EFFECTS OF NIGERIAN MARKET STORAGE CONDITIONS ON ASCORBIC ACID CONTENTS OF SELECTED TETRAPAK PACKAGED CITRUS FRUIT JUICE

Ojo Rotimi Johnson[1], Abaya Jemimah Yetu[1], Akintayo Christopher Oloruntoba[2] and Seriki Samuel[2]
[1]Department of Biochemistry, Faculty of Science and Technology, Bingham University Karu Nasarawa state, Nigeria
[2]Department of Physiology, College of Medicine Bingham University Karu Nasarawa state, Nigeria
E-Mail: saintlevites@yahoo.com

ABSTRACT

The effect of Nigerian market storage conditions on ascorbic acid, titratable acidity and pH values of selected tetrapak packaged citrus fruit juice in Nigerian markets were studied with increasing shelf life (storage time). Six different brands of tetrapak packaged citrus fruit juice were selected for this study after a market survey of tetrapak packaged citrus fruit juice commercially available in Nigerian markets. The selection was made to reflect brands that are readily available in the markets and frequently consumed. Selected samples have the same packaging materials, close manufacturing date and similar compositions. A typical Nigerian market condition was imitated during the research with the average maximum and minimum temperature of the storage being 28 ±5°C and Relative humidity of 75 ± 5% with less ventilated room. The analyses of the samples were carried out from 3 months to 10 months from the day of production. The results showed that there was gradual decrease in ascorbic acid contents and pH values with increase in storage period irrespective of the brands but at different rate depending on the brand while there was increase in total titratable acidity after storage for 10 months at ambient room temperature. Therefore, the effect of market storage conditions should be considered in production process and there should be proper education of the traders and consumers on proper storage procedure to minimize the ascorbic acid loss during storage and finally, nutritional labels must be printed on the container showing: best before date, storage conditions and nutritional facts to ensures adequate monitoring of the quality.

Keyword: ascorbic acid, tetrapak, citrus, Nigerian market, titratable acidity, storage.

INTRODUCTION

The richest natural sources of ascorbic acid (vitamin C) are fruits and vegetables, the vitamin content of which has been shown to be nutritionally superior when compared to many cereal and leguminous crops (FAO, 1992).

Fruits and vegetables supply more than 90% of vitamin C in human diets (Erentuk *et al*., 2005). In Nigeria however, vegetables cannot be relied upon to supply ascorbic acid since almost all vegetables are thoroughly cooked before consumption, thereby destroying the heat-labile ascorbic acid content but fruits are however consumed raw, thereby conserving the ascorbic acid content.

A high recommendation of daily intake for humans has been suggested, since stress is known to increase the requirement for ascorbic acid (Olivier, 2004). Besides its vitamin action, Ascorbic acid is valuable for its antioxidant effect, stimulation of the immune system and other health benefits. Ascorbic acid also reduces the risk of arteriosclerosis, cardiovascular diseases and some forms of cancer. Also, dehydroascorbic acid (DHA), an oxidation product, also exhibits biological activity (Lee and Adel, 2000). Ascorbic acid also inhibits reactive oxygen species which are generated in cells during oxidative metabolism and their harmful effects (Minnunni *et al.*, 1992; Knekt *et al.*, 1991; Ferraroni *et al.*, 1994; Incerpis *et al.*, 2007). Reactive oxygen species and/or free radicals when they are uninhibited cause damage to the cells by snatching electrons from their DNA, proteins, carbohydrates or unsaturated fatty acids resulting in change of structure and functions. Based on available biochemical, clinical and epidemiological studies, the recommended daily acceptance for ascorbic acid is suggested to be 100-120 mg/day to achieve cellular saturation and reduction in risk of some ailments in healthy individuals (Klimezak *et al*., 2007), since ascorbic acid is not synthesized in humans; this requirement must be satisfied through dietary intake.

Ascorbic acid is highly sensitive to various modes of deterioration. The main factors that can affect ascorbic acid loss include temperature, salt and sugar concentration, pH, oxygen, light, metal catalysts, initial concentration of ascorbic acid, the ratio of ascorbic acid to dehydroascorbic acid, microbial load and protection by the container (Zerdin *et al*., 2003). Ascorbic acid added to beverages is readily oxidized and lost during storage, at a rate depending on the conditions of storage. This fact is of great importance to the consumer who must know how to store the beverages and when to consume them in order to get the maximum benefit of vitamin C content (Kabasakalis *et al*., 2000). This is because prolonged storage and careless handling can destroy the nutrient content and other quality of the juice (Serrano *et al.*, 2006). Determination of the nutrient content of foods is extremely important because there is a relationship between dietary intake and human health (Gokmen *et al*., 2000).

This study was therefore undertaken to assess the effects of Nigerian market storage conditions on ascorbic acid contents of selected tetrapak packaged citrus fruit juice in northern Nigeria with increasing shelf life (storage time).

MATERIALS AND METHODS

Experiment site

The survey and the analyses were carried out for 11 months in the chosen markets and Department of Biochemistry, Bingham University Karu Nasarawa state, Nigeria, respectively between August, 2011 and June, 2012.

Survey and collection of samples

A survey of tetrapak packaged citrus fruit juice commercially available in Nigerian markets was carried out in Nasarawa state and Nyanya in Federal capital terriotory, Abuja, all located in Northern part of the country.

Six different brands of tetrapak packaged citrus fruit juice were selected for this study. The selection was made to reflect brands that are readily available in the market and frequently consumed. Selected samples have the same packaging materials, close manufacturing date and similar compositions. These samples are: 5-Alive citrust bust, Happy Hour Orange, Caprisone, Chivital, Dansa and Fumman.

Storage conditions of the samples

A typical Nigerian market condition was imitated during the research with the average maximum and minimum temperature of the storage being 28 ±5°C and Relative humidity of 75 ± 5% with less ventilated room.

Chemicals and reagents

Potassium iodide (BDH, England); sodium hydroxide (Hopkins and Williams Ltd, England), Iodine (Burgoyne, India), soluble starch (BDH, England), Vitamin C standard (cartivalues, USA), 1-Butanol (BDH, England); Thiobarbituric acid (Kem Light, India); Phenolphtalein (Qualikem, India).

Analytical procedure

Analyses were done in triplicates. The analyses of the samples were carried out from 3 months to 10 months from the date of production which is usually their shelf life. Safety precautions and good quality assurance procedures were taken to ensure reproducibility of the results, as a result of this between six to eight samples of each product was collected. After each determination, each open sample was discarded and not used for the next test. The ascorbic acid content was determined by redox titration method as described by Ajibola *et al*., (2009). The pH of the sample was determined by the method described by Rangana (1979) and Total titratable acidity by the methods and calculation of both Moneruzzaman *et al*. (2009) and Tsige *et al*. (2008).

Statistical analysis

The data are expressed as mean ± SD. Readings within a group were compared using the one-way ANOVA analysis and readings between groups were compared using the Independent sample test. Statistical analysis was performed using SPSS (Version 17). A level of $p < 0.05$ was considered to be significant.

Table-1. Effect of storage period on ascorbic acid contents of citrus fruit juice.

Samples	**3 Months** (Mg/100ml)	**6 Months** (Mg/100ml)	**9 Months** (Mg/100ml)	**10 Months** (Mg/100ml)	**% Decrease in ascorbic acid contents between 3 and 10 months after production**
Caprisonne	7.66 ± 0.13	6.52 ± 0.08	6.40± 0.08	6.19± $0.04^{b,f}$	19.19
Chivita	9.57 ±0.12	7.84 ± 0.12	7.33± 0.16	7.02 ± $0.12^{a,f}$	26.65
5-Alive	9.78± 0.08	8.13 ± 0.04	7.60 ± 0.11	7.20 ±$0.13^{a,f}$	26.38
Happy Hour	4.19 ± 0.06	3.67 ± 0.05	3.55 ± 0.04	3.29 ± $0.09^{d,f}$	21.48
Dansa	5.45 ± 0.18	5.08 ± 0.02	4.87 ± 0.08	4.72 ± $0.13^{c,f}$	13.39
Fumman	3.67± 0.21	3.47 ± 0.08	3.20± 0.12	2.94 ± $0.16^{d,f}$	19.89

Values are expressed as Mean ± Standard deviation, n = 3
Values with different superscript in the same column are significantly different at $p<0.05$
f = significant difference at $p<0.05$ compare to 3 months

According to the results of Table-1, the storage period had significant influence on the ascorbic acid content of the fruit juice and there was positive correlation was between ascorbic acid content and the storage temperature. Storage temperature had significant effect on the ascorbic acid content. That is both storage time and temperature had a significant the ascorbic acid content of the juice after 10 months of storage.

Table-2. Effect of storage period on Titratable acidity of citrus fruit juice.

Samples	**3 Months** (%)	**6 Months** (%)	**9 Months** (%)	**10 Months** (%)
Caprisonne	0.023 ± 0.007	0.029 ± 0.007	0.032 ± 0.007	0.037 ±0.006[a,f]
Chivita	0.081± 0.008	0.085 ± 0.002	0.093 ± 0.009	0.096 ± 0.008[c,f]
5-Alive	0.093 ± 0.010	0.095 ± 0.007	0.101 ± 0.008	0.103 ± 0.007[d,f]
Happy Hour	0.044 ± 0.009	0.046 ± 0.006	0.050 ± 0.009	0.055 ± 0.010[b,f]
Dansa	0.079 ± 0.008	0.084 ± 0.009	0.087 ± 0.008	0.094 ± 0.002[c,f]
Fumman	0.091 ± 0.008	0.096 ± 0.0014	0.097 ± 0.007	0.101 ± 0.008[d,f]

Values are expressed as Mean ± Standard deviation, n = 3
Values with different superscript in the same column are significantly different at p<0.05
f = significant difference at p<0.05 compare to 3 months

Temperature and storage period significantly affected the concentration of total titratable acidity Table-2 of the fruit juice and there was negative correlation between the total titratable acidity and the storage temperature. Storage temperature had significant effect on the titratable acidity. That is both storage time and temperature had a significant the effect on the total acidity of the juice after 10 months of storage.

Table-3. Effect of storage period on pH values of citrus fruit juice.

Samples	**3 Months**	**6 Months**	**9 Months**	**10 Months**	**% Decrease in pH values between 3 and 10 months after production**
Caprisonne	4.43 ±0.04	4.35 ± 0.05	4.26 ± 0.04	4.18 ± 0.03	5.64
Chivita	4.26 ±0.05	4.15 ± 0.04	4.04 ± 0.09	3.89 ± 0.07	8.67
5-Alive	4.27 ±0.08	4.20 ± 0.04	4.09 ± 0.13	3.92 ± 0.14	8.20
Happy Hour	4.33 ±0.07	4.18 ± 0.08	4.07 ± 0.12	3.96. ± 0.07	8.54
Dansa	4.29 ±0.09	4.18 ± 0.05	4.09 ± 0.09	3.93 ± 0.13	8.62
Fumman	4.58 ±0.07	4.42 ± 0.09	4.30 ± 0.05	4.22 ± 0.06	7.86

Values are expressed as Mean ± Standard deviation, n = 3

The storage period had significant influence on the pH of the fruit juice and there was positive correlation between pH and the storage temperature. Storage temperature had significant effect on the pH. That is both storage time and temperature had a significant on the pH of the juice after 10 months of storage.

DISCUSSIONS

Table-1, showed the effect of storage on ascorbic acid contents of selected tetrapak packaged citrus fruit juice with increase in shelf life(storage time). The ascorbic acid contents of all samples decreased during storage, this was similar to what Weichmann (1987) reported that ascorbic acid content of stored produce generally decrease more rapidly at higher storage temperature since it is thermo labile and also with the established fact that ascorbic acid degrades steadily during prolonged storage (Murcia *et al*., 2000), even in frozen products (Rickman *et al*., 2007). Greater loss of ascorbic acid was observed at the initial storage period. The loss may be due to the inability of the packaging materials to act as effective barrier against light, oxygen, temperature and other environmental factors (Gordon, 2006). This rapid decrease at the beginning of the storage can also be attributed to the immediate reaction of ascorbic acid with the dissolved oxygen (Polydera *et al*., 2003) while anaerobic degradation of ascorbic acid may be responsible for other loss during storage, which is especially observed in thermally preserved juices (Burdurly *et al*., 2006). The results showed that storage temperature and the environment in which the juice is stored affects its ascorbic acid content significantly (Table-1) resulting in ascorbic acid loss. This is consistent with reports that, climate, especially temperature affects ascorbic acid (vitamin C) level (Padayatty *et al*., 2003). The concentration of vitamin C decreased faster in the samples and the same pattern was observed throughout the storage period even in unopened containers (Li *et al*., 1989; Lee and Coates, 1999; Johnston and Bowling, 2002; Kabasakalis *et al*., 2000). The retention of vitamin C is often used as an estimate for the overall nutrient retention of food products because it is by far the least stable nutrient; it is highly sensitive to oxidation and leach into water-soluble media during storage (Davey *et al*., 2000; Franke *et al*., 2004). Ascorbic acid (Vitamin C) also, acts as chain breaking scavenger for peroxy radicals and also, donates hydrogen atom to the vitamin E derived phenolate radical, thus regenerating its activity (Buring and Hennekens, 1997; Lata, 2002) therefore decrease in its contents will affect its activities.

Titratable acidity is a measure of the total acid present in a juice. The predominant acid naturally occurring in orange juice is citric acid. There are also small amounts of malic acid and tartaric acid. The total acid may not be measured by pH because the acids concerned are “weak acids” and not completely ionized. The acid content was therefore measured using a titration with sodium hydroxide. The titratable acidity of all samples increased with storage time Table-2. The total titratable acidity content was significantly influenced by the storage conditions Table-2. Storage duration made juice to be more acidic. The maximum total titratable acidity (0.103 percent) was found in 5-Alive while it was minimum (0.055 percent) in Caprisonne under the control treatment. The increase in titratable acidity indicated fermentation of orange juice.

Orange juices analyzed that it had pH values between 3.89 and 4.22. The decrease in pH (Table-3) was related to deterioration of fruit characteristics (María Gil *et al*., 2006). The biochemical reactions occurring over the storage period together with microbial action in all fruit juices resulted in pH changes observed, this was observed by Teniola, (2000) in his research on ogi that the variation in pH levels noticed in stored product may be attributed to the difference in the trend of activities of microorganisms. The reduction in the pH (Table-3) of all samples which implies that the samples turned more acidic with increase in storage period may be attributed to increase in total acids which increased the hydrogen ion concentration (Pantastico, 1975). This result showed that extension in storage period at ambient temperature (28±5ºC) leads to decrease in pH. The low pH may give the juice a good potential of inhibiting the growth of pathogenic bacteria (Jay, 2000; Hatcher *et al*., 1992) although some moulds, yeasts and pathogenic microorganism could tolerate this and cause spoilage (Brown and Booth, 1991).

CONCLUSIONS

It is an established fact that orange juice quality depends on the raw material, processing conditions, storage conditions and packaging materials but storage temperature was the prime limiting factor for shelf life of this product. The results of this study showed that there were gradual decreases in ascorbic acid contents and pH values with increase in storage period irrespective of the brands but at different rate depending on the brand while there was increase in total titratable acidity after storage for 10 months at ambient room temperature 28 $\pm 5^{o}$C. Therefore, the effect of market storage conditions should be considered in production process and there should be proper education of the traders and consumers on proper storage procedure to minimize the ascorbic acid loss during storage and finally, nutritional labels must be printed on the container showing: best before date, storage conditions and nutritional facts to ensures adequate monitoring of the quality during storage.

REFERENCES

Ajibola V.O., Babatunde O.A. and Suleiman S. 2009. The Effect of Storage Method on the Vitamin C Content in Some Tropical Fruit Juices. Trends in Applied Sciences Research. 4: 79-84.

Brown M.H. and Booth I.R. 1991. Accidulants and Low pH: Food Preservative. In: Russell, N.J. and G.W. Gould (Eds.). Critical Reviews in Food Science and Nutrition. Blackie Glasgow and London 1st Fermentation. 28: 349-400.

Biljana R. C. and Marija R. J. 2009. Effect of preservation method and storage condition on ascorbic acid loss in beverages.

Burdurly H. S., Koca N. and Karadeniz F. 2006. Degradation of vitamin C in citrus juice concentrates during storage. Journal of Food Engineering. 74: 211-216.

Buring J. and Hannekens C. 1997. Antioxidant vitamins and cardiovascular disease. Nutr. Rev. 55: 553-560.

Davey M.W., Van Montagu M., Inze D., Sanmartin M., Kanellis A.and Smirnoff N. 2000. Plant L-ascorbic acid: Chemistry, function, metabolism, bioavailability and effects of processing. J. Sci. Food Agric. 80: 825-860.

Erentuk S, Gualaboglu M.S. and Gultekin S. 2005. The effects of cutting and drying medium on the vitamin C content of rosehip during drying. Journal of Food Engineering. 68: 513-518.

FAO. 1992. Food and Agricultural Organization Report: Use of Fertilizer in Nigeria. Paper 8. pp. 63-77.

Ferraroni M., Heinonen O.P., Albanes D., Heinonen M., Rikkala E. and Teppo L. 1994. Selected micronutrients intake and the risk of colorectal cancer. Br. J. Cancer. 70: 1150-1155.

Franke A.A., Custer L.J., Arakaki C. and Murphy S.P. 2004. Vitamin C and flavonoid levels of fruits P.S. and vegetables consumed in Hawaii. J. Food. Compos. Anal. 17: 1-35.

Gokmen V., Kahraman N., Demir N. and Acar J. 2000. Enzymatically validated liquid chromatographic method for the determination of ascorbic and dehydroascorbic acids in fruit and vegetables. Journal of Chromatography A. 881: 309-316.

Gordon L. R. 2006. Food Packaging Principle and Practice (2nd Ed). Taylor and Francis Group, New York, USA. pp. 202-203.

Hatcher W.S., Weihe J.L., Splittstoesser D.F., Hill E.C. and Parish M.E. 1992. Fruit beverages. In: Compendium of Methods for the Microbiological Examination of Food. Vanderzant, C. and Splittstoesser, D.F. (Eds).

Incerpis S., Fiore A.M., De Vito P. and Pedersen J.Z. 2007. Involvement of plasma membrane redox systems in hormone action. J. Pharm Pharmacol. 59: 1711-1720.

Jay J.M. 2000. Modern Food Microbiology, 6th Ed. Aspen Publishers, Inc., Maryland.

Johnston C.S. and Bowling D.L. 2002. Stability of ascorbic acid in commercially available orange juices. J. American Dietetic Assoc. 102: 525-529.

Kabasakalis V., Siopidou D. and Moshatou E. 2000. Ascorbic acid content of commercial fruit juices and its rate of loss upon storage. Food Chem. 70: 325-328.

Kabasakalis V., Siopidou D. and Moshatou E. 2000. Ascorbic acid content of commercial fruit juices and its rate of loss upon storage. Food Chemistry. 70: 325-328.

Klimezak I., Malecka M., Szlahta M. and Gliszezynska-Swiglo A. 2007. Effect of storage on the content of polyphenols, vitamin C and the antioxidant activity of orange juices. Journal of Food Composition and Analysis. 20: 313-322.

Knekt P., Jarvinen R., Seppanen R., Rissanen A. and Aromaa A. 1991. Dietary antioxidant and the risk of lung cancer. Am. J. Epidemiol. 1134: 471-479.

Lata B. 2002. The role of antioxidants and the factors influencing their content in fruit. Zeszyty-Naukowa. 10: 7-18.

Lee H.S. and Coates G.A. 1999. Vitamin C in frozen fresh squeezed unpasteurized polyethylene bottle orange juice: a storage study. Food Chem. 65: 165-168.

Lee S. K. and Adel A. K. 2000. Preharvest and post harvest factors influencing vitamin C content of horticultural crops. Post harvest Biology and Technology. 20: 207-220.

Li Z., Alli I. and Kermasha S. 1989. Use of acidification, low temperature and sorbates for storage of orange juice. J. Food Sci. 54: 674-678.

María G. I., Encarna A. and Adel K. A. 2006. Quality changes and nutrient retention in fresh-cut versus whole fruits during storage. J. Agric. Food Chem. 54: 4284-4296.

Minnunni M., Wolleb U., Mueller O., Pfeifer A. and Aeschbacher H.U. 1992. Natural antioxidants as inhibitors of oxygen species induced mutagenicity. Mutat. Res. 269: 193-200.

Moneruzzaman K.M., Hossain A.B.M S., Sani W., Saifuddin M. and Alenazi M. 2009. Effect of harvesting and storage conditions on the post harvest quality of tomato (*Lycopersicon esculentum* Mill) cv. Roma VF. Australian Journal of Crop Science. 3(2): 113-112.

Murcia M.A., L´opez-Ayerra B., Martinez-Tom´e M., Vera A.M. and Garc´ıa-Carmona F. 2000. Evolution of ascorbic acid and peroxidase during industrial processing of broccoli. J. Sci. Food Agric. 80: 1882-1886.

Padayatty S.J., Katz A., Wang Y., Eck P. and Kwon O. 2003. Vitamin C as an antioxidant: Evaluation of its role in disease prevention. J. Am. Coll. Nutr. 22: 18-35.

Pantatisco E.R.B. 1975. Post Harvest Physiology Handling of Tropical and Subtropical Fruits and Vegetables. AVI publishing company inc., West port, Conneticut. pp. 67-71.

Polydera A.C., Stoforos N.G. and Taoukis. 2003. Comparative shelf life study and vitamin C loss kinetics in pasteurized and high pressure processed reconstituted orange juice. Journal of Food Engineering. 60: 21-23.

Rangana. 1979. Manual analysis of fruits and vegetables product. Tata McGraw Hill Co. Ltd., New Delhi, India. 2-95: 634.

Rickman J.C., Barrett D.M. and Bruhn C.M. 2007. Review: Nutritional comparison of fresh, frozen and canned fruits and vegetables (Part 1). Vitamins C and B and phenolic compounds. J. Sci. Food. Agric. 87: 930-944.

Serrano M., Valverde J.M., Guillen F.,Castillo S., Martinez-Ronero D. and Valero D. 2006. Use of Aloe Vera gel coating preserves the functional properties of Table grapes. J. Agric. Food Chem. 54: 3882-3888.

Teniola O.D. 1990. Development of starter culture for the production and nutritional improvement of Ogi. Ph.D. Thesis, Botany and Microbiology Department, University of Ibadan, Nigeria.

Tsige K., Tsegaye G. and Ketema B. 2008. Microbiological safety of fruit juices served in cafes/restaurants, jimma town, southwest Ethiopia Ethiop. J. Health Sci. 18(3): 98-100.

Weichmann J. 1987. Post Harvest Physiology of Vegetables. Marcel Dekker Inc., New York, USA. pp. 42-44.

Zerdin K., Michael L. R. and Vermue J. 2003. The vitamin C content of orange juice packed in an oxygen scavenger material. Food Chemistry. 82: 387-395.