IJAMBR 7 (2019) 24-29 ISSN 2053-1818

International Journal of Applied Microbiology and Biotechnology Research
www.bluepenjournals.org/ijambr

Antibiogram profiling and cost effectiveness of urine microscopy, culture and sensitivity in Yenegoa, Nigeria

doi.org/10.33500/ijambr.2019.07.004

Arikekpar Ibemologi* and Alade Tolulupe

Department of Medical Laboratory Science, Faculty of Basic Medical Sciences, College of Health Sciences, Niger Delta University, Wilberforce Island, Amassoma, P. M. B. 071, Bayelsa State, Nigeria.

Article History
Received 26 October, 2018
Received in revised form 16 January, 2019
Accepted 21 January, 2019

Keywords:
Urinary tract infection, Uropathogen, Cost-effectiveness, Antibiotics susceptibility.

Article Type:
Full Length Research

ABSTRACT

The study was carried out in the Medical Laboratory of the Federal Medical Centre, Yenegoa, Bayelsa State between January - September 2010 with the aim to ascertain the antibiogram profile, prevalence rate of bacterial urinary tract infection in Bayelsa State and the cost effectiveness of such urine diagnosis. A total of 610 urine samples collected from adult patients were processed and microbial isolates identified using standard microbiological techniques. Results obtained show that 467(211 males and 256 females) had significant bacterial growth of 76.6% and are statistically significant ($p<0.05$). A total of 528 strains of some common bacterial uropathogens were isolated, 238 from males (45.1%) while females had 290 strains (59.9%). *Escherichia coli* was the predominant uropathogen (41.9%) followed by *Staphylococcus aureus* (25.6%), *Coagulase negative Staphylococcus* (13.8%), *Klebsiella pneumoniae* (9.4%), *Proteus mirabilis* (5.4%) and *Pseudomonas aeruginosa* (3.9%). The percentage susceptibility of all isolates to the antimicrobial agents were 66.7% for ciprofloxacin, 51% for nitrofurantoin, 43.2% for erythromycin, 40.3% for colistin, 39.2% for cotrimoxazole, 37% for nalixidic acid, 30% for gentamycin, 28% for streptomycin, 21% for tetracycline and 14.6% for amoxicillin. Comparing the 76.6% prevalence rate in this study to that obtained from the Bayelsa State Ministry of Health in 2008 and 2009 which was 67.5 and 75.2%, respectively indicates a rising trend in urinary tract infection (UTIs). The average cost of urine diagnosis was 700 naira/patient and the estimated cost for these 610 patients was 427,000 naira, which is more cost effective than empirical treatment because it could prevent waste of more resources due to revisits, re-evaluation and re-treatment of patients. Thus antibiotic susceptibility profiling is best based on the antibiogram of individual patient.

INTRODUCTION

Urinary tract infection (UTI) is one of the most common infections (Dilnawaz and Safia, 2005), with about 150 million people diagnosed each year worldwide (Gupta, 2001). The urinary tract is free of microbes above the entrance to the bladder, but the lower urethra hosts a number of genera of bacteria like *Lactobacillus, Staphylococcus* (coagulase-negative), *Corynebacterium, Haemophilus, Streptococcus* and *Bacteriodes* (Eugene and Denise, 2004). A current idea on antimicrobial susceptibility pattern is necessary for appropriate therapy of UTIs (Mohammed and Mohammed, 2007). UTIs are important complications of pregnancy, diabetes mellitus,

*Corresponding author. E-mail: ariko20002000@yahoo.com.

polycystic kidney disease, renal transplantation, and structural and neurological conditions and establish bacteriuria often at levels more than or equal to 10^5 colony forming units of bacteria/ml of urine. Although fungi and viruses are occasional etiological agents, UTIs are predominantly caused by bacteria (Jones et al., 2006). *Escherichia coli* dominates as the causative agent in most patient populations; Others would include *Staphylococcus* sp., *Neisseria gonorrhoea*, Enterobacteriaceae group, *Klebsiella* sp., *Proteus* spp., *Enterobacter* sp., and *Pseudomonas* sp., *Chlamydia trachomatis*, *Candida* spp., *Mycoplasma* (Ebie et al., 2001).

Most infections are caused by reversed ascent of bacteria from the faecal flora especially in the females who have a shorter and wider urethra that is close to the anus (Jones et al., 2006). Antimicrobial susceptibility testing of significant bacterial isolates to detect possible drug resistance in common pathogens and to assure susceptibility to drugs of choice for particular infections is paramount (James and Mary Jane, 2009).

The high consumption of inappropriately prescribed antibiotics combined with multiple pathology and frequent use of invasive devices is a major factor contributing to high levels of resistance (Monique, 2000).

There is an emergence of antibiotic resistance of UTIs in the developing world (El-Astal, 2005) and increasing resistance have noted against amoxicillin, cotrimoxazole, and lately, fluoroquinolones (Mohammed and Mohammed, 2007). It is important to consider a range of costs in treating UTIs and the economic analyses relevant to antibiotic therapy of UTIs include cost-benefit, cost-effectiveness, and cost minimization. Cost-benefit analysis involves quantifying the monetary costs of a treatment option. Cost-effectiveness involves benefits that are measured in events, such as physician visits or episodes of pyelonephritis. The most important predictor of high cost-effectiveness is high efficacy against the urinary pathogen. The lower the effectiveness of antibiotics against this pathogen, the greater the number of revisits and cases of progression to pyelonephritis. While, cost minimization analysis finds the lowest cost option among those found to be of equal benefit (Rosenberg, 1999).

Measurement of costs is the quantification of the resources used in providing the particular service; and the cost of using a particular service or treatment is not necessarily the price that is paid for that resource but the benefit foregone by not choosing the alternative (Ceri, 2005).

Hence, this research intends to determine the prevalence rate of bacterial UTIs, the most common bacterial etiologic agents of UTI in adults, the antimicrobial susceptibility to some commonly used antibiotics and the cost effectiveness of urine diagnosis of UTIs in Bayelsa State.

MATERIALS AND METHODS

Study location/design

In a prospective study from January-September 2010, a total of 610 suspected adult patients with UTI recommended for urine microscopy, culture and sensitivity (MCS) test at the Federal Medical Centre, Yenegoa, Bayelsa State, Nigeria were chosen for the study. Prevalence rates of UTIs in Bayelsa State for 2008 and 2009 were also collected from the State Ministry of Health through the Health Systems Development Statistics Unit.

Specimen collection

Freshly voided urine specimens were collected aseptically from patients with symptoms suggestive of UTIs into 20 ml sterile universal containers (Cheesborough, 2006).

Sample analysis

A loopfull (0.01 mL) of urine sample was aseptically cultured on blood agar and cystine lactose electrolyte deficient (CLED) using standard procedures and incubated aerobically at 37°C for 24 h; cultures are then observed for growth via formation of colonies. Bacteriuria was defined as culture of a single bacterial species from the urine sample at a concentration of $>10^5$ cfu/mL (Cheesborough, 2006).

Isolate identification

Identification of suspected isolates was carried out by Gram staining, biochemical tests and sugar fermentation test following standard procedures as described by Cheeseborough (2006).

Biochemical tests

All isolates were subjected to coagulase, indole, urease, citrate, oxidase and sugar fermentation tests using standard microbiological procedures. Any clumping within 10 s on a glass slide was reported as coagulase positive. Indole production was indicated by red-pink ring at broth surface within 10 min in a test tube. After incubation at 35°C for 48 h, citrate utilization was indicated by a bright blue color. After 12 h incubation at 37°C, urease production was indicated by a change in color to pink. Oxidase production was indicated by a deep purple color. Sugar fermentation was indicated after 24 h incubation at

Table 1. Prevalence of isolates according to gender.

Bacterial species	Males N (%)	Females N (%)	Total N (%)
E. coli	92(17.4)	129(24.4)	221(41.9)
S. aureus	62(11.7)	73(13.8)	135(25.6)
CON	32(6.1)	41(7.8)	73 (13.8)
Proteus	12(2.3)	17(3.2)	29 (5.4)
Klebsiella	32(6.1)	18(3.4)	50(9.5)
Pseudomonas	8(1.5)	12(2.3)	20(3.8)
Total	238(45.1)	290(54.9)	528(100)

CON, Coagulase negative *Staphylococcus*; **N,** number of isolates; **%,** isolates in percentages.

37°C, with acid production indicated by change in colour from red to yellow, and gas production was shown by a bubble trapped in the Durham's tube (Cheesborough, 2006).

Antibiotic susceptibility testing

The antibiotic susceptibility patterns of all isolates to 11 antibiotics were determined by the modified Kirby-Bauer diffusion technique (Cheeseborough, 2006). The concentration of the standard antibiotics discs used were amoxicillin 25 µg, colistin 25 µg, chloraphenicol 30 µg, ciprofloxacin 5 µg, erythromycin 5 µg, gentamycin 10 µg, Nalaxidic acid 30 µg, nitrofurantoin 200 µg, streptomycin 25 µg and tetramycin 10 µg. The Mueller Hinton's agar was prepared according to manufacturer's instructions and standard protocol for modified Kirby-Bauer diffusion technique was followed. The plates were then incubated at 37°C for 18 h. The diameter of the zone of inhibition produced by each antibiotic disc was measured, recorded and the isolates were classified as resistant or sensitive (Adedeji and Abdulkadir, 2009).

Costs

Costs were estimated from prices obtained from computer data base of hospital registry and laboratory department which included cost for card/registration and cost for urine MCS (Pateld et al., 2009). The average cost of MCS was determined in Nigerian local currency and estimated its equivalent in international dollars using the standardized conversion table. The average cost effectiveness ratio is defined as the total cost of all MCS studies divided by the number of MCS.

Statistical analysis of results

Data was analyzed using Graph Pad Prism 5.01. A p-value of less than or equal to 0.05 was considered to be statistically significant while p-value more than 0.05 was considered to be statistically not significant (NS). A 2×2 contingency table using Fisher's exact test was used to analyzed gram positive and negative bacterial isolates in both sexes and was found to be statistically significant with a relative risk of 1.003 and odds ratio of 1.004 (Table 1). The correlation analysis on the sensitivities and resistances of both Gram positive and negative bacterial isolates were all statistically significant. The r values ranged between 0 and 1, which implies that the sensitivities and resistances tend to increase and decrease together.

RESULTS AND DISCUSSION

A total of 610 urine samples were collected from patients and 467 (211 males and 256 females) showed a 76.6% significant bacterial growth. A total of 528 species of various bacteria were isolated consisting of 238 (45.1%) from males and 290 (54.9%) from females.

Prevalence rates of bacterial uropathogens collected from the Bayelsa State ministry of Health for 2008 and 2009 were found to be 67.5 and 75.2%, respectively. Prevalence rate of bacterial uropathogens in FMC in 2010 was 76.6% (Table 2). The antibiogram profile of bacterial isolates and their general susceptibility to antibiotics are depicted in Table 3. Isolates exhibited more susceptibility to ciprofloxacin (66.7%).

The average cost of one urine MCS was 4.52 dollars to a patient. Estimated cost of MCS for 610 consenting patients in this study was 2,757.2 dollars (Table 4). Females have shown in several studies to have higher occurrence of UTIs (El-Mahmood, 2009), and a study by Adedeji and Abdulkadir (2009) revealed that 61.9% of the infected population was of females. This was confirmed in this study because most of the isolates were from females (54.9%). This result corresponds to a similar study done in Yola from 3 tertiary institutions with 61.9% of the infected population being females (Adedeji and

Table 2. Prevalence rates of bacterial uropathogens from 2008 to 2009 and 2010 in FMC, Bayelsa State.

Hospital/Year	Urine cultures	Males	Females	Positive cultures	Prevalence (%)
HMB/2008	385	112	148	260	67.5
HMB/2009	331	85	164	249	75.2
FMC/2010	610	211	256	467	76.6

HMB, Hospitals Management Board; **FMC,** Federal Medical Centre.

Table 3. Antibiogram profile and general susceptibility of bacterial isolates to antimicrobials.

Antibiotics n (%)	EC (221)		STA (135)		CON (73)		KB (50)		PR (29)		PS (20)		Total
AMX	24(11)	197(89)	23(17)	112(83)	26(36)	47(64)	4(7)	47(93)	0(0)	29(100)	0(0)	20(100)	77(14.6)
COL	69(31)	152(69)	70(52)	65(48)	38(52)	35(48)	25(50)	25(50)	9(31)	20(69)	2(10)	18(90)	213(40.3)
COT	62(28)	159(72)	68(50)	67(50)	38(52)	35(48)	26(52)	24(48)	12(41)	17(59)	1(5)	19(95)	207(39.2)
CHL	100(45)	121(55)	64(47)	71(53)	24(33)	49(67)	27(54)	23(46)	17(59)	12(41)	2(10)	18(90)	234(44.3)
CPF	149(67)	72(33)	90(67)	45(33)	53(73)	20(27)	33(66)	17(34)	21(72)	8(28)	6(30)	14(70)	352(66.7)
ERY	92(42)	129(58)	47(35)	88(65)	46(63)	27(37)	20(40)	30(60)	17(59)	12(41)	6(30)	14(70)	228(43.2)
GEN	45(20)	176(80)	39(29)	96(71)	40(55)	33(45)	22(44)	28(56)	12(41)	17(58)	0(0)	20(100)	158(30)
NAL	82(37)	139(63)	37(27)	98(73)	44(60)	29(40)	15(30)	35(70)	16(55)	13(45)	2(10)	18(90)	196(37)
NIT	93(42)	128(58)	73(54)	62(46)	55(75)	18(25)	27(54)	23(46)	15(52)	14(48)	7(35)	13(65)	270(51)
STR	56(25)	165(75)	35(26)	100(74)	31(42)	42(58)	14(28)	36(72)	10(34)	19(66)	0(0)	20(100)	146(28)
TET	28(13)	193(87)	36(27)	99(73)	25(34)	48(66)	15(30)	35(70)	7(24)	22(76)	0(0)	20(100)	111(21)

AMX, Amoxycillin; **COL,** colistin; **COT,** cotrimoxazole; **CHL,** chloramphenicol; **ERY,** erythromycin; **GEN,** gentamycin; **NAL,** nalixidic acid; **NIT,** nitrofurantoin; **STR,** streptomycin; **TET,** tetracycline; **CPF,** ciprofloxacin; **S,** sensitive isolates; **R,** resistant isolates; **n,** number of isolates; **%,** percentage number of isolates; **EC,** *E. coli*; **STA,** *S. aureus*; **CON**, *coagulase negative Staphylococcus*; **KB,** *Klebsiella* spp.; **PR,** *Proteus* spp.; **PS,** *Pseudomonas* spp.; ***T,*** total number of susceptible isolates to antibiotics.

Table 4. Average cost of urine diagnosis per patient.

S/N	Cost item	Cost (Naira)	Cost (dollars)	Percentage of total cost
1	Hospital card	200	1.29	28.6
2	Cost of MCS	500	3.23	71.4
3	Average total cost per MCS	700	4.52	100

MCS, Microscopy, culture and sensitivity; 610 patients × \$4.52 = \$2,757.2.

Abdulkadir, 2009). This study reveals a rising trend in the prevalence of UTI in Bayelsa State evident in the prevalence rate of 76.6% in 2010 which is slightly higher than 67.5 and 75.2% prevalence rates recorded in 2008 and 2009 respectively by the Bayelsa State Hospital Management Board. The findings of this study accord with other works like Kolawale et al. (2009) (60%), Akortha and Ibadin (2008) (63.8%), El-Mahmood (2009) (67.2%) and Mbata (2007) (77.9%). However, the findings varies with those of Nwadioha (2010) (26%), Ebie et al. (2001) (35.5%) and Bankole (2011) (39.69%). This rising trend may be due to the huge amount of oil locations around communities and the workers activities with sex workers, frequent night Burial ceremonies and the aftermath of political meetings in hotels and guest houses. Again, high military presence due to youth restiveness and hardship may force some female undergraduates encourage prostitution. Other additional causes of antibiotics (fake and sub-standard drugs) and standard of medical intervention (quackery), self medication, non-compliance and non-adherence to drug prescriptions, re-infection from sex partners who were not initially treated alongside or from other multiple partners, reluctance to spend money for medical assistance and rather rely on herbs and also not being comfortable with using condoms or using initially and stopping later due to more familiarity with partner. The results of this study revealed that *E. coli* was the most predominant isolate (41.9%) which agrees with previous reports in Benin (Oladeinde et al., 2011), but differs from that of Akortha and Ibadin (2008) that incriminated *S. aureus* as the most common organism isolated. In this study, *S. aureus* was the next predominant isolate which was slightly similar to reports of Annabelle and Jennifer (1999) and Manikandan et al. (2011) but this differs from reports of El-Mahmood (2009) and Nwadioha (2010). Yearly, an estimated 150 million UTI cases have been documented with a cost burden in excess of 6 billion dollars worldwide. It is therefore important that costing is carefully considered when treating UTIs. Much research has not been reported about cost effectiveness of urine diagnosis but one study in north western Nigeria estimated that the average cost of one diagnosis was 4 dollars to the patient and suggested that empirical treatment based on community profiling of bacteria sensitivity to antibiotics may be superior to patient based MCS studies (Shaibu et al., 2008). Irene et al. (2010) also reported that the systematic screening of all women for asymptomatic *C. trachomatis* infection is not cost effective, because the cost exceeds the benefits. However, keeping the emerging antimicrobial resistance in mind, it is strongly suggested that the antibiotic therapy should only be commenced after the sensitivity report from the Microbiology laboratory (Rajesh et al., 2011). This would not only help in the prudent use of antibiotics but also would curb the dissemination of antimicrobial resistant strains in the community as well as in the hospital.

The results of this study clearly showed that the average cost of one urine MCS was 4.52 dollars to a patient and the estimated cost of MCS for 610 consenting patients was 2,757.2 dollars. In disagreement to the opinions of Shaibu et al. (2008) and Irene et al. (2010), urine MCS is more cost effective in the diagnosis and treatment of UTI because it could avert issues like symptoms reoccurrence, revisits, re-evaluation and re-treatment of patients or progression to pyelonephritis which definitely will give room to additional monetary and manpower expenses. Although the infectious disease society of America guidelines state that uncomplicated UTI should be treated empirically with trimethoprim sulfamethoxazole unless the community resistance among uropathogens exceeds 10-20% in which case a fluoroquinolone should be used (Thuan and Loren, 2001).

Conclusion

Here in Bayelsa State, such guidelines will be difficult to follow because such studies about community resistance among uropathogens are yet to be embarked upon and reported to the best of our knowledge. Thus, it would be better if antibiotics susceptibility profiling is based on the antibiogram pattern of individual patients.

Compliance with ethics and guidelines

Approval was sought and obtained from the Ethical Committee of the Hospital.

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