Role of aminoethoxyvinylglycine in creasing of sweet orange [*Citrus sinensis* (L.) Osbeck] fruit

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Key Message: Creasing in sweet oranges is a serious threat to oranges industry of world. In the current study, creasing was minimized with the application of ethylene inhibitor aminoethoxyvinylglycine.

Abstract: Creasing is a physiological disorder in the rind of sweet orange [*Citrus sinensis* (L.) Osbeck] fruit and causes serious economic losses. The current study was conducted at Westralian fruit farm Gingin, WA-Australia during year 2011 and 2012. In this study, aminoethoxyvinylglycine (AVG) a reversible ethylene inhibitor with different concentrations (0, 20, 40 and 60 mg/l) was sprayed at fruit set (FS), golf ball (GB) and colour break (CB) stage of fruit development to alleviate creasing as well as to promote rind properties of sweet orange cultivars viz. Washington Navel and Lane Late fruit. Both experiments were conducted under randomized complete block design (RCBD) with two factors (treatments and stages of AVG spray). The fruit were randomly collected from a tree (35-fruits/replication) and were collected randomly from the tree to check the albedo breakdown and fruit rind characteristics. The AVG spray was effective to reduce creasing and improve the fruit quality. The creasing was significantly reduced when AVG (60 mg/l) was sprayed at GB (27.95 and 24.30%) stage with respect to control (52.15 and 51.54%) in Washington Navel. The AVG application at FS stage (22.86%) was more effective than the control (51.43%) in cv. Lane Late sweet orange during second harvest season. In conclusion application of AVG significantly alleviates the creasing (%) and improves the textural properties of sweet oranges cultivars. © 2020 Department of Agricultural Sciences, AIOU

Keywords: Aminoethoxyvinylglycine (AVG), Creasing, Ethylene inhibitor, Fruit quality, Sweet orange

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Introduction

Sweet orange belongs to genus citrus (which is an important genus for flowering plants) and family Rutaceae. It is originated from subtropical as well as tropical regions in the world (Ismail & Zhang, 2004) and many species thought to be native of China. However, the genus citrus includes sweet orange, mandarins, lime lemon and grape fruit. According to area and production, citrus ranked second fruit in the whole world. Sweet orange is leading fruit group grown all over the world (FAOSTAT, 2013). Australia is the leading orange producing country in the world which produced 1000 metric tonnes during the year 2018-2019 as well as Australia is the 6th largest exporter of sweet oranges with value (\$230,387,000) during the year 2018 (Workman, 2019). Quality is determined by the fruit firmness, smoothness, gloss and colour. However, the sweet orange fruit is attractive due to its fruit rind. The citrus fruit rind is composed of white tissues known as albedo and orange tissues which are known as flavedo. In creasing, albedo tissues of fruit rind prone to rupture and groves occur on the surface of flavedo in oranges (Monselise et al., 1976). It affects diverse varieties of citrus like Washington Navel, Valencia, Navelina and Nova mandarins (Hussain, 2014; Saleem et al., 2014; Hussain & Singh, 2015 a&b). However, creasing was first time

reported by Le-Roux and Crous (1938) in South Africa during 1938. Now, creasing is prime issue in South Africa, USA, China, Spain and Uruguay as well as in Australia (Bower, 2004; Greenberg et al., 2006; Hussain, 2014; Hussain and Singh, 2015 a&b) and all these countries are famous for the production of orange in the world. According to an estimate, more than 50% Australian sweet orange loss occurs due to creasing (Bower, 2004). It is very difficult to detect creasing at the initial stage of fruit growth. However, it is easily detectable at maturity and the CB stage of fruit development (Monselise et al., 1976). There are many factors responsible for the development of creasing like fruit size, rind thickness, deficiency of elements, fruit position, load and irrigation (Bower, 2004; Treeby et al., 2007). Although, it has been previously described that ethylene is involved for creasing in oranges (Monselise et al., 1976; Pham, 2009: Hussain and Singh, 2015b), but the existing information is unconvincing.

It is well known that citrus fruit is in non-climacteric nature, which has limited both ethylene and respiration process. Ethylene is used as ripening and colour changing agents in fruits (Bleecker, 2000). Rath and Prentice (2004); Ladaniya (2007) stated that softening of fruits and vegetables is due to the presence of ethylene which also causes decaying in cell membranes. The application of ethephone or ethrel as ethylene source significantly

improved the ripening, respiration and color changes in Navel oranges (Augusti et al., 2002; Burg, 2004; Ladaniya, 2007). However, it is reported previously that ethylene plays a key role in creasing of oranges (Hussain, 2014; Hussain & Singh, 2015b).

Ethylene biosynthesis can be inhibited by the use of cobalt sulphate and polyamines (PAs). Rath et al. (2004) reported that AVG is used as an ethylene biosynthesis blocker which stops the activities of ethylene biosynthesis enzymes like 1-aminocyclopropane-1-carboxylic acid (ACC) synthase in plant tissues. AVG is commonly available in the market in the name of ReTainTM which is used for pre-harvest for improving physiological disorders such as fruit drop in different fruit plants especially temperate fruits (Rath et al., 2004). Therefore, it was hypothesized that AVG applications at fruit set, golf ball or at colour break stage retard the ethylene biosynthesis due to which it may reduce the creasing (%) by improving the textural properties of sweet orange including cv. Washington Navel and Lane Late.

Materials and Methods

The current research work conducted on commercial citrus groove which is located in Gingin (latitude 21° 31° S, longitude 55° 155° E) Western Australia during the year 2011 and 2012 as well as research started from fruit set (Fruit diameter: 15±5 mm), the golf ball (Fruit diameter 40±5 mm) or at the colour break stage (Fruit diameter: 80±5 mm). 25-year old uniform oranges trees of Washington Navel and Lane Late were used in both experiments. Trifoliate orange is a common rootstock which is used for grafting Navel oranges in Australia. The climate is dominated by cool wet winter and hot dry summer. The average annual maximum temperature (41.2 and 41 °C) was reported during the year 2011 and 2012, respectively. While minimum average annual temperature was (26.2 and 26.0 °C) in both years (Bureau of Meteorology [BoM], 2013). The average daily and annual rainfall is given in Fig. 1.



Fig. 1 Average annual rainfall and evapotranspiration (mm) in Gingin, Western Australia (Strobach et al., 2014)

In the 1st experiment, AVG (0, 20, 40 and 60 mg/l) was sprayed at FS (Fruit size: 15 ± 5 mm), GB (Fruit size 40 ± 5 mm) and CB stage (Fruit size: 80 ± 5 mm) of Washington Navel by spraying with hand sprayer. The data regarding on creasing percentage, textural properties of fruit rind and fruit quality parameters was studied. However, 2nd year research trial was done on Lane Late sweet orange by using the same concentration of AVG at same fruit developmental stages and same parameters were recorded as described in 1st experiment. Both experiments were conducted under randomized complete block design (RCBD) with two factors (treatments and stages of AVG spray) and replicated four times.

Creasing (%)

At ripening stage, 35 fruits randomly harvested from each replication from both cultivars, and creasing (%) was noted as described by Pham (2009):

 $Creasing (\%) = \frac{(Total number of creased fruit) \times 100}{Total number of fruit assessed}$

Textural properties

Textural properties were recorded with the help of textural analyzer.

Compression force (N)

The compression force measured with the help of textural analyzer. 10-fruit from each replication were selected with 75 mm height with 200 mm/min speed.

Rind hardness test (N)

Different 10-fruit per replication were selected for rind hardness. The fruit rind was peeled (2.5 cm wide \times 0.6 cm thick) by slicer (Zyliss slice 2 folding Mandolin Slicer, Swiss). A 4 mm diameter cylinder probe was used to check rind hardness, attached to the load cell and all the samples were placed onto the flat plate. Hardness was the first penetration of probe at speed of 100 mm/min and expressed as force (N).

Rind tensile strength test (N)

To measure the tensile strength, rind of oranges carefully removed at a size of $(2.5 \times 5 \text{ cm} \text{ area with } 6 \text{ mm} \text{ thickness})$ and was measured at the maximum load and limit points where the rind deflection occurred.

Measurement of fruit weight, fruit diameter and rind thickness

Ten fruit were selected from randomly harvested fruit to measure weight with electrical balance and mean weight was calculated by dividing ten on total fruit weight and expressed in gram (g). Similarly, the fruit diameter and rind thickness of selected fruit were determined by using digital Vernier calliper from each treatment and expressed in mm.

Statistical analysis

Two-way analysis of variance (ANOVA) was used to analyse the data, using GenStat 14th edition. The impacts of different treatments, time of application and their connections were evaluated using least significant difference (LSD) test at 5% probability level (Steel et al., 1997).

Results

Creasing

Creasing (%) significantly alleviated by the foliar spray application of AVG at FS, GB or CB stage in both cultivars of sweet oranges in both harvest seasons (Fig. 1). The results showed that exogenous applications of AVG reduced creasing (%) regardless of spray application at FS, GB and CB stage in Washington Navel in 2011 and 2012. The same trend was also noted in Lane Late sweet orange in harvest season 2. The result showed that creasing (%) significantly decreased by increasing the concentration of AVG in both cultivars of sweet oranges. When we compared the stage of treatment, GB was more effective stage of application than FS and CB in Washington Navel sweet orange during the years 2011 and 2012.

Fruit compression force

Foliar spray of AVG considerably enhanced the compression force of fruit irrespective of its application at FS, GB and CB stage in both seasons of harvest (Fig. 2 and Fig. 3) in both cultivars of sweet oranges. Similarly, the application AVG (40-60 mg/l) showed higher compression force than control in both cultivars at harvest season 1. However, in harvest season 2, all the spray applications prominently enhanced the fruit compression force compared with the control in both cultivars. Similarly, all spray applications of AVG were effective when sprayed at FS, GB and CB stages. However, the CB stage of application resulted higher fruit compression force (310.20 N) with respect to FS (262.40 N) and GB (269.20 N) in cultivar Washington Navel sweet orange in the year 2011.

The treatment and stage of spray application significantly affected the fruit compression force.

Rind hardness

The results showed that hardness of rind increased with the spray application of AVG at the FS, GB and CB during both harvest seasons of Washington Navel and Lane Late oranges (Fig. 2 and Fig. 3). AVG showed significant increase in mean hardness with respect to control in Lane Late in the year 2011 only. Similar findings were observed at treatment 60 mg/l of AVG than the untreated trees and 20-40 mg/l AVG in Washington Navel during harvest season in 2011.

Rind tensile strength

The data showed that AVG significantly improvement tensile strength at FS, GB and CB stage in harvest season 1 and 2 (Fig. 2 and Fig. 3). The results showed that 20-60 mg/l AVG resulted substantially improvement in tensile force than the control sweet orange Lane Late in harvest season 1. Similar findings were observed in harvest season 2 in both cultivars. When all the treatments were compared, the FS stage (71.42 N) was more effective stage of AVG application followed by its application at the GB (57.85 N) and CB stage (59.97 N) in sweet orange Lane Late in harvest season 1. The application of AVG at different stages considerably increased the tensile force in cultivars of sweet orange. However, the best treatment for rind tensile force was AVG (60 mg/l) as compared to all other treatments and control.

Fruit weight

The result showed a considerably increase in weight by the application of AVG at different fruit developmental stages such as FS, GB and CB in sweet oranges (Table 1). When average over treatments, then results showed that mean fruit weight increased with the increase of AVG concentrations in both cultivars during both seasons. However, the 60 mg/l AVG substantially enhanced the fruit weight (279.80 and 253.61 g) regarding control (263.61 and 232.20 g) in Washington Navel in both seasons. Similar findings were observed in cv. Lane Late. Similarly, when we compared the stage of application, the FS increased fruit weight than GB and CB stages irrespective of cultivars and seasons. The non-significant (p < 0.05) interactions were noted between treatments and stage application.

Fruit diameter

The results showed that application of AVG affected the fruit diameter in both cultivars on fruit developmental stages. When we compared the effect of AVG treatments related to its stage of application, then results showed that mean fruit diameter increased with the increase in AVG concentration in both cultivars in both seasons. AVG (60 mg/l) substantially enhanced the fruit size (84.69 and 81.98 mm) and (80.93 and 83.02 mm) regarding control (77.61 and 73.46 mm) and (75.76 and 78.67 mm) in both

cultivars. Similarly, when we compared the stage of application, the GB stage was more effective stage of AVG application regarding fruit diameter (82.71 mm) followed by CB (81.37 mm) and FS (81.01 mm) in Washington

Navel in both seasons. However, CB (82.93 mm) stage was more effective than FS (82.59 mm) and GB (79.38 mm) in Lane Late Sweet orange.



Fig.1 Effect of different concentrations of AVG sprayed at fruit set, golf ball or colour break on creasing (%) on sweet orange cv. Washington Navel and Lane Late during 2011 and 2012. n = 4 replications (35 fruit per replication), LSD ($P \le 0.05$) for Washington Navel 2011 treatments = 3.22, stage = 2.79, treatments x stage = ns; Washington Navel 2012 = treatments = 6.54, stage = 5.67, treatments x stage = ns; LSD (P < 0.05) for Lane Late 2011, treatments = 7.28, stage = ns, treatments x stage = ns; Lane Late 2012 = treatments = 8.40, stage = ns, treatments x stage = Ns; Ns = not-significant.

Rind thickness

The rind thickness was increased when AVG applied at FS, GB and CB stage in the both cultivars during both seasons (Table 3). When the effect of AVG treatments related to its stage of application was compared, the results showed that mean rind thickness increased with the increase in concentration in both cultivars during both seasons. However, AVG (60 mg/l) showed substantial improvement in thickness of rind (5.18 and 5.43 mm) and (4.77 and 5.18 mm) regarding control (4.69 and 4.92 mm) and 4.31 and 4.69 mm) during both harvest seasons in cultivars. Similarly, when we compared the stage of spray

application, the CB stage resulted higher rind thickness (5.10 and 5.28 mm) followed by GB (4.95 and 4.92 mm) and CB (3.61 and 4.87 mm) in cultivar Lane Late during both seasons. In Washington Navel, CB (5.28 mm) stage was more effective stage of AVG application followed by GB (4.92 mm) and FS (4.87 mm) in harvest season 1 only. In harvest season 2, GB (5.35 mm) stage was more effective than CB (5.15 mm) and FS (5.06 mm) stage in Washington Navel. However, GB stage was more effective stage of AVG application (5.35 mm) with respect to the CB (5.15 mm) and FS (5.06 mm) during the year 2012 in Washington Navel.



Fig. 2 Effect of different concentrations of AVG sprayed at fruit set, golf ball or colour break on rheological properties of rind sweet orange cv. on Washington Navel fruit during 2011 and 2012. n = 4 replications (5 fruit per replication). Vertical bars represent standard error means. LSD ($P \le 0.05$) for 2011; fruit compression force Treatments = ns, Stage = 19.33, Treatments x stage = ns; for rind hardness, Treatments = 2.67, Stage = ns, treatments x stage = ns; for rind tensile strength, Treatments = 7.24, Stage = ns, Treatments x stage = ns; ($P \le 0.05$) for year 2012; fruit compression force Treatments = 20.16, Stage = ns, Treatments x stage = ns; for rind hardness, Treatments = 2.81, Stage = ns, treatments x stage = ns; for rind tensile strength, Treatments = 5.39, Stage = ns, Treatments x stage = Ns = not-significant.

Discussion

The incidence of creasing was significantly alleviated by the treatments of AVG at FS, GB and GB in both cultivars of oranges in both consecutive years 2011 and 2012. The higher treatment of 60 mg/l AVG substantially reduced the creasing (%) in both cultivars of oranges during both harvest seasons. Similarly, the GB stage was more suitable stage for AVG application. However, the GB (22.86%) stage was more effective followed by FS (29.29%) as compared to control (51.43 and 59.29%) during harvest season 1 and harvest season 2 in cv. Lane Late. It has been reported previously that different factors like temperature, production, fruit position, rind thickness, rootstocks and endogenous production of ethylene are responsible for creasing in oranges (Pham, 2009).



Fig. 3 Effect of different concentrations of AVG sprayed at fruit set, golf ball or colour break on rheological properties of rind sweet orange cv. on Lane Late during 2011 and 2012. n = 4 replications (5 fruit per replication). Vertical bars represent standard error means. LSD ($P \le 0.05$) for 2011; fruit compression force Treatments = 26.13, Stage = ns, Treatments x stage = ns; for rind hardness, Treatments = 0.85, Stage = ns, treatments x stage = ns; for rind tensile strength, Treatments = 2.15, Stage = 1.86, Treatments x stage = ns; ($P \le 0.05$) for year 2012; fruit compression force Treatments = 20.15, Stage = 17.46, Treatments x stage = ns; for rind hardness, Treatments = 2.81, Stage = ns, treatments x stage = ns; for rind tensile strength, Treatments = 5.39, Stage = ns, Treatments x stage = ns = not-significant.

It has been previously claimed that incidence of creasing was substantially increased with the exogenous application of ethylene. It has been reported by Hyodo and Nishino (1981) who found softening enzymes in the creased oranges fruit. Similarly, the findings of Hyodo and Nishino, (1981); Saleem et al. (2014) also supported the findings of Hussain (2014); Hussain & Singh (2015 b). AVG is an irreversible inhibitor of ethylene biosynthesis,

which prevents ethylene production in plant tissues by retarding the activity of ACC synthase (Ladaniya, 2007). Similarly, another ethylene inhibitor putrescine significant reduces creasing in oranges (Hussain, 2014; Hussain & Singh, 2015a). Possibly, creasing (%) was decreased by the decline in the activities of softening enzymes with spray application of AVG.

Table 1 Effect of AVG spray on fruit weight of sweet orange cv. Washington Navel and Lane Late

Fruit weight (g)										
Washington Navel										
	Fruit set		Golf ball		Colour break		Mean (Treatments)			
Treatments	2011	2012	2011	2012	2011	11 2012 2011 20				
Control	270.20 ^{ab}	234.80 ^{ab}	257.20 ^b	242.50 ^{ab}	263.20 ^{ab}	219.20 ^b	263.61 ^b	232.20 ^b		
AVG (20 mg/l)	271.80 ^{ab}	237.50 ^{ab}	263.21 ^{ab}	244.20 ^{ab}	273.00 ^{ab}	234.00 ^{ab}	269.31 ^{ab}	238.61 ^{ab}		
AVG (40 mg/l)	274.51 ^{ab}	238.20 ^{ab}	272.50 ^{ab}	261.70 ^a	278.30 ^{ab}	245.81 ^{ab}	275.10 ^a	248.60 ^{ab}		
AVG (60 mg/l)	279.20 ^a	246.20 ^{ab}	281.00 ^a	265.50 ^a	279.00 ^{ab}	249.00 ^{ab}	279.80 ^a	253.61 ^a		
Mean (stage)	273.90 ^a	239.20 ^{ab}	268.51 ^a	253.50 ^a	273.41 ^a	237.00 ^b				
LSD $(p < 0.05)$										
Treatments (T)			10.8	16.95						
Stage (S)			Ns	14.68						
T×S			Ns	Ns						
Lane Late										
	Fruit set		Golf ball		Colour break		Mean (Treatments)			
Treatments	2011	2012	2011	2012	2011	2012	2011	2012		
Control	265.2 ^{ab}	270.20^{ab}	263.20 ^b	247.21 ^b	263.80 ^b	268.20 ^{ab}	264.10 ^b	261.90 ^b		
AVG (20 mg/l)	278.20 ^{ab}	281.80^{a}	272.0 ^{ab}	270.80 ^{ab}	269.51 ^{ab}	283.00 ^a	273.20 ^b	278.51 ^a		
AVG (40 mg/l)	292.00 ^{ab}	287.01 ^a	272.2 ^{ab}	272.50 ^a	273.20 ^{ab}	283.30 ^a	279.21 ^{ab}	280.90^{a}		
AVG (60 mg/l)	295.50 ^{ab}	294.20 ^a	298.2 ^a	276.00 ^a	280.49 ^{ab}	281.50 ^a	291.40 ^a	283.90 ^a		
Mean (stage)	282.80 ^a	283.31 ^a	276.41 ^a	266.61 ^b	271.80 ^a	279.00 ^a				
LSD (p < 0.05)										
Treatments (T)			16.92	12.92						
Stage (S)			Ns	22.37						
T×S			Ns	Ns						

n = 4 replications (10 fruit per replication), any two mean within a column and within a row followed by different letters are significantly different; Ns = not-significant

It has been reported that ethylene inhibitors such cobalt sulphate and putrescine are used to block the ethylene biosynthesis in the plant tissues. Similarly, AVG is also used to block the ethylene synthesis in fruit tissues through blocking the ACC synthase and ACC oxidase as well as minimised the activities of softening enzymes (Hussain, 2014). The current study revealed that AVG played a significant rule in textural properties of sweet oranges fruit rind. However, the treatments 20-60 mg/l of AVG enhanced the textural properties of fruit rind at all stages of spray application during both harvest seasons in both cultivars. It may be argued that AVG enhanced the textural properties of sweet oranges by suppressing the activities of cell wall degrading enzymes. Bregoli et al. (2002) found that AVG also inhibits the activities of S-adenosylmethionine decarboxylase (SAMDC). It has been previously reported that ethylene inhibitor significantly improved the textural properties of papaya (Hanif et al., 2020), mango (Zahedi et al., 2019), Kanzi apples (Gwanpua et al., 2016), strawberry (Paniagua et al., 2016), blueberries (Chen et al., 2015) and Murcot mandarin (Enab et al., 2020).

Fruit weight, diameter and rind thickness substantially increased with the increase in AVG when applied at FS, GB and GB during both seasons of application. Possibly, it may be argued that the application of AVG subsequently delayed fruit maturity and ripening processes in fruits which may improve fruit size, color development as well as rind thickness by the inhibition of ethylene production (Byers et al., 2005). The findings of Al-Husseini (2012) also supported the findings of current study. Similar findings were reported in various fruit crops like pear (Bal, 2019; Hosseini et al., 2018), cherry (Chea et al., 2019) and blueberries (Chen et al., 2015).

Conclusion

The creasing was significantly reduced by foliar spray of AVG and improved the textural properties of fruits as well fruit weight, diameter and rind thickness. The application of AVG (60 mg/l) was more effective in reducing creasing (%) by increasing the hardness, as well as strengthens the fruit rind. It is concluded that creasing was also decreased by the increase in rind thickness as well as fruit size.

Author Contribution Statement: Zahoor Hussain conducted the current research study. Zora Singh planned, designed as well as supervised this research study and provided the research facilities.

Conflict of Interest: The authors have no conflict of interest for this research study.

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Fruit diameter (mm)										
Washington Navel										
	Fi	Fruit set		Golf ball		Colour break		Mean (Treatments)		
Treatments	2011 2012		2011	2012	2011	2012	2011	2012		
Control	79.92 ^{bcc}	67.69 ^c	75.50 ^d	75.50 ^b	77.42 ^{cd}	77.18 ^{ab}	77.61 ^b	73.46 ^b		
AVG (20 mg/l)	79.92 ^{bcc}	¹ 81.80 ^{ab}	82.47 ^{ad}	79.56 ^{ab}	80.75 ^{a-d}	¹ 78.56 ^{ab}	81.05 ^{ab}	79.97 ^a		
AVG (40 mg/l)	81.40 ^{a-d}	82.13 ^a	85.55 ^{ab}	80.91 ^{ab}	83.36 ^{abc}	² 79.66 ^{ab}	83.44 ^a	80.90 ^a		
AVG (60 mg/l)	82.79 ^{abc}	83.39 ^a	87.30 ^a	82.23 ^a	83.97 ^{abc}	^e 80.91 ^{ab}	84.69 ^a	81.98 ^a		
Mean (stage)	81.01 ^a	78.76 ^a	82.71 ^a	79.55 ^a	81.37 ^a	78.92 ^a				
LSD (p < 0.05)										
Treatments (T)			3.59	1.59						
Stage (S)			Ns	Ns						
T×S			Ns	2.75						
Lane Late										
	Frui	Fruit set		Golf ball		Colour break		atments)		
Treatments	2011	2012	2011	2012	2011	2012	2011	2012		
Control	76.55 ^{ab}	80.80 ^{abc}	75.52 ^b	76.80 ^{cd}	75.20 ^b	78.41 ^{bcd}	75.76 ^b	78.67 ^b		
AVG (20 mg/l)	77.03 ^{ab}	86.10 ^a	79.58 ^{ab}	75.05 ^d	78.66 ^{ab}	83.11 ^{ab}	78.43 ^{ab}	81.42 ^{ab}		
AVG (40 mg/l)	77.80 ^{ab}	81.46 ^{abc}	79.72 ^{ab}	78.77 ^{bcd}	79.33 ^{ab}	84.02 ^{ab}	78.95^{ab}	81.40^{ab}		
AVG (60 mg/l)	79.34 ^{ab}	81.98 ^{abc}	82.69 ^a	80.88 ^{abc}	80.75^{ab}	86.19 ^a	80.93 ^a	83.02 ^a		
Mean (stage)	77.68 ^a	82.59 ^a	79.38 ^a	77.87 ^b	78.49^{a}	82.93 ^a				
LSD (p < 0.05)										
Treatments (T)			3.19	2.89						
Stage (S)			Ns	2.51						
T×S			Ns	Ns						

n = 4 replications (10 fruit per replication), any two mean within a column and within a row followed by different letters are significantly different; Ns = not-significant

Rind thickness (mm)									
Washington Navel									
	Fruit set		Golf ball		Colour break		Mean (Treatments)		
Treatments	2011	2012	2011	2012	2011	2012	2011	2012	
Control	4.57 ^{cd}	4.68 ^c	4.47 ^d	5.08 ^{abc}	5.04 ^{abc}	4.98 ^{bc}	4.69 ^b	4.92 ^b	
AVG (20 mg/l)	4.89 ^{bcd}	5.09 ^{abc}	5.03 ^{abc}	5.30 ^{ab}	5.23 ^{ab}	5.04 ^{abc}	5.05 ^a	5.14 ^{ab}	
AVG (40 mg/l)	4.97 ^{bc}	5.15 ^{abc}	5.08 ^{ab}	5.44 ^{ab}	5.47 ^a	5.16 ^{abc}	5.17 ^a	5.25 ^a	
AVG (60 mg/l)	5.05 ^{abc}	5.32 ^{ab}	5.10 ^{ab}	5.56 ^a	5.38 ^{ab}	5.40^{ab}	5.18 ^a	5.43 ^a	
Mean (stage)	4.87 ^b	5.06 ^b	4.92 ^b	5.35 ^a	5.28 ^a	5.15 ^{ab}			
LSD (p < 0.05)									
Treatments (T)			0.25	0.28					
Stage (S)			0.21	0.24					
T×S			Ns	Ns					
Lane Late					-				
	Fruit set		Golf ball		Colour break		Mean (Treatments)		
Treatments	2011	2012	2011	2012	2011	2012	2011	2012	
Control	3.42 ^b	4.57 ^{cd}	4.67 ^a	4.46 ^d	4.84 ^a	5.04 ^{abc}	4.31 ^b	4.69 ^b	
AVG (20 mg/l)	3.52 ^b	4.89 ^{bcd}	4.91 ^a	5.03 ^{abc}	5.13 ^a	5.23 ^{ab}	4.52 ^{ab}	5.05 ^a	
AVG (40 mg/l)	3.62 ^b	4.96 ^{bc}	5.04 ^a	5.08 ^{ab}	5.21 ^a	5.47 ^a	4.62^{ab}	5.17 ^a	
AVG (60 mg/l)	3.88 ^b	5.05 ^{ab}	5.20 ^a	5.10 ^{ab}	5.22 ^a	5.38 ^{ab}	4.77 ^a	5.18 ^a	
Mean (stage)	3.61 ^b	4.87 ^b	4.95 ^a	4.92 ^b	5.10 ^a	5.28 ^a			
LSD (p < 0.05)									
Treatments (T)			0.34	0.21					
Stage (S)			0.30	0.43					
T×S			Ns	Ns					

n = 4 replications (10 fruit per replication), any two mean within a column and within a row followed by different letters are significantly different; Ns = not-significant; treat = Treatments

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