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Antibiotic Susceptibility Pattern of Extended Spectrum Beta-Lactamase (ESBLs) Producing Enterobacteriacaea Isolated from Urine in a North-Eastern Tertiary Care Hospital in Nigeria.

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ABSTRACT

This study was done to evaluate the frequency of extended spectrum Beta-lactamase (ESBLs) producing uropathogens and their susceptibility pattern isolated from urine in Abubakar Tafawa Balewa University Teaching Hospital (ATBUTH) Bauchi. A total of 373 urine samples from out-patients and hospitalized patients were studied. Samples were inoculated on Cystine Lactose Electolyte-deficient (CLED) agar, Blood agar and MacConckey agar. 165 isolates were obtained which were further identified by standard Microbiological methods. Antimicrobial Susceptibility pattern was studied by Kirby-Bauer's disc diffusion method. Among the 165 uropathogens isolated from patients with UTI, the commonest isolate was *E. coli* (29.1%) followed by *Klebsiella pneumonae* (15.8%) with the least *Citrobacter spp.* 1(0.6%). The overall prevalence of UTI of this study is 44.0%. Extended spectrum beta lactamase (ESBL) was noted in 100 (77.0%). Among the ESβLs producers detected, *E. coli* recorded the highest occurrence rate of 46 (46.0%) followed by *K. pneumoniae* with 22(22.0%). Others were, *K.oxytoca* 15(15.0%), *P. aeruginosa* 4(4.0%). However, there was no evidence of ESβL production observed in the clinical isolates of *P. vulgaris* and *Citrobacter spp.* Multidrug resistance was found to be significantly (P<0.05) more in ESBL producing isolates (77.0%) than Non-ESBL producers (23.0%. Monitoring of ESBL production and antimicrobial susceptibility testing are necessary to avoid treatment failure in patients with UTI.

INTRODUCTION

Urinary tract infections (UTIs) are among the most common bacterial infection affecting human both in community and hospital settings, with high rates of morbidity imposing substantial economic burden especially in developing countries(1). A diagnosis of UTI is confirmed when a count, more than or equal to 10^5 organisms/mL in an adequately collected mid-stream clean catch urine sample (2). Although UTIs may be caused by a variety of pathogens, they occur most frequently in the Enterobacteriaceae family. There is an increasing frequency of antibiotic resistance among the uropathogens. Urinary tract infections (UTIs) are among the most common and readily treatable infectious diseases; however, timely control is threatened by an unsustainable rise in antibiotic resistance, especially among Enterobacteriaceae. Accordingly, these uropathogenic bacteria represent a major human health problem and socioeconomic burden. ESBL-producing bacteria can produce enzymes that resist a lot of antibiotics. ESBLs are enzymes that provide resistance to a broad-spectrum of beta-lactam antibiotics including penicillins, Sulbactam, first through third-generation

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cephalosporins. Cefoperazone, Ceftaroline, Aztreonam (2-4). This makes these bacteria especially hard to treat because a number of the usual antibiotics are ineffective against it. ESBL-producing bacteria are detected and described in many parts of the world (4), notably North America, Europe, Africa etc. This widespread presence of these resistant bacteria is a serious public health threat, leaving few viable remedies and elevating the likelihood that severe disease-difficult to treat-will occur. ESBL-producing organisms are also associated with a broad range of infections in almost all organs, including meningitis, pneumonia, urinary tract infection (UTI), septicemia and intra-abdominal infections [5–7]. They are capable of causing diseases such as osteomyelitis, endophthalmitis and pyomyositis wound infections (7). In health care facilities, there is significant potential for the spread of ESBL-producing pathogens which is of great concern.The spread of ESBL-producing pathogens in health care-related settings is also a major problem. The antibiotic resistance of ESBLs is associated with morbidity, mortality and difficulties in managing infectious diseases; patient stays longer time due to the spread out of illness have an increased hospital debt caused on healthcare system offering a more cost-effective costly within this context.

METHODOLOGY

Study Area

Abubakar Tafawa Balewa University Teaching Hospital (ATBUTH), Bauchi State, a referral center in North-eastern Nigeria.

Study Design

The study was a hospital based, descriptive and crosss-sectional

Study Population

Both out-patients and in-patients in ATBUTH were considered for the study

Sample size determination

$N= t^2 \times p(1-p)/m^2$

N= required sample size

t= confidence level at 99% (std value 2.575)

P= estimated prevalence 10% (0.1)

m= margin of error at 4% (0.04)

$$N= \frac{2.575^2 \times 0.10\ (1-0.10)}{(0.04)^2}$$

N= 372.09≈373 nearest 10.

Sampling Method

Convenient (non-probability) Sampling

Specimen Collection

First urine passed by the patient was targeted; usually the mid-stream urine (10-20ml) was collected in sterile, dry, wide-necked, leak-proof, screw- capped universal bottle and transported immediately to the hospital medical microbiology laboratory.

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Bacterial Identification

The specimen was inoculated on CLED agar, Blood agar, and Mac conkey in a culture plates. The plates were incubated at 37^0C for 24 hours. Preliminary identification of isolates obtained was carried out based on morphological features (texture, size, edge, elevation, odor, hemolysis, color) and chemical reaction (gram reaction) in accordance to microbiological standards (18). In addition, biochemical tests (citrate test, urea test, catalase and coagulase tests, motility, and triple sugar ion were performed on gram negative isolates using the standard procedures described by (18). The isolates were further characterized based on the Bergey's manual of systemic bacteriology.

Screening Test for ESBLs

The sensitivity of standard inocula of the isolates to Cefotaxime (30µg, Oxoid England) and Ceftazidime (30µg Oxoid England) discs was determined on prepared Mueller-Hinton agar plates using Kirby Bauer method as suggested by (19). Sterile swab sticks were used to inoculate each of Mueller Hinton agar plates with the test organisms by streaking the surface of the media evenly in three directions and left for 3- 5 minutes for the surface of the agar to air-dry. Sterile forcep was used to place the oxoid discs of Cefotaxime and Ceftazidime. Within 30 minutes of disc application, the plates were incubated aerobically at 35^oC for 24 hours after which the zones of growth inhibition were read by measuring the diameters.

Phenotypic confirmation test(s)

Double disc synergy test

This was performed on suspected ESBL producing isolates based on CLSI breakpoint above as described by (19). In this test, each of the identified Gram negative organisms was swabbed onto a Mueller-Hinton agar plate. A susceptibility disc containing amoxicillin-clavulanate (Augmentin) (30µg Oxoid England, LOT-1301407) was then placed at the center of the plate and discs containing Cefotaxime (30µg Oxoid England, LOT-1252576) and Ceftazidime (30µg Oxoid England, LOT-1295263) were placed 15mm (center to center) from the amoxicillin-clavulanate disc. After 30 minutes of pre-incubation time, the plates were incubated aerobically at 35^oC for 24hrs. A difference of ≥5mm between the zone diameters of either of the cephalosporin disc and their respective amoxicillin-clavulanate disc was taken to be phenotypic confirmation of ESBL production.

Antibiotic Susceptibility testing (agar diffusion method)

Kirby-Bauer disk diffusion method was employed. Discrete colonies were picked and emulsified in 3ml sterile aqueous normal saline. The suspension optical density is standardized to a McFarland density of 0.5 with the aid of a Densi ChekTM densitometer (bioMerieux, USA) apparatus. The suspensions were used within 15 minutes of standardization. Dry, sterile, absorbent cotton wool was dipped into the standardized suspension and excess moisture drained by pressing the wet cotton wool against the walls of the test tube. A second swab stick was then dipped into the suspension and then used to streak the surface of a Mueller-Hinton agar plate, which was earlier poured to a uniform depth of 5 mm and dried in the incubator for 15 minutes to reduce excess moisture. The inoculated plates were allowed to stand for 5 minutes and the antibiotic susceptibility discs were placed on the inoculated Mueller-Hinton agar plate. The plates were then incubated aerobically at 37^0C for 16 hours. After overnight incubation the zone of inhibitions were measured with the aid of a meter rule in two directions across each inhibition zone and the results averaged and recorded. CLSI 2024 guidelines for interpretative criteria for susceptibility to antibiotics were adopted. The antibiotic discs used in this study include: Amoxicillin (30ug), Augmentin (25ug), Chloramphenicol (30ug), (25ug), Cotrimoxazole (30ug), Gentamicin (10ug), Ofloxacin (10µg), Sparfloxacin (10µg), Ciprofloxacin (10 µg), Pefloxacin (10µg), Streptomycin (30 µg), Ampiclox (30µg), Zinnacef (20 µg), Cefotaxime (30µg), Ceftazidime (30µg), Amoxacillin clavulanic acid (30µg).

Data Analysis

Data collected was recorded into a computer and analysis was done using statistical package for social sciences

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version 17.0 (SPSS Chicago III, USA). Results were presented when necessary as tables, figures and photograph.

Ethical Consideration

The study was reviewed and approved by the ethical review committee of ATBUTH

RESULTS

Of the 373 urine samples collected for the study 99(60%) were from the outpatients while the inpatients constituted 66(40%) as shown in table 1. Table 2 shows the distribution of urine samples in relation to age and gender of the patients, with females having the highest frequency (64.9%) while males have the least with (35.1%). The age bracket of 25-30 has the highest number of samples collected followed by 17-24, with the least 0-16 (6.7). Out of the 373 urine sample collected 165(44.2%) uropathogens were obtained. Most of the uropathogens were isolated from the female patients (73.9%), while the uropathogens isolated from the male patients constituted 26.1%. In addition the age group 25-32 has the highest distribution rate of 21.0% of the uropathogens, followed by 33-40 (20.0%), with 0-16 age group having the least occurrence rate of the uropathogens isolated. Of the 165(44.2%) isolates obtained, *Escherichia coli* ranked highest 48(29.1%), others were *Klebsiella pneumoniae* 26(15.8%), *Klebsiella oxytoca* 25(15.2), *Proteus mirabilis* 19(11.5), *Proteus vulgaris* 5(3.0%), *Pseudomonas aeruginosa* 4(2.4%), *Enterobacter spp* 2(1.2%) and *Citrobacter spp* 1(0.6). Of the gram positive bacterial isolates, Coagulase negative Staphylococcus accounted for 21(12.7), *Staphylococcus aureus* 10(6.1%), and *Candida albicans* 4(2.4%) (Table 4). Table 5 shows the antimicrobial susceptibility test of gram negative isolates. It showed that most of the *E. coli* isolates were resistant to amoxicilin and Ceftazidime (54.2%), pefloxacin (50.0%). In addition 41.7% of the *E. coli* isolates were sensitive to both Augmentin and gentamycin respectively. 37.5% of the *E. coli* isolates were sensitive to Ciprofloxacin, streptomycin (35.5%) and (31.3%) to cotrimoxazole. Of the *Klebsiella pneumoniae* isolates 68.8% were resistant to cotrimoxazole and augmentin respectively, 54.5% were resistant to cofotaxime, 45.5% to ceftazidime, 27.3% to amoxicillin and 72.2% to ciprofoxacin. However, 27.3% were sensitive to cefotaxime, pefloxacin and amoxicillin respectively. *Proteus mirabilis* sensitive to Ciprofloxain, gentamycin amoxicillin and ceftazidime were 3(15.8%), 5(26.3%), 8(42.1) and 4921.1) respectively. Least susceptibIlity to ciprofloxacin (57.9%), 42.1% to gentamycin and 68.4% to third generation cephalosporins (cefotaxime). *Pseudomonas aeuroginosa* isolates were resistant to cotrimoxazole, amoxicillin, ceftotaxime and ceftazidime 4(100%), Chloramphenicol 2(50.0%), pefloxacin 3(75.0%), but 1(25.0%). Citrobacter spp were resistant to cotrimoxazole, chloramphenicol, ciprofloxacin and augmentin 1(100%) respectively. Likewise it was observed that the isolates were sensitive to cefotaxime and ceftazidime 1(100%).

On subjecting the Gram negative isolates to screening test for occurrence of ESBLs based on Clinical and Laboratory Standards Institute (CLSI) breakpoint, 100 (77%) were found to be positive. Among the ESBLs producers detected, *E. coli* recorded the highest occurrence rate of 46 (46%) followed by *K. pneumoniae* with 22(22%). Others were, *K.oxytoca* 15(15%), *P. aeruginosa* 4(4%). However, there was no evidence of ESβL production observed in the clinical isolates of *P. vulgaris* and *Citrobacter spp* (Table 6).

The Resistance pattern of ESBL producing isolates obtained from the hospital were higher than that from the community. 33% of the ESBLs producing isolates obtained from the in-patients were resistant to Cotrimoxazole as compared with that from outpatients (17%) (p- 0.217044), and amoxicillin compared to those obtained from the community(p- 0.23323).The sensitivity pattern of bacterial isolates: *E.coli, Klebsiella spp, Protues spp, Staphylococcus aureus ,Pseudomonas aeruginosa,Citrobacter spp and Enterobacter spp* from CA UTI was considered lower for the majority of the antibiotics as compared to isolates from the HA UTI. This difference was found to be statistically significant ($p<0.05$) Table7)

Table 1: Out Patient and In-patient distribution of Patients

Source	No. Of Samples n=373	No. Positive (%)
Outpatient	224	60

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Inpatient	149	40
Total		

Table 2: Distribution of samples in relation to age and sex

Age group	**Sex**		**Total**
	Male	Female	
0-16	9	16	25
17-24	25	45	70
25-32	28	47	75
33-40	18	38	56
41-48	12	27	39
49-56	20	31	51
57-64	9	19	28
>64	10	19	29
Total	131	242	373

Table 3: Table 2 Distribution of bacterial isolates in relation to patient demography

Demographic details	**No. Of Samples collected (n=373)**	**No. Of bacterial isolate (n=165)**	**Percentage (%)**
Gender			
Male	131	43	26.1
Female	242	122	73.9
Age			
0-16	25	08	5.0
17-24	70	32	19.0
25-32	62	35	21.0
33-40	56	33	20.0
41-48	45	17	10.0
49-56	40	19	12.0
57-64	36	9	6.0
>64	39	12	7.0

Table 4: Distribution of uropathogen isolates

Isolate	**No. Isolated n =165**	**% Occurrence**
Citrobacter spp	1	0.6

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Escherichia coli	48	29.1
Enterobacteriacaea spp	2	1.2
Klebsiella oxytoca	25	15.2
Klebsiella pneumonae	26	15.8
Proteus mirabilis	19	11.5
Proteus vulgaris	5	3.0
Pseudomonas aeruginosa	4	2.4
Staphylococcus aureus	10	6.1
Coagulase negative (CON)*Staph.*	21	12.7
Candida albicans	4	2.4

Table 5: Antimicrobial susceptibility pattern gram-negative bacteria isolated.

Bacteria	**Pat-tern**	**SXT no. (%)**	**CH no. (%)**	**CPX no. (%)**	**AU no. (%)**	**CN no. (%)**	**AM no. (%)**	**PEF no. (%)**	**S no. (%)**	**CTX no. (%)**	**CAZ no. (%)**
E. coli (n = 48)	S	15(31.3)	4 (8.3)	18(37.5)	20(41.7)	20(41.7)	16(33.3)	18(37.5)	17(35.4)	8(167)	9(18.8)
	I	3 (6.25)	20(41.7)	17(35.4)	8(16.7)	10(20.8)	6(12.5)	6(12.5)	11(22.9)	18(37.5)	11(22.9)
	R	30 (62.5)	24(50.0)	23(47.9)	20(41.7)	18(37.5)	26(54.2)	24(50.0)	20(41.7)	22(45.8)	26(54.2)
K.pneumoniae (n=22)	S	3 (13.6)	5(22.7)	2(9.1)	3(13.6)	8(36.3)	6(27.3)	2(90.1)	3(13.6)	4(18.2)	4(18.2)
	I	4 (18.2)	6(27.3)	4(18.2)	4(18.2)	7(31.8)	6(27.3)	6(27.3)	16(72.7)	6(27.3)	8(36.3)
	R	15(68.8)	11(50.0)	16(72.7)	15(688)	7(31.8)	10(45.5)	14(63.6)	3(13.6)	12(54.5)	10(45.5)
K. oxyto-ca (n=15)	S	2 (13.3)	4(18.2)	1(6.7)	2(13.3)	3(20.0)	2(13.3)	2(13.3)	1(6.7)	4(18.2)	3(20.0)
	I	4 (26.7)	5(33.3)	3(20.0)	2(13.3)	5(33.3)	4(18.2)	0(0.0)	3(20.0)	2(13.3)	4(18.2)
	R	9 (60.0)	6(40.0)	11(73.3)	11(73.3)	7(46.7)	9(60.0)	13(86.7)	11(73.3)	9(60.0)	8(53.3)
P.mirabilis (n=19)	S	10 (52.6)	6(31.6)	3(15.8)	5(26.3)	5(26.3)	8(42.1)	4(21.1)	7(36.8)	0(0.0)	4(21.1)
	I	7 (36.8)	4(21.1)	5(26.3)	4(21.1)	10(52.6)	3(15.8)	3(15.8)	3(15.8)	6(31.6)	3(15.8)
	R	2 (10.5)	9(47.4)	11(57.9)	10(52.6)	4(21.1)	8(42.1)	12(63.2)	9(47.4)	13(68.4)	12(63.2)
P.vulgaris	S	2 (40.0)	3(60.0)	1 (20.0)	2(40.0)	1 (20.0)	1 (20.0)	0(0.0)	1 (20.0)	1 (20.0)	1 (20.0)

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(n=5)											
	I	1 (20.0)	0(0)	1 (20.0)	0(0.0)	2(40.0)	1 (20.0)	2(40.0)	0(0.0)	0(0.0)	0(0.0)
	R	2 (40.0)	2(40.0)	3(60.0)	3(60.0)	2(40.0)	3(60.0)	3(60.0)	4(80.0)	4(80.0)	4(80.0)
P.aeruginosa (n=4)	S	0(0.0)	1(25.0)	0(0.0)	1(25.0)	1(25.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
	I	0(0.0)	1(25.0)	1(25.0)	0(0.0)	1(25.0)	0(0.0)	1(25.0)	1(25.0)	0(0.0)	0(0.0)
	R	4 (100)	2(50.0)	3(75.0)	3(75.0)	2(25.0)	4(100)	3(75.0)	3(75.0)	4(100)	4(100)
Citrobacter spp (n=1)	S	0(0.0)	0(0.0)	0(0.0)	0(0.0)	1	1	1	1	0(0.0)	0(0.0)
	I	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)
	R	1 (100)	1(100)	1(100)	1(100)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)	0(0.0)
Enterobacter spp (n=2)	S	0(0.0)	0(0.0)	1(100)	1(100)	1(100)	0(0.0)	1(100)	0(0.0)	0(0.0)	0(0.0)
	I	1 (50.0)	1(50.0)	1(50.0)	1(50.0)	1(50.0)	1(50.0)	0(0.0)	1(50.0)	0(0.0)	0(0.0)
	R	1 (50.0)	1(50.0)	0(0.0)	0(0.0)	0(0.0)	1(50.0)	1(50.0)	1(50.0)	2(100)	2(100)

SXT Cotrimoxazole; CH Chloramphenicol; CPX Ciprofloxacin; AM Amoxicillin; AU Augmentin; CN Gentamycin; PEF Pefloxacin; S Streptomycin; CTX Cefotaxime; CAZ Ceftazidime

R= Resistant; S = Sensitive; I = Intermediate.

Table 6 Distribution of ESBL producing bacteria isolated

ISOLATES	**No. TESTED (%) n=130**	**No. POSITIVE (%)**	**No. NEGATIVE (%)**
Escherichia coli	48(36.9)	46(46.0)	02(6.7)
Proteus mirabilis	19(14.6)	12(12.0)	07(23.0)
Proteus vulgaris	05(3.8)	0(0)	05(16.7)
Klebsiella oxytoca	25(19.2)	15(15.0)	10(33.3)
Klebsiella pneumoniae	26(20.0)	22(22.0)	04(13.3)
Pseudomonas aeruginosa	04(3.1)	04(4.0)	00(0)
Citrobacter spp	01(0.8)	00(0.0)	01(3.3)
Enterobacter spp	02(1.5)	01(1.0)	01(3.3)
Total	130	100(77.0)	30(23.0)

Table 7: Frequency of multiple resistance among ESBL producing bacterial isolates in relation to mode of CA and HA

Antibiotics	**Disc potency (µg)**	**ESBL-producing Isolates**		**P-Value**
		CA UTI n=74	**HA UTI n=56**	
Cotrimoxazole	30µg	17	25	0.639412

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Chloramphenicol	30µg	19	22	0.705457
Ciprofloxacin	30µg	12	20	0.157299
Augmentin	25µg	31	23	0.276303
Gentamycin	10µg	15	20	0.398025
Amoxicillin	30µg	24	33	0.233230
Pefloxacin	10µg	1	5	0.102470
Streptomycin	30µg	17	15	0.723674
Cefotaxime	30µg	21	29	0.257899
Ceftazidime	30µg	25	35	0.196706

UTI- Urinary tract Infection; CA- Community associated; HA- Hospital associated

DISCUSSION

This study revealed a prevalence of 44.2% UTI and 76.9% prevalnce of ESBL in patients attending ATBUTH. This prevalence was found to be higher than the prevalence of the study carried out by (8,9) which showed 13%, and 51.9% respectively but lower than the studies in Enugu (77.9%) and Yola (67.2%). The variation in prevalence may be attributed to the differences in study populations and in the criteria used by centers in selecting urine samples for culture as most of the request in this study came from the outpatient department which sees most of the cases coming in directly from the community. The study showed that UTI was more frequent in women (73.1%) than men (26.1%) which is in agreement with previous studies (10,11). The higher frequency in females has been attributed to the shorter female urethra and the proximity of this to the gastrointestinal outlet, hence making it easier for enteric flora to colonize this area(12,13). Other contributory factors may include the use of contraceptives, childbirth and menopause(14). The age group with the highest incidence was found to be among the sexually active age group . This finding is in line with that of (11), thus explaining the relatively high incidence rate within these age groups (11). The isolated pathogens in this study include both gram positive and gram negative bacteria. The gram negative constituted the highest incidence (78.8%) as compared to the gram positive (21.2%). This finding agrees with the reports of(10,15). *Escherichia coli* was the most frequently isolated common UTI pathogens (29.1%). This agrees with previous reports of (11,15) with incidences of 45.9% and 36.0%, respectively. The high prevalence of *E. coli* could be that, it is the most common commensal organism. *K. pneumoniae* which was the second most common uropathogen isolated in this study is an indication that the organism is achieving more prominence as etiological agents of UTI than previously reported by (2). Coagulase negative stapphylococcus with prevalence rate of 12.4% constituted the highest incidence in gram positive bacterial isolates. The frequency of antimicrobial resistance among microorganisms that cause UTI is increasing worldwide and is a major factor in selecting antibiotics for treatment. There are local variations in the antimicrobial susceptibility among urinary pathogens in different hospitals. The results of Gram-negative antibiotic susceptibility test revealed varied susceptibility ranging from sensitive, intermediate and resistant. All the Gram-negative organisms were variably resistant to Chloramphenicol, Ciprofloxacin, Augmentin and Amoxicillin.The gram-positive drug susceptibility pattern showed high resistant to quinolones, aminoglycoside, Cephalosporins. This finding is similar to that of (2,16,17).The resistance to these antibiotics may be attributed to the purchase of drugs over the counter, administration of false drugs in treating cases when no prior test is carried out and misuse of drugs. Effective management of patients suffering from UTIs commonly relies on the accurate identification of etiological agents and the selection of an appropriate antimicrobial agent. The multidrug resistant status of these isolates in this study indicate possible production of resistance enzymes like extended spectrum beta latamases (ESBLs) and carbapenemase. The result obtained from the antibiotic profile of both ESBL from the community goes a long way to describe the degree of abuse and misuse of common routine antibiotics in the society. The high resistance of ESBL bacterial isolates from the hospital to commonly used antibiotics is also attributed to the continued exposure of bacteria to routine antibiotics used in the hospital. This finding is in line with the findings of (5), who reported high resistance of ESBL and Non-ESBL bacterial isolates from the

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hospital to commonly used antibiotics.

CONCLUSION

The findings of this study showed an alarming rate of 77.0% ESBL-producing bacterial isolates in ATBUTH with high *E.coli* having the highest occurrence, followed by *K. pneumoniae* and least *Pseudomonas aeruginosa.*In addition the study showed that high resistance of these ESBLs- producing isolates to antibiotics of the class aminoglycosides, Flouroquinolones, betalactams, sulfonamides and third generation cephalosporins, which poses a great threat to public heath. As such continuous monitoring of antibiotic susceptibility as well as screening of ESBL should be carried out in order to prevent the further of antibiotic resistance and prevent therapeutic failure.

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