

EFFECT OF TREATING THE SEEDS AND SEEDLINGS WITH CHILLING AND DIETHYL SULPHATE ON THE GROWTH AND FLOWERING OF *ANTIRRHINUM MAJUS*

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ABSTRACT

Seeds and seedlings of *Antirrhinum majus* cv. Snowflake were chilled (4 and -20°C for seeds and 4°C for seedlings) or treated with different concentrations of the chemical mutagen diethyl sulphate (DES) (0.3, 0.6 and 0.9%) and then the treatments were transplanted in the plastic house and on the open field. The effect on the vegetative growth, flowering characters and induction of variations was assessed in three generations. In general, the decrease in plant height and inflorescence length was correlated with increasing the concentrations of DES and decreasing the temperature or the period of exposure to chilling. The variations included dwarf plant and leaf and floret changes.

INTRODUCTION

The genus *Antirrhinum* (Fam. *Plantaginaceae*, Olmstead *et al.* 2001) has about 20 species, most found around the Mediterranean Sea and in North America (Stubbe 1966). Among them, only *Antirrhinum majus* has been domesticated as an ornamental which commonly known as snapdragon. The cultivar Snowflake has white flowers, belongs to tall cultivars (80-100cm) and used as a cut flower or border plant.

Chilling (vernalization) is subjection of seeds or seedlings to low temperatures in order to hasten germination, vegetative growth and flowering. *Antirrhinum majus* can be vernalised as shown by Cremer *et al.* (1998).

Cheng *et al.* (2001) on *Paeonia suffruticosa* reported that the pre-chilling decreased the stem length and advanced the flowering date.

Shinoda and Murata (2004) found that low temperature hastened the flowering of *Allium caeruleum*, but reduced flowering percentage.

The chemical mutagen diethyl sulphate (DES) belongs to alkylating agents. Chemical mutagens induced mutations like dwarf plants (Sasi *et al.*, 2005 on bhendi), changes in leaves shape (Jabeen and Mirza, 2004 on *Capsicum annum*) and changes in florets (Bhate, 1999 on *Ipomoea purpurea*).

Pavadaï and Dhanavel (2004) determined the effects of ethyl methanesulfonate (EMS), diethyl sulfate (DES) and colchicine on the seeds of soyabean. Shoot length decreased with the increase in the concentration of EMS, DES and colchicine. Days to first flower increased with the increase in the concentration of EMS, DES and colchicine.

Sinha and Joshi (1987) mentioned that spike length was reduced by treated seeds of hexaploid triticales with chemical mutagens.

Number of florets per inflorescence was increased by chemical mutagens (Reddy, 2001 on seeds of triticales, barley and wheat).

The aim of this work was to induce variation in *Antirrhinum majus* cv. Snowflake using chilling or diethyl sulphate.

MATERIALS AND METHODS

The investigation was carried out during the years from 2001 till 2005 in the Central Laboratory for Agricultural Climate, Giza, Egypt. Three generations were conducted.

Plant materials were seeds and seedlings of *A. majus* cv. Snowflake (cut flower crop). Seeds were treated with DES (0, 0.3, 0.6 and 0.9%) and other seeds were exposed to low temperature (4°C and -20°C) on December 30, 2001 for one week and planted in seedling trays until the seedlings reached a height of six cm, then they were transplanted in plastic bags and arranged in blocks in plastic house or on open field. For seedlings, five groups of seeds were sown in seedling trays and put in a plastic house until the seedlings reached a height of six cm. First and second groups were put in refrigerator by 4°C for one or two weeks, while the other three groups were treated with 0.3, 0.6 and 0.9% DES.

In the three generations, treatments were as follow in both locations (plastic house and open field):

1. Untreated seeds (dry) (control).
2. Seeds treated with 4°C for one week.
3. Seeds treated with -20°C for one week.
4. Seeds treated with 0.0% DES (soaked).
5. Seeds treated with 0.3% DES (soaked for 22h).
6. Seeds treated with 0.6% DES (soaked for 22h).
7. Seeds treated with 0.9% DES (soaked for 22h).
8. Seedlings treated with 4°C for one week.
9. Seedlings treated with 4°C for two weeks.
10. Seedlings treated with 0.3% DES (25ml solution to the soil).
11. Seedlings treated with 0.6% DES (25ml solution to the soil).
12. Seedlings treated with 0.9% DES (25 ml solution to the soil).

Seeds which obtained from the selfing of M₁ and M₂ plants were sown in seedling trays until the seedlings reached a height of six cm then moved to plastic house or open field.

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In each location, plants were arranged according to a randomized complete block design containing three replicates. Every replicate contained 12 treatments.

Two weeks after transplanting to the plastic house or open field, the following data were recorded:

1. Plant height
2. Flowering date
3. Inflorescence length
4. Number of florets per inflorescence
5. Variations

Statistical Design and Analysis

Combined Analysis of Variance for Randomized Complete Block Design was used and data were subjected to analysis of variance. Separation of means among treatments was determined using LSD test at 5% (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

1. Plant Height

Data in Table (1) showed that plant height was significantly affected by both treatments groups (DES and chilling) as well as by location in the three generations. Plastic house treatments gave taller plants (35.42– 51.87– 73.50cm) than those on the open field treatments (27.90- 33.30- 67.04cm) in the three generations respectively. The data showed that the plant height for the seed treatments decreased with chilling in all generations. This result is in agreement with those obtained by Cheng *et al.* (2001) on *Paeonia suffruticosa* and Wang *et al.* (2003) on *Raphanus sativus*.

Generally, the increase of chilling period for the seedlings caused an increase in M_1 - plant height as compared with the control. This result was in accordance with that reported by Mi and Wang (1998) on rice seedlings.

Generally, there was an apparent trend towards decreasing the plants height with increasing the concentrations of DES in all generations.

For the seedlings treatments in the plastic house, the treatments of 0.3% DES produced the highest plants as compared with the control and the other two concentrations, while the seed treatment of 0.6% DES gave the shortest plants. The proportional decrease in plant height with increasing the concentrations of chemical mutagens found in the present study was supported by many investigators on

the other plants, i.e. [Pavadai and Dhanavel (2004) on soyabean and Bhat *et al.* (2007) on *Vicia faba*]. These results might be attributed to the physiological damage caused by DES and its hydrolysis products.

2. Flowering Date

The data revealed that the effects of locations and treatments were highly significant in all generations except for the effect of locations in the M_1 and the effect of treatments in the M_2 , where they were only significant, while the interaction between locations and treatments was not significant in all generations (Table 2). Plants on the open field flowered earlier than plants in the plastic house.

The data showed that the average number of days from seed sowing till the beginning of flowering decreased in the three generations with decreasing the degrees of temperature as compared with the control treatments. This result is in agreement with those obtained by Cremer *et al.* (1998) on *Antirrhinum majus*. On the other side, Pandey and Singh (1999) reported that chilling treatments caused delayed flowering date on strawberry.

Increasing the period of seedlings exposure to chilling resulted in an earlier flowering date as compared with the control treatments in all generations. Shinoda and Murata (2004) on *Allium caeruleum* and Khokhar *et al.* (2007) on onion suggested that increasing the exposure period of chilling caused a decrease in flowering date. But, Sudipta and Sharma (2005) on maize reported contrary conclusion.

According to the data obtained from seed treatments, increasing the DES concentration caused delayed flowering date in the M_3 .

In seedlings treatments, increasing the DES concentration caused delay in the flowering date in the M_1 and M_2 generations with some exceptions. The treatment of 0.3% DES in the M_1 generation flowered before the control treatment, the concentration of 0.6% DES delayed the flowering than 0.9% DES and the control treatment in the M_2 generation delayed the flowering date than 0.3 and 0.6% DES. In the M_3 generation, increasing the DES concentration caused later flowering date except by 0.3% DES treatment.

These results are in conforming to those of El-Nagar (2002) on carnation and Pavadai and Dhanavel (2004) on soyabean who found that chemical mutagens seemed to inhibit cell growth, decrease the rate of growth and delay the flowering.

Table (1): Plant height of *Antirrhinum majus* as affected by locations (plastic house and open field) and treatments (DES and chilling) in the three generations.

Plant height (cm)											
Treatments	M ₁ (2003)			M ₂ (2004)			M ₃ (2005)				
	Plastic house	Open field	Mean	Plastic house	Open field	Mean	Plastic house	Open field	Mean		
control	36.97	30.54	33.76 bc	54.18	36.05	45.12 ab	78.72	73.11	75.91 abc		
Seeds 4°C	35.15	28.23	31.69 cd	49.83	33.32	41.57 bcd	76.83	65.83	71.33 abcde		
Seeds -20°C	31.19	26.35	28.77 ef	48.02	30.51	39.26 cd	68.05	60.22	64.13 ef		
Soaked seeds	37.85	30.66	34.25 bc	54.45	33.21	43.83 bc	80.11	74.33	77.22 ab		
Seeds 0.3% DES	30.00	27.55	28.78 ef	48.61	33.05	40.83 bcd	71.22	67.05	69.18 cdef		
Seeds 0.6% DES	27.30	25.17	26.23 f	44.58	30.77	37.68 d	66.83	59.11	62.97 f		
Seeds 0.9% DES	31.27	21.49	26.38 f	45.67	30.21	37.94 d	68.05	58.61	63.33 f		
Seedlings 4°C one week	40.42	27.82	34.12 bc	57.28	33.18	45.21 ab	73.61	72.22	72.91 abcd		
Seedlings 4°C two weeks	43.36	32.15	37.75 a	60.70	38.54	49.67 a	81.44	75.00	78.22 a		
Seedlings 0.3% DES	39.92	28.74	34.33 b	57.26	33.96	45.61 ab	71.94	71.11	71.52 abcde		
Seedlings 0.6% DES	35.09	26.17	30.63 de	53.18	30.64	41.91 bcd	69.66	64.55	67.11 def		
Seedlings 0.9% DES	36.50	29.96	33.23 bcd	48.69	36.11	42.40 bcd	75.55	63.33	69.44 bcdef		
Mean	35.42 a	27.90 b	31.66	51.87 a	33.30 b	42.58	73.50 a	67.04 b	70.27		
LSD _{0.05} Treatments	2.63			5.68			7.99				
LSD _{0.05} Locations	2.28			2.05			4.34				
LSD _{0.05} T/L	3.72			N.S			N.S				

Table (2): Flowering date of *Antirrhinum majus* as affected by locations (plastic house and open field) and treatments (DES and chilling) in the three generations.

Treatments	Flowering date (days)											
	M ₁ (2003)				M ₂ (2004)				M ₃ (2005)			
	Plastic house	Open field	Mean		Plastic house	Open field	Mean		Plastic house	Open field	Mean	
control	192.49	186.75	189.62	bc	228.73	205.98	217.36	c	269.39	261.55	265.47	de
Seeds 4°C	179.90	174.19	177.04	ab	220.69	197.99	209.34	bc	261.20	257.00	259.10	abcd
Seeds -20°C	162.00	174.92	168.46	a	216.28	197.79	207.04	ab	260.40	245.72	253.06	abc
Soaked seeds	184.80	159.22	172.01	a	213.53	183.82	198.67	a	256.89	244.94	250.91	ab
Seeds 0.3% DES	203.99	204.59	204.29	c	219.65	200.38	210.02	bc	266.69	256.67	261.68	cd
Seeds 0.6% DES	218.75	185.16	201.96	c	224.83	201.05	212.94	bc	278.54	264.94	271.74	e
Seeds 0.9% DES	210.90	198.19	204.55	c	219.44	200.78	210.11	bc	281.28	266.92	274.10	e
Seedlings 4°C one week	174.40	165.87	170.13	a	221.25	201.07	211.16	bc	266.40	253.06	260.06	bcd
Seedlings 4°C two weeks	167.70	163.40	165.55	a	219.62	202.85	211.23	bc	261.20	257.00	259.73	abcd
Seedlings 0.3% DES	188.67	165.42	177.04	ab	223.53	199.87	211.70	bc	257.07	244.61	250.84	a
Seedlings 0.6% DES	205.00	191.00	198.00	c	219.68	205.78	212.73	bc	264.72	255.39	260.97	cd
Seedlings 0.9% DES	198.53	189.13	193.83	c	224.00	212.55	218.27	c	263.23	251.32	257.28	abcd
Mean	190.59 b	179.82 a	185.21		220.94 b	200.83 a	210.88		265.77 b	255.05 a	260.41	
LSD _{0.05} Treatments	15.42				9.50				9.16			
LSD _{0.05} Locations	7.57				5.60				6.00			
LSD _{0.05} T/L	N.S				N.S				N.S			

3. Inflorescence Length

The inflorescence length was highly significant affected by the treatments (DES and chilling) in all generations except in the M₃ generation, which was only significant, but it was not significant influenced by locations and interactions between locations and treatments in all generations except in the M₂ generation, where it was highly significant affected by location.

The data in Table 3 indicated that the plants in the plastic house had longer inflorescences than those on the open field. The decrease of chilling temperature by the seeds caused a decrease in the length of the inflorescence. Similar result was obtained by Noack *et al.* (1996) on *Hebe*, while that of other reporter on *Gypsophila paniculata* was in disagreement with this result (Cheong *et al.* 1999).

According to the data, the inflorescence length increased with increasing the period of exposure to chilling as compared with the control treatment in the M₃ generation. On the other side, in the M₂ generation, the control treatment had longer inflorescence as compared with the seedlings exposed to 4°C. In the M₁ generation, the seedlings treatment with 4°C for one week had shorter inflorescence as compared with the two other treatments (4°C for two weeks and the control).

Noack *et al.* (1996) indicated that increasing the exposure period to chilling decreased the length of the inflorescence of *Hebe*.

Generally, the inflorescence length decreased with increasing the concentrations of DES for seeds or seedlings treatments as compared with 0.0% DES for seeds and with the control for seedlings treatments in all generations with some exceptions.

Similar results were reported by Rachovska and Dimova (2000) on winter common wheat. They found that the chemical mutagens caused a decrease in the inflorescence length. Contradicting, EL-Nashar (2006) on *Amaranthus caudatus* and *A. hypochondriacus* mentioned that the chemical mutagens caused an increase in the inflorescence length.

4. Number of Florets per Spike

The number of florets per spike was affected by location, DES and chilling in the three generations. A highly significant response was recorded by the effect of the treatments and locations in the M₁, M₂ and M₃ generations except by the effect of location in the M₁ generation it was not significant. The interaction between locations and treatments was not significant in the three generations (Table4).

Plants in the plastic house produced more florets per spike than those on the open field, especially in M₂.

In all generations, chilling of seeds decreased the number of florets per inflorescence. Shinoda and Murata (2004) on *Allium caeruleum* reported the same

result. On the other hand, this finding disagrees with those reported by JeongSeob *et al.* (2003) on *Hepatica asiatica*.

The number of florets per inflorescence increased with increasing the chilling period as compared with the control treatments, but in the M₁ and M₂ generation the control treatment gave number of florets higher than seedlings exposed to 4°C for one week. This result coincides with those of Khokhar *et al.* (2007) on onion. But, Molina *et al.* (2005) on saffron reported contradicting conclusion.

In seeds treatments, the concentration of 0.0% DES (soaked seeds) had the largest number of florets per inflorescence as compared with the other concentrations (0.3- 0.6- 0.9%) in all generations.

The increase in florets number with increasing the concentration of the chemical mutagens has been reported by Reddy (2001) on triticale, barley and wheat. But, they are in disagreement with that which was obtained by Rachovska and Dimova (2000) on winter common wheat.

5. Variations

5.1. Changes in Growth Habit

Growth habit of some plants was changed showing an inhibition in the vegetative growth resulting in dwarfed growth. These changes in the growth habit were observed in the M₁ generation. There were two dwarf plants belongs to seedlings treated with 0.6% DES in the plastic house, one plant belongs to seeds exposed to -20°C and sown in the plastic house and one plant belongs to seeds treated with 0.9% DES and grown on the open field.

The gained selfed seeds of each dwarfed plant either in the M₁ and M₂ generation were separately planted to investigate their hereditary behaviour. The dwarfness seemed to be just a modification because it did not appear in the M₂ or M₃ progeny respectively.

This result is in agreement with those of Datta and Sengupta (2002) on coriander and Sasi *et al.* (2005) on *Abelmoschus esculentus*.

The dwarfed growth can be attributed to the effect of DES on the apical bud which inhibits its growth or to the effect of chemical genetic factors controlling the growth habit of the plant.

Goulas *et al.* (2002) reported that chilling induced dwarf development in white clover plants.

5.2. Changes in Leaf Shape

Chilling and DES treatments caused changes in leaf shape in all generations. This result is in agreement with those of Odeigah *et al.* (1999) on *Vigna unguiculata*.

Nishizawa and Yasukawa (1992) reported that chilling plants of strawberry produced differentiated leaves.

Table (3): Inflorescence length of *Antirrhinum majus* as affected by locations (plastic house and open field) and treatments (DES and chilling) in the three generations.

Treatments	Inflorescence length (cm)								
	M ₁ (2003)			M ₂ (2004)			M ₃ (2005)		
	Plastic house	Open field	Mean	Plastic house	Open field	Mean	Plastic house	Open field	Mean
control	7.78	6.99	7.39 b	14.03	7.95	10.99 b	8.00	6.55	7.27 bcde
Seeds 4°C	3.34	4.57	5.20 cd	12.37	7.06	9.72 b	7.50	5.99	6.74 cde
Seeds -20°C	3.50	5.64	3.95 d	9.29	6.23	7.76 bc	6.77	5.72	6.24 e
Soaked seeds	8.93	9.55	9.24 a	21.30	9.84	15.57 a	7.55	7.27	7.41 abcde
Seeds 0.3% DES	4.04	6.37	5.17 cd	9.85	5.69	7.77 bc	7.11	6.44	6.77 cde
Seeds 0.6% DES	6.50	3.84	4.57 d	7.52	4.87	6.19 c	6.89	6.33	6.61 de
Seeds 0.9% DES	4.72	6.19	5.46 cd	12.21	6.58	9.40 bc	7.89	6.05	6.97 cde
Seedlings 4°C one week	6.62	7.95	7.28 b	11.77	8.08	9.92 b	9.27	7.72	8.50 ab
Seedlings 4°C two weeks	9.26	7.01	8.13 ab	14.99	5.47	10.23 b	9.16	8.11	8.63 a
Seedlings 0.3% DES	6.97	7.54	7.26 b	11.45	8.17	9.81 b	9.00	6.94	7.97 abc
Seedlings 0.6% DES	5.59	7.42	6.50 bc	11.13	6.46	8.80 bc	8.72	6.66	7.69 abcd
Seedlings 0.9% DES	6.32	8.21	7.26 b	13.68	6.36	10.02 b	8.44	7.66	8.05 abc
Mean	6.13	6.77	6.45	12.47 a	6.90 b	9.68	8.02	6.79	7.41
LSD _{0.05} Treatments	1.73			3.24			1.36		
LSD _{0.05} Locations	N.S			2.06			N.S		
LSD _{0.05} T/L	N.S			N.S			N.S		

Table (4): Number of florets per spike of *Antirrhinum majus* as affected by locations (plastic house and open field) and treatments (DES and chilling) in the three generations.

Number of florets												
Treatments	M ₁ (2003)			M ₂ (2004)			M ₃ (2005)					
	Plastic house	Open field	Mean	Plastic house	Open field	Mean	Plastic house	Open field	Mean			
control	7.00	7.31	7.15 bcd	12.19	6.01	9.10 bcd	5.99	5.11	5.55 cde			
Seeds 4°C	3.51	2.83	4.84 efg	10.58	6.40	8.49 bcde	6.22	4.44	5.33 cde			
Seeds -20°C	4.93	3.44	4.19 efg	8.45	4.69	6.57 de	5.22	4.44	4.83 de			
Soaked seeds	5.87	8.96	7.42 abc	22.45	7.46	14.95 a	5.78	6.00	5.89 bcd			
Seeds 0.3% DES	2.83	4.78	3.80 fg	6.57	3.70	5.13 e	5.11	4.11	4.61 e			
Seeds 0.6% DES	2.48	3.90	3.19 g	8.85	4.97	6.91 cde	5.55	4.11	4.83 de			
Seeds 0.9% DES	2.83	4.78	5.46 def	10.59	5.38	7.98 bcde	5.11	4.78	4.94 cde			
Seedlings 4°C one week	6.23	7.50	6.87 bcd	13.77	4.18	8.98 bcd	8.44	6.55	7.49 a			
Seedlings 4°C two weeks	7.21	10.97	9.09 a	12.78	7.16	9.97 bcd	8.55	6.78	7.66 a			
Seedlings 0.3% DES	5.94	9.17	7.55 abc	14.88	5.74	10.31 bc	9.00	6.22	7.61 a			
Seedlings 0.6% DES	4.64	6.82	5.73 cde	10.43	6.86	8.64 bcde	6.89	5.44	6.16 bc			
Seedlings 0.9% DES	8.46	8.12	8.29 ab	15.71	5.94	10.82 b	8.66	5.55	7.11 ab			
Mean	5.31	6.96	6.13	12.27 a	5.71 b	8.99	6.71 a	5.29 b	6.00			
LSD _{0.05} Treatments	1.89			3.71			1.26					
LSD _{0.05} Locations	N.S			1.77			0.06					
LSD _{0.05} T/L	N.S			N.S			N.S					

5.3. Changes in Florets

Chilling caused initiation of petal-like structure from the style in the florets. This change was appeared in the plastic house plants which emerged from seeds exposed to -20°C in the M_1 , M_2 and M_3 generation, seedlings exposed to 4°C for one week in the M_1 , M_2 and M_3 generation and seedlings exposed to 4°C for two weeks in the M_1 and M_2 generation. This change was also found on the open field plants which emerged from seeds exposed to 4°C in the M_2 and M_3 generation, seedlings exposed to 4°C for one week in the M_2 generation and seedlings exposed to 4°C for two weeks in the M_1 and M_3 generation. M_1 and M_2 plants which had initiation of petal-like structure from the style in the florets were selfed and their seeds were sown, and the change appeared again in the most inflorescences of the M_2 and M_3 progenies, respectively, but this initiation of petal was shorter than his parent (Figure. 1).



Figure (1): Changes in the initiation of petal-like structure from the style

Modified petals originated from receptacle at the circumference of the stamens were observed in florets. Regarding chilling and DES in the M_1 generation, it was observed that the modified petals originated from receptacle at the circumference of the stamens were appeared in plants emerged from seeds exposed to -20°C in the plastic house, seedlings exposed to 4°C for two weeks in the plastic house, seedlings treated with 0.3% and 0.6% DES in the plastic house, seeds treated with 0.6% DES on the open field and from seedlings treated with 0.6% DES on the open field. In the M_2 generation, plants emerged from seeds exposed to -20°C in the plastic house, seedlings exposed to 4°C for one and two weeks in the plastic house, seedlings treated with 0.9% DES in the plastic house, seeds treated with 0.9% DES on the open field and from open field seedlings which exposed to 4°C for two weeks, gave plants with modified petals originated from receptacle at the circumference of the stamens. The results of the M_3 generation showed that plants emerged from exposed seeds to -20°C in the plastic house, seedlings exposed to 4°C for two weeks and seedlings treated with 0.6% and 0.9% DES in the plastic house, seeds treated with 0.9% DES and seedlings treated with 0.6% DES on the

open field, gave plants which had the same change in the two other generations. The plants in the M_1 and M_2 which had this change were selfed and their seeds were sown, and the change appeared again in most inflorescences of the M_2 and M_3 progeny, respectively, with an increase in the modified petals (Figure. 2).

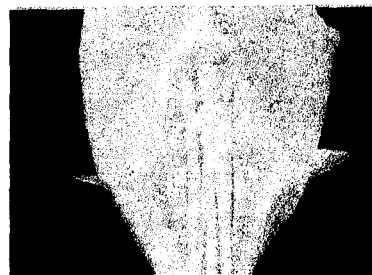


Figure (2): Changes in modified petals originated from receptacle at the circumference of the stamens

Minute modified petals formed from torus and located between calyx and corolla of *Antirrhinum majus* florets was appeared in the plastic house control plants in the M_2 and M_3 generation, plants emerged from seeds exposed to -20°C in the plastic house for the M_2 and M_3 generation, plants emerged from seeds treated with 0.3% DES in the M_2 generation and with 0.6% DES in the M_2 and M_3 generation, seedlings exposed to 4°C for one or two weeks in the plastic house for the M_2 generation, seedlings treated with 0.3% DES in the plastic house for the M_2 and M_3 generation, seedlings treated with 0.9% DES in the plastic house for the M_2 generation, seeds exposed to -20°C on the open field in the M_2 generation, seeds treated with 0.9% DES on the open field in the M_2 and M_3 generation, seedlings exposed to 4°C for two weeks on the open field for the M_3 generation and seedlings treated with 0.6% DES on the open field for the M_2 generation. This change was not found in the M_1 generation. The plants in the M_2 which had this change were selfed and their seeds were sown and the changes appeared again in most inflorescences of the M_3 progeny, but the minute modified petals were thin and shorter than those of the parents (Figure. 3).



Figure (3): Changes in formed minute modified petals from torus and located between calyx and corolla

The variation in florets as a result of chemical mutagens treatments was also found by Bhate (1999) on *Ipomoea purpurea*.

CONCLUSION

The results showed that growing the plants in the plastic house had resulted in better growth and flowering than growing the plants on the open field. Also, treating the seedlings with 4°C for two weeks gave the best results.

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الملخص العربي

تأثير معاملة البذور والشتلات بالبرودة و الداي إيثايل سلفيت على نمو و إزهار نبات حنك السبع

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أجرى هذا البحث فى المعمل المركزى للمناخ الزراعى التابع لمركز البحوث الزراعية - الجيزة فى الفترة من ٢٠٠١ الى ٢٠٠٥ و ذلك بغرض دراسة تأثير تركيزات مختلفة من المطفر الكيماوى كبريتات ثنائى كحول الإيثايل و معاملة البذور و الشتلات بدرجات حرارة منخفضة على النمو الخضرى و الزهرى وإحداث طفرات فى نبات حنك السبع *Antirrhinum majus* صنف Snowflake (إزهار قطف). و قد تمت زراعة ثلاثة أجيال فى كل من الصوبة البلاستيك و الحقل و كانت المعاملات بالنسبة للجيل الأول:

- ١- بذور غير معاملة.
- ٢- بذور معاملة ب ٤م.
- ٣- بذور معاملة ب - ٢٠م.
- ٤- بذور معاملة بمحلول صفر % كبريتات ثنائى كحول الإيثايل (منقوعة فى الماء).
- ٥- بذور معاملة بمحلول ٠,٣ % كبريتات ثنائى كحول الإيثايل (منقوعة لمدة ٢٢ ساعة).
- ٦- بذور معاملة بمحلول ٠,٦ % كبريتات ثنائى كحول الإيثايل (منقوعة لمدة ٢٢ ساعة).
- ٧- بذور معاملة بمحلول ٠,٩ % كبريتات ثنائى كحول الإيثايل (منقوعة لمدة ٢٢ ساعة).
- ٨- شتلات معاملة ب ٤م لمدة أسبوع.
- ٩- شتلات معاملة ب ٤م لمدة أسبوعين.
- ١٠- شتلات معاملة بمحلول ٠,٣ % كبريتات ثنائى كحول الإيثايل (٢٥ مل محلول مضافة إلى التربة).
- ١١- شتلات معاملة بمحلول ٠,٦ % كبريتات ثنائى كحول الإيثايل (٢٥ مل محلول مضافة إلى التربة).
- ١٢- شتلات معاملة بمحلول ٠,٩ % كبريتات ثنائى كحول الإيثايل (٢٥ مل محلول مضافة إلى التربة).

أما بالنسبة للجيل الثانى و الثالث فقد تمت زراعة البذور الناتجة من كل معاملة من الجيل الأول و الثانى فى كلا الموقعين للحصول على نباتات الجيل الثانى و الثالث على التوالى.

وقد كان تصميم التجربة فى صورة قطاعات عشوائية كاملة داخل الصوبة و فى الحقل و قد شمل كل موقع ثلاث مكررات كل منها به ١٢ معاملة. و قد تسبب تعريض البذور للبرودة الشديدة و زيادة تركيز المطفر الكيماوى فى تقليل طول النبات وطول الشمراخ، أما زيادة مدة تعرض الشتلات للبرودة فقد أدت الى زيادة طول النبات وطول الشمراخ وتأخر الإزهار .

و قد أظهرت النتائج بصفة عامة تفوق الزراعة فى الصوبة البلاستيك على الزراعة فى الحقل المكشوف و كذلك تفوق معاملة تعريض البادرات للبرودة (٤م) لمدة أسبوعين على بقية المعاملات.

و قد ظهرت تغيرات مورفولوجية مثل تقزم النباتات و تغير شكل الأوراق و تغيرات فى شكل الزهيرة.