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# IMPACT OF GIBBERELLIC ACID TREATMENTS ON GERMINATION OF CLEOPATRA MANDARIN SEEDS

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**ABSTRACT**: This study was carried out throughout two successive seasons (2021 and 2022) for investigating GA<sub>3</sub> pre-sowing soak solutions influence on Cleopatra mandarin seeds (*Citrus reshni L.*). It was aimed to enhance the Cleopatra mandarin seeds germination process, as well as seedlings vigor. Thus, seeds of Cleopatra mandarin rootstock were subjected to soaking for 24 hours in GA<sub>3</sub> solutions at 200, 250, and 300 ppm as well as soaking in tap water (control treatment), for investigating their effect on some germination and growth parameters. Data obtained during both experimental seasons revealed obviously that all GA<sub>3</sub> treatments increased significantly both germination measurements (total germinated seeds & germination %) and some growth parameters as compared to control (soak for 24 hours in tap water). However, soaking in GA<sub>3</sub> at 250 and 300 ppm concentrated solutions particularly were statistically the most effective in this study.

Key words: Cleopatra mandarin seeds, gibberellic acid, germination, rootstock, citrus.

# **INTRODUCTION**

Citrus is one of the most important fruit crops all over the world. It ranks the third position between fruit crops and only preceded with grapes and apples. In Egypt, it is the most important fruit crop. Cleopatra mandarin (Citrus reshni L.) has been established as rootstock over many years. Tree vigor and fruit quality of sweet orange and mandarin cultivars on Cleopatra mandarin rootstock is very good and its equal to those produced on sour orange rootstock, but fruit size is smaller. Cleopatra mandarin is considered one of the important species of citrus root-stocks, which studies have begun on it as a substitute for the sour orange rootstock in many countries. Cleopatra mandarin is more tolerant than sour orange for Tristeza, gummosis, phytophthora foot rot, exocortis, cold, calcareous soil, this rootstock grows well in sandy and heavy soils as well as its high tolerance to chlorosis and salinity. The trees budded on Cleopatra mandarin are slowly grown

until they reach the fruiting stage. Citrus seeds of all rootstock types germinate unevenly, which resulting poor seedling uniformity. Thus, it is highly interested by nursery owners to get higher and earlier germination (Llosa *et al.*, 2009; Anjum, 2010; Lacey, 2012; Sharaf *et al.*, 2016 and Al-Janabi, 2018).

GA<sub>3</sub> play important roles in many essential plant growth and development processes including seeds germination, stem elongation, and leaf expansion. They are often used to overcome seeds dormancy and can significantly improve seeds germination in many species, mainly through the activation of embryo growth, mobilization of reserves, and weakening of the endosperm layer. It has also been reported that seeds priming with GA<sub>3</sub> improves germination and the growth parameters of shoot length, root length, and seedling weight. (Khan and Chaudhry, 2006; Baskin and Baskin, 2014; Pallaoro *et al.*, 2016; Ma *et al.*, 2018 and Cipcigan *et al.*, 2020). This study aimed to enhance the Cleopatra mandarin seeds germination process, as well as seedlings vigor by using  $GA_3$  with different concentrations.

# MATERIAL AND METHODS

The present investigation has been carried out throughout two successive seasons (2021 and 2022) on Cleopatra mandarin seeds (*Citrus reshni L.*) to enhance the Cleopatra mandarin seeds germination process, as well as seedlings vigor by seeds soaking for 24 hours in GA<sub>3</sub> with different concentrations (200, 250 and 300 ppm) and soaking in tap water (control treatment). Also, before subjecting seeds to the differential pre-sowing treatments, they were soaked for 24 hours in Carpndazim fungicide solution at 50% concentration.

At late February 2021 and 2022 years seeds of Cleopatra mandarin rootstock were separately subjected to the pre-sowing treatments just before planting in germination boxes during  $1^{st}$ &  $2^{nd}$  experimental seasons, respectively. The seeds planted in an individual germination box previously filled with planting medium consisting of sand + clay mixture at equal proportion (v:v).

The experimental seeds were sowing in greenhouse at private orchard at Nobaria region, El Behera Governorate Egypt. The above treatments were applied as soaking in a solution of gibberellic acid in different concentrations. All the seeds of this study received the same horticultural practices except experimental treatments.

# The tested treatments were evaluated throw the following parameters:

#### **Germination parameters**

During each experimental season 10 seeds germinated measurements i.e., number and percentage of germinated seeds per every replicate (an individual germination box) were periodically counted at 30 and 60 days. Then average value of either number or percentage of germinated seeds per each investigated presowing soak treatment was estimated as an average of its five replicates during every experimental season.

#### Average plant height (cm) and leaves number

On August during 2021 and 2022 years, respectively the average seedling height in cm.

was counted for 10 seedlings per each replicate (an individual germination box), then an average plant height per every investigated pre-sowing soak treatment was estimated (as an average of its five replicates). Also, leaves number were counted.

#### Leaf area (cm<sup>2</sup>)

On August during 2021 and 2022 years, respectively, the leaf area  $(cm^2)$  was determined using discs of the leaf blades and total leaves area according to **Bremner and Taha (1966)**.

#### Leaf total chlorophyll

On August during 2021 and 2022 years, respectively, the leaf total chlorophyll was recorded in fresh leaves per each seedling using a portable chlorophyll meter SPAD 502 according to **Yadava**, (1986).

#### Leaf chemical composition

On August during 2021 and 2022 years, respectively, five dried leaves were finely grinded and digested using micro-keildahl unit. The percentage of nitrogen content was determined according to **Naguib** (1969). Phosphorus percentage was determined according to **AOAC**, 1985. Potassium percentage was determined according to **Brown** and Lillil (1964).

# Statistical analysis

The experimental design was randomized complete block design (RCBD) with five replicates and 40 seeds for each replicate. The data obtained were statistically analyzed using the analysis of variance method as reported by **Snedecor and Cochran, 1980**. The differences between means were differentiated by using Duncan's range test (**Duncan, 1958**).

# **RESULTS AND DISCUSSION**

The obtained data in Table (1) showed that, all gibberellic acid concentration had a great positive effect on germination parameters after 30 and 60 days in both seasons.

As for in the first season, the number of germination seedling after 30 days in Table (1) reached 58.56 and 55.85 with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 28.96. In addition, germination percentage after 30 days gained 29.28 and 27.93 % with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 14.48%. At

the same time, number of Germination seedling after 60 days were 169.20 and 146.24 with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 49.17. Moreover, germination percentage after 60 days gained 84.60 and 73.12% with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 24.59%. The same trend was obtained with germination parameters at the second season.

In this respect, our results are in agreement with those obtained by other researchers (Ollitrault *et al.*, 2007; Turgutoglu *et al.*, 2015; Sharaf, *et al.*, 2016 and Dilip *et al.*, 2017).

Moreover, the mod of action for gibberellins depends on their binding to a carrier protein in

the first place, which allows it to be inserted into the cell, after binding to a specific receptor, it passes into the cell nucleus and changes the synthesis of genetic material (RNA). At the same time, the gibberellins in the seed induce expression by the  $\alpha$ -amylase genes, an enzyme responsible for the hydrolysis of starch to produce simple sugars that will be the seedling's food source during germination until it can perform photosynthesis on its own. In addition, GA<sub>3</sub> can significantly improve seeds germination through activation the embryo growth, mobilization of reserves, and weakening of the endosperm layer (Pallaoro et al., 2016; Ma et al., 2018 and Cipcigan et al., 2020).

Table 1. Impact of gibberellic acid treatments on number of germination seedling and<br/>germination percentage after 30 and 60 days from sowing of Cleopatra mandarin seeds<br/>(2021-2022 seasons)

Characteristics Treatments	Number of sowing seeds at start	Number of germination seedling after 30 days	Germination % after 30 days	Number of germination seedling after 60 days	Germination % after 60 days
	First season 2021				
Control (water tap)	200	28.96 C	14.48 C	49.17 D	24.59 D
GA <sub>3</sub> at 200 ppm	200	48.46 B	24.23 B	107.30 C	53.65 C
GA <sub>3</sub> at 250 ppm	200	55.85 A	27.93 A	146.24 B	73.12 B
GA <sub>3</sub> at 300 ppm	200	58.56 A	29.28 A	169.20 A	84.60 A
		S	Second season 202	22	
Control (water tap)	200	32.41 C	16.20 C	47.63 D	23.82 D
GA <sub>3</sub> at 200 ppm	200	54.18 B	27.09 B	117.16 C	58.58 C
GA <sub>3</sub> at 250 ppm	200	63.40 A	31.70 A	149.15 B	74.57 B
GA <sub>3</sub> at 300 ppm	200	67.39 A	33.69 A	165.16 A	82.58 A

Mean followed by the same letter\s within each column are not significantly different from each other at 0.5% level.

The present data in Table (2) illustrated that, all gibberellic acid treatments had a great statically influence on plant height, leaves number, leaf area, total leaves area and total chlorophyll characteristics in both seasons.

Regarding, plant height achieved 9.71 and 9.02 cm with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 6.15 cm in the first season. In addition, leaves number earned 8.75 and 8.33 with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 6.37. At the same time, leaf

area was 8.83 and 8.51 cm<sup>2</sup> with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 7.22 cm<sup>2</sup> in the first season. Moreover, total leaves area attained to 77.32 and 70.96 cm<sup>2</sup> with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 46.04 cm<sup>2</sup> in the first season. The total chlorophyll acquired 68.48 and 67.28 with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 60.44. The same trend was obtained with plant height, leaves number, leaf area, total leaves area and total chlorophyll characteristics at the second season. These results are in congeniality with those found by **Francis and Sorrell (2001); Debaje** *et al.* (2010); Mahmoud (2012); Sharaf *et al.* (2016) and Dilip *et al.* (2017).

The existing data in Table (3) supply that, all gibberellic acid treatments had a major statically impact on N, P and K leaf composition in both seasons.

Generally, leaf nitrogen content reached to 1.771 and 1.755 % with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 1.427 % in the first season. In addition, leaf phosphorus content gained 0.393

and 0.384 % with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 0.324 %. At the same time, leaf potassium content was 2.171 and 2.130% with 300 and 250 ppm of GA<sub>3</sub>, respectively compared to the control, which was 1.885 % in the first season. the same trend was obtained with N, P and K leaf composition at the second season.

These results are in general agreement with the earlier findings of Abd El-momein *et al.*, 2007; Panigrahi and Strivastava, 2011; Mahmoud, 2012 and Sharaf *et al.*, 2016.

Characteristics Treatments	Plant height (cm)	Leaves number	Leaf area (cm <sup>2</sup> )	Total leaves area (cm <sup>2</sup> )	Total chlorophyll SPAD
		First season 2021			
Control (water tap)	6.15 C	6.37 C	7.22 D	46.04 D	60.44 C
GA <sub>3</sub> at 200 ppm	7.41 B	7.60 B	8.03 C	61.01 C	63.60 B
GA <sub>3</sub> at 250 ppm	9.02 A	8.33 A	8.51 B	70.96 B	67.28 A
GA <sub>3</sub> at 300 ppm	9.71 A	8.75 A	8.83 A	77.32 A	68.48 A
			Second seasor	n 2022	
Control (water tap)	6.42 C	6.39 C	7.24 D	46.26 D	59.64 C
GA <sub>3</sub> at 200 ppm	8.51 B	7.50 B	8.06 C	60.46 C	63.19 B
GA <sub>3</sub> at 250 ppm	10.18 A	8.47 A	8.48 B	71.85 B	67.96 A
GA <sub>3</sub> at 300 ppm	10.60 A	8.75 A	8.73 A	76.46 A	68.80 A

 Table 2. Impact of gibberellic acid treatments on plant height, leaves number, leaf area, total leaves area and total chlorophyll of Cleopatra mandarin seedling (2021-2022 seasons)

Mean followed by the same letter/s within each column are not significantly different from each other at 0.5% level.

Table 3. Impact of gibber	rellic acid treatments	s on leaves	nitrogen,	phosphorus	and po	tassium
contents of Cleop	oatra mandarin leave	s (2021-202	22 seasons)			

Characteristics	Nitrogen	Phosphorus	Potassium
Treatments	percentage	percentage	percentage
		First season 2021	
Control (water tap)	1.427 C	0.324 D	1.885 D
GA <sub>3</sub> at 200 ppm	1.589 B	0.357 C	1.995 C
GA <sub>3</sub> at 250 ppm	1.755 A	0.384 B	2.130 B
GA <sub>3</sub> at 300 ppm	1.771 A	0.393 A	2.171 A
		Second season 2022	
Control (water tap)	1.426 C	0.337 C	1.877 C
GA <sub>3</sub> at 200 ppm	1.565 B	0.355 B	2.000 B
GA <sub>3</sub> at 250 ppm	1.753 A	0.382 A	2.150 A
GA <sub>3</sub> at 300 ppm	1.777 A	0.391 A	2.178 A

Mean followed by the same letter/s within each column are not significantly different from each other at 0.5% level

# CONCLUSION

On the basis of present investigation, it is concluded that Cleopatra mandarin seeds treated by soaking in gibberellic acid with 250 or 300 ppm for 24 hours results in quicker germination, vigor vegetative growth and better germination percentage of seedlings.

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