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(RESEARCH ARTICLE)



Propagation spores of arbuscular mycorrhiza fungi and rooting colonization characteristic's on different host plants

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Abstract

This research aimed to study the propagation spores of arbuscular mycorrhiza fungi (AMF) on different host plants. This research conducted in completely randomized block design (CRBD) with factorial pattern and each treatment was repeated three times and each consist five pot cultures, the total of treatments are 150 pot culture. The first factor are: kinds of host plant (A) with five levels i.e.: *Pueraria javanica* (A₁), *Centrocema pubescens* (A₂), *Sorgum bicolor* (A₃), *Cajanus cajan* (A₄), *Zea mays* (A₅) and second factor are mycorrhiza fungi (B) which comprises two levels i.e.: *Glomus* sp (B₁), *Acaulospora* sp (B₂). The result of research showed that the highest average of spores number on *P. javanica* + *Glomus* sp as 38 spores and lowest on *C. pubescens* + *Glomus* sp and *Z. mays* + *Glomus* sp respectively as 28 spores. The percentage of roots colonization for *Glomus* sp on kinds of host plant i.e.: *P. javanica* as 60.36%, *S. bicolor* as 47.93%, *Z. mays* as 35.36%, *C. cajan* as 21.72%, *C. pubescens* as 20.55%. The percentage of roots colonization for *Acaulospora* sp on *S. bicolor* as 39.83%, *C. cajan* as 27.98%, *C. pubescens* as 26.57%, *P. javanica* as 23.82%, *Z. mays* as 21.20%. The highest average weight of shoots and roots on treatment of *P. javanica* (A₁) each 0.82 g and 0.91 g.

Keywords: *Acaulospora* sp; *Glomus* sp; Host plants; Root colonization; Spores abundance

1. Introduction

The arbuscular mycorrhiza fungi is one of the potential biological natural resources which are found in nature and can be found in various ecosystems. [1], the genus of mycorrhiza fungi i.e.: *Glomus* sp, *Gigaspora* sp, *Acaulospora* sp and *Scutellospora* sp. According to [2], the plant on the surface of the earth estimated as 80-90% to forming a symbiotic mutualism with the mycorrhiza fungi. According [3], the genotypic and species diversity of mycorrhiza fungi are broadly comparable across space.

The mycorrhiza fungi can be an alternative technology of forest and agricultural land rehabilitation, where the application is made possible by the use of local mycorrhiza fungi suitable with host plants to be introduced. [4], reports that's the effort that can be done is by utilizing the plant as a host of mycorrhiza fungi propagation and to increase the spores of mycorrhiza fungi needs to consider the supporting factors so that the results obtained can be optimal. According to [5], the factors that influence the mycorrhiza fungi's occupation are the maturity level of spores, host plants and the environment. [6], states that the selection of host plant is very influential on the production of spores and mycorrhiza infections or colonization in rooting host plants. It is similar to [7], that the abundance of

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arbuscular mycorrhiza fungi in the roots was related to some chemical and physical properties of the soil, thus suggesting an effect of arbuscular mycorrhiza fungi on improving the soil quality.

The host plants preferably by mycorrhizal fungi are capable of providing maximal symbiotic and colonization responses [8]. The criteria of host plant used as mycorrhiza fungi culture are: tolerant to grow in greenhouse conditions, have extensive roots suitable for the development of mycorrhiza fungi [9]. The kinds of host plant using for mycorrhiza fungi propagation or spores production are *Pueraria javanica* [1], *Centrocema pubences*, *Sorghum bicolor* [10], *Cajanus cajan* [11], maize [12], [13] and some kinds of weed like as *Eupatorium odorata*, *Imperata cylindrica* [14], *Amaranthus gracilis*, *Sida rhombifolia* [15], and *Ageratum conyzoides* [16]. The mycorrhiza fungi can be an alternative technology of forest and agricultural land rehabilitation, where the application is made possible by the use of mycorrhiza fungi suitable with host plants to be introduced. According to [17], that's arbuscular mycorrhiza fungi inoculation overall produces positive outcomes on plant production in both controlled and open field conditions, mainly due to several nutrition related benefits that this class of soil fungal symbiosis is able to provide to their host plant. The characteristic infection of mycorrhiza fungi to host plant rooting can be recommended for propagation mycorrhiza fungi.

2. Material and methods

2.1. Preparation of planting media

The zeolite (1-2 mm in size) using as carrier of mycorrhiza fungi spores was cleaned with sterile water to eliminating the fine powder. Prior to weaning, the surface of the zeolite filled culture pot is covered with paper to reduce the direct contact of the medium with the outside air. The seeds of the host plant used first are seeded in a sprout vessel (30 cm x 35 cm in size) separately. After the leaves appear, the sprouts are ready to be weaned into the culture pot.

2.2. Study area and experimental setup

Present study was conducted in Laboratory of Faculty of Forestry and Environmental Science Halu Ole University, Kendari Indonesia. This research conducted in completely randomized block design (CRBD) with factorial pattern and each treatment was repeated three times and each consist five pot cultures, the total of treatments are 150 pot culture. The first factor are: kinds of host plant (A) with five levels i.e.: *Pueraria javanica* (A₁), *Centrocema pubescens* (A₂), *Sorghum bicolor* (A₃), *Cajanus cajan* (A₄), *Zea mays* (A₅) and second factor are mycorrhiza fungi (B) which comprises two levels i.e.: *Glomus* sp (B₁), *Acaulospora* sp (B₂).

2.3. Preparation of pot culture and application mycorrhiza fungi

The making of pot culture refers to methods recommended by [1], with stages: (1) mycorrhiza fungi spores were placed on petrisdish as 10 spores, (2) seedlings of host plants inoculated with mycorrhiza fungi by putting sprouts in the planting hole, then sprayed with 10 spores of mycorrhiza fungi on each planting hole, then closed and watered.

2.4. Observation of variables

The variable were observed in this research include: (a) characteristic of mycorrhiza fungi (b) spores number, (c) percentage root colonization was calculated with formula proposed by [18].

Percentage root colonization = $\frac{\text{field of root view invected}}{\text{Total of root view}} \times 100 \%$, (d) root weight (e) shoot weight.

2.5. Data analysis

Data of each variable were observed were analyzed by variance of analysis. If the value of F count is greater than the value of F table, then continued with Duncan Multiple Range Test (DMRT) at 95% confidence level.

3. Result and discussion

Table 1 Recapitulation of the variance effect host plant and mycorrhiza fungi on the observation variable

Variable	Treatments		
	Host Plant (A)	Mycorrhiza Fungi (B)	Interaction (A*B)
Number of spores	ns	ns	ns
Root colonization	ns	ns	ns
Root weight	s	ns	ns
Shoot weight	s	ns	ns

Notes: ns = no significant, s = significant

The treatment of host plant and type of mycorrhiza fungi no significant to all the observations variable in the independent of treatment and interaction between host plant and mycorrhiza fungi, but host plant significant to root and shoot weight.

3.1. Number of spores and root colonization

Table 2 Effect of host plants and mycorrhiza fungi to spores number and root colonization

Treatments in Combination	Number of Spores (Spores/50 g soil*)	Average of Root Colonization (%*)
<i>Pueraria javanica</i> + <i>Glomus</i> sp	38	60.36
<i>Pueraria javanica</i> + <i>Acaulospora</i> sp	31	23.82
<i>Centrocema pubescens</i> + <i>Glomus</i> sp	28	20.55
<i>Centrocema pubescens</i> + <i>Acaulospora</i> sp	29	26.57
<i>Sorghum bicolor</i> + <i>Glomus</i> sp	33	47.93
<i>Sorghum bicolor</i> + <i>Acaulospora</i> sp	32	39.83
<i>Cajanus cajan</i> + <i>Glomus</i> sp	30	21.72
<i>Cajanus cajan</i> + <i>Acaulospora</i> sp	31	27.98
<i>Zea mays</i> + <i>Glomus</i> sp	28	35.36
<i>Zea mays</i> + <i>Acaulospora</i> sp	35	21.20

Notes: * = tabulated directly without statistical analysis

The highest of spores number on treatment of *P. javanica* + *Glomus* sp as 38 spores and lowest on treatment of *C. pubescens* + *Glomus* sp and *Z. mays* + *Glomus* sp respectively as 28 spores. It is indicated that *Glomus* sp suitable in *P. javanica* plants. According to [19], effects of arbuscular mycorrhiza fungi inoculation on plant growth in soils from organic farm varied depending on the plants species and the nutrient status of the soil. The variation difference of spore number each mycorrhizal fungi on different host plants is thought to be due to the difference in the mycorrhizal fungi response to the host plant. It is similar with result of research from [20], that's number of spores varied with the host plant i.e: peanut, maize and sorghum. [21], states that the root exudates of host plants greatly affect the germination of spores.

Table 2 showed that's on general colonization of mycorrhizal fungi at the root of the host plant is highest in the treatment of *P. javanica* + *Glomus* sp as 60.36% and lowest at the treatment of *C. pubescens* + *Glomus* sp as 20.55%. The root infected by *Glomus* sp according the kind of host plant i.e.: *P. javanica* as 60.36%, *S. bicolor* as 47.93%, *Z. mays* as 35.36%, *C. cajan* as 21.72%, *C. pubescens* as 20.55%. Whereas the root colonization for *Acaulospora* sp on the *S. bicolor* as 39.83%, *C. cajan* as 27.98%, *C. pubescens* as 26.57%, *P. javanica* as 23.82%, *Z. mays* as 21.20%. Thus then, the *Glomus* sp is more suited to the *P. javanica*, whereas the *Acaulospora* sp is more suited to the *S. bicolor*. In this study the percentage of mycorrhizal fungi infections at the roots of host plants varies. It is similarity with [22], that's characteristics of root surface besides the anatomical structure of the roots may also influence the early stages of the plant-fungus interaction.

The compatibility of mycorrhiza fungi and host plant can affect the ability of mycorrhizal fungi to assist the plant in absorbing phosphorus nutrients. This is appropriate statement to [23], root colonization was influenced by the kind of mycorrhizal fungi, host plant species for propagation media and environmental factors. According [5], that's the combination of host plant and kind of mycorrhiza fungi can be evaluated to obtain the best host growth stimuli, root colonization and sporulation. The indicators used as a sign of mycorrhizal fungi infections in rooting host plants are spores, vesicles and arbuscular [15].

The presence of spores in the roots of host plants allegedly associated with the development of mycorrhiza fungi so rapidly that if there is competition in obtaining root exudates then the less competitive to form self-defense in the form of resting spore. [24], that's the vesicles serve as food storage reserves. The arbuscular are not found in the root tissue of host plants because the age of the observed host plant is old in line with the age of the host plant, so that the arbuscula is no longer formed.[25], the arbuscular generally begin to form about 2 to 3 days after the roots are infected.

3.2. Weight of root and shoot

Table 3 Effect of host plant to average of root and shoot weight

Treatments	Average of Roots Weight (g)	Average of Shoots Weight (g)
<i>Pueraria javanica</i> (A1)	0.82 ^a	0.91 ^{ab}
<i>Centrocema pubescens</i> (A2)	0.79 ^{ab}	0.86 ^b
<i>Sorgum bicolor</i> (A3)	0.74 ^b	0.81 ^c
<i>Cajanus cajan</i> (A4)	0.73 ^b	0.79 ^c
<i>Zea mays</i> (A5)	0.72 ^b	0.73 ^d
	2 = 0.06	2 = 0.34
DMRT 95%	3 = 0.07	2 = 0.36
	3 = 0.07	3 = 0.38
	4 = 0.07	4 = 0.38

Notes: The numbers are followed by the same letters in the same column, no significant based DMRT at 95% confidence level.

The highest average of root weight at the treatment of *P. javanica* (A₁) is not significantly different with treatment of *C. pubescens* (A₂), but significantly different with the treatment of *S. bicolor* (A₃), *C. cajan* (A₄) and *Z. mays* (A₅). The average height root weight for *P. javanica* allegedly affected directly by *Glomus* sp. The highest average shoot weight of the host plant is in the treatment of *P. javanica* (A₁) which is significantly different with treatment of *C. pubescens* (A₂), *S. bicolor* (A₃), *C. cajan* (A₄) and *Z. mays* (A₅).

The result of research showed that's *Glomus* sp gives the highest average weight of the roots and shoot of *C. pubescens* (Table 3). It was indicated that between *P. javanica* and *Glomus* sp a match occurs, so that the relationship can run optimally mutualism. [13], stated that the higher dependence on mycorrhiza plants, the percentage increase in dry weight also higher. The selection of host can give good results in increasing spore production [1] and the role of host at the initial stage of infection is to provide root exudates to accelerate the rupture of the gastric tube spores of mycorrhizal fungi [26]. At a later stage of development the root exudates is exploited by the mycorrhizal fungi as a carbon source and the contribution of the mycorrhizal fungi is to help the absorption and supply of nutrients and water [27].

The host affects the colonization and sporulation of mycorrhizal fungi through root system and root exudates. The exudates is an organic compound in the form of liquid metabolites that stimulate spore germination, activate fungi transpiration and promote the growth of hyphae and root colonization [28]. According to [29], the characteristics and number of root exudates compounds emitted depend on plant species, plant age and environmental conditions of plant growth.

4. Conclusion

From the results of this research, the following conclusions can be drawn: (a) the highest average spore number is treatment of *P. javanica* + *Glomus* sp as 38 spores and lowest at the treatment of *C. pubescens* + *Glomus* sp and *Z. mays* + *Glomus* sp respectively as 28 spores. (b) The root colonized by *Glomus* sp based on the host plant species i.e. *P. javanica* as 60.36%, *S. bicolor* as 47.93%, *Z. mays* as 35.36%, *C. cajan* as 21.72%, and *C. pubescens* as 20.55%. (c) The percentage of root colonized by *Acaulospora* sp on the *S. bicolor* as 39.83%, *C. cajan* as 27.98%, *C. pubescens* as 26.57%, *P. javanica* as 23.82%, and *Z. mays* as 21.20%. (d) The highest average of root and shoot of host plant at the

treatment of *P. javanica* (A₁) respectively as 0.82 g and 0.91 g. The use of *P. javanica* as fungi mycorrhiza propagation is very effective to be developed in the long term, because the kinds of plants also function as ground cover plants, erosion prevention as well as sources of soil organic matter.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors declared that present study was performed in absence

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