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Full Length Research Paper

Antibiotic susceptibility patterns of bacterial isolates from hospitalized patients in Abakaliki

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Abstract

This study was designed to determine the antimicrobial sensitivity patterns of bacterial isolates from patients admitted in Federal Teaching Hospital (FETHA) Abakaliki in the year 2011-2012. Eight- five bacterial isolates were isolated from various clinical specimen namely urine (38), wound swab (17), blood (11), sputum (8), high vaginal swab (4), stool (4), Eye swab (3) and were analyzed using standard microbiology technique in the Department of Applied Microbiology Laboratory unit of Ebonyi State University Abakaliki. Standard microbiology techniques employed were culturing of the clinical specimen onto blood agar, MacConkey and Mannitol salt agar. Organisms were identified by their colonial morphology, Gram staining and appropriate biochemical test. Antimicrobial susceptibility patterns of the bacterial isolates recovered from different clinical specimen were determined using modified Kirby and Bauer method with the following antibiotics; ampicillin, sulfametoxazole/trimethoprim, ceftazidime, ciprofloxacin, amikacin, amoxicillin/clavulanic acid, gentamicin, tobramycin, ofloxacin, clindamycin, oxacillin, erythromycin and cefotazime. Among the 289 different clinical specimens collected 85 organisms were isolated which includes; five Gram negatives (*Escherichia coli* (25), *Klebsiella* spp. (24), *Proteus* spp. (7) *Citrobacter* spp. (6) and *Pseudomonas* spp. (6) and two Gram-positive organisms *Staphylococcus aureus* (15), and *Streptococcus* spp. (2). Antibiotics susceptibility studies showed that Gram-negative and Gram-positive organisms were all susceptible to amikacin. Individually *E. coli* and *Klebsiella* spp. were also susceptible to ciprofloxacin, gentamicin, ofloxacin and nitrofuraintoin, *Pseudomonas* and *Proteus* spp. were susceptible to ciprofloxacin and gentamicin while *Citrobacter* spp. to amoxicillin/clavulanic acid. Strains of *Staph. aureus* and *Streptococcus* spp were resistant to oxacillin. *Streptococcus* spp. were susceptible to ciprofloxacin, ceftazidime but resistant to amoxicillin/clavulanic acid, ceftazidime and ampicillin. In conclusion resistance observed in the present study among some commonly used antibiotics pose a serious problem although high susceptibility was observed with amikacin ciprofloxacin and gentamicin etc. Therefore, we suggest the need for continuous surveillance of sensitivity patterns of antimicrobial agents in our hospital to enable us know the trend of this problem.

Keywords: Bacteria, antibiotic susceptibility and clinical specimen.

INTRODUCTION

Infections with drug resistance organisms remain an important problem in clinical practice that is difficult to solve. Antimicrobial chemotherapy made remarkable advances, resulting in the overly optimistic view that infectious disease wound be conquered in the near future. However, in reality emerging and re-emerging infectious diseases have left us facing a counter charge from infections (Tomoo and Keizo, 2009).

It is said that evolution of bacteria towards resistance to antimicrobial drugs, including multi-drug resistance is

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unavoidable because it represents a particular aspect of the general evolution of bacterial that is un-stoppable (Courvalin, 2005). Antibiotic resistance emerges commonly when patients are treated with empiric antimicrobial drugs and to overcome these difficulties and to improve the outcome of serious infections in our institution, monitoring of resistance patterns in the hospital is needed (El- Azizi et al., 2005).

The present study was designed to determine the antibiotic sensitivity pattern of bacterial isolated from hospitalized patients in FEUTA in the year 2011-2012.

MATERIALS AND METHODS

Collection of clinical samples

Clinical samples (n=289) were collected from different hospital wards of a Federal Teaching Hospital Abakaliki in Ebonyi State from September 2011- November 2012. The sample includes; urine, stool, sputum, eye swab, wound swab and high vaginal swab. They were analyzed at the Applied Microbiology laboratory unit of Ebonyi State University Abakaliki by culture, Gram staining, biochemical test and antibiotics sensitivity testing (Konman et al., 1997).

Analysis of clinical samples

Clinical samples were inoculated onto blood agar, Mannitol salt agar and MacConkey agar plates and incubated aerobically at 37^0C for 18-24hrs. After incubation bacterial growth was observed for colony appearance and was Gram stained and subjected to further biochemical tests (Konman et al., 1997). Results were interpreted according to the guidelines of the Clinical and Laboratory Standards Institute (CLSI, 2006).

Antibiotic sensitivity studies

Antibiotic susceptibility of bacteria isolates to various classes of antibiotics was determined by the Kirby and Bauer disc diffusion method according to CLSI recommendations (CLSI, 2006). The following antibiotics were used against Gram–negative organisms; ampicillin (10µg) sulfametoxazole/trimethoprim (30µg), nitrofurantoin (15µg), cefotaxime (30µg), ciprofloxacin (10µg), amikacin (10µg), gentamicin (10µg) tobramycin (5µg), ofloxacin (10µg), ceftazidime (30µg), amoxicillin/clavulanic acid (30µg). While for Gram-positive organism additional antibiotics used different from the ones mentioned above are clindamycin (5µg), oxacillin (1µg) and erythromycin (10µg). Briefly, a sterile Muller-Hinton agar was prepared according to manufactures instruction and a 0.5 MacFarland equivalent standard of the test organisms were inoculated on the surface of the agar. Test antibiotics listed above were aseptically placed on the inoculated plates and incubated at 37^0C for 18-24 hrs. Inhibition zone diameter was measured and the organisms were identified as either resistance or susceptible based on CLSI standard (CLSI, 2007). Control strain was used to check for the quality of disc and reagents.

RESULTS

Our findings showed that out of 289 different clinical samples collected from patients admitted in FETHA and analyzed between September 2011-November 2012; 85(29.4%) bacteria were isolated. A total of 66 urine samples were collected and 38 (57.5%) of bacteria were isolated namely *kleb.* Spp. 15(39.4%), *E. coli* 10(26.3%), *Proteus* spp. 7(18.4%) and *Citrobacter* spp 6(15.7%). Thirty-Eighty wound swabs were collected and analyzed; 17(44.7%) bacteria were isolated which includes 15(88.2%) of *Staph aureus,* and 2(11.7%) of *Kleb,* spp. Of the 40 blood sample collected and analyzed; 11(27.5%) *E. coli* were isolated. A total of 52 sputum samples were collected and 6(75%) *Pseudomonas* spp and 2(25%) *Strep* spp. was isolated. Twenty-six high vaginal swab samples were collected and 4(15.3%) of *Kleb* spp. were isolated. Thirty-nine stool samples were collected and 4(10.2%) of *E. coli* were isolated while of the 28 eyes swab samples collected 3(10.7%) *Kleb* spp. was isolated.

Escherchia coli and *Klebsiella* spp. were the most frequently isolated bacteria pathogen from clinical samples. Table 1.

Antibiotic susceptibility studies showed that Gram-negative organisms were less susceptible to the antibiotic compared to the Gram-positive organism. Strains of *E. coli* were susceptible to six antibiotics namely; nitrofurantoin (72%), ciprofloxacin (81%), amikacin (95%), gentamicin (60%), tobramycin (68%), and ceftazidime (63%) Figure 1. Such was observed with *Klebsiella* spp. but the percentage of susceptibility was less Figure 2. *Pseudomonas* spp. were only susceptible to four antibiotic namely; cefotaxime 66% ciprofloxacin 83%, amikacin and gentamicin 100% Figure 3. *Proteus* spp. were 71% susceptible to ciprofloxacin, amikacin, gentamicin, and ofloxacin Figure 4. *Citrobacter* spp. were 83% susceptible to ampicillin, amikacin, amoxicillin/clavulanic acid Figure 5. *Staph. aureus* and and *Streptococcus* spp. were susceptible to all the antibiotics except with oxacillin and ceftazidime Figure 6 and 7. Increased resistance was also observed in *Streptococcus* spp. with ampicillin.

DISCUSSIONS

Antimicrobial chemotherapy has conferred huge benefits

Table 1. Organisms isolated from different clinical samples (n = 85)

Specimen	*Staph. aureus*	*E. coli*	*Klebsiella* Species	*Pseudomonas* Species	*Citrobacter* Species	*Proteus* Species	*Streptococcus* Species
Urine	-	10	15	-	6	7	-
Wound swab	15	-	2	-	-	-	-
Blood	-	11	-	-	-	-	-
Sputum	-	-	-	6	-	-	2
HVS	-	-	4	-	-	-	-
Stool	-	4	-	-	-	-	-
Eye swab	-	-	3	-	-	-	-
Total	15	25	24	6	6	7	2

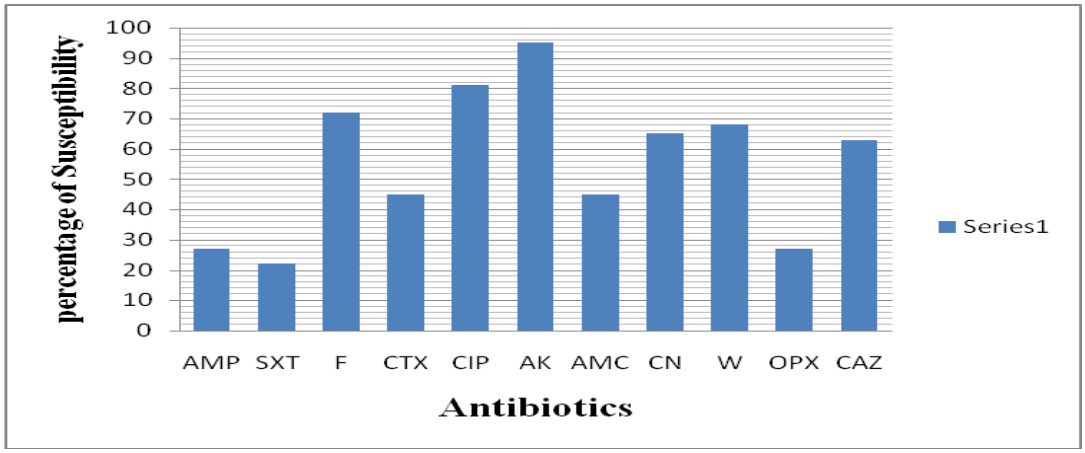

Figure 1. Percentage of antibiotics susceptibility of *E. coli* to antibiotics, Keys: AMP- ampicillin, SXT- sulphamethoxazole/trimethoprim, F- nitrofurainton, CTX-cefotaxime, CIP- ciprofloxacin, AK- amikacin, AMC- amoxicillin/clavulanic acid, CN- gentamicin, W- tobramycin, OFX- ofloxacin, CAZ- ceftazidime

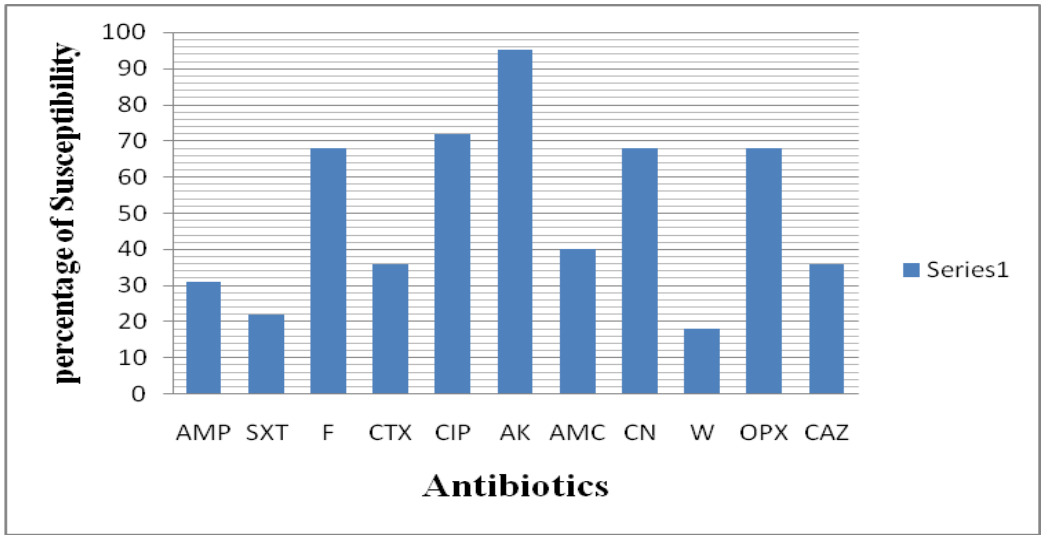

Figure 2. Percentage of antibiotics susceptibility of *Klebsiella* spp. to antibiotics
Keys: AMP- ampicillin, SXT- sulphamethoxazole/trimethoprim, F- nitrofurainton, CTX-cefotaxime CIP- ciprofloxacin, AK- amikacin, AMC- amoxicillin/clavulanic acid, CN- gentamicin, W- tobramycin, OFX- ofloxacin, CAZ- ceftazidime

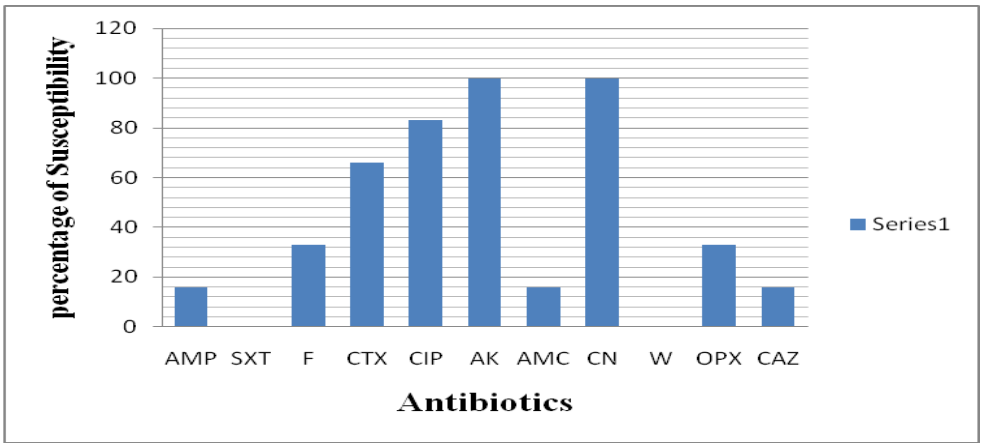

Figure 3. Percentage of antibiotics susceptibility of *Pseudomonas* spp. to antibiotics
Keys: AMP- ampicillin, SXT- sulphamethoxazole/trimethoprim, F- nitrofurainton CTX-cefotaxime, CIP- ciprofloxacin, AK- amikacin, AMC- amoxicillin/clavulanic acid CN- gentamicin, W- tobramycin, OFX- ofloxacin, CAZ- ceftazidime

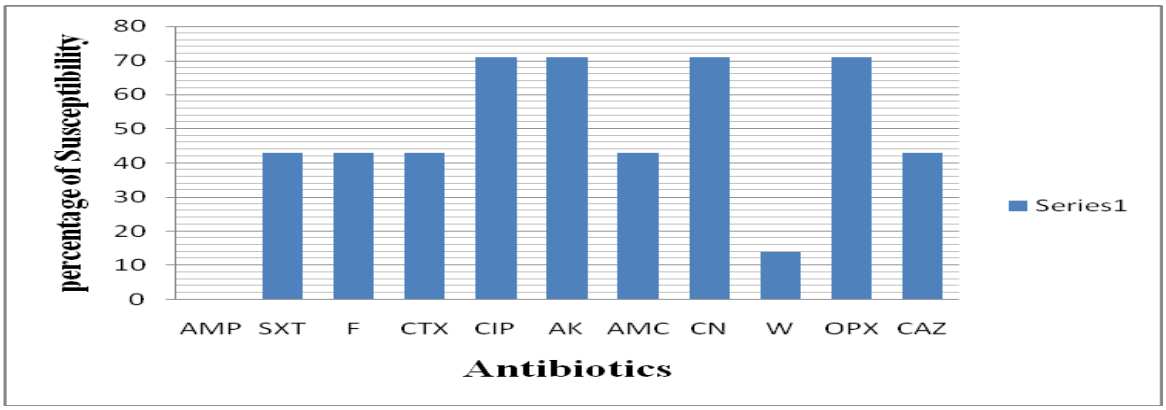

Figure 4. Percentage of antibiotics susceptibility of *Proteus* spp. to antibiotics
Keys: AMP- ampicillin, SXT- sulphamethoxazole/trimethoprim, F- nitrofurainton, CTX-cefotaxime CIP- ciprofloxacin, AK- amikacin, AMC- amoxicillin/clavulanic acid CN- gentamicin, W- tobramycin, OFX- ofloxacin, CAZ- ceftazidime

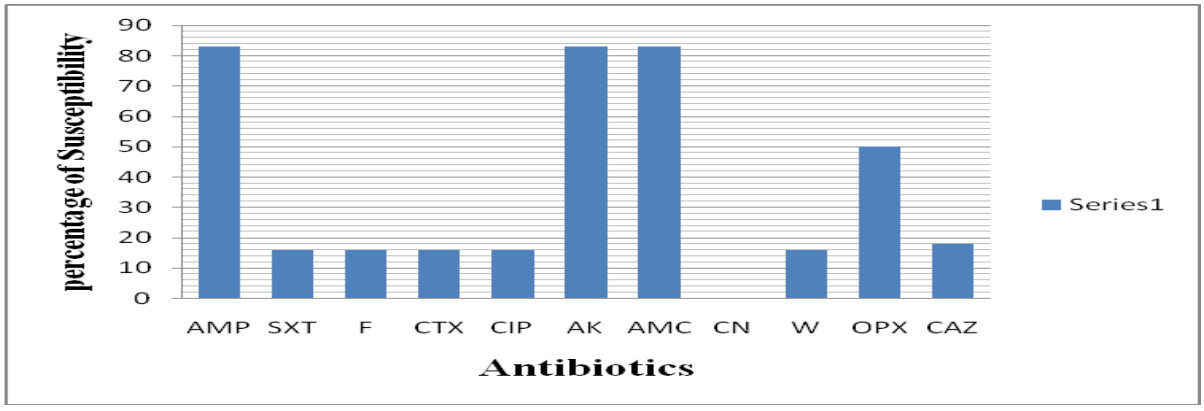

Figure 5. Percentage of antibiotics susceptibility of *Citrobacter* spp. to antibiotics
Keys: AMP- ampicillin, SXT- sulphamethoxazole/trimethoprim, F- nitrofurainton CTX-cefotaxime, CIP- ciprofloxacin, AK- amikacin, AMC- amoxicillin/clavulanic acid CN- gentamicin, W- tobramycin, OFX- ofloxacin, CAZ- ceftazidime

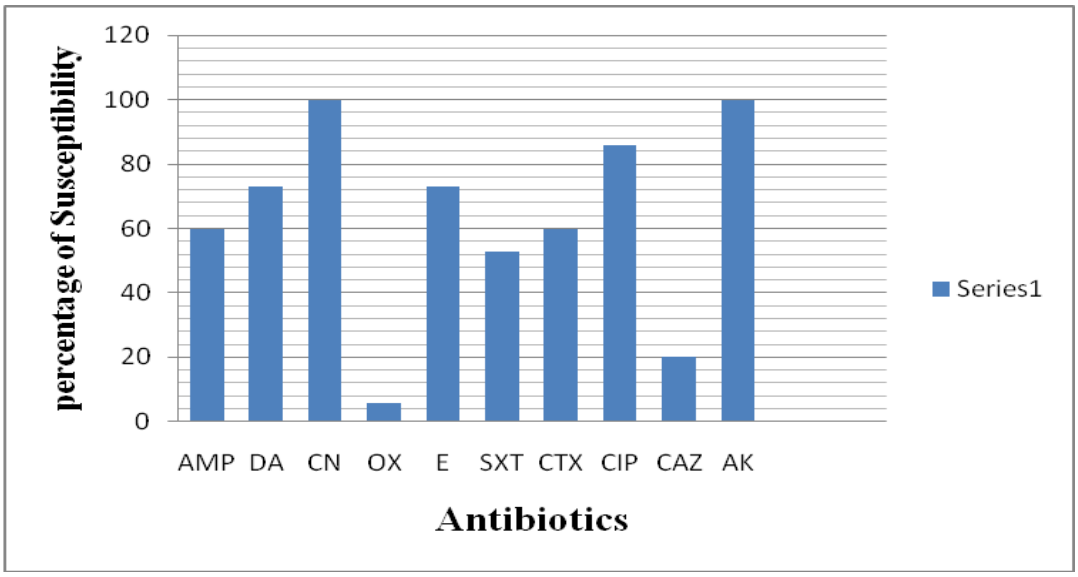

Figure 6. Percentage of antibiotics susceptibility of *Staph. aureus* to antibiotics
Keys: AMP- ampicillin, SXT- sulphamethoxazole/trimethoprim, DA-clindamycin
CTX-cefotaxime, CIP- ciprofloxacin, AK- amikacin, E- erythromycin, CN- gentamicin
OX- oxacillin, CAZ- ceftazidime

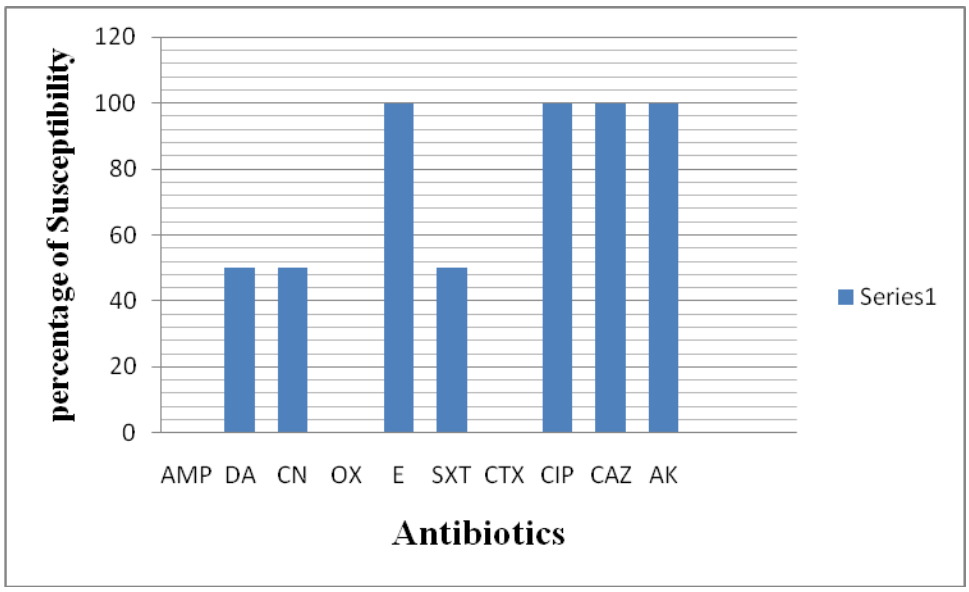

Figure 7. Percentage of antibiotics susceptibility of *Streptococcus* spp. to antibiotics
Keys: AMP- ampicillin, SXT- sulphamethoxazole/trimethoprim, DA-clindamycin
CTX-cefotaxime, CIP- ciprofloxacin, AK- amikacin, E- erythromycin, CN- gentamicin
OX- oxacillin, CAZ- ceftazidime

to human health as a variety of microorganisms that were elucidated to cause infectious diseases are controlled by the proper use of antibiotics. In the 20th century the discovery of antibiotic was viewed that all infectious disease will be conquered in the near future (Power, 2004). However, in response to the development of antimicrobial agents, microorganisms, that have acquired resistance to drugs through a variety of mechanisms have emerged and continue to plague human beings. In Nigeria, infectious diseases caused by drug resistant bacteria are one of the most important problems in daily clinical practices as observed in the present study.

Our findings showed that out of 85 bacteria isolated, *E. coli* (25) and *Kleb.* spp (24) are the most frequently isolated organism, followed by *Staph. aureus* (15), *Proteus* spp. (7), *Citrobacter* and *Pseudomonas* spp (6)

and *Streptococcus* spp. (2). *Kleb.* spp was the commonest Gram-negative organisms from urine while *Staph. aureus* was the most commonest bacteria from wound swab and also the most frequently isolated Gram-positive organisms. This finding is not in line with the work of Iffat et al., 2011; he showed that *E. coli* was the most frequently isolated organism from urine sample. Strains of *E. coli* showed high resistance to ampicillin (73%), sulphametoxazole/trimethoprim (78%), cefotaxime and amoxicillin/ clavulanic acid (55%). A study conducted by Nwadioha et al., 2010 also reported that *E. coli* were most frequently isolated from urine samples of HIV and non HIV patients and were highly susceptible to ceftazidime, ciprofloxacin and amoxicillin/clavulanic acid but were resistant to ampicillin, chloramphenicol and co-trimoxazole; this findings can be correlated with ours but not exactly. *Klebsiella* spp also showed marked resistance to ampicillin (69%), sulphametoxazole/trimethoprim (70%), ceftazidime (64%) and amoxicillin/clavulanic acid (40%). Sarathbadu et al., 2012 in his study reported that all *Klebsiella* species isolated from urine, pus and sputum were highly susceptible to amikacin. On the average this shows that more than 50% of the organism is resistance to the antibiotics. Similarly *Pseudomonas* spp. and *Proteus* spp. are resistance to ampicillin, sulphametoxazole/trimethoprim and ceftazidime while *Citrobacter* spp. is the only Gram-negative organism susceptible to ampicillin but were highly resistance to sulphametoxazole/trimethoprim, nitrofurantoin, cefotaxime, ciprofloxacin, gentamicin, tobramycin and ceftazidime. This pattern is comparable to other studies carried out in some other parts of the world (Khameneh and Afshar, 2009; Rahman et al., 2002; Anguzu and Olila, 2007). Our study showed a high cefotaxime, ceftazidine and amoxicillin/clavulanic acid resistance especially among *Klebsiella, Pseudomonas, Proteus* and *Citrobacter* spp. except for *E. coli* that is susceptible to ceftazidime. Figure 1. However, *Pseudomonas* was susceptible to cefotaxime, gentamicin, amikacin, and ciprofloxacin. Similar findings regarding drug resistance patterns of *Klebsiella*, *E. coli* and *Pseudomonas* have been reported by other researchers (Kamberovic et al., 2006; Kaul et al., 2007; Loureiro et al., 2002; Hasan et al., 2007) All the Gram-negative organisms were susceptible to ciprofloxacin and amikacin, only *E. coli* and *Kleb.* spp was susceptible to nitrofuraintoin. Susceptibility of *Kleb.* and *E. coli* to nitrofuraintoin is expected because most of the isolates were from urine and nitrofuraintoin is known to be active against urine-isolated pathogen. Shyamala et al., 2012 also reported high susceptibility of Gram-negative bacteria to amikacin in 2007. Alshara in 2011 also reported effectiveness of amikacin and cefotaxime against *E. coli* and other Gram negatives but our finding showed that cefotaxime was less effective except against *Pseudomonas* spp. Anguzu and Olila in 2007 reported that most of the Gram-negative bacteria isolated in their study were resistant to ampicillin, this findings can be compared with ours. Nnebe-agumadu et al., 2011 reported that all Gram-negative organisms isolated from otitis media patients were susceptible to ciprofloxacin and gentamicin but highly resistant to amoxicillin/clavulanic acid. Amoxicillin/clavulanic acid is among the last drug of choice against gram-negative organism and the sudden resistance to it by pathogenic bacteria of clinical important is a serious treat to the clinicians and should be seriously monitored. The study showed an alarming resistance of Gram-negative organism to beta lactam antibiotics which is a serious problem that should be looked into. This beta lactam antibiotics resistance could be by vertical as well as horizontal transfer of resistance genes.

Staphylococcus aureus resistance to antibiotics is a worldwide phenomenon especially when it involves methicillin resistance *Staphylococcus aureus* (MRSA). S*taph. aureus* isolates from wound swab in the present study was susceptible to all tested antibiotics except oxacillin. *Strep.* spp was also resistant to oxacillin. Although we did not test *Staph aureus* further for MRSA but their resistance to oxacillin may suggest that they are MRSA producers. Nwadioha et al., 2010 reported high susceptibility of *Staph aureus* to ciprofloxacin and gentamicin, this correlates with our findings too.

The results of the present study highlights the alarming increased resistance of bacteria to majority of the antibiotic used in this study. It is clear that the use of antimicrobial agents resulted in the selection of resistant bacteria. Therefore the proper use of commonly available antimicrobial agents as well as efforts to minimizing the spread of resistant bacteria through appropriate infection control would be quite important and may represent a first step in resolving the issue of resistant microorganisms. Moreover, the study indicates that both Gram-negative and Gram-positive organisms were susceptibility to amikacin and gentamicin compared to other antibiotic tested and therefore they may be considered the drugs of choice for the treatment of nosocomial infections in FETHA.

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