Coliforms Associated With Bacteriuria In Human Immunodeficiency Virus (HIV) Positive Patients Attending The Bamenda Provincialhospital

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Citation

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Abstract

The prevalence and antibiotic susceptibility pattern of coliform associated with bacteriuria among HIV positive patients attending the Bamenda Provincial Hospital was studied. Out of the 240 urine specimens from HIV positive patients, coliforms were isolated in 49 (20.4%). HIV positive patients with CD4 counts between 1-50 cell/mm^3 had more coliforms with significant bacteriuria than those with higher counts from 151-200 (22.5% vs. 6.1%, $p < 0.05$). Female HIV positive patients had a higher prevalence of coliform organisms than their male counterparts (21.4% vs. 18.9%, $P > 0.5$). Eleven different species of coliform organisms were isolated with Escherichia coli (30.6%) being the most frequently encountered. Resistance to Bacitracin (100%), Co-trimoxazole (85.7%) and Trimethoprim (77.5%) were the most common. Isolates were most sensitive to Ceftriazone (77.5%), Perfloxacin (73.5%) and Netilmicin (71.4%). Ceftriazone, Perfloxacin and Netilmicin are drug of choice in the treatment of urine infections caused by coliform organisms of HIV/AIDS patients.

INTRODUCTION

The Gram-negative bacilli of the genera Escherichia, Klebsiella, Enterobacter, Serratia, Citrobacter, and Proteus are members of the normal intestinal flora of humans and animals and may be isolated from a variety of environmental sources [1]. With the exception of Proteus, they are sometimes collectively referred to as the coliform bacilli i.e. gram negative, facultative anaerobic, non spore-forming rods that ferment lactose and are all motile (except for Klebsiella). Many of these microorganisms used to be dismissed as harmless commensals. Today, they are known to be responsible for major health problems worldwide. A limited number of species, including Escherichia coli, Klebsiella pneumoniae, Enterobacter aerogenes, Enterobacter cloacae, and Serratia marcescens, are responsible for most infections produced by this group of organisms [2]).

OBJECTIVE OF STUDY

The aim of this research project was to identify coliform bacteria in the urine of HIV positive patients and their antibiotic sensitivity pattern.

HYPOTHESIS

Bacteriuria due to coliforms occurs with increased frequency in HIV positive AIDS patients and represents an important cause of morbidity.

MATERIALS AND METHODS

STUDY AREA AND DESIGN

This study was based on laboratory investigations. A total of three hundred urine samples were collected. Two hundred and forty (240) urine specimens came from HIV /AIDS patients (obtained from both in – and outpatients for whom the CD4 count was requested for control and also from patients who came for voluntary testing) and sixty were obtained from healthy HIV negative individuals (control).

Demographic data including age and sex of patients was taken into consideration.

THE HIV TEST

The HIV test was carried out following the HIV test algorithm established by the Cameroon Ministry of Public Health in 2001. A rapid test to screen for the presence of HIV antibodies was performed using the rapid test kid,

Dermine™ [3]. All positive tests were confirmed with the ImmunoComb® II HIV 1 & 2 Bispot kit [4] according to manufacturer's instructions.

SAMPLE COLLECTION

All samples were collected by the mid-stream catch technique into sterile wide-mouth screw-capped bottles according to methods previously reported [5]. All specimens were examined within 60 minutes.

ISOLATION AND IDENTIFICATION OF ORGANISMS

Specimens were inoculated onto MacConkey agar plates which were prepared as previously described [6]; and incubated at 37ºC for 18 hours; after which the plates were then examined for reddish, mucoid, or dark-centered colonies.These suspect colonies were subjected to oxidase test to rule out oxidase positive gram-negative bacilli. Suspect colonies were sub cultured on chocolate agar plates to obtain pure colonies. They were gram stained, inoculated KIA slants on and tested for motility test (7).

SPECIES IDENTIFICATION

Species identification made use of the Analytical Profile Index (API) 20E Scheme. This was performed according to the manufacturer's instructions.

ANTIBIOTIC SUSCEPTIBILITY TESTING

The disc diffusion (Kirby-Bauer) technique was employed as previously described [58]. Mueller-Hinton (MH) agar, which is a standard medium for the disc diffusion method, was used. Antibiotics used in this study included the Quinolones: Ciprofloxacin and Perfloxacin; Aminoglycoside: Netilmicin; Macrolide: Erythromycin; Tetracycline: Doxycycline; the third generation cephalosporin: Ceftriazone and Cefotaxime; Bacitracin, Trimethoprime and Cotrimoxazole. Apart from bacitracin, these are drugs commonly used to treat infections due to enterobacteriaceae [9].They were compared with recommended standards which conform to those of the National Committee for Clinical Laboratory Standards, [10].

STATISTICAL ANALYSIS

Data was analyzed using SPSS. The chi-square (χ^2) test was employed where appropriate for statistical analysis. Differences were considered significant at $P \leq 0.05$ [11].

RESULTS

PREVALENCE OF COLIFORMS IN HIV /AIDS PATIENTS AND CONTROL.

A total of three hundred (300) urine samples were analyzed. Out of these, 240 came from HIV/AIDS patients, attendees of the laboratory who came for check up or patients referred to the laboratory from the wards. The remaining 60 individuals constituted the control group i.e. they were made up of healthy volunteers who came to donate blood; seronegative for both HIV and Hepatitis B surface antigens.

Of the 240 urine samples from HIV positive patients, coliform organisms were isolated from 49, giving an overall prevalence of 20.4%, compared to 2 out of 60 (3.3%) for the control. The difference was statistically significant ($P < 0.05$). Significant bacteriuria was observed in HIV positive patients (Table 1).

{image:1}

Out the 95 male and 145 female samples collected, 18(18.9%) and 31(21.4%) had coliform organisms isolated respectively. The difference was not statistically significant ($P > 0.05$) Out of the 18 male that had coliform, 7 (38.9%) were symptomatic whereas 19(61.3%) of females were symptomatic (Table 2).

{image:2}

Of the 50 hospitalized and 190 nonhospitalised cases sampled, coliform were isolated from 23(46%) and 26(13.7%) patients respectively. The difference was statistically significant ($P < 0.05$) (Table 3).

{image:3}

The prevalence of the organism was highest in the age group 30 – 39 (26.4%) and lowest in the age group ≥ 49(11.1%), but the difference was not statistically significant ($P > 0.05$) (Table 4).

{image:4}

Lactose fermentation, motility (except for Klebsiella) was positive for all isolates. Of the 49 coliform organisms isolated eleven different species were isolated, whereby Escherichia coli 15 (30.6 %) was more frequently encountered with followed by Serratia marcescens 6 (12.24%) and Citrobacter freundi, 6 (12.24%).

{image:5}

Out of the 49 species isolated, 38 (77.5%) were susceptible

to Ceftriazone, a third generation cephalosporin. 36 (73.5%) were sensitive to perfloxacin and 35 (71.4%) were sensitive to netilmicin, an aminoglycoside, but the difference was not statistically significant (P > 0.05).

All species isolated were resistant (100%) to Bacitracin. None of the isolates were completely sensitive to Trimethoprime and co-trimoxazole, however, 11(22.4%) and 7(14.3 %) of the isolates were intermediate to these antibiotics respectively.

Generally, isolates were resistant to at least three antibiotics (Table 5).

{image:6}

Of the 49 isolates 19(38.9%), corresponded to patients with CD 4 count from 1 to 50; 14(28.5%) to those with CD4 count from 51 to 100, 8(16.3%) to those with CD4 count from 101 to 150 and 8(16.3%) to those with count from 151 to 200. Generally, those with very low CD4 counts had significant bacteriuria than those with higher counts (p < 0.05). (Table 6).

DISCUSSION AND CONCLUSION

People with AIDS are predisposed to urinary tract infections by common bacteria and pathogens [12]. These pathogens may affect any urologic organ and treatment therefore should be culture specific and long-term [13]. Voiding dysfunction in patients with AIDS is usually a result of opportunistic infections [1415] that might include coliforms as well. Vulnerability to bacterial infection in the HIV/AIDS patient correlates inversely with the CD4 lymphocyte count. When the count falls to less than 200/mm³, the risk of bacterial and opportunistic infections, including UTI rises dramatically. This may be compounded by neutropenia that accompanies long-term treatment with some antiretroviral drugs. Patients with CD4 counts greater than 200/mm³ were often asymptomatic in this study. The intensive use of quinolones in the treatment of common infections has led to the spread of resistant microorganisms [16].

This study was conducted to determine the prevalence of coliform bacteria and their antibiotic sensitivity in urine of HIV/AIDS patient by employing biochemical profiles for species identification; and comparing this prevalence with non HIV healthy control. The organisms were isolated from urine with an overall prevalence of 20.4% in the HIV/AIDS patients compared to 3.3% in the control group (P < 0.05). This result tie with those previously reported [1718]. They found that bacteriuria was higher in HIV patients especially in those that were symptomatic. In the HIV/AIDS patients, the prevalence of the organisms was higher in females (21.4%) compared to 18.9% in males. However, the difference was not statistically significant (P > 0.05). This could be because of the anatomy of the female genitourinary system. The female urethra, being short and close to the anal opening predisposes her to infections by coliforms which can easily contaminate the urethral opening from one's anus [719].

Generally hospitalized patients (46.0%) were significantly (P < 0.05) prone to coliform infections than nonhospitalised patients (13.7%). This may be indicative of a nosocomial infection risk probably because patients might have acquired infection following hospitalization with factors such as prolonged hospitalization and impaired immunity compound risk of being infected [19].

There was no statistical significant difference between the prevalence of the organisms in the different age groups. (P > 0.05) however, a high occurrence was observed in the age group 30-39 (26.4%). This may be attributed to inadequate health education, poverty and risks associated with the acquisition of nosocomial infections. Most of the hospitalized cases fell in this age group. Reports have shown that inherent resistance (age, nutritional status, severity of underlying disease and breaks in the integrity of the skin or mucous membrane) of patience to infection is an important determinant of acquiring nosocomial infections [192021]. The age group ≥ 49 had the lowest occurrence probably because these individuals are less exposed to sexual hazards, injuries and contamination.

Those with very low CD 4 count had highly significant bacterial counts (P < 0.05). The high prevalence and significant counts of coliform with low CD4 count emphasizes the enumeration of CD4 cell to check disease progression in HIV. Those with very low CD4 counts were more immunocompromised than those with higher counts. This agrees with the previous findings [22].

Escherichia coli were the most encountered (30.6% of isolates) followed by Citrobacter freundi (12.2%) and Serratia marcescens (12.2%). Enterobacter aerogenes (2.0%) was the least common pathogen. This again ties with the results of De Pinho [17]. They as well found E. coli as the most prevalent organism in urine of HIV patients. Coliforms, being nosocomial pathogens with an increasing propensity to acquire resistance to a wide range of antibiotics poses a great

problem to empirical approaches to therapy.

Antibiogram of the organisms revealed good susceptibility with three antibiotics. 77.5% of the isolates were sensitive to the third generation cephalosporin, ceftriazone. This was probably due to the fact that the high cost of the drug limited its use in the locality thus bacteria could not acquire resistance due to abusive use of the antibiotic. Moreover, third generation cephalosporin are highly active against gram negative bacteria. Similarly, high susceptibility was observed for the quinolone, perfloxacin (73.5% of isolates) and the Aminoglycoside, Netilmicin (71.4% of isolates). Fluoroquinolones do have advantages in the treatment of complicated UTI secondary to host factors, resistant organisms, or difficult-to-treat pathogens such as Pseudomonas aeruginosa [23]. Isolates showed complete resistance to Bacitracin. The use of antibiotics in hospital and community at large serves as a major selective pressure for antibiotic resistant bacteria. Multidrug resistant nosocomial infections by these organisms are increasing world wide [24]. Complete resistance to bacitracin could be because the outer membrane of the gram negative cell wall prevented the antibiotic from penetrating to its site of action. Also, bacitracin has toxic effects unrelated to its antimicrobial activity that limits its use as a chemotherapeutic agent [9].

CONCLUSION

Results revealed that Escherichia coli, Serratia marcescens and Citrobacter freundi respectively were the most encountered organisms found in urine of HIV/AIDS patients. The majority of coliform organisms are now resistant to cotrimoxazole.

Antibiotic susceptibility testing revealed that, Ceftriazone, Perfloxacin, and netilmicin could be employed for the treatment of infections caused by coliforms in HIV/AIDS patients. Patients with very low CD4 counts were more immunocompromised hence posses significant bacteriuria. Bacteriuria in HIV/AIDS patients represents a relevant cause of morbidity.

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