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Growth and yield of Mung bean (*Vigna radiata* L.) as affected by *Rhizobium* sp. bacteria inoculant and frequency of watering

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Abstract. The aim of the study was to know the influence of the *Rhizobium* bacteria inoculant doses as biotic fertilizer and the frequency of watering on the growth and yield of the mung bean (*Vigna radiata* L.). The study was conducted in Green House of Faculty Animal and Agricultural Sciences, Diponegoro University. The experimental design of this research was Completely Randomized Design (CRD) The parameters observed were height of plant, number of leaves, days of blooming, and days of pod. The result showed that the dose of the *Rhizobium* inoculant had a significant effect on days of blooming. The frequency of watering had significant effect on height of plant, number of leaves, days of the bloom, days of the podded and weight of pods. In conclusion, mung bean that was treated by *Rhizobium* sp. bacteria inoculant (treatments of R2, R3 and R4) and treatment of F3 had same in days of blooming and days of podded with bacteria inoculant RHIZOKA (treatment of R1).

1. Introduction

Mung bean is a plant which is from the legume class. Mung bean is an important commodity of legume plants after soybeans and peanuts. Compared with other types of legumes, mung beans have the advantage of agronomic and economic aspects, namely, fewer pests and diseases, can be harvested at the age of 55-60 DAP and a higher selling price than soybeans [1]. Mung bean are the most widely used by humans. Improving the processed industry made from mung beans makes the demand for mung beans in Indonesia continue to increase. The high demand has not been accompanied by the availability of mung beans supply by farmers. In 2015 until 2017, the production and harvest area of mung beans decreased, respectively 3.57% and 7.36%. Increasing mung bean production can be optimized by using less productive lands, one of which is dry land. Water-deficient soil conditions can cause disruption to plant physiology and microorganisms in the soil. One technology that is expected in dealing with drought conditions is the use of *Rhizobium* sp. bacteria as biological fertilizer.

Global climate change which has an impact on drought also becomes one of the obstacles in the cultivation of mung beans. Drought is a condition where water needs are not met, so that it will affect plant metabolism, including microorganisms associated with these plants. In general, inoculation of *Rhizobium* sp. into the ground, intended to be able to associate with plants, thereby increasing the ability to bind N in the air. Therefore, it is necessary to test the association of *Rhizobium* sp. in mung bean plants in soil conditions with different levels of drought.



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2. Materials and Methods

The research was conducted in Green House of the Faculty of Animal and Agricultural Science, Diponegoro University, Semarang and continued with laboratory analysis at the Laboratory of Ecology and Plant Production, Faculty of Animal and Agricultural Science, Diponegoro University. The experimental design of this research was Completely Randomized Design (CRD) factorial with two factors namely dose of *Rhizobium* sp. bacteria inoculant (R) which consist of R0 = control, R1 = positive control RHIZOKA (1,8 mg/ plant of commercial inoculant), R2 = 5 mg/ plant inoculant of *Rhizobium*, R3 = 10 mg/ plant inoculant of *Rhizobium* and R4 = 15 mg/ plant inoculant of *Rhizobium* while frequency of watering (F) which consist of five standards that F1 = a time in a day, F2 = a time in two days and F3 = a time in three days. With 3 replications. The material needed is mung bean variety VIMA-3, isolate of *Rhizobium* sp. (from mung bean roots in Demak), ingredients for making 1 liter of Yeast Mannitol Agar (YEMA) + Congo Red (CR) medium consisting of 0.5 g K_2HPO_4 ; 0.2 g $MgSO_4$; 0.1 g NaCl; 10 g Mannitol; 1 g of yeast extract; 20 g of bacto agar; 2.5 ml of Congo Red 1%. Materials for making liquid media of *Rhizobium* sp., peat, NPK fertilizer, soil. The tools needed are 30 cm diameter polybags, barrels, stoves, hoes, sieves, stirers, autoclaves, scales, lamina air flow, test tubes, erlenmeyer, bunsen, analytical scale, ruler, oven, ose and shaker.

3. Results and Discussion

The presence of *Rhizobium* sp. bacteria associated with root nodules is influenced by environmental factors. *Rhizobium* sp. Bacteria. live at optimum environmental conditions. According to Surtiningsih and Nurhayati [2] which states that the *Rhizobium* sp. live at a temperature of 18-26° celcius, a minimum temperature of 3° celcius and a maximum temperature of 30° celcius. Temperature in the soil is influenced by the temperature and water received by the soil. This experiment examines the effect of *Rhizobium* sp. with different watering frequency.

Based on the analysis it is known that the treatment of watering frequency has a significant effect while the rhizobium inoculum dose treatment and the interaction of both have no significant effect.

Table 1. Height of Plant (cm) of Mung bean that Innoculated with Different Dosage of *Rhizobium* sp. Bacteria and Different Frequency of Watering

<i>Rhizobium</i> Inoculant Bacteria	Frequency of Watering			Mean
	F1	F2	F3	
R0 (control)	50.66 ab	44.00 abcde	36.66 cdef	43.78
R1 (positive control)	43.33 abcde	40.33 bcdef	32.66 ef	38.78
R2 (5 mg/ plant)	54.00 a	42.66 abcdef	33.00 ef	43.22
R3 (10 mg/ plant)	49.33 ab	42.00 bcdef	34.00 def	41.78
R4 (15 mg/ plant)	46.66 abc	44.66 abcd	31.33 f	40.89
Mean	48.80 a	42.73 b	33.53 c	(-)



Figure 1. Interaction of the Height of Plant on Mung Bean that Innoculated by *Rhizobium* sp.

Table 1. and Figure 1., it showed that the tallest of the height plant is F1 treatment (48.80 cm), and F3 (31.53 cm) is the shortest. Treatment of F1 had significant different with F2 and F3. The height of mung beans planted every day is higher than that of plants that are watered every other day and every three days. Daily watering treatment makes the plant height higher by 12.4% and 31% in F2 and F3, respectively. Mung bean plants that are watered once a day have the highest value. Research by Daba and Tadese [3] reported that *Moringa olifera* plants that are watered with sufficient water capacity consistently every day have optimum growth compared to those that are not.

From Table 1., it is known that the plant height in the *Rhizobium* dose treatment was not significantly different. Mung bean plant height given inoculum *Rhizobium* sp. with various doses or without given an inoculum there is no difference. From **Table 1.** we know that there is no significant interaction between the treatments that are given.

Table 2. Number of Leaves (trifoliat) of Mung bean that Innoculated with Different Dosage of *Rhizobium* sp. Bacteria and Different Frequency of Watering

Rhizobium Inoculant	Frequency of Watering			Mean
	F1	F2	F3	
Bacteria				
R0 (control)	5.00 ^a	5.00 ^a	4.33 ^{ab}	4.78 ^a
R1 (positive control)	5.00 ^a	4.66 ^{ab}	3.66 ^{abc}	4.44 ^{ab}
R2 (5 mg/ plant)	4.66 ^{ab}	4.00 ^{abc}	3.00 ^c	3.89 ^b
R3 (10 mg/ plant)	5.00 ^a	4.33 ^{abc}	4.33 ^{ab}	4.44 ^{ab}
R4 (15 mg/ plant)	5.00 ^a	4.66 ^{ab}	3.33 ^{bc}	4.33 ^{ab}
Mean	4.93 ^a	4.47 ^a	3.73 ^b	

^{a,b,c} Means in the same row with different letters show significant differences ($p < 0.05$)

From Table 2., it is known that the frequency of watering in F1 and F2 treatments did not differ, while the F1 treatment was significantly different from F3 and F2 treatment was significantly different from F3. The number of leaves on mung bean plants that are watered every day has a number of leaves more than 24% compared to mung beans plants that are watered once every 3 days. According to Kalaydjieva *et al.* [4] showed that irrigation influences the leaf growth of *Phaseolus vulgaris* plants. Water as a photosynthetic material, if the amount is reduced, it will inhibit photosynthetic synthesis. This can inhibit the process of growth and formation of organs from plants.

Based on Table 2., it is known that the administration of different dosage of inoculum does not have a significant effect on the number of leaves of mung bean plants. The results of the statistical

analysis showed no interaction of the dose of *Rhizobium* sp. Inoculum. and frequency of watering on the number of leaves of mung bean plants.

Table 3. Day of Blooming (day after plant) of Mung bean that Innoculated with Different Dosage of *Rhizobium* sp. Bacteria and Different Frequency of Watering

Rhizobium Inoculant Bacteria	Frequency of Watering			Mean
	F1	F2	F3	
R0 (control)	39.66 ^{ab}	41.66 ^{ab}	40.33 ^{ab}	40.56 ^a
R1 (positive control)	40.00 ^{ab}	40.00 ^b	42.33 ^{ab}	40.78 ^b
R2 (5 mg/ plant)	36.66 ^b	36.66 ^b	43.00 ^a	38.78 ^{ab}
R3 (10 mg/ plant)	38.00 ^b	43.33 ^{ab}	35.33 ^b	38.89 ^a
R4 (15 mg/ plant)	40.00 ^b	39.33 ^b	45.33 ^a	41.56 ^b
Mean	38.87 ^b	40.20 ^b	42.27 ^a	

Means in the same row with different letters show significant differences ($p < 0,05$)

From Table 3., it is known that the time of emergence of plant flowers treated with watering frequency treatment in F1 (39 HST) and F2 (40 HST) treatments did not differ, whereas F1 treatment was significantly different from F3 (42 HST) and F2 treatment was significantly different from F3. When the flowers appear on the mung bean plants that are watered every day and every two days have a shorter flowering time than the mung beans that are watered every 3 days.

The results of the statistical analysis showed that there was an interaction of *Rhizobium* sp. and the frequency of watering when the mung bean flower plants appear. The fastest time to appear was R3F3 (35 HST) treatment while the longest time to appear was R4F3 (45 HST). Mung bean plants which are treated with *Rhizobium* 5 mg inoculum / plants watered every three days, have a flower time that is equivalent to the mung bean plants that are applied with RHIZOKA with watering every day.

Based on Table 3., it is known that the administration of different dosage of inoculum does not have a significant effect on the flowering time of mung bean plants. Administration of *Rhizobium* sp. increasing nitrite so that it can be used by legume plants, but nitrite is not dominantly used in generative growth such as flower formation. According to Farias *et al.* [6] which stated that the administration of *Rhizobium* inoculums did not give significant effect during the flowering period of the *Vigna unguiculata* plant.

Table 4. Days of Podded (days after plant) of Mung bean that Innoculated with Different Dosage of *Rhizobium* sp. Bacteria and Different Frequency of Watering

Rhizobium Inoculant Bacteria	Frequency of Watering			Mean
	F1	F2	F3	
R0 (control)	38.33 ^c	43.66 ^{abc}	40.00 ^{bc}	40.66 ^{ab}
R1 (positive control)	43.33 ^{abc}	41.66 ^{abc}	45.00 ^{abc}	43.33 ^b
R2 (5 mg/ plant)	39.66 ^{bc}	39.00 ^c	44.00 ^{abc}	40.88 ^{ab}
R3 (10 mg/ plant)	40.33 ^{bc}	44.33 ^{abc}	49.66 ^a	44.77 ^a
R4 (15 mg/ plant)	40.33 ^{bc}	40.00 ^{bc}	48.33 ^{ab}	42.88 ^b
Mean	40.40 ^b	41.73 ^b	45.40 ^a	

Means in the same row with different letters show significant differences ($p < 0,05$)

From Table 4., it is known that the time of emergence of plant pods treated with watering frequency treatment in F1 (40 HST) and F2 (42 HST) treatments did not differ, while F1 treatment was significantly different from F3 and F2 treatment was significantly different from F3 (45 HST). When the pods appear on the mung beans plant watered every day and every two days have a shorter time of appearing pods than the mung beans plant watered once every 3 days.

The results of the statistical analysis showed that there was an interaction of *Rhizobium* sp. and the frequency of watering when the pods appear in the mung bean plants. The fastest time for pods to appear was R0F1 (38 HST) treatment while the longest pods appeared was R3F3 (50 HST) treatment. When pods appeared in R2F3 plants were not significantly different or the same as the treatment of R1F1, R2F1, R3F1 and R4F1. Mung bean plants which were applied with *Rhizobium* sp. 5 mg / pot with watering every three days has a pod emergence time equivalent to the treatment without *Rhizobium* application, RHIZOKA inoculum application and *Rhizobium* inoculum application that is watered every day. According to Ntambo *et al.* [5] which states that *Rhizobium* bacteria inoculation can increase the appearance of pods in soybean plants, this is related to increased nitrite intake so that plants can survive in unfavorable conditions.

Based on Table 4., it is known that the different dosage of inoculum does not have a significant effect on the time the pods appear on the mung bean plant.

Table 5. Weight of Pods (g) of Mung bean that Inoculated with Different Dosage of *Rhizobium* sp. Bacteria and Different Frequency of Watering

<i>Rhizobium</i> Inoculant Bacteria	Frequency of Watering			Mean
	F1	F2	F3	
R0 (control)	19.10 ^a	11.50 ^{bcd}	15.83 ^{ef}	15.80
R1 (positive control)	15.30 ^{ab}	9.17 ^{bcd}	2.50 ^f	9.00
R2 (5 mg/ plant)	14.00 ^{abc}	5.50 ^{de}	2.00 ^f	7.17
R3 (10 mg/ plant)	13.80 ^{ab}	7.67 ^{dc}	3.00 ^{ef}	8.17
R4 (15 mg/ plant)	11.67 ^{abcd}	12.50 ^{abcd}	1.30 ^f	8.50
Mean	14.80 ^a	9.27 ^b	4.93 ^c	

^{a,b,c} Means in the same row with different letters show significant differences ($p < 0.05$)

From Table 5., showed that frequency of watering treatment has interaction with weight of pods. It is known that weight of pods treated with watering frequency treatment in F1 (14.80 gram) was significantly different from F2 (9.27 gram) and F3 (4.93 gram) treatment.

The results of the statistical analysis showed that there was an interaction of *Rhizobium* sp. and the frequency of watering in the weight of the mung bean pods. Based on Table 4., it is known that the different dosage of inoculant also didn't have a significant difference for weight of pods in mung bean. Based on research by Hussain *et al.* [7] reported that phosphorus needed more than nitrogen when developing of pods.

4. Conclusion

There were significant interaction effect on the days of the bloom and days of the podded. Mung bean that was treated by *Rhizobium* sp. bacteria inoculant (treatments of R2, R3 and R4). It had same result of days of blooming and days of podded with bacteria inoculant RHIZOKA (treatment of R1). It recommend to use *Rhizobium* sp. inoculant to make faster blooming with 5 mg/ plant (treatment of R2).

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