

Bacterial Isolation from Hospitalized Diabetic Patients Infected with Urinary Tract Infection in Ibrahim Malik Hospital

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Abstract:

Background: *Diabetes is a serious disease when untreated, and may lead to several conditions such as urinary tract infection which may lead to nephropathy in uncontrollable diabetes.*

Methods: *A cross-sectional study was conducted at Ibrahim Malik Hospital, Khartoum, Sudan, during the period from December 2015 to April 2016. Consecutive patients (men and women) were approached to participate in the study, irrespective of UTI symptoms. Information's about duration, type of DM and Catheterization were obtained from each participant and registered in the questionnaires. Midstream urine samples were collected from infected patients, examined physically, chemically and then cultured for UTI diagnosis. Bacterial isolates examined for their susceptibility to antimicrobial drugs. Bacteriuria and positive urine culture also recovered from diabetics in emergency room showing no UTI symptoms.*

Results: *A total of (70) diabetic patients were enrolled in the current study, 30 (65.6%) were males and 40(71.1%) were females to assess the incidence of UTI in association with risk factors of diabetes. Type 2 diabetic patients showed high incidence rate of infection (75.6 %) more than those of type 1(56.0%).Prolonged duration of diabetes illness had imposed great burden on incidence of UTI and this was clear in percentage of (74.2%) for patients with 6 years duration while it was (64.3%) for those with 3 years and (63.6%) for 1 year duration. The predominant isolates were Escherichia coli[68.2%],and Klebsiella pneumoniae [40.2%].*

Conclusion: *In Sudan diabetic patients having UTI, E.coliwas the most frequent isolate followed by K. pneumoniae.*

Key words: Diabetes, Urinary tract infection, Bacteriuria, *E. coli*, *K. pneumoniae*, Sudan.

INTRODUCTION:

Diabetes mellitus (DM) is a group of metabolic disorders characterized by increased blood glucose level resulting from defects in insulin secretion, insulin action or both ⁽¹⁾.The chronic hyperglycemia in diabetes is associated with long-term damage, dysfunction, and failure of various organs, especially the eyes, kidneys, nerves, heart, and blood vessels. Diabetes mellitus has long been considered to be a predisposing factor for urinary tract infection (UTI) and the urinary tract is the principle site of the infection in diabetics with increased risk of complications of UTI^(2,3). Over time, patients with diabetes may develop cystopathy, nephropathy, and renal papillary necrosis, complications that predispose those to UTIs .Susceptibility increases with the longer duration and great severity of diabetes. The high urine glucose content and defective host immune factors predispose to infection. Hyperglycemia causes

neutrophil dysfunction by increasing intracellular calcium levels and interfering with action and, thus, diapedesis and phagocytosis ⁽⁴⁾.UTI simply means the presence of bacteria undergoing multiplication in urine within the urinary drainage system with infection expressed predominantly at a single site of the kidney (pyelonephritis), bladder (cystitis), prostate (prostatitis), and urethra (urethritis). UTIs are more common and tend to have a more complicated course in patients with DM. UTI in patients with diabetes mellitus occurs because of sweet urine which serves as media for growth of bacteria, defects in neutrophil function and increases adherence to uroepithelial cells⁽⁵⁾. Even though the precise mechanisms for the predilection of pathogens to cause UTI in diabetics remains unclear, a few researches have revealed that the reasons could be immunological impairments such as impaired migration of neutrophils, intracellular killing, phagocytosis, chemotaxis of polymorph nuclear leukocytes from diabetic patients and neuropathic complications such as impaired bladder emptying, as a result static pools of urine will remain in the bladder. In addition, a higher glucose concentration in the urine acts as a favorable culture medium for pathogenic bacteria and promotes rapid bacterial colonization and growth ^(6, 7).

METHODS:

Seventy voided midstream urine samples were collected in sterile containers after giving proper instructions; samples were processed in the laboratory within 2 hours of collection. Macroscopic examination of urine samples was carried out including color of specimen, consistency and results of urine strip for the presence of protein glucose, ketones, haemoglobin, bilirubin, urobilinogen, acetone, nitrate, leucocytesm pH and specific gravity. Then microscopically for a wet preparation to detect: Significant pyuria, i.e. WBCs in excess of 10 cells/l of urine, Red cells, Casts, Yeast cells, Trichomonas vaginalis

motile trophozoites, Schistoma haematobium eggs and bacteria (providing the urine is freshly collected) to diagnose urinary schistosomiasis microscopically, Detecting bacteria in centrifuged (fresh) urine indicates urinary infection, i.e. bacteriuria in excess of 10^4 cell /ml. Pyuria can be quantified by counting WBCs or estimating numbers by examining a drop of urine on a slide (1 WBC per low power field corresponds to 3 cells per high field).

Cultures were performed using semi-quantitative technique, urine sample were inoculated on cysteine-Lactose electrolyte deficient (CLED) medium plates with a calibrated loop (0.001ml) and incubated overnight at 37°C.

After appearance of significant bacterial growth, colonies were examined for their characters; gram stain was carried out to identify the microorganisms then confirmed by biochemical tests as catalase, oxidase, coagulase, kligler iron agar, peptone water (indole), motility, citrate utilization and urease broth test etc.

Antimicrobial susceptibility was performed using multidisc Kirby-Bauer disc diffusion method employing Mueller-Hinton Agar (Hi media laboratory) plate as described by the Clinical and Laboratory Standards Institute (CLSI).

The antibiotic discs representative of Penicillin group, cephalosporin's, amino glycosides, quinolones and Nitrofurantoin were obtained from Hi-Media Laboratories in the following concentrations: Ampicillin/sulbactam(20µg), Co-Trimoxazole(25µg) Amikacin (30µg), Chloramphenicol(30µg) Cephalexin(30µg), Tetracycline (30µg), Ciprofloxacin(5 µg), Nitrofurantoin(300µg), Sparfloxacin(5µg), Levofloxacin (5µg), Norfloxacin (10µg), Ofloxacin (5µg)

ANTIBIOTICS SUSCEPTIBILITY:

Bacterial suspension equivalent to McFarland tube (0.5) from each isolates was streaked on Mueller Hinton agar plate with sterile cotton swab. Antibiotics discs were applied on the agar using sterile forceps, plates were incubated overnight at 37 C. Diameter of the zone of inhibition around the disc was measured in millimeter using ruler and the isolates were classified as sensitive, intermediate and resistant.

ETHICAL CONSIDERATION:

After approval of the management of the hospital administration concerned authority the approval of the patient on their own free without using any mean of pressure and taking in account all the professional ethics and the samples were collected by the patient.

RESULTS:

Total of (70) diabetic patients were enrolled in the current study, 30 male and 40 female to assess the incidence of UTI in association with risk factors of diabetes. The overall prevalence of urinary tract infection in females was (71.1%) and in male (65.6%).

Regarding to the type of diabetes, patients with Type 2 showed high incidence rate of infection (75.6%) more than those of type 1(56.0%).Prolonged duration of diabetes illness had imposed great burden on incidence of UTI and this was clear in percentage of (74.2%), (64.3%), (63.6%), for patients with 6, 3, and1 year duration of illness respectively.

The catheterization considered as a high risk factor in incidence of UTI which was evident in the recovery of *Pseudomonas aeruginosa*& *S.aureus* with percentage of (17.6%,

95.9%) versus (no growth, 22.6%) in catheterized and non-catheterized respectively. (Table 3).

The most abundant isolated microorganism in females & males was *E. coli* an *klebsiellapneumoniae* in percentage of (44.4%), (25.9%) and (23.8%),(14.3%) respectively .

Related to Antimicrobial susceptibility pattern of bacterial isolates, Gram-negative bacteria (*E. coli*, *K. pneumoniae*, *pseudomonas aeruginosa* & *P. mirabilis*) were (100%) sensitive to (ciprofloxacin, fuicidic acid, chloramphenicol, levofloxacin, and Norfuration.and moderately sensitive to Co-trimoxazole, norfloxacin, chloramphenicol, ampicillin / sulbactm, Ofloxaccin, and Amikacin while all isolate were resistant to vancomycine *S.aureus* was sensitive tociprofloxacin, chloramphenicol, Tetracycline, Vancomycin and resistant for other antibiotics. (Table2).

Table (1) Correlation between risk factors for UTI with number and percentage of bacterial isolates

Classification		Name of the organism									
		<i>Escherichia coli</i>		<i>Klebsiellapneumoniae</i>		<i>Pseudomonas aeruginosa</i>		<i>Proteus spp</i>		<i>Staphylococcus aureus</i>	
		Count	%	Count	%	Count	%	Count	%	Count	%
Gender	male	5	23.8%	3	14.3%	2	9.5%	2	9.5%	9	42.9%
	female	12	44.4%	7	25.9%	1	3.7%	1	3.7%	6	22.2%
duration of UTI	15 day	4	40.0%	1	10.0%	2	20.0%	0	0.0%	3	30.0%
	1 month	10	38.5%	7	26.9%	1	3.8%	2	7.7%	6	23.1%
	2 month and more	3	25.0%	2	16.7%	0	0.0%	1	8.3%	6	50.0%
Type of DM	Type 1	5	35.7%	2	14.3%	0	0.0%	1	7.1%	6	42.9%
	Type 2	12	35.3%	8	23.5%	3	8.8%	2	5.9%	9	26.5%
Duration of DM	1 year	4	57.1%	0	0.0%	0	0.0%	1	14.3%	2	28.6%
	3 years	4	22.2%	7	38.9%	0	0.0%	0	0.0%	7	38.9%
	6 year and more	9	39.1%	3	13.0%	3	13.0%	2	8.7%	6	26.1%
treatment use	yes	16	34.8%	9	19.6%	3	6.5%	3	6.5%	15	32.0%
	no	1	50.0%	1	50.0%	0	0.0%	0	0.0%	0	0.0%
Catheterized	yes	5	29.4%	0	0.0%	3	17.6%	1	5.9%	8	47.1%
	no	12	38.7%	10	32.3%	0	0.0%	2	6.5%	7	22.0%

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Table (2): P value of different variables of risk factor affecting UTI

Classification		Result of culture						P. value	Remark
		Growth		no growth		Total			
		Count	%	Count	%	Count	%		
Gender	Male	21	65.6%	11	34.4%	32	100.0%	0.6	b
	Female	27	71.1%	11	28.9%	38	100.0%		
Type of DM	Type 1	14	56.0%	11	44.0%	25	100.0%	0.09	a
	Type 2	34	75.6%	11	24.4%	45	100.0%		
Duration of DM	1 year	7	63.6%	4	36.4%	11	100.0%	0.6	b
	3 years	18	64.3%	10	35.7%	28	100.0%		
	6 year and more	23	74.2%	8	25.8%	31	100.0%		
treatment of DM	Yes	46	70.8%	19	29.2%	65	100.0%	0.1	b
	No	2	40.0%	3	60.0%	5	100.0%		
Catheterized	Yes	17	89.5%	2	10.5%	19	100.0%	0.02	a
	No	31	60.8%	20	39.2%	51	100.0%		

Key: a = significant

b = not significant

Significant different consider as P. value (0,5)

Table (3) Antimicrobial susceptibility pattern of bacterial isolate

ANTIBIOTICS		Escherichia coli		Klebsiella pneumonia		Pseudomonas aeruginosa		Proteus spp		Staphylococcus aureus	
		Count	%	Count	%	Count	%	Count	%	Count	%
CP	Sensitive	17	100%	10	100%	3	100%	3	100%	15	100%
	Intermediate	0	0.0%	0	0.0%	0	0.0%	0	0.0%	0	0.00%
	Resistance	0	0.0%	0	0.0%	0	0.0%	0	0.0%	0	0.00%
FD	Sensitive	17	100%	10	100%	3	100%	3	100%	0	0.00%
	Intermediate	0	0.0%	0	0.0%	0	0.0%	0	0.0%	0	0.00%
	Resistance	0	0.0%	0	0.0%	0	0.0%	0	0.0%	15	100.0%
CH	Sensitive	6	35.5%	5	50%	0	0.00%	3	100%	15	100%
	Intermediate	9	52.9%	3	30%	2	66.6%	0	0.0%	0	0.00%
	Resistance	2	11.6%	2	20%	1	33.4%	0	0.0%	0	0.00%
BA	Sensitive	10	58.8%	5	29.4%	2	66.6%	0	0.0%	0	0.00%
	Intermediate	4	23.5%	2	28.6%	0	0.00%	1	14.3%	0	0.00%
	Resistance	3	17.7%	3	12.5%	1	33.4%	2	8.3%	15	100%
SC	Sensitive	7	41.4%	3	23.1%	2	66.6%	1	7.7%	0	0.00%
	Intermediate	6	35.2%	5	35.7%	1	33.4%	2	14.3%	0	0.00%
	Resistance	4	23.3%	2	9.5%	0	0.00%	0	0.0%	15	100%
LF	Sensitive	7	41.5%	6	35.3%	1	33.4%	3	17.6%	0	0.00%
	Intermediate	6	35.2%	3	27.3%	2	66.6%	0	0.0%	0	0.00%
	Resistance	4	23.3%	1	5.0%	0	0.00%	0	0.0%	15	100%

Table (4) Antimicrobial susceptibility pattern of bacterial isolate

ANTIBIOTICS		Escheriacoli		Klebsiellapneumoniae		Pseudomonas aeruginosa		Proteus spp		Staphylococcus aureus	
		Count	%	Count	%	Count	%	Count	%	Count	%
AK	sensitive	4	23.5%	4	40%	1	33.4%	0	0.0%	0	0.00%
	intermediate	4	23.6%	5	50%	2	66.6%	1	33.3%	0	0.00%
	resistance	9	52.9%	1	10%	0	0.00%	2	66.7%	15	100%
AS	sensitive	7	41.5%	3	30%	2	66.6%	0	0.00%	0	0.00%
	intermediate	8	47.3%	4	40%	0	0.00%	3	100%	0	0.00%
	resistance	2	11.2%	3	30%	1	33.4%	0	0.00%	15	100%
PR	sensitive	7	41.5%	3	30%	2	66.6%	0	0.00%	0	0.00%
	intermediate	4	23.5%	3	30%	0	0.00%	2	66.7%	0	0.00%
	resistance	6	35%	4	40%	1	33.4%	1	33.3%	15	100%
TE	sensitive	5	29.4%	5	50%	1	33.4%	0	0.00%	15	100%
	intermediate	5	29.1%	2	20%	1	33.3%	3	100%	0	0.0%
	resistance	7	41.5%	3	30%	1	33.3%	0	0.00%	0	0.0%
OF	sensitive	9	52.9%	5	50%	1	33.4%	2	66.7%	0	0.00%
	intermediate	3	17.6%	2	20%	1	33.3%	0	0.00%	0	0.00%
	resistance	5	29.5%	3	30%	1	33.3%	1	33.3%	15	100%
NX	sensitive	6	35.5%	1	10%	1	33.4%	3	100%	0	0.0%
	intermediate	9	52.9%	3	30%	1	33.3%	0	0.00%	0	0.0%
	resistance	2	11.6%	6	60%	1	33.3%	0	0.00%	15	100%
V	sensitive	0	0.0%	0	0.0%	0	0.0%	0	0.0%	15	100.0%
	intermediate	0	0.0%	0	0.0%	0	0.0%	0	0.0%	0	0.0%
	resistance	17	100%	10	100%	3	100%	3	100%	0	0.0%

DISCUSSION:

The main findings of the present study were that the incidence of urinary tract infection in diabetic females was higher than in males, were corresponding with the study reported for diabetic patients suffering from UTI in Saudi Arabia ⁽⁸⁾. This is probably due to the high level of recurrence of UTI in women ⁽⁹⁾.

Type of diabetes had significant correlation with incidence of UTI, patients of type 2 had high incidence than those of type 1 (P.value less than 0.05) which is attributable to presence of some factors as local complications related to neuropathy such as impaired bladder emptying ⁽⁸⁾ and higher glucose concentrations in urine ⁽¹⁰⁾. The distribution of type 1 and 2 DM in patients with UTI in Ethiopia, showed regional variations, where in rural parts type 1DM is more abundant than type 2 which was more common in small and big cities of the country ⁽¹¹⁾

Our result showed that patients suffering from diabetes for 6 years and more were more aprons to UTI than those with 3 and 1 year duration which may be due to poor controlled hyperglycemic.

Previous study conducted in Saudi Arabia indicated that the age did not influence the incidence of UTI. In the age <20 was (18.6%) and from (21–30) years was (23.3%).⁽¹²⁾

The catheterization considered as a high risk factor in incidence of UTI significantly was evident in the recovery of *pseudomonas aeruginosa* & *S.aureus*. (P.value less than 0.05).

CONCLUSION:

UTI is more common among patients with diabetes mellitus especially elderly women's who were more affected than the other age group. Our study concluded that there was significant correlation between incidence of UTI and effect of risk factors as type of DM, and catheterization. There was high proportion

of gram negative bacilli with predominant uropathogen being *E. coli* in diabetic patients when correlated with age, and duration of disease.

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