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RETARDATION OF RUTAB DEVELOPMENT OF ''BARHI'' DATES AND ENHANCING BUNCH RIPENING UNIFORMITY BY PREHARVEST TREATMENTS WITH CALCIUM AND ANTI-ETHYLENE COMPOUND

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ABSTRACT: "Barhi" date fruits are highly valued by the consumers especially at full maturity or the bisr stage. At the full yellow coloration, the fruit is not stringent, crispy and sweet. Thus, research attempts have been focusing on safe and feasible treatments that could be adopted on the field scale. In this study, the treatments included antiethylene (AVG) at 25 ppm, AVG at 50 ppm, AVG at 25 ppm+ calcium chloride 2% (W/V), AVG at 25 ppm + calcium sulphate 2% (W/V), AVG at 50 ppm + calcium chloride 2% (W/V), AVG at 50 ppm + calcium sulphate 2% (W/V), calcium chloride 2% (W/V), and calcium sulphate 2% (W/V). Treatments were conducted by a hand sprayer until the runoff point. The spray was done at the end of August during the two seasons 2017 and 2018 respectively. Bunches of each treatment were harvested at the commercial harvest on October 22 and Sep 4 during the two seasons 2017 and 2018. The data proved that AVG alone whether at 25 or at 50 ppm delayed the progress towards the rutab stage. In other words, such treatments alone or when combined with $CaCl_2$ at 2% extended the bisr stage and even reduced the full rutab (full ripening) stage. Electrolyte leakage of dates was significantly reduced by many treatments especially the combinations of AVG at 25 or at 50 ppm plus $CaCl_2$ in both seasons. However, such former combinations resulted in reducing carotene content in dates as compared with the control. In conclusion, this study proved the possibility of controlling the ripe of "Barhi" dates while reducing their loss by safe treatments.

Keywords: Calcium, AVG, Date Palm, Ripening.

INTRODUCTION

The date palm fruit (***Phoenix dactylifera* L.**) is considered one of the most important fruit crops in the world, and it is highly needed and consumed all over the world, especially in the Middle East as part of the Arab heritage. Egypt ranks the first country in the production of dates where it produces about 1.6 million tons per year (**FAOSTAT, 2017**). Dates are an essential healthy food in the human diet, as it has many medicinal uses including anticancer, antihyperlipidemic, hepatoprotective activities (**Biglari *et al.*, 2009**). The dates have been described as a wealth of healthy ingredients (**Farag, 2016*)***. Dates are considered as a good source energy as they have a high content of sugar (**Franz *et al.*, 2002 and Vayalil, 2002**).

The fruits are loaded with sugars, nutrients and antioxidants. The date fruit goes through five stages during its growth these stages, namely, are hababook, kimri, khalal, rutab and tamar. Most date varieties are harvested at the tamar stage when the fruits are fully ripe. "Barhi " dates has been considered one of the most demanded cultivars of dates, which is harvested and consumed at the khalal stage when reaching to physiologically maturity, solid, clear and bright yellow in color (**Barreveld, 1993)**, at this stage, fruits reaches to harvest maturity. Meanwhile farmers face some problems that

cause heavy losses due to non-uniform maturation of date fruits within the same bunch which makes dates advanced in growth, development and maturity such dates are subjected to abscission since they are ahead or reached to the rutab stage. There have been few attempts to increase dates ripening uniformity. Research attempts were few to achieve fruit within the same bunch which focused only on preharvest spray of ethephon that works on releasing ethylene compound, in order to enhance fruit ripening (**Farag and Kassem, 1998**).

A great potential excise to enhance the harvested khalal date fruit, to improve the quality and to reduce the losses whether pre or postharvest. The purpose of this study was to investigate the effects of preharvest application of the calcium and antiethylene AVG on 'Barhi' date fruit quality and bunch uniformity picked at khalal stage and stored at different storage temperatures. Aminoethoxy vinyl glycine (AVG) has been shown to inhibit ethylene action and to delay softening in many fruits (**Kim *et al*., 2004)**. The application of the antiethylene compound such as aminoethoxy vinyl glycine (AVG) results in delaying fruit ripening by inhibition of ACC synthesis, throw inhibiting the conversion of SAM to ACC, so it can inhibit ethylene production, and delay ripening (**Capitani *et al*., 2002**). Ripening of dates means reaching to the rutab stage. On the other hand, calcium proved to maintain the plasma membrane integrity and to preserve the structure of the cell walls in plant cells (**Farag, 2010**). Thus, the aims of this study were to increase the uniformity of "Barhi" dates at harvest, while enhancing fruit quality. In addition, providing the date growers with an applicable safe treatment that could be adopted on a commercial scale.

MATERIALS AND METHODS

The present study was conducted during the two successive seasons (2017 and 2018) using "Barhi" date palm cultivar. Trees were grown in MAFA farm, Nubaria region Beheira Governorate, Egypt. The palm trees were planted at five meters apart in sandy soil. Trees were of similar age (12 Years old), uniform in growth, free from insect's damage and diseases. Trees have received standard agricultural practices under drip irrigation system. Nine treatments were arranged in a randomized complete block design with four replicates per treatment each replicate was represented by one bunch (9×4×1 = 36 bunches). The treatments were as follow: Control (sprayed with tap water), antiethylene (AVG) at 25 ppm, AVG at 50 ppm, AVG at 25 ppm+ calcium chloride 2% (W/V), AVG at 25 ppm + calcium sulphate 2% (W/V), AVG at 50 ppm + calcium chloride 2% (W/V), AVG at 50 ppm + calcium sulphate 2% (W/V), calcium chloride 2% (W/V), and calcium sulphate 2% (W/V). All trees were sprayed during the Bisr stage (50-70% the yellow fruit color) at the end of August during the two seasons using a small handgun sprayer until the run-off. Ionic surfactant was used at (0.05 cm/L) with all treatments.

At harvest: bunches were carefully harvested on October 22 and Sep 4 during the two seasons, respectively. Seven strands were collected from each bunch to calculate bisr fruit percentage and rutub fruit percentage as follow:

- Bisr fruit percentage = the number of bisr fruits / the total number of fruits*100.

- Rutub fruit percentage = the number of rutub Fruit / the total number of fruits *100.

- Number of fruit/ strands= the total number of bisr fruit /7 strands.

- Fruit quality parameters

A sample of 20 fruits was taken from each seven strands for the determination of some physical and chemical characteristics of fruits.

1-Physical characteristics

Fruit weight (g), the average weight of 20 fruits was determined by using a digital analytical balance and fruit diameter (cm) as well as, fruit size (cm^3) was measured by using a graduated cylinder and flesh weight (g).

2-Chemical characteristics:

- Total Soluble Solids (TSS %): was measured using a hand refractometer.

- Vitamin C: was determined as mg/ 100 ml according to (**A.O.A.C., 1985**).

- Total acidity %: was measured as a malic acid according to (**A.O.A.C., 1985**).

- Total sugars: was estimated according to **(Smith, 1956)**.

- Carotene (µg/g): was measured according to **(Mustapha and Babura, 2009)**.

- Electrical conductivity (EC): conductivity of the flesh tissue was measured by using a conductivity meter.

3-Nutrient determination in fruits

Samples from date fruits were taken for their content of K, Ca and Mg. Date fruit samples were taken and placed in digestion glass in a muffle for 6 hours at temperature of 500^0C to determine of dry aching based on method described by **Chapman and Pratt (1978)**. After 6 hours, the muffle furnace is closed and waiting until the digestion glass cooled down with the digested samples. Then the weight of 1 gram of the digested sample was taken and dissolved by adding 5 ml diluted aqua regia for each vial. After half an hour dissolved sample was quantitatively transferred into a 100 ml glass for elemental determination. Concentrations of K, Ca was determined in the previously filtrates by the flame photometer (Jenway, PFP7) while Mg was determined by using Atomic Absorption Spectrophotometer (GBC 932 AA).

Statistical analysis

The data was laid out and analyzed in randomized complete blocks design (RCBD), the analysis was done using **(SAS, 2000)** program version. The means were compared according to the least significant difference (**LSD**) at 0.05 levels.

RESULTS

Physical characteristics

Data in Table 1 revealed that the use of the anti-ethylene compound, namely AVG resulted in a significant increase in bisr (Khalal) dates as compared with the control. The greatest increase was obtained with the application of AVG at 25 ppm plus $CaCl_2$ at 2% in both tested seasons. Moreover, all other AVG included treatments were able to increase the bisr fruits with various magnitudes when compared with the control in a consistent manner in both seasons. Moreover, $CaCl_2$–treated fruits were also able to cause a significant increase in bisr fruits while $CaSO_4$ resulted in a reduction of the bisr dates at harvest as compared with the control.

Data in Table 1 indicated that the control and Calcium sulphate treated fruits had the highest percentage of dates with partial rutab development in both seasons. Meanwhile, all other treatments resulted in less development of partially rutab stage with various magnitudes. The treatments of Calcium chloride at 2 % alone followed the control and calcium-sulphate treated dates in the descending order of the percentage of partially Rubab fruits. The least values of such character were obtained with the combination of AVG (25ppm) plus calcium chloride (at 2%) in both seasons. Meanwhile, the application of AVG (at 50ppm) alone resulted in about 13% of partially rutab fruits. Even AVG at 25 ppm resulted in similar percentage of such character to that obtained with AVG alone at 50 ppm.

Data in Table 1 also showed that the greatest percentage of full rutab dates was found in the control fruits. Meanwhile, the other treatments whether individually applied or in combinations between AVG and calcium compounds resulted in a significant reduction of the full rutab dates. However, the only treatment that resulted in a similar percentage of full rutab dates was calcium sulphate at 2% in a consistent manner in both seasons. Moreover, the combinations of AVG plus calcium chloride or plus calcium sulphate resulted in the greatest reduction of full rutab dates in both seasons except with AVG (25ppm) plus $CaSO_4$ (2%) in the second season. In addition, the magnitude of the reduction of the full rutab by AVG (50ppm) plus $CaSO_4$ (2%) was significant but lower than that obtained with AVG at 25 or 50 ppm plus $CaCl_2$ especially in the second season.

Table 1. Influence of preharvest application treatments on some physical characteristics of "Barhi" date palm during 2017 and 2018 seasons.

Treatments	% Bisr		% Partially Rutab		% Full Rutab	
	2017	2018	2017	2018	2017	2018
Control	69. 53 h	68. 34 g	18. 38 b	20. 82 a	12. 09 a	10. 84 a
AVG (25ppm)	82. 48 e	82. 51 d	11. 93 d	11. 94 d	5. 59 c	5.50 bc
AVG (50ppm)	80. 21 f	80. 55 e	13. 54 d	13. 85 c	6. 16 bc	5. 60 bc
AVG (25ppm) +Ca Cl_2 (2%)	96. 17 a	96. 34 a	2. 92 g	2. 72 f	0. 91 d	1. 12 d
AVG (25ppm) + Ca SO_4 (2%)	89. 60 c	88. 22 b	7. 94 f	7. 48 e	2. 42 d	4. 31 c
AVG (50ppm) +Ca Cl_2 (2%)	94. 09 b	95. 61 a	3. 85 g	3 .00 f	1. 92 d	1.07 d
AVG (50ppm) + Ca SO_4 (2%)	85. 18 d	86. 63 c	9. 83 e	8. 9 e	4. 83 c	4. 37 c
Ca Cl_2 (2%)	76. 57 g	75. 4 f	15. 93 c	17. 44 b	7. 49 b	7. 16 b
Ca SO_4 (2%)	65. 17 i	66. 93 h	21. 58 a	22. 18 a	13. 21 a	10. 89 a

Values within a column with similar letter (s) were not significantly different based on the least significant difference at 0.05%.

With regard to the influence of various applications on the number of fruits per each strand, some variations were found (Table 2). However, the absolute values of this number ranged between 17 to 22 fruits but there was specific trend that could be followed there were many treatments that had similar number of fruits per strand when compared with the control such as AVG at 25 ppm, AVG at 50 ppm in the second season alone and $CaCl_2$ alone in the second season.

The variations in fruit weight and size at harvest of '' Barhi'' dates response to various treatments was reported in Table 2. The data showed that fruit weight was similar for all the treatments and the control.

The variations in flesh weight at harvest of '' Barhi'' dates response to various treatments was reported in Table 2. The data indicated that flesh weight was similar for all the treatments and the control.

Chemical characteristics

The changes in TSS at harvest in response to various used treatments were shown in Table 3. The data indicated that the greatest TSS value was found with the control and with calcium sulphate. Moreover, the application of calcium chloride alone caused a significant increase in TSS only in the first season when compared with the control. Meanwhile, the individual application of AVG whether at 25 or 50 ppm resulted in a significant reduction of TSS values in both seasons. The addition of AVG (25ppm) to $CaSO_4$ former combination had a similar influence on TSS to that found by AVG (at 50ppm) plus $CaCl_2$. The greatest magnitude in the reduction of TSS was obtained with the combination of AVG (at 25 ppm) plus $CaCl_2$ at 2%.

The response of fruit acidity to various used treatments was shown in Table 3. The data indicated that the control and many applications had similar influence on such acidity such as the applications of AVG (at 25 ppm) and AVG (at 50 ppm) in addition to the application of AVG (at 50ppm) plus $CaSo_4$ (2%) in both seasons. Meanwhile, the two individual applications either $CaCl_2$ or $CaSo_4$ resulted in similar dues of juice acidity to that of the control in a consistent manner in both seasons. The greatest values of fruit acidity were obtained with the combination of AVG followed by $CaCl_2$ (at 2%).

Table 2. Influence of preharvest application treatments on some physical characteristics of "Barhi" date palm during 2017 and 2018 seasons

Treatments	Number of Fruits / Strand		Fruit weight (g)		Fruit size (cm^3)		Flesh weight (g)	
	2017	2018	2017	2018	2017	2018	2017	2018
Control	17.57de	18.67c	341.65b	443.47a	320a	380a	309.98bc	408.34a
AVG (25ppm)	16.82de	20.33bc	340.90b	416.18a	350a	408a	313.08bc	384.18a
AVG (50ppm)	23.05a	18.38cd	345.38b	400.32a	362a	400a	312.15bc	370.68a
AVG (25ppm) +Ca Cl_2 (2%)	15.71e	24.50a	357.03ab	394.63a	380a	365a	328. 95ab	365.67a
AVG (25ppm) + Ca SO_4 (2%)	21.39ab	20.23bc	337.01b	415.76a	360a	380a	304.74c	382.30a
AVG (50ppm) +Ca Cl_2 (2%)	18.38cd	17.24d	353.90ab	394.21a	370a	380a	322. 69bc	362.92a
AVG (50ppm) + Ca SO_4 (2%)	17.79de	22.05b	354.94ab	436.04a	367a	380a	322.75bc	403.12a
Ca Cl_2 (2%)	20.04bc	18.39cd	343.52b	414.16a	342a	441a	314.07bc	381.22a
Ca SO_4 (2%)	22.68a	16.86d	369.67a	421.96a	315a	394a	339.16a	387.97a

*Values within a column with similar letter (s) were not significantly different based on the least significant difference at 0.05%.

The effect of applied treatments before harvest on vitamin c content at harvest was shown in Table3. The data provided evidences that either $CaCl_2$ or $CaSo_4$ had a significant influence of vitamin c only in the second season while $CaCl_2$ in the first seasons had a similar vitamin c similar to that of the control. Moreover, treatments such as AVG at 25 ppm, AVG at 50 ppm, AVG (25 ppm) plus $CaCl_2$, AVG (25 ppm) plus $CaSo_4$, AVG (50 ppm) plus $CaCl_2$ and AVG (50 ppm) plus $CaSo_4$, all such treatments were able to cause a significant increase in vitamin c in the first season while their influence on vitamin c in the second season was not significant.

The effect of various used treatments on Electrical conductivity of "Barhi" date fruits was reported in Table 3. The data revealed that the control fruits in both seasons had greater values of electrolyte leakage when compared with all used treatments except $CaSo_4$ (2%) alone that gave greater EC than that of the control. Meanwhile, all applications resulted in a significant reduction in their EC values with different magnitudes. The lowest values of EC resulted from the applications of AVG (25 ppm) plus $CaCl_2$and the second formulation that contained AVG (50 ppm) plus $CaCl_2$. Moreover, to a lesser extend of EC reduction, it was found with AVG (at 25 ppm) plus $CaSO_4$. Furthermore, the individual treatment of either AVG 25 or 50 ppm each of them was able to reduce the EC of "Barhi" dates but to lesser magnitude than their combination treatments.

The response of total sugars to used treatments to "Barhi" dates during the two seasons 2017 and 2018 was reported in Table 3. The data showed that the control dates had greater total sugars than all other applications expect $CaSo_4$ alone especially in the second season. The highest reduction in total sugars occurred with the application of the combination of AVG (at 25 ppm) plus $CaCl_2$ (at 2%) in both season. Meanwhile, the individual treatments of AVG (at 25 ppm or at 50 ppm) and $CaCl_2$ (at 2%) caused a significant reduction in total sugars as compared with the control. Even the combination of AVG (at 25 ppm) plus $CaSo_4$ (at 2%) gave 25-30% total sugars but was lower than that obtained with $CaSo_4$ alone but varied from AVG (at 50 ppm) plus $CaSo_4$ only in the first season. The influence of applied treatments on carotene content in "Barhi" dates during the

two seasons 2017 and 2018 was reported in Table 3.

The data indicated that all treated dates in general had lower carotenes than the control. Moreover, the combination of AVG (at 25 ppm) plus $CaCl_2$ (at 2%) resulted in drastic reduction of carotene content as compared with other used treatments. Meanwhile, the sole application of $CaCl_2$ resulted in lower carotenes than the control in both seasons but was higher than that obtained with AVG at 25 ppm alone. In a similar manner, the combination of AVG (at 50 ppm) plus $CaCl_2$ (at 2%) followed AVG (at 25 ppm) plus $CaCl_2$ (2%) in the descending order of carotene reduction in significant manner by used treatments.

Table 3. Influence of pre harvest application treatments on some chemical characteristics of "Barhi" date palm fruits during 2017 and 2018.

Treatments	TSS (%)		Acidity (%)		Vitamin C mg/100mL		EC (%)		Total Sugars (%)		Carotene (µg/g)	
	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018	2017	2018
Control	39. 37 a	38. 55 ab	0. 208 c	0. 246 b	7. 66 d	9. 55 a	17. 15 b	17. 48 b	36. 55 a	35. 85 b	113. 15 a	116. 33 b
AVG (25ppm)	36. 40 bc	35. 62 cd	0. 255 c	0. 300 b	8. 02 c	10.00 a	12. 63 d	12. 65 e	33. 48 c	32. 74 d	81. 62 c	80. 62 e
AVG (50ppm)	37. 40 b	36. 95 bc	0. 235 c	0. 293 b	8. 06 c	8. 89 a	14. 41 c	14. 10 d	34. 71 b	33. 49 cd	83. 04 c	83. 43 d
AVG (25ppm) + Ca Cl2 (2%)	31. 28 e	32.00 e	0. 503 a	0. 566 a	11. 58 a	9. 78 a	7. 41 g	7. 38 h	25. 62 e	27. 38 f	52. 41 g	55. 37 i
AVG (25ppm) + Ca SO4 (2%)	34. 93 cd	34. 15 d	0. 346 b	0. 346 b	8. 70 b	9. 05 a	9. 62 8 f	9. 33 g	30. 08 d	29. 55 e	65. 80 e	64. 50 g
AVG (50ppm) + Ca Cl2 (2%)	34. 67 d	33. 88 d	0. 358 b	0. 363 b	8. 68 b	9. 56 a	8. 32 g	7. 90 h	29. 57 d	28. 10 f	61. 13 f	61. 58 h
AVG (50ppm) + Ca SO4 (2%)	36. 43 bc	35. 10 cd	0. 300 bc	0. 319 b	8. 14 c	9. 17 a	11. 01 e	11. 07 f	33. 00 c	29. 93 e	72. 47 d	69. 81 f
Ca Cl2 (2%)	37. 98 b	37. 15 bc	0. 228 c	0. 265 b	7. 61 d	7. 33 b	15. 23 c	15. 79 c	35. 33 b	34. 60 c	105. 39 b	110. 51 c
Ca SO4 (2%)	40. 05 a	39. 48 a	0. 205 c	0. 234 b	6. 64 e	6. 78 b	21. 17 a	21. 36 a	37. 9 a	37. 32 a	114. 29 a	121.8 2 a

*Values within a column with similar letter (s) were not significantly different based on the least significant difference at 0.05%.

DISCUSSION

The current research provided evidence about the possibility of controlling ripening of date palm fruits. Ripening of dates means reaching to the rutab stage **(Shahdadi *et al*., 2015).** That starts at the floral end of the fruits. Extension or prolonging the Bisr stage (also called khalal) is desired and demanded for Barhi dates fruits. The edibility of the "Barhi" dates at the full coloration and full size gives this cultivar its economic value since the disappearance of the astringent taste due to polymerization of soluble tannins **(Al-Farsi and Lee, 2008).** In a similar way, a few other date fruits such as Zaghloul and Samany dates could be marketed and consumed at the Bisr (khalal) stage. In another way, it has been reported that fruits of Barhi, Zaghloul or samany could be consumed at full coloration before the initiation of rutab development this ascribed to the convention of soluble tannins to the insoluble or polymerized tannins **(Al-Farsi and Lee, 2008 and Hammouda *et al*., 2013)**.

Many dates palm growers and producer are keen to have a safe method and applicable on a large scale to delay rutab development since many dates convert to the rutab stage during limited duration of time. The storability and the shelf life of rutab dates is much shorter than that of bisr stage. The fruit deteriorates rapidly at the full rutab stage which increases post-harvest losses since date fruits have been proved to be climacteric fruit (**Abd Elwahab *et al*., 2019).** That added another important aspect to necessary of profaning of the bisr stage as long as possible. The autocatalysis prosperity of climacteric fruits means that any external source of ethylene would lead to more production of internal ethylene which enhances the rapid development towards the soft and rutab date fruits. Furthermore, the crispy texture and the high content of carotene in the bisr dates and lower non-reducing sugar content of the bisr dates adds a lot to the attraction of consumers. Thus, bisr dates have more antioxidants and less sucrose than rutab fruits (**Farag, 2004**). In

addition to the crispiness of texture of the bisr dates. Since AVG (amino ethoxy vinyl glycine) is an anti-ethylene that has been used on a commercial scale for an extended period of time. Its application at the convenient time and at the desired concentration would inhibit the endogenous ethylene and the resulting endogenous ethylene as well. AVG was proved to inhibit to the conversion of S -adenosine methionine to ACC in the pass way of ethylene biosynthesis **(Martínez-Romero *et al*., 2007).** Meanwhile, calcium plays important roles in protecting the cell wall against degradation and break down. Calcium proved to be able to inhibit some enzymes responsible for the breakdown of cell wall polymers of polysaccharides. Its role maintaining the integrity of the plasma membrane has been reported by many researchers. That could explain the reduction obtained in the electrical conductivity by treatments such as calcium chloride (at 2%) alone or even with its combination with AVG (at 25 ppm) as compared with the control. In similar manner, a significant reduction occurred with application of AVG (at 50 ppm) plus $CaCl_2$ in a consistent manner in both seasons. No wonder the treatments that related rutab development, were also able to delay fruit characteristics related to ripening such as the reduction of TSS, an increased acidity and reduction total sugar as compared with the control.

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