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Prevalence and antibiotic susceptibility pattern of *E.coli* isolated from clinical samples in Kalaburagi region, Karnataka, India

Jahanara Kudsi, Chandrakanth Kelmani R

Medical Biotechnology and Phage Therapy Laboratory, Department of Post Graduate Studies and Research in Biotechnology, Gulbarga University, Gulbarga, Karnataka, India

Abstract
Antibiotic resistance is a major concern throughout world in all the microorganisms and especially in *E.coli* as it is a commensal bacteria and pathogenic variants are infectious. Presently it is important to design a systematic choice of antibiotics to avoid spread of multi drug resistance in *E.coli*. The aim of this study is to determine antibiotic susceptibility pattern of *E.coli* from different clinical samples in Kalaburagi region. About 150 isolates were isolated by standard methods and antibiotic susceptibility profiling was done.100% resistance to ampicillin and Nalidixic acid, 40% to amoxicillin/clavulinic acid, 80% to Cefotaxime, 70% to ceftazidime, 80% to ceforoxime, 81% to ciprofloxacin, 40% to levoflaxacin, 20% to gentamycin and 5% resistance to Imepenem was observed. Our study indicates that first three generation of beta-lactams antibiotic and fluroquinolones have limiting effect over MDR *E. Coli*. But most of strains were susceptible to Imepenem, gentamycin and to some extent amoxicillin clavulinic acid. MIC for cefotaxime and ceftaxime/clavulinic acid was performed to confirm the presence of ESBL enzymes in the resistance strains. Out of 150 strains 120 resistant strain were ESBL positive i.e., 60% strains. With proper data analysis this study can be used to decide an empirical antibiotic thearpy for *E.coli* infections.

Keywords: *E.coli*, antibiotic susceptibility test, prevalence

1. Introduction
E.coli is commensally growing bacteria found in human as well as animals. In humans, they are the major aerobic organism residing in the intestine, typically with around 106 to 109 colony forming units per gram of stool [1]. The organism is also found in soil and water, usually as a result of fecal contamination but the pathogenic variants can cause various types of infection including gastroenteritis, urinary tract infection, meningitis, peritonitis and septicaemia [2, 3]. Treatment to this infection is complicated due to emergence of multi drug resistance among pathogenic variants. In last 20 years of time span major increases in emergence and spread of multidrug-resistant bacteria and increasing resistance to antibiotic groups, such as fluoroquinolones and certain cephalosporins [4].

The β-lactams antibiotics, in combination with amino glycosides, are among the commonly prescribed antibiotics which are major part of empirical therapy. Because of injudicious and unnecessary use in developing countries, resistance to these drugs has become a major problem. A feature in the emergence of multidrug-resistant Gram-negative bacilli is the production of extended-spectrum b-lactamases (ESBLs) and enzymatic modification of amino glycosides, which are responsible for resistance to b-lactams antibiotics and amino glycosides, respectively [5]. CMY, CTX-M, and NDM types of b-lactamase are mostly responsible for the emerging resistance to the β-lactams antibiotics among *E. coli* [11].

E. coli possess a naturally occurring chromosomally mediated β-lactamase or plasmid mediated β-lactamases which confers resistance. These enzymes have been identified in large numbers and from different regions worldwide and are significantly detected in various E. coli strains [6]. Due to reduced antibiotic choice for infections caused by MDR-ESBL-producing bacteria, designing an empirical drug therapy is needed this antibiotic profiling will provide a guideline for proper choice of the same.

2. Materials and Methods
2.1 Isolation and identification of *E.coli*
In this study 150 strains of *E.coli* from 170 different clinical samples like urine, stool, blood, pus etc were isolated. The clinical samples were collected from hospitals and diagnostic centres in Kalaburagi region over a period of three months in 2013. Idedtification was done by culture on EMB agar. Isolation of strains was done by conventional morphological, cultural and biochemical characterisation. Standard strain of *E.coli* MTCC 443 were obtained from Medical and Phage Therapy Laboratory, department of Biotechnology, Gulbarga University, Kalaburagi.

2.2 Antbiotic susceptibility test
Antibiotic susceptibility test was done by Kirby-Bauer disk diffusion method as recommended by the Clinical Laboratory Standards Institute (CLSI) guidelines. Commercially available antibiotic disks (Himedia Labs, India) were used for antimicrobial susceptibility testing [7]. The following antibiotic disks were used, ampicillin (10 µg), nalidixic acid (100 µg), amoxicillin/clavulinic acid (20/10 µg), cefuroxime (30 µg), cefoxitin (30 µg), ceftazidime (30 µg), Cefotaxime (30 µg), imepenem (10 µg), gentamicin (10 µg), ciprofloxacin (30 µg), levofloxacin (5 µg) [8].

Inoculum of 0.5 McFarland standards turbidity was taken in a nutrient broth from isolated colony of E. coli selected from

18–24 hour agar plates. A sterile cotton swab was dipped into the inoculum suspension and inoculated on the dried surface of a Mueller-Hinton agar (MHA) plate by streaking the swab over it. Antibiotic discs were placed on agar and were incubated for 18 hours. Diameter of zone of inhibitions was measured and recorded. Strains resistant to more than four antibiotics were considered as MDR *E.coli.*

3. Result and discussion

3.1 Isolation and identification

150 strains of *E.coli* were isolated from 170 different clinical samples and identified by conventional methods. Highest number of strains was isolated from urine and least from blood sample as shown in table No 1.

3.2 Antibiotic profiles

All the isolated strains were subjected antibiogram. The resistance profile is as shown in the table No 2.

Antibiotic resistance of *E.coli* was 100% resistance to ampicillin and nalidixic acid,40% to amoxicillin/clavulinic acid, 80% to Cefotaxime, 70% to ceftazidime, 80% to ceforoxime,81% to ciprofloxacin, 40% to levoflaxacin, 20% to gentamycin and 5% resistance to imepenem was observed (Figures 1)

Table 1: Distribution of *E.coli* isolates in clinical samples and percentage of resistance.

Sl. No	Clinical samples	No. of strains isolated	% of resistance
1	Urine	70	46.6%
2	Stool	59	39.35%
3	Blood	5	4%
4	Pus	6	3.25%
5	Others	10	6.6%

Table 2: Antibiotic Susceptibility pattern of *E.coli* isolates

Antibiotic	No of resistant strains	% of resistance
Ampicillin	150	100%
Nalidixic acid	150	100%
Amoxicillin/clavulinic acid	60	40%
Cefotaxime	120	80%
Ceftazidime	105	70%
Cefuroxime	120	80%
Ciprofloxacin	120	80%
Levofloxacin	60	40%
Gentamycin	30	20%
Imipenum	7	5%

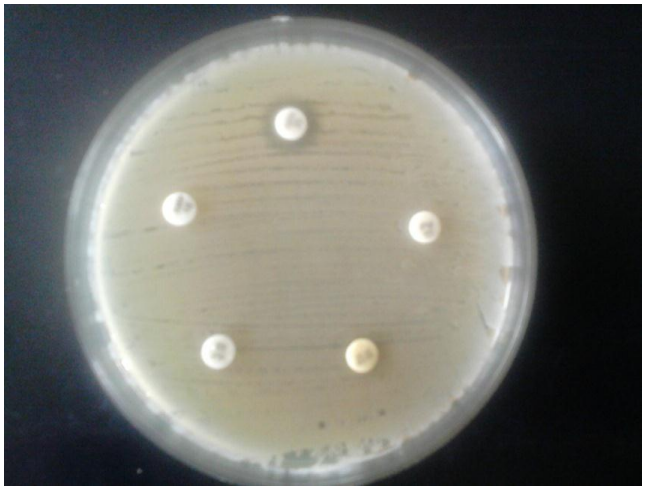

Fig 1: Antibiotic Susceptibility Pattern of E.coli Isolates

4. Discussion

As *E.coli* is commensal bacteria it becomes necessary to monitor antibiotic resistance development in this organism. *E.coli* also share human and animal host which elevates the difficulty to control the spread of multi drug resistant strains [10]. In developing country like India, with scarce economic resources it is necessary for proper evaluation before the treatment of highly resistant *E.coli* in clinical samples. The present data analysis suggests that Beta-lactams and quinolones may be no longer the choice of drug for treatment in MDR *E.coli* infections. Alternatives such as imepenem, gentamycin and amoxicillin/clavulinic acid should be considered for quick recovery of *E.coli* infections.

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