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Studies on Urinary Tract Infection among Diabetics in Some Eastern States of Nigeria

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Abstract
This study evaluated the incidence of Urinary tract infection (UTI) among diabetics in some Eastern States of Nigeria. Three hundred (300) midstream urine samples from diabetic patients with fasting blood sugar level between150mg/dl to 450mg/dl (8.3mmol/l to 25mmol/l), 150 males and 150 females) and 200 non diabetic subjects (100 each male and female) were cultured unto freshly prepared Eosine methylene blue (EMB) oxoid, MacConkey (Difco) and blood agar plates and incubated for 24hours at 37^{oc}. Urinalysis using Combi-9 urine test strip and microscopic examination of urine deposits were done. Gram staining and some biochemical tests, antimicrobial sensitivity tests were carried out on isolates using agar diffusion methods. Seventy eight (26%) urine samples from diabetic patients showed growth of specific organism with 45(15%) in female patients and 33(11%) from male patients. Out of the 200 control urine samples 30(10%) showed growth of specific organism (15 each from male and female subjects). *Escherichia coli* was the predominant isolate 55(70.5%), *Proteus mirabilis* 10(12.5%), *Klebsiella* 8(10%) and *Staphylococcus saprophyticus* 5(7%). While in the control *E.coli* was isolated 20(66.7%), *Proteus* 10(33.3%). All organisms isolated were 100% resistant to ampicilline, nitrofurantoin and100% sensitive to gentamicin, nalidixic acid and ofloxacin. The result of this work showed no significant difference between positive nitrite urine samples 60(20%) and specific bacterial growth 26% ($p>0.05$). No qualitative and quantitative differences were observed in the distribution of microbial pathogens of UTI in diabetic and non-diabetic male and female patients studied.
Keywords: Urinary tract infections, diabetics, antibiotic sensitivity.

INTRODUCTION

The urinary tract consists of various organs of the body that produce, store and get rid of urine. This includes kidney, ureter, bladder, and the urethra (National Kidney and Urologic Disease Centre, USA, 2003). The kidneys are the chemical filters of animate blood. About one quarter of the blood pumped by the heart goes through the kidney. The kidney also contains pressure sensitive tissues which help the body control blood pressure and some of the minerals and water are saved partly to keep the blood pressure in proper range. The waste products and the "extras" make up the urine which flows through the ureter into the bladder were it is held until you are ready to get rid of it through the urethra. Normal urine is sterile- that is when cultured it is free of microbial agents and simply contains fluids, salts, and waste products (National Kidney and Urologic Disease Center, USA, 2003). The bladder has an excellent mechanism to keep it free of any microbial agents that might enter. The defense against microbial agents is due to the hydrokinetic and microbial factors. The hydrokinetic aspect includes the periodic voiding of urine, the constant dilution of residual urine in the bladder (Sobel et al., 1997). The bacteriocidal mechanisms in the mucosa cause rapid clearing of microbial agents from the mucosa, the mechanism of which is unclear but may be immunological. The immune system of the mucus membrane is under the influence of many factors such as diet, microbial flora and changes in the hormonal level of the organism. Secretor immunoglobulin A (IgA) in the mucosal immunity probably exerts an important protective effects on the urinary tract by preventing microbial agents from adhering to the uroepithelial cells. Prostatic secretions and periuretheral gland secretions also possess antibacteriocidal factors (Teodosio, 1999). It is when these processes fail and the bacteria virulence traits that enables adherence, growth, and resistance to the host defenses resulting in colonization and infection of the urinary tract override that urinary tract infection (UTI) develops (Teodosio, 1999). Urinary tract infection (UTI) (significant bacteriuria) is defined as a quantitative urine culture yielding greater than or equal to 10^5 CFU per mililiter of urine (Evan et al., 1993; Robert and Edward, 1999, Oslen *et* al., 2000). UTI can be at a single site urethritis- this infection or inflammation of the urethra. Cystitis- this is an infection of the bladder. It is the most common form of UTI. It can be aggravated if the bladder did not empty completely when you urinate. Urethritis- it is infection of the ureter. It occurs if the bacteria entered urinary tract from above or if the ureter to bladder valves does not work properly and allows reflux from the bladder into the ureters. Pyelonephritis- It is an infection of the kidney. This can happen with infection from above or if reflux into the ureters is so bad that infected urine refluxes the way to the kidney (Robert and Edward(1999). UTI could be asymptomatic bacteriuria- two consecutive urine culture growing more than 10^5CFU of a single bacteria specie in a patient lacking symptoms such as pain when you want to urinate, urge to urinate frequently, feeling of heaviness in the lower abdomen, chills, flank pain, nausea and vomiting(John and Michael, 2000). Many microbial agents have been implicated in causing UTI. These agents range from

bacteria, fungi, protozoa, and few viruses. Up to 98% of them were bacteria in origin with up to 90% of them by *E. coli* (John and Michael, 2000). Other bacteria causes include *Staphylococcus saprophyticus* 5%, *Klebsiella* and *Proteus mirabilis* account for most of the remaining causes of UTI. *Pseudomonas* specie and Enterococci are rare bacteria agent and most often are detected in hospital acquired UTI. Candida specie is the most common fungi in urine which can cause UTI. Candida albicans causes 50 to 59% of cases of UTI (Rivert *et* al., 1985). Schistosoma haematobium is the most common protozoa found in urine and causes UTI by deposition of eggs in the bladder and ureter. Urinary schistosomiasis is found in 2-3% of patients between the ages of 10-30 years (Bello and Edungbola, 1992, Gabriel and Obrieby, 2000). Bacteria agents causing UTI in diabetes and non-diabetics are the same with *E. coli* being the predominant (Mario et al., 1999). Once UTI is established diabetic patients differ from normal host in severity (Tolkoff, 1997). Asymptomatic bacteriuria is the second most common cause of morbidity after respiratory tract infection (National Kidney and Urologic Disease Center, USA 2003). Women develop UTI much more than men due to their much shorter urethra. An estimated 40% of women report having had a UTI at some point in their lives (Kunin, 1994). People with diabetes have a higher risk of UTI because of changes in their immune system. Diabetes puts women but not men at significantly higher risk of asymptomatic bacteriuria (Kunin, 1994). The longer a woman has diabetes the higher the risk. Control of blood sugar appears to have no effect on this condition. The risk of asymptomatic, complicated UTI and UTI related abscesses are higher in diabetes (National Kidney and Urologic Disease Center, USA, 2003). UTI in diabetes leads to extensive renal parenchymal damage (Mustafa et al., 2005). UTI is associated with a number of serious side effects which includes carcinoma of the bladder, gram negative bacteriaemia, sepsis, pyrexia of unknown origin, end point renal failure, hypertension or hypotension, increased prematurity, low birth weight and fetal death (Elicia, 2005). It is widely recognised that many problems could be prevented by the application of knowledge already available. This work attempts to add to the body of knowledge about the pathogenesis, diagnosis and antibiotic sensitivity profile of bacterial agents associated with diabetes.

MATERIALS AND METHODS

Three hundred (300) midstream urine samples were collected from diabetic patients resident in three Eastern States of Anambra, Enugu and Abia (150 each from male and female subjects) with fasting blood sugar levels from 150mg/dl to 450mg/dl (7.8mmol/l to 24.94mmol/l) and 200 non diabetic subjects (100 each male and female) respectively. Fasting blood sugar test was done using Randox method. The midstream urine samples on arrival to the laboratory within 2 hours of collection was inoculated on blood, Eosine methylene blue (Oxoid), MacConkey (Difco) agar plates using modified semi quantitative method (Bachman, 1993, Machanda *et al.,* 2005; Fujita *et al.,*2005) and incubated for 24 hours at 37^{oc}. Media used were prepared according to manufacturer's instruction. Urinalysis was done using Combi-9 urine test strip and result recorded.

Initial reading of culture plates were done after 24 hours of incubation. Significant bacteriuria is a quantitative urine culture yielding greater than or equal to 10^5 CFU/ml of urine. All isolates were identified by Gram staining reaction, their colonial appearance on the media. The colonial morphology includes size, colour, opacity, shape, haemolysis, elevation and edge. MacConkey agar plate was used to distinguish between lactose fermenters "coliform type" colonies which gave pink colour on MacConkey plate. The EMB agar plate was used to differentiate *Esherichia coli* which gave metallic sheen colour from *Klebsiella* and *Proteus mirabilis* which gave pink colonies. *Staphylococcus* specie does not grow in this medium. The blood agar plate was used to detect the presence of haemolysis and also allows swarming of *proteus* and mucoid colonies of *klebsiella* to be easily seen. Isolates were stored on nutrient agar slants at 4^{oc} for further confirmation tests which include IMViC test, sugar fermentation for the production of gas, carbohydrate utilization, nitrate reduction, urease production and motility. Confirmation of typical colonies of *S. saprophyticus* on Baird-Parker agar was on the bases of the results of catalase, coagulase, phosphate reduction and carbohydrate utilization.

Antibiotic sensitivity test was done on identified isolates that have significant growth using agar diffusion method and incubated for 24 hours at 37^{oc}. The degree of sensitivity of the organism to the drugs was determined by measuring the easily visible zones of inhibition of growth produced by the diffusion of the antibiotic from the disc into the surrounding medium. Results were recorded.

Statistical analysis

The results obtained in this study were subjected to analysis of variance (ANOVA) and Duncan's multiple range tests.

RESULTS

The prevalence of UTI in diabetic subjects varied with sex, duration of diabetes and was not affected by control of blood sugar. In this work a prevalence of 78 (26%) UTI in diabetes was established with female 45(15%) and male 33(11%). In the 200 control group a prevalence of 30(10%) was established with female 15(5%) and male 15(5%). Positive nitrite test was 60(20%).

Table 1 shows the frequency of occurrence of aetiological agents causing UTI in diabetic and non-diabetic patients. The result reveal that *E. coli* had a significantly higher occurrence of 55(70.5%) and 20(66.7%) of organisms isolated in both diabetic and non- diabetic patients Table 2 depicts the distribution of isolates according to sex. According to this result, *E. coli* was isolated more in female 35(77.8%) than in male 23(69.7%). There was also higher prevalence of UTI in female 45(15%) than in male 33(11%).

Table 3 represents the prevalence of UTI according to blood sugar level. The results reveal that blood sugar levels do not affect the prevalence of UTI.

Table 4 shows the prevalence of UTI according to duration of diabetes. The results reveal that the longer a patient had diabetes the higher the prevalence of UTI. Patients who had diabetes between 12-15 years had the highest prevalence of 28(9.3%).

Table 5 represents the distribution of urinalysis parameters. The high protein positive 170(56.3%) and cast 120(40%) is worrisome and is presumptive diagnosis of increase renal parenchymal damage.

Table 6 shows antibiotic profile of the various isolates. Drugs with area of inhibition greater than 12mm using venier caliper were considered sensitive. All organisms isolated were 100% sensitive to gentamicin, nalidixic acid and ofloxacin and 100% resistant to ampicillin and nitrofurantoin

Table 1 Frequency of occurrence of bacteria aetiologic agents

Isolates	Frequency	Percentage
Escherichia .coli	55	70.5
Proteus miabilis	10	12.8
Klebsiella	8	10.3
Staphylococcus sapropyhticus	5	10.3
Total	78	100
Control group		
Escherichia coli	20	66.7
Proteus mirabilis	10	33.3
Total	30	100

Table 2 Distribution of isolates according to sex

Isolate	Male	Female
Escherichia coli	23(69.7%)	35(77.8%)
Proteus mirabilis	5(15.2%)	5(11.1%)
Klebsiella	3(9.1%)	3(6.7%)
Staphylococcus saprophyticus	2(6.0%)	2 (4.4%)
Total	33(100%)	45(100%)

Table 3 Prevalence of UTI according to blood sugar level

Fasting blood sugar	No. studied	Isolate	Prevalence
150 - 200mg/dl	60	14	4.7
201-250mg/dl	60	16	5.3
251- 300mg/dl	60	15	5.0
301- 350mg/dl	60	16	5.3
400-450mg/dl	60	17	5.7
Total	300	78	26%

There was no significant difference between different blood sugar levels and prevalence of UTI in diabetic subjects in the study $p>0.05$.

Table 4 Prevalence of UTI in diabetes according to duration of diabetes

Duration (years)	No. studied	Isolate	Prevalence
1-2	60	4	1.3
3-5	60	11	3.7
6-8	60	15	5.0
9-11	60	20	6.7
12-15	60	28	9.3
Total	300	78	26%

There was no significant difference in the prevalence of UTI according to duration of diabetes $p>0.05$

Table 5 Distribution of some urinalysis parameters in diabetic subjects

Sugar	Protein	Nitrite	Cast	WBC>4-5PHF
298(99.3%)	170(56.7%)	60(20%)	120(40%)	70(23%)

The high prevalence 40% of cast suggests increase risk of renal parenchymal damage.

Table 6 ANTIBIOTIC SENSITIVITY PROFILE

ORGANISM	SENSITIVITY /RESISTANCE PROFILE
Escherichia coli	100% sensitive to gentamicin, nalidixic acid, ofloxacin, 90.1% Pefloxacin, 77.8% ciprofloxacin, 74% augumentin, 63% Cotrimoxazole, 100% resistant to ampicilline, nitrofurantoin
Proteus	100% sensitive to gentamicin, nalidixic acid, ofloxacin, 85.7% Ciprofloxacin, 80% pefloxacin 100% resistant to ampicilline, nitrofurantoin, cotrimoxazol
Klebsiella	100% sensitive to gentamicin, nalidixic acid, ofloxacin, Pefloxacin, 60% Ciprofloxacin, augumentin, 20% cotrimoxazole. 100% resistant to ampicilline, nitrofurantoin
Staphylococcus saprophyticus	100% sensitive to gentamicin, ofloxacin nalidixic acid, 66% Pefloxacin ,augumentin 33% ciprofloxacin, cotrimoxazole. 100% resistant to ampicilline, nitrofurantoin,

DISCUSSION

People with diabetes have a higher risk of UTI because of changes in their immune system (National Kidney and Urologic Disease Centre, USA, 2003). This study reveals a high prevalence of 26% and 10 % UTI in diabetic and non diabetic patients respectively. *E. coli* was the predominant organism 55(70.5%) and 20(66.7) in diabetic and non diabetic patients respectively in the study. Edward *et* al; (2000) reported a prevalence of 13.1% and 6.8% UTI respectively in diabetic and non diabetic patients in USA. Mario e*t* al; (1999) in Italy reported 18.1% UTI in diabetics. Diabetes puts women (but not men) at significantly higher risk of asymptomatic bacteriuria 15% as revealed by this study. Dutch Diabetes Research (1996) in Germany reported a higher prevalence of 26% asymptomatic bacteriuria in women with diabetes and 6% in non diabetic women. The longer a patient has diabetes, the higher (9.3%) risk of UTI as revealed by this study. Control of blood sugar appears to have no effect on this condition. The risk of symptomatic complicated UTI and UTI related abscesses are higher in diabetic patients (USA National Kidney and Urologic Disease information clearing house 2003). UTI in diabetes leads to extensive renal parenchymal damage (Mustafa, *et* al; 2005). The high positive proteinuria 56.7% and cast 40% in this study is therefore worrisome.

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