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EFFECT OF SPRAYING WITH ORGANIC ACIDS AND VERNALIZATION ON GROWTH, PRODUCTION AND CHEMICAL CONTENT OF *HIBISCUS SABDARIFFA* L. PLANT

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ABSTRACT: This experiment was carried out at the Agricultural Research Station of the Agricultural Research Center in Gemmayzeh, Gharbia Governorate, Egypt during the two consecutive summer seasons of 2019 and 2020, with the aim to evaluate the effect of vernalization and some organic acids (citric and ascorbic acids) on roselle (*Hibiscus sabdariffa* L.) plants. The combined treatments were as follows, NPK 100% only (cont.) + vernalization (T1), cont. + non-vernalization (T2), ascorbic acid in addition to NPK 50% (AA) + vernalization (T3), AA + non-vernalization (T4), citric acid in addition to NPK 50% (CA) + vernalization (T5), CA + non-vernalization (T6), AA + CA + vernalization (T7), AA + CA + non-vernalization (T8). It was found that the effect of vernalization and some organic acids on plants gave highly significant values compared to control on all tested vegetative growth traits and chemical measurements. The best spraying treatments were recorded by citric acid on most vegetative measurements i.e. plant height (cm), weight of 100 seeds (g), fixed oil % etc. The vernalization recorded the best results on all parameters under study. Regarding the combined treatments between these two factors, the highest values of the studied characteristics (vegetative growth traits, fixed oil % and the organic acid i.e. stearic and linoleic acids) were obtained by T7 and T5. In general, applying T7 (ascorbic + citric acids (in addition to NPK 50%) + vernalization) is recommended to obtain the best results for the most studied measurements of roselle.

Keywords: roselle, ascorbic acid, citric acid, vernalization, sepals, oil.

INTRODUCTION

Medicinal plants are utilized for treating several disorders that are either rare or non-curable remedies by modern medicine systems (Abadi *et al*., 2015). *Hibiscus sabdariffa* L. is the most favorable crop for many countries as it is respectably easy to create. Firstly, the roselle sepals (calyx) have a high content of anthocyanin pigment and organic acids (Cisse *et al*., 2009) and have been used in Egyptian food as drinks, colour and flavour additives as well as it is considered because Egypt is the country that roselle originated (F.A.O., 2004 and Ismail *et al*., 2008). In addition, its seeds are utilized for their oil, and remedial properties and can be utilized as food and fibers. Also, it is utilized in food and clinical trials (Ali *et al*., 2005), additionally, it is utilized in hypertension treatment (Wahabi *et al*., 2010) as well as in hyperlipidemia treatment (Hopkins *et al*., 2013). Moreover, Da-Costa *et al.* (2014) and Singh *et al.* (2017) mentioned that roselle sepals have various restorative significant mixtures called phytochemicals, is notable for their therapeutic and healthful properties. Fasoyiro *et al.* (2005), Mahadevan and Pradeep (2009) and Shamkhi *et al*. (2012) reported that roselle leaf juice lessens hypertension, fortifies the heart’s serenity and diminishes blood consistency and is ample in vitamins C and B, as well as nutrients (Ca, Fe and P), anthocyanins, thiamine and riboflavin. Roselle seeds contain about 17-30% fatty oil, which is comparable in its properties to the seed oil of cotton (Louis *et al.*, 2013 and Hussin *et al.*, 1991). In general, roselle is rich in organic acids, minerals, vitamin C and total sugar in its calyx, leaves and seeds (Galaudu, 2006 and Cisse *et al*., 2009). Researchers have demonstrated that roselle tea contains an enzyme inhibitor so, it is possible that drinking a cup of hibiscus tea after meals can assist in weight loss (Tomes, 1990 and Bako *et al.*, 2014).

Ascorbic acid (AA) is one of the initial serious water-soluble antioxidants in plants as a coenzyme in responses by which carbohydrates, proteins and fats are metabolized and AA increased chlorophyll and productivity and improved plant growth. Recently, AA can be utilized as an antioxidant used to guard against adverse influences of the environment and oxidative stress (Chen and Gallie, 2006). Ascorbic acid influenced many biological processes (Inskbash and Iwaya, 2006). Moreover, applying AA at 200 ppm combined with a half dose of chemical fertilizer + 15 m^3 compost/fed enhanced growth (plant height and fresh and dry weights of branches/plant) and productivity (seed yield/plant, P, Zn leaf uptake and sepals anthocyanin content) of roselle plants (Youssef *et al.*, 2014). Gheeth *et al.* (2013) on peas, found that AA at 150 ppm improved all the traits being studied i.e. plant growth, yield and yield components and safe for animal feeding and human consumption. In addition, Azoz *et al.* (2016) found that using AA at 300 ppm promoted sweet basil vegetative growth, increased the productivity of fresh herbs and seeds, and improved volatile and fixed oil quality.

Citric acid (CA) is a six-carbon organic acid, that plays a central role in the CA-cycle in mitochondria by causing cellular energy through phosphorylative oxidation reactions. It's generated by the addition of acetyl-CoA to oxaloacetic acid that's converted to succinate. It occurs naturally in citrus fruits. Using citric acid as foliar spray increased yield components on strawberry fruits (Mandour *et al.*, 2019), apricot ‘Canino’ fruit yield and quality (Haggag *et al*., 2016), and dill fruits and essential oil yield than the control (Jafari and Hadavi, 2012 a). In addition, Jafari and Hadavi (2012 b) on basil and Miri *et al.* (2015) on thyme mentioned that using citric acid application enhanced growth, yield and essential oil content. However, foliar pre-harvest application of the combination between salicylic and citric acids within the soilless culture increased the vase life of cut rose flowers (Hajreza *et al.*, 2013).

Vernalization is typically defined as the promotion of flower formation by a period of low temperatures, generally between -5 and 15 °C, with a broad optimum between 1 and 7 °C or as the acceleration of the power to flower by a chilling treatment (Chouard, 1960). This competence to flower by vernalization can be stable through mitotic cell divisions (Lang, 1965). Runkle *et al*. (1998) recommend that the flowering of many herbaceous perennials is influenced by vernalization and photoperiod. Many researchers studied the effect of vernalization treatment on enhancing growth and increasing flowering of *Oenothera biennis* (Clough *et al*., 2001), *Osteospermum ecklonis* (Suzuki and Metzger, 2001), broccoli (Li *et al*., 2010) and on many plant species (Dennis and Peacock, 2007). Darapuneni *et al*. (2014) demonstrated that the choice for photoperiodic sensitivity and vernalization in flax genotypes and introgression of those parameters into recently adapted spring-genotypes is required for the development of high-yielding flax genotypes for southern Great Plains production areas. Khattab and El Sherif (2016) studied the influence of seed vernalization (room temperature, 6 and 12 °C) and duration (1, 2 and 3 weeks) as well as 3 planting times (April, May, and June) on yield components and composition of calyx extract of roselle, the results showed that vernalization resulted in a higher number of leaves, more fruits/plant and improved total calyx yield. Parikshit *et al.* (2017) on wheat noticed that vernalization with coldness application to water soaked-seeds, slightly germinated seeds, or seedlings hastened the flowering time of the plants, and also enhanced plant growth and productivity.

Therefore, the main goal of this study was to investigate the effect of vernalization and some organic acids on the growth, productivity and chemical constituents of roselle (*Hibiscus sabdariffa* L.) plants.

MATERIALS AND METHODS

This experiment was carried out at the Agricultural Research Station of the Agricultural Research Center in Gemmayzeh, El-Gharbia Governorate, during the two consecutive summer seasons of 2019 and 2020, with the aim of evaluating the effect of vernalization and some organic acids (citric acid (CA) and ascorbic acid (AA) on roselle (*Hibiscus sabdariffa* L.) plants.

Plant materials:

Seeds of roselle (*Hibiscus sabdariffa* L., cv. Sabahiya 17) were obtained from the Medicinal and Aromatic Plants Research Department, Horticulture Research Institute, Agricultural Research Center, El-Dokki, Cairo, Egypt.

Field preparation and seeds sowing:

The field was ploughed twice as cross-ploughing to get rid of the remains of the previous crops. The sector was divided into experimental plots, the area of the experimental unit was 2.16 m^2 (1.2 m length and 1.80 m width), each plot contained 3 rows, the space between hills was 30 cm, every hill contained 2 plants and every plot contained from 25 to 30 plants. Seeds were planted on the 14^{th} of May of each season. The physical and chemical properties of experimental farm soil are shown in Table (1).

Treatments:

1. Organic acids:

The produced plants were foliar sprayed two times with both ascorbic and citric acids (each at 150 ppm), the first one was done after 60 days from sowing, while the second one was done after 90 days from sowing. While, control treatment was sprayed with tap water. Both ascorbic and citric acids were obtained from Al-Gomhoria Chemicals Company Egyptian, Egypt. In this regard the organic acid treatments were as follows:

1. Control without organic acids (cont.).
2. Ascorbic acid at 150 ppm (AA).
3. Citric acid at 150 ppm (CA).
4. AA + CA (both at 150 ppm) (AA + CA).

NPK fertilization was applied at the recommended dose (100%) in case of control plants, while half dose (50%) was added in case of spraying with ascorbic, citric or ascorbic + citric acids.

2. Vernalization treatments:

The seeds were divided into two groups, the first one was subjected to vernalization treatment by soaking the seeds in water (at a temperature of 13 °C) for two weeks before sowing, while, the second group was sowed without vernalization. Vernalized and non-vernalized seeds were sowed on the same date (14^{th} of May).

3. Interaction treatments

1. Cont. + vernalization (T1).
2. Cont. + non-vernalization (T2).
3. AA + vernalization (T3).
4. AA + non-vernalization (T4).
5. CA + vernalization (T5).
6. CA + non-vernalization (T6).
7. AA + CA + vernalization (T7).
8. AA + CA + non-vernalization (T8).

Table 1. Physical and chemical properties of experimental farm soil (average of two seasons).

Mechanical analysis				
Clay (%)	Silt (%)	SP (saturation %)	ρb (Mg m^{-3})	Soil texture
51.14	37.17	68.00	1.12	Clayey

Chemical analysis											
pH	E.C. (dS/m)	OM (%)	Cations (meq/l)				Anions (meq/l)			Available K (ppm)
			Mg^{++}	Ca^{++}	K^{+}	Na^{+}	Cl^{-}	HCO_3^{-}	SO_4^{--}	
7.95	0.55	0.74	0.90	1.70	0.80	2.48	3.29	0.23	1.89	0.80

NPK fertilization:

The recommended amounts of mineral N, P and K fertilizers for roselle (*Hibiscus sabdariffa* L.) were applied to the control plants (without organic acids) at 163 kg/fed of ammonium nitrate (20.5% N), 150 kg/fed calcium superphosphate (15.5 % P_2O_5) and 75 kg/fed potassium sulphate (48% K_2O) as a source of N, P and K, respectively. Half of these amounts (50%) were applied to the plants treated with either ascorbic or citric acids.

Experimental layout:

These treatments were arranged in a split-plot design with three replications, organic acids (4 levels) represented the main plot, while vernalization (2 levels) represented the sub-plot.

Data recorded:

At the harvesting stage of roselle (*H. sabdariffa* L.), where the plants were approximately 120 days, the following data were recorded: plant height (cm), branch number/plant, number of fruits/plant, fresh weight of fruits (g/plant), number of days until flowering (the appearance of the first flower), fresh weight of sepals (g/plant), dry weight of sepals (g/plant), dry weight of sepals (kg/fed), weight of 100 seeds (g), weight of seeds (g)/plant, weight of seeds (kg/fed), fixed oil %, total carbohydrate % in sepals, anthocyanin content (μM) and vitamin C content (mg/100 g).

Chemical constituents:

A sample of dry sepals was randomly taken from each treatment for chemical analysis. Furthermore, seed fixed oil of roselle was extracted using petroleum ether in a soxhlet system HT apparatus according to the methods of A.O.A.C. (1990). Total anthocyanin in air-dried harvested roselle calyxes was determined according to the method described by Du and Francis (1973). Ascorbic acid was estimated using direct iodometric titration as described by Suntornsuk *et al.* (2002). The gas chromatography (GC) analysis of the fixed oil samples was carried out using gas chromatography instrument stands at the Medicinal and Aromatic Plants Res. Dept. Laboratory, Horticultural Research Institute according to the procedure described in the Egyptian Pharmacopoeia (2005).

Statistical analysis:

The present study's data were statistically analysed using Statistix computer program Version 9 (Analytical Software, 2008) and the differences between means were compared by L.S.D. test at a 5% probability level according to Snedecor and Cochran (1980).

RESULTS

Plant height (cm):

The findings in Table (2) presented that all organic acid treatments had a significant effect compared to control (untreated plants) on plant height (cm) in two seasons, the mastery was to CA which increased the plant height (cm) to the highest values (198.84 and 229.58 cm in both seasons, respectively).

Vernalization treatment, on the other hand significantly increased the plant height (cm) to 202.00 and 231.67 cm in the first and second seasons, respectively when compared with non-vernalization treatment 180.34 and 215.92 cm in the first and second seasons, respectively.

Table 2. Effect of ascorbic, citric acids, and vernalization on plant height (cm), number of branches/plant, number of fruits/plant and fresh weight of fruits (g/plant) of roselle (*Hibiscus sabdariffa* L.) plants in the two seasons of 2019 and 2020.

Treatments	Plant height (cm)		Number of branches/plant		Number of fruits/plant		Fresh weight of fruits (g/plant)	
	2019	2020	2019	2020	2019	2020	2019	2020
				Organic acids				
Cont.	182.17	216.75	7.17	8.34	41.84	72.16	313.20	654.00
AA	188.82	220.00	7.77	9.09	46.17	74.39	333.60	753.10
CA	198.84	229.58	9.36	10.33	50.83	82.64	396.50	795.80
AA + CA	194.84	228.83	9.28	9.86	52.50	84.31	400.80	803.20
L.S.D. 5%	2.33	2.08	0.93	0.53	2.10	4.63	11.80	30.99
				Vernalization				
vern.	202.00	231.67	10.17	10.14	55.66	90.94	431.80	834.90
n-vern.	180.34	215.92	6.62	8.67	40.01	65.81	290.30	668.20
L.S.D. 5%	1.13	4.26	0.33	0.19	0.60	2.61	20.39	40.23
				Organic acids × vernalization				
Cont. + vern. (T1)	193.00	223.83	9.33	8.67	49.67	87.44	367.40	765.40
Cont. + n-vern. (T2)	171.34	209.67	5.00	8.00	34.00	56.89	259.00	542.70
AA + vern. (T3)	197.33	227.00	9.66	9.67	55.78	87.11	391.60	829.10
AA + n-vern. (T4)	180.34	213.00	5.87	8.50	36.56	61.67	275.70	677.00
CA + vern. (T5)	216.00	239.67	11.55	12.00	60.33	99.45	501.00	885.40
CA + n-vern. (T6)	181.67	219.50	7.16	8.67	41.33	65.83	300.70	706.20
AA + CA + vern. (T7)	201.67	236.17	10.11	10.22	56.84	89.78	467.30	859.60
AA + CA + n-vern. (T8)	188.00	221.50	8.45	9.50	48.15	78.83	325.70	746.90
L.S.D. 5%	2.57	6.15	0.97	0.55	2.14	5.37	29.76	60.60

Cont. = without organic acids + NPK 100%; AA= ascorbic acid at 150 ppm + NPK 50%; CA= citric acid at 150 ppm + NPK 50%; AA + CA= ascorbic + citric acids both at 150 ppm + NPK 50%; vern.= vernalization; n-vern.= non-vernalization

Regarding the effect of the interaction treatments between organic acids and vernalization it could be noticed that CA + vern. (T5) resulted in the highest values as recorded 216.00 and 239.67 cm in both seasons, respectively. This was followed by AA + CA + vern. (T7) as recorded 201.67 and 236.17 cm in both seasons, respectively.

Number of branches/plant:

Data in Table (2) cleared that all organic acid treatments had a significant effect on number of branches/plant in both seasons compared to control (untreated plants), CA increased the number of branches/plant to the highest values (9.36 and 10.33 in the two seasons, respectively).

Vernalization treatment, on the other hand significantly increased the number of branches/plant to 10.17 and 10.14 in both seasons, respectively when compared with non-vernalization treatment (6.62 and 8.67 in the first and second seasons, respectively).

Regarding the effect of the interaction treatments between organic acids and vernalization, it could be noticed that CA + vern. (T5) resulted in the highest number of branches/plant as recorded 11.55 and 12.00 branches/plant in the two seasons, respectively. This was followed with AA + CA + vern. (T7) which occupied the second position as recorded 10.11 and 10.22 branches/plant in both seasons, respectively.

Number of fruits/plant:

The obtained results in Table (2) demonstrated that all organic acid treatments had a significant effect compared to control (untreated plants) on number of fruits/plant in both seasons. In this regard AA + CA increased the number of fruits/plant to the highest values as recorded 52.50 and 84.31 fruits/plant in both seasons, respectively.

On the other hand, vernalization treatment significantly increased the number of fruits/plant up to 55.66 and 90.94 fruits/plant in both seasons, respectively when compared with non-vernalization treatment which recorded 40.01 and 65.81 fruits/plant in the first and second seasons, respectively.

Regarding the effect of the interaction treatments between organic acids and vernalization it could be noticed that CA + vern. (T5) resulted in the highest number of fruits/plant as recorded 60.33 and 90.45 in the first and second seasons, respectively. This was followed by AA + CA + vern. (T7) which recorded 56.84 and 89.78 fruits/plant in both seasons, respectively.

The above-mentioned results are supported by Li *et al.* (2010) on broccoli plants, Lang (1965) and Khattab and El Sherif (2016) on roselle, Darapuneni *et al.* (2014) and Azoz *et al*. (2016) on basil plants.

Fresh weight of fruits (g/plant):

As for the effect of organic acids on fresh weight of fruits (g/plant) the results in Table (2) show that all organic acid treatments had a significant effect compared to control (untreated plants) in both seasons. In this regard AA + CA resulted in the highest values (400.80 and 803.20 g/plant) in the first and second seasons, respectively.

Concerning the effect of vernalization treatment, a significant increase was observed by vernalization treatment on the fresh weight of fruits (431.80 and 834.90 g/plant in both seasons, respectively) when compared with non-vernalization treatment (290.30 and 668.20 g/plant in the first and second seasons, respectively).

Regarding the effect of the interaction treatments between organic acids and vernalization it could be noticed that CA + vern. (T5) resulted in the highest values for the fresh weight of fruits (g/plant) as recorded 501.00 and 885.40 g/plant in both seasons, respectively. This was followed by AA + CA + vern. (T7) as resulted in 467.30 and 859.60 g/plant in both seasons, respectively.

These results are supported by Li *et al.* (2010) on broccoli, Lang (1965) and Khattab and El Sherif (2016) on roselle, Jafari and Hadavi (2012 a) on dill, Jafari and Hadavi (2012 b) on basil plants, Darapuneni *et al.* (2014) and Azoz *et al*. (2016) on basil plants.

Number of days until flowering:

The findings in Table (3) revealed that all organic acid treatments had a significant effect compared to control (untreated plants) on number of days until flowering in both seasons. AA + CA reduced the number of days required for flowering to the lowest values (122.33 and 120.83 days in both seasons, respectively).

Vernalization treatment, on the other hand significantly reduced the number of days required for flowering to 118.83 and 119.00 days in both seasons, respectively when compared with non-vernalization treatment (128.83 and 127.83 in the first and second seasons, respectively).

As for the effect of the interaction treatments between organic acids and vernalization, it could be noticed that AA + CA + vern. (T7) resulted in the lowest number of days as recorded 117.33 and 116.33 days in the first and second seasons, respectively. Many other combined treatments shared the previously mentioned treatment in its effect without a significant effect between them i.e. AA + vern. (T3) and CA + vern. (T5). In the contrary, cont. + n-vern. (T2) prolonged the period required for flowering as recorded 130.33 in both seasons. These results are in accordance with Khattab and El Sherif (2016) on roselle, Darapuneni *et al.* (2014), Runkle *et al.* 1998, Suzuki and Metzger (2001).

Fresh weight of sepals (g/plant):

For the fresh weight of sepals (g/plant), the findings in Table (3) revealed that all organic acid treatments had a significant effect compared to control (untreated plants) on fresh weight of sepals (g/plant) in both seasons, AA + CA resulted in the highest values in this regard (121.20 and 332.80 g/plant in both seasons, respectively).

A pronounced effect was observed of vernalization treatment as increased fresh

Table 3. Effect of ascorbic, citric acids, and vernalization on number of days until flowering, fresh weight of sepals (g/plant), dry weight of sepals (g/plant) and dry weight of sepals (kg/fed) of roselle (*Hibiscus sabdariffa* L.) plants in the two seasons of 2019 and 2020.

Treatments	Number of days until flowering		Fresh weight of sepals (g/plant)		Dry weight of sepals (g/plant)		Dry weight of sepals (kg/fed)	
	2019	2020	2019	2020	2019	2020	2019	2020
				Organic acids				
Cont.	125.33	126.00	71.90	264.80	18.28	66.70	406.00	1483.00
AA	124.33	124.83	91.30	306.80	22.83	77.10	507.00	1714.00
CA	123.33	122.00	120.00	332.60	30.18	83.50	677.00	1860.00
AA + CA	122.33	120.83	121.20	332.80	30.47	83.70	671.00	1856.00
L.S.D. 5%	0.65	0.33	15.65	16.85	3.93	4.22	87.40	93.80
				Vernalization				
vern.	118.83	119.00	149.30	350.70	37.50	88.10	833.00	1958.00
n-vern.	128.83	127.83	52.90	267.80	13.37	67.40	297.00	1498.00
L.S.D. 5%	0.471	0.2718	12.77	25.59	3.197	6.40	71.00	142.20
				Organic acids × vernalization				
Cont. + vern. (T1)	120.33	121.67	110.90	320.70	27.96	80.70	621.00	1793.00
Cont. + n-vern. (T2)	130.33	130.33	32.90	208.90	8.60	52.80	191.00	1172.00
AA + vern. (T3)	119.33	120.33	146.40	343.60	36.73	86.30	816.00	1917.00
AA + n-vern. (T4)	129.33	129.33	36.20	269.90	8.92	68.00	198.00	1511.00
CA + vern. (T5)	118.33	117.67	187.50	384.40	47.05	96.60	1046.00	2147.00
CA + n-vern. (T6)	128.33	126.33	54.90	281.20	13.89	70.80	309.00	1574.00
AA + CA + vern. (T7)	117.33	116.33	152.40	353.90	38.27	88.90	851.00	1975.00
AA + CA + n-vern. (T8)	127.33	125.33	87.60	311.30	22.08	78.10	491.00	1737.00
L.S.D. 5%	0.84	0.46	21.69	37.78	5.439	9.45	120.90	210.00

Cont. = without organic acids + NPK 100%; AA= ascorbic acid at 150 ppm + NPK 50%; CA= citric acid at 150 ppm + NPK 50%; AA + CA= ascorbic + citric acids both at 150 ppm + NPK 50%; vern.= vernalization; n-vern.= non-vernalization

weight of sepals to 149.30 and 350.70 g/plant in both seasons, respectively, compared with non-vernalization which resulted in fewer values (52.90 and 267.80 g/plant in the first and second seasons, respectively).

As for the effect of the interaction treatments between organic acids and vernalization, it could be noticed that the combined treatment of CA + vern. (T5) resulted in the highest values for fresh weight of sepals as recorded 187.50 and 384.40 g/plant in the first and second seasons, respectively. AA + CA + vern. (T7) came in the second rank as recorded 152.40 and 353.90 g/plant in both seasons, respectively. These outcomes are supported by Youssef *et al.* (2014), Chen and Gallie (2006), Lang (1965) and Khattab and El Sherif (2016) on roselle and by Azoz *et al.* (2016) on basil.

Dry weight of sepals (g/plant):

Data illustrated in Table (3) revealed that all organic acid treatments had a significant effect compared to control (untreated plants) on dry weight of sepals in both seasons the mastery was to AA + CA which increased the dry weight of sepals to the highest values (30.47 and 83.70 g/plant in both seasons, respectively).

On the other hand, vernalization treatment was more effective than non-vernalization treatment as significantly increased the dry weight of sepals to 37.50 and 88.10 g/plant in both seasons, respectively. While, non-vernalization treatment recorded only 13.37 and 67.40 g/plant in the first and second seasons, respectively.

The combined treatment of CA + vern. (T5) resulted in the highest values for the dry weight of sepals as recorded 47.05 and 96.60

g/plant in both seasons, respectively, followed by AA + CA + vern. (T7) (38.27 and 88.90 g/plant in both seasons, respectively.

Dry weight of sepals (kg/fed):

For the effect of the dry weight of sepals, the presented data in Table (3) revealed that all organic acid treatments had a significant effect compared to control (untreated plants) in both seasons. CA increased the dry weight of sepals to the highest value as recorded 677.00 and 1860.00 kg/fed in the two seasons, respectively.

The effect of vernalization treatment, on the other hand significantly increased the dry weight of sepals (kg/fed) to 833.00 and 1958.00 kg/fed in both seasons, respectively when compared with non-vernalization treatment (297.00 and 1498.00 kg/fed in the first and second seasons, respectively).

Regarding the effect of the interaction treatments between organic acids and vernalization, it could be noticed that CA + vern. (T5) resulted in the highest values as recorded 1046.00 and 2147.00 kg/fed in both seasons, respectively. This was followed by AA + CA + vern. (T7) which recorded 851.00 and 1975.00 kg/fed in both seasons, respectively. These outcomes are supported by Youssef *et al.* (2014), Chen and Gallie (2006), Lang (1965) and Khattab and El Sherif (2016) on roselle and Azoz *et al.* (2016) on basil.

Weight of 100 seeds (g):

Data illustrated in Table (4) revealed that all organic acid treatments had a significant effect compared to control (untreated plants) on weight of 100 seeds (g) in both seasons. In this respect, CA increased the weight of 100 seeds (g) to the highest significant values (3.98 and 4.06 g in both seasons, respectively).

Vernalization treatment, on the other hand significantly increased the weight of 100 seeds (g) to 4.03 and 4.13 g in both seasons, respectively when compared with non-vernalization treatment (3.76 and 3.87 g in the first and second seasons, respectively).

Regarding the effect of the interaction treatments between organic acids and vernalization, it could be noticed that CA + vern. (T5) resulted in the highest values for the weight of 100 seeds as recorded 4.18 and 4.23 g in both seasons, respectively. The second position was obtained by AA + CA + vern. (T7) as resulted in 4.05 and 4.16 g in both seasons, respectively.

Weight of seeds (g/plant):

The findings in Table (4) demonstrated that all organic acid treatments had a significant effect compared to control (untreated plants) on the weight of seeds (g/plant) in both seasons, the superiority was to AA + CA which recorded the highest values (81.60 and 57.62 g/plant in both seasons, respectively).

Vernalization treatment, on the other hand significantly resulted in the highest values (89.40 and 60.95 g in the first and second seasons, respectively) when compared with non-vernalization treatment (54.30 and 38.66 g in the first and second seasons, respectively).

As for the effect of the interaction treatments between organic acids and vernalization treatment, it could be noticed that CA + vern. (T5) resulted in the highest values as recorded 106.00 and 74.77 g in both seasons, respectively, followed by AA + CA + vern. (T7) as resulted in 92.60 and 62.90 g in both seasons, respectively.

Weight of seeds (kg/fed):

All organic acid treatments had a significant effect on weight of seeds (kg/fed) compared to control (untreated plants) in both seasons (Table, 4). AA + CA increased the weight of seeds (kg/fed) to the highest value as recorded 1814.00 and 1814.00 Kg/fed in the first and second seasons, respectively.

Vernalization treatment, on the other hand significantly increased the weight of seeds (kg/fed) to 1986.00 and 1986.00 Kg/fed in both seasons, respectively when

Table 4. Effect of ascorbic, citric acids, and vernalization on weight of 100 seeds (g), weight of seeds (g/plant) and weight of Seeds (kg/fed) of roselle (*Hibiscus sabdariffa* L.) plants in the two seasons of 2019 and 2020.

Treatments	Weight of 100 seeds (g)		Weight of seeds (g/plant)		Weight of Seeds (kg/fed)	
	2019	2020	2019	2020	2019	2020
			Organic acids			
Cont.	3.79	3.91	60.50	40.55	1345.00	1345.00
AA	3.87	3.98	65.70	46.48	1460.00	1460.00
CA	3.98	4.06	79.50	54.57	1767.00	1767.00
AA + CA	3.94	4.05	81.60	57.62	1814.00	1814.00
L.S.D. 5%	0.10	0.13	12.78	2.57	283.90	283.90
			Vernalization			
vern.	4.03	4.13	89.40	60.95	1986.00	1986.00
n-vern.	3.76	3.87	54.30	38.66	1206.00	1206.00
L.S.D. 5%	0.11	0.11	13.05	1.73	289.90	289.90
			Organic acids × vernalization			
Cont. + vern. (T1)	3.92	4.04	76.10	48.15	1692.00	1692.00
Cont. + n-vern. (T2)	3.66	3.77	45.00	32.95	999.00	999.00
AA + vern. (T3)	3.98	4.09	82.80	57.99	1840.00	1840.00
AA + n-vern. (T4)	3.76	3.87	48.60	34.97	1080.00	1080.00
CA + vern. (T5)	4.18	4.23	106.00	74.77	2355.00	2355.00
CA + n-vern. (T6)	3.78	3.89	53.00	40.47	1178.00	1178.00
AA + CA + vern. (T7)	4.05	4.16	92.60	62.90	2058.00	2058.00
AA + CA + n-vern. (T8)	3.84	3.95	70.60	46.25	1569.00	1569.00
L.S.D. 5%	0.16	0.18	20.63	3.21	458.30	458.30

Cont. = without organic acids + NPK 100%; AA= ascorbic acid at 150 ppm + NPK 50%; CA= citric acid at 150 ppm + NPK 50%; AA + CA= ascorbic + citric acids both at 150 ppm + NPK 50%; vern.= vernalization; n-vern.= non-vernalization

compared with non-vernalization treatment (1206.00 and 1206.00 Kg/fed in the first and second seasons, respectively).

Regarding the effect of the interaction treatments between organic acids and vernalization it could be noticed that CA + vern. (T5) resulted in the highest weight of seeds (kg/fed) as recorded 2355.00 and 2355.00 kg/fed in both seasons, respectively, followed by AA + CA + vern. (T7) as resulted in 2058.00 and 2058.00 kg/fed in both seasons, respectively. These findings are propped up to Miri *et al.* (2015) on thyme (*Thymus vulgaris* L.) and Azoz *et al.* (2016) on basil.

Fixed oil (%):

The data in Table (5) showed that all organic acid treatments had a significant effect compared to control (untreated plants) on fixed oil % in both seasons, CA increased the fixed oil % to the highest values (20.60 and 21.57% in both seasons, respectively).

Vernalization treatment significantly increased the fixed oil % to 21.10 and 21.97% in both seasons, respectively when compared with non-vernalization treatment (19.11 and 20.07%) in the first and second seasons, respectively.

Also, the effect of the interaction treatments between organic acids and vernalization was significant. It could be noticed that AA + CA + vern. (T7) resulted in the highest values as recorded 22.32 and 23.27% in both seasons, respectively. This was followed without significant differences by cont. + n-vern. (T2) 22.31 and 22.30% in both seasons, respectively.

Total carbohydrates (%):

As for the total carbohydrates % in sepals, data in Table (5) showed that all organic acid treatments had a significant effect compared to control (untreated plants). AA + CA resulted in the highest values (8.57 and 9.12 in both seasons, respectively).

Vernalization treatment significantly increased the carbohydrates % in sepals to 8.40 and 8.95% in both seasons, respectively when compared with non-vernalization

Table 5. Effect of ascorbic, citric acids, and vernalization on fixed oil (%), total carbohydrates (%) anthocyanin content (µM) and vitamin C content (mg/100 g) of roselle (*Hibiscus sabdariffa* L.) plants in the two seasons of 2019 and 2020.

Treatments	Fixed oil (%)		Total carbohydrates (%)		Anthocyanin content (µM)		Vitamin C content (mg/100g)	
	2019	2020	2019	2020	2019	2020	2019	2020
				Organic acids				
Cont.	20.41	21.19	8.47	9.02	0.80	0.86	1.53	1.59
AA	19.92	20.87	8.15	8.70	0.77	0.82	1.55	1.52
CA	20.60	21.57	7.40	7.95	0.70	0.76	1.39	1.44
AA + CA	19.49	20.44	8.57	9.12	0.81	0.86	1.64	1.69
L.S.D. 5%	1.10	1.77	0.52	0.52	0.05	0.05	0.18	0.25
				Vernalization				
vern.	21.10	21.97	8.40	8.95	0.80	0.85	1.60	1.61
n-vern.	19.11	20.07	7.90	8.45	0.75	0.80	1.45	1.51
L.S.D. 5%	0.75	0.78	0.66	0.66	0.06	0.06	0.17	0.21
				Organic acids × vernalization				
Cont. + vern. (T1)	18.50	19.46	8.28	8.83	0.78	0.85	1.58	1.63
Cont. + n-vern. (T2)	22.31	22.30	8.66	9.21	0.82	0.87	1.49	1.55
AA + vern. (T3)	18.35	19.30	8.83	9.38	0.83	0.88	1.68	1.65
AA + n-vern. (T4)	21.49	21.66	7.47	8.02	0.71	0.77	1.40	1.46
CA + vern. (T5)	18.88	19.87	7.71	8.26	0.73	0.79	1.45	1.51
CA + n-vern. (T6)	20.71	22.44	7.10	7.64	0.68	0.73	1.32	1.38
AA + CA + vern. (T7)	22.32	23.27	8.77	9.32	0.83	0.88	1.69	1.73
AA + CA + n-vern. (T8)	18.27	19.22	8.37	8.92	0.79	0.85	1.59	1.58
L.S.D. 5%	2.10	1.91	0.99	0.99	0.09	0.09	0.27	0.35

Cont. = without organic acids + NPK 100%; AA= ascorbic acid at 150 ppm + NPK 50%; CA= citric acid at 150 ppm + NPK 50%; AA + CA= ascorbic + citric acids both at 150 ppm + NPK 50%; vern.= vernalization; n-vern.= non-vernalization

treatment (7.90 and 8.45% in the first and second seasons, respectively).

Regarding the effect of the interaction treatments between organic acids and vernalization, it could be noticed that AA + vern. (T3) resulted in the highest values as recorded 8.83 and 8.38% in both seasons, respectively, followed by AA + CA + vern. (T7) 8.77 and 9.32 in both seasons, respectively.

Anthocyanin content (µM):

Table (5) demonstrated that all organic acid treatments had a significant effect on anthocyanin content (µM) in sepals in both seasons compared to control (untreated plants). AA + CA resulted in the highest value as recorded 0.81 and 0.86 µM in both seasons, respectively.

Vernalization was more effective than non-vernalization treatment on anthocyanin content (µM) in sepals as resulted in 0.80 and 0.85 µM in both seasons, respectively. while, non-vernalization treatment recorded 0.75 and 0.80 µM in the first and second seasons, respectively.

Regarding the effect of the combined treatments between organic acids and vernalization, it could be noticed that AA + vern. (T3) and AA + CA + vern. (T7) resulted in the highest anthocyanin content in sepals as recorded the same values (0.83 and 0.88 µM in both seasons, respectively). Cont. + n-vern. (T2) non-significantly followed the previously mentioned treatments as recorded 0.82 and 0.87 µM in both seasons, respectively.

Vitamin C content (mg/100 g):

The findings in Table (5) showed that all organic acid treatments had a significant effect on vitamin C mg/100 g in sepals in both seasons compared to control (untreated plants). AA + CA increased the vitamin C mg/100 g in sepals to the highest value (1.64 and 1.69%) in both seasons, respectively.

Vernalization treatment, on the other hand significantly increased the vitamin C mg/100 g in sepals to 1.60 and 1.69% in both seasons, respectively when compared with non-vernalization treatment (1.45 and 1.51%) in the first and second seasons, respectively.

Regarding the effect of the interaction treatments between organic acids and vernalization, it could be noticed that AA + CA + vern. (T7) resulted in the highest values as recorded 1.69 and 1.73% in both seasons, respectively, followed by AA + vern. (T3) as resulted in 1.68 and 1.65% in both seasons, respectively. Azoz *et al*. (2016) on basil plants.

Fixed oil organic acids:

The findings of Table (6) and Fig. (1) showed that AA + vern. (T3) resulted in the highest values for oleic and linolenic acids (7.07 and 10.22%, respectively), followed by CA + vern. (T5) as recorded 6.85 and 8.58%, respectively, while the lowest values for oleic and linolenic acids were obtained by CA + n-vern. (T6) as recorded 3.51 and 6.58%, respectively. It could be also noticed that, CA + vern. (T5) resulted in the highest values in terms of stearic and linoleic acids (13.09 and 62.34%, respectively), followed by AA + vern. (T3) as recorded 10.67 and 54.99%, respectively, while the lowest values for stearic and linoleic acids were obtained by CA + n-vern. (T6) as recorded 5.06 and 1.96%, respectively. In this respect, many investigators reported that ascorbic acid application resulted in an enhancement of growth, yield and chemical constituents of some different plant species, especially field crops. Among them, Abdel-Aziz (1999) on lentil plants, Nassar *et al.* (2016) on flax plant (*Linumus itatissimum* L.) and Nassar (2013) on mung bean and Azoz *et al*. (2016) on basil plants.

DISCUSSION

The results of the present study showed the superior effect of spraying with citric acid on plant height (cm), number of branches/plant, dry weight of sepals/(kg/fed), weight of 100 seeds (g), fixed oil %, and this may be explained by the fact that citric acid has a central role in CA-cycle inside mitochondria that causes cellular energy by phosphorylative oxidation reactions, it's generated by the addition of acetyl-CoA to oxaloacetic acid that's converted to succinate (Eidyan, 2014). These findings agreed with later studies which revealed that the CA effect isn't just due to pH change only, but there is a spread of physiological responses to CA application, and this corresponds with what they reached (Parikshit *et al.*, 2017 and Jafari and Hadavi, 2012 a on dill plants). Regarding the positive roles of ascorbic acid, Price (1966) reported that vitamin C

Table 6. Effect of ascorbic, citric acids and vernalization on fixed oil organic acids of roselle (*Hibiscus sabdariffa* L.) seeds in the two seasons of 2019 and 2020.

Treatments	Organic acids	Percentage (%)
AA + vern. (T3)	1-Oleic acid	7.07
	2-Stearic acid	10.67
	3-Linoleic acid	54.99
	4-Linolenic acid	10.22
CA + vern. (T5)	1-Oleic acid	6.85
	2-Stearic acid	13.09
	3-Linoleic acid	62.34
	4-Linolenic acid	8.58
CA + n-vern. (T6)	1-Oleic acid	3.51
	2-Stearic acid	5.06
	3-Linoleic acid	1.96
	4-Linolenic acid	6.58

AA= ascorbic acid at 150 ppm + NPK 50%; CA= citric acid at 150 ppm + NPK 50%; vern. = vernalization; n-vern.= non-vernalization

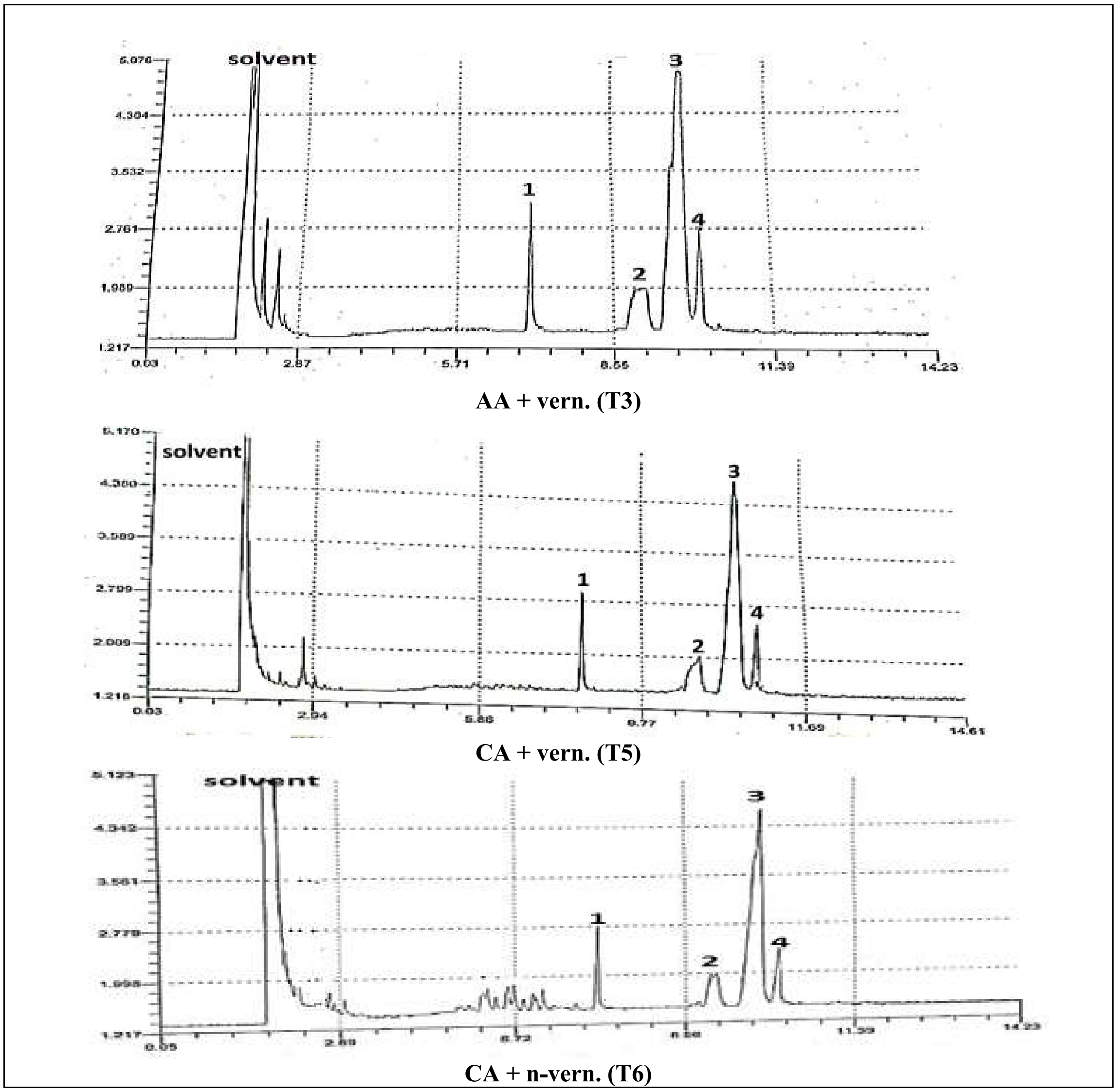

Fig. 1. Effect of ascorbic, citric acids and vernalization on fixed oil organic acids of roselle (*Hibiscus sabdariffa* L.) seeds in the two seasons of 2019 and 2020.
(1) oleic acid, (2) stearic acid, (3) linoleic acid, (4) linolenic acid, (AA) ascorbic acid at 150 ppm + NPK 50%, (CA) citric acid at 150 ppm + NPK 50%; (vern.) vernalization and (n-vern.) non-vernalization.

(ascorbic acid) increased nucleic acids content, especially RNA. It also influences the synthesis of enzymes and proteins and acts as co-enzyme in metabolic changes (Patil and Lall, 1973). Barth *et al.* (2006) reported the roles of ascorbic acid (AA) in controlling both flowering and senescence. Additionally, the redox status of (AA) may play a role in signaling in this interconnected phytohormone network. Mohsen and Ismail (2016) emphasized the positive role of foliar spray with ascorbic acid even under stress conditions, as it showed a positive influence on vegetative growth, flowering growth, chemical analysis of marigold (*Calendulaofficinalis* L.) and soil chemical properties.

As for the impacts of vernalization on growth and yield of plants. Vernalization treatment recorded the heights significant results in all studied characteristics of roselle because vernalization is the coldness treatment given to water-soaked seeds to hasten the time of flowering of plants which will develop from them. The increasing of vernalization duration also enhanced the plant height, other growth traits and yield components (number of seeds/plant, number of branches/plant and seed yield/plant). Furthermore, prolonged chilling provides competence to flower, other endogenous and/or environmental conditions, such as inductive photoperiods, are also required for flowering. A classic experiment illustrates that this competence to flower can be stable through mitotic cell divisions (Lang, 1965). These data side with those obtained by Zhang *et al*. (2014) on European turnips and Khattab and El Sherif (2016) on roselle. On the other hand, vernalization accelerates the transition to reproductive growth in most plants. These outcomes are supported by Li *et al.* (2010) on broccoli, Lang (1965), Chen and Gallie (2006), Youssef *et al.* (2014), Khattab and El Sherif (2016) on roselle plants, Darapuneni *et al.* (2014) on flax (*Linumus itatissimum* L.) and Azoz *et al*. (2016) on basil plants.

Regarding the interaction between the two applied factors employed in this study, the results showed that ascorbic (AA) and citric (CA) acids foliar spraying could promote vernalization treatment, and advance flower-bud differentiation stage with rapid climb and initial harvesting period. The mastery was to the treatments of CA + vern. (T5) and AA + CA + vern. (T7) which recorded the best results in the most studied traits. This may be explained by the different roles of vernalization, Ascorbic and citric acids as discussed previously. In addition, the improvement of vegetative growth is reflected directly in raising the plant production of secondary compounds as reported by Allen and Pilpeam (2006).

CONCLUSION

According to the results obtained from this study, vernalization treatment at 13 ℃ for 2 weeks of roselle seeds before sowing in addition to spraying with citric acid at 150 ppm only or with ascorbic acid at 150 ppm is recommended to contribute plant growth, enhance yield (total roselle dry sepals' weight per feddan) and improve the biochemical constituents of *Hibiscus sabdariffa* plant.

REFERENCES

Abadi, B.H.M.; Ganjali, H.R. and Mobasser, H.R. (2015). Effect of mycorrhiza and phosphorus fertilizer on some characteristics of black cumin. Biological Forum, 7(1):1115-1120.

Abdel-Aziz, E.A. (1999). Response of two lentil varieties to foliar spray with micronutrients mixture and ascorbic acid. J. Agric. Sci., Mansoura Univ., 24(10):5575-5585.

Ali, B.H.; Al Wabel, N. and Blunden, G. (2005). Phytochemical, pharmacological and toxicological aspects of *Hibiscus sabdariffa* L., a review. Phytotherapy Research, 19(5):369-375.

Allen, V. B. and Pilpeam, D.J. (2006). Handbook of Plant Nutrition. Taylor and Francis Group, NewYork, USA, 662 p.

Analytical Software (2008). Statistix Version 9, Analytical Software. Tallahassee, Florida, USA.

A.O.A.C. (1990). Official Methods of Analysis of the Association of Official Analytical Chemists, 15th Ed. Washington D.C., USA. 1298 p.

Azoz, S.N.; El-Taher, A.M.; Boghdady, M.S. and Nassar, D.M. (2016). The impact of foliar spray with ascorbic acid on growth, productivity, anatomical structure and biochemical constituents of volatile and fixed oils of basil plant (*Ocimum basilicum* L.). Middle East Journal of Agriculture Research, 5(4):549-565.

Bako, I.G.; Abubakar, M. S.; Mabrouk, M. A. and Mohammed, A. (2014). Lactogenic study of the effect of ethyl-acetate fraction of *Hibiscus sabdariffa* Linn (Malvaceae) seed on serum prolactin level in lactating albino rats. Advance Journal of Food Science and

Technology, 6(3):292-296.

Barth, C.; De Tullio, M. and Conklin, P.L. (2006). The role of ascorbic acid in the control of flowering time and the onset of senescence. Journal of Experimental Botany, 57(8):1657-1665.

Chen, Z. and Gallie, D.R. (2006). Dehydro ascorbate reductase affects leaf growth development and function. Plant Physiol., 142(2):775-787.

Chouard, P. (1960). Vernalization and its relations to dormancy. Annual Review of Plant Physiology, 11(1):191-238.

Cisse, M.; Dornier, M.; Sakho, M.; Ndiaye, A.; Reynes, M. and Sock, O. (2009). Le bissap (*Hibiscus sabdariffa* L.): composition et principals utilisations. Fruits, 64(3):179-193.

Clough, E.A.; Cameron, A.C.; Heins, R.D. and Carlson, W.H. (2001). Growth and development of *Oenothera fruticosa* is influenced by vernalization duration, photoperiod, forcing temperature, and plant growth regulators. Journal of the American Society for Horticultural Science, 126(3):269-274.

Da-Costa, R.I.; Bonnlaender, B.; Sievers, H.; Pischel, I. and Heinrich, M. (2014). *Hibiscus sabdariffa* L. A- phytochemical and pharmacological Review. Food Chemistry, 165:424-443.

Darapuneni, M.K.; Morgan, G.D.; Ibrahim, A.M. and Duncan, R.W. (2014).Effect of vernalization and photoperiod on flax flowering time. Euphytica, 195 (2):279-285.

Dennis, E.S. and Peacock, W.J. (2007). Epigenetic regulation of flowering. Current Opinion in Plant Biology, 10(5):520-527.

Du, C.T. and Francis, F.J. (1973). Anthocyanin of roselle. J. Food Sci., 38:810-820.

Egyptian Pharmacopoeia (2005). The Egyptian Pharmacopoeia, 4th Ed. Central Administration of Pharmaceutical Affairs (CAPA), Ministry of Health and Population, Cairo, Egypt. 1295 p.

Eidyan, B.; Ebrahim, H. and Noorollah, M. (2014). Pre-harvest foliar application of iron sulfate and citric acid combined with urea fertigation affects growth and vase life of tuberose (*Polianthes tuberosa* L.) 'Por-Par'. Hort. Environ. Biotechnol., 55(1):9-13.

F.A.O. (2004). Hibiscus: Post-Production Management for Improved Market Access. Food and Agriculture Organization of the United Nations, 19 p. http://www.fao.org/3/a-av006e.pdf

Fasoyiro, S.B.; Adeola, O.A.A. and Samuel, F.O. (2005). Chemical and storability of fruit flavored (*Hibiscus sabdariffa* L.) drinks. World J. Agric. Sci., 1:165-168.

Galaudu, M.S. (2006). Effect of moisture on the germination rate of roselle (*Hibiscus sabdariffa* L.) plant. Proc. the 23rd Annual Conference of the Horticultural Society of Nigeria, Port Harcourt, Nigeria, pp. 30-32.

Gheeth, R.H.M.; Moustafa, Y.M.M. and Abdel-Hakeem, W.M. (2013). Enhancing growth and increasing yield of peas (*Pissum sativum* L.) by application of ascorbic acid and cobalt chloride. J. Nov. Appl. Sci., 2:106-115.

Haggag, L.F.; Fawzi, M.I.F.; Shahin, M.F.M. and El-Hady, E.S. (2016). Effect of yeast, humic acid, fulvic acid, citric acid, potassium citrate and some chelated micro-elements on yield, fruit quality and leaf minerals content of 'Canino' apricot trees. International Journal of ChemTech. Research, 9(4):7-15.

Hajreza, M.R.; Hadavi, E.; Zeynanlou, A.A.; Mirzapour, M.H. and Naeini, M.R. (2013). Effect of different levels of citric acid and salicylic acid at pre-harvesting stage on vase-life of rose (*Rosa hybrida* L.) cut flower. J. Sci. Tech. Green. Cult., 4:99-109.

Hopkins, A.L.; Lamm, M.G.; Funk, J.L. and Ritenbaugh, C. (2013). *Hibiscus sabdariffa*

L. in the treatment of hypertension and hyperlipidemia: A comprehensive review of animal and human studies. Fitoterapia, 85:84-94.

Hussin, M.S.; El-Sherbeny, S.E.; El-Saeid, H.M. and Kandeel, M.M. (1991).Field experiments of foliar application with B-9 and micronutrients on *Hibiscus sabdariffa* L., 1. Growth yield and hormonal content. Egypt. J. Hort., 16(1):59- 68.

Inskbashi, Y. and Iwaya, M. (2006). Ascorbic acid suppresses germination and dynamic states of water in wheat seeds. Plant Production Sci., 9(2):172-175.

Ismail, A.; Ikram, E.H.K. and Nazri, H. S.M. (2008). Roselle (*Hibiscus sabdariffa* L.) seeds nutritional composition protein quality and health benefits. Food, 2(1):1-16.

Jafari, N. and Hadavi, E. (2012 a). Growth and essential oil yield of dill (*Anethum graveolens* L.) as affected by foliar sprays of citric acid and malic acid. Acta Hort., 955:287-290.

Jafari, N. and Hadavi, E. (2012 b). Growth and essential oil yield of basil (*Ocimum basilicum* L.) as affected by foliar sprays of citric and salicylic acid. Zeitschrift fur Arznei-und Gewurzpflanzen, 17:80-83.

Khattab, S. and El Sherif, F. (2016). Effect of seed vernalization temperature, duration and planting date on growth and yield of *Hibiscus sabdariffa* plants. Life Science Journal, 13(6):52-60.

Lang, A. (1965). Physiology of flower initiation. In: Lang, A. (ed), Differentiation and Development, Encyclopedia of Plant Physiology, vol. 15, Springer, Berlin, Heidelberg, Germany, pp. 1380-1536.

Li, D.; Yu, X.; Jiang, X.; Wang, L.I. and Wang, F. (2010). Effects of 5-azaC on vernalization and flower-ball development in broccoli (*Brassica oleracea* var. *italica*). Pak. J. Bot., 42(3):1889-1893.

Louis, S.J.; Kadams, A.M.; Simon, S.Y. and Mohammed, S.G. (2013). Combining ability in roselle cultivars for agronomic traits in Yola, Nigeria. Greener Journal of Agricultural Sciences, 3(2):145-149.

Mahadevan, N., Shivali and Kamboj, P. (2009). *Hibiscus sabdariffa* L., an overview. Natural Product Radiance, 8(1):77-83.

Mandour, M.A.; Metwaly, H.A. and Ali, A.M. (2019). Effect of foliar spray with amino acids, citric acid, some calcium compounds and mono-potassium phosphate on productivity, storability and controlling grey mould of strawberry fruits under sandy soil conditions. Zagazig Journal of Agricultural Research, 46(4):985-997.

Miri, S.M.; Ahmadi, S. and Moradi, P. (2015). Influence of salicylic acid and citric acid on the growth, biochemical characteristics and essential oil content of thyme (*Thymus vulgaris* L.). Journal of Medicinal Plants and Byproducts, 2:141-146.

Mohsen, M.A. and Ismail, H. (2016). Response of *Calendula officinalis* L. which grown in saline soil to plant growth promoters and some organic substances. International Journal of Pharma Technology Research, 9(4):153-172.

Nassar, R.M. (2013). Response of mungbean plant *Vigna radiata* L. Wilczek to foliar spray with ascorbic acid. Journal of Applied Sciences Research, 9(4):2731-2742.

Nassar, R.M.; Arafa, A.S. and Madkour, M.A. (2016). Pivotal role of ascorbic acid in promoting growth, increasing productivity and improving quality of flax Plant *Linum usitatissimum* L. Middle East Journal of Agriculture, 5(2):216-223.

Parikshit, S.; Jatav, A.L.; Poonam, S.; Abhishekh, S.; Sagar, K.S. and Udai, S.C. (2017). Effect of vernalization and fungicidal seed treatment on yield and quality of wheat *Triticum aestivum* L. Int. J. Curr. Microbiol. App. Sci., 6 (9):20-26.

Patil, B.N. and Lall, S.B. (1973). Effect of pre-sowing treatment with L-ascorbic acid and gibberellic acid on growth and physiological constituents of wheat. Botanique (Nagpur), 4:57-70.

Price, C.E. (1966). Ascorbic stimulation of RNA synthesis. Nature, 212:1481.

Runkle, E.S.; Heins, R.D.; Cameron, A.C.; and Carlson, W.H. (1998). Flowering of *Leucanthemum* × *superbum* 'Snowcap' in response to photoperiod and cold treatment. HortScience, 33:1003-1006.

Shamkhi, K.J.; Saad, T.M. and Aatashan, L.A. (2012). The effect of nitrogen and phosphorus levels on some yield components and the specific qualities of roselle (*Hibiscus sabdariffa* L.) plant tea. Al-Muthanna Journal of Agricultural Sciences, 1(1):16-26.

Singh, P.; Khan, M. and Hailemariam, H. (2017). Nutritional and health importance of *Hibiscus sabdariffa* L., a review and indication for research needs. J. Nutr. Health Food Eng., 6(5):2-12.

Snedecor, W.G. and Cochran, G.W. (1980). Statistical Methods, 7th Ed. Iowa State Univ. Press, Ames, Iowa, USA., 507 p.

Suntornsuk L.; Gritsanapun W.; Nilkamhank S. and Paochom A. (2002). Quantitation of vitamin C content in herbal juice using direct titration. J. Pharm. Biomed. Anal., 8(5):49-55.

Suzuki, A. and Metzger, J.D. (2001). Vernalization in a greenhouse promotes and synchronizes flowering of *Osteospermum ecklonis* Norl. HortScience, 36(4):658-660.

Tomes, D.T. (1990). Current research in biotechnology with application to plant breeding. In: Nijkamp, H.J.J.; Van Der Plas, L.H.W. and Van Aartrijk, J. (eds), Progress in Plant Cellular and Molecular Biology, Current Plant Science and Biotechnology in Agriculture, vol. 9, Springer, Dordrecht, Netherlands, pp. 23-32.

Wahabi, H.A.; Alansary, L.A.; Al-Sabban, A.H. and Glasziuo, P. (2010). The effectiveness of *Hibiscus sabdariffa* L. in the treatment of hypertension: A systematic review. Phytomedicine, 17(2):83-86.

Youssef, A.S.M.; Mady, M.A. and Ali, M.M. (2014). Partial substitution of chemical fertilization of roselle plant (*Hibiscus sabdariffa* L.) by organic fertilization in presence of ascorbic acid. Journal of Plant Production, Mansoura Univ., 5(3):475-503.

Zhang, J.; Dell, B.; Biddulph, B.; Khan, N.; Xu, Y.; Luo, H. and Appels, R. (2014). Vernalization gene combination to maximize grain yield in bread wheat (*Triticum aestivum* L.) in diverse environments. Euphytica, 198(3):439-454.

Zhang, N.; Zhao, J.; Lens, F., de Visser, J.; Menamo, T.; Fang, W. and Bonnema, G. (2014). Morphology, carbohydrate composition and vernalization response in a genetically diverse collection of Asian and European turnips (*Brassica rapa subsp. rapa*). Plos One, 9(12):1-29. doi:10.1371/journal.pone.0114241

تأثير الرش بالأحماض العضوية والإرتباع على النمو والإنتاج والمحتوى الكيميائي لنبات الكركديه

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أجريت الدراسة الحالية في مزرعة مركزالبحوث الزراعية بمحطة البحوث الزراعية بالجميزة بمحافظة الغربية بمصر خلال موسمي الصيف المتتاليين ٢٠١٩ و٢٠٢٠، بهدف تقييم تأثير معاملات الإرتباع و الرش ببعض الأحماض العضوية (حمض الستريك وحمض الاسكوربيك) على نباتات الكركديه. كانت معاملات التداخل على النحو التالي: الكنترول (الرش بالماء) + الإرتباع (المعاملة ١)، الكنترول + عدم الإرتباع (المعاملة ٢)، حمض الأسكوربيك + الإرتباع (المعاملة ٣)، حمض الأسكوربيك + عدم الإرتباع (المعاملة ٤)، حمض الستريك + الإرتباع (المعاملة ٥)، حمض الستريك + عدم الإرتباع (المعاملة ٦)، حمض الأسكوربيك + حمض الستريك + الإرتباع (المعاملة ٧)، و حمض الأسكوربيك + حمض الستريك + عدم الإرتباع (المعاملة ٨). وجد أن تأثير الإرتباع مع الأحماض العضوية علي نباتات الكركديه أعطت أعلى نتائج معنوية مقارنة مع الكنترول على جميع قياسات النمو الخضري المختبرة والقياسات الكيميائية حيث كان الرش بحمض الستريك أفضل المعاملات في معظم قياسات النمو الخضري مثل ارتفاع النبات (سم)، وزن ١٠٠ بذرة (جم) ونسبة الزيت الثابت. وكانت معاملة الإرتباع أفضل من معاملة عدم الإرتباع. بينما كانت أفضل معاملات التفاعل على جميع قياسات النمو الخضري والزيت الثابت والأحماض العضوية مثل حمض الستريك وحمض اللينوليك هي معاملتي ٥ و٧. ولذلك فإنه يمكن التوصية بتطبيق المعاملة ٧ (حمض الأسكوربيك + حمض الستريك + الإرتباع) للحصول على أفضل النتائج في معظم القياسات.