International Journal of Biosciences | IJB |
ISSN: 2220-6655 (Print), 2222-5234 (Online)
http://www.innspub.net
Vol. 17, No. 5, p. 195-201, 2020

INNSPUB

RESEARCH PAPER OPEN ACCESS

Effects of ectomycorrhizae on the wildling growth of six tree species from Dipterocarpaceae family

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Key words: wildling growth, ectomycorrhizae, Shorea, Dipterocrpaceae family.

http://dx.doi.org/10.12692/ijb/17.5.195-201 Article published on November 19, 2020

Abstract

The objectives of the research were to analyze the effects of ectomycorrhizae on wildling growth of six tree species from Dipterocarpaceae family, and to analyze the species that have the best growth response on the ectomycorrhizae. The method used was a factorial experiment in a completely randomized design. Factors applied were ectomycorrhizae and wildling species. Ectomycorrhizae factor consisted of without mycorrhizae (control) and with mycorrhizae. Species factor consisted of *Shorea leprosula*, *Shorea ovalis*), *Hopea mengarawan*, *Shorea palembanica*, *Shorea macroptera*, and *Shorea parvofolia*. The number of treatment was 12 combinations. Replication was four times, and the number of wildlings of each treatment was four wildlings. The total wildlings observed were 144 wildlings. The results showed that in terms of the height increment variable, the ectomycorrhizae factor provided significant effects on the growth of the wildlings. The average height increment of the six species of Dipterocarpaceae family with mycorrhizae was 3.60 cm while without mycorrhizae was 2.76 cm. In terms of diameter increment variable, the ectomycorrhizae and species factors independently affected the growth of the wildlings significantly. The average diameter increment of the six species of Dipterocarpaceae family with mycorrhizae was 0.11 cm while without a mycorrhiza treatment was 0.07 cm. The species that had the highest diameter increment (0.113 cm) was *Shorea ovalis,* while the smallest one was *Shorea macroptera* (0.063 cm). The survival percentage of wildling observed was 100%.

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Introduction

Dipterocarpaceae is a tree family that ecologically dominates tropical forests in Southeast Asia including Indonesia. In addition, according to Kettle (2010), the family has an important role in timber trade because it accounts for 80% of timber exports supporting the revenue of many developing countries. The family encompasses many genera including shorea. Some well-known species from the shorea genus are *Shorea leprosula* Miq., *Shorea ovalis* Korth., *Hopea mangerawan* Miq., *Shorea palembanica, Shorea macroptera* Dyer, and *Shorea parvifolia*. These species are in high demand because they supply raw materials for the wood industry and wood construction. Industrial and housing needs for these species are getting higher from year to year because the forests that produce the species decrease over time. As a result, demand and supply are not balanced. Demand is much higher than supply; therefore, the cultivation of these tree species is indispensable (Achmad, 2017a).

The problem faced in cultivating shorea seedlings is the provision of sufficient quantities of seedlings within a certain time. This is because the fruit season of the family does not occur every year. Shorea trees sometimes bear fruit only once in 4-13 years. In addition, the viability of seeds can decrease within a few days after the seeds fall from the mother tree (Achmad, 2017b). Therefore, it is not possible for seeds collected in one fruit season to be stored for planting the following year. One alternative to supply the seed needs is to use wildlings that are growing under the parent trees. Another problem in growing wildlings from the shorea trees is the lack of mycorrhizae that are symbiotic with wildling's roots. Although the roots of natural seedlings are known to be associated with mycorrhizae, the mycorrhizae are largely released when the wildlings are extracted from the soil surface. Therefore, natural seedlings or wildlings require additional mycorrhizae manually to grow well.

De la Cruz (1983) explained a number of techniques to inoculate mycorrhizal fungi. Those are (1) mycorrhizal soil, (2) mycorrhizal seedlings, (3) vegetative mycelia, (4) infected roots, (5) spore suspension, mycorrhizal capsules, and (7) mycorrhizal tablets.

The mycorrhizal tablets were prepared through dried spores of *Pisolithus tinctorius* or *Rhizopogon* sp. Mixec with suitable carries and compressed in the form of tablet. The carries can be dried-sieved soil or lime. Standard tableting machines used in the manufacture of medicine tablets can be used for this purpose. In addition, according to Hardiatmi (2008), the packaging of mycorrhizal inoculants in the form of tablets and capsules is intended to: (1) Inoculant savings and increased effectiveness. On practice previously, mycorrhizal transmission was carried out through use of the soil originating from forest stands and nurseries that are brought and moved to plant holes in the field. Moreover, this will be done for a very broad material of course it will be very troublesome and very not practical. Apart from that any land that comes from forest stands has not of course there are spores or mycorrhizal fungi hyphae, (2) make it easier to handle. Every plantation development area requires a very large number of mycorrhizal inoculants. When packed in capsule or tablet form will make it easier transportation and storage because it is usually the field of plants is in a remote location that lacks facilities, and (3) be specially produced. The land that will be used for plantation development, soil pH varies widely. If packaged in a tablet form, the components of the tablet can be adjusted in such a way as to match the pH of the local soil. When compared between the advantages and disadvantages of some of the inoculation methods mentioned above, the method of inoculation using spores, spore tablets or capsules has good prospects for development in Indonesia (Hendromono, 1987).

The objectives of the research were to analyze the effects of the ectomycorrhiza tablets on the wildling growth of six tree species from Dipterocarpaceae family, and to analyze the species that have the best growth response on the ectomycorrhiza tablets.

Materials and methods

Materials used were wildlings from six species of Dipterocarpaceae family, namely *Shorea leprosula* Miq., *Shorea ovalis* Korth., *Hopea mangerawan* Miq., *Shorea palembanica, Shorea macroptera* Dyer, and *Shorea parvifolia*. Wildling's age was one year. The number of wildlings used was 144 wildlings placed at seedbeds under trees. Another material used was mycorrhiza tablets with a weight of 0.5 gram/tablet. Equipment used was a measurer for measuring the height of wildlings, a caliper for measuring the diameter of the wildlings, polybags as containers of wildling's media, hoes, and big knives for field preparation.

Methods

The research procedure was done by preparing a nursery where the research was done. The activity in the nursery was a tool, equipment, and seedbed preparation. The next activity was collecting wildlings from under a big mother tree by extraction. After that, the wildlings were put in the polybags that had been arranged at seedbeds. Furthermore, the wildlings were provided mycorrhiza tables based on the instructions on the tablet's packaging, which is to make a hole until it touches the roots. The distance between the hole and the wildling stem was 1 cm. Mycorrhiza tablets with a diameter of 1 cm were inserted into the hole in a horizontal position so that they were easily dissolved. The number of tablets for one hole was one tablet. Mycorrhiza tablets were provided after one month of wildling being planted in a polybag. This was intended so that wildlings could adapt to the new environment.

The first factor (A) observed was providing a mycorrhiza tablet to a wildling. This factor consisted of without (A1) and with a mycorrhiza tablet (A2) (one tablet for one wildling). The second factor (B) was wildling species. This factor consisted of six species, namely *Shorea leprosula* Miq. (B1), *Shorea ovalis* Korth.(B2), *Hopea mangerawan* Miq. (B3), *Shorea palembanica* (B4), *Shorea macroptera* Dyer (B5), and *Shorea parvifolia* (B6). Each species consisted of four wildlings, and they were replicated three times so that it was needed wildlings as an amount of 144 stems (2 factors × six species × 4 wildlings × 3 replications). Variables measured were the height and diameter of wildlings, the number of leaves, and survival rates of wildlings.

Analysis

The effects of the treatments on the variables measured were analyzed using a factorial model in a completely randomized design.

The Statistical Package for Social Sciences (SPSS) software was used to conduct the analysis.

Results

Height increment

Based on the tests of between-subject effects or the F-test (Table 1), the mycorrhiza factor significantly affected the height increment of the six species of Dipterocarpaceae family (p-value = 0.000 < 0.05), while the species factor (p-value = 0.094), and its interaction with the mycorrhiza factor (p-value = 0.682) did not affect the height increment of the six species of the Dipterocarpaceae family.

Table 1. The tests of between-subjects effect on the height increment of the six species.

Source	Sum of Squares	df	Mean Square	F	Sig.
Species	2.222	5	.444	2.151	.094
Mycorrhiza	6.250	1	6.250	30.252	.000
Species * Mycorrhizae	.646	5	.129	.625	.682
Error	4.958	24	.207		
Total	378.250	36			

The average height increment of the six species of Dipterocarpaceae family with mycorrhizae was 3.60 cm while without mycorrhizae was 2.76 cm. For details of the average height increment of each species was described in Figure 1.

Table 2. The tests of between-subjects effect on the diameter increment.

Source	Sum of Squares	Df	Mean Square	F	Sig.
Species	.012	5	.002	5.661	.001
Mycorrhiza	.016	1	.016	37.885	.000
Species * Mycorrhiza	.001	5	.000	.428	.824
Error	.010	24	.000		
Total	.328	36			

Diameter increment

Based on the tests of between-subject effects or the F-test (Table 2), the mycorrhizae factor significantly affected the diameter increment of the six species of Dipterocarpaceae family (p-value = 0.000 < 0.05). Likewise, the species factor significantly affected the diameter increment of the six species of Dipterocarpaceae family (p-value = 0.001 < 0.05), but the interaction of the two factors (mycorrhizae and species) did not affect the diameter increment of the six species of the Dipterocarpaceae family (p-value = 0.824 > 0.05).

Table 3. Diameter increment of each species (cm) from the biggest to the smallest increment.

Species	Mean	Mark
Shorea ovalis	0.113	a
Shorea leprosula	0.104	ab
Hopea mengarawan	0.103	abc
Shorea parvifolia	0.083	bcd
Shorea palembanica	0.071	de
Shorea macroptera	0.063	de

Note: the species that have the same at least one letter in the mark were not different significantly at the 0.05 level.

The average diameter increment of the six species of Dipterocarpaceae family with mycorrhizae was 0.11 cm while without mycorrhiza treatment was 0.07 cm. The graph of diameter increment based on the mycorrhiza treatment and species can be seen in Figure 2.

Furthermore, the six tree species observed showed different response to mycorrhizae, especially on the diameter growth. Based on the least significance difference test at a critical level of 0.05, *Shorea ovalis* had the highest diameter increment, followed by *Shorea leprosula*, *Shorea mengarawan*, *Shorea parvifolia*, *Shorea palembanica*, and *Shorea macroptera*. The diameter increment for the six species was described in Table 3.

The Number of leaf increment and survival percentage

On the basis of the tests of between-subject effects or the F-test (Table 4), both factors (mycorrhizae and species) independently and their interactions did not affect the number of leaf increment of the wildlings. The survival percentage was 100%.

Table 4. The tests of between-subjects effect on the number of leaf increment.

Tests of Between-Subjects Effects					
Dependent Variable: Number of leaves					
Source	Sum of Squares	df	Mean Square	F	Sig.
Species	.932	5	.186	.344	.881
Mycorrhiza	.141	1	.141	.260	.615
Species * Mycorrhiza	.474	5	.095	.175	.969
Error	13.000	24	.542		
Total	102.438	36			

Discussion

Based on the F-test, the mycorrhizae factor affected the height increment of belangeran wildlings, and the mycorrhizae factor and the species factor independently affected the diameter growth of the wildlings. The mycorrhizae factor affected the growth of seedlings because the mycorrhizae can increase nutrient absorption, increase resistance to pathogen attack and increase resistance to drought (Simanungkalit, 2001). Likewise, Talanca (2010) stated that the arbuscular mycorrhizal fungi have crucial roles on plants. They can increase nutrient uptake, enhance resistance on root pathogen, and stimulate the growth of plants at marginal lands. In addition, irrespective of type of mycorrhizae inoculation, growth of seedlings increased significantly in comparison to control. Maximum growth was observed for the seedlings inoculated with ectomycorrhizae alone, followed by dual inoculations ectomycorrhizae and endomycorrhizae, and without mycorrhizae treatment or control (Tapwal *et al.*, 2015).

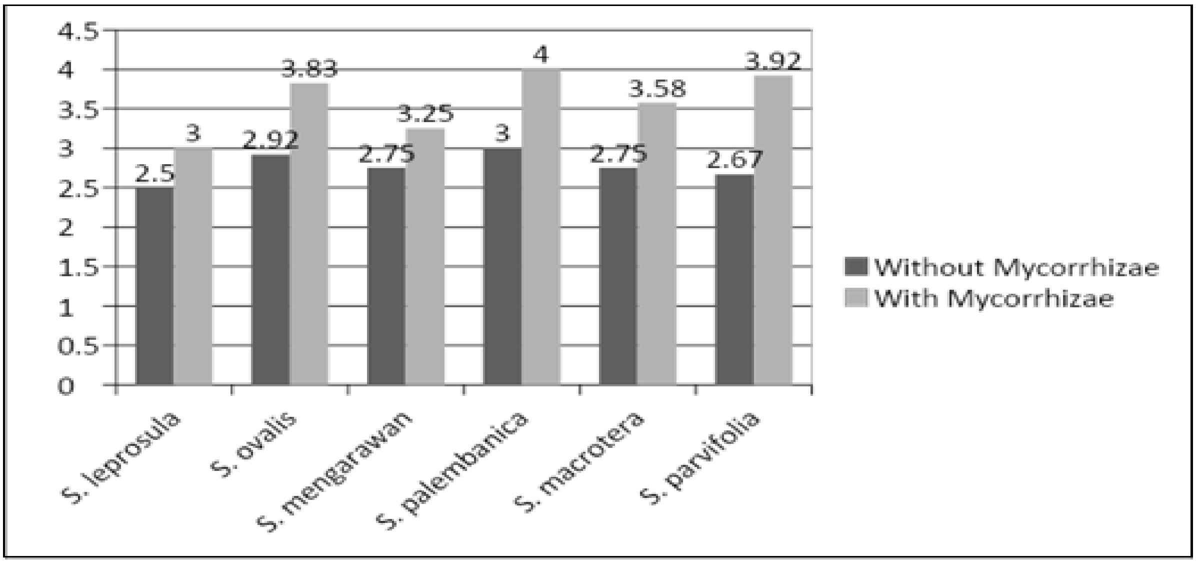

Fig. 1. Height increment (cm) with and without mycorrhizae of six-species of wildlings.

In details, Wu and Xia (2006) explained that arbuscular mycorrhizal (AM) fungus colonization significantly affected plant growth and biomass regardless of water status.

The soluble sugar of leaves and roots, the soluble starch of leaves, the total non-structural carbohydrates of leaves and roots, and the Mg2+ of leaves were higher in AM seedlings than those in corresponding non-AM seedlings. The levels of K+ and Ca2+ in leaves and roots were higher in AM seedlings than those in non-AM seedlings under water stress conditions. Moreover, AM colonization increased the distributed proportions of soluble sugar and non-structural carbohydrates to roots. AM seedlings had higher leaf water potential, transpiration rates, photosynthetic rates, stomatal conductance, relative water content, and lower leaf temperature than corresponding non-AM seedlings. Wu and Xia (2006) also found that AM colonization increased the osmotic adjustment originating not from proline but from the non-structural carbohydrates, K+, Ca2+ and Mg2+, resulting in the increase of drought tolerance. In addition, Hardiatmi (2008) stated that the advantages of using mycorrhizal fungi are plant roots in the mycorrhizal nursery can uptake more nutrients, especially phosphorus, with physical, chemical or enzymatic mechanisms, and plant roots absorb more water because they can enter soil cavities that are smaller than the diameter of the root hairs, plants are more resistant to root pathogens because there is physical defense as a result of the root surface is covered by a layer of mycorrhizal fungus hyphae, and there is a chemical defense due to the release of antibiotics from mycorrhizal fungi.

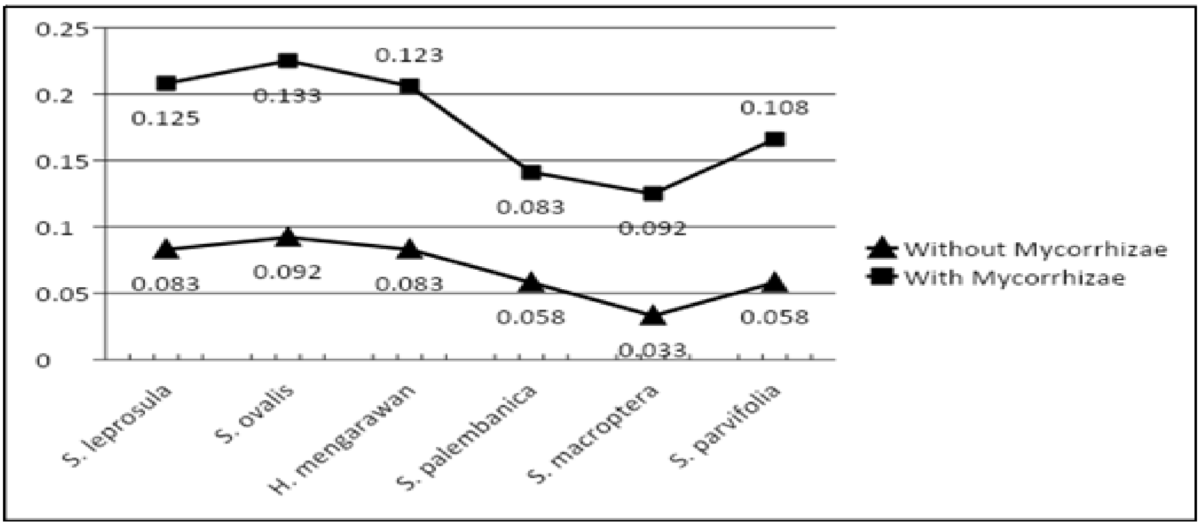

Fig. 2. Diameter increment (cm) with and without mycorrhizae of six-species of wildlings.

The previous and present study had proven that the mycorrhizae had important roles in stimulating the growth of seedlings in a nursery. The mycorrhizae can increase nutrient absorption, increase resistance to pathogen attack and increase resistance to drought.

In diameter growth, the six tree species from Dipterocarpaceae family had different response on the mycorrhizae. The same results had also been found by Gehring (2003) stating that rain forest seedlings highly varied in their growth responses to arbuscular mycorrhizal colonization and that some of this variability was pertaining to the light intensity of the environment. Given that seedlings may spend many years in the shaded understory, these differences among species could have important effects on long-term seedling performance and seedling community dynamics. Gehring (2003) and the present study used wildlings or natural seedlings collected from under mother trees as the research objects that had different ages and received different light intensity.

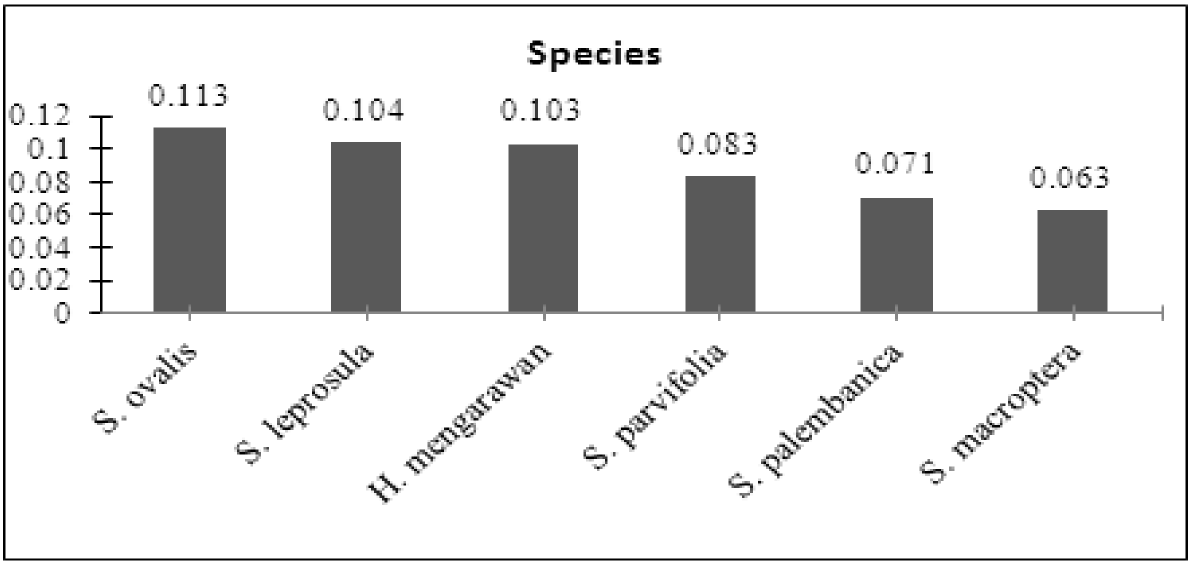

Fig. 3. Diameter increment of each species (cm) from the biggest to the smallest increment.

In addition, according to Omon (2002), factors affecting development of mycorrhizae could be light intensity, soil heat, humidity, soil pH, and oxygen supply. These factors were hypothesized as the causes of different responses of the six tree species of Dipterocarpaceae family on the mycorrhizae.

Acknowledgements

I would like to thank the Industrial Tree Plantations of PT. Janggala Semesta for facilitating the research. I also express my gratitude to Sulaiman Bakri and Djohansyah for their help in collecting and analyzing the research data.

Conclusions

The mycorrhizae factor stimulated the height growth of the wildlings of the six tree species of from Dipterocarpaceae family. Also the mycorrhizae and species factors independently stimulated the diameter growth of the wildlings. The species that was very sensitive on the mycorrhiza tablets in terms of diameter increment variable was *Shorea ovalis*. It is recommended the use of mycorrhizae in the form of tablet in a nursery with a dose of one tablet for one seedling. The mycorrhiza tablets are easy to transport, easy to inoculate, reduces disease-carrying, and allow transport between islands.

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