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Effect of Bio fertilizer on Germination and Morphological properties of *Arachis hypogaea (L.)*

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Abstract

Microbial inoculants are regarded as a suitable substitute for chemical pesticides. The goal of this study was to use individual and consortia microbial inoculants for the cultivation and crop development of various kinds of the ground nut *Arachis hypogaea (L.)*. Microbes. Eleven distinct groups were maintained in the pot culture method for 30 days. Root nodules and nitrogen-fixing bacteria (*Rhizobium*, *Azospirillum*, and *Azotobacter*), phosphate-utilizing bacteria (Phosphobacteria), and stress-resistant arbuscular mycorrhizal fungi were chosen for microbial fertilizer manufacture (T2-T11) in ground nut plants. Different varieties of groundnut *Arachis hypogaea* (L.) such as TMV10, TMV12, TMV (Gn) 13, ICGV 00348, VRI Gn 6was cultivated. Among the different microbial inoculants treated as fertilizers, T10 group showed significantly increased shoot and root length as 11.30±1.45cm, 6.66±0.27. The fresh and dry weight of the TMV (Gn) 13 groundnut seedlings were observed as 8.95±0.20 and 3.91±0.16g/seedling According to our findings, Rhizobium, Phosphobacteria, and AMF microbial inoculants employed as fertilizers resulted in considerable growth (germination %) and crop improvement through enhanced nutrient uptake with increased fresh biomass, shoot length, and root length.

Keywords: *Arachis hypogaea*, Microbial inoculants, Shoot length, root length, Nutrient analysis, weight, seedling.

Introduction

Groundnut (Arachis hypogaea L.) is a popular and widespread crop farmed in over 100 countries across six continents, and it is the world's fourth largest oilseed crop (Nwokolo and Smartt, 1996). It holds great promise as a legume crop due to its ability to provide nitrogen to the soil, low production costs, and wide range of applications. Groundnuts are grown in 108 countries worldwide and are grown in all subtropical and tropical countries. It is a significant cash crop farmed by millions of small farmers due to its economic and nutritional benefits (Kapulnik 1996). It is an important oilseed crop, accounting for approximately 53% of total global output crushed for high quality edible oil, 32% for confectionary use, and the remaining 15% for food and seed production (Kloepper 1993). Groundnuts include a lot of protein, minerals, and vitamins. It may fix atmospheric nitrogen as a legume crop, boosting soil fertility (Sable et al., 2017).Microorganisms have a crucial role in fixing, solubilizing, mobilizing, and recycling nutrients in the agricultural eco-system. These microbes naturally exist in soils. The needed rhizosphere microorganisms are extracted, artificially cultivated in sufficient numbers, and combined with the appropriate carriers in order to boost crop output (Alori and Babalola 2018; Sharma and Singhvi 2017). They are referred to as microbial inoculants or biofertilizers. *Rhizobium, Azotobacter Acetobacter Azospirillum*, phosphate-solubilizing microorganisms (PSM), arbuscular mycorrhizae (AM) fungi, plant growth-promoting rhizobacteria, bacteria that mobilize micronutrients like *Thiobacillus sp*., etc. (Vitorino and Bessa 2017).In order to increase the unit area productivity of agricultural land, the role of different crop nutrients in contributing increased crop yield play a vital role. Among the crop nutrients, nitrogen as well as phosphorus plays an important role in increasing the crop productivity. Plant development is extremely dependent on nutrients like N, P, and K (Karthikeyan *et al.*, 2010; Rani *et al.*, 2007). In today's intensive farming, the plants deplete the soil of many nutrients, which must be replaced. Microbes are a suitable alternative method to replace crop nutrients in such circumstances (Lipkie *et al.*, 2016; Kundan *et al.*, 2015).For agricultural output and plant growth, phosphorus is a crucial nutrient, yet most of it is inaccessible to plants (Wu *et al.*, 2019). By converting insoluble P into soluble forms that plants can absorb whereas phosphate solubilizing bacteria (PSB) like *Bacillus, Azospirillum, Rhizobium*, and *Serratia* can improve

plant development (Zhang *et al.*, 2019; Ramakrishna *et al.*, 2019).The main objective of this study to cultivate the different varieties of *Arachis hypogaea* (L.) with different microbial individual and consortia fertilizers in pot culture method. Based on the germination percentage, the selected significant variety was treated with different microbial inoculants. The morphological and nutrient contents of the selected variety were analyzed.

Materials and methods

Collection and Processing of seeds

Several types of ground nut *Arachis hypogaea (L.)* seeds (TMV10, TMV12, TMV (Gn) 13, ICGV 00348, VRI Gn 6) were acquired from a local private agro centre in Dharmapuri (Tamil Nadu, India). At various phases of growth, bacterial, fungal, and other microbial species may infect the sowing seed with diseases and pests. From the seedling stage prior to culture, the subsequent seed purification step can be omitted. For around 15 days, collect cow urine in a container. Completely dissolve 0.5L of cow urine in 1L of water. Mix carefully before adding 500mg of each variety of seeds. Allow the carcasses seeds and tiny airborne particles to drift in the solution after soaking the seeds in the liquid for 30 minutes. The following measurements such as shoot length, root length, fresh weight and dry weight of the samplings. From the samplings, the following nutrient parameters such as nitrogen, phosphorous, potassium, calcium, magnesium, copper, iron, manganese and zinc were analyzed by standard procedures (Ajay *et al.*, 2019; Valentine*et al.*, 2017; Krishna 1997). The results were statistically analyzed.

Experimental field preparation and setup

The test area was completely ploughed, levelled, and divided into test levels according to the plan. A sow was pre-irrigated to ensure enough soil moisture. The field is laid out in the form of a 1x1m2 split plot with three duplicates. In this study, a range of microbial fertilizers from the Forest Office (Tree Seedling branch), Dharmapuri (Tamil Nadu, India) were used.

Experimental design

Groundnut seeds of *Arachis hypogaea* L. (Order: Fabales, Family: Fabaceae) were grown for 30 days. The weeds in the field were manually picked twice on the 30th DAS following seeding. The eleven treatment groups such as *T1-Control, T2-Azotobacter, T3-AMF, T4-Rhizobium, T5-Azospirillum, T6-Phosphobacteria, T7-Azospirillum+Azotobacter, T8-Phosphobacteria+Azospirillum, T9-Azospirillum+Azotobacter+AMF, T10-Rhizobium+Phosphobacteria+AMF* and *T11-Rhizobium+Azospirillum+AMF* were organised. Seven duplicates were kept in each group.

Microbial inoculants preparation

To make a jaggery paste, cook 150 g of jaggery in a container with 1 L of water for 5 to 10 minutes over a low flame. Once the solution has cooled, combine 100 g of Rhizobium and AMF in the jaggery paste with 0.5 kg of seeds. Add the treated seeds to this container and mix vigorously to ensure that the microbes are evenly dispersed. After drying in the shade, these seeds are ready for planting. This will aid in pest and disease protection throughout the seedling stage. The same process was used for *Phosphobacteria*, *Azotobacter*, and *Azospirillium* samples for both individual and consortia groups. Before planting, single and mixed inoculants were made and combined with the seeds. Irrigation was used prior to seeding to guarantee consistent germination. Irrigation was carefully administered twice a day to avoid over flooding the land with water. Uniform irrigation was used three times each week.

Results and Discussions

Different varieties of groundnut *Arachis hypogaea* (L.) such as TMV10, TMV12, TMV (Gn) 13, ICGV 00348, VRI Gn 6. Was procured from standard laboratory and cultivated in the pot culture method. Eleven different experimental groups were used in this study (n=7). GroupT1-Controlwithout any additional microbial inoculants was maintained. Azotobacter bacterial inoculants used for Group T2. Arbuscular mycorhizal fungal inoculants used for Group T3. Rhizobium, Azospirillum and Phosphobacteria inoculants for Group T4, T5 and T6 respectively. For T7 and T8, *Azospirillum+Azotobacter* and *Phosphobacteria+Azospirillum* microbial inoculants were used respectively. Three different microbial species were

used for the remaining three groups as Azospirillum+Azotobacter+AMF for T9, Rhizobium+Phosphobacteria+AMF for T10 and *Rhizobium+Azospirillum+AMF* for T11 groups respectively. The consortia cultures were prepared to analyze the effect of microbial inoculants on the groundnut nutrient uptake, germination percentage and morphology of the seedlings.

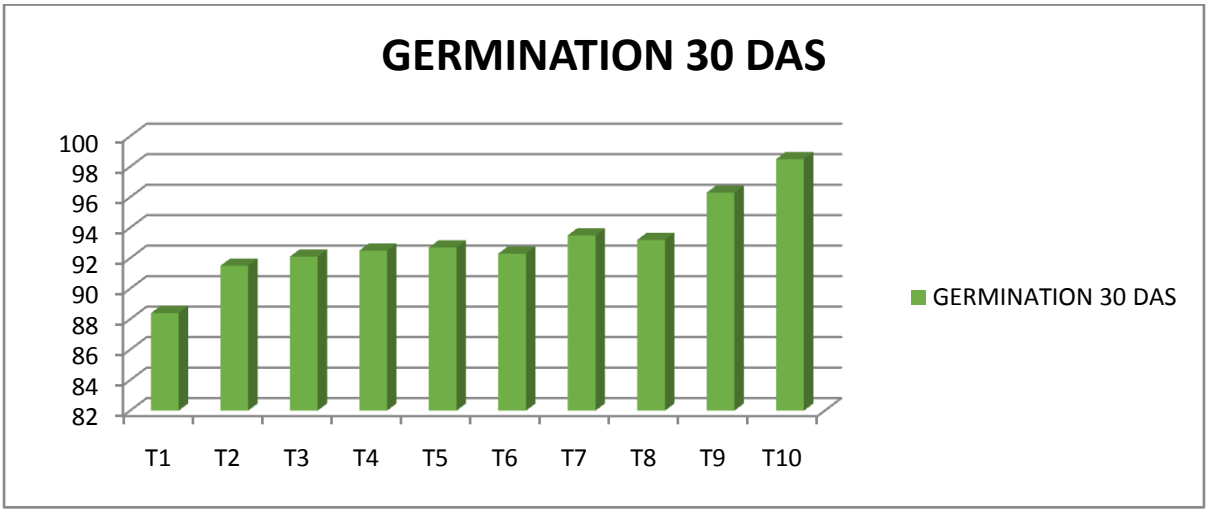
Germination study

Five different groundnut varieties were treated with ten different microbial fertilizers. One group (T1) maintained as control without microbial fertilizers. For TMV10 variety, the germination percentage ranged as 86.3±3.6% (T1) to 94.9±3.8% (T10) (Table 1). Similarly, for TMV12, TMV(Gn) 13, IGCV 00348 and VRI Gn6 varieties the germination percentage ranged as 86.8±3.1%(T1) to 95.4±3.9% (T10), 88.3±2.7%(T1) to 99.6±4.1% (T10), 82.5±1.9% (T1) to 94.5±2.2% (T10) and 85.7±1.1% (T1) to 95.7±1.9% (T10) respectively. Among the tested varieties with different microbial fertilizers, TMV (Gn) 13 showed significant germination percentage in the groundnut seedlings. In TMV(Gn)13 variety, the germination percentage were observed as 88.3±2.7, 90.8±3.6, 92.7±3.3, 92.8±3.5, 93.8±2.6, 94.8±3.0, 94.9±3.1, 95.4±3.1, 96.3±3.3, 97.9±3.8 and 99.6±4.1% for T1, T2, T4, T3, T5, T7, T6, T8, T11, T9 and T10 respectively. Groundnut propagates exclusively via seed, and this is a significant part of productivity (Grigoleto *et al.*, 2012). Germination tests conducted in this study revealed that the quality of groundnut seeds after harvesting might be impacted by the frequency of seed-borne fungus. According to Akonda *et al.* (2016), fungi such as *Fusarium* spp. and *A. niger* cause damage to the plumule, radicle, and hypocotyls of germinating seedlings, resulting in a decrease in percentage germination and seedling vigour. The 2% MI inoculation treatment resulted in the highest fresh (16.9 g) and dried (4.28 g) shoot weights, both of which were statistically different from the control (P=0.023 and P=0.025, respectively). In 2% of MI, fresh and dried root weights were both at their highest levels as 6.7 g and 2.23 g, respectively (Mridha *et al.*, 2016) which supported our microbial consortia (T10) results

Table: 1

BIOFERTILIZERS TREATMENTS	Germination (%)

T_1	88.4±1.2
T_2	91.5±0.86
T_3	92.1±1.03
T_4	92.5±1.53
T_5	92.7±1.21
T_6	92.3±1.40
T_7	93.5±1.18
T_8	93.2±1.07
T_9	96.3±1.16
T_{10}	98.5±1.49
T_{11}	96.7±1.23



Morphological parameters

Among the different ground nut varieties, TMV (Gn) 13 variety was used for further studies due to their high germination percentage. Shoot length and root length parameters of the TMV (Gn) 13 seedlings were analyzed at the end of the 30 days. The shoot length of the TMV (Gn) 13 groundnut seedlings were ranged as 5.76±0.23 to 11.30±1.45cm whereas the root length ranged as 2.08±0.08 to 6.66±0.27cm were observed (Table 2). The fresh and dry weight of the TMV (Gn) 13 groundnut seedlings were ranged as 1.72±0.11 to 8.95±0.20g/seedling and 0.56±0.09 to 3.91±0.16g/seedling respectively.

Among the different microbial inoculants treated as fertilizers, T10 group showed significantly increased shoot and root length as 11.30±1.45cm, 6.66±0.27. The fresh and dry weight of the TMV (Gn) 13 groundnut seedlings were observed as8.95±0.20 and 3.91±0.16g/seedling. The significant improved morphological parameters were due to the *Rhizobium*, *Phosphobacteria* and *AM* microbial proteins interaction with the phytohormones in the groundnut seedlings. The improved root growth in the treated seedlings may be the cause of the increased biomass output. Better root and shoot development as well as an increase in the number of phyllodes in the treated seedlings may also be caused by the microorganisms' secretion of growth-promoting chemicals (Khan *et al.*, 2011). Nevertheless, excessive chemical release might result in toxicity, which would reduce development, as was likely the case with seedlings treated with greater doses of MI (Khan *et al.*, 2014). The biologically active components of the inoculants, such as indole acetic acid (IAA) and gibberellins generated by *Rhodopseudomonas, Saccharomyces, Aspergillus* and *Lactobacillus* that promote plant development, may also be to blame for this promotion (Lim *et al.*, 1999). Root development (in terms of root length and diameter) increases a plant's ability and likelihood to absorb nutrients from a larger volume of soil, which results in more plant growth and biomass output (Chowdhury *et al.*, 1994).

Table: 2

BIOFERTILIZERS TREATMENTS	Root Length (cm)	Shoot Length (cm)	No. of Root Nodules
T_1	2.12±0.21	5.60±0.28	12.6±0.99
T_2	5.21±0.26	6.30±0.31	13.3±0.78
T_3	5.31±0.26	6.50±0.32	14.6±0.83
T_4	6.70±0.33	6.94±0.34	18.6±0.93
T_5	6.83±0.34	8.58±0.42	16.0±0.73
T_6	4.81±0.24	7.32±0.36	16.4±0.94
T_7	6.10±0.30	7.56±0.37	17.0±0.83
T_8	5.26±0.26	7.61±0.38	17.8±0.59
T_9	5.35±0.26	8.40± 0.42	16.0±0.63

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T_{10}	6.40±0.32	9.32±0.46	23.5±0.98
T_{11}	5.66±0.28	7.11±0.34	20.6±0.91

Table: 3

BIOFERTILIZERS TREATMENTS	**Fresh Weight (gm)**	**Dry Weight (gm)**
T_1	4.20±0.21	3.10± 0.22
T_2	5.20±0.26	5.07± 0.25
T_3	5.30±0.26	5.03±0.25
T_4	6.70±0.33	5.31±0.25
T_5	6.83±0.34	5.74±0.28
T_6	4.81±0.24	5.90±0.29
T_7	6.10±0.30	6.75±0.33
T_8	5.26±0.26	6.60±0.33
T_9	5.35±0.26	6.90±0.34
T_{10}	6.40±0.32	6.86±0.34
T_{11}	5.66±0.28	5.64±0.28

Table: 4

BIOFERTILIZERS TREATMENTS	**No. of Leaves**	**Total Leaf Area**
T_1	9.5±0.35	5.3±0.54
T_2	12.3±0.21	7.8±0.32

T_3	12.1±0.31	9.5±0.52
T_4	12.6±0.11	10.4±0.35
T_5	12.5±0.09	10.5±0.31
T_6	15.6±0.19	11.2±0.22
T_7	15.7±0.16	11.7±0.30
T_8	15.4±0.23	12.1±0.28
T_9	18.5±0.31	13.3±0.19
T_{10}	18.8±0.19	15.2±0.21
T_{11}	18.7±0.18	14.2±0.24

Conclusion

To guarantee their formation in the soil and practical use in conjunction with current cropping techniques, inoculants must be created and prepared. Microbial inoculants, which are primarily based on bacteria and fungi, are applied to soil as replacements for traditional inorganic fertilisers (bio fertilizers) or to perform certain tasks, such as bio control of diseases and pests (bio pesticides), bioremediation, and the improvement of soil properties. More effective inoculants products should result from new methods for strain selection and the formation of beneficial microbial consortia.

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