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Exogenous γ-aminobutyric acid and ascorbic acid application affect quality and biochemical changes of 'Hindi-Besennara' mangoes during ripening

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Effects of γ-aminobutyric acid (GABA) (10 and 20 mM), and ascorbic acid (AsA) (2 and 4%) dipping on quality and biochemical changes of 'Hindi-Besennara' mangoes were studied during ripening for 8 days at 20± 2°C and 60–70% RH. Weight loss increased during ripening to 12.5% and was lower in treated fruit than control with lowest value (8.8%) at 2% AsA. Unmarketable fruit percentage (UFP) was lower after 8 days in treated fruit (21.1 to 24.6%) than control (31.9%). Both GABA and AsA treatment retained higher green color, firmness, titratable acidity (TA) and membrane stability index (MSI) but lower total soluble solids (TSS) and TSS/TA ratio during ripening. Total phenol and flavonoid contents in both peel and pulp were higher during ripening than initial, fluctuated and were higher in treated fruit than control. Vitamin C content was higher in treated fruit than control except for 2% AsA. Radical scavenging capacity (RSC) was lower (higher DPPH IC_{50} values) during ripening in both peel and pulp than initial. RSC of peel showed no changes during ripening but was higher in treated fruit than control. However, RSC of pulp was not affected by treatments but was higher after 8 than 3 and 5 days during ripening. In both peel and pulp, polyphenoloxidase (PPO) activity gradually increased during ripening and was lower in treated fruit than control. While, peroxidase (POD) activity gradually increased during ripening and was higher in treated fruit than control. Polygalacturonase (PG), xylanase and α-amylase activities, in both peel and pulp, showed higher levels than initial, fluctuated during ripening and were lower in treated fruit than control. In conclusion, postharvest dipping in either 10 mM GABA or 2% AsA retard ripening, retained and improved quality of 'Hindi-Besennara' mangoes via inhibiting hydrolytic enzymes and enhancing fruit antioxidant system.

Keywords: Mango, Ripening, Quality, Antioxidant, Enzymes

INTRODUCTION

Mango (*Mangifera indica* L.) is commercially important fruit due to its favorable flavor and nutrition and especially health value as a good source for bioactive natural antioxidant compounds such as vitamins and phenolics (Sivakumar et al. 2011). It is classified as climacteric fruit where ethylene trigger its ripening. Mangoes can be harvested at the mature hard-green stage and resume normal ripening processes following harvest (Sivakumar et al. 2011). The relatively high perishability of mangoes following harvest are mainly due to rapid softening and incidence of physiological disorders that favors pathogens attack (Zhang et al. 2013). As a tropical fruit, mango is susceptible to chilling injuries (CI) which limit its storability, handling and marketing potential (Sivakumar et al. 2011). The approach of

regulating fruit ripening, inducing resistant to pathogens and reduce postharvest losses by physical, biological, and chemical elicitors is a promising alternative to synthetic fungicides (Zhang et al. 2013; Awad et al. 2017). GABA is a natural 4-carbon non-protein amino acid that rapidly accumulate in tissue in response to biotic and abiotic stresses suggesting a role in the defense system (Ramos-Ruiz et al. 2019). Pre-storage of 5 mM GABA dipping alleviated CI in peaches during storage at 1 ºC for up to 5 weeks via enhancing antioxidant enzymes and retaining energy (Yang et al. 2011). GABA dipping at different concentration from 100 to 1000 µg/ml, induced resistance to blue mold via activating a number of antioxidant defensive enzymes as well as their related genes in pears with little negative impact on edible quality (Yu et al. 2014). Wang et al. (2014) reported that vacuum infiltration with 20 mM GABA reduced CI and electrolyte leakage, and increased antioxidant systems of bananas stored at 7 ºC for 20 days. A postharvest treatment with 1 mM GABA increased GABA shunt that providing metabolites to produce energy, reduce power, and help zucchini fruit to cope with cold stress during cold storage (Palma et al. 2019). β-aminobutyric acid (BABA), an isomer for GABA, activated defense-related responses against fungal infection in mangoes (Zhang et al. 2013) and jujubes (Yan et al. 2015). AsA is a naturally occurring organic acid having strong antioxidant and anti-browning activity that frequently used to maintain quality of intact or minimally processed fresh fruit (Yen et al. 2002; Ling et al. 2007). Prasad et al. (2016) reported that pre-storage 150-200 ppm AsA dipping reduced lenticel browning and improved skin appeal of four different mangoes cultivars during ripening. In the Kingdom of Saudi Arabia (KSA), a considerable postharvest loss occur in 'Hindi-Besennara' mangoes, one of the most commercially growing cultivar, due to inappropriate postharvest handling especially with high climatic temperature and humidity which favor rapid softening, browning and pathogens attack (Awad et al. 2017). Therefore, this study aim to evaluate the response of 'Hindi-Besennara' mangoes to postharvest dipping in GABA or AsA as an attempt to regulate postharvest ripening, retain and improve quality at ambient conditions.

MATERIALS AND METHODS

Plant materials and experimental procedure

In 2019 growing season, uniform samples of 'Hindi-Besennara' mangoes were harvested at mature hard-green stage from a commercial orchard in Jizan region (17.4751° N, 42.7076° E), KSA. Fruit were packed in perforated cardbox (12 fruit of each box, about 3.0-3.5 kg) and transported to the postharvest laboratory at King Abdulaziz University in Jeddah within about 8 h at 15 ^{o}C. Fruit of uniform size, weight (200-250 g/fruit) and appearance and free of visual defects were selected for this experiment.

Fruit treatments

A completely randomized experimental design with three replicates (35 fruit of each) for each treatment was setup. Fruit of each treatment/replicate were drenched either into water (control), 10 or 20 mM GABA, 2 or 4% AsA for 10 min at ambient conditions for 10 min. The wetting agent Tween 20 at 1ml/l was added to all treatments solutions. After air draying of about 1 h, all treatments/replicates were stored at 20±2 ^{o}C and 60–70% (RH) in perforated cardboard cartons for 8 days. A separate three replicates (5 fruits of each) for each treatment were stored at the same conditions and periodically weighed (at 0, 3, 5 and 8 days) for loss in weight calculation and expressed in percentage. At the beginning of storage (0 days), another three samples (5 fruit of each) were randomly collected for initial quality and biochemical measurements as detailed below. After 3, 5 and 8 days during ripening, random samples (5 fruit) per replicate were taken for quality and biochemical measurements as indicated below. Following peel color and pulp firmness measurements, samples of both pulp and peel were taken, sliced and stored at –80 ^{o}C for enzyme, total phenol, total flavonoid and antioxidant capacity determinations. Additional portion of fruit pulp were directly used for TA, pH, TSS and vitamin C measurements.

Unmarketable fruit percentage (UFP)

Fruit that showed browning and shriveling of about 10% of its total surface area (visually estimated) or fungal infection symptoms was considered unmarketable and was calculated on initial fruit number basis for each samples and expressed in percentage.

Peel color measurement

Peel color was measured independently in 5 randomly selected fruit per replicate by a Minolta Chroma Meter CR-410 (Minolta Camera Co. Ltd., Osaka, Japan). The values of L*, a* and b* were measured in the middle of each of the five fruit/replicate. Chroma = $(a^{*2}+b^{*2})^{½}$ which

represented the hypotenuse of a right triangle with values ranging from 0 = least intense to 60 = most intense. The chroma values indicate the saturation of the color.

Firmness, TSS, TA, pH and vitamin C measurements in fruit pulp

Pulp firmness of fruit was measured independently in 5 fruit (two opposite measurements in the middle of each fruit) per replicate by a digital basic force gauge, model BFG 50N (Mecmesin, Sterling, Virginia, USA) supplemented with a probe of 11 mm diameter and the results were expressed as Newton. A homogeneous sample was prepared from these 5 fruit per replicate for measuring TSS content, TA, pH and vitamin C concentration. TSS content was measured in fruit pulp juice with a digital refractometer (Pocket Refractometer PAL 3, ATAGO, Japan) and expressed in percentage. TA was determined in distilled water diluted fruit juice (1: 2) by titrating with 0.1N sodium hydroxide up to pH 8.2, using automatic titrator (HI 902, HANNA Instrument, USA) and the results expressed as a percentage of citric acid. Fruit juice pH was measured by a pH meter (WTW 82382, Weilheim, Germany). Vitamin C was measured by titrating juice sample with freshly prepared dye solution of 2,6-dichlorophenol-indophenol until pink color and the results expressed as g Kg^{-1} on a fresh weight (FW) basis (Ranganna, 2000).

Leakage of ions from peel

Leakage of ions was measured in peel disks according to Awad et al. (2017) and was expressed as membrane stability index percentage (MSI %)

Preparation of methanol extract of peel and pulp

Two grams of fruit peel and pulp (randomly collected from 5 fruit/replicate) were extracted by shaking at 150 rpm for 12 h with 20 ml methanol (80%) and filtered with Whatman No. 1 paper. The filtrate designated as methanol extract that was used for total phenols, total flavonoids and antioxidant activity estimations.

Estimation of total phenol and flavonoid contents

Total phenol content was measured according to Hoff and Singleton (1977) as detailed in Awad et al. (2017). Total flavonoid content was determined according to Zhishen et al. (1999) as detailed in Awad et al. (2017).

Evaluation of DPPH radical scavenging assay of peel and pulp

Free radical scavenging activity of methanol extract of peel and pulp was determined using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) method (Ao et al. 2008) as described in in Awad et al. (2017).

Enzymes measurements of peel and pulp

Crude extract

One gram of fruit peel and pulp (randomly collected from 5 fruit/replicate) was homogenized with 20 mM Tris–HCl buffer, pH 7.2 using homogenizer. The homogenate was centrifuged at 10,000 rpm for 10 min at 4 ºC. The supernatant was designed as crude extract and stored at -20 ºC for peroxidase, polyphenoloxidase, polygalacturonase, xylanase and *α*-amylase assay.

Polyphenoloxidase assay

Polyphenoloxidase (EC 1.14.18.1) (PPO) activity was assayed with catechol as a substrate according to the spectrophotometric procedure of Jiang et al. (2002) as detailed in Awad et al (2017).

Peroxidase assay

Peroxidase (EC 1.11.1.7) activity (POD) was assayed according to Miranda et al. (1995) as detailed in Awad et al (2017).

Polygalacturonase, α-amylase and xylanase assays

Polygalacturonase (EC 3.2.1.15) (PG), *α*-amylase (EC 3.2.1.1) and xylanase (EC 3.2.1.8) activities were assayed by determining the liberated reducing end products using galacturonic acid, maltose and xylose, respectively as standards (Miller 1959) as detailed in Awad et al. (2017).

Statistical analysis

The data were statistically analyzed as a completely randomized design with three replicates by analysis of variance (two ways ANOVA) using the statistical package software SAS (SAS Institute Inc., 2000, Cary, NC., USA). Comparisons between means were made by the Duncan's multiple range test at $P \leq 5\%$.

RESULTS

In case of significant interactions between treatments and ripening period, we passed over the main effects and focused instead on the interactions and vice versa. Weight loss.

Table 1. Weight loss, firmness, total soluble solids (TSS), pH and membrane stability index (MSI) of 'Hindi-Besennara' mangoes during ripening period (RP) as affected by postharvest γ-amino butyric acid (GABA) and ascorbic acid (AsA) dipping.

	Weight loss	Firmness	TSS	pH	MSI
	(%)	(N)	(%)		(Index)
Initial	*0.0*	*7.80*	*3.4*	*2.44*	*46.6*
Treatments (T)					
Control	9.7a	2.50c	17.7a	5.60	11.6b
GABA (mM)					
10	9.0bc	2.87b	15.8c	5.66	17.4a
20	9.1b	2.94b	17.0ab	5.86	16.9a
AsA (%)					
2	8.8c	3.25a	15.9c	5.67	16.1a
4	9.0bc	3.22a	16.1bc	5.95	18.0a
F-test	***	***	***	NS	**
RP (d)					
3	6.0c	4.23a	15.3b	5.48b	25.4a
5	8.9b	2.59b	16.8a	5.62b	11.7b
8	12.5a	2.06c	17.5a	6.14a	11.0b
F-test	***	***	***	***	***
T x SL					
F-test	NS	NS	NS	NS	NS

Means within each column followed by the same letter are not significantly different at level $P \leq 0.05$. (**) and (***), significant at $P \leq 0.01$ and 0.001, respectively; (NS), not significant.

Table 2. The interaction effect between treatments and ripening period on unmarketable fruit percentage (UFP), titratable acidity (TA) and total soluble solids (TSS)/TA ratio of 'Hindi-Besennara' mangoes as affected by postharvest γ-amino butyric acid (GABA) and ascorbic acid (AsA) dipping.

	UFP	TA	TSS/TA
	(%)	(%)	(Ratio)
Initial	*0.0*	*3.14*	*1.1*
After 3 days			
Treatments			
Control	0.0e	1.06b	15.7fg
GABA (mM)			
10	0.0e	1.47a	10.5g
20	0.0e	1.48a	10.9g
AsA (%)			
2	0.0e	1.43a	9.5g
4	0.0e	1.47a	10.2g
After 5 days			
Control	6.7d	0.32f	55.3b
GABA (mM)			
10	5.1d	0.82c	19.3f
20	5.6d	0.54e	32.6d
AsA (%)			
2	5.2d	0.67d	25.8e
4	0.0e	0.47e	35.0d
After 8 days			
Control	31.9a	0.30f	63.0a
GABA (mM)			
10	24.6b	0.34f	48.0c
20	21.1c	0.31f	57.1ab
AsA (%)			
2	23.3bc	0.36f	48.5c
4	21.1c	0.32f	53.0bc

Means within each column followed by the same letter are not significantly different at level $P \leq 0.05$. percentage gradually increased during ripening reaching 12.5% after 8 days and was significantly lower in all treatments than control. The lowest weight loss value (8.8) was registered in 2% AsA treatment (Table 1).

Firmness gradually decreased during ripening and was higher in in all treatments than control. AsA treatments showed significant higher fruit firmness than GABA treatments. TSS increased during ripening and was significantly lower in all treatments than control, except for 20 mM GABA treatment that was similar to control. The pH values increased during ripening but were not significantly affected by the applied treatments. MSI of fruit peel sharply decreased during ripening and was significantly higher in all treatments than control (Table 1). UFP only recorded after 5 days during ripening in the control and GABA treatments, while 4% AsA treated fruit exhibited no browning, shriveling or fungal infection symptoms (Table 2). After 8 days during ripening, UFP was significantly lower in all treatments especially at 20 mM GABA and 4% AsA that showed the lowest value (21.1%) than control (31.9%). TA concentration decreased during ripening and was significantly higher in all treatments after 3 and 5 days than control.

Table 3. Minolta color values L*,a*,b* and chroma of 'Hindi-Besennara' mangoes during ripening period (RP) as affected by postharvest γ-amino butyric acid (GABA) and ascorbic acid (AsA) dipping.

	L*	a*	b*	Chroma
Initial	*50.6*	*-15.3*	*27.1*	*31.2*
Treatments (T)				
Control	51.2	-9.8a	27.0b	28.9b
GABA (mM)				
10	50.1	-11.4b	26.6b	29.0b
20	49.9	-11.8b	28.5a	30.9a
AsA (%)				
2	49.7	-11.8b	27.6ab	30.1a
4	50.3	-11.7b	27.7ab	30.2a
F-test	NS	***	*	**
RP (d)				
3	50.1	-12.8c	24.8c	27.9c
5	50.0	-11.7b	27.2b	29.6b
8	50.7	-9.4a	30.4a	31.9a
F-test	NS	***	***	***
T x SL				
F-test	NS	NS	NS	NS

Means within each column followed by the same letter are not significantly different at level $P \leq 0.05$. (*), (**) and (***), significant at $P \leq 0.05$, 0.01 and 0.001, respectively; (NS), not significant.

However, after 8 days of ripening, there were no significant differences among treatments in TA concentration. TSS/TA ratio increased during ripening and was lower in all treatments, after 5 and 8 days than control except for 20 mM GABA that exhibited a similar ratio to control (Table 2).

The L* values were not significantly affected either by ripening period or by the applied treatments. The values of a* increased during ripening and were lower in all treatments than control (Table 3). The values of b* significantly increased during ripening and were not affected by the applied treatments except for 20 mM GABA that showed higher values than control. The chroma values increased during ripening and were higher in all treatments except for 10 mM GABA that gave similar values to control (Table 3). Both of peel total phenol and pulp total flavonoid concentrations showed higher values during ripening than initial, fluctuated and were higher in all treatments than control after 3, 5 and 8 days of ripening (Table 4). Vitamin C concentration showed lower values during ripening than initial and was significantly higher in all treatments than control except for 2% AsA that exhibited a similar level to control after 5 days. There were no significant differences in vitamin C concentration among treatments after 3 and 8 days of ripening (Table 4). Both of total phenol concentration of fruit pulp and total flavonoid of peel showed higher values than initial, fluctuated during ripening, and were significantly higher in treated fruit especially at high rates than control (Table 5). RSC was lower (higher DPPH IC_{50} values) during ripening in both peel and pulp than initial. RSC of fruit peel showed no significant changes during ripening but was higher (lower DPPH IC_{50} values) in treated fruit than control. However, RSC of fruit pulp were not significantly affected by the applied treatments but was higher after 8 days of ripening than other periods (Table 5). PPO activity of fruit peel gradually increased during ripening and was lower in GABA and AsA treated fruit than control (Table 6). POD activity of fruit peel gradually increased during ripening and was higher in GABA and AsA treated fruit than control. PG activity of peel showed higher level than initial, fluctuated during ripening and was significantly lower in GABA and AsA treated fruit than control. α-amylase activity of peel showed higher level than initial, fluctuated during ripening and was significantly lower in GABA and AsA treated fruit than control. Xylanase activity of fruit peel slightly changed during ripening and was significantly lower in GABA and AsA treated fruit than control (Table 6).

Table 4. The interaction effect between treatments and ripening period on peel total phenol (TPC), pulp total flavonoid (TFC) and vitamin C concentrations (g $Kg^{-1}FW$) of 'Hindi-Besennara' mangoes as affected by postharvest γ-amino butyric acid (GABA) and ascorbic acid (AsA) dipping.

	TPC (peel)	TFC (pulp)	Vitamin C (pulp)
Initial	*2.47*	*0.22*	*0.61*
After 3 days			
Treatments			
Control	2.74e	0.28j	0.55ab
GABA (mM)			
10	3.18d	0.41i	0.58a
20	3.19d	0.50gh	0.53b
AsA (%)			
2	3.40bcd	0.45hi	0.45d
4	3.46bcd	0.59de	0.50bcd
After 5 days			
Control	3.38cd	0.56efg	0.38e
GABA (mM)			
10	4.49a	0.72b	0.48cd
20	4.66a	0.81a	0.51bc
AsA (%)			
2	4.56a	0.68bc	0.36ef
4	4.68a	0.80a	0.50bcd
After 8 days			
Control	2.84e	0.44hi	0.34ef
GABA (mM)			
10	3.24d	0.57def	0.33f
20	3.72b	0.61de	0.34ef
AsA (%)			
2	3.38cd	0.63cd	0.32f
4	3.59bc	0.59de	0.39e

Means within each column followed by the same letter are not significantly different at level $P \leq 0.05$.

Table 5. Pulp total phenol (TPC) and peel total flavonoid TFC) concentrations (g $Kg^{-1}FW$), and radical scavenging capacity (RSC) (DPPH IC_{50} values) of 'Hindi-Besennara' mangoes during ripening period (RP) as affected by postharvest γ-amino butyric acid (GABA) and ascorbic acid (AsA) dipping.

	TPC (pulp)	TFC (peel)	RSC (peel)	RSC (pulp)
Initial	*0.78*	*0.40*	*7.6*	*3.3*
Treatments (T)				
Control	1.20c	066c	9.6a	5.1
GABA (mM)				
10	1.47b	0.81b	7.6b	5.2
20	1.56a	0.87ab	8.1b	5.4
AsA (%)				
2	1.46b	0.82b	8.0b	5.1
4	1.60a	0.93a	7.8b	4.7
F-test	***	***	***	NS
RP (d)				
3	1.34b	0.69c	8.2	5.2a
5	1.76a	0.97a	8.1	5.4a
8	1.27b	0.79b	8.3	4.7b
F-test	***	***	NS	***
T x SL				
F-test	NS	NS	NS	NS

Means within each column followed by the same letter are not significantly different at level $P \leq 0.05$. (***), significant at $P \leq 0.001$; (NS), not significant.

Table 6. Antioxidant and hydrolytic enzymes activity (U min g FW) in peel and pulp of 'Hindi-Besennara' mangoes during ripening period (RP) as affected by postharvest γ-amino butyric acid (GABA) and ascorbic acid (AsA) dipping.

	PPO	POD	PG	α-amylase	Xylanase
Peel					
Initial	*33.3*	*1.17*	*0.57*	*0.39*	*0.60*
Treatments (T)					
Control	59.2a	1.99b	0.93a	0.50a	0.75a
GABA (mM)					
10	50.3b	2.71a	0.82b	0.44b	0.64b
20	49.1b	2.58a	0.81b	0.42bc	0.60bc
AsA (%)					
2	49.8b	2.45a	0.84b	0.42bc	0.62b
4	50.8b	2.61a	0.79b	0.40c	0.57c
F-test	***	***	***	***	***
RP (d)					
3	42.2c	1.98c	0.82b	0.40b	0.60b
5	53.7b	2.53b	1.04a	0.53a	0.62b
8	59.7a	2.90a	0.65c	0.38b	0.68a
F-test	***	***	***	***	***
T x SL					
F-test	NS	NS	NS	NS	NS
Pulp					
Initial	*40.7*	*0.97*	*0.49*	*0.28*	*0.55*
Treatments (T)					
Control	69.1a	1.91c	0.87a	0.37a	0.69a
GABA (mM)					
10	57.9b	2.32b	0.74b	0.31b	0.63b
20	54.2b	2.32b	0.69bc	0.29bc	0.58c
AsA (%)					
2	55.8b	2.35b	0.69c	0.30bc	0.61bc
4	54.2b	2.65a	0.61d	0.28c	0.57c
F-test	***	***	***	***	***
RP (d)					
3	51.5b	1.69c	0.78b	0.29b	0.58c
5	59.7a	2.30b	0.86a	0.38a	0.61b
8	63.5a	2.94a	0.52c	0.27c	0.66a
F-test	***	***	***	***	***
T x SL					
F-test	NS	NS	NS	NS	NS

PPO, POD and PG refereeing to polyphenoloxidase, peroxidase and polygalacturornase, respectively. Means within each column followed by the same letter are not significantly different at level $P \leq 0.05$. (***), significant at $P \leq 0.001$; (NS), not significant.

PPO activity of fruit pulp increased during ripening and was lower in GABA and AsA treated fruit than control (Table 6). POD activity of fruit pulp gradually increased during ripening and was higher in GABA and AsA treated fruit than control. PG activity of fruit pulp showed higher level than initial, fluctuated during ripening and was significantly lower in GABA and AsA treated fruit than control. α-amylase activity of fruit pulp showed higher level than initial, fluctuated during ripening and was significantly lower in GABA and AsA treated fruit than control. Xylanase activity of fruit pulp gradually increased during ripening and was significantly lower in GABA and AsA treated fruit than control (Table 6)

DISCUSSION

As a typical climacteric fruit, mango express

high metabolic activities leading to rapid softening with increase in TSS and a decrease in TA during ripening at ambient conditions (Sivakumar et al. 2011). Thus, searching for natural elicitors such as GABA and AsA to regulate fruit ripening and reduce postharvest losses are critically required. Weight loss is an important quality factor that affect fruit integrity and freshness. In the current experiment, 'Hindi-Besennara' mangoes exhibited a continuous weight loss increase during ripening. However, both GABA and AsA treated fruit showed slightly, but significantly, lower weight loss than control, with no significant difference between 10 and 20 mM (Table 1). The loss in fruit weight is mainly attributed to transpiration and respiration processes during ripening (Narayana et al. 1996; Razzaq et al. 2015). GABA and AsA treatments reduced weight loss during ripening possibly via slowing down metabolic activities of fruit. A postharvest 1 mM GABA dipping reduced weight and electrolyte leakage and increased chilling tolerant of zucchini fruit during cold storage (Palma et al. 2019). Unfortunately, respiration rate and ethylene production were not measured in the current experiment. Indeed, a high concentration of GABA (20 mM) suppressed ethylene biosynthesis in detached soybean leaves more effectively than the ACC oxidase inhibitors AVG and cobalt (Turano et al. 1997). In tomato plants, both *α*- and *β*- isomers (BABA) of γ-aminobutyric acid induced ethylene evolution, while GABA did not (Cohen et al. 1994). AsA dipping treatment, especially at 200 ppm, reduced lenticels browning, ethylene production and respiration rate in four mango cultivars stored at ambient conditions for 10 days (Prasad et al. 2016). In overall, both GABA and AsA treatments retarded ripening of 'Hindi-Besennara' mangoes during 8 days, as reflected by higher peel green color (lower a* values), MSI, firmness and TA, and lower TSS content and TSS/TA ratio than control especially after 3 and 5 days during ripening (Tables 1, 2 and 3). Similarly, 100 ppm AsA dipping delayed guavas ripening during storage at 6-8 °C (Gill et al., 2014), 1% isoascorbic acid dipping retarded litchis ripening at ambient conditions (Kumar et al. 2013), and 5% AsA mixed with Aloe vera gel dipping retarded strawberries ripening during storage at 1 °C (Sogvar et al. 2016). However, regarding GABA effects, it has been reported that 25-100 mM of its isomer BABA dipping reduced decay with no effect on firmness, respiration and ethylene production as well as TSS and TA in 'Guifei' mangoes during storage at 25 ^{o}C (Zhang et al. 2013). The observed positive effects of GABA and AsA on delaying fruit ripening might be due to their general preservative effects as anti-stress factors. Fruit ripening and senescence are considered an oxidative process in which the transition from maturation into ripening/senescence stage is accompanied by a progressive shift toward an oxidative state (Goulao and Oliveira, 2008). AsA has been found to retain cell wall integrity and play a protective role against reactive oxygen species (ROS) that rise in climacteric fruit during ripening (Akram et al. 2017). GABA and AsA treated fruit retained higher pulp firmness and peel MSI and lower UFP (Tables 1 and 2). These results might be ascribed to the inhibition of hydrolytic enzymes activities PG, α-amylase and xylanase as well as the activation of the antioxidant system as POD activity and increasing total phenol and flavonoid contents in both fruit peel and pulp (Tables 4-6). Accordingly, cell wall membrane stability is associated with the activities of cell wall hydrolytic enzymes namely cellulase, PG, α-amylase and xylanase (Hurber, 1983). AsA treatment at 4% reduced UFP compared to control (Table 2) confirming those of Gill et al. (2014) on guavas, Sogvar et al. (2016) on strawberries, and Kumar et al. (2013) on litchis. AsA protect fruit against oxidative damage and biotic stress by pathogen attack during ripening by enhancing antioxidant defense system that retard softening and decay (Ling et al. 2007). AsA treated fruit exhibited higher POD and lower PPO in both peel and pulp than control (Table 6). Similarly, dipping in 1% isoascorbic acid alone or in combination with 0.1% *N*-acetyl cysteine decreased PPO activity and browning of litchi fruit during shelf life at ambient conditions (Liu et al. 2006; Kumar et al. 2013). However, relatively low concentrations (100 to 200 ppm) of AsA dipping, decreased PPO, POD and lipoxygenase in four mango cultivars stored at ambient conditions for 10 days (Prasad et al., 2016). GABA and its isomer BABA have been suggested as potential natural elicitors inducing pathogens and chilling resistance as well as improving cell wall membrane stability of horticultural commodities during storage (Zhang et al. 2013; Wang et al. 2014; Yan et al. 2015; Ramos-Ruiz et al. 2019; Palma et al., 2019). Our results showed that GABA treated fruit exhibited lower UFP, retained higher antioxidant enzyme such as POD but lower PPO, as well as higher antioxidants (total phenol, flavonoid and vitamin C) and MSI during SL than control. In confirmation, dipping in 100 to 1000 µg/ml GABA induced resistance to blue mold via activating a number of antioxidant defensive enzymes as well as their related genes in pears with little negative impact on edible quality

(Yu et al. 2014). Vacuum infiltration with 20 mM GABA reduced CI and electrolyte leakage, and increased antioxidant systems of bananas stored at 7 ºC for 20 days (Wang et al. 2014). Similar results were also reported by using BABA isomer on both mango (Zhang et al. 2013) and jujube (Yan et al., 2015) fruits. Generally, the possible mode of action (s) of GABA and its isomers BABA includes a) induction of pathogenesis-related proteins, b) induction of phenylpropanoid biosynthetic pathway, and c) activation of antioxidative enzymes such as POD, superoxide dismutase and catalase that modulate ROS metabolism in several fruit including mangoes (Cohen et al. 2010; Yang et al. 2011; Zhang et al. 2013; Wang et al. 2014; Yan et al. 2015; Ramos-Ruiz et al. 2019; Palma et al. 2019). A part from the indirect effect on enhancing the antioxidant system of fruit, GABA itself possess a remarkable scavenging ability of ROS under stress conditions (Liu et al. 2011). Mangoes are rich in antioxidant compounds including phenolics, carotenoids and vitamins (Sivakumar et al. 2011; Razzaq et al. 2015; Al-Qurashi and Awad, 2018). Vitamin C content showed lower values during ripening than initial and was higher in all treatments than control except for 2% AsA that exhibited a similar level to control after 5 days (Table 4). However, vitamin C content has been reported to decrease in peaches, remain almost constant in kiwi, and to increase in tomatoes during ripening (Locato et al. 2013). Compared to initial, total phenol and flavonoid content increased with fluctuation during ripening and was higher in GABA and AsA treated fruit than control (Tables 4 and 5). In another study, total phenol content of 'Choke anan' mangoes peel decreased during storage at 6 °C for 10 days but was not changed in fruit stored at 12 °C while, DPPH-radical scavenging activity increased at both storage temperatures (Kondo et al., 2005). Both GABA and AsA treated fruit exhibited slightly but significantly higher FRSC (lower DPPH IC_{50} values) in fruit peel despite the higher contents of total phenol and flavonoid as well as vitamin C compared to control (Tables 4 and 5). In partial confirmation, 20 mM GABA treatment increased both total phenol content and antioxidant capacity as assayed by FRAP and DPPH scavenging activity in banana fruit peel during 20 days of storage at 7 ºC (Wang et al. 2014). AsA also retained higher antioxidants content (total phenol, flavonoid and vitamin C) during ripening (Tables 4 and 5). Similar results were recorded in 'Yali' pears treated by 10 mM AsA (Ling et al. 2007), guavas treated by 100 ppm AsA (Gill et al. 2014) and strawberries treated by 5% AsA mixed with Aloe vera gel (Sogvar et al. 2016). The increase in total phenol and flavonoid contents with the decrease in RSC during SL (Tables 4 and 5) suggest qualitative changes in antioxidant compounds toward a lower antioxidant potential. In another fleshy climacteric fruit, Fernando et al. (2014) found no significant correlation between total phenol content and antioxidant activity measured by DPPH and ferric reducing antioxidant power (FRAP) assays, except for vitamin C level and FRAP in 'Khai' banana pulp. It was reported that phenolics antioxidant capacity possibly has a concentration saturation limit above which the activity could not increase further with the concentration (Dani et al. 2012). In addition, other compounds such as cartotenoids, vitamins and some minerals are possibly synergistically contribute to RSC of fruit with phenolics. Accordingly, parallel several assays should be used to investigate the principles of antioxidant/oxidation activity of a certain horticultural commodity.

CONCLUSION

In overall, it is concluded that postharvest dipping in either 10 mM GABA or 2% AsA retarded ripening, retained and improved quality of 'Hindi-Besennara' mangoes via inhibiting hydrolytic enzymes and enhancing fruit antioxidant system. Also, such treatments decreased decayed and unmarketable fruit percentage (UFP) during 8 days ripening.

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

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AUTHOR CONTRIBUTIONS

All authors contributed equally to this manuscript

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