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Determination of water-soluble vitamins in 15 Iranian pomegranate cultivars and their variation after pasteurization and cold storage

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Abstract

The water-soluble vitamins (WSVs) of 15 pomegranate cultivars grown in Iran as well as their variations in fresh and heat-treated (90°C for 30 sec) juices were evaluated. The fresh and pasteurized juices of selected cultivars were stored respectively for 10 and 180 days at 4°C. The detected WSVs by HPLC-UV, in terms of quantity in the most of cultivars were ascorbic acid (10.4-35.4 mg/100 mL), pantothenic acid (114.9-301.5 µg/100 mL), thiamine (30.8-124.1 µg/100 mL), riboflavin (44.0-236.1 µg/100 mL), pyridoxine (12.0-90.3 µg/100 mL), and biotin (0.0-14.2 µg/100 mL). Significant reduction of WSVs in fresh (57.1-84.2%) and pasteurized (35.3-56.6) juices were observed at the end of storage period. The stability of ascorbic acid was lower than other WSVs in both fresh and pasteurized juices. Based on the results, WSVs are sensitive compounds that destroyed significantly during processing and storage, especially in unprocessed juice.

Introduction

Pomegranate fruit (*Punica granatum L.*) and its products are a rich source of bioactive compounds such as flavonoids, ellagitannins, mainly punicalagins and ellagic acid (Gil *et al.*, 2000). The antioxidant, antibacterial, antifungal, antiviral, anticancer, anticarcinogenic, and anti-inflammatory activities of pomegranate have been attributed to these compounds. Lastly, the pomegranate has been included in the group of the superfoods with health promoting effects (Johanningsmeier and Harris, 2011). Pomegranate fruit contains 85.4% water, 10.6% total sugar, 1.4% pectin and 0.2-1% polyphenol compounds (Prakash and Prakash, 2011). Pomegranate fruit is one of the most important commercial fruits in Iran and its total production in year 2013 was ~910,000 tons (Akhavan *et al.,* 2015). In the world, the pomegranate is consumed as fresh fruit, juice, jam and jelly, and pomegranate supplements.

The nine WSVs including thiamine, riboflavin, niacin, pantothenic acid, folate, biotin, pyridoxine, cyanocobalamin, and ascorbic acid are essential for normal growth, development, and maintenance of the human organism. Human body cannot synthesize vitamins and hence we have to get them through the daily diet in microgram to milligram amounts. Except of vitamin C, the other vitamins are generally served as coenzymes; vitamin C has been shown to be an efficient antioxidant (Sitrin, 2014).

Typically, fruit juices are processed at 90–100°C for a few seconds to inactivate spoilage microorganisms and enzymes and to enhance the shelf-life of refrigerated juices (Donahue *et al.,* 2004); however considerable alterations in physicochemical properties of fruit juices were observed (Patras *et al.,* 2010).

In recent years, the physicochemical properties of pomegranate fruit such as individual phenolic compounds, total phenolic, anthocyanin and tannin contents and also antioxidant activity (Gil *et al.,* 2000; Alighourchi *et al.,* 2008; Çam *et al.,* 2009; Tehranifar *et al.,* 2010; Fischer *et al.,* 2011; Mena *et al.,* 2011) as well as their variation during processing and storage (Martí *et al.,* 2002; Miguel *et al.,* 2004; Pérez-Vicente *et al.,* 2004; Alighourchi *et al.,* 2008; Mena *et al.,* 2012) have been investigated.

Based on these researches, the major physicochemical properties of pomegranate juices have been well documented, but no information is available about identification and quantification of WSVs, especially B-vitamins, as well as their variation during cold storage and pasteurization. On the other hand, limited studies have addressed the effect of cultivar, processing and storage on B-vitamins composition of pomegranate juice. Therefore, the aims of this study were: (i) to quantify major WSVs, (ii) to evaluate WSVs changes in fresh

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untreated juices, and (iii) to determine the stability of WSVs in pasteurized pomegranate juices during storage.

Materials and Methods

Plant material

Fifteen important pomegranate cultivars were harvested in September 2015 from mature fruits growing in the Agricultural Research Center of Yazd. The cultivars selected for this study included: Shirin Shahvare Yazd (No. 1, SSY), Gorche Shahvar Yazdi (No. 2, GSY), Malase Yazdi (No. 3, MY), Vahshe Kane Tehran (No. 4, VKT), Mesri Torshe Kazeron (No. 5, MTK), Jangali Pust Germeze Rodbare Torsh (No. 6, JPGRT), Torshe Mamoli Lasjer (No. 7, TML), Ardestani Torshe Semnan (No. 8, ATS), Khoram Dizin Torshe Gorgan (No. 9, KDTG), Toghe Gardan (No. 10, TG), Zaghe Yazdi (No. 11, ZY), Tabo Larze Mehr Mahi (No. 12, TLMM), Sefeede Robi Aval Brojen (No. 13, SRAB), Pust Syahe Yazd (No. 14, PSY), and Malase Porbarij Stahban (No. 15, MPS).

Chemicals

Water soluble standard (ascorbic acid, thiamine, riboflavin, pantothenic acid, pyridoxine, cyanocobalamin, biotin and folic acid) were purchased from Sigma–Aldrich Chemical Company (Louis, USA). Methanol (HPLC grade) and K_2HPO_4 (extra pure) were obtained from Merck (Darmstadt, Germany). The ultra pure water was prepared with the Purise system (Seoul, South Korea).

Preparation of raw pomegranate juice

Each pomegranate cultivar was washed in cold tap water and drained. They were manually cut-up and the outer leathery skin was removed. The arils were manually separated from the fruits and juices were obtained by squeezing of arils with hand press device. The juice samples (50 mL) were centrifuged (2 min at 10000 rpm at -4°C) with a refrigerated centrifuge (Sigma 3-30K, Germany), and then divided into small vials and kept frozen at –18 ºC upon analysis.

Heat treatment and storage conditions

After the separation and determination of WSVs in the 15 pomegranate cultivars, two pomegranate cultivars (MY and JPGRT) were selected based on the total production, industrial applications and fresh consumption. Samples were divided into two groups. One group (fresh untreated juices) was filled in sterile brown glass bottles and stored at 4°C in a refrigerator and removed at 0, 2, 4, 6, 8 and 10 days for analyses. Other group was pasteurized at 90°C for 30 sec. For this purpose, 10 mL of each pomegranate juices (MY and JPGRT) were completely sealed in Pyrex tubes and then pasteurized in a shaking thermostatic waterbath (Memmert, Germany). The juices were rapidly cooled in an ice bath and stored in a refrigerator at 4°C for 180 days. The pasteurized samples were analyzed on days 0, 20, 40, 60, 90, 120, 150, 180.

Table 1. Linear calibration equations of individual water-soluble vitamin standards that detected in pomegranate juices

WSV	t_R (min)	Linear range (mg/L)	Linear equation	R^2
C	1.90	10- 50	A=10358C-99574	0.985
B_5	6.45	0.1- 5	A=6685C-1823	0.988
B_6	8.40	0.05- 5	A=85520C+8573	0.991
B_1	12.40	0.1- 5	A=75375C-12414	0.995
Biotin	16.80	0.05- 5	A=16054C-1828	0.996
B_2	26.50	0.1- 5	A=53266C+79766	0.990

A: Peak area; C: Concentration (ppm); WSV: water-soluble vitamins; tR: retention time.

HPLC analysis, identification and quantification of water-soluble vitamins

Pomegranate juice consists of many components that cause chromatographic interferences with vitamins. For this reason the sample pretreatment with Sep-Pak C_{18} (500 mg) cartridges were conducted according to method of Ekıncı (2005). WSVs of pomegranate juices were determined using a Discovery C_{18} (4.6×150 mm, dp 5 µm) analytical column (Supelco, Bellefonte, USA), a Waters 600 HPLC system, 20 µL sample loop, and a UV–Vis detector (Waters model 2487). The mobile phase consisted of 50mM K_2HPO_4 buffer (pH 7) (A) and methanol (B). The elution was carried out at room temperature using 1% B for 5 min, 1-30% B in a linear gradient over 15 min, followed by 30% B for 5 min. Flow rate was 1 mL/min with UV–Vis detector at 220 nm (Anonymous, 2000). Calculation of the concentrations was based on the external standard method and WSVs were identified by comparison of their retention times with those of pure standards (Table 1). For each sampling point, there were two replicates.

Statistical analysis

One-way analysis of variance was used to analyze the data. A value of $p<0.05$ or less was taken to be statistically significant (using SPSS Version 16.0 software).

Table 2. Water soluble-vitamins content of 15 Iranian pomegranate cultivars (Mean±SD)

Name	Ascorbic acid (mg/100 mL)	Thiamine (µg/100 mL)	Riboflavin (µg/100 mL)	Pantothenic (µg/100 mL)	Pyridoxine (µg/100 mL)	Biotin (µg/100 mL)
SSY	28.29±0.97^{cd}	68.00±2.05^{e}	96.98±0.80^{h}	136.71±0.42^{g}	39.51±0.67^{f}	14.16±0.10^{a}
GSY	10.37±0.34^{g}	31.62±1.02^{i}	43.98±1.07^{k}	145.20±1.84^{f}	9.72±0.01^{k}	7.68±0.03^{i}
MY	27.34±0.44^{d}	65.99±0.86^{ef}	141.34±5.40^{e}	225.67±6.15^{d}	81.69±0.52^{b}	10.49±0.01^{d}
VKT	30.29±0.66^{b}	33.38±0.41^{i}	154.78±4.64^{c}	122.68±4.17^{h}	90.30±0.90^{a}	13.81±0.32^{b}
MTK	14.61±0.48^{f}	63.38±1.46f^{g}	140.98±3.04^{e}	259.76±5.67^{b}	33.32±0.77^{i}	13.68±0.21^{b}
JPGRT	34.68±0.40^{a}	95.30±1.02^{b}	236.15±2.66^{a}	230.02±5.15^{d}	35.38±0.73^{h}	7.75±0.06^{i}
TML	29.15±0.92^{bc}	124.12±3.10^{a}	154.48±1.44^{c}	128.61±2.44^{h}	81.67±1.47^{b}	8.94±0.09^{g}
ATS	14.81±0.46^{f}	62.49±1.26^{g}	124.07±1.73^{f}	301.47±3.10^{a}	58.27±0.20^{e}	11.75±0.01^{c}
KDTG	11.51±0.35^{g}	79.92±1.64^{d}	160.71±0.30^{b}	238.69±3.97^{c}	32.09±0.09^{i}	7.76±0.03^{i}
TG	16.75±0.08^{e}	77.01±1.49^{d}	147.39±2.47^{d}	114.85±0.28^{i}	10.35±0.06^{jk}	-
ZY	35.35±0.60^{a}	50.58±1.74^{h}	154.21±4.70^{c}	136.95±0.96^{g}	66.89±1.68^{d}	-
TLMM	27.00±0.15^{d}	48.76±0.97^{h}	54.90±1.46^{j}	145.87±2.20^{f}	74.63±1.10^{c}	9.43±0.08^{f}
SRAB	35.39±1.42^{a}	30.75±1.55^{i}	104.19±1.30^{g}	159.41±2.79^{e}	37.27±1.21^{g}	10.11±0.05^{e}
PSY	11.09±0.41^{g}	85.56±0.97^{c}	123.54±1.85^{f}	225.81±3.80^{d}	11.96±0.17^{j}	-
MPIE	27.02±0.89^{d}	34.01±0.57^{i}	64.17±1.20^{i}	146.10±3.05^{f}	35.74±0.22^{gh}	8.42±0.06^{h}

Values with different letters within a similar column are significantly different ($p<0.05$).

Results and Discussion

Water-soluble vitamins of pomegranate juices

Little information is available about WSVs, particularly B-vitamins, in fruits and their products as well as their variations during processing and storage. A major reason for the limited availability of studies is the lack of adequate analytical methodologies for routine analysis of these nutrients. The WSVs content of pomegranate cultivars is presented in Table 2. Six WSVs (Figure 1) including: ascorbic acid, pantothenic acid, pyridoxine, thiamine, riboflavin and biotin were identified in pomegranate juices, as reported previously in some fruits and vegetables (Goverd and Carr, 1974; Santos *et al.,* 2012). Ascorbic acid had the highest, and biotin had the lowest amount of WSVs. In addition, the WSVs of cyanocobalamin and folic acid in studied pomegranate cultivars were not detected. In terms of quantity, the main WSVs in most of the cultivars were ascorbic acid (10.37-35.39 mg/100 mL), pantothenic acid (114.85-301.47 µg/100 mL), thiamine (30.75-124.12 µg/100 mL), riboflavin (43.98-236.15 µg/100 mL), pyridoxine (11.96-90.30 µg/100 mL), and biotin (0.0-14.16 µg/100 mL) (Table 1). In this regard, the WSVs of thiamine (2.8-13.2 mg/100 mL), nicotinic acid (37-150 mg/100 mL), pantothenic acid (47-116 mg/100 mL) and riboflavin (0.3-2.6 mg/100 mL) in apple juice and also nicotinic acid (3-110 mg/100 mL), pantothenic acid (10-100 mg/100 mL), riboflavin (0.3-4.7 mg/100 mL) and no detectable thiamine in commercial vinegar were reported (Goverd and Carr, 1974). In addition, Eitenmiller *et al.* (1977) were detected the WSVs of ascorbic acid (10.5-19.2 mg/100 g), niacin (2750-5458 ng/g), thiamine (440-533 ng/g), riboflavin (292-720 ng/g), pantothenic acid (700-1380 ng/g), pyridoxine (77-213 ng/g), biotin (8-13 ng/g) and free folacin (11-13 ng/g) in blueberry juice.

According to Table 2, significant differences in WSVs content were observed among cultivars ($p<0.05$). Differences in ascorbic acid content among pomegranate cultivars have been previously reported (Dumlu and Gürkan, 2007; Opara *et al.,* 2009; Tehranifar *et al*., 2010), but the B-vitamins of pomegranate cultivar has not been published in the literature. It should be noted that the phytochemical content varies considerably among different cultivars or even different plants. The bioactive compounds of pomegranate fruit are affected by different factors such as cultivar, agro-climatic conditions, maturity, harvest season, irrigation and fertilization, storage and packaging (Mphahlele *et al.,* 2014).

Although the nutritional properties of pomegranate juice has been reported in several studies (Gil *et al.,* 2000; Johanningsmeier and Harris, 2011; Akhavan *et al.,* 2015), no research conducted on B-vitamins in pomegranate and majority of other fruits. But, the ascorbic acid content of pomegranate fruit has been reported in various studies (Dumlu and Gurkan, 2007; Opara *et al.,* 2009; Tehranifar *et al.,* 2010). In this regard, the ascorbic acid content of 9.91-20.92 mg/100 g juice in twenty Iranian pomegranate cultivars (Tehranifar *et al.*, 2010), 312 to 1,050 mg/100 g in Turkish pomegranate cultivars (Dumlu and Gurkan, 2007) and 52.8 to 72.0 mg/100 g fresh arils in Indian, Egyptian and Omani pomegranate cultivars (Opara *et al.,* 2009)

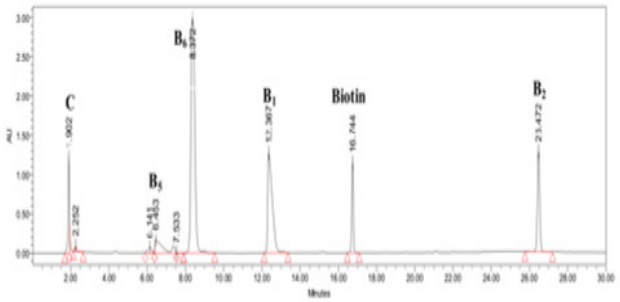

Figure 1. Separation of water-soluble vitamins in pomegranate juice of MY cultivar with HPLC-UV at 220 nm; 1: ascorbic acid 2: pantothenic acid (B5); 3: pyridoxine (B6); 4: thiamine (B1); 5: biotin; 6: riboflavin (B2)

were reported that ascorbic acid content of two later studies significantly higher than Iranian cultivars. The wide variations in the vitamin C content of the various fresh juices 33.13±11.55 mg/100g have been reported (Vikram et al., 2005). These different results may be due to stage of maturity and ripening of fruits and vegetables during harvesting, disinfecting treatments, storage, and processing treatments (Weatherspoon et al., 2005). There was a strong correlation between the date of commercial harvest and the vitamin C contents of pomegranate fruits (Kulkarni and Aradhya, 2005).

Water-soluble vitamins variation of raw juices during storage

Ready-to-drink pomegranate juice has a short shelf-life, is sold in grocery stores in Iran, without any specific processing and addition of preservatives. The detected WSVs in fresh untreated juices of MY and JPGRT cultivars significantly reduced during storage at 4°C for 10 days (Figure 2). The reduction trends of WSVs in some vegetables and fruit juices that stored in various conditions were previously reported (Goverd and Carr, 1974; Eitenmiller et al., 1977; Santos *et al*., 2012). The degradation percentages of WSVs in stored juices of MY and JPGRT cultivars were respectively as follows: ascorbic acid (78.8 and 89.7%), pantothenic acid (68.3 and 67.1), biotin (65.7 and 61.2%), riboflavin (53.4 and 65.2%), pyridoxine (53.3 and 61.0%) and thiamin (48.0 and 77.5%). The results showed that pomegranate cultivars can affect the degradation of vitamins.

The different trends of the vitamins variation in pomegranate juices represented that in actual food systems the factors such as food matrix, structural features, and combined conditions of processing and storage may affect the stability of WSVs. The factors of temperature, air or oxygen, light, moisture content, water activity, pH, degradative enzymes, and metal trace elements (particularly iron and copper) play a role in the degradation of vitamins during processing

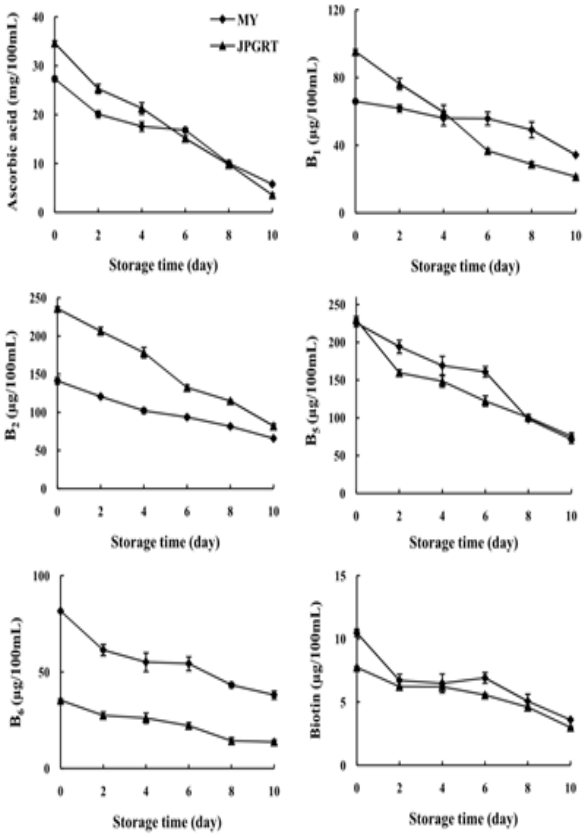

Figure 2. The variation of water-soluble vitamins in unpasteurized pomegranate juice of MY and JPGRT cultivars during storage at 4 °C for 10 days.

and storage (Ball, 2005). Also, the abundance of macro- and micro-nutrients in fresh juices is sufficient to support the growth of a wide range of foodborne pathogens and spoilage microorganisms that used certain basic micro-nutrients (vitamins and minerals) for growth and maintenance of metabolic functions (Weatherspoon *et al.,* 2005). Fruit juice extraction destroys the protective effects of plant cell walls, increase exposure of bioactive compounds to oxygen, light and release enzymes that catalyze their degradation. One such enzyme system is the ascorbic acid oxidase that oxidizes ascorbic acid to dehydroascorbic acid, thus vitamin C is lost. Enzymatic degradation may be a more serious problem than thermal decomposition in many foods that indicate the importance of thermal processing (Perera, 2007). But, low-temperature preservation inhibits microbial growth and slows down the rate of chemical and enzymatic reactions (Perera, 2007).

The results showed that the stability of ascorbic acid was less than the other vitamins. Although ascorbic acid is critical for the health, it should be noted that it is sensitive to processing and is unstable during storage period. Thus, this vitamin is often used as an indicator of fruits and vegetables quality, to evaluate the reduction of other vitamins and

nutritional compounds as well as sensory properties (Cortés *et al.*, 2008; Vervoort *et al.,* 2011). A decline in ascorbic acid content during storage is common among fruit and vegetables, but is influenced by the commodity, storage conditions, and cultivar (Lee and Kader, 2000). Completely degradation of ascorbic acid in unprocessed pomegranate juice (*Mollar* cultivar) over 4 days that kept at 4 and 25°C has been reported (Martí *et al.,* 2002). In contrast, vitamin C content was significantly reduced until 7 days and then remained constant up to 70 days in the new drink made of 75% of pomegranate juice and 25% of lemon juice (González-Molina *et al.,* 2009). Ascorbic acid is the least stable of all vitamins and is easily destroyed during processing and storage. The rate of destruction is increased by the action of metals (especially copper and iron) and enzymes (ascorbic acid oxidase, phenolase, cytochrome oxidase, and peroxidase). Also, exposure to oxygen, light and prolonged heating in the presence of oxygen are all harmful to the vitamin C content of foods (DeMan, 1999). Ascorbic acid is oxidized to dehydroascorbic acid by reactive oxygen species (Conte *et al*., 2008). Also, it is well-known that anthocyanins and ascorbic acid resulted in the degradation of both compounds through a condensation reaction in pomegranate juices (González-Molina *et al.,* 2009). In addition, light can cause degradation of light-sensitive vitamins, including ascorbic acid, thiamin, riboflavin and pyridoxine (DeMan, 1999; Weatherspoon *et al.,* 2005; Perera, 2007). The rate of vitamins destruction increases especially in the alkaline region. But, the pantothenic and folic acids are sensitive to acidic pH (Perera, 2007), that in accordance with our findings.

Water-soluble vitamins variation of juices after pasteurization and during storage

It should be noted that thermal processing is the most effective method for inactivation of microorganisms and enzymes and to increase the shelf-life in the food industry (Donahue *et al.,* 2004). In fruit juices, the enzymes can be inhibited by pasteurization, deaeration, or holding at low temperature for a short period (DeMan, 1999).

Significant changes ($p<0.05$) of WSVs in pasteurized juices (90°C for 30 sec) during storage at 4°C for 180 days were shown in Figure 3. Immediately after pasteurization, ascorbic acid showed the highest degradation rate (20%), followed by thiamine (13.5%), biotin (13%), pantothenic acid (12%), pyridoxine and riboflavin (10.5%). Also, the average degradation of water-soluble vitamins in the stored juices of MY and JPGRT cultivars were respectively as follows: ascorbic acid (52.3 and 60.9%), thiamine

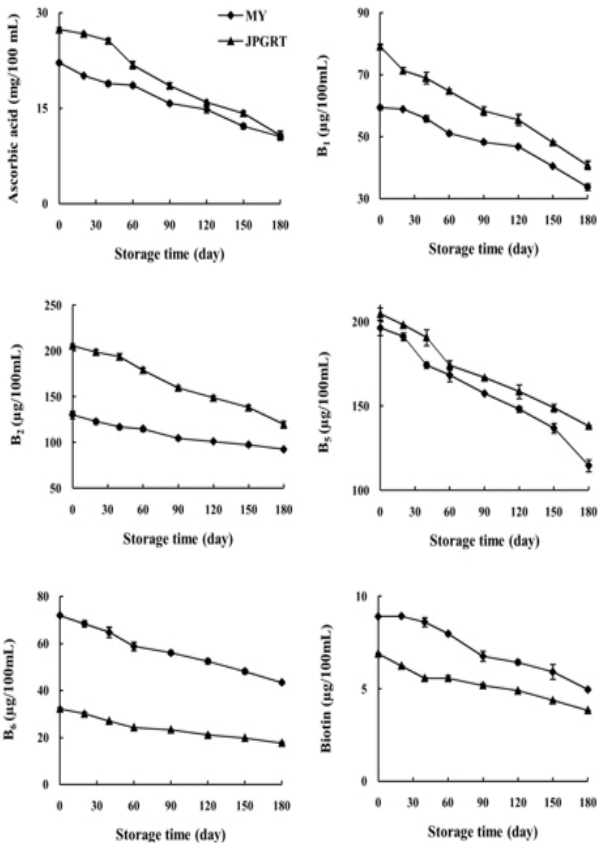

Figure 3. The variation of water-soluble vitamins in pasteurized pomegranate juices of MY and JPGRT cultivars during storage at 4°C for 180 days.

(43.1 and 48.4%), biotin (44.4 and 44.2%), pyridoxine (39.7 and 45.2%), pantothenic acid (41.6 and 32.5%) and riboflavin (28.8 and 41.8%). The losses of WSVs during commercial washing, blanching and domestic cooking have been reported (Ball, 2005). The vitamins destruction during thermal processing may depend on solubility in water. Fat soluble vitamins include A (in the presence of oxygen), D and E, and WSVs such as ascorbic acid, thiamine, riboflavin (in an acidic environment), nicotinic acid, pantothenic acid and biotin are sensitive to heat treatment (Awuah *et al.,* 2007).

Pasteurization and storage time caused significant reduction in WSVs content. The degradation rate of vitamins in pasteurized juices during storage at 4°C for 180 days was lower than in the unprocessed juices stored at the same temperature for 10 days. Lower degradation rate of pasteurized juices can be due to destruction of food-spoilage microorganisms and certain endogenous enzymes, responsible also for reduction of nutritional value (Ball, 2005). Because, microorganisms require certain basic nutrients (water, a source of energy, nitrogen, vitamins, and minerals) for growth and maintenance of metabolic functions. The amount and type of nutrients required

range widely depending on the microorganism. The Gram-negative bacteria are generally able to derive their basic nutritional requirements from the existing carbohydrates, proteins, minerals, and vitamins that are found in a wide range of food (Weatherspoon et al., 2005).

The considerable reduction of ascorbic acid compared with other WSVs, indicate the more susceptibility of vitamin C to chemical oxidation during processing, storage, and cooking (Ball, 2005). Thiamin is one of the more unstable vitamins. Various food processing operations may considerably reduce thiamin levels. Heat, oxygen, sulfur dioxide, leaching, and neutral or alkaline pH may all result in destruction of thiamin (DeMan, 1999). Also, riboflavin is stable to oxygen and acid pH but is unstable in alkaline medium and is very sensitive to light (DeMan, 1999). There are three compounds (pyridoxine, pyridoxal and pyridoxamine) with vitamin B_6 activity. Pyridoxine is stable to heat and strong alkali or acid. Pyridoxal and pyridoxamine are rapidly destroyed when exposed to air, heat, or light. Pyridoxamine is readily destroyed in food processing operations (DeMan, 1999).

The degradation rates of WSVs (except of thiamine, pyridoxine and free folacin) in blueberry juice were lower than 33% after UHT processing and 6 months storage at room temperature and 4°C (Eitenmiller *et al.,* 1977). But, the vitamin C content of various pomegranate juices of *Mollar* cultivar was significantly reduced (by ~55%) by thermal treatment, so that half-life values of ascorbic acid ranged from 21-95 days at 5°C and between 2-12 days for 25°C (Mena *et al.,* 2012). Also, the complete degradation of ascorbic acid in strawberry fresh juices during heating at 95°C for 2 and 4 h have been reported (Sadilova *et al.,* 2009). In this regard, keeping the 80% of the vitamin C content of pasteurized tomato juice at 90°C for 30 s have been reported (Odriozola-Serrano et al., 2007). Effect of Indian gooseberry juice pasteurization at 5 different temperatures (range 75-95°C) and kept at ambient temperature for 9 months showed that the ascorbic acid content in the samples decreased with increasing storage period, but the effect of pasteurization temperatures on ascorbic acid content was not significant. The highest decrease in ascorbic acid content of pasteurized gooseberry juices at the end of storage periods was 60%. Surprisingly, the pasteurization temperature compared with the storage period has slight effect on ascorbic acid content (Bhattacherjee *et al.,* 2011). Results indicated that ascorbic acid is sensitive to heat, so the temperature can be accelerated its degradation.

Conclusion

Fresh pomegranate juice can undergo quality degradation due to microbiological and enzymatic activities and chemical reactions. Maintaining nutritional compounds in pomegranate juice is problematic during processing and storage. In our study, there were significant differences in WSVs levels among pomegranate cultivars. In addition, the WSVs content of fresh and pasteurized juices significantly reduced during storage. Immediately after pasteurization, WSVs content decreased between 10-20%. The degradation rate of vitamins in pasteurized juices during 180 days at 4°C was lower than the fresh untreated juices that stored at the same temperature for 10 day. Although the pasteurization process reduced WSVs content, the unpasteurized juices showed much higher degradation of WSVs. Therefore, pasteurization and storage of fruit juices at refrigeration temperature due to inhibition of microorganism and enzyme activities and slow down the chemical reactions are recommended.

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