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Effect of Storage Temperature and Time on the Vitamin C Contents of Selected Fruits and Vegetables

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Abstract. The vitamin C contents of 5 fruits and 7 vegetables, as a whole and in diced form, were determined by HPLC during cold storage. Results showed a decrease in vitamin C contents during 15 days refrigeration (7 °C) as well as freezing at -20 °C. It was found that fruits are more stable than vegetables since the rate of degradation of vitamin C was higher in vegetables as compared to fruits, either during freezing or refrigeration. During 15 days freezing, fruits showed a decrease of 41.05 - 51.44%, whereas, this loss augmented to 54.12 – 89.10% in vegetables. In addition to this, it was also observed that fruits and vegetables which have peels are less vulnerable to vitamin C degradation; the ratio of degradation of vitamin C in all the fruits studied and potato was not more than 51.44%. In fruits, apple was more susceptible and in vegetables, potato was more stable to vitamin C degradation.

Keywords: fruits,vegetables, vitamin C, refrigeration, freezing

Introduction

Vitamin C is a water soluble vitamin, essential for the synthesis of collagen and inter-cellular material. Several substances have vitamin C activity, notably ascorbic acid and its sodium and calcium salts. Daily dietary intake of about 30-100 mg of vitamin C has been recommended for adults (Sweetman, 2007). Vitamin C functions in a number of biochemical reactions, mostly involving oxidation. Thus, it is required for and facilitates the conversion of certain proline residue in collagen to hydroxyproline in the course of collagen synthesis (Myllyla *et al.,* 1978), the oxidation of lysine side chains in proteins to provide hydroxytrimethylysine for carnitine synthesis (Hulse *et al.,* 1978), the synthesis of steroids by the adrenal cortex (Deana *et al.,* 1975), the conversion of folic acid to folinic acid etc. (Stokes *et al.,* 1975).

Humans are unable to form their own vitamin C, so a dietary source is necessary. Most dietary vitamin C is obtained from fruit and vegetable sources. Only small amounts are present in milk and animal tissues. Relatively rich sources include rose hips, black currant, citrus fruits, leafy vegetables, tomato and potato, green and red pepper etc. Vitamin C is readily destroyed during cooking. Considerable losses may also occur during storage (Sweetman, 2007). Studies indicate that vitamin C is by far the least stable nutrient during processing due to its sensitivity to oxidation and leaching into water soluble media during processing, storage and cooking of fresh, frozen and canned fruits and vegetables (Franke *et al.,* 2004; Lathrop and Leung, 1980). In various earlier studies, the effect of processing and storage on the vitamin C contents in a variety of fruit products had been determined (Gil-Izaquierdo *et al.,* 2002; Lima *et al.,* 1999; de-Dios and Viteri 1991); loss vitamin C after six days at 5 °C was noted to be 5- 25 % in various fruits and vegetables (Gil *et al.,* 2006). Vitamin C content of lemon juice decreased to about 36% of the initial value over a period of 12 weeks at various storage conditions (Abbasi and Niakousari, 2007). No considerable loss in vitamin C content of green beans occurred during blanching and freezing processes (Martins and Silva, 2003).

There has been an increasing demand for fresh cut fruits and vegetables for convenience as ready-to-eat products, along with their nutritional values requisite for healthy living (Martin *et al.,* 2002; Liu *et al.,* 2000; Block *et al.,* 1992). A major benefit of greater intake of fruits and vegetables may be the increased consumption of vitamins and their antioxidant activity which may reduce the risk of cancer, heart diseases as well as prevention of degenerative diseases (Grassmann *et al.,* 2002; Rimm *et al.,* 1996; Gaziano and Hennekens, 1993; Tee, 1992; Doll 1990). Among all other valuable nutrients like phenols, carotenoids and vitamins etc., vitamin C is the most perishable (Gil *et al.,* 2006). Degradation of vitamin C takes both aerobic and anaerobic pathways and depends upon many factors such as oxygen, heat, light, storage temperature and time (Uddin *et al.,* 2002; Nunes *et al.,* 1998; Sedas *et al.,* 1994). Many fruits and vegetables contain over 90% water and once harvested they begin to undergo higher rates of respiration, resulting in moisture loss and quality deterioration. Harvesting separates the source of energy required by the fruits and vegetables for the above mentioned activities resulting in reduction of their nutritional values. Storage and

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processing technologies are used to transform these fruits and vegetables into safe, delicious and stable products (Fennema, 1982). Refrigeration slows down degradation rate and lengthens the shelf life of fruits and vegetables (Rickman *et al.,* 2007).

The primary objective of the present study was to determine the vitamin C content of various fruits and vegetables (whole and diced) at various storage temperatures.

Materials and Methods

The study comprises of two parts; in the first part, whole and diced fruits and vegetables were stored at various temperatures for a definite period of time and in the 2nd part, the diced fruits and vegetables were stored at fixed temperature for varying periods and vitamin C content was determined at intervals.

Materials. Fruits and vegetables including mango, grapefruit, papaya, lemon, apple, spinach, potato, bitter gourd, capsicum and broccoli were purchased from the local market of Lahore city.

Chemicals and reagents. Ascorbic acid (BDH, England), 1-octanesulphonic acid sodium salt (Fisher Scientific, UK), citric acid (Scharlau, Spain), di- sodium EDTA, NaF, HPLC grade methanol (Fisher Scientific, UK), tri-ethanolamine (Merck, Germany), glacial acetic acid (Merck, Germany) were used in the study.

Preparation of samples. All fruits and vegetables under study were grouped into four lots and stored at various temperatures. Whole and freshly diced fruits and vegetables were analysed for vitamin C content at room temperature, at refrigeration temperature (7 °C) and at freezing temperature (-20 °C). One lot was immediately analyzed for vitamin C content and other lots (whole and diced) were stored at the above mentioned temperatures. In the 2nd part of the study pre-weighed diced fruits and vegetables were refrigerated at 7 °C and vitamin C content was analyzed for three weeks at one week intervals.

Extraction and analysis. The procedure was based on the method of Zapata and Dufour (1992) with some modifications. To 10 g of sample, 10 mL of extraction solution (0.1 M citric acid, 0.05% EDTA di-sodium salt, 5% methanol and 4 mM NaF) was added. The contents were homogenized on an ice bath and filtered through Whatmann filter paper 1. The filtrate was centrifuged at 10, 000 rpm for 10 min at 3 °C. pH of the filtrate was adjusted to 2.35-2.40 and passed through an activated Sep-Pak C_{18} cartridge and then filtered through a 0.22 µm filter paper. After 35 min, analysis was performed with HPLC (Perkin Elmer) with Total Chrom Workstation (TCW) software and Perkin Elmer Series 200 UV/Vis detector, a reverse phase C_{18} nucleosil column (25 cm x 4.6 mm ID, 5 µm particles); flow rate was 1 mL/min. Mobile phase was prepared by dissolving 1.1 g of 1-octanesulphonic acid sodium salt in 800 mL of water and by adding 24 mL of glacial acetic acid, 5 mL of tri-ethanolamine and 150 mL of methanol and total volume was made up to 1000 mL with distilled water. pH of the mobile phase was adjusted to 3.61 ± 0.1 with acetic acid or tri-ethanol amine. The detection was made at 348 nm for de-hydroascorbic acid (DHAA) and at 261 for ascorbic acid (AA). Vitamin C content (DHHA+AA) was expressed in mg/100 g of fresh weight.

Results and Discussion

Vitamin C content of fruits and vegetables remained stable during storage either in the form of ascorbic acid or de-hydroascorbic acid for a particular period of time at specific temperature. Beyond that time and temperature, vitamin C content drastically decreased though the ratio of degradation varied in various fruits and vegetables.

Effect of various temperatures on vitamin C content. When various fruits and vegetables (diced and whole) were frozen at -20 °C for fifteen days, it was observed that vitamin C content (AA+DHAA) decreased to 90% in vegetables, while this loss was limited to 51 % in the fruits. As Table 1 indicates, broccoli showed maximum loss after fifteen days storage at -20 °C (diced and whole) with slight variation. Several studies considered the effect of freezing on level of vitamin C in various vegetables (Korus *et al.,* 2002; Lisiewska *et al.,* 2002). Favell (1998) reported changes in ascorbic acid content on freezing in several vegetables; 20% loss was found in broccoli.

Minimum vitamin C loss was observed in grape fruit which showed 41.05% loss in vitamin C content during the storage as mentioned. A contrary situation was observed in the refrigerated samples. Vitamin C content increased in some refrigerated fruit samples and a tremendous increase was observed in some diced samples due to the moisture loss. As Table 2 indicates, among fruits, maximum increase was observed in vitamin C content of diced lemons which showed an increase of 64.37% after two day refrigeration and minimum increase was observed in papaya which showed an increase of 4.64 % in vitamin C content. Fruits stored without dicing either showed an increase or maintained the level. On the other hand, vegetables stored at 7 °C for two days showed a decrease in vitamin C content except potato. Capsicum showed maximum loss in diced samples which was 36.8 % and minimum loss was observed in broccoli which showed a decrease of 1.2%. Based on these observations, the 2nd part of the studies was conducted.

Table 1. Effect of freezing at -20 °C for 15 days on the vitamin C contents of whole and diced fruits and vegetables

Fruits/Vegetables	Vitamin C content in fresh fruits/vegetables				Vitamin C content in fruits and vegetables stored at -20 °C			
	Whole (mg/100g)	RSD* (%)	Diced (mg/100g)	RSD* (%)	whole (mg/100g)	Loss (%)	Diced (mg/100g)	Loss (%)
Mango	12.30	0.98	13.19	0.62	5.97	51.44	8.32	32.30
Grape fruit	23.88	2.54	22.95	0.81	14.08	41.05	13.8	42.2
Papaya	37.08	0.67	36.73	1.22	19.55	47.29	19.83	46.52
Apple	8.72	0.42	8.03	0.56	4.25	51.26	4.20	51.83
Tomato	41.06	1.02	42.35	3.02	10.62	74.12	11.34	72.38
Lemon	49.34	1.82	49.86	1.80	11.96	75.76	10.37	78.98
Bitter gourd	87.16	0.38	85.94	0.29	26.88	69.16	20.34	76.66
Broccoli	47.29	2.37	46.73	0.86	5.15	89.10	5.12	89.17
Capsicum	31.25	1.04	30.72	1.09	5.16	83.47	5.10	83.68
Potato	5.82	1.15	5.79	2.11	2.67	54.12	2.85	51.03
Spinach	9.55	0.67	9.60	1.69	3.11	67.43	2.95	69.10
Cabbage	8.55	0.49	8.48	2.54	1.93	77.42	1.95	77.19

* = every reading is an average of three independent measurements.

Table 2. Vitamin C content in fruits and vegetables refrigerated at 7 °C for two days

Fruits/vegetables	Vitamin C contents					
	Whole (mg/100g)	RSD* (%)	Vitamin C increase/decrease (%) as compared to fresh** items	Diced (mg/100g)	RSD* (%)	Vitamin C increase/decrease (%) as compared to fresh** items
Mango	14.40	2.13	15.85 I	14.25	1.54	15.85 I
Grape fruit	24.59	1.46	3.0 I	27.79	0.84	16.39 I
Papaya	35.26	0.57	4.9 I	38.807	1.21	4.64 I
Apple	8.85	1.58	1.49 I	9.35	0.43	7.22 I
Tomato	37.49	1.05	8.69 D	31.16	0.94	24.11 D
Lemon	79.65	0.76	61.0 I	81.10	1.02	64.37 I
Bitter gourd	70.76	1.69	51.05 D	19.75	0.86	16.46 D
Broccoli	31.33	0.76	33.74 D	46.73	1.25	1.2 D
Capsicum	15.29	2.02	51.05 D	19.75	0.73	36.8 D
Potato	5.85	0.56	0	5.88	0.92	1.81 I
Spinach	9.05	0.41	5.2 D	9.35	1.61	2.0 D
Cabbage	8.79	2.87	3.4 D	8.05	0.48	8.41 D

* = every reading is an average of three independent measurements; ** I = Increase; D = decrease.

Effect of storage time on vitamin C content. On storage of pre-weighed diced samples at 7 °C for three weeks, vitamin C content of both fruits and vegetables got reduced, though the ratio of degradation was higher in vegetables as compared to fruits. In case of fruits, maximum loss of vitamin C reached up to 50.62 % in apple after one week and in vegetables, this loss amplified to 77.04 %, 75.20 % and 74.76 % in broccoli, capsicum and bitter grourd, respectively (Fig. 1). These results are consistent with earlier studies on freezing, showing the highest average loss of vitamin C for spinach and broccoli and relatively lower, for legumes (Fennema, 1982). During the 1st week of study, diced samples lost about 30-77 % of vitamin C. This loss increased to 98.04 % during the 2nd week of study whereas only traces of vitamin C were left during the 3rd week of study (Fig. 1).

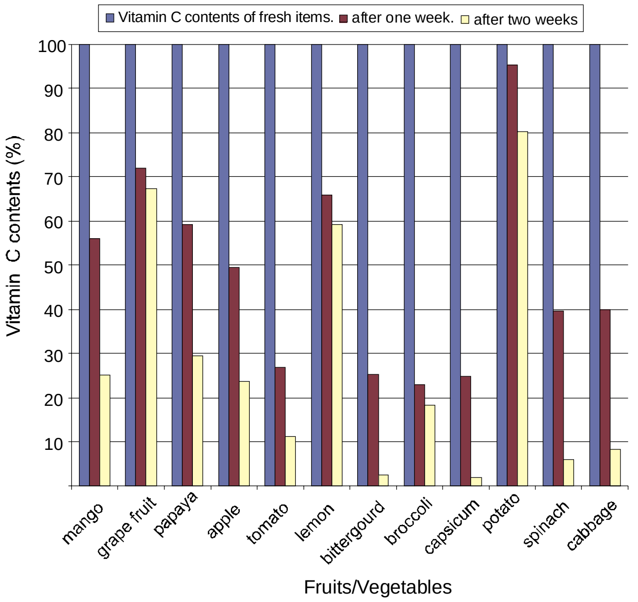

Fig. 1. Loss of vitamin C in diced fruits and vegetables during three week storge.

Conclusion

Due to much loss of moisture content of stored fruits and vegetables, vitamin C content increased when analyzed on the basis of wet weight, but in case of pre-weighed samples, decrease in vitamin C content occured. Thus comparison of nutritive components should always be made on dry weight basis. It was also noticed that the fruits and vegetables with removable peels are less susceptible to vitamin C degradation. Further studies are needed to confirm the role of peels in the stability of fruits and vegetables.

The results of study is important for the general public as well as the food industries since basically fruits and vegetables are eaten as major source of vitamins/antioxidants, minerals and dietary fibers. Accordingly, if stored fruits and vegetables are consumed, the expected health benefits may not be obtained.

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