

Bacteriological Profile and Antibiotic Susceptibility Patterns of Asymptomatic UTI among Female Students of R.R.M Campus Janakpur, Madhesh Pradesh, Nepal

Chandrika Sah^{1†} Ranjit Kumar Sah^{1†}

¹Ramswaroop Ramsagar Multiple Campus, Tribhuvan University, Janakpur, Nepal

[†]These authors contributed equally.

*Corresponding author: Chandrika Sah, Ramswaroop Ramsagar Multiple Campus, Tribhuvan University, Janakpur, Madhesh Pradesh, Nepal Email: linkchandrika4@gmail.com

ABSTRACT

Objective: The primary goal of this study was to identify common pathogenic microorganisms and their patterns of antibiotic susceptibility from asymptomatic UTI cases.

Methods: A Total of 151 mid-stream urine samples were collected from female students for bacteriological identification and antimicrobial susceptibility testing. Conventional biochemical tests identified the isolates, and their antibiotic susceptibility was determined according to the Clinical and Laboratory Standards Institute (CLSI) guidelines.

Results: Of the 151 samples analyzed, 30.46% exhibited significant bacterial growth, with *Staphylococcus aureus* being the most prevalent (30.43%), followed by *Escherichia coli* (17.39%) and *Klebsiella pneumoniae* (10.86%). Gram-negative bacterial isolates showed a higher level of resistance to amoxicillin (33%-100%), amoxicillin (60%-100%), amoxicillin-clavulanic acid (20%-100%), ceftriaxone (60%-100%), cotrimoxazole (40%-100%), nitrofurantoin (40%-100%), and ciprofloxacin (40%-100%). Piperacillin and Tazobactam, levofloxacin, imipenem, and amikacin were less resistant. Gram-positive bacterial uropathogens showed a high level of resistance to amoxicillin (65%-100%), ciprofloxacin (50%-100%), levofloxacin (50%-100%), and clindamycin (64%-100%). Imipenem, gentamicin, piperacillin, and tazobactam have shown reduced resistance.

Conclusion: Uropathogens were found to be highly prevalent, and bacterial isolates were shown to be highly resistant to routinely recommended medications. In order to prevent an asymptomatic infection from developing into a symptomatic UTI, routine UTI screening, regular health education on the risk of asymptomatic infectious diseases for females in the reproductive age group, and antibiotic susceptibility testing should be implemented.

Keywords: UTI, antibiotic resistance, asymptomatic, Janakpur Dham

INTRODUCTION

Urinary tract infections (UTIs) rank among the most prevalent bacterial infections in medical practice, affecting individuals of all ages and posing significant public health challenges due to their widespread occurrence, potential complications, and the growing issue of antimicrobial resistance (Hooton et al., 1996; WHO, 2025). The clinical spectrum of UTIs ranges from symptomatic infections to asymptomatic bacteriuria

(ABU), the latter often remaining undetected due to the absence of clear clinical symptoms, yet it can result in adverse outcomes if left untreated (Changizi Maryam et al., 2014). Adult women are particularly susceptible, with 40–50% experiencing at least one UTI episode in their lifetime, with incidence rising with age and during pregnancy (Geerlings, 2016).

Both community-acquired and hospital-acquired UTIs are primarily caused by Gram-negative bacteria

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such as *Escherichia coli*, *Klebsiella pneumoniae*, and *Proteus species*, with *Staphylococcus saprophyticus* and *Enterococcus faecalis* also contributing to the infection landscape (Foxman, 2014; Geerlings, 2016). Symptoms typically include painful urination and increased frequency, but a significant number of individuals may have substantial bacteriuria without symptoms, known as asymptomatic urinary tract infection (AUTI) or asymptomatic bacteriuria (ABU) (Foxman, 2014). The asymptomatic nature of ABU, particularly among young adults like college students, complicates timely diagnosis and treatment, potentially leading to complications such as pyelonephritis and the spread of antibiotic-resistant bacteria (Changizi Maryam et al., 2014; Nicolle et al., 2019).

Empirical antibiotic treatment is often initiated before culture results are available, contributing to selective pressures that foster the development and spread of multidrug-resistant (MDR) and extended-spectrum beta-lactamase (ESBL) producing organisms in both community and healthcare settings (Flores-Mireles et al., 2015; Fouad & Boraie, 2016). Variations in resistance patterns across regions underscore the need for localized surveillance to guide empirical treatment decisions and stewardship initiatives (Ronald & Pattullo, 1991). Despite the importance of such data, there is a lack of research on the prevalence and resistance patterns of AUTIs among college students in Nepal and similar settings.

METHODS

Study Design and Population

This study employed a cross-sectional design targeting students enrolled at the R.R.M. Campus in Janakpur, Nepal. The study population consisted of female students without signs or symptoms of urinary tract infection, aged 18 to 35 years.

Sample Collection and Laboratory Analysis

Urine samples were collected from participants who

provided consent, using the standard midstream clean-catch technique to reduce contamination. Each sample was analyzed with quantitative culture methods to detect significant bacteriuria ($\geq 10^5$ CFU/mL), and bacterial species were identified through established biochemical and microbiological techniques (Foxman, 2014; Ipe, 2013).

Antimicrobial susceptibility testing was performed using the Kirby-Bauer disk diffusion method, with results interpreted based on established clinical breakpoints. The antibiotics tested included commonly prescribed agents for UTIs, such as ampicillin, amoxicillin-clavulanic acid, nitrofurantoin, ciprofloxacin, and trimethoprim-sulfamethoxazole (Institute, 2023; Mandal et al., 2012; Odoki et al., 2019).

Data Analysis

Descriptive statistics were used to determine the prevalence of asymptomatic bacteriuria (ABU) in the study population, stratified by sex, age, and academic faculty. The distribution of bacterial isolates and their resistance patterns was analyzed to elucidate the local epidemiological landscape of the disease.

Ethical Considerations

Informed consent was obtained from all participants. Confidentiality and anonymity were maintained throughout data collection and analysis.

RESULTS

Out of 151 urine samples, 46 (just over 30%) showed bacterial growth, indicating a potential asymptomatic UTI. Another 35 samples (about 23%) had some bacteria, but not enough to be considered as significant bacteriuria. Rest of the 70 samples (over 46%) had no detectable bacteria at all. Which accounts nearly half of the students.

Among the isolated bacteria, the Gram-negative bacteria were found to be the highest in number, followed by Gram-positive bacteria (Table No. 1).

Table 1: Finding of Gram stain.

Gram Stain	Total No	Percentage (%)
Gram-Negative Bacteria	24	52.17
Gram-Positive Bacteria	22	47.82
Total	46	100

The analysis of student growth across faculties reveals a total of 46 students, with the following distributions: B.Ed. and BBA 17.39 % each, led by 8 students, and

B.Sc. CSIT 13.04% followed by B. Sc. Microbiology, B.Sc. Science and BBA contribute 5 students (10.86%) (Table No. 2).

Table 2: Positive Growth of Participants by Faculty.

Faculty of Significance	Numberof Positive Growth	Percentage (%)
B.Ed. (21)	8	38.09
BBS (31)	8	38.09
BBA (38)	5	13.15
B.Sc. Microbiology (16)	5	31.25
B.sc CSIT (6)	6	100
B.Sc. SCIENCE (15)	5	33.33
M.B.S(Master Finance) (14)	4	28.57
BIT (8)	3	37.5
BICTE (2)	2	100
Total (151)	46	30.46

Growth According to Marital Status.

The following are the growth results based on marital status (Table No. 3). A total of 6 married students

(total married, n=22) showed significant growth, while 40 unmarried students' sample didn't show any significant growth.

Table 3: Growth According to Marital Status.

Martial Status	Significant Growth (%)	Insignificant Growth (%)	No Growth (%)
Married (22)	6 (27.27)	6 (27.27)	10 (45.45)
Un Married (129)	40 (31.00)	29 (22.48)	60 (46.51)
Total (151)	46 (30.46)	35 (23.17)	70 (46.35)

Bacteriological Profile

Staphylococcus aureus emerged as the predominant isolate (30.4%), accounting for the majority of

positive cultures, followed by *E. coli* (17.3%), *Klebsiella pneumoniae* (10.8%), *Staphylococcus saprophyticus*, and *Enterococcus faecalis* (Table No. 4).

Table 4: The Bacteria Found in Urine Culture.

Identification of Bacteria	Culture Result	
	Numberof Cases	Percentage (%)
<i>Staphylococcus aureus</i>	14	30.43
<i>Escherichia coli</i> (ESBL Positive)	8	17.39
<i>Klebsiella pneumoniae</i> (SBL Producer)	5	10.86
Coagulase-negative staphylococci (CoNS)	4	8.69
<i>Acinetobacter</i> spp	3	6.52
<i>Enterococcus faecalis</i>	3	6.52
<i>Morganella</i> spp	2	4.34
<i>Pseudomonas</i> spp	2	4.34
<i>Pseudomonas aeruginosa</i>	2	4.34
<i>Citrobacter koseri</i>	1	2.17
<i>Escherichia coli</i>	1	2.17
<i>Enterococcus</i> spp	1	2.17
Total	46	100

Antimicrobial Resistance Patterns

The percentage distribution of resistance patterns of Gram-negative bacterial isolates is shown in Table No. 5. The susceptibility test result for *E. coli* shows that more than 50% isolates were resistant to amoxicillin, cotrimoxazole, ceftazidime, ciprofloxacin, and levofloxacin, while none of them were resistant

to piperacillin/tazobactam and imipenem. Most of the Gram-negative isolates were sensitive to the piperacillin/tazobactam and imipenem, while moderately sensitive to other antibiotics. Out of the tested antibiotics, Gram-positive bacterial isolates were highly resistant to ampicillin, amoxicillin/clavulanic acid, co-trimoxazole, and clindamycin (Table No. 6).

Table 5: Percentage distribution of Resistance patterns for each Gram-Negative isolate to commonly used antibiotics.

Antimicrobial Agent	<i>Citrobacter Koseri</i> Resistance (%) (N=1) (n)	<i>E. coli</i> Resistance (%) (N=9) (n)	<i>K. pneumoniae</i> Resistance (%) (N= 5) (n)	<i>P. aeruginosa</i> Resistance (%) (N=2) (n)	<i>Acinetobacter</i> spp Resistance (%) (N=3) (n)	<i>Morgenella</i> spp Resistance (%) (N=2) (n)	<i>Pseudomonas</i> spp Resistance (%) (N=2) (n)
AK	0	11.11 (1)	0	0	0	100	0
AMP	100	22.22 (2)	20 (1)	-	0	50 (1)	50 (1)
AMX	100	66.66 (6)	60 (3)	100	33.33 (1)	100	100
AMC	100	33.33 (3)	20 (1)	100	33.33 (1)	100	100
CTR	0	44.44 (4)	100	50 (1)	33.33 (1)	100	100
COT	0	66.66 (6)	40 (2)	50 (1)	33.33 (1)	0	100
CPM	0	55.55 (5)	80 (4)	50 (1)	0	100	0
CFM	0	11.11 (1)	-	0	-	-	-
CIP	0	77.77 (7)	40 (2)	50 (1)	100	0	0
GEN	0	22.22 (2)	20 (1)	100	33.33 (1)	100	0
NIT	100	22.22 (2)	40 (2)	50 (1)	100	0	100
LE	0	66.66 (6)	80 (4)	50 (1)	33.33 (1)	100	0
PIT	0	0	0	0	0	50 (1)	0
IPM	0	0	0	0	0	0	0
AZM	-	-	-	-	33.33 (1)	-	-
CX	-	11.11 (1)	-	-	0	-	-
CD	-	11.11 (1)	-	-	-	50 (1)	-
DO	-	-	-	-	-	0	-
MRP	-	11.11 (1)	-	-	33.33 (1)	0	-
E	-	-	-	-	66.66 (2)	-	-
CAZ	-	88.88 (8)	100	100	33.33 (1)	50 (1)	100
CAC	-	55.55 (5)	20 (1)	50 (1)	0	50 (1)	100
TE	-	-	0	-	33.33 (1)	-	-
P	-	11.11 (1)	20 (1)	0	33.33 (1)	50 (1)	0

Amikacin (AK), Ampicillin (AMP), Amoxicillin (AMX), Amoxyclav (AMC), Ceftriaxone (CTR), Co-Trimoxazole (COT), Cefepime (CPM), Cefixime (CFM), Ciprofloxacin (CIP), Gentamicin (GEN), Nitrofurantoin (NIT), Levofloxacin (LE), Piperacillin (PIT), Imipenem (IPM), Azithromycin (AZM), Cefotetan(CX), Clindamycin (CD), Doxycycline Hydrochloride (DO), Meropenem (MRP), Nevobiocin (NV), Erythromycin (E), Ceftazidime(CAZ), Clavulanic acid (CAC), Tetracycline (TE), Penicillin-G (P)

Table 6: Percentage distribution of Resistance patterns for each Gram-positive isolate to commonly used antibiotics.

Antimicrobial Agent	<i>Staphylococcus aureus</i> Resistance (%) (N=14) (n)	<i>Coagulase - negative staphylococci</i> Resistance (%) (N=4) (n)	<i>Enterococcus</i> spp Resistance (%) (N= 1) (n)	<i>Enterococcus faecalis</i> Resistance (%) (N=3) (n)
AK	0	-	0	66.66 (2)
AMP	78.57 (11)	100	-	0
AMX	64.28 (9)	100	100	66.66 (2)
AMC	7.14 (1)	-	0	33.33 (1)
CTR	21.42 (3)	0	0	100
COT	28.57 (4)	-	-	33.33 (1)
CPM	21.42 (3)	-	-	66.66 (2)
CFM	0	-	-	-
CIP	57.14 (8)	50 (2)	100	66.66 (2)
GEN	7.14 (1)	-	0	66.66 (2)
NIT	78.57 (11)	75 (3)	100	0
LE	50 (7)	50 (2)	100	66.66 (2)
PIT	0	-	0	0

Antimicrobial Agent	<i>Staphylococcus aureus</i> Resistance (%) (N=14) (n)	<i>Coagulase - negative staphylococci</i> Resistance (%) (N=4) (n)	<i>Enterococcus</i> spp Resistance (%) (N=1) (n)	<i>Enterococcus faecalis</i> Resistance (%) (N=3) (n)
IPM	0	-	-	0
AZM	57.14 (8)	75 (3)	-	33.33 (1)
CX	92.85 (13)	100	-	-
CD	64.28 (9)	100	-	66.66 (2)
DO	0	0	-	0
MRP	0	0	0	33.33 (1)
E	42.85 (6)	-	-	33.33 (1)
CAZ	7.14 (1)	-	100	-
CAC	-	-	-	33.33 (1)
TE	-	-	-	33.33 (1)
P	7.14 (1)	25 (1)	100	66.66 (2)

Amikacin (AK), Ampicillin (AMP), Amoxicillin (AMX), Amoxycylav (AMC), Ceftriaxone (CTR), Co-Trimoxazole (COT), Cefepime (CPM), Cefixime (CFM), Ciprofloxacin (CIP), Gentamicin (GEN), Nitrofurantoin (NIT), Levofloxacin (LE), Piperacillin (PIT), Imipenem (IPM), Azithromycin (AZM), Cefotetan(CX), Clindamycin (CD), Doxycycline Hydrochloride (DO), Meropenem (MRP), Nevobiocin (NV), Erythromycin (E), Ceftazidime(CAZ), Clavulanic acid (CAC), Tetracycline (TE), Penicillin-G (P)

DISCUSSION

Our study at R.R.M campus in Janakpurdham gives us a really in-depth look at asymptomatic Urinary Tract Infections (UTIs) are a significant health concern among students, with this study revealing that nearly one-third (30.46%) of the 151 participants had confirmed bacterial growth in their urine cultures. It's reassuring that the rest of the participants had no detectable bacteria, possibly due to good hygiene. Also, similar results were obtained. But higher than the study done at Pokhara University, Nepal, 4.12% (Sapkota et al., 2020). It might also be due to the variation in the methodology used, and sexual behavior, as individuals with frequent sexual practices are more exposed to UTI, which is due to ascending infection to the urinary tract from the genital area (Gebremariam et al., 2019).

The study found that UTIs are most prevalent among students aged 20-25 years (66.88%), which aligns with the typical university demographic. Second-year students formed the largest group affected (37.08%), and the issue wasn't confined to any specific academic discipline, with Management (BBA) students showing the highest representation (25.16%). However, the observed prevalence of 30.46% signifies a substantial and often undetected health concern, as such asymptomatic infections can serve as

a reservoir for the development and dissemination of antimicrobial resistance, a phenomenon increasingly documented among young adults (Kebede et al., 2025). This prevalence aligns with or even exceeds rates reported in other studies involving young adult populations, for instance, a study reporting a 24.6% overall UTI prevalence among college students, with asymptomatic bacteriuria accounting for a substantial proportion of these cases (Kebede et al., 2025). Further research indicates that inadequate water intake and unsatisfactory toilet habits are strong predictors of UTI, especially among female students, who are particularly vulnerable to these infections, with 60% experiencing an infection at some point in their lives (Vyas et al., 2015). This emphasizes the critical need for targeted health education and prophylactic strategies within university settings to mitigate the long-term sequelae of untreated bacteriuria, including potential renal damage and systemic infections (Tabassum et al., 2021). Given that urinary tract infections are among the most common medical conditions globally, with adult women being 30 times more likely to develop them, early detection and management are paramount to prevent severe morbidities (Abadi et al., 2023).

Among forty-six positive isolates, Initial Gram stain results showed a slight majority of Gram-negative bacteria (52.17%) over Gram-positive bacteria (47.82%). This is generally expected in UTIs, as Gram-negative bacteria, often originating from the GUT, are common culprits.

Unexpectedly, *Staphylococcus aureus* was the most common isolate, found in 30.43% of positive cultures. While it can cause UTIs, it's less common than *E. coli* in community-acquired infections. Its prevalence here might suggest alternative infection routes, such as spread from skin or nasal passages due to hygiene

or specific risk factors. *Escherichia coli* accounted for 21.74% of positive cultures. This finding was contradicted with the study conducted in Indonesia (Agustino Purba et al., 2012), Ethiopia (Gebremariam et al., 2019), and Nepal (Sapkota et al., 2020), where *E. coli* was the predominant isolate. While *Klebsiella pneumoniae* (SBL Producer) was identified in 10.86% of cases, its resistance profile is crucial for effective treatment. On *Acinetobacter* spp (6.52%) often seen in healthcare settings, their presence in community-dwelling students warrants attention. However, *Staphylococcus aureus* (4.34%) can cause UTIs, especially in those with urinary tract abnormalities. But *Pseudomonas* spp (including *Pseudomonas aeruginosa* 4.34%) can also be the cause of asymptomatic UTI in some cases. These are notorious for their inherent resistance to many antibiotics. And at last, *Citrobacter koseri* (2.17%) can be another opportunistic Gram-negative pathogen.

The high levels of resistance among Gram-negative isolates to both common and advanced antibiotics are a major concern. This emphasizes the critical need for culture-guided therapy to ensure effective treatment and prevent the spread of multidrug-resistant organisms. *E. coli* (N=9) was found to be resistant with the highest percentage to Ciprofloxacin (77.77%), 66.66% for amoxicillin, and Cotrimoxazole 55.55% for Cefepime and 44.44% for Ceftriaxone. Many Gram-negative isolates showed high resistance to commonly used antibiotics. Understanding antibiotic resistance is crucial for effective treatment, and the findings here are concerning. Similar findings were reported by study done in Nigeria (Okafor & Nweze, 2020), Ethiopia (Tigabu et al., 2020), and Saudi Arabia (Al Youssef et al., 2020).

Staphylococcus aureus (N=14) High resistance to Ampicillin (78.57%), Amoxicillin (64.28%) 0% to Amoxycylav, Imipenem, Meropenem. Coagulase-negative *staphylococci* (N=4) 100% resistant to Ampicillin, Amoxicillin, and Amoxycylav 0% to Imipenem, Meropenem, and Erythromycin. *Enterococcus* spp (N=1) & *Enterococcus faecalis* (N=3). Extensive resistance 0% resistant to Imipenem, Meropenem. *Staphylococcus aureus* showed very high resistance to Cloxacillin (92.85%), strongly suggesting the presence of Methicillin-Resistant *Staphylococcus aureus* (MRSA) strains, which significantly limit treatment options. Similarly, Coagulase-negative *staphylococci* showed 100% resistance to Cloxacillin, indicative of MR-CoNS.

Enterococcus spp and *Enterococcus faecalis* demonstrated high resistance to various antibiotics. These resistance rates were found to be similar to a previous study conducted in Ethiopia (Tigabu et al., 2020) and Saudi Arabia (Al Youssef et al., 2020).

Possible factors contributing to this resistance include inappropriate use and incorrect administration of these antibiotics, as well as other factors such as strain and geographic variation (Walsh et al., 2023).

CONCLUSION

A recent study on Urinary Tract Infections (UTIs) among students at R.R.M College revealed a significant health challenge, with roughly one-third of participants affected by these infections. Unlike typical patterns where *Escherichia coli* is the dominant cause, this study found *Staphylococcus aureus* to be the most frequently isolated bacterium. Notably, a significant proportion of the isolated bacteria exhibited resistance to ceftazidime, as well as penicillin, ampicillin, and amoxicillin.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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