

**EFFECT OF DIFFERENT LEVELS OF MYCOGOLD™ ON GROWTH AND
YIELD OF EGGPLANT (*Solanum melongena* L.)**

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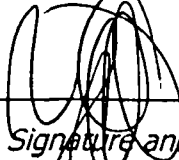
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ABSTRACT

A study on the effect of different levels of MycoGold™ on the growth and yield of eggplants was conducted from June to November 2015 at the Faculty of Sustainable Agriculture (FSA), Universiti Malaysia Sabah. This study observed the difference of growth and yield of eggplant supplied with different amounts of MycoGold™. The treatments involved in this study include control, NPK (12:12:17), NPK3M (NPK Blue + 300g MycoGold™), NPK4M (NPK Blue + 400g MycoGold™), and NPK5M (NPK Blue + 500g MycoGold™). The experimental design used was Completely Randomized Design (CRD). The parameters that were measured for this study include plant height, stem diameter, flowering time, fruit girth, fruit length, fruit dry weight, fruit fresh weight, and number of fruits per plant. Plant height and stem diameter data were collected once in two weeks after transplant. Flowering time was collected when all structures of flower was developed. Fruits were collected a total of four times and their parameters were recorded. Collected data was analysed using one-way Analysis of Variance (ANOVA) with SAS version 9.4 and each treatment has no significant effect on all measurable variables. It was found that colonization of mycorrhizae on eggplant roots were low in soil phosphorus level of 339.1 mg kg⁻¹.



Kesan Kadar Berlainan MycoGold Terhadap Pertumbuhan dan Hasil Terung

ABSTRAK

Sebuah kajian bertajuk kesan kadar berlainan MycoGold™ terhadap pertumbuhan dan hasil terung telah dijalankan daripada bulan Jun hingga November 2015 di Fakulti Pertanian Lestari (FPL), Universiti Malaysia Sabah. Kajian tersebut dijalankan untuk mengkaji perbezaan pertumbuhan dan hasil terung dalam kadar inokulasi MycoGold™ yang berlainan. Rawatan yang dibagi kepada terung merupakan kawalan, NPK (12:12:17), NPK3M (NPK Blue + 300g MycoGold™), NPK4M (NPK Blue + 400g MycoGold™), and NPK5M (NPK Blue + 500g MycoGold™). Rekabentuk eksperimen yang akan digunakan adalah Rekabentuk secara Rawak (CRD). Parameter yang diukur merupakan ketinggian tumbuhan, ketebalan batang, masa berbunga, lilitan buah, panjang buah, keberatan kering buah, keberatan basah buah, dan bilangan buah sepokok. Data ketinggian tumbuhan dan ketebalan batang direkod sekali bagi setiap dua minggu selepas anak benih diubahkan. Masa berbunga direkod semasa semua struktur bunga dibentuk. Buah dituai sebanyak empat kali dan semua parameter buah direkod. Data yang dikumpul dianalisis dengan menggunakan ANOVA sehalu dengan SAS versi 9.4 dan didapati semua rawatan tidak mempunyai perbezaan signifikan pada semua variasi yang dapat diukur. Kehadiran mikorhiza pada akar pokok terung rendah dalam tanah yang mengandungi 339.1 mg kg⁻¹ fosforus.

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LIST OF SYMBOLS, UNITS AND ABBREVIATIONS

AM	Arbuscular Mycorrhiza
AMF	Arbuscular Mycorrhizal Fungi
ANOVA	Analysis of Variance
CRD	Complete Randomized Design
EFSB	Eggplant Fruit and Shoot Borer
FSA	Faculty of Sustainable Agriculture
HCl	Hydrochloric acid
INVAM	International Culture Collection of Arbuscular Mycorrhizal Fungi
KOH	Potassium Hydroxide
ppm	parts per million (mg/kg)
SAS	Statistical Analysis System
™	Trademark
UMS	Universiti Malaysia Sabah
USDA	United States Department of Agriculture
VAM	Vesicular Arbuscular Mycorrhizae



LIST OF FORMULAS

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3.1 Mycorrhizae Root Colonisation Percentage (MRC %) $\text{MRC \%} = \frac{\text{Number of mycorrhizal root at intersections}}{\text{Total number of root at intersections}} \times 100$	20

CHAPTER 1

INTRODUCTION

1.1 Introduction

The demands on agriculture have shifted from just providing food to currently implementing sustainable practices on top of providing food (Lehmann, 2013). The increasing environmental awareness of the public in response towards the consequences of the Green Revolution movement that has swept the agriculture world in a haze of increased chemical inputs had placed agriculture in a spot where farming practices have to incorporate sustainability. To this, farmers seek alternatives from chemical inputs to curb problems such as soil degradation, pollution, rising fuel price and public demands (Zakaria, 2006). One of the alternatives includes the use of microorganisms such as mycorrhizae.

Mycorrhizae are a type of fungi that form a symbiotic relationship with plant roots whereby the mycorrhizae exchanges carbohydrates produced by the host plant with helping in uptake of water and immobile soil nutrients such as phosphorus (P), copper (Cu), and zinc (Zn) (Miyasaka *et al.*, 2003). Muchovej (n.d.) stated that this fungus takes over the role to the plant's root hairs and acts as an extension of the root system. An additional benefit of mycorrhizae according to Muchovej (n.d.) is that they reduce susceptibility of roots to soil-borne pathogens. Ortas (2011) reported early flowering time in relation to inoculation of mycorrhizae. Other than that, mycorrhizae inoculation has been found to be able to cure land contaminated by heavy metals (Hernandez-Ortega *et al.*, 2012).

Eggplant (*Solanum melongena* L.) is found in abundance in Malaysia and have different varieties, also it is considered as an important commercial crop planted in Cameron Highlands, the most productive upland vegetable production area in the state



of Pahang, Peninsular Malaysia (Mohd Rasdi *et al.*, 2012). It is also one of the most widely accepted vegetable crops in South East Asia (Edathil *et al.*, 1994). Other than a source of dietary nutrient, eggplants are found to have varying degrees of anti-oxidant compounds depending on their skin colour according to a research done by Somawathi *et al.* (2014).

The Department of Agriculture, Forestry and Fisheries of South Africa (2011) states that eggplant is usually grown in light or sandy loam soils that provide good drainage and favourable soil temperatures. They later stated that this is because of the roots of eggplants grows moderately deep and needs a loose soil for optimum growth. According to the soil texture triangle by USDA, sandy loam soils have 10% clay, 60% sand, and 30% silt. A study did by Motha *et al.* (2014) shows the mycorrhizal fungi infection with eggplant roots of up to 90% in soil samples containing that of 10% clay, 60% sand, and 30% silt. The soil available in FSA is not suitable to be used due to the high amount of clay content in the soil as it is part of Silabukan soil series (Bavani, 2010). Heavy clay and saturated soils should be avoided due to build-up of root-rotting diseases according to Chen *et al.* (2002).

Following that, the development of mycorrhizal fungi seems to differ based on a number of different factors such as plant, soil and climatic factors, and will in turn vary in its symbiotic relationship. For the case of soil texture, a study was done by Carrenho *et al.* (2007) and found that soil texture was one of the most important factor influencing colonization percentage, causing a lower percentage of root colonization in clayey soil compared to sandy soil for sorghum. Carrenho *et al.* (2007) relates this to the fact that clayey soil tend to be high in soil nutrients and hence, becomes a limitation towards the development of mycorrhizal fungi. Xu *et al.* (2015) suggests that low AM fungi colonization may contribute to improved soil structure even though it does not benefit in plant growth and nutrient absorption.

The type of mycorrhizal fungi that is important in agriculture is the arbuscular mycorrhizal fungi (AMF). It had been reported to be able to reduce chemical fertilizer inputs by half (Abdullahi and Sheriff, 2013). This enables farmers to produce optimum yield at a lesser cost, meanwhile improving the soil health through the soil remediating effects of AMF (Hernandez-Ortega *et al.*, 2012) and increase livelihood by using less chemical fertilisers. There is no need for further increased input of chemical fertilisers

for unfavourable soil conditions where nutrient availability is low. Currently in Malaysia, much work has been done on selection of effective indigenous mycorrhizae and exploiting their beneficial effects for enhancing crop production (Naher *et al.*, 2013). Mycorrhiza inoculum is also increasingly being utilized and accepted in the agriculture industry of Malaysia (Khairuddin, 2002).

With all the talk about the benefits of mycorrhizae towards improving agriculture yield and for sustainability, one problem remains where mycorrhizae spores needs to be applied to the soil for the hopes of an association forming. Mycorrhizal researches have been done since the 1980s, and currently there are several companies in Malaysia producing mycorrhizal products (Khairuddin, 2002). However, the use of mycorrhizal inoculants is mainly used in the oil palm sector to combat the *Ganoderma* disease, and also to pull forward the time taken for seedlings to be suitable for transplant (Zakaria, 2006). One of the locally produced products of mycorrhizal inoculant is MycoGold™ produced by Malaysian Agri Group of Companies.

1.2 Justification

This experiment is conducted in hopes to help increase production of eggplant with the help of mycorrhizae invasion. Poultry manure is added as a form of control as it proved that it is able to supply organic matter, add nutrients, and improve soil structure. Furthermore, it is recommended in the cultivation of eggplants (Ullio, 2003). Mycorrhizae are able to help plants in the uptake of nutrients and water. In this case where poultry manure is a control in supplying the soil with nutrients and increasing soil organic matter, mycorrhizae, namely MycoGold™ will be compared to that of NPK treated soil and control in agronomic parameters such as fruit yield, fruit dry weight, fruit fresh weight, plant height, stem diameter, and flowering time.

MycoGold™ is used as the source of mycorrhizal inoculant because it can be purchased and is sold in the local market of Sandakan. Furthermore, not many studies are done using MycoGold™ for the recommended rate needed for increased eggplant production, as it is more widely known to be used in the oil palm sector. One study was done on the recommended rate on roselle showed highest productivity at an application rate of MycoGold™ at 400g (Aulia *et al.*, 2009). Based on that, the levels of MycoGold™ application is at 300g, 400g, and 500g. MycoGold™ is also an excellent

source of mycorrhizal inoculant because it contains several mycorrhizae species collected from different local soil locations, and has a spore count of 200 to 250 units per 10 g (Zakaria, 2006). This proves that MycoGold™ is suitable for a wide range of plant species and can be used for eggplants as well.

Eggplants were chosen as the subject of study because it has a growing period of 120 days that fits the timeline for the project to be carried out. Other than that, eggplants are widely acceptable in Malaysia, and farmers can profit greatly from increased production. The heavy feeding characteristic of eggplant is also another factor towards why it was chosen as a subject. As with all heavy feeding plants, more chemical fertilizer would be applied to match the need for the plants' continuous growth and yield production in terms of quality and quantity. With the help of mycorrhizae, the plants may grow even better with decreased fertilization.

1.3 Objectives

1. To compare effect of MycoGold™ on the growth and yield of eggplant.
2. To identify the optimum rate of MycoGold™ for production of eggplant.

1.4 Hypothesis

H_0 : There is no significant difference in the growth and yield of eggplant treated with different levels of MycoGold™

H_a : There is significant difference in the growth and yield of eggplant treated with different levels of MycoGold™

CHAPTER 2

LITERATURE REVIEW

2.1 Mycorrhizae

Mycorrhizae are highly evolved and have symbiotic association that is non-pathogenic between roots of plants and certain specialized soil fungi that colonize the cortical tissues of roots during active plant growth periods in both natural environment and in cultivation (Motha *et al.*, 2014). Mycorrhizae exists in all soils, however the numbers are not enough to produce significant benefit on plant growth and health in soils that had been disturbed by human activities such as farming, therefore needs to be supplemented. Furthermore, different plant species or even genotypes, soil, and environmental factors have different responses towards mycorrhizae (Lehmann, 2013).

There are two major groups of mycorrhizal fungi according to Raminder *et al.* (2014). There are five main types of mycorrhizal fungi and they are Arbuscular endomycorrhiza, Ericoid endomycorrhiza, Orchid endomycorrhiza, Ectendomycorrhiza, and Ectomycorrhiza. However, they are only split into two major groups which are ectomycorrhiza and endomycorrhiza. Figure 2.1 shows the principle structural features of five types of mycorrhiza. Although there are principle key features to identify the different types of mycorrhizae associations, the same species of mycorrhiza can form different associations with different plants (Mohammadi *et al.*, 2011). Although said, there are reports of specificity in fungal symbiont for plant species (Mohammadi *et al.*, 2011).



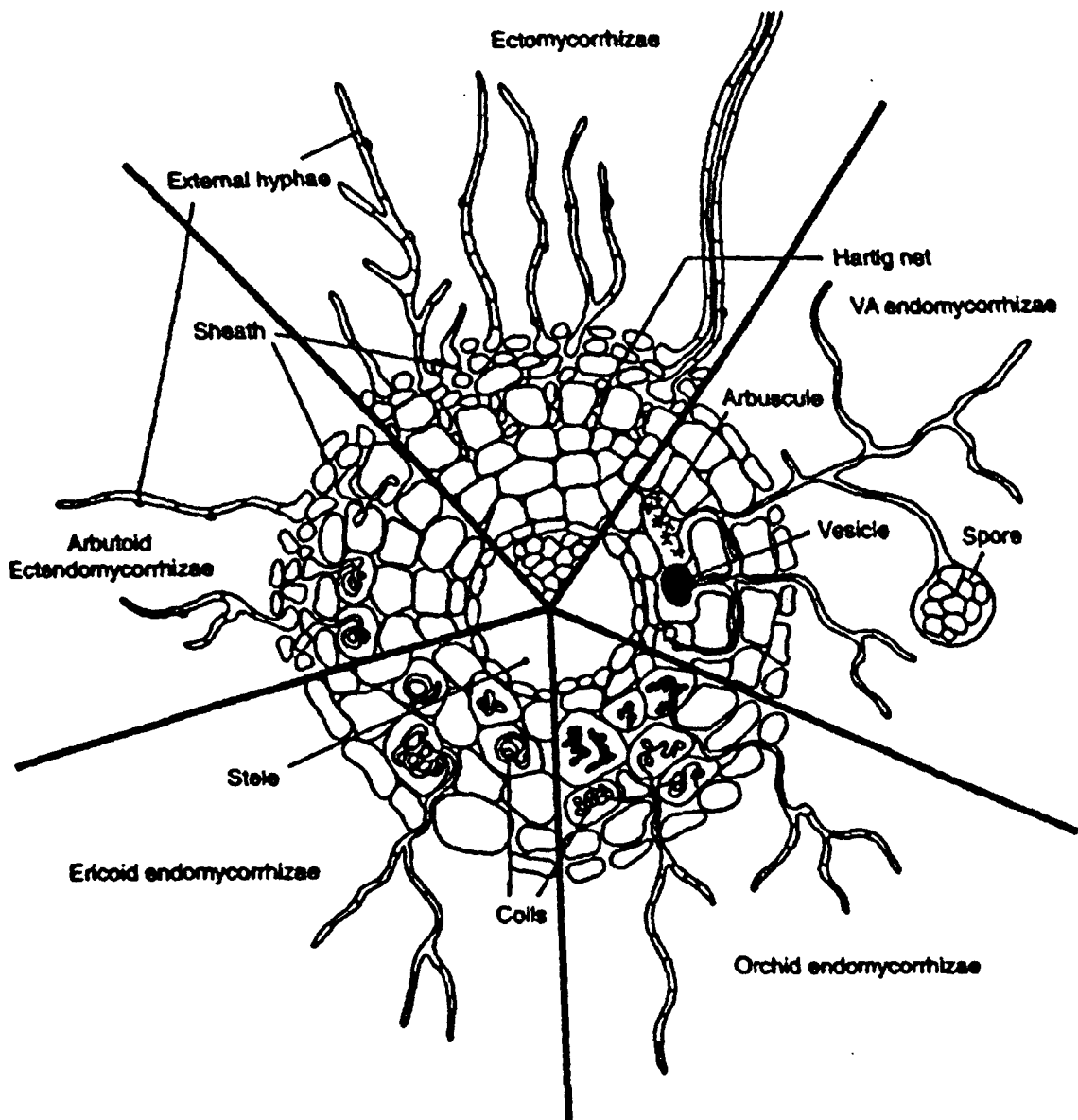


Figure 2.1 Principle structural features of five types of mycorrhiza
 Source: Selosse and Le Tacon, 1998

2.1.1 Arbuscular Endomycorrhizae

Endomycorrhizal fungi penetrate the plant cells where direct metabolic exchanges can occur and they colonise trees as well as shrubs and most herbaceous plants. Arbuscular mycorrhizal (AM) fungi are the most widespread in soils among all the other types of endomycorrhizal fungi (Raminder *et al.*, 2014). According to its official website, the product Mycogold™ that is sold in the local market of Sandakan contains a mixture of several species of AM fungi (*Acaulospora* spp., *Gigaspora* spp., *Glomus* spp., and *Scutellospora* spp.) up to 95% and another 5% of additives (sterilized sand and vermiculite) that prolongs the life of mycorrhizae in the packaging. The predominant

type of fungal infection for most agricultural crops is AM, up to 80% of plants (Raminder *et al.*, 2014).

AM was formerly known as vesicular arbuscular mycorrhizae (VAM) due to the occurrence of two structural characteristics, arbuscules and vesicles, that belongs to the family Endogonaceae (Raminder *et al.*, 2014; Priyadharsini and Muthukumar, 2015). Arbuscules are finely-branched structures formed within a cell that serves as a metabolic exchange site between plant and fungus; vesicles are sac-like structures that emerge from hyphae and acts as a storage organ for lipids. However because vesicles are absent in two of the seven genera containing these fungi, the term AM is preferred by most researchers instead of VAM.

Major effects of AM association with plants include enhanced uptake of immobile ions, nutrient cycling, rooting and plant establishment, improved soil quality and structure, plant tolerance to biotic and abiotic stresses, and increased plant community diversity (Priyadharsini and Muthukumar, 2015). They function by expanding the surface area of the root for resource exchange (Mohammadi, 2011).

2.1.2 Ericoid Endomycorrhizae

Ericoid mycorrhizal fungi forms symbiosis relationship with ericaceous plants and enable them to grow in highly pollute environments where metal ions reach toxic levels in the soil substrate (Perotto *et al.*, 2002). Recent studies are arguing that this mycorrhizal type is not a highly specific interaction as some ericoid mycorrhizal fungi are able to colonise plant from distant taxa. In addition to their general function of nutrient uptake, Perotto *et al.* (2002) reported that ericoid mycorrhizal fungi confer their host the ability to successfully compete with other plant species.

2.1.3 Orchid Endomycorrhizae

Orchid endomycorrhizae as the name suggests forms mutualistic interactions with members of the Orchidaceae, the world's largest family by growing into orchid tissues and form elaborate coiled structures known as pelotons within cortical cells (Dearnaley, 2007). Rasmussen (2002) stated that orchid mycorrhiza has an impact on plant fitness from the germination stage of growth through seedling stage, and in many cases

throughout life growth. Dearnaley (2007) also further stated that a majority of the world's orchids produce food for itself while a small number of species are myco-heterotrophic throughout their lifetime, meaning that they depend on their fungus for carbon and nutrient supply. Recent research suggests a third mode (mixotrophy) of obtaining food whereby orchids supplement their photosynthetic carbon product with carbon derived from mycorrhizal fungus (Dearnaley, 2007).

2.1.4 Ectendomycorrhizae

Ectendomycorrhiza is confined to *Pinus* and *Larix* spp. and is common in disturbed habitats as well as conifer nurseries, its occurrence is restricted and it has received little attention from the scientific community (Yu *et al.*, 2001). The unique combination of thin, fungal mantle (Hartig net) and intracellular hyphae is the characteristic of this association as they are usually well developed (Mohammadi *et al.*, 2011). These features led Egger and Fortin (1988) to suggest ectendomycorrhiza should be considered a variation of the other group of mycorrhiza, ectomycorrhiza. Another difference of ectomycorrhizae and ectendomycorrhiza is that the latter have a reduced or absent sheath.

2.1.5 Ectomycorrhizae

Ectomycorrhizal fungi are another type of mycorrhizae that is found in natural environments, however, this type is found mainly in forest ecosystems, estimated for a 30 percent of microbial biomass in temperate and boreal forests soils (Mohammadi, 2011). They are usually occurring in families of woody gymnosperms and angiosperms, namely *Pinaceae* and *Dipterocarpaceae* (Mohammadi, 2011). They may form visible structures such as mushrooms at the feet of trees they colonise (Raminder *et al.*, 2014). Raminder *et al.* (2014) also stated that this fungi grow between root cells without penetrating them, forming dense growth known as fungal mantle or sheath. Mohammadi *et al.* (2011) reviewed a different theory that this hyphae do penetrate inwards between cells of root to form an intercellular system called Hartig net, however usually they appear to have little to none intercellular penetration. The symbiotic relationships of these fungi are formed with most pines, spruces and some hardwood trees.

2.2 Mycorrhizae in Agriculture

Mycorrhizae have been a part of agriculture since the start of it, around 400 million years ago, though farmers do not realize it as they exist in soils (Lehmann, 2013; Mohammadi, 2011). The shift towards the use of microbes in Malaysia started in the late 1940s with the use of *Rhizobium*, with researches fore-fronted by the Malaysian Rubber Board (Khairuddin, 2002). It is then followed by researches on mycorrhizae in the 1980s, this time researches mainly done in Universiti Putra Malaysia (Khairuddin, 2002). A number of benefits have since been identified following mycorrhizae association with agricultural crops, and are utilized in the Malaysian agriculture field since then. The benefits include improved plant nutrition, soil fertility improvement, regulation of soil biological activity, root pest and disease control, water-use efficiency, soil structure improvement, and plant stress tolerance (Lehmann, 2013; Khairuddin, 2002).

Naher *et al.*'s (2013) study stated that mycorrhizal inoculation is generally accepted as substitute for the chemical fertilizers (especially phosphorus) to continuously enhance the productivity of maize crop. However, this seems to be questionable as mycorrhizal associations help in the uptake of nutrients, not supply them. With that being said, mycorrhizae can definitely not replace chemical fertilisers, or any sort of fertilizer at all. Furthermore, Miyasaka *et al.* (2003) stated that mycorrhizae only prove to be valuable to plants if a low but sufficient amount of P is available in the soil, and would otherwise be parasitic under extremely low nutrient contents. Following that, it would mean that mycorrhizal associations with plants in marginal lands would make the plants perform worse than without the association. Naher *et al.*'s (2013) theory seem to have missed the mark in pin pointing the function of mycorrhizae in agriculture.

Though most agricultural crops benefits from mycorrhizal associations, crops belonging to the families Amaranthaceae (spinach), Brassicaceae (cabbage), and Chenopodaceae (beet) do not form AM symbiosis (Brundrett, 2009; Hamel, 1996). Spore of AM fungi belonging to *Acaulospora*, *Entrophospora*, *Gigaspora*, *Glomus*, *Scdrocystis* and *Scutellospora* have been reported to occur in agricultural soils (Jansa *et al.*, 2002). These AM species are known for their association with crop growth and development, and among them, four of which is used in the production of MycoGold™.

The benefits of mycorrhizae association with crop may list on to a lot, however, these associations varies with a number of factors such as listed in Chapter 1 before. These factors include plant and soil.

2.2.1 Association of Mycorrhizae with Different Plant

Each species or variety of crop plants has different genotype which ultimately decides the degree of association with mycorrhizae. The Green Revolution has caused a string of plant breeding programmes to produce new and improved plant varieties that have desirable traits such as high yielding and stress tolerant. These improvements have caused a reduction in responsiveness of crop plants to mycorrhizae infection due to their changes in root structure (Lehmann, 2013). Carrenho *et al.* (2007) suggestion agrees with this as the degree of colonization for plants peanuts, sorghum and maize differs in his study was guessed to be due to the difference in root structure and development. Furthermore, as mentioned before, mycorrhizae associations occur at a minimal amount or none at all for crop families Amaranthaceae, Chenopodaceae, and Brassicaceae.

2.2.2 Association of Mycorrhizae in Different Soil

Different soil has different texture and nutrient content. All these are major factors contributing towards the degree of association of mycorrhizae with crop plants. A rather major point is the nutrient content of the soil, the nutrient in particular being N and P, which could decrease the colonization of mycorrhizae at high levels (Abdullahi and Sheriff, 2013). Low-input systems should be implemented to maximize the benefits of mycorrhizae associations. Abdullahi and Sheriff (2013) recommend reducing inputs by half of the recommended amount along with mycorrhizae inoculation to produce greatest yield.

Texture of soil also has an effect on degree of mycorrhizae colonization as reported by Carrenho *et al.* (2007) as their study showed a decrease in mycorrhizae colonization in clayey soil than in sandy soil. Carrenho *et al.* (2007) related this to the fact that clayey soil can hold nutrient ions better and hence have a higher soil nutrient

content than sandy soil. Ghorbani *et al.* (2012) relates soil structure with variation of pH, EC and available N, P, K contents.

2.2.3 Association of Mycorrhizae under Different Environmental Conditions

Some environmental conditions or cultural practice such as tilling, pesticide application, and cropping sequence are among the reasons for varying levels of mycorrhizal association (Hamel, 1996). Hamel (1996) reviewed that physical disturbances of the soil through tilling could greatly reduce mycorrhizal potential, either by damaging the structures while it is still living, or by exerting a selection pressure. A selection pressure causes maybe only one out of 3 AMF species to continue living.

Pesticide application for the removal of pests and weeds could also decrease the mycorrhizal fungi community in soil (Hamel, 1996). Some biocides function by removing competing organisms, or rather they function by directly killing any microorganisms. Herbicides on the other hand have a more indirect approach in the destruction of mycorrhizae. Herbicides, when it is a phytocide kill plants, and indirectly kill the mycorrhizae that are relying on the plant for food.

Leaving a field to rest after harrowing could reduce beneficial mycorrhizae spore count, as it may encourage saprophytic mycorrhizae (Hamel, 1996). Other than that, crop rotations with crops that do not form mycorrhizal associations like Brassicaceae would significantly reduce natural inoculum in the soil (Hamel, 1996).

2.3 Effect of Mycorrhizae Inoculation on Eggplant Growth

Mycorrhizae association with crop has reported to enhance plant nutrient acquisition like N, P, K, Mg, Cu, Ca, and Fe, improve soil quality and increase resistance to biotic and abiotic stress (Liu *et al.*, 2002; Priyadharsini and Muthukumar 2014). Mycorrhizal infection enhances plant growth by increasing water and nutrient uptake through increasing the area of root so that more nutrients can be absorbed and by mobilizing sparingly available nutrient sources especially for phosphorus (Marschner and Dell, 1994). Beneficial functions for crops that are performed by mycorrhizae are such as phosphate solubilizing bacteria (Panhwar *et al.*, 2009) and N₂ fixing bacteria (Naher *et al.*, 2009). Naher *et al.* (2013) believed that mycorrhizal colonization increases

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