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The effect of Salicylic acid and Aspirin Treatments on Enzymes Activity and Fruit Quality of Clementine Mandarin Fruits during Different Cold Storage Periods

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ABSTRACT

A considerable tendency is found nowadays to replace dangerous chemicals with natural compounds for having beneficial effects on catching free radical species or ROS. The activity of CAT, POX, APX enzymes and Fruit Quality were determined in cold stored Clementine Mandarin Fruits treated with Salicylic acid "SA" and Aspirin "ASA" over two seasons. Fruits were dipped in either SA or ASA solution at (*1mM, 2mM*) and stored for 45 days at 40°F with R.H. (85-90 %), where quality parameters and enzymes activity were evaluated. Results showed that *2mM* SA delayed weight loss and was more effective in reducing fruit decay% followed by *2mM* ASA up to 45 days. Data also revealed that decreasing in fruit SSC contents was accompanied by increasing SA dose as compared with the highest SSC values for control followed by low doses of SA and ASA. Meanwhile, total acidity slightly declined during experimental periods up to 45 days of storage with non significant differences. Accordingly, SSC / Acid values indicate that low doses of SA or ASA maintained high fruit quality over the same time period. High SA or ASA doses prevented the destruction of ascorbic acid content than that of lower concentrations; whereas the storage period progressed the decrease in Ascorbic acid content was faster with 2mM ASA. after 45 days of storage, CAT and APX activities and accumulation of phenol production recorded Maximum values with *2m*MSA followed by 2m MASA. Meanwhile, phenols were gradually decreased and equally affected by *1Mm* SA or *2mM* ASA treatment. Additionally, control treatment reflected the lower Phenol content with higher POX and MDA levels. So, it could be suggested that, high concentration (*2mM*) of SA or ASA were more effective in reducing fruit losses and decay and keeping quality of cold stored Clementine Mandarin fruits.

Key words: Mandarin fruits, Enzymes activity, MDA, SA, ASA, Cold storage, Fruit losses and quality.

Introduction

Clementine is a popular variety of mandarins (*Citrus reticulata* Blanco) that matures early with pronounced nutritional and medicinal values mainly attributed to the presence of bioactive compounds (Esna & Zokaee, 2011 and David *et al*., 2013). Mandarins as softer citrus fruit are spoil more quickly through the activity of both physiological and pathological disorders that develops storage weight loss, fruit decay and rind breakdown (Esna and Zokaee, 2011) and loss of flavor quality (David *et al*., 2013). Mandarins can be kept in good quality for only 15–30 days under ambient storage conditions (Chanikan *et al*, 2015). Avoiding spoilage aspects can be minimize by cold storage which extend the storage life of mandarins up to 45 days with good quality, availability period and remunerative prices to growers, (Obenland, *et al.,* 2011). California Citrus Research org. (CCR, 2017) found wide differences in sensitive of mandarin varieties to chilling ranged between 41 - 46 F (5-8 ^{o}C). Published studies cleared that cold storage reduced losses of fruit weight and decay %, decreased titratable acidity, continually increased TSS as well as TSS/acid ratio and slowed down enzyme activities which resulted in delaying declining of vitamin C contents (Roongruangsri *et al.,*2013 on tangarins, Chanikan *et al .,* 2015. on mango and Obenland, *et al.,* 2011 on mandarins). Salicylic acid as eco- friendly materials (endogenous hormone and simple phenolic compound, $C_7H_6O_3$) are involved in regulation of various processes in plant including induction of plant defense against cold stress that causes chilling injury in stored fruits

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i.e, Pineapple (Lu *et al.,* 2011); Pomegranates (Serrano *et al*., 2012); Peach (Khademi and Ershadi, 2013); fruits (Muzammil *et al*., 2014) ; Tomato, (Baninaiem, *et al*., 2016). Generally, environmental stress frequently reduced plant development through over production of reactive oxygen species (ROS) which damage various macromolecules and cellular structures (Apel and Hirt, 2004), cellular membrane, photosynthetic apparatus and enzymes (Lukatkin, 2003) and lead to the death of cells (Liu *et al.,* 2010; Mekki *et al.,* 2010; Orabi and AbdelHamid, 2016). These ROS, such as hydrogen peroxide, superoxide and hydroxyl ions, resulting in oxidative damage at the cellular level (Mekki and Orabi, 2007; Hussein and Orabi, 2008; Hussein *et al*., 2009; Goud and Kachole, 2011). It is worthy to mention that, ROS may play two very different roles: exacerbating damage or signaling the activation of defense responses (Yi *et al.,* 2014). Since, ROS in low concentration act as signaling molecules mediating a variety of physiological responses, including stomatal movement and gene expression (Yi *et al.,* 2014; Orabi *et al.,* 2017a). Meanwhile, over accumulation of ROS damage almost all cell components including membrane lipids, chloroplasts, pigments, enzymes and nucleic acids (Goud and Kachole, 2011; Orabi *et al.,* 2014). Plants protect cells and sub-cellular systems from the cytotoxic effects of these active oxygen radicals with both non- enzymatic and enzymatic antioxidant systems such as carotenoids, ascorbic acid, a-tocopherol, proline, SOD, peroxidase and catalase (Orabi 2004; Orabi and Mekki, 2008).

ROS radicals (H_2O_2, O_2 and OH) are recorded as important features of plant metabolism and in plant responses to stress and its oxidative stresses are imposed on fruit tissues (Raskin, 1992) The potentially toxic product (H_2O_2) server as a signal molecule during various abiotic stresses and could be reduced by a number of enzymes such as POX, CAT, APX and GR (Goud and Kachole, 2011). SA as (endogenous hormone and simple phenolic compound, $C_7H_6O_3$)) that are synthesized by plants involved in regulation of various processes of plant including producing higher polyamines level which reduces ethylene action and decreases activity of cell wall degrading enzymes that increase postharvest cycle and delay fruit ripening (Khademi and Ershadi, 2013). Meanwhile (Sayyari *et al.,* 2011) suggests that information on the role of SA in thermo-genesis and disease resistance meets some qualifying criteria for a plant hormone. Salicylic acid is not the same thing as aspirin. The two medicines are related, and have similar-sounding chemical names. Aspirin, a trade name for acetylsalicylic acid (ASA), undergoes spontaneous hydrolysis to SA. Exogenously applied it is rapidly converted to SA. Despite the fact that aspirin was not identified as a natural product, it is widely used by many scientists in their experiments according to the similarity of their physiological effects. Most investigators are focusing on the role of Aspirin in induction of disease resistance, while the roles of keeping certain quality characteristics of stored fruit are still not clear (Losanka *et al*., 1997). Recently, it has been observed that salicylic acid (SA) treatment could be used to reduce deterioration and chilling injury aspects in some fruits and vegetables (Orabi *et al.,* 2010; Sayyari *et al*., 2011; Orabi *et al.,* 2015).

Therefore, the objective of this study was to investigate the influence of salicylic acid and Aspirin (as eco-friendly materials) in different concentrations on the levels of bioactive compounds and storage quality of Calmantin mandarin fruit through different cold storage periods.

Materials and Methods

This investigation was carried out through (2015& 2016) seasons on mature Clementine mandarin fruits (*C. Clementina*) to study the effect of Salicylic acid and Aspirin concentrations on fruit quality during different cold storage periods.

Plant material: Uniform fruits were obtained from the selling port of experimental research station of NRC. Fruits were transported to the laboratory of ADS project, Faculty of Agriculture, Cairo University. On arrival, fruit uniform in size with orange color surface >1/3 and juice SSC 7.0 ± 0.1% were graded, washed, air dried and treated. Experimental fruits were divided into five similar groups (72 fruits/ each), dipped for 10 min in fresh solution of one of the following treatments:

1- Salicylic acid (1mM) 2- Salicylic acid (2 mM) 3- aspirin (1mM) 4- Aspirin (2mM) 5- Water for control. Thereafter, fruits were allowed to dry at room temperature, packed in carton boxes, where each treatment replicated three times for either physical or chemical properties and stored for 45 days at 40º F. with R.H. (85-90 %). Three representative replicates for each sampling date, were taken for physical and chemical analysis, as follows:

A-Physical properties:

1- Loss in fruit weight percentage: The initial weight of each box was individually recorded before storage and re- weighted at each sampling date. Fruit weight loss percentage was calculated according to the following equation: $\frac{Wi - Ws}{Wi} \times 100$

Where; Wi = initial fruit weight and Ws = fruit weight at the sampling date.

2-Fruit decay percentage:

Were calculated according to the equation:

$$\frac{\text{Total number of decayed fruits}}{100} \times \text{Initial number of stored fruits}$$

B-Chemical properties:

1- Soluble Solids Content (SSC %): was determined in fruit juice by a hand referactometer.
2-Titratable acidity %: (expressed as citric acid) was determined using 10*ml* of the extracted juice, diluted to 100 *ml* and titrated against 0.1 N NaOH to pH 8.1.
3- SSC/Acid ratio: was calculated by dividing SSC/Acid values as maturity index.
4- Ascorbic acid content (vitamin C): it was determined using phenol indophenol dye method, 10g of the fresh samples were blended with 3% metaphosphoric acetic acid extracting solution to homogenous slurry, and 5ml of the filtrate extract were titrated with standard Indophenol to pink end point.

Total phenols in fruit juice extract were determined by the colorimetric method of folin.– Denis as described by Daniel and George, (1972) results were expressed as mg gallic acid/ g fw.

Antioxidant Enzymes and lipid peroxidation were assayed as follow:

At the end of cold storage periods, extraction of the antioxidant enzymes were used for estimation the activity of antioxidant enzymes [Catalase(CAT, EC1.11.1.6.), Guaiacol Peroxidase(POX,EC 1.11.1.7), Ascorpate Peroxidase(APX EC1.11.1.11) as follow: 5g of frozen tissues were homogenized in pre-chilled mortar in presence of 10 *ml* of 50 mM potassium phosphate buffer (PH7) with 1% (W/V) insoluble polyvinyl pyrolidone (PVP) and 0.1 mM EDTA. The extraction procedures were repeated twice and supernatants were pooled, raised to a certain volume, referred as crude enzyme extract, all operation were carried out at -4°C for further analysis. APX and POX activity were determined according to Nakano and Asada, (1981)**.** One unit of APX was defined as the amount of enzyme that breaks down 1µ mol of ascorbate per min. The activity of POX was estimated by the increase of absorbance at 470 nm due to oxidation of guaiacol in the presence of H_2O_2 and formation of tetraguaiacol. Catalase Enzyme activity was assayed using a modification method of (Aebi, 1983). The activity of CAT was estimated by the decrease of absorbance at 240 nm for 1 min as a consequence of H_2O_2 consumption. Lipid peroxidation was determined by measuring Malondialdehyde (MDA) content (Dhindsa *et al.,* 1982). The activities was expressed as: µ mole/ g F. Wt.

Statistical Analysis:

The data obtained were subjected to standard analysis of variance procedure according to Snedecor and Cochran, (1980). The values of L.S.D. were calculated whenever F values were significant at 5% level.

Results and Discussion

Effect of Salicylic acid and Aspirin concentrations on:

1- Effect on fruit weight loss

Data in Table 1 show that different treatments significantly reduced fruit weight loss percentages during different storage periods than control. Maximum losses were observed in control fruits that lost

8.56% of their weight after 45 days of storage, meanwhile Salicylic acid at 2mM was the most effective treatment that delayed weight loss up to 45 days of storage to record 4.84%. Furthermore, no significant difference was detected between Salicylic acid 1mM and Aspirin 2mM treatment Particularly after15 days of storage where at the storage end period, Aspirin 2mM dominated in this respect and arranged the second (5.13%) followed by Salicylic acid 2mM (5.62%). Data concerning ASA and SA are in line with those of Sartaj *et al*., (2013) who reported that higher concentrations of salicylic acid that role as an electron donor produces free radical which decrease normal respiration and transpiration as well as stomata closing (Manthe *et al*., 1992).

2- Effect on fruit decay percentages

Data in Table 1 revealed that tested treatments significantly reduced fruit decay percentages during different storage intervals as compared with control. A negative relationship was observed between concentration of treatments and fruit decay percentages where control fruits recorded the highest decay values after 15, 30 and 45 days (3.44, 6.02 and 10.92%, respectively). Moreover, Salicylic acid 2mM and Aspirin 2mM treatments were significantly equal in their reducing effect up to 15 days, meanwhile Salicylic acid 2mM became the more effective in reducing decay than either Aspirin 2mM (4.09 % and 6.77%) or other treatments to record (3.86 and 6.43) after 30 and 45 storage days, respectively. Results appeared that exogenous application of SA and ASA on at tested concentrations provided efficient control of decay caused by postharvest pathogens. Khademi and Ershadi (2013) illustrated that SA and ASA treatments decreases activity of cell wall degrading enzymes that increase postharvest cycle and delay fruit ripening. Also, fruit ethylene production could be effectively decreased by SA and ASA treatments accompanied with cell swelling (Serrano *et al.,* 2012) and inducing systemic resistances against postharvest pathogen which extend storability of fruits with higher antioxidant activity that activates natural defense mechanism (Muzammil *et al.,* 2014).

Table 1: Effect of Salicylic acid and Aspirin concentrations on Loss in fruit weight and Decay % of mandarin fruits under cold storage conditions.

Treatments	Cold storage/ days					
	15	30	45	15	30	45
	Loss in fruit weight %			**Decay %**		
Salicylic acid 1mM	1.59 c	3.03 c	5.62 c	2.60 c	4.52 c	8.07 c
Salicylic acid 2mM	1.19 d	2.41 e	4.84 e	2.11d	3.86 e	6.43 e
Aspirin 1mM	1.64 b	3.25 b	6.05 b	2.86 b	5.54 b	9.58 b
Aspirin 2mM	1.47 c	2.98 d	5.13 d	2.29 d	4.09 d	6.77 d
Control	3.24 a	5.43 a	8.56 a	3.44 a	6.02 a	10.92 a

Similar letters in each column indicate non-significant differences at P _ 0.05

3- Effect on soluble solid contents (SSC).

As for SSC content, data in general showed gradual increase in SSC during storage periods of different treatments as well as control as shown in (Table 2). Significant variance was observed among 15, 30, 45 storage days and treatments. Where, control treatment significantly recorded the highest SSC values (9.65 and 10.15 %) for15 and 30 days and equally followed in descending order Salicylic acid 1mM and Aspirin 1mM as compared to high tested concentrations. Meanwhile, after 45 days of storage low concentrations of Salicylic acid 1mM followed by Aspirin 1mM dominated than high ones to record 11.79 and 11.28%. In addition, un-treated fruits were significantly higher than both Salicylic acid 2 mM and Aspirin 2mM. The increased values in soluble solids could be due to fruit weight loss and subsequently fruit juice concentration (Khademi and Ershadi, 2013). Where, the decrease in sucrose synthesis in treated fruits may result in lowered enzyme activity that reduced ethylene production (Alejandra *et al*., 2017). Contrarily, Ranjbaran *et al*., (2011) on grapes reported that SA treatment had no effect on SSC.

4- Effect on total acid contents (as citric acid): TA

After 15 days of storage, TA ratio was high in fruits treated with Salicylic acid 1mM and decreased with slower rate during 30 and 45 days to record non significant values as compared with other treatments and control. In general, TA was slightly declined during different experimental periods up to 45 days of storage; however either SA or ASA treatments had no significant influence on TA of fruits over the storage time. These results are in line with those reported by Alejandra *et al*., (2017) and Chanikan *et al*., (2015) who illustrated that the slight decline in TA was probably due to the slow rate of respiration and metabolic processes converting citric acid into sugars as a function of applied SA.

Table 2: Effect of Salicylic acid and Aspirin concentrations on SSC % and Total acidity % of mandarin fruits under cold storage conditions.

Treatments	Cold storage/ days					
	15	30	45	15	30	45
	SSC %			**Total acidity % (as citric acid)**		
Salicylic acid 1mM	9.60 b	10.22 a	11.79 a	1.03 ^{NS}	1.02^{NS}	0.96 ^{NS}
Salicylic acid 2mM	9.51 c	9.83 c	10.77 e	1.04 ^{NS}	1.01^{NS}	0.98 ^{NS}
Aspirin 1mM	9.57 b	10.16 a	11.28 b	1.02 ^{NS}	1.01^{NS}	0.95 ^{NS}
Aspirin 2mM	9.52 c	9.98 b	10.94 d	1.03 ^{NS}	1.00 ^{NS}	0.96 ^{NS}
Control	9.65 a	10.15 a	11.17 c	1.02 ^{NS}	1.00^{NS}	0.97 ^{NS}

Similar letters in each column indicate non-significant differences at *P _ 0.05*

5- Effect on SSC / Acid

Data in Table 3 revealed that there was a gradual increase in SSC/ acid with significant variance between treatments and also storage periods. The decline in total acid contents led to increases in SSC/acid over the same time period. After 15 and 30 days of storage, observed in fruits treated with Salicylic acid 1mM (12.28%) followed by Aspirin 1mM to arrange the second in this respect. In addition, the high concentration of Salicylic acid or Aspirin reflected lower SSC/acid values as compared with low concentration ones. This indicates that salicylic acid 1mM and Aspirin 1mMtreatments maintained high fruit quality. Our results agree with those of Sartaj *et al*., (2013) who found that 2mM of salicylic acid was effective in retaining keeping quality of apricot up to 12 days at ambient storage.

Table 3: Effect of Salicylic acid and Aspirin concentrations on SSC / Acid and Ascorbic acid Contents of mandarin fruits under cold storage conditions.

Treatments	Cold storage/ days					
	15	30	45	15	30	45
	SSC / Acid			**Ascorbic acid *mg/100 ml.***		
Salicylic acid mM_1	9.32 c	10.02 b	12.28 a	38.4 c	35.9 c	32.8 c
Salicylic acid mM_2	9.14 e	9.73 c	10.99 d	43.5 a	41.4 a	39.6 a
Aspirin mM1	9.38 b	10.06 b	11.87 b	35.7 d	34.4 d	29.9 d
Aspirin mM_2	9.24 d	9.98 b	11.40 c	41.2 b	39.5 b	36.8 b
Control	9.46 a	10.15 a	11.51 c	33.9 e	32.4 e	29.2 e

Similar letters in each column indicate non-significant differences at P _ 0.05.

6- Effect on Ascorbic acid

During storage, ascorbic acid (AA) appeared a consistent decrease in tested fruits with significant correlation between storage and treatments as shown in Table (3). AA content of both Salicylic acid **2**mM and Aspirin 2mM treatments slightly decreased with significantly higher contents than that of lower concentrations. Moreover, the decrease in AA content was faster with Aspirin 2mM (36.8 *mg*) as the storage period progressed to arrange the second after Salicylic acid 2mM (39.6 *mg*). Fruits treated

with low Salicylic acid concentration were richer in AA contents (32.8*mg*) than both Aspirin 1mM (29.9 *mg*) and control (29.2 *mg*). Lu *et al*. (2011) reported that SA delayed the decline of ascorbic acid content and prevented its destruction, so improve the fruit quality.

Effect on Enzyme activities and total phenolic contents

A- Effect on Total Phenolic Contents

The data in Table 4 shows the relationship between treatments was observed after 45 days of storage and partially induced accumulation of phenol compounds to record Maximum values in fruits treated with SA at high concentration of 2 mM. (1.66 *mg*) compared to different treatments and gradually decreased to record equal effect in fruits treated by Salicylic acid mM_1 (1.41 *mg*) or Aspirin ^{2} treatment (1.40 *mg*). Furthermore, control treatment induced the lower Phenol content (1.12 *mg*). Awad, (2013) reported that total phenol content in peach fruits were increased by increasing SA rates. Additionally, Lu *et al*., (2011) concluded that SA treatments inhibited the activities of PPO and PAL, thus reduced Total Phenol production and delayed conversion. The Phenolic Potential is to act as an antioxidant mostly due to their properties as hydrogen donors for reduction and quenchers of singlet O_2 (Rice- Evans *et al*., 1997). Phenolic synthesis is affected in general by different biotic / abiotic stress including chilling (Orabi *et al*., 2014,2015).Under low temperature stress, the increased synthesis of phenolic compounds is a response of plants to overcome Chilling injury through synthesizing poly phenolic phytotoxins, by an increase of the activity of phenylalanine ammonia lyase added to low level of poly phenol oxidase activity as a trial to reduce the oxidation of phenolic substrates to quinones (Lattanzio *et al*., 2009). Several authors demonstrated a great positive relationship exists between the content of total phenols and the antioxidant activity of fruits and vegetables (Maciel *et al*., 2011; Abd EL- Motty and Orabi, 2013; Orabi *et al.,* 2016, 2017b)

B- Effect on Catalase Enzymes activity

After 45 days of storage, significant reduction in CAT activity was associated with control fruits (12.42). In addition, Salicylic acid 2mM induced great enhancement in CAT activity of treated fruits (27.40). However, CAT value was significantly higher with little difference in fruits treated with Aspirin 2mM (21.66) followed by Salicylic acid 1mM (19.43). SA was found to enhance the efficiency of antioxidant system in plants as it applied exogenously at suitable concentrations (Hayat *et al.,* 2010).

C- Effect on Guaiacol Peroxidase (POX) activity

After 45 storage days, a significant correlation was found between reduction in (POX) activity and Aspirin or Salicylic acid concentrations. Meanwhile un-treated fruits followed by Aspirin 1mM recorded higher (POX) activity, moderate effect with no significant difference was observed between Salicylic acid 1mM and Aspirin 2mM in term of (POX) activity.

D- Effect on ascorbate peroxidase activity (APX):

Fruits treated with 2mM Salicylic acid exhibited APX activity to reach higher values than those of different treatments. Also aspirin at 2 mM enhanced APX activity to arrange the second in this respect. Contrarily, low concentration treatments of either Salicylic acid or Aspirin revealed less (APX) activity effect as compared with high ones. Where, fruits in control treatment recorded the lowest activity values. With regard to the activity of the antioxidant enzymes such as catalase (CAT) and ascorbate peroxidase (APX), activities dramatically increased due to cold stress. Cold- induced enhancement of CAT activity in mandarin fruits indicated that it had a higher capacity for the decomposition of H_2O_2 generated by SOD and can represent a central protective role in H_2O_2 scavenging process where it is the main scavenger of the strong oxidant H_2O_2 in peroxisomes to convert H_2O_2 to water and molecular oxygen (Bahari *et al*., 2013). Generally, increases of CAT activity are a strategy for improving chilling tolerance (Orabi, 2004; Orabi *et al*., 2010, 2015)

In concern to APX, the high activity of APX under chilling stress was in agreement with those reported by Orabi, (2004); Orabi *et al.,* (2010) APX is a primary enzyme of the ASA – GSH cycle that suppresses the accumulation of H_2O_2 to water(Orabi *et al.,* 2017c). APX has a high affinity for H_2O_2 than CAT and POX and it may have a more crucial role in the management of ROS stress or may be responsible for the fine modulation of ROS signaling ascorbate peroxidase enzymatic action attained through reduction of H_2O_2 using ascorbate as an electron donor (Orabi and El-Noemani, 2015).

The improvement of stress tolerance is mostly related to the enhancement of the activities of the antioxidant enzymes in plant (Orabi, 2004) as CAT & APX enzymes in this study to induce a protective effect on plants under stress whereas, high level of POX activity in untreated fruits was an indication for stress damage (Orabi and AbdelHamid, 2016) imply that fruits were damaged by chilling stress.

Table 4: Effect of Salicylic acid and Aspirin concentrations on Total phenols and enzymes activity of mandarin fruits under cold Storage conditions

Treatments	Cold storage after 45 days				
	Total phenols ***mg/g F.Wt.***	**Catalase Enzymes activity**	**POX** ***Guaiacol Peroxidase***	**APX** ***ascorpate Peroxidase***	**MDA** ***lipid peroxidation***
Salicylic acid 1mM	1.41b	19.43 c	0.66 c	4.20 c	5.42 c
Salicylic acid 2mM	1.66 a	27.40 a	0.48 d	4.63 a	4.28 e
Aspirin 1mM	1.29 c	15.29 d	0.70 b	3.70 d	6.16 b
Aspirin 2mM	1.40 b	21.66 b	0.61 c	4.51 b	4.94 d
Control	1.12 d	12.42 e	0.78 a	2.93 e	6.88 a

*enzymes activity as : µ mole / g f. wt.
** Similar letters in each column indicate non-significant differences at *P _ 0.05.*

E- Effect on MDA lipid peroxidation

Evidences suggest that pre-storage treatment with SA or ASA concs. was potentially effective in inhibiting MDA (lipid peroxidation) in treated mandarin fruits. MDA was highly activated in control fruits followed in descending order by Aspirin 1mM and Salicylic acid 1mM with significant differences. However, lipid peroxidation was highly reduced in fruits treated with Salicylic acid 2mM followed by Aspirin 2mM.

In this study, the increment in MDA content in untreated Mandarin fruit confirming the occurrence of Oxidative damage.

However, SA at 2 mM followed by 2 mM aspirin then 1 mM obviously reduced MDA accumulation and exhibiting a good potential to alleviate oxidative stress lowering of lipid peroxidation or ROS in general by these Treatments were reported by Mulu *et al*.,(2013) for barly; and Abd El hamid *et al.* (2016) for wheat under cold stress

SA produced the firmest fruits (Baninaiem *et al*., 2016). SA applicative is useful in inhibiting tissue softening in fruits by reducing cell wall hydrolases activities and maintaining consistency of the cell membrane (Supapvanich, 2015). Wei *et al.* (2011) mentioned that SA application enhances defense mechanisms and antioxidants production during fruit storage in order to decrease MDA content of the cell membrane to maintain cell membrane structure . Moreover Babalar *et al.,* (2007) and Shafiee *et al.,* (2010) suggested that pri and post SA application on strawberry could decrease the softening and keep fruits firm during storage. Generally, rapid softening of fruits during ripening was simultaneous with rapid decrease in the endogenous SA of fruits (Wang et al, 2006) SA through enhancing the antioxidant potential could maintain membrane consistency (Baninaiem *et al.,* 2016) through storage to prevent chilling injury (Solemani Aghdam *et al.,* 2012).

Salicylates are major component act as signal transduction pathways of plants to achieve disease resistance (Asghari and Soleimani, 2010). In general, SA applied to either plant's vegetative stage, fruit development stage or postharvest stage could control decay completely and increased fruit shelf life (Yao and Tian, 2005).

Conclusion

As a whole, the current study showed that postharvest treatments with Salicylic acid or Aspirin are effective to extend storability and postharvest life of mandarin fruits at 40°F. SA at 2 mM was the best treatment for reducing losses of fruit quality during the storage period

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