YIELD OF MUNGBEAN RELAY-PLANTED AT DIFFERENT DATES AND SPACINGS BETWEEN DOUBLE-ROWS OF MYCORRHIZA-BIOFERTILIZED MAIZE PLANTS

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ABSTRACT
This research aimed to examine yield performance of mungbean (Vigna radiata (L) Wilczek) cv. “Kenari” relay-planted at different dates and spacings between double-rows of maize plants that were either fertilized or unfertilized with mycorrhiza biofertilizer. The field experiment, conducted on dry land in Pringgabaya village, East Lombok, Indonesia, from August to December 2019, was arranged according to Split Split Plot design with three blocks and three treatment factors, i.e. application of mycorrhiza biofertilizer to maize plants as main plots (M0= without; M1= with biofertilizer), relay-planting dates of mungbean between double rows of maize plants as subplots (T1= -10; T2= 0; T3= +10 days of maize planting date), and mungbean row spacings as sub-subplots (S1= 50 cm; S2= 33 cm). Results indicated that yield components of mungbean relay-planted between double rows of maize were significantly affected by application of mycorrhiza biofertilizer to maize plants, relay-planting dates of mungbean, and row spacings of mungbean, except for the weight of 100 grains. There were significant interaction effects, but only between mycorrhiza application to maize and planting dates of mungbean on weight of dry filled-pods and grain number per clump as well as their intercropping ratios, indicating that without mycorrhiza application to maize plants, grain number of the mungbean plants per clump was the most badly affected by delaying planting dates of mungbean, but under micorrhiza biofertilized maize plants, grain number was not affected by the delay, especially between D-10 and D0, indicating positive effect of mycorrhiza biofertilizer in this intercropping system.

KEY WORDS
Additive intercropping, maize, mungbean, mycorrhiza biofertilizer, plant spacings.

Seeds of mungbean (Vigna radiata (L.) Wilczek) plants have a lot of health benefits for humans due to their high contents of protein, carbohydrates, vitamins, minerals, and other compounds, having antioxidant properties [1], and due to many essential compounds contained in the seeds, Shi et al. [2] have indicated that mungbean is becoming highly potential as functional food. Skylas et al. [3] also reported that in addition to high in proteins, seeds of various Australian mungbean varieties are high in essential amino acids ranging from 38.1-38.7% of the total amino acids, consisting of 18 types of amino acids. Based on results of grain analysis of 16 mungbean varieties reported by Li et al. [4], proximate composition the mungbean grains were crude protein 24.26–28.50%, crude fibre 3.21–4.18%, crude fat 0.57–1.86%, ash content 3.64–4.24%, moisture 7.49–8.45% and carbohydrates 54.25–58.69%, respectively.

Mungbean can be consumed by directly processing the seeds or consumed as mungbean sprouts, but the sprouts can contain higher vitamin C, vitamin A and proteins compared with the seeds, and they can be used to alleviate malnutrition or “hidden hunger”, especially in South Asia, Africa and the Pacific [5]. In India, mungbean is grown for human food, and it is mostly consumed as dhal, by cooking the split grains, i.e. processed mungbean grains by removing the seed coat [5]. In Indonesia, mungbean is also considered...
as functional food [6], but the most dominant utilization of mungbean grains is as vegetable in the form of mungbean sprouts [7].

In terms of agronomic properties, mungbean is a highly potential crop to be grown in rotation with rice and cereal crops where these cereal crops are predominantly cultivated, such as in the Indo-Gangetic Plains of India due to the potential of mungbean to fix atmospheric N\textsubscript{2} through biological nitrogen fixation and improve N content of the soil [5]. In Indonesia, mungbean is also mostly grown during the dry season in rotation with rice or maize when other crops are unable to grow well due to unavailability of irrigation water or rainfall during the peak of dry season, and these conditions are some limiting factors for improvement of mungbean production in Indonesia [7]. Based on the national statistical crop production data [https://bps.go.id/subject/53/tanaman-pangan.html#subjekViewTab3], in terms of the total harvested area in 2015, mungbean is the fifth widely grown crops after rice, maize, soybean and peanut, in which mungbean total harvested area was only 1.6% of that of rice crop (i.e. only 229,475 ha), indicating the least importance of mungbean among those food crops in spite of its high health values.

Nevertheless, mungbean can be a highly potential crop for expansion to marginal lands and semi-arid regions due to its short duration (it can be harvested in 55-65 days after seeding) and its tolerance to drought [8], [5]. Moreover, mungbean can also be grown in multiple cropping or intercropping with other crops [5]. Some researchers reported that cultivation of mungbean in intercropping with other crops can increase efficiency of land use indicated by a LER (land equivalent ratio) value of more than 1.00, such as those reported by Lingga \textit{et al.} [9], in which intercropping one row of sweet corn with six rows of mungbean resulted in the highest LER (LER>1). Lestari \textit{et al.} [10] also reported that among four mungbean varieties intercropped with maize, the Vima-3 variety of mungbean resulted in the highest LER, i.e. 1.56, but all mungbean varieties resulted in LER value of higher than 1.0, which means that growing mungbean in intercropping with maize can increase land use efficiency. In addition, Mayasari and Wangiyana [11] reported that different varieties of mungbean resulted in different yield relative to its monocrop, in which grain yield of “Vima-3” variety was not affected by intercropping but yield of “Vima-1” variety was significantly lower in intercropping while yield of “No.129” variety was higher in intercropping with red rice plants in aerobic irrigation system than in their monocrop.

This study aimed to examine the effect of mycorrhiza bio-fertilization of maize (\textit{Zea mays} L.) plants on yield components of mungbean plants (\textit{Vigna radiata} (L) Wilczek) cv. Kenari, which were relay-planted between double-rows of maize plants at different planting dates and row spacing of the mungbean plants.

**MATERIALS AND METHODS OF RESEARCH**

This study applied an experimental method by conducting a field experiment on the experimental farm of “IP2TP Balitbangtan BPTP NTB” located in Labuhan Haji village, Pringgabaya district, East Lombok, Indonesia, from August to December 2019. The maize variety used was “Srikandi Kuning”, which is a Quality Protein Maize (QPM) variety, and the mungbean variety was “Kenari”.

The experiment was arranged according to Split Split Plot (SSP) design, by testing 3 treatment factors, namely mycorrhiza application to maize plants (M0 = without or M1 = with application of mycorrhiza biofertilizer); relay-planting dates of mungbean between rows of maize (T1= planting mungbean 10 days before planting maize [D-10], T2= planting mungbean on the same day as planting maize [D-0], T3= planting mungbean 10 days after planting maize [D+10]); mungbean row spacing (S), by relay-planting either one (S1= 50 cm) or two (S2= 33 cm) rows of mungbean plants between double rows of maize (100 cm distance). Each treatment combination was made in three blocks (replications).

The land for the experiment was prepared by first conducting once tillage (once plowing & once harrowing), followed by plotting with plot size of 4 x 2 m\textsuperscript{2} surrounded with forrows of 40 cm width between plots or 50 cm width between blocks, and 25 cm depth. Maize was planted using a double row system (Fig. 1) with a planting distance of 100 cm
between double rows and 50 cm between rows of a double row and plant spacing of 20 cm within rows. Only one maize plant and two mungbean plants were allowed to grow per planting hole, by conducting thinning at 10 days of seeding of each crop. Mungbean was planted with plant spacing of 50 x 20 cm (for S1) and 33 x 20 cm (for S2) treatment, by seeding pre-germinated mungbean seeds of either one (S1) or two (S2) rows between double rows of maize with planting dates depending on the treatments, either -10, 0 or +10 days of planting maize (no mungbean was planted within the double row). For maize plants with mycorrhiza application, the planting holes were made deeper than for those without mycorrhiza, and then the mycorrhiza biofertilizer (Technofert biofertilizer, containing several species of arbuscular mycorrhizal fungi (AMF) supplied by “BPPT Serpong”, Indonesia) was placed at the bottom of the planting hole (8 g / planting hole) then covered with soil, and maize seeds were placed on it and were covered with soil.

![Figure 1](image1.png)

Figure 1 – Lay out of the plants in each treatment plot [A= maize plants (x) intercropped with two rows of mungbean (o); B= maize plants (x) intercroppe with one row of mungbean (o), C= monocrop maize]

Fertilization of maize plants was done twice, namely at 10 days after planting (DAP) using Phonska fertilizer (NPK 15-15-15) at a dose of 2.5 g/plant (equivalent to 250 kg/ha) and at 35 DAP using Urea fertilizer (45% N) at a dose of 1.25 g/plant (equivalent 125 kg / ha). For mungbean plants, fertilization was done only once at 10 DAP using Phonska fertilizer at a dose of 2.0 g/clump (equivalent to 200 kg/ha). Fertilizers were applied by dibbling then in the plant row as far as 7 cm from stem base of the plants at 5-7 cm depth. Weeding was done at 2 and 5 weeks after planting maize by hand weeding. Watering the plots was done six times by flooding the plots for 30 minutes and then draining the flood water for each time of watering. Plant protection was done by spraying suspension of Prevathon 50 SC at a concentration of 1 ml/2 liters of water for controlling caterpillar attack. Harvest was done at 105 DAP for maize ears and at 60 DAP for mungbean pods and stover.

The observations variables were yield components of mungbean, namely weight of dry filled pods, number of filled pods, number of seeds, weight of seeds, weight of dry stover and weight of 100 seeds, as well as intercropping ratios of those variables to their monocrop. Data were analyzed with Analysis of Variance (ANOVA) and Tukey’s HSD test at 5% level of significance, using the statistical software CoStat for Windows ver.6.303. The intercropping ratios agains monocrop were defined as the fractional ratio of mungbean yield components between intercropping and monocropping systems; thus, if intercropping ratio (IR) of grain yield is less than 1.00, it means that there is lower grain yield in intercropping than in its corresponding monocropping system. For showing interaction effects, graphs are presented using mean values and standard error (Mean ± SE) of each combination of treatments compared in the graphs, as suggested by Riley [12].

RESULTS AND DISCUSSION

The ANOVA results summarized in Table 1 show that mycorrhiza application to maize plants and relay-planting dates of mungbean between double rows of maize plants significantly affected all yield components of mungbean except the weight of 100 dry grains. Row spacing of mungbean plants relay-planted between double rows of maize plants also
significantly affected yield components of mungbean per clump, except for the weight of dry 100 grains and ratio of filled pod number between intercropping and monocropping system. There were also significant interaction effects between treatment factors, but only between mycorrhiza application to maize plants (M) and relay-planting dates of mungbean (D), and their interaction effects were significant only on weight of dry filled-pods and grain number per clump and their intercropping ratios (IR).

Table 1 – Summary of ANOVA results on the effects of mycorrhiza application to maize (Myc), and planting dates and spacing of mungbean between maize rows on yield components of mungbean and its intercropping ratio (IR).

| Observation variables | Main Effects | Interaction Effects |
|-----------------------|--------------|---------------------|
|                       | Myc | Date | Spacing | M*D | M*S | D*S | M*D*S |
| Weight of dry filled-pods | ** | *** | ** | * | ns | ns | ns |
| Filled pod number | * | *** | ** | ns | ns | ns | ns |
| Grain number per clump | ** | *** | ** | * | ns | ns | ns |
| Grain yield per clump | ** | *** | *** | ns | ns | ns | ns |
| Stover dry weight | ** | *** | * | ns | ns | ns | ns |
| Weight of 100 grains | ns | ns | ns | ns | ns | ns | ns |
| IR weight of dry filled-pods | ** | *** | ** | * | ns | ns | ns |
| IR filled pod number | * | *** | ns | ns | ns | ns | ns |
| IR grain number per clump | ** | *** | * | ns | ns | ns | ns |
| IR grain yield per clump | ** | *** | ** | ns | ns | ns | ns |
| IR stover dry weight per clump | ** | *** | * | ns | ns | ns | ns |

Note: ns = non-significant; *, **, *** = significant at p < 0.05; p < 0.01; p < 0.001, respectively.

Table 2 – Mean values of yield components of mungbean relay-planted between double rows of maize at different dates and spacings

| Treatments | Weight of dry filled-pods (g/clump) | Filled pod number per clump | Grain number per clump | Grain yield (g/clump) | Weight of dry stover (g/clump) | Weight of 100 dry grains |
|------------|-------------------------------------|-----------------------------|------------------------|------------------------|-------------------------------|--------------------------|
| Mycorrhiza on maize: | | | | | | |
| M0 | 5.54 | b | 8.40 | b | 66.56 | b | 3.61 | b | 8.39 | b | 5.78 | a<sup>1</sup> |
| M1 | 8.35 | a | 12.74 | a | 93.79 | a | 5.46 | a | 10.26 | a | 6.49 | a |
| HSD | 1.07 | | 2.31 | | 7.85 | | 0.78 | | 0.76 | | 2.99 |
| Mungbean planting dates: | | | | | | |
| D-10 | 8.77 | a | 13.69 | a | 97.25 | a | 5.96 | a | 10.51 | a | 6.23 | a |
| D0 | 6.95 | b | 10.02 | b | 80.38 | b | 4.55 | b | 9.45 | a | 6.14 | a |
| D-10 | 10.13 | c | 8.00 | c | 62.90 | c | 3.11 | c | 8.01 | b | 6.04 | a |
| HSD | 0.79 | | 1.98 | | 10.80 | | 0.70 | | 1.16 | | 0.77 |
| Mungbean spacing: | | | | | | |
| S1: 50 cm | 7.57 | a | 11.49 | a | 85.75 | a | 4.98 | a | 9.97 | a | 6.21 | a |
| S2: 33 cm | 6.33 | b | 9.65 | b | 74.60 | b | 4.09 | b | 8.06 | b | 6.07 | a |
| HSD | 0.68 | | 1.21 | | 7.54 | | 0.44 | | 1.08 | | 0.57 |

<sup>1</sup> Mean values in each column followed by the same letters are not significantly different between levels of each treatment factor based on Tukey’s HSD test.

Based on the main effects of each treatment factor on yield components of mungbean (Table 2), it is clear that application of mycorrhiza biofertilizer to maize plants significantly increased weight of dry filled-pods, filled-pod number, grain number, grain yield, and weight of dry stover, but this treatment did not significantly increase weight of 100 dry grains of mungbean. Based on the mean values in Table 2, it can be seen that increased grain yield of mungbean plants additively relay-planted between double rows of maize plants was mainly caused by an increase in filled-pod number and grain number per clump since the increase in weight of 100 grain was not significant. This means that the pod filling stage was the most affected (improved) growth stage of mungbean plants by application of mycorrhiza biofertilizer to maize plants in intercropping with mungbean additively relay-planted between double rows of the maize plants. Otherwise, without application of mycorrhiza biofertilizer to the maize plants, relay-planting mungbean between double rows of the maize plants would
significantly reduce both filled-pod number and grain number of those mungbean plants, especially under later planting dates, as can be seen from Table 2, that filled-pod number and grain number per clump were significantly lower on mungbean relay-planted on later planting dates. Thus, intercropping mungbean in additive series through relay-planting it between rows of mycorrhiza-biofertilized maize plants resulted in higher yield and yield components of mungbean compared with relay-planting it between rows of maize plants with no application of mycorrhiza biofertilizer.

In relation to these positive effects of involving mycorrhizal fungi in intercropping systems, several researchers have also reported that there were interspecific nutrient transfers between plants in intercropping through hyphae of arbuscular mycorrhizal fungi (AMF) colonizing roots of both plant species, such as those reported by Bethlenvalvay et al. [13] and Hamel et al. [14], in which nutrient transfer was measured using isotopic techniques. Red rice plants grown together in one pot with mungbean plants were also reported to show better growth and higher yield on pots fertilized with mycorrhiza biofertilizer compared with on pots without application of mycorrhiza biofertilizer [15].

Even though without application of mycorrhizal fungi in their experiment, Inal et al. [16], who measured levels of availability of nutrients in the rhizospheres and nutrient contents in the shoots, found that levels of nutrient availability were higher in rhizospheres of both peanut and maize in intercropping than in monocropping system, which resulted in higher nutrient contents in the shoots of both crops in intercropping compared with in monocropping system. Polthanee and Trelo-ges [17] also reported higher P and K contents of the leaves of mungbean in intercropping with maize than in monocropping system, in which the maize and legume crops were seeded on the same day, but intercropping significantly reduced seed yields of peanut, soybean and mungbean per plant, principally due to reduced number of pods per plant. Nevertheless, intercropping maize with these legume crops resulted in LER values of 1.66, 1.60, and 1.48 with peanut, soybean and mungbean, respectively.

| Treatments | Mycorrhiza on maize | Mungbean planting dates | Mungbean spacing |
|------------|---------------------|-------------------------|------------------|
|            | Weight of dry filled pods (g/clump) | Intercropping Ratio to Monocrop (IR) | Weight of dry stover (g/clump) |
|            | Filled pod number per clump | Seed number per clump | Grain yield (g/clump) | |
| M0         | 0.58 b                | 0.72 b                  | 0.67 b           | 0.54 b | 0.62 b |
| M1         | 0.87 a                | 1.10 a                  | 0.95 a           | 0.82 a | 0.76 a |
| HSD 0.05   | 0.11                  | 0.19                    | 0.09             | 0.12   | 0.06   |
|            |                      |                        |                  |        |
| D-10       | 0.90 a                | 1.14 a                  | 0.93 a           | 0.88 a | 0.75 a |
| D0         | 0.71 b                | 0.85 b                  | 0.82 b           | 0.66 b | 0.68 b |
| D+10       | 0.57 c                | 0.74 b                  | 0.68 c           | 0.50 c | 0.63 b |
| HSD 0.05   | 0.08                  | 0.16                    | 0.11             | 0.10   | 0.08   |

1/ Mean values in each column followed by the same letters are not significantly different between levels of each treatment factor based on Tukey’s HSD test.

In this study, additive intercropping by relay-planting mungbean between double rows of maize also reduced mungbean grain yield compared with its yield in monocropping system as indicated by IR values of less than 1.00 (Table 3). However, by planting mungbean 10 days prior to planting maize (D-10), intercropping resulted in higher filled-pod number of mungbean per clump compared with in monocropping system, and delaying planting dates significantly reduced filled-pod number as well as grain yield of mungbean per clump (Table 3).
In addition, mungbean plant spacing also significantly affected yield components of mungbean, except for the weight of 100 grains (Table 2). Reducing mungbean plant spacing, by relay-planting two rows of mungbean between double rows of maize plants also reduced intercropping ratios of yield components of mungbean per clump compared with relay-planting only one row of mungbean between double rows of maize plants, except for filled-pod number per clump (Table 3). Sarlak et al. [18] also reported that mixing ratio between sweet corn and mungbean in intercropping systems significantly affected LER values, in which mixing ratio of sweet corn and mungbean of 75%/25% with 8 corn plants per m² was found to result in the highest land equivalent ratio (LER), i.e. 1.08, and relative yield of mungbean biomass (RYm) was highest in this mixing ratio, i.e. 0.704, in which sweet corn and mungbean were planted on the same day.

However, there were significant interaction between mycorrhiza application to maize plants and relay-planting dates of mungbean especially on dry filled-pod weight and grain number per clump (Fig. 1 and Fig. 2) as well as their intercropping ratios (Fig. 3 and Fig. 4) of mungbean relay-planted between double rows of the maize plants. These mean that the effects of relay-planting dates on these yield components of mungbean depend on whether or not the maize plants were fertilized with mycorrhiza biofertilizer.

Based on Fig. 1 and Fig. 2, weight of dry filled-pods per clump (Fig.1) and grain number per clump (Fig. 2) were highly significantly reduced by delaying the dates of relay-planting mungbean between double rows of maize plants, when the maize plants were not fertilized with mycorrhiza biofertilizer. However, when the maize plants were biofertilized, there were no significant reduction in weight of dry filled-pods and grain number per clump, especially between D-10 and D0. Thus, it can be said that fertilization of maize plants with
mycorrhiza biofertilizer could increase yield components of mungbean plants that are relay-planted between rows of the maize plants even though in an additive series. Not only yield components of mungbean per clump were increased, but the intercropping ratios were also increased by application of mycorrhiza biofertilizer to the maize plants in intercropping with mungbean, as can be seen from Table 3. These mean that adaptation ability of mungbean plants to lower resources, such as sunlight intensity and nutrients in the soil due to relay-planting them in additive series between rows of the maize plants, were increased when they were grown under the canopies of maize plants fertilized with mycorrhiza biofertilizer compared with under canopies of those with no application of mycorrhiza biofertilizer. Intercropping ratios of grain number per clump of mungbean plants relay-planted between rows of mycorrhiza biofertilized maize plants were not significantly different between planting dates in contrast to significant reduction in the intercropping ratios with delayed planting dates of mungbean between non-mycorrhizal maize plants (Fig. 4). This indicates significant contribution of involving AMF in intercropping systems of maize and mungbean.

In relation to contribution of AMF to facilitation of growth and nutrient uptake by component crops in intercropping between upland rice and soybean, Li et al. [19] reported that this intercropping significantly improved the formation of arbuscular mycorrhizas, particularly in the upland rice roots, which increased total P uptake by 57% in rice, total P and N acquisition by 65% and 64% respectively in mungbean, and nodulation by 54% in mungbean. Not only biologically fixed N can be transferred between plants interconnected with AMF hyphal networks infecting roots of both crops, but also hydraulically lifted water [20], which could happen from mycorrhizal maize to mungbean plants in this case, as well as carbon and phosphorus transfer, such as between aerobic rice and water melon [21]. In addition, Sinclair and de Wit [22], from their investigation on 24 crop species including legume and cereal crops, concluded that seed production of legume crops is highly constrained by nitrogen nutrition of the legume crops because of their high protein contents of the grains. Thus, the possibility of higher P and N acquisition, and higher nodulation, as well as better water relation of mungbean in intercropping with maize plants fertilized with mycorrhiza biofertilizer would possibly increase the rates of seed-filling and grain formation by the mungbean plants, as can be seen from Fig. 2 that grain number per clump was significantly higher in mungbean relay-planted between rows of mycorrhiza biofertilized maize plants compared with between maize plants with no mycorrhiza application.

CONCLUSION

It can be concluded that application of mycorrhiza biofertilizer to maize plants increased yield components of mungbean additively relay-planted between double rows of maize plants at different relay-planting dates.

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