Journal Home page : www.jeb.co.in ★ E-mail : editor@jeb.co.in

JEB
Journal of Environmental Biology

ISSN: 0254-8704
CODEN: JEBIDP

The effect of phosphate bio-fertilizer (Barvar-2) on the growth of marigold

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Publication Info

Paper received:
21 December 2012

Revised received:
21 May 2013

Re-Revised received:
15 August 2013

Accepted:
17 September 2013

Abstract

The present study was conducted to study the individual and combined effect of bio-fertilizer (Barvar-2) and chemical phosphate fertilizer on the floral quality of marigold (*Tagetes erecta* L.). A factorial experiment was carried out which consisted of two factors: i) inoculation of seed, root and seed + root with bio-fertilizer (Barvar-2) and control; application of chemical phosphorus at 100 mg l^{-1}, 200 mg l^{-1}, 300 mg l^{-1} and 400 mg l^{-1} levels. In this study, flowering time, display life, fresh and dry weight of flower, available soil phosphorus, shoot phosphorus and carotenoid content were evaluated. Results showed that the combined effect of bio- and chemical fertilizer was insignificant ($p<1$ and 5%) for most of the characteristics studied except for shoot phosphorus and carotenoid content in petals. The lowest time to flowering (64.67 days) was obtained in seeds and transplant roots inoculation to bio-fertilizer × 400 mg l^{-1} P. Maximum display life (25.35), fresh weight (16.20 g), carotenoid content (3.903 mg g^{-1} d. wt.) and concentration of P in shoots (0.352%) were observed in transplant roots inoculation to bio-fertilizer × 400 mg l^{-1} P.

Key words

Phosphorus, Phosphorus bio-fertilizer, *Tagetes erecta* L., Vegetative and flowering traits

Introduction

Tagetes erecta commonly known as marigold is an annual herbaceous plant of ornamental and medicinal importance. It is one of the commercially exploited flowers of Asteraceae family and often used as cut flowers, garden displays, bouquets and in landscape gardening (Atiyeh *et al*., 2002). It has attracted attention of floriculturists due to short duration production of marketable flowers with wide range of attractive colours, shape and size. Since the growth and production of *T. erecta* is influenced by chemical fertilizers, their nutrient requirement is fulfilled especially by supplying phosphate fertilizers. However, indiscriminate and long term use of chemical fertilizers has not only led to imbalance of nutrients in soil resulting in degradation of soil structure but has also affected the growth and production of flowers (Singh *et al*., 2008; Mohammadi Torkashvand, 2009). Nowadays, a lot of emphasis is being paid on the use of bio-fertilizer to increase the production of crops. Bio-fertilizer usually consists of live or latent cells of micro-organisms which include biological nitrogen fixers, P-solubilizing, mineralization of nitrogen and transformation of several elements into available forms. VAM fungi, *Azotobacter, Azospirillum* and phosphate solubilizing bacteria are commonly applied bio-fertilizers in horticultural crops.

Bio-fertilizers are not only eco-friendly and cost effective but also increase phosphorus uptake, promotes growth and yield of plants by supplying nutrients in available form, provides resistance against pests and diseases and strengthens soil structure (Sharma, 2002). The natural uptake of phosphorus, present in chemical fertilizers, by plants is very low and most of the phosphorus remains fixed in calcareous soil. However, this problem can be overcome by using phosphorus solubilizing bacteria (PSB). PSBs' have the ability to convert unavailable form

of P into available form, thereby increasing P uptake and yield. A lot of work has been carried out on the effect of different organic manures individually or in combination with chemical fertilizers on the growth and yield characters of chrysanthemum, China aster, tuberose, marigold and several other ornamental plants (Meshram *et al*., 2008; Singh *et al*., 2003; Chandrikapure *et al*., 1999; Moghadam and Shoor, 2013; Mohanty *et al*., 2013).

Ratti *et al*. (2001) investigated some varieties of phosphate solubilizing bacteria on the yield of lemon grass and concluded that the plant height and biomass increased as compared to control. Gupta *et al*. (2002) found that inoculation of mint root by mycorrhiza fungi considerably increased the height and yield of plant. In another research, Kapoor *et al*. (2002, 2004) reported that the mycorrhizal inoculation of root significantly improved the quantity and quality of essential oil in dill and carom plants. Further, AM inoculation of plants along with phosphorus fertilization significantly enhanced growth, P-uptake and essential oil content of fennel plants. In light of above, the present study aimed to investigate the individual and combined effect of phosphate biological fertilizer (Barvar-2) and phosphate chemical fertilizer on the growth and yield characters of *T. erecta*.

Materials and Methods

The present study was conducted during the year 2010/2011 experimental season (April until August) at a greenhouse (photoperiod of 16 hr per day), located in Rasht city, Guilan province, Iran. Investigations were carried out on *Tagetes erecta* L. var. Tiashan, commonly known as marigold. Experiment was conducted in factorial arrangement based on randomized completely block design with two factors consisting; methods of bio-fertilizer application and different levels of Barvar-2. Barvar-2 is a phosphate bio-fertilizer and is environmentally safe that is applied increase the yield in crop and fruit trees. The first factor were to; seeds inoculation to bio-fertilizer (M_1), transplant roots inoculation to bio-fertilizer (M_2), seeds and transplant roots inoculation to bio-fertilizer (M_3) and control (without bio-fertilizer). The phosphate bio-fertilizer was purchased from Mabco Company. The second factor included treatment of (from Crystalon source with 12-12-12 ratio of NPK) chemical phosphorus at four levels 100 mg l^{-1} P (P_1); 200 mg l^{-1} P (P_2); 300 mg l^{-1} P (P_3) and 400 mg l^{-1} P (P_4), respectively.

The experiments were carried out at a temperature of 20 ± 2°C and 70% relative humidity. The seeds were planted in a medium containing coco peat and sand in 1:1 ratio (Table 1). After 45 days of planting, when transplanting had four leaves, transplants were transferred to the larger pots contain cocopeat, sand and mold (Table 1). Three months after planting, the first data about flower and shoot was recorded and during one month, all data including time of flowering, fresh weight of flower, and the percentage of flower dry weight were taken. Phosphorus concentration of medium and shoot plant; and the amount of carotenoid pigment were also measured at this experiment. Time of flowering base on counting the days from sowing seed to appearing flower was computed. The medium was extracted by AB-DTPA method and phosphorus was measures by spectroscopy method in extraction. Petals were extracted by acetone and carotenoides were measured by following the method of Mazumdar and Majmuder (2003).

MSTATC software was used for variance analysis of data by Least Significant Difference (LSD) test.

Results and Discussion

The analysis of variance showed that the effect of bio-fertilizer (Barvar-2) and chemical phosphorus fertilizer was significant at 1% probability level on the growth parameters of *Tagetes erecta*. Result of ANOVA showed that application of bio-fertilizer and chemical fertilizer significantly influenced flowering time in *T. erecta* while the effect of combined treatment on flowering time was insignificant. The mean comparison of data of combined treatment showed that lowest flowering time (64.53) was obtained in M_2P_4 treatment, lowest flowering time was found in B_2 treatment and longest in M_1 treatment, whereas application of chemical P fertilizer, the lowest (66.65) and longest (75.42) flowering time was observed in P_4 and P_1 treatment, respectively (Table 3)

Application of bio-fertilizer showed significant ($p<1\%$) effect on display life of *T. erecta* while insignificant effect on the same was observed on the application of chemical fertilizer and combined treatment of bio-and chemical fertilizer (Table 2). Maximum display life (25.35) in *T. erecta* was seen in M_2P_4 treatment and minimum (18.68) in M_1P_1 treatment, respectively. Among bio-fertilizer application, maximum display life (23.57) was found in M_2 treatment and minimum (20.62) in M_4 treatment, respectively. While P_4 and P_1 treatment of chemical phosphorus

Table 1 : The results of some physicochemical properties of the used media for plant seeds and plant roots

Properties	Medium (cocopeat plus sand) for plant seeds	Medium (cocopeat, sand and mold) for plant roots
pH	7.02	6.7
Available K (mg kg^{-1})	51.20	340.00
Available P (mg kg^{-1})	25.10	59.00
Total N (%)	2.70	4.30
Organic carbon (%)	1.00	7.80

Table 2 : Analysis of variance (ANOVA) of the effect of different treatments on traits

Source of variance	df	Mean square						
		Time of flowering	Display life	Flower fresh weight	Flower dry matter	Total shoot phosphorus	Total medium phosphorus	Carotenoid
M (Method)	3	76.976**	29.896**	68.174**	13.510ns	0.120*	4435.44**	0.807ns
P (Phosphorus)	3	174.312**	4.049ns	160.525**	5.238ns	0.220**	372.917ns	4.655**
M × P	9	3.152ns	6.486ns	2.719ns	8.125ns	0.230**	667.361ns	1.791**
Error	32	3.971	4.981	3.635	6.636	0.003	348.958	0.456
Total	48	258.411	54.412	235.053	33.509	0.573	5824.706	7.709
CV (%)	-	6.23	11.92	47.48	17.45	2.770	13.45	66.44

* and **: significant difference at 5% and 1% probability level, respectively, ns: not significant

Table 3 : Mean comparisons of the effect of different treatments on traits

Treatments	Time of flowering(day)	Display life(day)	Flower fresh weight(g)	Flower dry matter(%)	Total shoot phosphorus(%)	Total medium phosphorus(%)	Carotenoid (mg g^{-1} M.W.)
M1	73.07^{a}	20.44^{b}	6.69^{c}	17.10^{a}	0.343^{a}	0.187^{bc}	1.830^{a}
M2	68.03^{b}	23.57^{a}	9.83^{b}	15.92^{a}	0.344^{a}	0.212^{a}	1.343^{a}
M3	68.93^{b}	22.85^{ab}	11.73^{a}	14.51^{a}	0.340^{ab}	0.202^{ab}	1.545^{a}
M4	72.55^{a}	20.62^{b}	7.09^{c}	15.74^{a}	0.337^{b}	0.168^{bc}	1.242^{a}
P1	75.42^{a}	21.20^{a}	4.70^{d}	16.80^{a}	0.339^{b}	0.190^{a}	1.269^{b}
P2	71.77^{a}	21.67^{a}	7.48^{c}	15.36^{a}	0.341^{ab}	0.196^{a}	1.386^{b}
P3	68.75^{b}	22.05^{a}	9.83^{b}	15.51^{a}	0.337^{b}	0.198^{a}	0.928^{c}
P4	66.65^{c}	22.57^{a}	13.33^{a}	15.59^{a}	0.347^{a}	0.186^{a}	2.378^{a}
M1P1	79.00^{a}	18.68^{a}	1.93^{a}	19.38^{a}	0.344^{bcd}	0.180^{a}	2.073^{bc}
M1P2	73.80^{a}	20.80^{a}	4.87^{a}	17.38^{a}	0.332^{de}	0.180^{a}	1.640^{bcd}
M1P3	70.33^{a}	22.67^{a}	7.92^{a}	15.77^{a}	0.336^{cde}	0.190^{a}	0.973^{cd}
M1P4	69.13^{a}	19.60^{a}	12.03^{a}	15.87^{a}	0.352^{a}	0.200^{a}	2.633^{b}
M2P1	70.73^{a}	24.40^{a}	4.88^{a}	17.09^{a}	0.336^{cde}	0.220^{a}	1.020^{cd}
M2P2	69.73^{a}	21.80^{a}	8.62^{a}	15.62^{a}	0.348^{abc}	0.235^{a}	1.990^{bc}
M2P3	67.13^{a}	22.73^{a}	11.91^{a}	15.33^{a}	0.348^{abc}	0.210^{a}	0.763^{cd}
M2P4	64.53^{a}	25.35^{a}	13.91^{a}	15.64^{a}	0.344^{bcd}	0.185^{a}	1.600^{bcd}
M3P1	73.93^{a}	21.47^{a}	9.15^{a}	12.33^{a}	3.40^{bcde}	0.200^{a}	0.540^{d}
M3P2	70.45^{a}	22.40^{a}	9.87^{a}	15.10^{a}	0.340^{bcde}	0.205^{a}	0.773^{cd}
M3P3	66.65^{a}	23.22^{a}	11.68^{a}	15.33^{a}	0.328^{e}	0.220^{a}	0.963^{cd}
M3P4	64.67^{a}	24.33^{a}	16.22^{a}	15.28^{a}	0.352^{a}	0.185^{a}	3.903^{a}
M4P1	78.00^{a}	20.25^{a}	2.82^{a}	18.39^{a}	0.336^{cde}	0.160^{a}	1.443^{bcd}
M4P2	73.07^{a}	21.67^{a}	6.56^{a}	13.35^{a}	0.344^{bcd}	0.165^{a}	1.140^{cd}
M4P3	70.87^{a}	19.57^{a}	7.82^{a}	15.63^{a}	0.336^{cde}	0.173^{a}	1.010^{cd}
M4P4	68.27^{a}	21.00^{a}	11.14^{a}	15.57^{a}	0.332^{de}	0.175^{a}	1.373^{bcd}

*In each column, means with the similar letters are not significantly different at 5% level of probability using LSD test

fertilizer produced maximum (22.57) and minimum (21.20) display life in *T. erecta*, respectively.

The analysis of variance showed that fresh weight of *T. erecta* flowers were significantly affected by the application of bio-fertilizer and chemical phosphorus fertilize. Combined application of fertilizers showed insignificant effect on the fresh weight of flowers (Table 2). The mean comparison of data among different treatments showed that maximum (16.2 g) fresh weight of flowers was noted in M_3P_4 treatment and minimum (1.93 g) in M_1P_1 treatment, respectively. Among different treatments of bio-fertilizer and chemical phosphorus application, maximum fresh weight (11.73 g) was observed in M_3 treatment and minimum (6.69) in M_1 treatment while maximum and minimum fresh weight of flowers was found in P_4 and P_3 treatment of chemical fertilizer, respectively. Analysis of variance on dry weight data of flowers showed that all three types of treatments showed insignificant effect on the dry weight of *T. erecta* flowers. Results revealed that maximum dry weight of flowers was observed in M_1P_1treatment and minimum (12.33%) in M_3P_1treatment, respectively.

Application of different bio-fertilizer inoculations produced insignificant effect on carotenoid content in petals of *T. erecta*. While application of chemical phosphorus and combined

application of bio- and chemical fertilizer produced significant effect on the carotenoid content. The mean comparison of data showed that highest (3.903 mg g^{-1} d. wt.) carotenoid content was observed in petals of *T. erecta* treated with M_3P_4 while lowest (0.54 mg g^{-1} d. wt.) on application of B_3P_1 treatment, respectively. Application of different treatments of bio-fertilizer did not produce any significant difference on the carotenoid content; however among different treatments of chemical fertilizers, highest (2.37 mg g^{-1} d. wt.) carotenoid content was found in P_4 treatment and lowest (0.92 mg g^{-1} d. wt.) in P_3 treatment, respectively.

Table 2 shows that application of bio-fertilizer, chemical fertilizer and combined application of both significantly affected the phosphorus level in shoots of *T. erecta*. Mean comparison of data revealed that highest (0.352%) concentration of P in shoots was observed in M_1P_4 and M_3P_4 treatment while lowest concentration (0.328%) was found in M_3P_3 treatment, respectively. Application of bio-fertilizer significantly increased P content in shoots over control, whereas among different concentration of chemical fertilizer, maximum (0.347%) shoot P content was observed in P_4 treatment and lowest (0.337%) in P_3 treatment, respectively.

Analysis of variance showed that the effect of different treatments of bio-fertilizer on the content of phosphorus in medium was significant while different treatment level of chemical fertilizer and combined application of bio-and chemical fertilizer produced insignificant effect on phosphorus content in the medium (Table 2). Combined application of bio-and chemical fertilizer showed highest (0.235 %) phosphorus content in medium with M_2P_2 treatment and lowest (0.16%) with M_4P_1 treatment respectively. Different level of chemical phosphate treatment did not produce any significant effect on the total phosphorus content in the medium. However, different treatments of bio-fertilizer produced significant different showing highest (0.212%) phosphorus content in the medium with M_2 treatment and lowest (0.168%) with M_4 treatment, respectively.

Results of the present study showed that the floral characters of *T. erecta* were significantly influenced by the integrated application of Barvar-2, bio-fertilizer, and chemical phosphate fertilizer. Application of seed and root inoculated biofertilizer with 400 mg l^{-1} chemical phosphate fertilizer produced minimum flowering time and maximum fresh weight, dry weight and carotenoid content in *T. eracta* as compared to other treatments. Increased growth of *T. erecta* inoculated with bio-fertilizer might be due to addition of nitrogen through symbiosis and increased production of growth hormones like NAA, GA and cytokinins (Bashan, 2004). Several reports are available on the combined application of bio-fertilizer with chemical fertilizer on the enhanced growth and yield of ornamental flowers. Combined application of bio-fertilizers like VAM, *Azospirillum, Azotobacter*, phosphorus solubilizing bacterium along with chemical fertilizer have been found to increase plant height, minimum number of days to first flowering, maximum bud and flower size, maximum vase life and maximum cost benefit ratio and vegetative growth in ornamental flowers like rose (Singh *et al*., 2003), carnation (Gupta *et al.,* 2004), gladiolus corm (Dubey and Mishra, 2006), china aster (Chaitra and Patil, 2007), marigold and gaillardia (Deshmukh *et al*., 2008). Recently, Moghadam and Shoor (2013) and Mohanty *et al*. (2013) reported increase in yield attributes of *Petunia hybrid* and *Tagetes erecta*. The early flowering may be due to the presence of bio-fertilizers inoculated with *Azospirillum* and PSB which resulted in flower initiation and more flower duration. This may be due to easy uptake of nutrients and simultaneous transport of growth promoting substances like cytokinins to the axillary buds resulting in breakage of apical dominance. Combined application of *Azospirillum* + phosphate solubilizing + bacterium + vermicompost + 25% NPK significantly enhanced the growth, yield, and quality of petunia flowers. Similarly, combined application of poultryfarm + bio-fertilizer + vermicompost + 50% RDF increased floral characters in *T. erecta*. The increase in flower size, weight, number and duration might be due to the effect of balance nutrition supplied through combined application. Combined application of bio-and chemical fertilizer might have attributed to the translocation of nutrients from soil and enhanced supply of macro and micronutrients during entire growing season and microbial decomposition which might have favored the stimulation and production of axillary buds resulting in formation of more number of branches.

Yousefi *et al*. (2010) studied the impact of phosphate fertilizer of Barvar-2 on the yield of maize and concluded that the maize yield was increased by 50 kg ha^{-1}. Fallahi *et al*. (2009) reported that the treatment of nitroxin and PSB considerably influenced the fresh and dry weight of chamomile flowers. The results of present study are consistent with earlier studies of Fallahi *et al*. (2009), Raipoor and Asgharzadeh (2007) and Sanchez Govin *et al*. (2005). The increase in phosphorus uptake in roots inoculated with bio-fertilizer is due to the increase in P availability and the improvement of plant uptake capacity. Thus, application of biological phosphate fertilizers containing PSB and mycorrhiza fungi not only decreases the uptake of chemical phosphate fertilizer, but also increases plant growth. Khalaj (2009) investigated the effect of bio-fertilizer, Barvar-2 on phosphorus uptake in *Polianthes tuberosa* flower and concluded that the best results were obtained by applying bio- and chemical fertilizer in 50:50 ratios. Studies of El-Ghandour *et al*. (2009) showed that nutrient uptake of *Majorana hortensis* plants positively responded to bacterial inoculation and the concerned organic phosphorus sources. Many researchers have explained the role of phosphate solubilizing bacteria in enhancing availability of phosphorus in the soil through secretion of phosphatase enzyme which leads to transfer organic phosphorus to available form (Melero *et al*., 2006; El-Ghandour *et al*., 2009)

and consequently, enhancing phosphorus absorption and accumulation in plant tissues.

In conclusion, the highest time to flowering (79.00 day) and the lowest display life (18.68 day) were obtained in control plants. Minimum time to flowering (64.53 day) and the maximum display life (25.35 day) were obtained in plants grown under transplant roots inoculation with bio-fertilizer in 400 mg l^{-1} phosphorus. Therefore, to improve the quality of marigold, 400 mg l^{-1} P and root inoculation to bio-fertilizer can be used. Pay attention to increase in marigold yield and flower quality, the use of Barvar-2 fertilizer as a biological phosphate fertilizer can be saved chemical fertilizer.

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