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# Application of Mycorrhiza Biofertilizer to Increase Growth and Yield of Mungbean Grown following Paddy Rice during the Dry Season under Different Plant Spacing

**Wayan Wangiyana[1*], Nihla Farida[2], Dwi Ayu Sunarti[3]**
[1,2]Department of Agronomy, Faculty of Agriculture, University of Mataram, Mataram, NTB, Indonesia
[3]Graduate of the Faculty of Agriculture, University of Mataram, Mataram, NTB, Indonesia

**ABSTRACT:** In irrigated rice fields, secondary crops such as maize and pulse crops are usually grown during the dry season after harvest of paddy rice. One of the obstacles to the production of legume crops following paddy rice in the dry season is low P availability in addition to growth disturbances due to dry weather with low nighttime but high daytime temperatures.

**Purpose:** This research aimed to examine the effects of mycorrhiza biofertilizer in increasing yield of mungbean grown following paddy rice in the dry season under two planting densities.

**Methods:** The experiment was carried out in the experimental farm of the Faculty of Agriculture, University of Mataram, which is located in Narmada (West Lombok, Indonesia), from May to July 2023, which was arranged according to the Randomized Complete Block Design with six blocks and two treatment factors. The first factor was mungbean plant spacing (S1= 25x20 cm; S2= 35x20 cm) and the second one was application of mycorrhiza biofertilizer (M0= without; M1= with biofertilizer).

**Results:** The results indicated that the application of mycorrhiza biofertilizer significantly increased yield and yield components of mungbean, both per clump and per m2, in which the average grain yield was higher in the M1 (7.42 g/clump or 124.28 g/m2) than in the M0 treatment (4.76 g/clump or 78.59 g/m2). Based on the patterns of the interaction effects, the increase in AMF spore count due to the application of mycorrhiza biofertilizer was higher in S2 than in S1 treatment, whereas the increase in mungbean grain yield per m2 was higher in S1 (72.06%) than in S2 treatment (45.59%).

**Conclusion:** The plant density of mungbean grown following paddy rice during the dry seasons can be increased if mycorrhiza biofertilizer is applied to increase yield of the mungbean plants.

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**Corresponding Author: Wayan Wangiyana**

**KEYWORDS:** biofertilizer; dry season; mungbean; mycorrhiza; plant spacing.

## 1. INTRODUCTION

Mungbean seeds (*Vigna radiata* (L.) Wilczek) have many health benefits for humans due to their high content of protein, carbohydrates, vitamins, minerals, and other compounds, which have antioxidant properties [1], and because of the many essential compounds contained in the seeds, Shi et al. [2] have indicated that mungbeans have a great potential as a functional food ingredient. Skylas et al. [3] also reported that in addition to being high in protein, the seeds of various Australian mungbean varieties are also high in essential amino acids ranging from 38.1-38.7% of the total amino acids, consisting of 18 types of amino acids. Therefore, consuming mungbeans is very healthy for the human body.

Agronomically, mungbean plants are legume crops that are capable of forming a symbiosis with *Rhizobium* bacteria to fix $N_2$ gas from the atmosphere and change it into a form of N-available for mungbean plants, in addition to depositing available N compounds into the soil, especially in the rhizosphere of mungbean plants [4, 5]. Mungbeans are also suitable for agriculture development in dry land areas because they are classified as drought-tolerant and early-maturing crops [5].

If there is a competition for land use, especially in areas with adequate irrigation, farmers will definitely choose to grow higher priority crops such as rice and maize, making it less possible to produce mungbeans, even though this crop produces seeds with high nutritional and health value [2]. However, if there is further competition for land use and farmers grow crops in a monocropping system, it will be less likely for farmers to grow mungbeans, considering that the productivity and profits that can be obtained by growing mungbeans are still relatively low when compared to maize. Therefore, in rice fields, mungbeans are generally grown in the dry season, even after harvest of dry season rice, so mungbean productivity is generally very low due to drought stress.

In addition to drought stress, non-rice crops such as mungbeans grown in the dry season on land area previously used to grow flooded rice have the potential to experience P deficiency. In Australia, sunflowers grown following paddy rice often experience symptoms of P deficiency, especially plants growing around the rice stubbles, and this case is called "rice-stubble disorder", which is apparently due to the drying of former flooded rice fields causing available P to be fixed to become unavailable, especially fixed by $Fe^{3+}$ ions [6]. In conditions of changes in rice field land use patterns like this, non-rice crops require the formation of symbiosis with arbuscular mycorrhizal fungi (AMF). Soybeans grown on vertisol soil after harvest of paddy rice without tillage in the dry season had significantly higher yields if inoculated with *Rhizobium* inoculant compared to full dose application of NPK fertilizer, and even higher yields if *Rhizobium* inoculant application was accompanied by mycorrhiza biofertilizer application [7]. This study aims to determine the effect of mycorrhiza biofertilizer application on the growth and yield of mungbeans grown following paddy rice in the dry season at different plant spacing.

## II. MATERIALS AND METHODS

The field trial in this study was conducted in the experimental farm belonging to the Faculty of Agriculture, University of Mataram (Unram), which is located in Nyurlembang village, Narmada (West Lombok), Indonesia, which began in May 2023. The experiment was arranged according to a Randomized Complete Block Design, with two treatment factors, namely mungbean plant spacing (S1= 25x20 cm; S2= 35x20 cm), and application of mycorrhiza biofertilizer (M0= without; M1= with mycorrhiza biofertilizer). By combining the two treatment factors, there were four treatment combinations, each of which was made in six blocks (replications).

The experiment began with the formation of planting raised-beds. After clearing the land from grass and the residues of previous rice plants, the soil was cultivated with once plowing and once harrowing using a hand tractor, then planting raised-beds were made with a length of 2.6 m to grow 13 mungbean rows with a planting distance in rows of 20 cm, while the width of the raised-bed was 1.5 m. Between the raised-beds, a 40 cm wide and 20 cm deep ditch was made, and between the blocks a 50 cm wide ditch was made.

Planting of Vima-4 variety of mungbeans was done by dibbling 3-4 seeds per planting hole according to the plant spacing, which was finally thinned after 7 DAS (days after seeding) by leaving to grow only 2 plants per hole. Application of mycorrhiza biofertilizer was done when planting the seeds by applying "Technofert" biofertilizer at the bottom of the planting hole as much as 5 g/hole then covered with soil then the seeds were placed on top and covered again with soil. Mungbean plants were only fertilized with “Phonska” NPK fertilizer at 200 kg/ha which was dibbled after thinning had been completed, namely at 7 DAS.

Weeding and hilling were done at 3 and 5 weeks after seeding (WAS). Watering was done depending on the level of soil dryness, by flowing water through the ditch around the bed, and after all the beds were moistened, the standing water was discarded. Pest control was done according to the attack conditions, by spraying fungicides and insecticides depending on the presence or absence of attacks. Harvesting of mungbean pods was done according to the ripeness of the pods, by only harvesting pods that were already brown in color. For measuring the yield components, harvesting was carried out on 4 sample clumps per plot.

Observations variables included: plant height and number of trifoliate leaves at 56 DAS, number of productive branches, dry stover weight, number and weight of dry filled pods, weight of 100 grains, dry grain yield, number of root nodules, number of AMF spores and degree of AMF colonization on mungbean roots. Data were analyzed with ANOVA (Analysis of Variance) and Tukey's HSD using the statistical program “CoStat for Windows ver. 6.303”. The interaction pattern between treatment factors are presented in the form of a bar graph accompanied by error bars using the mean & standard error (SE) values.

## III. RESULTS

From Table 1, it can be seen that on average, the ANOVA results show that the application of mycorrhiza biofertilizer significantly increased plant height & number of trifoliate leaves at 56 DAS, as well as the number of productive branches per clump, dry stover weight per clump and per $m^2$, which indicates a positive effect of the application of mycorrhiza biofertilizer on these growth variables. In addition to the positive effect of mycorrhizal biofertilizer on the growth variables of the mungbean plants, the application of mycorrhiza biofertilizer was even more significant in increasing yield components of the mungbean plants, especially on dry grain yield per $m^2$, which almost doubled in the mungbean plants supplied with mycorrhiza biofertilizer (M1) compared to those without application of mycorrhiza biofertilizer (M0), which averaged 78.59 g/$m^2$ in M0 compared to 124.28 g/$m^2$ in the M1 treatment. In terms of AMF development and the number of root nodules, it can be seen from Table 2 that the application of mycorrhiza biofertilizer significantly increased the number of root nodules per clump, the number of spores and the degree of AMF colonization on the roots of mungbean plants.

**Table 1. Effect of mycorrhiza biofertilizer and planting distance on growth variables and yield components of post-rice mung beans**

| Observation variables | Main effect of mycorrhiza biofertilizer | | | | | Main effect of plant spacing | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | M0 | | M1 | | HSD | S1 (25 cm) | | S2 (35 cm) | | HSD |
| Plant height (cm) 56 DAS | 28.12 | b | 32.18 | a[1)] | 2.80 | 29.54 | a | 30.75 | a[1)] | 2.80 |
| Trifoliate leaf number/clump | 12.75 | b | 13.73 | a | 0.89 | 12.94 | a | 13.54 | a | 0.89 |
| Productive branch number/clump | 7.67 | b | 11.92 | a | 1.25 | 8.83 | b | 10.75 | a | 1.25 |
| Dry stover weight/clump (g) | 6.83 | b | 10.67 | a | 0.89 | 7.92 | b | 9.58 | a | 0.89 |
| Dry stover weight/m2 (g) | 114.76 | b | 180.48 | a | 16.01 | 158.33 | a | 136.90 | b | 16.01 |
| Filled pod number/clump | 10.67 | b | 17.75 | a | 1.41 | 12.42 | b | 16.00 | a | 1.41 |
| Filled pod number/m2 | 178.58 | b | 298.33 | a | 22.81 | 248.33 | a | 228.58 | a | 22.81 |
| Filled pod dry weight (g/clump) | 6.67 | b | 11.58 | a | 1.29 | 8.33 | b | 9.92 | a | 1.29 |
| Filled pod dry weight (g/m2) | 112.38 | b | 195.95 | a | 20.38 | 166.67 | a | 141.67 | b | 20.38 |
| Grain number/clump | 91.17 | b | 149.50 | a | 17.26 | 100.67 | b | 140.00 | a | 17.26 |
| Grain number/m2 | 1514.76 | b | 2498.57 | a | 296.98 | 2013.33 | a | 1999.99 | a | 296.98 |
| Weight of 100 grains | 5.06 | a | 5.47 | a | 0.47 | 4.99 | b | 5.53 | a | 0.47 |
| Grain yield (g/clump) | 4.76 | b | 7.42 | a | 1.20 | 5.07 | b | 7.11 | a | 1.20 |
| Grain yield (g/m2) | 78.59 | b | 124.28 | a | 20.44 | 101.30 | a | 101.57 | a | 20.44 |

[1)] Mean values followed by the same letter are not significantly different between treatments of each factor

**Table 2. Average AMF spore number, %-colonization, and nodule number on mungbean roots for each treatment level**

| Treatment factors | Number of AMF spore per 10 g rhizosphere soil | | %-colonization of mungbean roots by AMF | | Number of root nodule on mungbean roots per clump | |
|---|---|---|---|---|---|---|
| M0: without mycorrhiza | 114.08 | b | 29.21 | b | 14.33 | b[1)] |
| M1: with mycorrhiza | 159.08 | a | 36.67 | a | 23.50 | a |
| HSD 5% | 8.97 | | 2.58 | | 1.66 | |
| S1: 25x20 cm | 132.42 | a | 30.46 | b | 17.42 | b |
| S2: 35x20 cm | 140.75 | a | 35.42 | a | 20.42 | a |
| HSD 5% | 8.97 | | 2.58 | | 1.66 | |

[1)] Mean values followed by the same letter are not significantly different between treatments of each factor

However, there was an interaction effect of treatment factors on several observation variables (Figure 1 to Figure 4), which showed that the increase in the weight of 100 grains and grain yield per $m^2$ due to the application of mycorrhiza biofertilizer was higher under the narrower plant spacing (25x20 cm), namely 12.2% for the weight of 100 grains (Figure 3) and 72.1% for grain yield (Figure 2), compared with those in the wider plant spacing (35x20 cm). In contrast, the increase in spore count due to application of mycorrhiza biofertilizer is higher in the wider than in the narrower plan spacing (Figure 4).

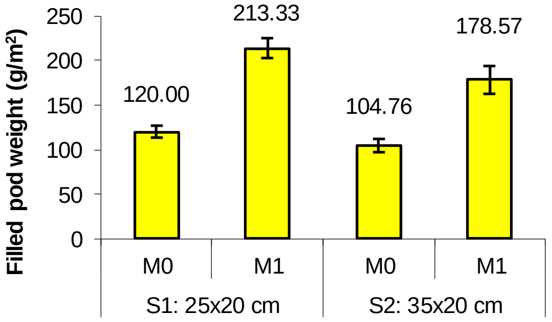


**Figure 1. Weight of dry filled pods (Mean ± SE) as affected by interaction effect of the treatment factors**

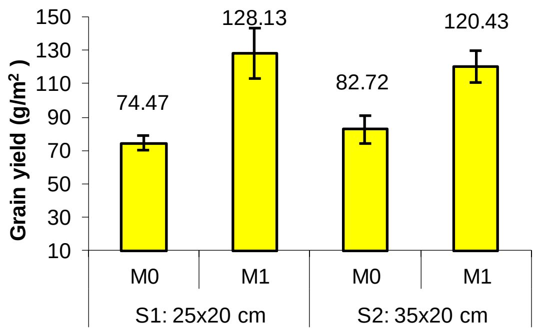


**Figure 2. Grain yield in g/$m^2$ (Mean ± SE) as affected by interaction effect of the treatment factors**

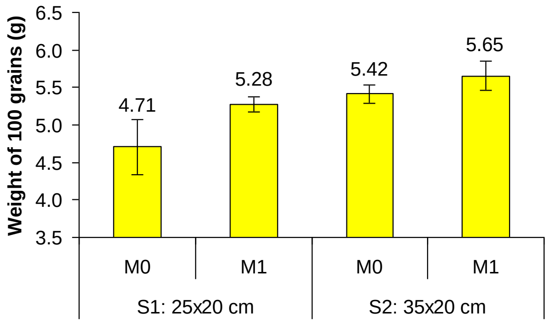


**Figure 3. Weight of dry filled pods (Mean ± SE) as affected by interaction effect of the treatment factors**

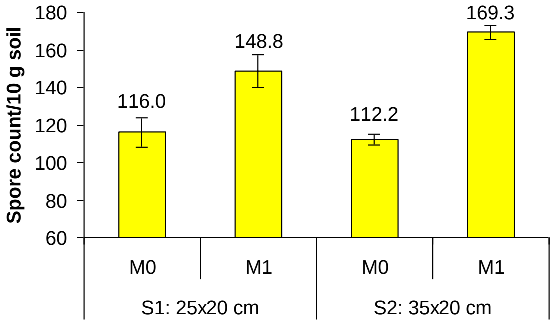


**Figure 4. Number of AMF spores in the soil (Mean ± SE) as affected by interaction effect of the treatment factors**

## IV. DISCUSSION

The significant increase in mungbean grain yield due to the application of mycorrhiza biofertilizer indicates a positive role of AMF contained in mycorrhiza biofertilizer in increasing the ability of the mycorrhizal mungbean plants to uptake more nutrients [8, 9]. Soybean plants that were direct-seeded after harvest of paddy rice in the dry season on vertisol land, even without NPK fertilizer application, also showed a significant increase in grain yield due to the application of mycorrhiza biofertilizer compared to the plants that received the recommended dose of NPK fertilizer, especially if mycorrhiza biofertilizer was combined with application of *Rhizobium* inoculant [7]. Gough et al. [10] also reported that AMF showed a synergistic effect with root nodule bacteria (*Bradyrhizobium* sp) in increasing root nodule formation, growth and yield of mungbean plants. This can happen because mungbean plants can form a tripartite symbiosis with AMF and root nodule bacteria [11].

In terms of AMF development and the number of root nodules, it can be seen from Table 2 that the application of mycorrhiza biofertilizer significantly increased the number of root nodules per clump, the number of spores and the degree of AMF colonization in the roots of mungbean plants. This indicates that the application of exogenous AMF inoculants does not have a problem with indigenous AMF in the soil. In addition, it was suspected that the indigenous AMF population was very low the rice land was always used to grow paddy (flooded) rice, which was reported to reduce the AMF population in the soil [12, 13]. In relation to the number of nodules, Gough et al. (2021) [10] also found that AMF inoculation can increase the number of root nodules in mungbean root system.

On the other hand, plant spacing in general only affected dry stover weight and yield components per clump, which were higher under the wider plant spacing (35x20 cm), but when calculated per $m^2$, there was no significant yield difference between the two treatments of plant spacing, although the weight of filled pods per $m^2$ was higher under narrower plant spacing (25x20 cm), as was also the weight of dry stover (Table 1). In relation to the development of AMF and the number of root nodules between plant spacing treatments, it can be seen from Table 2 that the number of root nodules and %-colonization of AMF in mungbean roots were higher under the wider plant spacing. This can occur because these two types of microbes live symbiotically in the cortex tissue of the roots and their development depends on the supply of carbohydrates from mungbean plants. Because the biomass of mungbean plants was higher under the wider plant spacing, it means that the supply of carbohydrates was higher under the wider plant spacing. In addition, both the number of root nodules and %-colonization of AMF were positively and significantly correlated with the dry weight of mungbean plants, with the regression equation $Y = 2.18 + 1.91 X$ ($R^2 = 66.8\%$; $p < 0.001$) for the number of nodules, and $Y = 16.66 + 1.86 X$ ($R^2 = 50.5\%$; $p < 0.001$) for %-colonization of AMF.

However, due to the significant interaction of the treatment factors on several observation variables (Figure 1 – Figure 4), it means that responses of those observation variables to the application of mycorrhiza biofertilizer varied depending on the plant spacing. Grain yield per $m^2$ (Figure 2), for example, showed a higher increase due to application of mycorrhiza biofertilizer in the narrower plant spacing (72.1%) than in the wider plant spacing (45.6%). The implication of this finding is that the application of mycorrhiza biofertilizer to mungbean plants in the dry season after paddy rice is very helpful in increasing grain yields per unit area, and with the application of this biofertilizer, the plant spacing can be narrowed, which in addition to significantly increasing grain yields, the narrower plant spacing has the potential to increase the ability of mungbean plants to compete with weeds due to the increase in the number of leaves per clump as well as per $m^2$ with the application of mycorrhiza biofertilizer. Growing mungbean during the dry season would make the plants to experience water stress, but through mycorrhizal symbiosis, the detrimental effects of water stress can be alleviated by the mycorrhizal fungi contained in the mycorrhiza biofertilizer [14]. Growing mungbean on the autoclave sterilized soil also reduced the tolerance of mungbean to water stress compared with growing it on the non-sterilized soil [15].

## V. CONCLUSION

The application of mycorrhiza biofertilizer significantly increased grain yield and yield components of mungbeans, both per clump and per $m^2$, in which the average grain yield was higher in M1 treatment (7.42 g/clump or 124.28 g/$m^2$) compared with in M0 treatment (4.76 g/clump or 78.59 g/$m^2$). Grain yield per $m^2$ did not differ significantly between plant spacing treatments, but due to the significant interaction, the increase in mungbean grain yield per $m^2$ due to the application of mycorrhiza biofertilizer (M1) was higher, namely 72.1%, in S1 (25x20 cm) than in S2 (35x20 cm), which was only 45.6%.

Therefore, with the application of mycorrhiza biofertilizer, it is better to narrow plant spacing so that there is an opportunity to increase the ability of mung bean plants to compete better with weeds due to the increase in the number of mungbean trifoliate leaves per $m^2$, which was higher at the narrower plant spacing treatment.

## VI. ACKNOWLEDGMENTS

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## VII. DISCLOSURE

We do not have any conflicts of interest in this work.

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