Mycorrhizal Species Diversity in Salam Rhizosphere (Syzygium Polyanthum) in Ex-Sand Mining Land in Cipancur, Ciawigebang District, Kuningan Regency

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Abstract. Mining activities on ex-excavated land cause critical land damage and will affect the ecosystem of the land. As one of the efforts to restore the condition of the former excavated land is by reclamation or revegetation. The use of Arbuscular Mycorrhizal Fungi (AMF) is an alternative that can be developed with various benefits including increasing plant growth, increasing soil nutrient absorption, and increasing plant tolerance to extreme conditions. This study aims to identify the types of mycorrhizae in the rhizosphere of salam (Syzygium polyanthum) in the ex-sand mines of Cipancur Village, Kalimanggis District, Kuningan Regency. The identification and observation of AMF colonization was carried out at the Laboratory of the Faculty of Forestry, Universitas Kuningan. The materials used were soil and root samples under a Salam tree (Syzygium polyanthum) stand. Spore morphology was identified using INVAM (2008). The results showed that the number of spores found under stands of Salam trees (Syzygium polyanthum) from 16 sample plots was 1,949 spores consisting of 3 types of AMF, namely Glomus sp., Gigaspora sp., and Acaulosprora sp.

Keywords: Arbuscular Mycorrhizal Fungi; Ex-Dug C; Reclamation

1 Introduction

Utilization of non-renewable natural resources through sand mining activities must be able to reduce the level of environmental damage for human welfare. Post-mining land damage begins with the destruction of the physical, biological, and chemical properties of the soil followed by changes in the shape and structure of the landscape [1]. The decrease in the quality of the topsoil on ex-mining land is characterized by damage to soil structure, accelerated erosion, excessive washing, soil compaction, decreased soil pH, accumulation of heavy metals in the soil, depletion of organic matter, decreased plant nutrients, and decreased exchange capacity. cations, decreased microbial activity [2]. Changes in the structure and function of the landscape, both in the form of natural events and human disturbances, will have an impact on changes in the composition of plant and animal species [3]. The return of the soil layer to the former post-mining excavation is not able to restore the land condition to the same as the condition before mining [4]. However, knowledge about functional soil microbial changes in land as a result of reclamation with vegetation succession is still inadequate [2].
The post-sand mining land in Cipancur Village has soil with a composition that is poor in nutrients. This is an obstacle for the success of revegetation activities. The existence of arbuscular mycorrhizal fungi (AMF) from plant rhizosphere in nature has an important role because of the mutualism symbiosis with almost 90% of land plant species [5]. Efforts to improve these conditions need to be carried out by land reclamation using biological agents with mycorrhizae in addition to improving the local ecosystem, intervention in the reclamation of ex-sand mining land is needed to accelerate the succession process [6].

One of the silvicultural techniques that can be used to implement it is the application of isomic technology (microbe isolates) or the utilization of potential soil microbes such as mycorrhizae [7]. All genera of arbuscular mycorrhizal fungi do not have the same morphological characteristics, so it is necessary to know their identity so that the existence and diversity of arbuscular mycorrhizae is required. Identification of mycorrhizae found in ex-sand mining areas is very important as an initial effort for land reclamation. The purpose of this study was to determine the diversity of mycorrhizal species in the rhizosphere of salam (Syzygium polyanthum) in the former sand mining area of Cipancur Village.

2 Methodology

Research site at the Silviculture Laboratory, Faculty of Forestry, Universitas Kuningan and soil samples on ex-sand mining land in Cipancur Village, Ciauwigebang District, Kuningan Regency.

The tools used to take soil samples and plant roots were plastic bags, hoes and markers. The tools used for observation in the laboratory are sieves graded 1mm, 425um, 106 um, 45um, and 0.0308 um. Centrifuge, test tube, test tube rack, computer, circular needle, dropper, tweezers, slide, backglass, digital microscope, digital scale, spray bottle, petri dish, measuring cup, small bucket, gloves [8].

The materials consisted of soil samples and roots of Salam trees (Syzygium polyanthum) with a solution of 60% granulated sugar, 10% KOH, 2% HCL, Melzer and Aquades reagents.

Soil samples were taken as much as 500 grams/sample. Isolation of spores from soil samples was carried out by referring to the wet pour and filter method [9] with modified centrifugation [10]. The AMF spores obtained from the isolation were then isolated and prepared with PVLG solution to be identified. The data obtained were analyzed descriptively and presented in the form of identification results of arbuscular mycorrhizal fungi, starting from spore morphology based on: color and size shape. The morphology of the spores was identified using [11] Mycorrhizal colonization was observed on the roots of the sample plants using the root staining technique. The method used for cleaning and staining the sample roots. The percentage of mycorrhizal colonization can be calculated using the colonized root length method [12]. The degree or percentage of infected roots is calculated using the following formula:

\[
\text{Infected roots} (\%) = \frac{\sum_{\text{infected roots}}}{\sum_{\text{all observed roots}}} \times 100\%
\]
3 Result and Discussion

The presence of AMF is characterized by the presence of internal hyphae, external hyphae, vesicles, arbuscules, or internal spores in the plant root system [13]. The results of the identification of AMF spores in the rhizosphere of salam (Syzygium pollyantum) in Cipancur Village were obtained as many as 1,949 different AMF spores. The types of spores obtained have the characteristics of different spore shapes and colors. The spores were then grouped based on shape and color so that three types of AMF spores were obtained, namely Acaulospora, Glomus, and Gigaspora.

Table 1. Total Spores found in the rhizosphere of Salam (Syzygium polyanthum)

<table>
<thead>
<tr>
<th>Sample</th>
<th>Total Spores FMA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Acaulospora</td>
</tr>
<tr>
<td>Salam 1</td>
<td>20</td>
</tr>
<tr>
<td>Salam 2</td>
<td>17</td>
</tr>
<tr>
<td>Salam 3</td>
<td>22</td>
</tr>
<tr>
<td>Salam 4</td>
<td>12</td>
</tr>
<tr>
<td>Total</td>
<td>71</td>
</tr>
</tbody>
</table>

Based on Table 1, the genus glomus was found in the rhizosphere of Salam (Syzygium polyanthum) as many as 1,313 spores, this is in line with research [14] which said that the distribution of the genus was very dominant in each tree stand. In line with research [15], the genus glomus in its distribution always dominates in every plant root. While the genus Acaulospora as many as 71 spores, and the genus gigaspora as many as 565 spores.

Figure 1. Graph of total spores in the rhizosphere of Salam (Syzygium polyanthum)
The genus Acaulospora is characterized by having a globus, sub-globus, irregular to elliptical shape. The spore wall consists of 2 layers where the innermost spore wall is equipped with a germination orb.

The color of the spores varies from yellow, orange, brownish, dark red to brownish red [11]. The acaulospora spores found were round and elliptical, orange and yellow in color, the spore wall consists of 2 layers.

The genus Glomus is characterized by a round shape, the spore wall consists of more than one layer. The color of the spores of the glomus genus varies from yellow, brownish yellow, yellowish brown, light brown, to dark brown and black. [11] The spores found are round to oval in shape, the color of the spores ranges from clear, yellow to brownish.

The genus Gigaspora is characterized by its characteristic bulbous suspensor. Gigaspora spores are spherical in shape and relatively large in size. The color of the spores varies from yellow, greenish yellow, brownish yellow to yellowish brown [11]. Gigaspora spores found are round, yellow in color, have only 1 layer of walls and are sized.

![Whole spores](INVAM, 2021)

**Figure 2.** Types of mycorrhizae found under stands of Salam trees (*Syzygium plectanthum*)
Environmental or edaphic conditions are very decisive in the distribution of arbuscular mycorrhizal genera. Genus *Glomus*, *Acaulospora* and *Gigaspora* are 3 different genera and indirectly have different environmental adaptations. According to [11], the level of adaptation of this genus has variations in tolerance and its own uniqueness in each genus.

The observations obtained after staining the roots showed the presence of round structures called vesicles and arbusculars. The appearance of these structures indicates that there has been infection or symbiotic colonization between the observed plant roots and AMF.

4 Conclusion

The diversity of species of Arbuscula Mycorrhizal Fungi (AMF) in stands of Salam trees (*Syzygium polyanthum*) from 16 sample plots was 1,949 spores consisting of 3 types of AMF, namely *Glomus* sp., *Gigaspora* sp., and *Acaulospora* sp.

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