Impact of Biological Fertilizers on Vegetative and Reproductive Traits of Stock (*Mathiola incana* L.)

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Leila Ahmadian¹, Mostafa Arab^{2*}, Mahmoud Reza Roozban²

1. Department of Horticulture, Science and Research Branch, Islamic Azad University, Tehran, Iran.

2. Department of Horticulture, College of Aburaihan, University of Tehran, Pakdasht, Tehran, Iran.

* Corresponding author email: mosarab@ut.ac.ir

ABSTRACT: Impacts of two kinds of biofertilizers (Nitroxin and Bio-phosphate), on vegetative and reproductive traits of two varieties of Stock (Mathiola incana L.) were evaluated in a factorial experiment with 12 treatment combinations (6 fertilizer treatments × 2 cultivar). Fertilizer treatments were including: 100% nitrogen and phosphate as chemical fertilizers (T1), 50% nitrogen and 100% phosphate and root inoculation and irrigation with Nitroxin in transplanting stage (T2), 100% nitrogen and 50% phosphate and root inoculation and irrigation with Bio-phosphate in transplanting stage (T3), 50% nitrogen and 50% phosphate and root inoculation and irrigation with Nitroxin and Biophosphate in transplanting stage (T4), 50% nitrogen and 50% phosphate, and root inoculation in transplanting stage and irrigation with Nitroxin and Bio-phosphate in transplanting and flowering stages (T5), Nitroxin and Bio-phosphate alone (T6). Chlorophyll content, leaf nitrogen and phosphorus concentrations, flowering time, inflorescence diameter, florets number and leaf area were the most important traits that were studied in this experiment. Based on the results, T1 (100% chemical fertilizers) and T5 (50% chemical fertilizers along with the use of bio fertilizers, irrigation and root inoculation in the transplanting and flowering stages were highest han other treatments for all traits except phosphorus. While treatment without chemical fertilizer (T6) for all traits resulted in lowest. Also, other treatments (T2, T3, and T4) have not desired effect on measured traits. This suggests that the use of bio-fertilizers Nitroxin and Bio-phosphate in transplanting and flowering stages can reduce utilization of chemical fertilizers by 50%.

Keywords: Bio-fertilizers,, Bio-phosphate, Nitroxin, Roots inoculation

INTRODUCTION

Iran is a land with diverse climatic conditions that is one of the most talented areas for floriculture, and also Iran is center of origin of many ornamental plants throughout world. So that only 4,700 hectares of lands devoted to the cultivation of flowers and ornamental plants. Nevertheless, Iran's position in terms of production and export of these products are 17 and 107, respectively. If the existing capabilities in production of ornamental plants in the country are properly identified and applied, in addition to supplying the domestic market, also can be obtained a greater share of world trade in ornamental plants. One way to achieving this goal, is using of technologies and the world's knowledge in the field of optimum nutrition in ornamental plants (2).

Use of the chemical fertilizers for years is considered as the most common source of plant nutrition. Excessive use of these fertilizers in successive years, causing an imbalance of nutrients in the soil, reducing crop yields, danger to human health and other living organisms. For this season, today, types of biofertilizers origin of bacteria, fungi, algae and other organisms that live in soil is taken into consideration, that their mechanisms of action will lead to increased nutrient uptake by plants from soil (3).

Stock is one of the ornamental plant that due to its fragrant and beautiful flowers have great importance in industry. There are few reports on the use of biofertilizers in the ornamental plants. Application of biological phosphorus in two white varieties of Stock increased significantly inflorescences length (8). Application of biofertilizers in Calendula also enhanced performance and improve quality of its medicinal, while in Chamomile flowers only increased flower yield (16). In Maize, application of biological fertilizers improved plant nutrition and soil conditions also (18).

The present research was done in order to evaluate the effect of Nitroxin, Bio-phosphateand chemical fertilizers on vegetative and reproductive traits of two varieties of Stock in greenhouse conditions.

Experimental site and soil

MATERIAL AND METHODS

This experiment was carried out during 2012-2013 at the research greenhouse in Pakdasht, Tehran, Iran. The experimental site was situated by 33° 28 E, 51⁰ 44 N, and at altitude of 1180 m above sea level with semi-arid climate. The Soil physico-chemical properties were analyzed before crop planting (Table 1).

Table 1. Physico- chemical properties of the experimental soil								
properties	EC	pН	TNV	OC	Ν	Р	К	
unit	dS/m	-	%	%	%	%	ppm	Silty
value	1.4	7.2	8.8	0.5	0.05	4.94	2042	Loam
			Source: Soil Fe	ertility Section	, Pakdasht			

Abbreviations: EC = Electrical conductivity, OC= Organic carbon, TNV=Total neutralizing value

Treatments detail and design

The experiment was planned in a factorial complete block design (RCBD) with 12 treatment combination and 4 replicates in greenhouse condition.

The possible treatment combinations were;

T1) 100% nitrogenandphosphate chemicalfertilizers.

T2) 50% nitrogen and 100% phosphate and rootinoculation and irrigation with Nitroxin in transplanting stage.

T3) 100% nitrogenand 50% phosphate and root inoculation and irrigation with Bio-phosphate in transplanting stage

T4) 50% nitrogen and 50% phosphate and root inoculation and irrigation with Nitroxinand Bio-phosphate in transplanting stage.

T5) 50% nitrogen and 50% phosphate, and root inoculation in transplanting stage and irrigation with Nitroxin and Bio-phosphate in transplanting and flowering stages.

T6) Nitroxin and Bio-phosphate alone.

Crop Planting

Stock seeds obtained from Pan American Company were sown on 2012 in Cultivation trays. After seed germination at 2 leaf stage, seedlings was transferred into main pots. The cropswas irrigated at twice per week and then due to excessive heatirrigated daily. All other agronomic practices were kept normal and uniform for all treatments.

Data Collection

Data collection started at the time of opening of 50% florets per inflorescence.Vegetative and reproductive parameters were determined in the following ways: leaf area was measured by Leaf Area Meter. A ruler was used for measuring the height from the ground level to the top-most leaf. Root fresh weight was weighed by digital scale. For measuring of root dry weight, roots was dried by oven at 70^oC for 48 h, then weighed.Percent total nitrogen (N) was determined by Kajeldal method. The content of available phosphorus (P) was determined calorimetrically (1). After flowering, length of inflorescence was measured by ruler. For determining of flowering time, number of days from seed planting time to flowering was recorded. After opening 50% florets, flowers were harvested in the early morning hours and were transported to the laboratory, and were placed in a glass containing distilled water. Life of postharvest of cut flowers from the beginning of harvest until time of loss of market friendly (loss of 50% of the total florets) was calculated. Diameter of the best midsection floretwere measured by using caliper. After full opening flowers, florets number on the main inflorescence was counted and recorded. Flowering stem diameter in the middle third of stem was measured with a caliper.

Statistical analysis

Data analysis was done by using SAS software. The ANOVA test was used to determine significant ($p\leq0.01$ or $p\leq0.05$) treatment effect and Duncan Multiple Range Test to determine significant difference between individual means.

RESULTS AND DISCUSSION

Effect of treatments and cultivar on vegetative traits

Based on the analysis of variance of growth characteristics, the effect of cultivar on the leaf area, phosphorus and nitrogen content and root fresh weight were significant at 1%. Effect of treatments on all traits except leaf area was significant at 1%. Interactions effect between fertilizer treatments and cultivars was not significant in any of the traits. (Table 2).

Mean comparison results showed that maximum values of root fresh weight (7.46gr), root dry weight (1.64gr), phosphorus content (0.785%) and nitrogen content (3.99%) were obtained from T1(100% chemicalfertilizers), while maximum values of plant height (47 cm) and leaf area (3025 cm²) were obtained from T5 (50% nitrogen and 50% phosphate, and root inoculation in transplanting stage and irrigation with Nitroxin and Bio-phosphate in transplanting and flowering stage). Minimum values for all traits were obtained from T6 (Nitroxin and Bio-phosphate alone). Comparison between two cultivars showed that at all traits, the red cultivar is better than the white cultivar (table 3).

An important role of bio-fertilizers is development of plant roots. Because this type of fertilizers contribute to the availability of nitrogen and phosphorus needed for plant growth and development. Also, other findings are based on effect of nitrogen fertilizers on root elongation and compression (12). Results showed that use of biofertilizers on seedling and flowering stages were similar for 50 % reduction in chemical fertilizers and using of 100 % chemical fertilizer on root dry weight, but application of fertilizers there was no favorable results in root transfer and inoculated roots stages.

Table 2. Analysis of eff	fects of different fertilizer treatments(biological and chemical) on thegrowth characteristicsof two
	varietiesof Stockflower

		Mean square	S					
S.O.V	df	Root dry weight (gr)	Root fresh weight (gr)	Plant height (cm)	Leaf area per plant (cm ²)	Phosphorus (%)	Nitrogen (%)	
Cultivar	1	• / • • ۵ ^{ns}	11/07**	۶/۷۵ ^{ns}	1/4.**	•/••1٣**	٠/٧٨**	
Treatment	5	•/٩٨**	1./047**	1.4/99**	γ/ΔΥ ^{ns}	•/•١٣٩**	۶/۰۵**	
Cultivar × treatment	5	•/•14 ^{ns}	.∕۲٧۶ ^{ns}	۲/۱ ^{ns}	۳/۱۲ ^{ns}	•/•••) ^{ns}	•/•Y ^{ns}	
Experimental error	36	•/•۴٧	1/19	17/14	0/98	•/••••	•/•٢	

*, ** = Significant at 5 % and 1%, respectively, NS= Non-significant

Table 3. Effect of different rates of the different fertilizer treatmentson the growth characteristics of two varieties of Stockflower

Factor		Root fresh weight (gr)	Root dry weight (gr)	Plant height (cm)	Leaf area per plant (cm ²)	Phosphorus (%)	Nitrogen (%)
	Т	1 ^{۷/۴۶} а	1/9°a	۴۵/۳۷ab	۲۷۱۲ab	۰/۲۸۵a	۳/۹۹a
Treatment	Т	2 ⁹ /1Ab	1/۲۷b	۴۳/۷۵ab	۲۳۰۰bc	۰/۷۸۳a	1/00C
	Т	3 [≁] / [≁] ab	1/3°a	40/10ab	TFFTab	·/347C	۳/۵۶b
	Т	4 ۵/۴۹bc	\/∙∻bc	۴۱/۳۷b	1.14cd	۰/۴۸d	۲/۴۳с
	Т	5 ^v /۳^a	\/Y∆a	۴۷a	۳۰۲۵a	•/ ⁹ 9b	۳/۴۷b
	Т	6 ۴/۴۸c	۰/۸ ⁹ C	۳۷c	۱۷۶۲d	•/۴۲1e	۱/۶۷d
Cultivar	White	۵/۲۸b	۱/۳۶a	۴۳/۶۶ a	1.41P	۰/۶۰۴b	۲/۸۲b
	Red	۶/۲۶a	1/۳۴a	41/91a	۲۷۲۹a	۰/۶۴۲a	۳/۰ ^v a

In a column, means with the same letters are not significantly different

It seems that the stock root sensitivity is reason for this shift. Studies on wheat (10), thyme (11) and pea (15), showed that use of bio-fertilizers (Azotobacter, Rhizobium) increased root dry weight.

Leaf area per plants is considered as an indicator of photosynthesis and carbon fixation..In similar circumstances, if leaf area is more, photosynthesis becomes greater and metabolic materials are greater available and consequently increases plant growth .In this experiment, even though the test F showed no significant difference between different levels of treatments, but mean comparison by Duncan's test revealed differences between treatments.Based on mean comparison results, in the T5 treatment, there was a greater height and leaf area per plant (Figure 1). Nigella seed inoculation with Azotobacter biofertilizers and Azospirillium also significantly increased plant height, leaf area, maximum dry matter accumulation and crop growth rate compared with control (4).

T1 and T5 and T3 treatments were superior in leaf N content than other treatments, which showed a higher level of access to nitrogen in this plants compared with plants of other treatments. Chemical and biological applications of nitrogen fertilizer was effective in increasing of leaf N content and seed of Rape, So that maximum leaf N concentrations was associated with use of bio-fertilizers and chemical fertilizers simultaneously (19).

T1 and T5 and t2 treatments were superior in leaf P content than other treatments, which showed a higher level of access to nitrogen in these plants compared with plants of other treatments. Previous studies indicated that application of phosphate biofertilizers in stock flowers increased vegetative growth, plant height, seed yield, dry matter, phosphorus and nitrogen (14).

Effects of fertilizer treatments and cultivars on reproductive traits

Reproductive traits analysis of variance showed that cultivar effects were significant in flowering time and number of florets traits at 1%, and in inflorescence diameter, flowering stem diameter and flower longevity was significant at 5%. Effect of fertilizer treatments was significant at 1% level for all reproductive traits and interaction effect was not significant in any of traits (table 4).

Means comparison of different treatments showed that greatest time to flowering (3/140) is related to T6, and maximum number of florets (25/35), inflorescence light (5/18 cm) and inflorescence diameter (53 mm) is related to T5. Flowering stem diameter was similar in treatments T2, T3 and T4 treatments and its was highest in this treatments. Maximum vase life (5/11) is related to T1. Comparison between two cultivars showed that red cultivar was better than white in all traits except number of florets and stem diameter (table 5).

On the one hand, increase in rate of plant growth causes bloom earlier than expected. On the other hand, in adequate nutrition situation vegetative and reproductive growth of plant will be completed soon (6). In this experiment, plants in T1, T2, T3 and T5 treatments were up earlier flowered than other treatments. It seems that, more availability of nutrients from using 100% chemical fertilizer and 50% bio or chemical fertilizers effective.

Number of florets per inflorescence is one of the important characteristics of Stock that is very effective in performance, marketability and beauty. The results showed that plants in T5, T1 and T2 treatments had a greater number of florets than other plants. In order to investigate effect of phosphorus and urea fertilizer and biofertilizer in medicinal plants Deracocephalum moldavica L. largest flower was obtained in treatment of triple superphosphate and urea (5). Use of biological fertilizers containing phosphate solubilizing bacteria, Azotobacter and mycorrhiza increased length of tuberose flower, number of flowering stem, number of florets and fresh weight of flowers and bulbs (13).

Table 4. Analysis of variance of effects of different fertilizer treatments on reproductive traits of two varieties

		Mean square	es				
\$.0.V	df	Flowering time	Inflorescence length (cm)	Inflorescence diameter (mm)	Number of florets	Flowering stem diameter (mm)	Vace life
Cultivar	١	۵۸۱/۰۲**	۰/۰ ^{۸۳} ns	۱۰۲/۰۸*	1.0/.1**	•/••1٣*	۶/۰۲*
Treatment	۵	**/۱۷**	Y4/YT**	۱ • ۶/ • ۸**	117/21**	•/••1٣**	17/47**
Cultivar × treatment	۵	۱/۲۷ns	۲/۱۳ns	۵/۶∧ns	۴/۸۲ns	۰/۰۰۰۰ ^۴ ns	۰/۵۲ns
Experimental error	36	۲/۶۰	۲/۵۹	27/17	٨/٢٧	•/•••٢	1/11

*, ** = Significant at 5 % and 1%, respectively, NS= Non-significant

Factor		Flowering time	Number of florets	Inflorescence length (cm)	Inflorescence diameter (mm)	Flowering stem diameter (mm)	Vace life
	T1	۱۳۶/۳ bc	۳۳/۵a	19/0b	۵۲ab	1/1″b	11/0a
Treatment	T2	186/8pc	۳۳/۵a	12/AYb	۴۹/۵ab	1/10a	1./17bc
	Т3	۱۳۷/۲bc	۲۸/۷۵b	19/97b	۵۰/۵ab	1/10a	۹/۳۷bc
	Τ4	۱۳۸b	۲۷/۳۷b	۱۵b	۴۷/۲۵bc	1/10a	9/17C
	Т5	180/0C	۳۵/۲۵a	14/0a	۵۳a	1/1″b	۱۰/۳۲b
	Т6	14./ra	۲۶b	17/10C	4°C	1/1″b	۲/۱۲d
	White	177/Ab	۳۲/۲a	١ŕa	۴۷/۷۵b	1/1fa	۹/۲۵b
Cultivar	Red	14.//a	19/10b	12/91a	۵۰/۶۶a	1/1″b	۹/۹۵a

In a column, means with the same letters are not significantly different

The results showed that diameter of inflorescence is higher than other treatments in T5, T3, T2 and T1 treatments. In another experiment, maximum diameter of inflorescence obtained with irrigation and seed inoculation with Nytroxin, and smallest diameter obtained in treatment of non-use Nytroxin (20).

Inflorescence is an important trait in double flower Stock as ornamental and increase the quantity and quality of seed production of single flower Stock. Also, This trait is effective in increasing of biomass. Inflorescence trait was improved in T5 and T1, T2 and T3 and T4 treatments, respectively. In another study application of biological phosphate on Stock increased inflorescence length, significantly (8).

Results in this study showed that T2 and T3 and T4 treatments have created largest flowering stem diameter and other treatments were similar. Previous studies indicated that interaction effect of vermicompost

(2/1 kg/m2) and Nitroxin (root seminal) in Stock flowers showed a significant increase in flowering stem diameter (20).

Effective factors in life of cut flower in Stock flower including light, temperature, nutrition and fertilization, irrigation and moisture, pest and disease control, developmental stage, and harvest time, harvesting techniques, breathing and loss of water. The results showed that maximum life of cut flowers was related to T1 and other treatments were similar. In one experiment carried out in order to investigate effects of different forms of application of phosphate fertilizers on yield and quality in Stock flowers and results showed that biological phosphate fertilizer (100 g in 2000 ml of water) treated with liquid fertilizer (50 mg, 200 mg in 50 ml water) and granular fertilizer (200 mg) resulted to highest postharvest life (20).

CONCLUSION

Superiority of T1 treatment (application of 100% nitrogen and phosphate chemical fertilizers) and T5(application of 50 % chemical fertilizers and then utilization of bio fertilizers in two stages) indicate this fact that any failure in important characteristics of Stock is achieved by 50% reducing nitrogen and phosphorus fertilizers and application of biological fertilizer, can supply plant fertilizer requirements.

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