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Effect of processing and storage on bioactive compounds and antioxidant activity of carrot juice

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Abstract

Fresh carrot juice is one of the widely consumed vegetable juice during winter season. Recipe for ready-to-serve carrot ginger juice was standardized with the addition of sugar, salt and ginger on the basis of sensory evaluation. The effect of processing and storage on bioactive compounds and antioxidant activity of control and ready-to-serve carrot ginger juice was studied. Among the various combinations prepared, 4% sugar, 0.6% salt, 0.8% ginger and 0.05% citric acid showed highest overall acceptability on the basis of sensory scores and was chosen for further analysis. Total phenolic content was determined by using Folin–Ciocalteu reagent and antioxidant activity was determined by using DPPH assay. During processing, significant losses were found in bioactive compounds and antioxidant activity of control and carrot ginger juice. The study revealed that carrot ginger juice was found to retain more antioxidant activity compared to control juice due to addition of ginger. Storage of six months had no significant effect on TSS and acidity of processed carrot juices. However, storage led to significant decrease in bioactive compounds and thus decreased antioxidant activity of carrot products.

Key words: Carrot juice, bioactive compounds, antioxidant activity, processing, storage

Introduction

Processed and refrigerated vegetable juices are a fast-growing segment within the beverage industry, due to the fact that a high consumption of vegetables has been associated with the prevention of cancer, cardiovascular diseases and other ailments. The beneficial properties of vegetables have been attributed to the presence of antioxidant compounds such as vitamin C, carotenoids and phenolic compounds. Carrot based products are of great health impact because of their high content of carotenoids, some of which also contribute to vitamin A activity such as β-carotene. They also act as free-radical scavengers (Bast *et al.*, 1998; Bramley, 2000). It has been shown that the presence of α- and β-carotene in blood has a protective effect against atherosclerosis (D'Odorico *et al.*, 2000). Carrots also contain substantial amount of vitamin C and phenols. Phenols are known to interrupt lipid peroxidation induced by reactive oxygen species. Vitamin C has been reported to prevent free-radical-induced damage to DNA quenching oxidants, thus, acting as antioxidants (Oviasogie *et al.*, 2009).

In recent years, carrot juice has become an important food commodity for these health reasons. Keeping in view the nutritional and therapeutic value of carrots and to make them available throughout the year, carrot juice in combination with other vegetable and fruit juices can be prepared, stored and evaluated for various characteristics. The importance of naturally occurring bioactive compounds in the maintenance of health is raising considerable interest among scientists, food manufacturers and consumers as the trend of the future is moving toward functional food with specific health effects. Processed carrot juice blended with ginger juice serve as a good source of carotenoids and polyphenols, providing consumers with nutritious and antioxidant rich ready to serve beverage round the year. Some studies have shown that absorption of carotenoids from unprocessed foods is low but it increases with mild processing (Failla and Chitchumroonchokchai, 2005). Thus, processing can have beneficial effects on human health. On the other hand, a significant loss of nutrients may occur as a result of thermal degradation or leaching during blanching, pasteurization and dehydration as well as during storage, home handling and cooking (Kalt, 2005). Hence, the present investigation was carried out to study the effect of processing and storage on bioactive compounds and antioxidant activity of carrot juice.

Materials and methods

Carrots were procured during the last week of January from the village Ranuuan, Samrala, Distt. Ludhiana, Punjab.

Carrot juice processing: Fresh carrots were washed thoroughly, peeled and cut from the top and bottom. Carrot juice was extracted in a screw type juicer extractor. Carrot juice was optimized with different sugar concentrations such as 2, 4, 6% and different ginger concentration (0.8 and 1%); constant salt (0.6%) and citric acid content (0.05%). Finally the particular level was selected on the basis of sensory evaluation (Table 1). All the ingredients were mixed and the juice was heated in a stainless steel vessel at 85°C for 3 min. Glass bottles of 200 mL capacity were pre-washed and pre-sterilized in boiling water. Bottles were filled with hot juice and corked. They were finally heated at 100 °C for 20 min in water and gradually cooled under running tap water. Control juice was prepared in the same manner without any added ingredients. Finally the bottles were kept at room temperature for six months.

Analytical methods: Fresh carrot juice, control and ready-to-serve processed carrot juice were evaluated for TSS, acidity,

ascorbic acid, total carotenoids, β-carotenoids, lycopene, total phenolic content and antioxidant activity. TSS, acidity, ascorbic acid, total carotenoids, β-carotenoids and lycopene content was determined as per Ranganna (1986). Total phenolic content was determined by using Folin–Ciocalteu reagent (Swain and Hillis, 1959). A standard curve was plotted by taking known amount of gallic acid as reference standard and concentration was calculated from the standard curve. Free radical scavenging activity was determined by DPPH (di phenyl picryl hydrazyl) method according to Brand-Williams *et al.* (1995) with some modifications. Methanolic extract of 5 g sample was taken for antioxidant activity analysis and calculated according to the following formula. BHT was taken as a standard at a fixed concentration of 5mg/mL.

$$\text{Radical scavenging activity(\%)} = \frac{\text{Control OD (0 min)} - \text{Sample OD (30 min)}}{\text{Control OD (0 min)}} \times 100$$

The sensory quality of processed carrot juice was assessed for appearence, flavor, mouthfeel and overall acceptability. Different attributes of the product were evaluated by a panel of seven judges using 9-point Hedonic rating scale (Amerine *et al.*, 1965). Results were analyzed statistically using completely randomized design as discussed by Singh *et al.* (1991).

Results and discussion

Optimization of carrot juice recipe on the basis of sensory evaluation: No significant effect of different levels of sugar, salt and ginger was found on appearance of the juice. However there was significant ($P \leq 0.05$) difference in the flavor, mouth feel and overall acceptability due to different levels of sugar and ginger. On the basis of sensory evaluation carrot juice with 4% sugar, 0.8% ginger and 0.6% salt received highest score in terms of overall acceptability and was chosen for further studies (Table 1).

Effect of processing on bioactive compounds and antioxidant activity of carrot juice

TSS and acidity: The TSS of fresh carrot was found to be 8.20 °Brix (Table 2). Total soluble solids for carrots ranged from 8.46 to 9.98 °Brix (Sandhu *et al.*, 1988). TSS of fresh juice was same as that of fresh carrot *i.e.* 8.20 °Brix. TSS of control juice was recorded to be 10.00 °Brix whereas TSS of carrot ginger juice was found to be13.00 °Brix due to addition of sugar. Acidity of 0.05 per cent was found in fresh carrots. Acidity in carrot cultivars was found to be 0.06 per cent in 'Sel 21', 0.04 per cent in 'PC-34', 0.06 per cent in 'Ambala local' and 0.05 per cent in 'Nantes' (Sra *et al.*, 2011). Heat processed control juice and carrot ginger juice had 0.06 and 0.08 per cent acidity, respectively. Increase in the acidity of altered juice compared to control sample was due to the addition of citric acid.

Total carotenoids, β carotene and lycopene: Table 2 depicts the effect of processing on total carotenoids and β-carotene of carrot products. Fresh carrots were found to have 13.35 mg/100g total carotenoid and 10.13 mg/100g β-carotene. The present study corroborates the findings of Sethi and Anand (1983) who reported the β-carotene content of raw carrot to be 11.88 mg/100g but Gopalan *et al.* (1997) reported lower (6.46 mg/100g) β-carotene content in fresh carrots. The variation in carotenoids may be due to variety, maturity, growing conditions, growing season and the part of the root sampled (Hart and Scott, 1995).There was a reduction in total carotenoids and β-carotene in carrot juices after processing. The total carotenoid content of raw juice, heat processed control and carrot ginger juice was 12.42 mg/100g, 10.10 mg/100g and 10.62 mg/100g, respectively. β-carotene content of raw juice, processed control and carrot ginger juice was found to be 9.86, 8.50 and 8.82 mg/100g, respectively. Lycopene content in raw juice, processed control and carrot ginger juice was 3.16 mg/100g, 2.93 mg/100g and 2.96 mg/100g, respectively. The decrease in lycopene content may be due to its co-planarity structure being very reactive to oxidation (Lin and Chen, 2005). Similar results were found by Dhaliwal and Hira (2001) who reported that fresh carrot: beetroot (95:5) had 3.56 mg/100g of β-carotene which reduced to 2.88 mg/100g after pasteurization, thus accounting for 19.10 % reduction. The loss of total carotenoids in juice during preparation may be due to oxidation or thermal degradation of unsaturated total carotenoids.

Ascorbic acid: Fresh carrots contained 4.31 mg/100g ascorbic acid (Table 2). According to Alasalvar *et al.* (2001) vitamin C content varied between 1.25 and 5.33 mg/100g, being lowest in white and highest in orange varieties of carrot. Ascorbic acid content in raw juice, processed control and carrot ginger juices were 2.24, 1.79 and 1.94 mg/100g, respectively. Losses of ascorbic acid in heat processed juice may be due to thermal degradation. Grewal and Jain (1982) also reported 27.62 % loss of ascorbic acid during processing of carrot juice beverage.

Total phenols: Raw juice contained 34.98 mg/100g galic acid equivalents (GAE) of total phenols while in control and altered juice it was found to be 29.95 mg/100g GAE and 34.84 mg/100g GAE, respectively (Table 2). Teixiera *et al.* (2009) found that fresh carrot juice had 334 mg L^{-1} of total phenol which reduced to 306 mg L^{-1} after heat treatment at 90° C for 30 sec. Carrot ginger juice had higher phenolic content than control juice due to the addition of ginger to carrot juice. Tarko *et al.* (2010) found that addition of any of the seasonings to apple chips such as cinnamon, garlic, ginger, pepper, mint, onion increased the values of total

Table 1. Optimization of carrot ginger juice recipe on the basis of sensory evaluation*

Sample	Appearance	Flavor	Mouth feel	Overall acceptability
2% sugar + salt + 0.8% ginger	8.42±0.60	7.85±0.37	7.85±0.37	8.05±0.23
2% sugar + salt + 1% ginger	8.42±0.44	8.00±0.40	7.85±0.55	8.09±0.23
4% sugar + salt + 0.8% ginger	8.57±0.44	8.42±0.53	8.28±0.39	8.42±0.18
4% sugar + salt + 1% ginger	8.57±0.34	8.28±0.48	8.00±0.57	8.28±0.26
6% sugar + salt + 0.8% ginger	8.57±0.44	8.00±0.57	8.00±0.40	8.14±0.20
6% sugar + salt + 1% ginger	8.57±0.53	8.14±0.55	8.14±0.55	8.28±0.31
CD ($P \leq 0.05$)	NS	0.14	0.13	0.14

*Mean ± SD (Standard deviation) of score by seven penalist

phenolic content which explains the higher content of phenolics in carrot ginger juice.

Antioxidant activity: Fresh carrots were found to have 23.83 % scavenging activity (Table 2). Radical scavenging capacity in raw carrots decreased continuously with increase in reaction time and maximum activity was shown at 30 minutes and became stable thereafter (Fig.1). Antioxidant activity of raw carrot juice was found to be 23.39 % (Table 2) and maximum antioxidant activity as determined by DPPH assay was shown at 30 minutes (Fig. 1). After processing the antioxidant activity of control and carrot ginger was found to be 21.73 and 23.37 %. Similar results were found by Teixiera *et al.* (2009) who observed that antioxidant capacity of fresh carrot juice was 211 milimol trolox L^{-1} which decreased to 199 milimol trolox L^{-1} after heat processing at 90° C for 30 seconds. Decrease in the antioxidant activity may be due to decrease in the levels of bioactive compounds such as ascorbic acid, total carotenoids and phenolic compounds. Duddone *et al.* (2009) found that antioxidant activity were correlated to phenolic compound concentration and Teixeira *et al.* (2009) found positive correlation between ascorbic acid and DPPH values ($R^2 = 0.807$) in processed carrot juice. Thus, bioactive compounds in carrot juice have positive relation with antioxidant activity, as a result antioxidant activity increases with increase in concentration of bioactive compounds and vice-versa. Carrot ginger juice had 1.64 % more antioxidant activity than control juice. This increase is due to the addition of ginger in carrot juice, as ginger is found to have good antioxidant properties (Shirin and Jamuna, 2010). Both control carrot juice and carrot ginger juice showed maximum antioxidant activity at 40 minutes (Fig. 1).

Effect of storage on bioactive compounds and antioxidant activity of carrot juice

TSS and acidity: As can be seen from Table 3, TSS of control juice and altered carrot juice decreased non significantly during storage period of six months. It decreased from 10.00 to 9.73 °Brix in control carrot juice and from 13.00 to 12.60 °Brix in carrot ginger juice. Similar results have also been reported by Aggarwal *et al.* (1995) who found negligible change in the total soluble solids of processed tomato juice from six varieties during six months storage. There was a slight increase in the acidity during storage. However, the overall increase was non significant. Carrot ginger juice also showed non significant increase in acidity during storage. Dhaliwal and Hira (2001) also observed non-significant change in the acidity of pasteurized carrot-beetroot juice during 6 month storage.

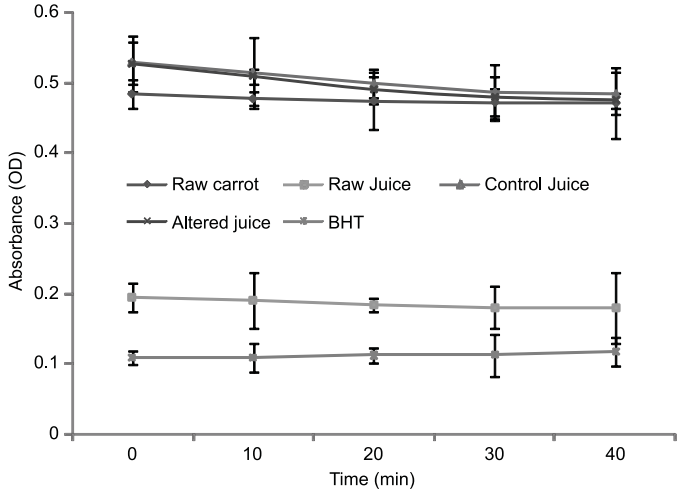

Fig. 1. Radical scavenging capacity of fresh and processed carrot juice

Table 3. Effect of storage on total soluble solids and acidity of carrot juice*

Storage months	TSS (°Brix)		Acidity (%)	
	Control	Carrot ginger juice	Control	Carrot ginger juice
0	10.00±0.03	13.00±0.02	0.06±0.01	0.08±0.02
1	9.94±0.10	13.06±0.05	0.06±0.03	0.09±0.04
2	9.87±0.11	12.87±0.11	0.07±0.04	0.10±0.02
3	9.87±0.11	12.87±0.10	0.08±0.02	0.11±0.03
4	9.80±0.03	12.80±0.02	0.08±0.01	0.12±0.03
5	9.73±0.09	12.67±0.11	0.06±0.03	0.10±0.02
6	9.73±0.11	12.60±0.00	0.05±0.01	0.10±0.01
F test	NS	NS	NS	NS

*Values are Mean ± SD (Standard deviation) of three replicates

Total carotenoids, β-carotene and lycopene: Effect of storage on total carotenoids, β-carotene and lycopene of control and carrot ginger juice is shown in Table 4. Total carotenoids decreased significantly ($P \leq 0.05$) from 10.10 mg/100g to 7.75 mg/100g in control juice and from 10.62 mg/100g to 7.97 mg/100g in carrot ginger juice, after six months storage at room temperature. β-carotene content showed significant ($P \leq 0.05$) reduction during storage in both control and carrot ginger juice. In control, juice β-carotene decreased from 8.50 mg/100g to 5.65 mg/100g and in carrot ginger juice from 8.82 mg/100g to 5.87 mg/100g. Dhaliwal and Hira (2001) also observed the decreasing trend in β-carotene content during six month storage of carrot: beetroot (95:5) beverage. The decrease in total carotenoids and β-carotene content may be due to oxidation of highly unsaturated carotenoid structure (Kidmose *et al.*, 2002). Chen *et al.* (1996) observed the stability of carotenoids during storage of carrot juice by subjecting the carrot juice to storage at different temperatures and in light and dark for three months. They found that α-carotene, β-carotene and vitamin A in carrot juice decreased with increasing storage temperature and light is more destructive to carotenoids than darkness. Teixeira *et al.* (2009) found that β-carotene concentration of carrot juice decreased throughout

Table 2. Effect of processing on physico-chemical and bioactive compounds of carrot and carrot juice*

Products	TSS (°Brix)	Acidity (%)	Ascorbic acid (mg/100g)	Total carotenoid (mg/100g)	β-carotene (mg/100g)	Lycopene (mg/100g)	Total phenols (mg/100g)	Scavenging activity (%)
Raw carrot	8.20±0.02	0.05±0.01	4.31±0.05	13.35±0.07	10.13±0.08	3.36±0.04	39.93±0.19	23.83±0.19
Raw juice	8.20±0.02	0.06±0.02	2.24±0.03	12.42±0.07	9.86±0.08	3.16±0.06	34.98±0.22	23.39±0.07
Control juice	10.00±0.03	0.06±0.01	1.79±0.03	10.10±0.06	8.50±0.05	2.93±0.03	29.95±0.25	21.73±0.28
Carrot ginger juice	13.00±0.02	0.08±0.02	1.94±0.05	10.62±0.06	8.82±0.04	2.96±0.03	34.84±0.26	23.37±0.32
CD ($P \leq 0.05$)	0.21	NS	0.06	0.11	0.09	0.05	0.15	0.19

*Values are Mean ± SD (Standard deviation) of three replicates

the storage following first order kinetics and found that higher degradation of β-carotene was found in heat-treated juices than high intensity pulse electric field treated juices. According to Aczel, (1972) and Drdak and Sorman (1979) time of storage and temperature considerably lowers the retention of β- carotene. Lycopene content decreased significantly ($P \leq 0.05$) from 2.93 mg/100g to 2.33 mg/100g in control juice and from 2.96 mg/100g to 2.42 mg/100g in carrot ginger juice, after six months storage. These results are in accordance with the findings of Lin and Chen (2005) who found decrease in lycopene content and its isomers in tomato juice during storage. According to them, in addition to isomerization, oxidative degradation is a major factor causing lycopene loss during storage of tomato juice. Also, Sharma and Le Magure (1996) stated that illumination could enhance the reaction rate of lycopene in the presence of air, and the degradation of all-trans-lycopene could be accompanied by the isomerization.

Ascorbic acid: The ascorbic acid content of control and altered juice decreased significantly ($P \leq 0.05$) during storage (Table 5). With the advancement of storage, ascorbic acid decreased from 1.79 to 0.57 mg/100g in case of control juice and from 1.94 to 0.72 mg/100g in carrot ginger juice. This decrease in ascorbic acid may be due to oxidation of ascorbic acid in presence of light. Similarly, Dhaliwal and Hira (2001) observed significant losses of ascorbic acid during storage of carrot juice. Higher losses have been found at elevated temperature storage. According to Kaur *et al.* (2004) the rate of ascorbic acid destruction increases with increased temperature in the presence of air.

Total phenols: The effect of storage on total phenolic content as investigated during six month storage of control and altered carrot juice has been presented in Table 5. There was significant ($P \leq 0.05$) reduction of total phenols during storage of both the juices. In case of control juice there was a decrease in total phenolics from 29.95 mg/100g to 15.97 mg/100g and from 34.84 mg/100g to 19.93 gm/100g in case of carrot ginger juice. These results are in agreement with Klimczak *et al.* (2007) who reported that hydrocinnamic acid in orange juice stored at 18, 28 and 38°C decreased by about 13, 22 and 32 per cent, respectively. Similarly, Teixeira *et al.* (2009) also found that total phenolic content in carrot juice decreased during storage of 56 days.

Antioxidant activity: During storage period, antioxidant activity of control and carrot ginger juice decreased significantly ($P \leq 0.05$) (Table 5). Scavenging activity in control carrot juice reduced from 21.73 per cent during zero month to 10.57 per cent after six months. In carrot ginger juice the scavenging activity decreased from 23.37 to 13.51 per cent after storage. The decrease in the antioxidant activity may be linked to a decrease in total phenolic content and vitamin C (Klimczak *et al.*, 2007). According to Teixeira *et al.* (2009) the antioxidant activity of carrot juice depleted with storage time regardless of the processing treatment. They found that DPPH values co-related well with vitamin C ($R^2 = 0.807$) which indicates that vitamin C is among one of the antioxidant compounds in carrot juices. Same authors reported that antioxidant capacity and β-carotene are also co-related well ($R^2 = 0.788$), suggesting that the variation in antioxidant capacity over time can be modulated by carotenoids. Klimczak *et al.* (2007) found that antioxidant activity of orange juices decreased by 18, 45 and 84 per cent after six months of storage at 18, 28 and 38°C, respectively.

Processing induces significant changes in chemical composition, influencing the concentration and bioavailability of bioactive compounds in carrots. It can have both positive and negative effects depending on process conditions (Miglio *et al.,* 2008). It is therefore desirable to assess the effect of processing on bioactive compounds and antioxidant activity of carrot products. Thus,

Table 4. Effect of storage on total carotenoids, β-carotene and lycopene content of carrot juice*

Storage months	Total carotenoids (mg/100g)		β-carotene (mg/100g)		Lycopene (mg/100g)	
	Control	Carrot ginger juice	Control	Carrot ginger juice	Control	Carrot ginger juice
0	10.10±0.06	10.62±0.06	8.50±0.05	8.82±0.04	2.93±0.03	2.96±0.03
1	9.65±0.07	9.83±0.05	7.95±0.05	8.03±0.06	2.82±0.02	2.89±0.03
2	9.26±0.05	9.30±0.03	7.19±0.03	7.37±0.05	2.68±0.04	2.74±0.05
3	8.53±0.03	9.09±0.03	6.70±0.06	6.72±0.07	2.60±0.02	2.56±0.02
4	7.99±0.04	8.33±0.03	6.39±0.05	6.57±0.06	2.54±0.01	2.51±0.03
5	7.82±0.04	8.11±0.06	5.86±0.05	6.14±0.03	2.42±0.04	2.47±0.03
6	7.75±0.06	7.97±0.04	5.65±0.06	5.87±0.06	2.33±0.02	2.42±0.02
CD ($P \leq 0.05$)	0.05	0.05	0.05	0.08	0.05	0.06

*Values are Mean ± SD (Standard deviation) of three replicates

Table 5. Effect of storage on ascorbic acid, total phenols and antioxidant activity of carrot juice*

Storage months	Ascorbic acid (mg/100g)		Total phenols (mg/100g)		Antioxidant activity (%)	
	Control	Carrot ginger juice	Control	Carrot ginger juice	Control	Carrot ginger juice
0	1.79±0.03	1.94±0.05	29.95±0.25	34.84±0.26	21.73±0.28	23.37±0.32
1	1.31±0.04	1.65±0.03	29.97±0.40	34.93±0.34	18.85±0.35	20.26±0.31
2	1.08±0.04	1.32±0.03	24.82±0.48	29.94±0.27	15.46±0.40	18.15±0.28
3	0.92±0.03	1.05±0.04	20.00±0.48	24.90±0.30	13.22±0.51	16.98±0.23
4	0.79±0.03	0.91±0.02	19.95±0.31	19.89±0.38	13.02±0.46	14.72±0.30
5	0.70±0.02	0.84±0.03	19.94±0.44	19.95±0.39	12.64±0.32	14.30±0.31
6	0.57±0.02	0.72±0.04	15.97±0.23	19.93±0.33	10.57±0.25	13.51±0.29
CD ($P \leq 0.05$)	0.06	0.08	0.44	0.40	0.88	0.99

*Values are Mean ± SD (Standard deviation) of three replicates

from the present investigation we conclude that during processing 79.94-86.76% ascorbic acid, 81.33-85.51% total carotenoids, 86.21-89.46% β-carotene, 92.73-93.68% lycopene, 85.63-98.6% total phenols and 92.91-99.81% antioxidant activity was retained in control and ready-to-serve carrot ginger juice, respectively. More antioxidant activity was found in ready-to-serve carrot ginger juice as compared to control juice due to addition of ginger which is known as a good source of antioxidant. During storage, mean sensory scores decreased non-significantly. No significant changes were observed in TSS and acidity of control and ready-to-serve carrot juice.

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