



Asian Journal of Chemical and Pharmaceutical Research

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Research Article

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Effect of Storage Duration on Physico-Chemical and Microbiological Quality of Fruit Served in Bahir Dar City, North-West Ethiopia

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ABSTRACT

Fruit is one of the major dietary sources of various antioxidant phytochemicals for humans. For physico-chemical and microbiological safety of fruits, a total of 120 fruit samples were collected. The fruits investigated in this study area were avocado, papaya, pineapple, and mango. The mean pH of fruits in this study ranged from (4.01±0.008, 4.07±0.006, 4.10±0.003, 4.12±0.005 and 4.15±0.002) as in the case of Pine-apple juices, to (5.00±0.05, 5.01±0.18, 5.04±0.14, 5.06±0.17 and 5.15±0.12) in avocados juices extracted from fruit stored for zero, one, two, three and four days respectively. At zero day of storage, fruits contained the maximum (0.204±0.06%) titratable acidity in pine-apple fruit while the minimum (0.10±0.01%) at 4th day of storage in mango fruit. The maximum total soluble solids (13.77±0.59%) were observed in papaya fruit stored at four days. The maximum mean reducing sugar (12.06±1.61%) was recorded at fourth days of storage in papaya fruit. In fruits stored at zero days had the highest content of ascorbic acid (86.03±0.15 mg/100g) in papaya fruit whereas fruits stored at fourth days had the lowest values (8.01±0.23 mg/100g) in mango fruit during the storage. The mean total viable count was highest in papaya fruit stored at 4th days (4.6± 0.04 x 10⁶ cfu/ml) and lowest in pine-apple fruit stored at zero day (1.2± 0.01 x 10² cfu/ml). The amount of yeast had increased from 8.0± 0.02 x 10² cfu/ml in pine-apple fruit stored at zero days to 1.01± 0.02 x 10⁵ cfu/ml in papaya fruit stored for four days. Similarly, the amount of mold had also increased from 3.1± 0.04x10² cfu/ml in mango fruit stored at zero days to 6.1± 0.05 x10⁵ cfu/ml in papaya fruit stored for four days. Therefore, the effect of storage duration should be considered in fruit juice vending houses and there should be proper education of the vendors and consumers on proper storage procedure to minimize nutritional losses during storage and adequate monitoring of the quality of fruits during storage.

Keywords: Storage duration, physico-chemical quality, Microbiological safety, Avocado, Papaya, Pineapple, and Mango fruits, Bahir Dar

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Article History: Received 28 December 2015, Accepted 31 January 2016, Available Online 12 April 2016

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Manuscript ID: AJCPR2877



PAPER-QR CODE

Citation: Mekonen Tekliye. Effect of Storage Duration on Physico-Chemical and Microbiological Quality of Fruit Served in Bahir Dar City, North-West Ethiopia. *A. J. Chem. Pharm. Res.*, 2016, 4(1): 49-54.

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1. Introduction

Fruit is one of the major dietary sources of various antioxidant phytochemicals for humans. A study by Costin *et al.*, 1999 indicates their particular importance. Fruit, which supply a number of vital nutrients essential to body, is an important constituent of human diet today, more with much awareness towards health and fitness. Fruits prior to postharvest storage were involved in the ripening process. This process particularly occurs with climacteric fruit, whereby the ripening is assisted by respiration and is associated with ethylene production (Alexander and Grierson, 2002).

Storage problems are complicated by the fact that various cultivars may vary significantly in their storage performance (Golias *et al.*, 2008). The postharvest losses may depend on external and internal conditions, which affect the fruit firmness, juice content, weight loss, pH, soluble solids content (SSC), and other quality parameters (Tu *et al.*, 2000). Citrus fruits for example are known for their high ascorbic acid content. Compared with other major fruits and vegetable antioxidants, ascorbic acid is more susceptible to significant losses during post harvest handling and storage (Wilhelmina, 2005).

Physiological properties of a product determine the kind of storage condition to be adopted. Deterioration of fruits during storage depends largely on temperature and humidity. Refrigeration and open-air storage at ambient temperature are common practice used for short-term storage of fruits (Mijinyawa, 2010).

In terms of chemical composition fruits consist of water and dry matter. The dry matter contains considerable amounts of dry vitamins, acids, sugars, polysaccharides, pectin, cellulose, polyphenols and minerals. Non-enzymatic antioxidants, subjects on a growing list, are a large group of compounds coming from food. These include glutathione and vitamin E (Tocopherols), vitamin C (ascorbic acid), carotenoids and bioflavonoids. Vitamin C is a common natural substance, especially in plants (Bates *et al.*, 2001).

The consequence of the problems is increasing loss of fruit due to microbial spoilage and the existence of some human pathogens (Abadias, *et al.*, 2008). Fruit safety has emerged as an important global issue with international trade and public health implication. In response to the increasing food borne illness, governments all over the world are intensifying their effort to improve fruit safety (Sudershan, *et al.*, 2009).

Generally, the shelf life of fresh fruits is restricted by enzyme and microorganism activity. Spoilage of fruit and vegetable juices is mainly due to the presence of

osmophilic microflora (Tahiri *et al.*, 2006). These microfloras (yeasts) cause fermentation and produce a buttermilk-like off-flavour and moulds (Tournas *et al.*, 2006).

The physicochemical characteristics of fruit considered in quality assessment include pH, titratable acidity (TA), total soluble solids (°Brix), dry matter contents, ascorbic acid, reducing sugar and °Brix (sugar)/acid ratio. The present study was, therefore, conducted to evaluate effect of storage duration on Physico-chemical and microbiological quality of fruit served in Bahir Dar City, Ethiopia.

2. Materials and Methods

Collection of samples

The samples were purchased from ten randomly selected fruit juice vendors among over 25 fruit juice vending houses between May, 2006 and June, 2007 at Bahir Dar City. A total of 120 fruit samples (30 each of avocado, papaya, pineapple, and mango) were collected from fruit juice vending houses. The fruit investigated in this study area were avocado, papaya, pineapple, and mango. Samples (5kg each) of these fruit were separately collected in a basket and transported to Food Chemistry and Analysis Laboratory, Bahir Dar University, Institute of Technology. Then the fruit samples were stored at room temperature for 0, 1, 2, 3 and 4 days. Physico-chemical and microbiological qualities of the fruits were determined at 0, 1, 2, 3 and 4 days of storage.

Preparation of juice

The fruits were washed thoroughly with clean water to remove dirty material, dust, pesticide etc. Then peeled off and cut into pieces and remove the seeds. The juice was extracted using electric juicer. The extracted juice was strained properly. Immediately the experiments were run for some parameters in each case.

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

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

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

Where,

TA = Titratable 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-Dichloro phenolindo phenol 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 (Ben and Gaweda, 1985).

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).

Experimental design:

The study will have a completely randomized design (CRD) for the determination of effect of storage durations on the physico-chemical and microbiological quality of fruit. The design represents five levels for the storage durations.

3. Results and discussion

The data recorded on the effect of storage durations on different parameters were presented in Table 1 and 2. The study explored the physico-chemical and microbiological quality parameters so as to assess the quality of the fruits in terms of the storage system of the fruits used in Bahir Dar City fruit juice vending houses.

pH and titratable acidity

The mean pH of fruit investigated in this study ranged from (4.01 \pm 0.008, 4.07 \pm 0.006, 4.10 \pm 0.003, 4.12 \pm 0.005 and 4.15 \pm 0.002) as in the case of Pine-apple juices, to (5.00 \pm 0.05, 5.01 \pm 0.18, 5.04 \pm 0.14, 5.06 \pm 0.17 and 5.15 \pm 0.12) in avocados juices extracted from fruit stored for zero, one, two, three and four days respectively. There was a significant increase in pH during storage (Table 1). This might be due to decrease in titratable acidity, as acidity and pH are inversely proportional to each other (Bhardwaj et al, 2005). However, during the processing, a large part of the quality characteristics of the fresh fruits undergo remarkable changes which could reduce the nutritional value of the products. Moreover, the fresh juice may be stored for a long time in unfavorable conditions that lead to undesirable quality changes. The decreasing trend of titratable acidity during storage period was reported by Upadhyay et al. (1994) also found the similar results. According to them, acidity was reduced during storage growth on attainment of maturity and

ripening. At zero day of storage, fruits contained the maximum (0.204 \pm 0.06%) titratable acidity in pine-apple fruit while the minimum (0.10 \pm 0.01%) at 4th day of storage in mango fruit. It was decreased with the advancement of storage period.

The changes in titratable acidity are significantly affected by the rate of metabolism (Clarke et al., 2003) especially respiration, which consumed organic acid and thus decline acidity during storage (Ghafir et al., 2009).

Moisture content

The changes in moisture content of the fruit samples during storage at room temperature are presented in Table 1. At 4th day of storage, the highest moisture content (88.49 \pm 0.17%) was recorded in mango fruit while lowest (70.62 \pm 0.32%) in avocado fruit. The increase in moisture content with storage time might partly be due to metabolic water (Ladele, et al., 1984) had reported that the moisture content of fruits increased during storage.

Total soluble solids (Brix %)

The maximum total soluble solids (13.77 \pm 0.59%) were observed in papaya fruit stored at four days. The total soluble solids of fruits increased gradually with increasing the storage duration. The total soluble solids increased during storage (Riveria, 2005). The increase in TSS could be attributed to the breakdown of starch (Beaudry et al., 1989) into sugars (Crouch, 2003) or the hydrolysis of cell wall polysaccharides (Ben and Gaweda, 1985). Total soluble solids (TSS) contents are related directly to both the sugars and fruit acids as these are the main contributors; Pectins, glycosidic materials and the salts of metals (sodium, potassium, calcium etc.), when present, will also register a small but insignificant influence on the solids figure. The TSS content is significantly influenced by the combined effect of stages of maturity and ripening conditions. Late season fruits exhibited the highest total soluble solids content of the fruits. The TSS increased with gradual passage of storage time, which might be due to hydrolysis of polysaccharides. Similar results were also reported by Deka and Sethi (2001).

Reducing Sugar content

Reducing sugar is very important component for a processed product with respect to quality, shelf life, taste, and discoloration during storage. The results showed an increasing trend of reducing sugar with the progress of storage period. The maximum mean reducing sugar (12.06 \pm 1.61%) was recorded at fourth days of storage in papaya fruit (Table 1). The results of these investigations are in conformity with the finding of Wright and Whiteman (1955) who reported that reducing sugars tended to increase during storage.

Ascorbic Acid

In fruits stored at zero days had the highest content of ascorbic acid (86.03 \pm 0.15 mg/100g) in papaya fruit whereas fruits stored at fourth days had the lowest values (8.01 \pm 0.23 mg/100g) during the storage (Table 1). The ascorbic acid content in the fruits decreased significantly

during the ripening storage period similar to observations of Aydin and Kadioglu (2001). The decrease of vitamin C was approximately linear during the storage period. It is also noticeable that vitamin C change was negatively related to the storage period. Storage contributed to a significant reduction of vitamin C amount found in our study. The decomposition of vitamin C during storage of fruit is attributed to ascorbic acid degradation due to oxidation that can occur because of the presence of catalysts and oxidizing enzymes. The values for vitamin C presented negative trend with the soluble solids (Harris (1975)).

Total viable count

Many microorganisms are found in fruit juice and soft drinks as environmental or raw material contaminations either during their growing in fields, orchards, vineyards or greenhouse or during harvesting, post-harvest handling and distribution. But relatively few can grow within the acidic and low oxygen environment, yeast are the most significant group of microorganisms associated with spoilage of fruit juice and soft drinks. Bacteria, yeast and mould have an important impact on fruit quality after harvest, with fermentation and spoilage being common symptoms among fruits in storage (Kamal, et al., 2014). The statistical mean for effect of storage at room temperature on microbiological characteristics of fruit is presented in Table 2. The mean total viable count was highest in papaya fruit stored at 4th days ($4.6 \pm 0.04 \times 10^6$ cfu/ml) and lowest in pine-apple fruit stored at zero day ($1.2 \pm 0.01 \times 10^2$ cfu/ml). Steady but

gradual increase was observed for total viable bacteria count in all samples throughout the storage period. The total viable count of the fruit increased significantly with increased storage period (Kamal, et al., 2014).

Yeast and mold count

The counts of molds and yeast in this study are shown in (Table2). Fruit juices contain a micro-flora which is normally present on the surface of fruits during harvest and postharvest processing which include transport, storage, and processing (Tournas, et al., 2006). The amount of yeast had increased from $8.0 \pm 0.02 \times 10^2$ in pine-apple fruit stored at zero days to $1.01 \pm 0.02 \times 10^5$ in papaya fruit stored for four days. Similarly, the amount of mold had also increased from $3.1 \pm 0.04 \times 10^2$ in mango fruit stored at zero days to $6.1 \pm 0.05 \times 10^5$ in papaya fruit stored for four days. The number of mold colonies was lower than the number of yeasts; this is normal as is in agreement with Jay (1992) who reported that yeasts are associated with the spoilage of fruits due to their ability to grow faster than the molds.

The presence of yeast and molds in many of the fruits suggest that handling of the fruits and extraction of the juices methods may fall short of acceptable standards (Al-Jadah and Robinson, 2002). Gradual increase was observed in yeast and mold count. In addition the mean values of yeast and mold counts were slightly higher than mean value of total viable bacteria count at the end of the storage period.

Table 1: Physico-chemical properties of fruit stored at different storage duration at room temperature

Storage period	Sample type	Moisture Content (%)	Total soluble solids (Brix %)	Titratable acidity (%)	PH	Reducing Sugar (%)	Vitamin C (mg/100g)
Day 0	Mango	86.31±0.01	11.01±0.23	0.105±0.01	4.02±0.23	10.11±0.11	10.15±0.25
	Avocado	70.62±0.32	8.73±0.25	0.081±0.01	5.00±0.05	4.17±0.20	15.58±0.26
	Papaya	81.47±0.41	11.82±0.85	0.16±0.02	4.09±0.04	10.78±1.52	86.03±0.15
	Pine-apple	84.44±0.14	11.58±1.13	0.204±0.06	4.01±0.008	9.14±0.30	26.41±0.12
Day 1	Mango	87.21±0.02	11.22±0.76	0.104±0.03	4.01±0.40	10.33±0.18	9.65±0.11
	Avocado	71.81±0.14	8.93±0.07	0.080±0.05	5.01±0.18	4.18±0.47	15.46±0.12
	Papaya	82.27±0.43	12.12±0.78	0.15±0.05	4.11±0.01	11.32±1.40	85.00±0.17
	Pine-apple	85.74±0.11	12.14±0.02	0.204±0.02	4.07±0.006	9.66±0.44	25.11±0.55
Day 2	Mango	87.63±0.00	11.41±0.88	0.101±0.03	4.33±0.22	10.53±0.34	9.23±0.14
	Avocado	72.53±0.16	9.37±0.15	0.080±0.01	5.04±0.14	4.81±0.66	15.03±0.21
	Papaya	82.61±0.34	12.82±0.09	0.14±0.07	4.13±0.08	11.54±1.71	84.43±0.07
	Pine-apple	86.42±0.11	12.68±0.08	0.203±0.02	4.10±0.003	9.98±0.22	24.76±0.12
Day 3	Mango	88.17±0.13	12.11±1.21	0.101±0.04	4.40±0.13	10.62±0.16	8.32±0.13
	Avocado	73.33±0.15	10.15±0.54	0.078±0.02	5.06±0.17	5.07±0.27	14.76±0.14
	Papaya	83.59±0.30	13.11±0.51	0.13±0.04	4.23±0.03	11.86±1.32	81.60±0.02
	Pine-apple	86.81±0.18	12.94±0.17	0.202±0.09	4.12±0.005	10.50±0.53	22.42±0.15
Day 4	Mango	88.49±0.17	12.22±1.05	0.10±0.01	4.06±0.41	11.32±0.13	8.01±0.23
	Avocado	74.05±0.12	9.82.28±0.16	0.074±0.03	5.15±0.12	5.59±0.33	14.01±0.10
	Papaya	84.33±0.25	13.77±0.59	0.12±0.06	4.58±0.06	12.06±1.61	80.01±0.03
	Pine-apple	87.15±0.11	13.43±0.05	0.201±0.05	4.15±0.002	11.14±0.42	21.02±0.13

Table 2: Microbial load of fruit stored at different storage duration at room temperature

Storage period	Sample type	Total plate Count Cfu/ml	Yeast CFU/ml	Mold CFU/ml
Day 0	Mango	$4 \pm 0.03 \times 10^4$	$3.2 \pm 0.05 \times 10^3$	$3.9 \pm 0.07 \times 10^3$
	Avocado	$3.2 \pm 0.07 \times 10^3$	$2.2 \pm 0.02 \times 10^3$	$4.0 \pm 0.03 \times 10^4$
	Papaya	$4.1 \pm 0.03 \times 10^3$	$3.40 \pm 0.03 \times 10^3$	$3.1 \pm 0.04 \times 10^2$

Day 1	Pine-apple	$1.2 \pm 0.01 \times 10^2$	$1.02 \pm 0.03 \times 10^3$	$3.1 \pm 0.06 \times 10^4$
	Mango	$3.26 \pm 0.05 \times 10^5$	$3.84 \pm 0.06 \times 10^3$	$4.1 \pm 0.02 \times 10^3$
	Avocado	$2.7 \pm 0.06 \times 10^4$	$3.4 \pm 0.04 \times 10^4$	$6.1 \pm 0.06 \times 10^4$
	Papaya	$3.1 \pm 0.04 \times 10^6$	$5.00 \pm 0.04 \times 10^4$	$4.0 \pm 0.07 \times 10^2$
Day 2	Pine-apple	$1.4 \pm 0.02 \times 10^3$	$1.2 \pm 0.03 \times 10^3$	$4.3 \pm 0.05 \times 10^4$
	Mango	$2.26 \pm 0.01 \times 10^6$	$3.0 \pm 0.04 \times 10^4$	$3.2 \pm 0.07 \times 10^4$
	Avocado	$1.5 \pm 0.01 \times 10^6$	$4.1 \pm 0.02 \times 10^4$	$4.9 \pm 0.01 \times 10^5$
	Papaya	$2.5 \pm 0.03 \times 10^5$	$3.2 \pm 0.04 \times 10^6$	$3.2 \pm 0.04 \times 10^3$
Day 3	Pine-apple	$2.3 \pm 0.06 \times 10^4$	$1.6 \pm 0.01 \times 10^3$	$6.4 \pm 0.03 \times 10^4$
	Mango	$1.6 \pm 0.02 \times 10^6$	$3.3 \pm 0.04 \times 10^4$	$4.2 \pm 0.05 \times 10^4$
	Avocado	$1.2 \pm 0.04 \times 10^5$	$4.5 \pm 0.02 \times 10^4$	$5.3 \pm 0.07 \times 10^5$
	Papaya	$3.6 \pm 0.07 \times 10^4$	$2.01 \pm 0.6 \times 10^7$	$3.6 \pm 0.02 \times 10^4$
Day 4	Pine-apple	$1.54 \pm 0.01 \times 10^4$	$2.9 \pm 0.03 \times 10^3$	$7.2 \pm 0.02 \times 10^4$
	Mango	$1.4 \pm 0.03 \times 10^5$	$2.57 \pm 0.02 \times 10^5$	$6.1 \pm 0.05 \times 10^5$
	Avocado	$1.8 \pm 0.02 \times 10^4$	$5.3 \pm 0.01 \times 10^5$	$5.9 \pm 0.04 \times 10^4$
	Papaya	$4.6 \pm 0.04 \times 10^6$	$1.01 \pm 0.02 \times 10^8$	$6.4 \pm 0.07 \times 10^4$
	Pine-apple	$1.4 \pm 0.02 \times 10^4$	$3.5 \pm 0.04 \times 10^3$	$3.5 \pm 0.04 \times 10^3$

4. Conclusion

During four days of storage duration of fruits at room temperature following conclusions were found: physico-chemical and microbiological quality of fruits changes significantly. Important has been generated on nutritional properties of the samples of fruits as they change with storage time. It can be concluded that these qualities generally change with time. The study revealed that there were continuous decreases in the quality parameters such as ascorbic acid and titratable acidity, while reducing sugar, total soluble solids, and moisture content increased with increase in storage period. The present research has shown that the storage of the fruits at room temperature cause significant change on the microbial load (total viable count (TVC), yeast and mould of the fruits increased over the storage period. Therefore, the effect of storage duration should be considered in fruit juice vending houses and there should be proper education of the vendors and consumers on proper storage procedure to minimize nutritional losses during storage and adequate monitoring of the quality of fruits during storage.

5. Acknowledgments

The authors are gratefully acknowledges the financial support provided by Bahir Dar University, Bahir Dar Institute of Technology.

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