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Bacteriological quality of street-vended beverages sold in two popular markets in Georgetown, Guyana

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ABSTRACT

In Guyana, street-vended beverages can often be bought in the local markets and some eating establishments; and are popular, sought-after beverages. The demand and consumption of local beverages are likely to increase for both locals and foreigners. The safe production of fruit beverages by the fruit juice vendor will become crucial to prevent spoilage and microbial contamination. This study analyses the bacteriological quality of street-vended juices from two of the main markets in Georgetown, by enumerating the total viable count and identifying coliforms, *Staphylococcus sp.*, *Salmonella sp.* and *Shigella sp.* This experimental study investigated street-vended juices. The standard plate count method was used to determine the total viable count on nutrient agar using serial dilutions. Selective media including MSA agar, EMB agar, and XLD agar; and other identification tests were used to select for and identify specific bacteria. The data were presented as mean±SD and numbers of positive samples. About 90% of samples were contaminated with bacteria with an average of 5.8±1.9 log cfu/mL or 2.1×10^7 cfu/mL. Of the 20 juices, 65% contained coliforms, 25% yielded *E. coli,* and 60% yielded *Klebsiella sp./Enterobacter sp.*, 65% yielded *Staphylococcus sp.*, 25% were suspected to be *Shigella sp.* and 5% were suspected to be *Salmonella sp.* Standards for ready-to-drink, non-carbonated beverages state that coliforms and other pathogens must be absent; therefore our findings are very worrying. We recommend that governmental and non-governmental agencies collaborate to bring more awareness about hygiene and storage techniques for street vendors.

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1. Introduction

Healthy eating and the increasing interest in natural and organic products have led to the exponential growth of fruit juice consumption (1). Fruit juices, especially street vended juices are a highly sought-after beverage in many developing and tropical countries (2). Street vended juices are nutrient-rich,

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affordable juices that are consumed by millions due to their fresh flavor (3). In 2016, it was reported that the global fruit and vegetable juices market was valued at USD 154 billion and it is expected that the juice market will continue to grow as the years progress (3).

In Guyana, street-vended juices can often be bought in the local markets or even in some eating establishments and are popular, sought-after beverages (4). However, with the presence of street-vended fruit juices becoming more frequent in the markets of Guyana, the microbial safety of these products must be taken into consideration, as failure to do so, can lead to a public health disaster. Furthermore, a comparison of visitor arrivals between the first half of 2021 and the corresponding period in 2022 has indicated that there is a 103% increase in visitors (5). This has been attributed to the burgeoning interest in the petroleum industry and the government's push to pursue the development of Guyana's potential as a leading territory for eco-tourism (6). Therefore, demand and consumption for local beverages by both locals and foreigners is likely to increase and the safe production of fruit juices by the fruit juice vendor or 'small man' will become even more critical. Street-vended juices are often unpasteurized or raw juices that lack any form of preservation measures. They are usually produced by extracting the "liquid or pulp of mature fruit, resulting in an unfermented, cloudy, untreated", ready-to-drink final product (7). Fresh, untreated juices are often more susceptible to spoilage and microbial contamination because of the absence of preservation processes such as pasteurization, proper hygienic practices for preparation and storage and the addition of additives (8). Although the appeal of these natural juices is linked to the absence of additives, colorants, artificial flavors, preservatives and heat, studies have shown that pathogenic microorganisms can contaminate the product because of the unhygienic conditions, under which they are processed (9).

In Guyana, fruit juices have become one of the most sought-after beverages due to their affordability and nutritional value (4). However, due to the lack of any implementation of preservation techniques such as pasteurizing, proper hygienic practices, and the addition of additives (treatment), fresh fruit juices are more susceptible than whole fruit to spoilage (8). Furthermore, untreated fruit juices are likely to undergo rapid microbial, chemical, enzymatic, and physical deterioration, with the major concern being microbial deterioration (8). According to Malik et al (10), fruit juices can be contaminated at any stage of the preparation process with several factors acting as sources of contamination, for example, the use of unhygienic water, flies, airborne dust, contaminated raw materials and equipment, improper handling, and unhygienic working environment. Because of the conditions under which street-vended fruit juices are made, stored and Pseudomonas sold, contamination can occur. As such, the consumption of these products can result in the likelihood of incidence of food-borne illnesses, which creates public health emergencies (11). Food-borne illnesses are diseases that mainly affect the gastrointestinal tract and are transmissible through the consumption of unhygienic, contaminated foods or drinks (3).Some of the pathogenic microorganisms that had been found in pasteurized and unpasteurized beverages included coliforms such as *E.coli*, and also *S. aureus*, *Pseudomonas, Salmonella, Aspergillus*, and *Rhizopus*, where higher microbial contamination was found in unpasteurized products (12). Contamination

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of juices with pathogenic bacteria like *E.coli* O157:H7 and *Salmonella sp.* has caused numerous illnesses and some fatalities. All reported cases of contamination by bacteria were due to fresh, unpasteurized juices (13). It is estimated that one-third of the population of developed countries is affected by microbiological food-borne diseases each year and that 1.9 million people globally in developing countries die (14). As recent as 2020, there was a Salmonella outbreak that was linked to the consumption of juices, smoothies, etc. from the Nektar Juice Bar in Woodbury, Minnesota (15).

Although the Public Health Department of the Mayor and City Council (M&CC) of Georgetown has a mandate to issue food handlers' certificates, it is unclear whether persons who manufacture and sell street-vended juice are also subject to these requirements. A review of the literature indicates that no study has been done in Guyana, concerning the potential of microbial infection from natural juices and with the increased demand by locals and tourists alike, a study on the investigation of microbial contamination of the street-vended juices would be very timely and relevant. The main purpose of this study was to analyze the bacteriological quality of street-vended juices from two of the main markets in Georgetown, Guyana by investigating the total viable bacterial count, total coliforms and identifying the following possible pathogens: *Escherichia coli*, *Staphylococcus sp*, *Salmonella sp.* and *Shigella sp.* This study also compared the total viable bacterial count, total coliforms and possible pathogens between the two markets.

2. Materials and Methods

2.1. Sample collection

This was an experimental study where 20 samples of street-vended juices were collected from vendors from two of the main markets in Georgetown. Ten (10) street-vended juices were purchased from the Stabroek Market and ten (10) from Bourda Market. It was difficult to obtain information regarding the population and location of all fruit juice street vendors in Georgetown, therefore the convenience sampling technique was utilized. A variety of juices were tested including cherry, passion fruit, tamarind, lemon and ginger, sugar cane, mauby and sorrel. The fruit juices were purchased from vendors however, no names were recorded. The type of container was noted, that is, whether it was in a bottle or a plastic bag. The samples were transferred to sterile closed containers and placed in a cooler with ice to maintain a temperature of 4^{o}C for one hour until arrival at the laboratory.

2.2. Enumeration of total viable count

The samples were investigated using the standard plate count method to ascertain the total viable count. The samples were mixed thoroughly. A 10-fold serial dilution was performed on each of the samples. An aliquot of 0.1 mL of appropriate dilutions was placed onto non-selective nutrient agar in duplicate using the spread plate method according to standard protocols and incubated at 37^{o}C for 18-24 h (9,16). Plates with colonies, otherwise known as colony-forming units (CFU) were counted and those with an average range from 25-250 cfu/mL per dilution were considered as important. Plates with more than 250 cfu/mL were considered too numerous to count (TNTC) and less than 25 cfu/mL were too few to be considered statistically significant. Colony-forming units were seen especially for the 10^{-5} and 10^{-7} dilutions. The pH of each juice was also recorded using paper pH strips (Fisherbrand™).

2.3. Presumptive identification of specific bacteria

The identification of specific bacteria was determined by inoculating various selective media in duplicate by adding an aliquot of 0.1 mL from the 10^{-1} dilution of each sample using the spread plate method. All inoculated plates were incubated at 37ºC for 18-24 h. Coliforms such as *Escherichia coli*, *Klebsiella sp.,* and *Enterobacter sp.* were identified using MacConkey (MAC) agar. Coliforms can ferment lactose and are observed as pink colonies on MAC agar. *Staphylococcus sp.* was identified using Mannitol Salt Agar (MSA) as yellow colonies. *Salmonella* and *Shigella sp.* were identified using *Salmonella* and *Shigella* (SS) agar. *Salmonella sp.* appears as colorless colonies with black centers, while *Shigella sp*. appears as colorless colonies on SS agar.

2.4. Confirmatory identification of specific bacteria

The identification of specific bacteria was confirmed by inoculating various selected media in duplicate using the streak plate technique according to Chees brough guidelines (16). Colonies grown on MAC agar, MSA and SS agar were streaked onto Eosin Methylene Blue (EMB) Agar, Baird Parker Agar (BPA) and Xylose Lysine Deoxycholate (XLD) agar respectively, for coliforms including *E. coli*, *Staphylococcus sp.*, *Salmonella sp.* and *Shigella sp*. All inoculated plates were incubated at 37ºC for 18-24 h. Coliforms appear as purple colonies and *E. coli* appears as colonies with a green metallic sheen on EMB agar. *Klebsiella sp.* and *Enterobacter sp*. are coliforms that appear as purple colonies on EMB. *Staphylococcus sp.* appears as black colonies surrounded by a light area on Baird Parker Agar (BPA), while *Salmonella sp.* appears as red-pink colonies with black centers, and *Shigella sp.* appears as red-pink colonies on XLD agar. Positively identified colonies were further confirmed by Gram stain. *Staphylococcus sp.* was subsequently identified using the catalase test. Because of the unavailability of plasma, the coagulase test was not done and so *Staphylococcus sp.* was identified up to the genus level. Therefore yellow colonies on MSA and black colonies surrounded by a light area on BPA, which appeared as Gram-positive cocci, and were catalase-positive, were recorded as *Staphylococcus sp*. *Klebsiella sp.* and *Enterobacter sp.* were not differentiated in our study due to the unavailability of media to conduct biochemical testing. Therefore, Gram-negative bacilli that appeared as pink colonies on MAC agar and purple colonies on EMB were reported as *Klebsiella sp. /Enterobacter sp.* It must be noted that biochemical and Enterotubes were unavailable to conduct further testing on the suspected colonies of *Salmonella sp.* and *Shigella sp*. Therefore Gram-negative bacilli that appeared as colorless colonies on SS agar and red-pink colonies on XLD agar were reported as suspected *Shigella sp*. Similarly, Gram-negative bacilli that appeared as colorless colonies with a black center on SS agar and red-pink colonies with black centers on XLD agar were reported as suspected *Salmonella sp*. No personal information was collected from the vendors and the research proposal was approved by the Ministry of Health (MoH) Institutional Review Board (IRB).

2.5. Statistical data analysis

Bacterial counts were tabulated in terms of mean± standard deviation (SD). A T-test was also conducted to compare the bacterial count between the two containers (bottle versus plastic bag). The T-test and ANOVA were conducted to determine if there was any statistical difference in the bacterial load of the street vended juices between the two markets. All statistical analyses were performed using SPSS version 22.0 (IBM Corp). A p-value of <0.05 was considered statistically significant.

3. Results

Higher microbial loads were seen with mauby and sugar cane juice (7.3±1.0 log cfu/mL and 7.6±0.4 log cfu/mL respectively), whilst the juices that had the least microbial load were sorrel and tamarind juices (Table 1). Based on the data, 90% of samples were contaminated with bacteria with an average of 5.8±1.9 log cfu/mL. Our calculations indicate that this represents 2.1×10^7 cfu/mL (Table 1).

The bacterial load of juices collected from the Stabroek market had an average of 44.4 cfu/mL, whilst that of the Bourda market had an average of 5.1 cfu/mL. A T-test and ANOVA analysis of the average total viable count between the two markets revealed a p-value of 0.03; therefore the difference was statistically significant.

Table 2 also revealed that several samples contained bacterial species identified as total coliforms such as *E.coli* and *Klebsiella sp.* /*Enterobacter sp.*, *Staphylococcus sp.*, and suspected *Shigella sp.* and *Salmonella sp.* Of the 20 juices, 65% were found to contain coliforms and 25% of those 65% samples yielded *E. coli* which is a fecal coliform. *Klebsiella sp. /Enterobacter sp.* was found in 60%, while *Staphylococcus sp.* was identified in 65% of the samples. Colonies suspected to be *Shigella sp.* and *Salmonella sp.* were found in 25% and 5% of the samples tested respectively.

The highest microbial load (7.1±0 cfu/mL) was noted for the street-vended beverage brought in the cup (Table 3). However, because we only had one sample in the cup, this type of container was not included in the T-test analysis. Rather the T-test analysis was done for beverages brought in plastic bags and plastic bottles. A p-value >0.05 was found. A comparison of the types of containers did not show any significant differences in terms of the total viable count. The mean microbial load for street-vended beverages bought in plastic bottles and plastic bags was 6.8±0.8 cfu/mL and 5.7±2.9 cfu/mL respectively (Table 3). This result suggests that the type of container used for selling street-vended juices does not matter when it comes to microbial contamination.

Table 4 revealed the specific bacteria identified for the juices sold in different types of containers. No major difference was noted between the two main types of containers in which the fruit juices were sold and the number of samples that tested positive for specific bacteria except for *Shigella sp*. Beverages sold in plastic bags were more likely to contain *Shigella sp.* when compared to plastic bottles. The pH for each juice was also recorded and was found to be an average of 5.2±0.2. A pH of 4.0 was recorded for one of the sorrel juices and one of the lemon and ginger juices, whilst the other juices had a pH of 5-6. A statistically significant association (p-value 0.005) between pH and the level of bacterial contamination was found.

Table 1. Average estimation of Total viable count

Types of Beverages	No. of samples (N)	Total Viable Count (TVC)		
		No. of Positive samples (%)	Average (log cfu/mL) ± SD	Average (cfu/mL)
Cherry	5	5 (100)	6.8 ± 0.8	1.5×10^7
Mauby	3	3 (100)	7.3 ± 1.0	7.0×10^7
Sorrel	2	1 (50)	3.1 ± 4.4	7.5×10^5
Sugar Cane	4	4 (100)	7.6± 0.4	4.6×10^7
Passion Fruit	2	2 (100)	5.8± 0.2	7.5×10^5
Lemon & Ginger	2	2 (100)	7± 0.1	1.1×10^7
Tamarind	2	1 (50)	3.1± 4.4	7.5×10^5
Total (N)/ Averages	**20**	***18 (90)**	****5.8 ± 1.9**	****2.1 x 10⁷**

*Total number (%) of samples ** Average/Mean value

Table 2. No. of samples contaminated with specific bacteria

Types of Beverages	No. of Samples (N)	No of positive samples (%)					
		Total Coliforms	*E. coli*	*Kleb sp. / Enter sp.*	*Staph sp.*	*Salmonella sp.*	*Shigella sp.*
Cherry	5	4 (80)	2 (40)	4 (80)	3 (60)	0	0
Mauby	3	3 (100)	1 (33.3)	3 (100)	2 (66.6)	0	2 (66.6)
Sorrel	2	1(50)	0	1 (50)	1 (50)	0	1 (50)
Sugar Cane	4	4 (100)	1 (25)	4 (100)	3 (75)	2 (50)	1 (25)
Passion Fruit	2	1(50)	1 (50)	0	2 (100)	0	1(50)
Lemon & Ginger	2	0	0	0	2 (100)	0	0
Tamarind	2	0	0	0	0	0	0
Total (N)/ Averages	**20**	***13 (65)**	***5 (25)**	***12 (60)**	***13 (65)**	***2 (5)**	***5 (25)**

*Total number (%) of samples

Table 3. Average estimation of TVC based on type of container

Types of Containers	No. of samples (N)	Total Viable Count		
		No. of Positive samples (%)	Average (log cfu/mL) ± SD	Average (cfu/mL)
Plastic Bag	11	9 (81.8)	5.7 ± 2.9	3.3×10^7
Plastic Bottle	8	8 (100)	6.8 ± 0.8	1.5×10^7
Paper Cup	1	1 (100%)	7.1 ± 0	1.3×10^7
Total (N)/ Average	**20**	***18 (90)**	****6.5 ± 0.7**	****2.0 x 10⁷**

*Total number (%) of samples ** Average/Mean value

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Table 4. No. of samples contaminated with specific bacteria by container type

Types of Containers	No. of samples (N)	No of Positive samples (%)					
		Total Coliform	*E. coli*	*Klebsiella sp./ Enterobacter sp.*	*Staph sp.*	*Salmonella sp.*	*Shigella sp.*
Plastic Bag	11	6 (54.5)	2 (18)	5 (45)	7 (63.6)	1 (9)	4 (36)
Plastic Bottle	8	6 (75)	3 (37.5)	6 (75)	5 (62.5)	1 (9)	1(12.5)
Paper Cup	1	1 (100)	0	1 (100)	1 (100)	0	0
Total (N)/ Average	**20**	***13 (65)**	***5 (25)**	***12 (60)**	***13 (65)**	***2 (5)**	*** 5 (25)**

*Total number (%) of samples

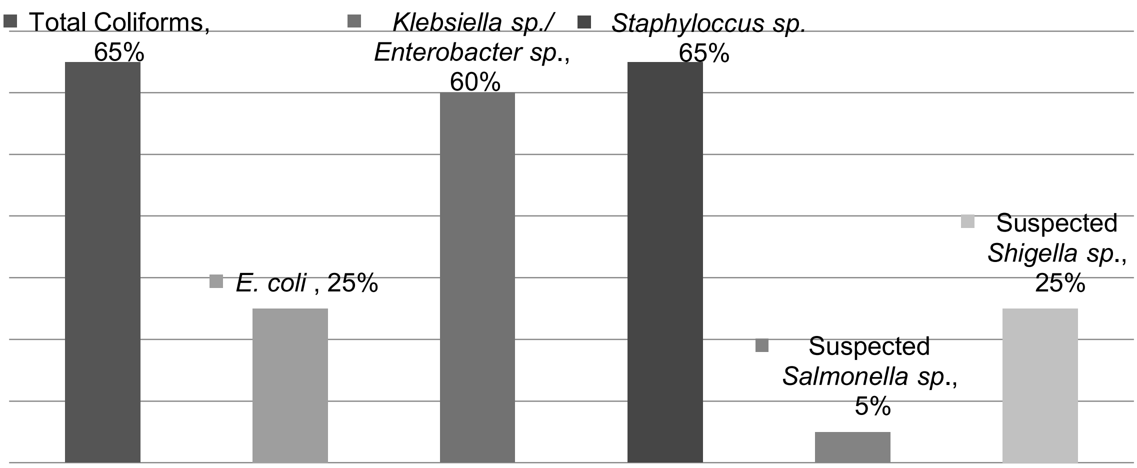

Figure 1. demonstrated that total coliforms and *Staphylococcus sp.* were the most common bacteria identified, followed by *Klebsiella sp.* / *Enterobacter sp.* Suspected *Salmonella sp.* was the rarest bacteria identified.

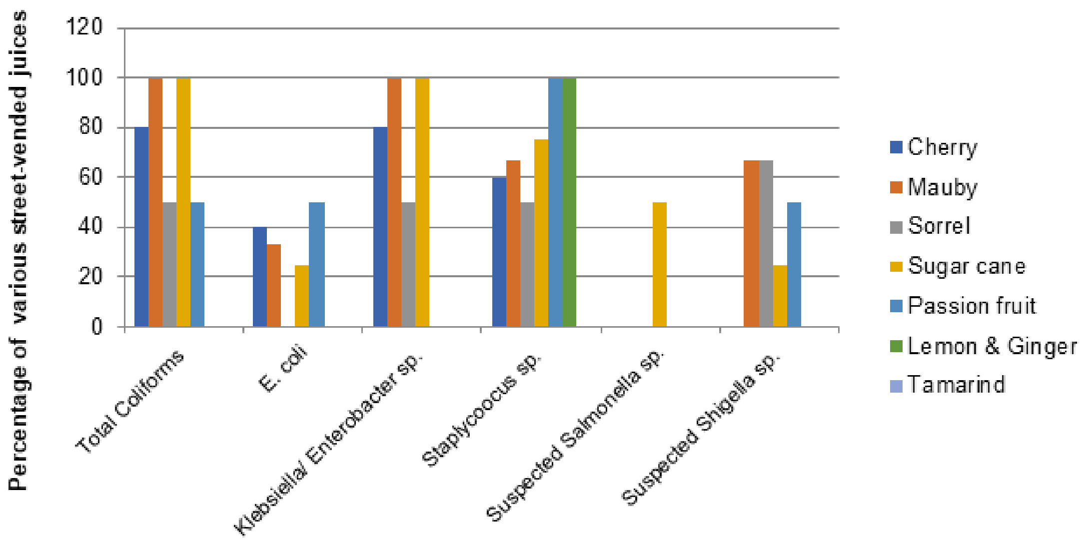

Figure 2. Prevalence of specific bacteria in the various types of beverages

4. Discussion

Natural juices offer an affordable and healthy option to obtain nutrients and are sought after by locals as well as tourists. The majority of street-vended beverages tested were found to be contaminated with bacteria (90%). Similar results were found in a study done in India (17), where most of the samples were contaminated with bacteria (91%). Another study done in India (18) showed that the majority of the samples tested were contaminated with bacteria (96%).

The fruit juices we sampled seemed to have a higher microbial load as compared to a study done in Kuala Lumpur, where the highest average count was 5.30±1.11 cfu/mL (19). However, the results from our study showed a lower microbial load than that of a study done in Mumbai India, where the average Total Viable Count (TVC) was 6.5 cfu/100 mL (7); a study done in Lahore, India, where the mean TVC was 6.80±1.91 log10 cfu/mL (20) and a study done in Bangladesh, where the average viable count ranged between 7.7×10^3 to 9×10^8 cfu/mL (21). In each of these studies, the authors found that the bacterial load exceeded the allowable limit for their respective territories.

The sugar cane juice had the highest bacterial load (7.6± 0.4 cfu/mL). Similarly, the study done in Mumbai, India (7) showed that sugar cane juices had the highest microbial load (7.19±1.4 cfu/100 mL). A recent study was conducted by Srisangavi et al (22) in Tamil Nadu, India using sugar cane juice, and a very high microbial load (525-1520 cfu/mL) was also found. Sugar cane juice is made by picking and washing the sugarcane and then passing it through a mill to extract the juice, which is then strained and bottled with no additives. In addition, no boiling is done during the processing of the sugar cane juice. Mwambeteand Mpenda (23) stated that the improper handling and processing of the sugar cane, the use of contaminated water for juice dilution and the use of unsanitary processing equipment such as the mill, may result in bacterial contamination.

This current study showed that samples with a pH of greater than 5 (pH 6) had significantly higher bacterial loads than samples with a pH of less than 5. Low pH can serve as a hindrance to bacterial growth. According to Sharma (17), high pH coupled with high temperature (>28°C) can promote the growth of bacteria. In this current study, the sorrel juice had the lowest pH and the lowest bacterial load. Therefore, the authors postulate that the low pH of the sorrel juice is one contributory factor for the low comparable bacteria load. A study done in Nagpur city in India (24) showed a very low microbial load (1.0×10^4-4.0×10^6 cfu/mL) in the fruit and vegetable juices test. The pH of those beverages was acidic (pH <3). In addition, studies have also shown that sorrel (*Hibiscus sabdariffa*) has antimicrobial properties (25). Furthermore, sorrel juices are prepared by soaking the sorrel in hot water. Tamarind juice was also the least contaminated fruit juice beverage. The preparation of tamarind juice requires the boiling of the fruit and the use of hot water for dilution. This boiling is similar to pasteurization and can kill microorganisms that might be present (7).

The results from this study showed that there is no significant association between the type of containers in which the fruit juices were sold and the level of bacterial contamination. This study indicated that the type of container in which street vended juices are sold, does not influence the total viable count of bacteria or the presence of bacteria such as total coliforms, *E. coli*, *Klebsiella sp.* / *Enterobacter sp.*, and *Staphylococcus sp.* and

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Salmonella sp. However, the presence of *Shigella sp.* may be linked to the type of container used. The authors did not find any study that investigates this aspect of street vended juices.

The location of street juice vending was found to be statistically significant, as the bacterial load of the street-vended fruit juices sold at the Stabroek market differed significantly when compared to those sold at the Bourda market. This difference in bacterial contamination could be attributed to the fact that the Stabroek market, when compared to the Bourda market, is larger and is known to have a greater amount of traffic than the Bourda market. In previous studies, it was discovered that areas with more traffic and congestion had a higher level of contaminated samples (10,17). It was further indicated that markets, bus parks, etc. are often unhygienic places for food handling due to overcrowding, poor sanitary conditions, dust, limited access to potable water, exposure to extreme heat without proper refrigeration for products, poorly kept storage containers, etc. Street-vended fruit juices sold in areas where these conditions are present can negatively impact the bacterial status of street-vended fruit juices (10,17).

A considerable number of street-vended juices were contaminated with total coliforms. Lower results were seen in a study done in Bangladesh, where 47% of the samples were contaminated with total coliforms (21) and higher results were noted in a study done in Kuala Lumpur, Malaysia, where about 71% of the samples showed the presence of total coliforms (19). According to the Codex standards (26), coliforms are considered indicators of quality. Their presence indicates fecal contamination. The fact that more than half of the samples we tested had total coliforms suggests that the street-vended juices in Guyana may not be safe. Total coliforms in street-vended juices may be due to fecal contamination that occurs during and immediately after production (27). The presence of coliforms may be due to the use of water that is not boiled, in the production of beverages or direct contamination from street vendors and producers (28). Coliforms, when present may reproduce rapidly when left at ambient temperature for a long time (29), thereby increasing the bacterial loads of the sample. Our study showed that mauby and sugar cane juice had the highest frequency of total coliforms.

E. coli is considered a more specific indicator to detect fecal contamination than fecal coliforms (30). This current study showed that a small number of samples tested had *E.coli* (25%). A lower result was revealed in a study done in Kuala Lumpur, where only 3% of the samples were contaminated with this bacterium (19). Higher results were noted in a study done by Lucky et al., where 50% of the beverages they tested were contaminated with *E. coli* (9). Studies conducted in India by Lakshmi et al. (18) and Sharma (17) also showed higher results, where 42% and 33% of the juice tested was contaminated with *E. coli* respectively. Even though, this current study had a lesser occurrence of *E. coli*, its presence within the samples is still of significance. Evidence of this bacterium in street vended juice is indicative of fecal contamination and poor sanitary practices by the producers and vendors (10). Certain strains of *E. coli* when consumed can lead to food-borne disease with symptoms such as vomiting, diarrhoea, and even death (31). *E.coli* positive samples indicate poor hygienic quality of the juices and suggest a high risk for transmission of diarrheal illnesses.

Of the twenty (20) samples, more than half of them had *Klebsiella sp./ Enterobacter sp.* (60%). *Klebsiella sp./ Enterobacter sp.* are often considered "opportunistic bacteria" and their presence is proof of the poor bacterial quality of the street vended fruit juices (32). A similar result to our study was found by Malik et al., where *Klebsiella sp.* and *Enterobacter sp.* were identified and *Klebsiella sp.* was found in 59% of the samples (10). A lower occurrence of *Klebsiella sp.* and *Enterobacter sp.* was noted in a study done by Tambekar et al., where *Klebsiella sp*. was found in 3% of the samples and *Enterobacter sp*. in 1% (2). Uddin et al. observed the presence of *Klebsiella sp.* in their study and indicated that its occurrence could be linked to many factors such as poor storage, insufficient refrigeration of the juices, the use of damaged stands or carts as well as unhygienic vending area (33). Mauby and sugar cane had the highest frequency of *Klebsiella sp./ Enterobacter sp.* in this current study.

Staphylococcus sp. was identified in more than half of the juices tested (65%). The samples we studied had a higher occurrence of *Staphylococcus sp.* than a study done in Ethiopia, where 12% of the samples were positive for *S. aureus* (34) and a lower occurrence than studies done in India, where 77% of the samples (18) and 100% of the samples were contaminated with *S. aureus* (9). In another study done in Dhaka, Bangladesh, all of the samples studied were found to be contaminated with *S. aureus* (21). *S. aureus* can be found in the nose, throat, hair and even the skin of humans. The presence of *Staphylocccus sp.* in fruit juices is primarily attributed to contamination via street vendors and producers. Food handlers, who fail to practice proper hygienic practices, e.g. wearing gloves when coming into direct contact with food, proper hand washing, etc., can contaminate the juice during processing, packaging and storage (9,33). Some *Staphylococcus* species may produce potent toxins that are associated with food-borne illnesses. *S.aureus* positive samples indicate poor beverage quality and the risk of staphylococcal food poisoning to consumers.

Salmonella sp. was suspected in two samples of the sugar cane juice and 5% of the juices overall. A higher occurrence of *Salmonella sp.* was found in a study done by Sharma, where *Salmonella sp.* was found in 19% of samples (17) and a study done by Tambekar et al., where *Salmonella sp.* was found in 16% of samples (2).*Salmonella sp.* can contaminate fruit juice production at any stage during the food supply chain and processing (35-37). The suspected occurrence of *Salmonella sp.* in the sugar cane juice is suggestive of poor food handling techniques, unsanitary utensils and processing equipment used by the vendor and producer thus resulting in contamination (35,36,38). *Salmonella sp.* is usually linked to animal products such as chicken, eggs, etc. (39). As such, finding its presence in juices specifically sugar cane juice is a matter of concern and indicates the need for more surveillance and implementation of better food handling techniques by both vendors and producers.

Shigella sp. was suspected to be in five of the twenty samples and 25% of the juices overall in this current study. *Shigella sp.* is usually found in salads, raw vegetables, dairy, etc. (36). The presence of *Shigella sp.* in fruit juices is evidence of the use of contaminated water, poor food handling, poor cold storage methods, and/or poor hygienic practices such as bare hand holding of raw fruits, etc. (36). The mauby juices were found to have the highest occurrence of suspected *Shigella sp.*

Of the seven types of street-vended fruit juices collected for this study, the sugar cane juices had the highest overall frequency of contamination with the bacteria investigated. Additionally, sugar cane juice was the only type of beverage suspected to be contaminated with *Salmonella sp*, where 50% of the samples were suspected to be contaminated. The high frequency of the investigated bacteria in the sugar cane juice samples can be a result of multiple factors such as improper handling and processing of the raw material, the use of contaminated water for juice dilution, and the use of unsanitary processing equipment (23).

It was difficult to obtain the GYS 494-2010 Standard which is the Guyana National Bureau of Standards specifications for fruit and vegetable juices, drinks and fruit nectars. This Standard is based on the CARICOM Regional Standard (CRS 27:2010), however, this document did not provide any details about the microbiological criteria for fruit juices. However, the Draft Standard for ready, non-carbonated non-alcoholic beverages for Tanzania (TZS 585:2011), indicated that coliforms must be absent from all beverages and a maximum of 1×10^2 cfu/mL for chilled beverages such as natural juices (40).

Street-vended fruit juices are often unpasteurized and manufactured under conditions that are devoid of proper hygiene, proper food handling practices, good water and quality, making them susceptible to microbial contamination (41). As such, while consumption of these juices is often seen as more nutritional and safer than juices with preservatives by consumers, this is not necessarily always the case (10, 41). The causes of bacterial contamination of the street-vended fruit juice samples in this research can be linked to many factors, with the practices of the producers and vendors of these juices being at the top of the list. As such, while this study provided some insight into the levels of bacterial contamination and the presence of total coliforms, *E. coli*, *Klebsiella sp.*/ *Enterobacter sp.*, *Staphylococcus sp.*, *Salmonella sp.* and *Shigella sp.*, more research should be done to understand the knowledge and practices of the vendors/ producers of street vended fruit juices sold the markets in Georgetown, to identify what other factors contributed to the contamination of these juices and what changes need to be made.

Our study was limited because we only looked at two markets in Georgetown and we were unable to confirm the identification of certain bacteria up to species level. However, we believe that our findings have direct implications for the public health environment as these are the two biggest markets in the country. In addition, we found the presence of fecal coliforms in more than half of samples and they should not be present at all. The strict guidelines required to avoid microbiological contamination and avert potential diarrhoeal outbreaks should not be compromised by the popularity of these beverages.

5. Conclusion

The findings in the present study are alarming since coliforms were found in 65% of samples, *E.coli* was identified in 25% of juices, *Klebsiella sp.*/ *Enterobacter sp.* was present in 60%, *Staphylococcus sp.* was present in 65%, suspected *Salmonella sp.* was found in 25% and suspected *Shigella sp.* was found in 5%. Therefore, interventions are needed to prevent the possibility of outbreaks. We recommend that government agencies such as the Public Health Department of M&CC, the Ministries of Health; and Tourism, Industry and Commerce, the GNBS and non-governmental agencies such as PAHO, collaborate to develop a robust program for education, training and monitoring. It is hoped that strategies to educate vendors about good hygienic practices and storage techniques; and to provide the relevant information from the GYS 494-2010 document, in an appropriate manner, will be implemented as soon as possible. We propose that there be continued education and training for those persons responsible for surveillance mechanisms for food-borne and water-borne diseases. One of the resources that provides guidelines about the prevention of contamination of fruit juices is provided by Codex Alimentarius (which is a collection of recommendations, codes of practice, and standards provided by a joint FAO/WHO alliance) and another is the Hazard Analysis Critical Control Point (HACCP) which provides comprehensive information about keeping foods (and beverages) safe for human consumption. Further studies should be done on a larger sample of juices and also the water sources used to make them, to enumerate the viable bacteria, total and fecal coliforms and to check for the presence of pathogenic bacteria such as *Pseudomonas sp*. The accessibility, low cost and nutritional benefits of street-vended juices cannot be overstated. However, these beverages must be free from coliforms and have a low microbial load. We found the opposite in our study, and it is hoped that there is an urgent Call to Action to improve the food safety mechanisms so that the local Guyanese population and the numerous visitors can have confidence in street-vended juices.

Conflict of interest

No financial interest nor any conflict of interest exists.

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