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Uropathogens from diabetic patients with asymptomatic bacteriuria and Urinary tract infections

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ABSTRACT

Urinary tract infections occur more frequently in diabetic than in non-diabetic patients and have been proven to be the primary cause of renal failure in diabetics. This study was to investigate the prevalence of asymptomatic bacteriuria (ASB) and urinary tract infections (UTIs) in clinically diagnosed patients with diabetes and to determine the uropathogens responsible for ASB and UTIs as well as their antimicrobial susceptibility pattern. One hundred and twenty five diabetic patients comprising 69 (55.2%) females and 56 (44.8%) males attending the Buea and Limbe Regional Hospital Diabetic Clinics, South West Region, Cameroon were studied. Midstream urine samples were collected from the study participants. The midstream urine samples were examined macroscopically, microscopically, and culturally using standard techniques on different agar cultures. Uropathogens isolated included *E. coli, Staphylococcus aureus, Streptococcus pyogenes, Klebsiella pneumoniae, Enterococcus faecalis, Proteus mirabilis, Pseudomonas aeruginosa* and fungal isolates. Isolates were tested against commonly used antibiotics and antifungals. Asymptomatic bacteriuria had a prevalence of 47.2% and urinary tract infections (UTIs) a prevalence of 34.4%. Isolates showed high resistance to ciprofloxacin (61.3%), ceftriaxone (70.1%), amoxicillin (96.2%), nystatin (72.3%) and ketoconazole (51.8%). We

came to the conclusion that a high prevalence of asymptomatic bacteriuria and UTIs was observed in both male and female diabetic patients in Cameroon. Improvement on management of diabetes mellitus and personal hygiene, as well as the appropriate use of duly prescribed antibiotic/antifungals would reduce prevalence of ASB and UTIs and hence prevent renal complications.

1. Introduction

Patients with diabetes mellitus (DM) have a high prevalence of asymptomatic bacteriuria (ASB) and urinary tract infections (UTIs). Diabetes has long term effects on the incidence of UTIs and has been reported to be around three to four times high in diabetic compared with non-diabetic patients (Adeyeba et al., 2007). It has been suggested that the presence of static pools of urine due to dysfunctional bladder contracting poorly serves as a favorable medium for bacterial growth (Andriole, 2002), while others suggest that hyperglycemic urine promotes rapid bacterial growth and colonization (Geerling et al., 2000).

In Cameroon, the prevalence of diabetes mellitus in the population is estimated at 8·8% (WHO, 2011). Studies on the distribution of uropathogens in the urine of these patients in the country are greatly limited. This study was aimed at determining the prevalence and susceptibility pattern of asymptomatic bacteriuria (ASB) and urinary tract infections (UTIs) in diabetic patients in Cameroon.

2. Materials and methods

2.1. Study setting and recruitment of study participants

This study which was approved by the Faculty of Health Sciences Institution Review Board of the University of Buea, Cameroon, was conducted using the Buea and Limbe Regional Hospital Diabetic and Hypertensive clinics from March to September 2011. Men and women aged 25–75 years, with confirmed diabetes were eligible to participate in the study provided they had been enrolled in the diabetic clinic for at least 6 months and receiving anti -diabetic medications. Patients who had received antibiotics or antifungal therapy in the preceding 21 days were not included in the study. Diabetic patients on wheelchair or those with severe psychiatric disorders and under urinary catheterization were also excluded from the study. Participants were required to sign an informed consent form which was duly explained to them.

2.2. Laboratory methods

Clean-catch midstream urine samples were collected from patients into labeled sterile MacCarthey bottles for urinalysis. Urine samples were then inoculated on MacConkey agar, Blood agar and cysteine lactose electrolyte deficient (CLED) media using calibrated loops to determine colony forming units. The plates were incubated at 37ºC aerobically for 24hrs. Cultures with colony counts $>10^5$ cfu/ml, for a single isolated uropathogen was considered significant bacteriuria (Harding et al., 2002). The organisms were identified using standard cultural, morphological and biochemical techniques. Sabouraud dextrose agar was used to isolate the fungal pathogens in urine. Urinalysis was carried out using dipstick (Combi 11) while antibiotic sensitivity test was done by the disc diffusion (Kirby-Bauer method) using Mueller-Hinton agar for bacterial isolates and Sabouraud dextrose agar for fungal isolates. The antibiotics used included gentamicin (10mcg), imipenem (10mcg), amikacin (30mcg), ciprofloxacin (5mcg), ofloxacin (5mcg), cephradine (30mcg), amoxicillin (75mcg) and cefotaxime (30mcg). Antifungals used included amphotericin B (30mcg), nystatin (10mcg), ketoconazole (10mcg), itraconazole (10mcg), miconazole (10mcg), and fluconazole (10mcg). The sensitivity plates were incubated aerobically at 37ºC for 24 hrs, and the zone of inhibitions were recorded.

2.3. Statistical analysis

Statistical analyses were performed using MINITAB version 15 statistical software for windows.

2.4. Limitations of the study

Molecular biology techniques or biochemical methods to identify fungal isolates up to the level of the species were not performed owing to the expensive nature of these analyses. These analyses will require a state-of -the art high-tech facility which is not readily available in developing countries like Cameroon. As a result of this, we worked with fungal isolates as a whole.

3. Results

There were a total of 125 study participant who constituted 69 (55.2%) females and 56 (44.8%) males clinically diagnosed diabetic patients. One hundred and two (81.6%) of the urine samples had significant bacteriuria with 59 (47.2%) of them having asymptomatic bacteriuria and 43 (34.4%) having urinary tract infections. Fungal isolates were observed in 7 (5.6%) of the 102 urine specimen.
Females (28.0%) were frequently seen with ASB than men (19.2%) and ASB was more common in the age group between 41 and 60 years (27.2%) (Table 1). Urinary tract infections on the other hand were slightly common in males (18.4%) than in females (16.0%) (Table 2). Fungi organisms were isolated only in the urine from females.

Table 1
Age and sex distribution of Asymptomatic Bacteriuria (ASB).

Age group	Male Number with ASB / %	Female number with ASB / %	Total
20 – 40	3 (2.4)	6 (4.8)	9(7.2)
41 – 60	14 (11.2)	20 (16.0	34(27.2)
> 60	7 (5.6)	9 (7.2)	16(12.8)
total	24 (19.2)	35 (28.0)	59(47.2)

Table 2
Age and sex distribution of Urinary Tract Infections (UTIs).

Age group	Male Number with UTIs / %	Female number with UTIs / %	Total
20 – 40	2 (1.6)	3 (2.4)	5 (4.0)
41 – 60	9 (7.2)	12 (9.6)	21 (16.8)
> 60	12 (9.6)	5 (4.0)	17 (13.6)
Total	23 (18.4)	20 (16.0)	43 (34.4)

Eight (8) different bacteria were isolated from the study participants: *Escherichia coli* (48.0%) were the most prevalent, followed by *Staphylococcus aureus* (19.6%) and *Proteus mirabilis* (8.9%). The least prevalent uropathogen was *Pseudomonas aeruginosa* (1.0%) which was found only in diabetic patients with ASB and not UTIs (Table 3).

Table 3
Uropathogens isolated from diabetic patients with ASB and UTIs.

Uropathogen	No. and % of occurrence (%)		Total
	ASB	UTIs	
Escherichia coli	27(45.8)	22(51.2)	49(48.0)
Staphylococcus aureus	14(23.7)	6(14.0)	20(19.6)
Fungal isolates	2(3.4)	5(11.6)	7(6.9)
Streptococcus pyogenes	4(6.8)	4(9.3)	8(7.8)
Klebsiella pneumoniae	2(3.4)	3(7.0)	5(4.9)
Enterococcus faecalis	1(1.7)	2(4.7)	3(2.9)
Proteus mirabilis	8(13.5)	1(2.3)	9(8.9)
Pseudomonas aeruginosa	1(1.7)	0(0.0)	1(1.0)
Total	59(57.84)	43(42.16)	102(100.0)

The results show that most of the bacterial isolates were highly sensitive to gentamicin (88.6%), imipenem (87.9%), nitrofuraton (79.5%) and amikacin (88.3%). Ciprofloxacin showed some resistance (53.3%), while amoxicillin was highly resistant (96.3%) to all the bacterial isolates (Table 4). Fungal isolates were sensitive to Amphotericin B (80.2%), Flucytosine (68.6%) and Fluconazole (72.0%) but demonstrated resistance to nystatin (72.3%), (Table 5).

Table 4
Antimicrobial susceptibility pattern of bacterial isolates from diabetic patients with ASB and UTIs.

Antibiotic	**% resistance of each antibiotic to**						
	EC	**SA**	**SP**	**KP**	**EF**	**PA**	**PM**
Imipenem	8.5	12.6	29.4	1.5	12.5	6.3	11.2
Nitrofuraton	15.6	41.2	10.7	12.1	24.0	14.0	26.2
Gentamicin	1.1	1.6	6.6	2.1	52.2	11.2	5.1
Amikacin	8.8	12.1	38.7	4.8	10.6	4.9	2.3
Ofloxacin	40.6	50.9	79.5	45.8	66.1	41.5	61.2
Cephradine	60.7	19.0	49.8	42.0	47.1	33.3	27.8
Ceftriaxone	33.2	70.1	51.3	30.5	39.7	43.2	50.0
Cefotaxime	58.7	64.1	62.8	33.6	51.2	42.9	30.2
Ciprofloxacin	68.2	59.0	64.5	33.4	99.8	20.1	28.0
Amoxicillin	98.0	97.1	98.5	89.0	96.2	97.0	98.2

Table 5
Antifungal susceptibility pattern of *Candida albicans* from diabetic patients.

Antifungal	**% Resistance to fungal isolates**
Amphotericin B	19.8
Flucytosine	31.4
Fluconazole	28.0
Itraconazole	43.7
Ketoconazole	51.8
Miconazole	51.2
Nystatin	72.3

4. Discussion

In this study, asymptomatic bacteriuria was present in 59 (47.2%) and urinary tract infections were present in 43 (34.4%) out of the 125 diabetic patients. The high prevalence of ASB (47.2%) recorded in this study was higher than results of previous studies that showed 36.15% in Nigeria (Ophori et al., 2010), 19% in Bahrain (Hajeri, 2008) and 21% in Karachi (Bagai et al., 2008). Other studies have reported much lower prevalence (Alebiosi et al., 2003).

Geerling et al. (2000), reported a prevalence of UTIs of 26.0% in diabetic patients. The result obtained in this study (34.4%) is higher than that of Geerling et al. (2000), the reasons advanced for this high prevalence are poor early diagnosis of diabetes and UTIs. Variations in prevalence have also been attributed to factors such as geographical variations, ethnicity of the study participants and variation in the screening tests used (Assel et al., 2009).

The uropathogen most associated with asymptomatic bacteriuria and urinary tract infection in the study participants was *E. coli* 49 (48.0%). This result is lower than the 59.6% obtained in the study performed by Ophori et al. (2010) in Nigeria. This result shows a contrast to that reported by Alebiosu et al. (2003) where *Klebsiella pneumoniae* was the most prevalent isolate. However, this result is consistent with the majority of reports where *E. coli* had been reported to be the major pathogen in ASB and UTIs (Olaitan, 2006; Hajeri, 2008; Bagai et al., 2008; Assel et al., 2009). Other prevalent uropathogens isolated were *Staphylococcus aureus* 20 (19.6%), *Proteus mirabilis* 9 (8.9%), *Streptococcus pyogenes* 8 (7.8%), fungal isolates 7(6.9%) *Klebsiella pneumoniae* 5(4.9%),

Enterococcus faecalis 3 (2.9%), and *Pseudomonas aeruginosa* 1 (1.0%). These pathogens were similar to those observed in other studies (Hansen et al., 1998; Stapleton, 2002; Boroumand et al., 2006; Ophori et al., 2010).

For the antimicrobial susceptibility pattern of the uropathogens, it was observed that *E. coli* was resistant at similar rates to Gentamicin, amikacin, Nitrofuraton and Imipenem. The same resistant pattern was observed with the remaining uropathogens. However, ciprofloxacin showed resistance to most of the bacterial isolates especially *Enterococcus faecalis*. Amoxicillin showed very high resistance to all the isolates. Fungal isolates showed a high resistance to nystatin. This high resistance can be accounted for by drug resistance due to inadequate treatment and recurrent infections and mono-therapy.

5. Conclusion

In conclusion, a high prevalence of ASB and UTIs was observed in both male and female diabetic patients. The prevalence of ASB (47.2%) and UTIs (34.4%) seen in this study is of major public health importance with the predominant uropathogen being *Escherichia coli*. The high level of resistance to clinically available antibiotics/antifungals is also of public health importance. The authors recommend improvement of personal hygiene and proper glycemic control which would reduce the prevalence of ASB and UTIs; and the appropriate use of duly prescribed antibiotic/antifungals.

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References

Adeyeba,O.A., Adesiji,Y.O., Omosigho, P.O., 2007. Bacterial urinary tract infections in patients with diabetes mellitus. Inter. J. Trop. Med. 2, 89-92

Alebiosu, C.O., Osinupebi, O.A., Olajubu, F.A., 2003. Significant asymptomatic bacteriuria among Nigerian type 2 diabetes. J. Nation. Med. Associ. 95 (5), 344-348.

Andriole,V.T., 2002. Assymptomatic bacteriuria in patients with diabetes, enemy of innocent visitor? New England J. Med. 347, 1617-1618.

Assel, M.T., Al-Meer, F.M., Al-Kuwari, M.G., Ismali,M. F., 2009. Prevalence and predictor of asymptomatic bacteriuria among pregnant women attending Primary health care in Qatar. J. Famil. Med. 4, 14-17.

Baqai, R., Aziz, M., Rasool, G., 2008. Urinary tract infections in diabetic patients and biofilm formation of uropathogens. Infec. Dis. J. Pakistan 17(1), 21-24.

Boroumand, M.A., Sam, L., Abassi,S. H., Salarifar, M., Kassanian, E., Forghani, S., 2006. Asymptomatic bacteriuria in type 2 Iranian diabetic women; A cross sectional study. BMC Womens Health 6, 4.

Geerling, S.E., Stolk, R.P., Camps, M.J., Netten, P.M., Hoekstra J.B., Bouter, K. P., Braven-boer, B., Collet, J.T., Jansz, A.R., Hoepelman, A.I., 2000. Asymptomatic bacteriuria may be considered a complication in women with diabetes: Diabetes Mellitus women asymptomatic bacteriuria Utrecht study group. Diabetes Care. 23, 744-749.

Hajeri,A., 2008. When to treat asymptomatic bacteriuria. Bahrain Med. Bull. 30(2), 1-4.

Hansen, D.S., Gottschau, A. and Kolmos, H.J. 1998. Epidemiology of Klebsiella bacteraemia: a case control study using E. coli bacteraemia as Control. J. Hospit. Infec. 38, 119-132.

Harding, G.K., Zhanel, G.G., Nicolle, L.E., Cheang, M., 2002. Antimicrobial treatment in diabetic women with asymptomatic bacteriuria. New England J. Med. 347, 1576-83.

Olaitan, J.O., 2006. Asymptomatic bacteriuria in female student population of a Nigerian University. Inter. J. Micro. 2(2), 4-9.

Ophori, E.A., Imade, P., Johnny, E.J., 2010. Asymptomatic bacteriuria in patients with type-2 diabetes mellitus. J. Bacterio. Res. 2 (2), 14-17.

Stapleton, A., 2002. Urinary tract infections in patients with diabetes. American Journal of Medicine 113 (1A), 80S-84S.

World Health Organisation (WHO), 2011. Non communicable disease (NCD) country profile. Cameroon. http://www.who.int/nmh/countries/cmr_en.pdf. Accessed at 8:14, 24/11/2012.