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Arbuscular mycorrhizal fungi to enhance the growth of tropical endangered species *Pterocarpus indicus* and *Pericopsis mooniana* in post gold mine field in Southeast Sulawesi, Indonesia

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Abstract. *Husna, Tuheteru FD, Arif A. 2021. Arbuscular mycorrhizal fungi to enhance the growth of tropical endangered species* Pterocarpus indicus *and* Pericopsis mooniana *in post gold mine field in Southeast Sulawesi, Indonesia. Biodiversitas 22: 3844-3853.* Gold mining activities contribute to the national economy, but have a serious impact on forest and environmental degradation and pose a threat to tree species in the tropics. Reforestation of post-gold mining with tropical legume tree species is threatened with extinction. Arbuscular mycorrhizal fungi are effective in conservation of endangered plants and restoration of degraded land. The objective of the study was to evaluate the effect of native AM Fungi inoculation on the growth of *Pterocarpus indicus* and *Pericopsis mooniana* in nurseries and post-gold mining fields. *P. indicus* and *P. mooniana* seedlings were inoculated with *Glomus claroideum* Schenk & Smith, *Glomus coronatum* Giovann., and mixed AMF (*G. claroideum*, *G. coronatum*). Uninoculated seedlings were used as control treatment, and they were maintained for 4 months under greenhouse conditions. After 4 months in greenhouse, seedlings were transferred to post-gold mine and planted for 4 months. The percentage of AMF colonization, plant growth, nutrient content and uptake of N, P, K, Fe, Mn were measured after 4 months both in the greenhouse and the field. The percentage of AMF colonization under greenhouse conditions in *P. indicus* and *P. mooniana* ranged 5.67-75.3% and 2.2-41.2%. All AMF colonization tended to have higher shoot height, leaf numbers and nodules, plant dry weight and N, P, K content under greenhouse conditions. Shoot height, stem diameter, leaf dry weight, N, P, K, Mn and Fe under field conditions had higher inoculated seedlings than control four months after planting. AMF could be used to conserve endangered tree species in post-gold mining reforestation in the tropics.

Keywords: Arbuscular mycorrhizal fungus, gold mining land, Indonesia, restoration, tropical trees species

INTRODUCTION

Deforestation and forest degradation, especially due to mining and other extractive activities, have serious impacts on biodiversity loss, especially in terms of threats to tropical tree species. Therefore, post-mining land restoration is needed to restore the richness of plant species and the function of forest ecosystems. Ecological restoration and conservation are the central themes of global environmental policy (FAO-UNEP 2020). Global experiences show that projects to restore degraded land and forests have been successful or even failed (Thomas et al. 2014). Post-mining land has physical, chemical and biological characteristics that do not support vegetation growth and development (Sheoran et al. 2010). Therefore, designing an appropriate restoration mechanism is very important. To support the program and stimulate plant growth, it is necessary to provide seeds with beneficial soil microbes, namely the Arbuscular mycorrhizal fungi (Asmelash et al. 2016). Arbuscular mycorrhizal fungi (AMF) are obligate fungi in symbiosis with 70-90 terrestrial plant families (Smith and Read 2008; Tuheteru et al. 2019), including legumes and endangered species and aquatic plants (Tuheteru and Wu 2017).

Adaptation and the success of seedlings in the field are vital components in restoration, reforestation and species conservation programs. Therefore, improving the quality of forestry plant seedlings by applying mycorrhizal fungi in the nursery is an important stage in ecosystem restoration activities (Urgiles et al. 2009). AMF increases the growth of various tropical tree species at the nursery scale (Turjaman et al. 2007; 2009; Wulandari et al. 2014; Husna et al. 2016; 2019a; 2021; Maulana et al. 2017; Prematuri et al. 2020). AMF has the potential to be developed for revegetation, reforestation and restoration and phytoremediation (Asmelash et al. 2016; Tuheteru et al. 2016; Husna et al. 2017; Wang et al. 2017; Dhalaria et al. 2020; Riaz et al. 2021). Several studies reported that the presence of AMF can accelerate plant growth in the land of various post-mining conditions in the world, including nickel mining (Husna et al. 2016; Amir et al. 2019), coal (Wulandari et al. 2016; Agus et al. 2018; Husna et al. 2019a), and gold (Husna et al. 2019b; 2021).

AMF is very important for the conservation programs of endangered plant species (Fuchs and Haselwandter 2008; Panwar and Tarafdar 2006; Zubek et al. 2009; Bothe et al. 2010; Husna et al. 2017; 2018). In Indonesia, the AMF application for the conservation of endangered species has been reported in species of *Aquilaria malacensis* and *A. crasna* (Turjaman et al. 2006a), *A. filaria* (Turjaman et al. 2006b) and *Gonystylus bancanus* (Muin 2003), *Kalapia celebica* (Husna et al. 2019b; 2021) and *Pericopsis mooniana* (Husna et al. 2016; 2019a). Post-mining land restoration and tree species conservation with AMF application improve nutrient and water uptake, increase plant resistance to biotic and abiotic stresses, so that it can support plant growth and development on a greenhouse or field scale.

Pterocarpus indicus and *P. mooniana* are tropical tree species, belongs to Leguminosae. They produce luxury wood (Soerianegara and Lemmens 1994). Both species are classified as vulnerable by IUCN and trade-in of the species is restricted by listing in Appendix 2 of CITES now. They are potential species for reforestation and forest plantation programs (Gazal et al. 2004; Thomson 2006; Husna et al. 2018). *P. indicus* can be found in lowland evergreen primary and some secondary forests from near sea level to 1,300 m elevation, grows best in full sunshine and adapts to a wide range of soil types (Thomson 2006; Manipol et al. 2020). *P. mooniana* has distribution in lowland evergreen primary (200-350 m elevation) and grows in relatively infertile sandy regosol (Soerianegara and Lemmens 1994).

Previous studies showed that native AMF improves plant growth and plant nutrient uptake of *P. mooniana* on post-mining soils of serpentine and nickel (Husna et al. 2016; 2018), inceptisol (Husna et al. 2015a) and overburdened coal mining land (Husna et al. 2019a). However, the study on tailings media and gold post mining land is still limited. We strongly believe that inoculation of native AMF enhances plant growth and nutrient uptake of *P. indicus* and *P. mooniana* as two threatened tropical tree species. The purpose of this study was to examine the effects of native AMF inoculation on the growth and nutrient uptake of *P. indicus* and *P. mooniana* under greenhouse and gold post mining land.

MATERIALS AND METHODS

Soil preparation

Gold tailing soil was collected on 1-2 March 2019 from the disposal site of PT. Panca Logam Makmur, Bombana District, Southeast Sulawesi, Indonesia and stored in a greenhouse. Chemical and physical properties of gold tailing soil were analyzed at the Soil and Plant Laboratory of SEAMEO BIOTROP, Bogor, Indonesia (Table 1). Gold tailing soil mixed with ultisol soil and cow manure compost in a mixture ratio of 4:2:1 (v/v/v). Chemical characteristics of cow manure compost were as follows: pH (H_2O): 7.75; total N: 7.9 g kg^{-1}; total K: 4.2 g kg^{-1} and total: P,1.92 g kg^{-1}.

Arbuscular mycorrhizal fungal inoculum and inoculation

The AMF inoculums used were *Glomus claroideum* Schenk & Smith (B), *Glomus coronatum* Giovann. (C), and mixed AMF isolated from the rhizosphere of *Pericopsis mooniana* (Husna et al. 2015b). AMF inoculums were propagated in zeolite media and *Pueraria javanica* host for 3 months in the greenhouse of the Indonesian Mycorrhizas Association, Southeast Sulawesi Branch, Kendari. Polyethylene pots (15x20 cm) were filled with 1000 g of sterilized gold tailing soil. AMF inoculation was obtained by placing ten grams (10 g) of inoculum of each species at 1-3 cm below the seedlings. Two new leaves of *P. indicus* and *P. mooniana* seedling were transplanted into the pots. The inoculated seedlings were maintained, watered, and observed for 4 months. Seedlings were watered daily with tap water to field capacity. Daily temperatures vary between 27 and 47^oC with relative humidity of 45–79%. Ten g of sterilized zeolite were placed into no-inoculated pots as a control treatment.

Table 1. Physical and chemical properties of gold tailing and post gold mining soil

Parameter	Unit	Gold tailing media		Post gold mining land	
		Value	Criteria[a]	Value	Criteria[a]
pH H_2O		6.1	low	6.9	Netral
C-organic	%	0.10	Very low	0.10	Very low
N Total	%	0.05	Very low	0.07	Very low
C/N ratio		2.1	Very low	1	Very low
P_2O_5	mg/kg	11.5	Medium	25.5	High
Ca	cmol/kg	1.45	Very low	2.13	Low
Mg	cmol/kg	0.73	Low	0.35	Low
Na	cmol/kg	0.13	Low	0.33	Low
K	cmol/kg	0.09	Very low	0.38	Low
CEC	cmol/kg	4.77	Very low	27.30	High
Base saturation	%	50.36	Medium	11.68	Very low
Al^{3+}Hdd	me/100 g	0.10	Very low	0.11	Very low
Texture					
Sand	%	79.2	Loamy Sand	42.2	Clay Loam
Silt	%	16.2		29.5	
Clay	%	4.6		28.3	
Mn Total	mg/kg	487	Very high	939.8	Very high
Fe2O3	%	0.73	Very high	4.61	Very high
Cd Total	mg/kg	0.6	Normal	0.51	Normal
Pb Total	mg/kg	9.8	Normal	16.3	Normal
Hg	mg/kg	<0.00004	Normal	-	

Note: [a]Balai Penelitian Tanah (Soil Research Center) (2009)

Seedling preparation

P. indicus and *P. mooniana* fruits were collected from trees growing on the Halu Oleo University Campus in Kendari, Southeast Sulawesi. These fruits are orthodox. The seeds are soaked in hot water with an initial temperature of 50ºC until cold for 24 hours and then germinated in plastic sprouts at the Plastic House of the Indonesian Mycorrhizal Association (IMA), Southeast Sulawesi Branch (03º57'55,9 " S-121º31'51,4 " E). The study was conducted from March to July 2019. The experiment consisted of four treatments of *P. indicus* and *P. mooniana* seedlings: (i) control (without inoculum), (ii) inoculated with *Glomus claroideum* Schenk & Smith, (iii) inoculated with *Glomus coronatum* Giovann., and (iv) inoculated with mixed AMF (*G. claroideum, G. coronatum*). Each treatment has 15 replications.

Transplanting in the field

The study site is located in the gold post-mining of PT. Panca Logam Makmur, Bombana, Southeast Sulawesi (04º39'24,6 " S-121º53'58.1 " E). The study site was cleared 3 months before the start of the study and had no vegetation. The Bombana region has a tropical climate with average annual rainfall varying from 1083 mm to 1325 mm and is located at 101 m elevation. The recapitulation of the average temperature in the morning is 37.43°C, in the afternoon is 37.43°C and in the late afternoon is 32.59°C. The average humidity in the morning is 38.32%, 38.24% in the afternoon and 45.47% in the late afternoon. In this study, the number of rain occurrences was 49 days.

The field experiment was designed by using a Randomized Block Design with four treatments with three blocks. Each block has four treatments, namely (1) control (non AMF inoculation), (2) *Glomus claroideum* inoculation, (3) *Glomus coronatum* inoculation., and (4) mixed AMF (*G. claroideum, G. coronatum*) inoculation. Planting was carried out at 2 x 2 m spacing with planting hole of 40x40x40 cm. Seedlings of *P. indicus* and *P. mooniana* aged 11 months were transferred and planted in holes according to treatment. Four kg of cow manure compost was applied to each planting hole in each treatment. Chemical fertilizer (N: P_2O_5: K_2O/16:16:16) was applied as much as 50 g per plant one day after planting. Maintenance was done by watering with tap water every two days and weeding. Temperature and humidity measurements were carried out every day.

Growth parameter

Parameters observed in this study include: *Plant height* and diameter were measured on the stem at a height of 1 cm above the soil medium at greenhouse condition and 3 cm above the soil at field condition. *The number of leaves and nodule*s. They were counted at the end of the study. *Shoots and roots* were separately harvested and oven-dried at 70ºC for 48 hours and then weighed. *Dried weight* of 10 leaves per plant from the field. They were randomly selected. *Phosphorus (P) concentration* of the plant was analyzed using SL-MU-TT-05 (Bray I/II) method, while nitrogen (N) concentration of the plant was analyzed using SN 13-4721-1998 (Kjeldahl) method. *K, Mn and Fe concentrations* were analyzed using HNO_3-$HClO_4$ method (Carter 1993). *The N, P, K, Mn, and Fe content* were calculated by multiplying the nutrients content of the plant by the plant dry weight. *The increase/decrease of nutrients content* of plant inoculated with mycorrhiza relative to control plants was calculated based on following formula = [nutrient content of mycorrhizal plant – nutrient content of non-mycorrhizal plant/nutrient absorption of non-mycorrhizal plant] x 100% (Wang et al. 2005).

Seed quality index (SQI) was calculated using formula as follows (Duryea and Brown 1984):

Seed Quality Index (SQI) = [Shoot dry weight + root dry weight]/[(height/diameter) + (dry weight of shoot/root dry weight)]. Seedlings have high quality if the value of SQI $\geq$ 0.09.

Determination of mycorrhizal colonization and Mycorrhizae Inoculation Effect (MIE)

The *P. indicus* and *P. mooniana* roots were rinsed under running water. The roots were then soaked in 10% KOH for 24 hours, followed by acidifying in 2% HCl for 30 minutes, and finally stained with trypan blue. The number of AMF colonies were calculated using formula by Brundrett et al. (1996):

[Σ number of fields of view colonized/Σ total observed field of view] x 100%.

Mycorrhizae Inoculation Effect (MIE) was calculated using formula by Habte and Manjunath (1991):

[total dry weight of mycorrhizal plant-total dry weight of non-mycorrhizal plant/total dry weight of mycorrhizal plant] x 100%.

Statistical analysis

Each unit of observation was analyzed by analysis of variance (ANOVA). Differences between treatments were analyzed using LSD at the 95% confidence level.

RESULTS AND DISCUSSION

Mycorrhizal colonization and plant growth in the greenhouse

Arbuscular mycorrhizal fungi colonized all seedlings of *P. indicus* and *P. mooniana* (Table 2). *P. indicus* seedlings inoculated with *G. coronatum* and mixed AMF had higher AM colonization than control. Shoot height, and the number of nodule of *P. indicus* colonized by AM fungi was higher than those of control seedling (Table 2). Stem diameter of *P. indicus* inoculated with AMF was not significantly different compared to control. AMF inoculation on *P. mooniana* with *G. claroideum, G. coronatum* and mixed AMF increased shoot height and stem diameter of *P. mooniana* significantly ($p<0.05$), however, shoot height of three types of AMF treatments were not significantly different ($p>0.05$). The number of leaves and nodule of *P. mooniana* colonized by *G.*

coronatum was higher than the other treatments. There was no difference in number of leaves and nodule between seedlings colonized by mixed AMF and control seedlings.

Inoculation of AMF increased the initial growth of two endangered trees species *P. indicus* and *P. mooniana* after four months of transplanting under greenhouse conditions (Table 2). The results of this study indicated that AMF inoculation in the greenhouse or nursery is very important to support the success of planting. The results of this study confirmed the results of the study conducted by Urgiles et al. (2009) who stated that colonization of native AMF colonization enhanced the growth of *C. montana* and *H. americanus* in the nursery condition. Previous studies showed that native AMF increased the growth of *P. mooniana* in greenhouse and nursery scale on inceptisol media (Husna et al. 2015), serpentine (Husna et al. 2016) and coal spoil (Husna et al. 2019). In gold tailings media, native AMF is effective in increasing the growth of *K. celebica* (Husna et al. 2019; 2021), *N. orientalis* (Tuheteru et al. 2020), *Paspalum* (Fiqri et al. 2016), *E. cladacalyx* (Madejon et al. 2012), and *D. viscosa*, *A, eucomus* and *I. cylindrica* (Orłowska et al. 2011). Inoculation of 2 g AMF improved the growth of *Pongamia pinnata* plants in open pit coal mine soil media (Agus et al. 2018). Inoculation of AMF (*G. decipiens* and *G. clarum*) increased early growth and nutrient uptake of *Mallotus paniculatus* and *Albizia* under nursery conditions (Wulandari et al. 2014).

P. indicus colonized by *G. claroideum* and mixed AMF had higher dry weight than control and *G. coronatum* inoculation treatment (Table 3). AMF inoculation by *G. claroideum*, *G. coronatum* and mixed AMF increased shoot dry weight and total dry weight of *P. indicus* significantly. Shoot dry weight and total dry weight of *P. indicus* seedlings inoculated by *G. claroideum*, *G. coronatum* and mixed AMF were not significantly different. *G. coronatum* inoculation on *P. indicus* did not increase total dry weight (Table 3). AMF inoculation by *G. claroideum*, *G. coronatum* and mixed AMF increased shoot, root and total dry weight of *P. mooniana*. Shoot, root and total dry weight of *P. mooniiana* seedlings in the three AMF inoculation treatments were not significantly different. The MIE of *P. indicus* and *P. mooniana* values ranged from 37,5 to 50.6% and 61,5 to 63,2% (Table 3). Shoot and roots ratio of *P. indicus* and *P. mooniana* was not different among the seedlings (Table 3). There was no significant effect on SQI of *P. indicus* between the non-inoculated and inoculated plants. AMF inoculation by *G. claroideum*, *G. coronatum*, and mixed AMF improve SQI of *P. mooniana.*

Table 2. The effect of four months AMF inoculation on mycorrhizal colonization, shoot height, stem diameter, leaf and nodule of *P. indicus* and *P. mooniana* in the greenhouse conditions

Plant species treatment	Mycorrhizal colonization (%)*	Shoot height (cm)*	Stem Diameter (mm)*	Leaf number*	Nodule*
P. indicus					
Control	5.67±1.113c	17.80 ± 0.723c	3.88 ± 0.444a	22.67 ± 4.645b	7.00 ± 1.155d
G. claroideum	52.7±19.047b	42.50 ± 0.759a	3.79 ± 0.204a	61.00 ± 11.00a	16.67 ± 1.443c
G. coronatum	69.9±19.047ab	37.47 ± 1.223b	3.85 ± 0.195a	51.33 ± 15.61ab	50.33 ± 0.881a
Mixed AMF	75.3±3.1535a	39.63 ± 1.954ab	4.34 ± 0.316a	72.33 ± 8.114a	34.67 ± 1.5b
P. mooniana					
Control	2.2±0.502 c	15.63 ± 0.726b	3.23 ± 0.117c	6.00 ± 0.577c	3.67 ± 1.667c
G. claroideum	39.6± 0.580ab	21.73 ± 0.384a	4.34 ± 0.072a	15.00 ± 0.577b	30.00 ± 6.244b
G. coronatum	36.5± 2.214b	21.23 ± 0.545a	3.82 ± 0.200b	14.00 ± 0.577a	64.33 ± 5.783a
Mixed AMF	41.2± 0.536a	21.90 ± 0.550a	3.86 ± 0.165b	16.00 ± 0.577bc	16.67 ± 4.409bc

Note: Average values followed by different letters in the same column are significantly different at LSD (P<0.05); *Mean±SE

Table 3. Total dry weight, shoot and root dry weight, and SQI of non-AMF and four months AMF inoculated of *P. indicus* and *P. mooniana* in the greenhouse

Plant species treatment	Dry weight (g/plant)*			MIE (%)*	Ratio of shoot and root *	SQI*
	Roots	Shoots	Total			
P. indicus						
Control	0.46 ± 0.02c	1.65 ± 0.141b	2.10 ± 0.323b	-	3.63± 0.174a	0.489± 0.041a
G. claroideum	0.91 ± 0.057ab	3.32 ± 0.360a	4.22 ± 0.420a	50.6±8.830	3.6 ± 0.302a	0.283± 0.013a
G. coronatum	0.76 ± 0.111b	2.72 ± 0.191a	3.47± 0.781ab	50.1±8.830	3.87± 0.774a	0.267± 0.071a
Mixed AMF	1.07 ± 0.129a	3.29 ± 0.264a	4.37± 0.394a	37.5±10.527	3.10± 0.115a	0.357± 0.038a
P. mooniana						
Control	0.65 ± 0.065b	2.40 ± 0.162b	3.05 ± 0.220b	-	3.72± 0.205a	2.40 ± 0.285b
G. claroideum	1.39 ± 0.157a	6.96 ± 0.379a	8.36 ± 0.285a	63,2±3.910	5.20± 0.907a	8.73 ± 1.673a
G. coronatum	1.71 ± 0.135a	6.60 ± 0.371a	8.31 ± 0.441a	62,9±4.024	3.90± 0.327a	5.83 ± 0.424a
Mixed AMF	1.55 ± 0.079a	6.42 ± 0.591a	7.97 ± 0.591a	61,5±2.184	4.17± 0.462a	5.96 ± 1.018a

Note: Average values followed by different letters in the same column are significantly different at LSD (P<0.05); *Mean±SE

Table 4. N, P, K, Mn and Fe concentration in the root of non-AMF inoculated and four months of AMF inoculation in *Pterocarpus indicus* and *Pericopsis mooniana* grown in the greenhouse

Plant species treatment	Concentration (mg g^{-1}) *				
	N	P	K	Mn	Fe
P. indicus					
Control	20.2 ± 0.75	3.1 ± 0.22	20.8 ± 2.53	0.69 ± 0.017a	0.19 ± 0.052
G. claroideum	20.6 ± 1.07	3.1 ± 0.08	18.2 ± 0.71	0.70 ± 0.010a	0.16 ± 0.053
G. coronatum	23.5 ± 1.06	2.8 ± 0.08	20.9 ± 0.71	0.41 ± 0.010b	0.16 ± 0.052
Mixed AMF	20.4 ± 0.08	3.5 ± 0.08	19.6 ± 2.22	0.23 ± 0.022b	0.17 ± 0.002
P. mooniana					
Control	17.3 ± 1.12	4.0 ± 0.37	17.8 ± 1.33	0.23 ± 81.388	1.57 ± 0.294
G. claroideum	15.3 ± 1.06	2.7 ± 0.16	15.4 ± 1.70	0.19 ± 37.239	1.02 ± 0.049
G. coronatum	17.0 ± 1.53	4.4 ± 0.73	13.8 ± 2.58	0.30 ± 73.053	1,23 ± 0.202
Mixed AMF	17.6 ± 1.25	4.5 ± 0.49	14.2 ± 1.67	0.19 ± 20.207	0.87 ± 0.083

Note: Average values followed by different letters in the same column are significantly different at LSD (P<0.05); *Mean±SE

Table 5. N, P, K, Mn and Fe contents in the root of non-AMF inoculated and four months of AMF inoculation in *Pterocarpus indicus* and *Pericopsis mooniana* in the greenhouse

Plant species treatment	Content (mg plant^{-1}) *				
	N	P	K	Mn	Fe
P. indicus					
Control	9.29 ± 0.58b	1.43 ± 0.11c	9,57 ± 1.45b	0.32 ± 0.006b	0.09 ± 0.019b
G. claroideum	18.7 ± 1.94a	2.82 ± 0.27ab	16,56 ± 2.16ba	0.64 ± 0.016a	0.15 ± 0.082ab
G. coronatum	17.8 ± 1.94a	2.13 ± 0.26bc	15,88 ± 2.16ba	0.31 ± 0.016b	0.12 ± 0.082b
Mixed AMF	21.8 ± 2.54a	3.75 ± 0.37a	20,97 ± 1.669a	0.25 ± 0.014b	0.18 ± 0.031a
P. mooniana					
Control	11.25 ± 1.92c	2.60 ± 1.03b	11.57 ± 0.80	0.38 ± 0.072	1.02 ± 0.295
G. claroideum	21.27 ± 2.14b	3.75 ± 7.24a	21.41 ± 3.79	0.10 ± 0.067	1.42 ± 0.013
G. coronatum	29.07 ± 0.90a	7.52 ± 7.20a	23.60 ± 4.87	0.13 ± 0.112	2.10 ± 0.049
Mixed AMF	27.28 ± 3.01ab	6.98 ± 5.78ab	22.01 ± 3.53	0.08 ± 0.001	1.35 ± 0.066

Note: Average values followed by different letters in the same column are significantly different at LSD (P<0.05); *Mean±SE

Mycorrhizal Inoculation Effect (MIE) in *P. indicus* was 50.6, 50.1 and 37.5 when inoculated with *G. clarodeum, G. coronatum* and mixed AMF, respectively; while MIE in *P. mooniana* was 63.2, 62.9 and 61.5, respectively. *P. mooniana* was more dependent on AMF colonization than *P. indicus*. Plant species that are highly dependent on AMF colonization have an MIE of 50-75% (Habte and Manjunath 1991). A high MIE value indicates that the growth and survival *P. mooniana* depend on the presence of symbiosis with AMF. This research is in line with research on *P. mooniana* plants in various soil media, including inceptisol (Husna et al. 2015), ultramafic (Husna et al. 2016) and coal spoil (Husna et al. 2019). A high dependency on AMF has also been reported for other endangered tree species, i.e., *Aquilaria malacensis* and *A. crasna* (Turjaman et al. 2006a) and *K. celebica* (Husna et al. 2021). The differences in MIE of tree species are closely related to the characteristics of the roots and root hairs (Habte and Manjunath 1991). *P. mooniana* has a high mycorrhizal dependence presumably due to its low branching and poor root hairs.

Nutrient content of shoot and root in plant grown in the green house

The N, P, K and Fe concentrations in the root of *P. indicus* dan *P. mooniana* among the inoculated seedlings were not significantly different (Table 4). The concentration of Mn in the root of *P. indicus* colonized by *G. coronatum* and mixed AMF was lower than control and *G. claroideum* inoculation (Table 4). The Mn concentration of *P. mooniana* root did not differ among seedlings.

AMF inoculation by *G. claroideum, G. coronatum* dan mixed AMF increased the N and K content in the shoot of *P. indicus*, although there was no difference between the AMF inoculation treatments in *P. indicus* seedlings. *P. indicus* colonized by *G. claroideum* dan mixed AMF had higher P and Fe content in the root than control and *G. coronatum* inoculation (Table 5). *P. indicus* colonized by *G. claroideum* had higher Mn content in the root than control, *G. claroideum* and mixed AMF (Table 5). AMF inoculation by *G. coronatum* and mixed AMF increased the N content of *P. mooniana* roots. There was no difference in root P content between seedlings colonized by mixed AMF and control treatment. The K, Mn, and F content in the root of *P. mooniana* did not differ among the seedlings.

Concentration of K and Mn in inoculated seedlings of *P. indicus* shoots was not significantly different (Table 6). The concentration of N in the shoot of *P. indicus* colonized by *G. coronatum* and control was higher than in the mixed AMF and *G. claroideum* inoculation (Table 6). Inoculation of the mixed AMF increased the shoot P and Fe concentration of *P. indicus*. There was no difference in shoot N, P, K, Mn and Fe concentration of *P. mooniana* among the inoculated seedlings (Table 6).

AMF inoculation by *G. claroideum*, *G. coronatum* dan mixed AMF increased the N, K and Mn content in the shoots of *P. indicus*. There was no significant difference between K content in the soot of *P. indicus* seedlings inoculated by *G. claroideum, G. coronatum* and control. *P. indicus* inoculated by mixed AMF had higher shoot P and Fe content than control and *G. coronatum* inoculation (Table 7). AMF inoculation by *G. claroideum*, *G. coronatum,* and mixed AMF increased shoot N, P, K and Mn content of *P. mooniana*. Shoot Fe content of *P. mooniana* did not differ among the seedlings.

The effect of AMF inoculation on changes in nutrient content

AMF inoculation increased the N, P, K, Mn content in the root as well as N, P, K, Mn and Fe contents in the shoot of *P. indicus* seedlings. The mixed AMF treatment decreased Fe content in the root by 55,3 % (Table 8). *P. mooniana* inoculated by G. *claroideum, G. coronatum* and mixed AMF had higher N, P, K, Mn and Fe content in the root and N, P, K and Fe in the shoot. The mixed AMF treatment decreased Mn content in the shoot by 11,3 % (Table 8).

The results of our study confirmed the positive effect of AMF inoculation on plant growth, nutrient uptake of *P. indicus* and *P. mooniana* grew in gold post-mine land. The results of this study indicate that local AMF is effective in increasing plant growth both in greenhouses and in the field. Corresponding to our results, that AMF inoculation of *G. macrocarpum* and *G. fasciculatum* significantly increased the survival rate and growth of *Cassia siamea* in Indian wasteland in field one year after transplanting (Giri et al. 2005). A previous study by Wulandari et al. (2016) showed that inoculation of the three native AMF increased the survival rate, plant growth and uptake of N, P of *Albizia saman* and *Paraserianthes falcataria* in post-opencast coal mine field (Wulandari et al. 2016). Local AMF was effective in increasing the growth of *Pericopsis mooniana* at 24 and 36 months after planting in nickel post mining land (Husna et al. 2017, 2018). The positive effect of AMF was shown on the growth of *Metrosideros laurifolia* in ultramafic soil in New Caledonia (Amir et al. 2019).

Table 6. The N, P, K, Mn and Fe concentration in the shoot of non-AMF inoculated and four months of AMF inoculation in *P. indicus* and *P. mooniana* in the greenhouse

Plant species treatment	Concentration (mg g^{-1})*				
	N	P	K	Mn	Fe
P. indicus					
Control	26.1 ± 1.04a	3.3 ± 0.11b	29.0 ± 0.91	0.128 ± 0.014	0.221 ± 0.046b
G. claroideum	22.9 ± 0.41b	2.4 ± 0.11c	24.8 ± 3.71	0.155 ± 0.017	0.161 ± 0.012b
G. coronatum	24.9 ± 0.21a	2.4 ± 0.08c	22.6 ± 2.31	0.169 ± 0.016	0.189 ± 0.021b
Mixed AMF	21.9 ± 0.39b	3.8 ± 0.09a	26.7 ± 2.31	0.184 ± 0.003	0.635 ± 0.069a
P. mooniana					
Control	21.8 ± 0.95	3.7 ± 0.08	29.3 ± 2.29	0.168 ± 0.041	0.154 ± 0.010
G. claroideum	25.5 ± 1.85	3.5 ± 0.42	30.2 ± 2.86	0.131 ± 0.005	0.136 ± 0.019
G. coronatum	25.9 ± 2.99	3.9 ± 0.46	27.6 ± 0.89	0.131 ± 0.018	0.154 ± 0.056
Mixed AMF	23.5 ± 2.46	3.9 ± 0.24	23.0 ± 4.16	0.149 ± 0.012	0.585 ± 4.568

Note: Average values followed by different letters in the same column are significantly different at LSD (P<0.05); *Mean±SE

Table 7. Shoot N, P, K, Mn and Fe contents of non-AMF inoculated and four months of AMF inoculation in *Pterocarpus indicus* and *Pericopsis mooniana* in the greenhouse

Plant species treatment	Content (mg plant^{-1})				
	N	P	K	Mn	Fe
P. indicus					
Control	43.07 ± 4.03b	5.45 ± 0.63b	47.85 ± 2.81b	0.21 ± 0.036b	0.36 ± 0.101b
G. claroideum	76.03 ± 7.09a	7.97 ± 1.01b	82.34 ± 18.03ab	0.51 ± 0.038a	0.53 ± 0.026b
G. coronatum	67.73 ± 4.52a	6.53 ± 0.59cb	61,47 ± 1.59ab	0.46 ± 0.065a	0.51 ± 0.072b
Mixed AMF	72.05 ± 6.67a	12,50 ± 1.32a	87.84 ± 11.29a	0.61 ± 0.038a	2.09 ± 0.257a
P. mooniana					
Control	52.32 ± 1.36b	8.88 ± 0.76b	70.32 ± 7.03b	0.40 ± 0.023b	0.37 ± 0.027
G. claroideum	177.48 ± 7.24a	24.36 ± 3.21a	210.19 ± 15.73a	0.91 ± 0.077a	0.95 ± 0.097
G. coronatum	170.94 ± 9.81a	25.74 ± 2.44a	182.16 ± 13,91a	0.86 ± 0.153a	1.02 ± 0.372
Mixed AMF	150.87 ± 21.55a	25.04 ± 3.68a	147.66 ± 36.79a	0.96 ± 0.142a	3.76 ± 0.317

Note: Average values followed by different letters in the same column are significantly different at LSD (P<0.05); *Mean±SE

Table 8. The effect of AMF inoculation on changes in nutrients contained in the root and shoot of 4-month old *P. indicus* dan *P. mooniana* seedlings in the greenhouse

Plant species/ treatment	Roots					Shoots				
	N	P	K	Mn	Fe	N	P	K	Mn	Fe
P. indicus										
G. claroideum	100.7	100.7	76.9	62.7	106.5	76.1	46.9	69.9	136.8	45.4
G. coronatum	90.2	50.6	64.5	35.0	2.4	57.6	21.7	26.6	116.5	41.7
Mixed AMF	72.5	59.9	75.0	60.1	-55.3	30.6	89.8	12.5	42.4	72.4
P. mooniana										
G. claroideum	85.2	486.3	88.6	60.8	35.7	241.6	173.7	197.1	119.6	152.8
G. coronatum	151.2	641.9	107.8	204.7	98.0	224.9	181.7	159.0	109.2	172.2
Mixed AMF	46.1	242.3	30.4	41.6	11.7	140.7	90.8	54.2	-11.3	43.0

Table 9. Mycorrhizal colonization, shoot height, stem diameter, leaf and leaf dry weight of *P. indicus* and *P. mooniana* grown with or without four-month mycorrhizal fungi colonization in post-gold minefield in Bombana, Southeast Sulawesi, Indonesia

Plant species treatment	Mycorrhizal colonization (%)*	Shoot height (cm)*	Stem diameter (mm)*	Leaf number*	Leaf dry weight (g)*
P. indicus					
Control	40.0 ± 9.01 b	125.9 ± 7.96 c	16.6 ± 0.98 b	572 ± 88.86	1.90 ± 0.36 c
G. claroideum	81.7 ± 8.90 a	197.3 ± 14.9 a	20.9 ± 1.01 a	585 ± 101.51	6.64 ± 1.02 ab
G. coronatum	84.2 ± 1.67 a	162.2 ± 6.27 b	19.5 ± 0.97 a	339 ± 99.24	4.39 ± 0.75 b
Mixed AMF	93.3 ± 2.20 a	141.1 ± 9.33 bc	18.8 ± 1.53 ab	829 ± 42.00	8.15 ± 0.34 a
P. mooniana					
Control	7.5 ± 3.82 b	5.3 ± 0.69 c	1.78 ± 0.25 c	31 ± 5.86 d	0.65 ± 0.16 b
G. claroideum	18.3 ± 4.64 b	7.6 ± 0.35 c	2.65 ± 0.45 c	82 ± 2.02 b	0.75 ± 0.01 b
G. coronatum	50.0 ± 10.90 a	39.1 ± 1.73 a	6.37 ± 0.10 a	105 ± 3.46 a	1.48 ± 0.28 a
Mixed AMF	50.0 ± 8.04 a	18.6 ± 0.03 b	3.65 ± 0.26 b	61 ± 2.31 c	1.11 ± 0.30 ab

Note: Average values followed by different letters in the same column are significantly different at LSD ($P<0.05$); *Mean±SE

Mycorrhizal colonization and plant growth in the post-gold minefield

The number of colonization of *G. claroideum*, *G. coronatum* and mixed AMF on *P. indicus* roots was higher than on *P. mooniana* roots at 4 months after planting in the post-gold minefield (Table 9). There was no significant difference in the percentage of colonization between *G. claroideum*, *G. coronatum* and mixed AMF on *P. indicus*. Seedlings in control treatment of *P. indicus* and *P. mooniana* were colonized by indigenous AM Fungi.

Colonization by the Arbuscular mycorrhizal fungi G. *claroideum* and *G. coronatum* increased shoot height and stem diameter of *P. indicus* (Table 9). However, colonization by mixed AMF did not increase shoot height and stem diameter of *P. indicus*. There was no significant difference in number of leaves on *P. indicus* seedlings colonized by AMF and control. Leaf dry weight of *P. indicus* colonized by AMF was higher than control.

Shoot height, stem diameter, and leaf dry weight of *P. mooniana* colonized by *G. coronatum* and mixed AMF was higher than that of control and *G. claroideum* colonization (Table 9). There was no significant difference in shoot height, stem diameter and leaf dry weight of seedling colonized by *G. claroideum* and control. Seedlings colonized by *G. coronarum* had higher number of leaf and leaf dry weight than seedlings in control treatment.

Pterocarpus indicus and *P. moniana* can adapt and grow on tailing soil media and post-gold post minefield with a seedling survival rate of 100%. These species are potential to be used as tree species for revegetation of post-gold mining land in the tropics. *P. indicus* can be grown in open land due to its light-demanding species, grows in various soil conditions and fast-growing species. Meanwhile, *P. mooniana* can be grown with enrichment planting technique because it requires shade at the beginning of its growth. Both species are suitable to be developed as revegetation species because they meet the requirements of economic importance, N-fixing capacity, fast-growing, tolerance to adverse climate and soil conditions, have a deep root system and are easy to propagate (Maiti 2013). The results of this study indicate that post-mining land can be used as an *ex-situ* conservation area for endangered tree species. Therefore, mining reclamation policies in tropics should oblige mining companies to use tropical tree species as one of the selected species for post-mining land reclamation.

Based on the results of this study, indicated that AMF has a very important role in ecological restoration and conservation of endangered tropical tree species. It is due to the ability of AMF to increase the surface area of root absorption by 100 or even 1000 times (Larcher 1995). Therefore, it can improve plant nutrient status and water status (Smith and Read 2008). AMF can also improve soil structure and plant resistance to environmental stress and reduce the need for fertilizer use.

Table 10. Shoot N, P, K, Mn and Fe content of *Pterocarpus indicus* and *Pericopsis mooniana* grown with or without four-month mycorrhizal fungi inoculation in post gold minefield in Bombana, Southeast Sulawesi, Indonesia

Plant species treatment	**Concentration (mg g^{-1})**				
	N	**P**	**K**	**Mn**	**Fe**
P. indicus					
Control	38.7±0.17	2.6±0.01	16.4±0.27	0.47±0.123	0.25±0.041
G. claroideum	41.1±0.33	3.3±0.01	23.5±0.26	0.39±0.103	0.27±0.046
G. coronatum	38.7±0.29	2.9±0.04	25.3±0.14	0.13±0.043	0.20±0.003
Mixed AMF	39.2±0,33	2.8±0.01	20.2±0.16	0.25±0.039	0.23±0.019
P. mooniana					
Control	14.3±0.78	1.3±0.07	7.9±0.45	0.044±0.028	0.12±0.060
G. claroideum	28.0±0.27	2.2±0.04	13.4±0.17	0.085±0.024	0.15±0.039
G. coronatum	29.1±0.54	2.0±0.01	14.8±0.16	0.139±0.056	0.18±0.038
Mixed AMF	24.6±0.20	1.7±0.01	11.3±0.15	0.084±0.011	0.14±0.027

Note: *Mean±SE

Table 11. Shoot N, P, K, Mn, and Fe contents of *Pterocarpus indicus* and *Pericopsis mooniana* grown with or without mycorrhizal fungi under post gold mining land conditions in Bombana, Southeast Sulawesi, Indonesia, for four months

Plant species treatment	**Content (mg plant^{-1})***				
	N	**P**	**K**	**Mn**	**Fe**
P. indicus					
Control	73.53±14.4 c	4.94±0.1 c	31.16±2.4 b	0.89±0.33	0.48±0.160 b
G. claroideum	272.90±59.1 ab	21.91±03.5 a	156.04±29.8 a	2.59±0.99	1.79±0.460 a
G. coronatum	169.89±29.2 bc	12.73±0.4 b	111.07±12.6 a	0.57±0.003	0.88±0.154 ab
Mixed AMF	319.48±1.53 a	22.82±0.9 a	164.63±12.0 a	2.04±0.76	1.87±0.248 a
P. mooniana					
Control	9.30±4.5 c	0.85±0.4 b	5.14±2.3 b	0.03±0.03 b	0.08±0.050 b
G. claroideum	21.00±2.4 c	1.65±0.2 b	10.05±1.4 b	0.06±0.01 b	0.11±0.011 b
G. coronatum	43.07±4.8 a	2.96±0.5 a	21.90±5.7 a	0.21±0.001 a	0.276±0.04 a
Mixed AMF	27.31±4.2 b	1.89±0.7 ab	12.54±5.2 ab	0.09±0.002 b	0.16±0.033 b

Note: Average values followed by different letters in the same column are significantly different at LSD ($P<0.05$); *Mean±SE

Shoot N, P, K, Mn and Fe contents in the post-gold minefield

There was no difference in shoot N, P, K, Mn and Fe concentration of *P. mooniana* and *P. indicus* among the inoculated seedlings (Table 10). AMF inoculation by *G. claroideum, G. coronatum* and mixed AMF increased shoot N, K and Mn content of *P. indicus*, but there was no significant difference in shoot K content of *P. indicus* seedlings inoculated *G. claroideum, G. coronatum* and control. *P. indicus* colonized by mixed AMF had higher shoot P and Fe content than control and *G. coronatum* colonization (Table 11). AMF inoculation by *G. claroideum*, *G. coronatum,* and mixed AMF increased shoot N, P, K and Mn content of *P. mooniana*. Shoot Fe content of *P. mooniana* was not significantly different among the seedlings.

Our results also showed the increase of shoots and roots N, P, K content of *P. indicus* and *P. mooniana* by AMF colonization in four-month-old in the greenhouse. AMF fungal inoculation improved shoot N, P and K uptake of *P. indicus* and *P. mooniana* in post-gold minefield. N, P and K are macronutrients that are essential for plant growth and development. Our results are in line with previous studies showing that AMF can increase N and P uptake in *P. mooniana* (Husna et al. 2015; 2016; 2019a) and *K. celebica* (Husna et al. 2021). Increased nutrient uptake by AMF in low fertility soils indicates that AMF can reduce fertilizer use. However, the applications of organic and inorganic fertilizers are strongly needed for planting activities on post-gold mining land (Muddarisna and Siahaan 2014; Sulakhudin et al. 2017).

Mixed AMF decreased Fe content in the roots of *P. indicus* and Mn content in the shoots of *P. mooniana* (Table 4) in plant grown in the greenhouse. Inoculation of *G. coronatum* decreased Mn content in the shoots of *P. indicus*. In gold tailings soil media, the application of mixed AMF also reduced Fe content by 15% in the roots of *K. celebica* (Husna et al. 2021), while *Glomus* sp. reduced Fe content by 13% in the roots of *N. orientalis* (Tuheteru et al. 2017).

In conclusion, the results of our study in the greenhouse and post-gold mining field demonstrated that AMF promoted growth and increased nutrient uptake of *P. indicus* and *P. mooniana* seedlings four months after planting. AMF has the potential to be developed as a biofertilizer in the cultivation of *P. indicus* and *P. mooniana* and the conservation of endangered tropical trees in the tropics and may be adopted in large scale nurseries along with reforestation.

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