Prevalence of antimicrobial resistant *E. coli* in water reservoirs

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**ABSTRACT**

**Background:** The antibiogram of the *E. coli* isolated from the different water bodies contaminated by undefined fecal sources have been investigated. **Aims and Objectives:** To isolate *E. coli* from water sources and to evaluate their antimicrobial resistance/sensitivity pattern. **Results:** A total of 296 *E. coli* strains isolated from pooled samples. Approximately 50% of these isolates expressed resistance to one or more antimicrobials during the study. High degree of resistance was recorded to Ceftazidime followed by Co-trimoxazole, however isolates were also sensitive to Kanamycin and Streptomycin. **Conclusion:** Isolation of drug resistant *E. coli* in water reservoirs highlights the importance of water disinfection before its utilization for drinking purpose.

**KEYWORDS:** Antibiotic susceptibility test; *Escherichia coli*; Multiple drug resistance

**INTRODUCTION**

Waterborne diseases are not unknown to us. Food and waterborne diseases are responsible for highest number of mortality in children globally. There are several pathogens which may cause gastrointestinal illness through contaminated water; however *E. coli* is the most common agent of Diarrhoea. It is prevailing ubiquitous in nature and is a normal inhabitant of the gastrointestinal tract of almost all animals and humans\(^1\)\(^-\)\(^2\). In reports, aquatic ecosystems of many countries reported with virulent *E. coli* species and it is indicator of faecal contamination of water\(^3\). Sporadic studies on occurrence of virulent *E. coli* strains and their antimicrobial resistance pattern isolated from aquatic reservoirs has been documented earlier \(^4\)\(^-\)\(^5\). Information on detection of *E. coli* from water reservoirs nearby Nagpur found scanty, therefore in the present study, water reservoirs around Nagpur were screened for the presence of the multidrug resistant *E. coli*.

**MATERIALS AND METHODS**

**Selection of Water Bodies**

Water samples were collected from different reservoirs by deep method. A total of 5 samples were collected at frequency of three days from the water bodies of Nagpur such as Salburdi River, Kanhan River, Kamptee River, Gorewada Lake, Khindsi Lake and Zilpi Lake. About 100 ml of water sample collected from five different points of each reservoir and pooled to 500 ml.

**Processing of Sample**

The samples (500 ml) were immediately passed through the membrane filtration, in an order to concentrate the available microflora available in water. The membranes from each source were transferred to the specific media such as McConkeys, Eosin Methylene Blue and Endo agar for the isolation of *E. coli*. After 24 hours of incubation, *E. coli* were identified based on colony characteristics, staining and biochemical tests. Confirmed isolates were tested for antibiogram.

**Antibiogram**

The *E. coli* strains isolated from selected water bodies were screened for the 16 antibiotics sensitivity, which are generally being used in the therapies using Kirby- Bauer (KB) antibiotic sensitivity test. The Octa discs supplied by HiMedia with the product codes as OD057R-1PK having the antibiotics: Ceftazidime (30µg), Ciprofloxacin (5µg), Cefotaxime (30µg), Nalidixic acid (30µg), Nitrofurantoin (300µg), Nor floxacin (10µg), Netrillin (30µg), Ofloxacine (5µg) and OD058R having antibiotics: Chloramphenicol (30µg), Ampicillin (10µg), Tetracycline (30µg), Gentamycin (10µg), Kanamycin (30µg), Co-trimoxazole (25µg), Amikacin (30 µg) and Streptomycin (10µg) were used in assay.

By using inoculum of 1.0 x 10\(^8\) CFU/ml of *E. coli* maintained as per standard McFarland turbidity scale was spread on the Muller Hinton agar and allowed to soak for 10 minutes and then the disc was placed...
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RESULT

Prevalence of E. coli in Water Bodies

With total isolation of 296 strains of E. coli from different water bodies, prevalence was recorded more in Gorewada Lake 95 (32.09%), for Salburdi River 70 (23.64%) for Kanhan River 38 (12.83%), for Kamptee River 35 (11.82%), for Zilpi Lake 31 (10.47%) and for Khindsi Lake 27 (9.12%) as aseptically on agar. Plates were incubated at 37°C for 24 hours. Results were interpreted as per the guidelines of manufacturer (Himedia) of antimicrobial discs. Based on the overall results, these isolates were classified as either resistant or susceptible.

Antibiotic Sensitivity and Multi Drug Resistance

Results highlighted that several E. coli strains have showed antibiotic resistance against all sixteen drugs but in variable combinations. Based on the total resistance in strains, highest resistance was recorded for Ceftazidime (52.36%) followed by Co-trimoxazole (49.66%), Ciprofloxacin (38.51%), Ampicillin (34.80%), Cefotaxime (32.77%), Nitrofurantoin (25.00%), Norfloxacin (20.95%), Nitrofurantoin (17.57%), Chloramphenicol (16.89%), Netilin (16.55%), Gentamicin (13.18%), Tetracycline (11.82%), Amikacin (11.82%), Kanamycin (9.12%) and Streptomycin (5.74%).

Along with the particular antibiotic resistance several strains have showed combination of multidrug resistance as depicted in Table 1. Results were interpreted as per the guidelines of manufacturer (Himedia) of antimicrobial discs. Based on the overall results, these isolates were classified as either resistant or susceptible.

![Table 1: Multidrug resistance pattern highlighted by number of E. coli strains](image)

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Drug resistance pattern</th>
<th>No. of isolates expresses pattern</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>OF, AMP, NIT, CTX, NA, K</td>
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<tr>
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<td>CAZ, COT, CIP, CTX, AMP, K</td>
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<tr>
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<td>4</td>
<td>CAZ, COT, NIT, CTX, AK, OF</td>
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<tr>
<td>5</td>
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</tr>
<tr>
<td>6</td>
<td>CIP, NIT, AMP, GEN, OF, C</td>
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<td>8</td>
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<td>9</td>
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<tr>
<td>16</td>
<td>AMP, NIT, CTX, C, GEN</td>
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</tbody>
</table>

Ceftazidime (CAZ), Ciprofloxacin (CIP), Cefotaxime (CTX), Nitrofurantoin (NIT), Nitrofurazone (NX), Netilin (NET), Ofloxacin (OF), Chloramphenicol (C), Ampicillin (AMP), Tetracycline (TE), Gentamicin (GEN), Kanamycin (K), Co-trimoxazole (COT), Amikacin (AK), Streptomycin (S)

DISCUSSION

The antibiogram of the E. coli isolated from the different water bodies contaminated by undefined fecal sources have been investigated. Their occurrence in water and harboring of antimicrobial resistance is alarming from public health view if water left untreated. Present study highlighted that these water bodies could act as pool for emergence of multidrug resistant strains of E. coli. Earlier report highlighted that such E. coli strains available in water bodies posses the resistance plasmid and hence pose a serious threat to the consumer by such pathogens.

The E. coli population available in the water samples showed the defined variations in number of isolates per unit volume sampled from different sources. This variation in load recorded may be due to the distance between the points of contamination and sampling point along with the dilution factor and water flow rate when taken together in consideration, which decides the rate of colony forming units available in the water. Similar observations in consistent with the present study have been documented in terms of prevalence of pathogens in different drinking water resources in Makkah City.

The microbial ecosystems of humans, swine, cattle, and food are undoubtedly remains connected; therefore it is tough to pinpoint the source of the antimicrobial resistance that we observed. E. coli have been reported by many workers to showcase the resistance against tetracycline, sulfamethoxazole, cephalothin, and ampicillin. Similar results were recorded in our study for the drug resistance in which maximum resistance recorded for Ceftazidime, Co-trimoxazole, Ciprofloxacin, Cefotaxime and Nitrofurantoin. Similar results were recorded earlier also in which the pattern of susceptibility and resistance of the tested drug showed defined variations in E. coli.

Multidrug resistant E. coli has become a major public health concern globally, causing a failure in treatment with consequent huge health burden. In our study, multi drug resistance has been recorded for the E. coli and all 16 drugs showing increasing resistance indicated that even new generation antibiotics could become ineffective in controlling E. coli infection. Similar results have been reported earlier in which the high resistance rates in MDR E. coli isolates to the first-line oral antimicrobial agents such as amoxicillin, cefuroxime, trimethoprim sulfamethoxazole, tetracycline, nalidixic acid and amoxicillin-clavulanate has been recorded. These findings represent an alarming rise in resistance of E. coli and comparable to other studies done in countries such as Sudan and elsewhere.

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