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Investigation of Uropathological Bacterial Profile and Antibiotic Susceptibility Patterns Among Pregnant Women in Rawalpindi and Islamabad: A Comprehensive Analysis

Amaila Qaisar^{1*}, Abeer Israr², Igra Saeed², Esha Ashfaq², Laiba Shaukat², Faiza Haider²

¹Senior Lecturer, Faculty of Rehabilitation and Allied Health Sciences, Riphah International University, Islamabad, Pakistan.

²Student, Faculty of Rehabilitation and Allied Health Sciences, Riphah International University, Islamabad, Pakistan.

*Corresponding Author: Amaila Qaisar, Senior Lecturer; Email: <u>amaila.qaiser134@gmail.com</u>

(†) Iqra Saeed is recognized as an equal contributor alongside 2nd author i.e. Abeer Israr, reflecting her significant contribution to the research.

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ABSTRACT

Background: Urinary tract infections (UTIs) represent a significant health concern for pregnant women, potentially leading to adverse outcomes for both mother and child. The anatomical and physiological changes during pregnancy increase the risk of UTIs, making the study of their bacteriological profile and antibiotic susceptibility patterns especially pertinent.

Objective: This study aimed to assess the bacteriological profile and antibiotic susceptibility patterns of UTIs among pregnant women in Rawalpindi and Islamabad, to identify the most prevalent pathogens and their resistance to commonly used antibiotics.

Methods: A cross-sectional study was conducted on 165 early morning mid-stream urine samples collected from pregnant women attending the Railway General Hospital, Rawalpindi, over four months. Samples were analyzed for UTI-causing organisms using Cysteine Lactose Electrolyte Deficient (CLED) medium, MacConkey agar, and blood agar plates. Gram staining and biochemical tests were utilized for bacterial identification, while the Kirby-Bauer disk diffusion method determined antibiotic susceptibility. Statistical analysis was performed using SPSS version 25.

Results: The study population predominantly consisted of residents from Rawalpindi (94.5%). The highest incidence of UTIs was observed in the age group of 31-36 years (34.5%). Staphylococcus aureus (28%) and Pseudomonas aeruginosa (30%) were the most prevalent pathogens. High resistance rates were noted against ampicillin and azithromycin among gram-positive isolates, whereas gram-negative bacteria showed considerable resistance to ciprofloxacin. However, gentamicin displayed relative effectiveness across various pathogens.

Conclusion: The study highlights a significant prevalence of antibiotic-resistant UTIs among pregnant women in the studied regions, with Staphylococcus aureus and Pseudomonas aeruginosa being the dominant pathogens. The findings call for an urgent need to implement effective antibiotic stewardship and screening programs during pregnancy to mitigate the risk of UTIs and their associated complications.

Keywords: Urinary Tract Infections, Pregnant Women, Antibiotic Resistance, Staphylococcus aureus, Pseudomonas aeruginosa, Antimicrobial Susceptibility, Rawalpindi, Islamabad.

INTRODUCTION

Urinary tract infections (UTIs), defined as infections caused by microorganisms within the urinary tract, represent a significant health concern globally, with a substantial proportion of women experiencing at least one UTI episode in their lifetime, estimated between 40% to 50% (1-3). The risk of UTIs in females is attributed to the anatomical structure of the female urogenital system (4-6), and this risk is further exacerbated during pregnancy due to increased vulnerability to bacterial contamination. A study from Dezful city, Iran, indicated that around 5% of pregnant women tested positive for UTIs (7), highlighting the prevalence of this condition within this demographic. Moreover, the distinction between asymptomatic bacteriuria (ASB), where significant bacterial presence is detected without clinical symptoms of UTI (8), and symptomatic UTIs, which present with a range of clinical signs, is crucial for appropriate management and treatment.



The predominant pathogens implicated in UTIs include Escherichia coli, Staphylococcus aureus, Klebsiella pneumoniae, Proteus species, Pseudomonas, Acinetobacter, and Streptococcus Group B (9, 10). Escherichia coli, in particular, has been identified as the most common causative agent, responsible for infecting approximately 56.8% of pregnant women (11), while Klebsiella pneumoniae is recognized for causing 10-15% of UTIs among this group and ranks as the second most prevalent pathogen after E. coli (12). Additionally, the impact of Staphylococcus aureus, including methicillin-resistant strains (MRSA), has been noted for its contribution to increased mortality and morbidity rates among pregnant women.

If left untreated, UTIs can lead to severe complications such as cystitis and acute pyelonephritis. A prospective multi-center cohort study in Ukraine reported that 51.7% of pregnant women experienced cystitis (13), and 30% of those with untreated ASB developed acute pyelonephritis (14). Furthermore, the progression of untreated UTIs can cause bacterial overgrowth, which may result in membrane rupture, premature labor, and miscarriage (19), underscoring the critical need for timely and effective treatment. The cornerstone of UTI management is antibiotic therapy. However, the increasing prevalence of antibiotic resistance has become a significant public health concern globally, as highlighted by the World Health Organization. The rise in antimicrobial resistance necessitates the timely screening of UTIs during pregnancy to reduce incidence rates effectively. Nonetheless, in developing countries such as Pakistan, screening for UTIs and antimicrobial resistance is often not prioritized, posing a significant risk to maternal and fetal health (15).

This study aims to ascertain the trimester with the highest incidence of UTIs among pregnant women, identify the most common uropathogens, and analyze their antibiotic susceptibility patterns to assess the risk of antibiotic resistance in Pakistan. By focusing on these objectives, the study seeks to contribute to the existing body of knowledge on UTIs in pregnant women and inform public health strategies to mitigate the impact of this condition and its associated complications.

MATERIAL AND METHODS

The research conducted was a cross-sectional study at the Railway General Hospital, Rawalpindi, Pakistan, from September to December 2023. This facility, affiliated with the Islamic International Medical College Trust (IIMCT), hosts a comprehensive array of departments dedicated to various medical disciplines, including Surgery, Internal Medicine, Pediatrics, and Intensive and Diagnostic Laboratories. The study, which spanned four months following the research board's approval, analyzed specimens from 165 patients. These early morning mid-stream urine specimens, ranging from five to ten milliliters each, were systematically collected for UTI analysis. Following collection, specimens were promptly labeled and dispatched to the laboratory for examination within thirty minutes of receipt.

Socio-demographic and clinical data pertaining to the participants were meticulously gathered from patient reports and history charts spanning the previous two years, in addition to current records from the microbiology lab of Railway General Hospital. This data encompassed age, marital status, occupation, residence, and relevant clinical information. A random systematic sampling method was employed to select participants for the study, focusing on pregnant women aged 19 to 45 who presented with clinical symptoms indicative of urinary tract infections. Conversely, individuals who were not pregnant, had been on antibiotics within the two weeks prior to the study, or lacked definitive clinical signs of urinary tract infections were excluded from the analysis (16-18).

Data collection was comprehensive, and the information obtained was entered into an Excel spreadsheet before being transferred to SPSS version 25 for analysis. Statistical tests utilized included chi-square, Cramer's V, and phi tests to examine various parameters such as age, residence, trimester of pregnancy, symptomatic versus asymptomatic patients, causative organisms of UTIs, and their antibiotic susceptibility profiles. The analysis sought to identify correlations and associations between these variables, which were visually represented through bar charts and pie charts.

The bacterial examination process entailed the use of a sterilized wire loop to inoculate specimens onto Cysteine Lactose Electrolyte Deficient (CLED) medium. Following incubation at 37°C for 24–48 hours, bacterial growth was assessed, and significant colonies were further cultured on MacConkey agar and blood agar plates (BAP) under similar conditions. The Gram staining technique was crucial for identifying the bacteria involved, distinguishing between Gram-positive bacteria, which retain a dark purple color post-alcohol treatment, and Gram-negative bacteria, which adopt a reddish-pink hue due to the lipid solubilization and subsequent dye loss.

Furthermore, various biochemical tests were conducted, including the Catalase Test, Coagulase Test, Oxidase Test, Indole Test, Urease Test, Triple Sugar Iron Test, Bile Esculin Agar Test, and API 20 E Test, facilitating the identification of bacteria such as Staphylococcus aureus, Pseudomonas aeruginosa, Acinetobacter baumannii, Escherichia coli, Klebsiella pneumoniae, and Enterococcus faecalis. The Antibiotic Susceptibility Test (AST), performed using the Kirby-Bauer disk diffusion method on Mueller



Hinton Agar (MHA), determined the effectiveness of antibiotics like chloramphenicol, ampicillin, linezolid, azithromycin, vancomycin, amikacin, fosfomycin, gentamicin, ciprofloxacin, amoxicillin clavulanate, cefotaxime, imipenem, azactam, and meropenem against these pathogens.

In adherence to ethical standards, this study was conducted following the Declaration of Helsinki guidelines, ensuring respect for participant privacy and the confidentiality of their medical information. The research board's approval was a testament to the study's ethical alignment, underscoring its commitment to contributing valuable insights into the bacteriological profile of UTIs and antibiotic susceptibility patterns among pregnant women in Rawalpindi and Islamabad.

RESULTS

In the study, a detailed examination of socio-demographic characteristics revealed a diverse age distribution among the pregnant women participants, with a concentration in the 31-36 age group, accounting for 34.5% of the total (Table 1). This was closely followed by the 25-30 age group, which constituted 28.5%. The geographical distribution predominantly featured residents from Rawalpindi (94.5%), indicating a significant regional concentration of the study population. An analysis of pregnancy stages highlighted a substantial majority in their 3rd trimester, representing 75.75% of participants, underscoring the critical timing of UTI susceptibility during pregnancy. The gestational diabetes prevalence was notably low at 3%, suggesting a limited influence on the UTI occurrence within this cohort. The symptomatic versus asymptomatic classification of UTI presence showed a nearly balanced distribution, with symptomatic patients slightly less at 47.3%, which included conditions such as abdominal pain and pain during urination.

The antimicrobial resistance pattern illustrated in Table 2 shed light on the challenging landscape of antibiotic effectiveness against UTI-causing bacteria. The data revealed a high resistance rate among Staphylococcus aureus and Escherichia coli strains to commonly used antibiotics like Ampicillin, where resistance reached as high as 97% in E. coli, indicating a concerning trend of antimicrobial resistance (AMR). In contrast, Linezolid showed remarkable efficacy, with nearly 98% sensitivity in Staphylococcus aureus and complete sensitivity in Enterococcus faecalis, pointing towards potential therapeutic avenues in the face of rising AMR.

The graphical representation of UTI organisms identified in the study (Figure 1) visually encapsulated the predominance of Pseudomonas aeruginosa (30%) and Staphylococcus aureus (28%), overshadowing other bacteria in terms of incidence. This emphasis on the high prevalence of these pathogens underscores the critical need for targeted interventions and monitoring within this demographic to mitigate UTI risks effectively.

Parameter	Frequency (N)	Percentage (%)
Age		
19-24	25	15.2
25-30	47	28.5
31-36	57	34.5
37-42	29	17.6
43-48	7	4.2
Residence		
Rawalpindi	156	94.5
Islamabad	9	5.45
Trimester		
1st Trimester	9	5.45
2nd Trimester	31	18.78
3rd Trimester	125	75.75
Gestational Diabetes		
Present	5	3.0
Absent	160	97.0
Symptomatic Patients	78	47.3
- Abdominal pain/pain	57	34.5
- PV leakage	16	9.7
- Anemia	3	1.8

Table 1: Socio-demographic Characteristics of the Study Participants

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Parameter	Frequency (N)	Percentage (%)	
- Burning micturition	2	1.3	
Asymptomatic Patients	87	52.7	

Table 2: Antimicrobial Resistance Pattern of Gram-positive and Gram-negative Bacteria

S.	Antibiotics	Gram-positive Isolates			Gram-negative Isolates		
No.		Percentage (%)			Percentage (%)		
		S. aureus	Ε.	Ε.	K. pneumoniae	Α.	Р.
			faecalis	coli		baumannii	aeruginosa
1	Ampicillin	88	90	97	Nil	Nil	Nil
2	Azithromycin	87	80	Nil	Nil	Nil	Nil
3	Ciprofloxacin	Nil	Nil	45	38	40	12
4	Imipenem	Nil	Nil	Nil	Nil	91	22

Furthermore, an expanded analysis including a variety of antibiotics across different bacteria (Figure 2) provided a comprehensive overview of the antimicrobial susceptibility landscape. The sensitivity and resistance percentages painted a vivid picture of the



ongoing battle against UTI pathogens, with notable findings such as the high resistance of Acinetobacter baumannii to Ciprofloxacin (99% resistance) and the effective sensitivity of Pseudomonas aeruginosa to the same antibiotic (88% sensitivity). These contrasting results highlight the nuanced nature of bacterial response to antibiotic treatments and the importance of tailored therapeutic strategies based on accurate susceptibility data.

> The aggregated data from these tables and figures enrich the understanding of UTI dynamics among pregnant women in the study region, emphasizing the importance of age, trimester, bacterial prevalence, and the alarming trend of antibiotic resistance. Such insights are invaluable for developing more effective diagnostic, prevention, and treatment strategies to combat UTIs in this vulnerable population.





DISCUSSION

In the present study, urine samples from 165 pregnant patients were analyzed, with a significant majority, 156 (94.5%), residing in Rawalpindi and the remainder, 9 (5.45%), hailing from Islamabad. The age distribution revealed a higher incidence of urinary tract infections (UTIs) among women aged 31-36 years (33.9%), followed by those in the 25-30 year range (28.5%). This distribution contrasts with findings by Kumar Devoor and Umashankar KM, who reported a 20.1% positivity rate for UTIs among pregnant women aged 21-25 years, indicating a discrepancy with our findings (17). Our study identified Staphylococcus aureus as the predominant gram-positive microorganism, corroborating the results of Ayoyi et al., who found a 29.7% positivity rate among 1020 pregnant women (18), and Akoachere et al., with a 24.1% prevalence among 133 participants (19). However, this sharply contrasts with a mere 3.3% prevalence reported in Saudi Arabia, highlighting regional differences in microbial prevalence (20).

Furthermore, Enterococcus faecalis was identified in 5% of our samples, a rate that exceeds findings by Ayoyi et al. (1.9%) and Mike-Ogburia et al. (1.3%), yet falls short of the 10.8% prevalence reported by Almutawif and Eid (21, 22). In terms of gram-negative organisms, Escherichia coli was the most prevalent in our study at 28%, a figure that, while lower than reported in studies by N. G. et al. (56.79%) and Begum et al. (60%), was higher than those found in research conducted in Iraq and Nigeria (6, 7). Klebsiella pneumoniae, the third most prevalent microorganism in our study (26%), showed higher prevalence rates than those noted in Nigerian studies but lower than the rates reported by Johnson et al. in southwestern Uganda (23).

The variation in the prevalence of Acinetobacter baumannii and Pseudomonas aeruginosa compared to studies in Dezful city, Iran, and Saudi Arabia, respectively, underscores the geographic variability in the distribution of uropathogens (24, 25). Literature review indicates that gram-negative species, particularly Escherichia coli, dominate among the microorganisms isolated from pregnant women, with prevalence rates varying significantly across different regions (3-6, 2).

This study concludes that pregnant women, particularly those in their third trimester and aged in their mid-20s to 30s, exhibit a heightened risk of developing UTIs. Staphylococcus aureus, identified as the most dominant organism, presents alongside high resistance rates to commonly used antibiotics such as ampicillin and azithromycin. Conversely, linezolid demonstrated high sensitivity against gram-positive isolates, indicating a potential therapeutic avenue. The moderate to high resistance observed among gram-negative organisms against several antibiotics, with gentamicin showing relative sensitivity, highlights the critical need for targeted antibiotic stewardship and awareness programs to mitigate the risk of antibiotic resistance during pregnancy.

This study's strength lies in its comprehensive analysis of uropathogens and antibiotic susceptibility patterns among pregnant women in Rawalpindi and Islamabad, contributing valuable insights into the regional microbial landscape and resistance profiles. However, limitations include the study's regional focus, which may not fully represent the broader demographic variations in UTI prevalence and antimicrobial resistance patterns. Moreover, the reliance on urine samples precludes a more nuanced understanding of UTIs that may benefit from a multi-faceted diagnostic approach.

Recommendations stemming from this study emphasize the importance of stringent antibiotic stewardship, including the supervision of antibiotic use and the establishment of policies to curb the unauthorized sale of antibiotics. Hospital and pharmacy data surveillance, alongside mandatory urine culture and susceptibility testing at each trimester, could significantly enhance the management of UTIs during pregnancy. Furthermore, the development of novel antimicrobial agents is urgently needed to address the evolving challenge of antibiotic resistance among prevalent uropathogens.

CONCLUSION

This study underscores the critical implications of urinary tract infections (UTIs) among pregnant women, particularly highlighting the prevalence of pathogens like Staphylococcus aureus and the challenges of antimicrobial resistance. The findings reveal a pressing need for heightened surveillance, targeted antibiotic stewardship, and the development of new therapeutic strategies to manage UTIs in pregnant populations effectively. Addressing these issues is paramount to safeguarding maternal and neonatal health, emphasizing the urgency of integrating comprehensive UTI management and prevention strategies within the broader context of prenatal care to mitigate the potential adverse outcomes associated with these infections.

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