

## Effect of Azophos (50:50), a biofertilizer formulation on cardamom (*Elettaria cardamomum* L. Maton) in primary nursery

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### ABSTRACT

An experiment was conducted in sandy clay loam soil to determine the influence of *Azospirillum*, *Azotobacter*, phosphobacteria (PSB), azophos (50:50) either individually or in combination, on growth and development of cardamom. Rate of germination and growth attributes such as shoot length, root length, number of leaves, chlorophyll pigments, plant fresh and dry weight were quantified. The results revealed that in general, earliness of germination was recorded in all biofertilizer treatments and germination percentage was higher in combined inoculation of *Azospirillum*, *Azotobacter* and phosphobacteria. This treatment improved shoot length, root length, number of leaves, plant biomass and chlorophyll content. Increment in growth attributes was lower in azophos (50:50) treatment in comparison with combined inoculation of *Azotobacter*, *Azospirillum* and phosphobacteria. However, growth attributes were on par when azophos (50:50) was combined with *Azotobacter* with that of *Azospirillum*, *Azotobacter*, phosphobacteria combinations.

**Keywords:** Azophos, biofertilizer, cardamom, seed germination.

### INTRODUCTION

Cardamom (*Elettaria cardamomum* L. Maton) popularly known as “Queen of spices” is an export oriented spice crop of India. The Indian cardamom industry faces stiff competition from other countries in the world market and it has to sustain productively with superior quality of produce. To achieve success in cardamom plantation programme, promotion of seed germination and establishment of seedling is one of the important phases because propagation through seed is highly preferred as does not transmit the ‘Katte’, a viral disease. In order to make the seeds germinate, they must be placed under favourable environmental conditions. Seed treatment often improves germination through control of seed surface mold; it provide good insurance against disease, soil borne organisms etc., it affords protection to weak seeds and make them produce healthy seedlings. In cardamom attempts have been reported on hastening of germination of seeds through the application of gibberellin (Kalavathy and Ranganayaki, 1993). However, there are no comprehensive reports on the efficacy of biofertilizers on seed germination and growth in cardamom. Thus, the present study was undertaken to throw light on the influence of biofertilizers viz., *Azospirillum*, *Azotobacter* and phosphobacteria on growth and development of cardamom.

### MATERIALS AND METHODS

Cardamom (cv. Malabar) seeds were obtained from mother plants from Pandiyan Estates (P) Ltd., Vellimalai (+1090m above MSL), Tamil Nadu, from the selected uniformly ripened capsules. Native efficient acid tolerant isolates of *Azospirillum*, phosphobacteria (PSB), *Azotobacter*, obtained from the Vellimalai cardamom plantations were used in the present study. *Azospirillum*, *Azotobacter* and phosphobacteria were loaded in sterilized rice husk ash as carrier. Azophos (50:50) formulation was developed by mechanical mixing of the individual broths sterilized rice husk ash in equal ratio on volume basis after attain the sufficient growth (Premalatha, 2000). These rice husk ash based bacterial inoculants were treated with seeds @ 2 g per 10 g of seeds.

Following treatments were included in the present study

T<sub>1</sub>: *Azospirillum*; T<sub>2</sub>: PSB; T<sub>3</sub>: *Azotobacter*; T<sub>4</sub>: *Azospirillum* + PSB; T<sub>5</sub>: *Azotobacter* + PSB; T<sub>6</sub>: *Azospirillum* + *Azotobacter* + PSB; T<sub>7</sub>: Azophos (50:50); T<sub>8</sub>: Azophos (50:50) + *Azotobacter* and T<sub>9</sub>: Control

One hundred and twenty seeds (3.0 g) were sown separately in each treatment in lines. The recommended nursery management practices were followed for the production of elite seedlings. The observations such as germination

percentage, shoot length, root length, number of leaves, plant bio-mass and plant dry weight were recorded at various intervals. Chlorophyll a, b and total chlorophyll were quantified following the method of Mahadevan and Sridar (1986).

## RESULTS AND DISCUSSION

### Effect of biofertilizers on seed germination

The germination of seeds differed significantly due to the inoculation of biofertilizers (Fig. 1) both in Madurai and Vellimalai locations. In general early germination was observed (on 25 DAS) in the seeds inoculated with biofertilizers. Combined inoculation of *Azospirillum*, *Azotobacter* and PSB registered highest seed germination per cent of 65.8 and 86.7 when compared to 20 and 68.7% germination in control at Madurai and Vellimalai locations, respectively on 65<sup>th</sup> days after sowing. Enhanced seed germination due to co-inoculation of *Azospirillum* and phosphobacteria and combined inoculation of *Azotobacter*, PSB, SSB and ZSB observed in the present study corroborates with earlier report in coffee (Raghu, 2000). *Azotobacter* treatment either individually or in combination with other biofertilizers was found to augment germination. Our results were in agreement with those reported in coffee (Glory Swarupa *et al.*, 1997) and in chilli (Bhat and Alagawadi, 1998). Azophos (50:50) treatment recorded relatively lesser seed germination when compare with combined inoculation of *Azospirillum* and phosphobacteria. On the other hand, azophos (50:50) in combination with *Azotobacter* improved seed germination percentage on par to that of *Azospirillum*, *Azotobacter*, phosphobacteria

combination (T6). The increased seed germination might be due to auxin and other growth promoting substances released by these organisms. Earlier, Bottini *et al.* (1989) reported the production of gibberellic acid by *Azospirillum* whereas Barea *et al.* (1976) reported the production of gibberellic acid by phosphobacteria. Martinez-Toledo *et al.* (1988) reported the production of auxin, gibberellin and cytokinins by *Azotobacter chroococcum*.

### Effect of biofertilizers on shoot length, root length, number of leaves and plant biomass

In the present investigation, all the treatments irrespective of the inoculants resulted in vigorous growth of seedlings with increased root length, shoot length, number of leaves and plant biomass in comparison with control (Tables 1 and 2). Irrespective of location the combined inoculation of *Azospirillum*, *Azotobacter* and phosphobacteria had recorded highest shoot and root length, number of leaves and plant biomass when compared to the other treatments and un-inoculated control. Similar positive influence of biofertilizers on growth attributes were reported by Rajagopal and Ramarethinam (1997) and Glory Swarupa (1996) in coffee. Azophos (50:50) treatment recorded relatively lesser plant growth parameter values when compare with combined inoculation of *Azospirillum* and phosphobacteria which is in contrary to the findings of Premalatha (2000) who reported improved growth and development in rice as compared to inoculation of *Azospirillum* and phosphobacteria. This deviation may be positively due to the rhizosphere effect/ niche-ecosystems difference between the crops supported by the views of Roberts *et al.* (1992) that inoculants may differ in their ability to colonize different plants. The interactions inoculated microorganisms with other components of the soil biota have not been well characterized but will have a profound effect on the ability of the inoculant to establish in soil and on roots. The vast majority of seed exudates (sugars, sugar alcohols, amino acids, organic acids, and various volatiles and enzymes) released during germination would stimulate the proliferation of the diverse microbial community present on the surface of seeds.

The enhancement in growth parameters registered with combined inoculation of biofertilizers was possibly due to the accumulation or uptake of nitrogen by *Azospirillum* or *Azotobacter*; uptake of phosphorous owing to phosphobacteria and also through the production of growth promoting substances like phyto-hormones, vitamins and antibiotic substances by the inoculants.

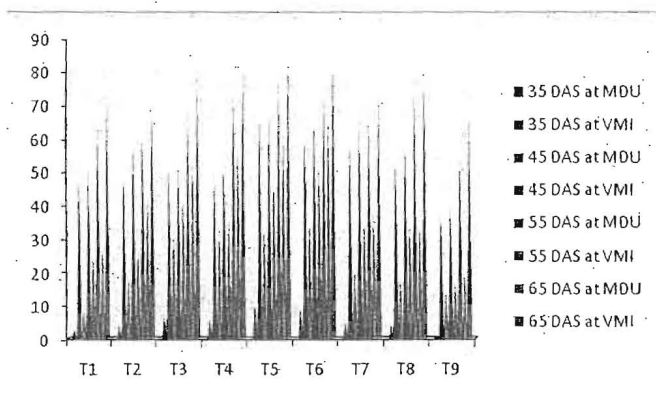


Fig. 1. Effect of biofertilizers on germination percentage in cardamom. Field experiment was conducted at Madurai (MDU) and Vellimalai (VMI) locations and rate of seed germination (%) after 35, 45, 55 and 65 days of sowing was represented.

**Table 1:** Effect biofertilizer inoculation on growth and development of cardamom in primary nursery at Madurai location

Treatments	Root length (cm)		Shoot length (cm)		No. of leaves		Plant biomass (g plant <sup>-1</sup> )		Plant dry weight (g plant <sup>-1</sup> )	
	DAS		DAS		DAS		DAS		DAS	
	100	125	100	125	100	125	100	125	100	125
T <sub>1</sub>	2.0	4.1	6.4	6.5	2.0	3.0	0.527	0.733	0.400	0.053
T <sub>2</sub>	2.3	3.5	6.1	6.9	2.0	3.0	0.560	0.693	0.043	0.050
T <sub>3</sub>	2.6	4.3	6.6	7.4	2.3	3.0	0.673	0.827	0.050	0.057
T <sub>4</sub>	2.5	4.3	6.3	7.2	2.0	3.0	0.713	1.067	0.050	0.073
T <sub>5</sub>	2.7	4.8	6.7	7.6	2.3	3.3	0.760	1.207	0.053	0.087
T <sub>6</sub>	2.8	6.4	6.7	7.7	3.0	4.0	0.847	1.747	0.060	0.120
T <sub>7</sub>	3.3	4.0	6.0	7.1	2.0	3.0	0.660	0.973	0.047	0.063
T <sub>8</sub>	2.9	4.9	6.5	7.4	2.3	3.7	0.787	1.603	0.057	0.113
T <sub>9</sub>	1.7	2.5	5.2	6.3	2.0	2.3	0.427	0.587	0.030	0.040
SEd	0.119	0.367	0.145	0.137	0.236	0.272	0.012	0.023	0.014	0.006
CD (0.05%)	0.245	0.758	0.300	0.284	0.487	0.562	0.026	0.047	0.030	0.040

DAS – Days after sowing

**Table 2:** Effect biofertilizer inoculation on growth and development of cardamom in primary nursery at Vellimalai location

Treatments	Shoot length (cm)		Root length (cm)		No. of leaves		Plant biomass (g plant <sup>-1</sup> )		Plant dry weight (g plant <sup>-1</sup> )		Chlorophyll content (mg g <sup>-1</sup> ) at 155 DAS		
	100 DAS	155 DAS	100 DAS	155 DAS	100 DAS	155 DAS	100 DAS	155 DAS	100 DAS	155 DAS	Chl- a	Chl- b	Total
T <sub>1</sub>	2.7	8.3	3.5	7.8	3.0	4.0	0.640	1.367	0.053	0.143	0.76	0.82	1.57
T <sub>2</sub>	2.4	8.5	3.5	9.5	3.0	4.3	0.820	2.037	0.067	0.210	0.69	0.84	1.53
T <sub>3</sub>	2.8	9.3	3.9	11.5	3.0	4.7	0.953	2.453	0.080	0.257	0.81	0.91	1.71
T <sub>4</sub>	3.0	10.0	3.9	11.6	3.0	5.3	1.050	2.873	0.087	0.300	0.91	1.09	1.99
T <sub>5</sub>	3.2	11.9	4.2	12.5	3.3	5.7	1.140	3.807	0.093	0.420	0.84	1.11	1.96
T <sub>6</sub>	3.3	12.7	4.3	13.3	3.3	6.0	1.530	4.400	0.107	0.450	0.94	1.30	2.24
T <sub>7</sub>	3.0	9.2	3.9	8.3	3.3	4.70	0.917	2.603	0.073	0.267	0.74	0.93	1.67
T <sub>8</sub>	3.1	12.0	3.8	12.3	3.3	5.3	1.187	3.433	0.093	0.373	0.78	1.14	1.92
T <sub>9</sub>	2.1	5.8	2.8	4.0	2.3	4.0	0.453	0.973	0.033	0.100	0.49	0.48	0.97
SEd	0.14	0.38	0.21	0.87	0.41	0.41	0.019	0.055	0.003	0.006	0.03	0.04	0.07
CD (0.05%)	0.28	0.78	0.43	1.80	0.84	0.83	0.038	0.112	0.005	0.013	0.07	0.09	0.15

DAS – Days after sowing

**Effect of biofertilizers on chlorophyll content**

The results revealed that chlorophyll content of inoculated seedlings recorded a marked increase as compared to un-inoculated control (Table 4). Combined inoculation of *Azospirillum*, *Azotobacter* and phosphobacteria recorded a higher total chlorophyll (2.2 mg g<sup>-1</sup>) and chlorophyll - b (1.3 mg g<sup>-1</sup>) at 155 DAS and the un-inoculated control recorded least total chlorophyll (0.97 mg g<sup>-1</sup>) and chlorophyll - b (0.48 mg g<sup>-1</sup>). The increase in chlorophyll - b content was relatively higher than chlorophyll - a. Our results were in agreement with those observed in coffee by Raghu (2000).

**CONCLUSION**

The results of the present study clearly indicate that combined inoculation of biofertilizers will be simple, less expensive and a promising technology for hastening germination of seeds and production of vigorous seedlings

which can be transplanted to secondary nursery with in a short period in primary nursery. Study also standardized the ratio of the mixed formulation of biofertilizers with respect to germination, growth and development in cardamom.

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**REFERENCES**

- Barea, J.M., E. Navare and E. Montoya. 1976. Production of plant growth regulators by rhizosphere phosphate solubilizing bacteria. *Journal of Applied Bacteriology*, **40**:129-134.

- Bhat, J.M. and A.R. Alagawadi, 1998. Seed borne nature of *Azotobacter chroococcum* in chilli (*Capsicum annum*) and its role in seed germination and plant growth. *Hindustan Antibiotic Bulletin*, **40(1-4)**: 20-30.
- Bottini, R., M. Fulchieri, D. Pearce and R.P. Pharis. 1989. Identification of gibberellins A<sub>1</sub>, A<sub>3</sub> and isoA<sub>3</sub> in cultures of *Azospirillum lipoferum*. *Plant Physiology*, **90**: 40-47.
- Glory Swarupa, S. 1996. Study on the effect of biofertilizers on the growth of C X R coffee seedlings. *Journal of Coffee Research*, **26(2)**: 62-66.
- Glory Swarupa, S., T. Basavaraj Naik, A.G.S. Reddy and T. Raju. 1997. Effect of biofertilizers on germination of coffee seed. *Indian Coffee*, **LXI (3)**: 3-5.
- Kalavathi, D. and P.R. Renganayaki. 1993. Effect of GA on seed germination in cardamom. *South Indian Horticulture*, **41**: 123-124.
- Mahadevan, A. and R. Sridhar. 1986. Methods in physiological plant pathology. Sivakami publications Madras. p.328.
- Martinez-Toledo, M.V., T. de La Rubia, J. Moreno and J. Gonzalez-lopez. 1988. Root exudates of *Zea mays* and production of auxins, gibberellins and cytokinins by *Azotobacter chroococcum*. *Plant and Soil*, **110**: 149-152.
- Premalatha. 2000. Studies on a new mixed biofertilizer formulation, azophos and its effect on rice. M.Sc.(Ag.) Thesis submitted to Tamil Nadu Agricultural University, Coimbatore. p.139.
- Roberts, D.P., C.J. Sheets and J.S. Hartung. 1992. Evidence for proliferation of *Enterobacter cloacae* on carbohydrates in cucumber and pea spermosphere. *Canadian Journal of Microbiology*, **38**: 1128-1134.
- Raghu, R. 2000. Studies on the biofertilizer consortium for coffee. M.Sc. (Ag.) Thesis submitted to Tamil Nadu Agricultural University, Coimbatore. p.164.
- Rajagopal, B. and S. Ramarethinam. 1997 Influence of combined inoculation of biofertilizers and digested organic supplement on the growth and nutrient uptake in organic tea cultivation. *Planter's chronicle*, **15**: 7-14.