

Antibiotic Resistance Pattern of Diarrheal pathogens in Stool Samples Obtained from Children in Thi-Qar Governorate

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Abstract

Background: Diarrheal diseases remain a major cause of morbidity among children worldwide, particularly in developing countries, with *Escherichia coli* being one of the most common bacterial pathogens. The increasing emergence of antimicrobial resistance among diarrheagenic *E. coli* poses a serious challenge to effective treatment. **Objective:** This study aimed to investigate the prevalence of bacterial pathogens, antimicrobial susceptibility patterns, and multidrug resistance among *E. coli* isolates obtained from children with diarrhea. **Methods:** A cross-sectional study was conducted on 100 stool samples collected from children aged 1–12 years presenting with diarrheal symptoms. Bacterial isolation was performed on MacConkey and Eosin Methylene Blue (EMB) agar plates, followed by identification using the VITEK 1 compact. Antimicrobial susceptibility testing was carried out using the disk diffusion method according to standard guidelines. Multidrug resistance (MDR) was defined as resistance to three or more antibiotic classes. **Results:** The results exhibited high positivity rates across all age groups, ranging from 82.6% in the 1–3 years group to 100% in the 10–12 years group. The highest number of positive samples was observed in the 1–3 years age group (57/69), while all samples in the 10–12 years group were positive (6/6). However, no statistically significant association was observed between age group and positivity status ($\chi^2 \approx 2.29$, $df = 3$, $p > 0.05$). This indicates that the differences in positivity rates among the studied age groups were not statistically significant. *E. coli* was the predominant isolate, followed by *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*. High resistance rates were observed against cefotaxime, tetracycline, and amoxicillin, whereas higher susceptibility was recorded for imipenem, ciprofloxacin, and amikacin. A considerable proportion of isolates were identified as multidrug-resistant, indicating widespread resistance among enteric pathogens. **Conclusion:** The findings highlight the high prevalence of antimicrobial resistance among diarrheagenic bacteria, particularly *E. coli*, in children. Continuous surveillance, rational antibiotic use, and implementation of antimicrobial stewardship programs are essential to control the spread of multidrug-resistant pathogens and improve clinical outcomes.

Keywords: *Escherichia coli*; Diarrheagenic *E. coli* (DEC); Antibiotic resistance; and Multidrug resistance (MDR)

الخلاصة:

لا تزال أمراض الإسهال سبباً رئيسياً للمراضة بين الأطفال في جميع أنحاء العالم، وخاصة في البلدان النامية، حيث تُعد الإشريكية القولونية من أكثر مسببات الأمراض البكتيرية شيوعاً. ويُشكل تزايد ظهور مقاومة المضادات الحيوية بين الإشريكية القولونية المسببة للإسهال تحدياً خطيراً أمام العلاج الفعال. **الهدف:** هدفت هذه الدراسة إلى التحقق من مدى انتشار مسببات الأمراض البكتيرية، وأنماط حساسية المضادات الحيوية، ومقاومة الأدوية المتعددة بين عزلات الإشريكية القولونية المأخوذة من الأطفال المصابين بالإسهال. **الطرق:** أجريت دراسة مقطعية على 100 عينة براز جُمعت من أطفال تتراوح أعمارهم بين 1 و 12 سنوات يعانون من أعراض الإسهال. تم عزل البكتيريا على أطباق أجار ماكونكي وأجار EMB، ثم تم تحديدها باستخدام نظام VITEK 1. أُجري اختبار حساسية المضادات الحيوية باستخدام طريقة انتشار القرص وفقاً للإرشادات القياسية. عُرِفَت مقاومة الأدوية المتعددة (MDR) بأنها مقاومة لثلاث فئات أو أكثر من المضادات الحيوية. **النتائج:** أظهرت النتائج معدلات إيجابية عالية في جميع الفئات العمرية، حيث تراوحت من 82.6% في الفئة العمرية من 1 إلى 3 سنوات إلى 100% في الفئة العمرية من 10 إلى 12 سنة. سُجِّلَ أعلى عدد من العينات الإيجابية في الفئة العمرية من 1 إلى 3 سنوات (57 من 69)، بينما كانت جميع العينات في الفئة العمرية من 10 إلى 12 سنة إيجابية (6 من 6). مع ذلك، لم تُلاحظ أي علاقة ذات دلالة إحصائية بين الفئة العمرية وحالة الإيجابية ($\chi^2 \approx 2.29$ ، $df = 3$ ، $p > 0.05$). يُشير هذا إلى أن الاختلافات في معدلات الإيجابية بين الفئات العمرية المدروسة لم تكن ذات دلالة إحصائية. كانت الإشريكية القولونية هي العزلة السائدة، تليها الزائفة الزنجارية ثم الكلبسيلا الرئوية. لوحظت معدلات مقاومة عالية ضد السيفوتاكسيم، التتراسيكلين، الأموكسيسيلين، بينما سُجِّلَت حساسية أعلى للإيميبينيم والسيبروفلوكساسين والأميكاسين. وتم تحديد نسبة كبيرة من العزلات على أنها مقاومة لأدوية متعددة، مما يشير إلى انتشار واسع للمقاومة بين مسببات الأمراض المعوية. **الخلاصة:** تُبرز هذه النتائج الانتشار الواسع لمقاومة المضادات الحيوية بين البكتيريا المسببة للإسهال، وخاصة الإشريكية القولونية، لدى الأطفال. ويُعد الترصد المستمر والاستخدام الرشيد للمضادات الحيوية وتطبيق برامج ترشيد استخدام المضادات الحيوية أموراً ضرورية للسيطرة على انتشار مسببات الأمراض المقاومة لأدوية متعددة وتحسين النتائج السريرية.

الكلمات المفتاحية: الإشريكية القولونية؛ الإشريكية القولونية المسببة للإسهال (DEC)؛ مقاومة المضادات الحيوية؛ ومقاومة الأدوية المتعددة (MDR).

Introduction

Diarrheal diseases remain a major global public health concern and are among the leading causes of morbidity and mortality in children under five years of age, particularly in developing countries. Recent global estimates indicate that diarrheal diseases account for a substantial proportion of pediatric deaths annually and contribute significantly to hospitalizations and long-term complications such as malnutrition, impaired growth, and developmental delays [1, 2]. The burden is further exacerbated by poor sanitation, unsafe drinking water, and limited access to adequate healthcare services in many low-resource settings [3]. Among the etiological agents responsible for diarrheal infections, bacterial pathogens play a significant role, with *E. coli* being one of the most frequently isolated organisms in children with diarrhea [4]. Diarrheagenic *E. coli* (DEC) comprises several pathotypes, including enteropathogenic (*EPEC*), enterotoxigenic (*ETEC*), enteroaggregative (*EAEC*), enterohemorrhagic (*EHEC*), and enteroinvasive (*EIEC*) strains, each characterized by distinct virulence mechanisms that contribute to disease pathogenesis [5]. These pathogens differ in their ability to adhere to intestinal epithelial cells, produce toxins, and induce cellular damage, leading to a wide range of clinical manifestations from mild diarrhea to severe disease. The pathogenicity of DEC is largely determined by the presence of virulence genes that encode factors involved in adhesion, colonization, invasion, and toxin production [6]. In addition, virulence-associated genes related to colonization, fitness, toxin production, and effector proteins enhance bacterial survival and pathogenic potential [7]. The coexistence of multiple virulence genes within a single isolate has been reported and is often

linked to increased disease severity and persistence [8].

E. coli strains are also classified into phylogenetic groups (A, B1, B2, C, D, E, and F), which are associated with differences in pathogenic potential. Pathogenic strains are typically associated with phylogroups B2 and D, while commensal strains belong mainly to groups A and B1 [9]. However, commensal strains can serve as reservoirs for resistance and virulence genes, which may be transferred to pathogenic strains through horizontal gene transfer [10]. This genetic exchange plays a critical role in the emergence of more virulent and resistant *E. coli* strains. Antimicrobial resistance (AMR) in *E. coli* has become a major global health challenge. The widespread misuse and overuse of antibiotics in both clinical and agricultural settings have accelerated the development of multidrug-resistant (MDR) strains, limiting therapeutic options [11, 12]. High resistance rates have been reported against commonly used antibiotics such as β -lactams, fluoroquinolones, and aminoglycosides [13]. Extended-spectrum β -lactamase (ESBL)-producing *E. coli* have emerged as critical priority pathogens due to their ability to hydrolyze a wide range of β -lactam antibiotics, including penicillins and cephalosporins [14]. Resistance is primarily mediated by β -lactamase genes such as *blaTEM*, *blaCTX-M*, *blaSHV*, and *blaOXA*, which are widely distributed among enteric pathogens [15]. The dissemination of these resistance genes is largely facilitated by plasmid-mediated horizontal gene transfer, enabling rapid spread among bacterial populations [16].

Other resistance mechanisms, including efflux pumps and aminoglycoside-modifying enzymes such as *aac(6')-Ib-cr*, further contribute to multidrug resistance phenotypes [17]. The emergence of

carbapenem-resistant strains, particularly those carrying New Delhimetallo- β -lactamase (NDM) genes, represents an additional threat to effective treatment [17]. Newly reported advances in diagnostic microbiology have enhanced the detection of diarrheagenic *E. coli* in children with diarrhea. Whereas traditional culture methods continue crucial for bacterial isolation, molecular techniques such as multiplex PCR, real-time PCR, and other nucleic acid-based procedures provide rapid, sensitive, and specific recognition of *E. coli* pathotypes and their virulence determinants. These methods enhance diagnostic precision and verify effective epidemiological monitoring and clinical management of pediatric diarrheal diseases. [18]. These approaches allow accurate tracking of transmission pathways and identification of high-risk clones. Previous studies conducted in different regions, including Kenya, have investigated antimicrobial resistance patterns in *E. coli* isolates using phenotypic methods. However, these approaches often lack sufficient resolution to identify the genetic determinants of resistance and virulence [15]. This study aimed to investigate the prevalence of bacterial pathogens, antimicrobial susceptibility patterns, and multidrug resistance among *E. coli* isolates obtained from children with diarrhea, in order to improve clinical management and support effective control strategies.

Materials and Methods

2.1 Study Design and Sample Collection

A cross-sectional study was conducted to investigate antibiotic resistance and virulence characteristics of *E. coli* isolated from children with diarrhea. A total of 100 stool samples were collected from children under five years of age presenting with acute diarrheal symptoms. The study population consisted of 57 male and 43 female children.

Samples were collected in sterile, leak-proof containers under aseptic conditions and transported immediately to the microbiology laboratory for processing to minimize contamination and bacterial overgrowth. Proper collection, handling, and transportation of stool specimens were performed according to standard microbiological guidelines [19]. This study was conducted at the Department of Microbiology, College of Veterinary Medicine, Shatrah University, Iraq.

2.2 Ethical Approval

Ethical approval for this study was obtained from the Department of Microbiology, College of Veterinary Medicine, Shatrah University, Iraq. The study protocol was reviewed and approved in accordance with institutional ethical standards for research involving human samples. Informed consent was obtained from the parents or guardians of all participating children before sample collection.

2.3 Isolation and Culturing of Bacterial Isolates

Stool specimens were inoculated onto selective and differential media, including MacConkey agar and Eosin Methylene Blue (EMB) agar (Oxoid, UK), to isolate Gram-negative enteric bacteria. The inoculated plates were incubated aerobically at 37°C for 18–24 hours. Colonies showing characteristic morphology of *E. coli*, such as pink colonies on MacConkey agar (lactose fermenters) and metallic green sheen colonies on EMB agar, were selected and subcultured to obtain pure isolates [19]. The samples were numbered from S1 to S100.

2.4 Identification of Bacterial Isolates

Presumptive *E. coli* isolates were identified using the automated VITEK I Compact system (bioMérieux, France) following the manufacturer's instructions. Bacterial suspensions were prepared in sterile saline and adjusted to a turbidity equivalent to 0.5 McFarland standard using a densitometer. The

suspensions were then inoculated into GN identification cards, and species-level identification was obtained automatically based on biochemical profiling [20, 21].

2.5 Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing (AST) was performed using the disk diffusion method. The susceptibility results were interpreted automatically by the system and categorized as susceptible, intermediate, or resistant according to the Clinical and Laboratory Standards Institute (CLSI) guidelines [22]. Antibiotics tested included commonly used agents such as Amikacin (AK), Amoxicillin (AMC), Ceftazidime (CA2), Colistin (CL), Gentamicin (CN), Cefotaxime (CTX), Ciprofloxacin (CIP), Cefepime (CPM), Imipenem (IPM), Tetracycline (TE), Nitrofurantoin (NIT/F).

2.6 Definition of Multidrug Resistance (MDR)

Multidrug resistance was defined as resistance to at least one agent in three or more antimicrobial classes, in accordance with internationally accepted criteria [23].

2.7 Statistical Analysis

Data obtained from laboratory investigations were analyzed using Statistical Package for the Social Sciences (SPSS) software version 25.0 (IBM Corp., USA). Descriptive statistics were used to summarize the data. Associations between categorical variables were evaluated using the Chi-square (χ^2) test. A p-value of less than 0.05 was considered statistically significant [24].

3. Results

3.1 Demographic Characteristics of the Study Population

A total of 100 stool samples were collected from children presenting with diarrheal symptoms, with ages ranging from 1 to 12 years. The study population consisted of 57 male and 43 female patients. All specimens were obtained from stool samples and subjected to microbiological analysis. The distribution of cases showed a slightly higher occurrence of diarrheal infections among males compared to females, Figure (1).

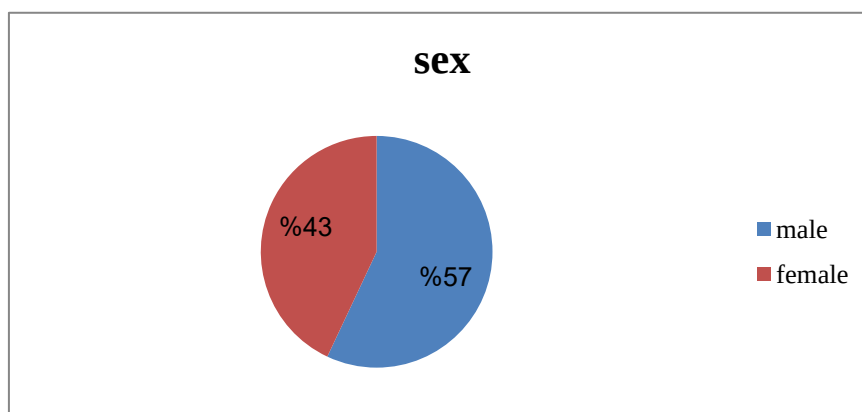


Figure 1. The study population consisted of 57 male and 43 female patients.

Based on age the samples were distributed into 4 groups including (1-4), (4-6), (7-9), and (10-12) and the results revealed that the distribution of positive and negative samples among different age groups was interpreted using the chi-square test. While

positivity rates increased slightly with age, the association between age group and positivity status was not statistically significant ($\chi^2 = 2.29$, $df = 3$, $p > 0.05$), as shown in table (1).

Table 1. Distribution of samples by age group

Age Group	Total Sample	Positive	Negative
1–3	69	57	12
4–6	14	13	1
7–9	11	10	1
10–12	6	6	0
Total	100	86	14
$\chi^2 = 2.29, df = 3, p > 0.05$			

The distribution of bacterial isolates among different age groups was analyzed using the chi-square (χ^2) test to evaluate the association between age and bacterial species. The results demonstrated that *Escherichia coli* was the most frequently isolated bacterium, particularly in the 1–3 years age group (20 isolates), followed by *Klebsiella pneumonia* (17 isolates) and *Pseudomonas aeruginosa* (11 isolates). Lower frequencies were observed for other bacterial species, including *Enterobacter*, *Raoultella*, *Citrobacter freundii*, *Proteus*, *Burkholderia cepacia*,

and *Pantoea*. Statistical analysis using the chi-square test showed no significant association between age group and distribution of bacterial isolates ($\chi^2 = 27.06, df = 24, p > 0.05$). Although the majority of isolates were detected in the 1–3 years age group, the variation in bacterial distribution among the studied age groups was not statistically significant. The highest frequency of isolates was observed in the 1–3 years age group, particularly for *E. coli* and *Klebsiella pneumonia*.

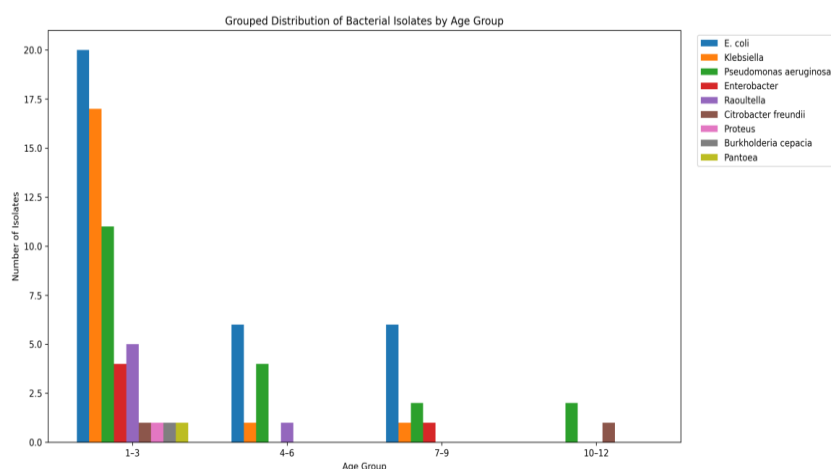


Figure 2. Distribution of bacterial isolates by age group

3.2 Bacterial Isolation and Identification

All collected samples were cultured and analyzed using the VITEK 1 Compact for bacterial identification. The results revealed the presence of various Gram-negative bacteria, as shown in Table (2). *E. coli* was the most frequently isolated organism among the samples. Other identified bacteria included *P. aeruginosa*, *K. pneumoniae*, *Raoultella*

ornithinolytica, *E. cloacae* complex, and *C. freundii*.

Some samples showed no bacterial growth or were recorded as negative. The predominance of *E. coli* highlights its significant role as a causative agent of diarrheal infections in the studied population.

Table 2: Frequency distribution of bacterial isolates identified by the VITEK 1 compact

Bacterial species	Number of isolates (n)	Percentage (%)
<i>Escherichia coli</i>	30	30.0
<i>Pseudomonas aeruginosa</i>	15	15.0
<i>Klebsiella pneumoniae</i>	14	14.0
<i>Raoultella ornithinolytica</i>	5	5.0
<i>Enterobacter cloacae</i> complex	2	2.0
<i>Citrobacter freundii</i>	1	1.0
<i>Pantoea</i> spp.	1	1.0
<i>Kluyvera cryocrescens</i>	1	1.0
Enterobacteriaceae spp.	3	3.0
No growth	11	10.0
Total	100	100.0

3.3 Colony Morphology

The bacterial isolates exhibited characteristic colony morphologies on selective and differential media (Figure 3). On MacConkey agar, lactose-fermenting organisms such as *E. coli* and *K. pneumoniae* produced pink to purple colonies, indicating lactose fermentation. In contrast, non-

lactose fermenting organisms, including *P. aeruginosa*, appeared as pale or colorless colonies. On EMB agar, *E. coli* isolates demonstrated a distinctive metallic green sheen, confirming strong lactose fermentation. These findings are consistent with standard microbiological identification characteristics.

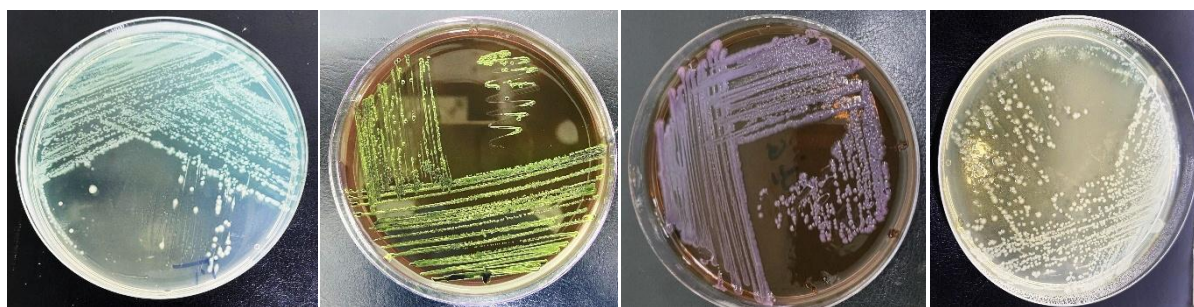


Figure 3: Colony morphology of bacterial isolates on MacConkey and EMB agar

3.4 Antimicrobial Susceptibility Patterns

Antimicrobial susceptibility testing was performed against a panel of commonly used antibiotics, as presented in Tables 3. The results demonstrated a high level of resistance among the isolates to amoxicillin (AMC), cefotaxime (CTX), and tetracycline (TE/DO). Moderate resistance was observed against gentamicin (GM) and ceftazidime

(CAZ). In contrast, higher susceptibility rates were observed for imipenem (IPM), ciprofloxacin (CIP), and amikacin (AK), indicating that these antibiotics remain relatively effective against the tested isolates. The variability in antimicrobial susceptibility patterns reflects the presence of both susceptible and resistant bacterial strains within the study population.

Table 3: Antibiotic susceptibility patterns of bacterial isolates

Antibiotic	Sensitive (n, %)	Resistant (n, %)	Total
Amikacin (AK)	26 (65.0%)	14 (35.0%)	40
Amoxicillin (AMC)	10 (25.0%)	30 (75.0%)	
Ceftazidime (CAZ)	22 (55.0%)	18 (45.0%)	
Colistin (CL)	12 (30.0%)	28 (70.0%)	
Cefotaxime (CTX)	8 (20.0%)	32 (80.0%)	
Ciprofloxacin (CIP)	30 (75.0%)	10 (25.0%)	
Cefepime (CPM)	18 (45.0%)	22 (55.0%)	
Imipenem (IPM)	34 (85.0%)	6 (15.0%)	
Gentamicin (GM)	20 (50.0%)	20 (50.0%)	
Tetracycline (TE)	6 (15.0%)	34 (85.0%)	
Nitrofurantoin (NIT)	10 (25.0%)	30 (75.0%)	
$\chi^2 = 57.71$, df = 10, P < 0.001			

3.5 Multidrug Resistance (MDR)

A considerable proportion of bacterial isolates were classified as multidrug-resistant (MDR), as they exhibited resistance to three or more classes of antimicrobial agents. MDR was predominantly observed among *E. coli*, *K. pneumoniae*, and *P. aeruginosa* isolates. The presence of multidrug-resistant strains indicates a widespread distribution of resistant pathogens in pediatric diarrheal cases and highlights the potential challenges in treatment and infection control. The results of the antibiotic susceptibility test exhibited a high prevalence of multidrug resistance (MDR) among the intestinal bacterial isolates. Several samples, such as S11, S17, S18, S24, S30, S38, S79, and S93, exhibited resistance to nearly all antibiotics included in the panel, suggesting not only MDR but potentially

extensive drug resistance. Overall, antibiotics such as CTX (cefotaxime), TE/DO (tetracycline/doxycycline), AMC (amoxicillin-clavulanic acid), and CL showed very high resistance rates across the isolates. In contrast, CIP (ciprofloxacin) and IPM (imipenem) demonstrated the highest effectiveness, with large inhibition zones observed in many samples, followed by CPM, which showed moderate to good activity. Antibiotics like AK, CN, and CA2 displayed variable effectiveness, indicating partial susceptibility among isolates. Overall, these findings highlight the widespread occurrence of MDR bacteria and the limited number of antibiotics that remain effective, emphasizing the importance of continuous monitoring and rational antibiotic use, figure (4a and 4b).

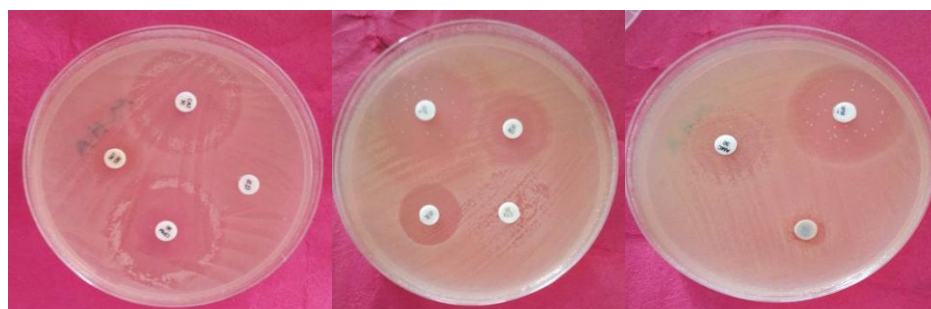


Figure 4a. The disk diffusion method of Antibiotic susceptibility test

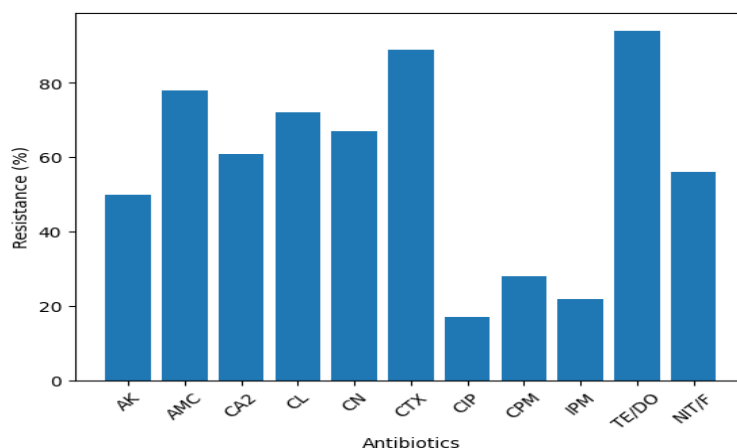


Figure 4b. Antibiotic resistance percentages

4. Discussion

The present study investigated the bacterial profile and antimicrobial susceptibility patterns of isolates obtained from children with diarrhea. The findings demonstrated that *E. coli* was the most predominant pathogen, followed by *P. aeruginosa* and *K. pneumoniae*. These results are in agreement with previous studies, which have consistently reported diarrheagenic *E. coli* as the leading cause of pediatric diarrheal infections worldwide [5]. Similar findings were also identified *E. coli* as the most common isolate among children with diarrhea [4].

The detection of other Gram-negative bacteria, including *K. pneumoniae* and *P. aeruginosa*, further supports the polymicrobial nature of diarrheal infections. Comparable observations have been reported in studies conducted in developing countries, where these opportunistic pathogens are frequently isolated alongside *E. coli* [8]. This may be attributed to environmental contamination and compromised hygiene conditions, which facilitate the transmission of multiple enteric pathogens. The antimicrobial susceptibility results revealed a high level of resistance to commonly used antibiotics, particularly cefotaxime (80%), tetracycline (85%), and amoxicillin (75%). These findings are consistent with previous reports that have documented increasing resistance to β -lactam antibiotics and tetracycline among enteric bacteria [11, 13]. The high resistance to cefotaxime observed in this study suggests the possible presence of extended-spectrum β -lactamase (ESBL)-producing strains. Similar findings were documented a high global prevalence of ESBL-producing *E. coli* [14]. Moderate resistance levels were observed for ceftazidime (45%) and gentamicin (50%), which is comparable to findings reported in other regional studies indicating variable susceptibility patterns among Gram-negative bacteria [13]. This variability

may reflect differences in antibiotic usage practices and local resistance trends.

In contrast, higher susceptibility rates were observed for imipenem (85%), ciprofloxacin (75%), and amikacin (65%), indicating that these antibiotics remain relatively effective. These findings are consistent with previous studies reporting that carbapenems, particularly imipenem, retain high activity against multidrug-resistant Gram-negative bacteria [25]. Similarly, ciprofloxacin and amikacin have been reported as effective treatment options in several studies, although emerging resistance remains a concern [8]. The high prevalence of multidrug-resistant (MDR) isolates observed in this study is particularly alarming. The occurrence of MDR strains among *E. coli*, *K. pneumoniae*, and *P. aeruginosa* indicates a significant public health concern. These findings are in agreement with studies, which reported a high frequency of MDRE. *coli* in pediatric populations [4, 8]. The presence of MDR organisms complicates treatment strategies and increases the risk of treatment failure.

The observed resistance patterns can be explained by the dissemination of resistance genes through horizontal gene transfer mechanisms, including plasmids and integrons, which facilitate the rapid spread of antimicrobial resistance among bacterial populations [26]. In addition, factors such as inappropriate antibiotic use, self-medication, and lack of regulation contribute significantly to the emergence of resistant strains [12]. Former study found that pathogenic isolates from clinical samples exhibited resistance to Bacitracin and Oxacillin whereas they were affected by Levofloxacin, Ceftriaxone, Cefotaxime, Ciprofloxacin, Meropenem, and Tazobactam; also Imipenem are more effective in contrast to other antibiotics [27]. Other study found that the percentage of antibiotic resistance of bacterial species obtained from different sources were

82.75%, 49.42%, 41.37%, 39.08%, 22.98%, 24.13%, 9.19%, 4.59%, and 4.59%; respectively against Amoxicillin, Tetracycline, Trimethoprim, Chloramphenicol, Nalidixic acid, Ciprofloxacin, Streptomycin, Gentamycin and Amikacin [28]. Amoxicillin, Penicillin, Gentamycin, Ceftriaxone, Nalidixic acid, Tetracycline, Ciprofloxacin, Erythromycin, Nitrofurantoin, Amikacin, Imipenim, Ceftazidime, Vancomycin, Ampicillin, Chloramphenicol, Piperacillin, Azithromycin, and Cefotaxime were resisted by different isolates that were obtained from clinical specimens and the percentages were (57) 68.70%, (23) 27.70%, (22) 26.50%, (19) 22.90%, (17) 20.4%, (14) 16.90%, (13) 15.70%, (10) 12%, (10) 12%, (8) 9.60%, (8) 9.60%, (7) 8.40%, (7) 8.40%, (6) 7.20%, (6) 7.20%, (4) 4.80%, (3) 3.60%, and (3) 3.60%, respectively [29]. However, other investigation exhibited that Ampicillin (AM-10 µg) was the least affect antibiotic (81.80%), and Amikacin (AK-30 µg) was the most affect antibiotic was 9%. Ceftriaxone was resistant by 13 isolates. Although CIP is the most commonly used medication among patients in Iraq, it has shown a high level of resistance[30]. Despite the valuable findings, certain limitations should be considered. The analysis was based on available and interpretable susceptibility data, and some samples lacked complete antibiotic profiles. Nevertheless, the study provides important insights into the current resistance patterns among enteric pathogens in the studied population. Overall, the findings of this study

are consistent with global trends showing increasing antimicrobial resistance among diarrheagenic bacteria.

5. Conclusion

The present study demonstrated that *E. coli* is the predominant bacterial pathogen associated with diarrheal infections in children, followed by other Gram-negative bacteria such as *P. aeruginosa* and *K. pneumoniae*. The antimicrobial susceptibility patterns revealed a high level of resistance to commonly used antibiotics, particularly β -lactams and tetracycline, while relatively higher susceptibility was observed for imipenem, ciprofloxacin, and amikacin. The high prevalence of multidrug-resistant (MDR) isolates identified in this study highlights a significant public health concern, as it limits therapeutic options and increases the risk of treatment failure. These findings reflect the growing global problem of antimicrobial resistance, particularly in developing regions where antibiotic misuse and inadequate infection control practices are common. Overall, the study emphasizes the urgent need for continuous surveillance of antimicrobial resistance patterns, rational use of antibiotics, and implementation of effective antimicrobial stewardship programs. Strengthening hygiene practices, improving sanitation, and promoting awareness about appropriate antibiotic use are essential measures to reduce the burden of diarrheal diseases and prevent the spread of resistant pathogens among children.

References

1. Troeger, C., Blacker, B.F., Khalil, I. A., Rao, P.C., Cao, S., Zimsen, S. R. M., et al. (2018). Estimates of the global, regional, and national morbidity, mortality, and aetiologies of diarrhoea in 195 countries. *The Lancet Infectious Diseases*, 18(11), 1211–1228.
2. GBD Diarrhoeal Diseases Collaborators. (2020). Global burden of diarrhoeal diseases, 1990–2019. *The Lancet*, 395(10239), 121–134.
3. Khalil, I. A., Troeger, C., Rao, P.C., Blacker, B.F., Brown, A., Atherly, D. E., et al. (2018). Morbidity and mortality due to diarrheal

- diseases. *The Lancet Infectious Diseases*, 18(11), 1229–1240.
4. Zelelie, T.Z., Eguale, T., Yitayew, B., Abeje, D., Alemu, A., Seman, A., Mihret, A., & Abebe, T. (2023). Molecular epidemiology and antimicrobial susceptibility of diarrheagenic *Escherichia coli* isolated from children. *PLoS ONE*, 18, e0288517.
 5. Gomes, T. A. T., Elias, W. P., Scaletsky, I. C. A., Guth, B. E. C., Rodrigues, J. F., Piazza, R. M. F., et al. (2016). Diarrheagenic *Escherichia coli*. *Frontiers in Cellular and Infection Microbiology*, 6, 25.
 6. Kaper, J.B., Nataro, J.P., & Mobley, H. L. T. (2019). Pathogenic *Escherichia coli*. *Nature Reviews Microbiology*, 17(2), 123–140.
 7. Pakbin, B., Brück, W. M., & Rossen, J. W. A. (2021). Virulence factors of enteric pathogenic *Escherichia coli*. *International Journal of Molecular Sciences*, 22, 9922.
 8. Mahmud, Z., Shabnam, S. A., Mishu, I.D., Johura, F.T., Mannan, S.B., Sadique, A., Islam, L.N., & Alam, M. (2021). Molecular characterization of multidrug-resistant *Escherichia coli* isolated from diarrheal patients. *Gene Reports*, 23, 101182.
 9. Haghighatpanah, M., Zeighami, H., Mozaffari Nejad, A.S., & Hajipour, N. (2022). Phylogenetic groups and antimicrobial resistance characteristics of *Escherichia coli*. *Arab Journal of Gastroenterology*, 23(2), 102–107.
 10. Byarugaba, D. K., Wokorach, G., Alafi, S., Erima, B., Najjuka, F., Mworozzi, E. A., Kibuuka, H., & Wabwire-Mangen, F. (2023). Genetic diversity and virulence-associated genes in *Escherichia coli*. *Microorganisms*, 11, 1868.
 11. Prestinaci, F., Pezzotti, P., & Pantosti, A. (2015). Antimicrobial resistance: A global multifaceted phenomenon. *Pathogens and Global Health*, 109(7), 309–318.
 12. Rhee, C., Aol, G., Ouma, A., Audi, A., Muema, S., Auko, J., Omere, R., & others. (2019). Inappropriate use of antibiotics for childhood diarrhea case management. *BMC Public Health*, 19, 468.
 13. Vila, J., Sáez-López, E., Johnson, J.R., Römling, U., Dobrindt, U., Cantón, R., & Giske, C.G. (2016). *Escherichia coli*: An old friend with new challenges. *International Journal of Antimicrobial Agents*, 48(4), 389–395.
 14. Bezabih, Y.M., Sabiiti, W., Alamneh, E., Bezabih, A., Peterson, G.M., Bezabhe, W. M., & Roujeinikova, A. (2021). The global prevalence of ESBL-producing *Escherichia coli*. *Journal of Antimicrobial Chemotherapy*, 76(1), 22–29.
 15. Tornberg-Belanger, S.N., Rwigy, D., Mugo, M., Kitheka, L., Onamu, N., Ounga, D., et al. (2022). Antimicrobial resistance including ESBL among *E. coli* isolated from Kenyan children. *PLoS Neglected Tropical Diseases*, 16, e0010283.
 16. Dwiyanto, J., Hor, J.W., Reidpath, D., Su, T.T., Lee, S. W. H., Ayub, Q., et al. (2022). Pan-genome and resistome analysis of ESBL-producing *Escherichia coli*. *PLoS ONE*, 17, e0265142.
 17. Breijyeh, Z., Jubeh, B., & Karaman, R. (2020). Resistance of Gram-negative bacteria to current antibacterial agents. *Molecules*, 25(6), 1340.
 18. Kalule, J. B., Bester, L. A., Banda, D. L., Derra, F. A., Msefula, C., Smith, A. M., et al. (2024). Molecular epidemiology and AMR perspective of diarrhoeagenic *Escherichia coli* in Africa: A systematic review and meta-analysis. *Journal of Epidemiology and Global Health*, 14, 1381–1396.

19. Cheesbrough, M. (2010). *District laboratory practice in tropical countries* (2nd ed.). Cambridge University Press.
20. Abdulhusein, S.H. (2026). Investigation of multidrug-resistant *Klebsiella pneumoniae* and *Acinetobacter baumannii* isolated from inpatients at Al-Nasiriyah Teaching Hospital. *International Journal of Medical Science and Dental Health*.
<https://doi.org/10.55640/ijmsdh-12-01-03>.
21. Pincus, D. H. (2010). Microbial identification using automated systems. *Clinical Microbiology Reviews*, 23(3), 550–566.
22. Clinical and Laboratory Standards Institute (CLSI). (2022). *Performance standards for antimicrobial susceptibility testing* (32nd ed.). CLSI supplement M100.
23. Field, A. (2013). *Discovering statistics using IBMSPSS statistics* (4th ed.). Sage Publications.
24. Magiorakos, A.P., Srinivasan, A., Carey, R. B., Carmeli, Y., Falagas, M. E., Giske, C. G., et al. (2012). Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria. *Clinical Microbiology and Infection*, 18(3), 268–281.
25. Dadgostar, P. (2019). Antimicrobial resistance: Implications and costs. *Infection and Drug Resistance*, 12, 3903–3910.
26. Bengtsson-Palme, J., Kristiansson, E., & Larsson, D. G.J. (2018). Environmental factors influencing the development and spread of antibiotic resistance. *Nature Reviews Microbiology*, 16(9), 556–564.
27. Kadim, BaidaaMezher. “Distribution of Antibacterial Resistance among Pathogenic Bacterial Isolates from Patient in Al-ShatrahGeneralHospital.” *University of Thi-Qar Journal of agricultural research* (2024), Vol: 13 (2), p: 153-163
28. DebMandal, M., Mandal, S., and Kumar Pal, N., (2011). Antibiotic Resistance Prevalence and Pattern in Environmental Bacterial Isolates. *The Open Antimicrobial Agents Journal*, 2011, 3, 45-52.
29. Alwatar, W. M.A., Suha, A.AlFakhar, SaadHasanMohammedAli, KhalilIsmailAbidMohammed, Jenan M.Mousa (2023). Prevalence of Antibiotics Resistance among Patients in Iraqi Hospitals. *Haya Saudi J Life Sci*, 8(7), 118-126.
30. Abdulhusein H.S., Kadim B.M., Al-A’Dhami R.M. Molecular identification, and phenotypic characterization of antibiotic-resistant bacteria from patients in Al-Nasiriyah General Hospital. *Infekc. bolezni (Infectious Diseases)*. 2026; 24(1): 130–141. DOI: 10.20953/1729-9225-2026-1-130-141.