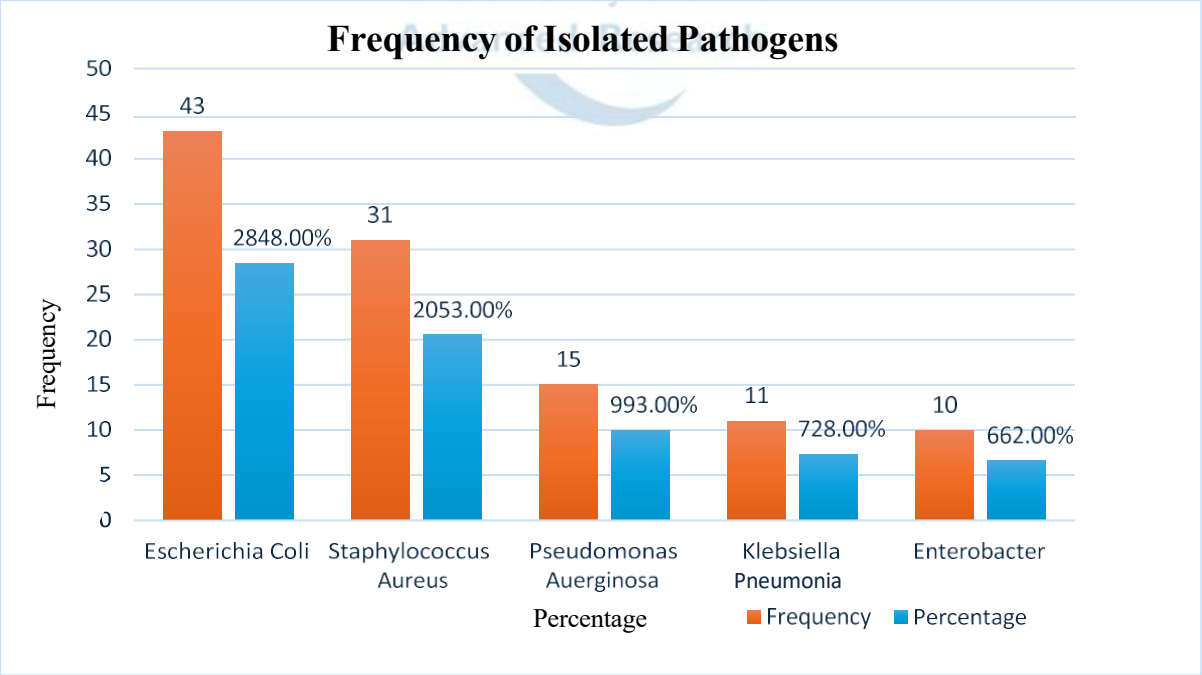


Figure 4: Bar Chart showing Frequency of Pathogens

DISCUSSION

Accurate identification of bacterial isolates in clinical samples is critical for diagnosing infections and guiding efficient treatment, culture, and biochemical tests to identify species and select appropriate antibiotics (17). The increase of multidrug-resistant (MDR) bacteria, caused by misuse
or overuse of antimicrobials, affects the management of common infections such as UTIs, wounds, and pus infections, especially in hospitalized or immunocompromised patients (18).

This study demonstrates that urinary tract and wound infections remain a major clinical concern. Escherichia coli was the predominant isolate, confirming its global role as the leading cause of UTIs, while

Staphylococcus aureus and *Pseudomonas aeruginosa* were also significant contributors, highlighting the persistent presence of both Gram-positive and Gram-negative pathogens.

Patients aged 16–45 years represented the majority, reflecting higher exposure to environmental risk factors. Female predominance aligns with known anatomical and physiological susceptibility to UTIs.

Antimicrobial susceptibility testing revealed alarming resistance patterns. Nalidixic acid and ceftriaxone were completely ineffective, with high resistance also observed for ampicillin, penicillin, and trimethoprim-sulfamethoxazole. Gentamicin and ciprofloxacin showed moderate resistance, whereas vancomycin, cefoxitin, and linezolid retained high efficacy, particularly against Gram-positive organisms.

A similar study at the Microbiology Laboratory of a tertiary care hospital in Karachi from October 2021 to December 2021 analyzed 2,209 clinical samples, with 14.9% pus and 15.9% urine. Among culture-positive isolates, Gram-positive cocci, *Staphylococcus aureus* and *Enterococcus spp*. were resistant to penicillin and erythromycin but sensitive to vancomycin, while Gram-negative rods *E. coli* and *Klebsiella pneumoniae* were resistant to ciprofloxacin and ceftriaxone and sensitive to polymyxin-B and tigecycline (19).

This study is limited by its single-center setting, small sample size, and testing of only commonly used antibiotics. Additionally, factors such as comorbidities and prior antibiotic use were not considered, which may affect infection outcomes. Healthcare facilities should strengthen infection control measures and perform regular antimicrobial susceptibility testing to guide treatment. Efforts to monitor and limit antibiotic overuse are essential, and larger, more comprehensive studies are needed to better understand resistance patterns.

CONCLUSION

This study concludes that infections remain major public health concern, with *Escherichia coli*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa* being the most common pathogens. The study found high infection rates, particularly among females and the 31–45 age group, with a culture positivity rate of 72.5%. Antibiotic susceptibility testing revealed significant resistance to commonly used drugs such as nalidixic acid, ceftriaxone, and ampicillin, while vancomycin and linezolid remained highly effective. These findings highlight the growing threat of antimicrobial resistance and underscore the need for routine surveillance, prudent antibiotic use, and targeted treatment strategies to improve patient outcomes.

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