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Assessment of microbial quality of street foods sold in Nugegoda, Sri Lanka.

G. G. N. T. Dharmarathne*, P. De A. Abeysundara

(Department of Food Science and Technology, Faculty of Applied Sciences, University of Sri Jayewardenepura, Gangodawila, Nugegoda, 10250, Sri Lanka.)

ganganidharmarathne@gmail.com

ABSTRACT: The purpose of this research was to assess the microbial quality of some selected street foods sold in Nugegoda, Sri Lanka. Street food samples (Fruit salad, Fruit juice, Wade, String hopper, and Dosai) were collected and tested for Total Plate Count (TPC), Total Yeast and Mold Count (TYMC), Total Coliform Count (TCC), Presence of Escherichia coli and Staphylococcus aureus using standard microbial testing methods. TPC and TYMC values of the tested street food samples were between 3.4 – 6.3 log CFU/ml and 3.8 – 6.4 log CFU/ml respectively. The highest TPC and TYMC values were observed in Fruit salad and Dosai samples respectively. Five (33.3%) food samples exceeded the permissible limit of TCC for RTS foods (10MPN/g). Eight (53.3%), samples were positive for E. coli while 5 (33.3%) food samples were positive for S. aureus. Overall, these data revealed the poor microbial quality in most of the street food samples tested suggesting there is an emerging need for close monitoring of microbial quality by relevant authorities in street foods sold in Nugegoda, Sri Lanka and improving food safety and hygiene practices among street food vendors.

KEYWORDS - food contamination, food quality, hygiene, microbial quality, street foods

I. INTRODUCTION

Street foods are Ready-To-Eat foods and beverages which are sold or prepared by vendors especially in streets and other similar public places. Street foods are very popular among people around the world due to their low price and convenience [1]. The number of people who involved in street food business has also increased remarkably over the last few decades due to low startup cost and comparatively loose legal restrictions in most low and medium income countries [2–4]. The number of people who depend on street foods increases with the burgeoning population and especially in many urban areas, people highly depend on street foods. Moreover, street foods are one of the convenient ways to fulfill most of the nutritional requirements among people from low-income families. The other important social group in which street foods are very popular is school children [5] and due to this reason street food vendors can be frequently found near schools, tuition classes and bus stands in Sri Lanka.

The World Health Organization (WHO) [6] reported that the microbial quality of street foods is doubtful and street foods contaminated with foodborne pathogens are the major source of foodborne diseases and food outbreaks around the world. Therefore, maintaining a good microbial quality in street foods is important to preserve the overall health and wellness of the society [2].

Street foods also represent the cultural, social, and economical characteristics of a country. Hence, they play a very important role in culinary tourism in countries where tourism is one of the major source of foreign income. Even within a small region of a country, the diversity among street foods can be high due to different preparation methods, ingredients, recipes, cultural believes, etc. The diversity of street foods makes them more attractive not only to local customers, but also to foreign tourists. Therefore, maintaining good microbial quality in street foods can also help in developing countries' economy via tourism.

Poor implementation of Good Manufacturing Practices (GMP) and personal hygiene practices by street food vendors are the major causes for most microbial contaminations in street foods [5,7,8]. A direct relationship can be identified between street food preparation/serving methods and potential microbial contaminations where many studies have reported foods that require lot of manual handling or do not cook at high temperature tend to have high microbial contaminations [9,10]. Many published studies show that street food can contain pathogenic microorganisms such as Escherichia coli, Enterobacter spp. Staphylococcus aureus, Salmonella spp., Bacillus cereus, Clostridium perfringens, and Listeria monocytogens [2,11,12].

The microbial quality of street foods is directly affected by the socio-economic characteristics such as educational level, age, income level, and dwelling areas of street food vendors [13]. Street foods can be easily contaminated by pathogenic microorganism due to unhygienic handling practices [14]. Studies have shown that the education level has a positive impact on food hygiene practices of vendors [15].

African and South-East Asian world regions where most nations are considered as developing nations, have been identified as the two world regions with highest food-borne disease cases per population [16]. Further, another two researches [10,17] also reported that food-borne illnesses are widespread in developing countries than in developed countries. Therefore, assessment of the microbial quality of street foods is a timely topic for Sri Lanka where popular street foods include roti, dosai, rice, kottu, wade, string hoppers, noodles, rolls, and pastry-like short eats [18]. There are only few studies have been published on the microbial status of street vended foods in Sri Lanka during the last decade. This study is an attempt to determine microbial quality in street foods sold in Nugegoda, Sri Lanka and thereby increase the awareness among the general public on the microbial safety of street foods.

II. MATERIALS AND METHODS

2.1 Study location and sampling

This study was conducted in the Microbiology Laboratory of the Department of Food Science and Technology of University of Sri Jayewardenepura, Sri Lanka. The selected area for this study was Wijerama junction area of Nugegoda city, in the Colombo district. Colombo district has the highest urban population (6,149,000) and the highest population growth rate among the 24 districts of Sri Lanka (Department of census and statistics, 2020). Wijerama junction is a place which is always crowded with university students, school children, tuition class students, and working people.

Five popular types of street foods (fruit juice, fruit salad, dosai, string hoppers, and wade) were selected for this study where 3 samples from each food type were analyzed. Ingredients, preparation method, serving method, and possible ways of contamination of selected street food types are shown in Table 1. The purchased food samples were transported immediately to the laboratory in a cooler box (0-4 °C) and stored at -4°C temperature until the analysis. However, all the samples were analyzed within 2 hours of sampling.

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Food type	Ingredients	Preparation method,	Serving	Possible ways of
	markey of	An approximate cooking	method	contamination
	15/10	temperature		
Fruit juice	Fresh fruits,	Mixed raw ingredients,	In cups or	Poor personal hygienic
	water, sugar, salt	27.8 − 30 °C	glasses	practices, Contaminated
				water and, utensils, Polluted
				air
Fruit salad	Fresh cut fruits,	Mixed raw ingredients,	In cups	Poor personal hygienic
	sugar, salt,	27.8 − 30 °C		practices, Contaminated
				water and, utensils, Polluted
				air
Wade	Chana dal, salt,	Deep-fried,	In paper	Poor personal hygienic
	curry leaves,	177 – 191 ℃	bags	practices, Contaminated
	chili, onions, oil			water and, utensils, Polluted
				air
String hopper	Rice flour, hot	Steamed,	In plates or	Poor personal hygienic

Table 1: Description of the analyzed RTE foods.

Assessment of microbial quality of street foods sold in Nugegoda, Sri Lanka

	water, salt	>100 °C	lunch sheets	practices, Contaminated water and, utensils, Polluted air
Dosai	Undu flour, wheat flour, yeast, salt	Fermented and cooked on a pan 80 -100 °C	In plates or lunch sheets	Poor personal hygienic practices, Contaminated water and, utensils, Polluted air

2.2 Determination of Total Plate Count and Yeast & Mold Count in street foods

Aseptic conditions were maintained throughout the analysis to avoid any external contaminations. Samples were homogenized and tenfold serial dilutions were made using peptone water (1%) up to 10^{-3} . The total plate count of street food samples was determined using Plate Count Agar (HIMEDIA –M091-500G) and incubated at $37 \pm 2^{\circ}$ C for 24-48 h as described in Sri Lankan Standard (SLS) code 516; Sec 1, Part 1; 2013. The Yeast and mold counts were determined using Potato Dextrose Agar (HIMEDIA- M096-500G) and incubated at 25-27°C for 48-72 h as described in as described in Sri Lankan Standard SLS 516; Sec 1, Part 2; 2013 methodology.

2.3 Determination of Total Coliform Count

Total Coliform Count (TCC) was determined using the Most Probable Number (MPN) three-tube method as described in SLS 516, 2013. In order to determine the presumptive TCC, tubes containing MacConkey broth (HIMEDIA- MH083-500G) and durham tubes were inoculated and incubated at $37 \pm 2^{\circ}$ C for 24-48 h. Tubes with color change (purple to yellow) and gas production were noted. Then, a loopful from tube with positive result in MacConkey broth was sub-cultured in Brilliant Green Bile Broth (BGB) (HIMEDIA-M1211-500G) and incubated at $37 \pm 2^{\circ}$ C for 24-48 h. Tubes with gas production was noted as positive result.

2.4 Presence of E.coli

A loop full from positive BGB tubes were streak platted on Eosin Methylene Blue (EMB) agar (HIMEDIA- M317-500G) and incubated at 44 - 44.5°C for 24-48 h. Plates containing colonies with specific morphological characteristics (metallic sheen, dark purple centers) were noted as presumptive *E. coli*. A loop full of colonies from previously positive plates of EMB agar media were sub-cultured in Tryptone water (HIMEDIA- M463-500G). Indole test was performed using Koyac's indole reagent, after 24hrs of incubated at 44 °C. Positive results were taken using the red ring formation on the top of the test tube.

2.5 Presence of Staphylococcus aureus

MacConkey agar (HIMEDIA- M008S-500G) media was used to enumerate *S.aureus* and round, convex and pink head colonies were morphologically recognized at the initial stage. Particular colonies with positive results were subjected to Grams' staining test to differentiate gram-positive bacteria from gramnegative bacteria. Finally, the coagulase test was performed using human plasma to confirm the presence of *S. aureus* in collected street food samples.

III. RESULTS AND DISCUSSION

In general, microbial quality and safety of street foods are questionable around the world. The consumption of microbiologically unsafe foods not only creates threat to the consumers but also can affect to the country's economy. Small children, elderly people, immune-compromised people such as pregnant women and cancer patients are the most vulnerable groups of people for foodborne diseases [12]. The main objective of this study was to assess the microbial quality of street foods sold in Wijerama junction area, Nugegoda, Sri Lanka. The tested street food samples in this study had a great diversity with respect to their raw materials, preparation method and cooking temperature. Fruit salads and Fruit juices are none-cooked preparations while Wade, Dosai, and String hopper are cooked at high temperature by deep-frying, pan cooking and steam cooking respectively.

However, the obtained data in this study revealed high bacterial loads in both cooked and none-cooked street food samples. Unsatisfactory levels of microbial contaminations in both fresh and cooked street vended foods have also been reported from various parts of the world by many other researchers [13,15,17,19].

3.1 Total Plate Counts of street food

TPC values of tested street food samples were in the range of 3.4-6.3 log CFU/ml (Table 2). The highest TPC value was observed in a Fruit salad sample (6.3 log CFU/ml). Two fruit juice samples and one fruit salad sample had unsatisfactory level of TPC values (> 1 x 10⁵ CFU/ml) and String hopper, Wade, and Dosai samples were below the marginal level (< 1 x 10⁵ CFU/ml). According to International Commission on Microbiological Specifications for Foods [20], more than 1 x 105 CFU/ml of TPC is considered as unsatisfactory level of microbial quality in majority of RTE foods. However, according to the Agri-Food & Veterinary Authority of Singapore, the maximum allowable limit for TPC in fresh foods cannot be taken into consideration as fresh products naturally tend to contain a high load of microorganisms [21]. Fruits salads and Fruit juices are generally categorized into raw food category. Therefore, the high TPC value of tested fruits juice and fruit salad samples cannot be considered as the sole indicator for poor microbial quality.

3.2 Yeast and Mold in tested street food samples

Eleven (73.3%) food samples were found to be positive for Yeast and mold and the results ranged between 3.7-6.4 log CFU/ml (Table 2). Dosai sample exhibited the highest TYMC value (6.4 log CFU/ml) among tested street food samples. Standard TYMC limits for street foods is not mentioned in the SLSI guidelines. Among the analyzed samples, dosai was reported to bear the highest TYMC (6.4 log CFU/ml). However, fermented and raw food items generally contain high load of microorganisms due to the fermentation process. Therefore high TYMC values for dosai, fruit juice, and fruit salad samples are quite acceptable. But out of three tested Wade samples, high yeast and mold counts (5.42 log CFU/ml, 3.80 log CFU/ml) were detected in two wade samples. Wade is a none-fermented deep fried food which is prepared by using ground Chana dal paste (Table 1). Therefore, it is unlikely for properly cooked wade to carry such high yeast and mold count. Therefore, it can be suspected that Wade sample was undercooked or old.

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Tested street food type	Ra <mark>nge</mark> of T <mark>otal</mark> plate count (log CFU <mark>/ml)</mark>	Range of Yeast and Mold count (log CFU/ml)
Fruit juice	$3.80\pm0.01 - 6.25\pm0.02$	4.25±0.35 - 6.33±0.01
Fruit salad	3.36±0.05 - 6.29±0.05	3.78±0.04 - 6.33±0.05
String hopper	$3.88\pm0.15 - 3.95\pm0.10$	-
Wade	3.52±0.06 – 3.58±0.19	3.79±0.11 – 5.42±0.03
Dosai	3.95±0.08 – 4.43±0.01	3.78±0.09 - 6.42±0.01

3.3 Total coliform count of tested street food samples

Coliform organisms are being used as a general indicator of the overall quality of foods and hygienic conditions during food processing [5]. Therefore, high counts of total coliform indicate poor sanitary conditions and unhygienic handling practices during and after food processing. In this study, 2 fruit juice, 1 fruit salad, and 2 string hopper samples (33%) exceeded the permissible limit of TCC (> 10MPN/ml) indicating that the food samples may also contain potentially hazardous microorganisms such as E. coli 0157, Salmonella, Staphylococcus, etc.

3.4 Escherichia coli contaminations in tested street food samples

Thirteen (86.7%) presumptive *E.coli* samples were initially identified. Among them, 53.3% of samples (fruit juice- 3; fruit salad-1; string hopper- 3; dosai- 1) were found to be positive for the Indole test which confirms the presence of E.coli in tested street foods (Table 3). According to the Sri Lankan standard (SLS 1162: 1997/ UDC664.696.1), E.coli should be absent in 1g of ready to serve food. Even though the level of E. coli in food samples were not enumerated, our findings on E.coli indicated alarming microbial quality in street foods sold in Wijerama junction area, Nugegoda. Similar situations seem to persist in other developing countries in the South Asian region such as Bangladesh [9], [22], Pakistan [23] where over 50% of the tested street food samples found to be positive for E. coli.

Large number of foodborne outbreaks have been reported in recent years as a result of emerging and re-emerging enterohaemorrhagic *E. coli*, *Campylobacter spp.*, *Listeria monocytogenes*, *Vibrio cholerae* and other foodborne pathogens [24]. Increased consumption of raw and minimally processed foods and unhygienic food preparation practices by food handlers are the major sources of E.coli contamination. WHO stated that cooking at a temperature above 70 °C for at least an hour, kills *E.coli*. In this study, string hopper (3), fruit juice (3), fruit salad (1) and dosai (1) samples were found to be positive for *E.coli*. Notably, string hopper and dosai are usually being cooked at a temperature close to 100 °C using steam cooking and pan cooking (Table 1). Therefore, E.coli positive results in string hopper and dosai samples can be due to incomplete cooking or post-processing contaminations.

3.5 Staphylococcus aureus contaminations in tested street food samples

Staphylococcus is an important foodborne pathogen that produces highly heat stable enterotoxin [25]. It is a gram-positive round-shaped bacteria that can be commonly found in the human upper respiratory system. Thus their presence in food indicates poor sanitary practices in food handlers [26]. Among the tested food samples, five (33.3%) samples including 3 string hopper, 1 fruit juice, and 1 dosai samples were found to be positive for Staphylococcus aureus (Table 2). S. aureus in string hopper can be due to excessive handling with bare hands during counting and serving.

Table 3: Summary of *E.coli* and *S. aureus* test results for tested street food samples.

Tested street food type	Presence/absence of <i>E.coli</i> in street	Presence/absence of Staphylococcus
	food samples	aureus in street food samples
Fruit juice	100	· _ #
Sample 1	Present	Present
Sample 2	Present	ND
Sample 3	Present	ND
Fruit salad		
Sample 1	Present	ND
Sample 2	ND	ND
Sample 3	ND	ND
String hopper	ASSESSED FOR THE PARTY OF THE P	
Sample 1	Present	Present
Sample 2	Present	Present
Sample 3	Present	Present
<u>Wade</u>		
Sample 1	ND	ND
Sample 2	ND	ND
Sample 3	ND	ND
<u>Dosai</u>		
Sample 1	Present	Present
Sample 2	ND	Present
Sample 3	ND	Present

Note: ND - Not detected

In general for most of the street food vendors there is no access to proper sanitary facilities such as potable water and toilets. Moreover, they don't have proper facilities to keep food covered while preparing and selling. These types of deficiency in facilities have made it difficult to improve the microbial quality in street foods sold in Sri Lanka. Similar types of insufficiencies in infrastructure in street food vending have also been observed by many other people around the world [27,28]. The poor microbiological quality of street foods can also be due to the lack of knowledge on good personal hygiene practices among street food vendors [3,23]. Some common mistakes that most of Sri Lankan street food vendors do during food handling include touching food with bare hands and not washing hands properly after handling money or after using the toilet. Another common hygiene mistake that these vendors do is not cleaning utensils such as cutting boards, spoons, knives, glasses and cups properly during food preparation or serving. Recent studies from Bangladesh [9], from Pakistan [29], and from India [30] reported that street food vendors in their countries also have similar poor hygiene practices.

A recent study carried out in the Colombo municipal area in Sri Lanka [18] revealed that the majority of street food vendors did not have an authorized license or proper training on food safety practices. Therefore, there is an emerging need to provide proper training on food safety practices and sanitary practices to street food vendors as many of them are unaware of GMP, and personal hygienic practices. Furthermore, according to the "Code of hygiene practices for the preparation and sales of street foods" published by Sri Lankan Standard Institute (SLS 1451:2013), all the ingredients and stages in food handling and preparation should be considered as Critical Control Points (CCPs) and necessary actions must be taken at each CCP to avoid possible microbial contamination and growth.

IV CONCLUSION

According to our research findings, a considerable amount of street food samples obtained from Wijerama junction area, Nugegoda had unsatisfactory microbial quality such as high TPC, TCC. Moreover, significant number of tested food samples were found to be positive for E. coli and S. aureus indicating incorrect food handling practices during processing and vending. Based on this research findings, it can be suggested that food safety and hygiene practices followed by these street food vendors need improvements, and enforcement of strict regulations on good manufacturing and hygienic practices among street food vendors is important.

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Assessment of microbial quality of street foods sold in Nugegoda, Sri Lanka

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Assessment of microbial quality of street foods sold in Nugegoda, Sri Lanka

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