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Improvement Pepper Plant Seedling (*Piper nigrum* L.) of Growth Through the Application of Mycorrhiza Fungi Isolated From Rootings of Broad Leaves Weed

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ABSTRACT

Pepper plant is one that holds important export commodity for the economy of the people of Indonesia. One attempt to improved crop production, namely pepper cultivation technology improvements that begins with the provision and improvement of growth of seedlings good quality before they are transplanting. This study aims to determine the improvement of growth pepper plant seedling before transplanting using mycorrhiza fungi isolated from rooting of broad leaves weed. The study was performed in Sindang Kasih village, District of West Ranomeeto and Laboratory of the Faculty of Forestry and Environmental Science Halu Oleo University, Kendari, Indonesia. The experimental design was a randomized complete block design (RCBD) with six treatments, each treatments was replicated with 3 replications. The treatments i.e. without mycorrhiza fungi (M0), mycorrhiza fungi 5 g polybag^{-1} (M1), mycorrhiza fungi 10 g polybag^{-1} (M2), mycorrhiza fungi 15 g polybag^{-1} (M3), mycorrhiza fungi 20 g polybag^{-1} (M4), and mycorrhiza fungi 25 g polybag^{-1} (M5). The variables observed for results were plant height, number of leaves, percentages of mycorrhiza fungi infection to plant roots. Results of study showed that inoculation of mycorrhiza fungi provide a noticeable effect on height increment of plants aged 35 to 56 day after planting (DAP) and the number of leaf at age of 77 DAP. Inoculation mycorrhiza fungi was gift impact on the plant height increment at age of 63 to 77 DAP, and infection of mycorrhiza fungi to roots of pepper plant seedling increment at age of 77 DAP. The highest of plant height and mycorrhiza fungi infection to plant roots is the treatment of mycorrhiza fungi propagules 10 g polybag^{-1}(M2), the highest number of leaves is the treatment of 5 g mycorrhiza fungi propagules 5 g polybag^{-1} (M1).

INTRODUCTION

Pepper plants (*Piper nigrum* L.) occupied the fifth foreign exchange Indonesian plantation commodity after oil palm, rubber, coffee and tea. The pepper plants were cultivated by farmers in the form of small farms, but productivity is still low. One cause of low production of pepper plant are preparation of seedling which is not good (Rachmawati and Halim, 2011) and the use of seeds have not qualified (Karmawati and Supriya, 2007). The pepper seed quality standard in Indonesia was prepared by a technical committee of seed and nursery products agriculture in an effort to improve the quality assurance. The quality standard is 100% pure seeds, 100% seed health, 5-7 the number of sections and number of leaves 5-8 strands (Standar Nasional Indonesia, 2006).

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The one effort to provide a good pepper plants that provided seeds of good quality. According to Rachmawati and Halim (2011), the quality of seed is one of the innovative reliable to improve the productivity of pepper. In providing quality seed pepper, then propagation medium is very important to note. According Haryadi *et al.* (1996), that the media planting is very influential on component nursery seedling growth of pepper plants. Media nurseries are a mixture of sand, cow manure and soil in the ratio 1: 1: 2. Preparation of a good pepper plant seeds can also be done by applying of mycorrhiza fungi in the growing medium. The result of research Rachmawati and Halim (2011), showed that the inoculation 10 g propagules of mycorrhiza fungi can increase in the number of leaves pepper plant seeds. Similarly, the results of research Halim *et al.* (2016a), the application of mycorrhiza fungi at a dose of 10-20 g able to spur the growth of the pepper plant seeds such as plant height, leaf number and the number of tendrils.

MATERIAL AND METHODS

Study area and Experimental Set Up:

The study was performed in net house Sindang Kasih village, District of West Ranomeeto and Laboratory of the Faculty of Forestry and Environmental Science Halu Oleo University, Kendari, Indonesia. The experimental design was a randomized complete block design (RCBD) with six treatments, each treatments was replicated with 3 replications.The treatments i.e. without mycorrhiza fungi (M0), mycorrhiza fungi 5 g polybag^{-1} (M1), mycorrhiza fungi 10 g polybag^{-1} (M2), mycorrhiza fungi 15 g polybag^{-1} (M3), mycorrhiza fungi 20 g polybag^{-1} (M4), mycorrhiza fungi 25 g polybag^{-1} (M5).

Planting Media and Pepper Plant Seedling Set Up:

The soil has been taken from the study area it cleared from debris such twigs, roots, leaves and small rocks. Cleared soil sifted into a polybag with a weight 7 kg soil and recommended dose basic organic manure was added to each polybag. The cuttings of pepper plant seeds used from climbing vines and tendrils hanging. The tendrils that have been taken, cut to obtain cuttings, then planted in seedling pots and treated to form buds on the segments. The cuttings that have sprouted then transplanted in a polybag which has been prepared in accordance with the treatment. Each polybag contains two pepper plant cuttings.

Mycorrhiza Fungi Propagules Set Up and Inoculation on Planting Media:

Mycorrhiza fungi used in this study were isolated from the roots of broadleaf (Halim *et al*., 2014). Before they were used in the study, it has to be propagated on *Ageratum conyzoides* and maize for 3 months long time to get enough the propagules as research needs (Halim *et al*., 2016b). At the time of transplanting peppers seedlings, mycorrhiza fungi propagules applied by making a hole in a polybag with ± 15 cm in depth and mycorrhiza fungi propagules are locations under plant seedlings (Halim, 2009).

Observation of Variables and Data Analysis:

The variables measured in this study were as follows:

1. Plant height and leaf number were measured on the intervals of each seven days which start from the 7th days after planting and continue up to 77 days after planting (DAP).
2. Percentage of mycorrhiza fungi colonization on root of the pepper plant. The root colonization calculation method colonized root length. Randomly take root pieces that have been colored with a length of ± 1 cm by 10 pieces and are arranged regularly on preparations, for each plant sample was made by 2 preparations. The root piece on a glass slide was observed for each field of view. The field of view that showed the presence of colonization was characterized by hyphae, arbuscular and vesicles. The degree of root colonization was calculated using the formula proposed by Mosee (1981):

$$\%\ Root\ colonization = \frac{\text{field of root view infected mycorrhiza fungi}}{\text{Total of root view}}\ x\ 100\%$$

Data of each variable were observed were analyzed by variance of analysis. If the F count is greater than the F table, then continued with Duncan Range Multiple Test (DRMT) at 0.05% confidence level.

RESULTS AND DISCUSSION

Recapitulation of the variance effect of mycorrhiza fungi to the observation variable (plant height, leaf number, the colonization of mycorrhiza fungi on the roots) are presented in Table 1.

Table 1: Recapitulation of the variance effect of mycorrhiza fungi to the plant height, leaf number and the colonization of mycorrhiza fungi in the roots

Observation of variables	Effect of treatment
Average of plant height	
7 DAP	ns
14 DAP	ns
21 DAP	ns
28 DAP	ns
35 DAP	*
42 DAP	*
49 DAP	*
56 DAP	*
63 DAP	*
70 DAP	*
77 DAP	*
Average of leaf number	
7 DAP	ns
14 DAP	ns
21 DAP	ns
28 DAP	ns
35 DAP	ns
42 DAP	ns
49 DAP	ns
56 DAP	ns
63 DAP	ns
70 DAP	ns
77 DAP	*
Percentage of mycorrhiza fungi colonization on root	*

Notes: ns = no significant, * = significant

The results showed that the inoculation of mycorrhiza fungi no significant effect on the average of plant height at age of 7, 14, 21 and 28 DAP. While the leaf number, inoculation of mycorrhiza fungi only significant effect at the age of 77DAP. The results showed that the inoculation of mycorrhiza fungi effect on plant height pepper at age of 35, 42, 49, 56, 63, 70 and 77 DAP (Table 2). The results also showed that at the beginning of vegetative growth, mycorrhiza fungi inoculation propagules not give real effect to the average of plant height. It is presumed in the early stages of plant growth, mycorrhiza fungi have not interact optimally with pepper plant roots so that the effect of mycorrhiza fungi on plant growth is not yet fully functional so that plant roots are still difficult to absorb the nutrients in the soil. Beltrano *et al*., (2013), suggest that hyphae of mycorrhiza fungi can avoid phosphorus depletion zones roots, or facilitate absorb phosphorus. It showed that the mycorrhiza fungi can improve plant growth.

Table 2: Impact of mycorrhiza fungi on average of height plant

Treatments	Height Plant (cm)						
	35 DAP	42 DAP	49 DAP	56 DAP	63 DAP	70 DAP	77 DAP
without mycorrhiza fungi (M0)	9.97bc	11.17bc	14.90ab	18.00bc	19.17bc	25.00bc	29.83b
mycorrhiza fungi 5 g polybag^{-1} (M1)	12.17bc	14.27abc	18.60a	21.43ab	25.43ab	27.40ab	31.60ab
mycorrhiza fungi 10 g polybag^{-1} (M2)	10.55bc	15.97ab	18.47a	24.80a	28.63a	31.47a	37.13a
mycorrhiza fungi 15 g polybag^{-1} (M3)	13.07ab	15.73ab	18.07a	18.07bc	18.07cd	18.97c	19.73c
mycorrhiza fungi 20 g polybag^{-1} (M4)	15.97a	17.63a	18.53a	18.97ab	19.97bc	20.70c	22.80c
mycorrhiza fungi 25 g polybag^{-1} (M5)	8.83c	9.68c	10.33b	11.67c	11.67d	11.67d	11.67d
DRMT 0.05%	2 = 3.60	2 = 4.75	2 = 5.68	2 = 6.14	2 = 6.82	2 = 5.99	2 = 6.60
	3 = 3.77	3 = 4.97	3 = 5.94	3 = 6.43	3 = 7.13	3 = 6.27	3 = 6.91
	4 = 3.87	4 = 5.10	4 = 6.10	4 = 6.60	4 = 7.33	4 = 6.44	4 = 7.09
	5 = 3.94	5 = 5.19	5 = 6.21	5 = 6.72	5 = 7.46	5 = 6.55	5 = 7.22
	6 = 3.99	6 = 5.25	6 = 6.28	6 = 6.79	6 = 7.54	6 = 6.63	6 = 7.31

Note: The numbers are followed by the same letters in the same column are not significantly different on DRMT 0.05%

Inoculated the mycorrhiza fungi no real effect on the number of leaves at 28, 35, 42, 49, 56, 63 and 70 DAP, but the real effect on the age of 77 DAP. The highest of average number of leaves of the plant at the age of 77 DAP obtained at treatment of M1 that is not different with the treatment of M0, M2 and M3, but significantly different with the treatment of the M4 and M5. The added of plant height in line with the number of leaves. Musfal (2008) reported that the plants were infected by mycorrhiza fungi capable of absorbing element Phosphor (P) is higher than with plants that are not infected by mycorrhiza fungi. The high of P uptake by plants infected by mycorrhiza fungi caused mycorrhiza fungi hyphae secrete the Phosphatase enzymes that P is bound in the soil will be dissolved and available for plants. This is similar result were reported recently with the results of research Rachmawati and Halim (2011) that the mycorrhiza fungi inoculations were low or high doses propagules capable of increasing the number of leaves pepper plant seeds.

Table 3: Impact of mycorrhiza fungi on average of leaves number at the age 77 DAP

Treatments	Leaves number (helai)	DRMT 0.05%
without mycorrhiza fungi (M0)	9.00ab	
mycorrhiza fungi 5 g polybag^{-1} (M1)	10.33a	2 = 2.37
mycorrhiza fungi 10 g polybag^{-1} (M2)	9.33ab	3 = 2.48
mycorrhiza fungi 15 g polybag^{-1} (M3)	8.33abc	4 = 2.55
mycorrhiza fungi 20 g polybag^{-1} (M4)	7.67bc	5 = 2.59
mycorrhiza fungi 25 g polybag^{-1} (M5)	6.33c	6 = 2.62

Note: The numbers are followed by the same letters in the same column are not significantly different on DRMT 0.05%

The percentage of mycorrhiza fungi infection in pepper plant roots was highest in treatment of 10 g polybag^{-1} (M2) is not significant to all other treatments, but significantly different from those without mycorrhiza fungi propagules (M0). The percentage of mycorrhiza fungi infection in pepper plant roots are presented in Table 4.

Table 4: The percentage of mycorrhiza fungi infection on pepper plant roots

Perlakuan	Percentage of mycorrhiza fungi infection (%)	DRMT 0.05%
without mycorrhiza fungi (M0)	0.00 b	
mycorrhiza fungi 5 g polybag^{-1} (M1)	36.67 a	2 = 9.59
mycorrhiza fungi 10 g polybag^{-1} (M2)	46.67 a	3 = 10.04
mycorrhiza fungi 15 g polybag^{-1} (M3)	30.00 a	4 = 10.31
mycorrhiza fungi 20 g polybag^{-1} (M4)	40.00 a	5 = 10.49
mycorrhiza fungi 25 g polybag^{-1} (M5)	33.33 a	6 = 10.61

Note: The numbers are followed by the same letters in the same column are not significantly different on DRMT 0.05%

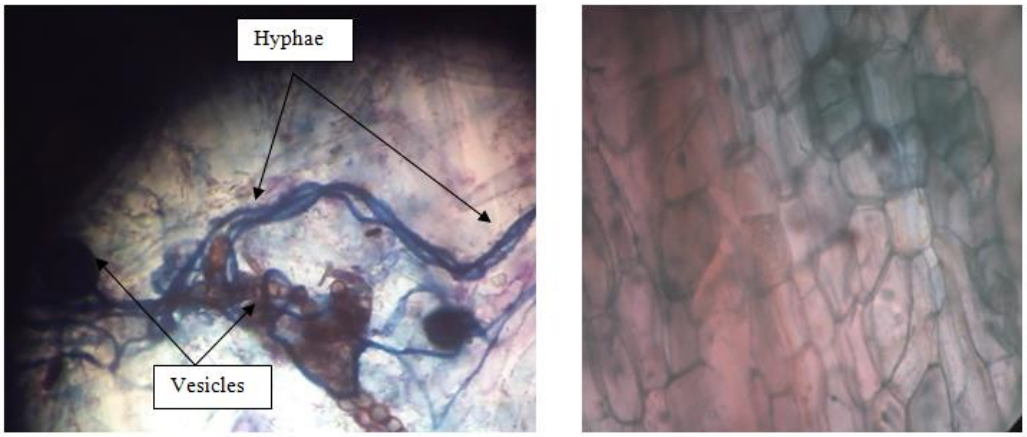

Fig. 1: The form mycorrhiza fungi colonization on pepper plant roots characterized by hyphae and visicles (a), the roots were not colonized by mycorrhiza fungi (b)

The results of research showed that the mycorrhiza fungi associated with roots of pepper plants are characterized by vesicles and hyphae contained in the root tissues of pepper seedlings (Figure 1). The percentage of mycorrhiza fungi infections on the roots of seedlings pepper plants highest in the treatment of 10 g polybag^{-1} (M2) as such as 46.67%, while the lowest occurred in the treatment of 15 g polybag^{-1} (M3) as such as 30.00%, although statistically all treatments only differing real with controls. External hyphae of mycorrhiza fungi that infect the roots of the plant will expand the field of root uptake of water and nutrients (Kabirun, 2002; Musfal, 2008). In addition, the size of hyphae a very fine in root hairs allow the hyphae can infiltrate into the pores of the soil of the most delicate that hyphae absorb water on soil moisture conditions were very low and greater water uptake by plants infected by mycorrhiza fungi will bring nutrients like N, P and K nutrient uptake by plants will increase (Kilham 1994).

Conclusions:

Plant height best until the end of the study were treated 10 g polybag^{-1} (M2) as such as 37.13 cm, number of leaves largest at the end of the study is the treatment of 5 g of polybag^{-1} (M1) as such as 10.33 strands with the percentage of mycorrhiza fungi infections on the roots of pepper plant seedlings top occurred in the treatment of 10 g polybag^{-1} (M2) as such as 46.67%.

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