

Original Research Article

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Evaluation of Bio-efficacy of Endomycorrhiza and Lipochitooligosaccharides as Seed Treatment on Chickpea

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ABSTRACT

A field experiment was carried out to study the bio-efficacy of endomycorrhiza and Lipochitooligosaccharides as seed treatment on chickpea at Agriculture Research Station, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Achalpur, Dist. Amravati, Maharashtra during Rabi season of the year 2019-20. A set of eight treatments consist of seed treatment kg^{-1} of seed as control (untreated), recommended (Rhizobium -25 g + PSB-25g + Trichoderma-04 g), endomycorrhiza (MycoMix-1g), endomycorrhiza (MycoMix-2g), endomycorrhiza (MycoMix-4g), endomycorrhiza and lipochitooligosaccharides (MycoMix - 1g + MON90505- 0.33 ml), endomycorrhiza and lipochitooligosaccharides (MycoMix 2g + MON90505- 0.33 ml) and endomycorrhiza and lipochitooligosaccharides (MycoMix- 4g + MON90505- 0.33 ml) were taken under Randomized Block Design with four replication. The experimental results indicated that, among the various treatments, seed treatment with endomycorrhiza and lipochitooligosaccharides (MycoMix-4g + MON90505 - 0.33 ml) produced significantly higher number of pods plant^{-1} and grain yield of chickpea (2025kg/ha). It helps to increase soil microbial count, dehydrogenase and phosphate activity. However, the yield levels under seed treatment with endomycorrhiza and lipochitooligosaccharides (MycoMix-4g + MON90505- 0.33ml) were at par with recommended seed treatment (Rhizobium 25 g +PSB 25g + Trichoderma -04g). It is further revealed from the data that seed treatment with endomycorrhiza and lipochitooligosaccharides (Mycomix + MON90505) increased 14.84 to 23.17 %, grain yield of chickpea as compared to control and 2.27 to 9.66 % increase as compared to recommended seed treatment for chickpea (Rhizobium 25g + PSB25g + Trichoderma 04g).

Keywords

MycoMix,
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Introduction

Chickpea (*Cicer arietinum* L.) is a very important legume food crop in arid regions considering as an essential source of protein,

lipid, carbohydrates and vitamins for human beings. The growth, development and yield of chickpea (*Cicer arietinum* L.) is strongly influenced by abiotic factors such as salinity and drought in the arid conditions. The use of

efficient plant growth promoting bacteria in chickpea production is the best solution to overcome those stresses. One of the most ancient and wide spread mutualistic association concerns the endomycorrhizal symbiosis, in which particular soil fungi, called arbuscular mycorrhizal (AM) fungi, colonize the root of most (74%) plant families on earth (Van der Heijden *et al.*, 2015).

In tropical agriculture, the major importance of mycorrhizal symbiosis will be in its role in the phosphorus nutrition of the plants, especially because (i) tropical soils are invariably poor in available phosphorus, (ii) many tropical soils are phosphorus fixing, and (iii) phosphatic fertilizers are expensive. In legumes, particularly, the triple symbiosis of legumes, mycorrhizal fungi and rhizobia deserves special attention because legumes are very important sources of protein and phosphorus is necessary for symbiotic nitrogen fixation. In phosphorus deficient soils, the leguminous host plants obtain phosphorus mainly through VA mycorrhizal association.

A symbiosis has been documented between Gram-negative soil bacteria, Rhizobiaceae and Bradyrhizobiaceae, and legumes such as soybean. The biochemical basis of these relationships involves the exchange of molecular signals, the compounds that transmit signals from plants to bacteria include flavones, isoflavones and flavanones and the compounds that transmit signals from bacteria to plants include the final products of expression of bradyrhizobial and rhizobial nod genes known as lipochitooligosaccharides (LHO). The symbiosis between these bacteria and legumes allows legumes to bind the atmospheric nitrogen necessary for plant growth, and thereby eliminate the need for nitrogen fertilizers. The symbiotic interaction is initiated when a plant releases flavonoid compounds that stimulate

rhizobial bacteria in the soil to produce "Nod-factors." Nod factors are lipochitooligosaccharide compounds (LCO's). Nod-factors are signaling compounds that induce the early stages of nodulation in plant roots, which lead to the formation of root nodules containing the nitrogen-fixing rhizobial bacteria. Although this process occurs naturally over time in legumes, agricultural procedures have been developed to begin the process earlier. These procedures include providing nitrogen-fixing bacteria to seeds or soil and applying nod factors directly to seeds or soil prior to or at sowing. Nod factors have recently been shown to also enhance the germination, growth and yield of legumes and nonlegumes through processes other than nodulation (Prithivaraj *et al.* 2003).

Although the effects of Nod factors on nodulation have been widely studied and reviewed by Ferguson and Mathesius (2003), the mechanisms for Nod factor effects independent of nodulation are not well understood. Nod factors have also been shown to enhance root development (Olah *et al.*, 2005). These LCO's are released by bacteria into the soil, bind to the roots of leguminous plants, and initiate a cascade of plant gene expression that stimulates formation of nitrogen-fixing nodule structures on legume roots. Alternatively, modified and synthetic LCO molecules can be produced through genetic engineering or chemical synthesis. Synthetic LCO's of the same molecular structure interact with plants and stimulate nodulation in the same manner as naturally produced molecules.

In the present study endomycorrhiza (MycMix) and lipochitooligosaccharides (MON90505) were used as seed treatment on chickpea.

Hence, an experiment was conducted to study the bio-efficacy of endomycorrhiza

(MycoMix) and lipochitooligosaccharides (MON90505) as seed treatment on chickpea.

Materials and Methods

An agronomic experiment was carried out at Agriculture Research Station, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Achalpur, Dist. Amravati, Maharashtra during *Rabi* season of the year 2019-20. The chickpea crop (var. JAKI-9218) was sown at 45 x 10 cm spacing in a gross plot size of 6 x 3.6 m with a seed rate of 75 kg ha⁻¹. The crop was raised using RDF @ 25:50:30 kg N:P:K ha⁻¹. The treatments were allotted in field follow the random methods. A set of eight treatments consist of seed treatment kg⁻¹ of seed as control (untreated), recommended (Rhizobium- 25g + PSB -25g + Trichoderma-04g), endomycorrhiza (MycoMix- 1g), endomycorrhiza (MycoMix-2g), endomycorrhiza (MycoMix-4g), endomycorrhiza and lipochitooligosaccharides (MycoMix-1g + MON90505- 0.33 ml), endomycorrhiza and lipochitooligosaccharides (MycoMix-2g + MON90505- 0.33 ml) and endomycorrhiza and lipochitooligosaccharides (MycoMix-4g + MON90505- 0.33 ml) were taken under Randomized Block Design with four replication.

The Rhizobium + PSB + Trichoderma, endomycorrhiza (MycoMix), endomycorrhiza and lipochitooligosaccharides (MycoMix + MON90505) were applied to seed as seed treatment before sowing. Observation on germination, crop vigor, root length, plant population, no of root nodules per plant, no. of pods/plant, No. of seeds/pod, grain yield/ha, microbial count, dehydrogenase activity and phosphatase activity were recorded treatment wise. Data obtained during the course of investigation were subjected to statistical analysis by statistical method as suggested by Panse and Sukhatme (1954) in RBD and conclusions were drawn.

Results and Discussion

Impact on germination, crop vigor and root length

Data on seed germination, crop vigor and root length are presented in Table 1. Highest crop vigor at 30 DAS was observed when chickpea seed is treated with endomycorrhiza and lipochitooligosaccharides (MycoMix @ 4 g / kg seed + MON90505 @ 0.33 ml/kg seed) (T₈). The significantly higher root length of chickpea @ 30 DAS was recorded in treatment T₈ (Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 4 g / kg seed + MON90505 @ 0.33ml/kg seed). But it was found at par with treatment T₇ (Endomycorrhiza and Lipochitooligosaccharides MycoMix @ 2 g / kg seed + MON90505 @ 0.33ml/kg seed), T₆ (Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 1 g / kg seed + MON90505 @ 0.33ml/kg seed) and T₅ (Mycomix @ 4 g / kg seed).

Mycorrhizal treatments significantly increased the shoot and root lengths reported by Salahedin moradi *et al.*, in chickpea in 2013.

Lipochitooligosaccharides have recently been shown to also enhance the germination, growth and yield of legumes and nonlegumes through processes other than nodulation (Prithivaraj *et al.*, 2003).

Lipochitooligosaccharides i.e. nod factors have also been shown to enhance root development (Olah, *et al.*, 2005).

Impact on plant population and no. of nodules per plant

Plant population of chickpea at 30 and 60 DAS showed non-significant effect due to various treatments (Table 1). Significantly

higher No. of nodules per plant at 30 and 60 DAS were recorded in endomycorrhiza and lipochitooligosaccharides (MycoMix @ 4 g / kg seed + MON90505 @ 0.33ml/kg seed) (T₈). However, it remained at par with other treatments except treatment T₃ (Endomycorrhiza and Lipochitooligosaccharides- MycoMix @ 1 g / kg seed) and control (T₁).

Impact on yield contributing characters and grain yield of chickpea

Significantly higher no. of pods/plant of chickpea were recorded in treatment T₈ (Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 4 g / kg seed + MON90505 @ 0.33ml/kg seed). However, it was found at par with treatment T₇ (Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 2 g / kg seed + MON90505 @ 0.33ml/kg seed), T₆ (Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 1 g / kg seed + MON90505 @ 0.33ml/kg seed), T₅ (Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 4 g / kg seed), T₄ (Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 2 g / kg seed) and T₂ (recommended practice). No. of seeds per pod does not affect significantly due to various seed treatments.

Grain yield of chickpea is significantly influence by various seed treatments. Seed treatment with endomycorrhiza and lipochitooligosaccharides (MycoMix @ 4 g / kg seed + MON90505 @ 0.33ml/kg seed) recorded significantly higher grain yield of chickpea (2025kg/ha) followed by T₇ (Endomycorrhiza and Lipochitooligosaccharides- MycoMix @ 2 g / kg seed + MON90505 @ 0.33ml/kg seed), T₆ (Endomycorrhiza and Lipochitooligosaccharides -MycoMix @ 1 g / kg seed + MON90505 @ 0.33ml/kg seed), T₅

(Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 4 g / kg seed), T₄ (Endomycorrhiza and Lipochitooligosaccharides-MycoMix @ 2 g / kg seed) and T₂ (recommended practice). However, these treatments were at par with each other. Lowest grain yield of chickpea was recorded in control treatment.

It is further revealed from the data that seed treatment with endomycorrhiza and lipochitooligosaccharides (MycoMix + MON90505) increased 14.84 to 23.17 %, grain yield of chickpea as compared to control and 2.27 to 9.66 % increase as compared to recommended seed treatment for chickpea (Rhizobium + PSB+ Trichoderma).

Erman *et al.*, (2011) showed that application of endomycorrhiza AM significantly increased the growth and yields of chickpea.

Lipochitooligosaccharides have recently been shown to enhance yield of legumes and nonlegumes through processes other than nodulation (Prithivaraj *et al.* 2003).

Impact on microbial count, dehydrogenase activity and phosphatase activity

Microbial count (fungi) was recorded at 50 % flowering showed significant results due various seed treatments. With the increase in dose of endomycorrhiza (MycoMix) for seed treatment either alone or in combination with lipochitooligosaccharides (MON90505), significant increase in microbial count in soil was recorded.

Significantly maximum dehydrogenase activity in soil per 24 hrs was recorded in seed treatment with endomycorrhiza and lipochitooligosaccharides (MycoMix @ 4 g / kg seed + MON90505 @ 0.33ml/kg seed). Similar trend was recorded in Phosphate activity per 24 hrs.

Table.1 Evaluation of bio-efficacy of Endomycorrhiza (MycoMix) and Lipochitoooligosaccharides (MON90505) as seed treatment on chickpea

Sr. No.	Treatment No.	Treatments	Germination % at 10 DAS	Crop vigor @ 30 DAS (as % against UTC, wherein UTC = 100 %)	Root length @ 30 DAS (cm)	Plant Population/ha @ 30 DAS	Plant Population/ha @ 60 DAS	No. of Nodules /plant @ 30 DAS	No. of Nodules /plant @ 60 DAS	No. of Pods/plant	No. of seeds/pod	Grain yield kg/ha	Microbial Count (Spores/g soil) ($\times 10^{-5}$) at 50 % flowering	Dehydrogenase activity at Maturity ($\mu\text{g TPF g}^{-1}$ soil 24 hr $^{-1}$)	Phosphatase activity ($\mu\text{g PNP g}^{-1}$ soil 24 hr $^{-1}$)
1	T1	UTC (Untreated control)	91.77	100.00	11.65	197915	192013	20.17	26.40	24.26	1.05	1644	10.11	34.85	174.03
2	T2	Recommended (Rhizobium 25g + PSB 25g + Trichoderma-04g)	93.13	120.00	13.80	201388	193633	30.75	38.33	30.27	1.10	1846	13.77	38.53	182.48
3	T3	Endomycorrhiza (MycoMix 1g)	92.03	120.00	12.15	199999	192591	26.91	33.91	28.03	1.10	1709	12.92	39.65	178.93
4	T4	Endomycorrhiza (MycoMix 2g)	92.29	120.00	12.28	200809	193170	28.50	35.83	29.86	1.10	1780	13.83	40.14	188.73
5	T5	Endomycorrhiza (MycoMix 4g)	93.07	120.00	13.40	200693	194212	30.00	36.75	30.10	1.10	1862	14.20	40.48	191.10
6	T6	Endomycorrhiza and Lipochitoooligosaccharides (MycoMix- 1g + MON90505- 0.33ml)	93.23	125.00	13.95	201272	195369	31.33	38.41	30.28	1.10	1888	14.68	41.42	193.40
7	T7	Endomycorrhiza and Lipochitoooligosaccharides (MycoMix - 2 g + MON90505- 0.33ml)	93.49	125.00	14.28	202892	196064	32.00	39.16	31.44	1.10	1923	15.06	41.59	199.98
8	T8	Endomycorrhiza and Lipochitoooligosaccharides (MycoMix- 4 g + MON90505 - 0.33ml)	93.65	130.00	14.48	203124	197800	32.75	39.91	31.80	1.10	2025	15.21	43.76	203.38
	SE (M)		0.57	-	0.73	1511	1424	1.80	1.65	1.05	0.06	82	0.25	0.43	2.64
	CD @ 5%		NS	-	2.13	NS	NS	5.29	4.84	3.10	NS	241	0.74	1.26	7.76

The dehydrogenase activity denotes soil microbial biomass. Mandal *et al.*, (2007) reported a closer relationship between the soil microbial biomass and crop yields under both greenhouses as well as field conditions.

Microorganisms in soil played an important role in nutrient cycling and plant nutrition, reduced pathogen populations, increased soil organic matter, total carbon, cation exchange capacity, and lowered bulk density thus improving soil quality (Bulluck *et al.*, 2002).

In conclusion, the results of present investigation suggest that, among the various treatments, seed treatment with endomycorrhiza and lipochitooligosaccharides (MycoMix@ 4 g / kg seed + MON90505 @ 0.33ml/kg) produced significantly higher number of pods plant⁻¹ and grain yield of chickpea. It helps to increase soil microbial count, dehydrogenase and phosphate activity. Seed treatment with endomycorrhiza and lipochitooligosaccharides (MycoMix @ 4 g / kg seed + MON90505 @ 0.33ml/kg) would be more beneficial to the farmers due to increased number of pods plant⁻¹ and grain yield. However, the yield levels under seed treatment with endomycorrhiza and lipochitooligosaccharides (MycoMix @ 4 g / kg seed + MON90505 @ 0.33ml/kg) were at par with recommended seed treatment (Rhizobium + PSB+ Trichoderma).

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