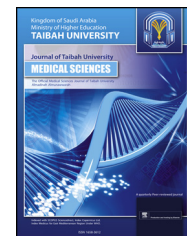




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Brief Communication

Prevalence of fluoroquinolone resistance in *Escherichia coli* in an Indian teaching hospital and adjoining communities



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Abstract

Multidrug resistant (MDR) strains of the Gram-negative pathogenic bacterium, *Escherichia coli*, particularly fluoroquinolone-resistant strains, are the major causative agents for hospital acquired (HA) infections, as well as epidemics linked to gastrointestinal (GI) and urinary tracts in the non-hygienic communities of most developing countries. The prevalence of multidrug resistance among 1642 strains of *E. coli*, isolated from clinical samples of patients with GI infections in a hospital over 39 months (November 2009–January 2013) is recorded, along with sensitivity patterns to 23 currently used antibiotics, including third-generation cephalosporins and fluoroquinolones with disc-diffusion method. A total of 1642 strains of *E. coli* were isolated from the clinical samples, of which 810 isolates were from CA samples and 832 isolates were from hospitalized patients during the study period. Of the 810 CA isolates, 567 strains were resistant to fluoroquinolone antibiotics; of the 832 HA isolates, 575 strains were fluoroquinolone-resistant, independently. Minimum inhibitory concentration values of fluoroquinolones (ciprofloxacin and levofloxacin) against the isolated *E. coli* strains confirmed the resistance in the current/coveted treatment options. Patients with other bacterial infections had relatively higher chances of becoming infected with fluoroquinolone-resistant *E. coli* strains. The data presented epitomize

the daunting state of the infection-dynamics of fluoroquinolone-resistant *E. coli* in hospitals and adjoining communities.

Keywords: *Escherichia coli*; Fluoroquinolone resistance; Hospital and community infection; Multidrug resistance

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Introduction

Suffering from several chronic uncontrollable bacterial infections often leads to terminal diseases, at least in immune-compromised/aged individuals, if the innards become infected. On analysis, it is found that drug-resistant bacteria are the causative organism of morbidity and mortality.¹ Indeed, pathogenic bacteria gain multidrug resistant (MDR) traits due to their simple genomes, and concomitantly natural consortia of bacteria help mediate their evolutionary exchanges of genetic materials.² As antibiotics are microbial in origin, targeted microbes develop resistance to the applied antibiotics intrinsically; the mutation frequency of antibiotic-resistance is recorded as one in $10^6 - 10^8$ cells.³ However, small and clean a hospital be it may, the chance of spreading pathogenic bacteria to health personnel, who often serve as reservoirs along with hospitalized patients, should be ample aside from the spread from fomites and devices.⁴ Furthermore, nosocomial infections of patients with burn and surgical injuries, as well as life-threatening urinary tract infections or even enteropathogenic episodes, frequently lead to

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bacteraemia/septicaemia.^{5,6} Any infection in a patient must be controlled forthwith, and the surveillance of a group of pathogens can be undertaken at a hospital for the estimation required for assurance on prescribed antibiotics. The evolutionary capabilities of a few pathogenic Gram-negative (GN) bacteria are so versatile that notorious pan-drug resistant (PDR; some strains of these bacteria are resistant to almost all contemporary antibiotics) strains have emerged; they are identified mainly as *Escherichia coli*, *Enterobacter aerogenes* and *Klebsiella pneumoniae*.^{7,8} The outbreak of accredited MDR bacterial strains and their rapid spread affects the cost of hospitalization and the public health sector, leading to the urgency behind the implementation of some avant-garde drugs as antimicrobials.

Fluoroquinolones are broad-spectrum antibiotics that are used to treat several GN and Gram-positive (GP) bacterial infections. Since 1960, fluoroquinolones have become prevalent in the treatment of urinary, respiratory, gastrointestinal, urogenital, intra-abdominal, and skin infections. *E. coli* infections, especially in the urinary and gastrointestinal tracts, are frequently addressed with fluoroquinolones. The emergence of quinolone-resistance during treatment was first reported in *Staphylococcus aureus*, particularly along with the methicillin-resistant *S. aureus* (MRSA) and *Pseudomonas aeruginosa*. Fluoroquinolone resistance emerged rapidly and spread to Gram-positive (GP) and GN bacteria in hospitals,⁹ with minimal inhibitory concentration (MIC) values inhibiting 90% of pathogen growth specifically over a broad range from ≤ 0.015 up to ≥ 128 mg/L fluoroquinolones.⁹ Drug-resistant subpopulations of pathogens became prevalent two decades ago and have remained almost unnoticed. Recent surveillance studies demonstrated that fluoroquinolone resistance rates have continued to increase, affecting patient management and necessitating the need for changes in the current treatment guidelines.¹⁰

The aim of this study was to evaluate the occurrence of fluoroquinolone resistance, particularly in *E. coli* causing nosocomial and community-acquired infections in patients over a period of 39 months (November 2009–January 2013). The antibiotic resistance patterns of these isolates against 23 antibiotics along with fluoroquinolones used in the study is crucial to hospital management, as empiric therapy is often needed for *E. coli* infections in the GI and urinary tracts.

Materials and Methods

Isolation, identification and antibiotic sensitivity of E. coli isolates

IMS and Sum hospital is a philanthropic teaching hospital situated in Bhubaneswar, the capital city of Odisha state, India. Patients from all walks of life, from slum-dwellers to socialites, attend this hospital. Clinical samples (urine and stool samples) collected from the outpatient department (OPD) were taken as community acquired (CA) samples. Collected clinical samples from wards, cabins, intensive care units (ICU) and neonatal intensive care units (NICU) of the hospital were referred to as hospital acquired (HA) samples for the isolation of *E. coli* strains. Of the total 12,846 clinical samples obtained over a period of 39 months,

only 7194 samples yielded both GN and GP bacteria. GN bacteria were cultured on MacConkey (MC) agar and cysteine deoxycholate electrolyte deficient (CLED) agar (HiMedia, Mumbai). The *E. coli* strain MTCC strain number 443, was used as the reference strain for all experiments. Antibiotic susceptibility tests were performed using Kirby–Bauer's disc diffusion method.¹¹ This study was performed after being approved by the institutional ethical board.

Determination of MIC values of fluoroquinolones antibiotics

The MIC values of two frequently used fluoroquinolone antibiotics, ciprofloxacin and levofloxacin against 50 drug-sensitive strains and 50 resistant strains of *E. coli* were determined by micro-broth dilution method using 96-well microtiter plates, as described elsewhere.¹² The results were interpreted using the standard breakpoint values suggested by the clinical and laboratory standards institute and the 2014 guidelines of the European committee of antimicrobial susceptibility testing.^{13,14}

Statistical analysis

Statistical analysis was performed using the Statistical Package for Medical Science version 17.0 (SPSS Inc., IL) and Microsoft Excel.

Results

Isolation and identification of E. coli strains

A total of 1642 strains of *E. coli* were isolated from clinical samples, of which 810 isolates were from CA samples and 832 isolates were from hospitalized patients, taken during the study period. Of the 810 CA isolates, 567 strains were resistant to fluoroquinolone antibiotics. Similarly, of the 832 HA strains, *E. coli* 575 strains were fluoroquinolone-resistant, independently. When grown on MC agar, medium-sized colonies coloured bright pink (due to inherent lactose fermentation) were seen, and when grow on CLED agar, large, round yellow coloured colonies were seen, confirming the presence of *E. coli*.

Antibiotic susceptibility pattern of E. coli isolates

The antibiotic susceptibility patterns of isolated *E. coli* strains were studied over the period of 39 months using 23 antibiotics. HA *E. coli* strains were found to be highly resistant to, in descending order, gentamicin (98%), cefepime (82%), nitrofurantoin (94%) and norfloxacin (87%) and were least resistant to imipenem (21%). Similarly, CA *E. coli* isolates were resistant to co-trimoxazole (94%), followed by gentamicin (91%), ciprofloxacin (89%), amikacin (87%) and cefotaxime (87%), and were least resistant to imipenem (17%) and levofloxacin (25%) (Table 1). The resistance percentage values clearly support that there is high occurrence of ESBL and fluoroquinolone-resistant strains of *E. coli* isolates in hospital and community settings. Moreover, resistance to aminoglycosides and carbapenems (imipenem and meropenem) were recorded, which would further complicate the treatment regimen for an infection, as

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