



## Maintaining the postharvest nutritional quality of peach fruits by $\gamma$ -Aminobutyric acid

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### Abstract

Fresh peaches are highly perishable and they deteriorate quickly during low temperature storage which results in losses in nutritional quality. To prevent these adverse effects, peach fruits were treated by dipping with  $\gamma$ -aminobutyric acid (GABA; 2, 4 and 6 mM) immediately after harvest and then stored at 1 °C for 7, 14, 21, and 28 days. Peach fruits treated with GABA exhibited higher flesh firmness and TSS. In addition, GABA treatments were effective in maintaining higher contents of healthpromoting molecules such as ascorbic acid, total phenols and flavonoids as well as DPPH scavenging capacity. These results suggest that GABA could have promising postharvest effect for maintaining quality and enhancing the health benefits of peach fruit consumption by increasing the antioxidant capacity.

*Keywords:* Peach; postharvest;  $\gamma$ -aminobutyric acid; nutritional quality

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### Introduction

Peach (*Prunuspersica* L) fruit is one of the most important horticultural crops in the world because of its attractive taste and nutritive value. It is highly perishable and deteriorates quickly during storage (Nunes, 2008). Rapid ripening in peach fruits that happens in the high temperature accompanied by low relative humidity during harvesting and marketing is responsible for the short postharvest life of the commodity. Under

normal storage conditions the life of fruit does not exceed 5 days (Tonini and Tura, 1998). Postharvest strategies have been presumed to extend the shelf life and quality of peaches such as heat (Cao et al., 2010), intermittent warming (Zhu et al. 2010), gamma irradiation (Hussain et al., 2010) and treatment with chemicals such as aminoethoxyvinylglycine, 1-methylcyclopropene (Hayama et al., 2008), calcium chloride (Manganaris et al., 2007), nitric oxide (Zhu et al., 2010), salicylic acid (Cao et al., 2010) and methyl jasmonate (Jin et al., 2009).

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Table 1

ANOVA for dependent variables for GABA treatments, storage time, and their interactions for peach fruits<sup>a</sup>

Quality attributes (Sensory and Nutritional)	Time	Treatment	Time × Treatment
Firmness	271.3**	30.1*	1.9 <sup>ns</sup>
Weight loss	18.3**	0.458 <sup>ns</sup>	0.138 <sup>ns</sup>
pH	0.737**	0.004 <sup>ns</sup>	0.005 <sup>ns</sup>
Total soluble solids (TSS)	33.99**	0.29 <sup>ns</sup>	1.86**
Total acidity (TA)	0.002**	0.005 <sup>ns</sup>	0.005 <sup>ns</sup>
Total phenols (TP)	3455.5**	982.4**	109.9 <sup>ns</sup>
Total flavonoids (TF)	1986.3**	248.2**	48.1**
Ascorbic acid (AA)	4.07**	1.6**	0.2 <sup>ns</sup>
Total antioxidant activity (TAA)	1820.7**	142.5**	40.5*

<sup>a</sup> \*\* and \* represent significance at the 0.01, and 0.05 levels, respectively, and NS represents non-significance at P < 0.05.

Fresh peaches are highly perishable and they deteriorate quickly during low temperature storage which results in losses in nutritional quality. To prevent these adverse effects caused by postharvest factors, use of environmentally friendly technologies such as  $\gamma$ -aminobutyric acid (GABA) as a safe signalling molecule was recommended (Yang et al., 2011; Shang et al., 2011). GABA, a four carbon non-protein amino acid, as a signalling molecule plays a crucial role in response to stress such as salinity, anoxia, hypoxia, drought, heat, and chilling (Kinnersley and Turano, 2000). In addition to anti-stress function of GABA, GABA plays an important role in human health due to its antihypertensive effects (Mae et al., 2012). Song et al. (2010) reported that GABA treatment mitigates oxidative stress produced by Al<sup>3+</sup> toxicity in barley seedlings by increasing the levels of SOD and CAT activities, which in turn reduces the accumulation of ROS and MDA. Yang et al. (2011) showed that the treatment of peach fruit with 5 mM GABA for 10 min reduces chilling injury and generally enhances the activities of antioxidant enzymes such as SOD, CAT, APX, GPX, GST, MDHAR, and DHAR. Shang et al. (2011) reported that the exogenous GABA treatment alleviated chilling injury and maintained higher fruit quality in peach fruit during cold storage. The effect of GABA treatment on alleviating chilling injury of peach may be attributed to its ability to enhance accumulation of endogenous GABA and proline contents in peaches, which was due to the increased glutamate decarboxylase (GAD), pyrroline-5-carboxylate synthetase (P5CS), and ornithine  $\delta$ -

aminotransferase (OAT) activities and decreased proline dehydrogenase (PDH) activity. Shang et al. (2011) suggested that the exogenous GABA treatment not only alleviated chilling injury but also maintained postharvest quality in peach fruit. The studies related to impact of exogenous GABA treatment on the postharvest behavior and nutritional quality of peach fruit has not been reported. Hence, the objective of this study was to evaluate the effect of GABA treatment on the total phenols, flavonoids and ascorbic acid contents, and DPPH scavenging capacity as well as flesh firmness, TSS, TA, and weight loss of peach fruits during storage at 1 °C for 28 days.

## Materials and Methods

### Fruits and treatment

Peach fruit (*Prunuspersica* L. Batsch) cultivar 'Anjiryemaleky' were harvested from a commercial orchard in Tabriz, Iran. The fruit were then transported to the laboratory and were divided into twelve groups each one containing 40 fruits. Fruit were dipped in distilled water (control) or in 2, 4, and 6 mM GABA for 10 min. The treated fruit were then air dried for 60 min and stored at 1 °C and 80–90% RH for 28 d. During cold storage control and GABA treated fruits were subjected to physicochemical analysis once a week.

### Quality attributes

Flesh firmness was measured using Effegi penetrometer (Model FT 011) with an 8 mm diameter flat probe. Total acidity (TA) was determined by titrating 10 mL of juice to pH 8.2

using 0.1 M NaOH and the results were expressed as gram of malic acid equivalent per 100 g of FW. Total soluble solids (TSS) were determined using a digital refractometer (Model PAL-1). The pH values of solutions were monitored with a pH meter (Model HI 9811). Weight loss was determined in each replication (five fruit per replication) and was recorded initially and weekly during storage. Weight loss was calculated as:  $(W_0 - W_f) / W_0 \times 100$  where  $W_0$  is the initial sample weight and  $W_f$  is the final sample weight. Results are reported as percentage weight loss.

### Total phenolics, flavonoids, and ascorbic acid contents

The total phenolic content of the extract was determined according to the Folin-Ciocalteu method as described by Singlton and Rossi (1965) with slight modifications. Briefly, 0.1 milliliter of extract was transferred into a test tube and mixed with 2 ml of 2%  $\text{Na}_2\text{CO}_3$  and allowed to stand for 2 min at room temperature. For each sample 0.1 milliliter of 50% (v/v) Folin-Ciocalteu reagent was added and the reaction mixture was mixed and allowed to stand for 30 min in the darkness. After incubation, absorbance was read at 720 nm. The absorbance values were converted to total phenolics and were expressed in micrograms equivalents of gallic acid (GAE) per 100 g FW. Different concentrations of gallic acid in 95% methanol were used as standards. The total flavonoid concentration of the peach extract was determined using a colorimetric assay (Kaijv et al., 2006). The absorbance of the solution versus a blank at 510 nm was measured immediately. The results were expressed as mM of quercetin equivalents per 100 g FW. Ascorbic acid content was estimated spectrophotometrically by dinitrophenylhydrazine (DNPH) method (Terada et al., 1978). The ascorbic acid content was expressed as ascorbic acid on a fresh weight basis, mg per 100 g fresh weight.

### DPPH radical scavenging activity

The DPPH radical scavenging activity of peach extracts was measured according to the method of Dehghan and Khoshkam (2012). Fruit extract was added to 2 ml of DPPH solution (0.1 mM in methanol). The mixture was shaken

vigorously and kept in the dark at room temperature for 30 min. Absorbance of the mixture (AS) was measured at 517 nm in a UV-visible spectrophotometer (T-60, PG Instrument UK). As a control, the absorbance of the blank solution of DPPH (2 ml) was also determined at 517 nm (AC). The percentage of radical scavenging activity (RSA %) was calculated according to the following equation:

$$\text{RSA \%} = \frac{100 (A_c - A_s)}{A_c}$$

### Statistical analysis

The experiment was arranged as split plots in time based on completely randomized design with three replications. Sources of variation were storage times and treatment. Analysis of variance (ANOVA) was carried out with SPSS software. Mean comparison were performed by Duncan's multiple range tests at  $P \leq 0.05$  and  $P \leq 0.01$  probability levels. ANOVA table with the significance of each factor and their interaction is shown in Table 1.

### Results

#### GABA treatments and quality attributes

As shown in Table 2, GABA treatments maintained significantly ( $p < 0.05$ ) higher firmness in peaches. GABA at 6 mM was more effective in maintaining firmness than 2 and 4 mM GABA treatment, and no significant difference was found between 2 and 4 mM GABA treatments (Table 2). As shown in Fig. (I), TSS in GABA treated peach fruits were significantly enhanced during postharvest storage ( $P < 0.01$ ). However, there was no significant difference in TSS of peaches between applied GABA concentrations (Fig. I). Also, GABA treatments had no significant effect on weight loss, TA, and pH (Table 2).

Table 2

Effect of GABA treatment at 0, 2, 4, and 6 mM on flesh firmness, total phenols (TP), and ascorbic acid (AA) content of peach fruit

GABA treatments	Quality attributes		
	Firmness	Total phenols (TP)	Ascorbic acid (AA)
Control	16.71c	64.10b	3.54c
2 mM	18.33b	68.72b	3.92b
4 mM	17.94b	80.74a	4.33a
6 mM	20.55a	82.62a	4.29a

Each value represents the mean values  $\pm$  SD (n = 3). Mean values in each column followed by the same lower-case letters are not statistically different by Duncan's multiple range test ( $P < 0.05$ ).

### GABA treatments and total phenols, flavonoids, and ascorbic acid contents

As shown in Table 2, the GABA treatments at 4 and 6 mM maintained higher total phenols content in peaches ( $P < 0.01$ ), and 2 mM GABA had no significant effect on total phenols content. Similar to TSS, the total flavonoids content of peach fruits treated with GABA was significantly enhanced during postharvest storage ( $P < 0.01$ ) (Fig.II). Moreover, the results showed that the contents of flavonoids of the peach fruits treated with 6 mM GABA were higher than those treated with 2 and 4 mM, which suggests that the effects of GABA on flavonoids content of the peach fruits was concentration dependent. As shown in Table 2, GABA treatments at 4 and 6 mM significantly enhanced total ascorbic acid content in peach fruits (Table 2;  $P < 0.01$ ).

### GABA treatments and DPPH scavenging activity

As shown in Fig. III, DPPH scavenging capacity of the peach fruits treated with GABA was

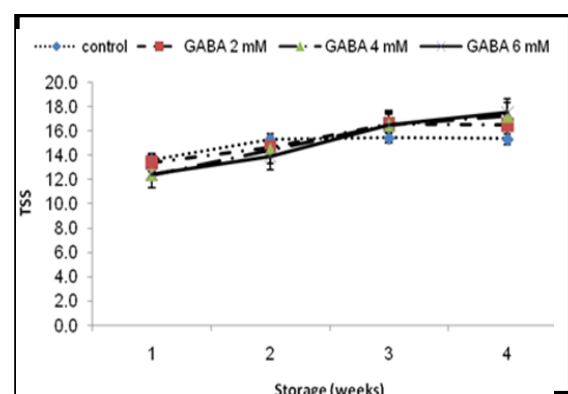


Fig. I. The effects of 0, 2, 4 and 6 mM GABA on TSS of peach fruits stored at 1°C for 4 weeks. Data shown are Mean values  $\pm$  SD (n=3).

significantly enhanced when compared with that of the control ( $P < 0.05$ , Fig. III), showing that GABA treatment stimulated the scavenging capacity of peach fruits on DPPH radical. Moreover, the results showed that the peach fruits treated with 4 and 6 mM GABA have higher DPPH scavenging capacity than fruits treated with 2 mM GABA, suggesting that treatment of 4 and 6 mM GABA might be optimal for enhancing the DPPH scavenging capacity of the peach fruits.

### Discussion

Shang et al. (2011) reported that the peach fruit treated with GABA exhibited significantly higher extractable juice. Also, GABA treatment maintained significantly higher TSS and TA in peaches (Shang et al., 2011). Phenols have been associated with a lowered risk of heart disease via their action towards low-density lipoproteins (LDL) (Vinson et al., 2001). Also, phenols are important because of their contribution to the nutritional quality attributes of fruits and vegetables such as color, stringency, bitterness, and flavor. Phenols and flavonoids are beneficial antioxidants and exhibit scavenging

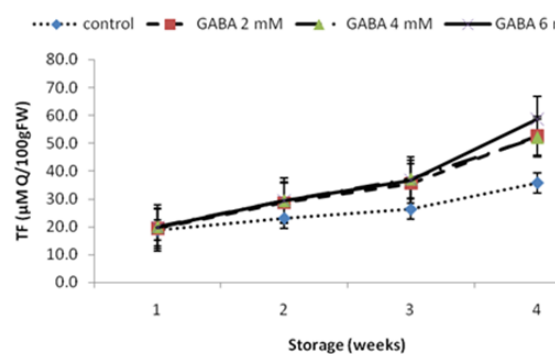


Fig. II. The effects of 0, 2, 4 and 6 mM GABA on total flavonoids (TF) of peach fruits stored at 1°C for 4 weeks. Data shown are Mean values  $\pm$  SD (n=3).

activity of ROS (Hassanpour et al., 2011). We report herein that GABA treatment significantly enhanced total phenols and flavonoids contents in peach fruits. We suggest that the higher total phenols and flavonoids contents in GABA treated peach fruits could be due to the stimulation of PAL enzyme activity and thus triggering the phenyl propanoid pathways. According to present results, we suggest that GABA treatment might be an efficient strategy for improving total phenols and flavonoids contents in fruits such as peach.

Oxidative damage plays an important role in disease initiation and progression in humans (Yamaguchi et al., 1998). Damage is generally reduced by endogenous antioxidants, but additional protection is necessary, and therefore fruits and vegetables phytochemicals are critical for disease prevention. Ascorbic acid (AA) as vitamin in fruits and vegetables is the water-soluble antioxidant (Kinsella et al., 1993). The role of AA is to reduce hydrogen peroxide ( $H_2O_2$ ), which preserves cells against ROS (Davey et al., 2000). Humans are not able to synthesize AA, and fruit and vegetables, especially citrus fruit, strawberries, cornelian cherry, peaches, peppers, tomatoes, cabbage, and spinach are main sources of ascorbic acid (Davey et al., 2000; Hassanpour et al., 2011).

Also, Rao et al. (2011) reported that the treatment of sweet pepper with  $CaCl_2$  inhibited the ascorbic acid oxidase (AAO) enzyme activity, which is responsible for AA oxidation, and enhanced AA content. Their results confirmed Ruoyi et al., (2005) finding which showed that inhibition of AAO activity by  $CaCl_2$  treatment caused delay in the senescence of pepper fruits during storage and preserve AA and anti-browning property (Rao et al., 2011). Also, Wang et al. (2006) suggested that enhanced ascorbate/dehydroascorbate (AA/DHA) and glutathione/glutathione disulfide (GSH/GSSG) ratios in peach fruit treated with SA attributed to increase of cytosolic  $Ca^{+2}$  concentrations and enhanced antioxidant system activity. With respect to enhancement of antioxidant systems activity by GABA, Song et al. (2010) reported that GABA treatment mitigates oxidative stress produced by  $Al^{3+}$  toxicity in barley seedlings by increasing the levels of SOD and CAT activities, which in turn reduces the accumulation of ROS

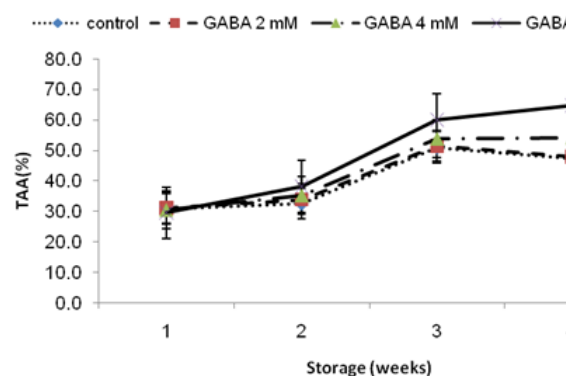


Fig. III. The effects of 0, 2, 4 and 6 mM GABA on DPPH scavenging activity of peach fruits stored at 1°C for 4 weeks. Data shown are Mean values  $\pm$  SD (n=3).

and MDA. Also, Yang et al. (2011) showed that the treatment of peach fruit with 5 mM GABA for 10 min reduces chilling injury and generally enhances the activities of antioxidant enzymes such as SOD, CAT, APX, GPX, GST, MDHAR, and DHAR. According to this report, we suggest that enhanced ascorbic acid content in GABA treated peach fruits may be due to activation of antioxidant systems or inhibition of AAO enzyme activity. Our result in the present research suggest that the GABA could be a potential postharvest treatment to improve the health benefits of peach fruits consumption by increasing the antioxidant capacity.

Our results indicated that GABA treatment significantly enhanced the total phenols, flavonoids, and ascorbic acid contents in the peach fruits. It is therefore presumed that GABA treatment may stimulate the antioxidant activities of the peach fruits. In order to examine this presumption, the effects of GABA on the antioxidant activities of the peach fruits were examined, using scavenging activity on DPPH radicals. The results indicated that the DPPH scavenging activity of the peach fruits treated with GABA were significantly enhanced, showing that GABA treatment stimulated the scavenging activity of fruits on DPPH radical. Taken together, our results suggested that GABA treatment significantly enhanced the antioxidant activity of the peach fruits. Since DPPH scavenging activity is mainly attributed to the phenols, flavonoids as well as ascorbic acid contents (Aghdam et al., 2013), it is suggested that phenols, flavonoids, and ascorbic acid contents make a significant

contribution to the peach fruits antioxidant activity.

## Conclusion

In this study, we report for the first time the positive effect of postharvest GABA treatment on enhancing antioxidant potential of peach fruits, which was accompanied by increased total phenols, flavonoids and ascorbic acid contents as well as improved DPPH scavenging capacity. We suggest that GABA may have potential for commercialization to maintaining postharvest quality attributes and enhancing nutritional value of peaches during cold storage.

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