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# Identification of bacterial isolates in urinary tract infections patients of Basrah province

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## ABSTRACT

**Introduction and aim.** Urinary tract infections (UTIs) are among the most common bacterial diseases worldwide that are caused primarily by members of the Enterobacteriaceae family. This study aimed to identify the most frequent bacterial agents associated with UTIs and analyze their patterns of antibiotic resistance using the Vitek®2 system.

**Material and methods.** The study included 200 urine samples collected from adult UTI patients of both sexes.

**Results.** The characterization of bacterial isolates revealed the following distribution: *Escherichia coli* (35 isolates, 50%), *Staphylococcus aureus* (18 isolates, 25.7%), *Klebsiella pneumoniae* (5 isolates, 7.14%), *Staphylococcus* spp. (4 isolates, 5.7%), *Streptococcus* spp. (3 isolates, 4.2%), *Pseudomonas* spp. (3 isolates, 4.2%), and *Proteus mirabilis* (2 isolates, 2.86%).

Antibiotic resistance testing showed that ceftazidime had the highest resistance rate (88.57%), while amikacin had the lowest (17.14%). Additionally, extended-spectrum  $\beta$ -lactamase (ESBL) production was detected in 35 *E. coli* isolates. Of these, 22 isolates (62.86%) tested positive for ESBL production, while 13 isolates (37.14%) were negative.

**Conclusion.** This study concluded that *E. coli* is the most prevalent bacterial species causing UTIs. Furthermore, the *E. coli* isolates demonstrated a high capacity for ESBL production, highlighting the need for effective antimicrobial management and monitoring.

**Keywords.** antibiotic susceptibility, *E. coli*, ESBLs

## **Introduction**

Urinary tract infections (UTIs) are inflammation of the urinary tract epithelium resulting from invasion by microorganisms.<sup>1</sup> It is an important and common disease affecting men and women of all ages.<sup>2,3</sup> UTIs are among the most common bacterial infections, affecting more than 150 million people annually.<sup>4</sup> *Escherichia coli*, a natural gut flora in humans, causes 70–95% of UTIs. Germs enter the urinary tract.<sup>5</sup> Uropathogenic *Escherichia coli* (UPEC) has the ability to penetrate the deeper layers, develop quiescent intracellular reservoirs, and remain there for months at a time before recurring infections.<sup>6</sup> UTIs can be caused by obstructions or defensive factors, such as urinary incontinence, retention, immunosuppression, renal failure, pregnancy, or the use of indwelling catheters.<sup>7</sup> Identifying UPEC strains based on their virulence factor-encoding genes and their function in illness progression is possible.<sup>1</sup> UPEC is caused by a variety of components, including secreted proteins, hemolysins, capsules, lipopolysaccharides, biofilm, fimbriae adhesions, and iron acquisition mechanisms.

The best type of medication for treating pathogenic bacteria is an antibiotic. When treating infections caused by Gram-negative bacteria, fluoroquinolones, cephalosporins,  $\beta$ -lactams, and  $\beta$ -lactamase inhibitors are often prescribed, alone or in combination.<sup>8</sup> One major problem threatening the global healthcare system is multidrug resistance (MDR). Usually, the widespread Enterobacteriaceae family is linked to the problem. Extended-spectrum  $\beta$ -lactamases (ESBLs) are the main antimicrobial resistance mechanism in this family of bacteria that renders  $\beta$ -lactam antibiotics ineffective.<sup>9</sup> Gram-negative bacteria can generate enzymes that hydrolyze  $\beta$ -lactam rings, making them resistant to one of the most effective medications.<sup>10</sup> ESBLs, which hydrolyze penicillins, monobactams, and cephalosporins, were first discovered in Germany in 1983. Since then, the number of diseases caused by bacteria with this resistance mechanism has grown. Furthermore, *E. coli* has evolved resistance to several antibiotics, leading to a major cause of illness. Multidrug-resistant *E. coli* bacteria, especially those producing ESBLs, pose a significant hazard to public health.

## **Aim**

The current study aimed to detect the most frequent bacterial agents associated with UTIs and to analyze the pattern of antibiotic resistance using the Vitek®2 system test.

## **Material and methods**

Urine samples were collected from Al Basrah General Teaching Hospital, Al Mawani General Teaching Hospital, and Al Sadir Teaching Hospital in Basrah Province between October 2023 and March 2024. A total of 250 individuals suspected of having UTIs, aged between 20 and 60 years, provided clinical samples. After collection, urine samples were centrifuged and cultured on blood agar, MacConkey agar mannitol salt agar, and eosin methylene blue agar. Biochemical tests were then performed for the identification of Gram-

positive bacteria. This study was conducted in accordance with the Declaration of Helsinki and received ethical approval No. (773) dated (10/3/2024) from the Basra General Health Directorate.

### ***Identification by Vitek®2 system***

The Vitek®2 system (bioMérieux, France) was used to identify bacterial species with high accuracy. The system includes 64 biochemical tests for bacterial diagnosis.<sup>11</sup>

### ***Bacterial DNA extraction***

Genomic DNA was isolated from bacterial isolates using the Wizard® Genomic DNA Purification Kit (Promega, USA).

### ***Detection of 16S rDNA***

The extracted bacterial DNA was amplified using PCR to target the 16S rDNA gene with a specific primer approximately 585 bp in length (Table 1). The primer sequence for 16S rDNA was used as described previously.<sup>12</sup> A standard molecular DNA ladder (2000 bp) was used to compare the PCR results.

**Table 1.** The bacterial 16S rDNA gene was amplified by PCR using specific primers

<b>Primer</b>	<b>Sequence of primer</b>	<b>Length (bp)</b>	<b>Product (bp)</b>
16SrDNA Forward	5-GAC CTC GGT TTA CTT CAC AGA-3	21	585
16SrDNA Revers	5-CAC ACG CTG ACG CTG ACC-3	18	

### ***Reagents***

The reagents and their volumes used in PCR amplification are described in Table 2.

**Table 2.** Reagents used in PCR amplification of 16SrDNA

<b>No.</b>	<b>Reagent</b>	<b>Volume</b>
1	Genomic DNA	1 µL
2	Forward primer	1 µL
3	Reverse primer	1 µL

4	Master Mix	12.5 $\mu$ L
5	Nuclease-free water	9.5 $\mu$ L
<b>Total volumes 25 <math>\mu</math>L</b>		

Thermal cycling condition: The program is described in Table 3.

**Table 3.** Program used in PCR amplification

Steps	Temperature	Time	No. of cycles
Initial denaturation	94°C	4 min	1
Denaturation	94°C	90 sec	30
Annealing	62°C	90 sec	
Extension	72°C	2 min	
Final extension	2°C	7 min	1

### ***Procedure***

Approximately 5  $\mu$ L of a 2000 bp DNA ladder and 5  $\mu$ L of 16S rDNA amplicons were subjected to gel electrophoresis for 45 minutes at 70 V using a casting tray with 1.5% agarose gel prepared in 1 $\times$ TBE buffer containing 0.2  $\mu$ L of ethidium bromide. The products were visualized under a UV light system.

### ***Antibiotic susceptibility test by Vitek®2 system***

The antibiotic susceptibility test was performed using the Antibiotic Susceptibility Kit Card (Vitek®2 AST, reference number 413083, bioMérieux, France). This kit includes a comprehensive range of antimicrobial tests.

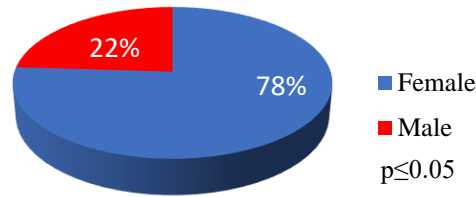
### ***Detection of extended spectrum $\beta$ -lactamase (ESBL)***

#### ***Double disk approximation method (DAM)***

Bacterial isolates, prepared as previously described, were spread onto Mueller-Hinton agar plates. A disk of Augmentin (amoxicillin-clavulanic acid) was placed at the center of the agar surface. Disks of ceftazidime (30  $\mu$ g), cefotaxime (30  $\mu$ g), amikacin (30  $\mu$ g), and ceftriaxone (30  $\mu$ g) were positioned around it at a distance of approximately 20 mm (center to center). After incubation at 37°C for 24 hours, bacteria were considered ESBL producers if the zone of inhibition around aztreonam or any of the antibiotic disks showed a clear-cut increase directed toward the Augmentin disk.<sup>13</sup>

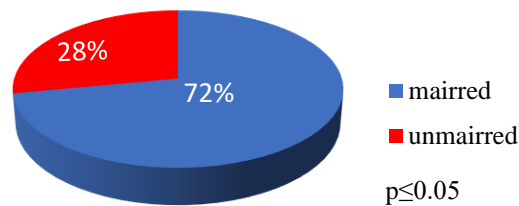
## Results

The present study included 100 patients with UTIs. The distribution of UTIs patients according to sex is shown in Figure 1. In this study, the proportion of female patients with UTIs was significantly higher than that of male patients, with 78 females and 22 males.



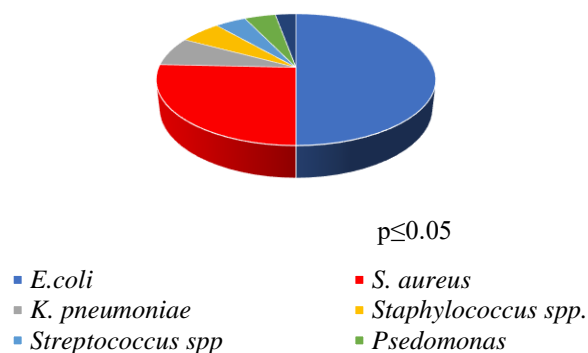
**Fig. 1.** Distribution of patients according to sex

The current data on the distribution of UTIs patients based on marital status showed that 28% were single and 72% were married, with a statistically significant difference ( $p \leq 0.05$ ) (Fig. 2).



**Fig. 2.** Distribution of UTIs patients according to the marital status

The present work detected uropathogenic *E. coli* (35 isolates, 50%), *S. aureus* (18 isolates, 25.71%), *K. pneumoniae* (5 isolates, 7.14%), *Staphylococcus* spp. (4 isolates, 5.71%), *Streptococcus* spp. (3 isolates, 4.29%), *Pseudomonas* (3 isolates, 4.29%), and *Proteus mirabilis* (2 isolates, 2.86%), as shown in Figure 3, with significant differences ( $p \leq 0.05$ ).



**Fig. 3.** Frequency of bacterial isolates in UTIs patients

The current investigation utilized the coagulase test, catalase test, mannitol salt agar, blood agar, and Gram's stain to identify Gram-positive bacteria (Table 4).

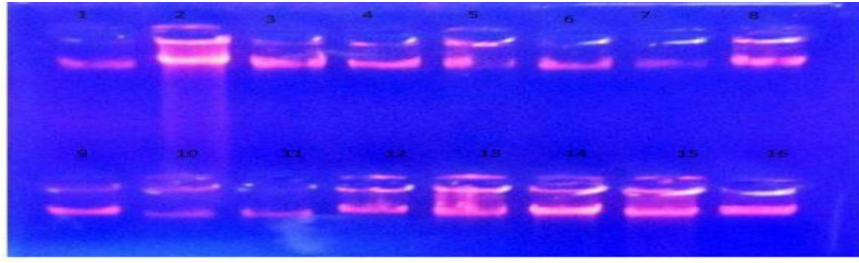
**Table 4.** Gram-positive bacteria are identified by biochemical tests

Type of isolate	Type of Test				Isolates number
	Gram's stain	Catalase test	Fermentation of mannitol	Coagulase test	
<i>Staphylococcus aureus</i>	+	+	+	+	18
<i>Staphylococcus spp.</i>	+	+	-	-	4
<i>Streptococcus spp.</i>	+	-	*N	N	3
Total			25		

\* N – not detectable

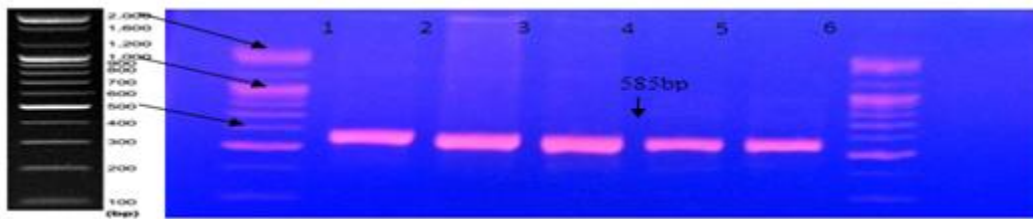
The present work indicated that the percentages of Gram-negative bacteria identified by the Vitek®2 system were the following: *E. coli* (35 isolates, 50%), *K. pneumoniae* (5 isolates, 7.14%), *Pseudomonas* (3 isolates, 4.29%), and *Proteus mirabilis* (2 isolates, 2.86%).

The 35 *E. coli* isolates were examined, and genomic DNA was extracted using the kit protocol method. The presence of genomic DNA was confirmed by gel electrophoresis on a 0.8% agarose gel, with the results visualized under UV light (Fig. 4).



**Fig. 4.** Total genomic DNA extracted from *E.coli* isolates, using 0.8% agarose gel, 70V, 45 min

PCR was used to amplify the 16S rDNA from the extracted DNA. The individual 16S rDNA band (585 bp) was compared to the standard molecular DNA ladder (2000 bp) (Fig. 5).



**Fig. 5.** PCR amplified products of 16S rDNA are seen in agarose electrophoresis patterns. Lane 1: 2000 base pair DNA ladder; Lane 2: 16S rDNA band of *E. coli* isolates, using 70V, 45 min, 1.5% agarose gel

In this study, the Vitek®2 system was used to detect and confirm antibiotic susceptibility tests, as shown in Table 5 and Figure 4.

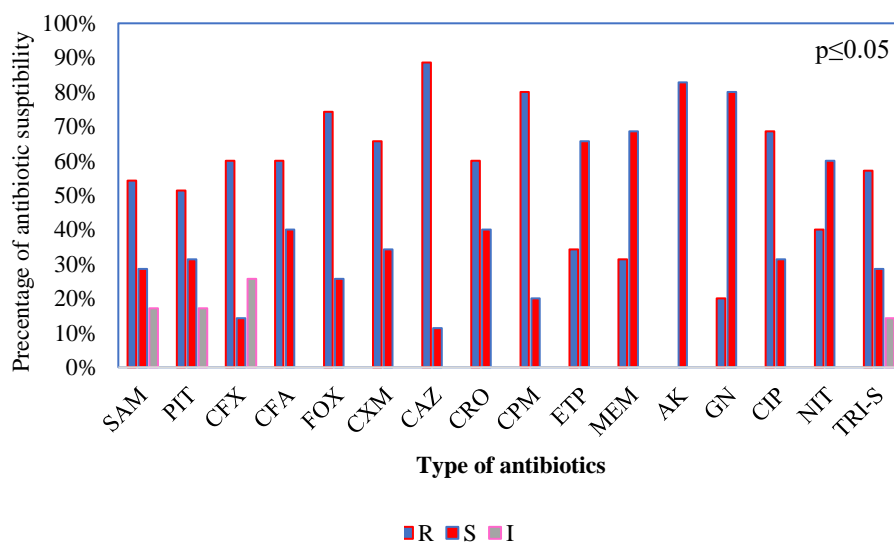
**Table 5.** Sensitive testing against different species of gram-negative bacteria and detection of ESBLs

Antibiotic group	Antimicrobial agent	<i>E. coli</i>			<i>K. pneumoniae</i>		<i>P. mirabilis</i>		<i>P. aeruginosa</i>	
		R	S	I	R	S	R	S	R	S
β-lactam combinations	Ampicilli/sulbactam (SAM)	19	10	6	4	1	2	-	-	-
		54.29	28.57	17.14	80%	20%	100	-	-	-
		%	%	%			%			
β-lactam combinations	Piperacillin/Tazobactam (PIT)	18	11	6	5	-	2	-	2	1
		51.43	31.43	17.14	100	-	100	-	66.7	33.3
		%	%	%	%		%		%	%
Cephalosporins	Cefuroxime (CFX)	21	5	9	4	1	--	2	2	1
		60%			80%	20%				



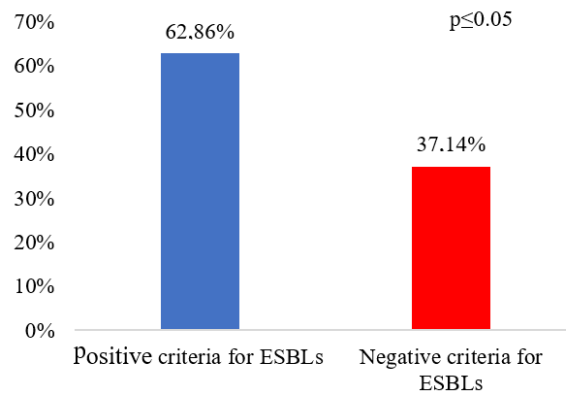
		14.29	25.71				100	66.7	33.3
		%	%				%	%	%
	Cefuroximeaxetil (CFA)	21 60%	14 40%	-	4 80%	1 20%	-	2 100%	66.7 80%
	Cefoxitin (FOX)	26 74.29%	9 25.71%	-	-	5 100%	-	2 100%	100%
	Cefixme (CXM)	23 65.71%	12 34.29%	-	-	5 100%	-	2 100%	1 33.3% 2 66.7%
	Ceftazidime (CAZ)	31 88.57%	4 11.43%	-	1 20%	4 %80	-	2 100%	1 33.3% 2 66.7%
	Ceftriaxone (CRO)	21 60%	14 40%	-	1 20%	4 %80	-	2 100%	1 33.3% 2 66.7%
	Cefepime (CPM)	28 80%	7 20%	-	-	5 100%	-	2 100%	1 33.3% 2 66.7%
β-lactam carabapenem	Ertapenem (ETP)	12 34.29%	23 65.71%	-	4 80%	1 20%	-	2 100%	3 100% -
	Meropenem (MEM)	11 31.43%	24 68.57%	-	1 20%	4 80%	-	2 100%	1 33.3% 2 66.7%
Aminoglycoside	Amikacin (AMK)	6 17.14%	29 82.86%	-	4 80%	1 20%	-	2 100%	2 66.7% 1 33.3%
	Gentamicin (GEN)	7 20%	28 80%	-	2 40%	3 60%	2 100%	-	- 100%

Fluroquinolones	Ciprofloxacin (CIP)	24 68.57 %	11 31.43 %	-	-	5 100 %	2 100 %	3 100 %	-
	Nitrofurantoin (NIT)	14 40%	21 60%	-	-	5 100 %	2 100 %	3 100 %	-
Sulfonamides	Trimethoprim sulfamethoxazole (TRI-S)	20 57.14 %	10 28.57 %	5 14.29 %	-	5 100 %	2 100 %	2 66.7 %	1 33.3 %



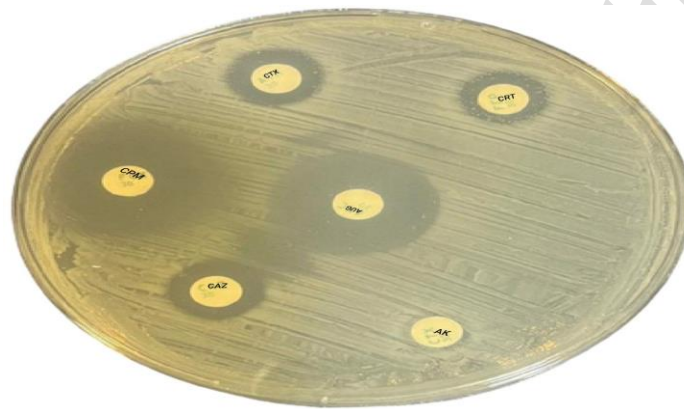
**Fig. 6.** Antibiotic susceptibility of *E. coli*

The Vitek®2 system was used to detect the ability to produce extended-spectrum  $\beta$ -lactamases (ESBLs) in 35 *E. coli* isolates. The results showed that 22 (62.86%) *E. coli* isolates tested positive for the ability to produce ESBLs, while 13 (37.14%) *E. coli* isolates tested negative (Fig. 7).



**Fig. 7.** Demonstrated that positive and negative results of ESBL by *E. coli*

Of the 35 *E. coli* isolates, 19 (54.29%) developed ESBLs with positive results, while 16 (45.71%) isolates showed negative results for ESBL production, as determined by the DAM method (Fig. 8).



**Fig. 8.** DAM test for ESBL detection using *E. coli* isolates

## Discussion

*E. coli* spreads in the urinary tract due to its natural habitat in the digestive tract, its proximity to the anus opening near the urinary tract opening, and its virulence factors, which allow it to adhere to surfaces, withstand urine-induced flow, and cause urinary tract infections. The current study revealed that women constituted the majority of cases compared to males, a finding that is consistent with most previous studies. A study by Kamel et al. showed that women represented the highest percentage of UTIs, with 77% of cases in women and 23% in males.<sup>14</sup> This finding is supported by most research due to the anatomical structure of females, where the urethra is shorter than that of males, thus reducing the distance bacteria must travel to reach the bladder.<sup>15</sup>

Current data on the distribution of UTI patients based on marital status showed that 28% were single and 72% were married, which is consistent with the study by Al-Musawi and Al-Husseini.<sup>16</sup> According to

Alhamdy and Shani, married women had a higher prevalence and incidence of UTIs, which may be due to hormonal impacts and behavioral habits.<sup>17</sup>

The present study employed several techniques for identifying Gram-positive bacteria, including Gram's stain, microscopic examination, blood agar culture, mannitol salt agar, coagulase test, and catalase test. *Streptococcus* species were catalase-negative, Gram-positive, and arranged in chains, while *Staphylococcus* species appeared as purple clusters resembling grapes. The colony size of *S. aureus* ranged from 1 to 4 mm, spherical, convex, and with sharp boundaries. It was golden yellow in color, Gram-positive when stained, catalase-positive, and surrounded by areas of  $\beta$ -hemolysis on blood agar. Nonmotile clusters of *S. aureus* resembled grapes.<sup>18</sup> Furthermore, *S. aureus* may ferment mannitol when grown on mannitol salt agar, resulting in a positive coagulase production test and changes the indicator's color from red to yellow by producing acid. However, some species of *Staphylococcus* do not change color on mannitol agar and show negative results from the coagulase test.<sup>19</sup> Since Gram-positive bacteria received limited attention in this study, simple methods, such as the color change from red to yellow due to acid production and a positive coagulase test, were sufficient for their identification. Other species of *Staphylococcus*, such as *S. alhilua* and *S. al-shujairib*, grow on mannitol agar without changing color and provide negative coagulase results.<sup>19</sup> In this study, different bacterial species were identified through biochemical analysis, with *E. coli* accounting for 35 isolates, *S. aureus* for 18 isolates, *K. pneumoniae* for 5 isolates, *Staphylococcus* spp. for 4 isolates, *Streptococcus* spp. for 3 isolates, *Pseudomonas* for 3 isolates, and *P. mirabilis* for 2 isolates. These were the most frequently occurring bacteria in UTIs, which are a prevalent issue affecting both men and women and are typically caused by bacterial infections.<sup>20,22</sup> The current study found that *E. coli* was the most common bacteria isolated from UTIs, accounting for 50% of cases. This is consistent with previous studies from Baghdad, Iraq, which reported an *E. coli* rate of 50%, and from Basrah, which reported a rate of 60%.<sup>20,21</sup> *E. coli* is dominant in UTIs due to its presence in the intestines and its ability to enter the urinary tract. It has virulence factors such as biofilm formation, fimbriae, alpha-hemolysin, cytotoxic necrotizing factor, adhesins, and iron acquisition systems, as well as antibiotic resistance genes. The majority of *E. coli* strains are resistant to  $\beta$ -lactam antibiotics.<sup>22,23</sup> *UPEC* has the ability to penetrate the urothelium's deeper layers, develop quiescent intracellular reservoirs, and remain there for months before causing recurrent infections.<sup>6</sup>

The results of the biochemical identification test, used to identify Gram-negative bacteria in the current investigation, were validated and supported by the Gram-negative identification card of the Vitek®2 System test. Based on the findings from the Vitek®2 System, 35 isolates were identified as *E. coli*, 5 isolates as *K. pneumoniae*, 4 isolates as *Pseudomonas*, and 2 isolates as *P. mirabilis*. Crowley et al. demonstrated that the Vitek®2 system accurately identifies bacteria down to the species level. The study evaluated the technique and concluded that the Vitek®2 GN identification method is a suitable automated method for the rapid identification of Gram-negative bacteria. Additionally, the results of Salumi and Abood demonstrate the

high quality and reliability of the Vitek®2 GN technology.<sup>24,25</sup> A study by Pincus also highlighted that Vitek®2 offers an efficient timeframe for detecting isolates without mutations, boasting a high degree of accuracy (99%) and an extremely narrow margin of error. Another study by Ossman et al. confirmed the utility of the Vitek®2 Compact method for identifying bacteria in UTIs samples, combining conventional and biochemical methods.<sup>26</sup> In the current study, the results of Gram-negative bacterial identification by the Vitek®2 system were completely consistent with the primary identification, in addition to saving time and effort, avoiding laboratory errors, and providing more accurate results.

16S rDNA was amplified using PCR with the extracted DNA. A single 16S rDNA band (585 bp) was identified by comparing it to the conventional molecular DNA ladder (2000 bp). The diagnostic gene, 16S rDNA, which is stable and exhibits some heterogeneity over time within bacterial species, was used for molecular diagnostics on all *E. coli* isolates using PCR technology. The results showed that the 16S rDNA gene sequence was present in all isolates at a 100% rate, and the molecular weight of the band was 585 bp when compared to the DNA ladder. The findings of this study are consistent with those of Maleki et al. in Iran and Lai et al.<sup>27,28</sup> The results align with the study conducted by Jenkins et al. in Malaysia.<sup>29</sup> This gene exhibits slight heterogeneity but contains conserved regions that overlap with variable portions, which are used to identify the bacterial genus and species. This makes it one of the most rapid, accurate, and sensitive methods for diagnosing bacteria, with minimal random variation in the genomic sequence over time.<sup>30,31</sup>

This study aims to determine the antibiotic resistance patterns of predominant uropathogens and confirm *E. coli*, the most common Gram-negative bacterium, as a frequent cause of urinary tract infections.<sup>32</sup> Appropriate treatment heavily depends on the proper prescription of both effective and efficacious antibiotics. The development of resistance by pathogenic bacteria has become synonymous with antibiotic use. Antibiotic resistance is a major global threat, particularly recognized in *E. coli* and other Gram-negative bacteria, making treatment difficult. Determining the resistance patterns of *E. coli* is crucial for guiding empirical and targeted therapy.<sup>34</sup>

The study revealed low resistance rates to aminoglycosides, specifically amikacin and gentamicin. These findings were consistent with prior studies, such as those by Mohamed and Aljanaby and Ayatollahi et al., which demonstrated high sensitivity to these antibiotics.<sup>35,36</sup> Similar observations were noted in research by Al-Khikani et al. regarding gentamicin. Aminoglycosides exert their action by binding to the 30S ribosomal subunit of bacteria, disrupting protein synthesis, and either terminating protein synthesis prematurely or incorporating the wrong amino acids.<sup>37</sup> Aminoglycosides are bactericidal, displaying concentration-dependent bacterial killing. However, it is still not fully understood whether there is an additional mechanism of action involved.

Carbapenem resistance was observed at varying levels across studies. The current study found a resistance rate of 34.8%, which is in agreement with Mujahid et al.<sup>38</sup> However, lower resistance rates of 2.4% were

reported by Jalil and Atbee.<sup>39</sup> These variations could be attributed to regional differences in antibiotic usage and resistance monitoring practices.

The resistance to trimethoprim-sulfamethoxazole was 40%, as also observed by Critchley et al.<sup>40</sup> The rise in resistance is fueled by the easy access to antibiotics without prescriptions, combined with a lack of knowledge, particularly in communities with low incomes. Educational programs focusing on accurate disease diagnosis, selecting the right antibiotics, and promoting responsible antibiotic use can greatly help reduce resistance. Ahmed and Andaleeb et al. also found high resistance rates against  $\beta$ -lactam and second- to fourth-generation cephalosporin antibiotics.<sup>41,42</sup> The development of resistance mechanisms involves the production of  $\beta$ -lactamases, such as cephalosporinase and penicillinase enzymes, which break down the  $\beta$ -lactam ring of antibiotics, rendering them ineffective, as mentioned in the studies by Paltansing and Al-Shoyaikh.<sup>43,44</sup>

The bacteria developed resistance to ciprofloxacin by producing biofilms, which prevented antibiotics from penetrating and killing them, allowing them to survive. According to Paltansing, resistance to quinolones is attributable to target site alterations or changes in outer membrane permeability.<sup>43</sup> The major mechanism involves DNA gyrase mutations, as described by Zaman et al.<sup>45</sup> Variations in antibiotic resistance and sensitivity among *E. coli* strains highlight the importance of antimicrobial susceptibility testing as a fundamental step in guiding effective treatment.

The results showed that out of 35 *E. coli* isolates, 22 (62.8%) were ESBL producers, while 13 isolates (37.14%) did not produce the enzyme. These results align with Veve et al., who reported that 83.7% of *E. coli* isolates were able to produce ESBL.<sup>46</sup> However, the present study disagrees with Talan et al., which found that only 14.8% of 453 *E. coli* isolates produced ESBL.<sup>47</sup> ESBL and carbapenemase infections have been on the rise in recent years and are leading causes of hospital- and community-acquired infections. As a result, a rapid and reliable test is required to determine the appropriate antibiotic. The Vitek®2 system demonstrated rapid and accurate detection, saving time and effort. The Vitek®2 compact system is a fully automated, standardized microbiological system that performs both drug susceptibility testing and bacterial identification simultaneously. The Vitek®2 ESBL test panel is used for the quick detection of ESBL production, in addition to accurate identification and susceptibility testing, by evaluating the inhibitory effects of cefepime, cefotaxime, and ceftazidime both alone and in combination with clavulanic acid. Both molecular methodology and the Vitek®2 ESBL test panel successfully detected the formation of ESBL in Enterobacteriaceae.<sup>48</sup>

The isolates were also tested for ESBL enzyme production using the Double Disk Approximation Method (DAM). Results showed that out of 35 *E. coli* isolates, 19 (54.29%) were ESBL producers, while 16 (45.71%) did not produce the enzyme. The results of this study agree with Samiyah et al., who found that 67% of their isolates were ESBL producers in Saudi Arabia, and with Mohammed et al. in Iraq.<sup>49,50</sup> A study

in India by Sudharani et al. found that 53% of their *E. coli* isolates had the ability to produce ESBL.<sup>51</sup> However, the present study disagreed with Riaz et al., in Pakistan.

$\beta$ -lactamase is a major virulence factor that destroys the  $\beta$ -lactam ring in some antibiotics, enhancing antibiotic resistance and pathogenicity in *E. coli*. ESBLs have emerged as a significant mechanism of  $\beta$ -lactam and other antibiotic resistance in Enterobacteriaceae. ESBL enzymes provide resistance to penicillins, cephalosporins, monobactams, and other antibiotics.<sup>46</sup> The World Health Organization has identified bacterial antibiotic resistance as one of its top health concerns. The extensive use of antibiotics without susceptibility testing is a major cause of the evolution of multidrug-resistant bacteria, which significantly hampers treatment efforts and may compromise the effectiveness of other treatments.<sup>54</sup>

### ***Study limitations***

Despite the significance of this study's findings on the isolation of *E. coli* from individuals suffering from UTIs and its resistance to antibiotics, several limitations exist. The study did not focus on the virulence factors that exacerbate infections or contribute to antibiotic resistance. This limitation can be addressed in future studies by employing advanced analyses, such as PCR for identifying genes responsible for virulence factors and whole-genome sequencing.

Whole-genome sequencing of the bacterial strains was not conducted due to financial and time constraints. Consequently, the diversity of *E. coli* strains and the mechanisms underlying antibiotic resistance development were not explored. Future studies should include such analyses to provide a comprehensive understanding of the genetics of these bacteria and their potential impact on therapy.

Although the number of participants in this study was statistically adequate, it may not have fully captured the variations in genetic patterns and resistance mechanisms prevalent in other regions or communities worldwide. Previous studies addressing these factors utilized larger sample sizes representative of diverse geographical areas. Future research should consider a broader and more diverse participant base to enhance the generalizability of the findings.

### ***Conclusion and future scope***

The current study identified *E. coli* as the primary causative bacterial species responsible for UTIs among hospitalized patients in Basrah. Antibiotic resistance patterns of the causal microorganisms revealed that *E. coli* exhibited the highest prevalence of resistance to ceftazidime, whereas amikacin showed the lowest resistance. Furthermore, 62.86% of the samples tested positive for ESBL production, resulting in resistance to third-generation cephalosporins.

These findings have significant practical implications. The high resistance to ceftazidime underscores the urgent need to reconsider its use as a first-line treatment for UTIs, particularly among patients in Basrah's

hospitals. Amikacin may serve as a suitable alternative, provided its use aligns with local antibiotic stewardship guidelines and individual patient needs.

The prevalence of *E. coli* strains producing ESBL highlights the necessity of routine screening for these strains in clinical laboratories and diagnostic centers. Such measures would enable physicians to select appropriate medications, avoid ineffective treatments, and mitigate complications through the timely identification of ESBL production.

These findings also emphasize the importance of antibiotic stewardship programs that prioritize local resistance patterns in Basrah. Implementing such programs can help regulate antibiotic prescriptions, reduce the overuse of cephalosporins, and curb the spread of resistant bacterial strains.

## **Conclusion**

The current study identified *E. coli* as the primary bacterial species responsible for UTIs among hospitalized patients in Basrah. Antibiotic resistance patterns of the causative microorganisms revealed that *E. coli* exhibited the highest prevalence of resistance to ceftazidime, while amikacin showed the least resistance. Additionally, 62.86% of the isolates tested positive for extended-spectrum  $\beta$ -lactamase (ESBL) production, resulting in resistance to third-generation cephalosporins.

These findings have significant practical implications. The notable resistance to ceftazidime underscores the urgent need to reconsider its use as a first-line treatment for UTIs, especially in Basrah's hospitals. Given that amikacin demonstrated lower resistance, it may serve as a viable alternative, provided its use aligns with local antibiotic guidelines and patient needs. The high prevalence of *E. coli* strains that produce ESBL highlights the necessity for routine screening of these strains in clinical laboratories and diagnostic centers. This would enable healthcare providers to select appropriate treatments, avoid ineffective therapies, and promptly identify ESBL production to prevent complications.

Furthermore, these findings emphasize the importance of antibiotic stewardship programs that focus on local resistance trends in Basrah. Such programs can help regulate antibiotic prescriptions, minimize the overuse of cephalosporins, and curb the spread of resistant strains, ultimately improving patient outcomes and reducing the public health burden of antimicrobial resistance.

## **Declarations**

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### ***Author contributions***

Conceptualization, Z.A.E. and W.S.S.; Methodology, Z.A.E.; Software, W.S.S.; Validation, Z.A.E., W.S.S. and M.A.I.A.; Formal Analysis, W.S.S.; Investigation, M.A.I.A.; Resources, M.A.I.A.; Data Curation,



Z.A.E.; Writing – Original Draft Preparation, Z.A.E.; Writing – Review & Editing, Z.A.E.; Visualization, M.A.I.A.; Supervision, Z.A.E.; Project Administration, Z.A.E.; Funding Acquisition, W.S.S.

### ***Conflicts of interest***

The authors have disclosed no conflicts of interest.

### ***Data availability***

The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

### ***Ethics approval***

This study was conducted in accordance with the Declaration of Helsinki, and based on ethical approval No. (773) dated (10/3/2024) from the Basra General Health Directorate.

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