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Antibiotic Susceptibility and Prevalence Escherichia coli Isolation Pattern from Urinary Tract Infections in a Central Indian Tertiary Care Hospital: A Hospital-Based Cross-Sectional Investigation

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Abstract

Background: Escherichia coli continue to be the most prevalent causal organism for UTIs, which are among the most prevalent bacterial infections observed in typical clinical settings. The dependability of traditional empirical regimens has been undermined by the increasing appearance of multidrug-resistant uropathogens. To support sensible prescribing, region-specific antibiograms are required, especially in central India where there is a dearth of available data.

Objectives: To ascertain E. coli frequency in urine culture isolates and to describe its profile of antibiotic sensitivity, including the prevalence of multidrug resistance (MDR).

Methods: Over the course of eighteen months, a cross-sectional research was carried out at the microbiology department of a tertiary care teaching hospital in Indore, Madhya Pradesh. Clinically suspected UTI patients provided 350 mid-stream clean-catch urine specimens. The calibrated loop technique was used to process the specimens on CLED and MacConkey agar, and ≥10 CFU/mL was considered serious bacteriuria. Standard biochemical processes were used to identify E. coli.Mueller-Hinton agar was tested for antibiotic susceptibility using the Kirby-Bauer disc diffusion method in accordance with CLSI M100 (2023) criteria. Non-susceptibility to at least one agent in three or more antimicrobial classes was the definition of MDR according to the Magiorakos criterion.

Results: 97 (27.7%) of the 350 specimens that were processed had substantial bacteriuria. Pseudomonas aeruginosa (9.3%) and Klebsiella pneumoniae (11.3%) were the next most common

isolates, with E. coli accounting for 60 out of 97 culture-positive samples (61.9%). 62.9% of all positive cultures came from people between the ages of 20 and 49. The 60 E. Coli isolates showed the greatest levels of resistance to ampicillin (86.7%), cotrimoxazole (66.7%), ciprofloxacin (63.3%), and third-generation cephalosporins (46.7 to 60.0%). The best activity was maintained by amikacin (21.7%), nitrofurantoin (15.0%), piperacillin-tazobactam (13.3%), and imipenem (6.7%). Outpatient isolates had a slightly higher MDR rate (97.6%) than inpatient isolates (89.5%), with 57 out of 60 isolates (95.0%) meeting the criteria for multidrug resistance.

Conclusions: Resistance to commonly prescribed oral agents has reached a level at which empirical use of ampicillin, cotrimoxazole or fluoroquinolones is no longer justifiable in this setting. While carbapenems, aminoglycosides, and β-lactamase-inhibitor combos ought to be preserved for difficult or hospital-acquired infections, nitrofurantoin is still a reasonable option for simple lower UTIs in the community. A functional antimicrobial stewardship program and routine urine culture prior to starting treatment are now required, not optional.

Keywords: Escherichia coli, multidrug resistance, tertiary care hospital,urinary tract infection, central India, antibiotic resistance, and antimicrobial stewardship.

1. Introduction

One of the most frequent bacterial illnesses that doctors treat is UTIs, in hospital wards as well as the community. Roughly 150 million episodes are reported worldwide every year, and the actual figure is almost certainly higher in low- and middle-income settings where many patients self-medicate before reaching a laboratory [1,2]. In India the burden is amplified by easy access to over-the-counter antibiotics, irregular prescribing practices and uneven implementation of infection-control measures [3,4].

Among the organisms responsible, Escherichia coli has held its position as the dominant uropathogen for several decades. Reports from different parts of the world place its share between 55% and 80% of all culture-positive UTIs [5-7]. Its prevalence is not accidental. UropathogenicE. coli (UPEC) carries a battery of virulence determinants, including type 1 and P fimbriae, haemolysins, capsular antigens and iron-acquisition systems, that allow it to ascend the urinary tract, attach to uroepithelial cells and resist host defences [8,9]. What is changing, however, is not the organism but its antibiotic profile.

Over the last two decades, resistance to first-line oral antibiotics has climbed steadily. Ampicillin, once a reliable choice, is now resisted by over 80% of urinary isolates in most Indian centres [10,11]. Resistance to cotrimoxazole regularly exceeds the 20% threshold beyond which Infectious Diseases Society of America (IDSA) guidelines advise against its empirical use [12]. The fluoroquinolones, which replaced these agents in the 1990s, are following the same trajectory, with ciprofloxacin resistance now commonly reported between 50% and 70% in Indian uropathogens [4,13]. Third-generation cephalosporins are increasingly affected by ESBL production, plasmid-encoded enzymes that break down these medications' oxyimino side chain[14,15].

Multidrug resistance (MDR), which Magiorakos and Colleagues characterise acquired resistance to at least one agent in three or more categories of antimicrobials, significantly complicates the situation [16]. MDR E. coli narrows therapeutic options dramatically and often forces clinicians to use carbapenems or aminoglycosides, drugs that were previously held in reserve. The cumulative result is rising treatment failure, prolonged hospitalisation and avoidable mortality [17,18].

Local antibiograms are critical in this scenario because resistance patterns vary not only between countries but also between hospitals within the same city. Empirical regimens chosen from textbooks or from data more than a few years old may already be obsolete by the time they reach the bedside. Central India, despite its large population and several busy referral hospitals, is underrepresented in the published literature on uropathogen resistance, and the few studies available come from limited geographical pockets [4,11,19].

Therefore, we conducted this study with the following objectives: first, to ascertain the prevalence of E. coli among patients with UTIs who had positive urine cultures in a tertiary care facility in central India; and second, to characterise the isolates' antibiotic susceptibility profile against a panel of agents that are clinically relevant.; and third, to estimate the burden of multidrug resistance and identify drugs that still retain useful activity. The findings are intended to inform empirical prescribing at the institutional level and to add to the regional surveillance data on antimicrobial resistance.

2. Material and Methods

2.1 Study design

The Department of Microbiology at IMCHRC, Indore (M.P., India), which is connected to Malwanchal University, was the site of this prospective, cross-sectional observational study. The hospital is a tertiary care teaching facility with 1,200 beds that serves people from various nearby rural regions as well as metropolitan Indore. The investigation was conducted over a period of eighteen months.

2.2 Ethics

The study was carried out in compliance with the 2013 amendment of the Declaration of Helsinki [20], and the Institutional Ethics Committee authorised the protocol prior to its implementation. Each participant, or their parent or legal guardian if they are younger than eighteen, provided written informed permission after being told of the study's goals and methods in a language they could comprehend. Prior to data processing, patient identities were eliminated, and confidentiality was upheld at all times.

2.3 Sample size and study population

The method of successive sampling was applied. Three hundred and fifty (n = 350) individuals with clinical signs indicative of a urinary tract infection (UTI) who were either admitted to the wards or attended the outpatient department across all age groups and genders were included. Dysuria, urgency, suprapubic discomfort, costovertebral angle soreness, temperature over 38°C, haematuria, cloudy urine, and, in older patients, unexplained altered sensorium were among the clinical symptoms taken into consideration.

The research excluded patients who had taken any antibiotics within the preceding 72 hours, those who were known to be receiving chemotherapy, and those who refused to provide their permission.

2.4 Specimen collection and processing

Each patient was provided with a sterile wide-mouthed leak-proof container and instructed in the mid-stream clean-catch method. Samples from patients with catheters were aspirated aseptically from the sampling port after being cleaned with 70% isopropyl alcohol. When this was not possible, specimens were chilled at 4°C and processed within four hours after collection. Otherwise, specimens were transferred to the laboratory within 30 minutes.

A 0.001 mL platinum loop was used in the calibrated loop procedure for quantitative urine culture. After being inoculated on MacConkey and CLED agar, the specimens were incubated aerobically at 37°C for 18 to 24 hours [21]. Colony counts of $\geq 10^3$ colony-forming units (CFU)/mL of a single organism were considered significant bacteriuria. Counts between 10^4 and 10^5 CFU/mL in symptomatic patients, or counts above 10^5 CFU/mL of more than two organisms, were considered insignificant or contaminated and excluded from further investigation.

2.5 Identifying the isolates

Colony morphology, Gram staining, and a conventional panel of biochemical reactions, such as methyl red, indole, citrate utilisation, urease, triple sugar iron, motility, and lysine decarboxylase,Voges-Proskauer were used to identify the isolates [22]. E. coli was identified by the characteristic IMViC pattern (++ - -), positive motility, and lactose fermentation with metallic sheen on EMB agar where applicable.

2.6 Testing of antibiotic susceptibility

In accordance with Clinical and Laboratory Standards Institute (CLSI) M100 (2023) guidelines [23], susceptibility testing was conducted using the Kirby-Bauer disc diffusion technique on Mueller-Hinton agar. A new overnight culture was used to create a 0.5 McFarland suspension of the experimental organism, which was then lawn-inoculated in 15 minutes. Using a disc dispenser, commercial antibiotic discs (HiMedia Laboratories, Mumbai, India) were applied to the agar surface with a minimum center-to-center spacing of 24 mm. For 16 to 18 hours, the plates underwent aerobic incubation at 37°C. Using a sliding calliper, zone diameters were assessed and categorised using CLSI 2023 breakpoints as susceptible (S),resistant (R), or intermediate (I).

Ampicillin (10 µg), ceftazidime (30 µg),cefotaxime (30 µg), ceftriaxone (30 µg), ciprofloxacin (5 µg), norfloxacin (10 µg), nitrofurantoin (300 µg), imipenem (10 µg), piperacillin-tazobactam (10 µg), amikacin (30 µg),amoxicillin-clavulanate (20/10 µg), and cotrimoxazole (1.25/23.75 µg)

2.7 Multidrug resistance definition

According to the worldwide expert suggestion of Magiorakos et al., developed resistance to a minimum of one agent across three or more antimicrobial categoriesis the definition of multidrug resistance [16]. Aminopenicillins, β-lactam/β-lactamase inhibitor combinations, carbapenems, aminoglycosides, fluoroquinolones, extended-spectrum cephalosporins, folate pathway inhibitors, and nitrofurans were the pertinent categories that applied to E. coli for the purposes of this study. Intermediate results were not counted as non-susceptible for the purpose of MDR classification.

2.8 Quality control

Three reference strains: Pseudomonas aeruginosa ATCC 27853, Klebsiella pneumoniae ATCC 700603, and Escherichia coli ATCC 2592 -as listed in CLSI M100 [23] were used for quality control. Reference strains were run on every new batch of Mueller-Hinton agar and on every new lot of antibiotic discs, and at least weekly thereafter. Out-of-range zone diameters triggered an investigation, repeat testing and corrective action before patient isolates were reported.

2.9 Statistical analysis

A randomly chosen 10% of the records were independently rechecked for transcription mistakes once the data was input into Microsoft Excel 2019. For analysis, the cleaned dataset was imported into IBM Corp., Armonk, New York, USA; IBM SPSS Statistics for Windows, version 27. For every variable, Descriptive statistics were calculated, such as frequencies and percentages for categorical variables and means with standard deviations for normally distributed continuous data. When expected cell frequencies were less than five, the Pearson chi-square test or Fisher's exact test were utilised to investigate relationships between categorical variables. A two-tailed p value of less than 0.05 was considered to be statistically significant.

3. Results

3.1 Demographic profile

Over the course of the 18-month period, 350 patients were enrolled (213 females, 60.9%; 137 males, 39.1%), resulting ratio in a male &female is 1:1.55. They were 40.2 ± 16.8 years old on average and their ages varied from 2 to 90. The 30 to 49-year age band contributed the largest share of patients (28.3%). Two hundred and thirteen patients (60.9%) were recruited from the outpatient department, while 137 (39.1%) were inpatients.

3.2 Culture positivity

Of the 350 urine specimens processed, 97 (27.7%) showed significant bacteriuria in figure 1. Culture positivity was almost identical between sexes, with 39 of 137 male specimens (28.5%) and 58 of 213 female specimens (27.2%) yielding growth ($\chi^2 = 0.064$; $p = 0.800$),df = 1;Table 1 and Figure 1 provide the age breakdown.

Table 1. Distribution of E. Coli isolates and culture-positive patients by age (n = 350).

Age group (years)	Total examined (n)	Culture +ve (n)	Positivity (%)	E. coli isolated (n)
0-9	12	3	25.0	2
10-19	33	6	18.2	4
20-29	53	18	34.0	14
30-39	73	22	30.1	11
40-49	76	21	27.6	14
50-59	54	12	22.2	6
60-69	31	12	38.7	7
≥70	18	3	16.7	2
Total	350	97	27.7	60

The 20 to 49-year band accounted for 61 of 97 positive cultures (62.9%), while the highest positivity rate was recorded in the 60 to 69-year group (38.7%).

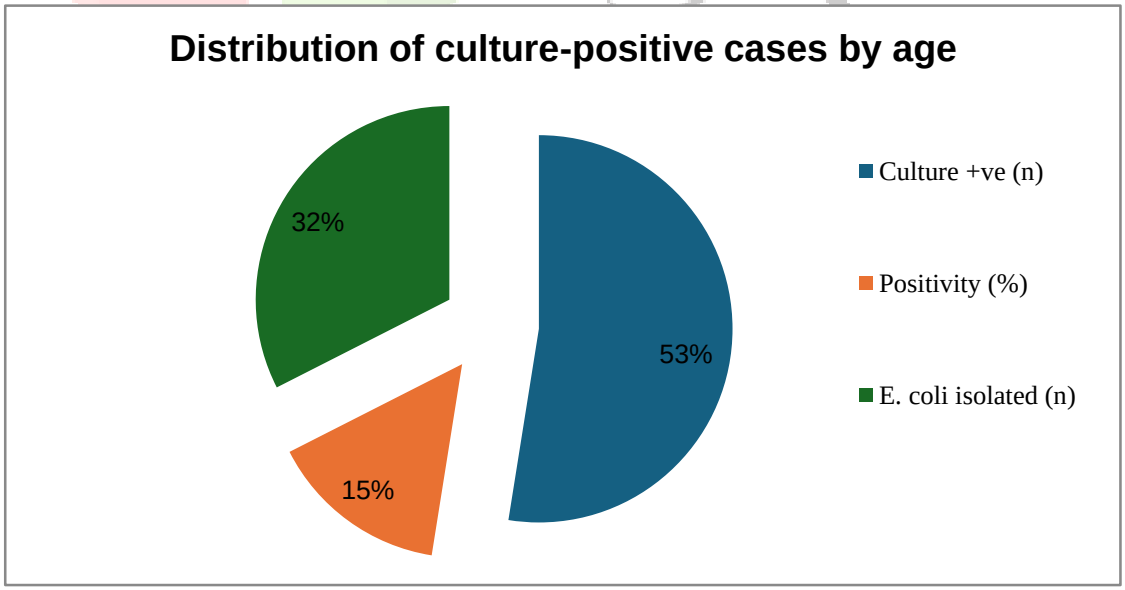

Figure 1. Distribution of E. Coli isolates and culture-positive patients by age

3.3 Uropathogen spectrum

E. coli was most common isolate found in 60 (61.9%) of the 97 culture-positive specimens. Of the 97 isolates, 92 (94.8%) were gram-negative organisms. Table 2 and Figure 2 show the distribution of organisms.

Table 2. Distribution of uropathogens within culture-positive specimens (n = 97).

Organism	Number isolated (n)	Percentage (%)
Escherichia coli	60	61.9
Klebsiella pneumoniae	11	11.3
Pseudomonas aeruginosa	9	9.3
Proteus mirabilis	4	4.1
Enterococcus species	3	3.1
Citrobacter species	3	3.1
Enterobacter species	3	3.1
Staphylococcus aureus	3	3.1
Candida species	1	1.0
Total	97	100.0

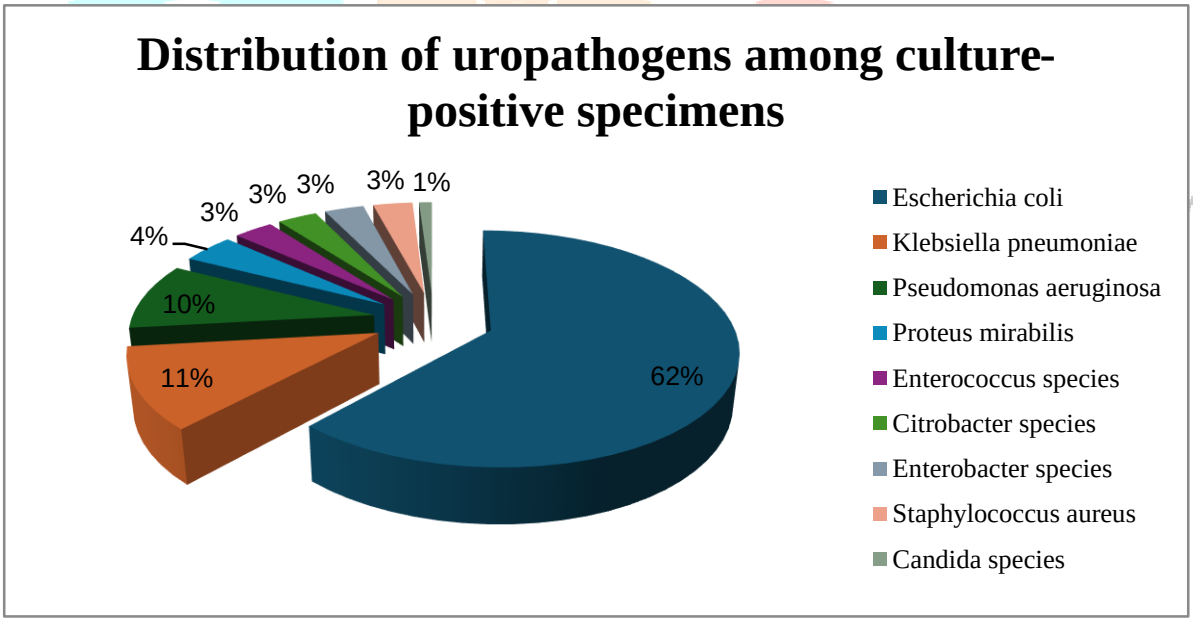

Figure 2. Distribution of uropathogens among culture-positive specimens

3.4 E. Coli's profile of antibiotic susceptibility

Testing the 60 E. Coli isolates' susceptibility to the 13-drug panel revealed a sharply polarised pattern. Resistance to ampicillin was almost complete at 86.7%, while only 5.0% of isolates were fully susceptible. Cotrimoxazole resistance reached 66.7%, well above the IDSA threshold beyond which empirical use is discouraged. Ciprofloxacin and norfloxacin retained activity against just one in four isolates each. Third-generation cephalosporins were resisted by 46.7% to 60.0% of isolates.Imipenem, however, had the maximum level of activity, with 88.3% susceptibility and only 6.7% resistance. Piperacillin-tazobactam (81.7% susceptible), nitrofurantoin (80.0%) and amikacin (71.7%) also retained reliable activity. Table 3 and Figure 3 show the whole antibiogram.

Table 3. E. Coli isolates' pattern of antibiotic susceptibility (n = 60)

Antibiotic (disc strength)	Susceptible n (%)	Intermediate n (%)	Resistant n (%)
Ampicillin (10 µg)	3 (5.0)	5 (8.3)	52 (86.7)
Amoxicillin-clavulanate (20/10 µg)	28 (46.7)	4 (6.7)	28 (46.7)
Cefotaxime (30 µg)	23 (38.3)	1 (1.7)	36 (60.0)
Ceftriaxone (30 µg)	24 (40.0)	8 (13.3)	28 (46.7)
Ceftazidime (30 µg)	23 (38.3)	2 (3.3)	35 (58.3)
Ciprofloxacin (5 µg)	15 (25.0)	7 (11.7)	38 (63.3)
Norfloxacin (10 µg)	20 (33.3)	8 (13.3)	32 (53.3)
Nitrofurantoin (300 µg)	48 (80.0)	3 (5.0)	9 (15.0)
Imipenem (10 µg)	53 (88.3)	3 (5.0)	4 (6.7)
Piperacillin-tazobactam (100/10 µg)	49 (81.7)	3 (5.0)	8 (13.3)
Gentamicin (10 µg)	32 (53.3)	5 (8.3)	23 (38.3)
Amikacin (30 µg)	43 (71.7)	4 (6.7)	13 (21.7)
Cotrimoxazole (1.25/23.75 µg)	16 (26.7)	4 (6.7)	40 (66.7)

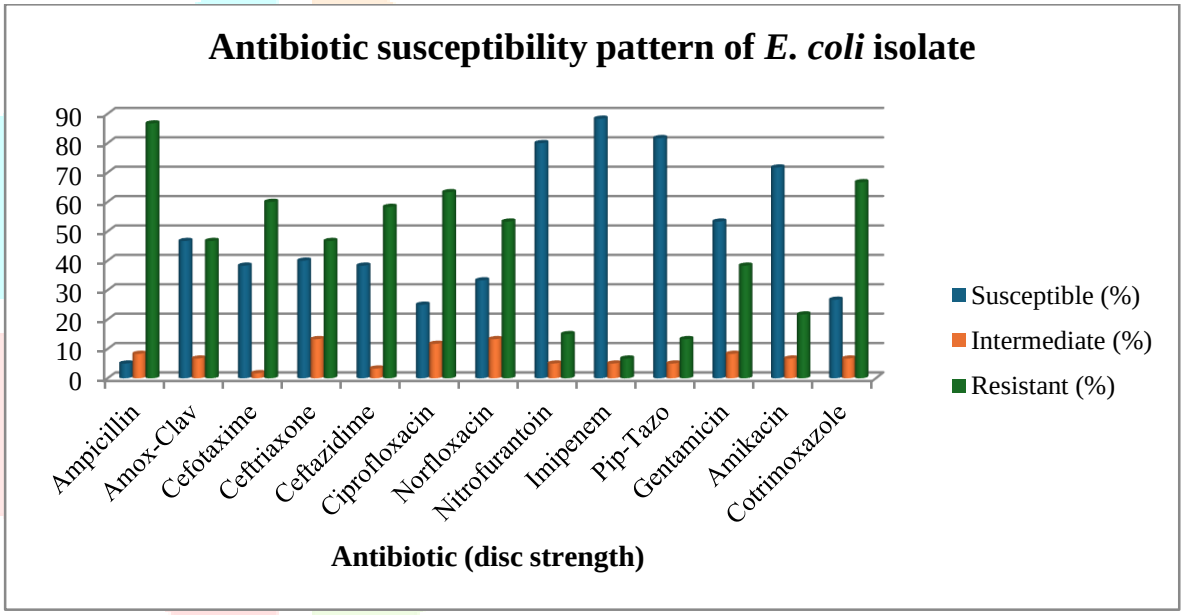

Figure 3. E. Coli isolates' pattern of antibiotic susceptibility

3.5 Burden of multidrug resistance

Applying the Magiorakos criteria [16], 57 of 60 E. coli isolates (95.0%) qualified as multidrug-resistant. The proportion of MDR isolates was higher among those obtained from outpatients (40 of 41, 97.6%) than from inpatients (17 of 19, 89.5%), Despite this, with p = 0.190, the difference was not statistically significant. Most MDR isolates simultaneously resisted aminopenicillins, fluoroquinolones, third-generation cephalosporins and cotrimoxazole, a combination that effectively eliminates the conventional oral options for empirical use.

3.6 Comparison with other Indian studies

Our findings sit comfortably within the range reported by recent Indian studies. E. coli prevalence in the present cohort (61.9%) is close to that reported by Akram et al. from Aligarh (61.0%) [10], Pelluri et al. from southern India (60.5%) [24] and Rana Pratap et al. from Bihar (64.6%) [11]. Resistance rates against ampicillin, cotrimoxazole and fluoroquinolones in our isolates are likewise comparable to, or marginally higher than, the rates reported by these centres. Sustained imipenem activity is also a common finding across Indian studies [4,25].

4. Discussion

The 27.7% culture positivity recorded in our cohort is consistent with figures reported from comparable Indian and overseas studies, which generally fall in the range of 21% to 32% [11,26,27]. Nearly three-quarters of clinically suspected cases yielded no growth, an observation with two practical implications. First, symptom-based diagnosis alone over-estimates the bacterial burden of UTI; many patients presenting with urinary symptoms have non-infective conditions such as interstitial cystitis, urolithiasis, urethral irritation, or are receiving an antibiotic that has already suppressed bacterial growth. Second, empirical antibiotic prescription for every symptomatic patient inevitably contributes to unnecessary exposure and selection of resistance. Our data argue for routine pre-treatment urine culture wherever feasible.

The continuing dominance of E. coli as the principal uropathogen (61.9% of our isolates) is unsurprising. Globally, the organism accounts for between 55% and 80% of culture-positive UTIs, and our figure sits squarely within that range [5-7,10,11]. Its biological advantages are well characterised: it is a natural resident of the human colon and is therefore continuously available to colonise the periurethral area; it carries a versatile set of adhesins and toxins encoded on pathogenicity islands; and it has a remarkable capacity for horizontal gene transfer, which allows it to acquire resistance determinants rapidly [8,9,28].

What is changing is not the organism but the drugs that work against it. The ampicillin resistance figure in our isolates (86.7%) effectively retires the drug from any empirical role in UTI management. This finding is not new in itself, but the consistency with which it is being reported from across India is striking [10,11,19]. The mechanism is dominated by plasmid-mediated β-lactamase production, particularly TEM and SHV enzymes, and is reinforced by decades of unrestricted clinical use [14,28].

Cotrimoxazole resistance at 66.7% in our isolates is similarly worrying. The IDSA guideline recommends that empirical use of cotrimoxazole should be avoided in regions where local resistance exceeds 20% [12]. By that yardstick our isolates have been beyond the threshold for many years, yet the drug continues to feature in some empirical regimens because it is cheap and widely available. Continued empirical use, in the face of a 66.7% resistance rate, is no longer defensible.

Fluoroquinolone resistance has followed the same trajectory. Ciprofloxacin, which was introduced into routine UTI practice in the late 1980s and was for a time the preferred oral agent for complicated infections, now retains activity against only one in four of our isolates. This decline reflects three converging pressures: extensive outpatient use without microbiological confirmation, easy over-the-counter availability in much of India, and a high frequency of incomplete treatment courses [4,13,15]. The cross-resistance between ciprofloxacin and norfloxacin observed in our data is expected, since both drugs share the same gyrA and parC mutational pathways.

Third-generation cephalosporin resistance in the 46.7% to 60.0% range is closely linked to ESBL production, which is plasmid-mediated and frequently co-carried with resistance determinants for aminoglycosides, fluoroquinolones and cotrimoxazole [15,16,29]. The clinical consequence is that even ceftriaxone, long considered a dependable parenteral option for complicated UTI, can no longer be relied on as monotherapy without susceptibility data. Amikacin, β-lactam/β-lactamase inhibitor combos, and carbapenems become the obvious backup options, with all the associated costs and toxicity.

The retained activity of nitrofurantoin (80.0% susceptibility) deserves attention. The drug has been in clinical use since the 1950s and its preservation against modern uropathogens is something of a pharmacological success story. Two features explain it: nitrofurantoin reaches high urinary concentrations without producing meaningful tissue or serum levels, which limits the selection pressure on extra-urinary flora; and its multiple intracellular targets make point-mutation resistance difficult to develop [30]. For uncomplicated lower UTI in non-pregnant women, nitrofurantoin remains a defensible first-line oral choice in this setting.

Imipenem retained the best overall activity in our panel (88.3% susceptibility). This is reassuring at one level but worrying at another. The wider use of carbapenems, driven by ESBL and MDR

organisms, is the major selective pressure for carbapenem-resistant Enterobacterales (CRE), a problem now well documented in Indian tertiary care [4,17]. The drug should therefore be reserved for documented carbapenem-sensitive ESBL infections or severe sepsis where empirical broad-spectrum cover is genuinely required. Routine use for uncomplicated infections accelerates the next wave of resistance and should be avoided.

The overall MDR rate of 95.0% in our isolates is at the higher end of published Indian figures, which range from 65% to 85% [10,11,24,25]. One possible explanation is referral bias: a tertiary care centre receives a disproportionate share of patients who have already failed earlier empirical regimens and are carrying selected resistant flora. Another is the unrestricted prescribing environment in the catchment population. Either way, the implication is the same. When 19 of every 20 urinary isolates resist three or more antibiotic classes, the empirical pool of usable oral drugs has shrunk to almost nothing, and pre-treatment culture is no longer a luxury.

A finding that did not reach statistical significance but is worth flagging is the marginally higher MDR rate among outpatient isolates (97.6%) compared with inpatient isolates (89.5%). The traditional view, that resistance is essentially a hospital problem, no longer fits the data. Outpatient resistance now equals or exceeds inpatient resistance in several Indian and international studies [4,15,18]. Likely drivers include over-the-counter antibiotic availability, self-medication, antibiotic use in food animals with subsequent environmental spread, and inadequate sanitation [3]. Empirical regimens for community-acquired UTI must be revised on the assumption that resistance is no longer confined to the hospital walls.

4.1 Strengths and limitations

This study's prospective design, parallel quality control using three ATCC reference strains, and utilisation of CLSI 2023 standards for susceptibility testing are its strong points, and the inclusion of both outpatient and inpatient populations, which captures the community-hospital interface.

There are a few restrictions to be aware of. Initially, the research was carried out at a single tertiary care facility and the resistance pattern reported here may not be directly generalisable to other regions or to primary and secondary care facilities. Second, the cross-sectional design provides a snapshot rather than a temporal trend; longitudinal surveillance would be needed to track the evolution of resistance. Third, ESBL production was detected phenotypically and resistance genes were not characterised at the molecular level. Fourth, clinical outcome data such as treatment response, recurrence and length of stay were not captured, so the link between in-vitro resistance and clinical failure could not be established directly. Fifth, the exclusion of patients on recent antibiotics, though methodologically necessary to avoid false-negative cultures, may have under-represented the most heavily resistance-selected sub-group.

5. Conclusion

In this hospital-based cross-sectional study from central India, E. coli remained the dominant uropathogen, accounting for 61.9% of all culture-positive UTIs. Resistance to ampicillin, cotrimoxazole, fluoroquinolones and third-generation cephalosporins has reached levels at which these drugs can no longer be used empirically with any confidence. Imipenem, piperacillin-tazobactam, nitrofurantoin and amikacin retained the best activity. Ninety-five percent of E. coli isolates met the criteria for multidrug resistance, with comparable rates in outpatient and inpatient settings, indicating that resistance is widely disseminated in the community as well as in the hospital.

Three practical recommendations follow. Empirical regimens for uncomplicated lower UTI in this region should default to nitrofurantoin rather than fluoroquinolones or cotrimoxazole. Complicated or hospital-acquired infections require culture-guided therapy, with carbapenems, piperacillin-tazobactam or amikacin held in reserve for confirmed indications. Sustained antimicrobial stewardship, regular institutional antibiograms and stricter regulation of over-the-counter antibiotic sales are needed to slow the further erosion of available options.

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Conflicts of Interest

The writers affirm that they have no conflicting interests.

Ethical Approval

Before the study started, the Institutional Ethics Committee of IMCHRC, Malwanchal University, Indore, examined and approved the protocol (Ref No: MU/Research/EC/Ph.D/2022/291B). All participants provided written informed consent.

Author Contributions

Aneesh Kumar Sharma planned the study, carried out the lab work, gathered and examined the data, and wrote the article. Kailash Jatav oversaw the research, helped with the study's design, analysed the data, critically edited the article, and gave the go-ahead for its publication. The final document was read and approved by both writers, who both agreed to take full responsibility for the work.

Data Availability

The corresponding author can provide the datasets created and examined for this work upon reasonable request.

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