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Study the effect of chemical and bio-fertilizers (Azotobacter and Phosphate Barvar-2) on quantity characteristics of Marigold (Calendula officinalis L.)

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Abstract

To study the effect of chemical and bio-fertilizers (Azotobacter and Phosphate Barvar-2) on quantity characteristics of Marigold (Calendula officinalis L.) an experiment was conducted in the Jiroft. Field experiment was arranged base on randomized complete block design with three replications. Treatments were involved: T1: nochemical, biological as control, T2: 100% NPK, T3: Azotobacter, T4: Azotobacter + 50% NPK, T5: Phosphate Barvar-2, T6: Phosphate Barvar-2+ 50% NPK, T7: Azotobacter Phosphate Barvar-2, T8: Azotobacter Phosphate Barvar-2 + 50% NPK and T9: Azotobacter Phosphate Barvar-2 + 100% NPK. result showed that the signification effect of chemical and bio-fertilizers on height, flowering stem length, stem diameter, number of branches, number of leaf, Fresh and dry weight plant, Fresh and dry weight flower and harvest index. So that most of these traits were obtained from treatment (T8): Azotobacter + fertile phosphate 2 + 50% NPK. Biochemical and chemical fertilizers had a significant effect on the percentage and yield of marigold essential oil. The highest amount of essential oil was obtained from Azotobacter + fertile phosphate 2 treatment. Also, the application of biochemical fertilizers had a significant effect on the number of days to flowering and the lowest time was related to T8 treatment. In general, the results showed that Applying bio-fertilizers (Azotobacter and Phosphate Barvar-2) benefited on growth and generative and yield Marigold. The use of bacteria not only increased yield, quality and early yield, but also reduced the use of chemical fertilizers.

Keywords: Azotobacter, Phosphate Barvar, bio-fertilizers.

Introduction

In recent years, following the crisis of environmental pollution and especially the pollution of water and soil resources that have contaminated human food sources and threatened the health of human societies, extensive efforts It has been started in order to find suitable solutions for improving the quality of soil, agricultural products and removing pollutants. The use of biological fertilizers, especially growth-promoting bacteria, is the most important strategy in the integrated management of plant nutrition for a sustainable agricultural system (Sharma, 2004). Therefore, biofertilizers are economically affordable and acceptable from an environmental point of view. Also, in order to avoid negative pressures on the environment and to improve the development plans, it is necessary to change the management of supplying fertilizer needs of plants. Among the bacteria that have been known in the last two decades, Azotobacter has attracted more attention due to its wide geographical distribution and the wide range of host plants, and to the biological fixation of nitrogen through free-associating bacteria such as Azotobacter in the ecosystem. Special attention has been paid to agriculture (Paliyath et al., 2008). Azotobacter is able to fix molecular nitrogen non-symbiotically. This bacterium can synthesize all kinds of amino acids, vitamins and plant growth stimulating hormones and all kinds of exopolysaccharides, the number of Azotobacter in the soil is usually less than 104 (Khosravi, 2013). The use of biological fertilizers with different methods to increase phosphorus sources has optimized the use of chemical fertilizers for the production of crops (Salimpur et al., 2010). Phosphate-dissolving bacteria are Bacillus lentus and Pseudomonas putida, which dissolve insoluble phosphorus compounds by using two mechanisms of organic acid secretion and acid phosphatase, respectively (Alijani, 1390). The research results indicate that the use of nitrogen-fixing and phosphate-dissolving bacteria in marjoram medicinal plant increased the yield and percentage of essential oil (Gharib et al., 2008). Phosphate Fertilizer 2 increased photosynthesis by providing the required phosphorus and consequently the yield components and finally seed yield (Khalilian 2015 and Ojaklou, 2016). Darzi (2006) reported improved yield in fennel following the application of phosphate biofertilizer. Nikzad et al. (2013) also reported an increase in the vegetative indices of basil due to the use of Fertilizer Phosphate-2. The studies conducted by Beykhormizi et al. (2009) showed that the use of 100 grams of fertile phosphate fertilizer 2 is equivalent to 100 grams of phosphorus chemical fertilizer. Also, this fertilizer increased the durability of the leaf surface and, as a result, higher photosynthesis of the plant. Afrasiabi et al. (2013) reported that the efficiency of phosphorus fertilizers in the soil due to stabilization is low and around 15-20%. Therefore, phosphate-dissolving bacteria in the rhizosphere environment can play an important role in making the phosphorus available in the soil usable. The results of a research also showed a significant improvement in dry matter due to the use of phosphate-dissolving bacteria in a medicinal plant from the Ferfion family compared to the control treatment (Annamalai et al., 2004). However, it has been reported that there is no significant difference in terms of biomass between the treatments inoculated with phosphate-dissolving bacteria and the control (Gewaily et al., 2006). The marigold plant is a plant from the chicory family (Asteracea), with the scientific name (Calendula officinalis), the marigold flower contains small amounts of volatile essential oil, saponin, resin, organic acids, a substance called calendolin, gum, glazing materials, albumin. And it is a coloring matter in dry petals and inulin (in the root), salicylic acid, lauric acid, palmitic acid and cholesterol (Falahtgar, 2012).

The purpose of cultivating this plant is to produce medicine and effective substances found in flowers and especially in petals (Martin, 1999). Considering the necessity of plant nutrition management in order to increase and sustain production and the importance of biofertilizers in sustainable agriculture and environmental preservation and the necessity of reducing the excessive use of chemical fertilizers and optimizing their use in the agricultural ecosystems of the country, research present with the aim of "examining the effects of chemical and biological fertilizers on the quantitative and qualitative characteristics of the medicinal plant marigold" in order to find a suitable combination of chemical and biological fertilizers in order to reduce the consumption and increase the efficiency of the use of chemical fertilizers to achieve sustainable development in the plant production sector Medicine was done in the country.

Materials and methods

This research was carried out in the agricultural year of 1399-1400 in the research farm of Jiroft Islamic Azad University located in Khizrabad. The experiment was conducted as a randomized complete block design with 3 replications. The experimental agents included: chemical fertilizers and biofertilizers of Aztobacter and Fertilizer 2 biophosphate as follows. (T1): No fertilizer use, (T2): 100% NPK, (T3): Azotobacter, (T4): Azotobacter + 50% NPK, (T5): Fertilizer phosphate 2, (T6): Fertilizer phosphate 2 + 50% NPK , (T7): Azotobacter + fertilizing phosphate 2, (T8): Azotobacter + fertilizing phosphate 2 + 50% NPK, (T9): Azotobacter + fertilizing phosphate 2 + 100% NPK were considered. In the beginning of March 2019, the intended land was prepared and chemical fertilizer treatments were applied in such a way that nitrogen, phosphorus and potassium were added to the soil of the plots in a ratio of 1-1-2 three days before planting the seeds and at a depth of 20 A centimeter was mixed with the soil. Marigold seeds were inoculated with biological fertilizers. Biofertilizers, which were applied at the rate of two liters per hectare, were cultivated in heaps. The distance between planting seeds on the lines was 30 cm and between the lines was 30 cm in each plot. The chemical fertilizers used were urea (46% nitrogen), triple superphosphate (23% P2O5) and potassium sulfate (48% K2O). The biofertilizer was Aztobacter with the brand name Azto Fervor-1. The type of microorganism in this fertilizer was Pantoea agglomrans with a population of 109 live cells per gram. Fertilizer-2 phosphate biofertilizer had Pseudomonas putida and Pantoea agglomerans microorganisms with a population of 109 live cells per gram. The biological fertilizers used in this research were obtained from Zistavar-e- Sabz Company. The application of fertilizers included placing the seeds in a bacterial solution for 30 minutes and immediately after the seeds dried in the shade and away from direct sunlight, planting was done. The time of the appearance of the first flower in each treatment was recorded, and at the end of the growth stage, the number of sub-branches, the height of the plant, the diameter of the plant, the number of leaves, the fresh and dry weight of the plant, the fresh and dry weight of the flowers, the biological yield, the harvest index was measured. became. The measurement of essential oil was done using a Cloninger device. The yield of essential oil in kg/ha was obtained by multiplying the dry weight of flowers by the percentage of essential oil. The data obtained from this experiment were statistically analyzed using Excel and SAS software, and the averages were compared using Duncan's multi-range test at a probability level of 5%.

Results and Discussion

Plant height: During this research, the final height of the marigold medicinal plant increased as a result of the combined use of biological and chemical fertilizers, and the highest amount was obtained from the treatment of Azotobacter + Fertilizer Phosphate 2 and 50% of the recommended amount of NPK fertilizer required in marigold cultivation. (Tables 1-3). It seems that chemical fertilizers, especially nitrogen, due to their role in the production and export of cytokinin hormone from roots to aerial organs, increase the speed of cell division and plant growth and height, and the intensive use of chemical fertilizers continues. This reduction is the result of reducing the biological activities of the soil, decreasing the physical characteristics of the soil and the absence of micronutrients in NPK fertilizers (Adediran et al., 2004).

Flowering stem length:

In this study, the average height of flowering stem increased as a result of the combined use of biological and chemical fertilizers, and the highest amount was obtained from the treatment of Azotobacter + fertile phosphate 2 and 50% of the recommended amount of NPK fertilizer required in marigold cultivation. (Table 1-3). With the continued use of chemical fertilizers, the amount of this trait decreased (Micherlich's law of diminishing returns (1909) and Macy's law of critical percentages (1936)). The number of sub-branches: in this experiment, the number of sub-branches of the marigold medicinal plant increased due to the combined use of biological and chemical fertilizers, and the highest amount with an average of 14 branches per plant was from the treatment of azotobacter + 2 fertilizing phosphates and 50% of the recommended amount of fertilizer. The required NPK was obtained in marigold cultivation and the amount of this trait was reduced by increasing the amount of complete chemical fertilizers (Tables 1-3). In this regard, it can be said that Mace's law of critical percentages, which states that in the range of luxury consumption, increasing units of a food item has little effect on performance, but the percentage of the food item is true, because it is sufficient There is no food substance in the tissue that causes a lot of plant growth. Plant diameter: The plant diameter of the marigold plant in this experiment was at statistical level in all treatments, and the Aztobacter treatment was at level b, and the control had the lowest plant diameter (Table 1-3). Optimal provision of water and nutrients in the form of chemical fertilizers and biofertilizers and their combined use made the plant thicker. Increasing the plant's access to nutrients with the combined use of biological and chemical fertilizers and increasing growth and photosynthesis due to the increase in the surface of the plant's leaves is one of the factors of increasing the yield in the combined nutrition system treatments. Nitrogen-fixing and phosphorus-dissolving bacteria provide more nitrogen and phosphorus to the plant in the presence of chemical nitrogen and phosphorus (Suneja et al., 1994). The number of leaves: During this research, the number of leaves of the medicinal marigold plant showed an increase due to the combined use of biological and chemical fertilizers, and the highest amount was obtained from the treatment of azotobacter + 2 fertilizing phosphates and 50% of the recommended amount of NPK fertilizer required in the cultivation of marigolds. (Table 1-3), the combined use of biological and chemical fertilizers increases the plant's access to nutrients, and their absorption by the plant, and as a result, increases the number of leaves and more photosynthesis. Plant growth promoting bacteria (PGPR) improve plant growth in various ways, including nitrogen fixation, synthesis and production of iron complexing siderophores, production of plant hormones, production of antibiotics and fungicidal compounds, and They are useful by solubilizing mineral materials (Herman et al., 2008) and in the presence of biofertilizers, nitrogen absorption from chemical fertilizers increases (Roesty et al., 2006).

Table 3-1 Comparison of the averages of stem height, length of flowering stem, number of sub-branches, diameter of plant and number of leaves of marigold under the influence of biological and chemical fertilizers

Number of leaves	Bush diameter (mm)	Number of sub-ranches	Flowering stem length (cm)	Stem height (cm)	treatment
90/33 f	6/2 c	9/33 e	9/33 f	40/40 d	T1
121/66 c	10/32 a	11/66 d	25/66 d	56/08 c	T2
105/66 e	8/3 b	11d	21/33 e	50/44 c	T3
118/33 cd	10/46 a	12/66 bc	30/66 b	68/88 b	T4
104 e	10/07 a	11 d	22 e	54/23 c	T5
124 c	10/18 a	12bcd	28 c	66/97 b	T6
114 d	10/18 a	12 bcd	24/66 d	65/08b	T7
171/33 a	10/52 a	14a	32/66 a	87/49 a	T8
147/66 b	10/46 a	13 b	31/66 ab	80/75 a	T9

Numbers with the same letters in each column have no significant difference based on Duncan's multi-range test at the 5% probability level. T1: No fertilizer use, T2: 100% NPK, T3: Azotobacter, T4: Azotobacter + 50% NPK, T5: Fertilizer phosphate-2, T6: Fertilizer phosphate-2 + 50% NPK, T7: Azotobacter + biophosphate, T8: Azotobacter + Fertilizer Phosphate-2 + 50% NPK, T9: Azotobacter + Fertilizer Phosphate-2 + 100% NPK

***Number of days until the appearance of the first flower**:*

In this experiment, the control treatment had the longest duration of flowering with an average of 73 days from the time of seed germination. and the shortest time until the appearance of the first flower in marigold (with an average of 60 days from the time of seed germination) (Table 3-2). As a result of the combined use of biological and chemical fertilizers in the form of azotobacter + fertilizing phosphate, 2 and 50% of the recommended amount of NPK fertilizer needed in marigold cultivation was obtained. Phosphorus is one of the key elements that plays a role in energy transfer, cell division, cell wall phospholipids structure, development of plant reproductive parts (Poriosov et al., 2009). The use of phosphate-dissolving bacteria caused a significant increase in available phosphorus in the soil due to the positive effect of bacteria on dissolving phosphorus faster (Hendawy, 2008).

Plant fresh weight:

In this experiment, the fresh weight of the marigold medicinal plant showed an increase due to the combined use of biological and chemical fertilizers, and the highest amount was from the treatment of Azotobacter + fertile phosphate 2 and 50% of the recommended amount of NPK fertilizer required in marigold cultivation. Spring was obtained (Table 3-2), and with the increase in the amount of complete chemical fertilizers, the amount of this trait decreased. Biological fertilizers have the highest efficiency and effectiveness in terms of producing growth-stimulating factors and providing food elements for agricultural plants, and they are one of the most effective helpers for plants to meet their needs. By introducing a large and active population of microorganisms in the activity area of the root, it increases the plant's ability to absorb most of the nutrients (Selispor, 2012, Kochaki et al., 2017, Khan et al., 2007) and microorganisms Biology increases the availability of elements in the soil and plant growth by making plant hormones (Foti et al. 2002). The increase in crop yields as a result of the use of biological fertilizers is due to the provision of micronutrient elements and growth regulators that are obtained from this series of fertilizers (Zodape, 2001).

Fresh and dry flower weight:

During this research, the dry weight of marigold medicinal plant increased due to the combined use of biological and chemical fertilizers, and the highest amount was from the treatment of Azotobacter + fertile phosphate -2 and 50% of the recommended amount of NPK fertilizer required. It was obtained in marigold cultivation (Table 3-2). These positive effects of the application of biofertilizers can be attributed to the increase in the absorption of water and nutrients through the further development of the roots and also to the improvement of the process of biological fixation of nitrogen, which causes an increase in the amount of photosynthesis and ultimately an increase in the number of spikes per meter. square and the amount of flowering.

Table 2-3 Comparison of the average number of days until the appearance of the first flower, plant fresh weight and marigold flower fresh weight under the influence of biological and chemical fertilizers

Flower fresh weight (kg/hectare)	Plant fresh weight (kg/hectare)	Number of days until the first flowering	treatment
1453/3 f	19402/3 f d	73 d	T1
2586 ed	23526/7 c	68 c	T2
2466/3e	23130 c	66c	T3
2751/3 cd	23673/7 bc	63/66 b	T4
2705/3 cde	23173 c	68 c	T5
2900/3c	23579 c	66/33 b	T6
2676/7 cde	23576/7 c	66b	T7
3495a	25303/7 a	60 a	T8
3183/3 b	24210 b	61/33 a	T9

Numbers with the same letters in each column have no significant difference based on Duncan's multi-range test at the 5% probability level. T1: No fertilizer use, T2: 100% NPK, T3: Azotobacter, T4: Azotobacter + 50% NPK, T5: Fertilizer phosphate-2, T6: Fertilizer phosphate-2 + 50% NPK, T7: Azotobacter + biophosphate, T8: Azotobacter + Fertilizer Phosphate-2 + 50% NPK, T9: Azotobacter + Fertilizer Phosphate-2 + 100% NPK

Plant dry weight (biological performance):

In this experiment, the integrated treatment of biofertilizers including phosphate-dissolving bacteria (Fertilizer Phosphate 2) and nitrogen-fixing bacteria (Azotobacter) and chemical fertilizers were able to increase the dry weight of the plant, which is considered biological performance in this research. ¬ should be increased (Table 3-3). It should be noted that the increase of chemical fertilizer to 100% of the recommended amount in marigold cultivation decreased the yield in the integrated treatment of biological and chemical fertilizers. In justification of this, we can refer to (Michlerich's Law of Diminishing Returns) which states that the increase of some factors of production compared to other factors of production, whose amount is constant, causes the total product to increase, but after a certain point, additional product which is obtained after the increase of additional units of production factors decreases. Harvest index: adding biofertilizers by allocating more of the materials to flowers and as a result improving the harvest index increased the harvest index in the treatment of Azotobacter + Fertilizer Phosphate 2 and 50% of NPK consumption (Table 3-3). If the allocation of photosynthetic materials between economic reservoirs is more than other reservoirs in the plant, then the ratio of flower yield to biomass yield will be higher (Ngoc-Son et al., 2001).

Table 3-3 Comparison of plant dry weight (biological function), flower dry weight and marigold harvest index under the influence of biological and chemical fertilizers

harvest index	Flower dry weight (kg/ha)	Plant dry weight (biological function) (kg/hectare)	Treatment
10/18 d	324/67 f d	3171/7 c	T1
10/98 cd	514/67 ed	4684 b	T2
10/65 cd	493/33 e	4630b	T3
11/75 bcd	553/33 cd	4709 b	T4
11/53 bcd	540/67 cde	4686/7 b	T5
12/27 abc	580 c	4726/7 b	T6
11/75 bcd	554/67 cd	4718/9b	T7
13/66a	697/67 a	5150/2 a	T8
13/14 ab	663/33 b	4816/8 ab	T9

Numbers with the same letters in each column have no significant difference based on Duncan's multi-range test at the 5% probability level. T1: No fertilizer use, T2: 100% NPK, T3: Azotobacter, T4: Azotobacter + 50% NPK, T5: Fertilizer phosphate-2, T6: Fertilizer phosphate-2 + 50% NPK, T7: Azotobacter + biophosphate, T8: Azotobacter + Fertilizer Phosphate-2 + 50% NPK, T9: Azotobacter + Fertilizer Phosphate-2 + 100% NPK

The percentage and yield of essential oil:

In this experiment, the percentage of essential oil of marigold medicinal plant increased due to the combined use of biological fertilizers, and the highest amount was obtained from the treatment of Aztobacter + fertilizing phosphate 2 (Table 3-4), due to the use of complete fertilizers. The amount of essential oil of marigolds was reduced chemically. And since the yield of essential oil is a result of the percentage of essential oil and the dry weight of flowers, the increase of each of these traits will increase the yield of essential oil of this plant. Considering that the performance of essential oil in medicinal plants is one of the key and important factors, with the increase in the absorption of nutrients in the leaves, the amount of chlorophyll and carotenoid increases, and due to the increase in photosynthesis, the growth and development of the plant increases and the yield of dry matter increases. increases. In this regard, increasing the yield of dry matter had a direct increasing effect on the yield of essential oil. It has been found that biological fertilizers increase the amount of essential oil (Shokooh et al., 2013). The results of other researchers also indicate this, so that the inoculation of basil plant (Vinutha, 2005) and marjoram plant (Fatma et al., 2006) with different species of Aztobacter increases biomass, growth rate.

Table 3-4 Comparison of the average percentage of essential oil and yield of marigold essential oil per hectare under the influence of biological and chemical fertilizers

Essential oil yield ($kgha^{-1}$)	Essence precentage	Treatment
35/10 f d	0/11 bc	T1
41/20 d	0/08 c	T2
70/76 c	0/14b	T3
70/16 c	0/12 b	T4
79/44 bc	0/14 b	T5
67/73 c	0/11 b	T6
115/67 a	0/22a	T7
95/37 b	0/13 b	T8
75/46 c	0/12 b	T9

Numbers with the same letters in each column have no significant difference based on Duncan's multi-range test at the 5% probability level. T1: No fertilizer use, T2: 100% NPK, T3: Azotobacter, T4: Azotobacter + 50% NPK, T5: Fertilizer phosphate-2, T6: Fertilizer phosphate-2 + 50% NPK, T7: Azotobacter + biophosphate, T8: Azotobacter + Fertilizer Phosphate-2 + 50% NPK, T9: Azotobacter + Fertilizer Phosphate-2 + 100% NPK

Conclusion

In this research, the use of bacteria not only increased the yield and quality, but also reduced the consumption of chemical fertilizers by half. Most of the use of chemical fertilizers in the plant is stored in luxury. And here it caused the decrease of most of the examined traits. Therefore, in order to maintain the stability of the agricultural ecosystem and the soil, and economically, it is better to use NPK chemical fertilizer in the amount of half of the recommended amount, along with phosphate-dissolving biofertilizer (Barvar-2) and Aztobacter. The use of renewable resources and institutions is one of the principles of sustainable agriculture and causes agricultural productivity and the least environmental risks. Biofertilizers increase the effect of chemical fertilizers in agricultural production by increasing the activity of growth-enhancing bacteria. By using chemical fertilizer and biological fertilizer in a combined way, suitable and ideal conditions are provided for plant growth and not only there is no irreconcilable effect between them, but they complement each other.

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