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A comparative study of bacterial isolates from the urine samples of AIDS and non-AIDS patients in Benue, Nigeria

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ABSTRACT

Objective: To determine the common bacterial causes of urinary tract infection and their antibiotic susceptibility pattern in AIDS patients versus non-AIDS patients. **Methods:** One thousand consecutive AIDS patients with signs and symptoms of AIDS and non-AIDS patients (served as control) each on admission were recruited into the study between January 2005 to January 2008, in Federal Medical Center, Makurdi. 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 National Committee for Clinical Laboratory Standards (NCCLS, now CLSI) criteria. The results were analysed using SPSS 11.0 statistical software. **Results:** Urine samples of AIDS patients with urinary infection had a more spectrum of micro-organisms including *Candida organisms*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*. Ceftriaxone, Ceftazidime or Ciprofloxacin had a remarkably high anti-bacterial activity across the two study groups. A general resistance was recorded in ampicillin, tetracycline and co-trimoxazole. There was no significant difference in antibiotic susceptibility patterns between AIDS and non- AIDS patients($P>0.05$). **Conclusions:** A reduction in unnecessary use of antibiotics as well as infection control should be encouraged in our health facilities.

1. Introduction

Opportunistic infections in HIV/AIDS cut across bacteria, fungi, parasites and viruses and immensely contribute to mortality and morbidity in this condition[1]. Much work on laboratory analysis has been documented on specimens such as sputum , blood, stool and cerebrospinal fluid from people living with AIDS (PLWA), showing a large spectrum of pathogens[2]. On the other hand, less work has been done on urine samples of HIV/AIDS patients especially on African continent[3].

Considering the role of opportunistic bacterial infections in the natural history of AIDS[4] 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.

The study was therefore to ascertain the bacterial isolates and their antibiotics susceptibility patterns in urine samples of AIDS versus non-AIDS patients in Benue, central 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 Benue, central Nigeria, which is a known HIV/AIDS endemic place[5].

2. Materials and methods

A prospective study of microscopy, culture, and sensitivity of urine samples of AIDS and non-AIDS patients from January 2003 to January 2009 was carried out in medical microbiology and parasitology laboratory of Federal Medical Center, Makurdi .

In the study, a group of patients on admission at FMC Makurdi confirmed to be HIV positive by western blot assay and with clinical features of AIDS were recuited; and another group of patients with age and sex matched on admission in the same center and confirmed to be HIV negative were recruited as control. All patients were conscious and not catheterized and willing to participate in the study. Consent was sought and obtained from the patients.

A well structured questionnaire was either self administered or interviewer administered depending on the literary level of the subjects. The items on the questionnaire included features associated with AIDS such as progressive

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weight loss(>10 kg loss of body weight in the past 1 year), persistent cough, prolonged diarrhea (>1 month), prolonged fever (>1 month) and some demographic data including age, sex, address and occupation.

The samples were early morning midstream urine (MSU) samples, collected in sterile universal bottles(about 15 mL) and processed immediately or stored at 4 ℃ within 4 hours. Urine samples were examined macroscopically and microscopically. Uncentrifuged urine was examined under ×40 objective for pus cells, red blood cells, casts and crystals and other important features and later centrifuged at 1 500 rpm and sediments examined for parasites. The uncentrifuged urine samples were inoculated with a calibrated sterile loop delivering 0.001 mL 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 ℃ for 18 to 24 hours. A number of more than 100 colonies per mL of urine were considered significant. A significant bacteriuria count was taken as any count equal to, or in excess of 105 per milliliter of urine. The colonies were identified by standard biochemical tests and antibiotics sensitivity of the bacteria was performed by Kirby-Bauer diffusion technique in accordance with National Committee for Clinical Laboratory Standards (NCCLS) criteria (NCCLS,2002)[6]. Control strains used were *Escherichia coli* (*E. coli*) NCTC 10418, *Pseudomonas aeruginosa* (*P. aeruginosa*) NCTC 10662 and *Staphylococcus aureus* (*S. aureus*) NCTC 6571.The results were analyzed using SPSS 11.0 statistical software; chi-square(χ^2) was used to compare association between proportion, and P values<0.05 was considered significant at 95.0% confidence level. Ethical approval was given by the ethics committee of the hospital.

3. Results

Two thousand subjects recruited from the AIDS group and the non-AIDS (served as a control) respectively. Out of the 2 000 subjects recruited from each group, AIDS and non-AIDS groups, 540 (27.0%) and 240 (12.0%) were diagnosed to have urinary tract infection(UTI) respectively. Age distribution of urinary tract infection (UTI) among study population was shown in Table 1. Significant difference was found (P<0.05).

The infection rate was found to be higher in females (n=432, 80.0%) than males (n=108, 20.0%) among the AIDS patients with UTI, similar with the 70.0% (n=168) and 30.0% (n=72) found among females and males, respectively among those without AIDS. 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 (P>0.05).

Table 2 showed the organisms recovered from the urine samples of both AIDS(Test, T) and the non-AIDS (Control, C) groups.

Table 3 showed *P. aeruginosa* as well as *S. aureus* was isolated from AIDS group of patients only. *P. aeruginosa* was very sensitive to Piperacillin (93%) and Ceftazidime (93%); moderately sensitive to Augmentin (51%) and gentamycin; resistant to Ampicillin (0%), Co-trimoxazole (0%) and Chloramphenicol (0%). *S. aureus* showed a high sensitivity to Cloxacillin (89%), Cefriaxone(92%) and Ciprofloxacin(92%); moderate sensitivity to Gentamycin (56%), Ampicillin (46%) and Chloramphenicol (50%) but showed resistance to Tetracycline (0%).

Table 3 and 4 showed that antibiotics sensitivity pattern of *E. coli* was similar in both AIDS and non-AIDS patients (P>0.05). *Proteus mirabilis* was highly sensitive to Ceftazidime (93%); moderately sensitive to Gentamycin (56%), Augmentin (56%), Nitrofurantoin (56%) and Nalidixic acid (56%); resistant to Co-trimoxazole (0%). There was no statistical significant difference between the AIDS and the non- AIDS groups(P>0.05).

Salmonella species as well as *Enterococcal* species were isolated from the two groups of patients. *Salmonella* species were highly sensitive to Ciprofloxacin (100%), Ceftizidime (100%) and Augmentin (79%); moderately sensitive to Chloramphenicol (66%), Ampicillin (53%), Co-trimoxazole (53%), Tetracycline (26%). *Enterococcus* was moderately sensitive to Ceftriaxone (73%), Ciprofloxacin (73%) and Gentamycin (64%); weakly sensitive to ampicillin (45%); resistant to Chloramphenicol (0%) and Tetracycline (0%). The difference in pattern of antibiotic sensitivity in the two groups was not statistically significant (P>0.05).

In summary, subjects recruited from AIDS group had a higher prevalence of UTI (27.0%) than the non-AIDS group

Table 1
Age distribution of urinary tract infection (UTI) among study population [n(%)].

Age(years)	Subjects	UTI in AIDS group	UTI in non-AIDS group
0-9	200(10.0)	32(16.0)	26(13.0)
10-19	300(15.0)	100(33.3)	30(10.0)
20-29	400(20.0)	140(35.0)	50(12.5)
30-39	500(25.0)	120(24.0)	59(11.8)
40-49	400(20.0)	90(22.5)	40(10.0)
50-59	140(7.0)	40(28.6)	15(10.7)
60-69	60(3.0)	18(30.0)	20(33.3)
Total	2 000(100.0)	540(27.0)	240(12.0)

Table 2
Organisms recovered from the urine samples of AIDS and non-AIDS patients in Benue, 2005-2008 [n(%)].

Isolates	UTI in AIDS group (n=540)	UTI in non-AIDS group(n=240)	Total
E. coli	135(25.0)	154(64.0)	289(37.0)
Klebsiella spp	108(20.0)	36(15.0)	144(18.0)
Candida spp	108(20.0)	0(0.0)	108(14.0)
S. aureus	54(10.0)	0(0.0)	54(6.9)
P. auruginosa	43(8.0)	0(0.0)	43(6.0)
Salmonella spp	38(7.0)	2(1.0)	40(5.1)
Proteus spp	27(5.0)	24(10.0)	51(6.5)
Coagulase negative Staphylococcus	16(3.0)	14(6.0)	30(3.8)
Enterococcal spp	11(2.0)	10(4.0)	21(2.7)

Table 3
Antibiotics sensitivity pattern of urinary bacterial pathogens isolated from the AIDS group of patients in Benue, 2005–2008 [*n*(%)].

Antibiotics	Bacterial isolates							
	E. coli (*n*=135)	*Klebsiella* (*n*=108)	*S. aureus* (*n*=54)	*P. aeruginosa* (*n*=43)	*Salmonella* spp (*n*=38)	*Proteus* spp (*n*=27)	CNS (*n*=16)	*Enterococci* (*n*=11)
Ampicillin	40(30)	–	25(46)	0(0)	20(53)	–	8(50)	5(45)
Augmentin	100(74)	60(56)	–	22(51)	30(79)	15(56)	–	–
Co-trimoxazole	80(59)	0(0)	–	0(0)	20(53)	0(0)	–	–
Ceftazidime	130(96)	100(93)	–	40(93)	38(100)	25(93)	–	–
Ceftriaxone	–	–	50(93)	–	–	–	16(100)	8(73)
Ciprofloxacin	120(89)	90(83)	50(93)	–	38(100)	–	15(94)	8(73)
Gentamycin	100(74)	60(56)	30(56)	22(51)	–	15(56)	8(50)	7(64)
Nitrofurantoin	100(74)	50(46)	–	–	–	15(56)	–	–
Nalidixic acid	100(74)	66(61)	–	–	–	15(56)	–	–
Tetracycline	–	–	0(0)	–	10(26)	–	5(31)	0(0)
Chloramphenicol	–	–	27(50)	0(0)	25(66)	–	5(31)	0(0)
Piperacillin	–	–	–	40(93)	–	–	–	–
Cloxacillin	–	–	48(89)	–	–	–	12(75)	–

CNS: Coagulase negative *staphylococci*, spp: species.

Table 4
Antibiotics sensitivity pattern of urinary bacterial pathogens isolated from non-AIDS group of patients in Benue ,2005–2008 [n(%)].

Antibiotics	Bacterial isolates						
	E. coli (*n*=154)	*Klebsiella* (*n*=36)	*P. vulgaris* (*n*=10)	*P. mirabilis* (*n*=14)	*Enterococci* (*n*=10)	*Salm.* spp (*n*=2)	CoNS (*n*=14)
Ampicillin	50(32)	–	–	3(23)	4(40)	1(50)	8(57)
Augmentin	110(71)	20(56)	6(60)	11(75)	–	2(100)	–
Co-trimoxazole	85(55)	0(0)	0(0)	4(27))	–	1(50)	–
Ceftazidime	150(97)	33(92)	8(80)	14(100)	–	2(100)	–
Ceftriaxone	–	–	–	–	7(70)	–	13(93)
Ciprofloxacin	140(91)	30(83)	–	–	7(70)	2(100)	13(93)
Gentamycin	110(71)	20(56)	6(60)	12(85)	6(60)	–	9(64)
Nitrofurantoin	120(78)	20(56)	5(50)	10(65)	–	–	–
Nalidixic acid	120(78)	22(61)	5(50)	9(55)	–	–	–
Tetracycline	–	–	0(0)	0(0)	0(0)	0(0)	3(21)
Chloramphenicol	–	–	–	–	0(0)	2(100)	6(43)
Piperacillin	–	–	–	–	–	–	–
Cloxacillin	–	–	–	–	–	–	9(64)

P. mirabilis: *Proteus mirabilis*, *P. vulgaris*: *Proteus vulgaris*, *Salm*. spp.: *Salmonella* spp., CoNS: Coagulase negative *Staphylococci*.

(12.0%). Analysis of urine samples from AIDS patients with UTI, showed a wider spectrum of micro-organisms including *P. aeruginosa*, *S. aureus* and *Candida* species. There was no statistical significant difference in the antibiotic susceptibility pattern of the AIDS and non-AIDS groups of patients (P>0.05). Ceftazidime and Ciprofloxacin showed highest antibacterial activity against gram negative bacteria, while Ceftriaxone and Ciprofloxacin showed a higher antibacterial activity against gram positive bacteria.

4. Discussion

The study was to determine the bacterial isolates of urinary tract infection and their antibiotics susceptibility pattern in AIDS versus non- AIDS patients in Benue ,central Nigeria.

The prevalence of UTI among AIDS patients in the study was 27.0% (80.0% females and 20.0% males) and non-AIDS was 12.0%; this difference was found to be statistically significant (P<0.05). The finding was similar to a work done in Jos, Nigeria with a prevalence of 24.0% for AIDS group of patients and 10.6% for non-AIDS group respectively[7]. A wider spectrum of micro-organisms which included *Candida* species, *P. auruginosa* and *S. aureus* isolated from urine samples of AIDS group of subjects with UTI. Hence the high rate of prevalence 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[8]. This finding stresses the importance of adopting sterile invasive /non 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[8].

The antibiotics susceptibility patterns of gram-negative bacteria, such as *Klebsiella*, *Proteus*, *Salmonella* species strains in the urine samples from AIDS and non-AIDS patients were highly sensitive to Ceftazidime, Ciprofloxacin and Augmentin. On the other hand, the gram-positive bacteria such as *S. aureus*, *Enterococci* and *Coagulase* negative *staphylococci* showed a high level sensitivity to Ceftriaxone, Ciprofloxacin and Gentamycin. The result of antibiotic sensitivity pattern was similar to a work done on hospital versus community-acquired UTI, that recorded similar high antibacterial activity range for Ceftriaxone, Ceftazidime and Ciprofloxacin against the two groups[9]. There was no statistical significant difference in the antibiotics sensitivity 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[10].

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)[11]. 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 or outside the health facilities.

Limitation in the study included 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 non-AIDS patients. The study has also shown that there is significant statistical difference between the prevalence of UTI in AIDS group of subjects and non-AIDS group. Lastly, there was a wider spectrum of micro-organisms in the urine samples from AIDS group of subjects than in non-AIDS group ($P<0.05$).

Conflict of interest statement

We declare that we have no conflict of interest.

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