

## Physico-Chemical and Microbiological Safety of Fruit Juices Served in Bahir Dar City, Northwest Ethiopia

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### Abstract

Fruit juices are becoming an important part of the modern diet in many communities. For physico-chemical and microbiological safety of fruit juices, a total of 120 fruit juice samples (30 each of avocado, papaya, mango and pine-apple) were collected. Questionnaire was used to obtain preliminary information on hygienic and safety practices of fruit juice makers and handlers. The mean pH of fruit juices investigated in this study ranged from  $4.13 \pm 0.001$ , as in the case of pine-apple juices, to  $5.09 \pm 0.14$  in avocados juices. The highest titratable acidity was recorded in pine-apple juices ( $0.203 \pm 0.164\%$ ) and the lowest in avocado juices ( $0.081 \pm 0.07\%$ ). The highest quantity of vitamin C ( $85.47 \pm 0.02$  mg/100g) was recorded in papaya juices and lower in mango juices ( $8.55 \pm 0.11$  mg/100g). The highest content of reducing sugar was observed in papaya juices ( $11.54 \pm 1.2\%$ ) while the lowest in avocado juices ( $4.15 \pm 0.25\%$ ). The total soluble solids content of pine-apple juices ( $13.43 \pm 0.08\%$ ) in this study was higher and lower in avocado juices ( $9.75 \pm 0.09\%$ ). The mean total viable count was highest in avocado juice ( $3.1 \times 10^7$  cfu/ml) and lowest in pine-apple juice ( $1.5 \times 10^5$  cfu/ml). Yeast count was highest in papaya juice ( $6.2 \times 10^4$  cfu/ml) and lowest in pine-apple juice ( $5.0 \times 10^3$  cfu/ml). Papaya and pine-apple juices recorded the lowest mold count ( $3.2 \times 10^4$  cfu/ml). In conclusion, the fruit juices had higher microbial loads than the specifications set for fruit juices in some parts of the world. Presence of pathogenic species in some of these fruit juices should be of paramount concern.

**KEYWORDS:** Fruit juice, hygiene, Bahir Dar, Microbial safety, physicochemical parameters

### INTRODUCTION

Fruit juices are available in essentially the same form almost anywhere in the world (Dennison, 1996). Due to their potential nutritional and biological fruit juices are foods with multiple implications for body balance (DHFS, 1998). Fruit juices are common beverages in many countries of the world. In hot climate areas, cafés, restaurants and road side stalls have local facilities to extract the juice from fresh fruits (Al-jedah, 2001).

Several factors encourage, prevent or limit the growth of microorganisms in juices; the most important are pH, hygienic practice and storage temperature and concentration of the preservative (Deanna & Jeffrey, 2007).

The quality of fruit juices is strictly maintained in developed countries under several laws and regulations but in many developing and underdeveloped countries, the manufacturer are not concerned about the microbiological safety and hygiene of fruit juices because of lack of enforcement of the law. Thus the transmission of certain human diseases through juice and other drinks in recent years is a serious problem (FDA, 2001).

The objective of this study was, therefore, to study the physico-chemical and microbiological safety of fruit juices served in Bahir Dar City, Northwest Ethiopia.

## **MATERIAL AND METHODS**

A total of 120 fruit juice samples (30 each of avocado, papaya, pineapple, and mango) were collected from ten randomly selected cafés from among over 25 cafés/restaurants in Bahir Dar City between may, 2006 and June, 2007. As some of the fruit juices vending cafés were serving either one, two, three, or four types of the fruit juices, only those serving the maximum number of fruit juices were considered and ten of them were selected for sampling.

The fruit juices investigated in this study area were juices made from avocado, papaya, pineapple, and mango. Samples (250 ml each) of these fruit juices were separately collected in sterile flask and transported to the laboratory. Samples were processed within four hours of collection.

Questionnaire was used to obtain preliminary information on the demographic characteristics of the fruit juice makers, servers, and cares being taken during processing of the fruit juices. All the personnel's involved in the processing and/or serving of the fruit juices in the selected cafés/restaurants were included.

### **Sample Processing and Microbial Analysis**

Ten milliliter of the sample was diluted in 90 ml of distilled water and mixed well. The samples were homogenized and appropriate dilutions of up to  $10^{-6}$  were plated in triplicates on surfaces of respective media for microbial count using the spread plate technique. Aerobic mesophilic bacteria (AMB) were counted on plate count agar (PCA) after incubation at 32°C for 48 hours (Subbannayya et al., 2007).

Similarly, yeasts and molds were isolated on Sabouraud's dextrose agar (SDA) incubated at 28°C for 5 days. Smooth (non-hairy) colonies without extension at periphery (margin) were counted as yeasts. Hairy colonies with extension at periphery were counted as molds.

### **Determination of pH, titratable acidity, moisture content, Ascorbic acid and Total Soluble Solid**

PH was measured using digital pH meter (Nig 333, Naina Solaris LTD, India) after homogenizing 10 ml of the fruit juices in 90 ml of distilled water (Erkmen and Bozkurt, 2004; Ferrati, 2005).

Standard method was used to measure titratable acidity (Antony and Chanrda, 1997; Ferrati, 2005).

The fruit juice sample (5ml) was homogenized in distilled water (20ml) and filtered through whatman No.1 filter paper. Two to three drops of phenolphthalein were added to 20ml of the filtrate as indicator and titrated against 0.05M NaOH to determine the end point of phenolphthalein.

Titrateable acidity was expressed as g lactic acid/100g of juice and calculated using the formula:

$$TA = \frac{MNaOH \times ml \text{ NaOH} \times 0.09 \times 100}{ml \text{ juice sample}}$$

Where, TA = Titrateable acidity; MNaOH = Molarity of NaOH used; ml NaOH = amount (in ml) of NaOH used; 0.09 = equivalent weight of lactic acid.

A moisture content of fruit juices was determined using standard AOAC methods (Horwitz, 2003).

Ascorbic acid was estimated by UV-spectrophotometer method according to AOAC. The reagents used for the estimation of vitamin C as follows: (i) metaphosphoric acid (6%; xylene (ii) standard ascorbic acid solution; and (iii) 2, 6- Dichlorophenolindophenol dye. For estimation of vitamin-C, the following steps were followed: standardization of dye solution, preparation of solution and measurements of absorption (AOAC, 2004).

Total Soluble Solid (TSS) content of fruit juices were determined using an Abbe refractometer whereby a drop of pulp solution was placed on its prism. The percentage of TSS was obtained from direct reading of the refractometer

#### **Statistical Data Analysis**

All experiments were done in triplicate. Data were expressed as mean  $\pm$  standard error (SE) of triplicate experiments. The Statistical Analysis was carried out by one way ANOVA (Bhat et al., 2008)

## **RESULT AND DISCUSSION**

In Bahir Dar city, street foods provide an affordable source of nutrients to many sectors of population. Street-vended fruit juices are well appreciated by consumers, because of their taste, low price, and availability at right time (FAO, 1988; Ohiokpehai, 2003). However, street foods are frequently associated with diarrhea diseases due to their improper handling and serving practices (WHO, 2002; Barro et al., 2006; Bello et al., 2013).

The socio-demographic profile of respondents is presented in Table 1. Altogether there were 25 respondents. The mean age of respondents was 23 $\pm$ 4. About 75% of the respondents were aged between 19 - 30 years. Majority of workers in juice houses were females (95%) and were educated up to secondary school level. Only 3(12%) respondents had undergone pre-placement training for food handling indicating that professional training takes its own part to reduce food borne illnesses (FDA, 2001). The source of fruits used for the processing of juices was majorly from the open market as this constituted 80% of this study, while only 20% got their fruits directly from producers who were their routine suppliers.

Fruit juices producers made use of both ripened and over-ripened fruits, but with preference to ripened fruits as this constituted 96% of the study. The temporary storage sites of fruits were shelves (44%), baskets (36%) and refrigerators (16%). However, 4% of the juice producers had no special storage site for their fruits.

**Table1. Demographic study of fruit juice processing conditions in Bahir Dar City**

<b>Characteristics</b>	<b>Number/ Percentage of Respondents</b>
<b>Source of fruits</b>	
Open market	20(80%)
Directly from producers	5 (20%)
<b>Nature of fruit used:</b>	
Ripened	24 (96%)
Over ripened	1(4%)
<b>Temporary storage site of fruit</b>	
Shelf	11(44%)
Basket	9 (36%)
Refrigerator	4 (16%)
No special storage	1 (4%)

Juice handlers were asked how they prepare fruits and all of them peeled and cut; mango, pineapple, avocado, and papaya fruits with knives before being juiced. But, observation reveals that majority of juice vending houses store fruits outside in a condition that is exposed to temperature abuse and dust. Reasons for proliferation of microorganisms in fruit juices could also be attributed to the fact that the most juice producers lacked special training in food hygiene and safety.

**Table 2: physico-chemical properties of fruit juices served in cafés/ restaurants in Bahir Dar city, 2006 – 2007.**

Sources of fruit juices	Sample size	Moisture content (%)	Total soluble solids (Brix %)	Titratable Acidity (%)	PH	Reducing Sugar (%)	Vitamin(mg/100g)
Mango	30	87.32±0.04	12±0.23	0.105±0.00	4.32±0.42	10.51±0.23	8.55±0.11
Avocado	30	69.81±0.17	9.75±0.09	0.081±0.07	5.09±0.14	4.15±0.25	16.41±0.15
Papaya	30	83.49±0.33	13.12±0.59	0.17±0.02	4.61±0.01	11.54±1.2	85.47±0.02
Pine-apple	30	86.36±0.15	13.43±0.08	0.203±0.164	4.13±0.001	9.95±0.31	27.21±0.00

**Table 3: Microbial load of fruit juice served in cafés/restaurants in Bahir Dar city, 2006 – 2007.**

Sources of fruit juices	Sample size	Total plate Count Cfu/ml	Yeast CFU/ml	Mold CFU/ml
Mango	30	1.7 x 10 <sup>5</sup>	3.2 x 10 <sup>4</sup>	4 x 10 <sup>3</sup>
Avocado	30	3.1x10 <sup>7</sup>	4.5x10 <sup>4</sup>	4x10 <sup>4</sup>
Papaya	30	3.9x10 <sup>6</sup>	6.2x10 <sup>4</sup>	3.2x10 <sup>4</sup>
Pine-apple	30	1.5 x 10 <sup>5</sup>	5.0 x 10 <sup>3</sup>	3.2 x 10 <sup>4</sup>

In spite of the potential benefits offered by fruit juices, concerns over their safety and quality have been raised. An estimate of the gross nutrient composition of the juices is shown in Table 2 and there appear to be some differences between the juices.

The mean pH of fruit juices investigated in this study ranged from 4.13±0.001, as in the case of Pine-apple juices, to 5.09±0.14 in avocados juices. The pH of fruit juices is usually too low with good potentials of inhibiting the growth of pathogenic bacteria (Uzeh et al., 2007). Although some molds and yeasts could tolerate the acidity. However, the low acidity (i.e. higher pH) and viscosity of avocado, besides its nutrient content, makes it good medium for growth of microorganisms.

The highest titratable acidity was recorded in Pine-apple and the lowest in avocado juice, the highest moisture content was recorded in mango juice and the lowest in avocado juices (Table2). Water used for juice preparation can be a major source of microbial contaminants such as total coliforms, faecal coliforms, faecal Streptococci, etc. (Doyle, Beuchat & Montville, 2001).

Ascorbic acid (vitamin C) content of different fruit juices is shown in Table 2. The ascorbic acid content of commercial fruit juices is lost with respect to time and temperature during processing and storage (Biljana & Marija, 2009). The highest

content of ascorbic acid was observed in papaya juices and lower in mango juices in this study.

The Total soluble solids (TSS) contents are significantly influenced by the combined effect of stages of maturity and ripening conditions. The TSS content of pine-apple juices in this study was higher and lower in avocado juices (Table 2).

It is estimated that reducing sugar and total sugar content are increased with the advanced ripening of fruits. Mango juices contained the highest quantity of reducing sugar while avocado juices contained the lowest quantity at all times of observation (Table 2).

The mean total viable count was highest in avocado juices ( $3.1 \times 10^7$  cfu/ml) and lowest in Pine-apple juice ( $1.5 \times 10^5$  cfu/ml).

Yeast count was highest in Papaya juices ( $6.2 \times 10^4$  cfu/ml) and lowest in Pine-apple juices ( $5.0 \times 10^3$  cfu/ml). Papaya and Pine-apple juices recorded the lowest mold count ( $3.2 \times 10^4$  CfU/ml) while avocado juice recorded the highest ( $4.0 \times 10^4$  cfu/ml) (table 3).

To the authors' knowledge, there was no specification set for the permissible level of microbes in fruit juices being served in Ethiopia.

However, the recommended specifications for fruit juices served in the Gulf region suggests that the maximum count permitted for total colony count of coliforms, yeast and molds are  $1 \times 10^4$ , 100, and  $1.0 \times 10^3$  CFU/ml, respectively (Gulf Standards, 2002). On the basis of the gulf standards, it is clear that the colony counts of almost all the microbial groups in our fruit juices exceeded the standard by considerable margin. Because of the poor monitoring system in developing countries like Ethiopia, it is very difficult to evaluate how the consumer can be affected by taking fruit juices. In addition, conditions under which the juice was processed, stored, and/or served might have contributed to the betterment of the product. In fact, its low pH did not inhibit the growth of acid tolerant yeasts and molds (Doyle, Beuchat & Montville, 2001).

## CONCLUSION

The fruit juices investigated in this study had higher microbial load than the specifications set for fruit juices in some parts of the world.

On the basis of the gulf standards, it is clear that the colony counts of the microbial groups in our fruit juices exceeded the standard by considerable margin. These high counts, however, may pose hazard to the health of consumers especially if pathogenic species are present in the fruit juices to be consumed. Street vendors were mostly uninformed of good hygienic practices (GHP) and causes of diarrhea diseases which could increase the risk of street food contamination.

They were also unaware of food regulations as well as lacking supportive services such as knowledge on safe fruit juice preparation, waste disposal systems which enhance their ability to provide safe food.

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