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PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Pakistan Journal of Biological Sciences 18 (1): 1-10, 2015
ISSN 1028-8880

ansinet
Asian Network for Scientific Information

RESEARCH ARTICLE OPEN ACCESS

DOI: 10.3923/pjbs.2015.1.10

Diversity of Arbuscular Mycorrhizal Fungi in the Growth Habitat of Kayu Kuku (*Pericopsis mooniana* Thw.) In Southeast Sulawesi

[1]Husna, [2]Sri Wilarso Budi, [2]Irdika Mansur and [2]Dan Cecep Kusmana
[1]Department of Forestry, Faculty of Foresty and Environmental Science of Halu Oleo University, Kendari, 93121, Southeast Sulawesi, Indonesia
[2]Department of Silviculture, Faculty of Forestry, IPB, Dramaga Bogor 16680, Indonesia

ARTICLE INFO
Article History:
Received: October 25, 2014
Accepted: December 11, 2014

Corresponding Author:
Husna,
Department of Forestry,
Faculty of Foresty and Environmental Science of Halu Oleo University,
Kendari, 93121, Southeast Sulawesi,
Indonesia Tel: +6285241627519

ABSTRACT
Arbuscular Mycorrhizal Fungi (AMF) are categorized as fungi which have symbioses with terrestrial plants and are distributed in various habitat types. The objectives of this research were to investigate the diversity of AMF in stands of kayu kuku (*Pericopsis mooniana* Thw.) in Southeast Sulawesi. Collection of samples of soil and root were conducted in six locations. Isolation of spores used the method of wet sieving and decanting, whereas AMF identification was conducted by observing morphology of AMF spores. Parameters of AMF diversity, namely species richness, diversity index, dominance index, evenness index and colonization were studied using method of infected root length. Research results showed that location differences affected significantly the spore density and parameters of AMF diversity, except colonization of AMF ($p<0.116$). Location around the Governor office showed the highest number of spores (208.6 spores/100 g of soil). Soil chemical properties, such as C, N, P and heavy metal contributed towards AMF spore density and diversity. Soil C and N correlated negatively with spore density. In terms of location, Glomeraceae constituted the genera with the largest number of species and possessed wide distribution in all research locations. In general, natural forest has higher AMF diversity index (Shannon-Weiner diversity index-H'), evenness (E) and species richness (S) as compared with location of PT. Vale Indonesia Tbk.

Key words: Arbuscular mycorrhizal fungi, *Glomus*, *Pericopsis mooniana* Thw, mining

INTRODUCTION

Arbuscular Mycorrhizal Fungi (AMF) are obligate fungi which have symbioses with 97% of terrestrial plants (Smith and Read, 2008). Phylum Glomeromycota possesses four orders (Glomerales, Diversisporales, Paraglomerales and Archaeosporales), 11 families, 18 genera and around 300 species which have been recognized (Schuβler and Walker, 2010). Such symbioses are generally mutualistic, although they are sometimes parasitic depending on the species of plants and AMF (Klironomos, 2003). AMF possess fungal structures in the form of arbuscule, vesicle and hyphae which are found in roots and external hyphae and spores in soil (Smith and Read, 2008). AMF constitutes an important component in natural and artificial environment (Bainard *et al*., 2011). Host plants obtain nutrients and water from the fungi and as a return the fungi obtain carbon from photosynthesis products of the host plants (Smith and Read, 2008).

The role of AMF in maintaining and improving ecosystem productivity occurs in various ways, such as: (1) Improvement of community structure and plant ecosystem productivity (Van der Heijden *et al*., 2006; Wolfe *et al*., 2007), (2) Improving plant health and soil fertility (Jeffries *et al*., 2003), helping the absorption of nutrients and water and protecting the roots from root pathogen (Smith and Read, 2008) and (3) Improving plant resistance towards heavy metal pollution (Amir *et al*., 2008), salinity (Giri and Mukerji, 2004) and drought (Auge, 2004). AMF could improve soil structure and aggregation (Nichols, 2008) colonization of AMF could increase plant productivity (Koide, 2010).

AMF has a wide range of biodiversity and could be found in various ecosystems. These species live together with other organism communities existing in rhizosphere in various ecosystems, such as coastal areas (Delvian, 2003; Puspitasari, 2005), sandbar in South Brazil (Sturmer *et al*., 2013), natural and degraded forests (Cuenca *et al*., 1998), lowland forest ecosystem in Indonesian Ujung Kulon National Park (Kramadibrata, 2012) and natural forest and secondary forest and other land use types such as agricultural land, animal husbandry land and agroforestry land in western part of Brazilian Amazon (Sturmer and Siqueira, 2011) and peat swamp area (Ekamawanti, 1997). Besides occurrence in various ecosystems, AMF are also reported to be in symbiosis with various species of terrestrial plants which comprise among others plant species threatened with extinction (Wang and Qiu, 2006). Wang and Qiu (2006) in their review results reported that there were 139 plant species which were threatened with extinction, among the 2469 species which are associated with AMF, either in dry land or wet land. In Indonesia, forest tree species in IUCN list, have been reported to form symbiotic relationship with mycorrhizal fungi. The tree species among others are *ramin* (*Gonystillus bancanus*) (Tawaraya *et al*., 2003), *gaharu* (*Aquilaria* sp.) (Turjaman *et al*., 2006), *eboni* (*Diospyros celebica* Back) (Rahman and Abdullah, 2002) and kayu kuku (*Pericopsis mooniana* Thw.) (Husna *et al*., 2006).

Kayu kuku (*Pericopsis mooniana* Thw.) belongs to subfamily Papilionaceae and grows naturally in Kolaka District, Southeast Sulawesi (Whitmore *et al*., 1989; Surianegara and Lemmens, 1994) produces fancy wood (Surianegara and Lemmens, 1994). This species has been categorized as threatened species with extinction and with vulnerable status (VU A1c, d ver 2.3) (IUCN., 2014). Efforts to save *Pericopsis mooniana* Thw. from extinction should be conducted (Hardiyanto and Na'iem, 2001). Establishment of Lamedai Nature Reserve in Kolaka District constitutes the only form of *in-situ* conservation for *P. mooniana* Thw. which has been conducted in Indonesia, while those of *ex-situ* conservations have been conducted in several regions of Southeast Sulawesi (Sultra), such as in nickel post mining reclaimed land of PT Inco Tbk (now is referred to as PT. Vale Indonesia Tbk. Pomalaa), campus area of Halu Oleo University (UHO) and urban forests around the governor Office of Southeast Sulawesi province.

Study on diversity of AMF of *P. mooniana* Thw. in Indonesia has been conducted. In *Pericopsis mooniana* Thw. which grow in Nature Reserve Lamedai Kolaka, there were generally found 4 genera of AMF, namely *Glomus*, *Scutellospora*, *Acaulospora* and *Gigaspora* (Husna *et al*., 2006). However, such study is still confined to the identification of AMF and has not covered the study on their diversity. Therefore, to obtain comprehensive information on diversity of AMF in *Pericopsis mooniana* Thw. rhizosphere, there is a need for study on AMF diversity in various *P. mooniana* Thw. growth sites in Southeast Sulawesi.

The objectives of this research were to study the diversity, colonization and density of AMF spore in *P. mooniana* Thw. stands in their distribution area in Southeast Sulawesi.

MATERIALS AND METHODS

Time and locations of research: Research was conducted for three months from February to April. Locations of soil sample collection in the natural habitat of kayu kuku, Lamedai Nature Reserve, Lamedai village (04°18'44.6'' S-121°3'22.8''E) and natural forest in Tanggetada village (04°0'44.6''S-121°2'18.0"E) and *Pericopsis mooniana* Thw. plantation forests which were located in Bali Jaya village (04°7'57.6''S-121°2'59.2"E). The three places belong to sub district of Tanggetada. Soil samples were also collected from *Pericopsis mooniana* Thw. revegetation area in block of zone III of Nirwana of PT. Vale Indonesia (Tbk) in Pomalaa subdistrict (04°2'30.2''S-121°38'57.6" E), Kolaka district; Campus of Halu Oleo University (UHO) (04°00'17.1''LS-122°31'06.1"BT) and urban forest around Southeast Sulawesi Governor Office, both of them in Kendari (04°01'19.5''S-122°32'17.5"E). Analysis of soil physical and chemical properties were conducted in Soil and Plant Laboratory of SEAMEO BIOTROP. Isolations of AMF spores were conducted in Biotechnology Research Center of IPB and identification of AMF species was conducted in Laboratory of Low Level Plants (Biological Research Center, Indonesian Institutes of Sciences) and Laboratory Wood Anatomy (Forest Research and Development Center, Bogor).

Exploration of mycorrhizal fungi

Collection of soil samples: Soil samples were collected randomly under the trees or stand of *P. mooniana* soil depth of 0-20 cm, as many as ten trees at each location. At each tree with a distance of 1-2 m from the based of the trees, four soil sample points were collected: West-East, North-South, it was taken as 250 g at each point and totally of samples weigh was 1 kg each tree. Soil samples were filled and labelled into plastic bags. All of soil samples for AMF isolation, identification and nutrients soil analysis were dried in the Laboratory.

Colonization of mycorrhizal fungi: For the first step, fresh fine roots were selected from sample plants. Afterwards, those roots were put inside KOH 10% solution for 24 h at room temperature. The KOH solution was afterwar discarded and the root samples were washed with flowing water until became clean. Roots were then soaked in HCl 2% solution for 30 min. The HCl solution was discarded. The root samples were subsequently soaked in staining solution (trypan blue 0.05%+glycerol 70%+aquadest 30%) for 24 h. The roots were washed again and put into glycerol 50% solution.

Enumeration of AMF colonization used the method of infected root length observation (Giovannetti and Mosse, 1980). From the roots which had been stained, ten root

samples which had length of ± 1 cm were taken and arranged in preparation glasses. Root segments in the preparation glasses were observed for each angle of view. Fields of view which showed colonization were marked with (+), whereas those which did not, were marked with negative sign (-).

Isolation of spores: Technique being used for AMF spore isolation was sieving and decanting method from Pacioni (1992) and was followed with centrifugation technique from Brundrett *et al*. (1996). The steps being conducted were as follows: (1) Mixing soil sample as much as 50 g with 200-300 mL of water and stirred, (2) The mixture was sieved in a set of sieves with sizes of 670, 125 and 45 μm in ordered sequence from upper to lower side, (3) Materials which passed the sieve of 125 and 45 μm were afterwards transferred to centrifuge tube, added with glucose 60% (w/v), (4) Centrifuge tube was tightly closed and centrifuged at speed of 2500 rpm for 3 min, (5) supernatant was decanted into sieve of 45 μm and washed with flowing water to eliminate glucose and (6) The remaining precipitate was decanted to Petri dishes and observed under compound microscope with 200X magnification to enumerate spore population and to make preparation for identification of AMF spores being found.

Identification of mycorrhizal fungi: Identification of AMF spore was performed by morphological observation color, shape, size, hyphal attachment, spore ornamentation and spore reaction towards Melzer's solution (Schneck and Perez, 1988; Schuβler and Walker, 2010). Spore enumeration was conducted under stereo microscope and spore identification was conducted under microscope Axio Imager A_1m/Axiocam MRc5 with 200 X magnification.

Parameters: Parameters of AMF diversity observed in this research include frequency of isolation, relative abundance, importance value, spore density, species richness, spore diversity (Shannon-Wiener diversity index, evenness and Simpson's index) (Yang *et al*., 2011) and root colonization (Brundrett *et al*., 1996) as shown in Table 1.

Data analysis: Data was analyzed using analysis of variance (F test) on variables such soil chemical properties, spore density and AMF colonization, species richness, Shannon-Weiner indexes, Evenness and Simpson Index. If test results showed significant effect, then there would be test of treatment differences using LSD at level of 95%. Correlation between soil chemical properties and spore density was conducted by using Pearson's correlation.

RESULTS

Soil properties: Soil physical and chemical properties in the research locations are presented in Table 2. Table 2 shows that the highest pH, C and total N were found in location of PT. Vale Indonesia Tbk. However, PT. Vale Indonesia Tbk also showed smaller amount of available P (0.94 ppm) as compared with those of other locations. Distribution area of *P. mooniana* Thw. which is dominated by sand fraction, except the location of PT. Vale Indonesia Tbk.

Colonization and density of AMF spores: All roots of the surveyed *P. mooniana* Thw. were colonized by local AMF. AMF structures found in roots were internal hypha, external hypha, hyphal coil, vesicles and arbuscules. Internal hypha structure was found to be dominant (77%) in the roots of *P. mooniana* Thw. in all locations, followed by that of vesicle

Table 1: Parameters of AMF variability and the calculation techniques

Parameters	Formula
Frequency of isolation (FI)	[number of samples where species and genera of AMF were found /total number of sample]×100%*
Relative Abundance (RA)	Percentage of number of spores from each species or genera*
Importance Value (IV)	(FI + RA)/2. If IV≥20 it is categorized as dominant species or genera*
Spore density	Number of spores per 100 g of soil*
Species richness	Number of species for each soil sample*
Shannon-Weiner diversity index	$H' = -\sum pi \ln pi$*
Evenness	$E = H'/H'max$
Simpson's index	$D = \sum[n_i(n_i-1)/N(N-1)]$*
AMF colonization	$\frac{\sum \text{field of view with mycorrhiza}}{\text{Total number of field of view observed}} \times 100\%$**

$Pi = n_i/N$, where n_i is the number of spore per species and N is total number of spores being identified. H'_{max}=ln S, where S is total number of species being identified. *Shi *et al*. (2006), Yang *et al*. (2011) and **Brundrett *et al*. (1996)

Table 2: Soil physical and chemical properties in various locations *P. mooniana* Thw. distribution in Southeast Sulawesi

	Soil chemistry				Soil texture		
Habitat	pH	C org (%)	N Tot (%)	P available (ppm)	Sand (%)	Silt (%)	Clay (%)
KG	4.5±0.09^{c}*	0.56±0.05^{d}	0.09±0.01^{c}	2.9±0.22^{bc}	61±0.31^{ab}	19±0.30^{bc}	21±0.28^{b}
UHO	5.1±0.12^{b}	1.04±0.08^{c}	0.16±0.03^{b}	4.26±0.29^{ab}	46±0.58^{b}	32±0.50^{ab}	22±0.47^{b}
Vale	5.8±0.09^{a}	2.43±0.10^{a}	0.35±0.04^{a}	0.94±0.13^{c}	13±0.33^{c}	42±0.35^{a}	45±0.42^{a}
CA	4.3±0.12^{c}	1.33±0.13^{b}	0.17±0.04^{b}	6.36±0.24^{a}	69±0.85^{a}	12±0.60^{c}	19±0.64^{b}
BJ	4.1±0.09^{c}	0.88±0.07^{cd}	0.13±0.03^{bc}	6.24±0.25^{a}	57±0.46^{ab}	21±0.49^{bc}	23±0.40^{b}
HA	4.2±0.08^{c}	1.00±0.06^{bc}	0.13±0.01^{bc}	5.38±0.22^{ab}	68±0.75^{a}	16±0.66^{c}	16±0.51^{b}
Pr>F	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

KG: Governor office, UHO: University of Haluoleo, Vale: PT. Vale Indonesia Tbk, CA: Nature reserve, BJ: Bali Jaya and HA: Natural forest, *Average ±SE

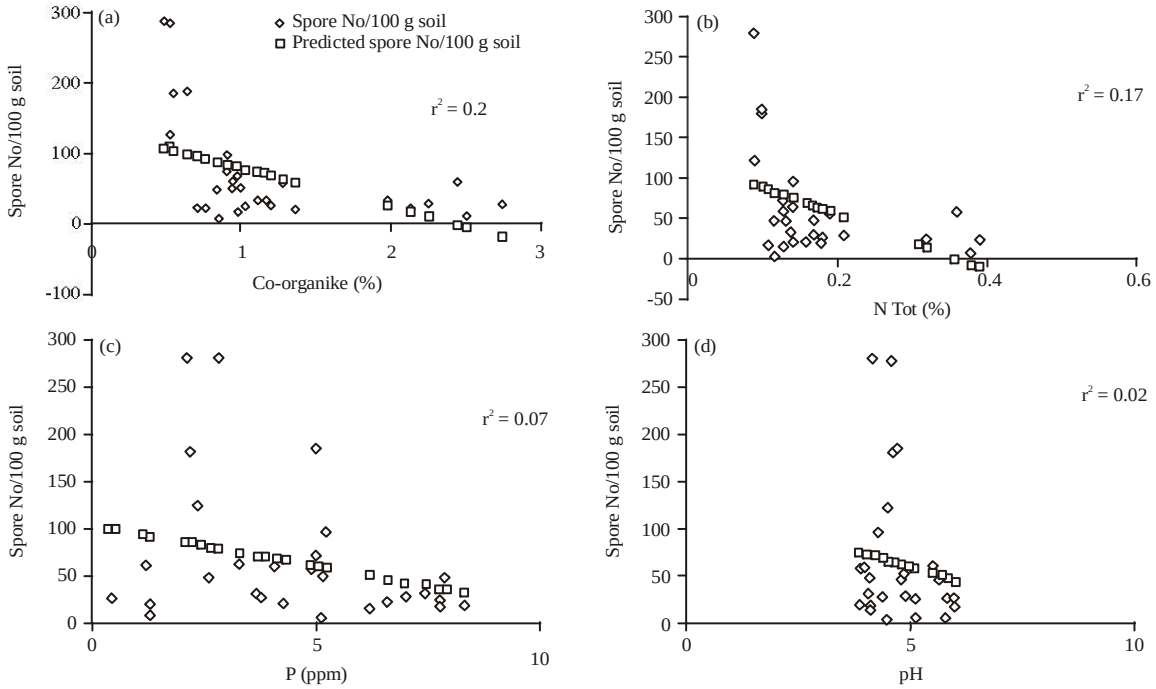

Fig. 1(a-d): Relation between soil properties and density of AMF spores, (a) Co-organic (%), (b) Total N (%), (c) Available P (ppm) and (d) pH

Table 3: Colonization and Structure of AMF in various habitats

		AMF structure				
Location	Colonization (%)	Internal hypha	External hypha	Vesicle	Arbuscule	Hyphal coil
KG	71±0.51^{a*}	77.0	3.3	17.9	0.0	1.8
UHO	60±0.63^{a}	81.3	7.7	9.0	0.0	2.0
Vale	70±0.76^{a}	72.4	6.2	19.2	0.9	1.4
CA	59±0.56^{a}	77.4	5.0	15.9	0.6	1.2
BJ	57±0.63^{a}	83.2	2.5	11.6	1.1	1.8
HA	73±0.75^{a}	67.4	9.3	18.0	1.5	3.8
	-	76.4	5.7	15.3	0.7	2.0

KG: Governor office, UHO: University of Haluoleo, Vale: PT. Vale Indonesia Tbk, CA: Nature reserve, BJ: Bali Jaya and HA: Natural forest, *Average ±SE

Table 4: Spore density, species richness, shannon-Weiner diversity index, evenness and simpson's index of AMF in various habitats

Locations	Spore density/100 g of soil	Species richness	H'	E	D
KG	208.6±1.66^{a*}	4.6±0.15^{ab}	1.05±0.09^{ab}	0.69±0.07^{a}	0.39±0.06^{ab}
UHO	41.2±0.77^{b}	4.8±0.18^{ab}	1.18±0.09^{a}	0.76±0.07^{a}	0.30±0.04^{ab}
Vale	25.6±0.87^{b}	3.4±0.23^{b}	0.68±0.11^{b}	0.46±0.08^{b}	0.52±0.09^{a}
CA	37.8±0.97^{b}	4.8±0.18^{ab}	1.40±0.08^{a}	0.90±0.04^{a}	0.25±0.04^{b}
BJ	13.8±0.51^{b}	3.6±0.19^{ab}	1.08±0.07^{ab}	0.86±0.07^{a}	0.27±0.08^{b}
HA	57.4±0.98^{b}	5.4±0.21^{a}	1.37±0.10^{a}	0.82±0.07^{a}	0.29±0.06^{ab}
Pr>F	<0.0001	0.023	0.001	<0.00	0.023

KG: Governor office, UHO: University of Halu Oleo, Vale: PT. Vale Indonesia Tbk, CA: Nature reserve, BJ: Bali Jaya and HA: Natural forest, *Average ±SE, H: Shannom-Weiner index, E: Evennes dan D: Simpson's index

(15%) and the least was that of arbuscule (0.7%) (Table 3). Colonization of AMF was not affected by difference in growth site ($p>0.05$) and there was no correlation between soil properties and AMF colonization (Table 3, 5). In this research, AMF spores were found with greatest abundance in location of Governor office (208.6 spores/100 g of soil). Organic C and total soil N were negatively correlated with AMF spore density (Table 5). Correlation between C and N was relatively high as compared with those of pH, available P and soil structure ($r^2 = 0.23$ and 0.17) (Fig. 1a-d). Colonization was not correlated with number of spores ($r = 0.141$; $p>0.467$).

Diversity of AMF: Total number of AMF species found in the rhizosphere of *P. mooniana* Thw. was as many as 11 species which belonged to 4 families and 7 genera, namely *Glomus* (4 species), *Acaulospora* (2 species) and *Rhizophagus*,

Septoglomus, *Claroideoglomus*, *Racocetra* and *Scutellospora* which comprised one species of AMF, respectively. *Glomus* cf. *canadense* and *Claroideoglomus etunicatum* (syn. *Glomus etunicatum*) constituted the dominant AMF species in the whole research area (Table 6). *Racocetra gregaria* (syn. *Gigaspora gregaria*) was found to be dominant in PT. Vale Indonesia Tbk and that of *Scutellospora* cf. *Auriglobosa* was dominant in natural forest. Habitat variation affected significantly the species richness. Natural forest habitat possessed the highest species richness although it did not differ significantly with other habitat, except with location of PT. Vale Indonesia Tbk (Table 4). Species richness were found to be 2-7 individuals per soil sample which ranged between 3.4 in PT. Vale Indonesia Tbk and 5.4 in Natural Forest (Table 4). Besides possessing low species richness, PT. Vale Indonesia Tbk also possessed low species diversity (Shannon-Wiener diversity index) and low evenness (E), namely 0.68 and 0.46, respectively. However, PT. Vale Indonesia Tbk possessed higher Simpson's Dominance Index (D) and did not differ significantly with those of Governor Office, Campus of Halu Oleo University and Tangketada Natural Forest.

At family level, relative density and frequency varied greatly between families at various locations. The highest FI and RA were shown by family Glomeraceae, followed by Claroideo Glomeraceae, Gigasporaceae and Acaulosporaceae. Glomeraceae was categorized as family with FI 100% at all research locations. FI from family Acaulosporaceae ranged between 10-30% at locations ofHalu Oleo University Campus, CA Lamedai and Bali Jaya (Fig. 2). For variable RA, family Glomeraceae was dominant with range between 57-71% at all locations. Unlike Glomeraceae, RA values for the other three families showed different trends. In terms of variable RA, Gigasporaceae showed the highest value in location of PT. Vale Indonesia Tbk (18.28±0.98%), whereas that of Acaulosporaceae being highest in Bali Jaya (11.3±0.913), while Claroideoglomeraceae showed highest RA in Governor Office (42.2±0.71%) followed by location of natural forest Tangketada with value of 35.4±0.96% (Fig. 3a-d).

DISCUSSION

Research results showed that roots of *P. mooniana* Thw. in six habitats in Southeast Sulawesi were colonized by AMF. Percentages of AMF colonization did not differ in all habitats, with percentage >50%. Colonization of AMF was marked with the finding of AMF structure in roots. AMF structure being found were internal hypha, external hypha, vesicle and arbuscule. Each structure possessed different roles

Table 5: Analysis of correlation (r) and regression (r^2 = figures in brackets) between environmental factors and AMF symbiosis of *P. mooniana* Thw. plants

Prarmeters	pH	C	N	P	Sand	Silt	Clay
Spore density	-0.12 (0.018)	-0.48** (0.23)**	-0.49** (0.17)*	-0.27 (0.07)	0.21 (0.04)	-0.21 (0.04)	-0.19 (0.04)
Colonization	0.04 (0.002)	0.08 (0.006)	0.07 (0.006)	-0.10 (0.01)	0.01 (0.0002)	-0.03 (0.0008)	0.006

** Highly significant with p<0.01

Table 6: Species of AMF in the rhizosphere of *k* in various growth sites

Species	KG	UHO	Vale	CA	Bali Jaya	HA
Glomeraceae						
Rhizophagus						
Rhizophagus diaphanum (Morton and Walker) Walker and Schussler	+	+	+*	+	+*	
Glomus						
Glomus cf. *versiforme* (Karst.) Berch	+*	+*		+	+*	+*
Glomus cf. *canadense* (Thaxt.)	+*	+*	+*	+*	+*	+*
Trappe and Gerd.						
Glomus cf. *boreale* (Thaxt.)						
Trappe and Gerd.					+	
Glomus cf. *halonatum* rose and trappe	+			+		+*
Septoglomus						
Septoglomus constrictum (Trappe)	+	+*	+	+*	+	
Sieverd. Silva and Oehl						
Claroideoglomeraceae						
Claroideoglomus						
Claroideoglomus etunicatum	+*	+*	+*	+*	+*	+*
(Becker and Gerd.) Walker and Schussler						
Acaulosporaceae						
Acaulospora						
Acaulospora scrobiculata Trappe		+			+	
Acaulospora cf. *delicata*						
Walker, Pfeitt and Bloss	+*					
Gigasporaceae						
Racocetra						
Racocetra gregaria Souta and Sieverd.	+		+*	+		+
Scutellospora						
Scutellospora cf. *auriglobosa* (Hall.)						
Walker and Sanders	+	+	+	+		+*

KG: Governor office, UHO: University of Halu Oleo, Vale: PT. Vale Indonesia Tbk, CA: Nature reserve, BJ: Bali Jaya and HA: Natural forest, +The species was found, *NP ≥ 20

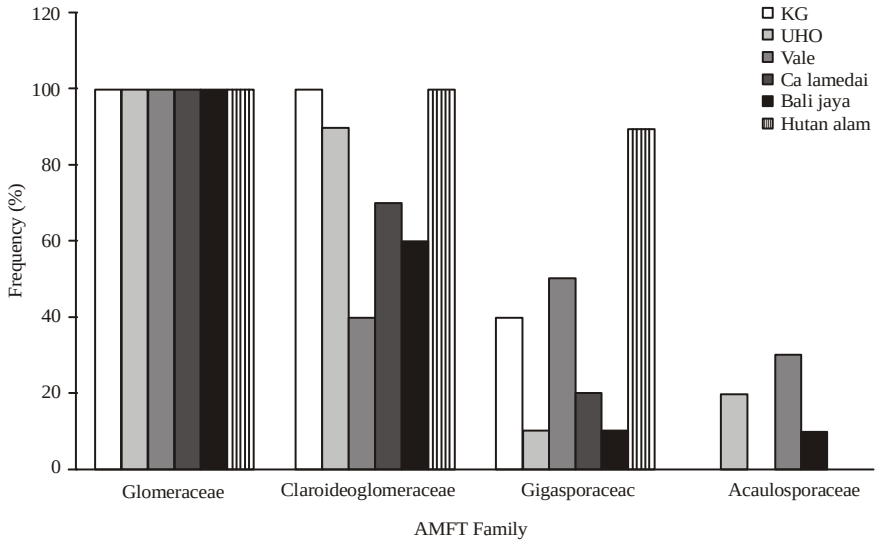

Fig. 2: Frequency of AMF families in various research locations

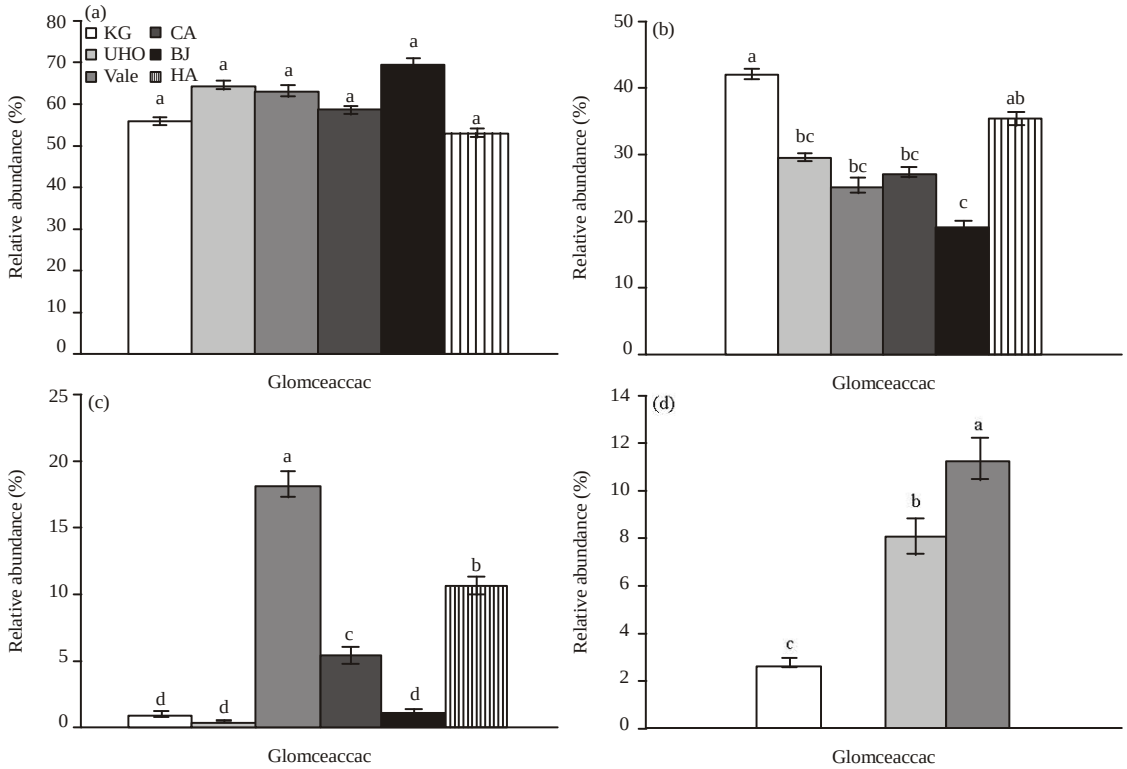

Fig. 3(a-d): Relative densities of AMF families in various research locations, (a) Glomeraceae, (b) Claroideoglomeraceae, (c) Gigasporaceae and (d) Acaulosporaceae

(Smith and Read, 2008). Colonization of AMF which occurred in all habitats showed that AMF possessed ability to colonize *P. mooniana* Thw. in various habitats.

Differences in habitat affected significantly the spore density ($p<0.001$). Number of spores ranged between 14-208 spores per 100 g of air dried soil. The greatest number (density) of spores per 100 g of air dry soil in *P. mooniana* Thw. stand was found in the office of Southeast Sulawesi Governor, Kendari (Table 4) (208 spores). The high density of AMF spores in the area (98% of spore were those of *Glomus* as the dominant species) was due to low soil fertility. This was proven from the correlation analysis of organic C and total N which were negatively correlated with spore density (Table 5) and also from the phenomenon of low content of

available P (Table 2). Negative correlation between soil organic C and AMF spore density was also reported by Gai *et al.* (2012). In relation with P availability in soil, several research results reported that the number of AMF spores were high at condition of low P availability (Lee *et al.*, 2009; Birhane *et al.*, 2010; Tian *et al.*, 2011). This was in agreement with Fig. 1d which shows the trend of decrease in number of spores in line with increase in available. Verma *et al.* (2008) and Pande and Tarafdar (2004) reported that areas with low soil fertility could increase the population of AMF. Muthukumar and Udaiyan (2002) reported that high availability of soil N could suppress the AMF colonization and spore production. This explanation was supported by Powell and Bagyaraj (1984) which showed that soils which were less fertile would decrease the root phospholipid membrane, increase root cell permeability, followed by leaking in roots and resulted in decrease in carbohydrate and amino acid and these phenomena would accelerate the formation of mycorrhiza. However, soil properties did not affect the level of AMF colonization in roots of *P. mooniana* Thw. in various research locations (Table 2 and 5). There was no correlation between AMF colonization and spore density. This was in agreement with statement of Ragupathy and Mahadevan (1991) that there was no correlation between number of spores and percentage of root colonization in host plants.

Total number of AMF species being found were 11 species and were categorized into seven genera, namely *Glomus*, *Rhizophagus*, *Septoglomus*, *Claroideoglomus*, *Racocetra Acaulospora* and *Scutellospora*. Total number of AMF species being found was smaller than those which had ever been reported for species *P. mooniana* Thw. (Husna *et al.*, 2006), *cacao* (Kramadibrata, 2009) and *Citrus retriculata* (Wang *et al.*, 2013). The genus *Glomus* dominated AMF species, wherein 5 species possessed high relative density and frequency in all research locations. According to Mosse (1981), naturally, the genus *Glomus* possessed very large distribution. Besides that, *Glomus* was categorized as genus with very many species of AMF (INVAM., 2003; Schuβler and Walker, 2010). *Glomus* also constituted the kind of AMF which was abundantly found in various types of habitat (Cuenca *et al.*, 1998; Sturmer and Siqueira, 2011; Gai *et al.*, 2012) and in various kinds of plant in terrestrial region (Shi *et al.*, 2006; Kramadibrata, 2009; Wang *et al.*, 2013) and in wet land (Miller and Bever, 1999; Escudero and Mendoza, 2005).

Domination of *Glomus* was also reported in various types of forest ecosystem and agriculture landin Indonesia. There were found 4 species of *Glomus* in natural forest ecosystem with pH of 3,89-6,10 in Gunung Halimun National Park (Suciatmih, 2002), 6 species of *Glomus* which associated with various plants in Ujung Kulon National Park (Kramadibrata, 2012), 7 species of *Glomus* in rhizosphere of soybean crop in Lampung and West Java (Kramadibrata *et al.*, 1995). Four species of *Glomus* in rhizosphere of Bisbul (*Diospyros blancoi*) in Bogor (Ningsih *et al.*, 2013) and 5 species of *Glomus* which were found in rhizosphere of 4 species of bamboo in Java island (Kramadibrata *et al.*, 2007).

Besides spore density, difference in location also affected the component of AMF diversity. Research results showed that natural habitat tended to possess higher species richness and AMF diversity as compared with those of *P. mooniana* Thw. development habitat. Plantation of *P. mooniana* Thw. such as that at PT. Vale Indonesia Tbk. revegetation area possessed low species richness, index of species diversity (Shannon-Wiener diversity Index) and Evenness (E). The low diversity of AMF at PT. Vale Indonesia Tbk. location was due to change in landscape and poor composition of vegetation species. Several study reported that number of spores and AMF diversity were low in soil which were heavily contaminated with heavy metal (Del Val *et al.*, 1999; Khan *et al.*, 2000). Besides *Glomus*, genus *Racocetra* (*Gigaspora gregaria*) was also dominant (importance value >20) and possessed highest frequency in PT. Vale Indonesia Tbk. Regvar *et al.* (2006) and Pawlowska *et al.* (1997) reported that *Gigaspora* was found in land contaminated with heavy metal.

Besides that, low value of species richness in PT. Vale Indonesia Tbk. and that of high value in natural habitat such as in natural forest Tangketada were probably related with species richness of the plants (Lee *et al.*, 2009; Kivlin *et al.*, 2011; Sturmer and Siqueira, 2011). However, plant species richness is not always correlated with species richness of AMF (Cuenca *et al.*, 1998). Species richness of AMF (5,5 species) in natural forest Tangetada Kolaka was still lower than those in young secondary forest in Brazil Amazon forest (12.8) (Sturmer and Siqueira, 2011) riparian forest in La Gran Sabana Venezuela (9) (Cuenca *et al.*, 1998) and forest in Mount Segrila Tibet (10 species) (Gai *et al.*, 2012). In general, distribution of AMF was greatly affected by many environmental factors such as soil type and texture, land degradation, humidity, temperature and nutrient availability (Kivlin *et al.*, 2011). Rillig *et al.* (2002) confirmed that the magnitude of abiotic factors, namely climate and soil properties which combined with other factors such as composition of host communities and intra and interspecific interaction, constituted the important factors determining the extent of AMF distribution.

Results of this study showed that difference in habitat of *P. mooniana* Thw. affected the species richness and AMF diversity. This was in agreement with statement of Opik *et al.* (2008) and research results of Opik *et al.* (2006) which showed that number of AMF species per species of host in particular location was possibly different between regions and habitat types. This statement was in agreement with research results of Wubet *et al.* (2006) which found differences in species richness of AMF species in rhizosphere of *Juniperus procera* in different locations and there was found variation of AMF species richness in two research locations in the rhizosphere

of *Prunus africana*. Opik *et al.* (2006) concluded further that AMF species richness was higher in natural forest as compared with other habitats (ecosystems), such as disturbed habitat due to human activities. The same phenomenon was reported by Wubet *et al.* (2006) which showed that AMF diversity was found to be high in natural habitat.

Variation of population and AMF diversity was affected greatly by variation of soils (mainly soil chemical properties), environmental condition (temperature and season), kinds of host and destruction regime (Opik *et al.*, 2006; Wubet *et al.*, 2004, 2006; Verma *et al.*, 2008). In relation with aspect of habitat destruction, Johnson *et al.* (2013) explained that high intensity land use could change soil properties and had implication on decrease of species richness and AMF diversity. Species richness and AMF diversity were probably related with host species, life cycle and specific site condition (Opik *et al.*, 2006).

Study on species richness variation and AMF diversity in various locations and rhizosphere in Indonesia, had been conducted. Difference in species richness and AMF diversity based on location had been reported in rhizosphere of soybean (Kramadibrata *et al.*, 1995). *Rambutan* (Muliawan *et al.*, 2002), bamboo (Kramadibrata *et al.*, 2007) and Bisbul (*Diospyros blancoi*) (Ningsih *et al.*, 2013). Besides location and rhizosphere, difference between natural habitat and degraded habitat, affected also the existence of AMF. Sabaruddin (2004) and Suciatmih (2002) reported that forests (natural habitats) possessed species richness and AMF diversity which were higher than disturbed habitat (agricultural land).

Table 6 shows that the genus of *Glomus* (*Glomus* cf. *Canadense* and *Claroideoglomus etunicatum*) dominated the distribution and diversity of AMF in growth sites of kayu kuku in Southeast Sulawesi. Domination of the two *Glomus* were possibly due to several factors which were among others: (1) The two species possessed small spore sizes, (2) Possession of sporulation (spore production) ability at various environmental conditions, (3) Possession of adaptive capability in various conditions of soil and climate and (4) Ability to produce inoculum (propagule) in rhizosphere of *P. mooniana* Thw. (Shi *et al.*, 2006; Verma *et al.*, 2008; Kivlin *et al.*, 2011; Shukla *et al.*, 2013).

On the basis of research results it could be concluded that difference in growth sites of *Pericopsis mooniana* Thw. affected the spore density of Arbuscular Mycorrhizal Fungi (AMF) and parameters of AMF diversity, except the AMF colonization. Soil chemical properties, such as C, N, P and heavy metal contributed toward spore density and component of AMF diversity. Soil C and N correlated negatively with spore density. Glomeraceae constituted the genus with the greatest number of species and possessed wide distribution in all research locations. Natural forests possessed high diversity index (Shannon-Weiner diversity index-H), evenness (E) and species richness as compared with those of PT. Vale Indonesia Tbk.

ACKNOWLEDGMENTS

The authors would like to thank DIKTI/Ministry of Education and Culture scholarship, PT. Vale Indonesia Tbk. and Bogoriense herbarium laboratory.

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