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Bacteriological assessment of mashed street food (Bharta) and their antibiotic susceptibility phenomena in Dhaka city, Bangladesh

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ABSTRACT

Chitoi Pitha (a cake made from rice powder) is one of the most popular traditional street foods in Bangladesh. What makes Chitoi Pitha truly special is the Bharta that accompanies it. Bharta means mashed raw food items. There are many types of Bharta, usually made with vegetables/fish/seeds. Among them, Mustard seed bharta, dried fish bharta, and Coriander leaf bharta are very popular, especially in the winter season. The raw bharta is tasty. Heating of bharta loses its taste and flavors. Since they are typically eaten raw and without heat treatment or thorough washing, they serve as vectors for the transmission of pathogenic microorganisms. These vectors spread multidrug-resistant pathogens among the large population. A total of 24 bharta samples were collected from 8 different areas of Dhaka city. Selective and non-selective media were used to isolate and enumerate the bacteria. Gram-negative and Gram-positive organisms were 63% and 37% respectively. From the 61 isolates, there were *Staphylococcus aureus* (36%), *E. coli* (31.1%), *Salmonella* spp. (13.1%), *Klebsiella* spp. *(*13.1%), and *Vibrio* spp. (6.5%). Antibiotic susceptibility testing showed Piperacillin, Imipenem, and Co-Trimoxazole were effective against most of the strains, but some of the organisms were multidrug resistant.

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1. Introduction

Street foods are defined as ready-to-eat foods and beverages prepared and/or sold by vendors and hawkers, especially in streets and other similar public places (1).

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Some street food concomitantly contains delicious sauces like bharta. Street food bharta is prepared by smashing the vegetable leaves or seeds/garlic/green chilli/dried fish. If it is cooked, it loses its taste and flavor. It is a raw item eaten with the main food. Bharta contains a huge number of contaminants due to the lack of a cooking procedure. Bharta is very tasty and is used

as a sauce. Sellers and consumers are unaware of the hidden microbial load in bharta. Bharta is a predominant vector for food-borne illness. Street food vending has become an important public health issue and a great concern to everybody (2).

The problem is severe in low-income countries due to difficulties in securing optimal hygienic food handling practices, and with the increase in the consumption of raw products of animal origin (3). Due to its low cost and convenience, street food is consumed by millions of low- and middle-income consumers, especially in developing countries (4). The street-vended foods are prepared under unhygienic conditions and displayed openly, leading to a high degree of contamination (5). The traditional processing methods that are used in preparation, inappropriate holding temperature, and poor personal hygiene of food handlers are some of the main causes of contamination of street-vended foods (6). Ready-to-eat street foods are also subjected to cross-contamination from various sources, such as utensils, knives, raw foodstuffs, flies that sporadically land on the foods, vendors' bare hands serving, and occasional food handling by consumers (7, 8). The bharta manufacturers and the customers have limited knowledge of pathogenic microorganisms. Bacteria, viruses, fungi, protozoa, and helminths are responsible for the contamination. Most of the street foods are cooked and fried. Thus, the fresh street food contains very few contaminants. Food products may become contaminated at different stages along the food chain, could be during production, processing, distribution, preparation, and/or final consumption (9). Globally, contaminated food causes 600 million foodborne diseases and 420,000 deaths annually (10). Prevailing poor food handling and sanitation practices, inadequate food safety legislation, weak regulatory systems, lack of financial resources to invest in safer equipment, and lack of education for food handlers are the reasons for the common occurrence of food-borne diseases in developing countries (11-13, 14). Currently, these bacterial pathogens are a great concern for public health due to the emergence of multidrug-resistant strains (9). The widespread use of antibiotics and the ability of bacteria to rapidly develop and acquire antimicrobial resistance have facilitated the emergence of resistant strains such as methicillin-resistant *S. aureus* (MRSA) (15-17). MRSA strains or multidrug-resistant *S. aureus* cause nosocomial infections responsible for rapidly progressive, potentially fatal diseases, including life-threatening pneumonia, necrotizing fasciitis, endocarditis, osteomyelitis, severe sepsis, and toxinoses such as toxic shock syndrome (18). Third-generation cephalosporin-resistant and carbapenem-resistant Enterobacterales (eg, *Escherichia coli* and *Klebsiella* spp.), multidrug-resistant *Pseudomonas aeruginosa*, and carbapenem-resistant *Acinetobacter baumannii* are the most problematic and have been identified as priority pathogens (19). The arbitrary use of antibiotics in recent decades has led to the emergence of multidrug-resistant organisms. Natural selection amplifies the resistant organisms in the antimicrobial adverse condition. The misuse of antimicrobial agents in hospitals, vast communities, in feed for livestock, poultry, and fish raises multidrug-resistant organisms in the environment. In 2019, antimicrobial resistance was directly responsible for 1.27 million deaths and associated with an estimated 4.95 million deaths

globally (20). The control method or measures also include education of those who prepare the food at home and other food handlers, prohibiting individuals with abscesses or other skin lesions from handling food, and placing food in a cold place at 4°C or lower temperature, which prevents bacterial multiplication and toxin formation (9). During winter, the street food consumption increases among city dwellers in Bangladesh. As a consequence, multidrug-resistant pathogens spread through street food bharta to the general population.

This study aimed to isolate and identify the pathogenic bacteria from selected popular bharta based on colonial morphology, Gram staining, and biochemical tests, and assess their antibiotic susceptibility patterns.

2. Materials and Methods

2.1. Sample collection time

The study was conducted during the period of January to December 2023.

2.2. Sample collection site

Samples were collected from different areas of Dhaka city at Banani, Farmgate, Uttara, Shahbagh, Mirpur, Sadorghat, Pallabi, and Dhanmondi (Fig.1). Three types of bharta samples, which are mostly consumed, were collected using an aseptic technique (Table 1 and Fig. 2).

2.3. Sample preparation

A freshly collected sample was dissolved in sterile normal saline and processed using a ten-fold serial dilution technique. The diluted samples (10^{-1} to 10^{-5}) were spread on Nutrient agar plates. After 24 h of incubation, the isolated single colonies were streaked onto the MacConkey agar, SS agar, Manitol salt agar, Thiosulfate-Citrate-Bile salt-Sucrose agar, Eosin methylene blue agar, and Cetrimide agar media.

A total count of organisms was conducted on nutrient agar media. A higher Colony Forming Unit (CFU/mL) indicates significant microbial contamination, whereas a lower CFU/mL count suggests better hygiene and food safety. Total colony count is crucial for assessing food quality, identifying contamination risks, and supporting food safety regulations.

2.4. Classification by gram staining

The isolated colonies on the nutrient agar plate were smeared on a glass slide and mixed with water until an even and thin film was formed; then the slide was air-dried. Gram staining of the dried smear was done using several reagents and light microscopy.

2.5. Cultivation on selective media

Selective media are specialized culture media designed to promote the growth of specific bacteria while inhibiting others. They contain inhibitory agents such as antibiotics, dyes, or salts that restrict unwanted microbial growth. Common selective media used in food microbiology include MacConkey Agar (for Gram-negative bacteria like *E. coli*), Mannitol Salt Agar (for *Staphylococcus aureus*), *Salmonella-Shigella* Agar (for *Salmonella* and *Shigella*), and Thiosulfate Citrate Bile Sucrose (TCBS) Agar (for *Vibrio cholerae*). The selective media help to isolate and identify food-borne pathogens accurately, ensuring reliable microbiological analysis and food safety assessments. The selective media containing the inoculated culture were incubated for 24 h at 37°C.

2.6. Biochemical tests for the identification of organisms

Biochemical tests reveal specific enzymatic and metabolic characteristics for bacterial identification. It is a conventional method and an inexpensive means of identifying bacteria. Several biochemical tests were performed to detect presumptive organisms (Fig.3).

2.6.1. Catalase test

The catalase test detects the presence of the catalase enzyme, which breaks down hydrogen peroxide (H_2O_2) into water and oxygen. A positive result is confirmed by the immediate formation of bubbles, indicating catalase-positive bacteria like *Staphylococcus aureus* and *Bacillus* spp. A negative result, with no bubble formation, suggests the presence of catalase-negative bacteria, such as *Streptococcus* spp. This test is crucial for differentiating between *Staphylococcus* and *Streptococcus* species.

2.6.2. Oxidase test

The oxidase test is used to determine the presence of cytochrome c oxidase, an enzyme involved in the bacterial electron transport chain. A positive result is indicated by a purple color within 30 s, confirming oxidase-positive bacteria like *Pseudomonas* spp. and *Vibrio* spp. In contrast, oxidase-negative bacteria, such as *Escherichia coli* and *Salmonella* spp., show no color change. This test helps to differentiate Gram-negative bacteria, particularly *Pseudomonas* spp., from *Enterobacteriaceae*.

2.6.3. Methyl Red (MR) test

The MR test determines whether bacteria perform mixed acid fermentation of glucose, producing stable acidic byproducts. After adding Methyl Red reagent, a positive result is indicated by a red color, confirming strong acid production (*e.g., Escherichia coli* and *Salmonella* spp.). A negative result, shown by a yellow color, suggests weak acid production or neutral pH (*e.g., Enterobacter* spp.). This test is part of the IMViC (Indole, Methyl Red, Voges-Proskauer, Citrate) test series, used for differentiating enteric bacteria.

2.6.4. Voges-Proskauer (VP) test

The VP test detects the production of acetoin, a neutral end-product of glucose fermentation. After adding VP reagents (α-naphthol and potassium hydroxide), a positive result is indicated by a red color, confirming acetoin production (*e.g., Klebsiella pneumoniae, Enterobacter* spp.). A negative result, with no color change or a copper-brown appearance, suggests bacteria that do not produce acetoin (*e.g., Escherichia coli*). This test is paired with the MR test to classify enteric bacteria.

2.6.5. Simmons citrate test

The citrate test assesses whether bacteria can utilize citrate as their sole carbon source. Growth on Simmons Citrate Agar with a blue color indicates a positive result (*e.g., Klebsiella* spp., *Salmonella* spp.), showing that the bacteria can metabolize citrate. A green color indicates a negative result

(*e.g., Escherichia coli*), meaning the bacteria cannot use citrate. This test is useful for differentiating fecal coliforms from non-fecal coliforms.

2.6.6. Triple Sugar-Iron (TSI) test

The TSI test differentiates enteric bacteria based on their ability to ferment glucose, lactose, and sucrose, as well as their ability to produce gas (H_2S).

2.6.7. Motility Indole Urease (MIU) test

The MIU test is a combination test that evaluates motility, indole production, and urease activity.

Motility: Bacteria that spread away from the stabbed line indicate a positive motility result (*e.g., Proteus* spp.), while non-motile bacteria remain confined inside the stabbed line (*e.g., Klebsiella pneumoniae*).

Indole Production**:** After adding Kovac's reagent, a red ring at the top confirms a positive result (*e.g., Escherichia coli*), indicating the breakdown of tryptophan into indole.

Urease activity: A pink color indicates a positive result (*e.g., Proteus* spp., *Klebsiella* spp.), showing the production of the urease enzyme, which breaks down urea into ammonia.

2.7. Antibiotic assay

The antibiotic susceptibility test (AST) evaluates the effectiveness of various antibiotics against isolated bacterial strains. In this study, the Kirby-Bauer disk diffusion method was used to assess bacterial resistance and susceptibility (21). Bacterial sensitivity to each antibiotic was evaluated by measuring the diameter of the zone of inhibition, and the results were categorized into resistant and sensitive.

2.8. Statistical analysis

All the data were analyzed using Microsoft Excel at a 95% confidence level. The error bar is added in the Figure. The error was less than 5%.

3. Results

3.1. Isolation of organisms

The diluted samples (10^{-1} to 10^{-7}) were spread on nutrient agar plates. The best-isolated colony was found in a 10^{-5} dilution. The isolated colonies were considered for further investigation.

3.2. Organism identification

A total of 61 bacteria were isolated from 24 samples. Bacteria were identified based on their colonial morphology, staining characteristics (Gram-positive and Gram-negative), and biochemical tests. Based on staining traits, 63% were Gram-negative and 37% were Gram-positive organisms.

3.3. Biochemical tests

Traditional biochemical tests were performed to identify the bacterial species (Table 2). These tests are based on the production of enzymes or on visualizing a biochemical change with a substrate. These techniques are fast and efficient in differentiating bacteria. Out of 61 isolates, 5 types of bacteria were identified, such as *Staphylococcus aureus* (36%), *E. coli* (31.1%), *Salmonella* spp. (13.1%), *Klebsiella* spp. (13.1%), *Vibrio* spp. (6.5%) (Fig. 4).

Table 1. Location and the name of Sample with their Identification Number (ID)

Location (Dhaka City)	Name of sample	ID
	Coriander Leaf Bharta	1
Banani- Kacha Bazar	Mustard Seed Bharta	2
	Dried Fish Bharta	3
	Coriander Leaf Bharta	4
Farmgate -Tajgoan Collage	Mustard Seed Bharta	5
	Dried Fish Bharta	6
	Coriander Leaf Bharta	7
Uttara- Diyabari	Mustard Seed Bharta	8
	Dried Fish Bharta	9
	Coriander Leaf Bharta	10
Shahabag -TSC	Mustard Seed Bharta	11
	Dried Fish Bharta	12
	Coriander Leaf Bharta	13
Mirpur- Love Road	Mustard Seed Bharta	14
	Dried Fish Bharta	15
	Coriander Leaf Bharta	16
Sadorghat- Victoria Park	Mustard Seed Bharta	17
	Dried Fish Bharta	18
	Coriander Leaf Bharta	19
Pallabi- Duyaripara	Mustard Seed Bharta	20
	Dried Fish Bharta	21
	Coriander Leaf Bharta	22
Dhanmondi- Jigatola	Mustard Seed Bharta	23
	Dried Fish Bharta	24

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Table 2. Biochemical test results for the identification of bacteria.

	Bio-Chemical tests									
Isolates No.	**Gram Staining**	**Catalase Test**	**Oxidase Test**	**MR**	**VP**	**SimonCitrate**	**TSI**	**MIU**	**H_2S**	**Presumptive Organism**
1	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
2	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
3	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas -	Motile Urease - Gas - Indole +	−	*E. coli*
4	-ve	+	−	+	−	−	S - Red B - Yellow Gas +	Motile Urease - Gas - Indole -	+	*Salmonella* spp.
5	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
6	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
7	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
8	-ve	+	−	−	−	+	S - Yellow B - Yellow Gas -	Non-Motile Urease + Gas + Indole +	−	*Vibrio* spp.

9	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
10	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas + Indole +	−	*E. coli*
11	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
12	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
13	-ve	+	−	+	−	−	S - Red B - Yellow Gas +	Motile Urease - Gas - Indole -	+	*Salmonella* spp.
14	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
15	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease + Gas - Indole +	−	*E. coli*
16	-ve	+	−	−	+	+	S - Yellow B - Red Gas +	Non-Motile Urease + Gas - Indole -	−	*Klebsiella* spp.
17	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
18	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas -	Motile Urease - Gas + Indole +	−	*E. coli*
19	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*

20	-ve	+	−	+	−	−	S - Red B - Yellow Gas +	Motile Urease - Gas - Indole -	+	*Salmonella* spp.
21	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
22	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
23	-ve	+	−	−	−	+	S - Yellow B - Yellow Gas -	Non-Motile Urease + Gas + Indole +	−	*Vibrio* spp.
24	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
25	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas + Indole +	−	*E. coli*
26	-ve	+	−	−	+	+	S - Yellow B - Red Gas +	Non-Motile Urease + Gas - Indole -	−	*Klebsiella* spp.
27	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
28	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease + Gas - Indole +	−	*E. coli*

29	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
30	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
31	-ve	+	−	−	+	+	S - Yellow B - Red Gas +	Non-Motile Urease + Gas - Indole -	−	*Klebsiella* spp.
32	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
33	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas -	Motile Urease - Gas - Indole +	−	*E. coli*
34	-ve	+	−	+	−	−	S - Red B - Yellow Gas +	Motile Urease - Gas - Indole -	+	*Salmonella* spp.
35	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
36	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
37	-ve	+	−	−	+	+	S - Yellow B - Red Gas +	Non-Motile Urease + Gas - Indole -	−	*Klebsiella* spp.
38	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*

39	-ve	+	−	+	−	−	S - Red B - Yellow Gas +	Motile Urease - Gas - Indole -	+	*Salmonella* spp.
40	-ve	+	−	−	−	+	S - Yellow B - Yellow Gas -	Non-Motile Urease + Gas + Indole +	−	*Vibrio* spp.
41	-ve	+	−	+	+	+	S - Yellow B - Red Gas +	Non-Motile Urease + Gas - Indole -	−	*Klebsiella* spp.
42	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
43	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease + Gas - Indole +	−	*E. coli*
44	-ve	+	−	+	−	−	S - Red B - Yellow Gas +	Motile Urease - Gas - Indole -	+	*Salmonella* spp.
45	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
46	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
47	-ve	+	−	−	−	+	S - Yellow B - Yellow Gas -	Non-Motile Urease + Gas + Indole +	−	*Vibrio* spp.
48	+ve	+	−	+	−	+	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
49	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
50	-ve	+		−	+	+	S - Yellow B - Red Gas +	Non-Motile Urease + Gas - Indole -	−	*Klebsiella* spp.

51	**+ve**	**+**	**-**	**+**	**-**	**+**	S - Red B - Yellow Gas -	Non-Motile Urease - Gas - Indole -	-	*S. aureus*
52	-ve	+	−	+	−	−	S - Red B - Yellow Gas +	Motile Urease - Gas - Indole -	+	*Salmonella* spp.
53	+ve	+	−	+	−	+	S - Red B - Red Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
54	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
55	-ve	+	−	−	+	+	S - Yellow B - Red Gas -	Non-Motile Urease + Gas - Indole -	−	*Klebsiella* spp.
56	+ve	+	−	+	−	+	S - Yellow B - Red Gas -	Motile Urease - Gas - Indole -	−	*S. aureus*
57	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas -	Motile Urease - Gas - Indole +	−	*E. coli*
58	-ve	+	−	−	+	+	S - Yellow B - Red Gas +	Non-Motile Urease + Gas - Indole -	−	*Klebsiella* spp.
59	-ve	+	−	+	−	−	S - Yellow B - Yellow Gas +	Motile Urease - Gas - Indole +	−	*E. coli*
60	+ve	+	−	+	−	+	S - Red B - Red Gas -	Non-Motile Urease - Gas - Indole -	−	*S. aureus*
61	-ve	+	−	+	−	−	S - Red B - Yellow Gas +	Motile Urease - Gas - Indole -	+	*Salmonella* spp.

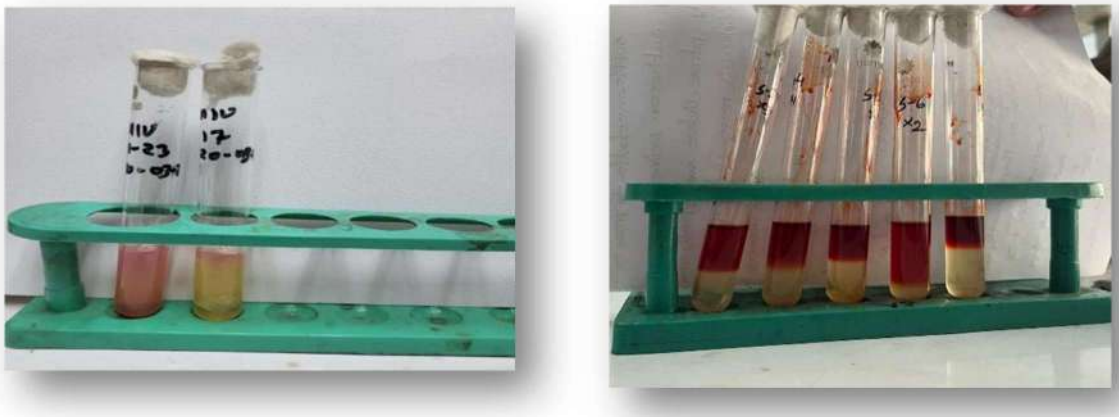
Table 3. Frequency of multidrug resistant organisms

Name of the Organism	3 Drugs Resistant	4 Drugs Resistant	5 Drugs Resistant	6 Drugs Resistant	7 Drugs Resistant	8 Drugs Resistant
S. aureus	3	3	2	0	0	1
E. coli	3	0	0	0	0	0
Salmonella spp.	1	0	0	0	0	0
Vibrio spp.	0	0	1	0	0	1
Klebsiella spp.	0	4	0	1	0	0

Figure 1. Sample collection site in Dhaka city, Bangladesh.

Figure 2. Different types of street Bharta in Dhaka city.

a b



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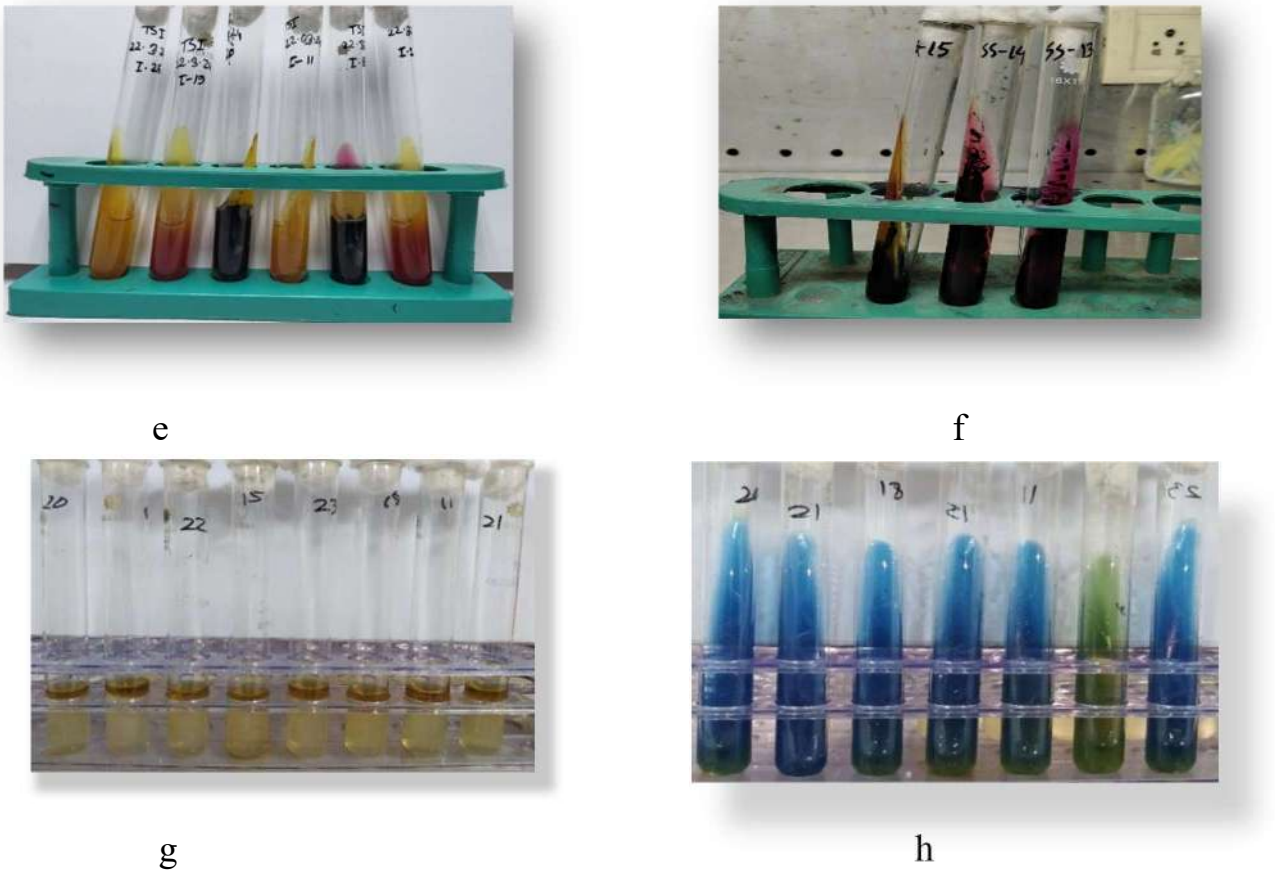


Figure 3. Several biochemical tests. (a) Catalase, (b) Oxidase, (c) Motility Indole Urea (MIU), (d) Methyl Red (MR), (e) Triple Sugar Iron agar (TSI), (f) H_2S gas, (g) Voges-Proskauer (VP), and (h) Simmons Citrate agar

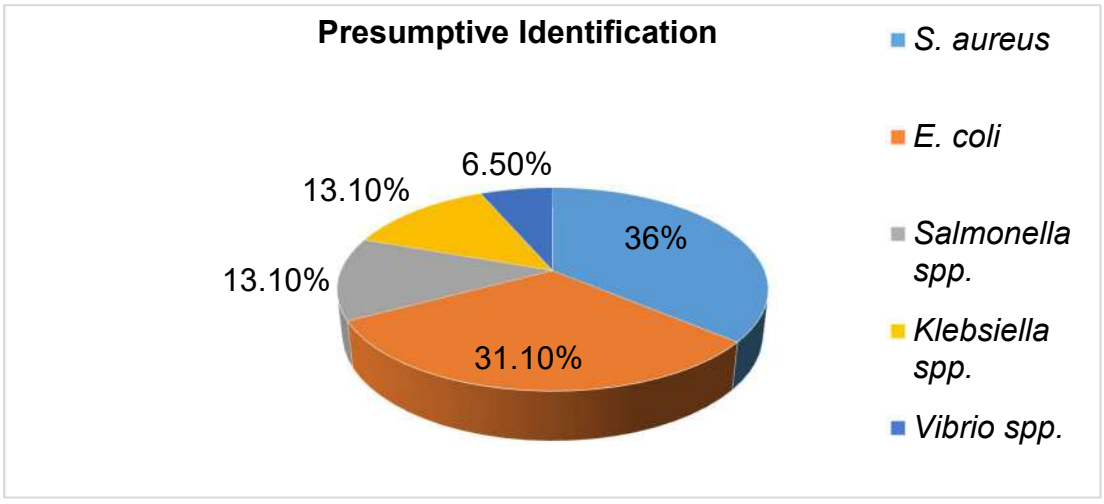
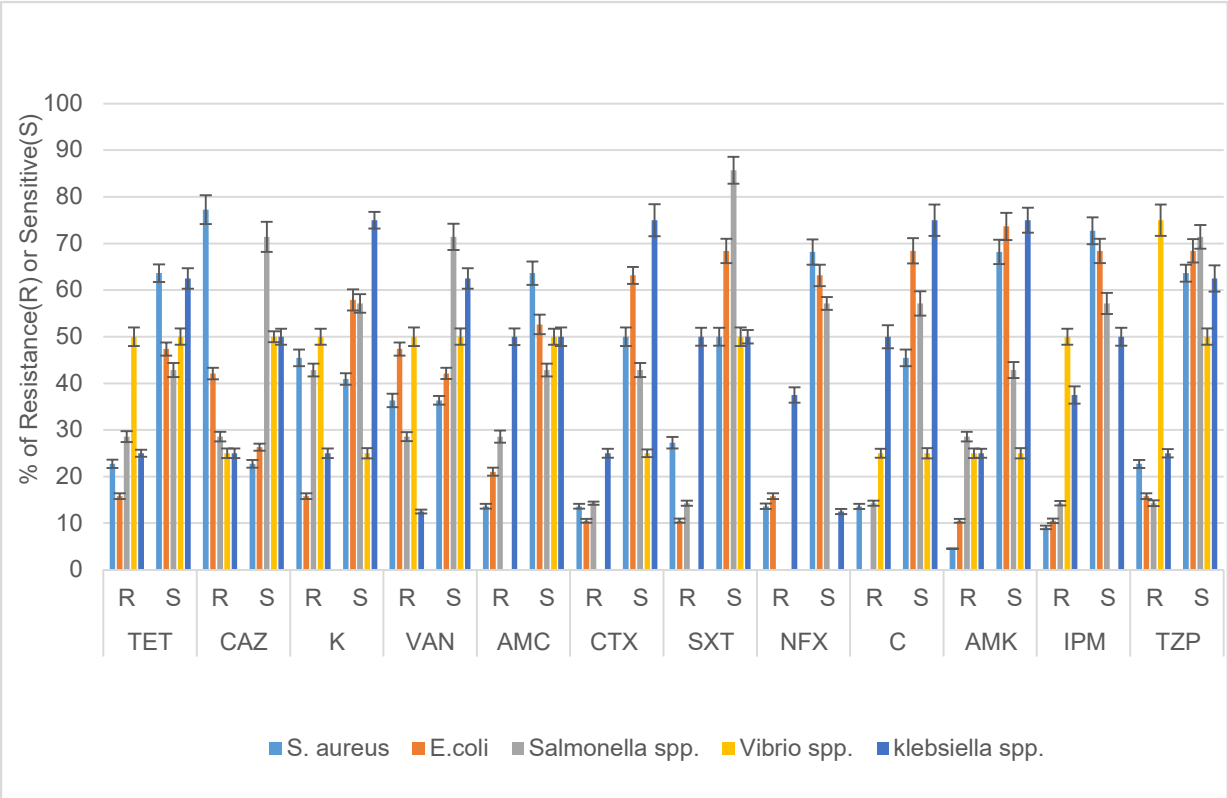


Figure 4. Presumptive identification of organisms based on their colonial characteristics, Gram staining, and biochemical tests.



Trimoxazole (SXT), Norfloxacin (NFX), Chloramphenicol (C), Amikacin (AMK), Imipenem (IPM), Tetracycline (TET), Ceftazidime (CAZ), Kanamycin (K), Vancomycin (VAN), Amoxycillin-clavulanic acid (AMC), Cefotaxime (CTX Piperacillin/Tazobactam (TZP)

Figure 5. Antibiotic susceptibility pattern of isolated organism

3.4. Antibiotic susceptibility tests

Results showed that *Staphylococcus aureus* exhibited the highest sensitivity to Imipenem (IPM;72%), Amikacin (AMK; 68%), Norfloxacin (NFX;68%), and the highest resistance to Ceftazidime (CAZ;77%) (Fig.5). *Escherichia coli* showed the highest sensitivity to Amikacin (AMK;73%) and resistance to Vancomycin (VAN;47%). *Salmonella* spp. showed the highest sensitivity to Co-Trimoxazole (SXT; 85%) and resistance to Kanamycin (K;42%). *Vibrio* spp. showed 50% sensitivity to Tetracycline (TET), Ceftazidime (CAZ), Vancomycin (VAN), Amoxycillin-clavulanic acid (AMC), Co-Trimoxazole (SXT), and the highest resistance To Piperacillin/Tazobactam (TZP;75%). *Klebsiella* spp. was 75% sensitive to Kanamycin (K), Cefotaxime (CTX), Chloramphenicol (C), Amikacin (AMK), and 50% resistant to Amoxycillin-clavulanic acid (AMC), and Co-Trimoxazole (SXT).

3.4.1. Multi-drug resistant (MDR) organisms

In literal terms, MDR means 'resistant to more than one antimicrobial agent', but no standardized definitions for MDR have been agreed upon yet by the medical community (22). Many definitions are being used in order to characterize patterns of multidrug resistance in Gram-positive and Gram-negative

organisms (23- 27). The multidrug resistance patterns of bacteria isolated from food samples varied across different species (Table 3). One strain of *Staphylococcus aureus* isolated from dried fish bharta showed 8 drug-resistant phenomena. The other two strains of *S. aureus* showed 5 drug-resistant phenomena. Three strains of *S. aureus* were shown 4 drugs resistant traits. One strain of *Vibrio* spp. isolated from Coriander leaf Bharta showed 8 drug-resistant characteristics, and the other one showed 5 drug-resistant traits. One strain of *Klebsiella* spp. was 6 drug-resistant, while the other 4 strains were 4 drug-resistant. *Salmonella* spp. was 3 drug-resistant in one case. Three cases of *E. coli* showed 3 drug resistance traits. Antibiotic susceptibility testing revealed that some isolates were highly resistant to commonly used antibiotics.

4. Discussion

This study revealed that the street food bharta contains harmful organisms and acts as a potential vector. Food safety knowledge is pivotal for preventing health-hazard pathogens. The illness exhibited mild to severe symptoms based on the immunity and the dose of infection. Higher doses of pathogens cause severe illness. Food service workers are unaware of the foodborne pathogens. There is no regulatory body to train them. Even consumers are also unconscious of foodborne illness. They do not know the cause of their illness. The physicians only treat the illness. They do not warn about the sources and occurrences of the diseases. In our study, biochemical tests and culture-based identification confirmed the presence of opportunistic and food-borne pathogens, some of which are known to cause diarrheal diseases, food poisoning, and severe infections. *Staphylococcus aureus* (36%), *E. coli* (31.1%), *Salmonella* spp. (13.1%), *Klebsiella* spp. (13.1%), and *Vibrio* spp. (6.5%) were identified from the bharta samples. Most of these contaminants are paralleled with other studies conducted in street foods (5). In our study, *Staphylococcus aureus* (36%) was prevalent may be due to poor food handling practices. The dominant pathogen during the monsoon season was *E. coli* (63%), which may be due to increased exposure to human sewage or contaminated water as fecal material gets mixed with water (28). Washing of utensils by contaminated water, poor personal hygiene, overcrowded, dusty, and poorly maintained shopping areas may cause transmission of pathogens. Some of them are multi-drug resistant. This type of drug-resistant organism is rising day by day. As a consequence, treatment of the disease will be difficult in the near future. There is no alternative to raising awareness the people about the risk of spreading multidrug-resistant foodborne pathogens.

5. Conclusion

Very rare studies have been carried out previously to detect the extent of microbiological contamination in different types of bharta items. The high bacterial load and pathogens observed in these isolates pose a severe public health risk. The study demonstrated the occurrence of multiple antibiotic-resistant bacterial isolates in bharta

in Dhaka city, Bangladesh. The presence of multiple antibiotic resistances among bacterial isolates in these food items underscores the significance of intensive surveillance and proper food handling practices. Traditional winter bharta has an increased risk of drug-resistant pathogen exposure to the large population. Thorough cooking and the use of food-grade antimicrobial preservatives can indeed help mitigate this risk. Implementing strict food safety measures, such as regular monitoring of food production processes, ensuring proper sanitation practices, and educating cooks and consumers about safe food handling practices, can all contribute to reducing the incidence of food-borne illnesses associated with bharta consumption. Continued research and surveillance efforts are crucial for understanding the extent of microbiological contamination in food items like bharta. In this study, five types of pathogenic organisms were isolated. Most of them are multidrug-resistant organisms. These organisms cause severe health issues among the large population silently.

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Author contribution

Hridoy Chandra Bhoumik:Writing - original draft; Formal analysis. Md. Iqbal Hossain: Investigation; Supervision; Visualization. Niloy Tarafder: Investigation. Antora Sutradhar: Investigation. Md. Abu Zihad: Investigation. Dayanidhi Sarkar: Writing - review & editing; Data curation; Conceptualization; Visualization; Investigation.

Conflicts of interest

The authors have no conflict of interest.

Data availability statement

Will be supplied on request

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Ethical consideration

Not needed ethical approval for this work

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