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RESEARCH ARTICLE

FIELD PERFORMANCE OF *SCHIMA WALLICHII* (DC.) KORTH. SEEDLINGS INOCULATED WITH ECTOMYCORRHIZAL FUNGI

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ABSTRACT

Symbiotic efficiency of ectomycorrhizal fungi on the growth and nutrient uptake of *Schima wallichii* seedlings were studied in two abandoned jhum land. Native ectomycorrhizal fungi (*Suillus bovinus*, *Boletus edulis* and *Scleroderma citrinum*) were inoculated using pure culture technique. Various growth characters viz. shoot height, leaf length, root length, root colar diameter and seedlings volume at various intervals responded significantly to all the fungal inoculants. A significant variation in phosphorus ($P<0.01$) content was observed between the treated and un-inoculated control. The formation and network of ectomycorrhizae appears to have an affirmative impact on the increased absorption of phosphorus resulting in better growth of the seedlings. Among the fungal inoculants, *Boletus edulis* successfully colonized and promoted the growth and nutrient uptake in *Schima wallichii* seedlings.

INTRODUCTION

Natural ecosystems depend on closed nutrient cycles to minimize losses of nutrients and optimize turnover within the soil-plant system. Any disturbance in the ecosystem generally disrupts these cycles resulting in rapid decline of plant productivity. The scale of human activity has become such that most of the ecosystems of the earth have been disrupted in some ways (Ehrlich, 1993). Deforestation and land degradation as a result of shifting cultivation (Jhum) is of great concern because of its impact on biodiversity and environmental issues. In Northeast India (with particular reference to Nagaland), jhum system of cultivation worked well when there was a balance between population and soil fertility as a result of longer fallow cycle. In recent years, the resilience of ecosystem has broken down and the land is increasingly deteriorating because of the increase in population, pressures on land, coupled with decreasing fallow cycles. In view of the unabated deforestation and decline in land productivity, plantation of native plant species has been encouraged to ameliorate the abandoned jhum lands. The chief advantage of using local tree species in reforestation programmes is that they are adapted to local environmental conditions, improve livelihood and economics of the local communities, who can utilize them and contribute to the conservation of biodiversity.

Degraded lands are generally poor in available nutrients limiting the growth of transplanted seedlings. Successful establishment of seedlings on such sites often fails without the presence of appropriate fungal partners (Kendrick, 2000; Lakhanpal, 2000). Ectomycorrhizal fungal inoculants provide the potential for improvement in quality and performance of out planted seedlings (Dominguez *et al*., 2006; Quoreshi *et al*., 2008; Rivero *et al*., 2009). In nature, a large diversity of ectomycorrhizal fungi is available with varying capacity for colonizing plant roots and promoting growth. Ectomycorrhizal fungi can be easily isolated from the field especially from fungal fruiting bodies (Dodd and Thomson, 1994) and profitably launch with the existing reforestation programmes. Furthermore, host fungus specificity exists for many mycorrhizal fungi (Lesueur *et al*., 2001; Wu *et al*. 2002; Cavallazzi *et al*., 2007) which imply a need to select appropriate fungal isolates before embarking on inoculation for a particular host plant. Field trials of inoculated seedlings is an important stratagem as the full benefits of mycorrhiza can become evident only when plants are transplanted to the field and exposed to various stresses (Baum *et al*., 2002; Perry etal., 1987; Quoreshi *et al*., 2008).

MATERIALS AND METHODS

Study Site: The field trials were carried out in two abandoned jhum land:

Plot A: At Yimyu (1080.52m above msl), in the district of Mokokchung, Nagaland;
Plot B: At Lumami (962.25m above msl), in the district of Zunheboto, Nagaland.

Three native ectomycorrhizal fungi viz. *Suillus bovinus*, *Boletus edulis* and *Scleroderma citrinum* were isolated and multiplied on Modified Melin Norkran's medium (Marx, 1969). Seedlings of *Schima wallichii* were raised in nursery beds and transplanted in both the plots. One month after transplanting, the selected isolates of fungi were inoculated (5mm diameter block) near the roots of the seedlings, with each treatment replicating 25 times. A set of seedlings were maintained as control without any fungal inoculum. After confirming the symbiotic association, different growth characters of the seedlings per treatment were recorded for one year at an interval of four months. Seedlings volume was calculated as [(root colar - diameter)^2 x height or D^2H].

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Percentage of mycorrhizal colonization/cm was calculated as follows (Sharma, 1981):

$$\text{Ectomycorrhizae (\%)} = \frac{\text{Total no. of mycorrhizal lateral rootlets}}{\text{Total No. of lateral rootlets}} \times 100$$

For the estimation of total phosphorus in plant tissue the wet tri-acid digestion procedure was followed as suggested by Allen (1974). Phosphorus was analyzed by molybdenum blue method (Jackson, 1973) and was calculated by the following formula:

$$P\ (\%) = \frac{c\ (\text{mg}) \times \text{solution volume (ml)}}{10 \times \text{aliquot (ml)} \times \text{sample weight}} \times 100$$

Where c = mg P obtained from the graph. The data was processed by analysis of variance (ANOVA).

RESULTS

All the fungal inoculants successfully established on *Schima wallichii* seedlings. The inoculated seedlings have superior growth response over seedlings not inoculated with ectomycorrhizal fungi (Table 1). There were statistically significant differences (Table 2) between the treated and control seedlings in terms of shoot height, leaf length ($P<0.05$), root length, root colar diameter and seedlings volume ($P<0.01$). Among the fungal inoculants, *Boletus edulis* proved superior for all the studied growth parameters in both the plots.

infected seedlings were able to generate numerous lateral roots of greater length, creating more sites and allowing a deeper penetration into soil where the unassociated roots cannot reach. The extensive development of the root system in the infected seedlings might have accounted for greater nutrient exploitation zone to the mycelium resulting in better growth of seedlings.

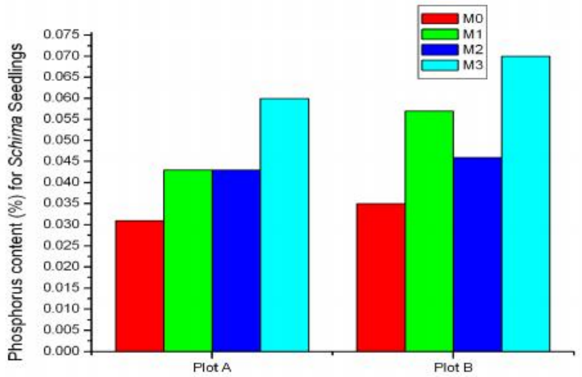

Figure 1 Phosphorus Content (%) in Schima wallichii Seedlings

M_0 = Control *M_1= Scleroderma citrinum*
M_2 = Suillus bovinus *M_3= Boletus edulis*

The capability of fungal inoculants to synthesize biologically active substances (Jackson *et al*., 1964); production and supply of growth regulators (Heinrich *et al*., 1989); and greater access to water (Stribley, 1987) can positively change the root physiology, growth,

Table 1 Effect of ectomycorrhizal on the growth of *S. wallichii* seedlings

Growth Parameters		Plot A				Plot B			
		M_4	M_8	M_{12}	M_{16}	M_4	M_8	M_{12}	M_{16}
Shoot height (cm)	M_0	7.4	11.8	39.5	102.3	7.5	11.8	33.6	110.6
	M_1	7.7	13.6	37.9	112.3	7.9	13.8	38.6	130.2
	M_2	7.5	13.0	34.1	107.1	7.7	13.6	35.0	109.0
	M_3	7.7	13.9	38.0	129.0	7.8	13.9	43.2	136.5
Leaf length (cm)	M_0	5.5	7.5	13.3	14.9	4.8	7.5	13.1	16.3
	M_1	6.9	10.3	16.8	18.5	5.3	10.6	16.8	19.6
	M_2	6.6	10.1	16.4	16.9	4.5	9.7	16.6	19.4
	M_3	6.8	10.4	16.5	19.5	5.3	10.7	16.9	20.0
Root length (cm)	M_0	7.0	11.2	14.6	28.1	7.1	11.3	14.5	28.3
	M_1	7.4	12.6	16.4	34.8	7.7	12.4	16.5	36.5
	M_2	7.3	12.4	16.4	35.5	7.4	12.5	16.4	36.5
	M_3	7.5	12.5	16.4	38.1	7.6	12.8	16.7	40.6
Root colar diameter (cm)	M_0	0.16	0.21	0.26	0.38	0.16	0.20	0.26	0.39
	M_1	0.18	0.24	0.32	0.50	0.19	0.26	0.32	0.52
	M_2	0.17	0.24	0.30	0.47	0.18	0.25	0.30	0.48
	M_3	0.19	0.24	0.34	0.53	0.19	0.26	0.34	0.55
Seedlings volume (cm^3)	M_0	0.18	0.49	0.99	4.06	0.18	0.45	0.98	4.31
	M_1	0.24	0.73	1.68	8.72	0.28	0.84	1.69	9.88
	M_2	0.21	0.71	1.46	7.86	0.23	0.78	1.48	8.42
	M_3	0.27	0.76	1.91	10.73	0.27	0.86	1.93	12.19

Each value is the mean of 5 replication.
M_0 = Control
M_1 = *Scleroderma citrinum*
M_2 = *Suillus bovinus*
M_3 = *Boletus edulis*

The uptake of phosphorus was greatly enhanced in seedlings colonized by ectomycorrhizal fungi (Figure 1). The results of ANOVA showed a significant variation in phosphorus ($P<0.01$) content between the treated and un-inoculated control (Table 3). *Boletus edulis* gave the best result with respect to uptake of phosphorus by colonized seedlings in both the plots.

DISCUSSION

Ectomycorrhizal fungi are well known to boost the ability of plants to deal with various environmental stresses. The seedlings infected with ectomycorrhizal fungi showed a better growth response as compared to unassociated seedlings. The development and morphogenesis (Barker and Tagu, 2000; Chalot *et al*., 2002; Luo *et al*,. 2009). Another benefit that seedling inoculation can bring is, the increased resistance against biotic (plant pathogen) and abiotic (presence of toxic elements) stresses (Chalot *et al*., 2002; Garbaye 2000; Hall 2002; Smith and Read, 2008).

The root systems of ectomycorrhizal seedlings were capable of increased absorption of phosphorus, which could be only due to the formation and network of ectomycorrhizae and their ability to solubilize unavailable nutrients. The beneficial role of mycorrhiza with respect to the uptake of nutrients appears to be related to the nutrient depletion zone. Phosphorous is less soluble and relatively immobile in soil. The development and extent of extraradical hyphae is thought to play a major role in overcoming the P_i depletion zone (Plassard and Dell, 2010). The extraradical mycelium extends beyond the phosphorous depletion zone thereby increasing the uptake volume of the integrated root fungus system. The production of organic acids by ectomycorrhizal fungi which may release phosphorus from organic and inorganic forms by lowering the soil pH or chelation of metal ions (Malajczuk and Cromack, 1982) and; the ability to store and efficiently translocate phosphorus into the host (Grove and Le Tacon, 1993) may also contribute to the increased uptake of phosphorus.

Table 2 Analysis of variance (F) values of mycorrhizal and non-mycorrhizal with various parameters.

	Source of variance	**Variation between mycorrhizal and non - mycorrhizal**
Schima wallichii	Shoot height	67.90*
	Leaf length	67.60*
	Root length	8.4**
	Root colar diameter	45.1**
	Seedlings volume	26.1**

*****= *Significant at p < 0.05 level* **= *Significant at p < 0.01 level*

Boletus edulis was the most efficient ectomycorrhizal fungi for *Schima wallichii* as it significantly increased the growth and uptake of nutrients by effectively colonizing the roots. The differential response of the host plant to different fungal inoculants could be attributed to the amount of ectomycorrhizae formed and the efficiency of mycobionts to improve the growth of host under varied ecological conditions (Browning and Whitney, 1993). The results are in agreement with the statement that ectomycorrhizal association in tree seedlings is far better than no such associations at all and; some species of ectomycorrhizal fungi have proven to be more beneficial to trees under certain environmental conditions than others (Agarwal and Sah, 2009).

Table 3 Analysis of variance (F) values of mycorrhizal and non-mycorrhizal with various parameters.

Analysis of Variance (F) Values of Mycorrhizal and Non-mycorrhizal with Various Parameters		
Schima wallichii	**Source of Variance**	**Variation BetweenMycorrhizal and Non- mycorrhizal**
	Phosphorus	11.2**

** = *Significant at p < 0.01 level*

CONCLUSION

Schima wallichii is an important plant which finds many uses for the local people of the region. It produces good firewood and yields a medium-weight to heavy hardwood which is moderately durable and resistant to dry wood termites. The local people use the wood for construction that is under cover such as columns, beams, for flooring, interior fitting, panelling, door and window frames etc. The experimental inoculation of seedlings with ectomycorrhizal fungi like *Boletus edulis* holds a great promise for effective use of abandoned jhum lands. Large scale reforestation programmes can be attempted by using *Schima wallichii* seedlings which is of great economic value to the local population.

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