Jessy. M D A. No. 219

Short scientific report *Journal of Plantation Crops, 2010, 38 (3): 223-227*

Effect of biofertilizers on soil microbial population and growth of young rubber (*Hevea brasiliensis*) plants

(Manuscript Received: 18-02-09, Revised: 16-05-10, Accepted: 25-10-10)

Keywords: Biofertilizers, *Hevea brasiliensis*, growth, microbial population

Although the phenomenon of microorganisms increasing availability of nutrients to plants was well known for a long time, the need for its commercial exploitation in agriculture was felt not very back. Increased awareness about soil health and sustainability and other environmental effects associated with input intensive agriculture has lead many farmers to look for alternate agricultural practices with minimum use of agrochemicals. Escalating price of fertilizers coupled with their non-availability during critical periods of crop requirement also necessitate use of other sources of mineral nutrients. Some bacteria and fungi are able to improve plant growth by solubilizing sparingly soluble inorganic and organic phosphates in the soil (Babana and Antocin, 2006). Microorganisms like *Azospirillum* have the potential to stimulate growth and development of plants by nitrogen fixation, hormone effects and root development (Mostajeran and Amooaghaie, 2007).

In rubber, girth of seedlings which received 50 per cent of the recommended dose of nitrogen with *Azotobacter* inoculation was reported to be comparable to plants which received 100 per cent of the recommended chemical fertilizer alone (Joseph *et al.*, 1997). However, the beneficial effect of different biofertilizers on soil microbial population and growth of plants in the main field was not studied so far. Hence, experiments were conducted in seedling nurseries and young rubber plantations to study the effect of different biofertilizers on soil microbial population and growth of plants.

Two experiments were conducted in seedling nurseries and one experiment in young rubber plantations at four locations.

Experiment I: Effect of phosphorus solubilizers on microbial population and growth of rubber seedlings

The experiment was conducted at two locations, Central nursery, Karikkattoor and Regional nursery, Perumpulickal and was repeated during two years. The treatments were different combinations of rock phosphate with P solubilizers viz., rock phosphate alone without P solubilizers, 75, 50 and 25 per cent rock phophate with P solubilizers, P solubilizers alone and a control without P solubilizers or rock phosphate. N, P and Mg were applied uniformly to all the plants as chemical fertilizers. During both the years, 10 kg biofertilizers was mixed with 100 kg cow dung and applied in one hectare at the time of bed preparation. The experimental design was RBD with four replications. Germinated seeds were planted at a spacing of 30 cm x 30 cm. Plot size was 4.14 m^2 and observations were recorded from 16 net plants excluding the border rows. Soil samples were collected before the commencement of the experiment for chemical analysis. Diameter of seedlings was recorded 4 and 8 months after planting and microbial population in the soil was recorded one, two and four months after biofertilizer application.

Experiment II: Effect of nitrogen biofertilizers on soil microbial population and growth of rubber seedlings

The experiment was conducted at Central nursery, Karikkattoor and was repeated during two years. There were 14 treatments, *Azotobacter* and *Azospirillum* from three sources (A- Indian Organic Chemicals, B- National biofertilizers and C- T Stanes), at 25 and 50 per cent replacement of N fertilizer, full dose of N as chemical fertilizer and a control without N fertilizer application.

In all the treatments, P, K and Mg were applied uniformly as chemical fertilizers. During both the years, 10 kg biofertilizers was mixed with 100 kg cow dung and applied in one hectare at the time of bed preparation. The experimental design was RBD with four replications. Germinated seeds were planted at a spacing of 30 cm x 30 cm. Plot size was 4.14 m^2 and observations were recorded from 16 net plants excluding the border rows. Soil samples were collected before the commencement of the experiment for chemical analysis. Diameter of seedlings was recorded five and six months after planting during first year and five and eight months after planting during second year. Soil microbial population was recorded one, two and three months after biofertilizer application in both years.

Experiment III: Effect of biofertilizer application on growth of young plants in the main field

The experiment was conducted at four locations in Central Kerala, Veloor, Peroor, Thodupuzha and Erumely. Chemical fertilizers were applied in one block and biofertilizers were applied in another block at the time of planting. In all the locations, the clone planted was RRII 105. Girth of 30 plants in each block was recorded at 15 cm height. Girth was recorded 6 months after planting and one year after planting at Thodupuzha, Erumely and Peroor, while at Veloor girth was recorded two years and three years after planting also. Soil samples (0-30 cm) were collected before commencement of the experiment to study the soil chemical properties. Soil samples (0-15 cm) were also collected for microbial analyses before commencement of experiment and after one, three and six months after biofertilizer application. A consortium of biofertilizers containing *Azospirillum*, *Azotobacter* and phosphorus solubilizers (15 kg) was mixed with 50 kg dried cow dung and 50 kg top soil and applied in one hectare (450 plants). Biofertilizers were incorporated into the soil at 30- 50 cm away from the plant base in a band.

In nurseries, analysis of soil samples indicated that organic carbon and available P status were in the medium range, available K status low and acidic reaction at both the locations (organic carbon content - 1.27 %, available P- 2.5 mg/100 g, available K – 2.50 mg/100g and pH- 4.7 at CN, Karikkattoor and the corresponding values were 1.12, 1.20, 2.50 and 4.4 at RN, Perumpulickal). In all the young rubber plantations, soil organic carbon status was in the medium range, available P and K status low, and soil was acidic in reaction (Table 1).

There was no significant difference between treatments with respect to the diameter of seedlings in experiment I (Table 2). The superior effect of biofertilizers over chemical fertilizers was not indicated at both locations and both the seasons. There was no significant difference between control and fertilizer applied treatments also. This might be due to the residual effect of rock phosphate applied during the previous years. Lack of consistent response to application of P solubilizers was

Table 1. Soil nutrient status in young rubber plantations

Location	Org. C (%)	P	K	Mg	pH
			Mg/100 g soil		
Veeloor	0.93	0.71	3.69	0.42	4.6
Peroor	0.96	0.50	2.85	0.92	4.6
Erumely	0.86	T	2.25	2.0	5.7
Thodupuzha	0.82	T	1.00	1.5	5.6

Table 2. Influence of P biofertilizer application on diameter (mm) of rubber seedlings

Treatments	RN, Perumpulickal Months after plating				CN, Karikkattoor Months after plating			
	Year I		Year II		Year I		Year II	
	4	8	4	8	4	8	4	8
Control	7.20	11.13	7.13	9.60	6.50	10.66	5.18	8.68
P solubilizers alone	7.59	11.05	6.90	9.25	6.88	10.78	4.98	8.47
25% P as chem. fert +P solubilizers	8.49	11.35	7.13	9.40	6.93	10.79	5.05	8.38
50% P as chem. fert +P solubilizers	7.71	11.46	7.05	8.88	7.28	10.86	5.05	8.66
75% P as chem. fert +P solubilizers	7.89	11.83	7.35	9.50	6.68	10.37	5.12	8.80
100% P as chem. fert	7.50	11.00	7.20	9.10	6.57	10.71	5.00	8.40
SE	0.36	0.43	0.35	0.41	0.18	0.38	0.11	0.15
CD (P=0.05)	NS	NS	NS	NS	NS	NS	NS	NS

NS = Not significant

earlier reported by Tandon (1987), positive effect was reported in only 10 out of 37 experiments. Kucey *et al.* (1989) also reported large variations in the effectiveness of P solubilizers. The population of P solubilizers was the highest one month after biofertilizer application at both the locations and decreased subsequently in all the treatments (Fig.1 and Fig. 2). The population of P solubilizers was higher in the treatment which received full dose of P fertilizer as rock phosphate compared to control which did not receive rock phosphate or biofertilizer.

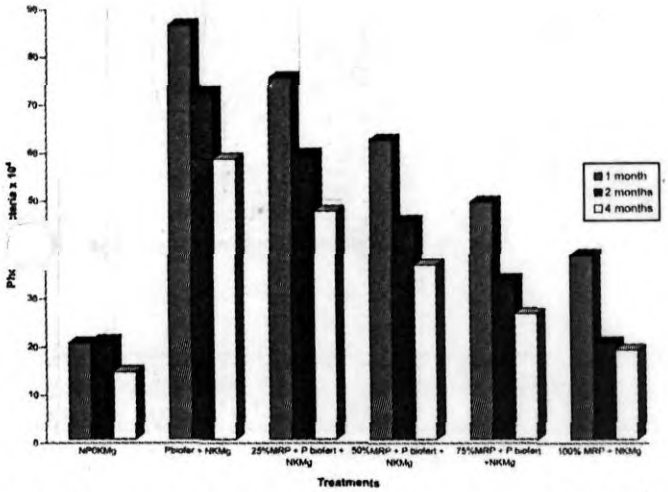

Fig. 1. Population of phosphobacteria (cfu/g soil) as influenced by P biofertilizer application in CN, Karikkattoor

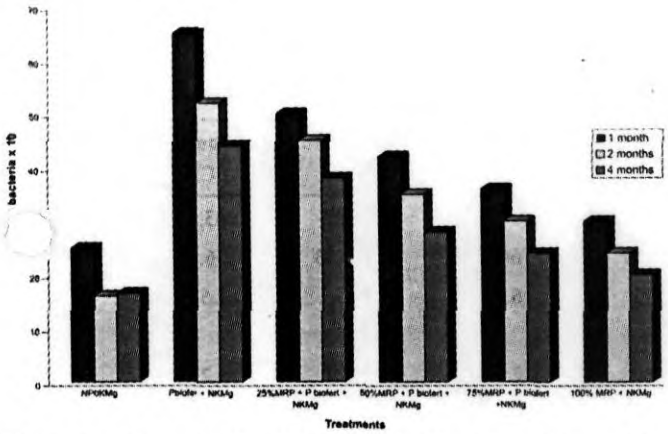

Fig. 2. Population of phosphobacteria (cfu/g soil) as influenced by P biofertilizer application in RN, Perumpulickal

In experiment II also, there was no significant difference between treatments with respect to the diameter of seedlings during both the years (Table 3). There was no response to application of N as chemical fertilizer also. However, the population of total bacteria, *Azotobacter* and *Azospirillum* was higher in the biofertilizer applied

Table 3. Influence of N biofertilizer application on diameter (mm) of seedlings

Treatments	Year I		Year II	
	5 MAP	6 MAP	5 MAP	8 MAP
AB a + 50% N	6.43	7.35	5.40	8.60
AB b + 50% N	6.27	7.23	5.13	8.70
AB c + 50% N	6.60	7.79	4.86	8.78
AS a + 50% N	6.23	7.77	5.00	8.79
AS b + 50% N	6.67	7.93	4.75	8.49
AS c + 50% N	6.33	7.40	4.81	8.10
AB a + 25% N	6.60	7.77	5.20	8.84
AB b + 25% N	6.27	7.40	4.89	8.61
AB c + 25% N	6.90	7.90	5.26	8.71
AS a + 25% N	6.70	8.30	4.92	8.90
AS b + 25% N	7.03	8.43	5.00	8.97
AS c + 25% N	6.43	7.37	4.89	8.91
100% n as CF	6.17	7.00	4.93	8.50
No N	6.40	7.87	4.81	8.37
SE	0.25	0.29	0.16	0.31
CD (P=0.05)	NS	NS	NS	NS

a - Indian organic chemicals Ltd, b - National biofertilizers c - T Stanes, AB - *Azotobacter* AS - *Azospirillum*, CF - Chemical fertilizer MAP = Months after planting NS = Not significant

treatments. *Azotobacter* counts were recorded only in plots which received biofertilizers containing *Azotobacter* (Table 4). Though the microbial population in the biofertilizer applied plots was higher one month after application, there was a decline in the population subsequently. This indicates the need for periodic fortification with biofertilizers to sustain soil microbial population at a higher level.

In experiment III, the effect of biofertilizer application varied between locations (Table 5). At Thodupuzha, plants supplied with chemical fertilizers had significantly higher girth compared to plants supplied with biofertilizers after one year. At Erumely, though chemical fertilizers applied plants had a significantly higher girth compared to biofertilizer applied plants after six months, both were comparable after one year. At Veloor and Peroor, there was no significant difference between treatments with respect to girth of plants. Compared to the pretreatment value, population of total bacteria and P solubilizers was higher after biofertilizer application at all the locations (Table 6). Population of non-symbiotic nitrogen fixers was observed only in biofertilizer applied plots. The microbial population followed the same trend as that of the other experiments,

Table 4. Soil microbial population (cfu/g soil) as influenced by N biofertilizer application in seedling nursery

Treatments	1 month after biofertilizer application			2 months after biofertilizer application			3 months after biofertilizer application		
	TBx10⁴	AS 10³	ABx10²	TBx10⁴	ASx10³	ABx10²	TBx10⁴	ASx10³	ABx10²
AB a + 50% N	260	2.1	8.2	255	1.5	7.5	224	1.0	4.5
AB b + 50% N	235	2.4	7.6	210	1.0	6.0	180	0.5	3.5
AB c + 50% N	255	1.9	9.1	215	1.5	8.6	147	1.0	3
AS a + 50% N	330	14.5		275	10.5		202	8.2	
AS b + 50% N	340	12.5		300	9.5		250	7.5	
AS c + 50% N	335	12		310	10		240	9.0	
AB a + 25% N	280	2.4	9.4	260	2	8.0	190	1.5	4
AB b + 25% N	295	2.8	8.8	244	2.3	7.5	173	1.8	4.5
AB c + 25% N	315	2.3	10.2	285	1.8	9.8	196	1.0	3
AS a + 25% N	375	15.2		345	10.6		265	8.4	
AS b + 25% N	365	13.9		350	12.2		265	9.0	
AS c + 25% N	380	12.5		340	10		279	8.0	
100% N as CF	195	2		174	2		144	1.5	
No N	255	2.5		220	1.9		209	1.8	

a - Indian organic chemicals Ltd, b - National biofertilizers, c - T Stanes, AB - *Azotobacter* AS - *Azospirillum*, TB - total bacteria, CF - Chemical fertilizer

Table 5. Influence of biofertilizer application on girth (cm) of young rubber plants

Location	6 months after planting			1 year after planting			2 years after planting			3 years after planting		
	CF	BF	t value	CF	BF	t value	CF	BF	t value	CF	BF	t value
Veloor				10.7	11.2	NS	16.9	16.8	NS	23.2	22.8	NS
Thodupuzha	5.2	4.6	NS	12.0	10.4	4.15**						
Erumely	6.2	5.6	2.02*	12.7	13.1	NS						
Peroor	8.4	8.4	NS	12.8	12.4	NS						

CF-chemical fertilizer BF- biofertilizer NS- Not significant

Table 6. Effect of biofertilizer application on soil microbial population (cfu/g soil) in young rubber plantations (mean of three locations)

Microorganism	Pretreatment		1 month after application		3 months after application		6 months after application	
	CF	BF	CF	BF	CF	BF	CF	BF
Fungus x 103	9.83	10.73	6.90	21.77	22.27	26.27	43.60	33.00
Total bacteria x 104	53.00	51.70	199.43	276.27	66.57	127.83	36.83	48.6
Phosphobacteria x 103	18.27	21.33	13.77	31.73	6.77	8.43	2.57	3.50
Non symb N fixers x 102				8.83		4.73		3.47
Actinomycetes x 104	10.53	11.50	4.33	13.50	2.07	21.27	5.23	3.57

CF - Chemical fertilizer BF - Biofertilizer

after the initial increase, the population decreased subsequently.

In all the experiments, application of biofertilizers temporarily enhanced soil microbial population. However, after the initial increase, the microbial population showed a declining trend subsequently. There was no significant difference between treatments in the seedling nurseries. In young rubber plantations, biofertilizer application was comparable with chemical fertilizers at Veloor, Erumely and Peroor and at Thodupuzha, chemical fertilizers was superior to biofertilizers with respect to girth of plants. The various experiments at different locations did not indicate a definite superiority of biofertilizers to chemical fertilizers.

However a possibility of reducing chemical fertilizer input with biofertilizers was indicated which needs further investigation.

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