An Assessment of Microbiological Quality of Some Commercially Packed and Fresh Fruit Juice Available in Dhaka City: A Comparative Study

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Fruit juices are regarded as the most preferred non-alcoholic beverage worldwide to all age groups. In general, microbial growth in fruit juice is restricted by using preservatives and also through other environmental factors during production. Still, the presence of some indicator organisms in fruit juice is responsible for causing food-borne diseases and even death. So, from public health point of view, it is quite important to know the microbiological quality of the fruit juices available in the market. Primary objective of this study was to assess the microbial quality of fresh and commercially packed available juices collected from different locations of Dhaka city. A total of six fresh juice and nine commercially packed juice samples were collected. Standard culture techniques were followed to assess total viable count (TVC), total staphylococcal count (TSC), total Bacillus count (TBC) and total fungal count (TFC) on different culture media. The TVC varied from the range of $10^2$ to $10^5$ cfu/ml with the highest of $2.4 \times 10^5$ cfu/ml. A large number of staphylococci and Bacillus was also found from several samples. Total coliform and fecal coliform was found in six and five (out of fifteen) samples, respectively. Among total coliforms, Klebsiella, Enterobacter along with E. coli were detected. From all the assessment it was determined that the microbial quality of commercially packed juice was fairer than that of fresh juice collected from local market.

In recent years, the increasing consumer awareness has emphasized the need for microbiologically safe food. Since the human food supply consists basically of plants and animals or products derived from them, it is understandable that our food supply can contain microorganisms in interaction with the foods. When the microorganisms involved are pathogenic, their association with our food is critical from a public health point of view. Serious health hazards due to the presence of pathogenic microbes in food can lead to food poisoning outbreaks (1). Freshly extracted fruit juices, which have always been considered as healthful drink, may not always be safe owing to the heavy load of microbes (2). In a situation where storage of juice is indispensable may be for a short duration, then the use of simple processing techniques soon after its extraction is essential for improving the keeping quality to ensure its safety, nutritive quality and acceptability while consumption. Preservation of fruit juice by pasteurization, refrigeration and sterilization are popular methods used to attain microbiological stability by destroying pathogenic microorganisms and to preserve the color, aroma and taste of fresh juice (3).

As indicated, the sources of fruits that can be made into juices are nearly limitless in plants and the plant parts appropriate for juice. However, the fruit producers and juice processors must adhere to some very important guidelines in fruit selection and juice manufacture (4). Environmental sources of contaminating organisms of juices are carefully considered as these microbes invade the drink preparations during processing, packaging and handling (5, 6).

The major ingredients of the juice such as water, sugar, natural fruit pulp etc may also carry some microbial contaminants. Food-borne illness is commonly caused by certain bacteria or their toxins, which are poisonous proteins produced by these bacteria (7). 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, Mycobacterium bovis, Listeria monocytogenes etc. In pregnant women, the fetus is most heavily infected, leading to spontaneous abortion, stillbirths, or sepsis in infancy (8). Contamination of juices with pathogenic microorganisms such as E. coli and Salmonella spp. has caused numerous illness and even some fatalities (9, 10).

The fruit producers and juice processors must adhere to some very important guidelines in fruit selection and juice manufacture. The juices contain mainly water, sugar, preservatives, color, fruit pulps and other additives (Table 1).
Table 1. The recommended microbiological standards for any fruit juice; all numbers are as per ml of juice consumed (11)

| Parameter                        | Total viable count | Coliform | Fecal coliform | Staphylococci |
|----------------------------------|--------------------|---------|---------------|---------------|
| Maximum count anticipated        | 5.0x10^3           | 10      | 0             | 100           |
| Maximum count permitted          | 1.0x10^4           | 100     | 0             | 1.0x10^5      |

The presence of microbial contaminants in water, sugar and pulp may cause spoilage of the drinks or gastrointestinal diseases to the consumers (4).

Industrially processed fruit juices were investigated for some physicochemical tests. They apply chemical preservatives that can inhibit all types of microbial growth. It is well known that the manufacturers commonly use Sulphur Dioxide (SO2) and benzoate as preservatives in processed fruit juices. Sulphur dioxide and benzoate can significantly damage the vegetative cells. The sulphites inhibit yeasts, moulds and bacteria and are most effective as inhibitors of browning in foods (8). They also reduce the number of growth of microbes and increase the shelf life of juice products.

The effect of storage temperature has also been reported in various research papers. At 4 °C nutritional values of juice were longer than 25 days in all system tested (12). In another study, while the refrigeration condition was able to stabilize Roselle fruit flavored drinks for a week, the freezing condition was able to preserve the drinks for more than a week (13).

The focus of this study was to compare the quality of fresh and commercially available fruit juices by assessing their microbial load and the presence of pathogenic bacteria.

Materials and Methods

Sample collection. Fifteen samples were collected, of which 6 fresh juice samples were collected from 5 different locations of Dhaka city and other 9 juice samples were commercially packed juices available in market. The table of collection sites is given below (Table 2).

| Parameter                        | Total viable count | Coliform | Fecal coliform | Staphylococci |
|----------------------------------|--------------------|---------|---------------|---------------|
| Maximum count anticipated        | 5.0x10^3           | 10      | 0             | 100           |
| Maximum count permitted          | 1.0x10^4           | 100     | 0             | 1.0x10^5      |

Microbial load in fresh juice samples. The mean bacterial count in fresh juice samples was 1.14x10^4 cfu/ml. The total viable count varied with different types of juices that ranged from 3.2x10^5 to 2.4x10^6 cfu/ml. Similarly, we found mean staphylococcal count as 7.2x10^5 cfu/ml with the range from 0 to 2.5x10^6 cfu/ml. In case of total Bacillus count, we found that the mean was 7.2x10^3 cfu/ml and it ranged from 0 to 2.5x10^3 cfu/ml in different fruit juice samples (Fig. 1).

Microbial load in commercially packed juice sample. The mean bacterial count in commercially packed juice samples was 1.8x10^5 cfu/ml. The total viable bacterial count varied with types of juices as well as different brands of same type of juices that ranged from 3.1x10^5 to 3.2x10^5 cfu/ml. Likewise, we found mean Staphylococcal count as 4.9x10^5 cfu/ml with the range from 0 to 2.3x10^5 cfu/ml. Moreover, mean Bacillus count was found 7.3x10^5 cfu/ml and it ranged from 1x10^5 to 2x10^5 cfu/ml in different types and brands of fruit juice samples (Fig. 1 & 2).

Total fungal count. The total fungal count was observed on potato dextrose agar (PDA) media. The single medium used for the enumeration of fungi showed a little range of growth from 2.3 x 10^2 to 9.0 x 10^6 cfu/ml (Table 3).
FIG. 1. Total viable bacterial count, total Staphylococcal count and total Bacillus count in (A) fresh juice (B) commercially packed juice samples

FIG. 2. Comparative mean viable bacterial count, Staphylococcal count and Bacillus count between fresh juice and commercially packed juice samples
### TABLE 3. Total fungal count of the tested samples in the PDA media

| Fresh juice | Sample no. | Average no. of fungal count (cfu/ml) |
|-------------|------------|-------------------------------------|
| Orange      | FO1        | 9.0 x 10⁶                            |
| Orange      | FO2        | 5.5 x 10³                            |
| Papaya      | FPA        | 4.5 x 10⁴                            |
| Pineapple   | FPI        | 5.4 x 10⁵                            |
| Sugarcane   | FS1        | 8.1 x 10⁴                            |
| Sugarcane   | FS2        | 2.3 x 10²                            |

| Commercially packed juice | Sample no. | Average no. of fungal count (cfu/ml) |
|---------------------------|------------|-------------------------------------|
| Orange                    | COA        | 8.4 x 10⁴                            |
| Orange                    | COB        | 2.2 x 10³                            |
| Orange                    | COC        | 5.7 x 10⁶                            |
| Orange                    | COD        | 2.2 x 10⁴                            |
| Mango                     | CMA        | 3.5 x 10⁶                            |
| Mango                     | CMB        | 2.5 x 10⁴                            |
| Mango                     | CMC        | 4.2 x 10⁴                            |
| Mango                     | CMD        | 8.2 x 10⁴                            |
| Apple                     | CAP        | 5.2 x 10⁴                            |

### TABLE 4. Presence or absence of total coliform and fecal coliform in the fruit juice

| Sample no. | Presence of total coliform | Presence of fecal coliform |
|------------|---------------------------|---------------------------|
| FO1        | +                         | Positive                  |
| FO2        | -                         | Negative                  |
| FPA        | +                         | Positive                  |
| FPI        | +                         | Positive                  |
| FS1        | +                         | Positive                  |
| FS2        | -                         | Negative                  |
| COA        | +                         | Negative                  |
| COB        | +                         | Positive                  |
| COC        | -                         | Negative                  |
| COD        | -                         | Negative                  |
| CMA        | -                         | Negative                  |
| CMB        | -                         | Negative                  |
| CMC        | -                         | Negative                  |
| CMD        | -                         | Negative                  |
| CAP        | -                         | Negative                  |

*: positive; -: negative

**Isolation of the total coliform and fecal coliform bacteria.** In the confirmatory test for the presence of coliform bacteria in the fruit juice, the samples were inoculated on the EMB agar media (Table 4).

**Identification of the total coliform.** The isolated coliforms were further inoculated in different biochemical media for the differentiation among the groups. On the basis of those tests, the bacteria were identified to the genus level. The result is given in the following table (Table 5).
DISCUSSION

Fruit juices are very popular among the people of all ages around the world. Also in Bangladesh, ready to eat packed fruit juice is becoming more and more popular as they are usually tastier than soft drinks. Advantage of packed fruit juices is that these are convenient to carry and can be kept for a considerable amount of time (14, 15). Fruit juices by their very nature contain various organisms. Clearly, many of these microorganisms will be harmless yeasts & saprophytic bacteria. By detecting this bacterial load in the juice, it apparently gives an idea about the quality of the sample. Therefore, aerobic plate count or total counts of some popular juice were carried out in the present experiment.

Total viable bacterial count in most of the fresh juice samples was higher than the commercially packed juice, as the highest count was found as $2.4 \times 10^4$ cfu/ml and $3.2 \times 10^3$ cfu/ml in fresh and packed juice, respectively. The scenario of total staphylococcal and Bacillus count was also similar. Though most of the juice samples showed equal or slightly higher count than the permitted count (11), these were unfavorable for consumption because six of them showed the presence of coliform. The presence of coliform in fruit juice is not allowed by safe food consumption standard (14). Coliforms were identified as E. coli, Enterobacter and Klebsiella, which can produce many systematic infections after consumption.

The fungal count was also much higher in many samples and varied from sample to sample. Though the fungal isolates were not identified, colony morphology and color showed the probability of the presence of pathogenic strain and could produce mycotoxins in juice samples.

The comparative study anticipated the safe consumption of commercially packed juice than the freshly packed juice marketed locally. This might be the reason of using automated machine and also some preservatives during fruit juice processing. But some preservatives of higher concentrations can be harmful for our health. Therefore studies on preservative concentrations should also be carried out.

The juices that are mishandled and mistreated during preparation or storage generally cause most outbreaks. The disease agents spread by juice like drink not only harm large groups of people but also sometimes result in serious disability and death. The lack of knowledge in cleaning, safe fruit juice preparation and contamination sources can contribute to the contamination process unknowingly to the people working in fruit juice industries. It is therefore, essential for the people who handle and prepare juices, to be properly trained in safe fruit handling technique.

The microbiological quality of processed fruit juices is the most important aspect to be taken care of by the manufacturer. Negligence in this area may result in serious contamination that ultimately represents a low quality product to the consumers. As these unwanted unhygienic conditions are usually due to the lack of knowledge and unawareness to the fundamental sanitary principles, it is preventable by proper training and monitoring. The government authorized institute (like BSTI) should take intensive investigation to control the microbial and chemical quality of the juices as well as the public awareness about the adulterated fruit juices should be increased.

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