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MORPHOLOGICAL IDENTIFICATION OF ARBUSCULAR MYCHORRIZAL FUNGI ISOLATED FROM THE RHIZOSPHERE OF Jatropha gossypiifolia L. IN TAMBORA INDONESIA

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Abstract

The morphological analysis allows identifying the characteristics of AMF spores, while molecular analysis identifies the species and taxonomy position. Thus, both methods are necessary to create a complete and successful taxonomy of AMF. Soil samples taken under Bellyache bush stands in Tambora, Dompu West Nusa Tenggara Indonesia (08˚26'50,278" S and 117˚59'1,999" E). Root sample taken randomly ± 1 g/sample then washed until clean and put in a bottle film. In summary, the roots are cleaned of cytoplasm with 10% KOH solution, then acidified with 1% HCl solution and then stained with 0.05% trypan blue. The results of calculating the percentage of root colonization show that the characteristics of each different mycorrhizae give percentages various colonization. Colonization percentage each the samples experienced differences, namely at P1 25%, P2 40%, P3 50%, P4 40%, P5 60%, P6 55%, P7 100%, P8 35%, P9 55% and P10 25%. Colonization percentage values on the root is moderate. The percentage is affected by circumstances location, such as litter thickness and soil color. On Tambora, Dompu District locations have a thin litter thickness and color reddish brown soil indicating level high levels of iron (Fe). Based on the results of research conducted then it can be concluded that the Genus of spores found at the research location both on soil samples and roots namely Glomus, Acaulospora and Gigaspora. Glomus spores have relative abundance highest. Percentage of mycorrhizal colonization in Tambora Dompu District is classified as moderate.

Keywords*: Mycorrhiza, Rhizosphere, Tambora, Jatropha gossypiifolia*

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INTRODUCTION

Arbuscular mycorrhizal fungi are a natural constituent of the soil of most ecosystems. They interact with the root of more than 80 % of terrestrial plants and can be considered functional extensions of plant roots considerably enlarging the volume for nutrient up take. Arbuscular mycorrhizal (AM) fungi provide an attractive system to advance plant-based environmental cleanup. During symbiotic interaction the hyphal network functionally extends the root systems of their hosts

Arbuscular mycorrhizal fungi (AMF) are one of the mycorrhizal types which can form internal and external hyphae structures that are beneficial for plants because they can improve the nutritional status of the plant and soil characteristics (physical, chemical, and biological (Bücking & Kafle, 2015) . AMF enhances nutrient uptake and allows plants to be more tolerant against drought, salinity, and other environmental stress (Herlina et al., 2016; Hoysted et al., 2018 ; Shao et al., 2021). Moreover, AMF is capable of protecting roots from pathogenic attack within the soil, improving plant growth and yield, and fixed solubilizing phosphate in soil ((Prasetya & Anderson, 2011; Rini et al., 2021). To successfully obtain higher effectiveness of AMF application, the utilized species needs to be determined.

Morphological identification of arbuscular mycorrhizal fungi is carried out by characterizing them based on the shape, color and size of the spores (Brundrett, et al. 1996) . Some species develop more than one spore morphology, and some morphologically questionable taxa can only be confirmed by molecular biological analysis. Morphological characterization of spores is used to identify arbuscular mycorrhizal fungi. This method is done because it is easy, fast and cost-effective (Proborini et al, 2021).

Although molecular analysis provides precise and specific results, spore morphology is still an important stage for AMF identification to complete the results of molecular identification (Covacevich et al. 2021). In addition, AMF morphology analysis is also essential for basic science purposes, especially when using AMF to be developed into biofertilizers which requires detailed traits information. The morphological analysis allows identifying the characteristics of AMF spores, while molecular analysis identifies the species and taxonomy position. Thus, both methods are necessary to create a complete and successful taxonomy of AMF.

MATERIAL AND METHOD

a. Soil and Root Sampling

Soil samples taken under Bellyache bush stands in Tambora, Dompu West Nusa Tenggara Indonesia (08˚26'50,278" S and 117˚59'1,999" E). This is done by determining 10 trees to be used place of sampling, with the process of taking sample. Soil and root samples were taken at the same time soil depth 0 - 20 cm and a hole diameter of 15 cm with a distance of ¾ from the outermost canopy. Land is taken randomly random with a hoe around the root area on four point repetition, then the land composed as much as 1 kg of soil.

b. Isolation and Identification of Mycorrhiza in Soil Samples

Work steps to identify mycorrhiza in the sample soil mix the soil sample as much as 50 g with 200 ml - 300 ml of water and stirred later leave it for 15-30 minutes. Then filtered in a set of filters with sizes of 53 µm, 45 µm, 38 µm sequentially from top to bottom. material that pass on the bottom filter then transferred to in a centrifuge tube. The filter results are added with glucose 60%. The centrifuge tube was tightly closed and centrifuged at 2500 rpm for 15 minutes. The remaining precipitate is poured into a petri dish then observed under a compound microscope calculating the spore population and making preparations for use Identification of mycorrhizal spores found. Identification mycorrhizal spores by means of morphological observations (color, shape, size, attachment hyphae and ornaments spores) and preparation of slide preparations.

c. Isolation and Identification of Mycorrhiza in Root Samples

Root sample taken randomly ± 1 g/sample then washed until clean and put in a bottle film. In summary, the roots are cleaned of cytoplasm with 10% KOH solution, then acidified with 1% HCl solution and then stained with 0.05% trypan blue. The stained root is cut into pieces ± 2 cm long, arranged on a glass slide 15 sheets, then observed below compound microscope binocular.

RESULTS AND DISCUSSION

Observations show that found three genera of mycorrhiza namely Glomus, Gigaspora, and Acaulospora with spore characteristics are presented in Table 1

Based on Table 1 shows that the characteristics observed microscopically are Observation of spore shape and color. Forms characteristics of the genus found in the location research, namely Glamus spores, Gigaspora, and Acaulospora. Glomus spores are found to be spherical, somewhat round, irregular and oval and has a brown color dark, black, brownish purple and purple. Not all spore's genus found to have substending hyphae. Glomus was high population also thought to be due to environmental conditions that are more suitable, optimal, and compatible in supporting the growth and development of spores. Supported again by the possibility of the absence of antagonistic mushrooms that inhibit mycorrhiza sporulation. Other factors, environmental differences from soil, nutrients, altitude, rainfall, allow the difference of spore's density (Proborini et al, 2021).

Table 1. Spore characteristics in the observed rhizosphere of *Jatropha gossypiifolia*

Trees	Spore characteristic			Spore Type
	Shape	Size (µm)	Colour	
J1	Globose	86	Red Brown	Glomus
J2	Globose	62	Dark Brown	Glomus
J3	Sub Globose	98	Brown	Glomus
J4	Globose	59	Light Brown	Glomus
J5	Globose	117	Light Brown	Acaulospora
J6	Globose	104	Light Brown	Acaulospora
J7	Sub Globose	72	Red Brown	Glomus
J8	Sub Globose	87	Dark Brown	Glomus
J9	Sub Globose	52	Red Brown	Glomus
J10	Globose	131	Light Brown	Gigaspora

The results of the observations in Figure 1 are found on soil samples showed that Gigaspora spores irregular and somewhat round in shape and has dark brown and bluish black. Gigaspora spores produced singly in the soil, with size which are relatively large and have the shape of globos or subglobos. The color of the spores in this genus varies from of yellow, greenish yellow, yellowish green, yellow brownish, to yellowish brown (Sari & Ermavitalini, 2014).

Figure 1. *Gigaspora* sp.

Acaulospora spores are formed by the sporiferous saccule originate from expansion of terminal hyphae. When the spores have fully formed, the contents of the saccule will be transferred to in the spore, then the saccule thins and gradually degraded. The genus Acaulospora has a globos shape, subglobos, irregular, to ellipsoid with two layers of walls spore, of which the innermost spore wall is provided with germination orb. Spore color varies from yellow to orange brownish, dark red, to brownish red. Acaulospora has a globular saccule subglobos, to irregular with colors varying from transparent, yellow, transparent pink, to white (Invam, 2022). According to Sastrahidayat, saccules are often found in Acaulospora (Sastrahidayat, 2011).

Identification results show a difference diversity of spore types found in the samples soil. According to Sundari et al. (2011), location factors and The rhizosphere is very influential on diversity mycorrhizal species and populations. This is due to the different types of soil and environmental conditions of each location.

Soil is a factor that influences presence of mycorrhiza. This can happen because any Soil type has organic content and soil pH different so that spores of the fungal genus can be found various mycorrhizae. One factor that effect on the type of spore is the soil fraction.

Soil characteristics at the sampling location in Tambora, Dompu district, area consist of regosol (volcanic), Mediterranean (volcanic) and alluvial (land) which are very sensitive to erosion and very unstable. This is a characteristic of soil types in volcanic areas.

Observations made on plant roots three types of mycorrhizal spores were found, namely Glomus, Gigaspora and Acaulospora. . Isolation and identification is done with observing the morphological characteristics of the spores in the form of shape, color and the presence of hyphae, arbuscular and vesicular. The three types of mycorrhiza have varied shapes including round, irregular and oval and has a color which are also different such as black, blue and clear.

The results of the isolation and identification of mycorrhizal spores dominating is the genus Glomus. This is due to because of the texture of the soil samples in Tambora, Dompu District is clay so that it is the genus Glomus donate. This is in accordance with (Widiastutik & Kramadibrata, 1998) which states that land which is dominated by the clay fraction is a condition suitable for spore growth and development genus Glomus. As for the genus Gigaspora and Acaulospora is found in loamy sand textures.

The results of observations that have been made indicating that the Glomus found had round shape and blue and has walls fine spore. Glomus is a genus that has wide dissemination, and fairly high adaptation to environmental conditions (Puspitasari *et al.*, 2012). Such that shown in Figure 2.

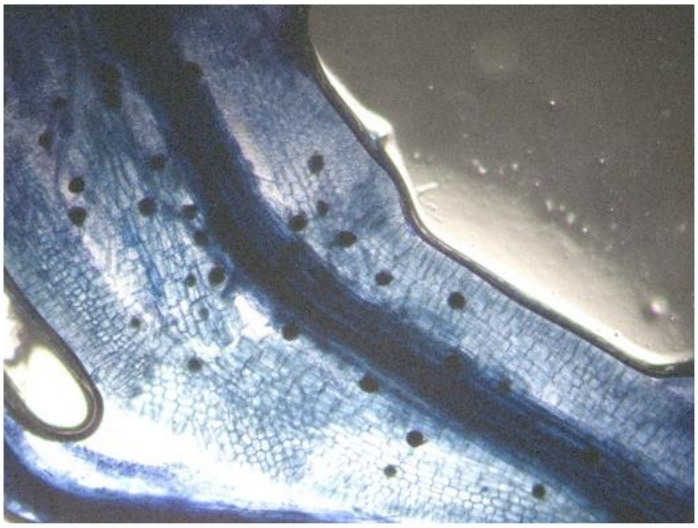

Figure 2. round shape and blue Glomus

Based on the results of observations in the laboratory Acaulospora spores were found at the location sampling. The spores are round and clear in color and has a smooth spore wall. As seen in Figure 3.

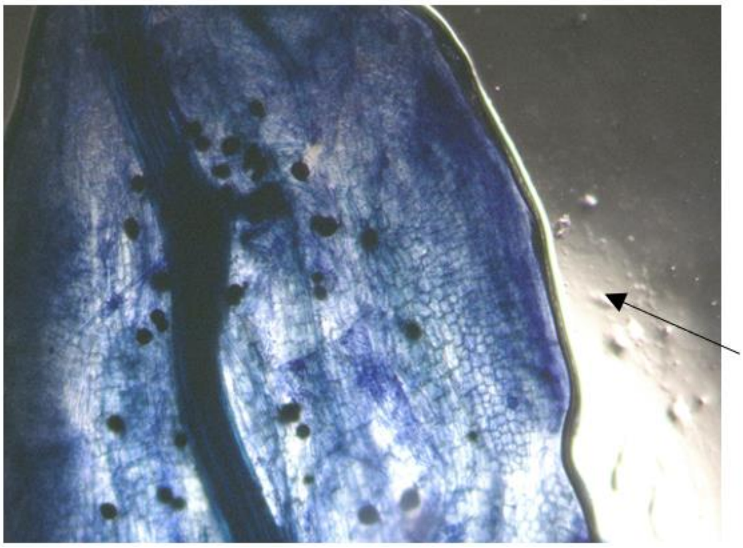

Figure 3. Acaulospora spores

Based on characteristics morphologically, Gigaspora spores have a spherical shape and the surface of the spore wall is relatively rough and wall spores are black, but there are no hyphae attached to the spore wall making it bulbous invisible suspension which can be seen in Figure 4.

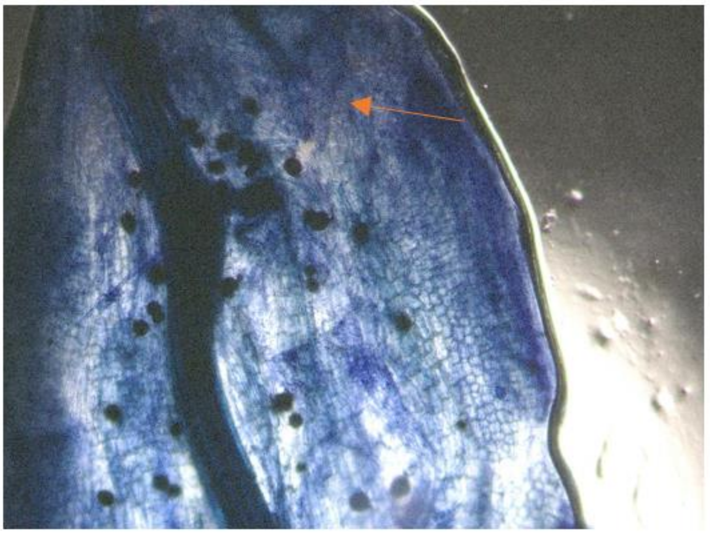
Figure 4. bulbous invisible suspension

According to Sundari *et al.* (2011), the existence of FMA on an area is influenced by environmental factors and types soil. Boan (1998) reports that the land that dominated by the dusty clay fraction, which is soil which is good for the development of the genus Glomus and soil the sandy ones of the genera Gigaspora and Acaulospora found in high numbers.

Soil type is an influential factor of mycorrhizal species. This can happen because any soil type has texture, organic content and pH different land. According to Simangunsong (2006), Glomus easier to find in the soil in the clay fraction while Gigaspora and Scutellospora are easier found in sandy soils. Gigaspora, Scutellospora and Acaulospora are generally only found in soil with an acidic pH while Glomus can be found on soils with acidic to neutral pH (Selvaraj et al., 2001).

The results of the research show that abundance relatively Glomus in Tambora by 73.3%, Gigaspora 20% and Schleroderma 6.6%. The high value of abundance Glomus is caused because this genus has a distribution broad (Tarmedi, 2006) and has levels high adaptation to the environment (Puspitasari et al., 2012). The Glomus genus is faster germinate due to smaller spore size causes the hydration phase to occur very quickly, so process-related enzymatic activity germination will take place more quickly (Saputra et al., 2015).

Glomus is the dominant genus in sampling locations with that abundance high while Gigaspora is only found in some samples. According to Simangunsong (2006), land which is dominated by the clay fraction conditions thought to be suitable for spore development Glomus, while sandy soils are suitable for Gigaspora development.

The results of calculating the percentage of root colonization show that the characteristics of each different mycorrhizae give percentages various colonization. Colonization percentage each the samples experienced differences, namely at P1 25%, P2 40%, P3 50%, P4 40%, P5 60%, P6 55%, P7 100%, P8 35%, P9 55% and P10 25%. Colonization percentage values on the root is moderate. The percentage is affected by circumstances location, such as litter thickness and soil color. On Tambora, Dompu District locations have a thin litter thickness and color reddish brown soil indicating level high levels of iron (Fe). Organic matter on there are not many soil samples because of the surrounding litter the sample tree decomposes slowly, so that the availability of soil nutrients is small so that the sample at this location is very dependent on a symbiotic fungus with it, to help meet these nutrient needs.

CONCLUSION

Based on the results of research conducted then it can be concluded that the Genus of spores found at the research location both on soil samples and roots namely Glomus, Acaulospora and

Gigaspora. Glomus spores have relative abundance highest. Percentage of mycorrhizal colonization in Tambora Dompu District is classified as moderate.

REFERENCES

Bücking H, Kafle A. 2015. Role of arbuscular mycorrhizal fungi in the nitrogen uptake of plants: current knowledge and research gaps. Agronomy 5 (4): 587-612. DOI: 10.3390/agronomy5040587

Chen M, Arato M, Borghi L, Nouri E, Reinhardt D. 2018. Beneficial services of arbuscular mycorrhizal fungi–from ecology to application. Front Plant Sci 9: 1270. DOI: 10.3389/fpls.2018.01270

Covacevich F, Hernández Guijarro K, Crespo EM, Lumini E, Rivero Mega MS, Lugo MA. 2021. Arbuscular mycorrhizal fungi from argentinean highland puna soils unveiled by propagule multiplication. Plants 10 (9): 1803

Duc NH, Csintalan Z, Posta K. 2018. Arbuscular mycorrhizal fungi mitigate negative effects of combined drought and heat stress on tomato plants. Plant Physiol Biochem 132: 297-307. DOI: 10.1016/j.plaphy.2018.09.011

Hu S, Hu B, Chen Z, Vosátka M, Vymazal J. 2021. Arbuscular mycorrhizal fungi modulate the chromium distribution and bioavailability in semi-aquatic habitats. Chem Eng J 420: 129925. DOI: 10.1016/j.cej.2021.129925

Anonim, 2021. INVAMInternational Culture Collection of (Vesicular) Arbuscular Micorrhizal Fungi. Arbuscular Mycorrhizal Fungi. Diperoleh dari University of Kansas https://invam.ksu.edu/, pada 27 Nopemer 2017.

Prasetya CAB, Anderson C. 2011. Assessment of the effect of longterm tillage on the arbascular mycorthiza colonization of vegetable crop grown in Andisol. Agrivita 33 (1): 85-92.

Puspitasari RT, N Sukarno, K Kramadibrata dan D Setiadi. 2010. Identifikasi jamur mikoriza arbuskula asal hutan pantai Ujung Genteng, Sukabumi – Jawa Barat. Prosiding Cakrawala Pemikiran Teori Evolusi Dewasa ini, Salatiga 24-25 November 2009. H. Semangun, FF Karwur, M Martosupono dan S Notosudarmo (Penyunting), 309-326. Universitas Kristen Satya Wacana Salatiga.

Puspitasari, D., Purwani, K.I., & Muhibbudin, A. (2012). Eksplorasi Vesicular Arbuscular Mycorrhiza (VAM) Indigenous pada Lahan Jagung di Desa Torjun, Sampang Madura. Jurnal Sains dan Seni ITS, 1(1)

Rini MV, Susilowati E, Riniarti M, Lukman I. 2020. Application of Glomus sp. and a mix of Glomus sp. with Gigaspora sp. in improving the Agarwood (Aquilaria malaccensis Lamk.) seedling growth in Ultisol soil. IOP Conf Ser Earth Environ Sci 449 (1): 012004. DOI: 10.1088/1755-1315/449/1/012004.

Sari, R.R., & Ermavitalini, D. (2014). Identifikasi Mikoriza dari Lahan Desa Cabbiya, Pulau Poteran, Sumenep Madura. Jurnal Sains dan Seni Pomits, Vol. 3(2): 67-70

Smith SE, Smith FA. 2012. Fresh perspective on the roles of arbuscular mycorrhizal fungi in plant nutrition and growth. Mycologia 104 (1): 1-13. DOI: 10.3852/11-229.

Tarmedi, E. 2006. Keanekaragaman Cendawan Mikoriza Arbuskuladi Hutan Sub Pegunungan Kamojang Jawa Barat (Skripsi). Program Studi Budidaya Hutan Fakultas Kehutanan Institut Pertanian Bogor.

Widiastutik, H., & K. Kramadibrata. (1998). Fungi Mikoriza Bervesikulaarbuskula di beberapa Tanah Masam ari Jawa Barat. Menara Perkebunan, 60(1):9-19