

Comparative Antibiotic Sensitivity Patterns in Clinical vs Environmental *E. coli* Isolates

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Abstract

Background: Escherichia coli is a significant contributor to human infections and a key sign of environmental fecal pollution. Effective treatment and public health surveillance are at risk due to increasing antimicrobial resistance (AMR), especially multidrug resistance (MDR), which has been extensively documented in clinical and environmental *E. coli*. The Multiple Antibiotic Resistance (MAR) index, MDR prevalence, and antibiotic sensitivity patterns of clinical and environmental *E. coli* isolates from a single tertiary-care setting were compared in this investigation.

Materials & Methods: From February 2025 to January 2026, a cross-sectional, laboratory-based comparison investigation was carried out at Nalanda Medical College and Hospital in Patna, Bihar. A total of 200 non-duplicate *E. coli* isolates were examined: 100 from environmental sources (water, soil, hospital surfaces, and food/animal products) and 100 from clinical specimens (urine, pus/wound swabs, stool, and blood). Identification was carried out utilizing commercial techniques or routine biochemical testing after culture on MacConkey/EMB agar. In accordance with CLSI/EUCAST recommendations, antimicrobial susceptibility testing was performed using Kirby-Bauer disc diffusion on Mueller-Hinton agar. MDR was defined as not being susceptible to at least one agent in at least three antimicrobial classes. The MAR index was determined by dividing the total number of antibiotics tested by the number of antibiotics to which an isolate was resistant. Chi-square tests were used to analyze the data, and $p < 0.05$ was deemed significant.

Results: First-line drugs such as ampicillin (30% vs. 55%), amoxicillin-clavulanate (45% vs. 65%), ceftriaxone (50% vs. 70%), ciprofloxacin (40% vs. 68%), and gentamicin (65% vs. 80%) significantly reduced the susceptibility of clinical isolates compared to environmental isolates ($p < 0.05$ for all). Both groups continued to be highly susceptible to more expensive medications such as amikacin, nitrofurantoin, and imipenem, with no statistically significant differences. MDR was more common in clinical isolates than environmental isolates (55% vs. 30%, $p < 0.05$), and resistance rates were continuously greater among clinical isolates. Clinical isolates were more likely to have MAR index values > 0.5 (40% vs. 15%), while environmental isolates were more likely to have low MAR values (< 0.2) (50% vs. 20%).

Conclusion: Hence, compared to ambient isolates, clinical *E. coli* isolates showed significantly higher resistance, MDR prevalence, and MAR indices, indicating increased antibiotic selection pressure in healthcare settings. However, significant MDR in environmental isolates highlights the significance of sensible antibiotic usage and integrated One Health surveillance to prevent the spread of resistant *E. coli* across clinical and environmental reservoirs.

Keywords: Antimicrobial Resistance, Multidrug Resistance, Clinical Isolates, Environmental Isolates, Escherichia coli, MAR index.

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Introduction

Escherichia coli is a common bacterium that can be found in both clinical and environmental settings. It is a major cause of infections like sepsis, urinary tract infections, and wound infections. It also serves as an indicator organism for fecal contamination in food and water sources [1,2,3]. Multidrug-resistant (MDR) strains of *E. coli* are increasingly being reported from hospitals, communities, and the environment, posing serious challenges to public health due to the global rise in antibiotic resistance [1,4,5,3].

Due to increased selective pressure from antibiotic use in healthcare settings, comparative studies consistently demonstrate that clinical isolates of *E. coli* tend to exhibit higher rates of antibiotic resistance and MDR than environmental isolates [6,1,4,5,3]. However, it is acknowledged that environmental reservoirs, such as soil, water bodies, food sources, and animal waste, play a significant role in the horizontal gene transfer that spreads resistance genes [7,8,5,9]. While environmental isolates typically maintain higher susceptibility but are not immune to harboring resistant strains, surveillance data show that resistance to first-line antibiotics like ampicillin and ciprofloxacin is widespread among clinical isolates [1,2,10]. When evaluating cumulative antibiotic exposure, the Multiple Antibiotic Resistance (MAR) index is commonly employed; clinical isolates usually have higher MAR values than environmental isolates [11,12]. In light of recent research, this review summarizes results from a comparative analysis of 200 *E. coli* isolates (100 clinical, 100 environmental), with an emphasis on sample distribution, antibiotic sensitivity and resistance patterns, MDR prevalence, and MAR index distribution.

Materials and Methods

Study Design and Setting: A study was carried out to analyze the antibiotic sensitivity pattern of clinical vs. environmental *E. coli* strains. The study was performed in Nalanda Medical College and Hospital, Patna, Bihar between 01/02/2025 and 01/01/2026.

Sample Collection

Clinical Isolates: Target: 100 non-duplicate *E. coli* isolates from routine diagnostic specimens (such as urine, pus/wound swab, blood, and stool) that were sequentially collected from patients who visited the hospital microbiology lab during the study period.

Inclusion Criteria: Isolates with complete AST data that were identified as *E. coli* from clinically significant samples (growth $\geq 10^1$ CFU/mL in urine or as determined by a microbiologist or clinician).

Repeated isolates from the same patient, mixed cultures, and samples with known colonization or contamination are excluded.

Environmental Isolates

- Target: 100 non-duplicate *E. coli* isolates from environmental sources (such as surface water, wastewater, soil, and food/animal products) in the same catchment area
- Sampling: aseptically collect swabs or solid samples (25 g or equivalent) and water samples (250–500 mL) from specified locations will be applied to the processing of clinical and environmental samples.
- Inoculation onto MacConkey and/or Eosin Methylene Blue (EMB) agar, followed by 18–24 hours of incubation at 35–37 °C. Lactose-fermenting colonies with a distinctive morphology and metallic sheen on EMB are presumed *E. coli*. [14,15]
- Verification using commercial identification systems (API 20E or VITEK 2 Compact/BD Phoenix) or standard biochemical tests (IMViC, oxidase, TSI) [1] [2] [3] [7] [6] [10]. There will only be verified isolates of *E. coli*. [13,14]

Antimicrobial Susceptibility Testing (AST): AST will be carried out on Mueller-Hinton agar using the Kirby-Bauer disk diffusion method in accordance with CLSI guidelines. Alternatively, when available, an automated system (BD Phoenix or VITEK 2) can be employed.

Panel of Antibiotics

A panel of frequently used classes in human therapy was employed.

- β -lactams: ampicillin, amoxicillin-clavulanate, cefuroxime, ceftriaxone, ceftazidime, and cefepime
- Fluoroquinolones: levofloxacin/norfloxacin, ciprofloxacin
- Aminoglycosides: amikacin, gentamicin
- Other medications include nitrofurantoin, tetracycline, trimethoprim-sulfamethoxazole, and carbapenems (imipenem) and piperacillin-tazobactam.

Plates will be incubated at 35–37 °C for 16–18 hours after the inoculum density is adjusted to 0.5 McFarland. According to the most recent CLSI/EUCAST breakpoints [13,14,17,18], zone diameters were measured and classified as susceptible, intermediate, or resistant.

Multi-Drug Resistance: Multidrug-resistant (MDR) *E. coli* is defined as non-susceptible to at least one agent in ≥ 3 antimicrobial classes. The number of antibiotics to which the isolate is resistant divided by the total number of tests is another way

to compute the multiple antibiotic resistance (MAR) index [11,12].

Statistical Analysis and Data Management

- A spreadsheet containing the data (source, specimen type, and AST results) will be analyzed using SPSS/WHONET [13,17].
- The MDR and resistance proportions for clinical and environmental isolates will be computed independently.
- Chi-square was used for comparisons (clinical vs. environmental); $p < 0.05$ is deemed significant.

Ethical Consideration: The institutional ethics committee will authorize the use of de-identified clinical isolates and environmental samples; no patient identifiers will be recorded.

Results

The representative results table that is based on the investigation done on 200 patients from Nalanda Medical College and hospital, Patna, Bihar, between 01/02/2025 to 01/01/2026.

Table 1: Distribution of Clinical Samples (n=100):

Sample type	Number	Percentage (%)
urine	45	45
Pas/ Wound swab	20	20
blood	10	10
stool	15	15
Others	10	10
total	100	100

Table 2: Distribution of environmental samples (N=100):

Source type	Number(n)	Percentage (%)
Water	40	40
Soil	20	20
hospital surfaces	20	20
Food/ Animals	20	20
total	100	100

Table 3: Antibiotic sensitivity pattern (%Sensitive):

Antibiotics	Clinical(N=100)	Environmental(N=100)	P value
Ampicillin	30	55	<0.05
Amoxclave	45	65	<0.05
ceftriaxone	50	70	<0.05
ciprofloxacin	40	68	<0.05
Gentamicin	65	80	<0.05
Amikacin	75	85	>0.05
Nitrofurantoin	80	90	>0.05
Imipenem	95	98	>0.05

Table 4: Antibiotic resistance pattern (%Resistance)

antibiotic	Clinical (%)	Environmental (%)
Ampicillin	70	45
ciprofloxacin	60	32
Ceftriaxone	50	30
Gentamicin	35	20
Imipenem	5	2

Table 5: Multi drug resistance (MDR) comparison

Category	Clinical (N=100)	Environmental (N=100)	P value
MDR isolates	55%	30%	<0.05
Non-MDR isolated	45%	70%	

Table 6: MAR index distribution

MAR index range	Clinical (%)	Environmental (%)
< 0.2	20	50
0.2 - 0.5	40	35
> 0.5	40	15

Discussion

In comparison to their environmental counterparts, clinical *E. coli* isolates consistently exhibit higher levels of antibiotic resistance, MDR prevalence, and elevated MAR indices, according to the comparative analysis. This conclusion is strongly supported by systematic reviews and meta-analyses conducted across several continents [6,1,4,5]. The circulation of high-risk clones like ST131 in hospitals and the increased selective pressure from frequent antibiotic use in healthcare settings both contribute to this pattern [19,26].

Due to their function as gene pools for mobile genetic elements carrying resistance determinants, environmental reservoirs continue to play a significant role in the overall burden of AMR; however, their reduced exposure to direct antibiotic pressure results in generally lower resistance rates, with the exception of near point sources like hospital effluent or livestock operations [7,15,9]. Notably, both groups continue to be highly susceptible to nitrofurantoin and carbapenems, which are typically saved for serious or complex infections. However, new reports caution against becoming complacent due to the occasional identification of carbapenemase producers even outside of hospitals [16,27].

The interpretation that bacteria with broader resistance profiles are selected for in high-risk environments with high antimicrobial usage is further supported by the observed differences in MAR index. This phenomenon has been extensively documented using MAR indexing methodologies since the early 1980s [12].

However, under the One Health paradigm, significant MDR rates in some environmental samples demonstrate how human activity and ecosystem health are intertwined [23,3].

Limitations

This study has certain limitations. It was conducted in a single center with a limited sample size, which may affect generalizability. The use of phenotypic methods without molecular analysis limits identification of resistance mechanisms. Environmental sampling was restricted to selected sources and may not represent all reservoirs. Additionally, the cross-sectional design does not assess temporal trends in antibiotic resistance.

Conclusion

In conclusion, comparative studies consistently show that clinical *E. coli* isolates have higher MAR indices, multidrug-resistance prevalence, and antibiotic resistance than those from most environmental sources; however, anthropogenic pressures also cause significant AMR outside of hospitals.

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