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Identification and Antibiogram of Bacteria Associated with Urine of Diabetic Patients in Wukari, North East, Nigeria

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Abstract

Diabetic and bacteriuria was investigated in Wukari community, North East, Nigeria in order to determine the various bacteria associated with the disease and there antibiogram. A total of 80 individuals who are diabetic comprising of 47 (58.8%) males and 33 (41.2%) females had their urine samples examined using standard bacteriological techniques. The bacteria isolated include: *Escherichia coli* 27 (28.1%), *Klebsiella pneumoniae* 25 (26.0%), *Pseudomonas aeruginosa* 13 (13.5%), *Proteus* species 20 (20.8%) and *Staphylococcus aureus* 11 (11.5%) in the decreasing order of isolates. Antibiotic susceptibility testing of isolates showed *Escherichia coli* isolates was 100% sensitive to Ceftazidime, Gentamycin, Ofloxacin, and Augumentin, 58.1% sensitive to Nalidixic acid and Nitrofurantoin, and 73.1% sensitive to Ceftriaxone. It was 100% resistant to Tetracycline, Cotrimoxazole and Amoxicillin, 42.86% resistant to Nalidixic acid and Nitrofurantion and 28.57% resistant to Ceftriaxone. *Staphylococcus aureus* isolated from diabetic females was 100% sensitive to Ceftazidime, Gentamycin and Augumentin, 66.67% sensitive to Cloxacillin, and Chloramphenicol, 33.33% sensitive to Ofloxacin, Tetracycline, Cotrimoxazole, Amoxcillin, Streptomycin and Erythromycin. It was however, 100% resistant to Ceftriaxone. *Klebsiella Pneumoniae* isolated from diabetic males was 100% sensitive to Ceftriazone, Nalidixic Acid, Gentamycin, Augumentin, Tetracycline, and Ofloxacin, but 100% resistant to Cotrimoxazole and Amoxicillin. *Proteus* species and *Pseudomonas aeruginosa* isolates from male and female were 100% sensitive to Ceftazidime, Nalidixic acid, Gentamycin, Ofloxacin, Tetracycline, Ofloxacin, Cloxacillin, Streptomycin, Chloramphenicol and Augumentin, 100% resistant to Cotrimoxazole, Amoxicillin, Ceftazidime, Gentamycin, Cotrimoxazole and Erythromycin. Thus for all the bacterial isolates, Ceftazidime and Amoxicillin had the highest and lowest antimicrobial activities respectively. Results from this finding underscore the need for frequent urine examination for diabetic individuals and illustrates that bacteriuria is a major difficulty encountered in the management of diabetic. Therefore, the need to incorporate antibacterial therapy to the treatment, along with other public health interventions such as access to health education, health communication and appropriate case management.

Keywords

Urine, Antibiogram, Bacteriuria, Wukari, Diabetic, North East, Nigeria

1. Introduction

Diabetes mellitus (DM) also known simply as diabetes, is a group of metabolic disease in which there are high blood sugar levels over a prolonged period [1]. This high blood sugar produces the symptoms of frequent urination increased thirst, and increased hunger untreated, diabetes can cause many complications serious long term complications include heart disease stroke, kidney failure, foot ulcers and damage to the eyes [2]. Diabetes is due to either the pancreases not producing enough insulin, or the cells of the body not

responding properly to the insulin produced [3]. There are three main types of diabetes mellitus which are Type 1 diabetes which results from the body's failure to produce enough insulin, Type 2 diabetes begins with insulin resistance, a condition in which cells fails to respond to insulin properly and gestational diabetes occurs when pregnant women without a previous history of diabetes develop a high blood glucose level [4]. Research studies have substantiated the significance of diabetes in relation to urinary tract infection (UTI) and sufficient evidence in the form of data has been provided to imply the role of diabetes in UTI [5]. Women with diabetes are more vulnerable to UTI than women without diabetes [3]. It is a known fact that the initiation of the infection begins as asymptomatic bacteriuria which develops in to symptomatic bacteriuria as the infection progresses [4]. Studies have highlighted the consequences of asymptomatic bacteriuria and its role in causing renal defect under untreated condition is substantiated. Occurrence of type I and type II diabetes enhances the factors associated with UTI [3, 5]. Presence of bacteria in urine in the absence of any clinical symptoms can be termed as asymptomatic bacteriuria and a population of ≥105 cfu/ ml from the mid-stream urine sample signifies the condition (Colgan *et al.,* 2011). The occurrence of asymptomatic bacteriuria among healthy women range from 2% to 5% and the incidence is three to four folds higher among women with diabetes [6]. Symptomatic bacteriuria is clinically significant and treatment can reduce the consequences of the condition and in turn lessens renal defects that could arise as a consequence of pyelonephritis and preterm delivery [7]. However, investigators found that the treatment among patients hospitalized for diabetes has not reduced the pervasiveness of asymptomatic bacteriuria [8]. Research studies also substantiated the relation between asymptomatic bacteriuria, diabetes and impaired kidney function [9]. Many other researchers have attempted to unveil the association between asymptomatic bacteriuria and diabetes and have successfully revealed the alliance of asymptomatic bacteriuria and the host factors among women with diabetes [10]. Various case control studies signify that the occurrence of bacteriuria and UTI is common among women and presence of debilitating diseases worsens the condition. Patients with diabetes are also susceptible to conditions like cytopathic, nephropathy, and renal papillary necrosis, complications that incline them towards UTI [6]. Diabetic cytopathic as a consequence causes vesicoureteral reflux which causes the backward flow of the urine from the bladder to ureter and kidney and this result in periodic infections [8]. Around 30% of women diagnosed with diabetes are prone to medical conditions like cystocele, cystourethrocele, or rectocele which may in turn cause persistent UTIs among diabetic females [9]. Medical condition like renal and perineal abscess, emphysematous pyelonephritis and emphysematous cystitis is often encountered among diabetic women with complicated UTI [6]. In addition, fungal infections, xanthogranulomatous pyelonephritis, and papillary necrosis are persistent among diabetic females [9]. Obstruction of the urinary tract as a result of diabetes can cause emphysematous UTIs which leads to necrosis and hemorrhagic infarction [6]. The emphysematous UTIs of the upper urinary tract are responsible for pyelonephritis, pyelitis and Emphysematous cystitis. The prime perpetrators accountable for this condition are *Escherichia coli, Klebsiella, pneumoniae,* and *Candida* [11]. The classical symptoms of untreated diabetes are weight loss, Polyuria (frequent urination), Polydipsia (Increased thirst), Polyphagia (increased hunger), blurry vision, head ache, fatigue, slow healing of cuts and itchy skin/skin rashes (diabetic dermatomes) [12]. People usually with type 1 diabetes may also experience episodes and diabetic ketoacidosis, a type of metabolic problems characterized by nausea, vomiting and abdominal pain, the smell of acetone on the breath, several cases a decreased level of consciousness [13]. Metformin is generally recommended as a first line treatment for type 2 diabetes, as there is good evidence that it decreased mortality. Routine use of aspirin, however, has not been found to improve outcomes in uncomplicated diabetes. Angiotensin converting enzyme inhibitors (ACELs). Types 1 diabetes is typically treat with a combination of regular and NPH insulin, or synthetic insulin analogy. Studies have also provided sufficient evidence that antibiotics is effective against the treatment of UTI, tropical creams, prolong course of daily antibiotics is effective [14]. There is no known preventive measure for type 1 diabetes [15]. Type 2 diabetes can often be prevented by a person being a normal body weight, physical exercise and following healthy diets rich diet in whole grains and fibre and choosing good fats, such as poly unsaturated fats found in nuts, vegetable oil, and fish. [16]. To this end, this research tends to evaluate the prevalence rate diabetes in Wukari community which will broaden the existing epidemiological picture of this disease in this part of the globe and has a direct consequence on planning adequate control programme.

2. Materials and Methods

2.1. Study Area and Population

This study was carried out in the Department of Microbiology, Federal University Wukari, Taraba State, Nigeria. Wukari metropolis is a large town which is the Headquarter of Wukari Local Government Area of Taraba State. Geographically, Wukari lies between latitude 7°55'42" North and longitude 9°47'59" East. It has an area of 4,308 km^2. Wukari is home to Federal University Wukari and Kwararafa University. The major languages spoken are Jukun, Kutep, Tiv, Hausa and Fulani [17].

2.2. Study Population (Individuals)

A total of 80 volunteers were recruited for this study. Their ages range between 40 - 70 years old. History and general body examination was taken to exclude individuals with other infections. the history of self-medication was excluded. The individuals were enlightened on the relevance of the study especially the public health significance. These urine samples

were transported to research laboratory of department of Microbiology, Federal University, Wukari for further procession.

2.3. Bacteriological Investigation

Standard bacteriological methods were used for the isolation of the various bacteria. Media used were Nutrient agar and MacConkey agar. For antibiotic susceptibility test, a small amount containing approximately 10 cfu/ml of peptone water culture containing the grown isolate was used to flood an already prepared Nutrient Agar. The excess was decanted and a multi-disc antibiotic was placed on the plate by means of sterile forceps. The plates were incubated at 37°C for 24 hours. This was observed for zones of inhibition and results were taken down accurately in millimeter. Zone size of less than 13mm was resistant to antibiotics while a zone size of 14mm and above indicated a sensitive antibiotic susceptibility testing

3. Results

In this study, a total of 80 samples were collected from diabetic individuals attending General Hospital, Wukari, Taraba State. The result of this study showed the bacteria isolates and their respective percentage occurrence in Table 1. Table 2, shows an identification characteristic of bacterial isolates from diabetic individuals and Table 3 shows antibiogram of various bacterial isolates from diabetic patients.

Table 1. *Frequency and percentage of bacteria isolates in diabetic individuals.*

Bacteria isolates	Number isolated	Percentage occurrence (%)
Escherichia coli	27	28.1
Klebsiella pneumoniae	25	26.0
Pseudomonas aeruginosa	13	13.5
Proteus species	20	20.8
Staphylococcus aureus	11	11.5
	96	99.9

Table 2. *Identification characteristics of bacterial isolates from diabetic patients.*

Colonial morphology	Gram Reaction	Cat	Cog	IN	UR	OX	Lac	Mal	Suc	Man	Glu	Isolates
Pinkish mucoid colonies	-	+	-	-	+	-	+	+	+	+	AG	*Escherichia coli*
Grey-white colonies	-	+	-	+	-	-	+	+	+	+	AG	*Klebsiella pneumoniae*
Blue-green colonies	-	+	-	-	-	+	-	-	-	-	A	*Pseudomonas aeruginosa*
Black colonies	-	+	-	+	+	-	+	+	+	+	AG	*Proteus* species
Smooth, round colonies	+	+	+	-	-	-	-	+	+	+	A	*Staphylococcus aureus*

KEY
+ Positive
- Negative
A – Acid, AG - Acid and Gas
OX Oxidase
Cat- Catalase, IN- Indole
Cog - Coagulase, Lac -Lactose
UR- Urease, Suc- Sucrose
Mal- Maltose, Glu - Glucose
Man- Mannitol

Table 3. *Antibiogram of various bacterial isolates from diabetic patients.*

Antimicrobial agent conc.	Antibiotic effect on bacteria									
	Escherichia coli		*Staphylococcus aureus*		*Klebsiella Pneumoniae*		*Proteus* species		*Pseudomonas aeruginosa*	
Ceftazidime (30µg)	-	7 (100)	-	3 (100)	-	2 (100)	-	1 (100)	1 (100)	-
Ceftriaxone (5µg)	2 (28.6)	5 (73.1)	3 (100)	-	-	2 (100)	-	-	-	-
Nalidixic acid (30µg)	3 (42.9)	4 (58.1)	NA	NA	-	2 (100)	-	1 (100)	NA	NA
Gentamycin (10µg)	-	7 (100)	-	3 (100)	-	2 (100)	-	1 (100)	1 (100)	-
Ofloxacin (5µg)	-	7 (100)	2 (66.7)	1 (33.3)	-	2 (100)	-	1 (100)	-	1 (100)
Nitrofurantoin (200µg)	3 (42.9)	4 (58.1)	NA	NA	-	2 (100)	-	-	NA	NA
Tetracycline (10µg)	7 (100)	-	2 (66.8)	1 (33.3)	-	2 (100)	-	1 (100)	1 (100)	-
Augumentin (30µg)	-	7 (100)	-	3 (100)	-	2 (100)	-	1 (100)	-	1 (100)
Cotrimoxazole (30µg)	7 (100)	-	2 (66.7)	1 (33.3)	2 (100)	-	2 (100)	-	1 (100)	-
Amoxicillin (30µg)	7 (100)	-	2 (66.7)	1 (33.3)	2 (100)	-	2 (100)	-	NA	NA
Cloxacillin (5µg)	-	-	1 (33.3)	2 (66.7)	-	-	-	-	-	1 (100)
Streptomycin (30µg)	-	-	2 (66.7)	1 (33.3)	-	-	-	-	-	1 (100)
Chloramphenicol (10µg)	-	-	1 (33.3)	2 (66.7)	-	-	-	-	-	1 (100)
Erythromycin (5µg)	-	-	2 (66.7)	1 (33.3)	-	-	-	-	1 (100)	-

KEY: R = Resistant (zone diameter of 0-13mm), S= Sensitive (between 14mm and above)
NA= Not applicable

4. Discussion

The bacteriuria of diabetic urine samples collected from individuals showed *Escherichia coli* 27 (28.1%), *Klebsiella pneumoniae* 25 (26%), *Proteus* species 20 (20.8%), *Pseudomonas aeruginosa*, 13 (13.5%) and *Staphylococcuss aureus* 11 (11.5%) isolates in their descending order of percentages, the results are similar to the finding of other studies [9]. However, a few studies indicated higher infections with *Proteus* species and *Klebsiella* species [7]. Some studies indicated a lower percentage of *Escherichia coli* on the other hand and some have given a higher percentage of *Escherichia coli* as compared to other organisms. This could be explained on the basis of sampling technique and the gender differences in different studies [13]. *Pseudomonas aeruginosa* and *Staphylococcus aureus* (13.5% and 11.5%) respectively remain the least isolated bacteria [18]. These results are consistent with other studies. The commonest bacterium isolated in this study which was *Escherichia coli* was also a common occurrence in some related studies [5]. However, in a research work carried out by [11], the reverse was true as *Staphylococcus aureus* was the most common bacteria isolated. *Escherichia coli* is a very common bacterium some are harmless while others cause serious illness [14]. Non-pathogenic strains of *Escherichia coli* those that do not because disease are normal inhabitants of the intestinal tract in humans but certain strains of *Escherichia coli* can cause severe diarrhea and infect the genital and urinary tracts (where they cause bladder or kidney infections) [12]. The antimicrobial susceptibility patterns of all isolates revealed varying percentage of susceptibilities by all isolates. Antibiotic susceptibility of *E. coli* showed that it was 100% sensitive to Ceftazidime, Gentamycin, and Ofloxacin. It was resistant to Tetracycline, Cotrimoxazole, and Amoxicillin. *Staphylococcus. aureus* showed 100% sensitivity to Ceftazidime, Gentamycin, Augumentin, 66.67% sensitivity to Cloxacillin, and Chloramphenicol, 33.33% sensitivity to Ofloxacin, Tetracycline, Cotrimoxazole, Amoxicillin, Streptomycin, and Erythromycin. It was however 66.67% resistant to Ofloxacin, Tetracycline, Cotrimoxazole, Amoxicillin, and Streptomycin. *Klebsiella Pneumoniae* was 100% sensitive to all antibiotics used with the exception of Cotrimoxazole, Amoxicillin, Cloxacillin, Streptomycin, Chloramphenicol and Erythromycin. *Proteus* species was 100% sensitive to Cotrimoxazole, Amoxicillin with the exception of other antibiotics. *Pseudomonas aeruginosa* was 100% sensitive to Ofloxacin, Augumentin, Cloxacillin, Streptomycin and Chloramphenicol. It was however resistant to the other antibiotics tested.

5. Conclusion

In conclusion, it is pertinent to state that since diabetes remains a burden in Africa and major health problem in developing countries especially in rural communities. Further studies are urgently required using a more sophisticated molecular and immunological tools to eliminate this problem. There is also need to incorporate antibacterial therapy to the integrated morbidity control approach of diagnosis and drugs treatment along with other public health interventions such as health education, health communication and appropriate case management. These strategies will improve the health of individuals in the study area.

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