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Extended Spectrum Beta Lactamase (ESBL) producing enteric bacteria from hospital wastewater, Ibadan, Nigeria

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ABSTRACT

This study was conducted to investigate the occurrence of antibiotic resistance, including β -lactamase and extended spectrum β -lactamase production among enteric bacteria isolated from hospital wastewater from selected hospital within Ibadan. Physico-chemical analysis of hospital wastewater samples was done, enteric bacteria were isolated and identified using convectional biochemical tests while the selection of potential ESBL-producing bacteria was carried out using disc diffusion method and ESBL detection using double synergy test. The turbidity of the wastewater samples ranged between 4.45-6.5 NTU and total suspended solids ranged between 3.4- 45.5 mg/L. While electrical conductivity was between 114.25-214 $\mu\text{s/m}$, the biological oxygen demand was between 25.8-31.25 mg/L and chemical oxygen demand ranged between 41.25-45.38 mg/L. Of the 200 bacteria isolated 35(17.5%) produced ESBL; 14(40%) from the tertiary hospital and 21(60%) from private hospital out of which 85.7%, 80% and 65.7% showed resistance to sulphamethaxazole/Trimetoprim, streptomycin and tetracycline respectively, while resistance to meropenem (8.6%) was low. Among the ESBL-producing isolates, *K. pneumonia* had the highest (15(42.8%)) rate of occurrence. This study revealed a need for hospital wastewater to be properly treated before discharged into water bodies and the environment to forestall the indiscriminate discharge of wastewater harbouring ESBL-producing bacteria.

Keywords: Extended Spectrum Beta Lactamase, Enteric bacteria, Antibiotics, Resistance, Hospital wastewater, Environment, physico-chemical, Wastewater treatment, *Klebsiella pneumonia*, *Escherichia coli*

1. INTRODUCTION

Hospital wastewater is a key source of environmental pollution especially in the developing countries of the world, where wastewater treatment plants are either not available or ill-equipped to handle effective treatment. Wastewater are generated from all the activities in the hospital including medical and non-medical activities from the operating, diagnosis, emergency, first aid, laboratory, radiology, kitchen and laundry activities (Majlesinasr, 1998).

Based on these activities, wastewater containing high content of enteric pathogens such as bacteria, viruses and helminthes which can be transmitted to humans through water is of great public health concern. The wastewaters that are generated in the wards during patient's treatment are contaminated with enteric pathogens and could be particularly problematic during outbreaks of diseases such as diarrhoea (Chitnis *et al.*, 2000; Stuart, 2002).

Uncontrolled and excessive use of antibiotics in human and animals' therapy has been one of the reasons for the increase in antibiotic resistance and the spread of resistance genes in environmental samples such as hospital wastewater (Iversen *et al.*, 2002). According to a previous report, hospital wastewater is a highly selective environment that contributes to the high rates of resistant bacteria that are being discharged into the natural environment. Effluents generated from hospitals contain antibiotic residues with a concentration that inhibits the growth of susceptible bacteria thereby leading to high numbers of resistant bacteria (Yang *et al.*, 2009).

Hence, effluents from hospital could augment the number and prevalence of antibiotics resistant bacteria in the environment where the effluents are discharged; this can be by both the means of introduction and selection for resistant bacteria (Al-Ahmad *et al.*, 1999).

In general, hospitals provide an environment conducive for multi-drug resistant bacteria, and especially the ESBL-producing bacteria making the treatment options limited and expensive (Magiorakos *et al.*, 2011). However, there is a dearth of information on the release and survival of this pathogens from the hospital wastewater, through the sewerage system and finally into treated effluent released by sewage treatment plants (STPs) into the environment especially in the developing countries of the world.

The aim of this present study was to determine the occurrence of antibiotic resistance and extended spectrum β -lactamase production among enteric bacteria isolated from hospital wastewater of selected hospital in Ibadan.

2. MATERIALS AND METHODS

2. 1. Description of the study area

The study sites were two selected hospitals. The first one is a private hospital that lies between longitude of 3°15'18.2E and latitude of 7°23'36.4N with no wastewater treatment facility before discharge into a neighbouring river.

The river also receives effluents from residential and industrial houses. The second is a tertiary hospital that lies between longitude 3°54'13.7E and latitude 7°24'19.5N.

Treatment facility for wastewater is available in this hospital before discharge into a neighbouring river.

2. 2. Sample collection

Wastewater samples were collected in sterile containers, preserved in ice pack and transported to the Environmental Microbiology laboratory, Department of Microbiology, University of Ibadan, for immediate analyses.

2. 3. Physico-chemical analysis of the wastewater samples

The physico-chemical analyses of water samples were performed using standard analytical methods. Parameters with extremely low stability such as temperature and pH were determined at the point of sample collection using thermometer and pH meter respectively. Total dissolved solid, electrical conductivity and dissolved oxygen were determined using an extech digital meter (Extech Instruments, USA). Chemical oxygen demand (COD), suspended solids, acidity, alkalinity, chloride, biochemical oxygen demand (BOD) and oil and grease were determined using standard methods as previously described (APHA, 2005).

2. 4. Isolation of the enteric bacteria

Serial dilution was carried out on the wastewater samples up to a 10 fold dilution factor and dilution 3 and 4 (10^{-3} and 10^{-4}) were chosen for media inoculation. The standard pour plate method was used for both the heterotrophic bacteria count and isolation of enteric bacteria. The total heterotrophic count was done on Nutrient agar while isolation of the enteric bacteria was on Eosin Methylene Blue Agar (EMB) and MaCconkey Agar. The plates were incubated for 24-48 hours at 35-37 °C. Distinct colonies were subcultured on freshly prepared media to obtain pure culture.

2. 5. Antibiotic susceptibility tests

Antibiotic susceptibility tests on the isolates was performed on Mueller Hinton Agar (Oxoid, England) using the standard disk diffusion technique as recommended by Clinical Laboratory Standard Institute (CLSI, 2017). The antibiotic discs that were used for the susceptibility test include: cefotazime (30 mg), Amoxicillin/clavulanic acid (30 mg), ceftazidime (30 mg), tetracycline (30 mg) ciprofloxacin (5 mg), chloramphenicol (30 mg), meropenem (10 mg), streptomycin (10 mg), sulphamethoxazole/trimethoprim (25 mg). The isolates were grown on Nutrient Agar for 18-24 hours; 3-5 colonies of the isolates were suspended in 0.85% saline solution and adjusted to achieve the turbidity equivalent of 0.5 McFarland standard. Using sterile swab sticks, the isolates were swabbed on the entire surface of the freshly prepared Mueller Hinton Agar and the antibiotics were aseptically placed on the swabbed Mueller Hinton agar plates using sterile forceps. The plates were incubated for 18-24 hours at 35-37 °C. Zones of inhibition were measured to the nearest millimeter and interpreted according to Clinical Laboratory Standards Institute (CLSI, 2017).

2. 6. Detection of ESBL-producing enteric bacteria using Double Disc Synergy Test (DDST)

Inoculum of 18-24 hours old cultures of isolates was introduced into test tubes containing normal saline and the turbidity of each was adjusted to 0.5 MacFarland standard. The standardized bacterial test suspension was inoculated on Mueller Hinton agar plates by uniformly swabbing the entire surface of the agar plates. The double disc synergy test was done

using discs of augmentin (20 mg amoxicillin + 10 mg clavulanate) and discs of ceftazidime (30 mg) and cefotaxime (30 mg); which were placed around augmentin disc keeping the distance 20 mm from it (center to center). The plates were incubated at 37 °C and after 18-24 hours of incubation, the plates were observed and the organisms was considered to be producing ESBL when the zones of inhibition around any of the cephalosporin discs showed a clear cut increase towards the augmentin (Jarlier *et al.*, 1988). The following were used as control: ESBL-positive *Klebsiella pneumoniae* ATCC 700603 and ESBL negative *Escherichia coli* ATCC 25922.

3. RESULTS

3. 1. Physico-chemical analysis

The results of the physicochemical analysis of the wastewater samples from the two study area are as shown in Table 1. The highest temperature value (29 °C) was obtained from the treated wastewater sample of the tertiary hospital, so also for the acidity (0.036 mg/L) and pH (6.6). The highest value for turbidity (6.5), total soluble solid (45.5 mg/L) and dissolved oxygen (7.94 mg/L) were obtained from the wastewater sample of the private hospital. The alkalinity values obtained from the three wastewaters samples were approximately the same (180 mg/L) while the highest values of 214 µs/m, 45.38 mg/L, 521.6 mg/L, 7.43 mg/L, 21.05 mg/L, and 5.49 mg/L for electrical conductivity, chemical oxygen demand, chloride, lead, sulphate and magnesium respectively were obtained from the untreated wastewater sample of the tertiary hospital.

Table 1. Results of the Physico-chemical analysis of the wastewater samples.

PARAMETERS	PRIVATE HOSPITAL	TERTIARY HOSPITAL	
		Untreated wastewater	Treated wastewater
PH	6.5	5.65	6.6
Temperature (°C)	28	27.8	29
Electrical Conductivity (µs/m)	114.25	214	117
Turbidity	6.5	6.28	4.45
Dissolve Oxygen (mg/L)	7.94	5.95	6.75
Biochemical Oxygen Demand (mg/L)	31.65	31.25	25.8
Chemical Oxygen Demand (mg/L)	44.55	45.38	41.25
Total Soluble Solid (mg/L)	45.5	3.4	2.92

Acidity (mg/L)	0.035	0.026	0.036
Alkalinity (mg/L)	180.38	180.55	180.30
Chloride (mg/L)	140.5	521.6	390.45
Sulphate (mg/L)	8.10	21.05	12.06
Phosphate (mg/L)	14.80	10.55	8.78
Nitrate (mg/L)	0.011	0.016	0.010
Oil & Grease (mg/L)	0.007	0.110	0.051
Lead (mg/L)	1.74	7.43	6.55
Cadmium (mg/L)	1.64	5.92	2.1
Arsenic (mg/L)	0.01	0.001	0.001
Cobalt (mg/L)	0.02	0.001	0.001
Copper (mg/L)	1.03	1.92	1.24
Manganese (mg/L)	0.13	2.65	1.2
Magnesium (mg/L)	2.1	5.49	3.77

3. 2. Frequency of occurrence of enteric bacterial isolates

A total of 200 enteric bacteria were isolated from the wastewater samples of the two hospitals; while 105 were from the private hospital, 95 were from the tertiary hospital. The bacterial isolates belonged to nine genera; *Escherichia* 49 (24.5%), *Klebsiella* 37 (18.5%), *Citrobacter* 6 (3%), *Enterobacter* 43 (21.5%), *Salmonella* 10 (5%), *Kluyyera* 6 (3%), *Serratia* 10 (5%), *Providencia* 12 (6%), and *Proteus* 27 (12.5%). Among the isolates obtained from the private hospital, *Escherichia* (27.6%) and *Proteus* (23.8%) had the highest percentage of occurrence while from the tertiary hospital, it was *Escherichia* (21.1%) and *Proteus* (21.1%). Furthermore, the isolate with the least rate of occurrence from the private hospital was *Providencia* (1.9%) while it was *Kluyyera* (1.1%) for the tertiary hospital (Table 2).

Table 2. Occurrence of enteric bacteria isolated from the wastewater samples n (%)

Genus	Sampling sites		
	Private hospital (n = 105)	Tertiary hospital (n = 95)	Total (n = 200)
<i>Escherichia</i>	29(27.6)	20(21.1)	49(24.5)
<i>Klebsiella</i>	22(21)	15(15.8)	37(18.5)

<i>Citrobacter</i>	3(2.9)	3(3.2)	6(3%)
<i>Enterobacter</i>	25(23.8)	18(18.9)	43(21.5)
<i>Proteus</i>	7(6.7)	20(21.1)	27(13.5)
<i>Salmonella</i>	7(6.7)	3(3.2)	10(5)
<i>Kluyyera</i>	5(4.8)	1(1.1)	6(3)
<i>Serratia</i>	5(4.8)	5(5.3)	10(5)
<i>Providencia</i>	2(1.9)	10(10.5)	12(6)

3. 3. Antibiotic Resistance profile of the enteric bacteria isolated from the wastewater samples

The susceptibility pattern of the isolates showed that 148 (74%) of the isolates were resistant to sulphamethazole/trimetoprim, 127 (63%) to streptomycin, 116 (58%) to tetracycline, and 87 (43%) to chloramphenicol. In addition, it was observed that 48 (24%), 57 (28%), 80 (40%), 87 (44%), and 134 (67%) showed resistance to ciprofloxacin, meropenem, ceftazidime, cefotaxime and amoxicillin clavulanate respectively (Table 3).

Table 3. Antibiotic susceptibility profile of enteric bacteria isolates from the wastewater samples (n = 200)

Antibiotics	R	I	S
SXT	148(78%)	4(2%)	48(24%)
S	127(63%)	19(10%)	54(27%)
TE	116(58%)	21(10%)	63(32%)
C	87(43%)	54(27%)	59(30%)
CIP	48(24%)	17(9%)	135(67%)
MEM	57(28%)	24(12%)	119(60%)
CEF	80(40%)	24(12%)	96(48%)
CTX	87(44%)	18(9%)	95(47%)
AMC	134(67%)	17(9%)	49(24%)

KEYS: R = Resistance, I = Intermediate, S = Susceptible, TE = Tetracycline, S = Streptomycin, C = Chloramphenicol, CIP = Ciprofloxacin, M = Meropenem, CEF = Ceftazidime, CTX = Cefotaxime, AMC = Amoxicillin/clavulanate, SXT = Sulphamethoxazole/Trimethoprim

3. 4. Detected ESBL-producing enteric bacteria

Out of the 200 bacterial isolates obtained from the wastewater samples of the two hospitals, a total of 35 (17.5%) were positive for ESBL production. Out of these, 21(60%) were from the private hospital while 14(40%) were from the tertiary hospital. The percentage and number of occurrence of ESBL-producing enteric bacteria isolates is shown in Table 4 with the genus *Klebsiella* having the highest percentage and number of occurrence.

Table 4. Occurrence of ESBL-producing enteric bacteria isolates from the wastewater samples n (%)

Isolates	Number of occurrence (n = 35)		
	Private hospital	Tertiary hospital	Total
<i>Serratia</i> spp.	0(0)	1(2.9)	1(2.9)
<i>Escherichia coli</i>	4(11.4)	3(8.6%)	7(20%)
<i>Proteus</i> spp.	3(8.6%)	3(8.6%)	6(17.1%)
<i>Enterobacter</i> spp.	4(11.4%)	2(5.7%)	6(17.1%)
<i>Klebsiella</i> spp.	10(28.6%)	5(14.3%)	15(43%)
Total	21(60%)	14(40%)	35(100%)

3. 5. Antibiotic resistance profile of the Extended Spectrum Beta-Lactamase producing bacteria

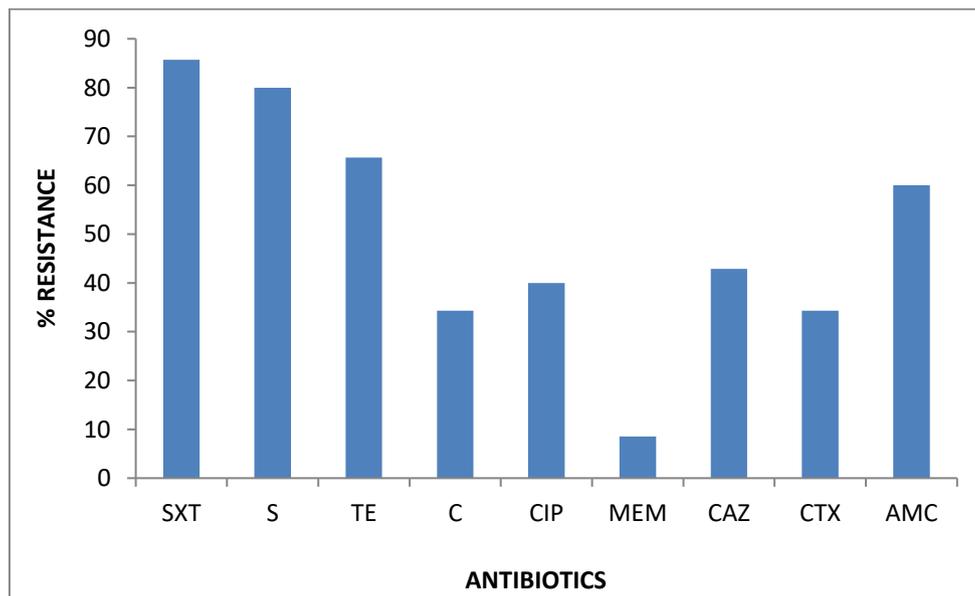


Figure 1. Percentage Frequency of Resistance of the extended spectrum beta lactamase producing enteric bacteria isolates to the antibiotics.

KEYS: TE = Tetracycline, S = Streptomycin, C = Chloramphenicol, CIP = Ciprofloxacin, M = Meropenem, CEF = Ceftazidime, CTX = Cefotaxime, AMC = Amoxicillin/clavulanate, SXT = Sulphamethoxazole/Trimethoprim

The resistant profile of the ESBL-producing bacteria is as shown in Figure 1. Resistance to sulphamethoxazole/trimethoprim was the highest while the lowest was to meropenem.

4. DISCUSSION

The physiochemical parameters of the wastewater samples obtained from the present study from which the pH values of the private and treated tertiary hospital wastewaters were within the range, 6.5 and 6.6 respectively, fell within the Standard Organization of Nigeria (SON) permissible limit (6.5-8.5) of effluent to be discharged into receiving water (SON, 2009). Similarly, the pH value from the untreated wastewater samples of the tertiary hospital which was the lowest (5.65) among the studied samples was below the local permissible limit (SON, 2009). The implication of this observation is that the presence of acid salts (such as ammonium and aluminium salts) is likely to be present in the hospital wastewater. However, these pH values were lower compared to the range of pH values (9.3-9.4) previously reported on samples collected from three rivers also in Ibadan, Nigeria (Falodun *et al.*, 2018). The difference could be due to the studied samples. But the pH values obtained from this study is in agreement with the pH 6.5 from a study on well water samples collected from an abattoir environment (Falodun and Ajala, 2018).

Furthermore, the temperature of all the wastewaters which were within the range 26 °C – 29 °C were within the international permissible limit of less than 35°C as recommended by the World Health Organization (WHO, 2006). These values are also in agreement with the temperature values from a study on samples collected from selected rivers and well water from abattoir environments in Ibadan (Falodun and Ajala, 2018; Falodun *et al.*, 2018). However, the turbidity of 6.5 NTU and 6.28 NTU obtained from the private and untreated tertiary hospital wastewaters respectively were above the S.O.N permissible limits of 5.0 NTU (SON, 2009). But the turbidity (4.45 NTU) obtained from the wastewaters of the treated tertiary hospital, fell within the permissible limit. This is an indication that treatment of the wastewater might have led to the reduction in the turbidity. however, the turbidity values obtained from this study is not in agreement but lower compared to the values reported from well water samples (9.4 NTU) and pond water samples (14.7 NTU) in the same city (Falodun and Ajala, 2018).

The values of the chemical oxygen demand (COD) of the three wastewater samples that were within the range 41.25 mg/L - 45.38 mg/L were all far below the Federal Ministry of Environment (FMENV) and WHO permissible limit of 80 mg/L and 60 mg/L respectively (FMENV, 2001; WHO, 2008). Likewise, the DO of the wastewater samples (range 5.95 mg/L and 7.94 mg/L) were below the permissible limit of 10 mg/L and 150 mg/L of the Federal Environmental Protection Agency (FEPA) and World Health Organization respectively set for hospital wastewater (FEPA, 1991; WHO, 2006). The BOD of 31.65 mg/L (private hospital) and 31.25 mg/L (untreated tertiary hospital) obtained from this study were slightly above the permissible limit of 30 mg/L of the World Health Organization (WHO, 2006). Although, the value of the BOD of the treated tertiary hospital wastewater was within the WHO permissible limit, if the indiscriminate discharge of hospital wastewater with high BOD into the environment is not controlled, over time, the dissolved oxygen of the receiving water bodies

may fall below the standard and consequently, oxygen level in the water body getting depleted, thereby greatly affecting the survival of aquatic organisms as well as making the water unsuitable for drinking or for any other use.

Furthermore, the heavy metals analysis in this study, that showed the value of lead in the three wastewater samples being within the range 1.74 mg/L and 7.43 mg/L were all above the international (World Health Organization) permissible limit of less than 1.0 mg/L (WHO, 2006). Similarly, the values of the dissolve zinc (4.5 mg/L) in the wastewater samples of the private hospital, 4.15 mg/L of the untreated tertiary hospital and 3.55 mg/L of the treated tertiary hospital were all above the permissible limit of less than 1.0 mg/L (WHO, 2006).

In addition, the Gram-negative bacteria isolated from the present study were similar to the ones previously reported from both clinical samples and hospital wastewater (Mansouri and Abbasi, 2010; Egbule, 2016). The high resistance of the isolates observed to tetracycline (58%) in this present study may be an indication that the drug might have been misused over a long period of time as previously noted (Newman *et al.*, 2006). However, this observation was not in agreement with a higher resistance (76.5%) to the antibiotics previously reported from Iran (Mansouri and Abbasi, 2010). Similarly, the observation from this study that 40% of all the isolates showed resistance to ceftazidime is lower compared to the 73.5% resistance to this antibiotic as well as resistance of 67% and 24% to amoxicillin/clavulanate and ciprofloxacin respectively is also lower compared to the 89% and 40.4% resistance to these antibiotics as previously reported from another study in Iran (Mansouri and Abbasi, 2010). Likewise, resistance of the Gram negative bacteria to ciprofloxacin (24%), ceftazidime (80.8%) and cefotaxime (44%) were comparably lower compared to the 54.8% (ciprofloxacin), 80.8% (ceftazidime) and 78.1% (cefotaxime) reported by Dalela (2012) in India.

The disparity in the observed resistance in the present study compared to the other studies may be due to the difference in the studied samples. While the present study was on hospital wastewater samples, the latter studies were on clinical isolates. Moreover, resistance to ceftazidime and cefotaxime as observed in this study may be due to the acquisition or possession of resistant gene against the antibiotics. While the observed resistance of the isolates to Amoxicillin/clavulanate, Streptomycin, chloramphenicol, meropenem, and ciprofloxacin, might be as a result of excessive use of these antibiotics in the treatment of bacterial infections through which the isolates might have received antibiotic pressure in the hospital environment during treatment. This is because, observation from previous studies had made it known that wastewater from hospitals is an environment that is highly selective and may serve as one of the contributing factors to the development of high resistant bacteria that are being discharged into the environment (Yang *et al.* (2009). Furthermore, the 37% of the isolates that were multidrug resistant (resistant to two or more classes of antibiotics), might be due to the acquisition of resistance genes through genetic exchange, mutation and physiological mechanisms such as possession of specific proteins and efflux pump as previously reported (Hermansson *et al.*, 1987; Herwig *et al.*, 1997).

However, a bit lower resistance (24%) exhibited by the isolates obtained from the wastewater samples of the hospitals in this present study to ciprofloxacin is similar to the findings of Brooks *et al.* (2004) that showed ciprofloxacin having high antimicrobial activity against enterobacteriaceae, including those that showed resistance to the third generation cephalosporins. Decreased resistance observed to meropenem can be attributed to its restricted use because it is extremely expensive and also a parenteral drug, unlike most drugs that have been abused (Sridhar, 2012).

In addition, the observation from the present study from which out of the ESBL-producing bacteria obtained from the wastewater samples, the predominant genera were *Klebsiella* and *Escherichia* is similar to the previous reports from studies on clinical isolates in which these two genera were found to be the dominant ESBL-producing bacteria (Umadevi *et al.*, 2011; Dalela, 2012). *Klebsiella* species being one of the dominant bacteria that produce ESBL is of public health concern because infections caused by this bacterium particularly respiratory tract infections are common due to the contagious nature and also the presence of capsules that gives some level of protection to the cells thereby leading to the resistance of the organism to conditions that are not favourable (Paterson and Bonomo, 2005). The observation from the present study in which the *Proteus* spp. and *Enterobacter* spp. as well as *Serratia* spp. being among the ESBL producers corroborates the assertion that the prevalence of ESBLs is increasing rapidly and have become widespread in hospitals as well as community settings (Pfaller and Segretti, 2006; Shakil *et al.*, 2010; Falodun *et al.*, 2018).

Furthermore, observation from this study showed that resistance of the ESBL isolates to the fluoroquinolones were high and this is similar to the outcome of another study from Tobago in which a significant high rate of resistance of *K. pneumonia* isolates obtained from clinical samples to the fluoroquinolones was reported. Hence, it was suggested that these antimicrobial agents when used against infections must be with caution (Akpaka and Swanston, 2008). Moreover, the susceptibility results showed that meropenem was the only antibiotic that was observed to be effective *in vitro* against the ESBL producing enteric isolates in this study as only 9% of the isolates showed resistance to the drug. Although, resistance of the ESBL producing bacteria to the carbapem (meropenem) in this study was low, it is however not in agreement with a study from which all the *E. coli* and *K. pneumoniae* that produced ESBL were found to be susceptible meropenem and imipenem (Akpaka and Swanston, 2008). More so, the observed resistance in this study was higher than the range of 1% to 3% resistance to imipenem that was recently reported on determination of water quality and ESBL detection of samples from three river water in Ibadan (Falodun *et al.*, 2018).

Carbapenems (such as meropenem) have been found effective in the treatment of ESBL-associated infections and are regarded as the antibiotics of last resort. However, there is need for continued surveillance and judicious use of these antibiotics as reports have been documented on the occurrence of carbapenem resistant *Escherichia coli*, *Klebsiella pneumonia* and *Klebsiella oxytoca* (Kuzucu *et al.*, 2011).

Furthermore, 60% of the ESBL-producing enteric bacteria in this study that showed resistance to Amoxicillin/clavulanate, 43% to ceftazidime and 40% to cefotaxime are not in agreement with the outcome of another study carried out on *Pseudomonas* spp. isolated from surface water in which all the isolates were reported to be completely resistant to the third generation cephalosporins (Nasreen *et al.*, 2015). However, a high percentage of the ESBL-producing isolates that exhibited resistance to non-beta lactam antibiotics such as streptomycin (80%), tetracycline (65.7%) and sulfamethoxazole/Trimethoprim (85.7%) is comparably similar to the report of a previous study in which 78.6% and 84.4% of the ESBL producing bacteria were found to be resistant to tetracycline and sulfamethoxazole/Trimethoprim respectively (Maina *et al.*, 2013). The low resistance of the isolates observed in this study to ciprofloxacin is reflective of the fact that has been established in several studies that ciprofloxacin is effective in the treatment of infections caused by ESBL-producing. This is also in agreement with the report of another study in which 83% of the ESBL bacteria isolated were found to be susceptible to ciprofloxacin (Maina *et al.*, 2013).

5. CONCLUSION

The findings from this study indicate the importance of safe disposal and better hospital wastewater treatment strategies to check the spread of antibiotic resistant pathogenic bacteria in the environment as these may pose public health challenge. ESBL-producing organisms are multidrug-resistant pathogens that are increasing rapidly and have become a major problem which makes it essential to report ESBL production along with the routine sensitivity reporting, which will help the clinicians in prescribing proper antibiotics. It is also very imperative that steps should be taken instantly to enforce hospital wastewater treatment before being discharged into the receiving water bodies in the environment. More so, efficiency of the wastewater treatment facility should be monitored on a regular basis.

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