

Phenotypic identification and antibiogram profile of *Citrobacter* species.

Oludare Temitope Osuntokun^{1*}, Taiye Anangwureyi Jemilaiye¹, Adisa O Thonda²

¹Department of Microbiology, Adekunle Ajasin University, Akungba Akoko

²Department of Biological Science, Kings University, Ode Omu, Nigeria

Abstract

The administration of antimicrobial agents on patients had a major impact on the rate of survival from infections. However, the evolving and the changing patterns of antimicrobial resistance posed a threat and caused a demand for review of the existing antibiotic policy or a demand for new antibacterial agents. This study was designed to determine the phenotypic identification in *Citrobacter* species and Antibiogram profile of *Citrobacter* species isolated from University College Hospital (UCH), Ibadan. Twenty clinical isolates of clinical specimens were recovered from patients using the laboratory services of UCH between December 2016 and March 2017. Preliminary confirmatory test to identify the isolates as *Citrobacter* was then performed using Oxidase strips and Microbact™ 12A and 24E (Oxoid, UK) Identification Kits. Antibiotic susceptibility tests (Antibiogram profile) was carried out on the identified *Citrobacter* isolates using some common antibiotics and Minimal Inhibitory Concentrations (MICs) of Piperacillin/Tazobactam tested against the *Citrobacter* spp were determined. Ofloxacin was active against all the four *Citrobacter* isolates (100%), with diameter zone of inhibition ranging from 18 mm to 25 mm. Closely followed is Piperacillin/Tazobactam which was active against three out of the four isolates (75%) with diameter zone of inhibition ranging from 26 mm to 40 mm. Two of the isolates (50%) showed susceptibility to Co-trimoxazole (24 mm and 30 mm), and Imipenem (18 mm and 35 mm). All the isolates were, however, resistant to Cephalosporins (Ceftazidime, Cefotaxime and Cefoxitin), Cloxacillin, Oxacillin, Piperacillin, Meropenem, Nystatin and combination of Ampicillin+Clavulanic acid. MIC values were quite high ranging from 100 µg/mL to 110 µg/mL. This study highlighted the prevalence of *Citrobacter* species among clinical samples in southern Nigeria and multi-drug resistant nature of the isolated *Citrobacter* species.

Keywords: Phenotypic identification, Antibiogram profile, *Citrobacter* species

Accepted on December 30, 2017

Introduction

The administration of antimicrobial agents on patients had a major impact on the rate of survival from infections. However, the evolving and the changing patterns of antimicrobial resistance posed a threat and caused a demand for review of the existing antibiotic policy or a demand for new antibacterial agents. Bacteria antimicrobial resistance is a worldwide problem that is exacerbated by the diminishing number of new antimicrobial drugs in the pharmaceutical pipeline [1]. This is an emerging public health problem, especially in hospitals of the developing and the newly industrialized countries of Africa, Asia and the Pacific [2,3].

In developing countries such as Nigeria, the spread of antimicrobial resistance is a matter of concern as it compromises the management of infectious diseases, the leading causes of morbidity and death in these countries [4,5]. Much of the current discourse on infectious disease and drug resistance as it affects Sub-Saharan Africa is limited to the pressing problems associated with emerging and re-emerging resistant organisms. Hence, the available therapeutic options for antibiotics-resistant organisms are severely limited, as these organisms frequently display a multidrug-resistant phenotype.

The excessive use of antimicrobials produces selective pressure

for resistance. Also, empirical treatment with ineffective antibiotics prescribed by physicians and poor patient adherence to antibiotic regimens could potentially lead to mutation and drug resistance [6]. Furthermore, the routine use of antibiotics in animal foods to promote their growth is another aspect that may enhance selective pressure for resistance [7,8]. The rate of emergence of antibiotic resistance in the isolates of *Enterobacteriaceae* and in other Gram-negative bacilli in many parts of the world is very alarming and this is a major threat to the successful treatment of infections in hospitals [9]. *Citrobacter* species are aerobic, motile, Gram negative bacilli of *Enterobacteriaceae*. *Citrobacter* species have been reported to cause a range of infections which include urinary tract infections, wound infections (surgical site infections), respiratory infections, middle ear infections, infections of meninges, osteomyelitis, peritonitis, endocarditis, and bacteremia [10].

The leading three species causing human infections are *C. freundii*, *C. koseri*, and *C. braakii*. Those predispose to *Citrobacter* infections are the neonates, who may develop sepsis and meningitis (usually less than 2 months of age), and the debilitated aged (≥ 65 years) or immunocompromised patients. Many *Citrobacter* infections are nosocomially acquired, however, they can also be community acquired. In the hospital settings, *Citrobacter* spp. might account for 3-6%

of all *Enterobacteriaceae* causing nosocomial infection. The significance of nosocomial infection lies not only in its ability to substantially alter morbidity and mortality statistics, but also in its economic implications. Nosocomial infections pose a problem of enormous magnitude globally as hospital localities have proven favourable in transmitting *Citrobacter* infection due to existing suitable pathogen-host-environment relationship [5].

The *Citrobacter* species are emerging as important nosocomial pathogens with antibiotic-resistant strains, though less frequent [11]. Patient with invasive devices are more prone for invasive *Citrobacter* infections and are associated with a high mortality rate with majority of patients perishing or wilting to *Citrobacter* bacteremia. The high mortality rate associated with *Citrobacter* infections may be due in part to ineffective empirical antibiotic therapy.

Antibiotic resistance has increased significantly among species of *Citrobacter*, making infections caused by these pathogens are difficult to treat [12]. *Citrobacter* species are often resistant to the routinely used antibiotics, like the Penicillins, Cephalosporins, Aminoglycosides and Tetracyclines. Cephalosporins form the major component of empirical antibiotic treatment especially in teaching hospitals. Indiscriminate use of cephalosporins, the 3rd generation cephalosporins, has contributed to the evolution of extended spectrum beta lactamases (ESBLs) production and Multiple Drug Resistance (MDR) [13]. MDR strains are becoming frequent in *Citrobacter* spp, where clinical isolates

resistant to a wide range of antibiotics are increasingly being reported [14,15].

Materials and Methods

Equipment and materials used

The equipment used for this study include: Autoclave (LDZX-30FBS, England), Incubator (Genlab, UK), Oven (Gulfex Medical and Scientific, England), Refrigerator (Haier Thermocool, China), Weighing balance, Hot plate, Bunsen burner, and Laminar flow hood.

The materials used for this study include: Test tubes, Beakers, Conical flasks, Inoculating loop, Sterile needles and syringes, McCartney bottles, Hand gloves, non-absorbent cotton wool, Aluminum foil, Alcohol (95%), Potassium hypochlorite solution, Paper tape, Petri dishes, Universal sterile bottles, Sterile distilled water, Mueller Hinton agar (Oxoid, UK), Microbact™ 12A and 24E identification Kits (Oxoid, UK), and some selected Antibiotics discs (Oxoid, UK) (Figure 1).

Sample collection and identification of bacteria isolates

Twenty (20) non-duplicate non-repetitive presumptive clinical isolates of clinical specimens were recovered from patients using the laboratory services of University College Hospital (UCH), Ibadan, between December, 2016 and March, 2017. The isolates were then transported using prepared Nutrient agar slant to the Microbiology laboratory of Adekunle Ajasin University for further microbiological analysis. The clinical

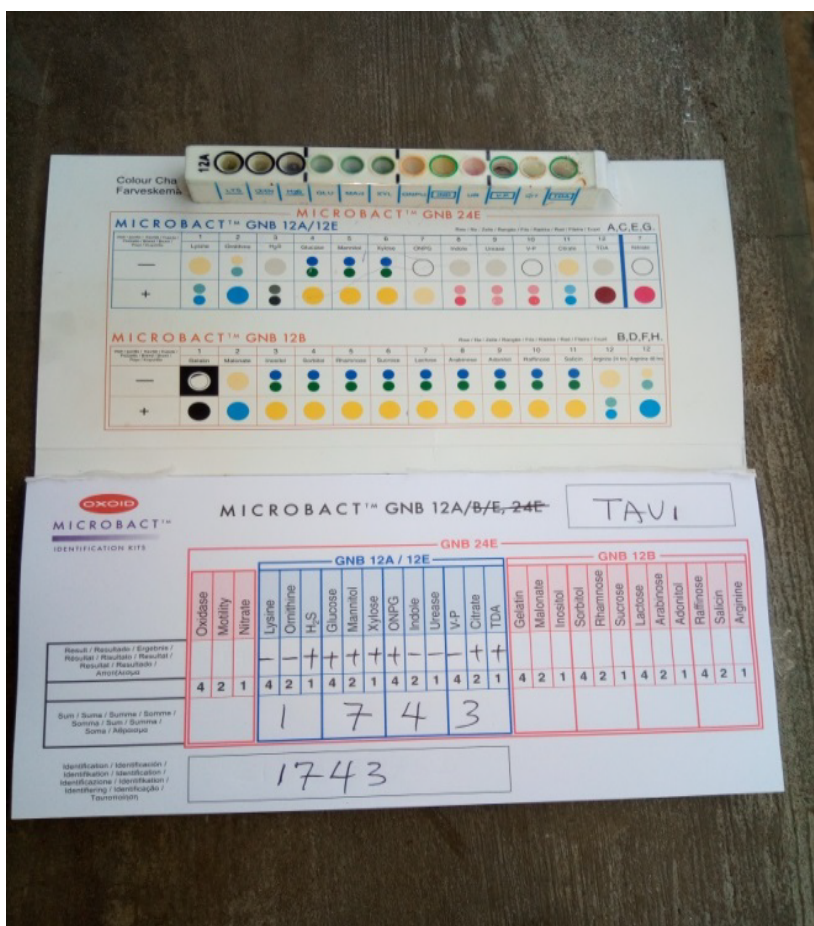


Figure 1. Used Microbact™ 12A test strip with its report form.

isolates were recovered from the following specimens: sputum, blood, urine and stool. Preliminary confirmatory test to identify the isolates as *Citrobacter* was then performed using Oxidase strips and Microbact™ 12A and 24E (Oxoid,UK) identification kits according to the manufacturer’s instruction. The recovered isolates were then prepared and sub cultured into nutrient agar slants and refrigerated at -4°C in readiness for further use (Figure 2).

Antibiogram profile

Antibiotic susceptibility tests were performed to determine the phenotypic resistance patterns of the identified *Citrobacter* spp isolates to some common antibiotics available in Nigeria. This test was carried out by Kirby-Bauer’s disc diffusion method on Mueller-Hinton agar (CM337-Oxoid,

UK) using 10⁴ normal saline dilution of a 24 hours broth culture (Figure 3). The results were interpreted according to the Clinical Laboratory Standard Institute (CLSI, 2016) guidelines and the diameter of zones of inhibition measured and recorded in millimeter. The choice of antibiotics was based on their mechanism of action on bacteria and also on the principle of the commonly prescribed antibiotics in Nigeria’s Tertiary Health Institutions. The standard antibiotic discs (Oxoid, UK) used with their concentrations include Ceftazidime (30 µg), Cefoxitin (30 µg), Cefotaxime (30 µg), Amoxycillin+Clavullanic acid (30 µg), Cotrimoxazole (25 µg), Ofloxacin (5 µg), Imipenem (10 µg), Piperacillin/Tazobactam (110 µg), Cloxacillin (5 µg), Oxacillin (1 µg), Piperacillin (100 µg), Meropenem (10 µg), Vancomycin (30 µg), Teicoplanin (30 µg) and Nystatin (100 IU) (Figure 3).



Figure 2. Used Microbact™ 24E test strip with its report form.



Figure 3. Plate showing antibiogram profile of one of the Citrobacter isolate.

Screening for ESBL producers

The *Citrobacter* spp isolates being resistant to the cephalosporins were further processed for ESBL detection by the combined disc potentiation method according to the Clinical Laboratory Standard Institute (CLSI, 2016) guidelines. Discs of Cefotaxime (30 µg), Ceftazidime (30 µg) and Cefoxitin (30 µg) were each placed 20mm away from a combination disc of Amoxicillin+Clavulanic acid (30 µg) on a Mueller-Hinton agar plate inoculated with a 24 hours broth culture of the isolates diluted to 10⁴ inoculum density with normal saline. The results were then read after 16-24 hours incubation.

Minimal inhibitory concentration (MIC)

Minimal inhibitory concentration (MIC) was carried out using the agar dilution method according to the Clinical Laboratory Standard Institute (CLSI, 2016). Various concentrations of Piperacillin/Tazobactam were tested against the clinical *Citrobacter* isolates that were susceptible to the antibiotic with a zone of diameter ≥ 18 mm. A 24 hour broth culture was inoculated into Mueller-Hinton agar plates containing varying concentration of Piperacillin/Tazobactam incubated aerobically

for 24 hour at 37°C under appropriate conditions.

Results

The distribution of the isolates among the collected clinical samples and profile of patients from whom samples were obtained is shown in Table 1. The distribution show that out of the 20 Gram negative isolates recovered, 4(20%) belonged to the *Citrobacter* species (*C. freundii*-3, *C. sakazakii*-1) with 50% of them recovered from urine sample. The antibiogram profile of the recovered isolates of *Citrobacter* spp to some selected antibiotics is represented in Table 2. Antibiogram of the study clearly demonstrated maximum sensitivity to Ofloxacin (100%), followed by Piperacillin/Tazobactam (75%) and Cotrimoxazole (50%).

Result of the screening for ESBL producers is shown in Table 3. Phenotypic resistance pattern of the isolates to glycopeptides is shown in Table 4. The result showed maximum resistance (100%) to Teicoplanin and Vancomycin. The Minimal Inhibitory Concentration (MIC) of Piperacillin/Tazobactam against the susceptible strains of the recovered *Citrobacter* spp is presented in Table 5.

Table 1. Distribution of isolates among clinical samples and profile of patients from whom samples were obtained.

S/No	Hospital Assigned Code	Clinical Sample	Sex of Patient	Age of Patient	Organism Isolated	Laboratory Assigned Code
1	UCH P1	Blood	----	----	<i>Proteus mirabilis</i>	Nil
2	UCH P2	Sputum	----	----	<i>Citrobacter freundii</i>	TAV 1
3	UCH P3	Ear swab	----	----	<i>Proteus mirabilis</i>	Nil
4	UCH P4	Blood	----	----	<i>Acinetobacter iwoffii</i>	Nil
5	UCH P5	Ear swab	----	----	<i>Citrobacter freundii</i>	TAV 2
6	UCH P6	Stool	----	----	<i>Salmonella marcescens</i>	Nil
7	UCH P7	Stool	-----	-----	<i>Klebsiella oxytoca</i>	Nil
8	UCH P8	Urine	----	----	<i>Citrobacter freundii</i>	TAV 3
9	UCH P9	Sputum	----	----	<i>Klbsiella pneumonia</i>	Nil
10	UCH P10	Blood	----	----	<i>Providentia stuartii</i>	Nil
11	UCH P11	Stool	Male	7 yrs	<i>Enterobacter agglomerans</i>	Nil
12	UCH P12	Urine	Female	Adult	<i>Acinetobacter iwoffii</i>	Nil
13	UCH P13	Urine	Female	37 yrs	<i>Hafnia alvei</i>	Nil
14	UCH P14	Urine	Male	27 yrs	<i>Citrobacter sakazakii</i>	TAV 4
15	UCH P15	Urine	Female	27 yrs	<i>Enterobacter agglomerans</i>	Nil
16	UCH P16	Urine	Female	27 yrs	<i>Acinetobacter iwoffii</i>	Nil
17	UCH P17	Stool	Female	22 yrs	<i>Tetmella ptyseos</i>	Nil
18	UCH P18	Urine	Female	21 yrs	<i>Yersinia pseudotuberculosis</i>	Nil
19	UCH P19	Urine	Female	22 yrs	<i>Salmonella liquefaciens</i>	Nil
20	UCH P20	Urine	Male	27 yrs	<i>Salmonella marcescens</i>	Nil

Table 2. Antibiogram profiles of citrobacter spp against common antibiotics (measured in mm) ± sem.

Antibiotic (Concentration In µg)	TAV 1	TAV 2	TAV 3	TAV 4
Ceftazidime (30 µg)	NI	40 ± 0.5	NI	NI
Cefoxitin (30 µg)	NI	NI	NI	NI
Cefotaxime (30 µg)	NI	NI	NI	NI
Cotrimoxazole (25 µg)	24 ± 0.3	NI	30 ± 0.2	NI
Ofloxacin (5 µg)	18 ± 0.1	20 ± 0.2	22 ± 0.2	25 ± 0.5
Imipenem (10 µg)	18 ± 0.2	NI	35 ± 0.5	NI
Piperacillin/Tazobactam (110 µg)	26 ± 0.5	40 ± 0.2	36 ± 0.3	NI
Cloxacillin (5 µg)	NI	NI	NI	NI
Oxacillin (1 µg)	NI	NI	NI	NI
Piperacillin (100 µg)	NI	NI	NI	NI
Amoxicillin+Clavulanic acid (30 µg)	NI	NI	NI	NI
Meropenem (10 µg)	NI	NI	NI	NI
Nystatin (100 iu)	NI	NI	NI	NI

Keys: NI=No inhibition, SEM=Standard error of magnitude.

Table 3. Phenotypic screening for ESBL producer among the *Citrobacter* isolates (measured in mm) \pm sem.

Isolates	Ceftazidime (30 μ g)	Cefotaxime (30 μ g)	Cefoxitin (30 μ g)	Amoxicillin+Clavulanic acid (30 μ g)
TAV 1	NI	NI	NI	NI
TAV 2	38 \pm 0.5	NI	NI	NI
TAV 3	NI	NI	NI	NI
TAV 4	NI	NI	NI	NI

Key: NI=NO Inhibition, SEM=Standard Error of Magnitude.

Table 4. Phenotypic resistance profile of *Citrobacter* isolates to glycopeptides.

Isolates	Teicoplanin (30 μ g)	Vancomycin (30 μ g)
TAV 1	13 \pm 0.5	NI
TAV 2	NI	NI
TAV 3	NI	NI
TAV 4	13 \pm 0.5	NI

Key: NI=No Inhibition

Table 5. MIC pattern of susceptible *Citrobacter* isolates to piperacillin/tazobactam.

Isolates	MIC values
TAV 1	110 μ g/mL
TAV 2	100 μ g/mL
TAV 3	110 μ g/MI

Discussion

This study was designed to determine the phenotypic resistance in *Citrobacter* species and Antibigram profile of *Citrobacter* species isolated from University College Hospital (UCH), Ibadan. The present study indicates that, of the 20 clinical isolates from the University College Hospital (UCH), Ibadan, 4 (20%) were *Citrobacter* species (*Citrobacter freundii*-3, *Citrobacter sakazakii*-1) with 1 (25%). This is much higher than that reported by Khanna A et al. [14] (4.70% of the *Citrobacter* spp out of a total of 1830 gram negative bacilli), but similar to the report of Avinash et al. (25.1% out of 1000 clinical samples). This high prevalence might be due to lack of preventive measures during the handling of the patients in these hospitals. A majority of the isolates were recovered from urinary samples (50%), while the remaining 50% were from sputum and ear swab. This low prevalence of *Citrobacter* spp from this research is similar to the report of Onoh RC et al. [16] who isolated five *Citrobacter* spp from a total of 252 bacterial isolates, cultured from urinary samples. Metri BC and Jyothi P [17] reported that invasive procedures like, catheterization helps the bacteria to colonize the urinary bladder. The presence of intact immune system in the host helps in defending against the pathogen to certain extent, but when the patients are immunocompromised, the condition would be fatal.

The study by Whitby has found *Citrobacter* in 12% of acute infections. Some studies reported that *Citrobacter* species appears to be one of the contributing organisms in causing urinary tract infection [17,18]. The antibiogram profile of the *Citrobacter* spp (Table 2) showed a significantly high level of resistance among all the isolates. In this study, Ofloxacin was active against all the four *Citrobacter* isolates (100%), with diameter zone of inhibition ranging from 18 mm to 25 mm. Closely followed is Piperacillin/Tazobactam which was active against three out of the four isolates (75%) with diameter zone

of inhibition ranging from 26 mm to 40 mm. Two of the isolates (50%) showed susceptibility to Co-trimoxazole (24 mm and 30 mm), and Imipenem (18 mm and 35 mm). The *Citrobacter* spp also demonstrated resistance to the β -lactam antibiotics. The rate of resistance among the isolates to Cephalosporins was high. Cefoxitin and Cefotaxime resistance were seen in all the four isolates (100%) while Ceftazidime resistance was observed in three of the isolates (75%). All the isolates were, however, resistant to Cloxacillin (100%), Oxacillin (100%), Piperacillin (100%) and combination of Ampicillin+Clavulanic acid (100%), Meropenem (100%) and Nystatin (100%). *Citrobacter sakazakii* demonstrated extreme multidrug resistance to all the drugs used except Ofloxacin. Over-prescription of antibiotics, lack of compliance, extensive use of antibiotics in Agriculture, poor infection control in health care settings, poor hygiene and sanitation and absence of new antibiotics being discovered have been linked to the development of resistant bacterial strains. Another likely cause of antibiotic resistance is poor drug quality which has been a major threat to healthcare system in Nigeria. In this study, *Citrobacter freundii* was the commonest species which was isolated, as also was the case in other studies [14,18]. ESBL production was found to be phenotypically absent in all the isolates (Table 3), using the combined disc potentiation method which differs from the 25% (1 out of 4) occurrence reported by Yusha'u (2010). It has been reported that the occurrence and distribution of ESBL producers among *Enterobacteriaceae* differs from country to country and hospital to hospital. However, the resistance shown by these *Citrobacter* spp isolates might be due to other mechanisms of resistance other than β -lactamase production. As shown in Table 5, the MIC values of Piperacillin/Tazobactam tested against the *Citrobacter* isolates are very high (100 μ g/mL and 110 μ g/mL). This may be a pointer that these isolates could develop resistance to these combined antibiotics if proper antibiotic administration measures are not put in place.

The strength of this study is the broad population of interest, involving both the in and out-patients samples reflects the circulating resistance, and the wide range of antibiotics used, involving almost all classes of antibiotics. There are however some limitations. This study had a relatively small sample size. This is due to the low prevalence of *Citrobacter* species among the isolates from the two tertiary health institutions, which has also been reported across the globe. Susceptibility testing was also carried out using antibiogram from few groups of antibiotics, excluding the aminoglycosides, tetracyclines, and macrolides.

Further area to be exploited for future research is the genetic diversity and the characterization of these antibiotic resistant *Citrobacter* strains. This will enable us confirm the mechanisms of resistance exhibited by these strains and the various genes responsible for their resistance.

Conclusion

Citrobacter species are emerging as important resistant pathogens. In this study, *Citrobacter freundii* was the commonest species isolated and the University Teaching Hospital (UCH), Ibadan in Southern Nigeria. Multidrug resistance is quite high among the *Citrobacter* species. These multidrug resistant

strains were mostly sensitive to a combination of Piperacillin/Tazobactam. Greater caution is required in the selection of the antibiotic/antibiotics to be used in treatment of patient with *Citrobacter* infections to avoid treatment failure. This bacterium can become resistant to these drugs at any time, and hence, the indiscriminate, inadequate and the prophylactic use of these antibiotics should be avoided. Infection control measures like disinfection of the wards and equipment, barrier precautions against colonized patients and hand hygiene measures should be strictly adhered to, to prevent the spread of the pathogen. The antibiogram is a cost effective, easy and rapid typing tool in a low resource country like Nigeria, where molecular typing tools are not easily accessible. Only in case of emergency, empirical therapy which is based on the antibiotic sensitivity pattern of that institution should be given to the patients.

Reference

1. Talbot GH, Bradley J, Edwards JE, et al. Bad bugs need drugs: an update on the development pipeline from the antimicrobial availability task force of the infectious diseases society of America. *Clin Infect Dis*. 2006;42(5):657-68.
2. Hsu LY, Tan TY, Jureen R, et al. Antimicrobial drug resistance in Singapore hospitals. *Emerg Infect Dis*. 2007;13(12):1944-47.
3. Okonko IO, Soley FA, Amusan TA, et al. Incidence of multidrug resistance (MDR) organisms in Abeokuta, southwestern Nigeria. *Global J Pharmacol*. 2009;3(2):69-80.
4. Mhada TV, Fredrick F, Matee MI, et al. Neonatal sepsis at Muhimbili National Hospital, Dar es Salaam, Tanzania: aetiology, antimicrobial sensitivity pattern and clinical outcome. *BMC Public Health*. 2012;12:904.
5. Roberts RR, Hota B, Ahmad I, et al. Societal costs of antimicrobial-resistant infections in a Chicago teaching hospital and implications for antibiotic stewardship. *Clin Infect Dis*. 2009;49(8):1175-84.
6. Okeke IN, Klugman KP, Bhutta ZA, et al. Antimicrobial resistance in developing countries Part 1: recent trends and status. *Lancet Infect Dis*. 2005;5(8):481-93.
7. Phillips I, Casewell M, Cox T, et al. Does the use of antibiotics in animals food pose a risk to human health? A critical review of published data. *J Antimicrob Chemother*. 2004;53(1):28-52.
8. Nawaz SK, Riaz S, Hasnain S. Screening for anti-methicillin resistant *Staphylococcus aureus* (MRSA) bacteriocin producing bacteria. *Afr J Biotech*. 2007;8(3):365-68.
9. Nada T, Baba H, Kawamura K, et al. A small outbreak of third generation, cephem-resistant *Citrobacter freundii* infections in a surgical ward. *Japan Infect Dis*. 2004;57(4):181-82.
10. Wang JT, Chang SC, Chen YC, et al. Comparison of antimicrobial susceptibility of *C. freundii* isolates in two different time periods. *J Microbiol Immunol Infect China*. 2000;33(4):258-62.
11. Thapa B, Adhikari P, Mahat K, et al. Multidrug resistant nosocomial *Citrobacter* in a hospital in Kathmandu Nepal. *Med Coll J*. 2009;11(3):195-9.
12. Metri BC, Jyothi P, Peerapur BV. Antibiotic resistance in *Citrobacter* spp. isolated from urinary tract infection. *Urol Ann*. 2013;5(4):312-3.
13. Menon T, Bindu D, Kumar CPG, et al. Comparison of double disc and 3 dimensional methods to screen for ESBL producer in tertiary care hospital. *Indian J Med Microbiol*. 2006;24(2):117-20.
14. Khanna A, Singh N, Aggarwal A, et al. The antibiotic resistance pattern in *Citrobacter* species: An emerging nosocomial pathogen in a tertiary care hospital. *J Clin Diagn Res*. 2012;6(4):642-44.
15. Avinash G, Sreenivasulu RP, Jithendra K, et al. Significance of *Citrobacter* as an emerging nosocomial pathogen with special reference to its antibiotic susceptibility pattern in a tertiary care hospital Nellore, AP, India. *Int J Curr Microbiol App Sci*. 2015;4(7):841-47.
16. Onoh RC, Umeora OIJ, Egwatu VE, et al. Antibiotic sensitivity pattern of uropathogens from pregnant women with urinary tract infection in Abakaliki, Nigeria. *Infect Drug Resist*. 2013;6:225-33.
17. Metri BC, Jyothi P. Antibiotic sensitivity pattern of *Citrobacter* spp. isolated from patients with urinary tract infections in tertiary care hospital in south india. *Int J Pharm Pharm Sci*. 2015;7(1):252-54.
18. Tula MY, Iyoha O. Distribution and antibiotic susceptibility pattern of bacterial pathogens causing urinary tract infection in Mubi General Hospital, Yola-Nigeria. *Br J Med Med Res*. 2014;4(9):224-30.

*Correspondence to:

Oludare Temitope Osuntokun
Department of Microbiology
Adekunle Ajasin University
Akungba Akoko
Nigeria
Tel: 08063813635; 08056096672
E-mail: osuntokun4m@gmail.com