

## Exploration Diversity of Arbuscular Mycorrhizal Fungal (AMF) Spores Indigenous at Coal Mine Area PT. KIM, Job Site Muara Bungo, Jambi

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### ABSTRACT

Tolerant plants that can still be living in the mining area, because of the possibility of symbiosis with microorganisms, including mycorrhizal types, the spores of living microorganisms in the root zone through the formation of roots and symbiotic hyphae penetrate the root epidermis cells. Interwoven hyphae that will serve to expand the field of far-reaching absorption of nutrients from the soil surface, especially the elements and phosphate can also act as a filter absorption of heavy metals by plants. Research on the exploration diversity of indigen MAF spores conducted in five locations (at Wika 1, Kelok S, Kampung Jawa, KIM 2, and East Pit ) mining area coal PT. KIM in Muara Bungo, Jambi, This research consists of two phases, the first phase of soil sampling in the field and the second stage is the isolation and identification of AMF infection as well as observations in the laboratory, analyzed the data description. The results showed that in the area; The MAF spores found were : 13 of *Glomus* , *Septoglomus constrictum*, *Rhizophagus Clarus*, and *Septoglomus deserticola* species. While the infected MAF on plant are: *Chromolaena odorata* (60%), *Mallotus panniculatus* (60%), *Cyperus sp.* (40%), *Hevea brassiliensis* (60%), *Axonopus Paspalum* (90%), *A.compressus* (80%), *Ipomea triloba* (70%) and *Clidemia hirta* (100%).

**Key words:** Spore, MAF, indigen, coal mine.

### INTRODUCTION

The impact of mining activities in the form of changes in the type of land cover / top soil due to land clearing. then the land becomes vacant, hard and dry and soil compaction occurs, so the ability to with stand low water, very poor nutrients such as nitrogen and phosphorus, the accumulation of toxic elements, and the soil pH reaction becomes sour. This is a common phenomenon that is encountered on land after mining such as coal mines (Margarettha, 2011). Acid soil conditions also disturb the balance of plant nutrients, while the solubility of the elements that poison the plant increases, and the concentration of Phosphate (P) is very low. This is due to the occurrence of P sorption were formed by ions of Al, Fe, and Ca to form the compound of Al-P, Fe-P and Ca-P were backwardly anmobil, so it can not be used by plants. (Tisdale at al., 1985).

In the critical condition of land, there are some plants that are tolerant are likely caused by the presence of symbiotic microorganisms. One of the microorganisms that can support the growth of plants are arbuscular mycorrhizal fungi (JMA). The fungi are microorganisms that live in symbiosis with rhizosfir plant roots. On heavy metal contaminated soil, mycorrhizal serves as a filter to prevent the plants develop symptoms of poisoning due to the accumulation of heavy metals

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(Gaur and Adholeya, 2004). In addition, these fungi also improve plant nutrition by increasing root absorption field (Prasad et al., 2010). The specialty of this fungus is its ability to help the plants to absorb nutrients, especially P nutrients through the external hyphae that infect plant roots forming internal hyphae, arbuscular and vesicles, so the plants get nutrition, the mechanism that is what helps the growth of plants although in environmental conditions in critical areas such as coal mining or disposal. This statement was supported by research and Malajczuk Dell (1998) who explains that the very poor nutrient environments or environments contaminated hazardous waste, mycorrhizal fungi are still showing their existence, these things can be applied in dealing with phytoremediation (Quoreshi, 2008). And according to Prasad et al., (2010) the use of indigenous arbuscular mycorrhizal types will increase the success of phytoremediation. Morton and Benny (1990) in Adyari, (2013). said that the arbuscular mycorrhizal fungus spores (AMF) is divided into several genera, namely *Gigaspora*, *Scutellospora*, *Glomus*, *Sclerocystis*, *Acaulospora* and *Entrophospora*.

Research Daft and Nicolson (1974) found associated with the roots of AMF indigenous herbaceous plants, including grasses that grow in the area of coal mining and rough asphalt in Pennsylvania and spores were identified gigantean *Gigaspora* spores were then collected and disseminated in the coal dumps. The result is seen the growth of plants, it is shown that the AMF essential for growth and survival of the plants in the coal dumping area, so as to save the land that is open and can avoid critical land or land becomes sour. In a study expected to get JMA spore isolates indigenous to serve and help the land reclamation mining areas.







## MATERIALS AND METHODS







The materials used are herbaceous plants rhizofir soil samples from the coal mining area of PT. KIM in Muara Bungo, Jambi, from five locations: Wika 1, Curved S, Kampung Jawa, KIM 2, and the East Pit. Chemicals Tryphan blue dye roots, laktofenol, 10% KOH, HCl 2%, for the examination of spores required Sucrose solution and Melzer. PVLG. The observed parameters: soil pH, percentage of root infection and spore identification. Method of rhizosphere soil sampling taken at random were then isolated by culture pots, root infection examined through a basic method of Phillip and Hayman (1970) were modified. Calculation of percentage of infection using a basic method of Giovannetti and Mosse (1980). Examination of spores using a basic method of wet sieving Brundett (1996). Identification of spores refers INVAM (2014) and Schenk and Perez (1990).


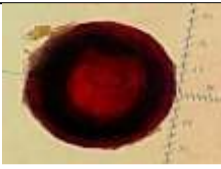
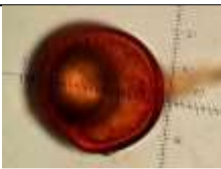
## RESULTS AND DISCUSSION

The observation of spores were obtained directly from the field, seems unclear, and to clarify isolation through pot culture, the results are listed in Table 1.

Table 1. Types of spores in five coal mining sites. PT. Kim, Job Site in Muara Bungo, Jambi

Location	Types of spore	Image	Deskription
Wika	<i>Glomus</i> sp 9		<ul style="list-style-type: none"> <li>- Color Reddish brown to dark brown.</li> <li>-Size spores 41µm x 46µm .</li> <li>-Wall thickness 3-4µm</li> <li>- Hyphae form a bulge</li> </ul>
Kelok S	<i>Rhizophagus clarus</i>		<ul style="list-style-type: none"> <li>- spherical shape</li> <li>-Color Reddish brown</li> <li>-Size 100-260 lm,</li> <li>- 3-layer wall</li> </ul>
	<i>Glomus</i> 10		<ul style="list-style-type: none"> <li>- Round shape.</li> <li>Spore-color light yellowish brown.</li> <li>-Size spores 31µm x 32µm.</li> <li>- wall thickness 3-4µm, with no visible hyphae</li> </ul>
	<i>Glomus</i> sp-8		<ul style="list-style-type: none"> <li>-Oval-shape.</li> <li>-Spore-color to dark reddish brown.</li> <li>-Size spores. 36µm x 37µm</li> <li>- Wall thickness 2-3 µm, with hyphae clearly visible</li> </ul>
	<i>Septoglomus constrictum</i>		<ul style="list-style-type: none"> <li>-round shape.</li> <li>- Spore color is reddish brown to almost black.</li> <li>-The size of 77µm x 72µm spores.</li> <li>- Wall thickness 2-3 µm, with clearly visible hyphae</li> </ul>
Kampung Jawa	<i>Septoglomus deserticola</i>		<ul style="list-style-type: none"> <li>-Oval-shape.</li> <li>Spore-color light yellowish brown.</li> <li>-Size spores 46µm x 50µm .</li> <li>-Wall thickness 3-4 µm, with no visible hyphae</li> </ul>

	<i>Glomus sp-11</i>		<ul style="list-style-type: none"> <li>-Round-shape.</li> <li>-Spore-color light yellowish brown.</li> <li>-Size spores 51µm x 52µm.</li> <li>-wall thickness 4-5 µm</li> <li>hyphae clearly visible -</li> </ul>
KIM 2	<i>Glomus sp-7</i>		<ul style="list-style-type: none"> <li>-Oval-shape.</li> <li>-Spore-color dark brown to reddish brown.</li> <li>-Size spores 45µm x 46µm .</li> <li>-wall thickness 2-3µm, with hyphae</li> </ul>
	<i>Glomus sp-12</i>		<ul style="list-style-type: none"> <li>-Oval-shape.</li> <li>-Spore-color light yellow to brownish yellow.</li> <li>-Size spores 45µm x 41µm.</li> <li>-wall thickness 3-4 µm, with hyphae</li> </ul>
	<i>Glomus sp-13</i>		<ul style="list-style-type: none"> <li>-Oval-shape like an egg.</li> <li>-Color Clear yellow to dark yellow.</li> <li>- Size spores 40 µmx38µm</li> <li>-Wall thickness 3-7 µm, with hyphae clearly visible</li> </ul>
Pit Timur	<i>Glomus sp-1</i>		<ul style="list-style-type: none"> <li>- Round to oval shape.</li> <li>-Color Brownish yellow spores.</li> <li>-Size Spores 55µm x 40 µm.</li> <li>-Wall thickness 3-4 µm, and has hyphae</li> </ul>
	<i>Glomus sp-2</i>		<ul style="list-style-type: none"> <li>-Oval-shape.</li> <li>-Color reddish brown spores.</li> <li>-Size spores 46µm x 38µm .</li> <li>-Wall thickness 1-2 µm, and has clearly hyphae</li> </ul>

	<i>Glomus sp-4</i>		-Round-shape slightly oval. Spore-color brownish yellow to lightbrown.  -Size spores 48µm x 52µm -Wall thickness 4-5 µm
	<i>Glomus sp-5</i>		-Round-shape. -Color Dark reddish brown spores. -Size spores 49µm x 40µm . -Wall thickness 3-4 µm hyphae clearly visible.
	<i>Glomus sp-6</i>		-Oval-shape. -Color Brownish yellow spores.  -Size spores 46µm x 35µm. -Wall thickness 3-4 µm, with hyphae clearly visible

Diversity of spores were seen more numerous in the East Pit area, kelok S and KIM 2, because in that location is an area of heavy metal contamination level is high enough, as said by Gadd (2005), that the AMF spores found in contaminated soil has heavy metal tolerance of higher than AMF spores are commonly found in soil.

### Rooting infection by AMF

The roots of infected plants in the study was from family Asteraceae, Convolvulaceae, Poaceae, Euphorbiaceae, Cyperaceae, and Melastomataceae. There Familia are a plant that is able to form a symbiosis with the AMF. Which is also supported by Smith and Read (2008) which states that a high percentage of root infection does not necessarily result in a high number of spores. In this study, the location of the stockpile Wika KIM 1 and 2 had an average infection percentage of 65% and 100%, although higher but the number of spores produced in lokasistockpile Wika little as 1, and KIM 2 obtained 7-5 spores. While in the east pit, AMF infection in plant roots by 40%, while the number of spores obtained was 17 spores. This suggests that the high AMF infection is not followed by the number of spores, in the opinion of Tri Puspitasari (2013), that the higher the contamination at a site, the higher the production of spores. This mechanism is used as a self defense AMF effort to remain in the soil.

The average percentage of infection in the plant at the location of the stockpile winding S is 66.67% with the number of spores in Kampung Jawa 24, and the percentage of infection by 70% the number of spores 29th Results obtained in

accordance with Adyari (2013) research report in which the results of the research percentage of plant roots AMF infection in contaminated locations (gold mining sites) supported a high number of spores.

AMF infection structures were visible on each root is vesicles and internal hyphae. organ vesicles are generally formed by glomus spores but not formed by the AMF of the genus Gigaspora and Scutellospora. This is consistent with the results obtained in this study, because spora AMF found is from the genus Glomus. Vesicles formed from the tip of the hyphae and contained in a cell or in the space between cells. The content of the vesicles composed of fat so that the organ is thought to function as a storage organ AMF (Peterson et al., 2014).

Arbuscular organs were only observed in the roots of *Mikania micrantha* preparations, *Axonopus compresus*, *M. panniculatus* and *Clidemia. hirta*. This organ is rarely teramati. Karena arbuscular AMF short-lived organs ie 4-15 days after penetration hyphae AMF (Smith and Read, 2008). Arbuscular structure will experience lysis old and looks like blobs that meet the root cortical cells. Arbuscular is the organ that serves as a place of exchange of nutrients between the AMF and host plants (Peterson et al.). More detail can be seen in Table 3.

Table 3. Infection Status JMA In Plants in Location Research

Location	plant species	Vesicula	Arbuscular	Hyphae	percentage
wika 1	<i>Mikania micrantha</i>	+	+	+	60%
wika 1	<i>Ipomoea triloba</i>	+		+	70%
Kelok S	<i>Axonopus compresus</i>	+	+	+	80%
Kelok S	<i>Chromolaena odorata</i>	+		+	60%
Kelok S	<i>Mallotus panniculatus</i>		+	+	60%
Pit Timur	<i>Cyperus sp.</i>	+		+	40%
K. Jawa	<i>Hevea brassiliensis</i>	+		+	60%
K. Jawa	<i>Axonopus paspalum</i>	+		+	90%
Kim 2	<i>Clidemia hirta</i>	+	+	+	100%



### The degree of acidity (pH)

Based on the analysis of the soil at five locations have different pH levels, low pH (acidic) at pit east, ie 4:44, then winding S, ie 4:49, while the highest is in the Javanese village location which has a pH of 6:23. While other sites have a pH of 5:15 to 5:58 (slightly acid), as shown in Table 4.

Table 4 Results of pH measurements in Coal Mining Area of PT. KIM

Lokasi	pH (H <sub>2</sub> O)	Kriteria
Wika 1	5.58	Agak Masam
K. Jawa	6.23	Agak Masam
Kelok S	4.49	Masam
KIM 2	5.15	Masam
Pit Timur	4.44	Sangat Masam

Source: Primary Data

Based on the results of the pH measurements at the site, has a fairly high acidity which can affect the growth of the JMA. Soil acidity can be caused by increased levels of free hydrogen ions which can affect the availability of nutrients, ie at a pH below 6, the elements P, Ca, Mg, and Mo less availability, in addition to the increased content of heavy metals in the soil can be toxic to plants (Kennedy, 1992).

### CONCLUSION

1. There arbuscular mycorrhizal fungi (AMF) in the coal mining area of PT. KIM, Job Site Muara Bungo, Jambi. soil pH of about 4.44 to 6.23.
2. Found 16 species spores in the area coal mines JMA PT. KIM, Job Site Muara Bungo, Jambi, namely: *Glomus* sp1, *Glomus* sp2, *Glomus* sp3, *Glomus* sp4, *Glomus* sp5, *Glomus* sp6, *Glomus* sp7, *Glomus* sp8, *Glomus* sp9, *Glomus* sp10, *Glomus* sp11, *Glomus* sp12, *Glomus* sp13, *Glomus* sp14, *Glomus* sp15, *Glomus* sp16.

sp10, *Glomus* sp11, *Glomus* sp12, *Glomus* sp13, *Septoglomus constrictum*, *Rhizophagus Clarus*, *Septoglomus deserticola*.

- Plants infected JMA research sites totaling 9 species, namely *Mikania micrantha*, *Ipomoea triloba*, *Axonopus compresus*, *Chromolaena odorata*, *Mallotus paniculatus*, *Cyperus sp.*, *Hevea brassiliensis*, *Axonopus Paspalum*, and *Clidemia hirta*.

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