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FOLIAR SPRAY WITH ABG-3168, GA_3, $CACL_2$ AND H_3BO_3 CORRELATED TO MATURITY, RIPENING AND STORABILITY OF ANNA APPLE FRUITS

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ABSTRACT

The influences of ABG-3168 (ReTain), GA_3, calcium and boron applications, at about four weeks before commercial harvest date, on maturity, ripening, pre-harvest drop and storability of Anna apple fruits (*Malus demestica* Borkh) were investigated during the two successive seasons of 2002 and 2003. The results showed that most maturity/ripening related fruit characteristics were significantly influenced by the tested treatments. ABG. ABG-treated fruits, at 250 and 500 ppm, were approximately one (Kg) firmer than non-treated ones on a given sampling date during both pre and post-harvest phases. Red color formation was inhibited, conversion of starch into sugar was less advanced and acidity was higher in this fruit compared with the control, indicating that maturation and ripening had been suppressed. ABG-treated fruits also contained a significant higher TSS concentration, only after shelf life, than the other treatments. Both the rate and the onset of red color accumulation were clearly stimulated and advanced (about one week) by boron application at both 50 and 100 ppm without hastening fruit ripening compared with the control. Pre-harvest fruit drop was effectively controlled by ABG application at both 250 and 500 ppm. While, $CaCl_2$ application at 2% greatly increased pre-harvest drop and caused leaf scorching. Gibberellic acid application, especially at 500 ppm, significantly increased fruit diameter in contrast to ABG one, especially at 500 ppm, which slightly decreased it compared with the control. Fruit weight was not significantly influenced by any of the applied treatments. The occurrence of browning was greatly decreased during storage and was completely eliminated during one week of shelf life by ABG application. However, the other treatments either had no significant effect or even increased the occurrence of browning. Our results herein pointed to ABG pre-harvest treatment as the best one to delay maturity and ripening and to improve the storability of Anna apple fruits under the Egyptian conditions. The efficacy of ABG treatment at the low concentration (250 ppm) was equal or even better than the high concentration (500 ppm). Also, the low concentration of ABG had less negative impact on fruit red colour formation and diameter.

INTRODUCTION

Anna apple is one of the most successful and rather-adapted apple cultivars to the warm winter climates due to its relative low chilling requirements (Trejo-Gonzalez and Valdez, 1991). Under the Egyptian conditions, the maturity and ripening of fruits occur during a hot and dry summer that causes several pre and post harvest physiological disorders e.g. dropping, bitter pit, rapid softening and browning. Such problems decrease marketable yield and limit fruit storability and shelf life.

The ability to control fruit softening during maturity and ripening stages is a demand if we planned to store the obtained fruits with minimal physiological and physical disorders. Ethylene is effectively connected with fruit maturity, ripening and post-harvest softening. Removal of ethylene from the atmosphere surrounding apple fruits or limiting their ability to synthesize ethylene and response to it slowed their ripening (Graell and Recasens, 1992; Klee and Clark, 2002). Lieberman (1975) found that rhizobitoxine, which is a metabolic product secreted by bacterium *Rhizobium japonicum* growing symbiotically in the root nodule of soybean, inhibited ethylene production of apple tissues by blocking the conversion of methionine to ethylene. They also reported that a series of structural analogs of rhizobitoxine had similar inhibitory properties on ethylene. Recently, Abbott Laboratories have developed (S)-trans-2-amino-4-(2-aminoethoxy)-3-butenoic acid hydrochloride which commercially is known as ABG-3168 or ReTain. It is a structural analog of aminoethoxyvinylglycine (AVG). Mitcham *et al.*, (1998) found that pre-harvest application of ReTain at 500 ppm on pear fruits suppressed ethylene production and greatly delayed maturity and ripening. They also found that ReTain maintained fruit firmness and green color following cold storage for four months. Awad and de Jager (2002) reported that pre-harvest application of ABG-3168 on Jonagold apples delayed the transition of anthocyanin accumulation by about two weeks compared to the control, however, it did not affect maturation and ripening parameters as determined by both streif and starch indices. Autio and Bramlage (1982) reported that AVG applied to apple trees at one week before harvest suppressed ethylene production and delayed ethylene peaks in fruits kept at room temperature. They also noticed large variations among cultivars in the response to AVG application. Walsh and Faust (1982) found that bloom application of AVG increased fruit set and yield of Delicious apple trees.

Treatment of some fruits with gibberellic acid (GA_3) also can delay ripening (Looney, 1994; Ben-Arie *et al.*, 1995). GA_3 treatment of mature green tomatoes delays red color development and the climacteric respiratory rise (Babbitt *et al.*, 1973; Ben-Arie *et al.*, 1995). Wills *et al.*, (1973) found that pre-harvest application of GA_3 to Jonathan apples reduced the incidence of fruit breakdown storage when it was applied close to harvest but it was ineffective with earlier application. El-Fakharany *et al.*, (1995) reported that GA_3 application at 50 ppm at full bloom increased fruit firmness and decreased TSS in Anna apples but it had no effect on acidity as compared to control. Gibberellins were also used to improve skin finish of apples, delay rind senescence of Citrus, and delay fruit ripening and improve storability of sweet cherries (Looney, 1994).

Calcium has long been associated with the regulation of ripening process of fruits and storage life (Ferguson, 1984). Treatment of fruits with calcium will slow their ripening and softening of fruit flesh due to lowering respiration rates and reduced ethylene production (Ferguson, 1984; Poovaiah *et al.*, 1988; Siddiqui and Bangerth, 1995; Eliwa *et al.*, 1999). calcium has also been utilized in post-harvest treatments (Mason *et al.*, 1975).

Treatment of apple trees with boron successfuly increased fruit size and decreased internal cork, cracking, russeting and fruit drop (Peryea, 1994). Pre-harvest boron application eliminated the incidence of brown heart in Conference pears during four months of controlled atmosphere storage.

Also, boron application reduced membrane permeability at harvest and during storage and increased boron and vitamin C content of inner cortex tissue (Xuan *et al.*, 2001).

There is a very limitted information about the influences of ABG as a potential fruit maturity/ripening regulator under a hot summer conditions. Therefore, the aim of this study was to investigate the influences of ABG on Anna apple fruits maturity, ripening and storability, under the Egyptian conditions. A comparison between its effect and those of GA_3, calcium, and boron was also considered.

MATERIALS AND METHODS

2.1. Plant material and experimental design

During the 2002 and 2003 seasons, 15-year-old Anna apple trees (*Malus demestica* Borkh) were selected. Such trees were on Malus rootstock, trained as modified leader and planted in a single row system with spacing of 2.5 x 4.0m at a commercial orchard in Kafer-Badawy, El-Dakahlia governorate. The selected trees were nearly uniform in vigour, grown in clay soil, drip irrigated and received the normal cultural practices. Four weeks before commercial harvest, the trees were subjected to a single foliar spray of 250 or 500 ppm (S)-trans-2-amino-4-(2-aminoethoxy)-3-butenoic acid hydrochloride (ABG-3168/ReTain, Abbott), 250 or 500 ppm GA_3, 1% or 2% calcium chloride, and 50 or 100 ppm boron as a boric acid. All sprays were included. with 0.1% Tween 20 as wetting agent and applied by a manual spryer until the entire tree was wet. The control treatment in this study was trees only sprayed with water and wetting agent. A complete randomized blocks design with 3 replicates (two trees/each) per treatment was adopted. At about 4 weeks before expected commercial harvest and until 3 weeks thereafter, samples of 10 fruits per replicate (5-fruits/tree) were weekly collected from trees periphery for maturity determination.

2.2. Determination and development of fruit maturity/ripening characteristics

At each sampling date, the following fruit maturity/ripening characteristics were measured: fruit weight and fruit diameter, flesh firmness at the blush and the non-blushed sides using a hand Effgi-penetrometer supplemented with probe of 8.0 mm diameter and the results were expressed as kg/cm^2. Total soluble solids concentration (TSS) was measured in juice using a hand refractometer. Titratable acidity was also determined in juice by titrating with 0.1N sodium hydroxide in the presence of phenolephthalene as an indicator (Ranganna, 1979) and the results expressed as malic acid percentage. Starch conversion stage was assessed after dipping a transverse fruit section into a solution of 1% iodine (w/v) and 4% potassium iodide (w/v); the image was scored on a 1-10 scale (1 = completely black, 10 = completely white). The percentage of blushed area of fruit skin was visually estimated (Awad and de Jager 2002).

2.3 . Fruit drop percentage

In both 2002 and 2003 seasons, four limbs per tree (representing all tree sides, with 15-25 fruits per limb) were selected and labelled with special care for avoiding picking any fruit. The total number of fruits on each branch was counted before treatments application and at one week before the expected commercial date of harvest and the percentage of pre-harvest fruit drop was then calculated.

2.4. Calcium and boron concentration

In the 2003 season, calcium and boron concentrations in the fruit flesh were measured in fruit samples collected at commercial maturity date. A representative sample of 0.5g was taken from fine powder of dried fruit flesh and digested with nitric/perchloric (wet ashing) according to Gavlak *et al.*, (1994). Calcium (mg/100g dw) and boron (ppm) concentrations were determined by an liberty ICP optical emission spectrometer (ICP-OES), Varian.

2.5 . Fruit storage and shelf life

At commercial maturity date, additional samples of 75 fruits per treatment (25 fruits of uniform size per replicate) were picked from the treated trees periphery, weighted and stored in carton boxes at 2°C±1 and 85-90% RH for two months. Fruits at harvest date, removal from cold storage and after one week of shelf life at 25°C±3 and 50-60% RH were subjected to measure fruit weight, flesh firmness, total soluble solids (TSS) and titratable acidity. Loss in weight was determined after both storage and after shelf life periods.

2.6. Statistical analysis

The obtained data were subjected to analysis of variance (ANOVA) and the treatments means were separated by *F*-test and the least significant difference (LSD) test at the 5% level using the statistical package SAS, release 5, 1996 (USA).

RESULTS

Because of similarity between the results of the two seasons, data are presented as the means for both years.

Developmental changes of fruit maturity/ripening characteristics

Fruit maturity development as estimated by starch index, blush%, firmness, acidity and TSS% concentration clearly changed from week 9 to week 12 following full bloom under all the tested treatments (Fig. 1 and 2). Firmness and acidity were gradually decreased but starch conversion, TSS% and blush% were gradually increased as these weeks of maturity advanced. There were significant differences between the different treatments (Fig. 1 and 2). Applications of ABG at 250 and 500 ppm and GA_3 at 500 ppm significantly slowed fewer starch conversion compared to all other treatments (Fig. 1a). Red colour formation, as estimated by blush%, was greatly suppressed by ABG application at both concentrations (Fig. 1b). However boron application significantly increased red colour formation compared to the control and the other treatments. ABG-treated fruits were significantly firmer and contain more acid than the control and the other treatments (Fig. 1c and 2a). TSS% was in GA_3 and $CaCl_2$-treated fruit, specially at the higher concentrations, lower than the other treatments (Fig. 2b). However boron-treated fruit contained TSS% higher than the other treatments. Generally, fruit weight and diameter were gradually increased from week 9 to week 12 following full bloom (Fig. 3a and b).

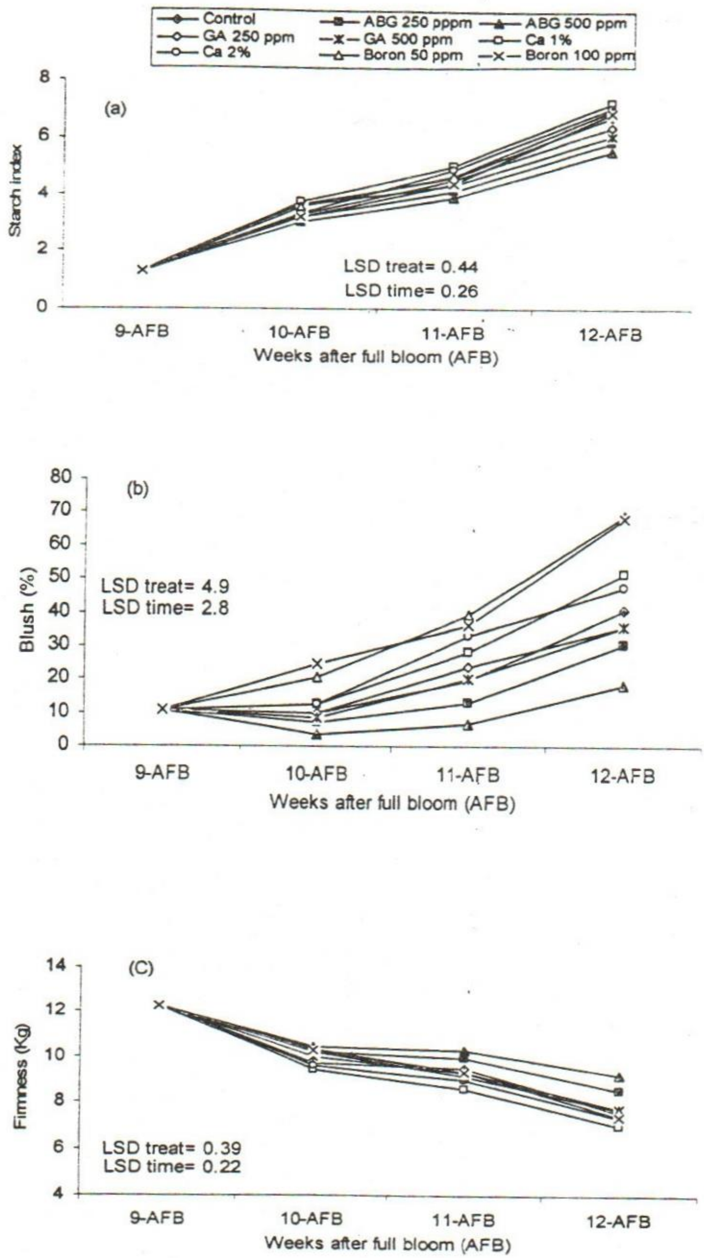


Figure 1: Diagrams showing developmental changes, at week 9 to week 12 following full bloom, in starch index (a), blush% (b) and flesh firmness (c) of Anna apple fruits as affected by pre-harvest spraying with 2 growth regulators and 2 mineral salts at 2 concentrations each. Data are the mean of the 2002 and 2003 seasons.

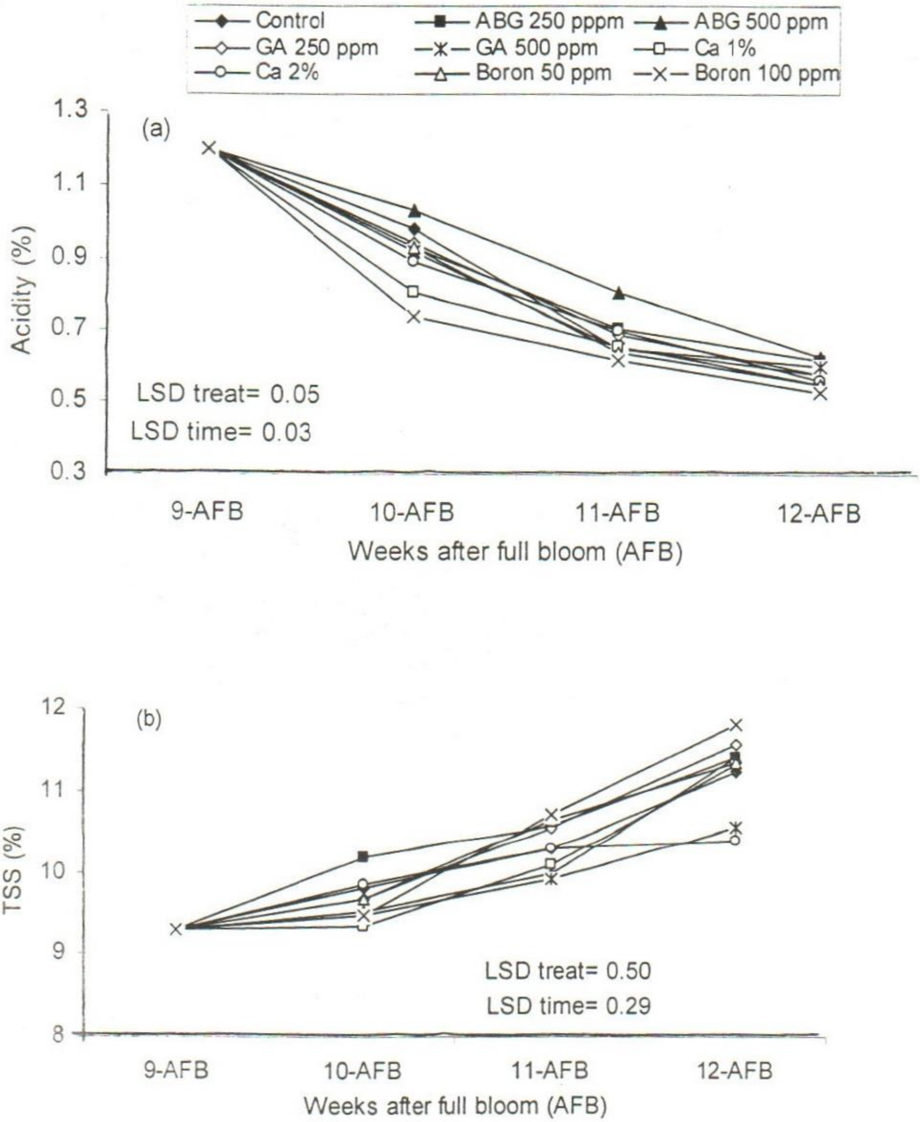
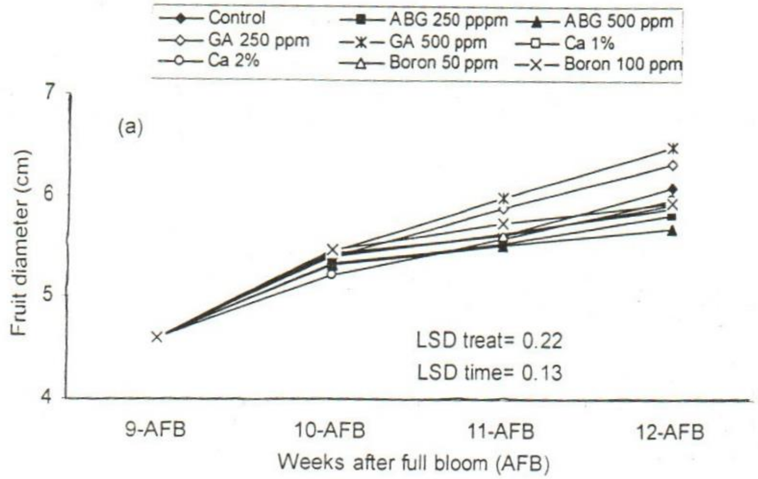


Figure 2: Diagrams showing developmental changes, at week 9 to week 12 following full bloom, in acidity% (a) and TSS % (b) of Anna apple fruits as affected by pre-harvest spraying with 2 growth regulators and 2 mineral salts at 2 concentrations each. Data are the mean of the 2002 and 2003 seasons.



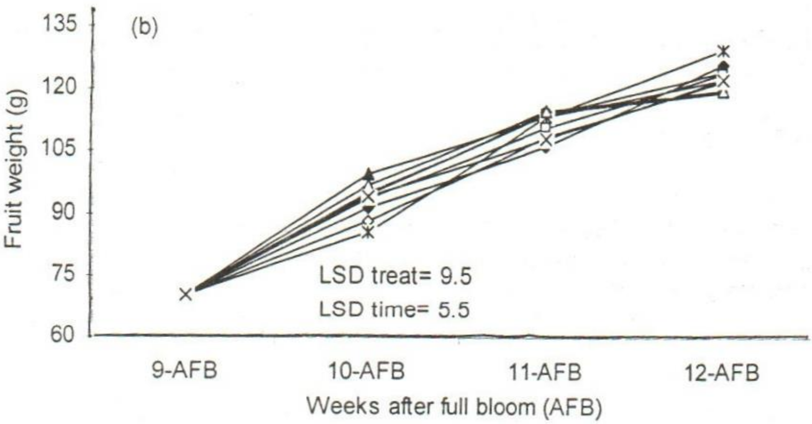


Figure 3: Diagrams showing developmental changes, at week 9 to week 12 following full bloom, in diameter (a) and weight (b) of Anna apple fruits as affected by pre-harvest spraying with 2 growth regulators and 2 mineral salts at 2 concentrations each. Data are the mean of the 2002 and 2003 seasons.

Application of GA_3, especially at the high concentration, significantly increased fruit diameter in contrast to ABG which slightly decreased it, especially at the high concentration, compared with the control (Fig. 3a). Fruit weight was insignificantly affected by any of the applied treatments (Fig. 3b).

Pre-harvest drop (%) and Calcium and Boron content of fruits

The results in Table (1) greatly indicated that foliar spraying Anna apple trees with ABG either at 250 ppm or 500 ppm was the most effective treatments to minimize pre-harvest fruit drop percentage (3.3-3.5%). While

$CaCl_2$ application at 2% recorded the highest pre-harvest drop (18.3%) compared with the control (13.1%) and the other treatments. GA_3 at both concentrations and boron at the high concentration significantly decreased pre-harvest drop compared with the control. The same table showed that $CaCl_2$ and boron pre-harvest applications significantly increased the concentration of calcium and boron, respectively in fruit flesh compared with the other treatments.

Table 1: Effect of foliar application with 2 growth regulators and 2 mineral salts at 2 concentrations each on pre-harvest drop (%) and Calcium and Boron content of Anna apple fruits.

Treatments	Pre-harvest drop (%)	Calcium (mg/100g dw)	Boron (ppm)
Control	13.1•	46.1••	0.34••
ABG at 250 ppm	3.5	50.4	0.32
ABG at 500 ppm	3.3	51.2	0.34
GA_3 at 250 ppm	9.7	47.6	0.36
GA_3 at 500 ppm	10.0	49.4	0.35
$CaCl_2$ at 1%	12.8	58.5	0.33
$CaCl_2$ at 2%	18.3	67.7	0.32
Boron at 50 ppm	13.8	48.4	0.43
Boron at 100 ppm	10.0	53.5	0.45
F-test	***	***	*
$LSD_{0.05}$	2.31	9.6	0.07

•[a]Data of pre-harvest drop percentage are the mean of 2002 and 2003 seasons. ••Data of calcium and boron are only for the 2003 season. * and * significant at level $P = 0.05$ and 0.001, respectively.**

Fruit storage and shelf life

Data of fruit maturity and ripening characteristics along with their changes during storage and shelf life as affected by the tested treatments were presented in Table (2). Such data proved that ABG treatment specially at the higher concentration (500 ppm) was the best among the other treatments and control to record significantly the greatest fruit firmness and relatively fruit TSS% at harvest time, during cold storage and shelf life. On the other hand, ABG-treated fruits at harvest time contained higher acidity%, but this increase was disappeared during storage and shelf life periods. Generally, fruit firmness and acidity% were decreased during cold storage and shelf life periods.

Concerning fruit loss in weight and browning% during cold storage and shelf life periods, the data in Table (3) showed that the loss in fruit weight and the incidence of browning were significantly decreased by ABG treatment during both storage and shelf life periods. The occurrence of browning was greatly decreased during storage and was completely eliminated during one week of shelf life. However, the other treatments had an increasing effect on the occurrence of browning during the same periods. Comparing with the control, such increasing effect was ranged from insignificant to significant differences.

Table 2:. Firmness, TSS% and acidity% of Anna apple fruits at harvest date, during storage and shelf life as affected by pre-harvest spraying with 2 growth regulators and 2 mineral salts at 2 concentrations each.

Treatments	Firmness (Kg/cm^2)			TSS (%)			Acidity (% malic acid)		
	At harvest	On removal from cold storage	After shelf life	At harvest	On removal from cold storage	After shelf life	At harvest	On removal from cold storage	After shelf life
Control	7.6	5.2	4.9	11.3	11.7	11.5	0.58	0.50	0.38
ABG at 250 ppm	8.5	5.8	5.4	11.4	12.9	12.2	0.62	0.50	0.37
ABG at 500 ppm	9.2	6.5	5.8	11.4	12.1	12.6	0.63	0.57	0.38
GA_3 at 250 ppm	7.6	5.2	4.7	11.6	11.0	11.7	0.58	0.49	0.33
GA_3 at 500 ppm	7.7	5.7	4.6	10.6	10.8	11.2	0.60	0.50	0.38
$CaCl_2$ at 1%	7.0	5.1	4.7	11.3	11.4	10.3	0.55	0.53	0.38
$CaCl_2$ at 2%	7.4	5.2	4.7	10.4	11.2	10.8	0.56	0.55	0.37
Boron at 50 ppm	7.7	5.5	4.6	11.4	11.5	11.7	0.55	0.47	0.37
Boron at 100 ppm	7.4	5.4	4.7	11.8	11.0	11.8	0.53	0.52	0.36
F-test	***	***	***	NS	NS	*	*	NS	NS
$LSD_{0.05}$	0.66	0.29	0.27	-	-	1.2	0.06	-	

Data are the mean of the 2002 and 2003 seasons. The fruits were stored at 2°C±1 and 85-90% RH for two months followed by one week of shelf life at 25°C±3 and 50-60% RH. NS, not significant; *, *, significant at level P = 0.05 and 0.001 respectively; (-), not calculated.**

Table 3: Loss in weight and browning percentage of Anna apple fruits during storage and shelf life as affected by pre-harvest spraying with 2 growth regulators and 2 mineral salts at 2 concentrations each.

Treatments	Loss in weight (%) On removal from cold storage	Loss in weight (%) After shelf life	Browning (%) On remov fromcoldstorage	Browning (%) After shelf life
Control	5.4	3.5	3.7	3.3
ABG at 250 ppm	3.7	2.9	1.0	0.0
ABG at 500 ppm	3.0	2.8	0.8	0.0
GA_3 at 250 ppm	6.1	4.1	5.0	2.5
GA_3 at 500 ppm	5.6	3.7	5.5	3.3
$CaCl_2$ at 1%	5.6	3.7	5.0	3.9
$CaCl_2$ at 2%	5.2	3.5	4.8	2.8
Boron at 50 ppm	5.3	3.6	3.6	3.5
Boron at 100 ppm	4.9	3.4	3.9	3.4
F-test	***	***	***	***
$LSD_{0.05}$	0.74	0.49	0.72	0.76

Data are the mean of the 2002 and 2003 seasons. The fruits were stored at 2°C±1 and 85-90% RH for two months followed by one week of shelf life at 25°C±3 and 50-60% RH. ***, significant at level *P* = 0.001.

4. Discussion

Our results show that maturity/ripening fruit characteristics were significantly influenced by the different treatments, especially ABG ones. Treatment with ABG significantly affected fruit firmness, red colour formation and starch content. ABG-treated fruits were approximately one (Kg) firmer than non-treated ones on a given sampling date during both pre and post-harvest phases (Figs. 1 and 2; Table 2). Red colour formation was inhibited, conversion of starch into sugar was less advanced, and acidity was higher in this fruit compared with the control. This means that fruit maturity and ripening had been suppressed. Although internal ethylene or ethylene production was not measured in our experiment, but ABG is most likely acting through ethylene pathway since it is well known that a series of structural analogs of rhizobitoxine had similar inhibitory properties on ethylene biosynthesis, especially when applied at an early stage prior to the accumulation of 1-aminocyclopropane-1-carboxylic acid (ACC) (Lieberman, 1975; Yang, 1980). These structural analogs are known to inhibit the conversion of S-adenosylmethionine (SAM) to ACC but it does not affect the conversion of ACC to ethylene (Yang, 1980; Masia, *et al.*, 1998). Ethylene is a powerful ripening hormone that is known to accelerate a number of ripening associated events in apple fruits such as softening, starch degradation, acidity loss, chlorophyll degradation and anthocyanin accumulation. Thus, the inhibition of ethylene biosynthesis in apple fruit tissues by ABG treatment can impede or delay the ripening process and senescence.

Both the rate and the onset of red colour accumulation (measured as blush percentage) were clearly stimulated and advanced (about one week) by boron application without hastening fruit maturity (Figs. 1 and 2; Table 2) compared with the control. In contrast, ABG treatment greatly inhibited both red color formation and fruit maturity. These results indicate that anthocyanin formation in apple skin is independently regulated maturity/ripening associated changes. This in turn confirming the conclusion of Awad and de Jager (2002) on Jonagold apples that anthocyanin formation can be regulated separately from other ripening events. Also, boron might have a direct effect on the activities of some relevant enzymes responsible for anthocyanin biosynthesis. Awad and de Jager (2002) also noted in Jonagold apples that the decrease in anthocyanin concentration in response to ABG application, which inhibits endogenous ethylene biosynthesis, was larger than the decrease in blush percentage. They suggested that not only fewer cells are involved in anthocyanin formation but also anthocyanin formation per cell might be affected by the ABG treatment.

One of the most frequently recurring problems in Anna apple growing under Egyptian conditions is the pre-harvest fruit drop which reduce potential yield (Eliwa, 1999). In our results, ABG application at both concentrations were the most effective treatments in controlling pre-harvest drop (Table 1). Such result is in agreement with those of Walsh and Faust (1982) who found that bloom application of AVG on Delicious apples increased both fruit set and fruit retention at harvest. This might be due to an interference with ethylene biosynthesis or action that obstruct or delay the formation of fruit abscission zone (Ferguson, 1984; Masia *et al.*, 1998). In our experiment, GA_3 application at both concentrations significantly decreased pre-harvest drop (Table 1). These results conflict with those of El-Fakharany *et al.* (1995) on Anna apples where they found that bloom application of GA_3 at 50 and 100 ppm had a little effect on fruit drop. However, the differences in the applied GA_3 concentrations and in the time of application may partly explain such conflictions. We also observed that the high rate of $CaCl_2$ application greatly increased pre-harvest drop compared with the other treatments. This might be due to a toxicity effect of such relative high rate of $CaCl_2$ on both fruit and leaves, especially under such hot summer conditions, since symptoms of toxicity e.g. leaf-scorching and leaf-fall were also observed in this treatment.

In our study, GA_3 application, especially at the high concentration, significantly increased fruit diameter in contrast to ABG which slightly decreased it compared with the control (Fig. 3a). It was reported by El-Fakharany *et al.*, (1995) that lower concentration of GA_3 application (50 or 100 ppm) at full bloom did not influence fruit diameter of Anna apples. Walsh and Faust (1982) also found that high levels of AVG application at full bloom caused excessive fruit set and decreased fruit size of Delicious apple, but the final yield was increased. These results suggest that the application of AVG and its structural analogs, especially at an early stage of fruit development, might have some inhibitory effect on cell division and/ or cell elongation.

Anna apple fruits are very susceptible to browning, especially when handled under hot summer conditions. This was explained by their relatively high level of phenolic substances and the high thermostability of the polyphenoloxidase enzyme (PPO) compared with other apple cultivars (Trejo-Gonzalez and Valdez, 1991). The occurrence of browning was greatly decreased during storage and was even completely eliminated during one week of shelf life by ABG treatments (Table 3). This result might be related to a delay in all senescence processes caused by the inhibition of ethylene production by ABG treatment that might increased the stability of cell walls and particularly the integrity of membranes thus, preventing the direct contact among enzymes e.g. PPO and peroxidase and their phenolic substrates (Masia *et al.*, 1998). In our study, boron treatment had no effect on the incidence of browning (Table 3). This is conflicting with those of Xuan *et al.* (2001) where boron sprays, several times before harvest, reduced membrane permeability and completely eliminated browning in Conference pears during four month of CA-storage. Loss in fruit weight was also significantly reduced by ABG treatment (Table 3). This might also be due to the inhibition of ethylene, respiration rate and general metabolic activity of fruits by the ABG treatments.

In conclusion, pre-harvest application of ABG has been clearly demonstrated in our study to delay maturity and ripening as well as greatly improve the storability of Anna apple fruits under Egyptian conditions. The efficacy of ABG treatment at the low concentration (250 ppm) was equal or even better than the high concentration (500 ppm). Also, the low concentration of ABG had less negative impact on fruit red color formation and diameter.

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تأثير مركب الريتان (ABG) وحمض الجبريليك والكالسيوم والبورون على اكتمال النمو والنضج والقدرة التخزينية لثمار التفاح الآنا.

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تم دراسة تأثير الرش بمركب الريتان وحمض الجبريليك والكالسيوم والبورون كل بمفرده قبل أربعة أسابيع من الحصاد التجاري على اكتمال النمو والنضج والقدرة التخزينية لثمار التفاح الآنا و كذلك دراسة تأثير هذه المعاملات على تساقط ما قبل الجمع خلال موسمى ٢٠٠٢، ٢٠٠٣.

وقد أوضحت النتائج أن معظم خصائص اكتمال النمو والنضج للثمار تأثرت معنويا بالمعاملات المستخدمة وخاصة مركب الريتان (ABG). فالثمار المعاملة بمركب الريتان بتركيزات ٢٥٠ أو ٥٠٠ جزء فى المليون كانت أكثر صلابة (حوالى ١ ك.جم) خلال مراحل ما قبل الجمع وأثناء التخزين بالمقارنة بمعاملة الكنترول. فلقد قللت المعاملة بهذا المركب من نسبة تكوين اللون الأحمر ومن تحول النشا الى سكر بينما أدت الى زيادة الحموضة بالمقارنة بالكنترول والتى توضح أن معدل اكتمال نمو ونضج هذه الثمار قد تأخر نتيجة لهذه المعاملة بمركب الريتان.

أدت المعاملة بالبورون بتركيزات ٥٠ أو ١٠٠ جزء فى المليون الى زيادة تكوين اللون الأحمر وكذلك التبكير فى تكوينه (حوالى اسبوعين) وذلك بدون زيادة فى معدل نضج هذه الثمار بالمقارنة بالكنترول. أدت المعاملة بمركب الريتان الى التقليل من تساقط الثمار قبل الجمع بفاعلية كبيرة بينما أدت المعاملة بكلوريد الكالسيوم بتركيز ٢% الى زيادة تساقط الثمار وكذلك حدوث احتراق لحواف الأوراق.

أدت المعاملة بحمض الجبريليك وخاصة عند تركيز ٥٠٠ جزء فى المليون الى زيادة قطر الثمار عند الجمع وذلك بعكس مركب الريتان وخاصة عند تركيز ٥٠٠ جزء فى المليون والذى أدى الى تقليل قطر الثمار ولكن بدرجة قليلة. لم تؤثر أى من المعاملات المستخدمة على وزن الثمار عند الجمع.

أظهرت النتائج أنه يمكن منع حدوث التلون البنى فى الثمار سواء أثناء التخزين أو خلال مدة اسبوع على ظروف الغرفة عن طريق المعاملة بمركب الريتان بتركيز ٢٥٠ أو ٥٠٠ جزء فى المليون. بينما باقى المعاملات الأخرى لم تؤثر أو حتى أدت الى زيادة حدوث التلون البنى.

وبصفة عامة يمكن القول بأن المعاملة بمركب الريتان قبل موعد الجمع التجارى المتوقع بعدة أسابيع أظهر امكانية كبيرة فى تأخير اكتمال النمو والنضج وتحسين القدرة التخزينية لثمار التفاح الآنا تحت ظروف جمهورية مصر العربية. كما أنه لوحظ أن التركيز المنخفض من مركب الريتان (٢٥٠ جزء فى المليون) أعطى تأثيرا مساويا أو حتى أفضل من تأثير التركيز المرتفع (٥٠٠ حزء فى المليون) من حيث التأثير السلبى على تكوين اللون الأحمر وقطر الثمار.