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PREVALENCE OF UROPATHOGENIC BACTERIA AND THEIR ANTIBIOTIC SUSCEPTIBILITY PATTERN AMONG DIABETIC AND NON-DIABETIC PATIENTS IN DISTRICT MARDAN.

Umar Aziz[1], Obaid Ullah[2], Shafiq Ur Rehman[3], Umar Farooq[4], Muhammad Haris[5], Muhammad Hilal Jan[1], Muhammad Awais[7], Hidayat Khan [8,]

ABSTRACT

BACKGROUND: Urinary tract infections are a common bacterial condition that can affect individuals of all ages caused by uropathogenic bacteria**. OBJECTIVE:** To find out the incidence of uropathogenic bacteria and their antibiotic resistance in people with diabetes and non diabetics at District Mardan. **METHODS:** Five-month prospective cross-sectional research work was done at Mardan Medical Complex, Mardan. Urine samples are taken and subjected to microbiological analysis. Antibiotic susceptibility was performed through Kirby-Bauer technique. **RESULTS:** Among the population urinary tract infections (UTI) were 17.0% with diabetes and 14.0% without diabetes respectively. Prominent isolated bacteria were *E. coli*, which accounted for 82.35% of individuals with diabetes and 57.1% of non-diabetes respectively. **CONCLUSION:** It highlights the importance of detection and prompt treatment to prevent complications and life-threatening situations associated with the co-occurrence of UTI and diabetes. This study will greatly assist in managing UTIs among the residents of District Mardan.

KEYWORDS: Antibiotic, Diabetes mellitus, prevalence, Urinary tract infection, Uropathogens.

1. College of Medical Technology, Bacha Khan Medical College, Mardan, KPK, Pakistan
2. College of Medical Technology, Bacha Khan Medical College, Mardan, KPK, Pakistan
3. College of Medical Technology, Bacha Khan Medical College, Mardan, KPK, Pakistan
4. Department of Biotechnology International Islamic University Islamabad, Pakistan
5. College of Medical Technology, Bacha Khan Medical College, Mardan, KPK, Pakistan
6. College of Medical Technology, Bacha Khan Medical College, Mardan, KPK, Pakistan
7. College of Medical Technology, Bacha Khan Medical College, Mardan, KPK, Pakistan
8. College of Medical Technology, Bacha Khan Medical College, Mardan, KPK, Pakistan

Corresponding Author: Umar Aziz[1] College of Medical Technology, Email: farhansf9@gmail.com

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INTRODUCTION

Urinary tract infections (UTIs) are common complication among populations with diabetes and non diabetic. Diabetes mellitus (DM) is a major global concern and an increasing public health issue, especially in emerging nations [1,2,3]. Diabetes mellitus is a metabolic illness that develops when there is inadequate insulin in the body, which leads to unnecessarily high blood glucose levels. A critical public health issue, particularly in emerging nations, is the emergence of diabetes mellitus (DM), which is occurring more often than ever before [4,5] . Untreated UTIs can result in serious problems among diabetic and non-diabetic patients which lead to kidney damage, renal scarring, and renal failure. Gram-negative bacteria such as *Pseudomonas aeruginosa*, Proteus, Acinetobacter, Klebsiella, Serratia, Enterobacter, and Citrobacter species, besides these, gram-positive bacteria can also cause

UTIs such as *Staphylococcus saprophyticus*, several Enterococcus species, and Coagulase-negative bacteria such as *Staphylococcus epidermidis, Staphylococcus haemolyticus* [6,7,8] . The most common signs of UTI are fever, hot urination, and soreness over the pelvic bone, pale, murky urine that smells bad, frequent urination, vomiting, and others. Antibiotic resistance is a serious problem for treating UTIs. The degradation of antibiotics by enzymes changes to the structural proteins of bacteria, or reduced membrane permeability due to antibiotic usage are common forms of antibiotic resistance. Antibiotic resistance may result from bacterial genome mutation or inappropriate use of antibiotics, Antibiotic sensitivity can determine the specific antibiotic used to treat UTI [9,10,11]**.** It is increasingly concerning that improper usage of antibiotics might result in urine bacteria becoming more resistant to conventional drugs. To guarantee appropriate use of the available antibiotics, local uropathogenic prevalence and susceptibility patterns must be assessed [12,13,14] . According to research, 1-2% of boys and 3-7% of girls under the age of six get UTIs [15]. Previous research indicates that 85% of urinary tract infections (UTIs) are caused by *Escherichia coli*, often known as *E. coli* [16.] Researchers examined the susceptibility of *E. coli* which is the most common cause of UTIs, to commonly used antibiotics. The study found that amikacin had a 97.8% sensitivity rate, gentamicin had a 97% sensitivity rate, ciprofloxacin had a 94% sensitivity rate, nitrofurantoin had an 87.1% sensitivity rate, nalidixic acid had a 93.7% sensitivity rate, trimethoprim-sulfamethoxazole had a 48.2% sensitivity rate, cephalexin had a 76% sensitivity rate, and ampicillin had a sensitivity rate of only 6.9% [17.]

It is crucial to investigate whether individuals, with diabetes and their caregivers experience UTIs caused by the bacteria. If these bacteria show varying responses to antibiotics. Conversely, limited research has been conducted in district Mardan on the prevalence and distribution of bacteria, among diabetic and non-diabetic populations. As a result, the goal of this research is to determine the resistance pattern and frequency of urinary bacterial infections in people with and without diabetes.

MATERIALS AND METHODS

STUDY DESIGNING

The cross-sectional study was conducted at the microbiology department of Mardan Medical Complex from July to November 2023, where patients with UTI were routinely diagnosed and treated. In our study, we use non-probability sampling specifically convenient sampling. The sample size for this study is based on the duration of the study. Patients aged 9 and above of both genders were included, while those over 70 were excluded.

Data Collection Procedure:

During collection, the containers are marked with a specific sample number, the date, and the time. After informing patients 5–10 ml of clean-catch midstream urine samples were obtained in a sterilized screw-capped wide-mouth container. After being collected, the urine samples were processed at the microbiology department section at Mardan Medical complex. After collecting the urine sample, A quantity of 0.002 ml urine are streak through a wire loop on Cystine Lactose Electrolyte Deficient (CLED) Media, incubate at 37°C for 24 hours in an aerobic environment. After 24 hours of incubation, the overall colony count will be determined. Mueller Hinton Agar (MHA) is used for the susceptibility pattern.

ETHICAL APPROVAL

The study conducted at Mardan Medical Complex, Khyber Pakhtunkhwa, was approved by the ethical committee. Before collecting samples, participants or their attendants were given a brief overview of the study's purpose, and written informed consent was obtained. All participants provided samples and were assured that their information would be kept confidential for research purposes, using relevant methods and guidelines.

DATA ANALYSIS PROCEDURE

The collected data were put and examined with SPSS statistical software. The frequency of isolates will be determined, as well as if there is a significant variation in isolates between diabetic and non-diabetic patients and their antibiotic susceptibility pattern. The threshold indicates statistical significance is a p-value below 0.05 [18].

RESULT

Sociodemographic data of study participants

A total of 200 urine samples, 100 from diabetics and 100 from non-diabetics, were taken as part of an extensive study from people ranging in age from 10 to 70. In the non-diabetic group, there were 39 males (39%), 61 females (61%) and 57 females (57%) among the patients with

diabetes. The purpose of collecting the samples was to look into the variations in urine composition among diabetic and non diabetes patients. The studyparticipants were carefully chosen based on a number of criteria, as indicated in (Table1).

Table 1. Age and gender of study participants

Prevalence of Urinary Tract Bacterial Pathogens in the Diabetic and Non-Diabetic Individuals

	Diabetic	Non-Diabetic		
	Gender	Gender		
Age Range	Male	Female	Male	Female
10 – 20	0	0	3	6
21 – 30	0	1	11	16
31 – 40	6	13	9	17
41 – 50	18	29	9	13
51 – 60	12	11	6	7
61 – 70	7	3	1	2

According to table 2, urinary tract bacterial pathogens were found in 17.0% of samples from diabetics and 14% of samples from non-diabetics. *E. coli* (14/17, 82.35% and 8/14, 57.1%), Klebsiella (0/17, 0% and 2/14, 14.2%), Enterococcus (0/17, 0% and 1/14,7.1%), Coliform (1/17, 5.8% and 3/14, 21.4%), GNR (1/17, 5.8% and 0/14, 0%), and Mix Growth (1/17, 5.8% and 0/14, 0%) were the proportions of the organisms for the participants among diabetic and non diabetes patients, respectively.

Table 2. Bacterial isolates were identified from diabetic and non-diabetic patients

Bacteria	Diabetic Group	Non-Diabetic Group		
	n	%	n	%
E.coli	**14**	**82.3%**	**8**	**57.1%**
Klebsiella	**0**	**0%**	**2**	**14.2%**
Enterococcus	**0**	**0%**	**1**	**7.1%**
Coliform	**1**	**5.8%**	**3**	**21.4%**
GNR	**1**	**5.8%**	**0**	**0%**
Mix Growth	**1**	**5.8%**	**0**	**0%**
Total	**17**	**100**	**14**	**100**

Escherichia coli, Klebsiella pneumonia, Enterococcus, Coli form, GNR (gram negative rods), Mix Growth *(S.saprophticus.proteus mirabill,p.aeruginosa)*

Susceptibility Profile of Bacterial Pathogens Isolated from Diabetic Patients

Effective antibiotics against isolated samples from diabetic patients were Fosfomycin, Imipenem, Meropenem, Sulbactam, and Piperacillin-Tazobactam, as shown in Table 3. Among isolated bacterial strains, *E. coli* showed high sensitivity (100%) to all used antibiotics, with exception of amikacin (42.9%) and nitrofurantoin (50%). Coliform bacteria exhibited 100% sensitivity to all administered antibiotics, with the exception of imipenem and sulbactam, which shows 100% resistance. Moreover, GNR showed high susceptibility to all used antibiotic, while sulbactam, showed 100% resistance.

Table 3. In vitro antimicrobial susceptibility pattern of the bacterial isolates from diabetic individuals

Antibiotics	Pattern (S or R)	*E.coli* (n=14)	Klebsiella (n=0)	Enterococcus (n=0)	Coliform (n=1)	GNR (n=1)
FOS	S	6	0	0	1	0
	R	0	0	0	0	0
IPM	S	8	0	0	0	0
	R	0	0	0	1	0
CAZ	S	10	0	0	1	1
	R	4	0	0	0	0
AK	S	6	0	0	1	0
	R	8	0	0	0	0
MRP	S	3	0	0	0	1
	R	0	0	0	0	0
SCF	S	4	0	0	0	0
	R	0	0	0	1	1
TZP	S	10	0	0	0	1
	R	0	0	0	0	0
F	S	2	0	0	1	1
	R	2	0	0	0	0

FOS=fosfomycin, IPM=imipenem,CAZ=Ceftazidime, AK=amikacin, MRP=meropenem, SCF=sulbactam, TZP= piperacillin-tazobactam, F= Nitrofurantoin

Susceptibility Profile of Bacterial Pathogens Isolated from Non-Diabetic Patients

In non diabetic patients the resistance pattern and sensitivity were shown in table 4. Among

the antibiotics fosfomycin showed 25% resistance, while other were highly susceptible to *E. coli*, it showed 100% sensitivity. Similarly Klebsiella was susceptible to all antibiotics in diabetic patients urine samples, but it was 100% resistant to ceftazidime. Enterococcus likewise demonstrated 100% antibiotic susceptibility, with the exception of 100% resistance to nitrofurantoin. Coliform demonstrated 100% susceptibility to all antibiotics, with the exception of meropenem and nitrofurantoin.

Table 4. In vitro antimicrobial susceptibility pattern of the bacterial isolates from Non-diabetic individuals

Antibiotics	Pattern (S or R)	*E.coli* (n=8)	Klebsiella (n=2)	Enterococcus (n=1)	Coliform (n=3)	GNR (n=0)
FOS	S	6	2	1	3	0
	R	2	0	0	0	0
IPM	S	8	2	0	3	0
	R	0	0	0	0	0
CAZ	S	0	0	0	3	0
	R	8	2	0	0	0
AK	S	0	2	2	3	0
	R	0	0	0	0	0
MRP	S	8	2	0	0	0
	R	0	0	0	3	0
SCF	S	8	2	1	3	0
	R	0	0	0	0	0
TZP	S	8	0	1	3	0
	R	0	0	0	0	0
F	S	8	2	0	0	0
	R	0	0	1	3	0

FOS=fosfomycin, IPM=imipenem,CAZ=Ceftazidime, AK=amikacin, MRP=meropenem, SCF=sulbactam, TZP= piperacillin-tazobactam, F= Nitrofurantoin

The overall susceptibility of bacterial pathogens was compared between diabetic and non-diabetic individuals. The results showed that *E. coli* has a susceptibility rate of 75% in diabetic patients, while it was 88% in non-diabetic individuals. Klebsiella was not found in diabetic patients, but it showed 88% susceptibility in non-diabetic patients. On the other hand, Coliform had a susceptibility rate of 75% in both diabetic and non-diabetic individuals. GNR was prominent bacteria found to have an 88% susceptibility rate in diabetic patients, but it was not found in non-diabetic individuals. Lastly, Enterococcus was not found in diabetic patients, but it showed an 88% susceptibility rate in non-diabetic patients as shown in (Figures 1 and 2)

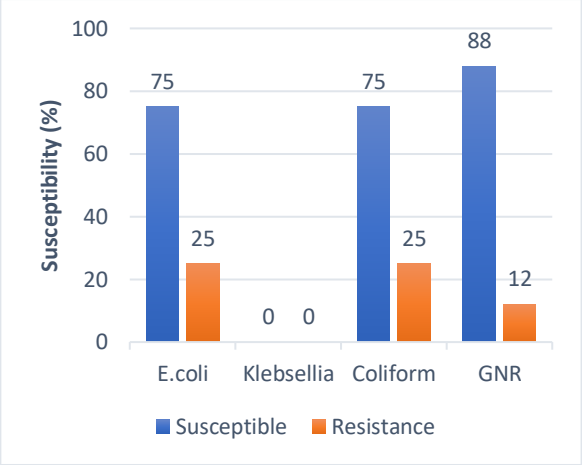

Figure 1. Overall susceptibility of bacterial isolates in Diabetic patients

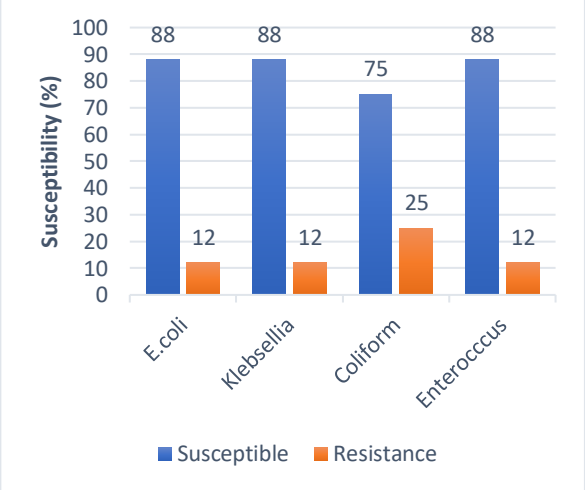

Figure 2. Overall susceptibility of bacterial isolates in Non-Diabetic patients

DISCUSSION AND RECOMMENDATIONS

Antibiotic resistance is a serious issue. Irrational use has made them ineffective. Monitoring global resistance rates is crucial to combat this menace[19]. Previous studies have demonstrated that among Pakistani individuals with diabetes, the incidence of urinary tract infections (UTIs) ranges from 50% to 53% [20]. However, our study produced lower percentages, only 17%, in comparison to the higher figures presented in those studies. It is important to note that our study was conducted over a period of 5 months with a sample size of 200 participants (100 diabetic and 100 non-diabetic), while the previous studies were conducted over a period of only 8 months with a total of 292 diabetic patients sample size. Therefore, it is crucial that we take these differences into consideration when making

any decisions based on these findings. According to a study conducted in Romania, it was found that UTIs were observed in 12% of patients with DM [21]. This figure is slightly lower than our study. According to a study that compared the incidence of UTI in females with and without diabetes, uncontrolled diabetes may increase the severity of UTI. *E. coli* was the most frequently isolated pathogen in both groups. Nevertheless, Candida was only found in the female diabetic group [22]. In our study, we isolated *E. coli* in both groups: 82.3% in diabetic and 57.1% in non-diabetic patients. Other bacterial strains isolated was *Klebsiella pneumonia* was 14.2% in non-diabetic patients. According to a recent study conducted in Pakistan, it was found that among non-diabetic individuals, approximately 5% of urinary samples tested positive for Pseudomonas, a type of bacteria that can cause infections in various parts of the body [23] . In our study, we found that *E. coli* was isolated the most (57.1%) in non-diabetic patients. It has been reported that *E. coli* has developed resistance to certain antibiotics commonly used for treating UTIs, indicating a growing concern for effective treatment options [24-28]. Urinary tract infections caused by *E. coli* are known to be challenging to treat with commonly used antibiotics due to high levels of resistance. However, the results of the current study are promising, as *E. coli* isolates were found to be susceptible to most of the antibiotics tested, with 75% of diabetic patients and 88% of non-diabetic patients showing susceptibility.

RECOMMENDATIONS

In order to gain an understanding of urinary tract infections (UTIs) and how common they are it is important to conduct more research studies, in different communities and environments. These studies should be properly planned, funded, and organized. Before diagnosing a UTI and prescribing antibiotics doctors should always request a urine culture and sensitivity test from a laboratory. This will ensure diagnosis and prevent the growth of antibiotic-resistant bacteria. It is essential for all institutional laboratories to use the urine culture test as it is considered the method, for diagnosing UTIs. Additionally conducting antimicrobial susceptibility testing before prescribing any drugs will help determine the most effective antibiotic treatment. Following these strategies will enable us to stop the spread of antibiotic-resistant bacteria and provide better treatment for patients suffering from UTIs.

CONCLUSION

In summary, our study found that women, with diabetes, had a higher rate of urinary tract infections (UTIs). Among both groups of individuals, the most commonly found bacteria causing UTIs were *E. coli*, Coliform, and Klebsiella. This research is valuable because it identifies the microorganisms for UTIs in people with and without diabetes as well as their susceptibility to antibiotics. The remarkable effectiveness of antibiotics like fosfomycin, imipenem, meropenem, sulbactam, and piperacillin-tazobactam against these bacteria is a discovery. Armed with this knowledge healthcare professionals can make decisions, about which antibiotics to use in treating UTIs.

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CONSENT TO PARTICIPATE: written and verbal consent was taken from subjects and next of kin.

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