

FIELD PERFORMANCE OF ARBUSCULAR MYCORRHIZAL
FUNGI ON VEGETATIVE GROWTH OF MAHOGANY
(*Swietenia macrophylla* King.) SEEDLINGS

By

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THESIS

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DEPARTMENT OF FOREST BIOLOGY AND TREE
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2018

DECLARATION

I hereby declare that this thesis entitled “Field performance of arbuscular mycorrhizal fungi on vegetative growth of mahogany (*Swietenia macrophylla* King.) seedlings” is a bonafied record of research done by me during the course of research and that the thesis has not previously formed the basis for the award of any degree, diploma, fellowship or other similar title, of any other University or society.

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CERTIFICATE

This is to certify that the project entitled, entitled “**Field performance of arbuscular mycorrhizal fungi on vegetative growth of mahogany (*Swietenia macrophylla* King.) seedlings**” is a record of research work done independently by **Mr. Satyabrata Nayak** (2015-17-013) under my guidance and supervision and it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to her.

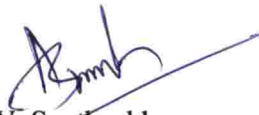
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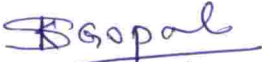
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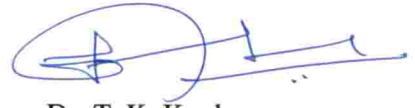
We, the undersigned members of advisory committee of **Mr. Satyabrata Nayak** (2015-17-013) a candidate for the degree of Master of Science in Forestry agree that this thesis entitled “**Field performance of arbuscular mycorrhizal fungi on vegetative growth of mahogany (*Swietenia macrophylla* King.) seedlings**” may be submitted by Mr. Satyabrata Nayak (2015-17-013), in partial fulfilment of the requirement for the degree.



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Finally, I bow my head before the ALMIGHTY.



Satyabrata Nayak

Dedicated

to

my beloved parents

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INTRODUCTION

INTRODUCTION

Swietenia macrophylla King. is an important tree species known all over the world for its timber. *S. macrophylla* has been planted extensively in Southern Asia including India, Sri Lanka, Indonesia and Phillipines (Krisnawati *et al.*, 2011).

Arbuscular Mycorrhizal Fungi (AMF) are found associated with about 80% of the plant species (Cekic *et al.*, 2012). They help in enhancing the biomass of the plants through improving the plant nutrition absorption (especially in phosphorous acquisition) improving soil structure, enhance resistance against drought and pathogens. The plant and AMF interaction has received greater attention especially under stress conditions, where without fungal symbionts, most trees cannot acquire soil resources to grow (Lambers *et al.*, 2008). However, it is true that without trees the AMF cannot carry out their life cycle. Arbuscular mycorrhizal fungi are thus considered as the primary vector for nutrient exchange between soil and the host plant. *Swietenia macrophylla* is mostly found to be associated with four genera of AMF which includes *Glomus*, *Acaulospora*, *Gigaspora* and *Ambispora* and the diversity of the fungi are found more in older trees than younger seedlings (Rodríguez-Morelos *et al.*, 2014).

Different studies have showed that inoculation of AMF during nursery stages have helped in increasing the growth and productivity of seedlings like *Tectona grandis* (Ajeesh, 2015), *Santalum album* (Binu *et al.*, 2015) *Populus x Canescens* (Beniwal *et al.*, 2010), *Fagus sylvatica* (Beniwal *et al.*, 2011), *Albizia lebbbeck*, *Acacia nilotica* (Rahangdale and Gupta, 1998) and *Dalbergia sissoo* (Sahgal *et al.*, 2004). Under field conditions, AMF help tree seedlings to withstand different stress conditions, increase their survival rate and productivity in many species like *Cassia siamea* (Giri *et al.*, 2005), *Sesbania aegyptiaca* and *S. grandiflora* (Giri and Murkhaji, 2004).

The benefits of AMF in *Swietenia macrophylla* seedlings in polybags has been established (Ajeesh *et al.*, 2017), no information on physiological and growth performance of *S. macrophylla* inoculated with AMF under field conditions are

available. Considering this, an experiment on *Swietenia macrophylla* King. was carried out with an objective to study the effect of arbuscular mycorrhizal fungi (AMF) on the growth and physiology of *Swietenia macrophylla* seedlings under field conditions.



REVIEW AND LITERATURE

REVIEW AND LITERATURE

Swietenia macrophylla King., known as more popularly “Big leaf mahoagany”, “Amazonian mahogany” or simply “mahogany”, is an important tropical tree species of great economic value. *S. macrophylla* naturally occurs in a wide range of soils and environmental conditions of the neo-tropical realm, which includes countries like Brazil, Belize, Bolivia, Costa Rica, Colombia, Guatemala, Ecuador, El Salvador, Honduras, Nicaragua, Peru, Panama, Mexico and Venezuela (Krisnawati *et al.*, 2011). The growing demand for timber and uncontrollable logging in its natural forests, has led to the decline in the population of *S. macrophylla* in most of these countries (de Barros *et al.*, 2013). *S. macrophylla* plantations are growing extensively in the Southern Asian and Pacific countries like India, Sri Lanka, Indonesia and Philippines for its demand and popularity (Soerianegara and Lemmens, 1993).

Application of chemical fertilizers in plantations during early stages of the plantations is essential to obtain better yield and maintaining the trees in a plantation. But these chemical fertilisers create problems for the natural environment by polluting the environment and misbalancing the nutrient cycles. The use of chemicals in soil amounts more cost and energy. So as an alternative, bio-fertilizers are preferable instead of chemical fertilizers (Guleri, 2001). Arbuscular mycorrhizal fungi (AMF) are one of the best bio-fertilizer available for its capability to transport essential nutrients and water to the plants.

Arbuscular mycorrhizal fungi are the obligate mutualistic symbionts that colonize roots of about 80 per cent of the plant species (Smith and Read, 1997; Giovannetti, 2008; Zobel and Öpik, 2014). These plants comprise of gymnosperms and angiosperms (Smith and Read, 2008). The application of AMF during seedling stage of trees help the plant in better establishment (Dhawal *et al.* 2016). The AMF obtain carbon from the plants for their life cycle, and simultaneously plants get numerous benefits from AMF such as nutrient and water (Walder *et al.*, 2012).

2.1 Benefits of AMF as bio-fertilizer

Arbuscular mycorrhizal fungi play an important role in soil aggregate stabilization, and better soil structure improves the water holding capacity, microbial activity and nutrient cycles (Posada *et al.*, 2018; Pal and Pandey, 2014). It helps in improving the soil fertility status by changing the soil physical, chemical and biological properties. Arbuscular mycorrhizal fungi are often considered as natural bio-fertilizers (Berruti *et al.*, 2016). The growth of external hyphae into the soil helps in formation of good soil structure by formation of micro-aggregates and protein secreted by AMF (Glomalin) works as a gluing agent to hold the particles together. The soil chemical properties are also altered by AMF. Arbuscular mycorrhizal fungi help in the mining of low available nutrients (phosphorus and micro-nutrientas) and alleviating toxic minerals from soil. Mycorrhiza also help in protecting plants from different disease and pest attack by altering the nutritional status of the plants and also helps in creating better environment for other beneficiary micro-organisms (Cardoso and Kuyper, 2006). Arbuscular mycorrhizal fungi can alter the pH of plant rhizosphere region which make suitable condition for better availability of some nutrients to plants (Giri *et al.*, 2005). Studies showed that AMF inoculation can reduce the acidity of the soil and can increase organic carbon level in soil (El Mrabet *et al.*, 2014).

2.2 AMF and nutrient uptake

The major role AMF play in soil, is to help the plant in nutrient uptake from the soil. By using the extended hyphal network, AMF efficiently transport mineral nutrients and water to the plant roots (George *et al.*, 1995, Babikova *et al.*, 2013; Kumar *et al.*, 2017; El Mrabet *et al.*, 2014). The nutrient exchange mainly occurs between the root cortical cells and the arbuscules of AMF. Arbuscules are highly branched hyphal structures of AMF present separately within the root cortical cells while not entering into the cytoplasm (Anwar *et al.*, 2008). These AMF are seen work more efficiently towards to the sparingly available nutrient sources like phosphorus (Tarafdar and Kumar, 1996). Phosphorus is an essential nutrient for plant to complete its life cycle and is non-substitutable. This major macronutrient

is not accessible for the plants due to its unique chemistry. Arbuscular mycorrhizal fungi are the key element for the plant to get access to P by extending the surface area of roots using their hyphae (Bucher, 2007).

Various studies on different tree species showed that increased phosphorous intake was recorded with application of AMF. Rajan *et al.* (2000) found that inoculation with different species of AMF increases the P level in *Tectona grandis*. Similar result were obtained in different experiments in different tree species e.g. *Acacia mangium* (Jeyanny *et al.*, 2011), *Casuarina equisetifolia* (Zhang *et al.* 2010), *Azadirchta indica* (Muthukumar *et al.*, 2001), *Casia siamea* (Giri *et al.*, 2005), *Prosopis cineraria* (Verma *et al.*, 2010), *Albizia saman* and *Paraserianthes falcataria* (Wulandari *et al.*, 2016). Studies showed the survival percentage was higher in inoculated seedlings in field conditions (Giri *et al.*, 2005; Wulandari *et al.*, 2016)

Studies showed not only phosphorous, but also other micro nutrients uptake were increased with AMF inoculation. In a pot culture experiment by Wu *et al.* (2011) on Peach (*Prunus persica*) seedlings using three AMF species (*Glomus mosseae*, *G. vesiforme*, and *Paraglomus occultum*), observed that inoculation with AMF helped in increasing K, Mg, Fe, and Zn concentration in both roots and leaves of the plant, whereas Ca concentration found higher in leaves and Cu and Mn concentration found higher in roots of AMF inoculated seedlings than the non-inoculated seedlings. This experiment showed the dependency of plants on the AMF to acquire the nutrients other than phosphorus. A study by Giri *et al.* (2005) on *Cassia siamea* seedlings showed that under nursery conditions, Cu, Zn, and Na uptake of seedlings increases when inoculated with two AMF species (*G. fasciculatum* and *G. macrocarpum*). Another study by Verma *et al.* (2010) concluded in increased uptake of Zn and Cu by *Prosopis cineraria* seedlings when inoculated with a mixed inoculum of AMF. Study by Rajan *et al.* (2000) showed different species of AMF inoculation increases the Zn and Cu content in *Tectona grandis* seedlings.

2.3 AMF and plant disease control

The use of chemical fertilizers for controlling various disease and pest attacks, have detrimental effect on the environment. The host root growths are modified by AMF, which change the physiological and biochemical characters by altering the nutrition status of the plant (Anwar *et al.*, 2008). Various studies show that use of AMF enhance the plant resistance towards to diseases and pests (Borowwicz, 2001; Veresoglou and Rilling, 2011; Eyles *et al.*, 2010; Birhane *et al.*, 2018; Wehner *et al.*, 2010).

2.4 AMF and drought tolerance

Water is the most important factor for seedling growth and establishment. The characteristic hyphal growth help the plant to get access to the water. The external mycelium of AMF allows the plant to spread beyond its root exploration zone (Khan *et al.*, 2000). Many studies showed the importance of AMF on improving the drought tolerance of host plants (Murkhaji and Kapoor, 1986; Cuenca *et al.*, 1997; Caravaca *et al.*, 2003). An experiment conducted by Khalvati *et al.* (2005) showed that under drought conditions AMF inoculated seedlings although showed no increase in plant biomass, but had more leaf elongation, photosynthesis rate, and stomatal conductance than the non-inoculated plants. He founded that hyphal network of AMF can store small amount of water and these water was transported to plants under the drought conditions, which further proved by the presence of large amount of the hyphal compartment in roots of the plants.

Zhang *et al.* (2010) studied the effect of AMF on *Casuarina equisetifolia* under drought condition in a greenhouse experiment. They found that, inoculation with AMF improved the drought tolerance of the seedlings by increasing the survival percentage and biomass of the seedlings compared to non-inoculated seedlings. The AMF inoculated seedlings also showed higher P, soluble sugar and soluble protein concentration. But, AMF showed no significant influence on seedling height.

2.5 AMF and heavy metal stress

Metals like Cd, Hg, Pb, and As are toxic to plants even in a very low concentration (Mertz, 1981). These toxic metals were stored in the mycorrhizal structures of spores and roots which make them unavailable to the plants (Meharg and Caimey, 2000; Hildebrandt *et al.*, 2007; Hailemariam *et al.*, 2018).

2.6 AMF and *S. macrophylla*

The relationship of *S. macrophylla* with arbuscular mycorrhizal fungi (AMF) has been poorly studied even though the species has been introduced for cultivation in plantations and agroforestry systems of South Asian tropical forests (Rodríguez-Morelos *et al.*, 2014). Studies showed that trees of meliaceae family have been associated with five genera of arbuscular mycorrhizal fungi (AMF) which are *Glomus*, *Acoulospora*, *Gigaspora*, *Entrophospora*, and *Scutellospora* (Shi *et al.*, 2006). *S. macrophylla* has been found to be associated with four genera of arbuscular mycorrhizal fungi (AMF) dominated with *Glomus*, *Acoulospora*, *Gigaspora* and *Ambispora* (Rodríguez-Morelos *et al.*, 2014). Recent studies showed that inoculation of AMF with *S. macrophylla* seedlings in nursery can increase the growth and its tolerance towards drought stress (Ajeesh *et al.*, 2017; Rajan, 2016).

2.7 AMF and growth in nursery

Tree seedlings inoculated with AMF in nursery helps them to grow more vigorously and made them healthy (Jha *et al.*, 2017) and make them suitable for planting in the field (Navarro-Garcia *et al.*, 2011). Many studies had been conducted to know the performance of various tree seedlings inoculated with AMF in nursery and increase in height, collar diameter, and the number of leaves were reported.

Ajeesh *et al.* (2017) showed that the height, collar diameter, number of leaves of *Swietenia macrophylla* and *Tectona grandis* seedlings increases with addition of different AMF species (*F. mosseae*, *G. intraradices*, and *G. proliferum*). Similar result on *S. macrophylla* seedlings with AMF association found by Rajan

(2016) under different drought conditions. Rajan *et al.* (2000) also found that the height and stem girth of *Tectona grandis* seedlings increased significantly by using nine different species of AMF. Ghosh and Verma (2006) reported that *Acacia mangium* seedlings showed significant higher shoot height, collar diameter, leaf area and chlorophyll content when inoculated with AMF in sterilized soil. Jha *et al.* (2017) conducted an experiment on growth of *Azadirachta indica*, *Jatropha curcas*, *Madhuca indica* and *Pongamia pinnata* seedlings using 11 AMF species. He found *A. indica* and *M. indica* had positive response in growth towards AMF inoculation but the *J. curcas* and *P. pinnata* showed no such responses.

Similar results were obtained with different tree species like *Casuarina equisetifolia* (Zhang *et al.* 2010), *Acacia mangium* (Jeyanny *et al.*, 2011; Ghosh and Verma, 2006), *Azadirachta indica* (Muthukumar *et al.*, 2001), *Prunus persica* (Wu *et al.*, 2011), *Tectona grandis* (Rajan *et al.*, 2000; Ajeesh *et al.*, 2017), *Citrus tangerine* (Wu *et al.*, 2006), *Casia siamea* (Giri *et al.*, 2005), *Santalum album* (Binu *et al.*, 2015), *Swietenia macrophylla* (Ajeesh *et al.*, 2017; Rajan, 2016), *Albizia saman* and *Paraserianthes falcataria* (Wulandari *et al.*, 2016). *Argania spinosa* (El Mrabet *et al.*, 2014), *Citrus* spp (Ortas and Ustuner, 2014), *Manihot esculenta* (Sridevi and Ramakrishnan, 2013), *Pinus halepensis* (Querejeta *et al.*, 1998), *Albizia saman* and *Paraserianthes falcataria* (Wulandari *et al.*, 2016) and *Bauhinia faberi* (Yamin *et al.*, 2016).

2.8 AMF and growth in field

Arbuscular mycorrhizal fungi (AMF) found to be have beneficial influence to all plant seedlings in both nursery and field (Calvet *et al.*, 2004). Arbuscular mycorrhizal fungi application in nursery helps the seedlings in better establishment in the field and withstand drought, pathogen attack and nutrient deficiency (Wilson *et al.*, 1991). A study by Jasper *et al.* (1989) showed that P intake by *Acacia* spp. increased with application of AMF. It was also found seedlings showed increased growth with addition of P fertilizers and AMF. However, without AMF inoculation seedlings had shown no substantial growth. The experiment proved the dependency of *Acacia* seedlings on AMF to absorb phosphorus from soil. As the soil had very

low phosphorous level, even after application of phosphorus fertilizer the seedlings were unable to absorb it from the soil.

Wulandari *et al.* (2016) conducted an experiment on *Albizia saman* and *Paraserianthes falcataria* inoculated with three native AMF species namely *Rhizophagus clarus*, *Gigaspora decipiens*, and *Scutellospora* spp. in nursery for six months period to get appropriate size seedlings for field planting. He found that shoot dry weight of *A. saman* had no difference among the inoculated and non-inoculated seedlings in nursery, however there was a better performance of shoot dry weight in inoculated *P. falcataria* seedlings than the non-inoculated seedlings. Increased P and N contents were also reported in the inoculated seedlings in nursery. After field planting it was found that after seven months of planting the inoculated seedling showed better height and stem diameter than the non-inoculated in *A. saman* seedlings. But there was no effect of AMF in *P. falcataria* seedlings in the field. The plant nutrient contents (P and N) was found to be improved in the inoculated seedlings in the field conditions.

Estaun *et al.* (2003) conducted an experiment on Olive trees (*Olea europaea*) with two AMF species (*Glomus mosseae* and *Glomus intraradices*) in both nursery and field conditions, to assess the long-term effect of AMF inoculation to olive plants in field. In the nursery experiment no significant difference was found between the inoculated and non-inoculated seedlings for the four months. In the field experiment it was found that the growth of the seedlings increased upto six months due to rapid establishment of the symbiosis in the inoculated plants, but after that the difference between growth parameters among the inoculated and non-inoculated seedlings diminished as the non-inoculated seedlings developed symbiosis with the native AMF species.

2.9 Consortium of AMF

In a natural ecosystem forest trees usually associated with more than one AMF species (Clapp *et al.*, 1995). Studies showed that consortium of AMF can get better result than the individual use of a single species (Shukla *et al.*, 2014; Wehner *et al.*, 2010; Neetu *et al.*, 2011; Ghosh and Verma, 2011; Jansa *et al.*, 2008; Muthukumar and Udaiyan, 2018).

Neetu *et al.* (2011) showed combination of two native AMF species (*Glomus mosseae* and *Acaulospora laevies*) and with other bio-inoculants (*Trichoderma viride* and *pseudomonas fluorescens*) to *Linum usitatissimum* resulted in increased both biometric and physiological growth parameters than the non-inoculated seedlings when applied either individually or in combinations. The phosphorous content recorded highest in case of combination of all the inoculants.

A study was conducted by Ortas and Ustuner (2014) to examine the difference between single and dual AMF inoculation on *Citrus aurantium* seedlings in nursery condition. The dual inoculation of AMF species found to have more P contents and root colonization percentage than the single inoculation of AMF. However, all the AMF inoculated seedlings showed increased height, diameter, shoot dry weight and nutrient contents than the non-inoculated seedlings.

Mixed application of AMF and other bio-inoculants also found to be helpful for the plants for their growth and development (Manjunath and Bagyaraj, 1984). A study by Muthukumar *et al.* (2001) showed that *Azadirchta indica* seedlings showed better shoot height, collar diameter, leaf number, phosphorous, nitrogen, and potassium content when inoculated with *Glomus intraradices*, *Azospirillum brasilense* and phosphorous solubilizing bacteria (PSB) together. It was also observed that root colonisation of the seedlings increased with application of *Azospirillum brasilense* and phosphorous solubilizing bacteria (PSB). Another experiment by Young (1990), showed mixed inoculation of AMF and phosphorous solubilizing bacteria (PSB) can increase the growth and P uptake of *Acaci mangium*, *A. confusa*, *Leucena leucocephala*, and *Liquidamber formosana* seedlings.

Muthukumar and Udaiyan (2018) also found that inoculation with AMF and PSB increased the growth and nutrient uptake of *Acacia auriculiformis* seedlings.

A field experiment was conducted by Siviero *et al.* (2008) to study the growth and survival percentage of *Schizolobium amazonicum* seedlings inoculated with combination of three AMF species (*Glomus clarum*, *Glomus intraradices* and *Glomus etunicatum*) and three N-fixing bacteria strains (two *Rhizobium* sp. and one *Burkholderia* spp.). They found that inoculation with *G. intraradices* was more effective among the AMF species and the bacteria strains showed no effect on the seedling growth. However, dual inoculation of AMF and bacteria strains showed the best result. The survival percentage found to be lowest in case of non-inoculated plants. The study showed that microbial combinations were more effective in stimulating the plant growth.

2.10 AMF and physiological factors

Arbuscular mycorrhizal fungi also help the plants to stimulate their growth by increasing the photosynthesis rate, resisting soil-borne pathogen attack and improving osmotic adjustment under water stress conditions (Al-Karki, 2006).

Photosynthesis increases when leaf area of plant increases. Using AMF as inoculation can improve plant nutrition and growth which resulted in larger leaf area and higher photosynthesis rate (Kaschuk *et al.*, 2009). Under drought conditions, AMF alter the water relationships of plants so that they can improve their resistance towards the drought (Auge, 2001; Smith and Read, 2008). During such situations plants get water through the hyphae of AMF which can reach up to micro-pores in soil. Stomatal conductance is also an important factor for plant as it determines the rates of CO₂ enters and water vapour exit through transpiration. As the water relation of plants positively influenced by AMF inoculation the stomatal conductance of AMF inoculated plants found to be higher than the non-inoculated plants (Auge *et al.*, 2015, Shinkafi, and Aduradola, 2009).

A study was conducted by Birhane *et al.* (2012) on *Boswellia papyrifera* seedlings to assess the effect of AMF on photosynthesis, water use efficiency and

growth of the seedlings. In that experiment it was found that transpiration rate was not affected by AMF inoculation, but stomatal conductance and photosynthesis rate were higher in inoculated seedlings than the non-inoculated seedlings. Wu *et al.* (2011) found that inoculation of AMF under different temperature stress in *Citrus* spp., seedlings showed different photosynthesis rate, transpiration rate and stomatal conductance characters. When temperature was low (15 °C), all the parameters showed no significant difference, but when temperature increased to 25°C there was a significant increase in photosynthesis rate, transpiration rate and stomatal conductance in inoculated seedlings than non-inoculated seedlings.

Ajeesh *et al.* (2017) and Rajan (2016) found that inoculation with AMF can increase the photosynthesis rate, stomatal conductance and transpiration rate under nursery conditions. Another experiment by Ginadaba *et al.* (2005) resulted in reduced photosynthesis rate and stomatal conductance with increase in drought stress in *Eucalyptus* seedlings.

Various studies recorded higher physiological characters were observed when inoculated with AMF in tree seedlings like *Bauhinia faberi* (Yamin *et al.*, 2016), *Dalbergia sisoo* (Bisht *et al.*, 2009) and *Azadirachta excelsa* (Huat *et al.*, 2002).

2.11 AMF and Site factors

The AMF population largely varies in the field due to various factors, like vegetation, soil moisture etc. (Schreiner, 2003). The AMF do not affect tree species equally due to their different physiological interaction with different plants (Huat *et al.*, 2002).

The growth of AMF also found adversely by precipitation, temperature, total nitrogen and available potassium (He *et al.*, 2016). The soil pH also plays an important role in altering the AMF population as *Glomus* species can be found abundantly at 6.8 pH, whereas *Acaulospora* species are found mostly at 4.5 to 4.9 pH (Porter *et al.*, 1987). A study by Giri *et al.* (2005) showed that the soil pH of rhizosphere region of *Cassia siamea* seedlings can be changed using AMF. The pH

was reduced from 8.5 to 7.8. The reduction in pH, resulted in availability of some nutrients to the seedlings, which helped them in increasing growth, biomass production and nutrient accumulation.

Spore density of AMF depends on the site vegetation i.e. Poor vegetation shows maximum AMF spore density while dense vegetation shows minimum AMF spore density. However, a reverse ratio was observed in case of root colonization (Birhane *et al.*, 2018).

Seasonal variation also affects the AMF colonization and species diversity (Pagano *et al.*, 2009). Unfavourable conditions increased the dependency of plants on micro-organisms, resulted in high spore density in poor vegetation. Similarly it was also observed that low soil nutrient content can lead to more spore density of the AMF (Thrall *et al.*, 2007).

Soil moisture content can also change the spore density. The spore density was generally high in dry conditions than the wet conditions (Birhane *et al.*, 2018). The root colonization percentage increases with increase in precipitation level due to availability of more photosynthetic materials to the roots which resulted in AMF sporulation in abundant manner (Kivlin *et al.*, 2013) The variation is due to more soluble nutrients released to soil during rainy season through litter decomposition (Scotti and Correa, 2004). The spore density was found to have negative relation with soil organic carbon, total nitrogen and potassium, whereas the root colonization percentage had positive relation with carbon content of the soil and negative relation with total nitrogen and potassium content (Birhane *et al.*, 2018). In neutral and slightly alkaline soil conditions, the spore density was found to be high (Sreevani and Reddy, 2004).

A study by Ouzounidou *et al.* (2015) showed that *Salvia hispanica* seedlings inoculated with AMF at different pH level showed different results. The growth of the seedlings were found to be high in neutral (7.1) and alkaline pH (8.2), while stunted growth was observed in acidic pH (5.1).



MATERIALS AND METHODS

MATERIALS AND METHODS.

The research project on 'Field performance of arbuscular mycorrhizal fungi on vegetative growth of mahogany (*Swietenia macrophylla* King.) seedlings' was conducted at College of Forestry nursery and Instructional farm of Kerala Agricultural University, Vellanikkara, Thrissur district, Kerala.

The nursery experiment was carried out to select most efficient arbuscular mycorrhizal fungi (AMF) treatments for the field experiment. In nursery, different species of AMF viz *Funneliformis mosseae*, *Acaulospora mellea*, and *Glomus etunicatum* were inoculated to *Swietenia macrophylla* seedlings either alone or in combination along with a non-inoculated control. The experiment was laid out in a factorial completely randomized design with 22 treatments combinations (Table 1) and three replications.

Table 1. Details of arbuscular mycorrhizal fungi and doses used in the nursery experiment.

Treatments	Individual and Combinations of arbuscular mycorrhizal fungi and doses
T1	<i>Funneliformis mosseae</i> 5g
T2	<i>Funneliformis mosseae</i> 10g
T3	<i>Funneliformis mosseae</i> 15g
T4	<i>Acaulospora mellea</i> 5g
T5	<i>Acaulospora mellea</i> 10g
T6	<i>Acaulospora mellea</i> 15g
T7	<i>Glomus etunicatum</i> 5g
T8	<i>Glomus etunicatum</i> 10g
T9	<i>Glomus etunicatum</i> 15g
T10	(<i>Funneliformis mosseae</i> + <i>Acaulospora mellea</i>) 5g
T11	(<i>Funneliformis mosseae</i> + <i>Acaulospora mellea</i>) 10 g
T12	(<i>Funneliformis mosseae</i> + <i>Acaulospora mellea</i>) 15 g

T13	(<i>Funneliformis mosseae</i> + <i>Glomus etunicatum</i>) 5 g
T14	(<i>Funneliformis mosseae</i> + <i>Glomus etunicatum</i>) 10 g
T15	(<i>Funneliformis mosseae</i> + <i>Glomus etunicatum</i>) 15 g
T16	(<i>Acaulospora mellea</i> + <i>Glomus etunicatum</i>) 5 g
T17	(<i>Acaulospora mellea</i> + <i>Glomus etunicatum</i>) 10 g
T18	(<i>Acaulospora mellea</i> + <i>Glomus etunicatum</i>) 15 g
T19	(<i>Funneliformis mosseae</i> + <i>Glomus etunicatum</i> + <i>Acaulospora mellea</i>) 5 g
T20	(<i>Funneliformis mosseae</i> + <i>Glomus etunicatum</i> + <i>Acaulospora mellea</i>) 10 g
T21	(<i>Funneliformis mosseae</i> + <i>Glomus etunicatum</i> + <i>Acaulospora mellea</i>) 15 g
T22	Control

The performance of the seedlings in nursery recorded and four of the best treatments and control were selected for planting in the field. The field experiment was laid out in a randomized block design with five treatments and three replications.

Mass multiplication of arbuscular mycorrhizal fungi (AMF)

Pure cultures of *Funneliformis mosseae*, *Acaulospora mellea*, and *Glomus etunicatum* were collected from TERI (The Energy Research Institute, New Delhi). Pure culture had a total spore count of 500 spores per 50g. Vermiculite was used for mass multiplication. The medium was sterilized using an autoclave to make it free from any microorganisms. Maize (*Zea mays*) seeds were used as host plants. The maize seeds were sterilized with 0.1 per cent mercuric chloride for five minutes and washed thoroughly with distilled water. The vermiculite were filled into growbags of capacity 5 kg and maize seeds were sown in it. The pure cultures of AMF were added in to the grow bags, the grow bags were irrigated regularly and at 10 days interval 50ml of 's solution (Hogland and Arnon, 1950) was applied.



Plate 1. Mass multiplication of AMF. A- Pure AMF culture. B- Mass multiplication using vermiculite as growth media and maize as host plant. C- Application of Hogland's solution as source of nutrients. D. Mixture of vermiculite and maize roots packed and stored



Plate 2. Soil fumigation. A- Transferring potting mixture (Sand, Coir pith and FYM mixture in 3:2:1) on to polythene sheet. B- Preparation of 5% formaldehyde. C- Spraying of formaldehyde on potting mixture. D. Mixing of soil and formaldehyde. E- Covering potting mixture with polythene sheet. F- Stones and soil placed over the edges to avoid direct contact to soil.

Root colonisation percentage and the total spore count were monitored for the *Zea mays* plants on the 45th day after sowing. When the root colonisation percentage found to be above 80 % and total spore count value recorded 100 spores for 10gs of medium, the shoot of the plants were removed and the roots were mixed up with vermiculite. This vermiculite and root mixture were then stored in a cool place and used as inoculant for further experiments.

Fumigation of soil mixture, Seedling preparation and AMF inoculation

Potting mixture was made by combining soils, coir pith and farm yard manure, which were collected, mixed up and transported on to a polythene sheet. The potting mixture then fumigated with 5 per cent formaldehyde and covered up with another polythene sheet for the next 25 days. Fumigated soil was then filed in to polythene bags.

S. macrophylla seeds were collected from Kerala Forest Research Institute. The seeds were sown directly in the polythene bags and irrigated frequently. The stored AMF inoculants were then weighed and added in the polythene bags which were arranged according to the experimental lay out. For control no AMF inoculants had been added in the polythene bags.

3.1 Experiment – 1

The experiment was conducted in nursery to find out best four treatments for the field plantation. The performance of the seedlings was recorded for three months using the following parameters:

3.1.1 Shoot height

The shoot height was measured non-destructively using a meter scale from the collar region to terminal bud at an interval of 30 days and expressed in centimetres.

3.1.2 Collar diameter

The collar diameter was measured non-destructively using digital vernier caliper and expressed in millimetres.

3.1.3 Number of leaves

The number of functional leaves were counted and recorded.

3.1.4 Root colonisation percentage

Fresh roots were collected from the nursery after 90 days and washed thoroughly. The roots were cut into 1 cm bits and treated with 10% KOH in a water bath for 60 minutes at 90 °C. The excess KOH was neutralized by using 2% hydrochloric acid for 10 minutes. The root materials are subsequently stained with trypan blue (0.05 %) in lacto-phenol for a period of 10 minutes at 80°C. The stained roots are then observed under compound microscope for the detection of mycorrhizal infection (Phillips and Hayman, 1970).

The root colonisation percentage was calculated using the formula:

$$\text{Root colonisation percentage} = \frac{\text{Number of positive AMF colonisation}}{\text{Total number of root segments observed}} \times 100$$

3.1.5 Total spore count

The numbers of spore of arbuscular mycorrhizal fungi were calculated using wet sieving and decanting technique (Gerdemann and Nicolson, 1963). 50 g of fresh soil samples were taken in a bucket (capacity of 1litre) and 500 ml of water was added. The soil was mixed thoroughly and the unnecessary debris were separated. The soil-water suspension was left for 5 to 10 minutes for settling down of heavy particles. After settling down, the soil water solution was carefully passed through a series of sieves ranging from 600, 300, 212, 150, 105 and 45µm kept one below another order. Each sieve was removed by washing down its content to the lower sieve till 105 µm. The contents of 105 and 45 µm were collected in a 100 ml beaker and transferred onto Whatman No. 1 filter paper placed in a petri dish. The petri

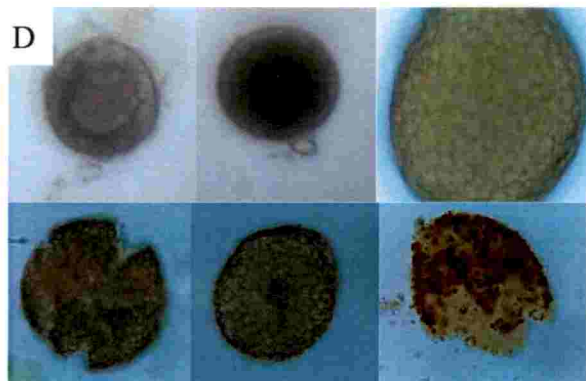
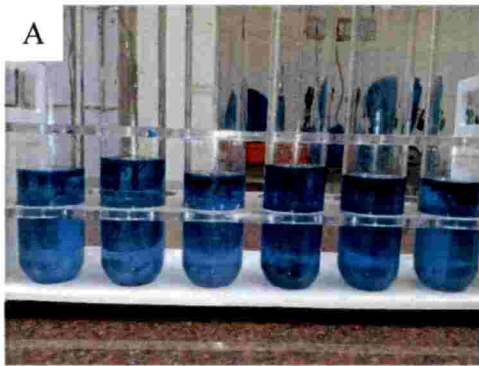


Plate 3. Root colonisation percentage and Total spore count estimation. A,B – Root colonisation percentage estimation using clearing and staining method. C. AMF spore observation using stereo microscope D. Different types of AMF spores.

dish was then observed under stereo microscope and the number of spores were counted and expressed per gram of inoculum.

3.2 Experiment – 2

On the basis of result of Experiment 1, four best performing treatments were selected for out-planting in the field along with the control. Non-destructive observations were taken in the field for 6 months for the following parameters:

3.2.1 *Shoot height*

The shoot height was measured from the collar to shoot tip using a meter scale at 30 days interval and expressed in centimetre.

3.2.2 *Collar diameter*

The diameter of collar was calculated using digital vernier caliper at 30 days interval and expressed in millimetre.

3.2.3 *Number of leaves*

The number of functional leaves were calculated at 30 days interval.

3.2.4 *Photosynthetic rate*

The rate of photosynthesis was calculated using infra-red gas analyser (LI-6400) and expressed in $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ at 30 days interval.

3.2.5 *Stomatal conductance*

The stomatal conductance was calculated using infra-red gas analyser (LI-6400) and expressed in $\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ at 30 days interval.

3.2.6 *Transpiration rate*

The rate of transpiration was calculated using infra-red gas analyser (LI-6400) and expressed in $\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ at 30 days interval.



Plate 4. A,B,C - Preparation of study site and plantation of seedlings.

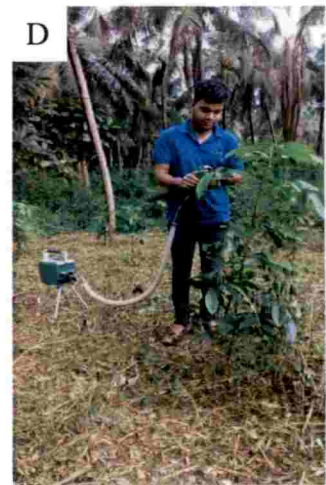


Plate 5. A,B,C,D – Recording physiological observations using IRGA

3.2.7 Leaf temperature

The leaf temperature was calculated using infra-red gas analyser (LI-6400) and expressed in °C at 30 days interval.

3.2.8 Absolute growth rate

The gain in height at a specific time period called absolute growth rate and was calculated using the formula:

$$\text{Absolute Growth Rate} = H_2 - H_1 / T_2 - T_1$$

Where,

H_2 – plant height at time T_2

H_1 – plant height at time T_1

3.2.9 Root colonisation percentage

The collection of root sample was done by non-destructive sampling using a borer near to the rhizosphere of the plant. Roots from different treatments were collected at 60 days interval and examined by using methods of clearing and staining (Phillips and Hayman, 1970).

3.2.10 Total spore count

The soil was collected through non-destructive sampling using a borer from the rhizosphere region. The number spores of arbuscular mycorrhizal fungi were calculated using wet sieving and decanting technique (Gerdemann and Nicolson, 1963) at 60 days interval.

3.2.11 Soil analysis

Soil samples were collected before and after the experiment for analysing the soil nutrient status like total nitrogen, available phosphorous, available potassium, and carbon content. The soil samples were air dried and passed through 0.2mm sieve (Anderson and Ingram, 1993).



Plate 6. A,B,C,D,E,F –Soil and plant nutrients estimation – instruments used.

a) The total nitrogen was calculated using Kjeldahl's method (Bremner, 1965).

One gram of soil sample was transferred into the digestion tube where 10ml of concentrated H_2SO_4 , 1g of digestion mixture along with 0.1g of Se powder were added. The contents were swirled and kept for pre-digestion overnight at room temperature. The content was digested and gently heated initially. When the frothing was ceased temperature was raised to 360 to 400⁰C. A light green coloured or colourless solution was obtained with the completion of digestion. The contents were cooled and transferred to a 100 ml volumetric flask and made up the volume. The aliquot (10 ml) was taken in a distillation flask and 10 ml of 40 % of NaOH was added. The receiving flask (100 ml conical flask) containing 10 ml 4 % boric acid was placed. The lid of the flask was closed and distilled by heating. The distillate was collected in the receiving flask with the evolution of ammonia which changed the pink colour of boric acid in to green. The distillation was continued till the evolution of NH_3 was ceased and the receiving flask was removed and the burner was put off. The distillate was titrated with standard acid (0.01 N HCl) till the green colour disappeared where one drop in excess turned the solution to pink and the blank was run simultaneously.

b) The available phosphorous was calculated using Bray and Kurtz method (Jackson, 1958).

Ten millilitre of sample solution of di- acid extract was pipetted to a 25ml volumetric flask. 5ml of reagent B was added to the distilled water to bring the final volume 25 ml, allowed 10 minutes for colour development and the blue colour intensity was measured at 660 nm in a spectrophotometer. If the colour was deep then a smaller volume the sample was taken. A standard curve was prepared by making up a range of P standards with concentrations of 0 to 1 ppm in the same manner as above (P concentration on X- axis and reading on Y-axis). Concentration of the unknown sample was obtained from the standard curve plotted with different P standards.

The available potassium determined by using ammonium acetate method and subsequently reading were taken using flame-photometer (Jackson, 1958).

c) Available Potassium (Ammonium acetate method):

The soil was ground to pass through 0.5mm sieve from where 0.5g of sample was taken and transferred to Kjeldahl's digestion flask (digestion tube). 10 ml of di-acid was added, was kept it for pre-digestion overnight and then kept on digestion unit at 300°C till it became a clear solution. It was filtered (to remove the sand particle) to 500ml standard flask after cooling and the volume was made up. It was used to read in flame photometer and the flame photometer was calibrated with standard solution of K (0, 5, 10, 20 ppm) and the instrument was set up as per the directions. (For manual model: The flame photometer was adjusted to get zero for blank reading with reagent blank solution. Then the reading was adjusted to get 100 for 20 ppm and 50 for 10ppm solution). After calibration readings of samples were taken.

d) The organic carbon was calculated using Walkley and Black's rapid titration method (Jackson, 1958).

The soil was ground and passed through 0.2 mm sieve from where 1g of soil was taken in a dry 500ml conical flask. 10 ml of 1N $K_2Cr_2O_7$ was pipetted in to it and swirled a little. The flask was kept on an asbestos sheet. Again 20 ml of concentrated H_2SO_4 was added and swirled again 2 to 3 times. The flask was allowed to stand for 30 minutes and thereafter 200 ml of distilled water was added to stop the reactions. 4 to 5 drops of ferronin indicator was added and the contents were titrated with ferrous ammonium sulphate solution till the colour was changed from dark green to chocolate brown colour. Simultaneously a blank was run without soil.

3.2.11.1 Soil pH

Ten gram of soil sample was taken in a beaker to which 25 ml of distilled water was added and stirred rapidly for 5 minutes. The pH was recorded in a digital pH meter.

3.2.11.2 *Electrical conductivity*

Ten grams of soil sample was taken in a beaker and 50 ml distilled water was added into it. The solution was stirred thoroughly. The electrical conductivity was calculated using digital conductivity meter and expressed in mS/cm.

3.2.12 *Plant nutrient status*

Plant leaves were collected, dried, grinded and sieved through a 0.2 mm sieve (Anderson and Ingram, 1993) to determine plant nutrient status viz. nitrogen, phosphorous, potassium and carbon content.

- a) The plant nitrogen was measured using Kjeldahl's method (Bremner, 1965).

Digestion:

1g of powdered plant sample was transferred into the digestion tube where 10ml of concentrated H_2SO_4 , 1g of digestion mixture along with 0.1g of Se powder were added. The contents were swirled and kept for pre-digestion overnight at room temperature. The content was digested and gently heated initially. The content was digested and gently heated initially. When the frothing was ceased temperature was raised to 360 to 400⁰C. A light green coloured or colourless solution was obtained with the completion of digestion. The contents were cooled and transferred to a 100ml volumetric flask and made up the volume. A suitable quantity of the aliquot (5-10 ml) was taken in a distillation flask and 10 ml of 40% of NaOH was added. The receiving flask (100 ml conical flask) containing 10 ml 4% boric acid was placed. The lid of the flask was closed and distilled by heating. The distillate was collected in the receiving flask with the evolution of ammonia which changed the pink colour of boric acid in to green. The distillation was continued till the evolution of NH_3 was ceased and the receiving flask was removed and the burner was put off. The distillate was titrated with standard acid (0.01 N H_2SO_4) till the green colour disappeared where one drop in excess turned the solution to pink and the blank was run simultaneously.

- b) The phosphorous was estimated using Vanadomolybdate phosphoric acid method in HNO₃ system (Jackson, 1958).

Preparation of standard curve:

0, 1, 2, 3, 4 and 5ml aliquot from 50 ppm standard P solution was transferred in to 50ml volumetric flask and 10 ml of vanadomolybdate reagent (Barton's reagent) was added to it. To obtain 0, 1, 2, 3, 4 and 5 ppm standards respectively the volume was made up to 50 ml with distilled water. The colour intensity was read at 420nm after 30 minute in a spectrometer where the absorbance value was plotted against concentration of P (X ppm) in the coloured solution in the graph paper to obtain the standard curve.

Sample preparation:

5ml of digest (diacid or triacid) was taken in a 50ml of volumetric flask to which 10 ml of Barton's reagent was added and diluted to 50ml with distilled water. The colour development was read at 420nm after 30 minute where the standard curve was prepared by using the standards and the concentration of the P was calculated from the curve.

- c) The potassium was calculated using flame photometry (Jackson, 1958).

Preparation of standard curve:

1.906g of KCl was dissolved in 1 litre of distilled water to get 100 ppm K. 10 ml of solution was pipetted out in to a 100ml volumetric flask and the volume was made up to 100 ml to get 100 ppm K. 2, 4, 6, 8 and 10 ml from the 100 ppm stock solution were pipetted out to different 100ml volumetric flask and volume was made up to 100 ml in order to get 2, 4, 6, 8 and 10 ppm K respectively.

Sample preparation:

5ml of digest (di-acid) was taken in a 50ml of volumetric flask and made the volume. This solution was feed to the flame photometer and read using the filter

for K (548nm). The standard curve was prepared by plotting the readings of known standards against their concentrations from where the concentration of K in the extract was calculated.

d) The carbon/ash content was determined by ash content method.

In a crucible C₄ was taken and kept inside the muffle furnace at 150⁰C for 30 minutes. After that the crucible were taken and kept inside the desiccator or oven for 2 hour and then took out the crucible from where 5g of sample were taken and kept inside the oven for 5 hours

3.3 Statistically analysis

In the first experiment completely randomized design was applied with two factors such as different species of mycorrhiza and different doses of mycorrhiza. In the second experiment randomized block design was applied. The data were analysed using one-way ANOVA in SPSS software version 21.0



RESULTS

RESULTS

4.1 EXPERIMENT 1

4.1.1 Height

The height of the *S. macrophylla* seedlings was not influenced by different species AMF during the course of the nursery experiment (Table 2). The seedling height at 30 days after inoculation showed an average height of 16.74 cm with a maximum height of 17.49 cm (*G. etunicatum*) and minimum height 16.68 cm (*F. mosseae*) with a standard deviation of 1.62 cm. At 60 days after inoculation, the shoot height had an average height of 23.82 cm and ranged from 24.55 cm (*F. mosseae* + *G. etunicatum*) to 22.78 cm (*A. mellea* + *G. etunicatum*) with a standard deviation 1.92 at 90 days cm. The shoot height was observed to have an average height of 31.64 cm which ranged from 32.97 cm (*F. mosseae* + *G. etunicatum*) to 30.38 cm (*A. mellea* + *G. etunicatum*) with a standard deviation of 3.14 cm.

The height of *S. macrophylla* seedlings was influenced by different doses of AMF. The shoot height was found significantly higher when inoculated with 15 g of inoculum throughout the experiment (Table 2). At 30 days after inoculation, the shoot height had a maximum value of 17.65 cm when inoculated with 15 g of inoculum and a minimum value of 16.01 cm when inoculated with 5 g of inoculum. The shoot height at 60 days after inoculation ranged from 24.88 cm (15 g) to 23.15 cm (5 g). The shoot height at 90 days after inoculation found to be range from 33.77 cm (15 g) to 30.16 cm (5 g).

Table 2. Shoot height of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in nursery.

Factors	Treatments	Height (cm)		
		30 days	60 days	90 days
Mycorrhiza	<i>F. mosseae</i>	16.68	23.51	31.72
	<i>A. mellea</i>	16.75	23.96	32.03
	<i>G. etunicatum</i>	17.13	24.31	31.54
	<i>F. mosseae</i> + <i>A. mellea</i>	16.88	23.82	31.64
	<i>F. mosseae</i> + <i>G. etunicatum</i>	17.49	24.55	32.97
	<i>A. mellea</i> + <i>G. etunicatum</i>	15.56	22.78	30.38
	<i>F. mosseae</i> + <i>A. mellea</i> + <i>G.</i> <i>etunicatum</i>	16.70	23.78	31.40
	F value	1.43^{ns}	0.90^{ns}	0.81^{ns}
Doses	5 g	16.01 ^b	23.15 ^b	30.16 ^b
	10 g	16.57 ^b	23.42 ^b	31.00 ^b
	15 g	17.65 ^a	24.88 ^a	33.77 ^a
	F value	6.50*	5.52*	11.24*
Interaction	F value	0.82^{ns}	0.99^{ns}	1.91^{ns}
Control	Mean	14.21	19.97	26.28
	F value	271.56*	365.78*	312.12*
	Mean	16.74	23.82	31.64
	SD	1.62	1.92	3.14

4.1.2 Collar diameter

The collar diameter of *S. macrophylla* seedlings was found to be significantly influenced by different species of AMF at 30 and 60 days of inoculation, but no significant difference was found at 90 days of inoculation (Table 3). The average diameter at 30 days was 1.69 mm ranging from 1.78 (*A. mellea*) to 1.60 (*F. mosseae* + *A. mellea* + *G. etunicatum*) with a standard deviation of 0.12 mm. At 60 days, the collar diameter ranged from 2.67 mm (*A. mellea*) to 2.52 mm (*A. mellea* + *G. etunicatum* and *F. mosseae* + *A. mellea* + *G. etunicatum*) with a mean value of 2.60 mm and standard deviation of 0.13 mm. After 90 days of inoculation, the collar diameter was found to be between 3.74 mm (*F. mosseae*) and 3.57 mm (*F. mosseae* + *A. mellea* and *F. mosseae* + *A. mellea* + *G. etunicatum*) with an average diameter of 3.64 mm and standard deviation of 0.22 mm.

It was found that different doses of AMF significantly influenced the collar diameter of *S. macrophylla* seedlings throughout the experiment (Table 3). Seedlings had significantly higher collar diameter when inoculated with 15 g of inoculum at all stages of the experiment. The collar diameter was found to be ranging from 1.77 mm (15 g) to 1.64 mm (5 g) at 30 days of inoculation. The collar diameter at 60 days varied from 2.69 mm (15 g) to 2.54 (10 g). At 90 days, the collar diameter ranged from of 3.79 mm (15 g) to 3.52 mm (5 g).

Table 3. Collar diameter of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in nursery.

Factors	Treatments	Collar diameter (mm)		
		30 days	60 days	90 days
Mycorrhiza	<i>F. mosseae</i>	1.71 ^{ab}	2.64 ^a	3.74
	<i>A. mellea</i>	1.78 ^a	2.67 ^a	3.70
	<i>G. etunicatum</i>	1.74 ^a	2.61 ^a	3.67
	<i>F. mosseae</i> + <i>A. mellea</i>	1.62 ^c	2.60 ^{ab}	3.57
	<i>F. mosseae</i> + <i>G. etunicatum</i>	1.75 ^a	2.65 ^a	3.68
	<i>A. mellea</i> + <i>G.</i> <i>etunicatum</i>	1.66 ^{bc}	2.52 ^b	3.56
	<i>F. mosseae</i> + <i>A. mellea</i> + <i>G.</i> <i>etunicatum</i>	1.60 ^c	2.52 ^b	3.57
	F value	8.20*	4.936*	1.42^{ns}
Doses	5 g	1.64 ^b	2.58 ^b	3.52 ^b
	10 g	1.67 ^b	2.54 ^b	3.61 ^b
	15 g	1.77 ^a	2.69 ^a	3.79 ^a
	F value	18.10*	19.41*	11.46*
Interaction	F value	5.40*	5.28*	1.76
Control	Mean	1.32	2.07	3.22
	F value	1043.4*	2150.5*	915.32*
	Mean	1.69	2.60	3.64
	SD	0.12	0.13	0.22

4.1.3 Number of leaves

Arbuscular mycorrhizal fungi did not influence number of leaves in *S. macrophylla* seedlings at 30 and 60 days of inoculation, but it had significant effect at 90 days of inoculation (Table 4). The number of leaves was found to range from 6.36 (*A. mellea*) to 5.91 (*A. mellea* + *G. etunicatum*) with an average value of 6.14 and standard deviation of 0.62 at 30 days of inoculation. At 60 days, the number of leaves ranged from 7.18 (*G. etunicatum*) to 7.07 (*A. mellea* + *G. etunicatum*) with an average value of 7.13 and standard deviation of 0.68. The number of leaves varied from 10.28 (*A. mellea*) to 9.01 (*A. mellea* + *G. etunicatum*) at 90 days of inoculation with a mean value of 9.69 and standard deviation of 1.08.

It was observed that different doses of AMF had significant effect on *S. macrophylla* seedlings throughout the experiment, where inoculation with 15 g of inoculum showed higher number of leaves than others (Table 4). The number of leaves at 30 days of inoculation showed a range of 6.63 (15 g) to 5.85 (5 g). At 60 days, the number of leaves ranged 7.88 (15 g) to 6.73 (10 g). The range of number of leaves was found to be 10.60 (15 g) to 9.19 (5 g) at 90 days of inoculation.

Table 4. Number of leaves of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in nursery

Factors	Treatments	Number of leaves		
		30 days	60 days	90 days
Mycorrhiza	<i>F. mosseae</i>	6.13	7.17	9.95 ^{ab}
	<i>A. mellea</i>	6.36	7.14	10.28 ^a
	<i>G. etunicatum</i>	6.19	7.18	9.97 ^{ab}
	<i>F. mosseae</i> + <i>A. mellea</i>	5.92	7.13	9.17 ^c
	<i>F. mosseae</i> + <i>G. etunicatum</i>	6.53	7.15	9.91 ^{ab}
	<i>A. mellea</i> + <i>G.</i> <i>etunicatum</i>	5.91	7.07	9.01 ^c
	<i>F. mosseae</i> + <i>A. mellea</i> + <i>G.</i> <i>etunicatum</i>	5.95	7.09	9.52 ^{bc}
	F value	2.15^{ns}	0.07^{ns}	6.50*
Doses	5 g	5.85 ^b	6.79 ^b	9.19 ^b
	10 g	5.95 ^b	6.73 ^b	9.27 ^b
	15 g	6.63 ^a	7.88 ^a	10.60 ^a
	F value	15.72*	41.07*	43.71*
Interaction	F value	1.42	0.89	6.12*
Control	Mean	6.06	6.90	8.71
	F value	464.48*	662.70*	761.10*
	Mean	6.14	7.13	9.69
	SD	0.62	0.68	1.08

4.1.4 Root colonisation percentage

Results of observation on root colonisation percentage is presented in Table 5. At 30 days of inoculation, the average root colonisation percentage was found to be 26.35 % with a maximum value of 31.1 % (*F. mosseae* + *A. mellea*, 5g) and minimum value of 21.6% (*G. etunicatum*, 5 g). The root colonisation percentage at 60 days ranged from 32.5 % (*G. etunicatum*, 15g) to 20.5 (*G. etunicatum*, 5 g) with an average of 26.79 %. The average root colonisation percentage at 90 days of inoculation was found to be 32.33 % with a range of 39.15 % (*G. etunicatum*, 10g) to 21 % (*A. mellea* + *G. etunicatum*, 5 g).

Table 5. Root colonisation percentage of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in nursery

Treatments	Root colonisation percentage (%)		
	30 days	60 days	90 days
<i>F. mosseae</i> 5g	25.6	22.1	35.2
<i>F. mosseae</i> 10g	27.2	23.2	35.68
<i>F. mosseae</i> 15g	28.3	27	36.49
<i>A. mellea</i> 5g	26.3	28.5	30.4
<i>A. mellea</i> 10g	23.5	29.6	35.5
<i>A. mellea</i> 15g	25.5	28.5	36.4
<i>G. etunicatum</i> 5g	21.6	20.5	30.23
<i>G. etunicatum</i> 10g	28.7	23.6	39.15
<i>G. etunicatum</i> 15g	30.3	32.5	38.56
(<i>F. mosseae</i> + <i>A. mellea</i>) 5g	31.1	30	30.2
(<i>F. mosseae</i> + <i>A. mellea</i>) 10 g	29.1	24.6	31.5
(<i>F. mosseae</i> + <i>A. mellea</i>) 15 g	23.5	24	29.2
(<i>F. mosseae</i> + <i>G. etunicatum</i>) 5 g	29.3	27	27.5
(<i>F. mosseae</i> + <i>G. etunicatum</i>) 10 g	25.5	28	28.5

(<i>F. mosseae</i> + <i>G. etunicatum</i>) 15 g	29.5	32	22
(<i>A. mellea</i> + <i>G. etunicatum</i>) 5 g	26	31	21
(<i>A. mellea</i> + <i>G. etunicatum</i>) 10 g	27	32	29.5
(<i>A. mellea</i> + <i>G. etunicatum</i>) 15 g	23.3	23	35
(<i>F. mosseae</i> + <i>G. etunicatum</i> + <i>A. mellea</i>) 5 g	26	22	39
(<i>F. mosseae</i> + <i>G. etunicatum</i> + <i>A. mellea</i>) 10 g	24.1	25	36
(<i>F. mosseae</i> + <i>G. etunicatum</i> + <i>A. mellea</i>) 15 g	22.1	28.5	32
Control	0	0	0
Mean	26.35	26.79	32.33

4.1.5 Total spore count

The total spore count is given in Table 6. The total spore count ranged from 70/10 g (*F. mosseae* + *G. etunicatum*, 15 g) to 44/10 g (*F. mosseae* + *G. etunicatum*, 10 g) with an average value of 58.57/10 g at 30 days of inoculation. At 60 days the total spore count was found to be range from 87 (*F. mosseae* + *G. etunicatum* + *A. mellea*, 5 g) to 59 (*G. etunicatum*, 10g) with an average value of 72.61. The average value of total spore count was found to be 103.95 at 90 days of inoculation with a range from 120 (*F. mosseae*, 15g) to 91 (*A. mellea* + *G. etunicatum*, 5 g).

Table 6. Total spore count of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in nursery.

Treatments	Total spore count (per 10 g)		
	30 days	60 days	90 days
<i>F. mosseae</i> 5g	56	73	97
<i>F. mosseae</i> 10g	59	71	101
<i>F. mosseae</i> 15g	63	79	120

<i>A. mellea</i> 5g	69	82	102
<i>A. mellea</i> 10g	54	71	96
<i>A. mellea</i> 15g	63	86	114
<i>G. etunicatum</i> 5g	60	70	93
<i>G. etunicatum</i> 10g	51	59	95
<i>G. etunicatum</i> 15g	69	83	115
(<i>F. mosseae</i> + <i>A. mellea</i>) 5g	68	75	97
(<i>F. mosseae</i> + <i>A. mellea</i>) 10 g	58	61	103
(<i>F. mosseae</i> + <i>A. mellea</i>) 15 g	52	68	101
(<i>F. mosseae</i> + <i>G. etunicatum</i>) 5 g	54	65	117
(<i>F. mosseae</i> + <i>G. etunicatum</i>) 10 g	44	63	97
(<i>F. mosseae</i> + <i>G. etunicatum</i>) 15 g	70	79	116
(<i>A. mellea</i> + <i>G. etunicatum</i>) 5 g	61	60	91
(<i>A. mellea</i> + <i>G. etunicatum</i>) 10 g	52	79	118
(<i>A. mellea</i> + <i>G. etunicatum</i>) 15 g	63	74	98
(<i>F. mosseae</i> + <i>G. etunicatum</i> + <i>A. mellea</i>) 5 g	51	87	99
(<i>F. mosseae</i> + <i>G. etunicatum</i> + <i>A. mellea</i>) 10 g	54	77	102
(<i>F. mosseae</i> + <i>G. etunicatum</i> + <i>A. mellea</i>) 15 g	57	63	111
Control	0	0	0
Mean	58.57	72.61	103.95

4.2 EXPERIMENT 2

4.2.1 Height

In the field experiment, it was observed that inoculation with AMF did not influence the height of the *S. macrophylla* seedlings significantly throughout the experiment (Table 7). The seedling height increased from 32.47 cm at 30 days to 92.22 cm at 180 days. With an average height of 32.47 cm, the seedlings at 30 days of planting showed a range of maximum shoot height 33.46 cm (*A. mellea*) to minimum 31.55 cm (*F. mosseae*) with a standard deviation of 1.89. At 60 days of planting, the seedlings height ranged from 39.15 cm (*A. mellea*) to 37.36 cm (*F. mosseae*) with an average of 38.18 cm and a standard deviation of 2.22. The seedling height at 90 days of planting showed an average height of 45.79 cm with a range of 46.76 cm (*A. mellea*) to 45.21 cm (non-inoculated) and had a standard deviation of 3.13. At 120 days of the planting the seedlings had an average height of 55.81 cm and range of 56.78 cm (*A. mellea*) to 54.38 cm (non-inoculated) with a standard deviation of 4.85. The seedlings showed an average height of 70.82 cm at 150 days of planting which was varied from 72.97 cm (*A. mellea*) to 68.85 cm (non-inoculated) with a standard deviation of 8.44. After 180 days of planting the height of the seedlings were found to be range from 95.35 cm (*G. etunicatum*) to 88.56 (non-inoculated) with an average height of 92.22 cm and a standard deviation of 14.01.

Table 7. Shoot height of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Height (cm)					
	30 days	60 days	90 days	120 days	150 days	180 days
<i>F. mosseae</i>	31.55	37.36	45.51	56.21	71.45	92.02
<i>A. mellea</i>	33.46	39.15	46.76	56.78	72.97	94.19
<i>G. etunicatum</i>	32.92	38.47	45.99	55.99	71.25	95.35
<i>F. mosseae</i> + <i>G. etunicatum</i>	32.54	38.19	45.49	55.25	69.61	90.99
Non-inoculated	31.90	37.75	45.21	54.83	68.85	88.56
F value	0.55^{ns}	0.45^{ns}	0.22^{ns}	0.18^{ns}	0.30^{ns}	0.32^{ns}
Mean	32.47	38.18	45.79	55.81	70.82	92.22
SD	1.89	2.22	3.13	4.85	8.44	14.01

4.2.2 Collar diameter

It was observed that inoculation with AMF did not influence the collar diameter of the *S. macrophylla* seedlings throughout the experiment (Table 8). The collar diameter increased from 4.23 mm at 30 days to 14.87 mm at 180 days. At 30 days of planting, the collar diameter of seedlings had an average value of 4.23 mm with a range of 4.59 mm (*F. mosseae*) to 4.07 mm (non-inoculated) with a standard deviation of 0.49. The collar diameter at 60 days of planting ranged from 5.95 mm (*F. mosseae*) to 5.32 mm (non-inoculated) with a standard deviation of 0.68 and an average value of 5.56 mm. The collar diameter varied from 7.54 mm (*F. mosseae*) to 6.91 mm (non-inoculated) at 90 days after planting with a mean of 7.21 mm and standard deviation of 1.00. At 120 days of planting the collar diameter of seedlings ranged from 9.43 mm (*G. etunicatum*) to 8.69 mm (non-inoculated) with an average

value of 9.14 mm and standard deviation of 1.44. The collar diameter at 150 days after planting had an average value of 11.65 mm and a range of 12.04 mm (*G. etunicatum*) to 11.04 mm (non-inoculated) with a standard deviation of 2.01. After 180 days, the seedlings showed an average collar diameter of 14.87 mm which ranged from 15.45 mm (*G. etunicatum*) to 14.04 mm (non-inoculated) with a standard deviation of 2.79.

Table 8. Collar diameter of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Collar diameter (mm)					
	30 days	60 days	90 days	120 days	150 days	180 days
<i>F. mosseae</i>	4.59	5.95	7.54	9.42	11.92	15.28
<i>A. mellea</i>	4.08	5.36	7.03	8.97	11.57	14.92
<i>G. etunicatum</i>	4.17	5.60	7.34	9.43	12.04	15.45
<i>F. mosseae</i> + <i>G. etunicatum</i>	4.24	5.58	7.26	9.21	11.70	14.68
Non-inoculated	4.07	5.32	6.91	8.69	11.04	14.04
F value	1.08^{ns}	1.37^{ns}	0.94^{ns}	0.87^{ns}	0.69^{ns}	0.83^{ns}
Mean	4.23	5.56	7.21	9.14	11.65	14.87
SD	0.49	0.68	1.00	1.44	2.01	2.79

4.2.3 Number of leaves

It was observed that the number of leaves of the *S. macrophylla* seedlings had not been influenced significantly when inoculated with AMF in the field condition (Table 9). The average number of leaves increased from 6.37 at 30 days to 16.07 at 180 days. At 30 days of planting the number of leaves of seedlings ranged from maximum 6.68 (*G. etunicatum*) to minimum 5.92 (*F. mosseae* + *G.*

etunicatum) with an average value of 6.37 and a standard deviation of 0.48. The number of leaves at 60 days of planting ranged from 7.71 (non-inoculated) to 6.61 (*F. mosseae* + *G. etunicatum*) with an average value of 7.30 and standard deviation of 0.74. The number of leaves at 90 days ranged from 8.86 (non-inoculated) to 7.80 (*F. mosseae* + *G. etunicatum*) with mean of 8.52 and standard deviation 0.82. The number of leaves at 120 days of planting showed an average value of 10.13 and ranged from 10.43 (*A. mellea*) to 9.86 (non-inoculated) with a standard deviation of 1.06. At 150 days of planting the number of leaves ranged from 13.34 (*A. mellea*) to 12.06 (*G. etunicatum*) with an average of 12.69 and a standard deviation of 1.39. With an average value of 16.07 the number of leaves observed at 180 days after planting ranged of 16.6 (*A. mellea*) to 15.71 (*G. etunicatum*) with a standard deviation of 1.30.

Table 9. Number of leaves of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Number of leaves					
	30 days	60 days	90 days	120 days	150 days	180 days
<i>F. mosseae</i>	6.50	7.14	8.71	10.32	12.80	15.92
<i>A. mellea</i>	6.29	7.45	8.51	10.43	13.34	16.76
<i>G. etunicatum</i>	6.68	7.59	8.73	10.11	12.16	15.71
<i>F. mosseae</i> + <i>G. etunicatum</i>	5.92	6.61	7.80	9.91	12.85	16.01
Non-inoculated	6.48	7.71	8.86	9.86	12.29	15.98
F value	1.69^{ns}	1.07^{ns}	1.07^{ns}	0.27^{ns}	0.48^{ns}	0.48^{ns}
Mean	6.37	7.30	8.52	10.13	12.69	16.07
SD	0.48	0.74	0.82	1.06	1.39	1.30

4.2.4 Photosynthetic rate

The *S. macrophylla* seedlings had shown significant effect in photosynthesis rate when inoculated with AMF at 90, 150 and 180 days of planting in the field (Table 10). Photosynthesis rate at 30 days of planting the seedlings showed an average of $4.06 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ and values ranged from $5.27 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*G. etunicatum*) to $3.26 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (non-inoculated) with a standard deviation of 1.15. The rate of photosynthesis at 60 days of planting ranged from $5.87 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*F. mosseae*) to $5.38 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (non-inoculated) with an average value of $5.21 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ and a standard deviation of 0.96. At 90 days of planting the photosynthesis rate was varied from $5.66 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*F. mosseae* + *G. etunicatum*) to $4.73 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*F. mosseae*) with an average of $5.30 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ and standard deviation of 0.77. The seedlings showed an average photosynthesis rate of $6.46 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ at 120 days of planting with a range of $8.32 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*F. mosseae* + *G. etunicatum*) to $5.77 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (non-inoculated) with a standard deviation of 1.45. At 150 days the photosynthesis rate ranged from $9.60 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*F. mosseae*) to $5.97 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*F. mosseae* + *G. etunicatum*) with an average of $8.07 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ and standard deviation of 2.06. The photosynthesis rate found to have an average value of $4.45 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ at 180 days of planting with a range of $6.57 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*G. etunicatum*) to $3.11 \mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ (*A. mellea*) with a standard deviation of 1.87.

Table 10. Photosynthetic rate of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Photosynthesis rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)					
	30 days	60 days	90 days	120 days	150 days	180 days
<i>F. mosseae</i>	4.59	5.87	4.73 ^b	6.23	9.60 ^a	5.93 ^a
<i>A. mellea</i>	3.57	4.04	5.60 ^a	6.19	6.31 ^{bc}	3.11 ^b
<i>G. etunicatum</i>	5.27	5.28	5.02 ^{ab}	5.79	9.41 ^a	6.57 ^a
<i>F. mosseae</i> + <i>G. etunicatum</i>	3.63	5.47	5.66 ^a	8.32	5.97 ^c	3.38 ^b
Non-inoculated	3.26	5.38	5.51 ^a	5.77	9.08 ^{ab}	3.25 ^b
F value	2.77 ^{ns}	2.02 ^{ns}	4.77*	3.27 ^{ns}	4.25*	5.25*
Mean	4.06	5.21	5.30	6.46	8.07	4.45
SD	1.15	0.96	0.77	1.45	2.06	1.87

4.2.5 Stomatal conductance

The stomatal conductance of *S. macrophylla* seedlings was not influenced by AMF inoculation throughout the field experiment except at 120 days (Table 11). The stomatal conductance of the seedlings at 30 days of planting had an average value of $0.09 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and it ranged from $0.11 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (*F. mosseae*) to $0.07 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (*A. mellea*) with a standard deviation of 0.01. At 60 days of planting, the seedlings showed an average stomatal conductance of $0.03 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ with a range of $0.08 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (*F. mosseae* + *G. etunicatum*) to $0.01 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (*A. mellea*) and a standard deviation of 0.04. At 90 days, the stomatal conductance ranged from $0.14 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (*F. mosseae*, *G. etunicatum* and *F. mosseae* + *G. etunicatum*) to $0.12 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ (non-inoculated) with a mean value of $0.13 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ and a standard deviation of 0.01. The stomatal

conductance at 120 days of planting showed a range of 0.19 mol H₂O m⁻² s⁻¹ (*F. mosseae* + *G. etunicatum*) to 0.12 mol H₂O m⁻² s⁻¹ (non-inoculated) with an average of 0.16 mol H₂O m⁻² s⁻¹ and a standard deviation of 0.03. At 150 days of planting, the stomatal conductance of the seedlings found to have an average value of 0.04 mol H₂O m⁻² s⁻¹ which varied from 0.07 mol H₂O m⁻² s⁻¹ (*F. mosseae*) to 0.03 mol H₂O m⁻² s⁻¹ (*A. mellea*, *G. etunicatum* and non-inoculated) with a standard deviation of 0.03. After 180 days of planting, stomatal conductance of the seedlings showed a range of 0.15 mol H₂O m⁻² s⁻¹ (*F. mosseae*) to 0.01 mol H₂O m⁻² s⁻¹ (*G. etunicatum*) with a standard deviation of 0.07 and an average of 0.07 mol H₂O m⁻² s⁻¹.

Table 11. Stomatal conductance of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Stomatal conductance (mol H ₂ O m ⁻² s ⁻¹)					
	30 days	60 days	90 days	120 days	150 days	180 days
<i>F. mosseae</i>	0.11	0.02	0.14	0.15 ^{bc}	0.07	0.15
<i>A. mellea</i>	0.07	0.01	0.13	0.17 ^{ab}	0.03	0.05
<i>G. etunicatum</i>	0.08	0.02	0.14	0.18 ^a	0.03	0.01
<i>F. mosseae</i> + <i>G. etunicatum</i>	0.09	0.08	0.14	0.19 ^a	0.04	0.08
Non-inoculated	0.09	0.02	0.12	0.12 ^c	0.03	0.05
F value	3.81^{ns}	1.26^{ns}	0.87^{ns}	9.26*	1.27^{ns}	3.15^{ns}
Mean	0.09	0.03	0.13	0.16	0.04	0.07
SD	0.01	0.04	0.01	0.03	0.03	0.07

4.2.6 Transpiration rate

It was found that inoculation with AMF did not influence the transpiration rate of *S. macrophylla* seedlings till 150 days of planting (Table 12). The transpiration rate at 30 days of plantation ranged from 1.77 mmol H₂O m⁻² s⁻¹ (*F. mosseae* + *G. etunicatum*) to 1.35 mmol H₂O m⁻² s⁻¹ (*A. mellea*) with an average of 1.60 and a standard deviation of 0.29. At 60 days the transpiration rate found to be have an average value of 0.75 mmol H₂O m⁻² s⁻¹ with a range of 0.89 mmol H₂O m⁻² s⁻¹ (*F. mosseae*) to 0.56 mmol H₂O m⁻² s⁻¹ (non-inoculated) and a standard deviation of 0.32. The transpiration rate had a range of 2.23 mmol H₂O m⁻² s⁻¹ (*F. mosseae* + *G. etunicatum*) to 1.65 mmol H₂O m⁻² s⁻¹ (*G. etunicatum*) with a standard deviation of 0.31 and an average of 1.84 mmol H₂O m⁻² s⁻¹ at 90 days of planting. At 120 days, the transpiration ranged from 3.39 mmol H₂O m⁻² s⁻¹ (*F. mosseae*) to 2.89 mmol H₂O m⁻² s⁻¹ (*A. mellea*) with a mean value of 3.08 mmol H₂O m⁻² s⁻¹ and a standard deviation of 0.32. The average transpiration rate found to be 2.91 mmol H₂O m⁻² s⁻¹ at 150 days of planting with a range of 3.24 mmol H₂O m⁻² s⁻¹ (*G. etunicatum*) to 2.60 mmol H₂O m⁻² s⁻¹ (*F. mosseae* + *G. etunicatum*) and a standard deviation of 0.37. At 180 days, the transpiration rate was found to be range from 6.41 mmol H₂O m⁻² s⁻¹ (*G. etunicatum*) to 4.57 mmol H₂O m⁻² s⁻¹ (non-inoculated) with an average value of 5.18 mmol H₂O m⁻² s⁻¹ and a standard deviation of 0.98.

Table 12. Transpiration rate of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Transpiration rate (mmol H ₂ O m ⁻² s ⁻¹)					
	30 days	60 days	90 days	120 days	150 days	180 days
<i>F. mosseae</i>	1.52	0.89	1.73	3.39	2.96	4.90 ^b
<i>A. mellea</i>	1.35	0.87	1.67	2.89	2.89	4.60 ^b
<i>G. etunicatum</i>	1.65	0.70	1.65	3.07	3.24	6.41 ^a

<i>F. mosseae</i> + <i>G.</i> <i>etunicatum</i>	1.77	0.76	2.23	3.02	2.60	5.42 ^{ab}
Non- inoculated	1.72	0.56	1.93	3.04	2.87	4.57 ^b
F value	1.68^{ns}	0.57^{ns}	2.92^{ns}	1.29^{ns}	1.23^{ns}	4.35*
Mean	1.60	0.75	1.84	3.08	2.91	5.18
SD	0.29	0.32	0.31	0.32	0.37	0.98

4.2.7 Leaf temperature

The leaf temperature in *S. macrophylla* seedlings was not significantly influenced by AMF inoculation (Table 13). At 30 days of planting, the leaf temperature was found to have an average value of 32.3 °C. The value ranged from 32.5 °C (non-inoculated) to 32.1 °C (*A. mellea*). The leaf temperature at 60 days of planting ranged from 36.1 °C (*F. mosseae*) to 35.6 °C (*A. mellea*) with an average value of 35.9 °C. At 90 days of planting, the leaf temperature was found to have an average value of 32.2 °C which varied from 32.4 °C (non-inoculated) to 32.0 °C (*A. mellea*). The seedlings showed an average leaf temperature of 34.9 °C at 120 days of planting, the leaf temperature at this stage ranged from 35.4 °C (*F. mosseae* + *G. etunicatum*) to 34.4 °C (*F. mosseae*). The average leaf temperature at 150 days of planting was found to be 34.9 °C which had a range of 35.4 °C (non-inoculated) to 34.5 °C (*F. mosseae*). The leaf temperature at 180 days of planting had an average value of 32.2 °C and a range of 32.4 °C (non-inoculated) to 32.1 °C (*F. mosseae*).

Table 13. Leaf temperature of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Leaf temperature (°C)					
	30 days	60 days	90 days	120 days	150 days	180 days
<i>F. mosseae</i>	32.2	36.1	32.0	34.4	34.5	32.1
<i>A. mellea</i>	32.1	35.6	32.0	35.3	35.1	32.1
<i>G. etunicatum</i>	32.4	35.9	32.3	34.7	34.5	32.3
<i>F. mosseae</i> + <i>G. etunicatum</i>	32.3	35.8	32.2	35.4	35.1	32.2
Non-inoculated	32.5	36.0	32.4	34.5	35.3	32.4
F value	0.355^{ns}	0.52^{ns}	0.62^{ns}	1.98^{ns}	1.50^{ns}	0.55^{ns}
Mean	32.3	35.9	32.2	34.9	34.9	32.2
SD	0.80	1.07	0.63	0.58	0.53	0.82

4.2.8 Absolute Growth Rate

There was no significant difference in the absolute growth rate of *S. macrophylla* seedlings when inoculated with AMF throughout the experiment (Table 14). The absolute growth rate increased from 0.18 cm/day at 60 days to 0.71 cm/day at 180 days. The absolute growth rate at 60 days of planting ranged from 0.19 cm/day (*F. mosseae*, *A. mellea* and non-inoculated) to 0.18 cm/day (*G. etunicatum* and *F. mosseae* + *G. etunicatum*) with an average of 0.18cm/day and a standard deviation of 0.02. At 90 days the average AGR 0.25 cm/day the absolute growth rate varied from 0.27 cm/day (*F. mosseae*) to 0.24 cm/day (*F. mosseae* + *G. etunicatum*) with standard deviation 0.33 at 90 days of planting. At 120 days, the absolute growth rate had an average of 0.33 cm/day with range from 0.35 cm/day (*F. mosseae*) to 0.32 cm/day (*F. mosseae* + *G. etunicatum* and non-inoculated). The seedlings showed an average absolute growth rate of 0.50 cm/day

ranging from 0.54 cm/day (*A. mellea*) to 0.46 cm/day (non-inoculated) with a standard deviation of 0.12 at 150 days after planting. The absolute growth rate observed at 180 days showed an average value of 0.71 cm/day and had a range of 0.80 cm/day (*G. etunicatum*) to 0.68 cm/day (non-inoculated) with a standard deviation of 0.19.

Table 14. Absolute growth rate of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Absolute Growth Rate (cm day ⁻¹)					
	30 days	60 days	90 days	120 days	150 days	180 days
<i>F. mosseae</i>	0	0.19	0.27	0.35	0.51	0.68
<i>A. mellea</i>	0	0.19	0.25	0.33	0.54	0.70
<i>G. etunicatum</i>	0	0.18	0.25	0.33	0.50	0.80
<i>F. mosseae</i> + <i>G. etunicatum</i>	0	0.18	0.24	0.32	0.48	0.71
Non-inoculated	0	0.19	0.25	0.32	0.46	0.65
F value	0	0.25^{ns}	0.27^{ns}	0.31^{ns}	0.33^{ns}	0.68^{ns}
Mean	0	0.18	0.25	0.33	0.50	0.71
SD	0	0.02	0.04	0.06	0.12	0.19

4.2.9 Root colonisation percentage

At 120 and 180 days, there were significant difference in root colonisation percentage between the treatments (Table 15). The root colonisation percentage increased from 3.5 % at 120 days to 6.69 % at 180 days. At 120 days of planting seedlings inoculated with *F. mosseae* + *G. etunicatum* (4.45 %) showed highest root colonisation percentage followed by *A. mellea* (3.64 %), *G. etunicatum* (3.56 %), *F. mosseae* (3.47 %) and non-inoculated seedlings (2.36 %) with an average of

3.5 % and a standard deviation of 0.84 %. After 180 days of planting, seedlings inoculated with *F. mosseae* + *G. etunicatum* (8.76 %) had shown highest root colonisation percentage followed by *A. mellea* (7.84 %), *G. etunicatum* (6.73 %), *F. mosseae* (6.12 %), and non-inoculated seedlings (3.98 %) with a standard deviation of 1.78 %.

4.2.10 Number of spores

The number of spores/10 g of soil differed significantly between different treatments (Table 16). At 60 days of planting the number of spores found to be highest in seedlings inoculated with *F. mosseae* (26.79/ 10 g) and lowest in the non-inoculated (7.02/ 10 g) seedlings with an average count of 20.44 and standard deviation of 7.47/ 10 g. The number of spores found at 120 days of planting was highest in *F. mosseae* (33.12/ 10 g) and lowest in the non-inoculated seedlings (14.39/ 10 g) with an average count of 27.94/ 10 g and standard deviation of 7.22/ 10 g. At 180 of days of planting the seedlings inoculated with *G. etunicatum* had shown the highest spore count (40.33/ 10 g) and the non-inoculated seedlings showed the lowest value (19.93/ 10 g) with an average of 34.40/ 10 g and standard deviation of 8.64/ 10 g.

Table 15. Root colonisation percentage of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Root colonisation percentage (%)		
	60 days	120 days	180 days
<i>F. mosseae</i>	0	3.47 ^a	6.12 ^c
<i>A. mellea</i>	0	3.64 ^a	7.84 ^{ab}
<i>G. etunicatum</i>	0	3.56 ^a	6.73 ^{bc}
<i>F. mosseae</i> + <i>G. etunicatum</i>	0	4.45 ^a	8.75 ^a
Non-inoculated	0	2.36 ^b	3.98 ^d
F value	0	5.57*	25.86*
Mean	0	3.5	6.69

SD	0	0.84	1.78
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Table 16. Number of spores per 10g of soil of *S. macrophylla* seedlings as influenced by arbuscular mycorrhizal fungi in field.

Treatments	Number of spores		
	60 days	120 days	180 days
<i>F. mosseae</i>	26.79 ^a	33.12 ^a	37.67 ^a
<i>A. mellea</i>	22.79 ^{bc}	30.19 ^a	36.63 ^a
<i>G. etunicatum</i>	25.07 ^{ac}	30.40 ^a	40.33 ^a
<i>F. mosseae</i> + <i>G. etunicatum</i>	20.54 ^c	31.59 ^a	37.45 ^a
Non-inoculated	7.02 ^d	14.39 ^b	19.93 ^b
F value	45.72*	55.08*	7.26*
Mean	20.44	27.94	34.40
SD	7.47	7.22	8.64

4.2.11 Soil nutrient status

Soil nutrient status analysed before and after the experiment, is presented in Table 17. The soil pH, electrical conductivity, total nitrogen, available phosphorous, available potassium, and organic carbon found to be higher when soil samples were collected after the experiment than the soil sample collected before the experiment.

Table 17. Chemical properties of soil before and after the experiment.

Chemical properties	Soil collected before the experiment				Soil collected after the experiment			
	Plot 1	Plot 2	Plot 3	Mean	Plot 1	Plot 2	Plot 3	Mean
Soil pH	5.58	5.40	5.40	5.46	5.67	5.94	6.21	5.94
Electrical conductivity (mS/cm)	11.85	7.55	8.99	9.46	11.87	9.50	16.40	12.59
Total nitrogen (%)	0.084	0.140	0.056	0.093	0.084	0.168	0.056	0.102
Available phosphorous (Kg/ha)	9.74	10.86	4.48	8.36	24.75	20.60	56.22	33.85
Available potassium (Kg/ha)	157.92	115.36	117.60	130.29	198.24	293.44	290.08	260.58
Organic carbon (%)	0.93	1.13	0.75	0.93	1.45	1.08	1.18	1.23

4.2.12 Plant nutrient status

It was found that there were a significant difference in plant phosphorous contents at 180 days of the experiment (Table 18). The average plant phosphorous content was 0.35 %, which ranged from 0.50 % (*F. mosseae* + *G. etunicatum*) to 0.21 % (non- inoculated) with a standard deviation of 8.64.

There were no significant difference found in plant potassium contents at 180 days of plantation (Table 18). The potassium contents ranged 1.86 % (*F. mosseae* and *F. mosseae* + *G. etunicatum*) to 1.68 % (*A. mellea*) with an average value of 1.76 % and a standard deviation of 0.11.

S. macrophylla seedlings showed no significant difference in plant nitrogen contents when inoculated with AMF at 180 days of plantation (Table 18). The nitrogen contents ranged from 2.76 % (*G. etunicatum*) to 2.36 % (non-inoculated) with a standard deviation of 0.10.

The seedlings showed a significant difference in plant carbon content when inoculated with AMF at 180 days plantation (Table 18). With an average of 52.33 % the plant carbon content ranged from 53.28 % (*G. etunicatum*) to 51.20 % (non-inoculated) with a standard deviation of 0.82.

Table 18. Plant chemical analysis of *S. macrophylla* influenced by arbuscular mycorrhizal fungi at 180 days of planting.

Treatments	Plant phosphorous (%)	Plant K (%)	Plant N (%)	Plant C (%)
<i>F. mosseae</i>	0.41 ^{ab}	1.86	2.61	52.93 ^a
<i>A. mellea</i>	0.28 ^{bc}	1.68	2.44	52.19 ^b
<i>G. etunicatum</i>	0.33 ^{abc}	1.70	2.76	53.28 ^a
<i>F. mosseae</i> + <i>G. etunicatum</i>	0.50 ^a	1.86	2.53	52.05 ^b
Non-inoculated	0.21 ^c	1.73	2.36	51.20 ^c
F value	4.13*	0.46^{ns}	1.33^{ns}	21.79*
Mean	0.35	1.76	2.54	52.33
SD	0.13	0.19	0.18	0.82



DISCUSSION

DISCUSSION

The present study entitled ‘Field performance of arbuscular mycorrhizal fungi on vegetative growth of mahogany (*Swietenia macrophylla* King.) seedlings’ was aimed at evaluation three species of arbuscular mycorrhizal fungi (*Funneliformis mosseae*, *Acaulospora mellea* and *Glomus etunicatum*) in mahogany.

5.1 EXPERIMENT 1

Inoculation with AMF helps in seedlings to get the access of sparingly available nutrients in soil (Tarafdar and Kumar, 1996). Arbuscular mycorrhizal fungi (AMF) inoculation also improves the seedling establishment in nursery as well as in the field (Giri *et al.*, 2005). Various studies have been conducted in different tree species using AMF as a bio-inoculant showing higher plant growth in inoculated seedlings than the non-inoculated seedlings e.g. *Acacia mangium* (Jeyanny *et al.*, 2011; Ghosh and Verma, 2006), *Casuarina equisetifolia* (Zhang *et al.*, 2010), *Hevea brasiliensis* (Moreas *et al.*, 2010), Citrus (Ortas and Usttuner, 2014), *Azadirchta indica* (Muthukumar *et al.*, 2001), *Prunus persica* (Wu *et al.*, 2011), *Tectona grandis* (Rajan *et al.*, 2000; Ajeesh *et al.*, 2017), *Citrus tangerine* (Wu *et al.*, 2011), *Casia siamea* (Giri *et al.*, 2005) and *Swietenia macrophylla* (Ajeesh *et al.*, 2017; Rajan, 2016). Some studies also showed that the combination of two or more AMF species are capable of giving better results than single species (Shukla *et al.*, 2014; Wehner *et al.*, 2010; Neetu *et al.*, 2011 and Jansa *et al.*, 2008). Again it was observed that the native AMF species can help the seedlings to grow vigorously than the non-native AMF (Caravaca *et al.*, 2003). In the above light, the present study included three native AMF species namely *Funneliformis mosseae*, *Glomus etunicatum* and *Acaulospora mellea*.

The present study was conducted to determine the growth performance of *S. macrophylla* seedlings in the nursery as well as in the field using the three species of AMF either individually or combined. The results of nursery experiment showed that the inoculation with three different species of AMF in *Swietenia macrophylla*

seedlings had no effect on height, collar diameter and number of leaves of the seedlings (Table 2). However, the present study indicated that all AMF inoculated seedlings significantly differed in height, collar diameter and number of leaves respect to control. This is similar with the findings of Binu (2002) where sandalwood (*Santalum album*) inoculated with different species of AMF (*Glomus fasciculatum*, *G. intraradices* and *G. mosseae*) showed no effect on the seedling height, collar diameter and number of leaves in nursery for the early 90 days of the experiment. This indicates that while AMF had a positive effect on height, collar diameter and number of leaves of seedlings, they did not differ among themselves. The result was supported by the work of Querejeta *et al.* (1998) where *Pinus halepensis* seedlings were inoculated with *Pisolithus arhizus*. Since height, collar diameter and number of leaves increased due to AMF, it clearly indicates its use in producing seedlings with higher quality as seedlings quality is normally measured as a function of height, collar diameter and number of leaves of seedlings.

The difference in AMF dosage application in the soil in nursery had different effects on the growth of the seedlings. The requirement of AMF (*Glomus mosseae*) inoculum for optimal growth of three tree species (*Acacia auriculiformis*, *Leucena leucocephala*, and *Delonix regia*) was studied by Ghosh and Verma (2011). In that study, they used 5 different dosages of inoculum (5 g, 10 g, 15 g, 20 g and 25 g). They found that the growth of the seedlings increased when the amount of inoculum increased up to 15 g of inoculum, after that, higher doses inoculum (20 g and 25 g) did not affect the performance of the seedlings. In the present study, 3 doses (5 g, 10 g, and 15 g) of inoculum was used in the nursery. The study showed that the height, collar diameter and number of leaves of the nursery grown seedlings were significantly influenced by AMF when inoculated under different dosages (Table 2, 3 and 4). A study was conducted by Ajeesh *et al.* (2017) on teak (*Tectona grandis*) inoculated with different doses (10 g, 25 g, and 50 g) of AMF (*F. mosseae*, *G. intraradices*, and *G. proliferum*) also resulted in higher performance of the seedlings inoculated with 50 g of *G. proliferum*. A direct relation between the amount of dose and growth of seedlings was observed.

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In the present study, it was found *S. macrophylla* seedlings showed mean root colonization percentage as 26.35 %, 26.79 % and 32.33 % of root colonization at 30 days, 60 days and 90 days respectively (Table 5). The highest root colonization percentages obtained were 29.5% (*F. mosseae* + *G. etunicatum* at 15 g), 32.5% (*G. etunicatum* at 15 g) and 36.49 % (*F. mosseae* at 15 g) at 30, 60 and 90 days respectively. The mean spores counted for 10 g of soil were 58.57, 72.61, and 103.95 at 30, 60, and 90 days of inoculation (Table 6) respectively. Root colonisation percentage of AMF in tree species varied between 4 to 95 % (Birhane *et al.* 2018), 0 to 95% (Birhane *et al.*, 2010), and 3.5 to 96.3% (Carrenho *et al.*, 2007) depending on the soil physicochemical properties of the surrounding host plants (Wu *et al.*, 2009).

Considering the result obtained, four best-performing treatments (*F. mosseae* 15 g, *A. mellea* 15 g, *G. etunicatum* 15 g and *F. mosseae* + *G. etunicatum* 15 g) were chosen for field planting.

5.2 EXPERIMENT 2

Healthy seedlings production in the nursery is pre-requisite to establish a good plantation (Jha *et al.*, 2017). During out-planting, the seedlings may face transplantation shocks in the field which may cause the seedlings to be weak and ultimately die (Hartmann and Kester, 1986, Ghosh and Verma, 2011). So pre-conditioning of young seedlings in the nursery is of utmost requirement before any plantation especially using suitable AMF (Jha *et al.*, 2014), which not only make the seedlings stronger but also helps their establishment in the field (Navarro-Garcia *et al.*, 2011). Different studies showed that inoculation with AMF resulted in increased height, collar diameter and number of leaves in *Citrus* spp. (Ortas and Ustuner, 2014), *Argania spinosa* (El Mrabet *et al.*, 2014), *Manihot esculenta* (Sridevi and Ramakrishnan, 2013), *Pinus halepensis* (Querejeta *et al.*, 1998) and *Bauhinia faberi* (Yamin *et al.*, 2016).

In the present study, the height, collar diameter and the number of leaves in *S. macrophylla* were not significantly influenced by AMF (Table 7, 8 and 9). Jha *et*

al. (2017) used 11 AMF species combinations with four tree species (*Azadirachta indica*, *Jatropha curcas*, *Madhuca indica* and *Pongamia pinnata*). Two (*Azadirachta indica* and *Madhuca indica*) of the tree species showed positive response towards the AMF, where as other two did not show any significant differences. The result obtained from this study showed that the AMF had no significant influence on the growth of the seedlings. The result obtained may be because of poor establishment of AMF under the field conditions during the study period due to acidic pH and low moisture availability (Ouzounidou *et al.*, 2015). Studies showed that the spore germination and root colonisation percentage of AMF decreases with soil pH and moisture level (Vosatka *et al.*, 1999; Wang *et al.*, 1993; Rajan, 2016).

Different studies showed that the physiological parameters like photosynthesis rate, stomatal conductance, transpiration rate and leaf temperature had been significantly affected by AMF (Wu *et al.*, 2011; Yamin *et al.*, 2016; Bisht *et al.*, 2009; Auge *et al.*, 2015 and Huat *et al.*, 2002). In the present study, it was found that in the early stage of the experiment, the AMF had no significant on photosynthesis, but in the later stages, there was a significant difference between different treatments (Table 10). At 180 days, the inoculated seedlings had the higher photosynthesis rate than the non-inoculated seedlings. Seedlings inoculated with *G. etunicatum* recorded for highest photosynthesis rate. The study showed that the non-inoculated seedlings showed lower stomatal conductance than that of the inoculated seedlings at 120 days of the experiment (Table 11). The stomatal conductance was found higher in *G. etunicatum* and combination of *F. mosseae* + *G. etunicatum*. It was also found that the transpiration rate and leaf temperature showed no significant influence among the treatments throughout the experiment, however, there was significant difference found in transpiration rate at 180 days of the experiment where, *G. etunicatum* showed the highest value of transpiration rate. Site factors like the soil type, soil depth, seasonal difference, precipitation, temperature and existing vegetation have a great role in influencing the AMF growth and establishment (Abbott and Robson. 1991). The plantation was established in the month of January 2017, so in the early stages of the plantation,

the seedlings were dependent upon weekly irrigation till the following four months (Appendix I). That may be the reason of changes in the physiological parameters like photosynthesis rate and transpiration rate which were found to have a significant effect in the later stages of the field experiment. Since AMF are heterotrophic, they need carbohydrate for growth and reproduction (Peterson *et al.*, 1984; Birhane *et al.* 2018). So, it may be the reason during early stages of the plantation photosynthesis rate was low in seedlings, but during later stages of the experiment, seedlings used increase photosynthesis and carbon supply.

During the study period, it was found, that the root was infected in both inoculated and non-inoculated plants (Table 15). There was a significant difference between the inoculated seedlings and the non-inoculated seedlings. The root colonization percentage was found highest in seedlings inoculated with both *F. mosseae* and *G. etunicatum*. The soils collected from rhizospheric regions were found to have spores in both inoculated and non-inoculated plants (Table 16). Initially, *F. mosseae* inoculated seedlings showed highest total spore count during the 60 and 120 days of planting. At 180 days *G. etunicatum* inoculated seedlings showed highest spores per 10 g of soil. The spore density and root colonization percentage depends on various factors like vegetation, precipitation, soil nutrients and soil pH (Birhane *et al.*, 2018; Abbott and Robson. 1991).

Rajan (2016) showed that the root colonisation percentage of AMF declines with increase in drought stress. So the result obtained in the present study showed very low root colonisation percentage among the seedlings as these seedlings were under water deficit condition in the early stages of the experiment. It was also observed that the seedlings inoculated with AMF showed more root colonisation than the non-inoculated seedlings.

5.3 SOIL NUTRIENT STATUS

The development of AMF in soil is heavily dependent on edaphic properties such as pH and soil humidity (Posada *et al.*, 2018). The soil physicochemical properties can alter the occurrence, distribution and effectiveness

of AMF (Aliasgharzadeh *et al.*, 2001). The spore density and root colonization percentage of AMF are correlated to total nitrogen, available phosphorus and available potassium in soil and soil pH; hence AMF referred as an indicator for soil and ecosystem health (Birhane *et al.*, 2018). A study by Jeyanny *et al.* (2011) showed that AMF inoculation in seedlings at nursery stage alter the soil chemical properties by increasing the soil pH, EC, available P, exchangeable Ca and Mg contents than the non-inoculated seedlings. However, the most suitable condition for AMF development in soil is pH 6-7 (Aditya *et al.*, 2010).

Before out-planting of the seedlings, soil samples were collected and tested for their soil pH, electrical conductivity, total nitrogen, and available phosphorus and organic carbon. After a period of 6 months completion of plantation, again the soil samples were collected and tested for the same. A clear increase in pH, electrical conductivity, total nitrogen, and available phosphorous and organic carbon (Table 17) in the soil were noticed. The similar results were found in different studies done by Muthunkumar *et al.*, 2001; Wu *et al.*, 2011; Rajan *et al.*, 2000; Giri *et al.*, 2005, Verma *et al.*, 2010; Querejeta *et al.*, 1998; Jha *et al.*, 2017.

The soil pH increase during the experiment, indicated that the soil reduced its acidic nature in the rhizosphere of the plant. The similar study by Giri *et al.* (2005), they observed AMF decreasing the alkalinity of soil to make the rhizosphere more suitable for the plant and increased the availability of nutrients. The present study also showed that all the nutrient availability in the soil increased during the end of the experiment.

5.4 PLANT NUTRIENT STATUS

The beneficial micro-organisms like AMF are the key elements for the plant to get access to P by extending the surface area of roots using their hyphae (Dhawal *et al.* 2016). Arbuscular mycorrhizal fungi also help the plants to stimulate their growth by increasing the photosynthesis rate, resisting soil-borne pathogen attack and improving osmotic adjustment during stress conditions (Al-Karki, 2006). Studies showed that under field conditions inoculation with AMF increased

phosphorous content in foliar region of plants *Argania spinosa* (El Mrabet *et al.*, 2014), *Bauhinia faberi* (Yamin *et al.*, 2016), *Olea europaea* (Estaun *et al.*, 2003), *Citrus* spp. (Ortas and Ustuner, 2014), *Casuarina equisetifolia* (Vasanthakrishna *et al.*, 1995), *Prunus persica* (Wu *et al.*, 2011), *Tectona grandis* (Rajan *et al.*, 2000) and *Acacia mangium* (Jeyanny *et al.*, 2011).

In the present study, the plant phosphorous had significantly higher value in inoculated seedlings than that of the non-inoculated seedlings. *F. mosseae* + *G. etunicatum* showed the highest plant phosphorus level in the experiment followed by *F. mosseae*. The obtained result was similar to Sumana and Bagyaraj (2003), where phosphorous levels were calculated in *Azadiracta indica* seedlings inoculated with different AMF (*Glomus fasciculatum*, *Glomus geosporum*, *Glomus deserticola* and *Funneliformis mosseae*) and the maximum result of phosphorous was found in the seedlings where *F. mosseae* was used as an inoculum.

Different studies also showed that the plant nitrogen and potassium increased with inoculation of AMF (Estaun *et al.*, 2003; Wu *et al.*, 2011). In the present study, AMF found to have no significant influence on plant nitrogen content. Similar result was observed by Zhao *et al.* (2006) on *Camptotheca acuminata* seedlings inoculated with *Acaulospora mellea*, *Glomus diaphanum* and *Sclerocystis sinuosa*.

CONCLUSION

Three different AMF species (*Funneliformis mosseae*, *Acaulospora mellea* and *Glomus etunicatum*) individually or in combinations, had shown significant influence on height, collar diameter and number of leaves of *Swietenia macrophylla* seedlings under nursery conditions when compared to control, but they did not differ among them efficiently. Different doses of AMF application had a significant effect on height, collar diameter and number of leaves of the seedlings and it was found the growth of the seedlings increased with increase in doses. Under field transplantations, AMF had no significant influence on both biometric (height, collar diameter and number of leaves) and physiological (photosynthesis rate, transpiration rate, stomatal conductance and leaf temperature) parameters of the

seedlings. However, at later stages of field plantation there was a significant influence of AMF on photosynthesis rate and transpiration rate of the seedlings. The possible reason for it, may be that the AMF could not establish in the field. The reason supported by the fact that AMF requires a neutral soil for its development and the soil pH was observed acidic (5.46) in nature. The soil collected from the rhizosphere of the seedlings showed a relatively high pH (5.94). This concluded that AMF helped in altering the pH of the soil to make it more favourable for the plant. It was also observed that soil nitrogen, phosphorus, potassium and carbon contents increased in the rhizosphere region of plants after AMF application. From the nutrient analysis of the seedlings showed that AMF helped the plant in absorbing more nutrients from the soil. The plant P and C content was found significantly higher in inoculated seedlings. The root colonisation percentage was found to be low throughout the experiment. But, there was a significant increase in root colonisation percentage of inoculated seedlings than the non-inoculated seedlings. This showed that seedlings inoculated with AMF have better root colonising capacity than non-inoculated seedlings. From the study, it was found that under field conditions, although there was no influence of AMF on seedling growth parameters, but it helped them to increase their nutrient acquisition capacity by enhancing the nutrient level in the rhizosphere of the plants. Further study may be required to identify better native species of AMF for *S. macrophylla*.



SUMMARY

SUMMARY

An experiment was conducted to analyse the effect of arbuscular mycorrhizal fungi on the growth of *Swietenia macrophylla* seedlings under field condition. The experiment was conducted in two phases. The first phase was in nursery and the second phase was under field condition. In nursery AMF were applied individually and in combinations with three different doses i.e. 5 g, 10 g and 15 g. The best four treatments from the nursery experiment were selected for the field. The treatments were laid in a factorial completely randomized design in nursery and in a randomized block design in field. The three species of arbuscular mycorrhizal fungi used in the experiment were *Funneliformis mosseae*, *Acaulospora mellea* and *Glomus etunicatum*.

The salient findings of the study are given below.

1. In the nursery experiment, seedlings inoculated with AMF showed higher height, collar diameter and number of leaves than the control.
2. The individual or combinations of AMF species had similar influence on seedling height, collar diameter and number of leaves.
3. The different doses of AMF showed significant influence on seedling height, collar diameter and number of leaves. The highest dose (15 g) showed highest increase in the growth parameters.
4. The root colonisation percentage and total spore count were recorded at 90 days of inoculation showed that all the treatment combinations had similar root colonisation and spore numbers.
5. The nutrient status of the soil was calculated prior to the filed plantation. The soil pH was recorded 5.46. The soil total nitrogen content, available phosphorus content, available potassium content and organic matter content were found to be 0.093 % , 8.36 kg/ha, 130 kg/ha and 0.93 % , respectively.
6. In the field experiment, the individual or combinations of AMF had no influence on seedling height, collar diameter and number of leaves.

7. The physiological parameters like photosynthesis rate, transpiration rate, stomatal conductance and leaf temperature of the seedlings, were found not influenced by AMF inoculation under the field conditions.
8. Root colonisation percentage and total spore count of AMF was found to be have significantly higher in inoculated seedlings than the control.
9. The soil pH was calculated at 180 days of planting. The soil pH found to be 5.94.
10. The soil nutrient status were recorded at 180 days of the planting. The soil total nitrogen content, available phosphorus content, available potassium content and organic matter content were found to be 0.102 %, 33.85 kg/ha, 260.58 kg/ha and 1.23%, respectively.
11. The plant nutrient status was calculated at 180 days of planting. The AMF was found to have no significant influence on plant potassium and nitrogen contents.
12. The AMF inoculated seedlings showed significantly higher plant phosphorous and carbon content than the non-inoculated seedlings.

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APPENDICES

APPENDICES

APPENDIX - I

Month-wise meteorological data

Year	Month	Maximum temperature (°C)	Minimum temperature (°C)	Rainfall (mm)	Relative humidity (%)	Mean evaporation (mm)	Number of Rainy days
2016	Jan	33.2	23.0	28.3	56	5.1	1
	Feb	35.3	23.5	011.4	57	5.1	1
	Mar	36.3	25.2	009.8	67	4.7	1
	Apr	35.8	26.2	025.8	69	4.7	2
	May	34.0	24.2	270.7	78	3.8	9
	Jun	29.8	21.7	654.7	89	2.1	22
	Jul	29.9	21.6	390.4	85	2.5	19
	Aug	30.4	23.2	183.5	83	2.9	19
	Sep	30.3	23.6	086.0	82	2.9	10
	Oct	31.5	22.7	037.0	81	2.8	4
	Nov	32.9	22.2	013.8	69	3.0	1
	Dec	32.4	22.3	052.9	69	3.3	3
2017	Jan	34.1	22.9	000.0	53	4.7	0
	Feb	36.0	23.2	000.0	51	5.7	0
	Mar	36.1	24.7	013.2	67	4.5	1
	Apr	35.7	26.0	019.1	70	4.0	1
	May	34.6	24.9	167.5	72	3.6	11
	Jun	30.4	23.5	630.2	87	2.5	25
	Jul	30.8	22.8	385.5	85	2.7	22
	Aug	30.1	23.3	478.0	87	2.6	17
	Sep	31.5	22.9	413.9	84	2.8	18
	Oct	31.7	22.3	183.4	81	2.3	10
	Nov	33.0	21.8	058.3	73	3.0	5
	Dec	32.4	21.1	011.5	63	3.9	2

APPENDIX - II
ANOVA TABLES

EXPERIMENT 1

1. Height in nursery
 - a. 30 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	18344.07	833.82	388.34	S
Error	44	94.47	2.14		
Total	65	18438.544			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	19.198	3.20	1.432	NS
Doses	2	29.065	14.532	6.504	S
Treatments * Doses	12	22.018	1.835	0.821	NS
Error	42	93.849	2.234		
Total	63	17831.864			

(S- significant at level 0.05, NS- non-significant)

- b. 60 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	135.36	6.44	2.02	S
Error	44	133.53	3.17		
Total	65	37180.302			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	17.764	2.961	0.905	NS
Doses	2	36.144	18.072	5.521	S
Treatments * Doses	12	39.037	3.253	0.994	NS
Error	42	137.474	3.273		
Total	63	35983.084			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	417.05	19.86	3.05	S
Error	44	272.64	6.49		
Total	65	65783.717			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	32.377	5.396	0.812	NS
Doses	2	149.458	74.729	11.249	S
Treatments * Doses	12	152.727	12.727	1.916	NS
Error	42	279.010	6.643		
Total	63	63710.263			

(S- significant at level 0.05, NS- non-significant)

2. Collar diameter in nursery

a. 30 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	187.86	8.53	1615.44	S
Error	44	0.233	0.005		
Total	65	188.099			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	0.253	0.042	8.200	S
Doses	2	0.186	0.093	18.104	S
Treatments * Doses	12	0.334	0.028	5.402	S
Error	42	0.216	0.005		
Total	63	182.882			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	1.64	0.078	12.35	S
Error	44	0.26	0.006		
Total	65	442.324			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	0.186	0.031	4.936	S
Doses	2	0.244	0.122	19.416	S
Treatments * Doses	12	0.398	0.033	5.285	S
Error	42	0.264	0.006		
Total	63	429.421			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	2.32	0.111	3.27	S
Error	44	1.41	0.034		
Total	65	871.988			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	0.293	0.049	1.428	NS
Doses	2	0.784	0.392	11.467	S
Treatments * Doses	12	0.724	0.060	1.766	NS
Error	42	1.436	0.034		
Total	63	840.867			

(S- significant at level 0.05, NS- non-significant)

3. Number of leaves in nursery

a. 30 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	2505.74	113.89	495.60	S
Error	44	10.11	0.23		
Total	65	2515.859			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	3.071	0.512	2.154	NS
Doses	2	7.470	3.735	15.721	S
Treatments * Doses	12	4.065	0.339	1.426	NS
Error	42	9.979	0.238		
Total	63	2405.312			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	20.334	0.968	4.44	S
Error	41	9.14	0.218		
Total	65	3382.123			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	0.094	0.016	0.072	NS
Doses	2	17.770	8,885	41.073	S
Treatments * Doses	12	2.313	0.193	0.891	NS
Error	42	9.085	0.216		
Total	63	3238.978			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	21	62.81	2.991	11.40	S
Error	44	11.01	0.262		
Total	65	6220.129			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	11.716	1.953	6.502	S
Doses	2	26.256	13.128	43.713	S
Treatments * Doses	12	22.076	1.840	6.126	S
Error	42	12.613	0.300		
Total	63	5991.797			

(S- significant at level 0.05, NS- non-significant)

Factorial CRD and CONTROL

1. Height

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	19.198	3.20	1.432	NS
Doses	2	29.065	14.532	6.504	S
Treatments * Doses	12	22.018	1.835	0.821	NS
Control * Rest	1	606.68	606.68	271.566	S
Error	42	93.849	2.234		
Total	63	18438.544			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	17.764	2.961	0.905	NS
Doses	2	36.144	18.072	5.521	S
Treatments * Doses	12	39.037	3.253	0.994	NS
Control * Rest	1	1197.218	1197.218	365.786	S
Error	42	137.474	3.273		
Total	63	37180.302			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	32.377	5.396	0.812	NS
Doses	2	149.458	74.729	11.249	S
Treatments * Doses	12	152.727	12.727	1.916	NS
Control * Rest	1	2073.454	2073.454	312.126	S
Error	42	279.010	6.643		
Total	63	65783.717			

(S- significant at level 0.05, NS- non-significant)

2. Collar diameter

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	0.253	0.042	8.200	S
Doses	2	0.186	0.093	18.104	S
Treatments * Doses	12	0.334	0.028	5.402	S
Control * Rest	1	5.217	5.217	1043.4	S
Error	42	0.216	0.005		
Total	63	188.099			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	0.186	0.031	4.936	S
Doses	2	0.244	0.122	19.416	S
Treatments * Doses	12	0.398	0.033	5.285	S
Control * Rest	1	12.903	12.903	2150.5	S
Error	42	0.264	0.006		
Total	63	442.234			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	0.293	0.049	1.428	NS
Doses	2	0.784	0.392	11.467	S
Treatments * Doses	12	0.724	0.060	1.766	NS
Control * Rest	1	31.121	31.121	915.323	S
Error	42	1.436	0.034		
Total	63	871.998			

(S- significant at level 0.05, NS- non-significant)

3. Number of leaves

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	3.071	0.512	2.154	NS
Doses	2	7.470	3.735	15.721	S
Treatments * Doses	12	4.065	0.339	1.426	NS
Control * Rest	1	110.547	110.547	464.483	S
Error	42	9.979	0.238		
Total	63	2515.859			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	0.094	0.016	0.072	NS
Doses	2	17.770	8,885	41.073	S
Treatments * Doses	12	2.313	0.193	0.891	NS
Control * Rest	1	143.145	143.145	662.708	S
Error	42	9.085	0.216		
Total	63	3382.123			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	6	11.716	1.953	6.502	S
Doses	2	26.256	13.128	43.713	S
Treatments * Doses	12	22.076	1.840	6.126	S
Control * Rest	1	228.332	228.332	761.106	S
Error	42	12.613	0.300		
Total	63	6220.129			

(S- significant at level 0.05, NS- non-significant)

EXPERIMENT 2

1. Height in field

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	7.066	1.767	0.551	NS
Replication	2	17.354	8.677	2.705	NS
Error	8	25.659	3.207		
Total	14	15873.036			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	5.633	1.408	0.455	NS
Replication	2	39.129	19.564	6.323	0.023
Error	8	24.754	3.094		
Total	14	21942.075			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	4.493	1.123	0.221	NS
Replication	2	92.212	46.106	9.082	S
Error	8	40.615	5.077		
Total	14	31596.423			

(S- significant at level 0.05, NS- non-significant)

d. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	7.225	1.806	0.187	NS
Replication	2	245.816	122.908	12.738	S
Error	8	77.192	9.649		
Total	14	47061.622			

(S- significant at level 0.05, NS- non-significant)

e. 150 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	31.625	7.906	0.301	NS
Replication	2	757.347	378.674	14.404	S
Error	8	210.318	26.290		
Total	14	76248.374			

(S- significant at level 0.05, NS- non-significant)

f. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	85.785	21.446	0.324	NS
Replication	2	2132.608	1066.304	16.085	S
Error	8	530.350	66.294		
Total	14	130333.270			

(S- significant at level 0.05, NS- non-significant)

2. Collar diameter in field

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.547	0.137	1.085	NS
Replication	2	1.915	0.958	7.598	S
Error	8	1.008	0.126		
Total	14	272.457			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.745	0.186	1.375	NS
Replication	2	4.796	2.398	17.693	S
Error	8	1.084	0.136		
Total	14	471.331			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.743	0.186	0.941	NS
Replication	2	11.833	5.916	29.963	S
Error	8	1.580	0.197		
Total	14	795.937			

(S- significant at level 0.05, NS- non-significant)

d. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	1.189	0.297	0.879	NS
Replication	2	25.320	12.660	37.452	S
Error	8	2.704	0.338		
Total	14	1284.136			

(S- significant at level 0.05, NS- non-significant)

e. 150 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	1.812	0.453	0.694	NS
Replication	2	49.972	24.986	38.281	S
Error	8	5.222	0.653		
Total	14	2095.407			

(S- significant at level 0.05, NS- non-significant)

f. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	3.716	0.929	0.838	NS
Replication	2	97.117	48.559	43.805	S
Error	8	8.868	1.109		
Total	14	3429.727			

(S- significant at level 0.05, NS- non-significant)

3. Number of leaves in field

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	1.009	0.252	1.698	NS
Replication	2	1.149	0.575	3.867	NS
Error	8	1.189	0.149		
Total	14	613.403			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	2.344	0.586	0.078	NS
Replication	2	0.998	0.499	0.918	NS
Error	8	4.350	0.544		
Total	14	807.918			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	2.124	0.531	1.074	NS
Replication	2	3.495	1.747	3.534	NS
Error	8	3.955	0.494		
Total	14	1099.964			

(S- significant at level 0.05, NS- non-significant)

d. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.731	0.183	0.276	NS
Replication	2	9.729	4.864	7.336	S
Error	8	5.304	0.663		
Total	14	1555.221			

(S- significant at level 0.05, NS- non-significant)

e. 150 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	2.703	0.676	0.484	NS
Replication	2	13.469	6.734	4.828	S
Error	8	11.158	1.395		
Total	14	2443.126			

(S- significant at level 0.05, NS- non-significant)

f. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	1.942	0.486	0.486	NS
Replication	2	13.913	6.957	6.963	S
Error	8	7.992	0.999		
Total	14	3901.701			

(S- significant at level 0.05, NS- non-significant)

4. Photosynthesis rate in field

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	8.447	2.112	2.778	NS
Replication	2	4.264	2.132	2.807	NS
Error	8	6.077	0.760		
Total	14	266.473			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	5.708	1.427	2.028	NS
Replication	2	1.584	0.792	1.127	NS
Error	8	5.620	0.703		
Total	14	420.926			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	2.009	0.502	4.776	S
Replication	2	5.632	2.816	26.775	S
Error	8	0.841	0.105		
Total	14	430.893			

(S- significant at level 0.05, NS- non-significant)

d. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	13.462	3.366	3.271	NS
Replication	2	7.846	3.923	3.812	NS
Error	8	8.232	1.029		
Total	14	656.418			

(S- significant at level 0.05, NS- non-significant)

e. 150 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	37.953	9.488	4.256	S
Replication	2	3.722	1.861	0.835	NS
Error	8	17.834	2.229		
Total	14	1038.321			

(S- significant at level 0.05, NS- non-significant)

f. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	33.169	8.292	5.250	S
Replication	2	3.451	1.726	1.092	NS
Error	8	12.637	1.580		
Total	14	346.383			

(S- significant at level 0.05, NS- non-significant)

5. Stomatal conductance in field

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.002	0.001	3.816	NS
Replication	2	0.0001	0.00005	0.286	NS
Error	8	0.001	0.0002		
Total	14	0.127			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.008	0.002	1.266	NS
Replication	2	0.004	0.002	1.214	NS
Error	8	0.012	0.002		
Total	14	0.042			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.001	0.0002	0.874	NS
Replication	2	0.001	0.0005	2.228	NS
Error	8	0.002	0.0002		
Total	14	0.290			

(S- significant at level 0.05, NS- non-significant)

d. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.010	0.003	9.264	S
Replication	2	0.002	0.001	3.853	NS
Error	8	0.002	0.0002		
Total	14	0.415			

(S- significant at level 0.05, NS- non-significant)

e. 150 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.004	0.001	1.271	NS
Replication	2	0.010	0.005	5.940	S
Error	8	0.007	0.001		
Total	14	0.050			

(S- significant at level 0.05, NS- non-significant)

f. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.032	0.008	3.158	NS
Replication	2	0.024	0.012	4.787	S
Error	8	0.020	0.003		
Total	14	0.153			

(S- significant at level 0.05, NS- non-significant)

6. Transpiration rate in field

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.337	0.084	1.680	NS
Replication	2	0.465	0.233	4.631	S
Error	8	0.402	0.050		
Total	14	39.764			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.222	0.056	0.577	NS
Replication	2	0.529	0.265	2.749	NS
Error	8	0.770	0.096		
Total	14	10.125			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.708	0.177	2.294	0.148
Replication	2	0.102	0.051	0.658	NS
Error	8	0.618	0.077		
Total	14	52.543			

(S- significant at level 0.05, NS- non-significant)

d. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.414	0.103	1.294	NS
Replication	2	0.443	0.221	2.767	NS
Error	8	0.640	0.080		
Total	14	144.409			

(S- significant at level 0.05, NS- non-significant)

e. 150 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.622	0.155	1.239	0.368
Replication	2	0.364	0.182	1.452	NS
Error	8	1.003	0.125		
Total	14	129.594			

(S- significant at level 0.05, NS- non-significant)

f. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	7.025	1.756	4.350	S
Replication	2	3.223	1.612	3.992	NS
Error	8	3.230	0.404		
Total	14	416.378			

(S- significant at level 0.05, NS- non-significant)

7. Leaf temperature

a. 30 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.287	0.072	0.355	NS
Replication	2	7.141	3.571	17.650	S
Error	8	1.618	0.202		
Total	14	15716.690			

(S- significant at level 0.05, NS- non-significant)

b. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.379	0.095	0.522	NS
Replication	2	14.434	7.217	39.755	S
Error	8	1.452	0.182		
Total	14	19386.488			

(S- significant at level 0.05, NS- non-significant)

c. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.357	0.089	0.629	NS
Replication	2	4.203	2.102	14.791	S
Error	8	1.137	0.142		
Total	14	15596.639			

(S- significant at level 0.05, NS- non-significant)

d. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	2.252	0.563	1.980	NS
Replication	2	0.274	0.137	0.481	NS
Error	8	2.275	0.284		
Total	14	18298.691			

(S- significant at level 0.05, NS- non-significant)

e. 150 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	1.487	0.372	1.508	NS
Replication	2	0.597	0.298	1.211	NS
Error	8	1.971	0.246		
Total	14	18335.680			

(S- significant at level 0.05, NS- non-significant)

f. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.265	0.066	0.555	NS
Replication	2	8.383	4.192	35.090	S
Error	8	0.956	0.119		
Total	14	15619.573			

(S- significant at level 0.05, NS- non-significant)

8. Absolute growth rate

a. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.0002	0.00006	0.256	NS
Replication	2	0.008	0.004	17.805	S
Error	8	0.002	0.0002		
Total	14	0.548			

(S- significant at level 0.05, NS- non-significant)

b. 90 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.001	0.0002	0.273	NS
Replication	2	0.016	0.008	6.204	S
Error	8	0.010	0.001		
Total	14	0.986			

(S- significant at level 0.05, NS- non-significant)

c. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.002	0.001	0.311	NS
Replication	2	0.046	0.023	12.166	S
Error	8	0.015	0.002		
Total	14	1.749			

(S- significant at level 0.05, NS- non-significant)

d. 150 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.010	0.002	0.339	NS
Replication	2	0.169	0.085	11.767	S
Error	8	0.058	0.007		
Total	14	3.997			

(S- significant at level 0.05, NS- non-significant)

e. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.036	0.009	0.680	NS
Replication	2	0.404	0.202	15.186	S
Error	8	0.107	0.013		
Total	14	8.180			

(S- significant at level 0.05, NS- non-significant)

9. Root colonisation percentage

a. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	6.625	1.656	5.579	S
Replication	2	0.888	0.444	1.495	NS
Error	8	2.375	0.297		
Total	14	193.837			

(S- significant at level 0.05, NS- non-significant)

b. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	39.736	9.934	25.869	S
Replication	2	1.941	0.971	2.527	NS
Error	8	3.072	0.384		
Total	14	716.104			

(S- significant at level 0.05, NS- non-significant)

10. Total spore count

a. 60 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	742.217	185.554	45.721	S
Replication	2	8.049	4.024	0.992	NS
Error	8	32.467	4.058		
Total	14	7.54.469			

(S- significant at level 0.05, NS- non-significant)

b. 120 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	7.4.386	176.096	55.086	S
Replication	2	1.062	0.531	0.166	NS
Error	8	25.574	3.197		
Total	14	12445.565			

(S- significant at level 0.05, NS- non-significant)

c. 180 days

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	808	2020.133	7.269	S
Replication	2	16.271	8.135	0.293	NS
Error	8	222.475	27.809		
Total	14	18803.472			

(S- significant at level 0.05, NS- non-significant)

11. Plant nutrient contents

a. Phosphorous

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	610.788	152.697	4.133	S
Replication	2	149.999	74.999	2.030	NS
Error	8	295.545	36.943		
Total	14	7507.352			

(S- significant at level 0.05, NS- non-significant)

b. Nitrogen

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.031	0.008	0.469	NS
Replication	2	0.013	0.006	0.386	NS
Error	8	0.130	0.016		
Total	14	15.543			

(S- significant at level 0.05, NS- non-significant)

c. Potassium

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	0.094	0.024	2.750	NS
Replication	2	4.379	2.189	243.222	S
Error	8	0.069	0.009		
Total	14	31.980			

(S- significant at level 0.05, NS- non-significant)

d. Carbon

Source of variation	Degrees of freedom	Total sum of squares	Mean sum of squares	F value	Sig.
Treatments	4	3.569	0.892	21.790	S
Replication	2	0.399	0.200	4.873	S
Error	8	0.328	0.041		
Total	14	14931.828			

(S- significant at level 0.05, NS- non-significant)

FIELD PERFORMANCE OF ARBUSCULAR
MYCORRHIZAL FUNGI ON VEGETATIVE GROWTH
OF MAHOGANY (*Swietenia macrophylla* King.)
SEEDLINGS

By

SATYABRATA NAYAK
(2015-17-013)

ABSTRACT OF THE THESIS

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ABSTRACT

A study was conducted to determine the efficiency of arbuscular mycorrhizal fungi (AMF) on the vegetative growth of mahogany (*Swietenia macrophylla* King.) seedlings under field conditions at College of Forestry nursery and Instructional farm of Kerala Agricultural University, Vellanikkara, Thrissur district, Kerala during 2015-2017. Three AMF species used in the study were *Funneliformis mosseae* (Fm), *Acaulospora mellea* (Am) and *Glomus etunicatum* (Ge).

The study was carried out in two parts. The first experiment was conducted in nursery for three months and the second experiment was conducted in the field for six months. In the nursery experiment, the AMF were applied to the seedlings either individually or in combinations (Fm, Am, Ge, Fm + Am, Am + Ge, Ge + Fm, Fm + Am + Ge) at three different doses (5 g, 10 g and 15 g) and laid out in a factorial CRD with control as 21+1 treatment combinations and three replications. The purpose of the nursery experiment was to choose the best four treatments for out-planting in the main field.

It was found that the AMF species used in the nursery significantly influence the biometric characters (height, collar diameter and number of leaves) of the seedlings when compared to control. However, the AMF treatments did not differ among themselves. There was significant differences among different doses of AMF. The seedlings growth increased when the doses of AMF increased. From the result, Fm (15 g), Am (15 g), Ge (15 g) and Fm + Ge (15 g) were chosen as treatments for seedlings for the second experiment.

The field experiment was laid out in a randomized block design with five treatments (four chosen treatments from nursery + control) with three replications. The observations like height, collar diameter, number of leaves, photosynthesis rate, stomatal conductance, transpiration rate and leaf temperature were recorded at 30 days interval. Soil nutrient analysis was done before and after the planting of

the seedlings. Plant's nutrient analysis were done after the completion of the experiment.

It was found that inoculation with AMF, did not result any significant differences in height, collar diameter, number of leaves, stomatal conductance and leaf temperature. However, the AMF had significant influence on photosynthesis rate and transpiration rate on the seedlings at later stages of the experiment. From soil nutrient analysis, it was found that total N, available P, available K and organic carbon content increased with AMF inoculation. The soil pH changed from 5.46 to 5.94. From plant nutrient analysis, it was found, that there were no significant effect of AMF on K and N content of the seedlings. However, the P and C content of the seedlings increased due to AMF. The root colonisation percentage and total spore count were found to be low in all treatments. These values were significantly higher in seedlings inoculated with AMF than the non-inoculated seedlings.

From the study, it was concluded that increasing doses of AMF can increase *S. macrophylla* seedlings growth in nursery. The AMF did not affect the growth of seedling during first six months of field experiment. But, some physiological parameters like photosynthesis rate and transpiration were influenced by the AMF at later stages of the growth. It may be pointed to the fact that AMF experimented could not establish in the field as evidence by low root colonisation percentage. This may be because of the soil reaction which was acidic in nature and was not suitable for AMF to establish since AMF require neutral medium for its better growth and development. Arbuscular mycorrhizal fungi helped in improving the pH of soil along with other nutrient contents (N, P, K and organic C). The seedlings were benefitted from AMF as their P uptake was increased. Further studies are required to identify better native species of AMF for *S. macrophylla* in acidic ultisols of Kerala.

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