

Original Research Article

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Antibiotic Sensitivity Profile of *Escherichia coli* Associated with Specific Infections in a Nigerian Tertiary Health Institution

M.S. Odimayo^{1*}, S.I. Nwadioha², Joseph Adejoke Adijat¹ and Daramola Gabriel Olugbenga³

¹Department of Medical Microbiology and Parasitology, College of Medicine, Ekiti State University, Ado Ekiti, Nigeria

²Department of Medical Microbiology and Parasitology, College of Health Sciences, Benue State University, Nigeria

³Department of Medical Microbiology and Parasitology, Ekiti State University Teaching Hospital, Ado Ekiti, Nigeria

*Corresponding author

ABSTRACT

To determine the prevalence of *Escherichia coli* and the drug susceptibility profile. A twenty –six month retrospective review of culture results of clinical specimens in Ekiti State University Teaching Hospital Medical Microbiology Laboratory from January, 2014 to February, 2016 was done. A total of five hundred and twenty seven (26.4%) *Escherichia coli* isolates were recovered from 2000 clinical samples for cultures submitted to the laboratory during the 26 months review period. About fifty eight per cent of the isolates were from clinical samples of female patients with 33.6% increased incidence in the age group of 45 years and above. About sixty per cent (n=315/527) of the isolates were from urine samples while 0.4% least isolation rate (n=2/527) was from cerebrospinal fluid samples. The following antibiotics were found to be effective against *Escherichia coli* isolates; Nitrofurantoin (83.5%)- primarily for urinary tract infection, Ceftriaxone (73.6%), Gentamicin (56.7%) and averagely Ciprofloxacin (49.3%). On the other hand, the following antibiotics showed poor activities; Ampicillin (1.3%), Cefuroxime (20.%) and Amoxicillin-clavulanic acid (30.4%). A high rate of multi-drug resistance was noted in this study, especially to commonly used first line antibiotics. Ceftriaxone, Gentamicin and Ciprofloxacin are choices to be considered when treating infections due to *E.coli* in this geographical location.

Keywords

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Antimicrobial
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Introduction

Escherichia coli, abbreviated and often referred to as *E. coli* is a gram-negative, facultative anaerobic, rod-shaped bacterium of the genus *Escherichia* that is widely distributed in nature and commonly found in the lower intestine of warm-blooded animals and humans. *E. coli* is the bacterial species

most commonly recovered in the clinical laboratories especially from urine samples (Nsofor *et al.*, 2003). It has been incriminated in infectious diseases involving virtually every human tissue and organ system (Washington *et al.*, 2006). *E. coli* is one of the main causes of both nosocomial

and community acquired infections in humans, a leading cause of urinary tract, ear, wound and other infections in humans including gram negative sepsis, endotoxin-induced shock, meningitis, etc (Monica Cheesbrough, 2010). *E. coli* is the most frequent cause of community-acquired UTIs and is frequently found to be responsible for intra-abdominal and soft tissue infections.

This hard organism can easily be isolated in the laboratory on some differential/ indicator media such as MacConkey, Cysteine Lactose Electrolyte Deficiency (CLED) media where it grows as Lactose fermenting colony. They are highly motile on wet mount. On Gram staining, they are gram negative rods. They are characteristically identified as positive to Indole reaction test.

The discovery of antimicrobial agents had a major impact on the rate of survival from infections caused by this organism though a rise in bacterial resistance to antibiotics complicates treatment of these infections. The changing patterns of antimicrobial resistance however necessitated the need for new antibacterial agents. Antimicrobial resistance in *E. coli* has been reported worldwide and increasing rates of resistance among *E. coli* is a growing concern in both developed and developing countries (Bell *et al.*, 2002).

Pathogenic isolates of *E. coli* have been reported to have a relatively large potential for developing resistance. Secretion of Extended Spectrum Beta-Lactamases (ESBL) is the main mechanism of developing resistance to antibiotics by the organism. β -Lactamases continue to be the leading cause of resistance to β -lactam antibiotics among gram-negative bacteria. *E. coli* strains involved in mixed infections as well as strains isolated from burns have an increased resistance to antibiotics. Most infections caused by ESBL-producing

Escherichia coli (ESBLEC) or *Klebsiella pneumoniae* had mostly been described as nosocomially acquired or nursing home related. Some studies suggest that infections due to ESBL-producing organisms might be an emergent problem in outpatients in different countries (Daza *et al.*, 2011; Garner *et al.*, 1998). In a nationwide study of ESBL-producing organisms carried out in Spain, 51% of ESBL-producing *E. coli* (ESBLEC) strains were isolated from outpatients. Oteo *et al.*, in a study carried out in some Spanish hospitals reported that antimicrobial resistance, particularly to fluoroquinolones and third-generation cephalosporins was increasing in *E. coli*.¹¹ In Nigeria there are some reports of high level resistance in *E. coli* infections from extraintestinal sources (Akinduti *et al.*, 2011).

There is a geographical variation in the antibiotic susceptibility pattern of isolates to commonly used antibiotics. This, with the frequency of isolation of *E. coli* in clinical microbiology laboratory because of its involvement in a wide spectrum of human infections and the ease of its isolation, and the emergence of resistance in strains irrespective of the clinical sample and whether patient is hospitalised or out-patient is the basis of carrying out this study in this environment.

The study therefore is aimed at determining the antimicrobial susceptibility pattern of *E. coli* isolated from various clinical samples.

Materials and Methods

A retrospective review of culture results of all clinical samples submitted to the Medical Microbiology laboratory of Ekiti State University Teaching Hospital from January, 2014 to February, 2016 was done. A total of 2000 samples were received in the laboratory for bacteria culture and

antimicrobial susceptibility testing during the period under review.

Culture

Specimens were inoculated on the appropriate media for the isolation of *Escherichia coli* according to standard culture techniques for culturing on solid agar and broth media was done. Gram staining of isolates was done and biochemical tests such as Triple Sugar Iron (TSI) slope inoculation, Indole test and Motility test were carried out on the isolates from purity plates.

Antibiotic susceptibility testing

Antibiotic susceptibility testing of isolates was done on dried Mueller Hinton agar plates using the modified Kirby Bauer method according to the Clinical and Laboratory Standards Institute (CLSI), 2006 guidelines on Antimicrobial Susceptibility Testing (Clinical Laboratory Standard Institute, 2008). Standard antibiotic impregnated discs were used which include Ciprofloxacin (5 mcg), Gentamicin (10 mcg), Ofloxacin (30mcg), Augmentin (30mcg), Cotrimoxazole (25mcg), Ceftriaxone (30mcg), Ampicillin (10mcg), Cefuroxime(30mcg), Nitrofurantoin (300mcg). Type culture of *Escherichia coli* ATCC 25922 served as control strains.

Data Analysis

The chi square test and percentage proportion were used in analysing collated data. P-value of < 0.05 was considered to indicate statistically significant differences where applicable.

Results and Discussion

Table 1 shows that a total of 527 *E. coli* isolates were recovered from 2000 clinical samples submitted to the laboratory during the 26 months review period. Three hundred

and six (58.1%) of the isolates was from female patients. As depicted in table 2, the majority of this isolates were from urine samples (59.8%) and least from cerebrospinal fluid samples (0.4%). Figure 1 shows that the age group most affected is the >45 years (33.6%) while the less than 15 years were the least affected (10.6%).

Table 3 shows the antibiotic sensitivity pattern of *E. coli* from clinical samples. Eight antibiotics were tested against each isolate and Nitrofurantoin was included for urine isolates only. All *E. coli* from CSF were sensitive to Gentamycin and Ciprofloxacin and resistant to all other antibiotics used. Likewise, *E. coli* isolated from blood and sputum were all sensitive to ceftriaxone. None of those recovered from CSF, blood and sputum were sensitive to Augmentin, Ampicillin and Cefuroxime. Urine isolates were noted to be generally most sensitive to Nitrofurantoin (83.5%) while isolates from swab specimen were most sensitive to Ceftriaxone (72.1%).

Infectious diseases are a common cause of hospital visit and death in the tropics. A larger group of diseases is caused by bacteria and in the past, bacterial diseases were a major cause of death before the advent of antibiotics. The rising incidence of resistance to commonly prescribed antibiotics by bacterial agents has become a global concern.

Of all the clinical specimens submitted to Ekiti State University Teaching Hospital Microbiology laboratory during the period under review, *E. coli* was found to be a common bacterial isolate, found in a variety of clinical samples like stool, CSF, blood, sputum, urine, semen and swab samples. This amount to an isolation rate of 26.4%($n=527/2000$) The 26.4% finding however is much lower than 63% isolation rate recorded in Osogbo, S/W Nigeria , 37%

¹⁶ at Owerri, S/E Nigeria, but higher than 14% and 14.2% in Kano, N/E Nigeria and Ethiopia consecutively. A high *E. coli* prevalence of 59.8% was isolated from urine samples at a greater frequency than from other clinical specimens. Similar results of 53%, 55%, 45.5% were reported in other studies.

A fifty eight per cent peak incidence of *Escherichia coli* was recovered from female subjects in the study. This is however not surprising for UTI is more common in females than in males because of differences in anatomic structure and changes during sexual maturation, pregnancy, and childbirth (Olowe *et al.*, 2008; Nsofor *et al.*, 2016). Lack of antimicrobial properties of prostatic fluid as in males, hormonal changes which affect the mucosal adherence of bacteria and trauma of urethra during sexual intercourse are also highlighted factors predisposing the female to UTI (Nwadioha *et al.*, 2010). Various studies has also implicated *E. coli* as the commonest cause of UTI (Olowe *et al.*, 2008; Nsofor *et al.*, 2016; Kibret *et al.*, 2011). From the study, *E. coli* was recovered 33.6% mostly from 45years of age and 10 % least in childhood (0-15years old). This is similar to the findings of Kaniz and co-workers in Bangladesh where it was reportedly commoner in the 31-45 years age group and least in the 0-15 years age group. The finding is in keeping with a similar report in Abeokuta, Nigeria where *E. coli* recovery in urine samples was 58.4% highest recovery rate in the age group 16-45 years. It may be attributed to frequent sexual intercourse, use of contraceptive spermicidal agents, diaphragms and menopause for women and bladder stasis for older men with prostatic hypertrophy.

Antibiotics commonly prescribed for various infectious disease conditions by physicians in this hospital and environ include

Gentamycin, Augmentin, Ciprofloxacin, Ceftriaxone, Cefuroxime and Ceftazidime hence the need to study the susceptibility of *E. coli* to this commonly prescribed drugs. In this study, *E. coli* isolates from CSF and blood specimens were found to be resistant to almost all antibiotics tested while isolates from urine samples were found to be sensitive to all antibiotics used. Of all the antibiotics tested against urine pathogen, nitrofurantoin is found to be 83.5% highly sensitive. A similar finding was reported in Ethiopia where significantly high degree of sensitivity rate to nitrofurantoin (96.4%) was recorded. The high degree of sensitivity to Nitrofurantoin by urinary tract bacterial pathogens is probably due to its narrow spectrum of activity, narrow tissue distribution and almost no effect outside of the urinary tract.

Resistance against common penicillin derivative like ampicillin was reported in this study. This maybe due to overuse of this drug which is readily available, affordable and can be procured over the counter without prescription. A below average sensitivity was also reported to ofloxacin (42.5%) and ciprofloxacin (49.3%). Increased resistance of *E. coli* against Quinolones may be due to the overuse of these drugs for the treatment of UTI or generalized use of fluoroquinolones in animals feed which in turn lead to the transmission of resistance mechanism to strains from animals to human (Kaniz, 2016).

From this study, it can be inferred that ampicillin (1.3%), cefuroxime (20.5%) and augmentin (30.4%) are less effective on *E. coli* in this environment. Gentamycin, ceftriaxone and, to a lesser extent, ciprofloxacin are choices to be considered when treating infections due to *E. coli* in the study environment.

Table.1 Distribution of *E. coli* Isolated From Different Specimens Between Jan. 2014 to Feb. 2016 at Ekiti State University Teaching Hospital, Ado Ekiti.

Sample	Male n (%)	Female n (%)	Total n (%)	χ^2	p value
Urine	138 (62.4)	177 (57.8)	315 (59.8)	22.215	0.001*
Sputum	9 (4.1)	8 (2.6)	17 (3.2)		
Swab	53 (24.0)	112 (36.6)	165 (31.3)		
Semen	8 (3.6)	-	8 (1.5)		
Stool	7 (3.2)	6 (2.0)	13 (2.5)		
Blood	5 (2.3)	2 (0.7)	7 (1.3)		
C.S.F.	1 (0.4)	1 (0.3)	2 (0.4)		
Total	221 (41.9)	306 (58.1)	527 (100.0)		

Table.2 Quarterly isolation rate of *E. coli* from different samples

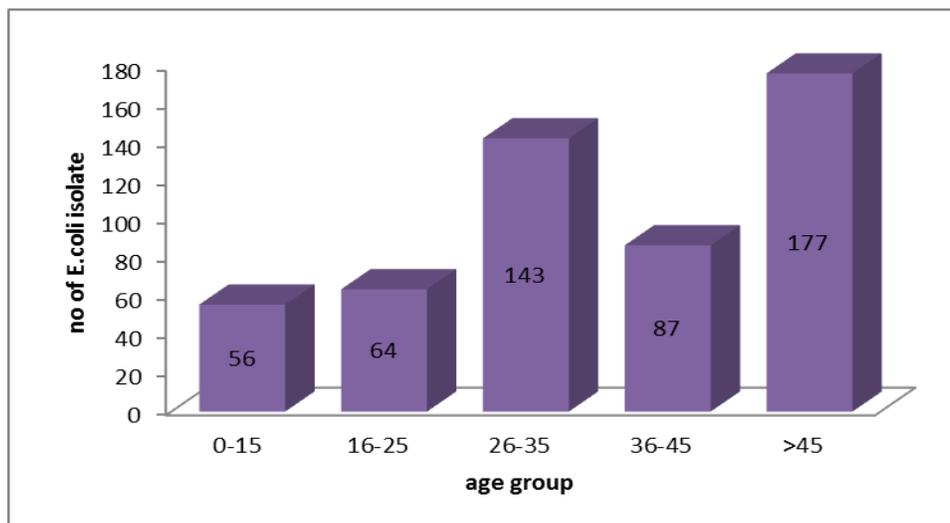
Period	Urine	Sputum	Swab	Semen	Stool	Blood	CSF	Total(%)
Jan.-March, 2014	38	2	16	0	2	0	1	59(11.2)
April – June, 2014	44	4	26	1	1	1	0	77(14.6)
July – Sept., 2014	24	3	19	1	1	0	0	48(9.1)
Oct. –Dec., 2014	33	0	13	1	1	1	0	49(9.3)
Jan.-March, 2015	31	1	23	1	2	0	0	58(11)
April – June, 2015	38	2	16	0	1	0	0	57(10.8)
July – Sept., 2015	25	1	20	0	4	2	0	52(9.9)
Oct. –Dec., 2015	61	4	20	0	1	3	0	89(16.9)
Jan.-Feb., 2016	21	0	12	4	0	0	1	38(7.2)
Total(%)	315 (59.8)	17 (3.2)	165 (31.3)	8 (1.5)	13 (2.5)	7 (1.3)	2 (0.4)	527 (100)

Table.3 Antibiotic Sensitivity Pattern of *E. coli* Isolated from the Clinical Specimens

Antibiotics	Urine n=315(%)	Swab n=165(%)	CSF n=2 (%)	Blood n=7 (%)	Sputum n=17(%)	Total N=527 (%)
Gentamycin	185(58.7)	103(62.4)	2(100)	0(0)	9(52.9)	299(56.7)
Ofloxacin	189(60)	17(10.3)	0(0)	5(71.4)	13(76.5)	224(42.5)
Ciprofloxacin	156(49.5)	93(56.4)	2(100)	2(28.6)	7(41.2)	260(49.3)
Augmentin	111(35.2)	49(29.7)	0(0)	0(0)	0(0)	160(30.4)
Cotrimoxazole	163(51.7)	85(51.5)	0(0)	0(0)	7(41.2)	255(48.4)
Ceftriaxone	245(77.8)	119(72.1)	0(0)	7(100)	17(100)	388(73.6)
Ampicillin	4(1.3)	0(0)	0(0)	0(0)	0(0)	4(1.3)
Cefuroxime	35(11.1)	73(44.2)	0(0)	0(0)	0(0)	108(20.5)
Nitrofurantoin	263(83.5)	*NA	NA	NA	NA	263(83.5)

*NA: not applicable

Fig.1 Age Distribution of Patients whose Clinical Specimens were Infected with *E. coli*



A similar study carried out in Bangladesh, Nepal, India and North America concluded that Co-trimoxazole and Nalidixic acid were virtually useless against *E. coli*. Of note in this study is the high rate of resistance to antibiotics and this further confirms earlier reports that various reservoir sources of multi-resistant clones of *E. coli* carrying mobile genetic elements capable of epidemic spread are present in our locality. This has a public health implication as an outbreak of *E. coli*, especially the Extended Spectrum Beta -Lactamase enzyme (ESBL), Carbapenemases enzyme multi-drug resistant strains is a possibility (Jacobson *et al.*, 2008).

In conclusion, a high rate of multi-drug resistance was noted in the study, especially to commonly used first line antibiotics. Gentamycin, ceftriaxone and ciprofloxacin are choices to be considered when treating infections due to *E. coli* in this geographical location.

Recommendation

Regular antimicrobial susceptibility surveillance is recommended and common antibiotics to which resistance is being noted

could be withdrawn from circulation.

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