Research Journal of Microbiology

ISSN 1816-4935

Academic Journals Inc.

www.academicjournals.com

Research Journal of Microbiology 2 (12): 966-971, 2007
ISSN 1816-4935

Bacteriuria in Primary Health Care Units in Makurdi Metropolis, Middle-Belt, Nigeria

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Abstract: To investigate the rate of significant bacteriuria among patients attending primary health care clinics within Makurdi metropolis, aseptically collected early morning midstream urine from individuals seeking medical attention, were bacteriologically examined. Out of the 500 specimens collected, 70.8% had significant bacteriuria. The bacterial isolates were *Staphylococcus aureus* (42.4%), miscellaneous coliforms (27.1%), *Escherichia coli* (21.5%), *Strept coccus pyogenes* (6.8%), *Klebsiella aerogenes* (1.1%), *Pseudomonas aeruginosa* (0.6%) and *Salmonella* sp. (0.6%). Bacteriuria was significantly correlated with sex of subjects ($r_{500} = 0.356$; $p = 0.000$) and was approximately 5 times as high in women as in men (odds ratio = 5.0; 95% confidence interval). The attributable risk was 30.15%. Gram-positive bacteria predominated in the males and were responsible for 60% of bacteriuria in males. In females, the Gram-negatives accounted for 66.7% of the bacteria isolated. Antibiotic susceptibility testing of *Staphylococcus aureus* and the coliform bacteria to commonly used antimicrobial drugs showed high rates of resistance. The results of this study could be a useful guide in diagnosis of urinary tract infections and in planning control and therapeutic strategies.

Key words: Bacteriuria, attributable risk, odds ratio, risk factor, antibiotic resistance, Middle-Belt, Nigeria

INTRODUCTION

The presence of 10^5 colony forming units (cfu) in one milliliter of early morning midstream urine connotes significant bacteriuria which is an important criterion for diagnosing urinary tract infections. A high prevalence of significant bacteriuria has been recorded in parts of the world and in parts of Nigeria (Aboderin *et al*., 2004; Iduoriyekemwen *et al*., 2006). Organisms implicated are usually enteric bacteria and members of the genus *Staphylococcus* (Aboderin *et al*., 2004; Das *et al*., 2006; Akinloye *et al*., 2006; Hooton *et al*., 2000). Those that are at a greater risk are young women, parous women, diabetic women, enuretic children from poor socio-economic families and pregnant women in whom the condition may lead to onset of pyelonephritis and fetal mortality (Olusanya *et al*., 1992; Orrett *et al*., 1995; Hooton *et al*., 2000; Aboderin *et al*., 2004; Akinloye *et al*., 2006; Iduoriyekemwen *et al*., 2006; Ishay *et al*., 2006). According to Aboderin *et al*. (2004) and Akinloye *et al*. (2006) significant bacteriuria is a major risk factor for developing urinary tract infection in pregnancy. Some factors that predispose to bacteriuria are poor toilet habit (Umeh *et al*., 2006), increased sexual activity (Abbey, 1987), overcrowding (Orrett *et al*., 1995), previous history of urinary tract infection (Abbey, 1987; Caksen *et al*., 2001; Orrett *et al*., 1995), urethral instrumentation and use of immunosuppressive drugs (Laupland *et al*., 2002).

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Abuse and indiscriminate use of antibiotics are responsible for antibiotic resistance. Organisms that show multiple resistance to antibiotics traditionally used in treating infections of the urinary tract are common (Aboderin *et al*., 2004; Chan *et al*., 1993). Studies have revealed an increasing resistance to first-line drugs such as cotrimoxazole and ampicllin used in treating uropathogens (Brown *et al*., 2003; Aboderin *et al*., 2004; Chan *et al*., 1993; Glikberg and Brawer-Ostrovsky, 1993; Adeyemo *et al*., 1994). Brown *et al*. (2003) speculated that the increasing use of broad spectrum antibiotics may result in changes in the microbiological and antibiotic sensitivity pattern of pathogens isolated from urinary tract infection.

Although much information on bacteriuria is available, little is known about the condition in most parts of the Middle-Belt region of Nigeria. The purpose of this study was to investigate the rate of bacteriuria in patients attending primary health care clinics within Makurdi metropolis, the organisms responsible and their antibiotic susceptibility pattern. The results of the study may provide baseline information which may be useful for rational therapy and adequate control measures.

MATERIALS AND METHODS

Study Area

The study area, Makurdi metropolis is the capital of Benue State (lying between latitudes 6° 18′ and 8° 12′ North and longitudes 6° 42′ and 9° 94′ East)and is located in the middle belt region of Nigeria. Its population is 114,376 (1991 National Census). Makurdi lies in the tropical guinea savanna of West Africa.

Study Population

Adult male and female patients within the age range of 18 and 75 and who were attending primary health care clinics within Makurdi metropolis were used for the study.

Microbiological Examination of Urine

Early morning midstream urine samples from 500 adult patients attending primary health clinics within Makurdi town were examined as described by Tugrul *et al*. (2005) and Olaitan (2006). A loopful of 5 mm wide standard wire containing 0.005 mL of urine was inoculated on MacConkey agar (Oxoid CM 109) plate and on blood agar plate. The plates were incubated aerobically for 24 h at 37°C. Bacterial counts exceeding 10^5 cfu mL^{-1} of urine indicated significant bacteriuria. Pure culture of each isolate was used for further identification.

Statistical and Mathematical Analyses

Associations, correlations, mean differences between variables and odds ratio were statistically determined using SPSS Version 10.0. Attributable risk was calculated based on rates of infection in males and in females.

Antibiotic Susceptibility Tests

Sixty four isolates of *S. aureus* and *E. coli* isolated from bacteriuric urine specimens were used for the antibiotic susceptibility testing. Reference cultures of *S. aureus* (NVRIVP-765) and *E. coli* (NVRIVP-820) obtained from the National Research Institute Vom, Plateau State, Nigeria, were used for comparison.

The Kirby-Bauer method of antibiotic susceptibility testing was used. Nutrient agar medium (Oxoid CM 3) was used for Gram-negative bacilli and blood agar medium containing 5% defibrinated blood in nutrient agar (Oxoid CM 3) for the Gram-positive cocci. A 6 h pure culture of each organism in peptone water (Oxoid CM 9) was inoculated onto an agar plate using sterile cotton swab in order

to obtain a uniform growth. The surface of agar plate was allowed to dry and antibiotic discs impregnated with ampicillin (Beecham, 25 μg), ampiclox (Beecham, 30 μg), amoxil (Beecham, 30 μg), ceporex (Glaxo, 30 μg), chloramphenicol (Alcon, 30 μg), ciproxin (Bayer, 10 μg), erythromycin (Remedica, 15 μg), fortum (Glaxo, 10 μg), furadantin (Maxi-care, 200 μg), gentamicin (Nicholas, 10 μg), nalidixic acid ((Maxi-care, 30 μg), norbactin (Ranbaxy, 30 μg), septrin (Wellcome, 30 μg) and streptomycin (Maxi-care, 25 μg), were placed on the agar plates using sterile forceps. After incubation (37°C, 18 h), the plates were examined for zones of inhibition. Zones less than 12 mm in diameter were considered resistant, while zones greater than 13 mm were considered sensitive (Benson, 1990).

Preparation of Antibiotic Discs

Antibiotic brands commonly used in chemotherapeutics within the study area were used to prepare the antibiotic discs. A modification of the process described by Stokes (1970) was used. Discs about 5 mm were cut from good quality blotting paper and sterilized in the oven at 160°C for 1 h. One drop (containing about 0.02 mL) of a sterile solution of the required antibiotic was dropped to each disc. The discs were then dried in the incubator with the lids of the Petri dishes tilted. Discs were stored in screw-capped bottles at 4°C.

RESULTS AND DISCUSSION

Out of the 500 specimens of urine examined bacteriologically, 70.8% had significant bacteriuria, thus indicating a high rate of urosepsis in the study population. The bacteria isolated were *Staphylococcus aureus* (42.4%), miscellaneous coliforms (27.1%), *Escherichia coli* (21.5%), *Streptococcus pyogenes* (6.8%), *Klebsiella aerogenes* (1.1%), *Pseudomonas aeruginosa* (0.6%) and *Salmonella typhi* (0.6%). Bacteriuria was significantly associated with the gender of patients ($r_{500} = 0.275$; $p = 0.000$) (Table 1). Bacteriuria was more common in women than in men, the infection rate being significantly higher in women than in men ($t_{500} = 24.773$; $p = 0.000$) (Table 2). The estimated odds ratio was 5.00 (95% confidence interval) and implies that the risk of bacteriuria was five times higher in women than in men. The risk of infection attributable to gender was 30.15%.

Gram-positive bacteria occurred more frequently in males than in females, whereas the Gram-negatives occurred more in females than in males ($t_{500} = 7.392$; $p = 0.000$). For example, the rate of *S. aureus* in males was 51.4%, but it was only 29.2% in females. Likewise, the rate of the coliforms including *E. coli* was 48.6% in females, but was 39.1% in males (Table 3).

Table 1: Correlation coefficients of the variables

Variables	Gender	Bacteriuria (Yes or No)	Gram reaction of isolates	Microorganism isolated
Gender	1.000			
Bacteriuria (Yes or No)	0.275**	1.000		
Gram reaction of isolates	0.356**	0.852**	1.000	
Microorganism isolated	0.291**	0.698**	0.792**	1.000

N = 500; **: Correlation is significant at the 0.01 level (2-tailed)

Table 2: Frequency of bacteriuria among the patients

*Bacteriuria	Male (n = 338)	Female (n = 162)	Total (N = 500)
Absent	128 (37.9)	18 (11)	146 (29.2)
Present	210 (62.1)	144 (88.9)	354 (70.8)
Risk ratio	1.6	8	
Odds ratio	Risk ratio in females ÷ Risk ration in males = 8/1.6 = 5.0		
Attributable risk	(Incidence of disease among females – Incidence of disease among males)*100/Incidence of disease among females = 30.15%		

*: t-test = 24.773 (p = 0.000), Values in parenthesis are in percentage

Table 3: Frequency of isolated bacteria according to sex

Isolated bacteria	Sex of patients: Male (%) (n = 210)	Female (%) (n = 144)	Total (%) (n = 354)	t-test N = 354
Gram-positive	60.0	33.3	49.2	-3.173; p = 0.002*
Gram-negative	40.0	66.7	50.8	
Total	100.0	100.0	100.0	
Staphylococcus aureus	51.4	29.2	42.4	-9.914; p = 0.000
Coliforms	22.9	33.3	27.1	
Escherichia coli	16.2	29.2	21.5	
Streptococcus pyogenes	8.6	4.2	6.8	
Klebsiella aerogenes	-	2.8	1.1	
Pseudomonas sp.	-	1.4	0.6	
Salmonella typhi	1.0	-	0.6	
Total	100.0	100.0	100.0	

*p = level of significance (2-tailed)

Table 4: Antibiotic susceptibility pattern of the bacteriuric isolates to commonly available antibiotics

Antibiotics	*Staphylococcus aureus* (%) (n = 32)	*S. aureus* (NVRIVP-765)	Coliforms (%) (n = 32)	*E. coli* (NVRIVP-820)
Ampicillin	100	R*	100	R
Ampiclox	94	R	100	R
Ceporex	97	S**	100	S
Chloramphenicol	97	S	97	R
Erythromycin	91	S	97	R
Fortrum	91	S	97	S
Amoxil	88	R	81	R
Septrin	78	R	78	R
Furadantin	72	S	50	R
Nalidix acid	34	S	28	R
Streptomycin	34	R	22	S
Ciproxin	0	S	0	S
Gentamicin	0	S	0	S
Norbactin	0	S	0	S

*: R-resistant; **: S-susceptible

The rates of resistance to commonly prescribed antibiotics, namely, ampicillin, ampiclox, ceporex, chloramphenicol, erythromycin, fortrum, amoxil, septrin and furadantin were high and ranged from 50 to 100% in both bacteria. On the other hand, none of these isolates were resistant to ciproxin, gentamicin and norbactin, even though low rates of resistance to nalidixic acid and streptomycin were observed. There was no significant difference in the resistance pattern of clinical isolates of *S. aureus* and the reference organism NVRIVP-765 ($t = -1.749$; $p > 0.05$), nor of *E. coli* and NVRIVP-820 ($t = 0.000$; $p > 0.05$) (Table 4).

Out of the 500 urine samples examined, 70.8% had significant bacteriuria. As in previous studies (Aboderin *et al*., 2004; Umeh, 2001; Umeh *et al*., 2006; Monane *et al*., 1995; Das *et al*., 2006), the incidence of significant bacteriuria was high and implies increased risk of urinary tract infection.

Present results show that bacteriuria was significantly associated with sex, the rate being five times (odds ratio, 5.0) as high in women as in men. As much as 30.15% (attributable risk value) cases of bacteriuria could be as a result of part of the study population being female. In other words, the infection would have been less by 30.15% if females were not included in the study. Among other factors, the structure of the female genitourinary tract as seen in the proximity of the female introital meatus to the anus and the vagina (Slack, 1997) and their short urethra (1.5 inches in women and 8.0 inches in men) is likely the most significant factor that predisposes women to a higher risk of bacteriuria. The predominance of *Staphylococcus aureus* and *Escherichia coli* among the urinary pathogens isolated agrees with results of other studies (Umeh, 2001; Aboderin *et al*., 2004; Umeh *et al*. 2006; Olaitan, 2006; Das *et al*., 2006; Hooton *et al*., 2000; Akinloye *et al*., 2006) and thus suggests that the organisms implicated in cases of significant bacteriuria are not dependent on geographical locations.

The antibiotic susceptibility pattern of *Staphylococcus aureus* and the coliform bacteria (the major bacteria isolated in this study) indicates a high rate of resistance to commonly prescribed antibiotics such as ampicillin, ampiclox, ceporex, chloramphenicol, erythromycin and fortrum. The high rate of resistance to antibiotics may be attributed to indiscriminate use of antibiotics which is a common practice in this part of the world. Most of the antibiotics tested are those that could easily be purchased across the counter in many drug stores and in most cases without doctor's prescription. In such cases the correct dose of the antibiotics is not usually taken. The public needs to be educated on the dangers of indiscriminate use and abuse of antibiotics.

CONCLUSIONS

Present findings have underscored the clinical importance of bacteriuria in the area studied. The resistance of bacteriuric isolates to commonly available antibiotics is high and this fact must be taken into account when selecting treatment strategies. The results of this study may also be a useful guide when planning control programmes against infections of the urinary tract. As has been suggested (Das *et al*., 2006), continuous and regular monitoring of the antibiotic resistance pattern of bacteria isolated in cases of significant bacteriuria is necessary for optimal treatment of affected individuals.

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