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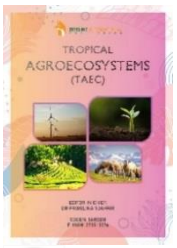

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RESEARCH ARTICLE

BIOINOCULATION RESPONSE ON GROWTH OF *ADENENTHERA PAVONINA* SEEDLINGS UNDER NURSERY CONDITIONS

Sujata Dash, Nibha Gupta

Regional Plant Resource Centre, Bhubaneswar -751015 Odisha, India.
****Corresponding Author Email: nguc2003@yahoo.co.in***

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ABSTRACT

The present study consists of the experiments on application of mineral solubilizing and plant growth hormone producing microbial strains in individual and /or combination under glass house conditions and their impact on enhancement in plant productivity of *Adnenthera pavonina*. Among microbial strains taken for the study, 6 phosphate solubilizing fungi, 4 iron ore leaching fungi, 5 phsophate solubilising bacteria and 11 isolates of Rhizobia had been used. Screening of ten fungal species and 5 bacterial isolates for their effects on growth of *Adnenthera pavonine,* done under pot culture in green house conditions, elucidated the best performance of Penicillum *crysogenum* Thom. 1 and Cunninghmella *elegans* Lendn. Combination of selected microbial inoculants for the individual test tree species were evaluated for their individual performance and /or in combination with the other selected inoculants in specific ratio of their inoculums. Under the dual inoculation experiment, selected mineral solubilisers (fungi/bacteria) were evaluated along with the combination of different isolates of *Rhizobium* and resulted selection of *Penicillium chrysogenum* and Cunnighmella *elegans* in 1:1 combination for better growth and development of seedlings under nursery conditions. The impact of Rhizobium inoculation alone or in combination with these fungal strains could not be observed and interpreted. However, the outcome the present study done under controlled experimental conditions may provide the plausible approach towards the development of nursery package and practice for development of quality planting material of this tree speices may be useful for the transplantation programe of revegetation of degraded land and environment.

KEYWORDS

Adenanthera pavonina, Microbial inoculants, Rhizobium & Phosphate solubilizing microbes.

1. INTRODUCTION

Adenanthera pavonina (L.) (family Leguminosae, subfamily Mimosoideae) has long been an important woody tree in Southeast Asia and the Pacific Islands and economically important in agroforestry, fuel wood, shade and avenue plantation (Atkins 1994; Clark and Thaman 1993; Benthall 1946). It has also been useful as green manure and fodder and also has antiseptic, anti inflammatory and medicinal properties. Soil microorganisms play an important role in ameliorating soil condition that facilitates plant productivity (Giri et al., 2009). Nitrogen phosphorus are major plant nutrients which occupy a key place in balanced use of fertilizer. Phosphorus is important requirement of legumes for their nitrogen fixation process (Huda et al., 2007). Microorganisms are useful for mineral solubilization of soil bound mineral content and make nutrients available to their host / or its surroundings thereby enhances the plant growth (Kaushik, 1998; Tilak et al., 2005; Hameeda et al., 2008; Gupta et al., 2007). Microbial application at the nursery stage was also found to be useful in enhancing productivity in some forest trees like *Albizzia, Acacia Dalbergia* etc (Dash et al., 2013; Gupta et al., 2011; Sahgal et al., 2004; Thatoi et al., 1993; Varma, 1989). In view to this we screened few phosphate solubilising bacteria and fungi and iron ore solubiliser under pot culture condition on *Adnenthera pavonina* in order to assess the growth performances in nursery condition.

2. MATERIALS AND METHODS

The experiment was set at the glass house of Regional Plant Resource Centre in the temperature of 35±2ºC & 80±5% relative humidity in Pot size: 8x11" polybags contains 2.5 kg soil. The soil contains 83.8% of sand, 8.8% of slit and 7.4% of clay. Textural class of the soil was loamy sand the soil pH was 6.27. Salt content of the soil was 0.504. Average nitrogen (N), average phosphate (P2O5) and average potassium (K2O) of the soil was 168.7Kg/Ha, 237.2kg/Ha and 645.12kg/Ha respectively. Microbial strains used :Six phosphate solublizing fungi, *Penicillium grisefulvum,* , *Penicillium restrictum* , *Penicillium Crysogenum* , *Aspergillus species* 1 , *Aspergillus ornatus* and *Aspergillus wentii* , four iron leaching fungi *Penicillum*

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expansum , *Paecilomyces variotii* , *Cunnighamella elegans* and *Penicillium Crysogenum* , five phosphate solublizing bacteria and 11 Rhizobium were used for the inoculation studies.

Microbial cultures - Fungal inoculum: 25ml of 7 days old culture prepared in Czapek dox medium (4.5 pH) was added to each pot prior to seed sowing. Bacterial culture: 25 ml of 5 days old culture prepared in potato dextrose broth (7.0 pH) was added to each pot prior to seed sowing. Dual inoculation: In second phase of experiment, two best microbial strains were selected for each tree legume on the basis of their performances in previous experiment. The experiment was set up according to first experiment done on screening except the different combination of microbial inoculum used for specific tree legumes. [1] Control: without any inoculation [2] PC: 50ml in each pot. [3] CE: 50ml in each pot [4] PC+CE: 50ml + 50ml in each pot [5] PC (more)+CE: 100ml + 50ml each pot [6] PC+CE (more): 50ml + 100ml in each pot. Triple inoculation: In second phase of experiment, two best microbial strains were selected for each tree legume on the basis of their performances in previous experiment. The experiment was set up according to first experiment done on screening except the different combination of microbial inoculum used for specific tree legumes.

Experimental schedule – April to August, in 20 replications watering schedule: daily through sprinkler mist system. Nutrient supplementation: Monthly 25ml Hoagland solution /pot. Seed sowing: Pretreated, water soaked and healthy seeds (3 no.) per pot. Final observation was recorded on 120 days of growth. Growth analysis: Growth parameters were recorded for the experimental plants such as shoot height (in cm), root length (in cm), number of leaf, leaflets and branches, total seedling height (in cm), collar diameter (in mm), fresh and dry biomass of leaf, stem, root, shoot and seedling (in gram) (Al-Garni, 2006; Sah et al., 1998; Tewari et al., 2006). Analysis of quality index: To quantify the morphological quality of the seedling the quality index was calculated (Dickson et al., 1960). Analysis of RGR: Relative Growth Rate (RGR), was calculated. (Kathirvelan and Kalaeselvan, 2006; Groneveld, 1998; Carswell et al., 2000; Poorter and Garnier, 1996; Storkey, 2004; Margolis and Vezina, 1988; Cal et al., 2007; Wayne Polley et al., 2002). Statistical analysis for one way ANOVA was done by following (Sockal et al., 1981).

3. Results

3.1 Screening experiments

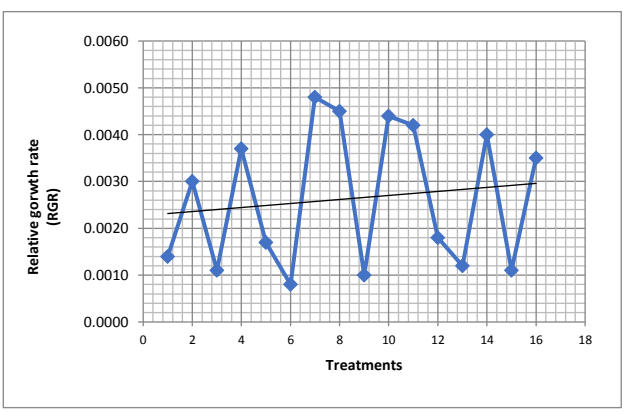
The plants of *Adenanthera pavonina* grown under different treatment along with control untreated plants exhibited good growth in terms of plant height, biomass and plant parts. Besides this, significant variations could be observed among all the treatments in effecting plant growth performances. Table 1 represents the data of growth measurements of seedlings in the nursery after four months period. It is evident that there are significant differences in seedlings height of control and those of inoculated seedlings. However, *Penicillium chrysogenum* showed better effects in enhancing plant growth as compared to other microbial inoculants. Bacterial strains PB 6 also performed well in improving plant health. Differential response on account of varying inoculants was observed in leaf, leaflets and branches numbers. *Penicillium chrysogenum* inoculated plants that showed highest number of leafs, leaflets and fresh biomass. Mean Biomass (fresh and Dry) and collar diameter measured after four months indicates the maximum increment in growth of plants inoculated with these fungi.

Table 1: Screening of bioinoculants for growth of Adnenthera pavonina grown under nursery conditions

	NO. OF LEAVES	NO. OF LEAFLETS	ROOT LENGTH (CM)	SHOOT HEIGHT(CM)	FRESH BIOMASS OF LEAVES(g)	DRY BIOMASS OF LEAVES(g)	TOTAL SHOOT FRESH BIOMASS(g)	TOTAL SHOOT DRY BIOMASS(g)
T1	15.80 ± 4.20	84.80 ± 24.84	5.46 ± 1.34	37.68 ± 14.03	4.63 ± 3.44	1.08 ± 0.87	6.40 ± 4.44	1.61 ± 1.23
T2	15.20 ± 3.11	78.20 ± 6.87	7.78 ± 1.28	44.34 ± 4.14	8.85 ± 3.13	2.28 ± 0.90	10.91 ± 3.81	3.07 ± 1.21
T3	15.40 ± 1.14	74.40 ± 17.98	8.08 ± 0.77	35.44 ± 8.89	4.55 ± 3.39	0.73 ± 0.21	5.95 ± 4.39	1.33 ± 0.65
T4	25.40 ± 7.66	161.60 ± 45.46	10.80 ± 0.92	51.08 ± 7.71	12.82 ± 6.65	3.06 ± 0.91	16.06 ± 8.29	4.14 ± 1.21
T5	16.00 ± 2.91	113.40 ± 34.53	7.88 ± 2.11	41.28 ± 9.61	5.26 ± 2.73	1.41 ± 0.57	6.80 ± 3.50	1.92 ± 0.82
T6	16.80 ± 2.94	67.20 ± 11.86	8.00 ± 0.60	34.98 ± 8.68	3.71 ± 2.76	0.92 ± 0.79	4.66 ± 3.42	1.22 ± 0.97
T7	19.20 ± 3.56	118.60 ± 36.08	7.68 ± 1.92	52.94 ± 5.97	12.04 ± 4.85	4.15 ± 0.90	15.02 ± 6.02	5.54 ± 1.11
T8	18.20 ± 5.35	104.80 ± 9.41	9.40 ± 2.02	56.88 ± 4.55	13.26 ± 2.86	4.00 ± 1.26	16.91 ± 3.79	5.04 ± 1.78
T9	12.00 ± 3.08	46.40 ± 20.25	9.66 ± 0.86	28.06 ± 11.18	2.27 ± 2.05	0.57 ± 0.61	2.87 ± 2.53	0.84 ± 0.87
T10	19.60 ± 3.20	105.00 ± 16.01	10.82 ± 3.90	58.34 ± 15.13	16.18 ± 9.15	3.16 ± 1.61	20.97 ± 11.34	4.87 ± 2.57
T11	17.20 ± 6.26	91.20 ± 13.51	7.12 ± 4.17	58.04 ± 7.81	12.06 ± 4.76	3.63 ± 1.00	15.62 ± 6.06	4.93 ± 1.41
T12	16.00 ± 3.46	64.00 ± 20.08	9.16 ± 3.34	36.10 ± 9.30	4.59 ± 2.53	1.52 ± 0.43	5.66 ± 3.07	2.01 ± 0.57
T13	15.00 ± 4.58	77.00 ± 34.92	10.06 ± 2.37	33.96 ± 19.15	5.28 ± 4.72	0.99 ± 1.26	5.89 ± 6.79	1.38 ± 1.63
T14	13.80 ± 3.76	80.80 ± 19.22	9.84 ± 2.06	53.20 ± 3.65	9.02 ± 4.48	3.07 ± 0.95	12.19 ± 4.82	4.36 ± 1.57
T15	14.40 ± 2.60	65.40 ± 27.40	10.94 ± 1.32	34.50 ± 9.28	3.87 ± 3.02	0.73 ± 0.62	4.80 ± 3.70	1.29 ± 0.47
T16	16.40 ± 4.72	118.20 ± 42.71	10.48 ± 1.75	52.52 ± 6.04	11.17 ± 2.70	3.03 ± 0.53	14.86 ± 3.00	4.02 ± 0.66

Abbreviations for Table 1: T1 – Uninoculated control, T2 Penicillium grisefulvum , T3 Penicillium restrictum , T4 Penicillium Crysogenum , T5 Aspergillus species 1, T6 Aspergillus ornatus , T7 Aspergillus wentii , T8 Penicillum expansum, T9 Paecilomyces variotii, T10 Cunnighamella elegans , T11 Penicillium Crysogenum A , T12 (Streptomyces sp.1), T13 Micrococcus sp.1, T14 Micrococcus sp.2 , T15 Micrococcus sp.3 , T16 Streptomyces species 2

The application of *Cunnighamella elegans and Penicillium chrysogenum* showed maximum plant height (58.34 ± 15.13 cm, and 58.04 ± 7.81 cm) as compared to other treatments. Due to enhancement in dry biomass, stem height and leaf area, the relative growth rate (RGR) and quality index was also changed in all the treatments (Figure 1). Inoculation with *Aspergillus wenti* and *Penicillium expansum* exhibited good relative growth rate may be due to high dry biomass of the inoculated plants. Bacterial strains PB 6 also performed well in improving plant health. Penicillium chrysogneum showed good effect on number of leaves, number of primary leaflets, root length, fresh biomass of leaves, total shoot fresh biomass where as *Cunnighamella elegans* exhibited best result in case of shoot height, fresh biomass of leaves, total shoot fresh biomass, collar diameter and better result in case of number of leaves, root length and good result incase of total shoot dry biomass. Finally, two fungal strains were selected for further experimentations on dual inoculations.



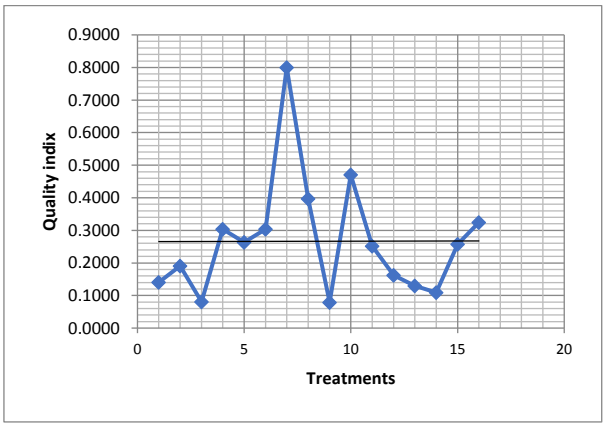


Figure 1: Effect of bioinoculation on Relative growth rate and quality index of Adenenthera pavonina

Abbreviations: T1 – Uninoculated control, T2 Penicillium grisefulvum, T3 Penicillium restrictum, T4 Penicillium Crysogenum, T5 Aspergillus species 1, T6 Aspergillus ornatus , T7 Aspergillus wentii , T8 Penicillum expansum, T9 Paecilomyces variotii, T10 Cunnighamella elegans , T11 Penicillium Crysogenum , T12 (Streptomyces species 1), T13 Micrococcus luteus, T14 Micrococcus luteus , T15 Micrococcus varians , T16 Streptomyces species 1

3.2 Dual inoculation experiments

Combined inoculation of both the fungal strains yielded maximum biomass, root length and shoot height as compared to other treatments. Fungal strain Penicillium chrysogenum and *Cunnighamella elegans* showed better effects in enhancing plant growth as compared to other treatments in combination (equal amount). Variation in inoculants concentration did not significantly influence the plant height and leaf numbers but impacted fresh biomass of leaf and shoot number. The plants of *Adnenthera pavonina* grown under different treatment along with control untreated plants exhibited good growth in terms of plant height, biomass and plant parts. Besides this, significant variations could be observed among all the treatments in effecting plant growth performances. However, fungal strain *Penicillium chrysogenum* and Cunighamella *elegans* showed better effects in enhancing plant growth as compared to uninoculated control. Combination of these two fungal isolates in 1:1 ratio showed better growth performance than the other treatments in pot culture. Finally, we have selected both fungal inoculants in 1:1 ratio for further experimentations on dual inoculations with *Rhizobium* (Table 2).

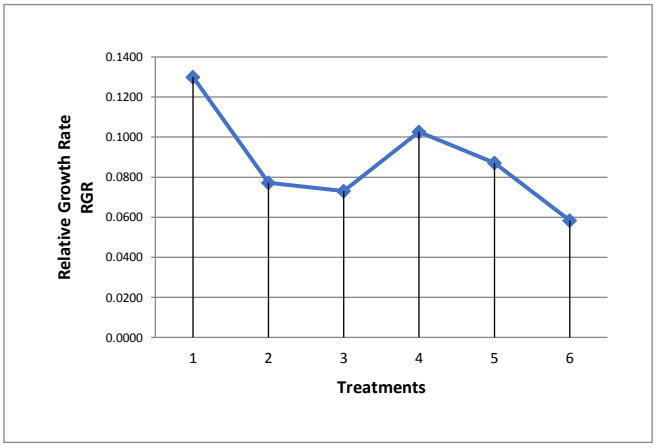
Table 2: Effect of dual inoculation on growth performance of Adnenthera pavonine

S. No.	Morphological parameters	T1	T2	T3	T4	T5	T6
1	**Leaf no.**	29.20 ± 6.69	28.40 ± 5.41	25.20 ± 8.29	33.40 ± 16.64	33.60 ± 11.67	46.80 ± 8.41
2	**Leaflet no.**	104.60 ± 20.04	83.00 ± 17.36	105.60 ± 24.43	117.00 ± 32.86	95.00 ± 38.11	138.60 ± 20.61*
3	**Root length(cm)**	22.84 ± 5.35	27.78 ± 8.89	37.82 ± 8.89*	33.10 ± 8.27	18.94 ± 1.18	21.14 ± 6.18
4	**Shoot height (cm)**	72.16 ± 22.91	97.32 ± 12.73	96.28 ± 20.77*	93.86 ± 24.09	76.98 ± 2.73	71.14 ± 7.52
5	**Fresh biomass leaf (g)**	14.42 ± 11.71	13.85 ± 6.87	16.88 ± 7.88	18.85 ± 6.30	8.42 ± 3.97	8.31 ± 0.87
6	**Dry Biomass leaf (g)**	7.25 ± 3.26	4.81 ± 2.97	4.44 ± 2.27	5.49 ± 0.95	4.48 ± 0.89	4.10 ± 0.11
7	**Fresh biomass Shoot (g)**	24.55 ± 16.59	24.37 ± 8.67	29.09 ± 10.64	33.51 ± 8.37	19.19 ± 2.18	15.15 ± 3.00
8	**Dry biomass shoot (g)**	14.51 ± 6.51	9.50 ± 4.15	9.31 ± 3.80	11.66 ± 1.51	10.26 ± 2.32	7.33 ± 0.14

Abbreviations for table -2: T1- Uninoculated control, T2- Penicillium Crysogenum (PC), T3- Cunnighamella elegans (CE), T4- PC+CE (50ml each), T5 - PC+CE (100+50ml), T6- PC+CE (50ml+100ml)

*-P value summary, t test unpaired p<0.05, One way ANOVA p<0.05, ns- not significant, * - P<0.05, **-P<0.01, *** P<0.001, **** P<0.0001, values are means of 20 replications

Inoculum density of *Penicillium chrysogenum* (1) used in combination with *Cuniinghamella elegans* in 1:1 ratio produced maximum effect. It was also observed that pots inoculated with these two fungi yielded significantly (P< 0.05) highest leaf no. as compared to others. Observation in respect of quality index and relative growth rate of *these plants* grown with selected fungal isolates are presented in figure 2. All inoculated seedlings showed better survival, shoot and root condition and vigor compared to control. The mean ratio of dry weight to fresh weight of shoot of seedling in inoculated than in the control were proved that higher biomass production could be possible by inoculating *this tree speces* with microbial inoculants in nursery.



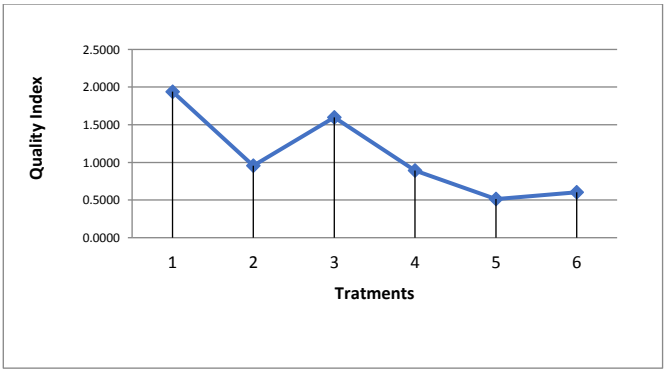


Figure 2: Effect of dual bioinoculation on Relative growth rate and quality index of Adenenthera pavonina Abbreviations for table -2 : T1- Uninoculated control, T2- Penicillium Crysogenum (PC) , T3- Cunnighamella elegans (CE) , T4- PC+CE(50ml each) ,T5 - PC+CE (100+50ml) , T6- PC+CE (50ml+100ml)

Combined inoculants using selected fungal strains (*PC and CE)* in combination under various ratio resulted in enhancement of plant growth compared to the control. These fungal strains yielded maximum biomass (33.51 ± 8.37 Leaf No. (117.00 ±32.86), and dry biomass (5.49± 0.95) as compared to other treatments. The quantity of inoculum did not affect the plant height and biomass substantially except leaf and leaflet numbers which increased due to high inoculum density of *Penicillium chrysogenum* when used in combination in 1:2 ratio (leaf no. 138.60 ± 20.61). The significant differences (P<0.05) among various treatments over control is observed. It was apparent that the ratio of dry weight to fresh weight of shoot of seedlings were higher in inoculated than in the non inoculated

seedlings indicating the higher biomass production in inoculated seedlings.

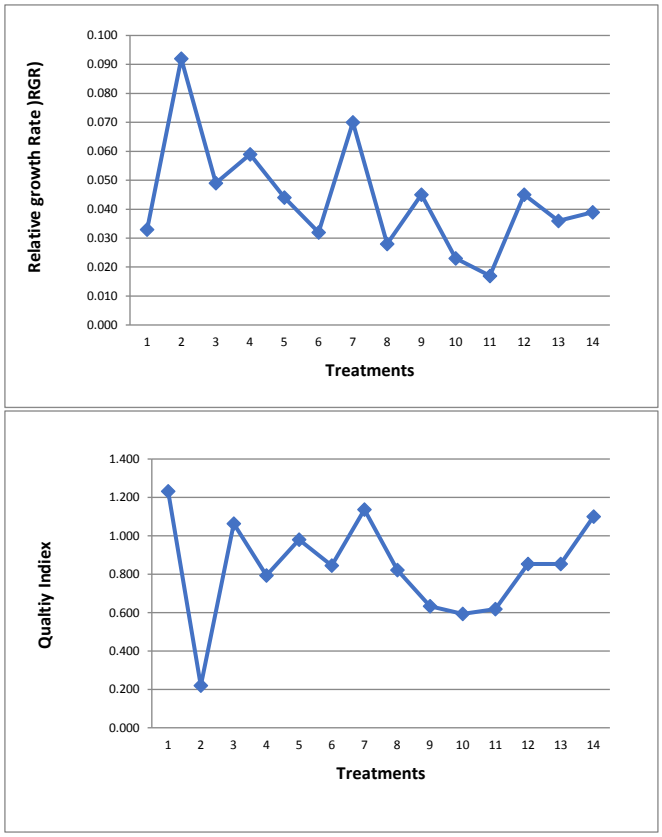


Figure 3: Effect of Rhizobium inoculation on Relative growth rate and quality index of Adenenthera

1- uninoculated control, 2 - Penicillium Crysogenum (PC), 3- Cunnighamella elegans (CE),4-14 – Rhizobium isolates R1-R11

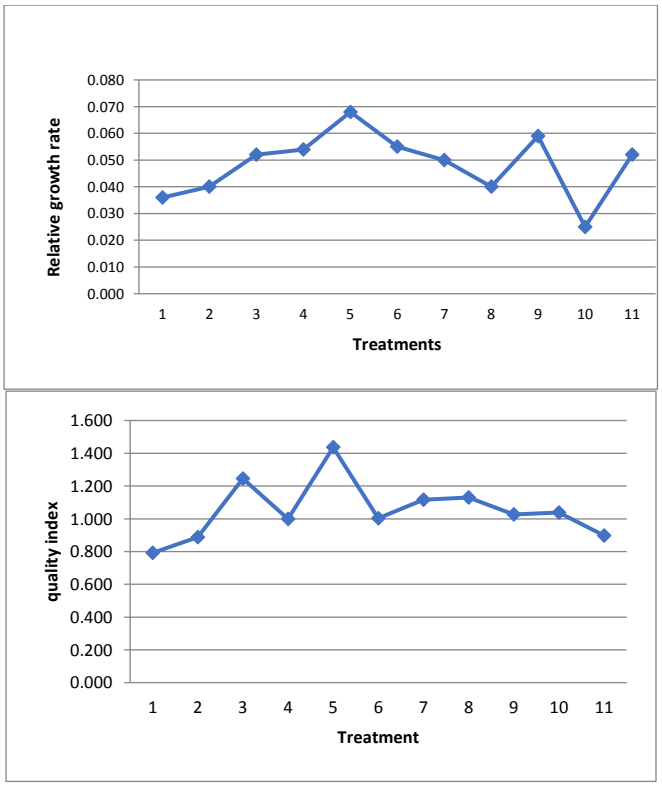


Figure 4: Effect of dual inoculation of Penicillium Crysogenum (PC) and Rhizobium isolates (1-11) on Relative growth rate and quality index of Adenenthera pavonina

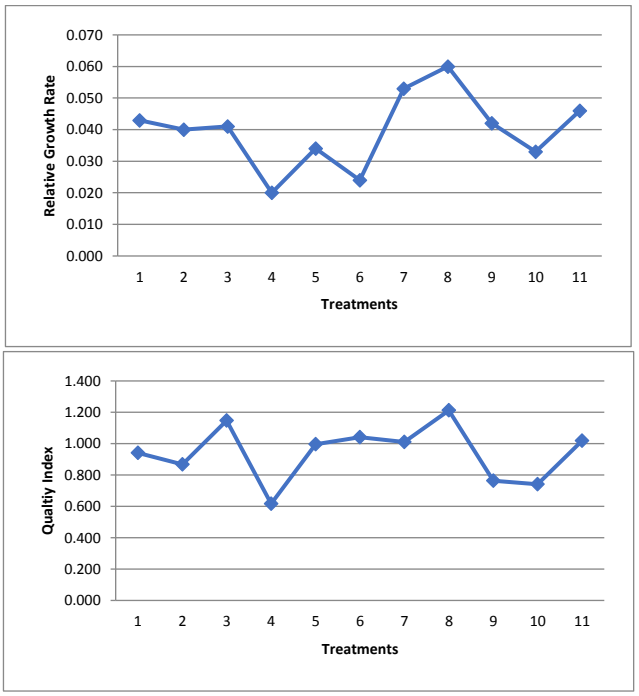


Figure 5: Effect of dual inoculation of Cunninghamella elegans (CE) and Rhizobium isolates (1-11) on Relative growth rate and quality index of Adenenthera pavonina

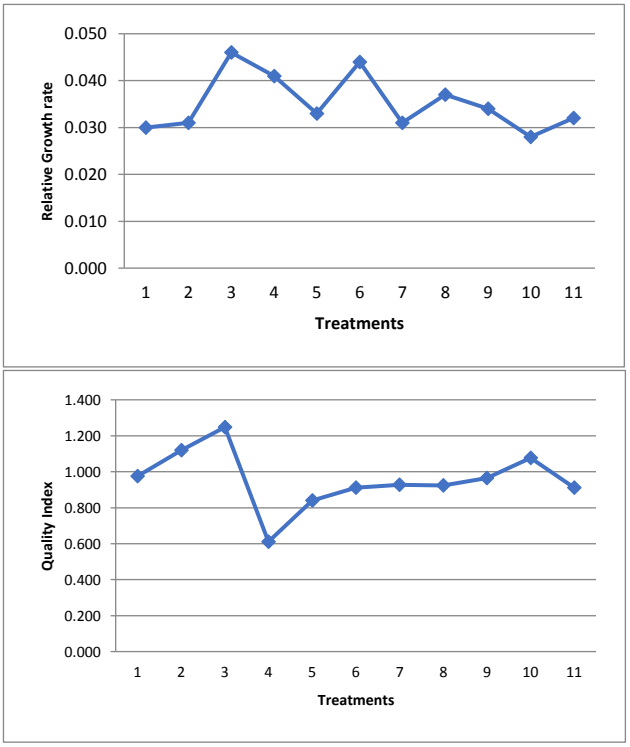


Figure 6: Effect of dual inoculation of Penicillium Crysogenum (PC) + Cunninghamella elegans (CE) and Rhizobium isolates (1-11) on Relative growth rate and quality index of Adenenthera pavonina

3.3 Combined inoculation of mineral solubilizers and Rhizobium

The plants of *Adenanthera pavonina* grown under different treatment (47) along with control untreated plants exhibited good growth and significant variations could be observed among all the treatments in effecting plant growth. However, fungal strain *Penicillium chrysogenum* showed better effects in enhancing plant growth as compared to other treatments in combination (Table 3-6). *Rhizobium* inoculations did not show much effect independently on plant growth. However, their grouping with *Penicillium chrysogenum* positively improved leaf number, root length and biomass. For the third experiment the *Penicillium chrysogneum* and *Cunnughamella elegans* were used in combination of 11 *Rhizobium* strains.

the data obtained on single inoculation of *Penicillium chrysogenum* showed improved number of leaflets, shoot height, fresh biomass of leaves, dry biomass of leaves, total shoot fresh biomass, total shoot dry biomass, relative growth rate (RGR) and collar diameter. Individual inoculation of Cunnighamella elegans exhibited best result in case the collar diameter, number of leaflets, total shoot fresh biomass, total shoot dry biomass and Relative Growth Rate (RGR). The combination of Penicillium chrysogenum+ R5 and R6 separately depicted better result on growth parameters. Though the test plants were found to be infected with Rhizobium but a very poor performance was observed as far as root nodule formation is concerned. Hence total number of nodules, size and structure could not be recorded.

Table 3: Effect of Rhizobium isolates on growth performance of Adnenthera pavonina grown under nursery conditions

TREATMENT	SHOOT HEIGHT (IN CM)	ROOT LENGTH (IN CM)	TOTAL SEEDLING HEIGHT (IN CM)	NUMBER OF LEAVES	NUMBER OF PRIMARY LEAFLETS	FRESH BIOMASS OF LEAVES (IN GRAM)	FRESH BIOMASS OF STEM (IN GRAM)	FRESH BIOMASS OF ROOT (IN GRAM)	TOTAL SHOOT FRESH BIOMASS (IN GRAM)	TOTAL SEEDLING FRESH BIOMASS (IN GRAM)	DRY BIOMASS OF LEAVES (IN GRAM)	DRY BIOMASS OF STEM (IN GRAM)	DRY BIOMASS OF ROOT (IN GRAM)	TOTAL SHOOT DRY BIOMASS (IN GRAM)	TOTAL SEEDLING DRY BIOMASS (IN GRAM)
C1	46.04 ± 7.13	15.18 ± 8.95	63.60 ± 11.10	19.60 ± 3.13	72.20 ± 13.41	4.77 ± 2.09	3.73 ± 0.63	3.94 ± 1.97	8.29 ± 2.71	12.22 ± 4.45	2.09 ± 0.57	1.64 ± 0.44	2.11 ± 0.88	3.73 ± 0.94	5.85 ± 1.73
C2	79.70 ± 4.29**	26.56 ± 2.87*	106.26 ± 3.80**	23.40 ± 8.11	140.20 ± 31.44**	14.76 ± 4.45**	6.71 ± 2.60*	4.17 ± 1.86	21.19 ± 6.98**	25.37 ± 8.20**	6.37 ± 2.50**	3.73 ± 1.59**	1.78 ± 0.39	10.45 ± 4.40**	12.23 ± 4.79 **
C3	70.04 ± 12.57	25.06 ± 3.91*	95.10 ± 9.48*	23.40 ± 7.92	116.80 ± 45.28*	10.64 ± 6.80	6.66 ± 2.99*	3.59 ± 1.45	17.30 ± 9.79*	20.89 ± 11.17	3.48 ± 1.68	3.53 ± 1.62**	1.73 ± 0.41	6.24 ± 2.60	7.97 ± 2.83
R1	64.42 ± 22.90	25.46 ± 6.90*	89.88 ± 28.55	19.80 ± 6.14	84.20 ± 22.20	6.67 ± 4.42	4.31 ± 1.94	3.27 ± 0.82	10.99 ± 6.09	14.26 ± 6.82	1.91 ± 0.94	2.33 ± 1.29	1.58 ± 0.35	3.35 ± 0.91	4.93 ± 1.23
R2	76.58 ± 22.91**	23.60 ± 2.43	100.08 ± 24.90**	23.20 ± 8.73	99.00 ± 16.17	9.68 ± 7.06	4.16 ± 1.23	3.34 ± 1.11	13.84 ± 8.16	17.18 ± 9.12	2.57 ± 0.41	2.44 ± 0.59	1.87 ± 0.19	4.65 ± 0.23	6.53 ± 0.42
R3	52.98 ± 19.30	27.16 ± 6.75**	80.14 ± 25.01	20.60 ± 8.36	74.40 ± 30.68	6.30 ± 4.13	3.65 ± 0.81	2.76 ± 1.42	9.95 ± 4.80	12.71 ± 6.05	1.58 ± 0.11	2.03 ± 0.35	1.38 ± 0.12	3.49 ± 0.16	4.88 ± 0.10
R4	54.90 ± 13.79	22.60 ± 5.23	77.50 ± 15.92	23.40 ± 4.67	78.00 ± 17.72	7.54 ± 3.09	4.54 ± 0.92	2.49 ± 0.42	12.08 ± 4.00	14.57 ± 4.31	4.76 ± 2.00	2.44 ± 0.50	1.60 ± 0.06	7.33 ± 2.57	8.93 ± 2.55
R5	50.30 ± 11.58	24.82 ± 4.24*	75.12 ± 15.19	22.40 ± 1.14	75.20 ± 20.41	4.76 ± 1.93	3.96 ± 1.15	2.98 ± 1.68	8.72 ± 2.93	11.70 ± 4.51	1.67 ± 0.44	1.74 ± 0.52	1.42 ± 0.38	3.14 ± 0.55	4.56 ± 0.64
R6	59.54 ± 17.22	25.98 ± 8.72*	85.52 ± 23.98	22.20 ± 2.49	88.80 ± 28.89	6.62 ± 3.43	4.41 ± 1.23	2.51 ± 1.31	11.03 ± 4.49	13.54 ± 5.57	2.88 ± 1.89	2.24 ± 0.74	0.88 ± 0.30	4.85 ± 2.80	5.73 ± 2.92
R7	41.06 ± 14.41	22.64 ± 7.34	63.70 ± 21.50	23.00 ± 4.06	71.60 ± 34.67	4.08 ± 3.35	3.39 ± 0.24	3.25 ± 2.13	7.47 ± 3.35	10.72 ± 5.39	1.15 ± 0.12	1.65 ± 0.02	1.31 ± 0.13	2.80 ± 0.12	4.11 ± 0.22
R8	77.06 ± 23.23**	23.58 ± 6.25	100.64 ± 28.90**	22.80 ± 3.90	81.80 ± 39.83	2.93 ± 1.71	4.36 ± 2.04	2.33 ± 0.25	7.30 ± 2.13	9.63 ± 2.36	1.56 ± 0.58	2.06 ± 0.96	1.26 ± 0.26	3.22 ± 1.06	4.49 ± 1.32
R9	75.16 ± 4.22**	24.86 ± 4.00*	100.02 ± 6.86**	22.40 ± 2.30	93.80 ± 22.77	8.69 ± 2.70	3.10 ± 0.37	3.72 ± 3.09	11.78 ± 2.91	15.50 ± 5.65	3.53 ± 1.06	1.49 ± 0.18	1.28 ± 0.36	4.97 ± 1.28	6.25 ± 1.51
R10	40.36 ± 2.26	22.24 ± 1.29	62.60 ± 1.32	22.60 ± 1.52	60.60 ± 9.07	6.66 ± 1.38	3.49 ± 1.63	2.90 ± 0.36	10.15 ± 1.51	13.04 ± 1.43	3.00 ± 0.39	1.50 ± 0.87	1.69 ± 0.16	3.90 ± 0.66	5.59 ± 0.66
R11	67.86 ± 2.41	22.52 ± 12.45	90.38 ± 12.73	22.20 ± 2.28	83.20 ± 20.66	7.08 ± 2.99	4.15 ± 1.04	3.68 ± 0.61	11.23 ± 3.98	14.91 ± 4.46	2.34 ± 0.32	2.29 ± 0.56	1.84 ± 0.37	4.27 ± 0.64	6.11 ± 0.93

C1- uninoculated control, C2- Penicillium Crysogenum (PC), C3- Cunnighamella elegans (CE)

*-P value summary, t test unpaired p<0.05, One way ANOVA p<0.05, ns- not significant, * - P<0.05, **-P<0.01, *** P<0.001, **** P<0.0001, values are means of 20 replications

Table 4: Effect of dual inoculation of rhizobium and selected phosphate solubilizaing fungi.

TREATMENT	SHOOT HEIGHT (IN CM)	ROOT LENGTH (IN CM)	TOTAL SEEDLING HEIGHT (IN CM)	NUMBER OF LEAVES	NUMBER OF PRIMARY LEAFLETS	FRESH BIOMASS OF LEAVES (IN GRAM)	FRESH BIOMASS OF STEM (IN GRAM)	FRESH BIOMASS OF ROOT (IN GRAM)	TOTAL SHOOT FRESH BIOMASS (IN GRAM)	TOTAL SEEDLING FRESH BIOMASS (IN GRAM)	DRY BIOMASS OF LEAVES (IN GRAM)	DRY BIOMASS OF STEM (IN GRAM)	DRY BIOMASS OF ROOT (IN GRAM)	TOTAL SHOOT DRY BIOMASS (IN GRAM)	TOTAL SEEDLING DRY BIOMASS (IN GRAM)
PC + R1	46.12 ± 13.41	27.88 ± 3.49**	74.00 ± 14.32	22.60 ± 3.21	69.60 ± 24.70	5.27 ± 3.16	4.08 ± 1.27	3.17 ± 1.10	9.34 ± 4.43	12.51 ± 5.44	2.17 ± 0.81	2.02 ± 0.73	1.42 ± 0.32	3.93 ± 1.22	5.35 ± 1.54
PC+ R2	53.24 ± 17.72	24.48 ± 4.86	77.72 ± 22.58	23.40 ± 2.88	71.00 ± 21.83	6.78 ± 4.45	4.79 ± 2.43	2.73 ± 0.64	11.57 ± 6.84	14.30 ± 7.30	2.76 ± 0.80	2.28 ± 1.36	1.29 ± 0.24	4.45 ± 1.18	5.74 ± 1.18
PC + R3	52.28 ± 21.69	23.42 ± 5.48	75.70 ± 26.94	21.40 ± 4.40	69.80 ± 28.46	8.37 ± 6.69	4.54 ± 1.41	3.05 ± 1.04	12.91 ± 8.04	15.96 ± 9.02	3.28 ± 1.16	2.42 ± 0.65	1.68 ± 0.29	5.61 ± 1.75	7.29 ± 1.96
PC + R4	54.98 ± 11.47	26.70 ± 2.88**	81.68 ± 11.37	24.40 ± 3.44	72.20 ± 12.93	6.85 ± 2.51	5.19 ± 2.07	3.23 ± 1.01	12.04 ± 4.41	15.27 ± 5.38	3.45 ± 1.52	2.56 ± 0.68	1.46 ± 0.48	5.84 ± 1.82	7.31 ± 1.88
PC + R5	68.00 ± 10.51	30.46 ± 7.97**	98.46 ± 12.32*	26.60 ± 5.50	110.20 ± 18.55	11.25 ± 4.12	6.02 ± 1.25	4.09 ± 1.20	17.28 ± 5.33*	21.36 ± 6.23	4.56 ± 1.75	3.17 ± 0.37*	2.28 ± 0.97	7.71 ± 2.27*	9.99 ± 2.97
PC + R6	75.88 ± 1.30**	24.28 ± 2.19	100.16 ± 3.27**	28.20 ± 9.09*	68.20 ± 36.15	10.71 ± 3.35	5.38 ± 3.18	3.79 ± 1.04	16.10 ± 6.14	19.89 ± 7.03	4.60 ± 0.47	2.91 ± 1.55	1.62 ± 0.17	6.62 ± 0.72	8.25 ± 0.88
PC + R7	68.94 ± 11.04	24.28 ± 3.26	93.22 ± 12.96*	26.20 ± 3.03	93.00 ± 17.41	9.55 ± 3.25	5.67 ± 3.04	4.85 ± 2.19	15.22 ± 5.48	20.07 ± 7.63	3.45 ± 1.40	3.00 ± 1.59	1.92 ± 0.33	5.49 ± 0.68	7.40 ± 1.01
PC + R8	56.06 ± 8.59	20.24 ± 2.95	76.30 ± 8.55	23.00 ± 3.16	84.20 ± 18.83	9.32 ± 8.63	4.02 ± 0.99	4.46 ± 2.60	13.34 ± 9.08	17.80 ± 11.42	2.55 ± 1.75	2.21 ± 0.65	2.14 ± 0.93	4.63 ± 2.31	6.78 ± 1.91
PC + R9	53.48 ± 14.61	23.64 ± 3.86	77.12 ± 17.97	24.20 ± 1.48	84.40 ± 25.67	8.49 ± 1.26	4.18 ± 0.76	3.23 ± 0.31	12.67 ± 1.82	15.90 ± 1.88	3.97 ± 0.79	2.02 ± 0.52	1.54 ± 0.11	6.19 ± 0.94	7.73 ± 1.01
PC + R10	44.82 ± 46.05	17.52 ± 2.61	62.34 ± 17.20	22.60 ± 1.95	67.40 ± 16.46	4.06 ± 1.42	3.67 ± 0.99	2.70 ± 0.48	7.73 ± 2.19	10.43 ± 2.45	1.31 ± 0.17	1.78 ± 0.50	1.55 ± 0.25	2.88 ± 0.23	4.43 ± 0.41
PC + R11	54.72 ± 18.25	22.02 ± 3.04	76.74 ± 20.48	22.00 ± 4.74	81.00 ± 28.00	6.82 ± 3.56	4.28 ± 0.87	2.71 ± 0.40	11.11 ± 4.43	13.82 ± 4.54	3.19 ± 0.88	2.27 ± 0.32	1.42 ± 0.25	5.53 ± 1.16	6.95 ± 1.22

PC- Penicillium Crysogenum ,*-P value summary, t test unpaired p<0.05, One way ANOVA p<0.05, ns- not significant, * - P<0.05, **-P<0.01, *** P<0.001, **** P<0.0001, values are means of 20 replications , ±- Standard Deviation

Table 5: Effect of dual inoculation of rhizobium and iron ore leaching fungi

TREATMENT	SHOOT HEIGHT (IN CM)	ROOT LENGTH (IN CM)	TOTAL SEEDLING HEIGHT (IN CM)	NUMBER OF LEAVES	NUMBER OF PRIMARY LEAFLETS	FRESH BIOMASS OF LEAVES (IN GRAM)	FRESH BIOMASS OF STEM (IN GRAM)	FRESH BIOMASS OF ROOT (IN GRAM)	TOTAL SHOOT FRESH BIOMASS (IN GRAM)	TOTAL SEEDLING FRESH BIOMASS (IN GRAM)	DRY BIOMASS OF LEAVES (IN GRAM)	DRY BIOMASS OF STEM (IN GRAM)	DRY BIOMASS OF ROOT (IN GRAM)	TOTAL SHOOT DRY BIOMASS (IN GRAM)	TOTAL SEEDLING DRY BIOMASS (IN GRAM)
CE + R1	48.42 ± 17.54	22.76 ± 6.11	71.18 ± 23.38	23.00 ± 4.18	72.80 ± 27.73	8.63 ± 5.45	4.35 ± 1.72	2.57 ± 0.64	12.97 ± 6.69	15.54 ± 7.21	2.73 ± 2.36	2.10 ± 0.63	1.31 ± 0.31	4.71 ± 2.93	6.02 ± 3.23
CE + R2	34.98 ± 18.60	18.28 ± 5.44	53.26 ± 23.91	21.80 ± 5.07	56.20 ± 30.87	8.99 ± 5.97	3.70 ± 1.35	2.37 ± 0.45	12.68 ± 6.84	15.05 ± 7.08	2.84 ± 2.94	1.91 ± 0.69	1.19 ± 0.23	4.43 ± 2.94	5.62 ± 2.78
CE + R3	60.00 ± 8.80	28.00 ± 7.60**	88.00 ± 3.53	24.40 ± 3.78	94.60 ± 21.58	7.50 ± 3.65	4.03 ± 1.39	4.47 ± 2.94	11.53 ± 4.68	16.00 ± 7.25	2.72 ± 2.02	2.01 ± 0.73	2.05 ± 1.00	4.71 ± 2.92	6.76 ± 3.92
CE + R4	51.34 ± 17.75	21.02 ± 4.84	72.36 ± 20.34	20.40 ± 3.91	62.60 ± 21.30	3.88 ± 2.57	4.00 ± 1.83	2.84 ± 0.54	7.88 ± 4.39	10.72 ± 4.79	1.26 ± 0.20	2.13 ± 1.16	1.31 ± 0.27	2.86 ± 0.21	4.17 ± 0.39
CE + R5	70.68 ± 14.31*	22.30 ± 7.65	92.98 ± 19.62	24.80 ± 3.56	97.20 ± 27.36	7.90 ± 5.05	3.69 ± 0.48	3.64 ± 1.46	11.60 ± 5.23	15.24 ± 6.08	2.01 ± 0.78	2.07 ± 0.54	1.97 ± 0.58	3.91 ± 1.29	5.88 ± 1.84
CE + R6	39.86 ± 7.47	17.48 ± 2.25	57.34 ± 8.52	25.20 ± 3.90	110.80 ± 52.18	3.39 ± 0.70	3.13 ± 0.84	3.03 ± 0.70	6.52 ± 1.25	9.55 ± 1.44	1.63 ± 0.48	1.47 ± 0.16	1.61 ± 0.14	3.07 ± 0.60	4.68 ± 0.67
CE + R7	63.28 ± 8.15	22.92 ± 3.23	86.20 ± 9.55	23.20 ± 3.56	78.00 ± 12.94	6.31 ± 0.80	4.21 ± 0.39	2.86 ± 0.44	10.52 ± 0.75	13.38 ± 1.05	3.52 ± 0.51	2.28 ± 0.44	1.55 ± 0.37	5.65 ± 0.50	7.20 ± 0.87
CE + R8	68.62 ± 19.49	23.34 ± 6.21	91.96 ± 25.45	22.80 ± 2.77	102.00 ± 26.01	11.06 ± 9.45	5.54 ± 3.11	3.40 ± 1.41	10.60 ± 12.51	20.00 ± 13.75	3.66 ± 2.48	2.98 ± 1.89	2.02 ± 1.08	6.31 ± 4.20	8.33 ± 5.28
CE + R9	72.78 ± 8.40*	20.44 ± 2.56	93.22 ± 7.74*	22.80 ± 2.17	85.40 ± 31.87	7.10 ± 3.68	5.14 ± 1.49	2.79 ± 0.58	12.24 ± 5.00	15.04 ± 5.56	2.30 ± 0.71	2.70 ± 0.65	1.30 ± 0.05	4.58 ± 1.08	5.87 ± 1.04
CE + R10	47.02 ± 11.40	17.66 ± 10.34	64.68 ± 17.57	22.80 ± 1.92	59.00 ± 10.44	4.82 ± 2.13	3.84 ± 0.54	2.35 ± 0.38	8.66 ± 2.49	11.01 ± 2.86	1.57 ± 0.38	1.97 ± 0.26	1.09 ± 0.12	3.64 ± 0.30	4.83 ± 0.32
CE + R11	52.64 ± 17.23	23.54 ± 6.69	76.18 ± 21.95	24.20 ± 3.03	79.60 ± 25.21	6.59 ± 3.29	4.20 ± 1.24	3.25 ± 0.73	10.80 ± 4.44	14.04 ± 4.59	3.21 ± 1.70	1.90 ± 0.47	1.70 ± 0.55	5.13 ± 2.27	6.83 ± 2.26

CE- Cuninghamella elegans ,*-P value summary, t test unpaired p<0.05, One way ANOVA p<0.05, ns- not significant, * - P<0.05, **-P<0.01, *** P<0.001, values are means of 20 replications , ±- Standard Deviation

Table 6: Effect of triple inoculation of Rhizobium, PC and CE on growth of Adenenthera pavonine

TREATMENT	SHOOT HEIGHT (IN CM)	ROOT LENGTH (IN CM)	TOTAL SEEDLING HEIGHT (IN CM)	NUMBER OF LEAVES	NUMBER OF PRIMARY LEAFLETS	FRESH BIOMASS OF LEAVES (IN GRAM)	FRESH BIOMASS OF STEM (IN GRAM)	FRESH BIOMASS OF ROOT (IN GRAM)	TOTAL SHOOT FRESH BIOMASS (IN GRAM)	TOTAL SEEDLING FRESH BIOMASS (IN GRAM)	DRY BIOMASS OF LEAVES (IN GRAM)	DRY BIOMASS OF STEM (IN GRAM)	DRY BIOMASS OF ROOT (IN GRAM)	TOTAL SHOOT DRY BIOMASS (IN GRAM)	TOTAL SEEDLING DRY BIOMASS (IN GRAM)
PC +CE + R1	40.94 ± 3.74	20.80 ± 3.26	61.74 ± 2.38	23.60 ± 4.98	68.60 ± 21.40	4.46 ± 2.60	3.64 ± 0.43	2.70 ± 0.44	8.10 ± 3.03	10.81 ± 3.41	1.66 ± 0.79	1.79 ± 0.32	1.61 ± 0.06	3.46 ± 1.17	5.07 ± 1.22
PC + CE + R2	52.06 ± 12.96	20.02 ± 2.70	72.08 ± 15.15	21.80 ± 1.92	101.40 ± 37.39	7.91 ± 6.38	3.95 ± 1.14	3.71 ± 1.63	11.86 ± 7.38	15.57 ± 8.85	2.06 ± 0.98	2.10 ± 0.53	1.58 ± 0.85	3.82 ± 1.24	5.40 ± 2.03
PC + CE + R3	59.96 ± 14.78	22.20 ± 4.98	82.16 ± 19.20	24.20 ± 4.15	100.60 ± 26.61	7.53 ± 2.92	4.08 ± 0.76	4.71 ± 2.19	11.61 ± 3.46	16.32 ± 5.58	2.76 ± 1.92	2.00 ± 0.44	2.24 ± 0.95	4.86 ± 2.41	7.10 ± 3.36
PC + CE + R4	68.74 ± 20.19	22.62 ± 2.91	91.36 ± 22.89	24.00 ± 4.06	91.40 ± 19.58	6.03 ± 1.20	7.76 ± 1.59	2.85 ± 0.76	10.79 ± 2.35	13.65 ± 3.07	2.73 ± 0.66	2.41 ± 0.64	1.16 ± 0.13	4.79 ± 1.14	5.95 ± 1.26
PC + CE + R5	46.00 ± 4.91	25.54 ± 3.37*	71.54 ± 3.45	22.20 ± 4.60	67.40 ± 7.06	8.10 ± 6.39	3.67 ± 0.41	2.82 ± 0.28	11.78 ± 6.26	14.60 ± 6.37	1.78 ± 0.29	1.85 ± 0.38	1.45 ± 0.19	3.81 ± 0.65	5.26 ± 0.51
PC + CE + R6	54.74 ± 3.50	21.94 ± 7.26	76.68 ± 8.21	22.40 ± 2.07	72.20 ± 9.26	6.49 ± 1.43	3.38 ± 0.88	2.92 ± 0.35	9.87 ± 1.90	12.79 ± 2.08	3.09 ± 0.82	1.69 ± 0.42	1.55 ± 0.10	4.71 ± 1.17	6.26 ± 1.23
PC + CE + R7	41.26 ± 10.78	24.00 ± 3.56	65.26 ± 8.83	22.60 ± 2.07	80.00 ± 22.72	4.85 ± 1.99	3.53 ± 0.35	2.62 ± 0.21	8.37 ± 2.33	11.00 ± 2.33	1.66 ± 0.49	1.82 ± 0.26	0.54 ± 0.40	3.45 ± 0.74	4.99 ± 0.82
PC + CE + R8	45.84 ± 9.99	25.08 ± 1.36*	70.92 ± 10.79	22.20 ± 1.10	80.60 ± 17.26	5.28 ± 1.58	3.80 ± 0.50	2.93 ± 0.57	9.08 ± 2.07	12.01 ± 2.57	2.36 ± 0.16	1.89 ± 0.21	1.67 ± 0.18	4.26 ± 0.08	5.93 ± 0.20
PC + CE + R9	45.66 ± 14.66	23.18 ± 5.17	68.84 ± 19.46	24.40 ± 3.21	72.40 ± 22.74	4.90 ± 2.61	2.94 ± 0.87	2.74 ± 0.87	7.44 ± 3.11	10.18 ± 3.77	1.96 ± 0.43	1.54 ± 0.76	1.65 ± 0.20	3.86 ± 0.70	5.51 ± 0.51
PC + CE + R10	48.30 ± 8.42	23.06 ± 6.17	71.36 ± 10.95	22.40 ± 2.07	62.40 ± 11.26	3.78 ± 0.88	2.95 ± 1.12	3.23 ± 1.22	6.73 ± 0.54	9.96 ± 1.68	1.60 ± 0.28	1.49 ± 0.64	2.17 ± 1.07	3.35 ± 0.26	5.52 ± 1.33
PC + CE + R11	46.68 ± 10.70	19.02 ± 3.53	65.70 ± 11.26	22.00 ± 2.45	67.00 ± 9.64	4.52 ± 0.75	3.69 ± 0.34	2.47 ± 0.39	8.21 ± 0.94	10.68 ± 1.15	2.23 ± 0.22	1.72 ± 0.48	1.38 ± 0.23	3.74 ± 0.18	5.12 ± 0.40

PC- Penicillium Crysogenum, CE- Cunnighamella elegans, R- Rhizobium isolates (1-11)

*-P value summary, t test unpaired p<0.05, One way ANOVA p<0.05, ns- not significant, * - P<0.05, **-P<0.01, *** P<0.001, **** P<0.0001, values are means of 20 replications

4. Discussion

The plants of *Adnenthera pavonina* grown under different treatment along with control untreated plants exhibited good growth in terms of plant height, biomass and plant parts with significant variations. The shoot height measurements of seedlings in the nursery stage (4-month-old) indicates that microbial applications are effective for development and growth of host plants. Amongst different parameter studied, difference in the biomass yield between treated and control was most significant. Inoculations with bacterial strains did not show better performance in seedling growth compared to control. *Penicillium chrysogenum* and *Cunninghamella elegans* showed to be the best inoculants for *this tree species in* increasing the plant dry biomass of leaves and total shoot dry biomass, as the biomass increment was approximately more than three

times higher. Knowledge on physiological variables like relative growth rate, net assimilation rate and leaf area ratio may be the useful tool for determining the proper growth and development of seedling (Wayne polley et al., 2002).

No significant observations have been made on above parameters during this study. It is reported that the stem and branching characters at early age of the plants may cause the difference in dimension of form of tree crown (Jankiewicz and Stecki, 1976). Microbial inoculations may also increase the root shoot ratio, leaf numbers and length of the branches of host plants (Al-Garni, 2006). Weih and Nordh stated that total leaf area of the pot grown plants is better predictor of shoot biomass and branches in the field than the pot grown plants (Weih and Nordh, 2005). Most of the tropical soils are phosphate fixing, use of mineral solubilisers from microbial origin may make them available to the host plants. Microbial inoculants are also found to be useful in enhancing growth of many forest tree seedlings grown under stress conditions (Dabas and Kaushik, 1998; Dash et al., 2013). It is evident from present study that after inoculation the increment of seedling height of inoculated seedling is very high compared to control.

Again the *P. chrysogenum* was found to be superior than other. Knowledge on physiological variables like RGR, NAR and LAR could be the useful tool for assessing the growth and development of seedling. In *P. chrysogenum* inoculations RGR increased over the control. Successful reforestation programme have included application of biofertilizers at the nursery state so as to produce seedlings which are value added with microsymbionts. In the present study, nodulation was very poor. Since, study was carried out for only four months; role of inoculated *Rhizobium* and their effects on performance of host plants is difficult to be interpreted. Many of the tropical leguminous trees are reported to be fixing atmospheric nitrogen through nodule development, endowed with VA mycorrrhizal and other microbial association. Plantation of such species enriches poor soils, seedlings fortified with microbial inoculations may lead to successful in establishment of plants in poor sites (Ghaham, 1976; Sah et al., 1998; Sahgal et al., 2004).

The effects of mineral solubiliers need to be evaluated under field conditions, on a long-term basis, before the application these inoculations to plantation seedlings is considered as a viable proposition. Screening of ten fungal species and 5 bacterial isolates for their effects on growth of this tree species done under pot culture in green house conditions, elucidated the best performance of Penicillium Crysogenum and Cunnighamella elegans were the most effective inoculants for the plant Adenanthera pavonina. The present study done under controlled greenhouse conditions has provided an experimental approach to adopt pre inoculation of the symbiotic microbes to seedlings in nursery for better plantation results. This aspect may become a very practicable and cost-effective proposition in plantation programs in tropical conditions. However, the outcome of present study is very important due to the requirement of microbial manifestation in tree improvement programe in stress environment.

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