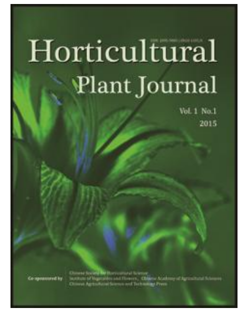
Accepted Manuscript



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PII: S2468-0141(17)30171-1
DOI: http://dx.doi.org/doi: 10.1016/j.hpj.2017.07.003
Reference: HPJ 65

To appear in: *Horticultural Plant Journal*

Efficient Microorganism Compost Benefits Plant Growth and Improves Soil Health in Calendula and Marigold

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Abstract

Organic farming needs to be promoted as a means of sustaining soil health and reducing the cost of farming, especially for small farmers. The present study deals with the effect of organic compost prepared using Efficient Microorganism (EM) consortium and applied along with full or half of the recommended dose of chemical fertilizers on growth of Calendula and Marigold plants, soil physico-chemical parameters and soil enzyme activities. Soil enzyme activities were improved with increase in the rate of EM compost application in both Calendula and Marigold. Carotenoid pigment increased by 46.11% and 12.19% with application of EM compost over the control in Calendula and Marigold flowers respectively. Soil humus, available nitrogen and organic carbon content also increased due to the supplementation of EM compost resulting in better soil fertility. For Calendula, the treatment T5 (Half dose NPK + EM compost 20 000 $kg \cdot hm^{-2}$) was found to be the most promising in terms of acid phosphatase (82.63 µg *p*-Nitrophenyl Phosphate$\cdot g^{-1} \cdot h^{-1}$), dehydrogenase (10.46 µg Triphenyl Formazan$\cdot g^{-1} \cdot d^{-1}$) and *β*-glucosidase (0.30 $IU \cdot g^{-1}$) activities. In Marigold, treatment C (Half dose NPK + EM compost 5 000 $kg \cdot hm^{-2}$) was most promising in terms of amendment in soil enzyme activities.

***Keywords*:** Calendula; Marigold; EM compost; soil fertility; plant growth; soil enzyme activity; soil physic-chemical parameter

1. Introduction

With the escalation of urbanization, there is an increased demand for beautiful landscaping in urban areas. Calendula and Marigold are the most important commercially grown flowers of North India. Besides their use in landscaping, these flowers are also used for garland making, general decoration and religious functions. To fulfill the increasing demand of these flowers, local farmers use high quantities of chemical fertilizer (Shober et al., 2010). The lack of balanced fertilization leads to small leaves, off-color foliage, less elongation, poor inflorescence etc.; hence, optimal availability of nutrients assumes immense importance for these flowers.

Most of the peri-urban farmers rely solely on chemical fertilizers to meet the high nutrient requirements of ornamental crops (Melvin and James, 2001). Although, chemical fertilizers optimize plant growth as well as increase flower yield, this subsequently deteriorates soil health in the long run. In contrast, organic fertilizers are slow-release fertilizers which benefit the ornamental plants by maintaining soil fertility, minimizing burn injury and reducing chemical leaching into land, drains and groundwater. Because of its slow releasing property, organic fertilizers release nutrients throughout the season (http://homeguides.sfgate.com/slow-fertilizers-beneficial-ornamental-plants-46828.html). According to the research undertaken at the Georgia College of Agricultural and Environmental Sciences, one time application of a slow organic fertilizer lasts an entire growing season. As Calendula and Marigold are annual flowering plants and need high dose of nitrogen (N), phosphrorus (P), and potash (K), integration of organic and chemical fertilizers may provide steady delivery of nutrients for better yield of ornamental flowers.

Efficient Microorganism (EM) compost is a bio-organic fertilizer prepared by combination of microbial inoculants, which stimulates the plant growth and soil fertility. This concept was developed by Professor Teruo Higa, University of the Ryukyus, Okinawa, Japan, in 1971 (Higa and Wididana, 1991), which formed the basis for the development of EM compost and its methodology was optimized by our group (Sharma et al., 2014). Application of this bioaugmented compost resulted in better growth of tomato and increased the microbial activity in soil. Besides improvement in soil fertility, EM compost application enhanced the lycopene content of tomato fruits (Verma et al., 2015). The use of organic EM compost not only helps in the balancing nutrients supply but also reduces the cost of cultivation, which is supported by several reports on integrated use of EM compost and chemical fertilizer on horticultural crops (Lee and Cho, 1993; Daly and Stewart, 1999) and paddy (Lee and Cho, 1993). Since limited information is available on the use of organic EM compost in ornamental crops, and flower crops being the source of income for many farmers of peri-urban areas, the present investigation was undertaken to optimize integrated doses of EM compost and chemical fertilizer for Calendula and Marigold crop, through the evaluation of flower yield along with soil physical chemical and biological parameters.

2. Materials and methods

2.1. Site and experimental design

The experiment was carried out from December 2014 to March 2015 at farms (latitude of 28°40´N, longtitude of 77°12´E) of Indian Council of Agricultural Research-Indian Agricultural Research Institute, New Delhi, India. The physico-chemical characteristics (Table 1) [Electrical conductivity (EC), pH, organic carbon (Walkley and Black, 1934), available N (Subbiah and Asija, 1956), available P (Olsen et al., 1954) and available K (Hanway and Heidel, 1952)] of experimental field soil were determined before planting the Marigold (var. Pusa narangi) and Calendula (var. Pacific beauty mix) seedlings. All the treatments were taken in triplicate using randomized block design (RBD). Individual plot size was 2.0 m × 2.0 m.

2.2. Preparation of Efficient Microorganism (EM) compost

The EM consortium consisting of *Candida tropicalis* (Y6), *Phanerochaete chrysosporium* (VV18), *Streptomyces globisporous* (C3), *Lactobacillus* sp. and photosynthetic bacteria was applied to paddy straw amended with poultry droppings in compost pits following the protocol developed earlier (Sharma et al., 2014). The physico-chemical characteristics of the mature EM compost used was: pH 7.8; EC 0.38 $S\cdot m^{-1}$; humus 7.55%; C/N ratio 15.66; available P 0.31%; total C 26%; total N 1.66%.

2.3. Treatment details

The experiment was carried out with five treatments replicated thrice including different combinations of EM compost and chemical fertilizer for both the Calendula and Marigold crops. Recommended doses of nitrogen, phosphorus and potash were used for both the flower crops as a full dose of NPK, as the control. The recommended dose of chemical fertilizer for Calendula was N 200 $kg\cdot hm^{-2}$, P 100 $kg\cdot hm^{-2}$ and K 200 $kg\cdot hm^{-2}$. The recommended dose of chemical fertilizer for Marigold was N 120 $kg\cdot hm^{-2}$, P and K 80 $kg\cdot hm^{-2}$ each. The dose of EM compost was calculated on the basis of original nutrient content of EM compost to substitute the desired amount of chemical fertilizers. EM compost was added to soil in the beginning of the experiment while preparing the field.

The treatments for Calendula field were: T1: Full dose of NPK (N_{200}:P_{100}:K_{200}); T2: Full dose NPK + EM compost 5 000 $kg\cdot hm^{-2}$; T3: Half dose NPK + EM compost 10 000 $kg\cdot hm^{-2}$; T4: Half dose NPK + EM compost 15 000 $kg\cdot hm^{-2}$; T5: Half dose NPK + EM compost 20 000 $kg\cdot hm^{-2}$.

Treatments for Marigold field were: A: Full dose NPK (N_{120}:P_{80}:K_{80}); B: Full dose NPK + EM compost 2 500 kg·hm^{-2}; C: Half dose NPK + EM compost 5 000 kg·hm^{-2}; D: Half dose NPK + EM compost 7 500 kg·hm^{-2}; E: Half dose NPK + EM compost 10 000 kg·hm^{-2}.

2.4. Flower growth and carotenoid pigment estimation

The numbers of flowers per plant were counted at regular intervals. Carotenoid pigment was extracted from the fresh flowers by using extractant (acetone:petroleum ether; 1:1, v/v). Total carotenoid was estimated by the method of Verma et al. (2015). The carotenoid content was expressed as mg·g^{-1} of fresh flower.

2.5. Estimation of soil enzyme activities and microbial biomass

A set of five soil cores (5 cm diameter, 0–15 cm depth) were taken from each plot and pooled together. The samples were stored at 4 °C for further microbiological analyses. Dehydrogenase activity was estimated by the method of Casida et al. (1964) using 3% Triphenyl Tetrazolium Chloride (TTC) as a substrate. The enzymatic activity was expressed as μg of triphenyl formazon (TPF) released·g^{-1} soil·d^{-1}. Acid phosphatase (EC 3.1.3.2) and alkaline phosphatase (EC 3.1.3.1) activity were assayed by method of Tabatabai and Bremner (1969) using *p*-nitrophenol phosphate as a substrate, at pH 6.0 and 11.0, respectively. The enzymatic activity was expressed as μg of *p*-nitrophenol released·g^{-1} soil·h^{-1}. Fluorescein diacetate (FDA) hydrolyase was estimated by method of Green et al. (2006) using FDA as a substrate. The activity was expressed as μg of fluorescein released·g^{-1} soil·h^{-1}. *β*-glucosidase (EC 3.2.1.21) was assayed by method of Wood and Bhat (1988) using *p*-nitrophenyl-*β*-*D*-glucopyranoside as a substrate. Enzyme activity was expressed in IU·g^{-1}. Microbial biomass C (MBC) was estimated by the method of Nunan et al. (1998), using aliquots of K_2SO_4 extracts through dichromate digestion and expressed as μg C·g^{-1} soil.

2.6. Statistical analysis

ANOVA (Analysis of variance) was performed in accordance with randomized block design using SPSS 16 statistical package to quantify the source of variation and critical difference (*CD*) calculated at probability level of 0.05%. Standard deviation (*SD*) values were calculated using Microsoft Excel and depicted in graphs as error bars. Duncan's multiple range test (DMRT) analysis was done and represented in tables with alphabets with 'a' representing highest value ($P < 0.05$).

3. Results

3.1. Effect on number of flower per plant and carotenoid pigment content in flowers

In Calendula, the flower number was positively influenced in response to application of EM compost with half dose of chemical fertilizers. The highest number of Calendula flowers was observed in treatment T4 (24.93 flowers per plant) receiving 15 000 kg·hm^{-2} EM compost along with half dose of chemical fertilizer. Similarly, in Marigold field, the treatment D (9.86 flowers per plant) receiving 7 500 kg·hm^{-2} EM compost with half dose of chemical fertilizers showed the highest number of flowers (Fig. 1). Calendula and Marigold plots receiving the full dose of chemical fertilizers (Treatment T1 and A) and EM compost supplemented with half dose of NPK (treatment T4 and D) showed the highest number of flowers.

T1: Full dose NPK; T2: Full dose NPK + EM compost 5 000 kg·hm^{-2}; T3: Half dose NPK + EM compost 10 000 kg·hm^{-2}; T4: Half dose NPK + EM compost 15 000 kg·hm^{-2}; T5: Half dose NPK + EM compost 20 000 kg·hm^{-2}; A: Full dose NPK; B: Full dose NPK + EM compost 2 500 kg·hm^{-2}; C: Half dose NPK + EM compost 5 000 kg·hm^{-2}; D: Half dose NPK + EM compost 7 500 kg·hm^{-2}; E: Half dose NPK + EM compost 10 000 kg·hm^{-2}.

Compost application modulated the carotenoid content of the Calendula and Marigold flowers. The increase in carotenoid pigment content was observed in all the treatments receiving EM compost over the control (Fig. 2). In case of Calendula flowers, with the increase in the rate of EM compost application, carotenoid pigment also increased. The highest carotenoid pigment was recorded in treatment T4 (1.0 mg·g^{-1}) which was at par with treatment T5 receiving 20 000 kg·hm^{-2} EM compost. Similar pattern of carotenoid pigment content was observed in Marigold flowers, with the highest value recorded in treatment D (2.0 mg·g^{-1}) receiving 7 500 kg·hm^{-2} of EM compost with half dose of chemical fertilizer. Carotenoid pigment was increased by 46.11% and 12.19% with application of EM compost over the control in Calendula and Marigold flowers respectively.

3.2. Effect of EM compost on physico-chemical parameters of soil

Application of EM compost improves the humus content, organic carbon and available nitrogen status of soil which in turn increased the soil fertility. Application of EM compost in both fields resulted in increased organic carbon, and humus status of soil (Table 2, Table 3) over the control receiving chemical fertilizer only.

3.3. Effect of EM compost on microbial enzyme activity in Calendula and Marigold field

In Calendula, soil enzyme activities, eg., dehydrogenase, *β*-glucosidase and acid phosphatase activity enhanced with the increase in the rate of application of EM composts along with half dose chemical fertilizer (Table 2). Highest dehydrogenase activity (10.46 μg TPF·g^{-1}·d^{-1}), acid phosphatase activity (82.63 μg *p*NP·g^{-1}·h^{-1}) and *β*-glucosidase activity (0.30 IU·g^{-1}) were observed in T5 treatment receiving 20 000 kg·hm^{-2} EM compost with half of the recommended dose of NPK.

In the present study, dehydrogenase activity increased with an increase in the dose of EM compost. Lowest dehydrogenase activity was observed in T1 (full dose of NPK) and EM compost amendment enhanced the dehydrogenase activity. A similar trend was observed in case of alkaline phosphatase activity. Highest alkaline phosphatase (157.20 μg *p*NP·g^{-1}·h^{-1}) and FDA hydrolase (1.11 μg Fluorscein released·g^{-1}·h^{-1}) activity was recorded in the treatment receiving EM compost of 10 000 kg·hm^{-2} along with half dose of NPK (T3). In terms of FDA hydrolase activity, there was a decline in activity in treatment T5, when EM compost application increased to 20 000 kg·hm^{-2} as compared with T3 treatment but alkaline phosphatase activity was statistically at par with T3. The activity of all the five enzymes was higher in the treatments receiving compost (T2–T4), at different rates, over the control (T1, recommended dose of chemical fertilizer).

In Marigold field, acid phosphatase activity improved with the increase of EM compost dose. The highest activity of acid phosphatase (49.27 μg *p*NP·g^{-1}·h^{-1}) was observed for treatment E receiving highest dose of EM compost 10 000 kg·hm^{-2} with half of recommended dose of NPK over the treatment A (Table 3). The highest activities of alkaline phosphatase (116.91 μg *p*NP·g^{-1}·h^{-1}) and dehydrogenase (2.88 μg TPF·g^{-1}·d^{-1}) were observed in treatment C receiving 5 000 kg·hm^{-2} EM compost with half of the recommended dose of NPK. Activities of FDA hydrolase and *β*-glucosidase were the highest in treatment D (0.50 μg Fluorscein released·g^{-1}·h^{-1} and 2.49 IU·g^{-1} respectively). Similarly, all the treatments involving the application of EM compost (B–E) showed higher microbial activity over the control (A, recommended dose of chemical fertilizer).

Microbial biomass C also increased with supplementation of organic EM compost (Fig. 3), resulting in higher enzyme activity in all treatments receiving EM compost.

4. Discussion

Extensive use of chemical fertilizers reduces soil organic matter resulting in lower soil biological activity and deterioration in soil physical properties (Gajalakshmi and Abbasi, 2002). Application of compost revitalizes the soil biological activity and reverses the negative effect of chemical fertilizers. Amendment of compost with chemical fertilizer improves soil biomass, soil respiration, enzymatic activities, nitrification rate etc. (Raviv, 2005). Compost can also be used successfully for biological control of diseases in horticultural crops as it supports a rich diversity of microorganisms, hence extends a potentially antagonistic community to phytopathogens (Coventry et al., 2002). EM compost is the promising soil supplement because it contains beneficial microbes and increases the soil fertility by slow release of organically bound nutrients. EM derived compost favours the plant growth parameters of various crops like *Vigna mungo*, tomato, rice and vegetables etc. (Lee and Cho, 1993; Raja Namasivayam and Bharani, 2012; Verma et al., 2015). EM compost, when applied with chemical fertilizer further improved the soil nutrient status, stimulated the economic yield and cost benefit ratio of cultivated crops.

In the present study, two ornamental crops (Calendula and Marigold) were selected to evaluate the impact of EM compost integrated with reduced dose of chemical fertilizers. The flowers were analysed for carotenoid content which is an important component of the photosynthetic machinery of plants. The results illustrated that the EM compost amendment significantly stimulated the photosynthetic ability of Calendula and Marigold plants, particularly in terms of carotenoid content. Moreover, carotenoid content was also positively correlated with the number of flowers in both Calendula ($r = 0.723$; $P < 0.05$) and Marigold ($r = 0.689$; $P < 0.05$). This revealed the positive impact of soil fertility on photosynthetic ability leading to improved growth characteristics of the plant. Similar observations were reported by several researchers in *Dracocephalum moldavica* (Hussein et al., 2006), *Tagetes erecta* (Khalil et al., 2002) and *Hippeastrum vittatum* (Ashry et al., 1995) wherein the amendment with compost improved the pigment content in flowers. Gajalakshmi and Abbasi (2002) studied the effect of water hyacinth compost on the growth and yield of a flowering plant of *Crossandra undulaefoila* and reported that compost amendment in soil significantly improved height, number of leaves, favourable shoot:root ratio, greater biomass per unit time and larger inflorescence length.

Carbon and nitrogen are among the essential nutrients for plant growth. Other than C and N, humus is the major factor which affects the plant growth by improving soil fertility and increasing the availability of nutrient elements (Verma et al., 2015). Due to the presence of diverse species of microorganisms in the compost, mineralization level of nutrients and subsequently nitrogen-absorbing capacity of plant also improves (Sharma et al., 1997). Amendment of EM compost increased organic carbon and available nitrogen and humus status of soil in both Calendula and Marigold. Similar pattern of increasing in these parameters was observed by Raja Namasivayam and Bharani (2012) with the application of EM compost.

Soil enzymes are mainly used to assess soil health because it refers to the sustenance of agricultural productivity of soil (Gil-Sotres et al., 2005). High phosphatase activity in treatment plot of Calendula and Marigold showed that the consortia of efficient microbes present in compost might have helped to mineralize the P resulting in better uptake by plants (Meena et al., 2016).

Dehydrogenase displays the oxidative activity of viable soil microflora. In the present study, dehydrogenase activity increased with the increase of EM compost dose. This showed that the amendment of EM compost, enhanced the microbial activity in soil (Masto et al., 2006). Similar trend was observed in case of alkaline phosphatase activity. In both the field experiments, the value of alkaline phosphatases was much higher as compared to acid phosphatases. This might be due to the alkaline pH of soil. A similar trend was also observed by Mandal et al. (2007).

Microbial biomass C also increased with supplementation of organic EM compost. This proves that the amendment of EM compost provided organic carbon to the resident microflora for their multiplication, leading to the enhancement of soil microbial enzyme activities (Kanchikerimath and Singh, 2001). These results also highlight that dehydrogenase and phosphatase activities are closely related with microbial organic carbon and these observations were also consistent with the reports of Masto et al. (2006) and Parham et al. (2003).

5. Conclusions

The present study revealed the efficiency of EM compost as soil supplements, to increase the flower number and pigment content of Calendula and Marigold flowers. Moreover, the application of EM compost enhanced soil biological health by improving soil enzyme activities which participate in geochemical cycling.

Acknowledgments

The authors are thankful to the Division of Microbiology, Division of Horticulture and Landscaping, Indian Council of Agricultural Research–Indian Agricultural Research Institute, New Delhi for providing the necessary facilities for undertaking this study. We also thank Dr. Radha Prasanna for giving critical inputs especially in English editing.

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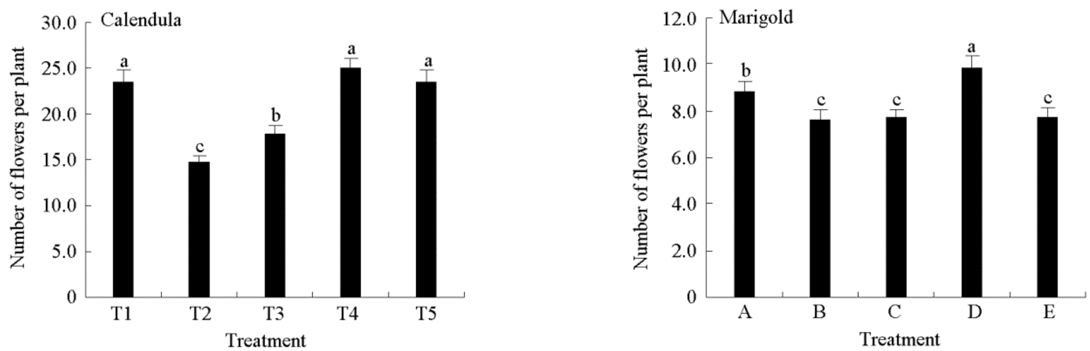

Fig. 1 Effect of EM compost along with chemical fertilizers on inflorescence of Calendula and Marigold flower yield

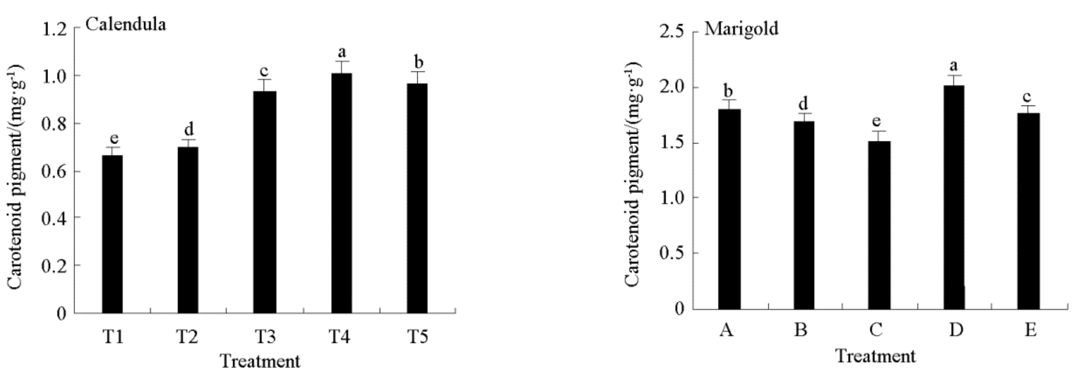

Fig. 2 Effect of integrated use of chemical fertilizer and EM compost on content of carotenoid pigment in flowers of Calendula and Marigold with application of EM compost

T1: Full dose NPK; T2: Full dose NPK + EM compost 5 000 kg·hm^{-2}; T3: Half dose NPK + EM compost 10 000 kg·hm^{-2}; T4: Half dose NPK + EM compost 15 000 kg·hm^{-2}; T5: Half dose NPK + EM compost 20 000 kg·hm^{-2}; A: Full dose NPK; B: Full dose NPK + EM compost 2 500 kg·hm^{-2}; C: Half dose NPK + EM compost 5 000 kg·hm^{-2}; D: Half dose NPK + EM compost 7 500 kg·hm^{-2}; E: Half dose NPK + EM compost 10 000 kg·hm^{-2}.

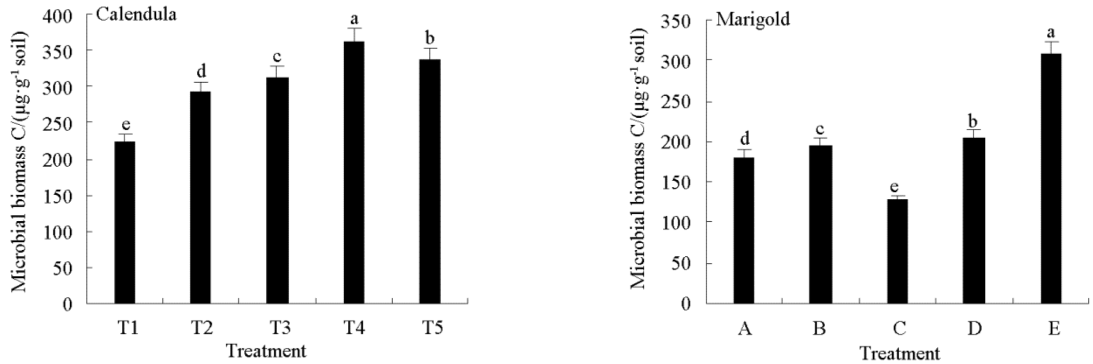

Fig. 3 Effect of EM compost with chemical fertilizer on the microbial biomass C of Calendula field and Marigold field

T1: Full dose NPK; T2: Full dose NPK + EM compost 5 000 kg·hm^{-2}; T3: Half dose NPK + EM compost 10 000 kg·hm^{-2}; T4: Half dose NPK + EM compost 15 000 kg·hm^{-2}; T5: Half dose NPK + EM compost 20 000 kg·hm^{-2}; A: Full dose NPK; B: Full dose NPK + EM compost 2 500 kg·hm^{-2}; C: Half dose NPK + EM compost 5 000 kg·hm^{-2}; D: Half dose NPK + EM compost 7 500 kg·hm^{-2}; E: Half dose NPK + EM compost 10 000 kg·hm^{-2}.

Table 1 Physio-chemical parameters of the soil at the beginning of the experiment

Field	Electrical conductivity/(S·m^{-1})	pH	Available N/(kg·hm^{-2})	Available P/(kg·hm^{-2})	Available K/(kg·hm^{-2})	Organic carbon/%
Marigold	0.01	9.17	225.79	26.88	584.64	0.35
Calendula	0.02	8.26	219.52	38.08	601.44	0.36

Table 2 Effect of EM compost amendment with NPK on physio-chemical parameters and soil enzyme activity in Calendula field

Treatment	Alkaline phosphatase /(μg *p*NP·g^{-1}·h^{-1})	Acid phosphatase/(μg *p*NP·g^{-1}·h^{-1})	Dehydrogenase (μg TPF·g^{-1}·d^{-1})	FDA hydrolase/(μg Fluorscein released·g^{-1}·h^{-1})	*β*-glucosidase/(IU·g^{-1})	Humus/%	Available nitrogen /(kg·hm^{-2})	Organic carbon/%
T1	115.13 c	63.94 d	7.64 b	0.93 ab	0.20 b	1.77 c	200 d	0.55 a
T2	155.89 a	67.88 c	9.92 a	1.04 a	0.25 a	1.65 d	238 a	0.55 a
T3	157.20 a	76.14 b	9.36 a	1.11 a	0.23 b	2.06 a	225 b	0.53 a
T4	144.45 b	75.21 b	9.56 a	0.94 ab	0.23 b	1.93 b	225 b	0.54 ab
T5	152.08 a	82.63 a	10.46 a	0.78 b	0.30 a	1.30 e	213 c	0.55 a
SEM	1.93	1.43	1.41	0.66	0.03	0.04	2.08	0.02
CD ($P < 0.05$)	5.32	3.94	3.89	1.82	0.09	0.12	5.75	0.05

Note: T1: Full dose NPK; T2: Full dose NPK + EM compost 5 000 kg·hm^{-2}; T3: Half dose NPK + EM compost 10 000 kg·hm^{-2}; T4: Half dose NPK + EM compost 15 000 kg·hm^{-2}; T5: Half dose NPK + EM compost 20 000 kg·hm^{-2}. All the lowercases denote ranking in descending order, based on Duncan's multiple range test. Means with different letters are significantly different.

Table 3 Effect of EM compost amendment with NPK on physio-chemical parameters and soil enzyme activity in Marigold field

Treatment	Alkaline phosphatase/(μg pNP·g^{-1}·h^{-1})	Acid phosphatase /(μg pNP·g^{-1}·h^{-1})	Dehydrogenase /(μg TPF·g^{-1}·d^{-1})	FDA hydrolase/(μg Fluorscein released·g^{-1}·h^{-1})	β-glucosidase /(IU·g^{-1})	Humus/%	Available nitrogen /(kg·hm^{-2})	Organic carbon /%
A	99.87 c	38.94 c	2.72 b	0.46 a	2.28 b	1.47 d	225 c	0.54 b
B	100.21 c	48.38 a	2.86 a	0.37 c	1.86 c	1.89 a	238 b	0.54 b
C	116.91 a	46.67 b	2.88 a	0.47 a	2.36 a	1.66 b	238 b	0.54 b
D	108.86 b	39.39 d	2.69 bc	0.50 a	2.49 a	1.62 bc	275 a	0.53 c
E	115.97 a	49.27 a	2.63 c	0.43 b	2.13 b	1.58 c	213 d	0.55 a
SEM	10.75	5.22	1.81	0.08	1.36	0.22	3.23	0.01
CD ($P < 0.05$)	29.67	14.41	5.00	0.22	3.76	0.60	8.91	0.01

Note: A: Full dose NPK; B: Full dose NPK + EM compost 2 500 kg·hm^{-2}; C: Half dose NPK + EM compost 5 000 kg·hm^{-2}; D: Half dose NPK + EM compost 7 500 kg·hm^{-2}; E: Half dose NPK + EM compost 10 000 kg·hm^{-2}. All the lowercases denote ranking in descending order, based on Duncan's multiple range test, means with different letters are significantly different.