(January – March,2014)



GLOBAL JOURNAL OF BIOLOGY, AGRICULTURE & HEALTH SCIENCES (Published By: Global Institute for Research & Education)

www.gifre.org

Effect of co-inoculation of antagonists and beneficial microorganisms on growth and yield of groundnut under greenhouse conditions

Krishna Naik, L. Raghunandan, B.L. & Shivaprakash, M.K. Dept of Agril. Microbiology, University of Agricultural Sciences, GKVK, Bangalore 560 065, Karnataka.

Abstract

Under glasshouse conditions growth parameters *viz.*, plant height, number of branches per plant and number of compound leaves per stem and yield parameters *viz.*, number of pods per plant, pod yield, shelling percentage, kernel yield and oil content were found maximum in pathogens uninoculated treatments receiving single biocontrol agent followed by other treatments which received two biocontrol agents with beneficial microorganisms and pathogens. The combined inoculation of fungal and bacterial biocontrol agents enhanced the growth parameters significantly compared to inoculation of single biocontrol agent. In general performance of fungal biocontrol agents was better compared to bacterial biocontrol agents in increasing the growth and yield parameters of groundnut.

Introduction

Groundnut (*Arachis hypogaea* Linn.) is an important oilseed crop of tropical and subtropical regions of the world. It is native of South America and belongs to annual legume group. It is regarded as 'King of oilseed crops' on account of its diversified uses. Cultivation of this crop is mainly confined to the geographical belt between 40°N and 45°S latitude. The low productivity in groundnut is attributed to many production constraints. Among them, biotic factors particularly diseases play a major role. The crop is known to be attacked by a number of fungal, bacterial and viral diseases. The literature reveals that the yield losses caused by major fungal diseases like leaf spots, rust and soil borne diseases like stem rot, root rot, collar rot and pod rot singly or in combination is as high as 15 to 70 per cent during both the kharif and rabi-summer seasons (Subramanyam *et al.*, 1984).

Biological control of plant pathogens has been considered as a potential control strategy in recent years. Use of fungal and bacterial bioagents in the management of soil borne plant pathogens is gaining importance. Several fungal and bacterial bioagents controlling soil borne fungal pathogens such as *Sclerotium rolfsii*, *Rhizoctonia bataticola*, *Fusarium oxysporum*, *Aspergillus niger* etc., have been identified (Kim *et al.*, 1999). *Trichoderma* spp. *Pseudomonas fluorescens* and *Bacillus subtilis* are the most commonly used biocontrol agents and have long been known as effective antagonists against plant pathogenic fungi. These bioagents are found in nearly all agricultural soils and are easy to isolate and mass multiply. They affect wide range of plant pathogenic fungi including *Phythium* spp., *Rhizoctonia* spp., *Fusarium* spp. *Sclerotium* spp., *Botrytis* spp., etc (Papavizas, 1985). In spite of enormous scientific literature on biocontrol of plant pathogens with *Trichoderma* spp., *Bacillus* spp. and *Pseudomonas* spp. the most effective species against wide range of plant of fungal and bacterial bioagents is lacking and needed to be found out.

Other properties of biocontrol agents like their compatibility with useful microorganisms like *Rhizobium*, *Bacillus megaterium* and *Glomus fasciculatum* which play an important role in enriching the soil and their ability to promote plant growth, to solubilize insoluble mineral nutrients and to mobilize them to plant roots are the most demanding studies to popularize these very common biocontrol agents and beneficial microorganisms. Keeping these points in view, an attempt was made to study the influence of combined inoculation of antagonists and beneficial microorganisms on growth and yield of groundnut.

Materials and Methods

In vivo evaluation of fungal and bacterial biocontrol agents for antagonism against soil borne fungal pathogens of groundnut

In glasshouse, a pot experiment was conducted in the Department of Agricultural Microbiology, UAS, GKVK, Bangalore to evaluate antagonistic effect of selected fungal and bacterial biocontrol agents along with beneficial microorganisms against soil borne fungal pathogens of groundnut.

Fungal biocontrol agents

Trichoderma harzianum (PDBC TH 10) Trichoderma viride (PDBC TV32) Trichoderma virens (PDBC TVS 12) Bacterial biocontrol agents Bacillus subtilis Pseudomonas fluorescens (pf 1) Test pathogens selected Aspergillus flavus Aspergillus niger Sclerotium rolfsii Rhizoctonia bataticola Fusarium oxysporum Beneficial microorganisms Rhizobium sp. (groundnut strain NC 92) Bacillus megaterium sub sp. phospaticum PB Glomus fasciculatum

Preparation of inoculum

Preparation of inoculum of pathogen

Fungal pathogens were first grown on PDA plates. A mixture of 110 gm. of crushed sorghum, 80 gm. of sand and 10 gm. of dhal powder (i.e., Sorghum : Sand : Dhal Powder :: 11% : 8% : 1% w/w/w) were mixed by adding tap water to sticky consistancy. Then the mixture was filled to autoclavable polybags and the opening of the bag was sealed using rubber band with cotton plug and autoclaved. After autoclaving five mycelial discs of 5 mm. diameter were cut from the margin of actively growing pathogens and transferred aseptically to the polybags containing sterilized sorghum, sand and dhal mixture and were incubated at 27° C for 15 days. The bags were carefully shaken periodically in order to permit uniform growth

Preparation of inoculum of fungal biocontrol agents

The fungal biocontrol agents (*Trichoderma* spp.) were mass multiplied by following the similar procedure as that of fungal pathogens

Preparation of inoculum of bacterial biocontrol agents and beneficial bacteria

Bacterial biocontrol agents and beneficial bacteria were initially grown on respective media plates viz., *Bacillus subtilis* and *Bacillus megaterium* on nutrient agar medium, *Pseudomonas fluorescence* on King's B medium and *Rhizobium* on yeast extract mannitol agar medium and subsequently they were transferred to their respective broth medium aseptically and incubated at 27°C on a rotary shaker at 150 rpm for five days. Shaking was stopped, allowed till good turbidity was formed and then each broth culture was thoroughly mixed in a mixer grinder. Fifty millilitres of required broth culture was individually formulated using lignite as a carrier material (Lignite: Liquid culture of biocontrol agent / beneficial microorganism @ 2:1 w/v) with 10gm. carboxyl methyl cellulose (CMC) per kg carrier material as adhesive.

Preparation of inoculum of Glomus fasciculatum

The starter inoculum of *Glomus fasciculatum* (Thaxter sensu Gerd.) was mixed separately with top 4 cm. layer of sterilized, sieved sand and soil (1:1) mix, taken in 20 kg. capacity battery box. Rhodes grass (*Chloris gayana* Kunth.) seeds were sown. The plants were grown for 90 days and were fed with Ruakura plant nutrient solution once in a week (Smith *et al.*, 1983). After 90 days of growth, shoot portion was removed. Root system was finely chopped and mixed with the substrate (sand) in which the plants were growing and air dried. This inoculum consisted of VAM hyphae, spores and root bits containing vesicles, arbuscules and hyphae. The number of infective propagules in the inoculum was determined by the most probable number (MPN) method, with four fold dilutions (Sieverding, 1991).

Preparation of pots and sowing

A new groundnut variety, GPBD4 was used in the present investigation. It is a high-yielding, improved Spanish bunch groundnut (*Arachis hypogaea* sub sp. *fastigiata* var *vulgaris*) a cross between KRG1 and ICGV86855 developed at the Department of Genetics and Plant Breeding, UAS, Dharwad, Kanrataka, India

Hundred grams of the sand sorghum based inoculum from each of the pathogen, from each of the fungal biocontrol agent and hundred grams soil based inoculum of *Glomus fasciculatum* were mixed with 20 kilograms of sieved sterilized soil and later filled in surface sterilized (0.1% HgCl2) battery boxes according to the treatment requirements. Uniform sized, bold and healthy seeds were treated with lignite based inoculum of biocontrol agents (*B.subtilis* and *P.fluorescens*) and beneficial bacteria (*Rhizobium* sp. and *Bacillus megaterium*) @ 3g per kilogram seeds. Six treated seeds were sown in each battery box and watering was done with over head sprinkler. Germination counts were recorded ten days after germination.

Details of treatments imposed

- T1 Control = No pathogens, no biocontrol agents and no beneficial microorganisms
- T2 BMO = Only beneficial microorganisms (*Rhizobium* sp., *Bacillus megaterium* + *Glomus fasciculatum*)
- T3 P = Pathogens (Aspergillus flavus + Aspergillus niger + Sclerotium rolfsii + Rhizoctonia bataticola + Fusarium oxysporum f.sp. ciceri)
- T4 P+TH10+BMO =P + Trichoderma harzianum (TH10) + BMO
- T5 P+TV32+BMO = P + Trichoderma viride (TV32) + BMO
- T6 P+TVS12+BMO =P + *Trichoderma virens* (TVS12) + BMO
- T7 P+Bs+BMO = P + Bacillus subtilis + BMO
- T8 P + Pf1 + BMO = P + Pseudomonas fluorescens (Pf 1) + BMO
- T9 P+TH10+BS+BMO
- T10 P+TH10+Pf1+BMO
- T11 P+TV32+BS+BMO
- T12 P+TV32+Pf 1+BMO
- T13 P+TVS12+BS+BMO

T14 P+TVS12+Pf1+BMO T15 TH10+BMO T16 TV32+BMO T17 TVS12+BMO T18 BS+BMO T19 Pf1+BMO

During the experimental period, germination counts were recorded initially and observations on plant height, number of branches per plant and number of compound leaves per main stem were recorded at 30, 60 and 90 days after sowing. Number of days taken to fifty per cent flowering was also recorded.

The plants were harvested on 110^{th} day after sowing by depotting and roots were washed with slow running water to remove soil particles and organic debris. Fresh and dry weight (recorded after drying to a constant weight in a oven at 60° C) of plant samples including shoot, root yield and yield components, mycorrhizal spore number per 50 g. soil, AM fungal root colonization of plant was recorded. Extramatricular chlamydospore produced by AM fungi in rhizosphere soil was estimated by wet sieving and decantation method (Gerdeman and Nicolson, 1963). Mycorrhizal root colonization was determined by Grid intersect method as outlined by Giovanetti and Mossae (1980). Per cent oil content of groundnut seeds was determined by nuclear magnetic resonance (NMR) procedure (Jambunathan *et al.*, 1985).

Results and Discussion

Effect of antagonists and beneficial microorganisms on growth parameters of groundnut under glasshouse conditions

The germination percentage of groundnut seeds was found to be inhibited to the greater extent in the treatment inoculated with only pathogens (83.33%) followed by the treatment inoculated with pathogens + BS + BMO (88.88%). In all the other treatments the germination was higher and on par with each other. Hundred per cent germination was recorded in the treatments which received TH 10 + BMO, TVS 12 + BMO, TV 32 + BMO and pathogens + TH 10 + BS + BMO treatments.

At 30 DAS maximum plant height (24.07cm) was observed in TH 10 + BMO treatment followed by TV 32 + BMO (22.13cm). The lowest plant height was observed in control (15.40cm) and in pathogens control (15.60cm) treatments. At 60 DAS plant height was minimum in pathogens control (23.93cm) followed by the treatments receiving P + BS + BMO (24.07 cm), P + Pf1 + BMO (24.07 cm) and in control treatment (24.80cm). Where as in all the other treatments plant height was maximum and on par with each other. At 90 DAS highest plant height was observed in TH 10 + BMO (51.13cm) and TV 32 + BMO (50.07cm) treatments followed by P + TVS 12+Pf1+BMO treatment (44.07cm) and lowest plant height was observed in control (25.27cm) treatment followed by P+BS+BMO (26.33cm) and pathogens control (27.73 cm) treatments.

The 50 per cent flowering was observed earlier in the treatments inoculated with TH 10+BMO (29.33 days) followed by Pf1+BMO and P+TH10+BMO (29.67 days) treatments. Where as delayed flowering was observed in pathogens control treatment (33.67 days) followed by P + BS + BMO (33.33 days) and P + Pf + BMO treatments (32.67 days).

At 30 DAS maximum number of branches were observed in TVS 12 + BMO treatment (3.27) followed by the treatments inoculated with TH 10 + BMO (3.13) and TV 32 + BMO (3.07). Minimum number of branches were observed in pathogen control treatment (2.00) followed by P + TV 32 + BMO (2.13) treatments. At 60 DAS highest number of branches were observed in P + TH10 + BS + BMO (6.0) and TH 10 + BMO (5.86) treatments and minimum number of branches were observed in pathogens control and P + BS + BMO (3.40) treatments. At 90 DAS also minimum number of branches were observed in pathogens control (3.53) followed by control (4.67) and highest number of branches were observed in TH 10 + BMO (7.40) and TVS 12 + BMO (7.33) treatments.

The number of compound leaves was found to be minimum in pathogens control treatment at all the stages of the crop. At 30 DAS maximum number of compound leaves were observed in TVS 12 + BMO (8.00) and Pf1 + BMO (7.80) treatments, whereas at 60 DAS they were maximum in P+ TH 10 + Pf1 + BMO and TH 10 + BMO (10.13) treatments at 90 DAS maximum number of compound leaves were observed in P + TVS 12 + Pf 1+ BMO as well as in TH 10 + BMO (12.93) treatments followed by TVS 12 + BMO (12.8) and TV 32 + BMO (12.60) treatments.

The plant height, number of branches per plant and number of compound leaves per main stem at different stages of growth were found maximum in pathogens uninoculated treatments receiving single biocontrol agent with beneficial microorganisms followed by other treatments which received two biocontrol agents with beneficial microorganisms and pathogens. Minimum plant height, less number of branches per plant and least number of compound leaves per main stem were observed in pathogens control followed by other treatments where single biocontrol agent with beneficial microorganisms and pathogens were co-inoculated. Kleifield and Chet (1992); Dileep Kumar and Dube (1992) obtained similar observations of increased seed germination, growth and yield of chickpea and soybean by seed treatment with fluorescent pseudomonads. Patel *et al.*, (1998) observed significantly increased plant height, number of branches, leaves per plant, number of pods per plant, grains per pod and pod yield in gardenpea by inoculation of *Rhizobium* and phosphate solubilizing microorganisms. The results obtained in the present study are in accordance with the results obtained by Kleifield and Chet (1992) and Dileep Kumar and Dube (1992).

Effect of co-inoculation of antagonists and beneficial microorganisms on yield parameters of groundnut under glasshouse conditions

Number of pods per plant varied from 7.00 to 14.27. Maximum number of pods (14.27) were observed in TH 10 + BMO followed by TVS 12 + BMO (13.40) and TV 32 + BMO (13.27) treatments. Lowest number of pods (7.0) were observed in pathogen control followed by P + TV 32 + BMO (8.67) treatment.

Minimum numbers of pods per plant were obtained in pathogens control treatment followed by other treatments, where single biocontrol agent with beneficial microorganisms and pathogens were coinoculated. Maximum pods per plant was found in treatment receiving single biocontrol agent with beneficial microorganisms followed by other treatments which received pathogens with two biocontrol agents and beneficial microorganisms. Geeta (1993) and Patel *et al.*, (1998) reported that by inoculation of *Rhizobium* and phosphate solubilizing microorganisms with 50 per cent N and P increased plant height, number of branches, leaves per plant, number of pods per plant, grains per pod and pod yield significantly in gardenpea.

Fresh pod yield per pot varied from 15.30 g/pot in pathogens control treatment to 134.62 g/pot in TH10 + BMO treatment. Similarly, dry pod weight per pot was minimum in pathogens control (7.10 g) and maximum in TH 10 + BMO (74.24g) and in other treatments where *Trichoderma* spp. were combined inoculated with beneficial microorganisms. The pod yield (fresh and dry) was found to be significantly lower in treatments where single biocontrol agent and beneficial microorganisms were combined inoculated with pathogens.

Fresh pod yield per pot was found to be minimum in pathogens control followed by other treatments which received single biocontrol agent with beneficial microorganisms and pathogens. The highest fresh pod yield per pot was obtained in the treatments receiving single fungal biocontrol agent with beneficial microorganisms followed by other treatments receiving pathogens with two biocontrol agents and beneficial microorganisms. Dry pod yield per pot also showed similar trend. Geeta (1993); Patel *et al.*, (1998) and Baig *et al.*, (2002) obtained significant increase in the growth and yield of groundnut by inoculation with *Rhizobium* and PGPR.

The kernal weight was found to be maximum in treatments where biocontrol agents (fungal/ bacterial) were coinoculated with beneficial microorganisms without pathogen inoculation. The treatments where only fungal or bacterial biocontrol agents were inoculated along with beneficial microorganisms in pathogen inoculated treatments showed significantly lower kernal weight which was on par with kernal weight obtained in pathogens control (31.22g) treatment.

Minimum weight of hundred kernels was observed in pathogens control followed by other treatments where pathogens with single biocontrol agent and beneficial microorganisms were coinoculated. Maximum weight of hundred kernels was obtained in the treatments receiving single fungal biocontrol agent with beneficial microorganisms followed by other treatments where two biocontrol agents with beneficial microorganisms and pathogens were coinoculated. The shelling percentage also followed similar trend that of 100 kernal weight. Lourduraj *et al.*, (1996) obtained increased nodule number per plant, 100 seeds weight, shelling percentage and seed yield by inoculation of *Rhizobium* strains and application of 100 or 50 per cent NPK to groundnut.

Shelling percentage was found to be maximum (80.05%) in TH10 + BMO and TVS 12 + BMO (79.17%) treatments and it was minimum in pathogen control treatment. Moderate to high shelling percentage was observed in treatments where both fungal and bacterial biocontrol agents were coinoculated with beneficial microorganisms. The treatments which received both fungal and bacterial biocontrol agents with beneficial microorganisms and pathogens gave higher shelling percentage compared to the treatments which received BMO + Pathogens with single biocontrol agent.

Per cent oil content in groundnut seeds was maximum (48.44%) in TVS 12 + BMO treatment followed by other treatments where single biocontrol agent (fungal/ bacterial) and beneficial microorganisms were co-inoculated in pathogen free pots. The lowest oil percentage was observed in pathogens control (45.02%) and P + BS + BMO (45.45%) treatments. Per cent oil content was found to be minimum in pathogens control followed by other treatments which received pathogens with single biocontrol agent and beneficial microorganisms. Maximum oil content was recorded in the treatments receiving single biocontrol agent with beneficial microorganisms followed by the other treatments which received two biocontrol agents with beneficial microorganisms and pathogens. Sankar and Jeyarajan (1996) in a similar study in sesamum reported that seeds treated with *Trichoderma harzianum* and *T.viride* significantly increased root length, shoot length, yield and oil content over control.

Fresh and dry biomass of plant was found to be highest in TH 10 + BMO treatment (48.15 and 11.18 g/pl respectively) followed by TV 32 + BMO treatment (41.62 and 9.61 g/pl). Lowest plant fresh and dry biomass was observed in pathogens control treatment (25.61 and 5.83 g/pl) followed by P + Pf1 + BMO (25.96 and 5.97 g/pl) and P + BS + BMO (27.35 and 6.09 g/pl) treatments. Fresh and dry root biomass per plant was found to be highest in TVS 12 + BMO (5.09 and 1.86 g/pl) and TH 10 + BMO (5.05 and 1.84 g/pl) treatments respectively. Lowest root fresh and dry weight per pant was observed in P + Pf 1+ BMO (1.90 and 0.70 g/pl) and pathogen control (1.92 and 0.69 g/pl) treatments respectively.

Shoot and root fresh as well as dry weight per plant was found to be maximum in treatments which received single biocontrol agent with beneficial microorganisms followed by other treatments receiving two biocontrol agents with beneficial microorganisms and pathogens. Minimum shoot fresh and dry weight was obtained in the pathogens control followed by other treatments where single biocontrol agent with beneficial microorganisms and pathogens. We wight agent with beneficial microorganisms and pathogens were coinoculated. Meyer and Linderman (1986) and Tomar *et al.*, (2003), reported an increase in root dry weight, shoot dry weight and nodulation in clover by inoculation of *Pseudomonas putida* and VAM fungi, which is in accordance with the present study.

The VAM colonization varied from 71.60 to 91.27 per cent in VAM inoculated treatments. Maximum colonization was observed in treatments where single biocontrol agent (fungal/ bacterial) plus beneficial microorganisms were applied without pathogen inoculation followed by other treatments where fungal and bacterial biocontrol agents were co-inoculated along with pathogens. The control and other treatments with only fungal or bacterial biocontrol agent along with pathogens inoculation showed lower colonization by VAM. Similar trend was observed with respect to the number of VAM spore/ 50g soil. The VAM spore number varied from 71 to 128 per 50 g soil in the VAM inoculated treatments. The maximum spore number was observed in P + TH 10 + Pf1 + BMO (128 spores/ 50g soil) followed by P + TV 32 + Pf1 + BMO (126 spores/ 50g soil) and P + TVS 12 + Pf 1+ BMO and TH 10 + BMO (125 spores/ 50g soil). The lowest

G.J.B.A.H.S., Vol.3(1):187-194

(January – March,2014)

spore count (71 spores/ 50g soil) was recorded in P + BS + BMO treatment. The VAM colonization or spores were not detected in treatments where VAM inoculum was not applied.

Increased growth and yield parameters of groundnut obtained in the present investigation by coinoculation of beneficial microorganisms and biocontrol agents might be due to cumulative effects of greater availability of nitrogen, phosphorus and other nutrients to the plants by microbial inoculants (Menaria *et al.*, 2003), suppression of plant pathogens by inoculated biocontrol agents (Windham *et al.*, 1986 and Alagawadi and Gaur, 1988), production of growth promoting substances by microbial inoculants (Windham *et al.*, 1986) and synergistic interactions with other beneficial microorganisms, which have an additive effect on growth and yield parameters (Rakesh Kumar *et al.*, 2004 and Selvadurai *et al.*, 1991).

References

Alagawadi, A.R. and Gaur, A.C., 1988, Associative effect of *Rhizobium* and phosphate-solubilizing bacteria on the yield and nutrient uptake of chickpea. *Plant Soil.*, **105**: 241-246.

Baig, M.M.V., Mia Baig and Muley, S.M., 2002, Enhanced growth of groundnut by plant growth promoting Rhizobacteria. *International Arachis News*, **22**: 60-63.

Dileep Kumar, B.S. and Dube, H.C., 1992, Seed bacterization with a fluorescent pseudomonas for enhanced plant growth, yield and disease control. *Soil Biol. Biochem.*, **24**: 539-542.

Geeta, K.N., 1993, Effect of seed treatment with molybdenum, zinc and calcium on the growth and yield of groundnut (*Arachis hypogaea* L.) *M.Sc.* (*Agri.*) *Thesis*, University of Agricultural Sciences, Bangalore.

Gerdemann, J.W. and Nicolson, T.H., 1963, Spores of mycorrhizal Endogone species extracted from soil by wet sieving and decanting. *Trans. British Mycol. Soc.*, **46**: 235-244.

Giovannetti, M. and Mosse, B., 1980, An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots. *New Phytol.*, **84**: 489-500.

Jambunathan, R., Raju, S.M. and Barde, S.P., 1985, Analysis of oil content of groundnuts by nuclear magnetic resonance spectrometry. *Journal of Scientific and Food Agriculture*, **36**: 162-166.

Kim, B.S., Moon, S.S. and Hwang, B.K., 1999, Isolation, identification and antifungal activity of a macrolide antibiotic, oligomycin A. produced by *Streptomyces libani*. *Canadian J. Bot.*, **77**: 850-858.

Kleifeld, O. and Chet, I., 1992, *Trichoderma harzianum* – intraction with plants and effect on growth response, *Plant Soil*, **144**: 267-272.

Lourduraj, A.C., Geetalakshmi, V., Devasenpathy, P., Nagarajan, P. and Myilswami, V., 1996, Performance of different rhizobial cultures in groundnut. *Madras Agril. J.*, **83**: 163-165.

Menaria, B.L., Pushpendra Singh and Nagar, R.K., 2003, Effect of nutrients and microbial inoculants on growth and yield of soybean (*Glycine max* (L.) Merril). J. Soil and Crops, **13**: 14-17.

Meyer, J.R. and Linderman, R.G., 1986, Response of subterranean clover to dual inoculation with vesicular arbuscular mycorrhizal fungi and a plant growth promoting bacterium *Pseudomonas putida*. *Soil Biol. Biochem.*, **18**: 185-190.

Papavizas, G.C., 1985, *Trichoderma* and *Gliocladium*: Biology, Ecology and potential for biocontrol. *Ann. Rev. Phytopathol.*, 23: 23-54.

Patel, T.S., Katare, D.S., Khosla, H.K. and Dubey, S., 1998, Effect of biofertilizers and chemical fertilizers on growth and yield of gardenpea (*Pisum sativum* L.). Crop Research, 15: 54-59.

Rakesh Kumar, B.L., Jalali and Harichand, 2004, Effect of different VAM fungi on nodulation, nitrogenase activity and rhizosphere microflora of chickpea. *Legume Res.*, 27: 50-53.

Sankar, P. and Jeyarajan, R., 1996, Biological control of sesamum root rot by treatment with *Trichoderma* spp and *Bacillus subtilis*. *Indian. J Mycol. Pl. Pathol.*, **25**: 217-220.

Selvadurai, E.L., Brown, E.A. and Hamilton, J.T.G., 1991, Production of Indole -3- Acetic acid analogues by strains of *Bacillus cereus* in relation to their influence on seedling development. *Soil Biol. Biochem.*, 23: 401 – 403.

Sieverding, E., 1991, Vesiclar-arbuscular mycorrhizal management in tropical agro systems. Technical Cooperation. Federal Republic of Germany Eschborn. Fried and Bremar.

Smith, G.S., Johnston, C.M. and Cornforth, I.S., 1983, Comparison of nutrient solutions for growth of plants in sand culture. *New Phytol*, **94**: 537-548.

Subramanyam, P., Williams, J.H., McDonald, D. and Gibbons, R.W., 1984, Stdies on sclerotial root rot disease of groundnut (*Arachis hypogaea* L.) caused by *Sclerotium rolfsii* Sacc. *Ann. Appl. Biol.*, **40**: 467-476.

Tomar, A., Kumar, N., Pareek, R.P. and Chandra, R., 2003, Residual effect of blackgram inoculated *Rhizobium*, VAM and PSB on succeeding wheat crop. *Indian J. Pulses Res.* **16**: 141-143.

Windham, W.T., Elad, Y. and Baker, R., 1986, A mechanism of increased plant growth induced by *Trichoderma* spp. *Phytopath.*, **76**: 18-21.

Annexure

Table 1. Effect of coinoculation of beneficial microorganisms, biocontrol agents and soil borne fungal pathogens on per cent germination and plant height of groundnut under glasshouse conditions

	Trootmonto	Plant height (cm)			
	Treatments	Germination (%)	30 DAS	60 DAS	90 DAS
T ₁	Control	94.44 ^{ab}	15.40 ^f	24.80 ^{bc}	25.27 ^g
T ₂	BMO	94.44 ^{ab}	17.33 bcdef	26.47 abc	35.40 ^{de}
T ₃	Р	83.33 ^c	15.60 ^{ef}	23.93 ^c	27.73 ^{fg}
T_4	P+TH10+BMO	94.44 ^{ab}	20.93 abcde	28.40 abc	37.73 ^{cd}
T ₅	P+TV32+BMO	91.66 ^{abc}	19.00 abcdef	28.40 abc	36.73 ^{cd}
T_6	P+TVS12+BMO	91.66 ^{abc}	16.67 ^{cdef}	27.93 ^{abc}	32.47 ^e
T ₇	P+BS+BMO	88.88 ^{bc}	15.73 ^{def}	24.07 ^c	26.33 ^{fg}
T ₈	P+Pf1+BMO	91.66 ^{abc}	16.40 ^{cdef}	24.07 ^c	29.13 ^f
T ₉	P+TH10+BS+BMO	100.00 ^a	21.33 ^{abc}	30.40 ^a	40.00 ^c
T ₁₀	P+TH10+Pf1+BMO	97.22 ^{ab}	19.87 ^{abcdef}	29.73 ^{ab}	38.27 ^{cd}
T ₁₁	P+TV32+BS+BMO	97.22 ^{ab}	19.93 ^{abcdef}	28.73 abc	36.33 ^d
T ₁₂	P+TV32+Pf1+BMO	97.22 ^{ab}	20.93 abcde	26.87 abc	36.67 ^{cd}
T ₁₃	P+TVS12+BS+BMO	94.44 ^{ab}	16.60 ^{cdef}	27.33 ^{abc}	35.20 ^{de}
T ₁₄	P+TVS12+Pf1+BMO	91.66 ^{abc}	17.33 ^{bcdef}	27.53 ^{abc}	44.07 ^b
T ₁₅	TH10+BMO	100.00 ^a	24.07 ^a	31.27 ^a	51.13 ^a
T ₁₆	TV32+BMO	100.00 ^a	22.13 ^{ab}	30.67 ^a	50.07 ^a
T ₁₇	TVS12+BMO	100.00 ^a	21.13 ^{abcd}	30.73 ^a	46.67 ^b
T ₁₈	BS+BMO	94.44 ^{ab}	18.40 bcdef	28.53 abc	32.80 ^e
T ₁₉	Pf1+BMO	97.22 ^{ab}	20.93 abcde	28.67 abc	40.00 ^c
	LSD (P <u><</u> 0.05)	8.55	4.54	4.39	3.20

Note: Control = No pathogens, no biocontrol agents, no beneficial microorganisms

BMO = Only beneficial microorganisms - (Rhizobium sp.+ Bacillus megaterium + Glomus fasciculatum)

P = (Only pathogens) - Aspergillus niger, Aspergillus flavus, Sclerotium rolfsi, Rhizoctonia bataticola and Fusarium oxysporum TH10 = Trichoderma harzianum (PDBC TH10)

TV32 = Trichoderma viride (PDBC TV32)

TVS12 = Trichoderma virens (PDBC TVS12)

Pf1 = Pseudomonas fluorescens-1

 Table 2. Effect of coinoculation of beneficial microorganisms, biocontrol agents and soil borne fungal plant pathogens on growth parameters of groundnut under glasshouse condition

			No. of branches/pl			No. of compound leaves /		
	Treatments	Days to 50%					main sten	
		flowering	30 DAS	60 DAS	90 DAS	30 DAS	60 DAS	90 DAS
T_1	Control	30.67 ^{def}	2.33 defg	3.93 ^{fg}	4.67 ^h	5.20 ^f	8.07 ^f	10.33 ^{efg}
T_2	BMO	30.33 ^{def}	2.60 bcdefg	4.80 ^{cde}	5.33 ^{def g}	5.87 ^{def}	8.87 ^{cde}	10.87 ^{def g}
T_3	Р	33.67 ^a	2.00 ^g	3.40 ^g	3.53 ⁱ	4.00 ^g	7.33 ^{gh}	9.80 ^g
T_4	P+TH10+BMO	30.67 def	2.60 bcdefg	4.87 ^{cd}	5.47 ^{cdefg}	5.47 ^{ef}	7.93 ^{fg}	10.80 ^{def g}
T_5	P+TV32+BMO	31.67 ^{bcd}	2.13 ^{fg}	4.47 ^{def}	5.20 efgh	4.27 ^g	8.33 ^{ef}	10.40 ^{def g}
T_6	P+TVS12+BMO	31.33 ^{cde}	2.40 cdefg	4.20 ^{ef}	5.40 defg	5.87 ^{def}	7.20 ^h	10.00 ^{fg}
T ₇	P+BS+BMO	33.33 ^{ab}	2.20 efg	3.40 ^g	4.93 ^{gh}	5.13 ^f	8.53 ^{def}	10.27 ^{efg}
T_8	P+Pf1+BMO	32.67 ^{abc}	2.27 defg	4.00 ^{fg}	5.00 ^{fgh}	5.33 ^f	8.93 ^{cde}	10.87 ^{defg}
T ₉	P+TH10+BS+BMO	29.67 ^{ef}	3.00 ^{abc}	6.00 ^a	5.93 bcd	6.67 ^{bc}	9.53 ^{abc}	11.06 ^{def}
T ₁₀	P+TH10+Pf1+BMO	30.00 ^{def}	3.00 ^{abc}	5.20 ^c	6.13 ^{bc}	5.67 ^{ef}	10.13 ^a	12.40 ^{abc}
T ₁₁	P+TV32+BS+BMO	31.33 ^{cde}	2.60 bcdefg	5.20 ^c	5.60 cdefg	5.87 ^{def}	9.00 ^{cde}	10.20 ^{efg}
T ₁₂	P+TV32+Pf1+BMO	30.00 ^{def}	2.73 abcdef	5.13 ^{cd}	5.73 ^{cde}	6.13 ^{cde}	9.20 bcd	11.80 bcd
T ₁₃	P+TVS12+BS+BMO	31.67 ^{bcd}	2.87 ^{abcd}	4.80 ^{cde}	5.73 ^{cde}	5.87 ^{def}	9.07 ^{cde}	11.07 ^{def}
T ₁₄	P+TVS12+Pf1+BMO	31.00 ^{cdef}	2.73 abcdef	5.00 ^{cd}	5.93 bcd	6.47 bcd	10.07 ^a	12.93 ^a
T ₁₅	TH10+BMO	29.33 ^f	3.13 ^{ab}	5.86 ^{ab}	7.40 ^a	6.87 ^{bc}	10.13 ^a	12.93 ^a
T ₁₆	TV32+BMO	30.33 ^{def}	3.07 ^{ab}	5.33 ^{bc}	6.40 ^b	6.67 ^{bc}	9.87 ^{ab}	12.60 ^{ab}
T ₁₇	TVS12+BMO	30.00 ^{def}	3.27 ^a	5.13 ^{cd}	7.33 ^a	8.00 ^a	10.07 ^a	12.80 ^a
T ₁₈	BS+BMO	30.67 ^{def}	2.87 ^{abcd}	5.20 ^c	5.67 ^{cdef}	7.07 ^b	9.07 ^{cde}	11.33 ^{cde}
T ₁₉	Pf1+BMO	29.67 ^{ef}	2.80 abcde	5.20 ^c	5.87 bcde	7.80 ^a	9.87 ^{ab}	11.07 ^{def}
	LSD (P <u><</u> 0.05)	1.61	0.54	0.59	0.59	0.67	0.65	1.05

Note:

DAS = Days after sowing; PI = Plant

*Mean values in each column with same superscript(s) do not differ significantly by DMRT (P = 0.05)

Bs = Bacillus subtilis

Table 3. Effect of coinoculation of beneficial microorganisms, biocontrol agents and soil borne fungal plant pathogens on yield parameters of groundnut under glasshouse condition

	Treatments	No.of Pods/pl.	Pod yield	Pod yield (dry)	100 Kernal wt.	Shalling %	% Oil content
	Treatments	No.or Pouspi.	(fresh) / pot (g)	/ pot (g)	(g)	Shelling %	% On content
T ₁	Control	9.33 ^{ef}	81.08 ^{hi}	44.68 ^{ijk}	34.70 ^{cdef}	74.95 ^{defg}	46.13 ^{de}
T ₂	BMO	10.33 ^{de}	97.00 ^{efg}	53.48 ^{fgh}	35.92 ^{abcde}	75.13 ^{defg}	46.89 ^c
T ₃	Р	7.00 ^g	15.30 ^k	7.10 ^m	31.22 ^g	72.33 ^g	45.02 ^g
T ₄	P+TH10+BMO	9.33 ^{ef}	87.30 ^{gh}	48.14 ^{hij}	34.92 bcdef	75.07 defg	46.04 def
T_5	P+TV32+BMO	8.67 ^f	71.82 ^{ij}	39.61 ^{ki}	34.30 defg	74.74 defg	45.85 ^{ef}
T ₆	P+TVS12+BMO	8.47 ^f	63.11 ^j	34.80 ¹	32.15 ^{fg}	72.47 ^{fg}	46.03 def
T ₇	P+BS+BMO	8.67 ^f	75.03 ^{ij}	41.38 ^{jkl}	33.10 ^{efg}	73.02 ^{efg}	45.10 ^g
T ₈	P+Pf1+BMO	9.73 ^{ef}	92.60 ^{fgh}	51.08 ^{ghi}	35.10 ^{abcdef}	75.11 ^{defg}	45.45 ^{fg}
Г ₉	P+TH10+BS+BMO	11.20 ^{cd}	103.89 ^{def}	53.30 ^{fgh}	36.50 ^{abcde}	75.38 ^{cdefg}	47.68 ^b
T ₁₀	P+TH10+Pf1+BMO	12.53 ^{bc}	118.70 ^{bc}	65.46 bcd	38.00 ^{abe}	78.28 ^{abcd}	48.17 ^{ab}
T ₁₁	P+TV32+BS+BMO	11.40 ^{cd}	112.17 ^{cd}	61.51 ^{cde}	36.90 ^{abed}	76.30 bcde	47.01 ^c
T ₁₂	P+TV32+Pf1+BMO	12.47 ^{bc}	116.30 bcd	64.23 bcde	37.92 ^{abe}	77.19 ^{abcd}	46.93 ^c
T ₁₃	P+TVS12+BS+BMO	11.80 ^c	111.65 ^{cd}	61.57 ^{cde}	37.50 ^{abed}	77.00 ^{abcd}	46.57 ^{cd}
T ₁₄	P+TVS12+Pf1+BMO	12.07 ^{bc}	113.83 ^{bcd}	62.78 bcde	37.60 ^{abed}	77.00 ^{abcd}	48.13 ^{ab}
T ₁₅	TH10+BMO	14.27 ^a	134.62 ^a	74.24 ^a	38.67 ^a	80.05 ^a	48.40 ^{ae}
Г ₁₆	TV32+BMO	13.27 ^{ab}	122.00 ^{bc}	66.79 ^{bc}	38.38 ^{ab}	78.81 ^{abe}	48.29 ^{ab}
Г ₁₇	TVS12+BMO	13.40 ^{ab}	125.93 ^{ab}	69.45 ^{ab}	38.57 ^a	79.17 ^{ab}	48.44 ^a
Г ₁₈	BS+BMO	11.27 ^{cd}	105.74 ^{de}	58.31 ^{def}	36.60 ^{abed}	75.93 bcdef	47.98 ^{ab}
T ₁₉	Pf1+BMO	11.40 ^{cd}	105.49 ^{de}	57.80 ^{efg}	36.98 ^{abed}	76.97 ^{abcd}	48.07 ^{ab}
	LSD (P <u><</u> 0.05)	1.20	11.45	6.43	3.02	3.06	0.63

Note:

DAS = Days after sowing; PI = Plant

*Mean values in each column with same superscript(s) do not differ significantly by DMRT (P = 0.05)

 Table 4. Effect of coinoculation of beneficial microorganisms, biocontrol agents and soil borne fungal pathogens on fresh and dry weight of shoot, root of groundnut under glasshouse conditions

	Sh	oot	Ro	Root		
Treatments	Fresh wt/pl	Dry wt/pl	Fresh wt/pl	Dry wt/pl		
	(g)	(g)	(g)	(g)		
T ₁ Control	28.05 ^{fgh}	7.19 ^{def}	2.28 ^{ij}	0.83 ^{hi}		
T ₂ BMO	30.57 defgh	7.25 ^{def}	3.24 ^g	1.18 ^f		
T ₃ P	25.61 ^h	5.83 ^f	1.92 ^j	0.69 ^j		
T ₄ P+TH10+BMO	29.57 ^{ef gh}	6.83 ^{def}	2.81 ^h	1.03 ^g		
T ₅ P+TV32+BMO	28.43 ^{fgh}	6.53 ^{ef}	2.54 ^{hi}	0.92 ^{gh}		
T ₆ P+TVS12+BMO	29.54 ^{efgh}	6.78 ^{def}	2.48 ^{hi}	0.91 ^{gh}		
T ₇ P+BS+BMO	27.35 ^{gh}	6.09 ^f	1.93 ^j	0.72 ^{ij}		
T ₈ P+Pf1+BMO	25.96 ^{gh}	5.97 ^f	1.90 ^j	0.70 ^{ij}		
T ₉ P+TH10+BS+BMO	32.02 defgh	7.36 ^{def}	3.91 ^{de}	1.41 ^{cd}		
T ₁₀ P+TH10+Pf1+BMO	34.75 ^{cdef}	7.98 ^{cde}	3.99 ^{de}	1.46 ^{cd}		
T ₁₁ P+TV32+BS+BMO	32.71 defg	7.47 ^{def}	3.70 ^{ef}	1.35 ^{de}		
T ₁₂ P+TV32+Pf1+BMO	34.39 ^{cdef}	7.92 ^{cde}	4.18 ^{cd}	1.52 ^c		
T ₁₃ P+TVS12+BS+BMO	31.76 defgh	7.31 ^{def}	3.49 ^{fg}	1.27 ^{ef}		
T ₁₄ P+TVS12+Pf1+BMO	34.89 ^{cdef}	8.02 ^{cde}	4.51 ^{bc}	1.65 ^b		
T ₁₅ TH10+BMO	48.15 ^a	11.18 ^a	5.05 ^a	1.84 ^a		
T ₁₆ TV32+BMO	41.62 ^b	9.61 ^b	4.77 ^{ab}	1.75 ^{ab}		
T ₁₇ TVS12+BMO	41.11 ^{bc}	9.47 ^{bc}	5.09 ^a	1.86 ^a		
T ₁₈ BS+BMO	35.88 ^{bcde}	8.20 bcde	4.00 ^{de}	1.45 ^{cd}		
T ₁₉ Pf1+BMO	36.66 bcd	8.46 bcd	4.02 ^{de}	1.46 ^{cd}		
LSD (P <u><</u> 0.05)	5.97	1.43	0.36	0.13		

Table 5. Effect of coinoculation of beneficial microorganisms, biocontrol agents and soil borne fungal plant pathogens on
mycorrhizal spore count and root colonisation in groundnut under glasshouse condition at harvest

Treatments –	AM Fungi				
Treatments -	Spore No. / 50 g soil	Colonization (%)			
۲₁ Control	0.00 ^g	0.00 ^f			
BMO	105.00 ^{bc}	85.33 ^{abcd}			
B P	0.00 ^g	0.00 ^f			
₄ P+TH10+BMO	112.00 ^{ab}	76.00 ^{de}			
₅ P+TV32+BMO	92.00 ^{cde}	76.65 ^{cde}			
6 P+TVS12+BMO	83.00 ^{def}	71.60 ^e			
7 P+BS+BMO	71.00 ^f	82.32 ^{abcd}			
в P+Pf1+BMO	79.00 ^{ef}	82.58 ^{abcd}			
P+TH10+BS+BMO	106.00 ^{bc}	86.87 ^{ab}			
• P+TH10+Pf1+BMO	128.00 ^a	88.20 ^{ab}			
1 P+TV32+BS+BMO	95.00 ^{cd}	79.00 ^{bcde}			
² P+TV32+Pf1+BMO	126.00 ^a	88.32 ^{ab}			
з P+TVS12+BS+BMO	101.00 ^{bc}	87.37 ^{ab}			
4 P+TVS12+Pf1+BMO	125.00 ^a	91.00 ^a			
15 TH10+BMO	125.00 ^a	91.27 ^a			
16 TV32+BMO	116.00 ^{ab}	88.07 ^{ab}			
7 TVS12+BMO	116.00 ^{ab}	87.50 ^{ab}			
8 BS+BMO	108.00 ^{bc}	85.98 ^{abc}			
19 Pf1+BMO	112.00 ^{ab}	86.83 ^{ab}			
LSD (P<0.05)	14.63	8.69			

Note: *Mean values in each column with same superscript(s) do not differ significantly by DMRT (P = 0.05)