

Identification and Antibiotic Susceptibility Profiling of *Escherichia coli*
Isolated from Water Sample and Vendor's Hand Swab in Dhaka
Metropolitan, Bangladesh

By

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A thesis submitted to the Department of Mathematics and Natural Sciences in partial
fulfillment of the requirements for the degree of
Bachelor of Science in Microbiology

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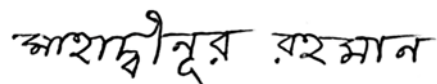
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Declaration

It is hereby declared that

1. The thesis submitted is our own original work while completing a degree at Brac University.
2. The thesis does not contain material previously published or written by a third party, except where this is appropriately cited through full and accurate referencing.
3. The thesis does not contain material which has been accepted, or submitted, for any other degree or diploma at a university or other institution.
4. We have acknowledged all main sources of help.

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Ethics Statement

The ethical permission to conduct this study was obtained from the departmental review board of Brac University.

Abstract/ Executive Summary

The study was focused to look at the microbial composition of water used in vegetable markets and hand swabs of vegetable market vendors to find out whether it can be a source of *E. coli* that are resistant to antibiotics. The samples were collected from four different markets located in four distinct areas of Dhaka. This research aimed to find out the degree of antibiotic resistance among *Escherichia coli* isolated from water and hand swab samples. From the four selected areas 36 samples (24 water samples and 9 hand swab samples) were collected . To be more specific there were two types of water samples, one is vegetable wash water and other is the source water. From analyzing the samples it indicates that vegetable washed water samples contain the most bacterial growth among three types of sample. Most bacterial isolates displayed modest intermediate levels which is between 3.45% and 41.38%, with Gentamicin and Kanamycin having the highest levels. On the contrary, two of the antibiotics Erythromycin and Ampicillin have shown considerably low levels of sensitivity to the isolates while expressing 89.66% and 100.00% of resistance respectively. The lowest rate of intermediate, on the other hand, is within 3%-40% across all isolates. Different antibiotic resistance characteristics might be seen in other isolates. Overall, among the isolates, there were 48.30% isolates showing sensitivity, 48.30% with MDR patterns, and 3.4% with extended drug resistance (XDR). It is alarming that some strains of *Escherichia coli* can cause severe illness like septicemia, meningitis, rheumatoid arthritis, reactive arthritis, atherosclerosis, and haemolytic uremic syndrome.

Keywords: *Escherichia coli*; Antibiotic susceptibility; Isolates; MDR; XDR; PCR.

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List of Acronyms

<i>E.coli</i>	<i>Escherichia coli</i>
MAC	MacConkey Agar
PCR	Polymerase Chain Reaction
EMB	Eosin Methylene Blue Agar
MDR	Multi Drug Resistant
XDR	Extensively Drug Resistant
NA	Nutrient Agar

Chapter 1

Introduction

1.1 Outbreaks and Disease Burden Linked to *E. coli* in Food, Water, and the Market Environment:

Safe water is fundamental for maintaining food hygiene, yet in many urban markets across developing nations, including Bangladesh, water often becomes a major vehicle of microbial contamination (Parveen 2008; Sharif et al. 2025) . In crowded market environments, water is widely used for rinsing vegetables, cleaning surfaces, and washing hands, but it is frequently sourced from untreated supplies or stored under unhygienic conditions(Zahra et al. 2025) . Such practices provide opportunities for the introduction and persistence of fecal contaminants, which may in turn spread to fresh produce and ultimately to consumers. Contaminated water thus represents a critical pathway for the transmission of foodborne pathogens and a significant public health risk.

Among microbial indicators of fecal contamination, *Escherichia coli* (*E. coli*) holds particular importance. While many strains are harmless commensals, pathogenic and antibiotic-resistant variants pose severe threats to human health (Rakib et al. 2015; Saima et al. 2023). The presence of *E. coli* in market water or on vendors' hands not only signals poor sanitation but also highlights the risk of direct transfer of pathogens into the food chain (Talukdar et al. 2013) . In urban settings such as Dhaka, where vendors often reuse wash water and lack access to regulated sanitation facilities, the likelihood of *E. coli* contamination is markedly increased (Luna-Guevara et al. 2019) . This contamination may originate from multiple sources, including polluted municipal supplies, cross-contamination from storage containers, or direct introduction via handling.

Compounding the issue is the rising prevalence of antibiotic resistance among *E. coli* strains in Bangladesh. The misuse of antibiotics in human medicine, agriculture, and aquaculture has accelerated the emergence of multidrug-resistant variants, some of which have already been detected in community and environmental samples. When such resistant strains are introduced into market water systems, they can spread rapidly to consumers through contact with food or hands, leading to infections that are difficult to treat. Therefore, assessing the presence and antibiotic susceptibility of *E. coli* in market water and hand swabs is essential to understanding contamination dynamics, guiding food safety practices, and shaping effective public health interventions. Multiple large, well-documented outbreaks illustrate how fecal contamination in agricultural water, processing water, or handling can seed *E. coli* through the supply chain into consumers. In the United States, the 2006 multistate spinach outbreak (*E. coli* O157:H7) caused more than 200 illnesses and three deaths; investigations pointed to environmental contamination in the growing region, including water and animal vectors near fields (CDC MMWR, 2006; FDA/CIDRAP, 2007; Jay et al., 2007). Subsequent produce-linked events—such as the 2018 romaine lettuce outbreak from the Yuma growing region (including five deaths) and a 2021 baby-spinach outbreak—underscore the recurring role of water and post-harvest handling in pathogen transfer to leafy greens (CDC, 2018; CDC, 2021).

Earlier, the 1996 Odwalla unpasteurized apple-juice outbreak demonstrated how minimally processed beverages can carry *E. coli* O157:H7 when source fruit or process water are contaminated, resulting in severe illness and a child's death (CDC/MMWR & Ann Intern Med reports).

These outbreak patterns are mechanistically consistent with experimental and field studies showing that Shiga toxin–producing *E. coli* can be transferred from contaminated manure or irrigation water to leafy tissues and even internalize within plants, making post-wash decontamination challenging (Solomon et al., 2002; Chekabab et al., 2013).

Contemporary risk assessments emphasize that water used post-harvest (washing, fluming, cooling) is a critical cross-contamination node for fresh produce; poor water quality or suboptimal water management can disseminate pathogens rapidly across large product lots (EFSA, 2025).

In Bangladesh, environmental monitoring and market-adjacent studies intensify the concern. ESBL-producing, multidrug-resistant *E. coli* have been characterized in urban surface waters in Dhaka, highlighting a plausible environmental reservoir intersecting with produce-handling and handwashing practices (Hossain et al., 2025).

Market-relevant hygiene studies similarly report multi-antibiotic-resistant bacteria on food handlers' hands and utensils in Dhaka restaurants, aligning with risks from vendor handwashing in unsafe water or from re-used wash basins (Akter et al., 2025; Hassan et al., 2017).

Together with evidence of microbial contamination in municipal and peri-urban waters around Dhaka, these findings support a credible pathway from environmental waters and handling surfaces to ready-to-eat foods (Parveen et al., 2008; Sharif et al., 2025).

1.2 Factor Behind Contamination:

Several factors contribute to the contamination of water in market environments, creating favorable conditions for the survival and spread of *Escherichia coli*. Poor sanitation infrastructure, such as leaking sewage lines and unprotected water sources, often allows fecal matter to mix with water supplies. Inadequate storage practices, where water is kept in uncovered or unclean containers, further increase the risk of microbial growth. Repeated use of the same water for washing vegetables or cleaning utensils enables cross-contamination, while insufficient hand hygiene among vendors can directly introduce bacteria into the water (Parveen 2008; Zahra et al. 2025). Environmental conditions such as warm temperatures, high humidity, and the presence of organic matter also promote bacterial persistence. Together, these factors make market water a highly vulnerable medium for transmitting *E. coli* to both vendors and consumers.

1.3 Objectives : The objectives of this study are:

1. To isolate and identify *Escherichia coli* from vegetable wash water, source water, and hand swab samples collected from selected markets in Dhaka city.
2. To determine the prevalence of *E. coli* contamination in different types of samples and across various market areas.
3. To evaluate the antibiotic susceptibility patterns of the isolated *E. coli* strains using standardized antimicrobial testing methods.
4. To assess the extent of multidrug resistance (MDR) and extensively drug resistance (XDR) among the isolates.
5. To generate baseline data that may guide public health interventions, food safety policies, and antimicrobial resistance (AMR) surveillance in urban market environments of Bangladesh.

Chapter 2

Materials and Methods

2.1 Area and Sample Collection:

To conduct the study, 4 different areas have been selected in the time period of March 2024 and February 2025 inside Dhaka city. Water samples have been collected from several vegetable markets that are quite popular among the people of the certain regions which are selected for sampling. The popularity of the markets is an indication that the majority people of those chosen areas are the consumers of those markets. Next, the samples are taken to the BRAC University Research Laboratory the same day the samples were collected within an isolated airtight icebox maintaining all the necessary protocols.

The 4 areas are: 1. Rampura, 2. Badda, 3. Gulshan 1 and 4. Gulshan 2.



Figure 1: Here three of the above pictures represent hand swap sample, wash water sample and source water sample respectively.

2.2 Preparation of Samples:

After the gathered samples were brought inside the lab, they were removed inside a laminar airflow to avoid further contamination. Then from each 1ml of sample were diluted with 9ml of peptone water for enrichment. The samples were then kept in a shaker incubator for 24 hours at 37 degrees Celsius for the second step. (Sun et al. 2018)

2.3 Enumeration of *E.coli*:

After 24 hours of incubation, each sample was 3 fold diluted. 0.1mL of each dilution was transferred and disseminated on MacConkey agar using a sterilized glass spreader under laminar airflow. The plates were then incubated at 37°C for 24h (Hudzicki 2016) . MacConkey (MAC) agar was used to recover presumptive *E. coli* from enriched, serially diluted samples. The colonies exhibiting dry pinkish texture and displaying the required colony morphology were selected for suspected *E. coli* count. The overall bacterial count was represented as colony-forming units per gram of wash water, source water, hand swab samples.



Figure 2 : *E. coli* colonies in MacConkey (Hi-media) Agar

2.4 Isolation and Identification:

Suspected isolates from MacConkey agar plates were then subcultured into NA agar plates for purity (37 °C, 24 h). After that, presumptive isolates were then confirmed on Eosin Methylene Blue (EMB) agar, a selective-differential medium in which *E.coli* typically produces a green metallic sheen due to acid-driven dye precipitation (Berry et al. 1984) ; Gram-positive growth is inhibited by the dyes. Here colonies exhibiting lactose fermentation causing change of color to green metallic sheen and displaying the required colony morphology were selected. Final identification followed standard cultural, morphological, and basic biochemical characteristics.



Figure 3 : *E.coli* isolates in EMB agar

Antibiotic susceptibility was assessed by the Kirby–Bauer disk diffusion method on Mueller–Hinton agar (MHA), recommended by CLSI because it is nonselective, nondifferential, has good batch reproducibility, contains starch that absorbs bacterial toxins, and permits reliable antibiotic diffusion (Hudzicki 2016; CLSI 2024a) . After 24 h growth, test organisms were uniformly swabbed onto MHA with sterile cotton swabs; antibiotic disks were applied using flame-sterilized tweezers, and plates were incubated at 37 °C for 24 h. Zones of inhibition were measured to determine susceptibility, recognizing that effective antibiotics produce clear no-growth zones. Results were interpreted against standard

breakpoints (e.g., Oxoid/NARMS) to classify isolates as sensitive, intermediate, or resistant, and all measurements were recorded for analysis and visualization.

2.5 DNA Extraction:

The necessary samples are prepared for extraction by combining each organism's bacterial growth with 1000 μL of Brain Heart Infusion (BHI) medium in an autoclaved Eppendorf (Khehra et al. 2023). These Eppendorf are then cultivated for 24 hours at 37 degrees Celsius in a shaker incubator. The samples are prepared for DNA extraction the next day. (Odetoyin et al., 2022) The entire operation is being carried out using the boiling technique. The samples are first centrifuged at 4 degrees Celsius for 5 minutes before being thrown away. After that, 50 μL of nuclease-free water was added, pipetted to resuspend it for a better mix, and then it was brought to a boil for five minutes at 100 degrees Celsius.

2.6 Amplification of suspected *E.coli*:

It's a lab procedure that quickly creates millions to billions of duplicates of a DNA fragment. According to Britannica, T. Editors of Encyclopaedia 2023, this process swiftly and efficiently creates a large number of duplicates that aid in examinations such as molecular biology, forensic analysis, and medical diagnostics.(Khehra et al., 2023) Replication cycles utilizing a PCR equipment (The Applied Biosystems 2720 Thermal Cycler) were completed in a matter of hours.

To carry out the process each primer was added to a 25 μ L PCR mixture that includes 5 μ L of nuclease-free water, 2 μ L of forward primer, 2 μ L of reverse primer, 12 μ L of PCR master mix (Emerald Amp), and 4 μ L of template DNA. The mixture was vortexed for 3-5 sec to ensure correct mixing. Following that, PCR was carried out while preserving the PCR settings for each target gene. All the PCR products were to be sorted at - 20°C for additional examination after the PCR ended.

2.7 Targeted gene and primer:

The suspected bacterial isolates were screened for confirmation by using primers dedicated for *E. coli* (ECO-1 & ECO-2 primers). PCR amplification was done with the following set of primers mentioned in the table below.

Target Gene	Primer	Sequence	Amplicon Size	Conditions	Reference
16srRNA	ECO-1	F- 5'-GACCTCGGTTTAGTTCACAGA-3'	585 bp	7 min at 95°C; 1 min at 94°C; 1 min at 55°C; 1 min at 72°C; 7 min at 72°C; 35 Cycle	(Ahmed et al., 2018)
	ECO-2	R-5'-CACACGCTGACGCTGACCA-3'			

Table 1 : Primer and PCR condition for this study (Ahmed et al.
2018)

2.8 Agarose Gel Electrophoresis: One of the best techniques for separating DNA fragments with sizes between 100 bp and 25 kp is agarose gel electrophoresis. In the agarose gel matrix, the biomolecules are sorted by size by pushing charged molecules across the material using an electric field. There are several physical methods available for determining the size of DNA, including agarose gel electrophoresis. This method involves passing DNA through a heavily cross-linked agarose substrate while an electric current is present. Due to the negative charge of the DNA phosphates in solution, the molecule will gravitate toward the positive (red) pole. (Khehra et al., 2023)

Three factors affect how quickly DNA moves across a gel, providing information about the size of the DNA and confirming its existence when a certain DNA ladder is utilized. The necessary agarose powder (usually 1.5–2%) is boiled with 2000 μL of TAE Buffer and 98% distilled water to create a gel for the tests. Then, as a fluorescent tag, 4 μL of ethidium bromide, an intercalating agent, was added. Using autoclaved tips, the previously completed PCR products were subsequently placed onto an agarose gel. In this case, TAE (tris-acetate-EDTA) was used as a running buffer to aid in the migration of DNA to the positive electrode (Khehra et al. 2023) . Agarose gel electrophoresis was carried out each time with 100 V of electricity and 5 μL of Biolab 100 bp ladder. It was possible to continue tracing the DNA profession across the gel since the DNAs are colored with MIDORI Green. The material that is used to stain the DNA after it has been processed through the gel attaches specifically to DNA molecules and either fluoresces a particular color when exposed to UV light or reflects a particular color when exposed to visible light. Thus, the band sizes are determined and recorded with the help of the used DNA ladder.

2.9 Antibiotic Susceptibility Test:

Different antibiotics work in different ways. These cause different mechanisms of bacterial resistance to antibiotics to arise. Therefore, in order to properly gather information about the *E. coli* bacterium, eleven antibiotics in all have been employed in this experiment.(Magiorakos et al., 2012) The following table describes the antibiotic disks utilized in this investigation, along with their mode of action and resistance mechanisms:

Antibiotic Group	Antibiotic Name	Disc code	Disc potency (µg)	Sensitive (mm)	Intermediate (mm)	Resistant (mm)
Aminoglycosides	Amikacin	AK	30	≥17	15-16	≤ 14
	Gentamicin	GEN	10	≥15	13-14	≤ 12
	Kanamycin	KAN	30	≥18	14-17	≤ 13
	Streptomycin	S	10	≥15	12-14	≤ 11
Carbapenem	Imipenem	IMI	10	≥23	18-22	≤ 19
	Meropenem	MEM	10	≥23	18-22	≤ 19
β-lactam	Ampicillin	AMP	10	≥17	12-16	≤ 13

Fluoroquinolone	Ciprofloxacin	CIP	5	≥ 21	16-20	≤ 15
Macrolides	Erythromycin	E	15	≥ 23	14-22	≤ 13
Phenicol	Chloramphenicol	C	30	≥ 18	13-17	≤ 12
Polymyxin	Colistin	CT	10	≥ 17	12-16	≤ 11
Tetracycline	Tetracycline	TE	30	≥ 15	12-14	≤ 11

Table 2 : Concentrations and diffusion zones of the antibiotics.

After a 24-hour incubation period, the sample organisms are first scattered onto nutritional agar (NA) culture plates using autoclaved cotton swabs. The swabs are soaked in distilled water and then mixed with the liquids. The cotton swabs are then swiped from different directions to guarantee that the bacteria are dispersed uniformly over the plates. Finally, the plates are held for a brief period of time to allow the medium to absorb the suspension.

The next step is to use a sterilized tweezer to insert the necessary antibiotic disks on the plates. The discs are then set on top of the media and gently pushed against it to guarantee direct contact. After that, the tweezer is heated and dipped in alcohol to sanitize it for future usage. Therefore, following the same procedure, all of the antibiotic disks are put on the MHA plates and incubated at 37 degrees celsius for 24 hours.

Bacterial growth was seen surrounding each disc when the MHA plates were removed after a day of incubation. There should be "no growth" surrounding the tested organism if the specific antibiotic is working on it. In a similar vein, an antibiotic that is less effective will cause a noticeable zone of inhibition. In these situations, the zone of inhibitions is assessed using a scale that provides a clear understanding of the permissible growth constraint zones surrounding the discs.

Oxoid Limited (England) uses a standard table to assess if the results are resistant, intermediate, or sensitive. The National Antimicrobial Resistance Monitoring System advises using a break point to identify if *E. coli* are susceptible or resistant. After that, all of the data collected from the tested sample is arranged in a Microsoft Excel spreadsheet for recording and creating factual visualizations in the form of graphs or charts.

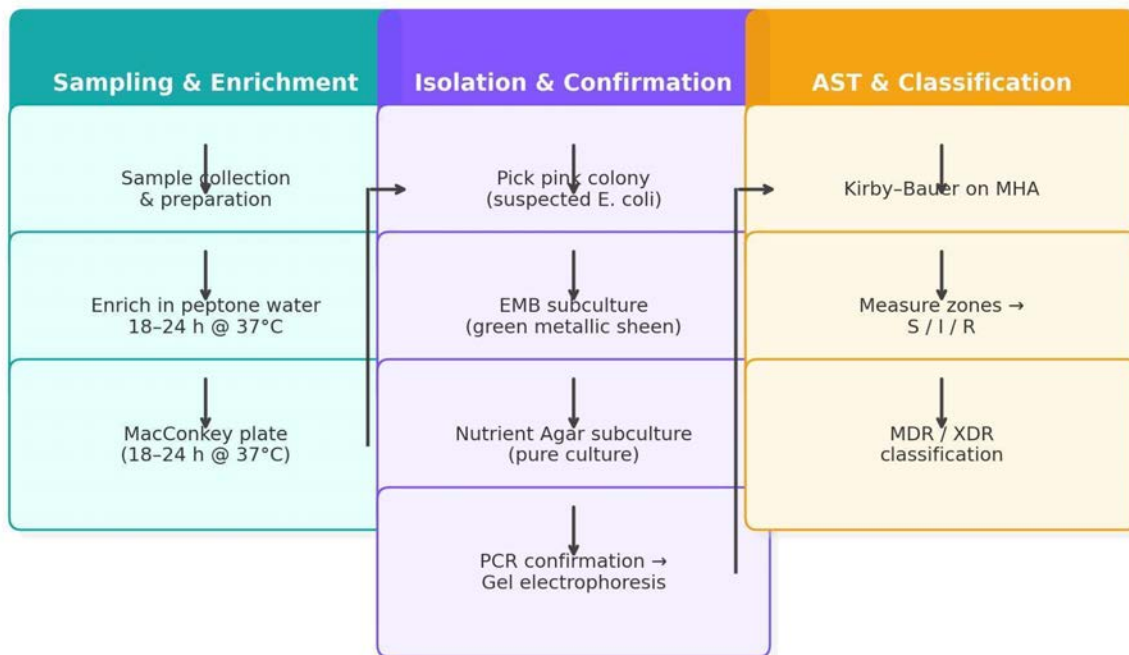


Figure 4 : Flowchart detailing the methods used in this study.

2.10 Agarose Gel Electrophoresis Result:

After performing a gel agarose test, the buffer was taken after the UV light in order to see if there are any bands visible according to the given primer. A picture is attached below to show the bands got from the result:

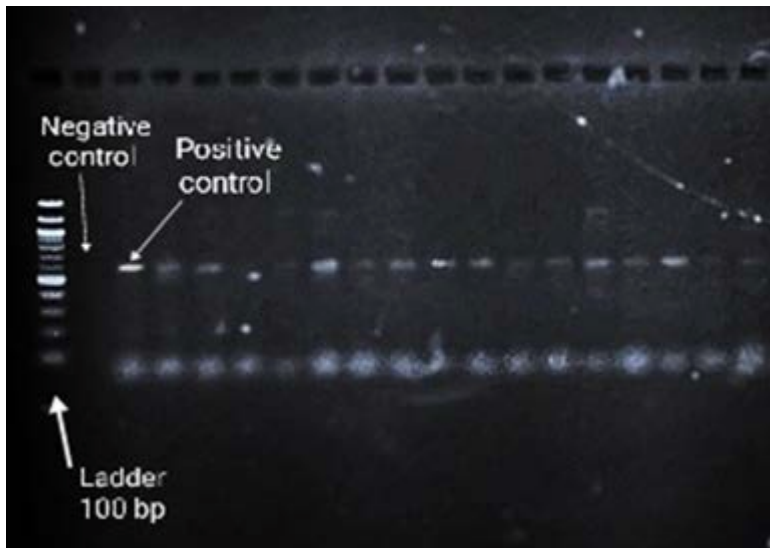


Figure 5 : *E. coli* confirmatory PCR result. [Here the ladder was a 100 bp DNA marker. Lane 1 is laddering, lane 2 was negative control, lane 3 was positive control. From lane 5-16 different samples were loaded). Amplicon size 585 bp.]

Chapter 3

Results

3.1 Positive *E. coli* isolates based on sample:

While analyzing 36 samples 29 positive isolates were found in total within the four regions of Dhaka city. In the collected samples except the source water samples most of the hand swab and wash water samples contain positive *E. coli* isolates though the percentage of positive isolates vary based on area. The highest percentage of positive isolate belongs to Badda which is 38.09%. Then comes Rampura with the score of 35% and Gulshan-2 and Gulshan-1 gradually hold the third and fourth place. Here the higher the percentage of an area the riskier the region to spread contamination to local people.

Area Name	Number of <i>E. coli</i> Sample	Number of isolates in the sample	<i>E. coli</i> isolate	Percentage of <i>E. coli</i> isolate (%)
Badda	6	21	8	38.09%

Rampura	6	20	7	35.00%
Gulshan-1	6	23	7	30.43%
Gulshan-2	6	22	7	31.81%
Total	24	86	29	Average = 33.83%

Table 3 : Positive *E. coli* isolates from samples.

3.2 Test analysis of Antibiotic Susceptibility:Antibiotic susceptibility testing of *E. coli* isolates revealed highly variable resistance patterns across different drug classes.(CLSI, 2024a) Out of 29 positive isolates, the majority exhibited sensitivity to aminoglycosides, carbapenems, and phenicols, while showing strong resistance against β -lactam (Ampicillin) and macrolides (Erythromycin).(Bulik et al., 2010) Gentamicin demonstrated the highest sensitivity with 96.55% of isolates susceptible, followed by Amikacin (89.66%) and Meropenem (82.76%). In contrast, all isolates (100%) were resistant to Ampicillin, and a striking 89.66% were resistant to Erythromycin.(Devirgiliis & Barile, 2024).

Intermediate responses were most notable in Colistin, with 82.76% of isolates falling in the intermediate category, suggesting reduced but not complete resistance.(Matuschek et al., 2018). Kanamycin displayed a mixed profile: nearly half of the isolates (48.28%) were sensitive, while 41.38% showed intermediate resistance and 10.34% were resistant. Streptomycin results indicated a balanced distribution, with 51.72% sensitive, 17.24% intermediate, and 31.04% resistant.

Overall, the findings highlight a concerning trend of multidrug resistance. While certain antibiotics remain effective (e.g., Gentamicin, Amikacin, Ciprofloxacin), the high resistance rates to Ampicillin and Erythromycin underscore the challenge of therapeutic management. The dominance of intermediate resistance in Colistin also signals the potential emergence of resistance against one of the last-resort antibiotics.

A data combining AST results from the positive isolates:

Antibiotic	Sensitive	Intermediate	Sensitive
Streptomycin	15 (51.72%)	5 (17.24%)	9 (31.04%)
Amikacin	26 (89.66%)	2 (6.89%)	1 (3.45%)
Gentamicin	28 (96.55%)	1 (3.45%)	0 (0.00%)
Kanamycin	14 (48.28%)	12 (41.38%)	3 (10.34%)
Meropenem	24 (82.76%)	3 (10.34%)	2 (6.90%)
Imipenem	24 (82.76%)	3 (10.34%)	2 (6.90%)
Ampicillin	0 (0.00%)	0 (0.00%)	29 (100.00%)
Ciprofloxacin	23 (79.31%)	4 (13.79%)	2 (6.90%)
Erythromycin	0 (0.00%)	3 (10.34%)	26 (89.66%)
Chloramphenicol	20 (68.97%)	5 (17.24%)	4 (13.79%)
Tetracycline	25 (86.21%)	2 (6.90%)	2 (6.90%)
Colistin	2 (6.90%)	24 (82.76%)	3 (10.34%)

Table 4 : Antibiotic Susceptibility profile of positive *E. coli* isolates

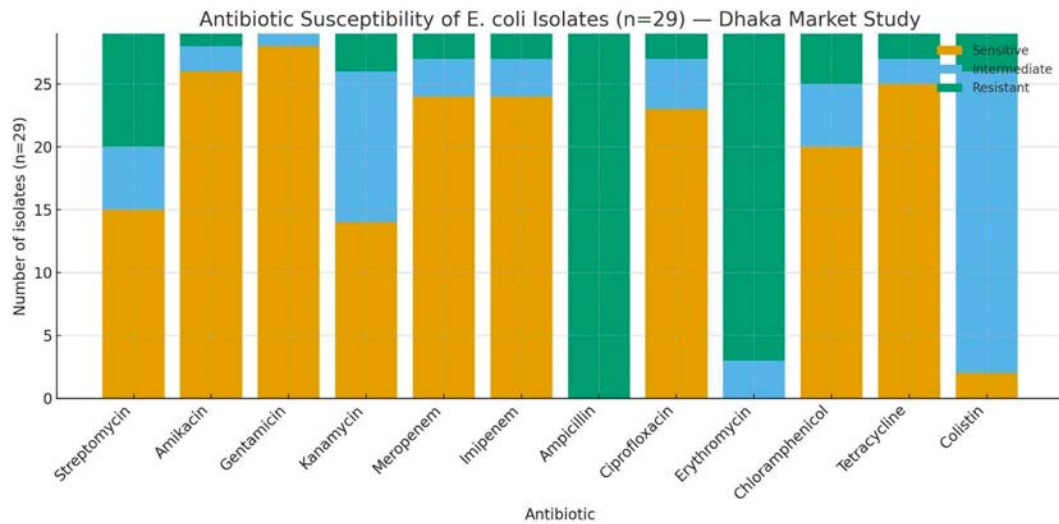


Figure 6 : A graph showing resistance results. (Yellow = Sensitive, Blue=Intermediate, Green = Resistant)

3.3 MDR, XDR and sensitive pie chart:

Distribution of Sensitive, MDR, and XDR Cases

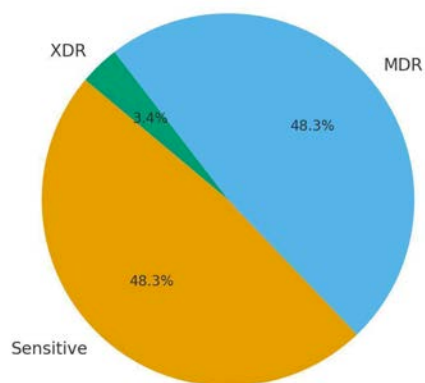


Figure 7 : Multi-drug resistant and sensitive isolates percentage

Chapter 4

Discussion :

This study demonstrates that market-associated water and vendor hands in Dhaka constitute important reservoirs and transmission interfaces for *Escherichia coli*. Sampling was deliberately designed to capture both environmental and human-contact routes—source water available to vendors, vegetable wash water in routine use, and hand swabs—thereby linking potential contamination pathways within the same marketplace micro-ecology. In total, 36 samples were collected, of which 24 were *E. coli* positive, yielding 29 confirmed isolates for downstream testing; notably, vegetable wash water showed the highest apparent bacterial load among the sample types, underscoring the role of water re-use and inadequate storage in amplifying contamination pressure in stalls.(Zahra et al. 2025; Parveen 2008)

Spatially, the proportion of *E. coli*–positive isolates varied across the four surveyed areas, with Badda showing the highest share (38.09%), followed by Rampura (35.00%), Gulshan-2 (31.81%), and Gulshan-1 (30.43%). While the study was not powered to attribute causality to specific infrastructural or behavioral factors by location, these differences plausibly reflect heterogeneity in water sourcing, storage practices, and stall hygiene, and they highlight sub-districts where targeted interventions could yield disproportionate benefits.

Phenotypic susceptibility patterns revealed a striking divergence across antibiotic classes. Aminoglycosides (Gentamicin 96.55% sensitive; Amikacin 89.66%) and carbapenems (Meropenem and Imipenem, each 82.76% sensitive) retained substantial activity against the isolates, whereas β -lactam Ampicillin and the macrolide Erythromycin showed complete and near-complete resistance, respectively (Ampicillin 100% resistant; Erythromycin 89.66% resistant). Mixed or transitional profiles were observed for Kanamycin (48.28% sensitive; 41.38% intermediate) and Streptomycin (51.72% sensitive; 31.04% resistant), while Colistin displayed a predominance of intermediate responses (82.76%), suggesting reduced susceptibility and the potential emergence of mechanisms affecting this last-resort agent. These data collectively portray a market-level *E. coli* population under strong β -lactam and macrolide selection pressure, with narrower—but still meaningful—therapeutic windows preserved for aminoglycosides and carbapenems.

The resistance landscape aligns with concerns about antibiotic use patterns in the community. Complete resistance to Ampicillin and very high resistance to Erythromycin are consistent with widespread availability and use of these agents, while the residual effectiveness of aminoglycosides and carbapenems—though clinically valuable—raises stewardship concerns if such drugs become default empiric choices. The high proportion of intermediate responses to Colistin further signals that even reserve agents may be compromised if environmental selection pressures persist.

Beyond single-drug patterns, the detection of multidrug-resistant (MDR) and extensively drug-resistant (XDR) phenotypes in the isolate set indicates a community-level risk of horizontal gene transfer and persistence in high-turnover water systems typical of wet markets. Because wash water is repeatedly used to rinse produce and hands, it can function as both sink and source, facilitating re-seeding of stalls and utensils and increasing the likelihood that resistant organisms reach consumers. This dynamic blurs the traditional boundary between “clinical” and “environmental” AMR and argues for integrated market-hygiene and antimicrobial-use policies. (Sharif et al. 2025; Saima et al. 2023)

Methodologically, the identification pipeline combined selective/differential culturing with confirmatory steps (EMB for colony morphology and sheen, followed by PCR using ECO-1/ECO-2 primers targeting 16S rRNA), providing internal validity to isolate designation prior to AST. While the Kirby–Bauer disk diffusion method on Mueller–Hinton agar remains a standardized approach for surveillance, interpretation of borderline categories (e.g., “intermediate,” especially for agents like Colistin) should be conservative and ideally corroborated by additional methods in future work (Matuschek et al. 2018) . The study’s strengths include triangulating three contamination points within the market environment and aligning isolate confirmation with established microbiological practice; limitations include modest sample size, confinement to four areas, and phenotypic resistance profiling without genotypic characterization of resistance determinants.

Taken together, the findings support three priority actions tailored to Dhaka's market context: (i) strengthen hygiene infrastructure and routines, with special attention to the quality, storage, and turnover of wash water; (ii) expand antibiotic stewardship and regulatory controls to curtail indiscriminate community use of β -lactams and macrolides; and (iii) institutionalize ongoing AMR surveillance in market water and among food handlers, with escalation to molecular typing where feasible to track transmission (Zahra, M. A. et al. 2025). Absent such measures, the continued circulation of resistant *E. coli* in market water systems is likely to narrow treatment options and sustain the risk of hard-to-treat infections in the community.

Conclusion :

This study shows that market water and vendor contact in Dhaka are key routes for *Escherichia coli* transmission, with vegetable wash water posing the greatest contamination risk. From 36 samples, 29 *E. coli* isolates were confirmed. Susceptibility testing indicated high effectiveness of aminoglycosides (Gentamicin 96.55%, Amikacin 89.66%) and carbapenems (Meropenem/Imipenem 82.76%), but complete resistance to Ampicillin (100%) and very high resistance to Erythromycin (89.66%). Colistin showed predominantly intermediate responses (82.76%). Overall, 24.09% of isolates were MDR and 2.2% were XDR. Immediate priorities include safer water sourcing and turnover, hygienic storage/handling, vendor hand hygiene, and community antibiotic stewardship, supported by routine AMR surveillance in market water. While the sample size and phenotypic focus are limitations, the evidence is clear: targeted, low-cost improvements in water handling and hygiene can substantially reduce the risk of resistant *E. coli* entering the food chain.

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