

Microbiological quality analysis of commercial fruit juice in Dhaka City, Bangladesh

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The aims of this study were to evaluate the microbiological quality of different commercially available fruit juice of Dhaka city. In this study total 20 fruit juice samples of five different types were repeatedly subjected to bacteriological and mycological screening for three months. Isolates were identified and confirmed using cultural and biochemical characteristics. Current study showed high bacterial and fungal loads consisting of specific pathogens such as coliform and *Staphylococcus* spp., fecal coliform and other pathogens are totally absent. The presence of *Staphylococcus aureus* portends health risk to consumers as some species produce potent toxins associated with food borne illnesses. And high bacterial count indicated unhygienic conditions of the processing area, and that good manufacturing practices during processing are not maintained properly. Recommendations are required to reduce the microbiological contamination and promote quality assurance of the products.

Key words: Fruit juice; Microbiological quality; Public health

Fruit juices are most common refreshing drinks in summer season (1). They taste good and also contain nutrients (1-3). Juices are fat-free and contain naturally occurring phytonutrients, antioxidant, vitamins and minerals which are essential elements for better health. Eventually it becomes an important part of the modern human diet (4-5). Vitamin C in orange juice acts as an antioxidant photochemical which help to improve the blood lipid profiles in hyper-cholesteromic patients (6). Fruit juices promote detoxification in the human body (7).

Fruit juices are available in packed form in different concentration though out the year. As fresh juice, they also rich in nutrient and to ensure the microbiological quality as well as same taste, color and aroma of fresh juice food industry employ pasteurization, sterilization or addition of preservative to fruit juice (1, 8-10). But different pathogenic microbes can get entry to packed juice during processing, packaging and handling (11). Different studies on fruit juice around the world reported that consumption of contaminated juices is responsible for number of diseases (1, 12-16). As juice contain high amount of nutrients and sugar make juice more susceptible to microbial contamination (1-3, 18). But to prevent microbial growth, several factors such as pH, temperature and concentration of preservative may play good role (9, 17-18).

The most common food borne pathogenic bacteria are *Bacillus cereus*, *Clostridium botulinum*, *Escherichia coli*, *Shigella* spp., *Salmonella* spp., *Vibrio parahaemolyticus*, *Staphylococcus aureus*, *Campylobacter jejuni*, *Streptococcus pyogenes*, *Listeria monocytogenes* etc. (1). Microbial contamination of juice can heavily infected the fetus, leading to spontaneous abortion, stillbirths, or sepsis in infancy (1). Coliforms, faecal coliforms contamination may come from the water used for juice preparation where environmental fomites may have a role in spreading of *Salmonella* spp., *Shigella* spp., *Vibrio* spp., *Escherichia coli*, and other diseases causing microbes (1, 18-19). Sometime growth of spoilage yeasts, such as *Saccharomyces cerevisiae*, *Candida lipolytica* and *Zygosaccharomyces* spp. can changes in pH which make juice more susceptible growth of pathogens (1, 18, 20). The existence of microorganisms and their parts including bacterial toxin or poisonous protein of certain foods can cause food borne illness and also responsible for food spoilage (14, 18). Consequently, commercially available juice could be maintaining their quality by following different recommended guidelines like FDA (Food and Drug Administration), FAO (Food and Agriculture Organization).

Dhaka is one of the densely populated city and have hot climate during summer resulting in increased consumption of fruit juices which may consequently result in food borne illnesses. So maintaining the quality of processed fruit juices is important issue at present. In order to develop awareness among the people about fruit juices in transmitting diseases this study was attempted to

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analyse the microbiological status of some brands of fruit juice processed and sold in Dhaka, Bangladesh.

MATERIALS AND METHODS

Sampling and sample processing. Five different fruit juice samples from four different brands were used in this study within a time frame of November 2017 - March 2018. Samples were randomly collected from different super shops in Dhaka city. Dates of manufacturing and expiry were checked prior to microbiological tests. Prior to the microbiological assay for the estimation of bacterial and fungal load, samples were well mixed with normal saline (in 1:10 ratio). Serial dilutions were then consecutively prepared up to 10^{-5} (21).

Enumeration of total viable bacteria and fungi . An aliquot of 0.1 ml of each suspension from the dilution 10^{-2} and 10^{-4} was introduced onto the nutrient agar (NA) plates and Sabouraud dextrose agar (SDA) plates by means of spreading in order to isolate and quantify the total viable bacterial count (TVBC) and fungi, respectively (18, 21, 24). The NA plates were incubated at 37 °C for 18 to 24 hours and the SDA plates were incubated at 25 °C for 48 to 72 hours, respectively.

Enumeration of specific pathogens. From the dilution of 10^{-2} of each sample, 0.1 ml of suspension was spread onto MacConkey agar, mannitol salt agar (MSA), and Cetrimide agar, for the enumeration of *Escherichia coli*, *Staphylococcus* spp., *Pseudomonas* spp., consecutively. All the plates were incubated at 37 °C for 24 hours. Appearance of the typical colonies such as pink colonies on MacConkey agar, yellow colonies on MSA, colonies with greenish pigmentation on cetrimide agar, was analytical for the growth of *E. coli* or *Klebsiella* spp., *Staphylococcus* spp., *Pseudomonas* spp., consecutively (18, 21). Finally the confirmative biochemical tests were conducted to ensure the identity of the isolates (1-2, 22-24). Enumeration of bacterial load was performed by standard method (25).

RESULTS AND DISCUSSION

Refreshing drinks including the fruit juices are very popular among the people of all ages around the world. Also in Bangladesh, ready to eat refreshing fruit juice

or sherbet is becoming more and more popular as they are usually tastier than soft drinks. After several study on food products from local market, they concern about the food safety and security issues in Bangladesh (1, 2, 22, 23). Nevertheless, researchers reported that microbial contamination in raw juice could take place from principal sources: from the raw materials; during handling; and from the unhygienic storage conditions (26-27).

Current investigation also showed a situation of huge microbial contamination in most of the samples. All samples were found to harbor the total viable bacteria within a range of 10^2 - 10^6 cfu/ml (Table 1). Out of 20 (twenty) fruit juice samples only 7 (seven) exceeded the microbial limit (28-30). And total fungal count of tested samples was within a range of 10^2 - 10^3 cfu/ml. Here all the samples were found to have fungi in the acceptable limit (28-30). This study reported that total viable bacterial count in most of the mango and lichi juice samples was higher than the others packed juice due to the variation in pH or chemical composition because juice of orange, grape and apple contain low pH.

Unlikely, pathogenic bacterial were found which were biochemically identified (Tables 1 and 2). Although fecal coliform, *Salmonella* spp. and *Vibrio* spp. was not encountered in any sample, only 6 samples were found to be contaminated with coliform within a range of 10^1 - 10^2 cfu/ml. *Staphylococcus* spp. were found in all the samples

TABLE 1. Microbial load in the commercial juice samples

Juice sample	Brand	Microbial load (cfu/ml)			
		Total Viable Bacteria	Fungal count	Total coliform	<i>Staphylococcus</i> spp.
Mango	A	2.8×10^6	1.2×10^3	2.0×10^2	1.1×10^2
	B	1.9×10^5	1.5×10^3	4.2×10^2	2.1×10^3
	C	2.7×10^6	2.2×10^3	0	2.6×10^3
	D	5.3×10^6	2.5×10^3	0	1.1×10^2
Lichi	A	1.3×10^6	1.2×10^3	2.0×10^2	2.1×10^3
	B	1.2×10^4	2.5×10^3	0	2.6×10^3
	C	3.5×10^4	1.1×10^2	0	1.1×10^2
	D	4.2×10^5	2.1×10^3	0	2.1×10^3
Orange	A	4.5×10^5	2.6×10^3	2.1×10^2	2.6×10^3
	B	2.3×10^3	1.5×10^2	0	1.1×10^2
	C	3.5×10^4	2.1×10^2	0	2.1×10^3
	D	1.4×10^3	1.1×10^2	0	2.6×10^3
Grape	A	3.5×10^4	2.2×10^2	0	1.1×10^2
	B	3.2×10^4	2.6×10^2	1.0×10^2	2.1×10^3
	C	2.5×10^3	1.1×10^3	1.1×10^1	2.6×10^3
	D	3.2×10^4	3.1×10^2	0	1.1×10^2
Apple	A	3.3×10^3	2.1×10^3	0	2.1×10^3
	B	1.2×10^2	1.0×10^2	0	2.6×10^3
	C	2.1×10^2	1.7×10^2	0	1.1×10^2
	D	3.7×10^3	2.3×10^2	0	2.1×10^3

Fecal coliform, *Staphylococcus* spp. and *Vibrio* spp. were absent in all the samples.

Microbiological standard recommended for any types of juice (Gulf Standards (29)

Maximum bacterial load anticipated: Total viable count - 5.0×10^3 , Coliform - 10, Fecal coliform - 0, Staphylococci - 100

Maximum bacterial load anticipated: Total viable count - 1.0×10^4 , Coliform - 100, Fecal coliform - 0, Staphylococci - 1.0×10^3

TABLE 2. Confirmative biochemical tests for the isolates

Assumed Organism	TSI			H ₂ S reaction	Indole test	MR test	VP test	Citrate test	Motility	Oxidase test
	Slant	Butt	gas							
<i>Klebsiella</i> spp.	Y	Y	-	-	-	-	-	-	-	+
<i>Staphylococcus</i> spp.	Y	R	+	+	-	+	-	+	+	-

TSI = Triple Sugar Iron Test

Y = Yellow (Acid)

R = Red (Alkaline)

MR = Methyl red

VP = Voges Prokauer

in a range of 10^2 - 10^3 cfu/ml. The presence of coliforms and *Staphylococcus* spp. in juice may be introduced due to improper processing as they are recognized to be the natural flora of fruits (31).

CONCLUSIONS

This work assessed the bacteriological condition of available local refreshing drinks. Cheap price and ease of availability make these drinks and sherbets highly demanding to people of all income groups. It was also found that the presence of pathogenic organisms such as total coliform, salmonella and fungus were within the acceptable range and considered safe for consumption. The Government-authorized institute such as Bangladesh Council of Scientific and Industrial Research (BCSIR) and BSTI should undertake pre-emptive investigations to check the microbial and chemical quality of the fruit juices as well as initiate increased public awareness programs on contaminated and adulterated juices. The present study revealed that all the studied samples had a higher microbial load than the specification set for these kind of drinks. Continuous scrutinizing of the quality of such drinks for human consumption is suggested to evade the outbreak of any bacterial pathogen.

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