Effect of seed vernalization temperature, duration and planting date on growth and yield of *Hibiscus sabdariffa* plants.

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Abstract: An experiment was conducted at the Experimental Station, Faculty of Agriculture, Suez Canal University, Ismailia, Egypt. The present study was aimed at determining the effects of seed vernalization temperature (room temperature, 6 and 12^{0}C), duration (one, two and three weeks) and three planting date (April, May, and June) on the growth behavior, yield and composition of calyx extract of Roselle (*Hibiscus sabdariffa* L.) variety "Sabahia 17". The results showed that early (April 13th) or late (June 13th) planting date causes a decrease in the growth and yield. Pre-sowing cold treatment of seeds increased the plant yield. The highest calyx yield per plant was observed from the pre-sowing cold treatment of seeds at 12 ^{0}C for one week. Vernalization of seeds at 12°C for one week and planting at 13th May is the most suitable practice for growing rosella in Ismalia, Egypt as it resulted in higher number of leaves, more fruits/plant and improved total calyx yield as compared to other treatment parameters.
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1. Introduction

Crop production is inherently sensitive to climate variability. Unfavorable environmental conditions can produce lower growth and yields of the crops. Planting date affects the day and night temperature, day light intensity and photoperiods which affected the flower inductions and yield production (Naeve et al. 2004). Late planting date often affects the crop vegetative growth and reproduction (Board et al. 1992, 1999; Boquet, 1990; Kantolic and Slafer, 2001). Plant activities such as growth rate, protoplasmic streaming, enzymes activities, or some other vital processes, are extremely sensitive to temperature (Noggle and Fritz, 1982). Vernalization accelerates the transition to reproductive growth in many plant species (Dennis and Peacock, 2007; Jane et al., 2008; Leopold and Kriedemann, 1983**)**. Rosella (*Hibiscus sabdariffa* L.) is an annual herbaceous shrub belonging to the family Malvaceae (Mccaleb, 1998). Roselle is widely grow throughout many tropical areas and native to tropical Central and West Africa (Tindall, 1983). Throughout the growing and fruiting periods, roselle plant is tolerant to relatively high temperatures (Tomes, 1990). The plant has an overall growing period of 4-6 months from planting of the seeds to harvesting of the calyx. Roselle is cultivated for its fleshy fruits, leaves, stems, flowers (calyces), seeds and fiber (Galaudu, 2006). The roselle calyx has high content of organic acids and anthocyanin (Cissé et al. 2009; Gomez-Leyva et al. 2008), and has been used as drinks, flavour and colour additives in food industries, as well as ethno medicine for hypertension, pyrexia liver disorders, anti-microorganism growth, diuretic, sedative, and digestive disorder (Akindahunsi and Olaleye, 2003; Da-Costa-Rocha et al. 2014). Egypt is considered as the country in which roselle originated (El–Sayed et al. 1998; FAO, 2004; Leung and Foster, 1996; Ismail et al. 2008). *H. sabdariffa* is grown in various parts of Egypt, for example Ismailia city. It is usually planted annually from May (El Sherif et al. 2011). Optimization of roselle planting date is an excellent approach to increase both crop yield and economic benefit (Naeve et al. 2004). Many research groups have studied the effect of planting date on growth, yield and anthocyanin content of roselle (Asante and Amankwa, 1992; Nilmar et al. 2004**;** Seghatoleslami et al. 2013). However, to our knowledge, this is the first investigation to study the effects of pre-sowing cold treatment (vary temperature and duration) of seeds and planting date on the growth, flowering and yield of rosella plant in the Ismailia city, Egypt.

2. Materials and methods

2.1 Plant material and experimental design

The experiment was conducted during 2014 and 2015 seasons, at the Faculty of Agriculture Experimental Station, Suez Canal University, Ismailia, Egypt. Roselle (*Hibiscus sabdariffa* L.) variety "Sabahia 17" was used in this experiment. Seeds of roselle were obtained from the Medicinal and Aromatic plants Department, Horticulture Research

Institute, Agriculture Research Center (ARC), Giza, Egypt. Dry seeds were exposed to low temperatures (6^0C or 12^0C) in the incubator for three duration treatments (one, two and three weeks) in addition to untreated seeds (control), which are stored at room temperature (about 25^0C). Seeds were sown in sandy soil at three different planting dates (April 13th; May 13th and June 13th) for both seasons. Each treatment was planted in 6 rows, in a plot of 4 m long and 0.6 m wide, making an area of 14.4 m^2. Hills were 50 cm apart, with 4 seeds per hill and then thinned three weeks later to one plant/ hill. Other agricultural practices such as, irrigation and weeding were carried out as recommended. The chemical contains of the irrigation water is tabulated in Table (1).

Table (1) Chemical content of the irrigation water

Salinity Level ppm	Cations				Anions				EC ppm	SAR
	Ca^{+2}	Mg^{+2}	Na^{+}	K^{+}	CO_3^{-2}	HCO_3^{-}	SO_4^{-2}	Cl^{-}		
(300)	1.14	1.44	1.78	0.29	n.d	2.66	0.46	1.42	300	1.7

2.2 Measurement of Vegetative growth parameters

In October 13th, before harvesting, the growth parameters such as plant height (cm), number of branches/ plant, stem diameter (cm), main root length/ plant (cm), root numbers /plant, and leaves, stems and roots dry weight / plant (g) were measured using eight random plants from each treatment.

2.3 Measurement of yield

Plants were harvested when practically there were no developing calyxes, almost at the end of the plant cycle, which is in the middle of December in both seasons; the fruits number / plant and calyx dry weight / plant (g) were recorded.

2.4 Chemical analysis of leaves

Plant leaf samples harvested in October 13th were dried at 70 ºC for 24 h; the dry matter was ground and digested according to Piper (1947). Nitrogen determination was determined using the modified micro- Kjedahl method, as described by Jackson (1967). The percentage of phosphorus was estimated calorimetrically as according to the method of Murphy and Reily (1962). The Potassium (K) content was determined by using Atomic Absorption /Flame spectrophotometer (3300) as according to Wilde et al. (1985).

2.5 Anthocyanin determination in calyxes

Total anthocyanin in air dried harvested rosella calyx harvested in the middle of December was determined according to the method described by Fuleki and Francis (1968) and modified by Du and Francis (1973).

2.6 Statistical analysis

The experiment design was split split plot with 20 replicates. Planting date represented the main plots, while pre-sowing cold treatment of seeds were considered as sub-plots and duration of cold treatments were considered as sub-sub plots. Data were statistically analyzed using ANOVA\MANOVA of Statistica 6 software, StatSoft company (Statsoft, 2001), the significance of differences among means was carried out using the Least Significant Test (L.S.D) at $p = 0.05$.

3. Results and Discussions

Early germination was observed in seeds, which were vernalized before planting as compared to non-vernalized seeds, regardless of the planting date. It was also observed that the longer was the vernalization treatment, the earlier was plant germination, and the germination rate was 100% in all vernalization treatments (data not shown). Data cited in Table (2, 3, 4 and 5) reveal significant differences due to the three factors of treatments, i.e. planting date, pre-sowing cold treatment of seeds and durations as interaction significantly affected the growth of roselle plant.

3.1 Plant growth

Data in Table (2 a and 3a) showed that the second planting date (13th May) significantly increased the stem diameter (cm), number of branches, number of roots, root length and dry weight of leaves, stem and root (g) of roselle plants in both season. The plant height significantly decreased with delay in planting date in both seasons (Table 2 a). The first planting date (13th April) increased the plant height. Prolonged growth period allowed the plants to perfectly use nutrients, water and radiation which improved the photosynthesis and the growth of the plants (Seghatoleslami et al. 2013). A reduction in plant growth by delay planting date had already been reported by (Castro et al. 2004; Rahman et al. 2014; Seghatoleslami et al. 2013; Silva 1982).

Data cited in tables (2a and 3a) showed that rosella plants produced from seeds vernalized at 12 ^{o} C significantly increased the plant height, number of branches and dry weight of leaves, stem and root of rosella plant in both seasons. While seeds having no vernalization treatment produced maximum root number, root length and stem diameter. Similar results were also reported by Esmat et al. (2013).

Tables (2d and 3d) showed significant interaction effect between the cultivation periods x vernalization

duration. Roselle plants performed differently depending on the cultivation period and the vernalization duration. Roselle plants presented highest number of branches; stem diameter and dry weight of leaves, stem and root in plants planted on 13th May from seeds vernalized at 12 ° C for one week (Table 2d and 3d). Our results indicated that cultivation period, vernalization period and vernalization duration affect the plant growth, which is in agreement with that reported by Pereira (2000) and Resende et al.(2004).

3.2 Mineral element contents

Result in table (4) showed that higher N, P and K contents were observed in plant cultured on the second planting date 13th May in both season, while the lowest N, P and K contents were observed in plant produced in the third planting date (13th June) in 2014 and 2015 respectively. Seeds cold pretreatment at 12°C increased N and P content in the roselle leaves compared to control in both seasons. Seeds cold pretreatment at 12°C increased K content in the rosella leaves in the first season, while in the second season, highest K content was observed in control treatments (Table 4b). The highest N, P and K content were found in plants produced from seeds vernalized at 12°C for one week and planted at 13th May in both (Table 4d).

3.3 The calyx yield

Data in Table (5a) indicated that maximum number of fruits (64.93 and 68.53 fruit /plant) and calyx dry weight (22.42 and 21.4 g) were recorded in the second plant date 13th May in the first and second season respectively and the lowest fruit number per plant (60.28 and 59.03) and calyx dry weight (17.67 and 18.44 g) were recorded in the third planting date (13th June) in 2014 and 2015 respectively. Yield of calyx was highest in the plants planted in 13th May (298.91 and 285.28 kg /fed) followed by those planted in 13th April (276.17 and 264.92 kg /fed) and then those planted in June (235.6 and 245.84 kg /fed). Lower yields with later plantings were presumably due to the short growing seasons. Fruit weight increased as plant dry weight increased, indicating that the dependence of fruit production on vegetative growth in this environment. Okosun et al. (2006) indicated that calyx harvest index may have an inverse relationship with dry matter in roselle. Same results were recorded with the late planting by Ahmed et al. (2010) and Ngalamu et al. (2012) in their experiments.

The results of the effects of seeds cold pretreatment on the fruit number and calyx dry weight of roselle plant were presented in Table (5b), pre-sowing cold treatment of seeds at 12°C significantly increased the fruit numbers and calyx dry weight as compared to control treatment (without seed cold treatment). It was apparent that maximum number of fruit and calyx dry weight (69.56 and 67.03 g) and/or (22.02 and 21.97 g) were recorded in the plant germinated from the12°C cold treated seeds in the first and second season respectively. On the other hand, the lowest numbers of fruit (56.48 and 56.76) were recorded in the plant raised from the 6°C treated seeds.

Data in Table (5d) showed that rosella plants produced from seeds vernalized at 12°C for one week and planted in the 13th May gave the maximum number of fruit and calyx dry weight in both season. The lowest number of fruit and calyx dry weight were produced from plants produced from seeds vernalized at 6°C for one week and planted on the 13th April.

3.4 Anthocyanin content

The concentrations of anthocyanin are given in figure (1 and 2). Results indicated that anthocyanin content of rosella calyxes were increased by the second plant time 13th May and / or when the seeds of the plant treated with 12°C in the both seasons (figure 1). The highest anthocyanin content was found in plants produced from seeds vernalized at 12°C for one week and planted at 13th June and the lowest anthocyanin content was found in plants produced from seeds vernalized at 6°C for one week and cultured at 13th April in both seasons (figure 2).

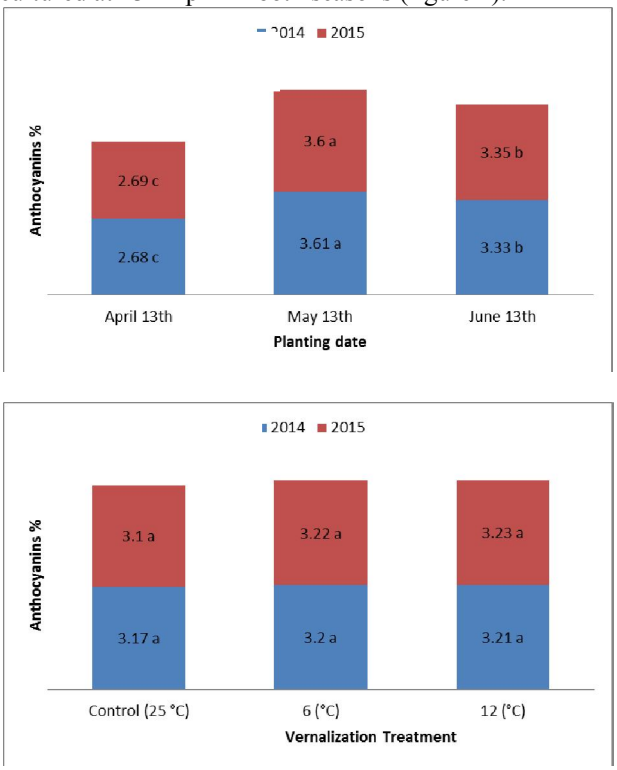
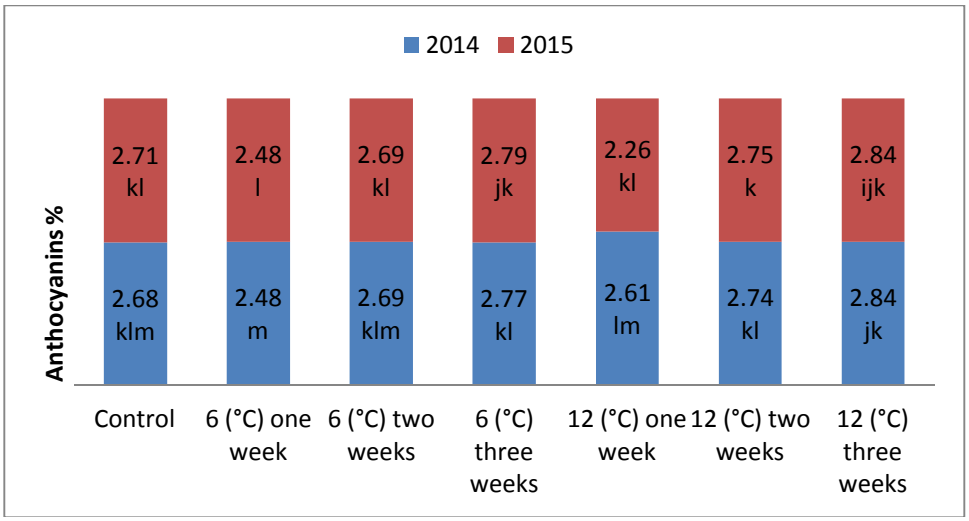
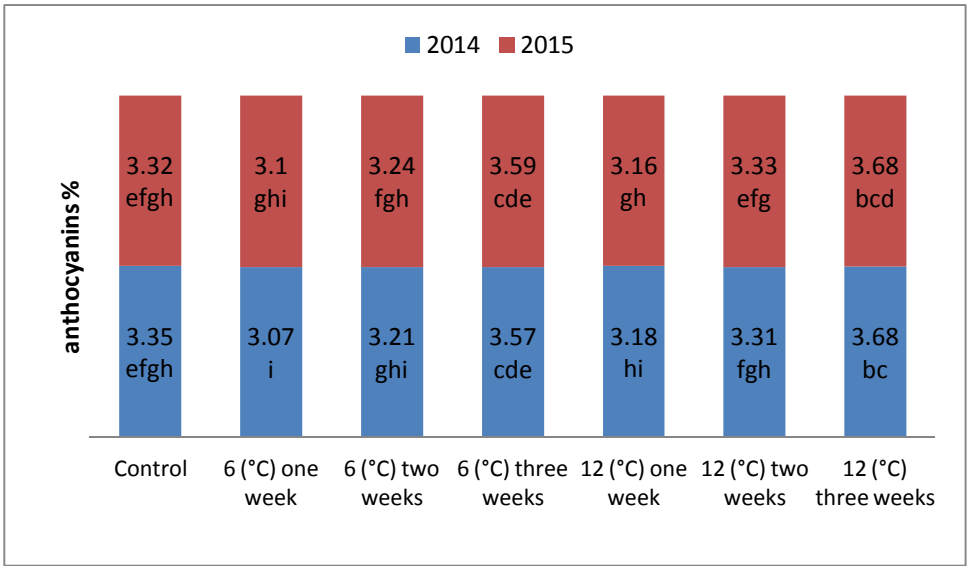


Figure (1) Effect of planting date (A) and seed vernalization temperature (B) on anthocyanin (%) content of calyx of roselle plants during 2014and 2015 seasons.

(A)





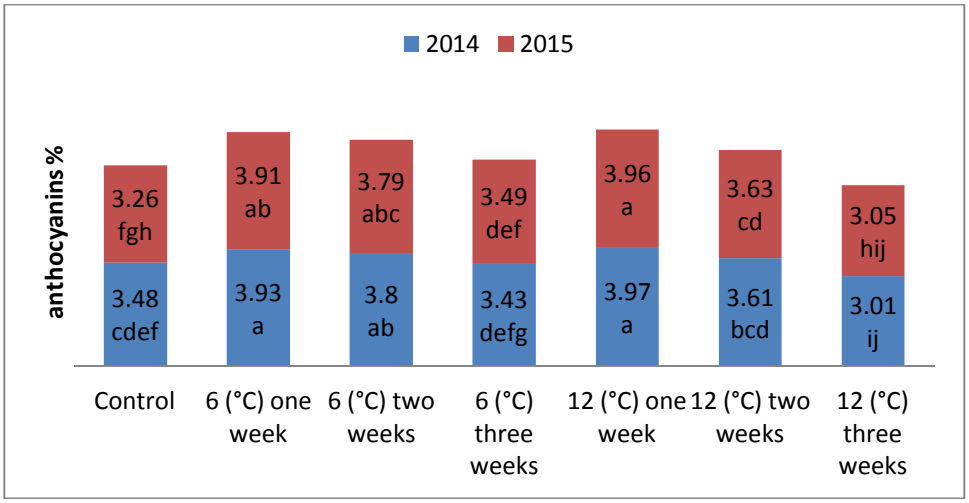


Figure (2) Effect of interaction of seed vernalization temperature, duration and planting date (April 13 th) (A), (May 13 th) (B) and (Jun 13 th) (C) on anthocyanin (%) content of roselle calyx plants during 2014 and 2015 seasons.

Table (2) Effect of planting date, seed vernalization temperature and duration, on plant height, branch number and stem diameter of roselle plants during 2014 and 2015 seasons.

Treatments			Plant height (cm)		Branch number (n)		Stem diameter (cm)	
Seasons			2014	2015	2014	2015	2014	2015
Planting date	Vernlization (0C)	Time						
(a) Effect of planting date (month)								
April			151.61 a*	156.85 a	12.73 b	13.36 b	2.06 a	1.75 ab
May			149.91 a	143.13 b	15.59a	20.13 a	3.75a	1.97 a
Jun			139.73 b	132.68 c	9.38 c	10.85 c	1.76 a	1.53 b
(b) Effect of Vernalization (^{0}C)								
	Control (25)		136.97 c	143.69 b	12.06 ab	14.72 a	1.94 a	1.96 a
	6		143.28 b	135.44 c	11.85 b	14.69 a	1.70 a	1.58 b
	12		153.14 a	153.11 a	13.39 a	14.89 a	1.91 a	1.87 a
(c) Effect of vernalization duration (week)								
		Control	136.97 c	143.69 b	12.06 b	14.72 b	1.94 a	1.96 a
		one	156.58 a	147.50 a	13.67 a	16.33 a	1.82 c	1.79 b
		two	137.17 c	138.92 c	11.92 b	14.33 b	1.63 d	1.64 c
		three	150.88 b	146.42 ab	12.28 b	13.69 b	2.00 b	1.76 b
(D) Effect of interaction								
April	Control (25)	Control	145.00 de	160.08 bcd	14.75 bc	11.00 fghi	2.26 b	1.91 bcd
April	6	one	148.50 de	152.50de	8.00 g	20.00 bc	2.01 b	1.82 bcde
April	6	two	132.50 fg	156.00 de	15.00 b	23.67 a	2.31 b	1.85 bcd
April	6	three	186.00 a	165.00 abc	3.50 i	22.67 ab	1.88 b	2.05 abc
April	12	one	152.00 cd	157.00 cd	10.00 f	13.50 efg	1.77 b	1.80 bcde
April	12	two	130.50 g	157.50 cd	12.10 de	13.00 fgh	2.10 b	1.86 bcd
April	12	three	163.00 b	166.00 ab	8.00 g	17.00 cde	1.84 b	2.13 ab
May	Control (25)	control	147.33 de	137.50 g	6.00 h	18.58 cd	2.98 a	2.30 a
May	6	one	161.00 b	117.00 i	11.00 ef	7.00 j	2.03 b	1.87 bcd
May	6	two	134.00 fg	117.00 i	11.00 ef	13.00 fgh	1.52 b	1.61 def
May	6	three	145.50 de	135.50 g	15.00 b	9.00 ij	1.37 b	1.49 efg
May	12	one	150.25 cd	157.00 cd	19.0a	25.50a	2.98 a	2.30 a
May	12	two	166.00 b	172.50 a	13.00 cd	11.50 fghi	1.86 b	1.97 abc
May	12	three	146.00 de	148.50 ef	15.00 b	11.00 ghi	1.73 b	1.78 cde
Jun	Control (25)	Control	118.58 hi	133.50 gh	15.42 b	14.58 defg	1.44 b	1.76 cdef
Jun	6	one	115.00 i	110.00 i	13.67 bcd	12.67 fgh	1.73 b	1.71 cdef
Jun	6	two	126.50 gh	140.50 fg	12.00 de	9.50 hij	1.08 b	0.94 h
Jun	6	three	140.50 ef	125.50 h	17.50 a	14.67 def	1.40 b	1.24 gh
Jun	12	one	178.50 a	140.00 g	12.50 de	12.00 fghi	1.89 b	1.44 fg
Jun	12	two	133.50 fg	141.50 fg	12.00 de	13.50 efg	2.06 b	1.58 def
Jun	12	three	158.50 bc	138.00 g	19.23 a	17.00 cde	1.53 b	2.02 abc

* Means followed by the same letter within a column are not significantly different at 0.05 level of probability accor ding to L.S.D. test

Table (3) Effect of planting date, seed vernalization temperature and duration, on root number, root length, fresh and dry weight g / of leaves, stem and root of roselle plants during 2014and 2015 seasons.

Treatments			Root number/plant (n)		Root length (cm)		Dry weight of leaves		Dry weight of stem		Dry weight of root	
Seasons			2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Planting date	Vernlization (0C)	Time										
(a) (a) Effect of planting date (month))												
April			14.40 b*	13.85 b	34.66 b	37.78 b	27.46 b	36.15 b	156.43 c	226.93 b	22.73 b	25.60 b
May			16.06 a	18.03 a	39.26 a	42.03 a	75.10 a	88.72 a	297.33 a	325.21 a	27.83 a	31.80 a
Jun			14.09 b	13.65 b	31.43 c	29.23 c	30.60 b	27.56 c	243.75 b	197.31 b	21.13 b	19.36 c
(b) Effect of Vernalization (^{0}C)												
	Control (25)		16.64 a	16.31 a	36.04 a	38.97 a	38.43 b	42.39 b	213.70 a	306.18 a	22.07 a	25.22 a
	6		15.04 ab	13.93 a	35.64 a	36.38 a	40.42 b	47.90 ab	225.33 a	243.65 a	23.96 a	25.05 a
	12		14.27 b	15.67 a	34.39 a	35.72 a	49.67 a	55.60 a	243.85 a	243.46 a	24.24 a	26.22 a
(c) Effect of vernalization duration (week)												
		Control	16.64 a	16.31 a	36.04 ab	38.97 a	38.43 b	42.39 d	213.70 b	306.18 a	22.07 b	25.22 ab
		one	13.54 c	16.00 a	33.97 bc	37.44 a	57.75 a	54.74 a	321.13 a	241.06 c	30.68 a	26.57 a
		two	15.50 b	18.67 a	33.58 c	38.11 a	37.86 b	51.98 b	202.09 b	234.89 c	21.80 b	25.60ab
		three	14.92 b	16.69 a	37.49 a	32.60 b	39.53 b	48.53 c	180.54 c	254.72 b	19.81 c	24.65 b
(D) Effect of interaction												
April	Control (25)	control	17.75 abcd	17.00 b	46.63 ab	47.75 bc	21.00hij	15.44 i	193.88 efgh	347.75 bc	26.00 cd	32.13 bcd
April	6	one	15.50 cdef	16.67 b	32.50 efg	56.50 a	44.32 f	75.60 c	197.85 efgh	294.16 cd	18.08 fg	27.28 defg
April	6	two	18.67 ab	23.00 b	28.50 gh	51.50 ab	89.88 b	105.30 a	361.12 b	383.70 ab	39.16 a	30.43 bcde
April	6	three	16.50 bcde	18.00 b	47.75 a	32.25 ghijk	45.29 f	93.05 b	192.15 efgh	291.43 cd	37.25 ab	35.53 ab
April	12	one	9.33 ig	19.33 b	29.50 fgh	42.50 cd	67.75 de	48.50 def	178.25 ghij	340.25 bc	22.25 de	34.75 abc
April	12	two	13.50 fgh	19.50 b	34.00 def	36.50 efg	72.90 d	80.43 c	223.85 ef	285.63 cd	21.60 def	28.10 cdef
April	12	three	12.00 gh	16.67 b	38.50 cd	35.00 efghi	83.55 bc	83.33 bc	269.73 cd	243.35 def	16.60 ghi	34.02 bc
May	Control (25)	Control	18.25 abc	18.50 b	30.50 efgh	38.92 def	59.02 e	72.23 c	214.25 efg	349.30 bc	18.45 efg	23.79 efgh
May	6	one	13.33 fg	14.00b	33.50 ef	30.00 ijkl	22.50 hi	40.00 efg	147.75 ijkl	208.75 efg	21.25 ef	22.50 fgh
May	6	two	18.75 ab	24.00 b	46.00 ab	34.50 fghij	15.50 ij	21.25 hi	142.50 jkl	182.75 fgh	26.00 cd	21.67 ghi
May	6	three	11.50 hi	21.00 b	43.00 bc	40.00 de	20.00 hij	30.32 gh	164.32 hijk	249.50 de	33.25 b	32.00 bcd
May	12	one	19.50 a	24.17 a	43.50 ab	37.50 defg	125.40 a	105.61 a	594.65 a	436.95 a	40.52 a	40.75 a
May	12	two	15.00 def	18.00 b	34.00 def	51.00 b	16.55 ij	38.17 fg	163.82 hijk	207.00 efg	13.60 hij	28.75 cde
May	12	three	15.50 cdef	14.50 b	30.67 efgh	27.00 klm	26.75 gh	52.50 de	116.95 lm	92.75 i	17.83 fg	12.63 j
Jun	Control (25)	Control	13.92 efgh	11.50 b	31.00 efgh	30.25 hijkl	35.26 fg	39.5 efg	232.98 de	221.50 defg	21.75 def	19.75 hi
Jun	6	one	9.00 j	11.50 b	29.50 fgh	22.67 m	19.05 hij	19.00 hi	89.25 m	135.75 hi	12.17 j	16.05 ij
Jun	6	two	14.33 efg	13.00 b	27.00 h	25.67 lm	11.83 j	12.50 i	135.50 kl	134.25 hi	13.20 ij	12.67 j
Jun	6	three	17.00 bcd	14.00 b	33.00 efg	34.33 fghij	15.33 ij	21.50 hi	195.00 efgh	167.00 gh	15.25 ghij	17.00 hij
Jun	12	one	15.33 def	14.33 b	35.33 de	35.50 efgh	38.25 f	15.00 i	278.00 c	176.00 gh	28.50 c	20.25 hi
Jun	12	two	12.00 gh	14.50 b	32.00 efg	29.50 jkl	20.50 hij	54.25 d	185.75 fghi	216.00 efg	17.25 fgh	21.67 ghi
Jun	12	three	17.00 bcd	16.00 b	32.00 efg	27.00 klm	75.50 cd	35.17 g	586.15 a	338.75 bc	40.00 a	28.25 cdef

* Means followed by the same letter within a column are not significantly different at 0.05 level of probability accor ding to L.S.D. test

Table (4) Effect of planting date, seed vernalization temperature and duration, on N, P and K % of roselle plants during 2014and 2015 seasons.

Treatments			N%		P%		K%	
Seasons			2014	2015	2014	2015	2014	2015
Planting date	Vernlization (0C))	Time						
(a) Effect of planting date (month)								
April			0.09 ab*	0.08 b	0.50 ab	0.47 a	2.08 a	2.72 a
May			0.10 a	0.09 a	0.62 a	0.51 a	2.26 a	2.08 b
Jun			0.06 b	0.07 c	0.41 b	0.43 a	1.93 a	1.86 c
(b) Effect of Vernalization (^{0}C)								
	Control (25)		0.07 a	0.07 a	0.48 b	0.46 a	2.04 a	2.35 a
	6		0.08 a	0.08 a	0.49 b	0.47 a	2.10 a	2.16 b
	12		0.09 a	0.08 a	0.58 a	0.49 a	2.13 a	2.19 ab
(c) Effect of vernalization duration (week)								
		control	0.07 ab	0.06 b	0.48 b	0.46 a	2.04 b	2.35 a
		one	0.09 a	0.08 a	0.52 a	0.48 a	2.35 a	2.31 a
		two	0.06 b	0.07 ab	0.50 ab	0.47 a	2.00 b	2.06 b
		three	0.08 ab	0.08 a	0.45 b	0.47 a	2.01 b	2.17 b
(D) Effect of interaction								
April	Control (25)	control	0.09 ab	0.08 bcd	0.41 b	0.39 cd	1.20 fghi	2.86 a
April	6	one	0.07 ab	0.08 bcd	0.37 b	0.40 cd	1.98 cdefgh	2.81 a
April	6	two	0.06 b	0.06 cd	0.41 b	0.40 cd	2.16 cde	2.91 a
April	6	three	0.09 ab	0.06 cd	0.43 b	0.37 cd	1.69 ghij	2.57 ab
April	12	one	0.10 a	0.09 abc	0.44 b	0.57 abcd	1.60 hij	1.55 f
April	12	two	0.10 a	0.08 bcd	0.43 b	0.49 bcd	2.06 cdefg	2.56 ab
April	12	three	0.09 ab	0.07 cd	0.39 b	0.51 abcd	2.76 ab	2.59 a
May	Control (25)	control	0.06 b	0.07 cd	0.54 b	0.61 ab	2.73 ab	1.98 cde
May	6	one	0.07 ab	0.08 bcd	0.55 b	0.57 abc	2.36 c	2.08 cd
May	6	two	0.06 b	0.11 ab	0.52 b	0.49 abcd	2.13 cdef	2.01 cde
May	6	three	0.09 ab	0.09 abc	0.50 b	0.51 abcd	2.22 cde	1.79 def
May	12	one	0.10 a	0.12 a	0.80 a	0.70 a	2.87 a	2.64 a
May	12	two	0.10 a	0.08 bcd	0.44 b	0.46 bcd	2.00 cdefg	1.65 ef
May	12	three	0.07 ab	0.08 bcd	0.42 b	0.36 d	1.86 efghi	1.84 def
Jun	Control (25)	control	0.08 ab	0.05 d	0.52 b	0.46 bcd	1.59 ij	2.23 bc
Jun	6	one	0.08 ab	0.06 cd	0.59 ab	0.40 cd	1.45 j	1.95 cdef
Jun	6	two	0.07 ab	0.08 bcd	0.57 b	0.42 cd	1.92 defghi	2.10 cd
Jun	6	three	0.07 ab	0.07 cd	0.57 b	0.43 cd	2.38 bc	1.99 cde
Jun	12	one	0.08 ab	0.06 cd	0.61 ab	0.44 bcd	2.06 cdefg	1.98 cde
Jun	12	two	0.08 ab	0.08 bcd	0.47 b	0.41 cd	2.20 cde	2.05 cde
Jun	12	three	0.06 b	0.07 cd	0.53 b	0.42 cd	2.31 cd	2.08 cd

* Means followed by the same letter within a column are not significantly different at 0.05 level of probability accor ding to L.S.D. test

Table (5) Effect of planting date, seed vernalization temperature, duration and, their interaction on number of fruit, dry weight of Calyx and Anthocyanin % of roselle plants during 2014and 2015 seasons.

Treatments			Number of fruits		Dry weight of Calyx		Dry weight of Calyx /fed	
Seasons			2014	2015	2014	2015	2014	2015
Planting date	Vernlization (0C)	Time						
(a) Effect of planting date (month)								
April			62.08 a*	68.53 a	22.42 a	21.40 a	298.91 a	285.28 a
May			64.93 a	68.53 a	22.42 a	21.40 a	298.91 a	285.28 a
Jun			60.28 a	59.03 b	17.67 c	18.44 b	235.65 c	245.84 b
(b) Effect of Vernalization (degree celcius)								
	Control (25)		57.11 b	66.94 a	16.55 b	19.41 ab	220.61 b	258.74 ab
	6		56.48 b	56.76 a	19.34 b	17.95 b	257.91 b	239.29 b
	12		69.56 a	67.03 a	22.02 a	21.97 a	293.62 a	292.87 a
(c) Effect of vernalization duration (week)								
		control	57.11 bc	66.94a	16.55 c	19.41 b	220.61 c	258.74 b

Treatments			Number of fruits		Dry weight of Calyx		Dry weight of Calyx /fed	
Seasons			2014	2015	2014	2015	2014	2015
Planting date	Vernlization (0C)	Time						
		one	74.17 a	63.92 ab	23.32 a	22.59 a	310.90 a	301.19 a
		two	61.42 b	61.06 bc	20.09 b	19.12 b	267.90 b	254.96 b
		three	53.47 c	60.58 c	18.64 bc	18.16 b	248.49 bc	242.08 b
(D) Effect of interaction								
April	Control (25)	control	47.75 efg	60.08 defg	18.49 fghij	24.18 bcd	246.08 fghij	322.33 bcd
April	6	one	35.00 gh	31.67 i	9.55 m	12.33 h	127.33 m	164.44 h
April	6	two	65.00 cd	75.50 bc	29.55 bc	17.30 efg	393.99 bc	230.66 efg
April	6	three	84.00 b	68.00 bcde	24.63 de	27.10 abc	328.44 de	361.32 abc
April	12	one	92.00 b	92.50 a	14.25 jkl	17.85 ef	189.99 jkl	237.99 ef
April	12	two	61.00 cdef	75.50 bc	22.85 defg	20.67 de	304.66 defg	275.64 de
April	12	three	60.50 cdef	71.00 bcd	20.94 efghi	20.43 de	279.24 efghi	272.44 de
May	Control (25)	control	61.33 cdef	78.50 ab	13.67 jklm	13.51 gh	182.20 jklm	180.08 gh
May	6	one	59.00 cdef	57.00 efg	25.95 cd	11.56 h	345.99 cd	154.06 h
May	6	two	63.00 cd	73.00 bcd	15.59 jk	19.00 e	207.91 jk	253.33 e
May	6	three	71.00 c	63.33 cdef	21.75 defgh	26.70 abc	289.99 defgh	356.04 abc
May	12	one	129.50 a	88.50 a	37.2 a	28.65 a	495.99 a	381.99 a
May	12	two	32.50 h	36.00 i	10.93 lm	13.20 h	145.73 lm	175.99 h
May	12	three	52.50 def	50.00 gh	17.05 ijk	18.50 e	227.33 ijk	246.66 e
Jun	Control (25)	control	62.25 cde	62.50 cdefg	17.52 hijk	20.54 de	233.55 hijk	273.82 de
Jun	6	one	48.50 ef	52.00 fgh	15.80 jk	13.20 h	210.66 jk	175.99 h
Jun	6	two	35.33 gh	31.00 i	12.93 klm	19.48 e	172.44 klm	259.73 e
Jun	6	three	47.50 fg	41.00 hi	18.33 ghij	14.85 fgh	244.44 ghij	198.04 fgh
Jun	12	one	66.50 c	67.50 bcde	33.55 ab	24.00 cd	447.32 ab	319.99 cd
Jun	12	two	64.00 cd	75.33 bc	23.35 def	26.43 abc	311.33 def	352.44 abc
Jun	12	three	67.50 c	64.50 bcdef	18.07 hij	27.95 ab	240.97 hij	372.66 ab

* Means followed by the same letter within a column are not significantly different at 0.05 level of probability accor ding to L.S.D. test

5. Conclusion

Hibiscus sabdariff variety "Sabahia 17" plant was selected to study the effects of planting date and seed vernalization temperature and duration on growth and yield. Based on the results of the experiment, it is recommended that planting rosella plant in May is the suitable time to produce the highest growth and yield of plant. Vernalization of seeds at 12°C for one week and planting in 13th May is the most suitable practice for growing rosella in Ismalia, Egypt.

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