



Seed Germination and Growth of Some Citrus Rootstocks Seedlings as Influenced by Some Stimulate Substances.

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ABSTRACT

This study was conducted during two consecutive seasons (2020 and 2021) in a plastic house located at the citrus seedling production unit of the Horticulture Research Institute, Agriculture Research Center, Giza Governorate, Egypt. To investigate the effects of different pre-sowing soak treatments on seed germination and growth performance of four citrus rootstocks: Rangpur lime (*Citrus limonia* L.), Carizzo citrange (*Citrus sinensis* Osb. X *Poncirus trifoliata* L.), Cleopatra mandarin (*Citrus reshni* L.) and sour orange (*Citrus aurantium* L.). Five pre-sowing treatments were applied by soaking rootstock seeds for 24 hours as follows: tap water (control), acetic acid at 1%, yeast extract at 1%, algae extract at 1%, and GA₃ at 20 ppm. The same five previous soaking treatments with the same doses were used as foliar treatments monthly until the grafting time.

The results revealed that the type of rootstock, pre-sowing treatment, and foliar fertilization significantly affected seed germination percentage and seedling attributes in both seasons. Concerning the effect of pre-sowing and foliar treatment, data showed that soaking seeds in acetic acid at 1% for 24 hours were more effective than other treatments in enhancing the germination percentage and all seedling traits, i.e., seedling height, number of leaves, root length, and number of roots. Regarding the effect of rootstock type, it was quite clear that sour orange rootstock significantly achieved the highest percentage of germination and all seedling characteristics.

Keywords: Citrus- Rootstocks- Germination- Seedlings - Characteristic

INTRODUCTION

Citrus rootstock production technology has recently gained great importance and is a successful strategy for optimization of citrus nursery tree production system. (Girardi et al., 2007). The importance of rootstocks is well documented, they are used not only as an effective means of controlling important biological pests, but they can also be used effectively to uptake of water and nutrition from the soil (Singh and Chahal, 2021).

Rangpur lime (*Citrus limonia* L.) rootstock is semi-vigorous, productive, tolerant to salt, drought and greening diseases and resistant to tristiza. Carizzo citrange (*Citrus sinensis* Osb. X *Poncirus trifoliata* L.) resist some species of phytophthora. It grows well on heavier clay loams to loamy soils. Cleopatra mandarin

(*Citrus reshni* L.) rootstock is moderately vigorous, cold hardiness, tolerant to tristiza, phytophthora foot rot and calcareous soil. Sour orange (*Citrus aurantium* L.) rootstock is still the most important rootstock because of its resistance to gummosis and compatibility with most commercial citrus varieties. (El-Sayed and El-Fangary, 2008 and Dilip et al., 2017)

Citrus seeds of all rootstock types characteristically germinate unevenly, resulting in poor seedling uniformity (Aubert and Vullin, 1998). Several factors such as temperature variations, oxygen levels, water availability and lack of inhibitory environmental conditions affect seed germination percent and seedling vigour (Yildiztugay and Kucukoduk, 2012). Therefore, seeds require pre-treatments



before sowing to obtain rapid, uniform and high germination rates. Numerous pre-sowing treatments have been used to improve seed germination and produce vigour seedlings such as water, Gibberellic acid, yeasts, algae and acetic acid (Aydn and Uzun, 2001, Bassal, 2009, Sharaf et al., 2016 and Masserano et al., 2022).

Water treatment can enhance seed germination by elevating the water and O₂ permeability of the seed (Msanga and Maghembe, 1986).

Acetic acid may successfully accelerate seed germination by increasing seed membrane permeability to facilitate proton transport in the seed cells (Sholberg et al., 2006).

Acetate is created when acetaldehyde is oxidized, and convert to Acetyl-CoA by acetate-activation enzymes. Acetyl-CoA is an essential acetyl donor for many biological reactions, including primary and secondary metabolism, protein acetylation, and transcriptional activity. Acetate metabolism is critical for the regulation of gene

This study was carried out during two consecutive 2020 and 2021 seasons in the plastic house located at the citrus seedling production unit of Horticulture Research Institute, Agriculture Research Center, Giza Governorate, Egypt. To investigate the effect of different pre-sowing soak treatments on the seed germination and growth performance of four citrus rootstocks as the first factor namely Rangpur lime (*Citrus limonia* L.), Carizzo citrange (*Citrus sinensis* Osb. X *Poncirus trifoliata* L.) Cleopatra mandarin (*Citrus reshni* L.) and sour orange (*Citrus aurantium* L.).

The rootstocks seeds were obtained from citrus seedling production unit of Horticulture Research Institute; from fully

expression, acetate homeostasis is maintained through acetate activation and that it is essential for plant development under normal conditions. Kudo and Kim (2023).

Algae extract comprise a great number of bioactive compounds, carbohydrates, minerals, trace elements and growth hormones that act as bio-stimulants of seed germination and enhance plant growth (Khan et al., 2009).

Yeast produces plant stimulating hormones, enzymes, vitamins, amino acids and minerals that is important to embryo germination, growth and differentiation of plant tissues (Sacakli et al., 2013), Gibberellic acid (GA₃) has a positive effect on seed germination and seedling growth. (Sanaullah et al., 2020).

The goal of the present study was to assess the impact of pre-sowing soak and foliar application with different solutions on the seed germination and growth performance of some citrus rootstocks.

developed homogenized fruits of adult trees in the middle of February for each season of the study. The seeds were extracted out, cleaned, dried by air, and stored at 3 °C.

The second factor is tap water (control), acetic acid at 1% (1% AA), yeast extract at 1% (1% YE), algae extract at 1% (1% AE) and GA₃ at 20 ppm. Yeast extraction was prepared by dissolving ten grams of dry yeast and one gram of sugar in 50 ml of warm distilled water (30 °C), and after 4 hours, the volume was supplemented to one liter, then filtered to obtain the yeast extraction, which contains several compounds according to (Somer, 1987) as shown in **Table (1)**.

Table1. Chemical analysis of the active dry yeast.

N (%)	Poly-sac charides (%)	Fats (%)	Protein (%)	Fiber (%)	Ash (%)	Thiamin (B1) (mg)	Riboflavin (B2)(mg)	Niacin (B4)(mg)	Vitamin (B6)(mg)	Vitamin (B12)(mg)
7.3	32.3	3.5	35	1.1	6.7	2.33	5.41	36.7	4.41	0.02



Algae extract contains of macro- and micronutrients as shown in (Table, 2) as recorded by (Aly and Mona, 2008). One hundred grams of *Spirulina platensis* algae powder was extracted with hot water and

kept overnight after vigorous shaking. The solution was decanted and filtered through Whatman filter paper and stored in refrigerated conditions at 4°C until usage (Ramamoorthy and Sujatha, 2007).

Table 2. Chemical analysis of *Spirulina platensis* algae extract

Components	(%)	Vitamins	(%)	Minerals	(%)	Colorants	(%)
Moisture	3.5	Provitamin A	0.21	N	15.3	Phycocyanin	15.6
Ash	0.7	Vitamin B1	0.02	P	0.92	Carotenoids	0.46
Lipids	9.5	Vitamin B2	0.03	K	1.83	Chlorophyll a	1.30
Protein	63.5	Vitamin B6	0.01	Ca	0.17		
Fiber	3.0	Vitamin B12	0.02	Mg	0.25		
		Vitamin E	0.14	Fe	0.05		

Seeds were sown in germination trays consisting of a 1:3 v/v mixture of peat moss and sand at April then, trays were watered after sowing, and subsequently irrigated once every two days, after three months. Selected vigor seedlings were planted in 4 kg plastic bags in a planting medium mixture of (peat moss and sand) in a ratio of 1:3.

The same previous five soaking treatments with the same doses were used as foliar application monthly until grafting time.

The planted bags were placed in the nursery in a factorial randomization completely design with 20 treatments each have 3 replicates and 20 seedlings per replicate.

The seed germination and vegetative growth of seedlings measurements were evaluated as following:

I- Seeds germination phase parameters: Three months after sowing, seed germination measurements were evaluated as following:

Germination percentage: Number of seeds germinated in each treatment was recorded; germination percentage was calculated using the following equation by Ranal and Santana (2006): Germination % = (number of germinated seeds / total number of seeds) X 100

Damping off percentage: Damping off percentage in each treatment was calculated

using the following equation by Kaster (1975): Damping off percentage = (Number of damped off seedlings / Initial number of seeds) x 100.

Albino percentage: Albino percentage in each treatment was calculated using Kaster (1975) equation:

Albino percentage = (Number of albino seedlings / Initial number of seeds) X 100.

Germination speed: Speed of germination (SG) was calculated according to (ISTA., 1999) using the following formula: $SG = n1/d1 + n2/d2 + n3/d3 + \dots$

Note, n = Germinated seed's number, d= Day's number.

Implant growth parameters: Seedling height (cm), number of leaves, root length (cm) and number of roots were assessed after three months of planted.

II- Seedlings phase parameters:

1- Seedlings vegetative growth parameters: Stem length (cm), stem diameter (cm.), number of leaves/ seedling and leaf area (cm²) a sample of ten leaves was collected to measure averaged leaf area by portable area meter – model LI 3000 made in U.S.A.

2- Seedlings chemical parameters:

Plant pigments determination: Chlorophylls a & b contents as mg/100 g leaves fresh weight were estimated. Fresh leaves were extracted with dimethyl formamide solution and placed overnight at (5°C). Chlorophyll A and B as well as carotenoid were measured by spectrophotometer at wave lengths



663, 647 and 470 nm respectively according to the following equation which described by Norani (1982).

Chl. a = $12.70 A_{663} - 2.79 A_{647}$.

Chl. b = $20.76 A_{647} - 4.62 A_{663}$.

Total Chls. = $17.90 A_{647} - 8.08 A_{663}$.

Total carotenoid = $(1000 \times A_{470}) - [(3.72 \text{ Chl. a}) - (104 \text{ Chl. b})] / 229$.

Total indoles and phenols: Total indoles was determined as mg/g dry weight according to Larsen (1962) and total phenols was determined as mg. /g. dry weight by using the Folin calorimetric method by Horwitz (2006)

Minerals content: 0.5 gram of dried leaves samples was digested using the H_2SO_4 and H_2O_2 as described by Cottenie (1980). The extracted samples were used to determine the following minerals:

Leaf nitrogen content: Nitrogen content (gm/100gm D. wt) was determined in the digested solution by the modified micro-Kjeldahl method as described by (Van Schouwenburg and Walinga, 1978).

Leaf phosphorus content: Content of phosphorus (gm/ 100gm D.wt) was determined calorimetrically according to the method of (Jackson, 1958).

Leaf potassium content: Content of potassium (gm/ 100gm D.wt) was determined using the flame photometer method according to (Piper, 1950).

Experimental design and statistical analysis:

The experiment was conducted in a factorial randomization completely block design. The statistical analysis was carried out according to Snedecor and Cochran (1980). Averages were compared by Duncan's multiple range tests ($P \leq 0.05$ level) (Duncan, 1965).

RESULTS AND DISCUSSIONS

Implantation phase:

Germination percentage and Germination speed

Data presented in **Table (3 a)** disclose that highest significant germination percentage and germination speed was recorded for Sour orange seeds. Cleopatra mandarin show significant increment in germination speed for both seasons. Also, it could be noticed that, soaking seeds in 1% AA significantly increase germination percentage and germination speed for the both seasons, Whereas, for the second season soaking in either 1% AA or GA_3 at 20 ppm cause significantly increment in germination percentage, Moreover, results revealed that Sour orange seeds soaked in 1% AA show significantly increase in germination percentage, whereas, Carrizo citrange seeds soaked in 1% AA show significantly increase in germination speed for both seasons .

Varying in germination percentages with different rootstocks documented by many authors. (Sharaf et al., 2016 and Bisi et al., 2020)

The results are in line with El-Sayed and El-Fangary (2008) who found that sour orange rootstock seeds gave the highest germination percentage as compared with Rangpur lime, Volkamer lemon and Cleopatra mandarin rootstocks, and as compared with Carrizo citrange rootstock (Bassal, 2009). Seed germination percentage could be enhanced by several pre-sowing treatments as GA_3 (Sharaf et al., 2016, Dilip et al., 2017 and Singh et al., 2017), and AA (Dorna et al., 2021 and Masserano et al., 2022).

For Cleopatra mandarin and Rangpur lime rootstocks, GA_3 pre-sowing soak treatment showed highest seed germination percentage (Sharaf et al., 2016). Moreover, application of 1% AA significantly increase



seed germination percent and germination rate (Leif Marvin, 2015)

The variation in germination and growth attributes among studied rootstocks may be

due to the difference in genetic behavior of each genotype (Sharaf et al., 2016 and Singh and Chahal, 2021).

Table (3 a): Effect of pre-sowing soak treatments on seed germination phase of some citrus rootstocks in 2020 and 2021 seasons.

Treatments	Season 2020					Means	Season 2021					Means
	Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm		Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm	
Rootstock	Seed germination %											
Rangpur lime	61.3fj	69.7ad	65.3ch	64.0di	66.7cf	65.4B	63.1fj	71.4ad	67.2ch	65.7di	68.5cf	67.2B
Carizzo citrange	52.7k	63.0ej	58.7ik	56.7jk	59.0hk	58.0D	54.2k	64.8dj	60.2ik	58.3jk	60.5hk	59.6C
Cleopatra mandarin	57.0jk	65.7cg	61.0fj	59.3gj	62.3fj	61.1C	58.7jk	67.6cg	62.6fj	60.8gk	63.9ej	62.7C
Sour orange	67.3bf	76.0a	71.3ac	69.0be	73.7ab	71.5A	69.2bf	77.6a	73.2ac	70.7ae	75.5ab	73.2A
Means	59.6C	68.6A	64.1B	62.3BC	65.4B		61.3C	70.4A	65.8B	63.9BC	67.1AB	
Rootstock	seed germination speed											
Rangpur lime	0.95i	2.89c	2.00de	2.24d	1.76f	1.97A	0.95i	2.89c	2.00de	2.24d	1.76f	1.97A
Carizzo citrange	0.73j	4.47a	0.48k	0.48k	0.96i	1.42B	0.73j	4.47a	0.48k	0.48k	0.96i	1.42B
Cleopatra mandarin	1.26g	3.52b	1.74f	1.85ef	1.20g	1.91A	1.26g	3.52b	1.74f	1.85ef	1.20g	1.91A
Sour orange	1.10h	3.21bc	1.92e	1.81ef	1.73f	1.95A	1.10h	3.21bc	1.92e	1.81ef	1.73f	1.95A
Means	1.01D	3.52A	1.54B	1.60B	1.41C		1.01D	3.52A	1.54B	1.60B	1.41C	

Means within column or row and interactions with the same letter (s) are not significantly differences by using Duncan's.

Percentage of damping off and albino implants

Obtained data in **Table (3b)** reveals that Cleopatra mandarin seedling had the lowest significant damping off and albino percentage for both seasons. Moreover, soaking seeds in 1% algae extract significantly reduced both damping and albino implant percentages. Furthermore, obtained results reveal that soaking Cleopatra mandarin seeds in 1% algae

extract reduced significantly damping off and albino implants percentage for both seasons.

The role of algae extract in reducing both damping off and albino percentage may be explained by the secretion of natural hormones, cytokinins, vitamin B12 and essential amino acid as tyrosine and phenylalanine (Aly et al., 2008, Bassal, 2009, Akgul, 2019, Prisa, 2019 and Think, 2019).

Table (3b): Effect of pre-sowing soak treatments on seed germination phase of some citrus rootstocks in 2020 and 2021 seasons

Treatments	Season 2020					Means	Season 2021					Means
	Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm		Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm	
Rootstock	Damping off percentage											
Rangpur lime	22.00ab	13.60e	11.45g	8.25ij	13.10ef	13.68B	23.50a	13.50de	12.40e	8.50hi	13.30de	14.24A
Carizzo citrange	23.75a	15.75de	12.50f	9.75h	16.70d	15.69A	23.90a	13.75d	11.70f	9.10h	14.10d	14.51A
Cleopatra mandarin	19.00c	9.30hi	8.90i	5.60k	9.00i	10.36D	20.10b	8.40hi	7.80i	4.90k	9.00h	10.04B
Sour orange	20.00b	12.00g	10.10h	7.00j	13.40e	12.50C	18.75c	9.10h	8.70 hi	5.50j	10.30g	10.47B
Means	21.19A	12.66B	10.74C	7.65D	13.05B		21.56A	11.19B	10.15C	7.00D	11.68B	
Rootstock	Albino percentage											
Rangpur lime	3.40de	2.40f	1.95g	1.25i	2.70ef	2.34B	3.55f	2.10j	1.80k	1.10m	2.45i	2.20B
Carizzo citrange	9.00a	4.30c	3.95cd	3.75d	5.10bc	5.22A	8.65a	4.00d	3.75e	3.25g	5.00c	4.93A
Cleopatra mandarin	2.90e	0.60l	0.55m	0.40n	0.85k	1.06D	2.75h	0.70op	0.60p	0.30q	0.95mn	1.06D
Sour orange	5.50b	1.50h	0.80k	0.75kl	1.10j	1.93C	5.60b	1.35l	0.65p	0.80o	0.90n	1.86C
Means	5.20A	2.20C	1.81D	1.53E	2.44B		5.14A	2.03C	1.70D	1.36E	2.32B	

Means within column or row and interactions with the same letter (s) are not significantly differences by using Duncan's.



Implant vegetative growth parameters:

Data presented in **Table (4)** show that, Sour orange record the highest significant implant height, number of leaves, root length and number of roots for both seasons. Moreover, data reveal that soaking seeds in either 20 ppm GA₃ or in 1% AA significantly increase all vegetative growth parameters with no significant between both treatments. AS for soaking Sour orange seeds in GA₃ at 20 ppm significantly increase vegetative growth parameters for both seasons.

Sour orange rootstock had the highest values of seedling vegetative growth parameters compared with Rangpur lime, Volkamer lemon, Cleopatra mandarin, Troyer citrange and Carrizo citrange

rootstock. El-Sayed and El-Fangary (2008) and Bassal (2009)

Seed pre-sowing and seedling treatment with Gibberellin improves seedling vegetative growth parameters. (Sharaf et al., 2016, Dilip et al., 2017, Singh et al. 2017, Khopkar et al., 2017 and Sanaullah et al., 2020), As application of gibberellin increases the plasticity of cells wall and reduces cell water potential through starch hydrolysis to sugar, hence, allowing entry of water into cell causing elongation (Arteca, 1996). The growth performance of mung bean plants under salt stress was improved by foliar application of AA, by enhancing growth associated characteristics, such as biomass; shoot height, primary root length, and total leaf area (Rahman, et al., 2019).

Table (4): Effect of pre-sowing soak and foliar applications on seed germination phase some citrus rootstocks seedlings in 2020 and 2021 seasons

Treatments	Season 2020					Means	Season 2021					Means
	Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA3 at 20ppm		Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA3 at 20ppm	
Rootstock	Implant height											
Rangpur lime	7.62 d	7.94 bd	7.87 bd	7.81 cd	8.49 ad	7.95 B	7.84 c	8.15 bc	8.10 bc	8.02 bc	8.70 ac	8.16 B
Carizzo citrange	7.94 bd	8.29 ad	8.14 ad	8.11 ad	8.74 ab	8.24 AB	8.17 ac	8.51 ac	8.34 ac	8.35 ac	8.92 ab	8.46 AB
Cleopatra mandarin	7.76 d	8.17 ad	8.01 bd	7.89 bd	8.61 ab	8.09 AB	7.98 bc	8.37 ac	8.23 ac	8.10 bc	8.84 ab	8.30 AB
Sour orange	8.13 ad	8.54 ac	8.43 ad	8.25 ad	8.94 a	8.46 A	8.36 ac	8.75 ac	8.65 ac	8.45 ac	9.13 a	8.67 A
Means	7.86 B	8.24 AB	8.11 B	8.02 B	8.69 A		8.09 A	8.45 AB	8.33 B	8.23 B	8.90 A	
Rootstock	Number of leaves											
Rangpur lime	2.98 h	4.03 cf	3.95 ef	3.20 gh	4.17 ce	3.67 C	3.07 h	4.14 cf	4.05 df	3.28 gh	4.28 be	3.76 B
Carizzo citrange	3.90 ef	4.31 be	4.27 be	4.19 ce	4.68 ab	4.27 A	3.99 ef	4.41 ae	4.36 be	4.30 be	4.82 ab	4.37 A
Cleopatra mandarin	3.34 gh	4.20 ce	4.05 cf	3.59 fg	4.28 be	3.89 B	3.43 gh	4.31 be	4.14 cf	3.66 fg	4.40 ae	3.99 B
Sour orange	3.99 d-f	4.49 ab	4.44 ad	4.28 be	4.81 a	4.40 A	4.08 c-f	4.60 ac	4.56 ad	4.37 ae	4.91 a	4.51 A
Means	3.55 D	4.26 AB	4.18 B	3.81 C	4.49 A		3.64 B	4.36 A	4.28 A	3.90 B	4.60 A	
Rootstock	Root length (cm)											
Rangpur lime	4.35 b	8.75 a	5.75 b	4.67 b	6.17 b	5.94 B	4.50 b	8.95 a	5.91 b	4.81 b	6.34 b	6.10 B
Carizzo citrange	5.69 b	9.82 a	6.21 b	6.11 b	9.03 a	7.37 A	5.86 b	10.04 a	6.36 b	6.29 b	9.26 a	7.56 A
Cleopatra mandarin	4.88 b	8.97 a	5.89 b	5.24 b	8.81 a	6.76 AB	5.02 b	9.18 a	6.05 b	5.38 b	9.03 a	6.93 A
Sour orange	5.82 b	10.08 a	6.47 b	6.25 b	9.42 a	7.61 A	5.98 b	10.32 a	6.64 b	6.40 b	9.65 a	7.80 A
Means	5.19 B	9.41 A	6.08 B	5.57 B	8.36 A		5.34 B	9.62 A	6.24 B	5.72 B	8.57 A	
Rootstock	Number of roots											
Rangpur lime	7.87ef	8.93ce	8.53ef	8.45ef	8.82 ce	8.52 B	8.10h	9.15cf	8.77fh	8.68 fh	9.06cg	8.75 B
Carizzo citrange	8.42ef	10.02ab	9.17be	9.04ce	9.21 be	9.17 A	8.67fh	10.31ab	9.40bf	9.31 cf	9.45bf	9.43 A
Cleopatra mandarin	7.93ef	9.15be	8.70de	8.51ef	8.99 ce	8.65 B	8.16gh	9.41bf	8.93ch	8.73 fh	9.21cf	8.89 B
Sour orange	8.62ef	10.29a	9.56ad	9.25be	9.61ac	9.46 A	8.86dh	10.50a	9.80ad	9.47 be	9.85ac	9.70 A
Means	8.21 C	9.59 A	8.99 B	8.81 B	9.16 AB		8.45 C	9.84 A	9.23 B	9.05 B	9.39 AB	

Means within column or row and interactions with the same letter (s) are not significantly differences by using Duncan's.

Seedlings phase parameters:
Seedlings vegetative growth parameters: Presented data in **Table (5)**

showed that Carizzo citrange seedling had the highest significant stem length and diameter, while sour orange seedling



showed the highest significant increment in leaf area and stem diameter. However, Cleopatra mandarin showed the highest significant leaf number for both seasons. Moreover, obtained data showed the 1% AA application achieved the highest significant value for all vegetative growth parameters. While, 20 ppm GA₃ application increase stem length significantly and 1% YE application show highest significant stem diameter for both seasons. On the other hand, the results revealed that spraying sour orange seedlings with 1% AA significantly increase both stem diameter and leaf area, while foliar application of 1% AA to Cleopatra mandarin seedlings significantly increased number of leaves/plant for both

seasons of study. That effect for acetic acid in augmenting vegetative growth may related to its role improving cell membrane integrity, nutritional balance and roots nutrients absorb selectively in mung bean, which enhance biomass production and shoot growth (Rahman, et al., 2019)

Improving cell membrane integrity and nutritional balance, and content of amino acid may have the positive effect of yeast extraction attributed to its high nutrient, protein, vitamin B and cytokinin content, which improve plant nutritional status and enhance cell division and enlargement (Abd El-Moniem et al. 2008, Ahmed et al., 1997, Mostafa and El-Hosseiny, 2001).

Table (5):Effect of pre-sowing soak and foliar applications on vegetative growth phase of some citrus rootstocks seedlings in 2020 and 2021 seasons.

Treatments	Season 2020					Season 2021						
	Water (Contro)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA3 at 20ppm	Means	Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA3 at 20ppm	Means
Rootstock	Stem length (cm.)											
Rangpur lime	34.33h	44.33d	41.50e	40.00f	43.67 d	40.77 D	34.67 h	44.85 e	41.00 g	40.25 g	44.50 e	41.05 D
Carizzo citrange	41.33ef	55.67a	45.50c	44.50d	54.50 b	48.30 A	42.50 f	58.60 a	46.90 d	46.00 d	56.25 ab	50.05 A
Cleopatra mandarin	34.99h	46.85c	42.00e	41.67e	45.98 c	42.30 C	35.75 h	48.33 c	42.33 f	43.50 ef	47.50 c	43.48 C
Sour orange	38.51g	54.00b	43.33d	42.50de	53.50 bc	46.37 B	39.67gh	55.33 b	43.67 ef	44.67 e	55.00 b	47.67 B
Means	37.29 D	50.21 A	43.08B	42.17 C	49.41 A		38.15 D	51.78 A	43.48 B	43.61 C	50.81 A	
Rootstock	Stem diameter (cm.)											
Rangpur lime	0.31d	0.44b	0.41bc	0.38c	0.33 cd	0.37 C	0.28 f	0.46 b	0.40 d	0.38 d	0.34 e	0.37 C
Carizzo citrange	0.46b	0.58a	0.54a	0.53ab	0.48 b	0.52 A	0.45 c	0.58 a	0.55 a	0.52 ab	0.49 b	0.52 A
Cleopatra mandarin	0.36c	0.47b	0.44b	0.42bc	0.37 c	0.41 B	0.38 d	0.50 b	0.45 c	0.41 c	0.39 d	0.43 B
Sour orange	0.42bc	0.56a	0.53ab	0.51ab	0.45 b	0.49 A	0.43 c	0.57 a	0.55 a	0.53 ab	0.47 b	0.51 A
Means	0.39 D	0.51 A	0.48A	0.46 B	0.41 C		0.39 D	0.53 A	0.49 AB	0.46 B	0.42 C	
Rootstock	leaf area (cm²)											
Rangpur lime	5.94e	12.73c	6.54e	7.33de	9.95 d	8.50 B	6.33 de	13.50 ab	6.90 d	8.10 c	10.13 c	8.99 B
Carizzo citrange	4.93f	11.17cd	5.89e	5.98e	9.00 d	7.39 C	5.00 e	12.00 b	6.10 de	6.94 d	9.75 c	7.96 C
Cleopatra mandarin	5.33ef	13.75b	6.75e	7.56de	10.96 cd	8.87 B	5.54 e	14.19 a	7.45 d	8.23 c	12.00 b	9.48 B
Sour orange	7.00e	14.85a	7.35de	8.76d	11.91 c	9.97 A	7.25 d	14.93 a	8.00 cd	9.22 c	13.85 a	10.65 A
Means	5.80 D	13.13 A	6.63CD	7.41 C	10.46 B		6.03 E	13.66 A	7.11 D	8.12 C	11.43 B	
Rootstock	Number of leaves / seedling											
Rangpur lime	12.50f	23.67b	18.86cd	19.00cd	13.00f	17.41 C	12.77 e	24.75 b	19.33 cd	20.33 c	13.46 e	18.13 C
Carizzo citrange	15.75e	24.75b	20.09cd	21.00c	16.00e	19.52 B	16.09 d	25.25 b	20.86 c	21.75 c	17.33 d	20.26 B
Cleopatra mandarin	17.00d	28.67a	21.77c	21.33c	17.75d	21.30 A	18.33 cd	29.00 a	22.00 c	22.75 c	18.86 cd	22.19 A
Sour orange	10.75fg	22.50c	16.25e	18.09d	11.00fg	15.72 D	11.86 ef	24.33 b	17.00 d	18.50 cd	12.75 e	16.89 D
Means	14.00 C	24.90A	19.24B	19.86 B	14.44 C		14.76 C	25.83 A	19.80 B	20.83 B	15.60 C	

Means within column or row and interactions with the same letter (s) are not significantly differences by using Duncan's.

Plant pigments:

Data presented in **Table (6)** demonstrate that plant pigment concentration varies within different studied rootstocks; however, the highest leaf total chlorophyll and total

carotenoids content was recorded for sour orange rootstock in both seasons. Besides, data showed that 1% AA treatment achieved the highest significant value of total chlorophylls, chlorophyll a, chlorophyll b,



and carotenoids, also, the obtained result disclose that, the highest significant value for leaf chlorophyll a and chlorophyll b content was obtained for Cleopatra mandarin, Carizzo citrange rootstocks treated with 1% acetic acid, respectively for both seasons; while the foliar application of 1% AA to Sour orange rootstock seedling increase significantly total chlorophylls content. Furthermore, the effect that acetic acid stimulates the production of jasmonic

acid (JA) may be connected to the action of AA application. Kudo and Kim (2023). Jasmonic acid increased the efficiency of photosynthetic processes and improved photosynthetic pigments. Increased activity of enzymes involved in the manufacture of chlorophyll, such as α -amino levulinic acid dehydratase and proto-chlorophyllide reductase, may have contributed to the higher amount of chlorophyll pigments (Sirhindi et al., 2020).

Table (6):Effect of pre-sowing soak and foliar applications on leaves pigment content of some citrus rootstocks seedlings in 2020 and 2021 seasons.

Treatments	Season 2020					Season 2021					Means	
	Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm	Water (Control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm		
Rootstock	Leaf chl. A											
Rangpur lime	0.46 g	0.65 d	0.57 e	0.61 de	0.51 f	0.56 C	0.43 f	0.64 d	0.52 e	0.58 de	0.47 f	0.53 C
Carizzo citrange	0.51 f	0.75 bc	0.63 d	0.69 c	0.57 e	0.63 B	0.53 e	0.69 c	0.60 de	0.65 d	0.56 e	0.61 B
Cleopatra mandarin	0.60 de	0.94 a	0.79 b	0.88 ab	0.69 c	0.78 A	0.60 de	0.95 a	0.79 b	0.91 ab	0.72 c	0.79 A
Sour orange	0.56 e	0.89 ab	0.76 bc	0.81 b	0.65 d	0.73 AB	0.59 de	0.90 ab	0.77 bc	0.82 b	0.60 de	0.74 AB
Means	0.53 E	0.81 A	0.69 C	0.75 B	0.60 D		0.54 E	0.80 A	0.67 C	0.74 B	0.59 D	
Rootstock	Leaf chl. B											
Rangpur lime	0.32 e	0.41 d	0.37 de	0.40 d	0.33 e	0.37 C	0.30 e	0.38 d	0.34 e	0.42 cd	0.31 e	0.35 C
Carizzo citrange	0.39 de	0.58 a	0.47 bc	0.53 b	0.41 d	0.48 A	0.41 cd	0.57 a	0.45 c	0.55 a	0.37 d	0.47 A
Cleopatra mandarin	0.40 d	0.49 b	0.41 d	0.46 bc	0.38 de	0.43 B	0.38 d	0.53 ab	0.46 c	0.51 b	0.41 cd	0.46 A
Sour orange	0.37 de	0.52 b	0.45 c	0.48 bc	0.39 de	0.44 B	0.36 de	0.54 ab	0.42 cd	0.50 b	0.39 d	0.44 B
Means	0.37 C	0.50 A	0.43 B	0.47 AB	0.38 C		0.36 C	0.51 A	0.42 B	0.50 A	0.37 C	
Rootstock	Total chls.											
Rangpur lime	1.41 i	1.57 f	1.50 g	1.55 fg	1.44 i	1.50 C	1.44 h	1.62 e	1.53 f	1.59 ef	1.47 g	1.53 CD
Carizzo citrange	1.49 h	1.64 e	1.53 g	1.58 f	1.47 h	1.54 C	1.49 g	1.68 de	1.57 ef	1.63 e	1.53 f	1.58 C
Cleopatra mandarin	1.55 fg	1.80 c	1.65 e	1.72 d	1.57 f	1.66 B	1.57 ef	1.85 b	1.68 de	1.74 cd	1.62 e	1.69 B
Sour orange	1.63 ef	1.89 a	1.75 d	1.85 b	1.66 e	1.76 A	1.60 e	1.92 a	1.77 c	1.90 a	1.72 d	1.78 A
Means	1.52 E	1.73 A	1.61 C	1.68 B	1.54 D		1.53 E	1.77 A	1.64 C	1.72 B	1.59 D	
Rootstock	Total carotenoids											
Rangpur lime	0.39 i	0.49 g	0.46 gh	0.44 h	0.40 i	0.44 C	0.37 i	0.52 fg	0.45 g	0.42 h	0.38 i	0.43 D
Carizzo citrange	0.45 h	0.58 e	0.48 g	0.53 f	0.44 h	0.50 B	0.46 g	0.63 e	0.53 fg	0.56 f	0.48 g	0.53 C
Cleopatra mandarin	0.63 d	0.79 a	0.71 b	0.73 b	0.67 c	0.70 A	0.65 e	0.84 a	0.74 c	0.79 b	0.69 d	0.74 A
Sour orange	0.58 e	0.82 a	0.65 c	0.72 b	0.61 de	0.68 A	0.55 f	0.76 bc	0.63 e	0.69 d	0.58 ef	0.64 B
Means	0.51 D	0.67 A	0.58 C	0.61 B	0.53 D		0.51 D	0.69 A	0.59 C	0.62 B	0.53 D	

Means within column or row and interactions with the same letter (s) are not significantly differences by using Duncan's.

Total indoles and phenols:

It is obvious from the obtained data in **Table (7)** that the highest significant value of leaves total indole and lowest significant value of leaves total phenol was achieved by sour orange rootstock. Furthermore, data reveal that, foliar application of 1% AE increased leaves total indoles, also both 1% AE and 1% AA applications reduced leaves total phenols contents with non significant

difference for both seasons. Moreover, Carizzo citrange and sour orange seedling treated with 1% AE achieved significantly the highest leaves total indole content and lowest total phenol content in both seasons.

Leaf macro elements content:

Data presented in **Table (8)** illustrated that sour orange seedling had the highest significant percentage of leaf nitrogen and potassium content, while the highest



significant percentage of leaf phosphorus content was obtained from Rangpur lime and Cleopatra mandarin rootstocks in both seasons.

Moreover, obtained data showed that 1% AA application achieved the highest significant value for all leaf macro elements

content. In the same concern, Kudo and Kim (2023). Pointed out that acetic acid helped protein structure stabilization, metabolic function optimization, and K⁺ homeostasis maintenance, which is essential for cell proliferation

Table (7): Effect of pre-sowing soak and foliar applications on leaves total indole and phenol of some citrus rootstocks seedlings in 2020 and 2021 seasons.

Treatments	Season 2020						Season 2021					
	Water (control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm	Means	Water (Control)	AA at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm	Means
Leaves total indole												
Rangpur lime	0.88 h	1.23d	1.09 f	1.28 cd	0.95 g	1.09 C	0.93 h	1.24 f	1.12 g	1.31 e	0.98 h	1.12 C
Carizzo citrange	1.27 cd	1.55 ab	1.43 bc	1.58 a	1.31 cd	1.43 AB	1.32 e	1.57 b	1.46 c	1.59 b	1.34 e	1.46 AB
Cleopatra mandarin	1.19 e	1.42 bc	1.37 c	1.49 b	1.24 d	1.34 B	1.17 g	1.47 c	1.39 d	1.54 bc	1.23 f	1.36 B
Sour orange	1.34 c	1.56 ab	1.48 b	1.61 a	1.36 c	1.47 A	1.36 de	1.58 b	1.52 bc	1.64 a	1.40 d	1.50 A
Means	1.17 CD	1.44 AB	1.34 B	1.49 A	1.22 C		1.20 CD	1.47 AB	1.37 B	1.52 A	1.24 C	
Leaves total phenol												
Rangpur lime	1.58 a	1.28 c	1.41 b	1.23 d	1.53 a	1.41 A	1.52 a	1.25 c	1.39 b	1.19 d	1.48 a	1.37 A
Carizzo citrange	1.30 c	1.01 g	1.12 f	0.91 h	1.25 c	1.12 C	1.32 c	0.98 f	1.09 e	0.93 g	1.28 c	1.12 C
Cleopatra mandarin	1.43 b	1.09 f	1.19 e	0.96 gh	1.37 b	1.21 B	1.44 b	1.10 e	1.21 d	0.98 f	1.36 bc	1.22 B
Sour orange	1.23 d	0.95 h	1.07 f	0.88 h	1.19 e	1.06 D	1.19 d	0.91 g	1.03 e	0.84 h	1.16 de	1.03 D
Means	1.39 A	1.08 D	1.20 C	1.00 E	1.34 B		1.37 A	1.06 D	1.18 C	0.99 E	1.32 B	

Means within column or row and interactions with the same latter (s) are not significantly differences by using Duncan's.

Table (8): Effect of pre-sowing soak and foliar applications on leaf macro elements of some citrus rootstocks seedlings in 2020 and 2021 seasons.

Treatments	Season 2020						Season 2021					
	Water (control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm	Means	Water (control)	Acetic acid at 1%	Yeast extract at 1%	Algae extract at 1%	GA ₃ at 20ppm	Means
Nitrogen percentage												
Rangpur lime	1.44 g	1.87 c	1.49 fg	1.50 f	1.46 g	1.55 B	1.42 g	1.90 c	1.51 f	1.53 e	1.45 g	1.56 B
Carizzo citrange	1.50 f	2.23 ab	1.61 e	1.72 d	1.49 fg	1.71 A	1.53 e	2.24 a	1.64 d	1.66 d	1.55 de	1.72 A
Cleopatra mandarin	1.45 g	1.98 b	1.51 f	1.62 e	1.45 g	1.60 AB	1.47 f	2.04 b	1.54 e	1.53 e	1.49 f	1.61 AB
Sour orange	1.52 f	2.31 a	1.56 ef	1.67d	1.54 ef	1.72 A	1.51 f	2.28 a	1.59 de	1.63 d	1.54 e	1.71 A
Means	1.48 D	2.10 A	1.54 C	1.63 B	1.49 D		1.48 D	2.12 A	1.57 B	1.59 B	1.51 C	
Phosphorus percentage												
Rangpur lime	0.07 de	0.16 ab	0.12 b	0.11 bc	0.07 de	0.11 A	0.07 e	0.18 a	0.11 c	0.11 c	0.07 e	0.11 B
Carizzo citrange	0.05 e	0.13 b	0.09 cd	0.10 c	0.07 de	0.09 AB	0.04 fg	0.12 bc	0.10 cd	0.09 d	0.05 f	0.08 C
Cleopatra mandarin	0.08 d	0.18 a	0.13 b	0.13 b	0.09 cd	0.12 A	0.09 d	0.18 a	0.16 ab	0.14 b	0.10 cd	0.13 A
Sour orange	0.03 f	0.11 bc	0.10 c	0.09 cd	0.04 ef	0.07 B	0.05 f	0.13 b	0.13 b	0.11 c	0.06 ef	0.10 B
Means	0.06 C	0.15 A	0.11 B	0.11 B	0.07 C		0.06 D	0.15 A	0.13 B	0.11 C	0.07 D	
Potassium percentage												
Rangpur lime	0.65 de	0.84 c	0.76 cd	0.74 cd	0.68 d	0.73 BC	0.63 f	0.86 b	0.73 e	0.72 e	0.67 ef	0.72 D
Carizzo citrange	0.79 c	1.03 a	0.93 b	0.89 c	0.82 c	0.89 AB	0.83 bc	1.07 a	0.91 b	0.87 b	0.84 bc	0.90 B
Cleopatra mandarin	0.67 d	0.99 a	0.79 c	0.78 c	0.67 d	0.78 B	0.67 ef	1.02 a	0.83 bc	0.80 c	0.71 e	0.81 C
Sour orange	0.84 c	1.14 a	0.97 b	0.95 b	0.85 c	0.95 A	0.87 b	1.18 a	0.99 ab	0.95 ab	0.86 b	0.97 A
Means	0.74 C	1.00 A	0.86 B	0.84 B	0.76 C		0.75 C	1.03 A	0.87 B	0.84 B	0.77 C	

Means within column or row and interactions with the same latter (s) are not significantly differences by using Duncan's

Conclusions

From the obtained findings in this study, it can be concluded that seeds soaked and

seedlings treated with 1% acetic acid for 24 hours were more effective enhancing germination percentage and overall performance for all rootstocks which is a



natural substance. Moreover, sour orange rootstock was more effective than other

rootstocks in responding to the effect of the conducted treatments.

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إنبات بذور ونمو شتلات بعض أصول الموالح تحت تأثير بعض المواد المشجعة

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أجريت هذه الدراسة خلال موسمين متتاليين (2020 و 2021) في صوبة بلاستيك بوحدة إنتاج شتلات الموالح بمعهد بحوث البساتين، مركز البحوث الزراعية - محافظة الجيزة لدراسة تأثير معاملات النقع المختلفة للبذور قبل الزراعة على نسبة الإنبات ومظاهر النمو لأربعة من أصول الموالح وهي: ليمون الرانجبور، كاريزو سترانج ، واليوسفي كليوباترا والنارنج. تم إجراء خمس معاملات ما قبل الزراعة عن طريق نقع بذور الأصول المستخدمة لمدة 24 ساعة على النحو التالي: ماء الصنبور (الكنترول)، حمض الخليك بتركيز 1% ، مستخلص الخميرة بتركيز 1%، مستخلص الطحالب بتركيز 1% والجبرلين بتركيز 20 جزء في المليون. تم استخدام نفس معاملات النقع الخمس السابقة وبنفس التركيزات رشاً على الأوراق شهرياً للشتلات حتى موعد التطعيم.

أوضحت النتائج أن نوع الأصل والمعاملة ما قبل الزراعة وكذلك الرش الورقي أثرت بدرجة معنوية على نسبة الإنبات للبذور وصفات البادرات في كلا الموسمين . أظهرت البيانات أن البذور المنقوعة و كذلك الشتلات المعاملة رشاً على الأوراق باستخدام حمض الخليك بتركيز 1% لمدة 24 ساعة كانت الأكثر فاعلية مقارنة بالمعاملات الأخرى من حيث تحسين نسبة الإنبات للبذور وجميع الصفات المدروسة للبادرات متمثلاً في ارتفاع البادرة، عدد الأوراق، طول الجذر وعدد الجذور والصيغات ومحتوى الأوراق من العناصر. وفيما يتعلق بتأثير نوع الأصل، كان من الواضح أن أصل النارنج حقق أعلى نسبة إنبات وأعلى صفات للبادرات مقارنة بجميع الأصول الأخرى.