



Detection of Extended Spectrum β -Lactamase Producing *Escherichia coli* in Neonatal Unit of a Tertiary Health Institution, South-South Nigeria

I. O. Idang¹, A. E. Moses^{1*}, A. N. Umo¹ and E. E. Ikpeme²

¹Department of Medical Microbiology and Parasitology, Faculty of Clinical Sciences, University of Uyo, Uyo, Nigeria.

²Department of Paediatrics, Faculty of Clinical Sciences, University of Uyo, Uyo, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. Author IOI performed the investigation and wrote part of the manuscript. Author AEM designed the study and wrote the protocol. Author ANU did literature search and also wrote part of the manuscript. Author EEI did the literature search and also wrote part of the manuscript. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JAMPS/2015/18742

Editor(s):

(1) Palmiro Poltronieri, National Research Council of Italy, CNR-ISPA, Italy and Institute of Sciences of Food Productions, Operative Unit in Lecce, Italy.

Reviewers:

(1) Anonymous, Brazil.

(2) Ronald Bartzatt, Chemistry Department, University of Nebraska, USA.

(3) Anonymous, University of Delhi, India.

Complete Peer review History: <http://www.sciencedomain.org/review-history.php?iid=1016&id=36&aid=9620>

Original Research Article

Received 7th May 2015

Accepted 25th May 2015

Published 6th June 2015

ABSTRACT

Aims: To determine the prevalence of multidrug resistant (MDR) and Extended Spectrum β -lactamase producing (ESBL) *E. coli* in neonates, their mothers and healthcare workers (HCWs) in the neonatal wards of the University of Uyo Teaching Hospital (UUTH), Uyo-Nigeria.

Study Design: It is a cross sectional hospital-based study involving neonates, their mothers and healthcare workers in the respective neonatal units of the hospital.

Place and Duration of Study: This study was conducted at the University Uyo Teaching Hospital, Uyo between May and November, 2012.

Methodology: The study involved 310 neonates admitted in Sick Babies' Unit, Special Care Babies' Unit and Neonatal ward; their mothers (168) and Healthcare Workers (35). Body sites of

*Corresponding author: E-mail: amoses264@yahoo.com;

neonates, palms of consented mothers and healthcare workers (HCWs) in these units were swabbed. Stool samples were also collected from the neonates and their mothers. Isolation and identification of *E. coli* were carried out using standard microbiological procedures. Antimicrobial susceptibility testing was done using the modified Kirby-Bauer disc diffusion method. Extended spectrum β -lactamase test was conducted using the disc diffusion synergy method following CLSI 2011 guidelines.

Results: Neonatal and maternal *E. coli* isolates were 38(12.3%) and 82(48.8%) respectively, and HCWs, 3(48.8%). Among the neonatal and mother isolates, resistance to Amoxycillin/Clavulanic acid ranged from 1.2% - 2.6%; Ceftriaxone and Ceftazidime resistance, 29.3% - 39.5% and resistance to Cefepime, 21.1% - 25.6%. Isolates from palms of HCWs were highly resistant to all antibiotics. Fourteen neonatal strains exhibiting multidrug resistance (MDR) had 100% homologous drug combinations with 18 maternal isolates. Neonatal isolates that are ESBL producers were 15(39.5%); mothers, 26(31.7%), and HCWs, 3(100%).

Conclusion: The importance of maintaining good hand hygiene by HCWs in neonatal units is highlighted in this study. There were high rates of MDR and ESBL-producing *E. coli* among neonates, mothers and HCWs in UUTH, Uyo. Similarities in drug combinations observed among the MDR strains obtained from subjects indicate possible transmission of *E. coli* from mothers and HCWs to neonates necessitating epidemiological concern. The practice of proper hygiene by mothers and HCWs should not be made optional. The high resistance strains observed may limit therapeutic options for serious neonatal infections with fatal consequences. Increased ESBL surveillance in neonatal units of hospitals is advocated.

Keywords: *E. coli*; multidrug resistance; ESBL; neonates; mothers; healthcare workers.

1. INTRODUCTION

Escherichia coli is a gram-negative rod shaped bacterium that is commonly found in the lower intestine of warm-blooded organisms [1,2]. It is the predominant facultative anaerobe of the human colonic flora. The organism typically colonises the infant gastrointestinal tract within hours of life, and therefore *E. coli* derive mutual benefit [3,4]. Most neonates may have swallowed the *E. coli* from the mothers' faeces along with the amniotic fluid during child birth and this may be related to the length of time it took doctors/midwives to deliver the baby. *E. coli* usually remains harmlessly confined to the intestinal lumen. However, in the debilitated or immune suppressed host, or when gastrointestinal barriers are violated, even non-pathogenic strains of *E. coli* can cause infection, such as meningitis, hemolytic uremic syndrome and peritonitis [3].

Results of antimicrobial susceptibility testing are normally used for multiple purposes, one of which is to assist clinicians in the design of empirical therapies for suspected infected individuals [5]. Treatment of infections including those caused by *E. coli* is becoming difficult because of multidrug resistance exhibited by the causative organisms [6,7]. More so, incidence of extended spectrum β -lactamase (ESBL) producing strains of *E. coli* has been steadily

increasing over the past few years resulting in limitation of therapeutic options [8]. This study was therefore carried out to determine the presence of multidrug resistance and ESBL producing *Escherichia coli* strains in neonates, their mothers and Healthcare workers (HCWs) in the neonatal wards of University of Uyo Teaching Hospital (UUTH), Uyo.

2. MATERIALS AND METHODS

2.1 Study Population

This study involved 310 neonates aged ≤ 28 days admitted in Sick Babies' Unit (SBU), Special Care Babies' Unit (SCBU) and Neonatal ward of University of Uyo Teaching Hospital (UUTH), Uyo. One hundred and sixty eight mothers and 35 Healthcare workers (HCWs) made up of nurses and ward orderlies working in these neonatal wards were included in this study following informed consent.

2.2 Sample Collection

Body sites of neonates (skin and ear) commonly exposed to touch by caregivers and rectum were swabbed using sterile swab sticks dipped in sterile normal saline as earlier described [9]. Rectal swabs were collected from neonates who were unable to provide stool samples. Palms of

consented mothers and HCWs were aseptically swabbed using sterile swab sticks dipped in sterile normal saline [9]. Stool samples were also collected from mothers in sterile universal bottles. All samples were transported to the laboratory within 2 hours of collection for processing.

2.3 Processing of Samples

All swabbed samples were cultured in Brain heart infusion broth at 37°C for 24 hours and later subcultured on Eosin Methylene Blue (EMB) agar (Lab M, UK) at 37°C for 24 hours. Isolates suspected to be *E. coli* strains with green metallic-sheen colour were isolated on EMB agar. Pure and discrete colonies were subcultured on nutrient agar slant and peptone water for conventional biochemical tests such as indole, citrate utilization, motility, oxidase, urease, reaction in Kligler's iron agar and sugar fermentation in order to identify putative *E. coli* isolates using standard procedures [10].

2.4 Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing was carried out with commercially prepared antibiotics disc using the Kirby Bauer disc diffusion method according to the guidelines of the Clinical Laboratory Standard Institute (CLSI) [11]. *E. coli* ATCC 25922 was used as reference strain. The antibiotics discs used were gentamicin (10 µg), ciprofloxacin (5 µg), ofloxacin (5 µg), amoxicillin/clavulanic acid, ceftazidime (30 µg), cefepime, ceftriaxone (30 µg) and cefuroxime sodium (Oxoid, UK). The diameters of the zones of inhibition were measured after 24 hours of incubation at 37°C.

2.5 Extended Spectrum β-lactamase Test

Isolates with zone diameters suspicious of ESBL production as pre-determined by the susceptibility test results were subjected to the CLSI ESBL phenotypic confirmatory test [11]. The phenotypic confirmatory test was conducted by placing amoxicillin (20 µg)/clavulanic acid (10 µg) (Oxoid, UK) combination disc at the center of each inoculated Mueller–Hinton agar plate. Cefotaxime (30 µg), ceftazidime (30 µg), aztreonam (30 µg) and cefpodoxime (10 µg) (Oxoid, UK) single discs were then placed 20 mm (center to center) from the amoxicillin/clavulanic acid combination disc and incubated at 37°C overnight (18-24 hr). Enhancement of the zones of inhibition of the

cephalosporin beta-lactam antibiotic discs towards the amoxicillin/clavulanic acid disc caused by the synergy with clavulanate was taken as an evidence of ESBL production. The result of the susceptibility test was interpreted according to the CLSI guidelines [11]. *E. coli* ATCC 25922 (β-lactamase negative), *Klebsiella pneumoniae* ATCC 700603 (ESBL-producer) were used as reference strains for quality control.

3. RESULTS

Out of 310 neonates examined, 38(12.3%) pure *E. coli* isolates were obtained from SBU and SCBU but none from neonatal ward. A total of 82(48.8%) pure *E. coli* isolates were isolated from stool of mothers and 3(8.6%) pure *E. coli* isolates from palms of HCWs. None of the mothers' palm yielded *E. coli*. (Table 1).

3.1 Antimicrobial Resistant Pattern of *E. coli* Isolates

Among the neonates and mothers, *E. coli* resistance to amoxicillin/clavulanic acid ranged from 1.2% - 2.6%. Third generation cephalosporins (ceftriaxone and ceftazidime) had 29.3% - 39.5% resistance while 21.1% - 25.6% were resistance to cefepime, a fourth generation cephalosporin. *E. coli* isolates from HCWs showed 2 (66.7%) resistance to amoxicillin/clavulanic acid and gentamicin and 100% resistance to all other antimicrobial agents (Table 2).

Table 1. Distribution of *E. coli* isolates from neonates, mothers and HCWs in UUTH, Uyo

Source/Location	No. tested	No. of <i>E. coli</i> (%)
Neonates		
SBU	115	20(17.4)
SCBU	115	18(15.7)
Neonatal Ward	80	0
Sub Total	310	38(12.3)
Mothers		
SBU	62	34(54.8)
SCBU	57	37(64.9)
Neonatal Ward	49	11(22.5)
Sub Total	168	82(48.8)
HCWs		
SBU	15	1(6.7)
SCBU	15	2(13.3)
Neonatal Ward	5	0
Sub Total	35	3(8.6)
Total	513	123(24.0)

Key: SBU= Sick Babies' Unit; SCBU= Special Care Babies's Unit

Table 2. Antimicrobial resistant pattern of *E. coli* isolates from neonates, mothers and HCWs in the Hospital

Antimicrobial agent µg/disc	No. <i>E. coli</i> resistance (%)		
	Neonates (n=38)	Mothers (n=82)	HCWs (n=3)
Ofloxacin (OFX) 5 µg	15(39.5)	26(31.7)	3(100)
Gentamicin (CN) 30 µg	15(39.5)	26(31.7)	2(66.7)
Cefepime (FEP) 30 µg	8(21.1)	21(25.6)	3(100)
Ciprofloxacin(CIP) 5 µg	15(39.5)	26(31.7)	3(100)
Cefuroxime Sodium (CXM) 30 µg	16(42.1)	27(32.9)	3(100)
Ceftriaxone (CRO) 30 µg	15(39.5)	26(31.7)	3(100)
Amoxicillin/Clavulanic acid (AMC) 30 µg	1(2.6)	1(1.2)	2(66.7)
Ceftazidime (CAZ) 30 µg	15(39.5)	24(29.3)	3(100)

3.2 Multidrug Resistance Pattern

The distribution of multidrug resistance (MDR) strains of *E. coli* among neonates, mothers and HCWs shown in Table 3 reveals some similarities among the *E. coli* isolates based on their resistotype drug combinations. Fourteen (14) MDR strains from neonates had 100% homologous resistotype drug combination with 18 *E. coli* isolates from mothers and one strain from the HCWs. Similarly, 6 MDR strains from mothers had homologous drug combination with an isolate from neonate while one MDR strain from mother had eight similar drugs in combination with one MDR strain from HCW. However, seven MDR strains from neonates and mothers had 86% homologous drug combination with 33 MDR strains from neonates- 14, mothers- 18 and HCWs- 1; and these 33 MDR strains had 87.5% homologous drug combinations with 2 MDR strains from mother - 1 and neonate - 1.

3.3 Extended Spectrum β -lactamase Producers

Fifteen (39.5%) of the *E. coli* isolates from neonates were ESBL producers. In mothers, 26(31.7%) were ESBL producers while all *E. coli* isolates from the HCWs were ESBL producers (Table 4).

4. DISCUSSION

Findings on the prevalence of *E. coli* isolates among neonates, mothers and HCWs in UUTH, Uyo has earlier been documented [9]. The detection of pure *E. coli* isolates from palms of 3 HCWs (8.6%) in this study underscores the importance of hand hygiene in infection control efforts in the hospitals, especially those working with and handling neonates. There are ample evidence to suggest that transient carriage of

microorganisms on the hands of HCWs is a more important means of microbial transmission among patients [6,12,13]. The elimination of these microbial agents is possible by observing a good hand hygiene such as routine hand washing with chlorohexidine or alcohol-based antiseptics [6] as well as use of barrier precautions (gloves and gowns) and body substance isolation to provide extra measure of protection against cross-infection [14,15]. However, its been suggested that barrier precautions could fail if colonized patients are not identified and isolated promptly [14].

The high presence of multidrug resistance observed among neonates, mothers and HCWs in this study and the fact that similar MDR pattern were observed in neonates and mothers, suggest that these strains were circulating among the subjects in this study. For instance, the resistance drug combination among the 14 MDR strains from neonates were 100% homogeneous with 18 MDR strains from the mothers. Also, these 14 MDR strains had 100% similar drug combinations with one of the strains from HCWs. The primary source of resistance may have resulted from selective pressure imposed by extensive use of antimicrobials by mothers and HCWs. This is in agreement with the findings of other workers who reported from their findings that most neonates are colonised by the predominant strains of *Escherichia coli* present in their own mother's faecal flora [2,16,17].

In this study, the antimicrobial pattern of *E. coli* strains show varied degree of resistance to routinely therapeutic used antimicrobial agents such as gentamicin, ciprofloxacin, ofloxacin, amoxicillin/clavulanic acid, including 3rd generation ceftazidime, ceftriaxone, cefuroxime sodium and 4th generation cefepime. These third

Table 3. Multidrug resistance (MDR) pattern of *E. coli* strains isolated from neonates, mothers and HCWs in the Hospital

Strains	No. of drug combination	Drug combination
Neonates		
AST45,AST28,AST85,ASK80,ASK23, AE82, BST21, BSK63, BSK15, BSK26, BSK6, BE56, BE61, BE24 (n=14) ^a	7	OFL, CAZ, CRO, CN, CXM, CIP, FEP
BR33 (n=1) ^b	6	OFL, CAZ, CRO, CN, CXM, CIP
Mothers		
AM122, CM136, BM40, AM17, AM50, AM122 (n=6) ^b	6	OFL, CAZ, CRO, CN, CXM, CIP
AM33 (n=1) ^c	8	OFL, CAZ, CRO, CN, CXM, CIP, AMC, FEP
BM30, AM5 (n=2) ^d	6	OFL, CRO, CN, CXM, CIP, FEP
AM37, BM54, BM64, AM112, BM83, BM151, AM120, BM96, CM135, BM160, AM115, BM109, AM143, AM144, AM125, AM149, BM77, BM166 (n= 18) ^a	7	OFL, CAZ, CRO, CN, CXM, CIP, FEP
HCWs		
BH30 (n=1) ^a	7	OFL, CAZ, CRO, CN, CXM, CIP, FEP
BH10 (n=1) ^e	7	OFL, CAZ, CRO, CXM, CIP, AMC, FEP
AH14 (n=1) ^c	8	OFL, CAZ, CRO, CN, CXM, CIP, AMC, FEP

Key: CN- Gentamicin, CIP- Ciprofloxacin, OFL- Ofloxacin, AMC- Amoxycillin/Clavulanic acid, CAZ- Ceftazidime, FEP- Cefepime, CRO- Ceftriaxone, CXM- Cefuroxime sodium. Groups ^{a, b, c, d and e} are MDR *E. coli* strains with homogeneous drug combinations

Table 4. ESBL producing *E. coli* strains isolated from neonates, mothers and HCWs in UUTH, Uyo

Sources of <i>E. coli</i>	No. of <i>E. coli</i>	ESBL positive isolates (%)
Neonates	38	15 (39.5)
Mothers	82	26 (31.7)
HCWs	3	3 (100)

generation cephalosporins are among the most widely used antibiotics in many hospitals in our locality and studies have shown that relationship exist between 3rd generation cephalosporins use and acquisition of ESBL [18,19]. Hence, the indiscriminate use of cephalosporins either through self-medication or over-prescribed by health practitioners could be responsible for the high rate of selection of ESBL producing microorganisms as confirmed by other researchers [20-22]. The production of ESBL by *E. coli* strains in this study may have contributed to the resistance observed among routinely used antibiotics as reported in other studies [21]. The positive rate of ESBL producing *E. coli* isolates (39.5%) among neonates in this study is comparable to 36.5% reported by Shakil et al. [23]. However, this is almost 2-fold less than 63.6% reported by Durrane et al. [24] in Karachi, Pakistan.

Also, six MDR strains obtained from mothers in this studies had similar drug combination with an isolate from neonate while one maternal MDR strain with eight resistant drug combination was similar to one MDR strain obtained from an HCW. This indicates a possibility that similar *E. coli* strains were being circulated within the different neonatal units/wards of the hospital by either the mothers or HCWs attending to these babies. This agrees with the study of other workers who reported an outbreak of ESBL producing *E. coli* in a Neonatal Care Unit in Basel, Switzerland, which began with transmission from a mother to her new born twins during vaginal delivery. Subsequently, infection was spread by an HCW who came in contact with the neonates to other neonates and then to other HCWs [25].

Among the HCWs in this study, all the *E. coli* isolates were found to be ESBL producers and this is higher than but comparable to 90% reported in Bamako, Mali [26]. In this study, the prevalence of ESBL producing *E. coli* (31.7%) in mothers was lower but comparable to 47% reported in India [27]. The importance of these findings is borne out of the fact that multidrug resistance and ESBL producing *E. coli* have been reported elsewhere as a major cause of

neonatal meningitis and haemolytic-uremic syndrome (HUS) in neonates [3,28]. Moreso, a recent study conducted in UUTH, Uyo reported 20% prevalence of ESBL producing organisms from children under 1 year of age [29]. This observation has serious therapeutic implications that may impact on clinical outcomes in infants. ESBL producing organisms have been associated with increased mortality and morbidity among babies [6].

5. CONCLUSION

The importance of maintaining good hand hygiene and barrier precautions by healthcare workers in neonatal units is highlighted in this study. The practice will avoid transfer of hand carriage organisms to babies by HCWs and will go a long way to aid infection control in our hospitals. Also, excessive use of broad spectrum antibiotics through self-medication and general abuse of antibiotics available as over the counter drugs in our locality may have contributed to the resistance observed in this study [30]. A high level of MDR strains and ESBL production was generally observed among the neonatal and maternal isolates, most especially isolates from HCWs. The observed similarities in multidrug resistance pattern in terms of drug combinations are of epidemiological concern as this may limit therapeutic options for serious neonatal infections caused by these strains thereby leading to fatal consequences. Policies that would encourage the practice of good personal and interpersonal hygiene by mothers and healthcare workers should be instituted in the children's wards/clinics of all healthcare facilities while increased ESBL surveillance in neonatal units/wards of hospitals is strongly advocated.

ETHICAL APPROVAL

All authors hereby declare that the protocol of this study have been examined and approved by the ethics committee of the University of Uyo Teaching Hospital and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

ACKNOWLEDGEMENTS

The technical assistance of Drs Eno Nyong and Ifeanyi Onwuezobe of University of Uyo Teaching Hospital, Uyo in the course of this study are highly appreciated.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Vogt RL, Dipplod L. *Escherichia coli* O157:H7 outbreak associated with consumption of ground beef, June-July 2002. Public Health Report. 2005;120(2): 174-78.
2. Sharma S, Bhat GK, Shenoy S. Virulence factors and drug resistance in *Escherichia coli* isolated from extraintestinal infections. Indian Journal of Medical Microbiology. 2007;25:369-73.
3. Todar K. Lectures in Microbiology: All about *E. coli*. Department of Bacteriology, University of Wilconson-Madison; 2009. Available:<http://www.textbookofbacteriologynet/E.coli.html> (Accessed on: 30th Nov., 2013).
4. Willey JM, Sherwood, LM, Woolverton CJ. Prescott, Harley and Klein's Microbiology. 7th ed., McGraw-Hill International Edition, New York. 2008;157-1032.
5. Chitnis SV, Chitris V, Sharma N, Chitnis DS. Current statue of drug resistance among gram negative bacilli isolated from admitted cases in a tertiary care centre. Journal association of Physicians India. 2003;51:28-31.
6. Paterson DL, Bonomo RA. Extended spectrum beta-lactamases: A clinical update. Clinical Microbiology Reviews. 2005;18:657-86
7. Schwaber MJ, Carmeli Y. Mortality and delay in effective therapy associated with extended-spectrum β -lactamase production in Enterobacteriaceae Bacteremia: A systematic review and meta-analysis. Journal of Antimicrobial Chemotherapy. 2007;60:913-920.
8. European Centre for Disease Prevention and Control. European Antimicrobial Resistance Surveillance Network (EARS-NET) homepage on the Internet. Stockholm, Sweden: European Centre for Disease Prevention and Control. Available:http://www.rivm.nl/earss/databas_e/ (Accessed on: September 16, 2013)
9. Moses AE, Idang IO, Onwuezobe IA. *Escherichia coli* colonisation in neonates,

- mothers and healthcare workers: The University of Uyo Teaching Hospital experience. *World Journal of Biomedical Research*. 2014;1(1):27-31.
10. Cheesbrough M. *Medical laboratory manual for tropical countries Vol. 2*, Cambridge University Press, UK. 2012; 400-80.
 11. Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing; Twenty First Informational Supplement; M100-S21 Clinical Laboratory and Standards Institute, Wayne, PA. 2011;31:42.
 12. Gupta A, Della-Latta P, Todd B, San-Gabriel P, Huas J, Wu F. Outbreak of extended-spectrum beta-lactamase-producing *Klebsiella pneumoniae* in a neonatal intensive care unit linked to artificial nails. *Infection Control Hospital Epidemiology*. 2004;25:210-215.
 13. Quinn JP. Clinical strategies for serious infection: A north american perspective. *Diagnosis of Microbiology Infectious Disease*. 1998;31:389-395.
 14. Goldmann DA. The role of barrier precautions in infection control. *J Hosp Infect*. 1991;18(Suppl A):515-23.
 15. Lynch P, Jackson MM, Cummings MJ, Stamm WE. Rethinking the role of isolation practices in the prevention of nosocomial infections. *Ann Intern Med*. 1987;107(2): 243-6.
 16. Harbarth S, Samore MH. Antimicrobial resistance determinants and future control. *Emerging Infectious Diseases*. 2005;11: 794-801.
 17. Van de Sande-Bruinsma N, Grundmann H, Verloo D, Tiemersma E, Monen J, Goossens H. Antimicrobial drug use and resistance in Europe. *Emerging Infectious Diseases*. 2008;14:1722-30.
 18. Livermore DM. Beta-lactamases in laboratory and clinical resistance. *Clinical Microbiology Review*. 1995;8:557-584.
 19. Arriffin H, Navaratnam P, Mohammed M, Arasu A, Abdullahi WA, Lee CL, Peng LH. Ceftazidime-resistant *Klebsiella pneumoniae* blood stream infection in children with febrile neutropenia. *International Journal of Infectious Diseases*. 2000;4:21-5.
 20. Ghatole M, Manthalkar P, Kandle S, Yemul V, Jahagirdar V. Correlation of extended-spectrum beta lactamases production with cephalosporin resistance in gram-negative Bacilli. *Indian journal of Pathology and Microbiology*. 2004;47:82-4.
 21. Ramesh N, Sumathi CS, Balasubramanian K, Palaniappan KR, Kannan VR. Urinary tract infection and antimicrobial susceptibility pattern of extended spectrum of beta lactamase producing clinical isolates. *Advances in Biological Research*. 2008;2:78-82.
 22. Gonzales R, Malone DC, Maselli JH, Sande MA. Excessive antibiotic use in acute respiratory infections in The United States. *Clinical Infectious Diseases*. 2011; 33:757-762.
 23. Shakil S, Akram M, Ali SM, Khan U. Acquisition of extended-spectrum β -lactamase producing *Escherichia coli* strains in male and female infants admitted to a neonatal intensive care unit: Molecular epidemiology and analysis of risk factors. *Journal of Medical Microbiology*. 2010;59: 948-54.
 24. Durrane T, Asad S, Anita Z. Antimicrobial resistance among neonatal pathogens in developing countries. *Pediatric Infectious Journal*. 2009;28:519-21.
 25. Tschudin Sutter S, Frei R, Battagay M, Hoesli I, Widmer FA. Extended spectrum β -lactamase producing *Escherichia coli* in neonatal care unit. *Emerging Infectious Diseases*. 2010;16(11):1758-1760.
 26. Tande D, Jallot N, Bougoudogo F, Montagnon T, Gouriou S, Sizun J. Extended-spectrum β -Lactamase-producing Enterobacteriaceae in Melian orphanage. *Emerging Infectious Diseases*. 2009;15(3):472-74.
 27. Rizvi M, Khan F, Shukia I, Malik A, Shaheen. Rising prevalence of antimicrobial resistance in urinary tract infections during pregnancy: Necessity for exploring newer treatment options. *Journal of Laboratory Physicians*. 2011;3(2):98-103.
 28. Todar K. Pathogenic *E. coli*. Online textbook of Bacteriology, Department of Bacteriology, University of Wisconsin-Madison; 2007. Available:http://www.textbookofbacteriology.net/E_coli.html (Accessed on: 16th Oct., 2010)
 29. Azekhueme I, Moses AE, Abbey SD. Extended spectrum β - lactamases in clinical isolates of *Escherichia coli* and

- Klebsiella pneumoniae* from University of Uyo Teaching Hospital, Uyo-Nigeria. Journal of Advances in Medical and Pharmaceutical Sciences. 2015;2(3):117-235.
30. Arikpo G, Eja M, Enyi-Idoh K. Self-medication in Rural Africa: The Nigerian Experience. The Internet Journal of Health. 2009;11(1). Available: <https://ispub.com/IJH/11/1/5032>. (Accessed on: 16th Dec., 2014)

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Peer-review history:
The peer review history for this paper can be accessed here:
<http://www.sciencedomain.org/review-history.php?iid=1016&id=36&aid=9620>