

Effect of Biological agents as substitutes for chemical seed treatment on groundnut cultivation

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Abstract

Field trials were conducted during the years 2009-2010, at a private farm in Puttaparthi, Anantapur District, India, to study the effect of biological agents as effective seed treatment substitutes over commonly used chemicals on ground nut plants, *Arachis hypogea*. The biological agents (BA) tested for this purpose involved, BGA, *Azotobacter* and *Azospirillum*. Seeds were treated prior to sowing with each of these 3 agents both individually and in combination. Commonly used chemical for seed treatment, was used as the control. The treated seeds were sown on the soil combination, 3:1 FYM: NPK. BGA proved to be the best among the 3 biological agents. The effect of *Azotobacter* was marginal when used alone. However, the same was augmented in the presence of BGA. *Azospirillum* seemed to have a negative effect on growth and yield, when applied alone and in combination with BGA.

Keywords: Biofertilizer; *Arachis hypogea*; yield FYM, NPK, BA, BGA

Introduction

With the introduction of green revolution technologies, modern agriculture is getting more and more dependent upon the steady supply of synthetic inputs (mainly fertilizers), which are products of fossil fuel (coal+ petroleum). Adverse effects are being noticed due to the excessive and imbalanced use of these synthetic inputs. This situation has led to identifying harmless inputs like biofertilizers. Use of such natural products like biofertilizers in crop cultivation will help in safeguarding the soil health and also the quality of crop products.

Biofertilizers are ready to use live formulations of such beneficial microorganisms which on application to seed, root or soil mobilize the availability of nutrients by their biological activity in particular, and help build up the microflora and in turn the soil health in general.

Biofertilizers are biologically active products or microbial inoculants of bacteria, algae and fungi (separately or in combination), which may help biological nitrogen fixation for the benefit of plants. Biofertilizers include the following, symbiotic nitrogen fixers *Rhizobium* spp. asymbiotic free nitrogen fixers (*Azotobacter*,

Azospirillum, etc.), algae biofertilizers (blue green algae or BGA in association with *Azolla*), phosphate solubilising bacteria, mycorrhizae, organic fertilizers.

Blue-green algae are considered the simplest, living autotrophic plants, i.e. organisms capable of building up food materials from inorganic matter. Certain blue-green algae live intimately with other organisms in a symbiotic relationship. Some are associated with the fungi in form of lichens. The ability of blue-green algae to photosynthesize food and fix atmospheric nitrogen accounts for their symbiotic associations and also for their presence in paddy fields. Blue-green algae are of immense economic value as they add organic matter to the soil and increase soil fertility. Also, they have been shown to have anti-fungal activity. This makes them suitable to be used as biofertilizers and substitutes for chemicals used in seed treatment.

Nagaraj *et al.* (2001) have shown that 7.5 t/ha FYM + NPK (25:32:20) produced the highest pods and seed yield/ha. Subramaniyan *et al.* (2000) have shown that increasing organic manure in the form of FYM to 15t/ha, can maximize pods, seeds yield/ha in groundnut. Abd El Rasoul *et al.* (2002) have shown significant increase in peanut yield

using bio and organic fertilizers. Kumaran (2001) has shown in his investigation on response of groundnut to organic manure & fertilizer levels that utilization of recommended dose of NPK (25:75:37) + 10 t/ha FYM gave the highest number of pods/plant, haulm and seed yield. Venkataramana *et al.* (1990) showed significant increase in pods, seeds and haulm yields /ha by FYM upto 15 t/ha. The integrated nutrient management by bio-organic and chemical fertilizers has recorded 35% increase in yield by Thakare *et al.* (2003). Laxminarayana *et al.* (2004) have also shown significant improvement in groundnut yield on application of organic and inorganic manures.

The use biofertilizers along with FYM has also been in studied. Use of BGA for rice cultivation is worth mentioning. El Habbasha *et al.* (2005) have shown that use of bio-fertilizers have improved the yield of groundnuts considerably. Badole *et al.* (2001a, 2001b & 2004) have stated that combined application of 5t/ha FYM + $\frac{3}{4}$ recommended dose of NPK increased no of pods/plant, seed index, seed protein & oil content. They have the same result on addition of *Azotobacter* as biofertilizer. El kramany *et al.* (2007) have shown that combining $\frac{1}{4}$ recommended dose of NPK with $\frac{3}{4}$ FYM and bio fertilizer microbein resulted in highest seed & pod yield, oil and protein content in poor sandy soils.

In India, losses in the range of 10 to 50% have been reported due to fungal attack on ground nut by Tikka disease. Tikka disease is associated with rust the losses may reach upto 10 % (Bhale *et al.*, 1998).

Groundnut (*Arachis hypogea*) is considered one of the most important oilseed crops in the world. Being the main crop grown in Rayalseema District of Andra Pradesh, the cultivated area of groundnut in Puttaparthi, Anantapur district is quite high. Owing to poor soil conditions, use of chemical fertilizers has increased greatly in this area. Hence, the coincident application of organic manures and bio-fertilizers is frequently recommended for improving biological, physical and chemical properties of soil and also to increase the productivity.

This work in an attempt to try to use BGA, along with *Azotobacter* and *Azospirillum* as substitutes for chemical seed treatment.

Materials and Methods

A series of field trials were carried out in a farm in Puttaparthi Mandal, Anantapur District, during both the growing periods, Jan – May and July – Dec, each year. Soil was cleaned and doweeded prior to cultivation.

The effect of algal agents comprising of blue green algae was tested against nitrogen fixers like *Azospirillum* & *Azotobacter* and chemicals such as Bavistin® commonly used for seed treatment.

The experimental land was divided into blocks. The soil was treated with 75% FYM and 25% NPK.

The seeds were treated with 50% Bavistin®, at 2g/Kg seed. Seeds were also treated with chosen biological agents, at the rate of 2.5 g/Kg of seeds for *Azotobacter*, *Azospirillum* and 2g/Kg for BGA. A solution was prepared by mixing the bio-fertilizers in water in the ratio 1:5. The seeds were soaked in the bio-fertilizer solution overnight and seeds were shade dried.

The experiment was carried out in RBD design with split plot arrangement using three replications. Each plot had 5 rows (4 cm in length and 50 cm apart). Seeds were sown 10cm apart in third week of Jan (season1) and July (season 2). 12 seeds were sown in each row in the plots, thereby maintaining 60 plants for each treatment. Each plot received seeds treated with particular biological agent.

Commercial samples of all biological agents were obtained from Agricultural Bacteriologist, College of Agriculture, Pune. Gypsum was added to the crops at the end of 60 days and 75 days.

The different parameters mentioned below were assessed at harvest, 120 days after plantation.

1. Pod yield– no of pods/plant; 2. Seed yield - no of seeds/pod; 3. quality of produce – no of good seeds/pod; 4. weight of pods/plant (g/plant); 5. seed index – 100 seed weight (g); 6. protein and oil content of seeds – seed weight X oil% or protein in seed. Oil and protein% in seeds were calculated as described by Chapman *et al.* (1978). 7. Disease-

scoring – The disease severity was recorded at 60 and 75 DAS as per the modified scale given by Subbarao *et al.* (1990).

All the data was statistically analysed. The least significant difference was used to compare means.



Control



BGA



BGA + Azotobacter

Results and Discussion

This experiment dealt with studying the effect of different biological agents over chemicals used for seed treatment and comparing the effect of different biological agents as an effective seed treatment alternative.

1. Yield: At the outset, among the different biological agents, Azotobacter and BGA have proved to better replacements to chemicals in terms of yield. Use of Azospirillum resulted in crippled plants, with shrunken pods, thereby reducing the yield of the plant.

Plants grown from seeds treated with Azotobacter alone, retained yield values same as the ones grown from chemical treatment. However, a slight improvement to 27.3 pods/plant and 51.9 seeds/plant was observed in the combined treatment of Azotobacter + BGA, from 24.4 pods/plant and 30.9 seeds/plant in the chemical treatment (Table 1).

Table 1. Treatment combinations for seed treatment

Treatment No	Treatment
CONTROL	Chemical seed treatment with Bavistin®
T1	Only BGA
T2	Only <i>Azotobacter</i>
T3	Only <i>Azospirillum</i>
T4	BGA + <i>Azotobacter</i>
T5	BGA + <i>Azospirillum</i>
T6	<i>Azotobacter</i> + <i>Azospirillum</i>
T7	BGA + <i>Azotobacter</i> + <i>Azospirillum</i>



BGA + Azotobacter

BGA

BGA proved to be the most effective among all agents used, recording an increase in the yield of seeds/plant as well as pods/plant. A highest of 30.1 pods/plant and 67.7 seeds/plant was recorded in plants grown from seeds treated with BGA, as against 24.4 pods/plant and 39.0 seeds/plant obtained from the control, treated with Bavistin® (Table 2). Apart from an increase in the no of pods/plant, presence of 3 seeds in each pod was a common feature in this treatment as against other treatments.

Table 2. Effect of Seed treatment on Yield, Weight and Nutritive value of *Arachis hypogea*

Treatment	Yield		Weight		Nutritive value	
	Pods/plant yield	seeds/plant yield	pod weight (g)	seed weight (g)	protein content	oil content
CONTROL	24.4	39.0	56.3	35.2	25.3	55.0
T1	30.1	67.7	109.3	48.5	30.5	67.6
T2	25.1	42.8	62.1	36.5	25.8	55.5
T3	12.4	13.7	24.7	22.0	19.1	39.4
T4	27.3	51.9	80.4	42.3	28.4	59.8
T5	18.1	24.3	30.0	22.3	20.3	39.8
T6	18.9	28.3	36.3	24.4	19.5	39.2
T7	21.7	33.6	35.9	23.1	22.4	45.0
S.Em ±	2.31	3.97	5.21	3.10	2.00	3.18

2. Weight of seeds – In consensus to yield, *Azotobacter* and BGA proved to be better than Bavistin® in terms of weight of seeds and pods. Again, owing to shrunken seeds in *Azospirillum* treatment, lower weights of seeds and pods were observed.

Azotobacter showed a slight increase in weight of seeds and pods over the control. However the effect was augmented in the presence of BGA, raising the values to 59.8g/plant.

BGA treatment proved to be most effective for increasing the weight of pods as well as seeds, (Table 2). No diseased or shrunken seeds were observed in these plants. The weight of seeds/plant was drastically enhanced from 56.3g/plant to 109.3g/plant (Fig.2).

Azospirillum treatment resulted in relatively shrunken seeds, smaller in size than the control, thereby reducing the weight of seeds and pods as well. Addition of BGA to *Azospirillum*, improved its quality slightly, though not significantly.

3. Protein & Oil content – Maximum protein and oil content of the seeds was recorded in BGA treatment, of the value, 30.5 and 67.6 resp as against that in control of 25.3 and 55.0 resp. for protein and oil content (Table 2).

The effect of *Azotobacter* treatment was parallel to that obtained when treated with Bavistin® and a slight improvement in the values was recorded when *Azotobacter* was combined with BGA, from 25.8 and 55.5 for protein and oil content when used individually to 28.4 and 59.8 rep when used in combination.

4. Physiological features - Use of biological agents hastened the process of emerging, flowering and maturation in *Arachis hypogea*, in comparison to Bavistin®.

The number of days required for plant emergence has been reduced drastically in BGA application over all other treatments. It has been reduced by half, from 12 days in the control to 6 days in BGA application. Similarly, the days for flowering has also been reduced greatly to 26 days as against the average of 35 days for Bavistin® and other agents, giving the crop an advantage. (Table 3)

While, *Azotobacter* could reduce the days to maturity by 10 days and *Azospirillum* by 14 days, BGA reduced the number of days for crop maturity by almost 30days (Fig. 4). The economical value of the crop has been thus increased manifold with application of BGA.

5. Disease Resistance – Groundnut plants are prone to fungal attack by *Cercospora arachidicola* as leaf spot. Tikka disease is associated with rust the losses may reach upto 10 %. Pod yield is determined by the duration of which leaves remain healthy and related with the time of defoliation. The disease also affects the quality of pods. It has been reported that apart from magnesium deficiency, increased application of nitrogen and phosphorus increases the incidence of infection.

Disease rating was scored at 2 intervals, end of 45 days from sowing and again at the end of 75 days. In both the study periods, treatment with Bavistin® recorded a score of 5.22 and 5.11

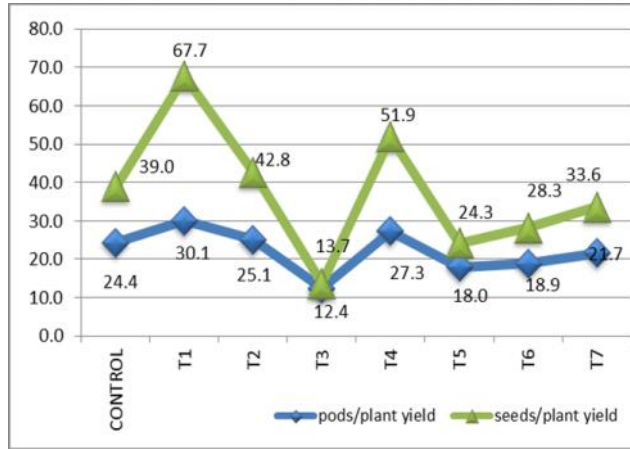


Fig. 1. Effect of Seed treatment on yield

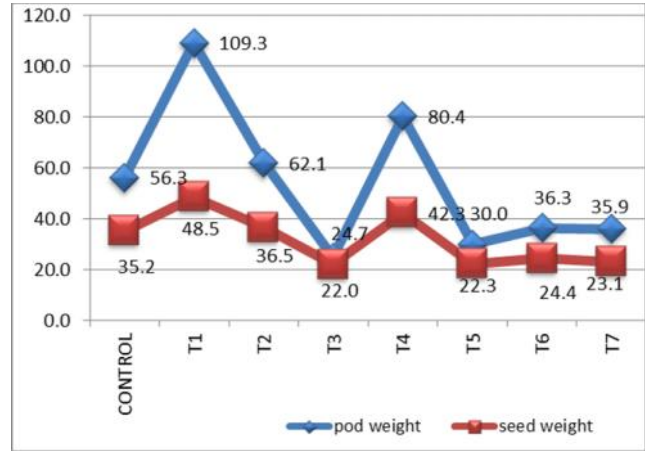


Fig. 2. Effect of Seed treatment on weight



Fig. 3. Effect of Seed treatment on oil and protein

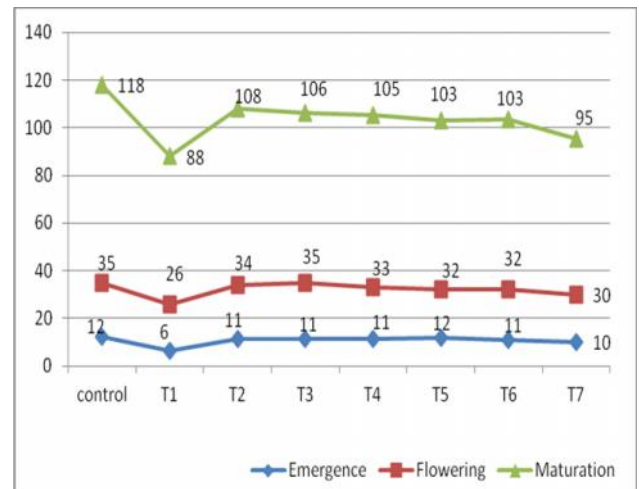


Fig. 4. Effect of Seed treatment on physiological features

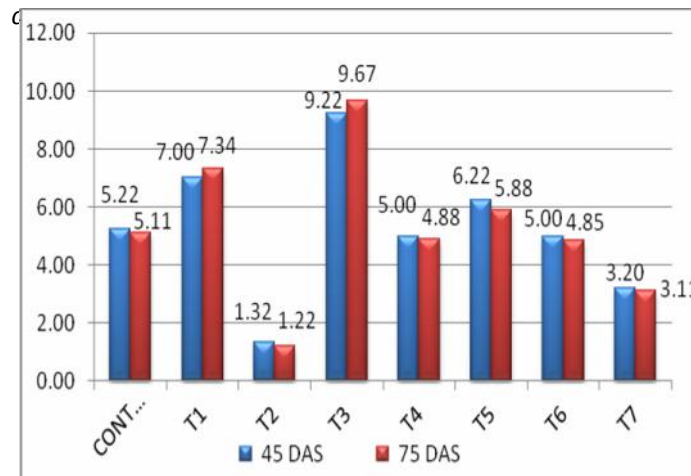


Fig. 5. Effect of Seed treatment on % Disease Index

Table 3. Effect of Seed treatment on Disease index and physiological parameters of *Arachis hypogea*

Treatment	% Disease Index		Physiological features		
	45 DAS	75 DAS	Days to emergence	Days to flowering	Days to maturation
CONTROL	5.22	5.11	12	35	118
T1	1.32	1.22	6	26	88
T2	7.00	7.34	11	34	108
T3	9.22	9.67	11	35	106
T4	5.00	4.88	11	33	105
T5	6.22	5.88	12	32	103
T6	5.00	4.85	11	32	103
T7	3.20	3.11	10	30	95
S.Em ±	1.23	1.22	2.12	2.19	2.03

respectively. Both *Azotobacter* and *Azospirillum* showed increased incidence of the disease, as indicated by the disease score of 7.00 and 9.22, 45 after sowing for *Azotobacter* and *Azospirillum* and an increase to 7.34 and 9.67 for both at 75 days after sowing (Table 3).

In BGA treatment, however, it was witnessed that, the incidence of leaf-spot disease very low, giving rise to healthy plants with a dense growth accompanied by healthy pods. The disease score recorded in case of BGA proved to be much lower than Bavistin®, of the order 1.32 in 45 days after sowing and 1.22 in 75 days after sowing.

Further, the diseased sate of the plant was reduced to great extent when BGA was used along with *Azospirillum* and *Azotobacter*, bringing them down to almost 6.22 in 45 days after sowing and 5.88 in 75 days after sowing when BGA was combined with *Azospirillum* and 5.00 in 45 days after sowing and 4.88 in 75 days after sowing when BGA was combined with *Azotobacter* as against their original values (Fig.5).

The combined application of BGA with *Azotobacter* and *Azospirillum* has further improved the % disease index to 33.20 and 3.11 at 45 days and 75 days after sowing, much lesser than that obtained with Bavistin®, thereby suggesting the positive role of biological agents over chemicals to be a potent fungicide.

Conclusion

BGA has proved to be a very effective seed treatment replacement to chemicals used for the

same, by enhancing the yield, weight, protein and oil content and also inducing resistance to Tikka disease. The resistance to Tikka disease imparted by BGA can be attributed to primarily 2 reasons, a) anti-fungal effect of BGA: b) early maturation of plants, thereby providing lower exposure to the fungal attack. However, the exact reason can known only by way of further investigation.

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