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ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF URINARY PATHOGENS ISOLATED FROM TWO TERTIARY HOSPITALS IN SOUTHWESTERN NIGERIA

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ABSTRACT

Background: Urinary tract infection (UTI) is among the most common reasons for patients to seek health assistance that is commonly encountered in office practices. This is also a leading cause of Gram negative sepsis in hospitalized patients.

Objectives: This study was carried out in order to isolate, characterize and identify the pathogens associated with UTI in two teaching hospitals at Osun state, Nigeria and to determine their antimicrobial susceptibility patterns.

Methods: This was a prospective observational study involving standard microbiological procedures for analysing urine samples of inpatient and outpatient cases of UTI. Identification of these pathogens was performed using Microbact/API identification system.

Results: Out of the 300 urine samples cultured, 88 (29.3%) yielded significant growth of urinary pathogens while 212 (70.7%) yielded either insignificant growth or no growth of any urinary pathogen. *Escherichia coli* 19 (21.6%) were the commonest pathogen isolated followed by *Klebsiella pneumonia* 14 (15.9%), *Staphylococcus aureus* 12(13.6%), *Candida albicans* 12(13.6%), *Pseudomonas aerugenosa* 9 (10.2%), *Klebsiella oxytoca* 8 (9.1%), *Staphylococcus saprophyticus* 6 (6.8%), *Serratia rubidaea* 3 (3.4%), *Enterobacter agglomerans* 2 (2.3%), *Acinetobacter iwoffii* 1 (1.1%), *Acinetobacter baumannii* 1 (1.1%), *Providencia retgerri* 1 (1.1%). The susceptibility of Gram negative bacteria (GNB) were mainly toward parenteral antibiotic rather than oral one, while most of the common antibiotic showed a resistant pattern. UTI was more prevalent among patients within hospital setting 71(80.7%) than out-patients 17(19.3%).

Conclusion: This study justifies the necessity to treat patients with UTI based on antimicrobial susceptibility test result in order to prevent evolution of resistant pathogens. Since UTI has large impact on the socio-economy and emergence of bacterial resistance, periodic surveillance of antibiotic susceptibility is strongly recommended.

Keywords: Microbact; antimicrobial resistance; UTI and Osobgo.

SENSIBILITE ANTIMICROBIENNE DE PATHOGÈNES URINAIRES ISOLES AU NIVEAU DE DEUX HOPITAUX TERTIAIRES AU SUD-OUEST DU NIGERIA

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RÉSUMÉ

Contexte: L'infection des voies urinaires (IVU) est parmi les raisons les plus courantes pour les patients à demander de l'assistance médicale qui est couramment rencontré dans les clientèles privés. C'est aussi la principale cause de septicémie de bactéries à Gram négatif chez les patients hospitalisés.

Objectifs: Cette étude a été réalisée afin d'isoler, caractériser et identifier les agents pathogènes associés aux infections urinaires dans les deux centres hospitaliers universitaires de l'Etat d'Osun au Nigéria et pour déterminer leurs profils de sensibilité aux antimicrobiens.
Méthodes: C'est une étude prospective observationnelle impliquant des procédures microbiologiques standard pour analyser des échantillons d'urine de patients hospitalisés et ambulatoires de cas d'infection des voies urinaires. L'identification de ces agents pathogènes a été réalisée en utilisant le système d'identification Microbact /API.
Résultats: Sur les 300 échantillons de culture urinaire, 88 (29,3%) ont eu une croissance importante de pathogènes urinaires tandis que 212 (70,7%) ont eu une croissance soit insignifiante ou pas de croissance d'aucun agent pathogène urinaire. *Escherichia coli* 19 (21,6%) était le germe pathogène le plus fréquent isolé suivi de *Klebsiella pneumonie* 14 (15,9%), *Staphylococcus aureus* 12 (13,6%), *Candida albicans* 12 (13,6%), *Pseudomonas aerugenosa* 9 (10,2%), *Klebsiella oxytoca* 8 (9,1%), *Staphylococcus saprophyticus* 6 (6.8%), *Serratia rubidaea* 3 (3,4%), *Enterobacter agglomerans* 2 (2,3%), *Acinetobacter iwoffii* 1 (1,1%), *Acinetobacter baumannii* 1 (1,1%), *Providencia retgerri 1* (1,1%). La sensibilité de bactéries à Gram négatif (BGN) était essentiellement à un antibiotique parentéral plutôt que par voie orale, alors que la plupart d'antibiotiques fréquents a montré une résistance. L'infection de voies urinaires était plus fréquente chez les patients hospitalisés 71 (80,7%) par rapport aux patients externes 17 (19,3%).
Conclusion: Cette étude justifie la nécessité de traiter les patients souffrant d'infection urinaire en se basant sur le résultat du test de sensibilité aux antimicrobiens afin de prévenir l'évolution de pathogènes résistants. Depuis, l'infection de voies urinaires a un impact important sur le développement socio-économique et l'émergence de la résistance bactérienne; la surveillance périodique de la sensibilité aux antibiotiques est fortement recommandée.

Mots-clés: Microbact; la résistance aux antimicrobiens; Infection des voies urinaires et Osobgo.

INTRODUCTION

Urinary tract infections (UTI) are among the most common conditions encountered in office practices. This is also leading cause of Gram negative sepsis in hospitalized patients [1]. UTI is the most common reason for patients to seek health assistance, accounting for about seven million patients visit every year. Majority are otherwise healthy women who typically present with dysuria each year. Approximately 20% of women develop UTI sometime during their life time. Above Age 50, the incidence of UTI is similar in men and women [2].

UTI encompasses both asymptomatic microbial colonization of the urine and symptomatic infection with microbial invasion and inflammation of urinary tract structures. Microbiologically, growth of more than 10^5 colony forming unit (cfu) /mL from a properly collected midstream urine samples indicates infection. However, a smaller number of bacteria (10^2 -10^4/mL) may signify infection in specimen from suprapubic aspiration, catheter samples and immunocompromized patients. The etiology of UTI and the antimicrobial susceptibility of urinary pathogens in both the community and hospitals have been changing, and in recent years antibiotic resistance has become a major problem worldwide due to several factors related to the genetic nature of the organisms and selective antimicrobial pressure in humans and animals.

Drug resistance is a large and growing problem in infection that accounts for most of Africa's disease burden e.g. malaria, TB, HIV, respiratory and diarrheal diseases. Currently, the prevalent pathogens of UTI have been resistant to most chemotherapeutic agents making antimicrobial susceptibility highly unpredictable without laboratory support. These would have profound impact on future management of infection with these drugs [3,4].

Furthermore, prevalence of the urinary pathogens and their susceptibility reactions to antibiotics differ from places to places with time. It is essential to know the current trends of UTI in these two places to ease diagnosis and thus establish the suitable antimicrobial agents for such infections in order to facilitate quick recovery, prevent/minimise complications of antimicrobial resistance.

Variability of Pathogens in UTI and Antimicrobial Susceptibility in Nigeria and Neighbouring countries

The prevalence of antimicrobial resistance in urinary pathogens is increasing worldwide. along with temporal and local variables affecting the pattern of pathogens. An accurate bacteriological records of UTI from studies locally and regionally, may provide guidance for empiric therapy with antimicrobial agents in UTI before the culture and sensitivity patterns are available [5,6,7].

In our regional context, variable results on the prevalence of pathogens and their antimicrobial susceptibility pattern in UTI were obtained in recent and past studies: at Abeokuta, Nigeria, Ojo *et al.*, 2007 reported that the overall prevalence of UTI was 52% with *E.coli* (28%), *Klebsiella pneumoniae* (16%) and *Pseudomonas aeruuginosa* (8%). The organisms were susceptible to ofloxacin, nitrofurantoin, ciprofloxacin and gentamicin (P= 0.0001) but resistant to ampicilin (P = 0.153) [7].

Similarly, Okesola and Oni, 2005 (6) at the University College Hospital (UCH), Ibadan, Nigeria, showed that the most prevalent bacterial pathogen in urine were *Staphylococcus aureus* (47.5%), *Pseudomonas aeruginosa* (24.6%), *Klebsiella species* (23%), *Proteus spp* (3.3%), *E. coli (*1.6%). Older generation antibiotics like streptomycin and chloramphenicol were more resistant then the newer gentamycin and ciprofloxacin [8].

Jumbo *et al*., 2011 from Calabar, Nigeria said the incidence of UTI was 7.7% (Male 46.7% and Female 53.3%). 69.2% were of community acquired while 30.8% were nosocomial in origin [9].

Obiogbolu *et al.*, 2009 from Akwa metropolis, South-eastern Nigeria also reported that *E.coli* was the commonest urinary tract pathogen and the study indicated a high incidence of UTIs (54%) in pregnant women [10].

At Imo State University Teaching Hospital, Orlu, Nigeria, it was also showed that *E. coli* accounted for 52.5% infection; *S. aureus* (33.9%), *P. mirabili*s (8.5%), *Enterococcus species* (5.0%) and *N. gonorrhea* (1.7%). These organisms were most sensitive to the quinolones compared to the penicillins and aminoglycosides [11].

Similar results were obtained at Port Harcourt, where most of the pathogens were resistant to tetracycline, ampicillin and cotrimazole but exhibited sensitivity to nitrofurantoin, gentamicin and nalidixic acid [11].

Kenechukwu *et al*., 2006 at Enugu, Nigeria, reported the commonest isolates to be *E.coli*, *Staphylococcus aureus, Streptococcus faecalis* and *Proteus spp*. The Gram positive organisms were very sensitive to Augmentin and the fluoroquinolones. *Escherichia coli* showed the highest sensitivity to nitrofurantoin (76%) and the fluoroquinolones (74%). The study clearly showed that nitrofurantoin is a very effective first line drug for UTIs [11].
From Makurdi, North central Nigeria, Umeh *et al*., 2007 reported that bacteriuria was approximately 5 times high in women as in men. The attributed 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 a higher resistance pattern [12].

In similar studies carried out in Kano Nigeria, Adeleke and Asani, 2009 reported that UTI was caused predominantly by *Staphylococcus aureus* (67.9%), *Klebsiella species* (17.9%), and *Pseudomonas* (14.2%). There was high in-vitro resistance of these organisms to nalidixic acid and ampicillin but sensitive to cefotaxime, ceftriazone and ciprofloxacin [13].

At Yola, Adamawa, Nigeria, it was reported that 37.1% of uropathogens were obtained from male suspects, while the remaining 62.9% were from female subjects. Gram negative bacteria had the highest frequency of occurrence with 63.6% than Gram positive with 36.4% with *E.coli* as the commonest [14].

Zaria *et al.* also reported from Maiduguri, Nigeria , a high incidence of UTI both in pregnant and non-pregnant women, *E.coli* being the prominent organism, and highly sensitive to quinolone group while resistant to Co-amoxicillin, Cotrimoxazole and Nalidixic acid [15]. Similar results were observed in Kaduna, Nigeria [16].
Apart from Nigeria, in Koforidua, Ghana, the most common urinary pathogens isolated were *E.coli* (58.6%) and *Klebsiella spp. (*20.6%). The majority of isolates (78%) were from females. The highest percentage of isolates (7.9%) was in age group 21-30 yrs. All pathogens were resistant to Ampicillin (95.8%) and cotrimoxazole (97.7%) but were sensitive to Nitrofurantoin (71.2%) and Gentamicin (51.75%) [17].

In Karachi, Pakistan, prevalence of asymptomatic bacteriuria (ASB) was 6.2%. *E.coli* and *Staphylococcus saprophyticus* showed 66.67% resistance to ampicillins and sulphonamides, while *Enterobacters* showed 100% resistance to ampicillins, cephalosporins and nitrofurantoin [18]. In Tehran, Iran, the most prevalent urinary pathogen was *E.coli* (56.6%) [19].
At Al-Khobar, Saudi Arabia, Bukharie and Saeed, 2001 reported that *E.coli* and *Klebsiella spp.*were also the most common pathogens of urine, accounting for 79% of the Nalidixic axid and Nitrofurantoin were least resistant (13%) while cotrimoxazole was most (39%) with Augmantin 19%. The best susceptibility pattern was seen with quinolones [20].

MATERIALS AND METHODS

This prospective observational study was carried out at Ladoke Akintola University Teaching Hospital (LTH), Osogbo and Obafemi Awolowo University Teaching Hospital Complex (OAUTHC) Ile Ife. The study subjects were from both out-patients clinics and in-patient general wards of these two hospitals.

Study Urine Collection

Each subject was given a sterile plastic bottle to collect mid-stream urine (MSU) after they were taught how

to collect it aseptically. 300 sterile urine samples were thus collected for subsequent bacteriological analysis [21].

Urinalysis
5mls each of the urine samples was centrifuged at 2,500g for 5 minutes. The supernatant was discarded and the deposit re-suspended with the small amount of urine left in the tube by tapping the base of the centrifuge tube. A drop was placed on cleaned grease free slide, covered with cover slip and examined under microscope using 10x and 40x objective lenses for the presence of white blood cells, red blood cells, epithelia cells, casts, crystals, bacteria, yeast cells and Trichomonas [21].

Culture and Sensitivity
A calibrated standard wire loop was used for inoculating culture plate. A loopful (0.002mL) of well mixed un-centrifuged urine was plated on a dried CLED and MacConkey agar media. These plates were incubated aerobically at 37^0C for 24hours. Colony counts were determined at the end of incubation period. Each urine sample with over 10^5 CFU per milliliter was followed up as significant bacteria growth and isolated colonies were sub-cultured.

Subculture of isolated colony
To obtain pure isolates, discrete colonies of pathogens isolated were inoculated in a well dried MacConkey agar media. The plates were incubated aerobically at 37^0C for 24hours. After overnight incubation, colonies were subjected to biochemical tests for identification [21].

Characterization of Isolates
The various isolates obtained were subjected to morphological, physiological, and biochemical tests.
Morphological characterization
The isolated colonies were examined and recorded based on the type of growth, elevation, size, colour, margin, edge, consistency, opacity, and change in medium [21].

Gram staining
A thin smear of each isolate was made on clean grease free glass slide, air dried and heat fixed by passing it gently over flame and then Gram stained. Gram positive cells stained purple while the Gram negative cells stained pink [21].
Biochemical tests
The *Candida albicans* were identified by performing Germ Tube Test (GTT) on any isolate whose Gram result shows yeasts. Catalase test was done on the Gram positive cocci to differentiate *Staphylococcus spp* from *Streptococcus spp.* Coagulase test was done to identify *S. aureus* which produces the enzyme coagulase.

Oxidase test was done on the Gram negative bacilli (GNB) to identify *Psuedomonas spp* from other Gram Negative bacilli. MICROBACT (API) identification system was used to identify the species of the oxidase negative GNB.

Microbat identification
MICROBACT™ GRAM-NEGATIVE 12A was used. It is a range of simple, standardised systems for the rapid identification of Gram-negative bacteria.

Principle
Each kit contains 12 miniature biochemical tests. Organism identification is based on pH change and substrate utilisation. Microbact™ Gram-negative 12A (microplate format) was used alone for the identification of oxidase-negative, nitrate-positive glucose fermenters (comprising 15 genera) and is useful for screening pathogenic Enterobacteriaceae from enteric and urine specimens.

Antibiotic Susceptibility Test
Antibiotic susceptibility of pure culture of confirmed isolate was performed on diagnostic sensitivity test agar (Mueller Hinton agar) by the Kirby Bauer disc diffusion method, using the appropriate Gram positive and Gram negative discs. Isolates were considered sensitive after incubation for 24 hours at 37^0C by measuring zone of inhibition with meter rule which was then compared with zone diameter interpretative to National committee for clinical Laboratory standard (CLSI chart) for different organisms and different antibiotics [22].

To guarantee precision and reliability of antibiogram data, quality control strains of *Staplylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, and *Pseudomonas aeruginosa* ATCC 27853 supplied by department of Pharmaceutics, Obafem Awolowo University, Ile-Ife, Nigeria were used.

Statistical Analysis
Data were analyzed using SPSS version 18.0 window based program. Discrete variables were expressed as percentages and proportions were compared using the Chi-square test. Statistical significant difference was considered at value of $P \leq 0.05$.

RESULTS

Table 1 shows the prevalence of UTI organisms in the two major tertiary institutions in Osun State. Of the 300 subjects, 100 (33.3%) were from LTH and 200 (66.7%) from OAUTHC. 88 (29.3%) of the 300 samples yielded significant growth of organisms; giving UTI a prevalence of 29.3% in Osun State.

The male and female ratio were comparable, however, there was a propensity of higher incidence of UTI in female (Table 2 and Table 3).

From the 88 samples that yielded significant growth, 19 (21.6%) were *Escherichia coli,* 14 (15.9%) *Klebsiella pneumonia,* 12 (13.6%) *Staphylococcus aureus,* 12(13.6%) *Candida albicans,* 9 (10.2%) *Pseudomonas aeruginosa,* 8 (9.1%) *Klebsiella oxytoca,* 6 (6.8%) *Staphylococcus saprophyticus,* 3 (3.4%) *Serratia rubidaea,* 2 (2.3%) *Enterobacter agglomerans,* 1 (1.1%) *Acinetobacter iwoffii,* 1 (1.1%) *Acinetobacter baumannii* and 1 (1.1%) *Providencia retgerri*. (Fig. 1 and Fig. 2). The infection was found to be more common among the in patients than the out patients (Fig. 3).

Contrary to the expectation, UTI was more common in non-pregnant women than the pregnant patients (66.7% vs 33.3%, Table 4).

Table 3 shows organisms in female infections with *K. pneumoniae* 10 (21%) out of of the 48 isolates from female patients. Making *K. pneumoniae* highest UTI organisms in females followed by *C.albicans* 9 (19%), *E.coli* 8 (17%), *K.oxytoca* 6 (13%), *S.*aureus 6 (13%), *S.saprophyticus* 5 (10%), *P. aeruginosa* 2 (4%), *S.rubidaea* 1 (2%) and *E.agglomerans* 1 (2%).

TABLE 1: PREVALENCE OF UTI IN OSUN STATE AT LARGE

Hospital	Growth (H%) (O%)	No Growth (H%) (O%)	Total (H%) (O%)
LTH	40 (40) (13.3)	60 (60) (20)	100 (100) (33.3)
OAUTHC	48 (24) (16)	152 (76) (50.7)	200 (100) (66.7)
Total (Osun)	88 (29.3)	212 (70.7)	300 (100)

H = Hospital O = Overall and Total (Osun) = H (LTH) + H (OAUTHC). Ladoke Akintola University Teaching Hospital (LTH), Osogbo and Obafemi Awolowo University Teaching Hospital Complex (OAUTHC)

TABLE 2: AGE GROUP AND SEX DISTRIBUTION OF SUBJECTS ATTENDING TERTIARY HEALTH INSTITUTIONS IN OSUN STATE

Age Group	Gender		Total (%)
	Male (%)	Female (%)	
0 – 9	16 (5.3)	10 (3.3)	26 (8.7)
10-19	7 (2.3)	8 (2.7)	15 (5.0)
20-29	44 (14.7)	46 (15.3)	90 (30.0)
30-39	31 (10.3)	53 (17.7)	84 (28.0)
40-49	9 (3.0)	4 (1.3)	13 (4.3)
50 & above	53 (17.7)	19 (6.3)	72 (24)
Total	160 (53.3)	140 (46.7)	300 (100)

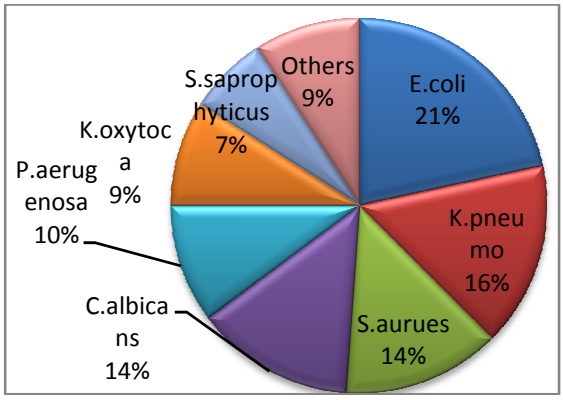


FIG. 1: MICROBIAL ISOLATES FROM SAMPLES

TABLE 3: MICROBIAL ISOLATES FROM URINE OF SUBJECTS ATTENDING BOTH HOSPITALS

Isolate	Male (%)	Female (%)	Total (%)
Escherichia coli	11 (12.5)	8 (9.1)	19 (21.6)
Klebsiella pneumonia	4 (4.5)	10 (11.4)	14 (15.9)
Staphylococcus aureus	6 (6.8)	6 (6.8)	12 (13.6)
Candida albicans	3 (3.4)	9 (10.2)	12 (13.6)
Pseudomonas aeruginosa	7 (8.0)	2 (2.3)	9 (10.2)
Klebsiella oxytoca	2 (2.3)	6 (6.8)	8 (9.1)
Staph. Saprophyticus	1 (1.1)	5 (5.7)	6 (6.8)
Serratia rubidaea	2 (2.3)	1 (1.1)	3 (3.4)
Enterobacter agglomerans	1 (1.1)	1 (1.1)	2 (2.3)
Acinetobacter iwoffii	1 (1.1)	0 (0.0)	1 (1.1)
Acinetobacter baumannii	1 (1.1)	0 (0.0)	1 (1.1)
Providencia retgerri	1 (1.1)	0 (0.0)	1 (1.1)
Total	40 (45.5)	48 (54.5)	88 (100)

Table 5 shows the pathogens isolated in relation to different age groups. Of the 88 samples that yielded significant growth, the highest number of isolates 31 (35.2%) was found among the age group 50 and above, followed by the age group 30-39 with 26 (29.5%) isolates, 22 (25%) in age group 20-29, 6 (6.8%) in 0-9 years, 2 (2.3%) in 10-19 and the least 1 (1.1%) seen in age group 40-49.

TABLE 4: ISOLATES FROM FEMALES RELATING PREGNANT TO NON-PREGNANT WOMEN

Isolate	Pregnant	Non-pregnant	Total (%)
E.coli	1	7	8 (16.7)
K.pneumo	2	8	10 (20.8)
S.aureus	3	3	6 (12.5)
C.albicans	3	6	9 (18.8)
Pseudomonas spp	1	1	2 (4.2)
K.oxytoca	3	3	6 (12.5)
S.saprophyticus	2	3	5 (10.4)
S.rubidaea	0	1	1 (2.1)
E.agglomerans	1	0	1 (2.1)
Total (%)	16 (33.3)	32 (66.7)	48 (100)

TABLE 5: PREVALENCE OF ISOLATES AMONG AGE GROUPS

Isolate	Age						Total
	0 - 9	10 – 19	20 – 29	30 - 39	40 - 49	50 >	
E.coli	2	0	6	3	0	8	19
K.pneumo	0	0	2	6	0	6	14
S.aureus	0	0	2	4	0	6	12
C.albicans	1	2	1	6	0	2	12
Pseudomonas spp	1	0	2	1	0	5	9
K.oxytoca	1	0	4	3	0	0	8
S.saprophyticus	0	0	3	1	0	2	6
S.rubidaea	0	0	0	1	0	2	3
E.agglomerans	0	0	1	1	0	0	2
A.iwoffii	0	0	1	0	0	0	1
A.baumannii	0	0	0	0	1	0	1
P.retgerri	1	0	0	0	0	0	1
Total (%)	6 (6.8)	2 (2.3)	22 (25)	26(29.5)	1 (1.1)	31(35.2)	88 (100)

Gram Positive Isolates

All the Gram positive bacteria were resistant to tetracycline and cotrimoxazole. *Staphylococcus aureus* showed varying susceptibility patterns to the antibiotics; augmentin and erythromycin (66.7%), chloramphenicol (58.3%), vancomycin (50%), gentamicin (33.3%) and amoxicillin, cloxacillin and ceftazidime (16.7%). *Staphylococcus saprophyticus* was resistant (100%) to amoxicillin, gentamicin and ceftazidime, mildly sensitive (66.7%) to erythromycin and cloxacillin and moderately sensitive (83.3%) to

augmentin, chloramphenicol and vancomycin (Table 6).

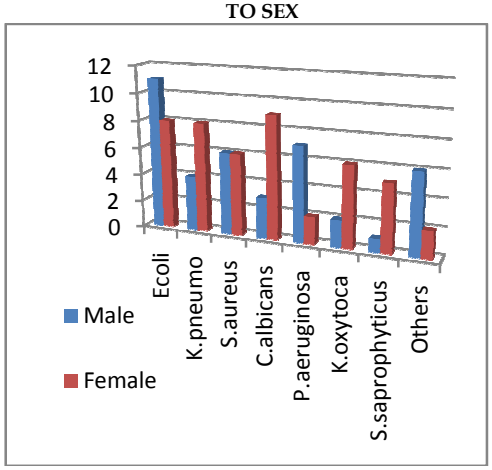
Gram Negative Isolates

All organisms were sensitive to imipenem except *Pseudomonas aeruginosa* which had 22.2% resistance. All the organisms were resistant (100%) to cotrimoxazole. *Escherichia coli* was moderately sensitive to gentamicin (84.2%) and ofloxacin (73.7%), mildly sensitive (36.8%) to nitrofurantoin and tetracycline, (31.6%) to ciprofloxacin, very low sensitivity (21.1%) to ceftazidime, (10.5%) to amoxycillin and (5.3%) to nalidixic acid with 100% resistance to augmentin. *Klebsiella pneumonia* was moderately sensitive to gentamicin (64.3%) and 57.1%) to ofloxacin and ciprofloxacin, mildly sensitive to ceftazidime (50%) and nitrofurantoin (42.9%), very low sensitivity (21.4%) to augmentin, (7.1%) to amoxycillin and nalidixic acid with 100% resistance to tetracycline. *Pseudomonas aeruginosa* was moderately sensitive (88.9%) to gentamicin and ofloxacin, (77.8%) to imipenem, mildly sensitive (55.6%) to nitrofurantoin, ceftazidime and ciprofloxacin, (44.4%) to nalidixic acid and tetracycline with 100% resistance to amoxycillin and augmentin.

Klebsiella oxytoca was moderately sensitive (62.5%) to ofloxacin and ceftazidime, mildly sensitive (50%) to nitrofurantoin and gentamicin, (37.5%) to augmentin and ciprofloxacin, very low sensitivity (25%) to nalidixic acid and tetracycline and (12.5%) to amoxicillin. *Serratia rubidaea* was highly sensitive (100%) to gentamicin, moderately sensitive (66.7%) to ofloxacin and ciprofloxacin, mildly sensitive (33.3%) to amoxicillin, nitrofurantoin, augmentin, tetracycline and ceftazidime with 100% resistance to nalidixic acid.

Enterobacter agglomerans was highly sensitive (100%) to nitrofurantoin, gentamicin and ceftazidime, mildly sensitive (50%) to amoxicillin, nalidixic acid, ofloxacin, augmentin and ciprofloxacin with 100% resistance to tetracycline. *Acinetobacter iwoffii* was highly sensitive (100%) to gentamicin, nalidixic acid, ofloxacin, tetracycline and ceftazidime with 100% resistance to amoxycillin, nitrofurantoin, augmentin and ciprofloxacin. *Acinetobacter baumannii* was highly sensitive (100%) to gentamicin and ofloxacin with 100% resistance to amoxycillin, nitrofurantoin, nalidixic acid, augmentin, tetracycline, ceftazidime and ciprofloxacin. *Providencia retgerri* was highly sensitive (100%) to ceftazidime and ciprofloxacin with 100% resistance to amoxycillin, nitrofurantoin, gentamicin, nalidixic acid, ofloxacin, augmentin and tetracycline. (Table 7)

FIG. 2: ISOLATES FROM SAMPLES IN RELATION TO SEX



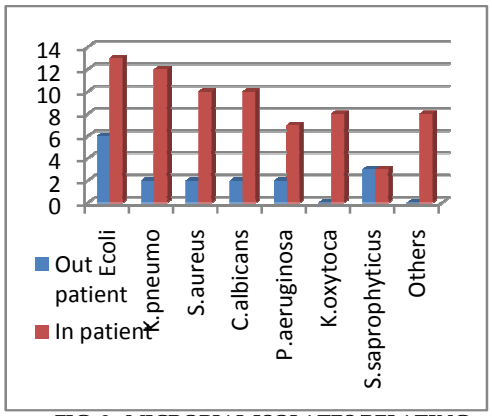


FIG. 3: MICROBIAL ISOLATES RELATING HOSPITAL ACQUIRED TO COMMUNITY ACQUIRED UTI

TABLE 6: ANTIBIOTIC SUSCEPTIBILITY PATTERNS OF THE GRAM POSITIVE ISOLATES

Isolate		AUG	AMX	ERY	TET	CXC	GEN	COT	CHL	VA	CAZ
Staph.aureus N=12	S	8(66.7)	2(16.7)	8(66.7)	0(0.0)	2(16.7)	4(33.3)	0(0.0)	7(58.3)	6(50)	2(16.7)
	R	4(33.3)	10(83.3)	4(33.3)	12(100)	10(83.3)	8(66.7)	12(100)	5(41.7)	6(50)	10(83.3)
S.saprophyticus N=6	S	5(83.3)	0(0.0)	4(66.7)	0(0.0)	4(66.7)	0(0.0)	0(0.0)	5(83.3)	5(83.3)	0(0.0)
	R	1(16.7)	6(100)	2(33.3)	6(100)	2(33.3)	6(100)	6(100)	1(16.7)	1(16.7)	6(100)

AUG = Augmentin, AMX = Amoxycillin, ERY = Erythromycin, TET = Tetracycline, CXC = Coxacillin, GEN = Gentamicin, COT = Cotrimoxazole, CHL = Chloramphenicol, VA = Vancomycin, CAZ = Ceftazidime

TABLE 7: ANTIBIOTIC SUSCEPTIBILITY PATTERNS OF THE GRAM NEGATIVE ISOLATES

Isolate		AMX	COT	NIT	GEN	NAL	OFL	AUG	TET	CAZ	IPM	CIP
E.coli	S	2(10.5)	0(0.0)	7(36.8)	16(84.2)	1(5.3)	14(73.7)	0(0.0)	7(36.8)	4(21.1)	19(100)	6(31.6)
N=19	R	17(89.5)	19(100)	12(63.2)	3(15.8)	18(94.7)	5(26.3)	19(100)	12(63.2)	15(78.9)	0(0.0)	13(68.4)
K.pneumoniae	S	1(7.1)	0(0.0)	6(42.9)	9(64.3)	1(7.1)	8(57.1)	3(21.4)	0(0.0)	7(50)	14(100)	8(57.1)
N=14	R	13(92.9)	14100)	8(57.1)	5(35.7)	13(92.9)	6(42.9)	11(78.6)	14100)	7(50)	0(0.0)	6(42.9)
Pseudomonas	S	0(0.0)	0(0.0)	5(55.6)	8(88.9)	4(44.4)	8(88.9)	0(0.0)	4(44.4)	5(55.6)	7(77.8)	5(55.6)
spp N=9	R	9(100)	9(100)	4(44.4)	1(11.1)	5(55.6)	1(11.1)	9(100)	5(55.6)	4(44.4)	2(22.2)	4(44.4)
K.oxytoca	S	1(12.5)	0(0.0)	4(50)	4(50)	2(25)	5(62.5)	3(37.5)	2(25)	5(62.5)	8(100)	3(37.5)
N=8	R	7(87.5)	8(100)	4(50)	4(50)	6(75)	3(37.5)	5(62.5)	6(75)	3(37.5)	0(0.0)	5(62.5)
S.rubidaea	S	1(33.3)	0(0.0)	1(33.3)	3(100)	0(0.0)	2(66.7)	1(33.3)	1(33.3)	1(33.3)	3(100)	2(66.7)
N=3	R	2(66.7)	3(100)	2(66.7)	0(0.0)	3(100)	1(33.3)	2(66.7)	2(66.7)	2(66.7)	0(0.0)	1(33.3)
E.agglomerans	S	1(50)	0(0.0)	2(100)	2(100)	1(50)	1(50)	1(50)	0(0.0)	2(100)	2(100)	1(50)
N=2	R	1(50)	2(100)	0(0.0)	0(0.0)	1(50)	1(50)	1(50)	2(100)	0(0.0)	0(0.0)	1(50)
A.iwoffii	S	0(0.0)	0(0.0)	0(0.0)	1(100)	1(100)	1(100)	0(0.0)	1(100)	1(100)	1(100)	0(0.0)
N=1	R	1(100)	1(100)	1(100)	0(0.0)	0(0.0)	0(0.0)	1(100)	0(0.0)	0(0.0)	0(0.0)	1(100)
A.baumannii	S	0(0.0)	0(0.0)	0(0.0)	1(100)	0(0.0)	1(100)	0(0.0)	0(0.0)	0(0.0)	1(100)	0(0.0)
N=1	R	1(100)	1(100)	1(100)	0(0.0)	1(100)	0(0.0)	1(100)	1(100)	1(100)	0(0.0)	1(100)
P.retgerri	S	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1(100)	1(100)	1(100)
N=1	R	1(100)	1(100)	1(100)	1(100)	1(100)	1(100)	1(100)	1(100)	0(0.0)	0(0.0)	0(0.0)

AMX = Amoxycillin, COT = Cotrimoxazole, NIT = Nitrofurantoin, GEN = Gentamicin, NAL = Nalidixic acid, OFL = Ofloxacin, AUG = Augmentin, TET = Tetracycline, CAZ = Ceftazidime, IPM = Imipenem, CIP = Ciprofloxacin

DISCUSSION

The study showed that, from 300 urine samples cultured 88(29.3%) yielded significant growth of urinary pathogens while 212 (70.7%) yielded either insignificant growth or no growth of any urinary pathogen. This 29.3% UTI prevalence from the study was lower in comparison to the finding of Yengkokpam *et al*. (40.4%) from India. However, this was in agreement with other study conducted by Chhetri *et al*. (21.8%), Rai *et al*. (28.6%), and Kumari *et al*. (25.7%) from Nepal [23,24,25,26]. All the significant culture growths were monomicrobial. There was higher number of males 160 (53.3%) attending the hospitals on UTI cases than females 140 (46.7%). The age group, 20-29, recorded the highest attendance with 90 (30%) while 40-49 age group had the lowest attendance 13 (4.3%).

The isolated organisms in this study showed that *Escherichia coli* were the most common UTI pathogens and this agrees with previous report of Obiogbolu *et al.* and Kumari *et al.* They found that *E.coli* was the commonest urinary tract pathogen which indicated a high incidence of UTIs (54%) and (42%) respectively [10,26]. Incidence value of *E.coli* in this work is lower than in many previous works. This is probably because some organisms were identified in this work, which were not identified in the previous works. This may be due to the Microbact identification system used and emergence of new strains of pathogens.

Also, the result showed that females 48 (54.5%) were more infected than males 40 (45.5%). This is in accordance with the patterns that UTIs follow universally. Close proximity of perineal structures along with the short urethra predisposes female to be at a higher risk than male. Moreover, the use of diaphragm with spermicide and indiscriminate use of antimicrobial agents may kill or reduce the number of the normal floras of the vagina thereby giving room for pathogens to have a field day [27]. In addition, the antibacterial properties of prostatic fluid play a preventive role in male [28]. Some species of organisms were found to be more in males (*E.coli* and *P.aeruginosa*), others are more in females (*K.pneumoniae, C.albicans, K.oxytoca* and *S.saprophyticus*) while some were equally distributed (*S.aureus*).

Interestingly, UTI was more prevalent in the patients within the hospital settings, 80.4% in-patient versus 19.3% out-patient cases. Patients' comorbidity, immune status and catheter association might account for this [27,28]. All isolated strains were highly prevalent among in-patients than out-patients except for *S.saprophyticus* with equal distribution. From the studies, it was shown that the most common pathogen of female UTI is *K.pneumoniae* (21%), followed by *C.albicans* (19%), *E.coli* (17%), *K.oxytoca* (13%), *S.aureus* (12%), *S.saprophyticus* (10%), *P.aeruginosa* (4%), *S.rubidaea* (2%) and *E.agglomerans* (2%). The study also

revealed that UTI was more prevalent in non-pregnant 32 (66.7%) than in pregnant 16 (33.3%) when female population was considered in general.

The high prevalence of UTI observed in the aged people may be solely due to the inability of their immune systems to fight or resist bacterial infection (Table 5).

Antibiotic sensitivity pattern of organisms is changing rapidly in recent period. It is especially true for developing countries (like Nigeria) where antibiotics are prescribed not only by the medical practitioners but also purchased directly over the counter from pharmacy.

Most of the gram positive cocci (GPC) were sensitive to common antibiotics except tetracycline and cotrimoxazole (Table 6). *Staphylococcus aureus* was moderately sensitive to augmentin, erythromycin, chloramphenicol, vancomycin, gentamicin, amoxicillin, cloxacillin and ceftazidine. *Staphylococcus saprophyticus* was highly resistant to amoxicillin, gentamicin and ceftazidime, mildly sensitive to erythromycin and cloxacillin and moderately sensitive to augmentin, chloramphenicol and vancomycin. This implies that, in UTI with *Staphylococcus spp* within Osun state (or Nigeria as a whole), tetracycline and cotrimoxazole should not be given empirically. But drugs like augmentin, erythromycin and vancomycin could be used instead.

All the gram negative bacteria (GNB) were sensitive to imipenem except *Pseudomonas aeruginosa,* and were largely resistant to cotrimoxazole. This finding is in consonance with the findings of Theodros [29]. It is obvious that cotrimoxazole is no more useful against GNB causing UTI as all the isolates were resistant to it. Previously this antibiotic was used as the drug of choice for empirical treatment of UTI. The broad spectrum activity of fluoroquinolones has made them as one of the best therapeutic options for UTI. In the present study the isolates showed low degree of susceptibility to fluoroquinolones which indicates that they can no more be opted for treating UTI empirically.

Resistance of the isolates to some of the antibiotics agrees with the reports of Khoshbakht *et al.* and Tula et al. [30,31]. This could be attributed to indiscriminate use of these agents by the general physicians. Strict control of antibiotic use and prophylaxis could reverse the situation. This is also applicable to other bacteria isolated. Most of them were resistant to commonly used antibiotics.

Administration of fake drugs in treating cases has been a problem of effective treatment in Nigeria. The circulation of fake drugs should be checked if emergence and resurgence of resistant strains of these isolated pathogens were to be stemmed. Therefore, government support and subvention to National Agency for Food and Drug Administration Control (NAFDAC) must be sustained.

CONCLUSION

Gram-negative bacilli (Enterobacteracea) were mainly responsible for urinary tract infections and most of the strains were multi-drugs resistant. The most common isolated bacteria from urinary tract infections was *E. coli* and the most effective antimicrobial agents against GNB were imipenem, gentamicin, ofloxacin, nitrofurantoin, tetracycline and ciprofloxacin against Gram-negative bacilli. Augmentin, erythromycin, chloramphenicol and vancomycin were most effective for Gram-positive organisms. These justify the necessity to treat UTI based on antimicrobial susceptibility in order to prevent evolution of resistant mutant strains.

Since UTI has a large socio-economic impact and many factors may contribute to the emergence of bacterial resistance in different places with time, the following are strongly recommended: periodic surveillance of antibiotic susceptibility in a systematic manner under supervision of a joint scientific experts and physicians.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

INFORMED CONSENT

The purpose of this work was explained to the clients before they voluntarily consented to participate in the research. The consent forms were appropriated filled by the investigators after which each client signed their corresponding forms.

ETHICAL APPROVAL

Ethical approval for this study was obtained from the ethical and human research committees of the Ladoke Akintola University of Technology Teaching Hospital and Obafemi Awolowo University Teaching Hospital before embarking on the research.

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