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Full Length Research Paper

Antibiotic susceptibility pattern of uropathogenic bacterial isolates from Aids patients in a Nigerian tertiary hospital

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To determine the antibiotic susceptibility pattern of bacterial isolates causing urinary tract infections in AIDS patients, 500 AIDS patients and 500 non-AIDS patients (served as control) each on admission were recruited into the study between 2007 and 2008 in Aminu Kano Teaching Hospital, Kano, Nigeria. Urine samples were collected with sterile universal bottles and analysed with appropriate laboratory methods and antibiotic susceptibility test was carried out by disk diffusion technique in accordance with CLSI (formerly, NCCLS) criteria, 2006. The results were analyzed using SPSS 11.0 statistical software, p-value <0.05 were considered significant at 95% confident limit. *Escherichia coli* were the most common isolate in the both groups. A wider spectrum of micro-organisms were isolated from AIDS group patients compared to control group. Most of the urinary bacterial isolates (n=350) from both groups were highly sensitive to ceftazidime (95%) and ciprofloxacin (88%). There was generally low sensitivity of bacterial isolates to most antibiotics in common use such as co-trimoxazole (40%), tetracycline (25%), chloramphenicol (33%) and ampicillin (40%) with no significant difference between the AIDS and control group. There was no significant difference in the antibiotic susceptibility pattern between the AIDS group and the control group in the study. Rational use of antibiotics with regular antibiotics susceptibility surveillance studies is recommended to maintain high antibiotic therapeutic profile.

Keywords: Urinary bacterial isolates, antibiotic susceptibility pattern, AIDS patients.

INTRODUCTION

Opportunistic pathogens of bacterial, fungal, parasitic and viral origin immensely contribute to mortality and morbidity in AIDS patients (UNAID, 2005). Microbiologic analysis of clinical specimens from this group such as cerebrospinal fluid, sputum, stool, blood have shown an expanded spectrum of pathogens however, less work has been done on urine samples of HIV/AIDS patients, especially, on the African continent (Wilson and Gaido, 2004; Abdul, 2005) .

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Considering the role of opportunistic bacterial infections in the natural history of AIDS (UNAID, 2004) and the limited reports on the antibiotics susceptibility patterns of urinary bacterial isolates of such patients, it is timely to draw up a tool for prompt diagnosis and specific treatment based on a good knowledge of common urinary bacterial isolates and their antibiotics susceptibility patterns in this locality.

We therefore, embarked on this study to ascertain the bacterial isolates and their antibiotics susceptibility patterns in urine samples of AIDS patients in Aminu Kano Teaching Hospital, Kano north- western Nigeria. The findings will be useful to the relevant health personnel

who are often constrained by time in the course of effective management of AIDS patients in this region.

MATERIALS AND METHOD

Study Design

A prospective study of microscopy, culture, and susceptibility of urine samples of AIDS patients and a control group (non- AIDS patients) in Aminu Kano Teaching Hospital, Kano, from September 2007 to September 2008 was carried out in medical microbiology and parasitology laboratory.

A group of patients on admission at Aminu Kano Teaching Hospital, Kano confirmed to be HIV positive by western blot assay and with clinical features of AIDS and another group of patients with age and sex matched on admission in the same center and confirmed to be HIV negative to serve as control were recruited into the study. All the patients were conscious and not catheterized and willing to participate in the study.

Consent was sought and obtained from the patients in writing. Patients were assured of confidentiality of information. A well structured questionnaire was either self administered or interviewer administered depending on the literary level of the subject. The items on the questionnaire included features associated with AIDS such as progressive weight loss (>10kg/body weight in the past 1 year), persistent cough, prolonged (>1 month) diarrhea, prolonged (>1 month) fever and some demographic data including age, sex, address and occupation.

Laboratory Procedure

The samples were early morning midstream urine (MSU) samples, collected in sterile universal bottles (about 15ml) and processed immediately or stored at 4 ◦C within 4 hours.

Urine samples were examined macroscopically and microscopically. Uncentrifuged urine was examined under x 40 objective for pus cells, red blood cells, casts and crystals and other important features and later centrifuged at 1500 rpm and sediments examined for parasites. The uncentrifuged urine samples were inoculated with a calibrated sterile loop delivering 0.001ml of urine onto Cysteine Lactose Electrolyte Deficient (CLED) agar, Blood agar and Chocolate agar plates. Two sets of the culture plates were incubated, one for aerobic and the other for anaerobic incubation at 36 ◦C for 18 to 24 hours. A number of more than 100 colonies per ml of urine were considered significant. A significant bacteriuria was taken as bacteria count equal to, or in excess of 10^5 per milliliter of urine. The colonies were identified by standard biochemical tests and antibiotics susceptibility of the bacteria was performed by Kirby-Bauer diffusion technique in accordance with CLSI(formerly, NCCLS) criteria, 2006 (CLSI,2006) . The following quality controlled antibiotic disks for locally available antibiotics for urinary pathogens were used: ampicillin (25 µg), augmentin (30 µg), gentamicin (10 µg), co-trimoxazole (25 µg), ceftazidime (30 µg), ceftriaxone (30 µg), nitrofurantoin (300 µg), nalidixic acid(30 µg), ciprofloxacin(5 µg), tetracycline(30 µg), piperacillin(5 µg), cloxacillin(5 µg), chloramphencol (30µg). Control strains used were *E. coli* NCTC 10418, *Pseudomonas aeruginosa* NCTC 10662 and *Staphylococcus aureus* NCTC 6571.The results were analyzed using SPSS 11.0 statistical software; chi-square(X^2) was used to compare association between proportions and p- values, 0.05 was considered significant at 95.0% confidence level. Ethical approval was given by the ethics committee of the hospital.

RESULT

Five hundred subjects recruited from the AIDS group and 500 subjects from the non- AIDS group (served as a control) respectively with a total of 1000 subjects. In a total of 1000 subjects, 250 (25%) subjects were diagnosed to have urinary tract infection(UTI) in AIDS group while 100 (10.0%) subjects were diagnosed to have UTI in the control group (non-AIDS group); this difference was found to be statistically significant ($p<0.05$) (Table 1).

The infection rate was found to be higher in females (n=175/250; 70.0%) than males (n=75/250; 30.0%) among the AIDS patients with UTI, similar with the 65.0% (n=65/100) and 25.0% (n=25/100) found among females and males respectively among the control group with UTI. These gender differences were found to be statistically significant within the two respective groups ($p<0.05$) but statistically insignificant between same gender among the two groups under study ($p>0.05$). Table 2 shows the organisms recovered from the urine samples of both AIDS and the control groups.

Tables 3 and 4 show the commonest organisms recovered from the urine of subjects that had UTI in both AIDS (Test – T) and the non- AIDS (control - C) groups and their antibiotic susceptibility patterns. *E. coli* was the most common isolate in the both groups. The difference in pattern of antibiotic susceptibility in the two groups was not statistically significant ($p>0.05$). But a wider spectrum of micro-organisms were isolated from AIDS group patients compared to control group.

DISCUSSION

The work was designed to determine the bacterial isolates of urinary tract infection and their antibiotics susceptibility pattern in AIDS patients as compared to the control group on admission in Aminu Kano Teaching Hospital in Kano, north western Nigeria.

The rate of UTI among AIDS patients in the study was 25.0% (70.0% females and 30.0% males) and the control group was 10.0%; this difference was found to be statistically significant ($p<0.05$). The finding was similar to a work done in Jos, north -central Nigeria with a prevalence of 24.0% for AIDS group of patients and 10.6% for non- AIDS group respectively (Jombo et al., 2006) . UTI was common in age fifty years and above in both the test and the control groups. The incidence of urinary tract infection increases greatly with age. A significant number of infections occur in men only after the age of 50 years when prostatic hypertrophy or other urinary tract abnormality may occur (Abdul, 2005). The short female urethra greatly predisposes to infection of the urinary tract by the ascending route and the great majority of symptomatic infections are in females. A wider

Table 1. Age distribution of urinary tract infection among study population

Age (Years)	Total no. of Subjects n (%)	* UTI in AIDS group of patients n (%)	UTI in **control Group n (%)
0-9	100 (10)	10 (10)	8 (8)
10-19	150 (15)	35 (23)	15 (10)
20-29	200 (20)	50 (25)	10 (5)
30-39	250 (25)	60 (24)	20 (8)
40-49	200 (20)	40 (20)	15 (7.5)
50-59	7 0(7)	40 (57)	20 (28.6)
60-69	30 (3)	15 (50)	12 (40)
TOTAL	**1000 (100)**	**250 (25)**	**100 (10)**

P<0.05

* UTI – urinary tract infection; **Control group – non- AIDS patients

Table 2. Organisms recovered from the urine samples of AIDS patients and the control group in Aminu Kano Teaching Hospital, Kano, 2007-2008

Isolates	*UTI in AIDS group of patients No. (%)	UTI in **control group No. (%)	Total No. (%)
Escherichia coli	75(30)	65(65)	140(40)
Klebsiella spp	55(23)	15(15)	70(20)
Staphylococcus aureus	40(16)	0	40(11)
Pseudomonas auruginosa	25(10)	0	25(7)
Salmonella spp	16(6)	1(1)	17(5)
Proteus spp	25(10)	9(9))	34(10)
Coagulase Negative staphylococcus	8(3)	5(5)	13(4)
Enterococcal spp	6(2)	5(5)	11(3)
TOTAL	**250(25)**	**100(10)**	**350(100)**

* UTI – urinary tract infection; **Control group – non AIDS patients

Table 3. Antibiotics susceptibility pattern of urinary bacterial pathogens isolated from the AIDS group of patients in Aminu Kano Teaching Hospital, Kano, 2007-2008

ANTIBIOTICS (susceptibility%)	BACTERIAL ISOLATES							
	E.coli 75 (%)	*Klebsiella* 55 (%)	*S. aureus* 40 (%)	*P. aeruginosa* 25 (%)	*Salmonella spp* 16 (%)	*Proteus spp* 25 (%)	*CNS.* 8 (%)	*Enterococci* 6 (%)
AMPICILLIN	15(20)	-	16(40))	0(0)	8(50)	-	4(50)	2 (33)
AUGMENTIN	56(75)	33(60)	-	13(52)	13(81)	14(56)	-	-
CO-TRIMOXAZOLE	30(40)	0(0)	-	0(0)	8(50)	0(0)	-	-
CEFTAZIDIME	72(96)	50(90)	-	23(92)	16(100)	24(96)	-	-
CEFTRIAXONE	-	-	36(90)	-	-	-	8(100))	4(67)
CIPROFLOXACIN	68(91)	44(80)	36(90)	-	16(100)	-	7(88)	4(67)
GENTAMICIN	53(71)	33(60)	20(50)	13(52)	-	14(56)	4(50)	3(50)
NITROFURANTOIN	53(71)	22(40)	-	-	-	14(56)	-	-
NALIDIXIC ACID	45(60)	33(60)	-	-	-	14(56)	-	-
TETRACYCLINE	-	-	0(0)	-	4(25)	-	2(25)	0(0.0)
CHLORAMPHENICOL	-	-	20(50)	0(0)	10(63)	-	2(25)	0(0.0)
PIPERACILLIN	-	-	-	23(92)	-	-	-	-
CLOXACILLIN	-	-	36(90))	-	-	-	6(75)	-

*E. coli – Escherichia coli, S. aureus –Staphylococcus aureus, CNS – Coagulase Negative Staphylococci, spp –species . – (Not tested)

Table 4. Antibiotics sensitivity pattern of urinary bacterial pathogens isolated from control group of patients in Aminu Kano Teaching Hospital, Kano, 2007- 2008.

ANTIBIOTICS (susceptibility %)	BACTERIAL ISOLATES						
	E. coli 65 (%)	*Klebsiella Spp.* 55 (%)	*P. vulgaris 3* (%)	*P. mirabilis* 6 (%)	*Enterococci* 5 (%)	*Salm. spp 1* (%)	*CoNS. 5* (%)
AMPICILLIN	20(30)	-	-	-	2((40)	0(0)	3(60)
AUGMENTIN	46(70)	28(50)	2(67)	5(75)	-	1(100)	-
CO-TRIMOXAZOLE	33(50)	0(0)	0(0)	1(17)	-	0(0)	-
CEFTAZIDIME	59(90)	50(90)	3(100)	6(100)	-	1(100)	-
CEFTRIAXONE	-	-	-	-	4(80)	-	4(80)
CIPROFLOXACIN	59(90)	44(80)	-	-	4(80)	1(100)	4(80)
GENTAMICIN	46(70)	28(50)	1(33))	5(83)	3(60)	-	3(60)
NITROFURANTOIN	46(70)	28(50)	1(33)	4(64)	-	-	-
NALIDIXIC ACID	46(70)	33(60)	1(33)	3(60)	-	-	-
TETRACYCLINE	-	-	0(0)	0(0)	0(0.0)	0(0)	0(0)
CHLORAMPHENICOL	-	-	-	-	0(0.0)	0(0)	0(0)
PIPERACILLIN	-	-	-	-	-	-	-
CLOXACILLIN	-	-	-	-	-	-	4(80)

**E. coli – Escherichia coli*, *P. mirabilis – Proteus mirabilis*, *P. vulgaris – Proteus vulgaris*, *Salm. spp.–Salmonella spp.*, CoNS – *Coagulase Negative Staphylococci*

spectrum of micro-organisms which included *Pseudomonas auruginosa* and *Staphylococcus aureus* were isolated from urine samples of AIDS group of subjects with UTI in the study. Again, the finding was similar to a study on UTI among AIDS patients carried out in Benue, central Nigeria 2010, which recorded a wider spectrum of micro-organisms such as *Candida species, Pseudomonas auruginosa* and *Staphylococcus aureus* (Okwori et al., 2010). Hence the high rate of UTI coupled with wide spectrum of opportunistic pathogens found in the urine samples of the AIDS subjects may not be unconnected with the general depression of the immunity of these patients against several infections. This finding stresses the importance of adopting sterile invasive procedures in management of AIDS patients, such procedures as; intravenous injections, intramuscular injections, vaccinations, catheterization, endotracheal intubation among others.

Also, prompt and effective treatment of opportunistic infections in AIDS patients would reduce the cycles of opportunistic infections with attendant complications.

The study recorded a preponderance of *E. coli* (40%) isolates from both the AIDS and the control groups. The antibiotics susceptibility patterns of Gram -negative bacteria, such as *E. coli, Klebsiella, Proteus,* and *Salmonella* species in the urine samples from AIDS patients and the control group were highly susceptible to ceftazidime, ciprofloxacin and augmentin. On the other hand, the Gram- positive bacteria such as *Staphylococcus aureus*, *Enterococci* and *Coagulase negative staphylococci* showed high level susceptibility to ceftriaxone, ciprofloxacin and gentamicin. Generally, the bacterial isolates showed little or no susceptibility to tetracycline, chloramphenicol, ampicillin and co-trimoxazole. In a similar study on AIDS group in Jos, north-central Nigeria, showed a 41.6% preponderance of *E.coli* with antibiotic susceptibility above 90% to ceftazidime, augmentin, norfloxacin and gentamicin and very low susceptibility to ampicillin, tetracycline, mecillinam and piperacillin (Jombo et al., 2006) . An earlier study on UTI in a general population carried out in Kano, north-western Nigeria, recorded a 51% high rate of E. coli isolates, in contrast to 40% in the present study (Nwadioha et al., 2010). Findings in Nsukka, south-eastern Nigeria, showed a higher proportion of Gram-negative bacteria were resistant to cephalexin and co-trimoxazole (80%) each, ampicillin (73.3%), gentamicin and nalidixic acid (70%) each, while Gram-positive bacteria were resistant to streptomycin and erythromycin by 40.5% each (Chah et al., 2003). In United States of America, resistance of over 60% was recorded by several bacterial isolates against the antibiotics tested such as ciprofloxacin, cephalexin in a study (Ishikawa et al., 2004). In our study, the antibacterial activity of ciprofloxacin was very high(88%) compared to the activity of nalidixic acid(55%), a first generation quinolone. This calls for caution in the use of ciprofloxacin(a modified florinated structured quinolone) in the locality.

Bacterial resistance to ciprofloxacin is very high in the Far East and United States of America. In China,

gonococal resistance to ciprofloxacin rose from 17.6% to 72.7% during 1996 – 2001 in Guangzhou (Ishikawa et al., 2004).

The findings in our study showed that there was no statistical significant difference in the antibiotics susceptibility profile of bacteria recovered from AIDS subjects compared to the non- AIDS subjects($p>0.05$). It shows that bacterial infections in AIDS patients have no special advantage over non-AIDS patients in the directions of the speed of acquisition of resistant genes. Interestingly, it is a welcome development in a time antibiotics prophylaxis is increasingly used in management of AIDS patients (WHO, 2004).

The profile of antibiotics susceptibility in the study goes a long way to describe the degree of abuse and misuse of routine antibiotics used in our society (Ampicillin, Tetracycline, Co-trimoxazole) (Ozumba, 2006). Education of the public on the exercise of restraints on abuse and misuse of antibiotics will reduce the current pattern of bacterial antibiotic resistance. Proper disinfection of hospital ware, proper disposal of hospital wastes and implementation of effective infection control policies in the hospitals will reduce the rate of antibiotics resistance of bacteria in / outside the health facilities.

Limitation in the study included occasional supply of contaminated/ stale urine specimens in which it was difficult sometimes to discern contamination from original infection and occasional power failures. A multi-center study program in order to test the findings from the present study will therefore be recommended.

The study therefore has shown that there is no significant statistical difference in antibiotics susceptibility pattern of bacteria isolates in urine samples from AIDS patients as compared to the control group but wider spectrum of micro-organisms were isolated from urine samples of AIDS group patients**.** The study has also shown that either Ceftriaxone or Ciprofloxacin can be used as first line of choice in the treatment of UTI in AIDS patients where antibiotics susceptibility results are inadvertently unavailable.

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