The associative effect of arbuscular mycorrhizae with *Trichoderma viride* and *Pseudomonas fluorescens* in promoting growth, nutrient uptake and yield of *Arachis hypogaea* L.

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Abstract: A pot experiment was performed to examine the consortium impact of two different indigenous AM fungi i.e. *Glomus mosseae* and *Acaulospora laevis* either alone and/or in combination with *Trichoderma viride* and *Pseudomonas fluorescens* on growth enhancement of groundnut under polyhouse conditions. All the treated/ combinations used significantly influenced the morphological, physiological as well as yield parameters over uninoculated control. Various parameters like plant height, shoot biomass, root biomass, root length, AM spore number, root and shoot phosphorus, protein percent and oil content were found to be in *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* treatment, whereas other parameters like, leaf area, stomatal conductance and leaf chlorophyll content were found to be maximum in triple inoculation of *G. mosseae* + *T. viride* + *P. fluorescens*. The presence of *G. mosseae* + *P. fluorescens* was efficient in increment in both (acidic and alkaline) phosphatase activity. These results emphasize the need for the incorporation of suitable bioinoculant with soil inoculation for ensuring better growth and improved productivity of this important oil yielding plant.
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1. INTRODUCTION

Arachis hypogaea L. commonly known as groundnut is one of the most important oil seed crop produced in the world. The oilseeds are second most important crop after food grains in terms of area under cultivation, production and value in the agricultural economy of India[19, 20].

To minimize the gap between the demand and supply of oil seeds, intensive efforts are being made to increase their production. As ever-increasing population and urbanization cannot allow increase in the land area under the cultivation of oilseeds anymore due to the pressure on land. Hence, there is an urgent need to improve the yield per unit area. To achieve this objective, agricultural scientists have laid more emphasis on biofertilizers like arbuscular mycorrhizal (AM) fungi, *T. viride* and *P. fluorescens.*

The arbuscular mycorrhizal symbiosis is recognized for its multiple positive effects on plant growth and for its important contribution towards the maintenance of soil quality. These fungi are known to improve the nutritional status of the host, particularly phosphorous (P) and thereby enhancing their growth, development and yield[34, 45, 46]. *Pseudomonas fluorescens* commonly known as mycorrhiza helper bacteria are found to be associated with mycorrhiza to promote the symbiosis between fungus and plant by stimulating fungal growth or protecting the fungus against other fungal competitors. It acts as a systemic bio-control agent against various fungal and bacterial diseases by producing a number of secondary metabolites including antibiotics, siderophores and hydrogen cyanide[17]. *Trichoderma viridae* has also been recognized as potent biological agent to contol plant diseases by producing antibiotics and cell walls degrading enzymes that can kill the pathogen[38].

Keeping in view the above information, present investigation was undertaken to investigate the efficacy of two dominant AM fungi i.e. *Glomus mosseae* and *Acaulospora laevis* alone and in combination with two different bioinoculants i.e. *T. viride* and *P. fluorescens* to find out the best combination having the maximum capability of enhancing the growth and yield of *A. hypogaea* in pot experiment under glasshouse conditions.

2. MATERIALS AND METHODS

2.1 Isolation of dominant AM fungi from soil samples

Two dominant AM fungi i.e. *Glomus mosseae* (Nicol. and Gerd.) and *Acaulospora laevis* (Gerd. and Trappe) were isolated from the rhizospheric soil of groundnut plants using 'Wet Sieving and Decanting Technique' of Gerdemann and Nicolson[18]. Spores were then picked by hypodermic needle under stereobinocular microscope.

2.2 Mass multiplication of AM spores

The starter inoculum or pure culture of each selected dominant AM fungus was raised by ''Funnel Technique'' of Menge and Timmer[26] using wheat as host for three months.

2.3 Mass culture of *T. viride*

The inoculum of *Trichoderma viride* (MTCC Number-2074) obtained from Institute of Microbial Technology (IMTECH), Chandigarh and then further mass produced by using wheat bran and saw dust medium which was prepared by using wheat bran, saw dust and distilled water in the ratio of 3:1:4[29].

2.4 Mass culture of *P. fluorescens*

Pseudomonas fluorescens (MTCC No. 103) was also procured from IMTECH, Chandigarh, India and mass multiplied in nutrient broth medium (1.25 g peptone, 0.75 g beef extract, 1.25g NaCl, 250 ml distilled water) for 24 h for suitable bacteria growth.

2.5 Preparation of pot mixture

Surface sterilized seeds of groundnut were grown in earthen pots (25×25 cm) under polyhouse conditions. To each pot 10 percent of inoculum of each AM fungi (*G. mosseae* and *A. laevis*) was added alone and in combination with *T. viride* and *P. fluorescens.* Different treatments used during the present investigation were as follows:

1. Control (autoclaved sterile sand: soil without any bioinoculant)
2. *Glomus mosseae* (G)
3. *Acaulospora laevis* (A)
4. *Trichoderma viride* (T)
5. *Pseudomonas fluorescens* (P)
6. *G. mosseae* + *A. laevis* (G + A)
7. *G. mosseae* + *T. viride* (G + T)
8. *G. mosseae* + *P. fluorescens* (G + P)
9. *A. laevis* + *T. viride* (A + T)
10. *A. laevis* + *P. fluorescens* (A + P)
11. *T. viride* + *P. fluorescens* (T + P)
12. *G. mosseae* + *A. laevis* + *T. viride* (G + A + T)
13. *G. mosseae* + *T. viride*+ *P. fluorescens* (G + T + P)
14. *G. mosseae* + *A. laevis* + *P. fluorescens* (G + A + P)
15. *A. laevis* + *T. viride* + *P. fluorescens* (A + T + P)
16. *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* (G + A + T + P)

Five replicates of each treatment were taken.

2.6 Quantification of AM spores

AM spores were quantified by grid line intersect method[2].

2.7 Identification of AM fungi

For identification of AM spores, the keys of Walker[44], Scheneck and Perez[37], Morton and Benny[27], Mukerji[28] were followed.

2.8 Analysis of various growth, biochemical and yield parameters

Plants were harvested after 120 days by uprooting them from the soil and various morphological and physiological parameters were measured. After harvest, roots and shoots were weighed separately to determine fresh weight, and then placed in an oven to dry at 70°C until a constant dry weight was obtained. Leaf area (cm^2) was assessed by using leaf area meter (Systronics 211, Ahmedabad, India).

The chlorophyll content was estimated by using Arnon's method [6] by using 80% acetone as solvent. Protein was estimated by the method of Bradford [11] using coomassive brilliant blue G-250 dye. The stomatal conductance was measured by using Porometer (AP4- Delta T devices, Cambridge, UK in morning and evening. The phosphorus content of roots and shoots of all experimentally plants was estimated by 'Phospho-vanadomolybdate yellow colour method [21]. Phosphatase activity was assayed by using p-nitrophenyl phosphate (PNPP) as substrate, hydrolyzed by the enzyme to p-nitrophenol. For this, ice cold sodium acetate buffer (0.05 M with pH 4.8) for acid phosphatase and sodium carbonate-bicarbonate buffer (0.05 M with pH 10) for alkaline phosphatase activity was used [42]. Oil was extracted by petroleum ether of boiling range between 40-60^{0}C using the Soxhlet's procedure [5].

2.9 Mycorrhizal root colonization

Mycorrhizal root colonization was studied by 'Rapid Clearing and Staining Method' of Phillips and Hayman [33]. Percentage AM colonization of roots was calculated using: (Number of root segments colonized / number of root segments studied) × 100.

2.10 Statistical analysis

All results were analyzed using Analysis of Variance (ANOVA), followed by post hoc test through computer software SPSS 11.5 version. Means were ranked at $P \leq 0.05$ level of significance using Duncan's Multiple Range Test for comparison.

3. RESULTS AND DISCUSSION:

Analysis of the present investigation showed that soil inoculated with AM fungi and bioinoculants increased plant growth, nutrient uptake and yield parameters of groundnut crops.

3.1 Plant growth parameters

All the different plant growth parameters significantly increased in all the inoculated treatments in comparison to uninoculated control (Table 1). The change in plant height was highest in plants treated with *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens*

followed by *G. mosseae* + *T. viride* and least in control. The longest roots were also recorded with *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* followed by *G. mosseae* + *T. viride* + *P. fluorescens*. The shoot biomass and root biomass (fresh and dry) was recorded to be maximum in *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* treated plants and lowest in control. It could be due to AM fungi colonization of root system or as external hyphae of the AM fungi greatly increase the effective absorbing surface area of the plant [10, 8]. Kapoor and Mukherji [24] found that the maximum increment in root length is due to the network of mycorrhizal mycelia, able to extend deeper in soil to invade nutrient depletion zone. El-Azouni *et al*.[16] and Abo-Ghalia *et al*.[1] also observed maximum growth enhancement with different AM fungi. Work done by Sivakamasundari and Usharani[39] indicated that mixed combination of *Glomus fasciculatum* and *Pseudomonas fluorescens* on the maximization of growth and yield of maize.

Table-1: Influence of AM Fungi, *T. viride* and *P. fluorescens* on different growth parameters of Groundnut after 120 days.

Sr. No.	Parameters→ Treatments ↓	Plant height (cm)	Shoot biomass (g)		Root biomass (g)		Root length (cm)	Root colonization (%)	AM spore number/ 10 g soil
			Fresh	Dry	Fresh	Dry			
1	**Control**	20.22±3.10^{f}	14.13±0.041^{f}	1.21±0.050^{f}	0.62±0.264^{f}	0.05±0.018^{f}	0^{f}	0^{f}	35.22±5.202^{f}
2	**G**	41.10±3.14^{c}	23.69±0.031^{bc}	2.97±0.038^{d}	1.30±0.220^{de}	0.22±0.027^{d}	18.48±1.62^{c}	53.45±2.98^{c}	81.73±6.369^{d}
3	**A**	31.42±2.75^{e}	19.42±0.025^{d}	1.85±0.038^{ef}	1.14±0.185^{ef}	0.10±0.015^{e}	15.52±1.79^{d}	35.40±1.52^{e}	60.34±5.201^{ef}
4	**T**	33.12±2.31^{e}	20.71±0.050^{e}	2.55±0.046^{e}	1.18±0.251^{def}	0.12±0.018^{e}	0^{f}	0^{f}	61.91±4.858^{ef}
5	**P**	29.34±2.99^{ef}	19.00±0.038^{d}	1.80±0.070^{ef}	1.10±0.266^{ef}	0.08±0.025^{ef}	0^{f}	0^{f}	49.11±5.429^{f}
6	**G+A**	51.36±4.94^{b}	27.77±0.038^{bc}	3.20±0.063^{bc}	1.56±0.408^{bcd}	0.42±0.018^{c}	19.46±1.82^{bc}	90.31±2.58^{ab}	110.31±5.789^{ab}
7	**G+T**	59.28±2.21^{ab}	31.10±0.044^{ab}	3.86±0.025^{b}	1.60±0.255^{bcd}	0.48±0.018^{bc}	25.32±1.56^{ab}	73.93±2.36^{bc}	102.47±5.458^{b}
8	**G+P**	45.10±0.64^{c}	25.00±0.044^{c}	2.85±0.056^{d}	1.49±0.203^{cde}	0.34±0.031^{cd}	21.42±1.42^{b}	58.34±2.08^{c}	87.90±5.359^{d}
9	**A+T**	43.54±2.26^{c}	24.81±0.035^{c}	2.75±0.035^{e}	1.41±0.267^{cde}	0.34±0.025^{cd}	20.42±2.22^{b}	56.63±1.58^{c}	86.41±6.221^{d}
10	**A+P**	38.48±2.09^{d}	22.00±0.049^{d}	2.84±0.021^{d}	1.25±0.263^{de}	0.18±0.032^{de}	16.38±1.52^{d}	47.37±1.98^{d}	74.23±5.275^{e}
11	**T+P**	35.58±2.08^{de}	21.15±0.049^{cd}	2.30±0.053^{e}	1.20±0.233^{def}	0.15±0.021^{de}	0^{f}	0^{f}	70.11±5.511^{e}
12	**G+A+T**	55.32±1.83^{ab}	28.89±0.037^{b}	3.37±0.018^{b}	1.66±0.197^{bc}	0.52±0.02b^{c}	23.38±1.57^{b}	83.40±1.75^{b}	124.80±6.786^{ab}
13	**G+T+P**	57.32±2.58^{ab}	32.00±0.053^{ab}	3.93±0.065^{ab}	2.03±0.217^{a}	0.75±0.018^{b}	28.28±1.75^{ab}	77.80±2.77^{bc}	107.37±4.799^{b}
14	**G+P+A**	48.5±2.31^{bc}	26.00±0.041^{bc}	3.00±0.072^{c}	1.51±0.327^{bcd}	0.38±0.037^{c}	22.42±1.58^{b}	63.39±1.59^{bc}	92.61±4.349^{c}
15	**A+T+P**	54.36±1.38^{ab}	28.00±0.066^{b}	3.40±0.072^{b}	1.70±0.187^{b}	0.62±0.031^{b}	26.36±1.41^{ab}	69.13±1.87^{bc}	99.82±5.419^{c}
16	**G+A+T+P**	64.24±3.06^{a}	34.12±0.037^{ab}	4.27±0.073^{a}	2.10±0.222^{a}	0.82±0.032^{a}	29.34±2.27^{a}	93.18±2.76^{a}	130.38±6.670a
L.S.D(P≤0.05)		6.8218	1.4866	0.2946	0.6368	0.104	4.4254	7.4288	15.3298
ANOVA($F_{15,32}$)		104.898	411.028	244.332	10.365	166.509	40.544	238.614	101.195

†G: *Glomus mosseae*, A: *Acaulospora laevis*, T: *Trichoderma viride*, P: *Pseudomonas fluorescens*, ±: Standard deviation

‡Mean value followed by different alphabet/s within a column do not differ significantly over one other at P≤0.05 (Duncan's Multiple Range Test)

3.2 AM spore number and root colonization

As depicted from Table- 1, the percent of AM root colonization and AM spore number in soil significantly increased in all the inoculated plants after 120 days of inoculation. The plants grown in the soil without AM inoculum showed absence of colonization and spores. Maximum spore population was observed in the plants treated with *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* followed by the combination of *G. mosseae* + *A. laevis* + *T. viride*. Similarly, the intensity of mycorrhizal root colonization was found highest in the plants inoculated with *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* followed by dual inoculation of *G. mosseae* + *A. laevis* and *G. mosseae* + *A. laevis* + *T. viride*. It was found that root colonization and AM spore number were greatly enhanced synergistically by the interaction of AM fungi, *P. fluorescens* and *T. viride*. *Trichoderma viride* is highly effective in root colonization by producing secondary metabolites and these metabolites enhance AM spore number and colonization [35, 40]. These phosphate solubilizing bacteria behaved as mycorrhizal helper bacteria, promoting root colonization when associated with mycorrhizal fungi [7]. Narula *et al.*[31] reported that highest root colonization allows more mycorrhiza–host contact, helping in exchange of nutrients which can result in improved plant growth. The results of the present study are similar to that of Sultana and Pindi[41] who reported significant effect of various bioinoclants with AM fungi on root colonization in cotton plant. Present results are well corroborated by the findings of other authors[22, 25, 34, 43, 45].

3.3 Leaf area, stomatal conductance and leaf chlorophyll content

Data in Table-2 clearly indicate that bioinoculants had significant promotive effect on leaf area in groundnut with AM fungi. Maximum increment in leaf

area was found in triple inoculation of *G. mosseae* + *T. viride* + *P. fluorescens* followed by consortium of *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens*. Plants inoculated with AM fungi, either alone or in combination with *T. viride*, *P. fluorescens* brought about significant changes in chlorophyll a, b and total chlorophyll content. The results attribute best results in G. *mosseae* + *T. viride* + *P. fluorescens* followed by mixed consortium of *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* and least in control. Stomatal conductance was also found to be enhanced after 120 days of inoculation. Higher stomatal conductance was observed on lower surface than upper surface in all the plants.

Table-2: Influence of AM Fungi, *T. viride* and *P. fluorescens* on different physiological parameters of Groundnut after 120 days.

Sr. No.	Parameters→ Treatments ↓	Leaf area (cm^2)	Chlorophyll content (mg g^{-1} Fresh weight)			Stomatal conductance ($mmol^{-2}$ s^{-2})			
			Chlorophyll a	Chlorophyll b	Total chlorophyll	Morning		Evening	
						Lower	Upper	Lower	Upper
1	**Control**	17.33±0.66^{f}	1.117±0.003^{f}	0.411±0.004^{f}	1.528±0.003^{f}	66.80 ± 5.89^{f}	15.44 ± 0.49^{f}	54.00 ± 3.53^{f}	11.40 ± 1.43^{f}
2	**G**	23.04±0.28^{c}	2.257±0.004^{d}	0.739±0.002^{e}	2.996±0.003^{d}	286.00 ± 9.90^{b}	45.94 ± 2.42^{bc}	188.20 ± 4.76^{bc}	28.30 ± 1.41^{b}
3	**A**	19.41±0.34^{e}	1.860±0.004^{e}	0.515±0.006^{ef}	2.375±0.004^{e}	187.60 ± 8.17^{e}	24.30 ± 2.85^{e}	95.00 ± 3.16^{e}	16.74 ± 1.34^{ef}
4	**T**	20.75±0.30^{de}	2.100±0.006^{de}	0.860±0.004^{d}	2.960±0.003^{d}	192.20 ± 6.97^{de}	26.34 ± 1.75^{de}	100.60 ± 3.64^{d}	17.92 ± 0.95^{e}
5	**P**	19.57±0.33^{e}	1.560±0.003^{ef}	0.700±0.003^{e}	2.260±0.003^{de}	176.40 ± 7.19^{e}	23.54 ± 1.46^{e}	90.00 ± 5.33^{e}	16.52 ± 1.42^{e}
6	**G+A**	25.40±0.48^{b}	2.699±0.003^{c}	1.384±0.003^{c}	4.083±0.003^{bc}	242.60 ± 6.65^{cd}	38.40 ± 1.37^{cd}	152.60 ± 4.15^{c}	23.66 ± 1.50^{bc}
7	**G+T**	23.47±0.43^{c}	2.729±0.005^{bc}	1.392±0.003^{c}	4.121±0.004^{bc}	293.80 ± 7.42^{b}	50.66 ± 1.65^{b}	201.20 ± 6.49^{b}	29.48 ± 1.64^{b}
8	**F+P**	23.91±0.63^{c}	2.470±0.006^{cd}	1.099±0.004^{cd}	3.569±0.003^{cd}	230.40 ± 11.69^{d}	36.42 ± 1.52^{cd}	137.60 ± 4.44^{cd}	19.94 ± 1.19^{cd}
9	**A+T**	22.21±0.68^{cd}	2.389±0.004^{cd}	0.990±0.005^{cd}	3.379±0.004^{cd}	196.80 ± 8.67^{de}	27.38 ± 1.53^{d}	112.60 ± 4.92^{d}	18.38 ± 1.15^{cd}
10	**A+P**	21.88±0.52^{d}	2.186±0.004^{de}	0.732±0.004^{e}	2.918±0.005^{d}	195.20 ± 11.32^{de}	25.76 ± 1.87^{de}	100.60 ± 6.18^{d}	16.26 ± 1.34^{e}
11	**T+P**	20.46±0.33^{de}	2.114±0.005^{de}	0.861±0.003^{d}	2.975±0.004^{d}	237.60 ± 7.12^{d}	38.00 ± 1.71^{cd}	147.80 ± 4.65^{c}	20.44 ± 1.90^{c}
12	**G+A+T**	26.10±0.28^{ab}	2.763±0.003^{bc}	0.985±0.004^{cd}	3.748±0.004^{c}	270.60 ± 9.91^{c}	46.02 ± 2.88^{bc}	185.80 ± 4.8^{bc}	26.50 ± 1.19^{bc}
13	**G+T+P**	27.40±0.56^{a}	3.150±0.004^{a}	1.980±0.004^{a}	5.130±0.003^{a}	322.20 ± 11.88^{a}	54.72 ± 2.36^{a}	229.00 ± 4.30a	38.26 ± 1.34^{a}
14	**G+P+A**	24.80±0.41^{b}	2.512±0.006^{c}	0.798±0.003^{d}	3.310±0.001c^{d}	252.20 ± 11.94^{cd}	42.30 ± 1.54^{c}	169.20 ± 3.96^{c}	24.48 ± 1.59^{bc}
15	**A+T+P**	24.08±0.62^{b}	2.873±0.005^{b}	1.750±0.004^{bc}	4.623±0.005^{b}	205.20 ± 6.37^{de}	35.32 ± 1.58^{cd}	124.40 ± 4.77^{d}	18.60 ± 1.17^{cd}
16	**G+A+T+P**	26.80±0.34^{ab}	2.901±0.003^{ab}	1.869±0.005^{b}	4.770±0.004^{ab}	303.60 ± 9.37^{ab}	53.44 ± 1.30^{ab}	210.60 ± 4.77^{ab}	31.16 ± 2.23^{ab}
L.S.D($P\leq0.05$)		1.2384	0.102	0.2744	0.376	11.4006	2.3577	5.9397	1.8427
ANOVA($F_{15,32}$)		173.561	861.876	91.337	207.748	242.029	201.226	572.405	113.121

†G: *Glomus mosseae*, A: *Acaulospora laevis*, T: *Trichoderma viride*, P: *Pseudomonas fluorescens*, ±: Standard deviation,

‡Mean value followed by different alphabet/s within a column do not differ significantly over one other at $P\leq0.05$ (Duncan's Multiple Range Test)

It was also maximum in plants inoculated with *G. mosseae* + *T. viride* + *P. fluorescens* but more in the morning than in the evening on lower surface of leaf. Similar was the case with upper surface of leaf i.e. more in the morning than evening. AM fungi increased the water uptake through improved hydraulic conductivity helping in increasing leaf area, leaf stomatal conductance and photosynthetic activity of plant [12]. The increase in total chlorophyll content may be due to increased uptake of P which increases the photosynthetic activity of the plants. The results are in accordance with the study of Nagarajan and Mahadevan[30] who observed that inoculation of *Helianthus annuus* with AM fungi (*G. mosseae, G. deserticola, G. aggregatum* and *Gigaspora*) showed significant increase in chlorophyll content over control.

Akladious and Abbas[3] and Bhatti *et al*.[9] also revealed that the AM fungi along with *Trichoderma* treatment significantly increased photosynthetic pigments of plants.

3.4 Plant nutrient uptake

As Table-3 shows, plant phosphorus (P) concentrations were much higher in the inoculated plants than the non-inoculated ones. Results reveal that higher content of P was recorded in roots than shoots, the maximum P content in shoot and root was noticed in the treatment of *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* followed by *G. mosseae* + *T. viride* + *P. fluorescens* and *A. laevis* + *T. viride* + *P. fluorescens* respectively. The superiority effect of phosphorus content was recorded in consortium and lowest in non-inoculated control plants. A variety of studies conducted by Doley and Jite[14], Kaushish *et al*.[25], Prasad *et al*.[34] suggest that P uptake by plant root can be enhanced when they are inoculated by AM fungi.

Table-3 Influence of AM Fungi, *T. viride* and *P. fluorescens* on phosphorus content, phosphatase activity and plant yield of Groundnut after 120 days.

Sr. No.	Parameters→ Treatments ↓	Phosphorus (%)		Phosphatase (IU/g Fresh Weight)		Yield			
		Shoot	Root	Acidic	Alkaline	No. of Pod	Weight of Pod	Protein content (%)	Oil content (%)
1	**Control**	0.46±0.016^{f}	0.69±0.047^{f}	1.03±0.030^{f}	1.27±0.055^{f}	4.0±1.58^{e}	09.98±1.350^{f}	23.8±0.32^{f}	40.3±0.32^{f}
2	**G**	0.84±0.032^{de}	0.99±0.016^{e}	1.24±0.032^{cd}	1.89±0.072^{c}	7.0±1.87^{cd}	14.98±1.173^{de}	28.6±0.32^{d}	47.6±0.47^{d}
3	**A**	0.55±0.022^{ef}	0.67±0.038^{ef}	1.14±0.029^{de}	1.53±0.053^{d}	5.0±1.22^{d}	11.50±1.336^{ef}	25.3±0.37^{e}	43.9±0.442^{ef}
4	**T**	0.68±0.025^{e}	0.79±0.035^{ef}	1.17±0.023^{d}	1.58±0.063^{d}	8.0±2.55^{bcd}	15.99±1.354^{de}	26.0±0.51^{de}	44.3±0.255^{e}
5	**P**	0.51±0.016^{ef}	0.63±0.032^{f}	1.10±0.007^{de}	1.46±0.070^{e}	5.0±0.71^{d}	12.11±0.695^{e}	24.6±0.46^{ef}	42.6±0.524^{ef}
6	**G+A**	1.18±0.032^{cd}	1.29±0.032^{d}	1.41±0.031^{bc}	2.02±0.070^{bc}	10.0±1.87^{bc}	18.99±1.376^{cd}	31.4±0.41^{c}	50.9±0.55^{c}
7	**G+T**	1.24±0.038^{cd}	1.38±0.047^{cd}	1.51±0.019^{b}	1.33±0.047^{ef}	11.0±2.55^{bc}	20.87±0.615^{c}	32.6±0.37^{bc}	51.20±0.43^{bc}
8	**G+P**	1.06±0.041^{d}	1.18±0.032^{de}	1.30±0.025^{cd}	1.94±0.051^{c}	8.0±1.87^{bcd}	16.18±1.028^{d}	30.5±0.37^{cd}	49.1±0.41^{d}
9	**A+T**	0.98±0.016^{de}	1.13±0.016^{de}	1.28±0.035^{cd}	1.91±0.071^{c}	7.0±1.87^{cd}	16.00±0.910^{d}	29.3±0.33^{d}	48.3±0.37^{d}
10	**A+P**	0.76±0.024^{de}	0.89±0.025^{e}	1.23±0.013^{cd}	1.68±0.067^{cd}	6.0±2.55^{cd}	13.81±0.963^{e}	27.5±0.37^{d}	44.99±0.24^{e}
11	**T+P**	0.70±0.032^{e}	0.83±0.035^{e}	1.20±0.029^{d}	1.64±0.073^{cd}	5.0±1.22^{d}	12.77±1.249^{e}	26.9±0.36^{de}	46.30±0.37^{d}
12	**G+A+T**	1.30±0.035^{c}	1.46±0.035^{c}	1.47±0.029^{bc}	2.00±0.081^{bc}	12.0±1.87^{bc}	25.20±0.750^{b}	34.6±0.43^{ab}	53.65±0.03^{ab}
13	**G+T+P**	1.42±0.016^{b}	1.57±0.025^{b}	1.56±0.016^{b}	2.09±0.085^{bc}	14.0±1.87^{b}	23.22±1.147^{bc}	32.0±0.37^{bc}	53.10±0.55^{b}
14	**G+P+A**	1.10±0.036^{d}	1.31±0.012^{cd}	1.71±0.0158^{a}	2.54±0.063^{ab}	9.0±2.45^{bcd}	17.34±1.133^{cd}	33.7±0.37^{b}	50.4±0.35^{c}
15	**A+T+P**	1.37±0.041^{c}	1.49±0.035^{c}	1.35±0.018^{c}	2.15±0.086^{b}	13.0±2.55^{bc}	24.18±0.825^{bc}	30.9±0.37^{cd}	52.90±0.32^{bc}
16	**G+A+T+P**	1.52±0.016^{a}	1.68±0.032^{a}	1.68±0.016^{ab}	2.59±0.069^{a}	19.0±2.24^{a}	28.12±1.152^{a}	35.1±0.35^{a}	54.20±0.48^{a}
L.S.D($P\leq0.05$)		0.0932	0.1938	0.0878	0.1768	5.0374	3.6994	1.1416	1.3808
ANOVA($F_{15,32}$)		434.741	108.172	168.805	152.469	20.741	65.878	317.931	305.208

†G: *Glomus mosseae*, A: *Acaulospora laevis*, T: *Trichoderma viride*, P: *Pseudomonas fluorescens,* ±: Standard deviation

‡Mean value followed by different alphabet/s within a column do not differ significantly over one other at $P\leq0.05$ (Duncan's Multiple Range Test)

3.5 Root phosphatase activity

As illustrated in Table-3, the root phosphatase activity was appreciably higher in inoculated groundnut plants after 120 days over control. Highest value in acidic activity was observed in the plants inoculated with *G. mosseae* + *P. fluorescens* + *A. laevis* but in case of alkaline phosphatase the highest value was recorded in combination *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens*. According to the results attributed, alkaline phosphatase activity was found higher as compared to acidic phosphatase activity. *G. mosseae* and *P. fluorescens* was efficient in increment in both (acidic and alkaline) phosphatase activity. It was found in the present investigation that plants with higher mycorrhizal root colonization also had maximum phosphatase activity. The increase of acidic and alkaline phosphatase activities in mycorrhizal plants have been reported by several other researchers [23, 13, 32, 43] and it is mostly likely due to higher phosphatase activity of the internal hyphae produced by mycorrhizal fungi[36].

3.6 Yield parameters

The synergistic effects between AM fungi and bioinoculants in increasing yield parameters were also observed in Table 3. After 120 days, average number of pods/plant and weight/pod were recorded to be highest in consortium of *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* and lowest in control. Among all the bioinoculants treatments *G. mosseae* + *A. laevis* + *T. viride* + *P. fluorescens* revealed the maximum value of protein percentage followed by *G. mosseae* + *A. laevis* + *T. viride*. Similarly, the best value of oil content was related to *G. mosseae* + *A. laevis* + *T.*

viride + *P. fluorescens* followed by *G. mosseae* + *A. laevis* + *T. viride*. *G. mosseae* has been found to be more efficient than *A. laevis*, *T. viride* and *P. fluorescens,* when used singly. *Trichoderma* is the best strain of AM symbionts when mixed together for inoculating the *Glycine max* to get higher oil content[15]. The results observed by Amiri *et al*.[4] indicated that AM fungi significantly increased protein and oil content of *Glycine max.* The beneficial effects of various bioinoculants with AM fungi have been reported to increase oil and protein content in various oil yielding plants like *Helianthus annuus*[40], *Glycine max*[45], *Ocimum basilicum*[47] which is in conformity with our results**.**

4. CONCLUSION:

AM fungi are the important basis of sustainable agricultural systems and over the past few years the enormous advances in research on mycorrhizal physiology and ecology have led to a greater understanding of the many roles of AM fungi in the ecosystem. The results of the present study clearly brought out the significant effect of different bioinoculants on various growth as well as physiological parameters of the selected oil yielding plants that may create the possibility of AM fungal application to improve their cultivation and propagation of these valuable oil yielding plants under polyhouse conditions. This combination can be further tested under field conditions and can be recommended to the farmers after proper confirmation.

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