

Research Paper

OPTIMIZATION OF *Eucalyptus pellita* F. Muell GROWTH USING ARBUSCULAR MYCORRHIZAL FUNGI AND COFFEE HUSK COMPOST ON SUBSOIL MEDIA

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ARTICLE HIGHLIGHTS

- Mycorrhizal fungi and coffee compost boost tree seedling growth in poor subsoil
- The method helps grow *Eucalyptus* faster for land restoration efforts.
- Reduces the need for topsoil, promoting sustainable nursery practices.
- An optimal mix of mycorrhizal fungi and compost supports robust *Eucalyptus* development.

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ABSTRACT

Arbuscular mycorrhizal fungi (AMF) and coffee husk compost are used to improve the physical and chemical conditions of the subsoil, enhancing plant growth. This study aimed to evaluate the response of plant growth and analyze the effect of AMF and coffee husk compost doses on optimal growth acceleration of *Eucalyptus pellita* seedlings in subsoil media. Seedlings were grown in subsoil with varying doses of AMF (0; 2.5; and 5 g/polybag) and coffee husk compost at different ratios (1 : 1; 2 : 1; and 3 : 1). AMF and coffee husk compost application significantly affected all parameters of seedling growth, including height, number of leaves, diameter, biomass, seedling quality index, and shoot/root ratio. Inoculation of AMF at a dose of 2.5 g/polybag and subsoil media enriched with coffee husk compost at a ratio of 2 : 1 was an efficient treatment to increase the seedling growth

Keywords: Arbuscular Mycorrhizal Fungi, biomass, coffee husk, compost, growth, subsoil



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INTRODUCTION

Deforestation and land degradation present substantial environmental issues, resulting in diminished productivity and the expansion of essential land areas. In Indonesia, a significant 14.01 million ha are designated as important land, comprising several land-use categories such as Other Use Area (APL), productive forests, protected forests, and conservation zones (Kementerian Lingkungan Hidup dan Kehutanan 2020). The urgent need for reforestation and land rehabilitation necessitates the production of high-quality seedlings on a large scale.

Eucalyptus pellita F. Muell is a rapidly growing species that is very adaptable to diverse

environmental circumstances and soil types, including those with low fertility, making it suitable for restoration efforts (Arisandi *et al.* 2019; Sulichantini 2016; Hutapea *et al.* 2023; Amezcua *et al.* 2018). Conventional nursery methods frequently utilize topsoil as the principal growth substrate. The widespread utilization of topsoil leads to horizontal land degradation, underscoring the necessity for sustainable alternatives. Subsoil, although accessible, poses difficulties due to its intrinsic constraints, such as low fertility, inadequate nutrient composition, coarse texture, and restricted biological activity, which impede proper root growth. To address these limits and guarantee successful seedling production, effective, economical, and eco-

friendly solutions are essential. The utilization of organic supplements, including compost and arbuscular mycorrhizal fungi (AMF), presents a viable approach to improve seedling development and survival in subsurface environments. Compost enhances the physical, chemical, and biological characteristics of soil, augmenting nutrient availability and promoting beneficial microbial activity (Prayudyaningsih 2016; Widuri & Ishak 2013; Paranoan *et al.* 2022). AMF, conversely, augments nutrient and water absorption, boosts drought resilience, and offers protection against pathogens and heavy metals, hence facilitating growth in poor settings (Corryanti 2000; Setiadi 2000; Setiawati *et al.* 2000; Yunita *et al.* 2023). Moreover, AMF inoculation can expedite seedling growth, resulting in earlier planting and enhanced adaptation to field conditions (Basri 2018; Dewi *et al.* 2017; Corryanti *et al.* 2007; Lumbantoruan *et al.* 2021; Yelli *et al.* 2022).

This study examined the efficacy of coffee husk compost, an accessible and sustainable organic resource, combined with AMF inoculation to enhance the growth of *E. pellita* seedlings in subsoil media. This method seeks to convert subsurface into a viable growing medium, diminishing dependence on topsoil in nursery operations and fostering sustainable land restoration. This study aimed to assess the growth response of *E. pellita* seedlings inoculated with AMF and grown on subsoil media supplemented with coffee husk compost.

MATERIALS AND METHODS

Experimental Design

A 3 x 4 factorial experiment was conducted using a Completely Randomized Design (CRD). Each treatment combination has 20 replicates, totaling 240 *Eucalyptus pellita* seedlings. The factors were: A) Arbuscular Mycorrhizal Fungi (AMF) inoculation dose (A0 = 0 g; A1 = 2.5 g; A2 = 5 g) and B) Subsoil: coffee husk compost ratios (V : V) (B0 = 1 : 0; B1 = 1 : 1; B2 = 2 : 1; B3 = 3 : 1). The applied treatment combinations were: 1) A0B0 = no AMF inoculation, the planting media was just subsoil, no coffee husk compost; 2) A0B1 = no AMF inoculation, the planting media was subsoil : coffee husk compost (1 : 1); 3) A0B2 = no AMF inoculation, and the planting media was subsoil : coffee husk compost (2 : 1); 4) A0B3 = no AMF inoculation, and the planting media was subsoil : coffee husk compost (3 : 1); 5) A1B0 =

2.5 g AMF inoculation, and the planting media was only subsoil without coffee husk compost; 6) A1B1 = 2.5 g AMF inoculation, and the planting media was subsoil : coffee husk compost (1 : 1); 7) A1B2 = 2.5 g AMF inoculation, and the planting media was subsoil : coffee husk compost (2 : 1); 8) A1B3 = 2.5 g AMF inoculation, and the planting media was subsoil : coffee husk compost (3 : 1); 9) A2B0 = 5 g AMF inoculation, and the planting media was only subsoil without coffee husk compost; 10) A2B1 = 5 g AMF inoculation, and the planting media was subsoil : coffee husk compost (1 : 1); 11) A2B2 = 5 g AMF inoculation, and the planting media was subsoil : coffee husk compost (2 : 1); 12) A2B3 = 5 g AMF inoculation, and the planting media was subsoil : coffee husk compost (3 : 1).

Experimental Procedure

Coffee husk compost was prepared using a modified method of Riga *et al.* (2022), involving a 60-day composting process with EM-4 bioactivator and cow manure. *E. pellita* seeds were germinated in sterilized fine sand. Subsoil was sterilized with fumigan containing 98% Dazomet before mixing with compost. Polybags (12 x 17 cm) were filled with the respective subsoil : compost mixtures. Three-week-old seedlings were transplanted and inoculated with AMF during transplanting. Seedlings were maintained in a greenhouse with twice-daily watering.

Observed Variables

Seedlings' height, stem diameter, and leaf number were measured monthly for three months. At three months, biomass (dry weight), seedling quality index (Dickson *et al.* 1960), and AMF root colonization percentage (Kormanik & McGraw 1982; Mosse 1980) were determined. Biomass and AMF colonization were assessed on 36 randomly selected seedlings. Chemical properties of the subsoil were analyzed to examine the nutrient availability, as well as the nutrient content of coffee husk compost.

Data Analysis

Data were analyzed using Analysis of Variance (ANOVA), and significant differences were further analyzed using Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Subsoil Media and Coffee Husk Compost

Our study indicated that subsoil had deficiencies in terms of macronutrient content which was below the quality standards of fertility. The levels of N, P, and K nutrients were very low, measured at 0.06%, 8.45%, and 0.14%, respectively (Table 1). Soil with very low macronutrient content required additional fertilization as an intervention. Rosniawaty *et al.* (2020) explained that subsoil had a low level of fertility, emphasizing the need for improvement by adding organic materials in solid and liquid form. These measures were expected to improve the physical, chemical, and biological properties of the soil. Fertilization with nitrogen, phosphorus, and potassium, is essential to support healthy plant growth.

Based on nutrient content analysis, robusta coffee husk compost prepared with EM4 bioactivator met the quality standards outlined in SNI 19-7030-2004, Regulation of the Minister of Agriculture Number 28/Permentan/SR.130/5/2009 and Number 70/Permentan/SR.140/10/2011. These standards include quality criteria, such as water, organic matter, and essential nutrient contents (N, P, K), which are important indicators in ensuring the effectiveness of compost as a source of plant nutrients.

EM4, a bioactivator used for producing solid compost, often called bokashi, offers distinct advantages. In addition to accelerating the composting process, it has been proven to eliminate odors associated with composting when the process was conducted approximately (Wellang *et al.* 2015).

Compost made from coffee husk has numerous advantages for enhancing the biological, chemical, and physical characteristics of soil. The study of nutrient composition in coffee husk compost revealed a comparatively high N content of 1.22%, a moderate P value of 1.36%, and a comparatively high K level of 1.73%. These findings are consistent with the assertion of Simarmata (2016) that coffee husk compost includes nitrogen (N) and potassium (K) in addition to the organic components' cellulose, pectin, hemicellulose, and lignin. Coffee husk waste can improve soil structure since it contains 45.3% organic material, 2.98% nitrogen, 0.18% phosphorus, and 2.26% potassium, according to Efendi (2017). Additionally, coffee husk compost contains carbon, nitrogen, phosphate, and potassium (Novita *et al.* 2018).

Eucalyptus seedlings require a sufficient amount of nutrients to grow. Lingga *et al.* (2013) found that the presence of essential nutrients like nitrogen, phosphorus, and potassium is closely related to plant growth. Nitrogen is the primary ingredient in amino acids, proteins, and the formation of cell protoplasm, which can stimulate plant growth. Furthermore, potassium functions as an activator of several enzymes involved in the production of protein and carbohydrates. Plants will use the photosynthate that they have created to elongate and divide their cells, which will increase their height. Gardner *et al.* (1991) emphasized that the main factors influencing plant height are cell growth and proliferation.

Table 1 Nutrient content in subsoil and coffee husk compost

No.	Variable	Sample code			Quality compost criteria	
		SS	CH	SNI 19-7030-2004	Regulation of the Minister of Agriculture No. 28/Permentan/SR.130/5/2009	Regulation of the Minister of Agriculture No. 70/ Permentan/SR.140/10/2011
1	pH (H ₂ O)	5.65 SA	6.55	6.8 - 7.49	4 - 8	4 - 9
2	C	0.85 VL	15.10	9.8 - 32	≥ 12	≥ 15
3	N	0.06 VL	0.86	≥ 0.4	< 6	-
4	P	8.45 VL	0.79	≥ 0.1	< 6	-
5	K	0.14 VL	1.18	≥ 0.2	< 6	-
6	C/N	11 M	18	10 - 20	15 - 25	15 - 25

Notes: SS = subsoil; CH = coffee husk compost; VL = Very Low; SA = Slightly Acid; M = Medium; Land value classification based on Hardjowigeno (2010); Sutarmanto and Miftakhurrohmat (2019).

Growth of Plant Height

The height increase of *E. pellita* seedlings was monitored biweekly over 3 month-period. The results showed that AMF inoculation isolated from the soil of a former limestone, comprising *Gigaspora* sp. and *Acaulospora* sp., had a better height increase than that in the control. At a 2-week period no significant differences were observed between AMF-inoculated seedlings and the control. The effects of the AMF inoculation became apparent after 2 weeks (Fig. 1).

Prayudyarningsih (2014) investigated the effect of AMF inoculation on the growth of *Acacia auriculiformis* seedlings. The results showed that inoculation with *Glomus* sp., the non-indigen (positive control), gave the best growth performance, achieving an average height of 22.12 cm. The growth of *E. pellita* seedlings was significantly affected by the combination of AMF inoculation and coffee husk compost application. The interaction of treatments showed differences in response to the increase in seedling height. Furthermore, the Duncan test showed that A1B2 treatment was the most effective. This treatment produced the highest average height growth of *E. pellita* seedlings (Table 2).

Adding both AMF and compost to the growing media accelerates the growth of *E. pellita* plants in comparison to the control treatment (Table 2). The application of coffee husk compost and subsoil medium (2 : 1) with 2.5 g of AMF increased the average plant height by 73.25% above the control. In the meantime, the average plant height increased by 63.70% when 2.5 g of AMF

were added to subsoil medium and coffee husk compost (1 : 1). Compost and AMF work together to improve soil porosity, enrich nutrients in the media, and enhance the absorption of water and nutrients, particularly phosphorus. This finding is similar to the work of Nursayuti (2017), who found that the type of planting media, specifically the amount of nutrients in the soil that plants can absorb, significantly impacts growth. Compost and AMF work well together to improve nutrient absorption, particularly for nitrogen, which is crucial for plants' generative growth (Ariandi & Sudantha, 2018). Given the availability of minerals in the soil, adding AMF and compost to less fertile soil will boost plant growth and yield (Wahab *et al.* 2023).

Compared to the control, the average plant height increased by 52.01% when AMF was added to subsoil planting media without compost. Inoculating plant roots with AMF is one method of promoting plant growth (Setiadi 2000). In order to promote greater leaf growth, which is directly linked to increased plant height, AMF helps plants absorb nitrogen (Husna *et al.* 2015). It is well known that the presence of AMF in plant roots can improve nutrient and water absorption. The external hyphae of AMF improve the range of nutrients that can be absorbed and offers a more effective surface (wider and better spread) for absorbing nutrients from the soil, which increases nutrients. The surface area of plant roots that absorb AMF in symbiosis is 18 times larger than that of roots that do not have mycorrhizae (Prayudyarningsih 2014; Prayudyarningsih & Sari 2016).

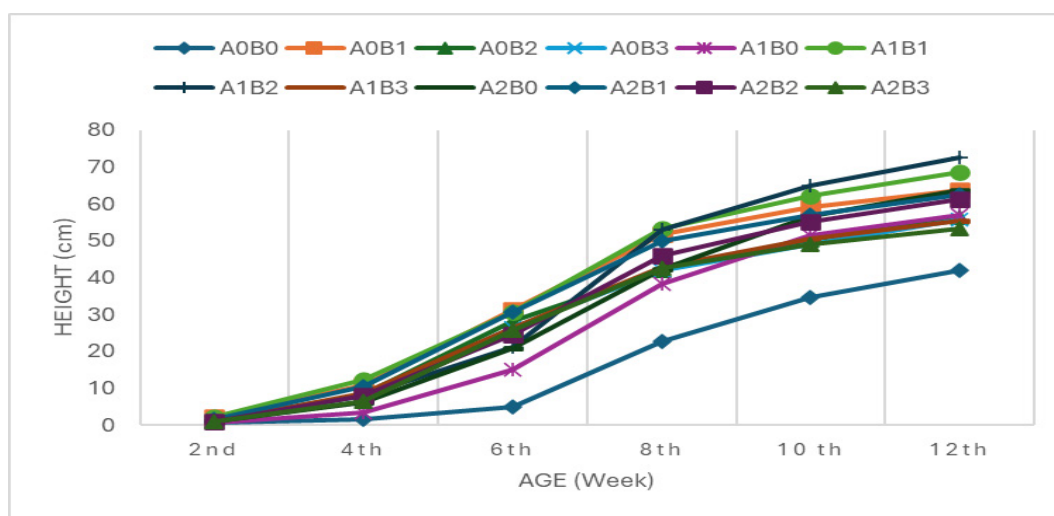


Figure 1 Effect of AMF inoculation and coffee husk compost application on the height growth of *Eucalyptus pellita* seedlings up to 12 weeks of age

Table 2 Effect of AMF inoculation and coffee husk compost application on the height of *Eucalyptus pellita* seedlings up to 12 weeks of age

No	Treatment	Average height growth (cm)
1	A1B2	72.61a
2	A1B1	68.61a
3	A2B0	63.71b
4	A0B1	63.71b
5	A2B1	62.38b
6	A2B2	61.13bc
7	A1B0	56.81cd
8	A0B2	56.30d
9	A0B3	55.78d
10	A1B3	55.35d
11	A2B3	53.12d
12	A0B0	41.91e

Notes: Numbers followed by the same letter are not significantly different at the 95% confidence level.

Stem Diameter

AMF inoculation and coffee husk compost application which was observed every 4 weeks showed an increase in the diameter of seedlings, which was a positive effect than that of the control. However, after 12 weeks, the increase in the diameter due to AMF inoculation was very different from the control. The most pronounced growth during this period was observed in the A2B0 treatment (Fig. 2).

The treatment combinations that produced the largest diameter increases were A2B0, A0B2, A2B3, A1B1, A0B1, A1B3, and A2B1 (Table 3). The highest AMF inoculation dose of 5 g with a

1 : 1 dose of coffee husk compost was particularly effective. Similarly, the study of Putra (2015) reported significant results with a 15 g AMF inoculation on Tanjung and Glodokan plants, achieving diameter increase of 0.158 cm and 0.165 cm, respectively.

Based on the Analysis of Variance, there was a significant difference in the response to the increase in diameter of *E. pellita* due to the interaction of the inoculation treatment of AMF with various doses of coffee husk compost. The Duncan test identified A2B0 as the most effective treatment, achieving the highest average diameter increase (Table 3).

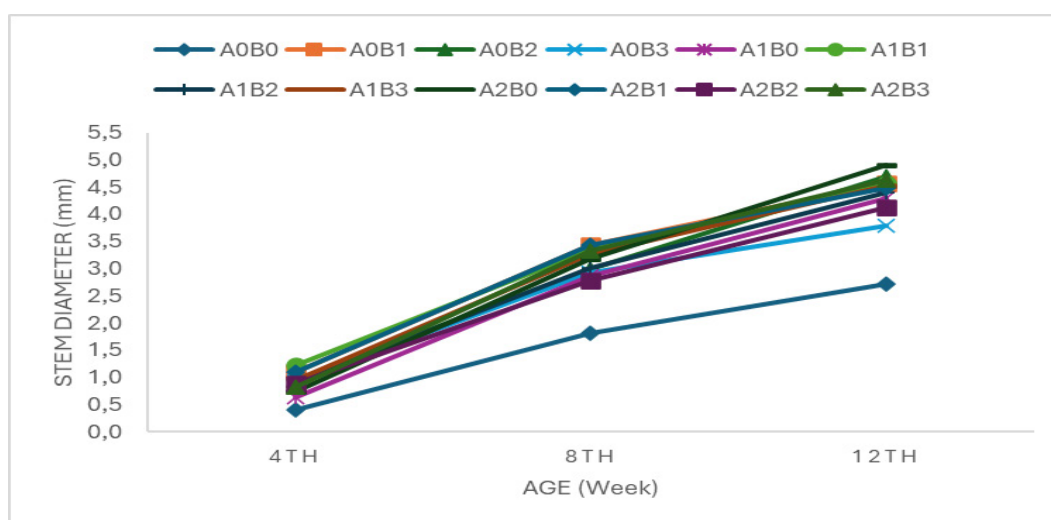


Figure 2 Effect of AMF inoculation and coffee husk compost application on the diameter of *Eucalyptus pellita* seedlings up to 12 weeks of age.

Table 3 Effect of AMF inoculation and coffee husk compost application on diameter increase of *Eucalyptus pellita* seedlings

No	Treatment	Average diameter increase (mm)
1	A2B0	4.89a
2	A0B2	4.68ab
3	A2B3	4.63ab
4	A1B1	4.57ab
5	A0B1	4.56abc
6	A1B3	4.52abc
7	A2B1	4.48abc
8	A1B2	4.40bc
9	A1B0	4.28bc
10	A2B2	4.13cd
11	A0B3	3.79d
12	A0B0	2.72e

Notes: Numbers followed by the same letter are not significantly different at the 95% confidence level.

Number of Leaves

Observations conducted every 2 weeks showed that *E. pellita* seedlings inoculated with AMF and coffee husk compost application had a greater leaf production compared to the control. The best result in the 12th week was achieved from the A1B1 treatment, producing 34 leaves (Fig. 3).

Analysis of variance showed that treatment interactions influenced leaf production. The 2.5 g AMF inoculation combined with subsoil media and coffee husk compost at a ratio of 1 : 1 (A1B1) significantly increased the number of leaves, outperforming the 5 g AMF inoculation (Table 4).

Treatment combination that produced the highest number of leaves was A1B1 (33.80 leaves). However, no significant differences were observed between A1B2 (32.40 leaves) and A0B1 (32.10 leaves). Duncan's test results showed that all treatments with AMF inoculation and coffee husk compost application enhanced plant growth compared to those without these additions. Those combinations improved overall plant biomass, including leaf production, by optimizing nutrient availability and enhancing root enzyme activity for nutrient absorption (Diagne *et al.* 2020).

Leaves significantly influence the growth and development of plants. Generally, leaves are essential organs that sustain several critical physiological processes to plant growth and survival. Leaf growth is influenced by the availability of nutrients

like nitrogen (Firmansyah & Sumarni 2013). An average of 33.80 and 32.40 strands of leaves were produced by adding 2.5 g of AMF isolates and coffee husk compost in subsoil media at ratios of 1 : 1 and 1 : 2. This demonstrates how using AMF and coffee husk compost can greatly boost plant leaf growth (Table 6). Compost not only provides the nutrients that plants require, but it also fosters the growth of microbial colonies, including AMF, by offering a favorable environment and a wealth of food sources.

Plant roots can absorb more nutrients, including nitrogen (N), with AMF inoculation (Xie *et al.* 2014), because AMF and plant roots can develop a symbiotic interaction that improves nutrient absorption. AMF promotes plant growth by increasing the absorption of nutrients from compost. *Eucalyptus* seedlings may produce more leaves when they are inoculated with AMF and placed in media that contains compost. When it comes to boosting leaf growth, 2.5 g of AMF is more effective than 5 g. The ideal mycorrhizal colony density, which is adequate to establish a productive symbiotic connection with plant roots, may be achieved by 2.5 g AMF inoculation. Adequate nitrogen requirements will promote the growth of vegetative tissues, including leaves, increasing the quantity of leaves (Fitriana *et al.* 2012). The ability for photosynthesis will rise with more leaves, increasing the amount of photosynthate produced and distributing it throughout the plant (Prayudyaningsih & Sari 2016).

The availability of certain nutrients that promote plant growth and development is increased by mycorrhizal inoculation. Mycorrhizal plants are better at absorbing water, which also allow them to absorb soluble nutrients like sulfur (S), potassium (K), and nitrogen (N), increasing the absorption of these components (Baon 1999). As one of the primary constituents of many different compounds found in plant tissue, nitrogen is also crucial for

developing the vegetative organ. AMF inoculation, however, is more effective in increasing the growth of *Eucalyptus* seedlings when it is coupled with the application of coffee husk compost fertilizer, as seen by a 30.50% increase in number of leaves as compared to the control (Table 4). This suggests that the coffee husk compost fertilizer and AMF treatment work well together.

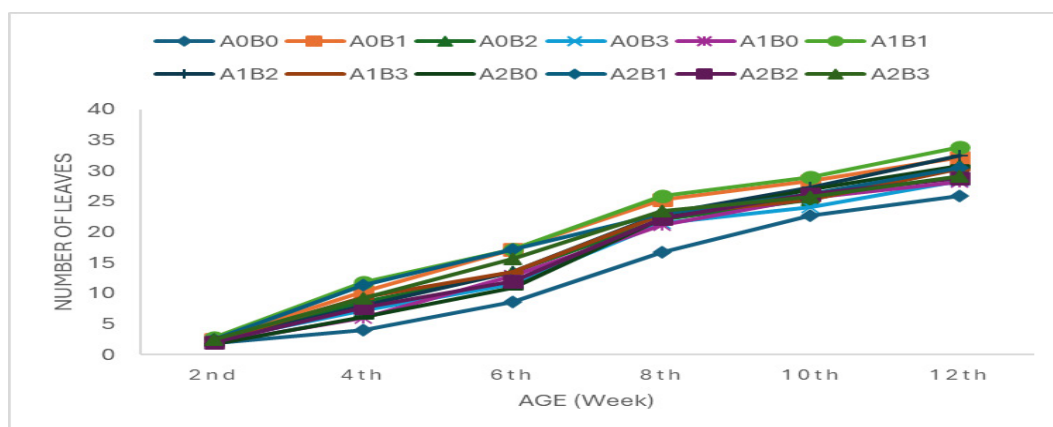


Figure 3 Effect of AMF inoculation and coffee husk compost application on the increase in the number of leaves of *Eucalyptus pellita* seedlings up to 12 weeks of age.

Table 4 Effect of interaction between AMF inoculation and coffee husk compost application on the increase in the number of leaves of *Eucalyptus pellita* seedlings up to 12 weeks of age

No	Treatment	Average number of leaves
1	A1B1	33.80a
2	A1B2	32.40ab
3	A0B1	32.10ab
4	A0B2	30.75bc
5	A2B0	30.70bc
6	A2B1	30.40bcd
7	A1B3	30.30bcd
8	A2B3	29.05cde
9	A2B2	28.70cde
10	A0B3	28.30de
11	A1B0	28.10e
12	A0B0	25.90f

Notes: Numbers followed by the same letter are not significantly different at the 95% confidence level.

Shoot-Root Ratio

The shoot-root ratio is among the criteria for assessing seedling quality. It is influenced by the interactions between AMF doses and coffee husk compost application. Seedlings treated with 2.5 g AMF inoculation and 1 : 1 coffee husk compost application showed the highest shoot-root ratio value and were better than the control (Table 5).

Further test on the positive interaction between AMF inoculation and coffee husk compost application showed that the highest average shoot-root ratio was observed in A1B1 (5.40), which was significantly different from A1B2 (4.34), A0B3 (4.03), and A2B2 (4.01).

The shoot-root ratio and seedling quality index serve as markers for assessing the quality of seedlings (Prayudyaningsih & Sari 2016). Before seedlings are planted in the field, the shoot-root ratio and seedling quality index must be taken into consideration because those are crucial indicators of the seedlings readiness to be planted. Both indicators offer details on effective field growth as well as seedling quality and transplantation readiness. This study showed that when AMF inoculation and compost application are combined, the *Eucalyptus* seedlings have a seedling quality index greater than 0.09 and a shoot-root ratio between 2 and 5, which is typically seen in seedlings with the best development and survival potential.

Table 5 The interaction of AMF inoculation and coffee husk compost application on shoot-root ratio of *Eucalyptus pellita* seedlings up to 12 weeks of age.

No	Treatment	Shoot-root ratio
1	A1B1	5.40a
2	A1B2	4.34ab
3	A0B3	4.03abc
4	A2B2	4.01abc
5	A2B1	3.69bc
6	A2B0	3.41bc
7	A2B3	3.28bc
8	A0B2	3.18bc
9	A1B3	3.18bc
10	A1B0	2.95 bc
11	A0B1	2.61 bc
12	A0B0	2.47 c

Notes: Numbers followed by the same letter are not significantly different at the 95% confidence level.

Table 6 Effect of AMF inoculation and coffee husk compost application on the seedling quality index of *Eucalyptus pellita* seedlings up to 12 weeks of age

No	Treatment	Seedling quality index average
1	A2B1	0.51a
2	A0B2	0.49ab
3	A2B3	0.46ab
4	A2B0	0.426abc
5	A1B3	0.413abc
6	A1B1	0.393bcd
7	A1B0	0.383bcde
8	A0B1	0.34cde
9	A1B2	0.316cde
10	A0B3	0.293de
11	A2B2	0.276ef
12	A0B0	0.18f

Notes: Numbers followed by the same letter are not significantly different at the 95% confidence level.

Seedling Quality Index

The seedling quality index is a comparison between the total dry weight with the seedling strength, and the shoot-root ratio, which serves as a parameter for describing the morphological and physiological characteristics of seedlings. Analysis of Variance showed that treatment interactions affected the seed quality index, signifying differences in response (Table 6).

Based on the results of further tests using the Duncan method, AMF inoculation and coffee husk compost application showed different response to the seedling quality index. The highest seedling quality index was observed in A2B1 (0.51), while the lowest was recorded in A0B0 (0.18). This signified a positive interaction between AMF and coffee husk compost.

An indication of seedling quality is the seedling quality index value (Prayudyaningsih & Sari 2016), which is an essential parameter for evaluating seedling quality and preparedness for transplantation and field growth. AMF inoculation and compost application together yield a seedling quality index of greater than 0.09 (Table 6). Meanwhile, Orpa *et al.* (2019) explained that seedlings with a seedling quality index greater than 0.09 are prepared for field transplantation. Therefore, a certain dose of AMF and compost inoculation together can enhance seedling quality. The high-quality seedlings are very viable in the field and exhibit a balanced ratio between the transpiration process and the area of photosynthesis of plants that can absorb water and minerals.

Regarding Technical Guidelines for evaluating the quality of forest plant seedlings, the Directorate of Land Rehabilitation and Social Forestry's Regulation Number: P.05/V-SET/2009 is the basis for this assessment. *E. pellita* F. Muell seedlings must meet the following standard requirements: height > 20 cm, diameter > 2 mm, number of leaves/LCR > 3 pairs, and age > 3 months. According to the standard for maximizing nutrient absorption and boosting growth efficiency, the use of coffee husk compost in conjunction with AMF inoculation can greatly shorten the nursery period. As a result, this combination of treatments increases production efficiency and prospective crop yields in the field by enabling seedlings to reach the ready-to-plant stage faster. According to these guidelines, *Eucalyptus*

seedlings can have their nursery period shortened by one month when AMF inoculation and coffee husk compost application are used together.

Biomass

Biomass is the dry weight of root, stem, and leaf samples after oven drying. Analysis of Variance showed different biomass responses based on AMF and coffee husk compost doses. Duncan's test results showed that the highest biomass value of 8.45 was shown by the A1B2 treatment, comprising 2.5 g of AMF inoculation and a 2 : 1 coffee husk compost ratio. In contrast, the lowest at 3.59 was recorded for the control treatment, A0B0 (Table 7).

Biomass indicates the nutritional state of plants, the rate of photosynthesis, and plant respiration, which can be used as an indicator to assess the quality of plant development (Prayudyaningsih & Sari 2016). Plant development is boosted when photosynthesis is functioning properly, as seen by an increase in plant dry weight (biomass). This can happen as a result of the improved condition of the planting media due to the addition of compost, which raises the nutrient content of soil, and AMF inoculation, which facilitates nutrient absorption.

Percentage of AMF Colonization

The level of mycorrhizal colonization is an indicator of the success of mycorrhizal development in the roots and the rhizosphere. This colonization reflects how well AMF has succeeded in establishing a symbiotic relationship with the plant, the success of which can be demonstrated through increased plant growth and resistance to various environmental conditions. Our study showed that the interaction between AMF inoculation and coffee husk compost application did not have a significant effect on the percentage of AMF colonization.

In general, there were variations in the percentage of AMF colonization in each treatment (Fig. 7). Plants inoculated with 5 g of AMF in subsoil media using coffee husk compost application with a ratio of 2 : 1 (A2B2) had the highest colonization percentage, with a percentage exceeding 50%. This shows that the combination of treatments supports AMF colonization well. When mycorrhiza is absent in subsoil media (A0B0), a lower colonization percentage of less than 10% was observed.

Studies showed that AMF inoculation coupled with organic materials application, such as vermicompost, can increase colonization and nutrient uptake. In ginger plants, AMF inoculation elevated the uptake of phosphorus and other nutrients, as well as increased root development

(Sarathambal *et al.* 2024). In addition, studies on tea plants showed that the combination of AMF and plant hormones also increased the percentage of colonization and overall growth (Gao *et al.* 2023).

Table 7 Effect of AMF inoculation and coffee husk compost application on the biomass of *Eucalyptus pellita* seedlings up to 12 weeks of age.

No	Treatment	Seedling biomass (g)
1	A1B2	8.45a
2	A1B1	8.26ab
3	A0B2	7.86abc
4	A2B0	7.05abcd
5	A1B2	7.03abcd
6	A2B3	6.77bcd
7	A1B0	6.52cde
8	A1B3	6.46cde
9	A0B3	5.65de
10	A0B1	5.65de
11	A2B2	4.97ef
12	A0B0	3.59f

Notes: Numbers followed by the same letter are not significantly different at the 95% confidence level.

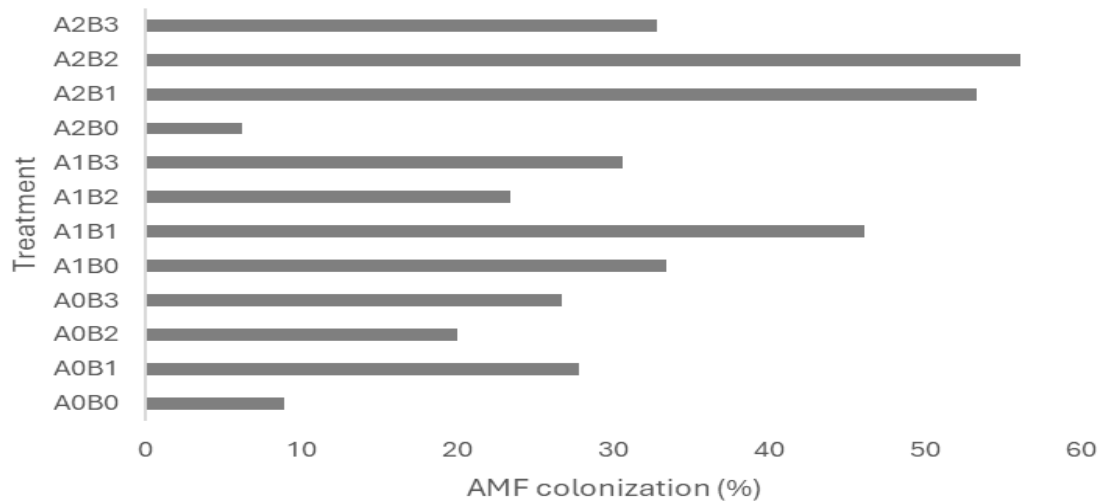


Figure 4 Percentage of AMF colonization in *Eucalyptus pellita* seedlings up to 12 weeks of age.

CONCLUSION

The application of AMF had a very significant effect on the growth of *Eucalyptus* seedlings. Coffee husk compost significantly influenced all growth parameter, including plant height, number of leaves, diameters, and seedling biomass, as well as seedling quality index and the shoot-root ratio. Inoculation of 2.5 g AMF with planting media consisted of subsoil and coffee husk compost at a ratio of 2 : 1 showed the best treatment to increase the growth of *Eucalyptus* seedlings.

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