

Enhanced growth and nutrient uptake of *Pterocarpus indicus* in asphalt post-mining soil media using local arbuscular mycorrhizal fungi

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Abstract. Husna, Tuheteru FD, Nurdin WR, Ramadin, Arif A. 2025. Enhanced growth and nutrient uptake of *Pterocarpus indicus* in asphalt post-mining soil media using local arbuscular mycorrhizal fungi. *Biodiversitas* 26: 1956-1963. Local Arbuscular Mycorrhizal Fungi (AMF) were previously reported to be effective in increasing the growth and nutrient uptake of legume plant in various degraded soil media, such as serpentine, coal spoil, and tailings. The entire previous studies related to AMF were not conducted in asphalt post-mining soil media. Therefore, this study aimed to examine the effect of local AMF inoculation on the growth and nutrient uptake of *Pterocarpus indicus* in asphalt post-mining soil media under Indonesian Mycorrhizal Association nursery conditions. A total of 7 treatments were tested in a 3-month experimental period by using seedlings of *P. indicus*. These included (A) control (no inoculation), as well as soil media inoculated with (B) *Glomus coronatum*, (C) *Glomus claroideum*, (D) *Glomus halonatum*, (E) mixed AMF (*G. claroideum* + *G. coronatum*), (F) mixed AMF (*G. claroideum* + *G. halonatum*), and (G) mixed AMF (*G. coronatum* + *G. claroideum* + *G. halonatum*). The parameters observed were AMF colonization percentage, nutrient content, plant growth, as well as the uptake of N, P, K, and Zn. After 3 months of experiment, the AMF colonization percentage in *P. indicus* ranged between 55.00% and 69.33%. *G. claroideum* and *G. coronatum* colonization showed higher plant growth in terms of height, diameter, number of leaves, and nodules, dry weight, as well as N, P, K, and Zn content at nursery conditions. *P. indicus* and mixed AMF consisting of *G. claroideum* and *G. coronatum* can be used for the restoration program in asphalt post-mining land in the tropics.

Keywords: Glomeraceae, land restoration, tree species conservation, tropical legume tree

INTRODUCTION

Arbuscular Mycorrhizal Fungi (AMF) are essential soil microbes for supporting the successful development of plant life on degraded land (Tuheteru et al. 2022). These are often found in symbiosis with 97% of plant families, helping to enhance growth, water and nutrient status, as well as resistance against biotic and abiotic stresses (Begum et al. 2019; Diagne et al. 2020; Wang et al. 2024). Additionally, some reports show that AMF can be developed as a tool and technology to support restoration programs (Asmelash et al. 2016; Wang 2017; de Moura et al. 2022) and the conservation of endangered species (Husna et al. 2021a,b). Providing AMF at nursery conditions is crucial to improving the quality of forestry seedlings, specifically in ecosystem restoration (Urgiles et al. 2009). Various studies and review results showed that AMF could increase tropical plant growth in post-mining soil media at nursery conditions (Husna et al. 2021a,b; de Moura et al. 2022; Wulandari et al. 2024).

AMF is considered an efficient tool for achieving the successful restoration of post-mining areas (de Moura et al. 2022; Amir et al. 2023). This has been reported to be capable of enhancing growth in varying conditions of post-mining soil media, including serpentine and nickel extraction sites (Husna et al. 2016; Amir et al. 2019), coal (Wulandari et al. 2024), and gold minings (Husna et al. 2021a,b). Moreover, AMF effectiveness of restoration

programs is highly dependent on mined minerals and the types fungi of used. Wulandari et al. (2024) found that AMF effectiveness varied with types/species, signifying a need to conduct screening in selecting the best option for reclamation. Previous studies showed that using local AMF is more effective than the exotic types (Husna et al. 2016; 2019; 2021b; Tuheteru et al. 2020a). Some of the local AMF tested at nursery and field conditions were isolated from the rhizosphere of *Pericopsis mooniana* Thwaites (legum) obtained from various field conditions. These were proven to be effective in increasing legume plant growth at nursery conditions in overburdened coal mining land (Husna et al. 2019), gold mine tailings (Husna et al. 2021a), serpentine and nickel post-mining soil media (Husna et al. 2016), as well as at field conditions in both gold (Husna et al. 2021b) and nickel post-mining land (Husna et al. 2017a). However, local AMF has not been tested on asphalt post-mining soil media for the enhancement of legume growth and nutrient uptake. Soil characteristics of asphalt post-mining land include slightly alkaline pH (7.7-8.1), available P, very low organic N and C content, as well as loamy texture (Albasri et al. 2023).

Selecting plant types cultivated for revegetation activities on asphalt post-mining land is crucial in addition to using soil microorganisms. An example of a potential legume generally selected for revegetation activities is *Pterocarpus indicus* Willd., a tropical tree species (Fabaceae) that produces luxury wood (Soerianegara and Lemmens

1994). *P. indicus* is categorized as vulnerable by the International Union for Conservation of Nature (IUCN) and considered a suitable species for reforestation programs and plantation forestry (Husna et al. 2021b). This plant distributes primarily across lowland evergreen forests and some secondary forests with altitudes reaching 1,300 masl. *P. indicus* has the best growth in sunny areas, shows adaptability to various soil types (Manipol et al. 2020; Husna et al. 2021c), and thrives naturally on Buton Island, Southeast Sulawesi Province, Indonesia (Powling et al. 2015).

Previous studies reported that local AMF supported improvement in the growth and nutrient uptake of legume on both mineral and post-mining soil media. Local AMF application was found to increase the growth and nutrient uptake of *P. indicus* on gold mine tailings and post-mining land, excluding asphalt post-mining land (Husna et al. 2021b). Therefore, this study aimed to examine the effect of local AMF inoculation on the growth and nutrient uptake of *P. indicus* in asphalt post-mining soil media under nursery conditions. The inoculation of local AMF is expected to improve plant growth and nutrient uptake of *P. indicus* at nursery conditions.

MATERIALS AND METHODS

Soil preparation

Soil media of the asphalt post-mining land (overburden) was collected on the 14th of June 2024 from PT. Wijaya Karya, Buton District of Southeast Sulawesi, Indonesia at

Geographical coordinates of 122° 53' 38.76"–50° 23' 8.28" and then kept in nursery conditions. Furthermore, the chemical and physical characteristics were analyzed at the Soil Laboratory of Soil Research Center, Bogor, West Java, Indonesia. The asphalt post-mining soil media were mixed with husk-charcoals and river sands at a ratio of 6:3:1 (v/v/v). The properties of asphalt post-mining land and soil media mixtures are comprehensively presented in Table 1.

Inoculation of AMF

The local inocula used were *Glomus claroideum*, *Glomus coronatum*, *Glomus halonatum*, and AMF mixtures (equal in volume, 1:1) isolated from the rhizosphere of *Pericopsis mooniana* (Husna et al. 2015). These samples were selected due to the proven effectiveness in increasing legume growth in various media conditions. Propagation of AMF was conducted in zeolite media for 3 months using *Pueraria javanica* as the host in the greenhouse of the Indonesian Mycorrhizal Association, Southeast Sulawesi Branch, Kendari.

Approximately 1,000 g of sterile soil media from the asphalt post-mining area were put into polyethylene pots measuring 15 × 20 cm. For inoculation, 10 g of each AMF species was positioned 1 to 3 cm underneath the seedlings. Subsequently, 2 new leaves of the respective *P. indicus* seedlings were planted into the pots and maintained with daily watering to ensure field capacity over 3 months. In the control treatment, 10 g of sterilized zeolite was added to pots without AMF inoculation.

Table 1. Soil physical and chemical properties

Parameter	Unit	The asphalt post-mining soil media	The asphalt post-mining soil media were mixed with husk-charcoals and river sands with a mixing ratio of 6:3:1
pH H ₂ O		7.6	7.8
C-organic (Walkley & Black)	%	7.19	5.41
N Total (Kjeldahl)	%	0.19	0.20
C/N ratio		38	27
P ₂ O ₅ (HCl 25%)	mg/100 g	55	54
K ₂ O (HCl 25%)	mg/100 g	9	15
P ₂ O ₅ (Olsen)	ppm	8	25
K ₂ O (Morgan)	ppm	53.7	142.7
Ca (NH ₄ -Acetat 1N, pH 7)	cmol/kg	19.54	11.07
Mg (NH ₄ -Acetat 1N, pH 7)	cmol/kg	1.42	0.34
K (NH ₄ -Acetat 1N, pH 7)	cmol/kg	0.09	0.23
Na (NH ₄ -Acetat 1N, pH 7)	cmol/kg	0.17	0.42
CEC (NH ₄ -Acetat 1N, pH 7)	cmol/kg	26.45	15.85
Base saturation (NH ₄ -Acetat 1N, pH 7)	%	80	79
Al ³⁺ (KCl 1N)	cmol/kg	0.00	0.00
H ⁺ (KCl 1N)	cmol/kg	0.05	0.04
Sand	%	16	52
Silt	%	44	24
Clay	%	40	24
Fe (DTPA)	ppm	5.3	4.2
Mn (DTPA)	ppm	10.0	11.2
Cu (DTPA)	ppm	1.0	0.5
Zn (DTPA)	ppm	0.3	0.4
Pb (Morgan Wolf)	ppm	0.4	0.7
Cd (Morgan Wolf)	ppm	0.2	0.2
CaCO ₃ (Titrimetri)	%	0.06	0.04

Preparation of seedlings

Pterocarpus indicus seeds collected from the parent plant at the vicinity of Universitas Halu Oleo located in Kendari, Southeast Sulawesi, Indonesia were soaked in hot water at 50°C until cooled down for 24 hours, then a germination process was carried out using a plastic sprout maker. A total of 7 treatments were implemented in the experiment carried out over 3 months by using nine *P. indicus* seedlings per treatment. These included (A) control (no inoculation), as well as soil media inoculated with (B) *G. coronatum*, (C) *G. claroideum*, (D) *G. halonatum*, (E) mixed AMF (*G. claroideum* + *G. coronatum*), (F) mixed AMF (*G. claroideum* + *G. halonatum*), and (G) mixed AMF (*G. coronatum* + *G. claroideum* + *G. halonatum*), which had 3 replications individually and 3 unit plant, total 63 seedlings.

Growth parameters

Growth parameters measured were plant height and diameter at a stem height of 1 cm above the soil media. At the final stage of this study, the number of leaves and nodules was counted. Harvested shoot and root were separated, then oven-dried at 70°C for 48 hours, and the dry weight was measured. Additionally, seed quality index (SQI) was assessed using the formula $SQI = [\text{Shoot dry weight} + \text{root dry weight}] / [(\text{height/diameter}) + (\text{dry weight of shoot/root dry weight})]$ suggested by Dickson et al. (1960). Seedlings were considered to have high quality when the value of $SQI \geq 0.09$, while Shoot Root Ratio (SRR) was calculated as the ratio of shoot dry weight to root dry weight. The Bray I/II and Kjeldahl methods were used to analyze the plant concentration of phosphorus (P) and nitrogen (N), respectively. Potassium (K) and zinc (Zn) concentration was determined with the $\text{HNO}_3\text{-HClO}_4$ method described by Carter (1993). The content of N, P, K, and Zn was determined by multiplying plant nutrient concentration with the dry weight. The increase or decrease of nutrient content in samples inoculated with AMF relative to the control was calculated by using $[\text{nutrient content of mycorrhizal plant} - \text{nutrient content of non-mycorrhizal plant}] / \text{nutrient content of non-mycorrhizal plant} \times 100\%$ (Wang et al. 2005).

AMF colonization and Mycorrhizae Inoculation Effect (MIE)

Pterocarpus indicus root was rinsed under running water, followed by a 24-hour soaking in 10% KOH, acidification for 30 minutes in 2% HCl, and staining with trypan blue. Colonies of AMF were determined using the formula $[\sum \text{number of fields of view colonized} / \sum \text{total observed field of view}] \times 100\%$ suggested by Brundrett et al. (1996). In this study, the Relative Field Mycorrhizal Dependency (RFMD) or Mycorrhizae Inoculation Effect (MIE) was calculated with $[\text{total dry weight of mycorrhizal plant} - \text{total dry weight of non-mycorrhizal plant}] / \text{total dry weight of mycorrhizal plant} \times 100\%$ applied by Plencette et al. (1983), then host species MIE was categorized using the procedure of Habte and Manjunath (1991). The Mycorrhizal Growth Responses (MGR) were using $[\text{total dry weight of mycorrhizal plant} - \text{total dry weight of non-}$

$\text{mycorrhizal plant}] / \text{total dry weight of non-mycorrhizal plant}] \times 100\%$ as suggested by Cavagnaro et al. (2003).

Statistical analysis

The F test for comparing means of variance was first implemented, followed by DMRT at a 95% confidence level when the F test results showed a significant effect.

RESULTS AND DISCUSSION

AMF colonization and plant growth

Pterocarpus indicus seedlings were inoculated with AMF in all treatments excluding the control, as shown in Table 2. Common AMF structures observed were internal and external hyphae as well as vesicles. *P. indicus* seedlings inoculated with AMF treatments experienced higher colonization compared to control. Shoot height, plant diameter, number of leaves, and nodules colonized by *G. claroideum* and *G. coronatum* were higher in the inoculated media. Treatments B and C did not show significant differences from E, while seedlings colonized by mixed AMF and control had similar length and width of leaves. The observed performance of the 3-month-old *P. indicus* seedlings is comprehensively presented in Figure 1.

The initial growth of 3-month-old *P. indicus* in this study was generally found to increase with AMF inoculation. Increased development in plant inoculated with AMF is attributed to the absorption of nutrients, specifically P, N, and other microelements (Smith and Read 2008). AMF expands root tissue by forming hyphae capable of covering a wider area for nutrient absorption as well as increases plant resistance to abiotic (drought and salinity) and biotic stresses (Begum et al. 2019). The presence of these fungi improves soil structure or aggregation through the production of glycoprotein compounds (glomalin), which can increase aeration and water retention (Zhou et al. 2023). Application of local AMF enhances the growth of *P. mooniana* on serpentine, coal spoil, and gold tailings media (Husna et al. 2016; 2019; 2021a,b), as well as *P. indicus* (Husna et al. 2021b), *Kalappia celebica* (Husna et al. 2021a), and *Nauclea orientalis* on gold tailings media (Tuheteru et al. 2017).

Glomus claroideum and *G. coronatum* are more effective in increasing plant growth. Husna et al. (2021b) reported these 2 fungi to significantly increase the growth and dry weight of *P. indicus* as well as *P. mooniana* in soil media and gold post-mining land. Additionally, AMF type was found to greatly influence the effect on plant growth in asphalt post-mining soil media, corresponding with the study by Wulandari et al. (2024).

AMF inoculation in this study increased the number of root nodules of *P. indicus* aged 3 months (Table 2). The high number of root nodules in plant inoculated with AMF is strongly associated with improved P uptake. The presence of local AMF and rhizobia in root nodules can synergize to promote enhanced plant growth and N acquisition (Liu et al. 2023). Observations from previous studies related to *P. mooniana* (Husna et al. 2016; 2017b; 2019; 2021a,b; Kusuma et al. 2022) and *P. indicus* (Husna

et al. 2021b) were consistent with the current results on the use of local AMF.

P. indicus colonized by *G. coronatum* had a heavier shoot and total dry weight than control and other treatments (Table 3), while treatments B, C, and F increased the shoot dry weight. MIE values ranged from 77.8% to 88.3% or exceeded 75% of the criteria outlined by Habte and Manjunath (1991), which reported the growth and success of *P. indicus* in asphalt post-mining soil media to highly dependent on the symbiosis with AMF. Mycorrhizal dependence shows the importance of this symbiotic relationship for plant growth and health. Additionally, the shoot-root ratio was not significantly different among the seedlings, and AMF treatments significantly affected SQI compared to the control. Enhanced growth of *P. indicus* inoculated with AMF was followed by increased dry weight, and this study showed that the dry weight of treated plant was higher. Plant can have better access to water and

nutrient through significant improvement of the root system. *Glomus* is an important genus of the phylum Glomerimycota with potential to increase plant growth due to possessing (i) a small spore size; (ii) the ability to sporulate (spore production) in various environmental conditions; (iii) the ability to adapt to various soil and climatic conditions, as well as (iv) the ability to produce inocula or propagules (Husna et al. 2015; Tuheteru et al. 2019, 2020b).

This study corresponds with previous reports about various post-mining land conditions and soil media, including *P. mooniana* cultivated in ultramafic (Husna et al. 2016) and coal spoil (Husna et al. 2019). In addition, high AMF dependency was observed on endangered species *Aquilaria malaccensis* and *Aquilaria crassna* (Turjaman et al. 2006), as well as *K. celebica* (Husna et al. 2021b). Taproot plant may depend more on AMF than counterparts with fibrous root systems (Yang et al. 2015).

Table 2. AMF colonization, shoot height, stem diameter, leaves parameter, and nodules of *P. indicus* cultivated with or without AMF at nursery conditions after 3 months of experiment

Treatment	Mycorrhizal colonization (%) [*]	Shoot height (cm) [*]	Stem diameter (mm) [*]	Leaf			Nodule [*]
				Number	Length (cm)	Width (cm)	
A	4.17 ± 4.17b	9.94 ± 0.66e	2.02 ± 0.03d	12 ± 0.52c	5.52 ± 0.29	3.30 ± 0.15	0.33 ± 0.33f
B	60.83 ± 13.41a	22.01 ± 1.51a	3.44 ± 0.10a	30 ± 2.25a	6.94 ± 0.36	4.31 ± 0.34	45.67 ± 3.38a
C	55.83 ± 3.00a	19.09 ± 1.00ab	3.60 ± 0.20a	29 ± 0.80a	6.85 ± 0.34	4.15 ± 0.14	37.00 ± 4.00ab
D	69.33 ± 10.24a	14.76 ± 0.80cd	2.69 ± 0.10c	24 ± 0.87ab	5.93 ± 0.37	4.07 ± 0.24	14.67 ± 3.673
E	55.0 ± 9.46a	13.49 ± 0.80d	3.37 ± 0.03ab	27 ± 6.50ab	5.64 ± 0.41	3.90 ± 0.26	22.00 ± 3.06de
F	50.83 ± 6.01a	18.37 ± 0.71b	3.11 ± 0.10b	30 ± 1.02a	6.05 ± 0.54	3.98 ± 0.35	33.00 ± 2.52bc
G	56.67 ± 5.07a	17.48 ± 1.29bc	3.09 ± 0.06b	19 ± 1.83bc	6.23 ± 0.16	4.11 ± 0.21	24.3 ± 2.33cd
Pr>F	0.0017	<.0001	<.0001	0.003	0.100	0.06024	<.0001

Notes: A: control (no inocula); B: *G. coronatum*; C: *G. claroideum*; D: *G. halonatum*; E: mixed AMF (*G. claroideum*, *G. coronatum*); F: mixed AMF (*G. claroideum*, *G. halonatum*); and G: mixed AMF (*G. coronatum*, *G. claroideum*, *G. halonatum*); mean values followed by different letters in the same column represent significant difference ($P < 0.05$); *Mean±SE

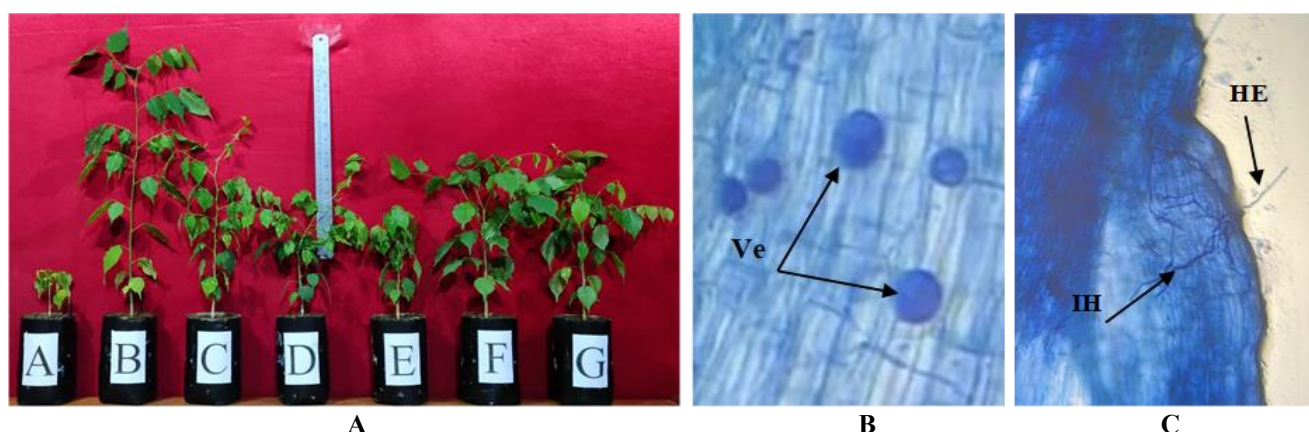


Figure 1. Performance of 3-month-old *P. indicus* seedlings (A) and AMF colonization (B and C). A: control (no inocula); B: *G. coronatum*; C: *G. claroideum*; D: *G. halonatum*; E: mixed AMF (*G. claroideum*, *G. coronatum*); F: mixed AMF (*G. claroideum*, *G. halonatum*); and G: mixed AMF (*G. coronatum*, *G. claroideum*, *G. halonatum*); Ve: Vesicle, IH: Intraradical hyphae; EH: Extraradical hyphae. AMF structure (Figures B and C) were observed under compound microscope and 100× magnification

Shoot and root nutrient concentration and content

Root concentration of N and P was not significantly different among seedlings, as presented in Table 4. Inoculation of *G. coronatum* and *G. claroideum* caused a higher Ca concentration in the root of *P. indicus*. All AMF inoculation increased root Zn concentration without showing significant differences between seedlings colonized by treatment G and control. This increased root N, P, Ca, and Zn, while *P. indicus* colonized by *G. coronatum* had higher root N and Ca content than other treatments (Table 4). Inoculation with *G. coronatum* and *G. claroideum* enhanced root P and Zn content of *P. indicus*.

Significant differences were not found in the shoot N, Ca, and Zn concentration of inoculated seedlings (Table 5). *P. indicus* colonized by mixed AMF (*G. claroideum* + *G. coronatum*), *G. halonatum*, and mixed AMF (*G. claroideum* + *G. coronatum* + *G. halonatum*) had higher shoot P concentration compared to other treatments. The entire AMF inoculation generally increased shoot N, P, Ca, and Zn content of *P. indicus*. *G. coronatum* and *G. claroideum* increased root N and K content, while *P. indicus* colonized

by these 2 fungi had higher root P content than control, *G. halonatum*, and mixed AMF (*G. claroideum* + *G. coronatum* + *G. halonatum*).

The increase and decrease of nutrient content

AMF inoculation increased nutrient uptake of *P. indicus* in both root and shoot compared to the control (Figure 2). Inoculation with *G. coronatum* and *G. claroideum* produced higher nutrient uptake values than other treatments.

N, P, Ca, and Zn content and uptake of the root and leaves of *P. indicus* were significantly enhanced by applying local AMF (Tables 4 and 5; Figure 2). The symbiosis of AMF with plant root is essential in supporting plant growth by increasing the uptake of water and P as well as other soil nutrients, including N, Ca, and Zn (Smith and Read 2008). Increased uptake of P and other nutrients has been reported in investigations related to the inoculation of local AMF with *P. mooniana* (Husna et al. 2016; 2017b; 2019; 2020; 2021a,b) and *P. indicus* (Husna et al. 2021b).

Table 3. Dry weight, MIE shoot-root ratio, and SQI of *P. indicus* cultivated with or without AMF at nursery conditions after 3 months of experiment

Treatment	Dry weight (g/plant)*			MIE (%)*	MGR (%)	Shoot root ratio*	ISQ*
	Roots	Shoots	Total				
A	0.35 ± 0.03e	0.15 ± 0.04d	0.50 ± 0.07e	-	-	2.43 ± 0.33	0.046 ± 0.00b
B	2.97 ± 0.09a	1.31 ± 0.10a	4.29 ± 0.15a	88.3 ± 1.19	758 ± 101.46	2.29 ± 0.17	0.206 ± 0.04a
C	2.58 ± 0.15b	1.12 ± 0.10ab	3.71 ± 0.18b	86.3 ± 1.93	642 ± 125.72	2.35 ± 0.28	0.196 ± 0.03a
D	1.69 ± 0.06d	0.67 ± 0.06c	2.36 ± 0.09d	78.3 ± 3.10	372 ± 51.13	2.57 ± 0.23	0.153 ± 0.00a
E	1.96 ± 0.06cd	0.88 ± 0.12bc	2.85 ± 0.17cd	82.4 ± 1.15	470 ± 51.97	2.31 ± 0.30	0.207 ± 0.01a
F	2.09 ± 0.13c	0.99 ± 0.09abc	3.08 ± 0.19c	84.5 ± 0.48	516 ± 43.99	2.13 ± 0.16	0.160 ± 0.01a
G	1.83 ± 0.14cd	0.76 ± 0.16c	2.59 ± 0.28cd	77.8 ± 2.01	418 ± 15.14	2.58 ± 0.40	0.143 ± 0.01a
Pr>F	<.0001	<.0001	<.0001			0.9139	<.0001

Notes: A: control (no inocula); B: *G. coronatum*; C: *G. claroideum*; D: *G. halonatum*; E: mixed AMF (*G. claroideum*, *G. coronatum*); F: mixed AMF (*G. claroideum*, *G. halonatum*); and G: mixed AMF (*G. coronatum*, *G. claroideum*, *G. halonatum*); mean values followed by different letters in the same column represent significant difference ($P < 0.05$); *Mean±SE

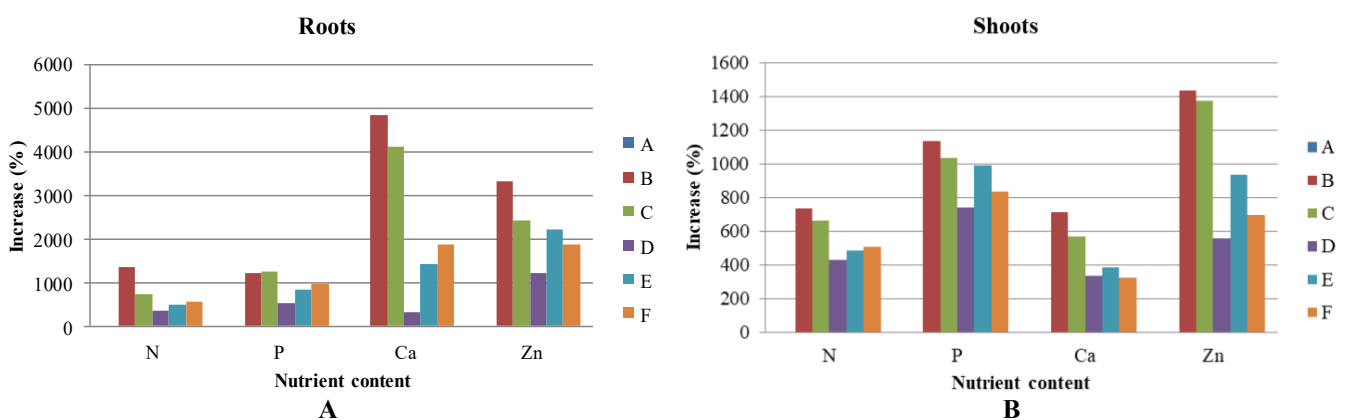


Figure 2. A. The increase or decrease of nutrient content in the root; B. Shoot of 3-month-old *P. indicus* seedlings at nursery conditions. Notes: A: control (no inoculum); B: *Glomus coronatum*; C: *Glomus claroideum*; D: *Glomus halonatum*; E: mixed AMF (*G. claroideum*, *G. coronatum*); F: mixed AMF (*G. claroideum*, *G. halonatum*); and G: mixed AMF (*G. coronatum*, *G. claroideum*, *G. halonatum*)

Table 4. N, P, Ca, and Zn concentration and content in the root of *P. indicus* cultivated with or without AMF at nursery conditions after 3 months of experiment

Treatment	Concentration (mg g ⁻¹) *				Content (mg plant ⁻¹ x10 ⁻³) *			
	N	P	Ca	Zn	N	P	Ca	Zn
A	2.23 ± 0.033	0.00029 ± 0.000012	0.017 ± 0.0008b	0.00023 ± 0.000038b	34.7 ± 0.033d	0.044 ± 0.0013d	2.72 ± 0.06e	0.03 ± 0.0021d
B	3.60 ± 1.150	0.00046 ± 0.000107	0.102 ± 0.0014a	0.00079 ± 0.000036a	4953.3 ± 0.000a	0.583 ± 0.0012a	134.35 ± 0.17a	1.03 ± 0.0146a
C	2.50 ± 0.058	0.00052 ± 0.000020	0.102 ± 0.0009a	0.00067 ± 0.000073a	2788.7 ± 0.058b	0.588 ± 0.0059a	114.3 ± 0.42b	0.76 ± 0.0169ab
D	2.33 ± 0.033	0.00041 ± 0.000035	0.016 ± 0.0074b	0.00057 ± 0.000094a	1567.3 ± 0.033c	0.280 ± 0.0033c	11.89 ± 0.14e	0.39 ± 0.0017c
E	2.26 ± 0.033	0.00046 ± 0.000015	0.043 ± 0.0202b	0.00074 ± 0.000207a	2000.7 ± 0.088bc	0.412 ± 0.0041b	41.18 ± 0.26cd	0.69 ± 0.0080bc
F	2.27 ± 0.033	0.00047 ± 0.000009	0.052 ± 0.0162b	0.00061 ± 0.000063a	2240.3 ± 0.088bc	0.469 ± 0.0035ab	53.43 ± 1.74c	0.59 ± 0.0245bc
G	2.27 ± 0.067	0.00043 ± 0.000049	0.033 ± 0.0213b	0.00049 ± 0.000098ab	1736.3 ± 0.067c	0.343 ± 0.0058bc	31.96 ± 3.34d	0.39 ± 0.0066c
Pr>F	0.3251	0.0760	0.0008	0.0313	<.0001	<.0001	<.0001	0.0003

Notes: A: control (no inocula); B: *G. coronatum*; C: *G. clarioideum*; D: *G. halonatum*; E: mixed AMF (*G. clarioideum*, *G. coronatum*); F: mixed AMF (*G. clarioideum*, *G. halonatum*); and G: mixed AMF (*G. coronatum*, *G. clarioideum*, *G. halonatum*); mean values followed by different letters in the same column represent significant difference (P < 0.05); *Mean±SE

Table 5. N, P, Ca, and Zn concentration and content in the shoot of *P. indicus* cultivated with or without Arbuscular AMF at nursery conditions after 3 months of experiment

Treatment	Concentration (mg g ⁻¹) *				Content (mg plant ⁻¹ x10 ⁻³) *			
	N	P	Ca	Zn	N	P	Ca	Zn
A	2.23 ± 75.12	0.00035 ± 0.01c	0.103 ± 0.68	0.00050 ± 0.00	781.3 ± 69.86c	0.12 ± 0.02c	36.10 ± 3.53d	0.17 ± 0.02c
B	2.20 ± 558.11	0.00049 ± 0.04b	0.098 ± 11.12	0.00087 ± 0.07	6548.7 ± 196.91a	1.48 ± 0.02a	292.96 ± 12.61a	2.61 ± 0.48a
C	2.30 ± 183.86	0.00052 ± 0.05b	0.093 ± 4.73	0.00095 ± 0.14	5950.0 ± 392.44a	1.36 ± 0.21ab	240.44 ± 10.25ab	2.51 ± 0.56a
D	2.43 ± 164.33	0.00059 ± 0.05ab	0.092 ± 1.96	0.00066 ± 0.10	4132.0 ± 204.86b	1.01 ± 0.09b	157.55 ± 3.52bc	1.12 ± 0.02bc
E	2.33 ± 53.01	0.00067 ± 0.01a	0.089 ± 5.87	0.00089 ± 0.17	4587.3 ± 281.80b	1.31 ± 0.10ab	174.38 ± 10.05b	1.76 ± 0.18ab
F	2.27 ± 177.17	0.0053 ± 0.04b	0.075 ± 7.29	0.00089 ± 0.08	4756.0 ± 463.21b	1.12 ± 0.14ab	153.15 ± 29.23bc	1.35 ± 0.46b
G	2.33 ± 250.15	0.00057 ± 0.06ab	0.034 ± 4.98	0.00079 ± 0.08	4274.7 ± 339.70b	1.05 ± 0.14b	69.89 ± 68.56cd	1.44 ± 0.11b
Pr>F	0.2130	0.0028	0.0611	0.2538	<.0001	<.0001	0.0003	0.0025

Notes: A: control (no inocula); B: *G. coronatum*; C: *G. clarioideum*; D: *G. halonatum*; E: mixed AMF (*G. clarioideum*, *G. coronatum*); F: mixed AMF (*G. clarioideum*, *G. halonatum*); and G: mixed AMF (*G. coronatum*, *G. clarioideum*, *G. halonatum*); mean values followed by different letters in the same column represent significant difference (P < 0.05); *Mean±SE

This study showed differences in the P, N, Ca, and Zn uptake ability of the plant root and shoot based on each AMF type. *G. claroideum* and *G. coronatum* only trigger lower P uptake compared to other AMF treatments, excluding the control. Walder and van der Heijden (2015) previously found different AMF species with varying capacities for P uptake. The currently obtained results confirmed a strong relationship between growth and dry weight as well as nutrient uptake in *P. indicus* treated with *G. claroideum* and *G. coronatum*.

In conclusion, this study found that AMF enhanced growth and improved nutrient uptake of *P. indicus* seedlings 3 months after transplantation at nursery conditions. AMF inoculation showed potential as a powerful tool for enhancing plant growth, nutrient uptake, and soil health, therefore considered crucial in the conservation efforts of endangered tropical plant and ecosystem management restoration.

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