

## Microbiological quality of tea vendors at Dhaka, Bangladesh

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### ABSTRACT

Tea is a popular drink in Bangladesh and taken by a large number of people. Many people are engaged in small business for selling food and drinks on the roadsides as mobile and permanent vendors. This study was carried out to determine the microbiological quality of tea vendors in Saver, Dhaka. A total of 10 street tea vendors (5 permanent and 5 temporary) were randomly selected for this study. Tea cup, vendor hand and water samples were collected from each tea vendors. All samples were assessed for the presence of total bacterial count, total Enterobacteriaceae, total coliform and total *Staphylococcus aureus*. The presence of *Salmonella*, *Shigella* and *Vibrio* species were determined following appropriate enrichment and culture method. Cup and hand samples collected from mobile vendors showed significantly higher count of total heterotrophic bacteria ( $p < 0.05$ ). Cup swabs also showed significantly higher count of *Staphylococcus aureus* in mobile vendors than permanent ones ( $p < 0.05$ ). There was no significant differences between the counts of total Enterobacteriaceae, coliforms and *Staphylococcus aureus* in mobile and permanent vendors ( $p > 0.05$ ). *Salmonella* and *Shigella* could not be detected in any of the samples. Proportion of *Vibrio* spp. were 20% higher in cup and hand samples collected from mobile vendors as compared to the other group. All vendors did not have adequate knowledge and training on food safety and hygiene which is also reflected by poor microbiological quality of vendor samples. Contamination of tea vendor samples with pathogenic and indicator bacteria indicate their poor quality and unacceptability as drinks. This could be a threat to the consumer's health and require immediate attention in order to control any outbreak of food and water borne diseases.

Keywords: Bacteria, tea, vendors, Bangladesh.

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### INTRODUCTION

In developing countries, various kinds of juice and drinks sold by the street vendors are widely consumed by millions of people. Tea is one of the most popular drinks in Bangladesh. It is not only famous in urban area but also in villages. In Bangladesh, this drink is traditionally consumed by many people at the streets or from roadside shops. According to FAO, street food is a food obtained from a street side vendor, often from a makeshift or potable stall (FAO 2007). A number of studies showed heavy bacterial contamination

of water used by street vendors. A study carried out in Trinidad and Tobago revealed that 57.5% of water used by vendors was contaminated by coliforms (Agard et al., 2002; Mankee et al., 2003). A study in Columbia revealed that more than 30% of a group of food handlers were carriers of pathogenic organisms including *Salmonella typhi*, *Staphylococcus aureus*, and *Shigella* spp. (Buchanan, 1998).

During winter season (during the months of October through February) a large number of population of all income and age groups consumes

tea in order to relieve of cold (Ahmed et al. 2009). This drink is sold all over the year. Comparatively this drink is cheaper in suburban area than the urban areas. They simply prepared tea by adding water and sugar. They usually use untreated water to make tea. The drinks are nutritious for people and tea provide extra energy for some time when they take it. This is also a good medium for the growth of many microorganisms some of which may be pathogenic to people. The pathogenic organism can cause various food borne diseases. Tea is getting more popularity day by day. Tea is getting attention as a important vehicles in food borne disease statistics (Sivapalasingam et al. 2004). Consumption of tea that contains caffeine provides potential health benefits to the general population (Allothman et al. 2012). However, tea could be good for health only when it will free from pathogenic microorganisms.

Contamination of drink products can result in many health problems ranging from mild bloating and gas to serious incidents of food poisoning and dehydration. Unsafe and non-hygienic tea consumptions cause serious outbreak of food borne illness (Sivapalasingam et al. 2004). There have been some notable outbreaks of illness in recent years that demonstrate the increasingly important role of tea, fresh fruits and vegetables in food borne disease (Sandeep et al. 2001). There are several reports of illness due to the food borne diseases associated with the consumption of fruit juices at several places around the globe (Mosupye and Holy 2000; Muinde et al. 2005; Chumber et al. 2007). Usually raw materials equipment's, hand of the handlers, containers etc are responsible for contamination. Contamination from raw materials and equipment, additional processing conditions, improper handling, prevalence of unhygienic conditions contributes substantially to the entry of bacteria pathogens in tea (Oliveira et al. 2006; Nicolas et al. 2007). Water used for tea preparation can be major source of microbial contaminations such as total coliforms, fecal coliforms, fecal streptococci etc. (Tasnim et al. 2010).

Street vended tea and others dry food are found to sell in almost all areas of Dhaka city. Most of the time consumers are not conscious about the safety, quality and hygienic of the drink. Tea can be

potential risk factors for food borne diseases. In view of the threat posed by the bacterial pathogens in street vended drinks and flourishing demands of these drinks, the present study was undertaken to assess the bacteriological load and their safety for human consumption in terms of bacterial pathogens.

## MATERIALS AND METHODS

### Sample collection and preparation

Total 10 street tea vendors sample (tea cup, vendor hand, water) was collected from tea vendor of different areas in Dhaka city during May, 2014 to August 2014. This study was carried out in the laboratory, Department of Microbiology, Stamford University. Samples were collected aseptically early in the morning in a sterile plastic bag and transported to the laboratory for examination as soon as possible with a cooler box. During transporting of samples temperature was less than 10°C. We were collected samples from tea cup, vendor hand and waste water of each one vendor (total 10 samples).

### Media

Manitol Salt Agar, Nutrient Agar, Thiosulfate-Citrate-Bile-Sucrose Agar (TCBS) agar, *Salmonella Shigella* (SS) agar, Modified Faecal Coliform (m-FC) agar, Selenite Cystine, Broth (SCB), Alkaline Peptone Water (APW), Violet Red Bile Glucose (VRBG) agar, Buffered Peptone Water (BPW).

### Maintenance of stock culture

The isolated organisms were preserved for longer periods. For this purpose the organisms from pure culture were inoculated into the tubes containing Luria broth & 10% glycerol then kept in the refrigerator at -20°C.

### Antibiotic sensitivity assay

Bacterial susceptibility to antimicrobial agent was determined against *S. aureus* by in vitro agar disc-diffusion method which is known as the Kirby Bauer method (Barry et al, 1985). Commercially available antibiotic discs eg. Aztreonam (10µg),

Amoxicillin (25 $\mu$ g), Ciprofloxacin (5 $\mu$ g), Cefixime (15 $\mu$ g), Ceftazidime (10 $\mu$ g), Cefuroxime sodium (10units), Meropenem (10 $\mu$ g), Oxacillin, Sulfamethoxazole (125 $\mu$ g) and Tetracycline (30 $\mu$ g) were used in this study. A suspension of the test organism was prepared containing 10<sup>5</sup> to 10<sup>6</sup> cfu/ml by adjusting the turbidity of the culture broth with normal saline to match the equivalent turbidity standard of Mc Far land (0.5 standard). A sterile cotton swab was dipped into suspension and pushing and rotating the swab firmly against the inside of the tube above the fluid level removed excess fluid. The swab was used to spread the bacterial suspension evenly over the entire surface

of a Mueller-Hinton agar (P<sup>H</sup> 7.3) plate to obtain uniform inoculum. Antibiotic discs were placed aseptically on the surface of the inoculated plates at appropriate spatial arrangement by means of sterile forceps. The plates were then inverted and incubated at 37°C for 18 to 24 hours. After incubation the plates were examined & the diameters of the zones of complete inhibition were measured in mm. The zone diameters for individual anti-microbial agents were used to determine susceptible, intermediate and resistant categories.

## RESULTS AND DISCUSSION

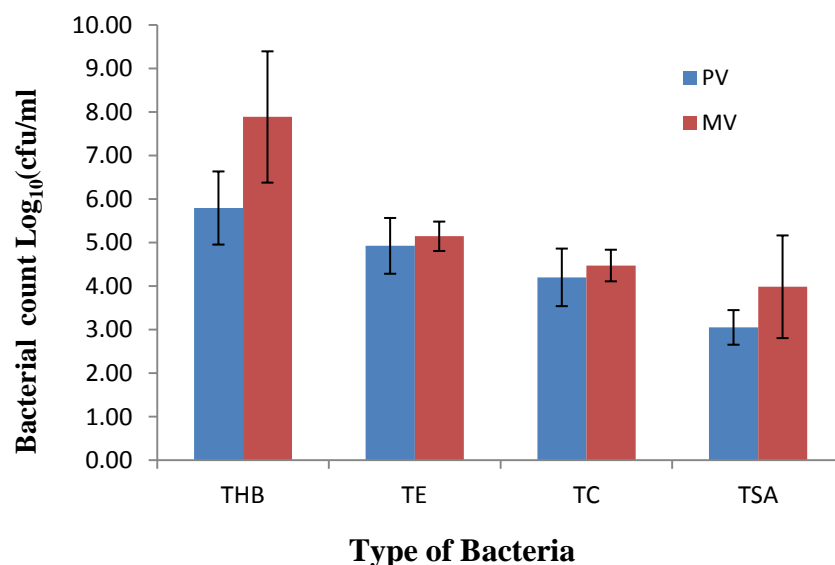


Figure A

Bacterial loads in cup samples collected from PV and MV. PV, Permanent Vendor; MV, Mobile Vendor; THB, Total heterotrophic bacteria log<sub>10</sub>cfu/ml; TEC, Total Enterobacteriaceae log<sub>10</sub>cfu/ml; TCC, Total coliform count log<sub>10</sub>cfu/ml; TSA, Total *S. Aureus*.

Figure A showed the bacterial loads of cup samples collected from both permanent and mobile vendors. Total heterotrophic bacterial (THB) and TSA counts were significantly high in mobile vendors than the permanent vendors

( $p < 0.05$ ). However, there was no significant difference in TE and TC counts of these two categories ( $p > 0.05$ ). All samples of both the categories were found to be contaminated.

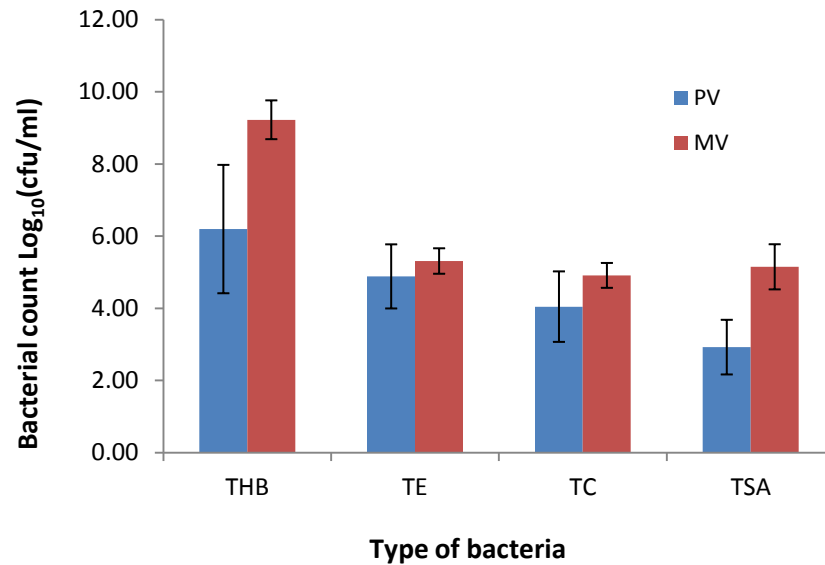


Figure B  
Bacterial loads in hand samples collected from PV and MV.

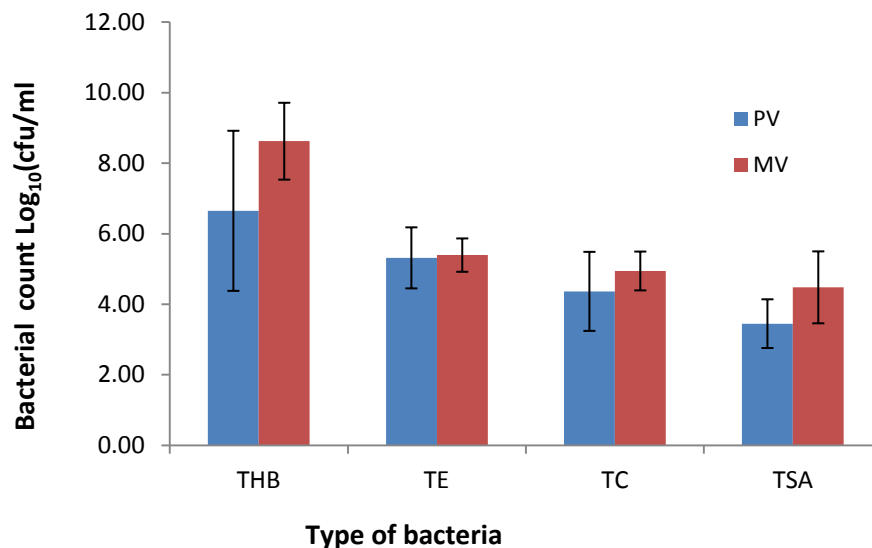


Figure C  
Bacterial loads in water samples collected from PV and MV.

Figure B demonstrated the bacterial loads of THB, TE, TC and TSA in hand samples of permanent and mobile vendors. Total heterotrophic bacterial count was significantly high in mobile vendors ( $p < 0.05$ ). However, there was no significant difference in TEC, TCC and TSA count of these two categories ( $p > 0.05$ ). All samples of both the categories were found to be contaminated.

Figure C illustrated the bacterial loads of THB, TEC, TCC and TSA in water samples of permanent and mobile vendors in Savar area. All samples of both the categories were found to be contaminated with all types of microorganisms. There was no significant difference in THB, TEC, TCC and TSA counts of these two categories ( $p > 0.05$ ). This indicates that both types of vendor's samples were contaminated.

Table 1  
Presence of pathogenic bacteria in cup samples collected from permanent and mobile vendor in Dhaka area.

	Sample	<i>Vibrio</i> spp.	<i>Salmonella</i> & <i>Shigella</i>
Permanent Vendor	S <sub>1</sub> C	A	A
	S <sub>2</sub> C	P	A
	S <sub>3</sub> C	A	A
	S <sub>4</sub> C	A	A
	S <sub>5</sub> C	P	A
	Percentage	2/5 (40 %)	0/5 (0%)
Mobile Vendor	S <sub>6</sub> C	P	A
	S <sub>7</sub> C	P	A
	S <sub>8</sub> C	A	A
	S <sub>9</sub> C	P	A
	S <sub>10</sub> C	A	A
	Percentage	3/5 (60 %)	0/5 (0%)

A, Absent; P, Present

Table 2  
Presence of pathogenic bacteria in hand samples collected from permanent and mobile vendor in Dhaka area.

	Sample	<i>Vibrio</i> spp.	<i>Salmonella</i> & <i>Shigella</i>
Permanent Vendor	S <sub>1</sub> H	A	A
	S <sub>2</sub> H	P	A
	S <sub>3</sub> H	A	A
	S <sub>4</sub> H	A	A
	S <sub>5</sub> H	P	A
	Percentage	2/5 (40%)	0/5 (0%)
Mobile Vendor	S <sub>6</sub> H	P	A
	S <sub>7</sub> H	P	A
	S <sub>8</sub> H	P	A
	S <sub>9</sub> H	A	A
	S <sub>10</sub> H	A	A
	Percentage	3/5 (60 %)	0/5 (0%)

Table 3  
Presence of pathogenic bacteria in water samples collected from permanent and mobile vendor in Dhaka.

	Sample	<i>Vibrio</i> spp.	<i>Salmonella</i> & <i>Shigella</i>
Permanent Vendor	S <sub>1</sub> W	A	A
	S <sub>2</sub> W	P	A
	S <sub>3</sub> W	P	A
	S <sub>4</sub> W	A	A
	S <sub>5</sub> W	P	A
	Percentage	60 %	0%
Mobile Vendor	S <sub>6</sub> W	P	A
	S <sub>7</sub> W	P	A
	S <sub>8</sub> W	A	A
	S <sub>9</sub> W	P	A
	S <sub>10</sub> W	A	A
	Percentage	60 %	0%

Table 4  
Characterization of isolated organism by biochemical test .

Colonies on Media	TSI										Suspected Organism
	Slant	Butt	Gas	H <sub>2</sub> S	Motility test	Citrate Utilization	Indole Production	MR Test	VP Test		
M-FC (Blue)			+	-	-	-	+	+	-		<i>E. coli</i>
	Acid	Acid									
TCBS (Yellow)			-	-	+	+	+	-	-		<i>Vibrio cholera</i>
	Acid	Acid									
TCBS ( Green)				-	+	-	+	-	+		<i>Vibrio parahaemolyticus</i>
	Acid	Acid									
MSA (Yellow with yellow halo)			-	-	-	-	-	+	-		<i>Staphylococcus aureus</i>
	Acid	Acid									

Table 5  
Antibiotic sensitivity patterns of *S. aureus* isolated from tea vendors in Savar area.

Sample ID	Oxacillin (1µg)	Tetracycline (30µg)	Cefuroxime Sodium (30µg)	Ciprofloxacin (5µg)	Amoxicillin (10µg)	Ceftazidime (30µg)
S <sub>1</sub> C	14 (S)	21 (S)	20 (S)	20(I)	17 (R)	15 (I)
S <sub>2</sub> W	11 (I)	18 (I)	14 (R)	21 (S)	21 (S)	17 (I)
S <sub>3</sub> W	14 (S)	17 (I)	19 (S)	8 (R)	9 (R)	16 (I)
S <sub>4</sub> C	14 (S)	17 (I)	18 (S)	7 (R)	7 (R)	17 (I)
S <sub>5</sub> H	11 (I)	19 (S)	15 (I)	18 (I)	25 (S)	12 (R)
S <sub>6</sub> H	23 (S)	34 (S)	30 (S)	30 (S)	43 (S)	26 (S)
S <sub>7</sub> C	13 (S)	26 (S)	18 (S)	23 (S)	20 (S)	14 (R)
S <sub>8</sub> W	19 (S)	15(I)	17 (I)	20 (S)	31 (S)	17 (I)
S <sub>9</sub> H	18 (S)	25 (S)	21 (S)	27 (S)	19 (R)	16 (I)
S <sub>10</sub> C	16 (S)	11 (I)	23 (S)	23 (S)	11 (R)	15 (I)
% of S	80	50	70	60	50	10
% of I	20	50	20	20	0	70
% of R	0	0	10	20	50	20

Table 1, 2 and 3 demonstrate the presence or absence of *Vibrio*, *Salmonella* and *Shigella* species in cup, hand and water samples of permanent and mobile vendors, respectively. Presence of *Vibrio*

spp. was 20% more in mobile vendors when they were isolated from cup and hand samples. However, both the category showed same proportion (60%) of contamination in case of

water samples. *Salmonella* and *Shigella* species could not be isolated from any of the samples tested. Table 4 bearing the result of biochemical test of isolated colonies. And, table 5 shows the detail of the Antimicrobial Sensitivity Test result of the isolated organism (*Staphylococcus aureus*).

Presence of total coliforms indicates the possible presence of other enteric pathogens such as, *Salmonella*, *Shigella*, *Vibrio*, *Campylobacter*, *Helicobacter* and parasites. However, in this study we could only determine the presence of *Vibrio*, *Samonella* and *Shigella* species due to the limitation of time. It was found that none of the samples showed the presence of *Shigella* and *Salmonella* after appropriate enrichment techniques. But, *Vibrio* spp. was found in most of the samples. This was possibly due to the collection of samples within the season of cholera.

The presence of pathogenic and indicator bacteria indicates the poor hygienic condition of the vendors. The sources of water used by the vendors were possible contaminated and it appears for the wash water count that the bacteria load was initially high. So, it is important for the vendors to use good quality water and edible disinfectants for preparation of tea and cleaning utensils, respectively. The bucket of the vendors was possibly not cleaned and may have built up the bacterial count. Although, presence of non-pathogenic bacteria may not be harmful to the customers, presence of pathogenic bacteria will be detrimental to the health of the pedestrian and customers taking food from these vendors.

All 30 samples among 10 vendors were highly contaminated with coliforms that exceed the WHO limits for fecal coliforms. Due to the shortage of potable water, many vendors tend to re-use the water, especially for cleaning utensils and used dishes. The TEC bacterial count was in a range of 3.50 to 5.77 Log<sub>10</sub> cfu/ml (PV cup sample), 4.71 to 5.60 Log<sub>10</sub> cfu/ml (MV cup sample); 4.30 to 5.64 Log<sub>10</sub> cfu/ml (PV hand sample), 4.68 to 5.60 Log<sub>10</sub> cfu/ml (MV hand sample); 4 to 6.24 Log<sub>10</sub> cfu/ml (PV water sample), 4.85 to 5.83 Log<sub>10</sub> cfu/ml (MV water sample). The presence of such high count of Enterobacteriaceae in vendor samples indicates the presence of other related

enteric pathogens in the served tea and related foods.

The total *S. aureus* count was in a range of 1.90 to 3.74 Log<sub>10</sub> cfu/ml (PV cup sample), 4.44 to 5.71 Log<sub>10</sub> cfu/ml (MV cup sample); 2.55 to 3.44 Log<sub>10</sub> cfu/ml (PV hand sample), 2.77 to 5.30 Log<sub>10</sub> cfu/ml (MV hand sample); 2.38 to 4.13 cfu/ml (PV water sample), 2.71 to 5.30 Log<sub>10</sub> cfu/ml (MV water sample). The serving utensils used at the vending site are often contaminated with *Staphylococcus sp.*, which may have been transmitted from the vendors hands when they touch the food preparation areas, dish washing cloth and the water during dish washing and hand washing which indicates cross contamination between dish water, food preparation surfaces, and the food itself.

According to the demographic data it was found that the street tea vendors were less educated, rough methods and work under crude unsanitary conditions were cause of heavy contaminations of pathogenic bacteria in street vended tea which might leads to water borne diseases in the people.

## CONCLUSION

These findings demonstrate that street vended Tea sold on Dhaka contain some pathogenic organisms which are likely to be a potential hazard to the health of the Community people in Dhaka. This study also reveals that lack of knowledge regarding the food contamination by the vendors. Lack of vendor's hygiene practice can also make the consumption of street food a high risk to health. Therefore, health education of the vendors on personal hygiene, safer food handling practice and the proper disposal of waste would improve food quality and thereby reduce the risk of contamination of street Tea vendor. Infrastructure developments for access to potable water, public toilet, washing and waste disposal facilities would reduce the health hazards.

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