

Contribution of *Rhizobium*-Mycorrhiza-Merapi-indigenous *Rhizobacteria* Association on Growth and Yield of Three Cultivars Soybean Cultivated on Coastal Sandy Soil

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ABSTRACT

A study was conducted to examine the effect of inoculum association between *Rhizobium* sp., mycorrhizae and Merapi-indigenous *Rhizobacteria* on the growth and yield of 3 soybean cultivars, and to determine the best inoculum and cultivars for soybean cultivation on coastal sandy soil. The study was conducted in the Agro-biotechnology and Research Laboratory and experimental station of Faculty of Agriculture, Universitas Muhammadiyah Yogyakarta during the period of September 2015 to June 2016. Experiments were conducted by using coastal sandy soil as planting medium in polybags by employing 4 x 3 factorial experiments, arranged in completely randomised design, and placed under the field condition. The first factor used was inoculation treatment consisted of 4 combination of inoculums: (1) *Rhizobium* sp. - mycorrhizae, (2) *Rhizobium* sp. - Merapi-indigenous *Rhizobacteria*, (3) *Rhizobium* sp. - mycorrhizae - Merapi-indigenous *Rhizobacteria*, and (4) without inoculation. The second factor was soybean cultivars consisted of 3 varieties: (1) Grobogan, (2) Detam-1, and (3) Petek. Observation was carried out on nodulation, mycorrhizal effect, *Rhizobacteria* population dynamics, plant growth and yield. The results showed that *Rhizobium* sp.-mycorrhizae inoculated on Petek increased root growth, leaf area and yield (5,97 tonnes/ha). *Rhizobium* sp.-mycorrhizae inoculation only increased diameter of nodules. It was also observed that the best soybean cultivar for coastal sandy soil was Petek.

Keywords: Soybean cultivars, *Rhizobium* sp., Mycorrhizae, *Rhizobacteria* indigenous of Merapi, Coastal sandy soil

ABSTRAK

Tujuan penelitian adalah mengetahui pengaruh asosiasi inokulum pada pertumbuhan dan hasil 3 varietas kedelai dan menetapkan asosiasi inokulum dan varietas yang sesuai untuk pengembangan kedelai di lahan pasir pantai. Penelitian dilaksanakan di Laboratorium Agrobioteknologi, Laboratorium Penelitian dan lahan percobaan Fakultas Pertanian, Universitas Muhammadiyah Yogyakarta pada bulan September 2015 hingga Juni 2016. Penelitian menggunakan rancangan percobaan faktorial (4 x 3) yang disusun dalam Rancangan Acak Lengkap (RAL) dengan media tanam pasir pantai. Faktor pertama adalah perlakuan inokulum yang terdiri dari 4 macam yaitu (1) *Rhizobium* sp.-Mikoriza, (2) *Rhizobium* sp.-*Rhizobacteria* indigenous Merapi, (3) *Rhizobium* sp.-Mikoriza-*Rhizobacteria* indigenous Merapi dan (4) tanpa inokulum. Faktor kedua adalah kultivar kedelai yang terdiri dari 3 varietas yaitu (1) Grobogan, (2) Detam-1 dan (3) Petek. Pengamatan dilakukan terhadap aktivitas nodulasi, pengaruh mikoriza, dinamika populasi *Rhizobacteria*, pertumbuhan perakaran, pertumbuhan vegetatif, dan hasil. Hasil penelitian menunjukkan bahwa inokulasi *Rhizobium* sp. - mikoriza hanya berpengaruh terhadap diameter nodul dan inokulasi *Rhizobium* sp.-mikoriza pada varietas Petek nyata meningkatkan pertumbuhan perakaran, luas daun dan hasil biji (5,97 ton/ha).

Kata kunci: Varietas kedelai, *Rhizobium* sp., Mikoriza, *Rhizobacteria* indigenous Merapi, Lahan pasir pantai

INTRODUCTION

Soybean has an important role as a protein source (Rahmat and Yuyun, 1996). Central Bureau of Statistics (Badan Pusat Statistik) [2015] stated that soybean production decreased by 5,38% yearly during the period of 2009 until 2013. Production of soybean was 850.000 tonnes in 2012, but soybean demand was forecasted to be 2.4 million tonnes. It was suggested that demand for tempeh and tofu reach 1.6 million tonnes and black soybean for soy sauce about

650,000 ton (Aditama, 2011). These figures suggest that, to catch up with the demand, soybean production needs to be increased by about 1.55 million tonnes.

Intensification such as high-yielding cultivars and biological fertilizer may provide ways to increase soybean production. Several soybean cultivars, such as Grobogan contains high protein and also high-yielding (Erliana *et al.*, 2009), while local cultivar from Boyolali, Petek, is known as

drought resistant cultivar, and Detam-1 cultivar contains high protein (45.36%) as well as high-yielding (3.45 tonnes/ha, [Balitkabi, 2008]).

Extensification may be conducted by using marginal area (Arie, 2013), such as coastal area which has the potential use of 1.060.000 ha. One of the drawbacks of coastal area is that it has low fertility, low nutrient and high porosity. Application of biological fertilizer may provide the alternative way to resolve coastal sandy soil problem. Some microorganisms are known to improve soil fertility such as *Rhizobium* sp., mycorrhiza and several indigenous *Rhizobacteria* such as Merapi-indigenous *Rhizobacteria*. *Rhizobium* sp. is known to reserve Nitrogen, while mycorrhiza reserve Phosphorus, and an indigenous *Rhizobacteria*, Merapi-indigenous *Rhizobacteria*, has been demonstrated to improve plant resistance to drought (Gunawan, 2014; Muhammad *et al.*, 2014).

Double inoculation of soybean with *Rhizobium* sp. and mycorrhiza resulted in the increase of nitrogen on coastal sandy soil and kept humidity of rhizosphere (Gunawan, 2014). On the other hand, single inoculation using *Rhizobium* sp. did not increase plant dry weight and leaf area yet (Lilik, 2005). Previous study also demonstrated that double inoculation using *Rhizobium* sp. and mycorrhiza, and single inoculation using only *Rhizobium* sp. resulted not significantly different effect on the number of pod, grain weight and yield. Double inoculation of soybean using *Rhizobium* sp.-mycorrhiza cultivated on coastal sandy soil produced only 25% of yield potential. Similarly, osmotolerant *Rhizobacteria-Rhizobium* sp. association did not increase soybean growth and yield (Ngadiman *et al.*, 2014). It is anticipated that inoculation of Grobogan cultivar using *Rhizobium* sp.-mycorrhiza – Merapi-indigenous *Rhizobacteria*, cultivated on coastal sandy soil, will increase soybean growth and yield which.

This study was, therefore, conducted to examine the contribution of inoculum association between *Rhizobium* sp., mycorrhizae and Merapi-indigenous *Rhizobacteria* on the growth and yield of 3 soybean cultivars, and to determine the best inoculum and cultivars for soybean cultivation on coastal sandy soil.

MATERIALS AND METHODS

Materials used in this study were *Rhizobium* sp., Merapi-indigenous *Rhizobacteria* MB and MD isolates (Agung_Astuti, 2012 personal communication), crude inoculum of mycorrhiza, Grobogan, Petek and Detam-1 soybean cultivars. The study was conducted in the Agro-biotechnology Research Laboratory and experimental station of Faculty of Agriculture, Universitas Muhammadiyah Yogyakarta during the period of September 2015 to June 2016.

Experiments were conducted by using coastal sandy soil as planting medium in polybags, arranged in completely randomized design (CRD) of 4 x 3 factorial experiments and placed under the field condition. The first factor used was inoculation treatment consisted of 4 combinations of inoculum: (1) *Rhizobium* sp. – Mycorrhiza, (2) *Rhizobium* sp. – Merapi-indigenous *Rhizobacteria*, (3) *Rhizobium* sp. – Mycorrhiza – Merapi-indigenous *Rhizobacteria*, and (4) without inoculation. The second factor was soybean cultivars consisted of 3 varieties: (1) Grobogan, (2) Detam-1, and (3) Petek. Observation was carried out on nodulation, mycorrhizal effect, *Rhizobacterial* population dynamics, plant growth and yield.

The final results were analysed using ANOVA (analysis of variance) with α of 5%. The significantly different treatment was further tested by Duncan's multiple range test (Duncan Multiple). Periodic observation data were presented on graphs and histograms.

RESULTS AND DISCUSSION

The Effect of treatment on root nodulations

Rhizobium sp. infection is indicated by the formation of root nodule which indicates the compatibility with the plant. Table 1 shows the average number of nodules parameter presented following inoculation.

Table 1. Average number of nodule, nodule effectiveness, nodule weight and diameter in the ninth week

Treatment	Number of nodule*	Effective nodule percentage (%)***	Weight of nodule (g)*	Diameter of nodule (mm)*
Inoculum:				
<i>Rhizobium</i> sp.- mycorrhiza	8,76 a	65,42 a	0,38 a	3,74 a
<i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	2,78 a	37,78 a	0,05 b	1,59 a
<i>Rhizobium</i> sp.- mycorrhiza- <i>Rhizobacteria</i>	2,56 a	49,17 a	0,12 b	2,46 a
Without inoculum	1,78 a	20,83 a	0,15 b	1,22 a
Soybean cultivar:				
Grobogan	6,42 p	49,17 p	0,31 p	2,86 p
Detam-1	4,08 p	31,11 p	0,14 p	1,72 p
Petek	1,33 p	49,62 p	0,08 p	2,18 p
Interaction	(-)	(-)	(-)	(-)

Note: numbers followed by the same letter showed no significant difference based on the F test α 5% and DMRT.

(-) No interaction between treatments

* square root data transformation

*** arc-sin and square root data transformation

It was observed that soybean cultivar did not significantly affect nodulation as *Rhizobium* sp. was found effectively nodulated all soybean cultivar. Soybean cultivar which was inoculated by *Rhizobium* sp.-mycorrhiza showed significantly different number of nodule (Lilik, 2005; Yudhy and Inorah, 2009). Ayu *et al.* (2013) suggested that the formation of nodule and nodule activity were influenced by phosphor from the mycorrhiza activities. The average of nodulation activities is presented in Table 1. It was the more effective nodule formed, the more nitrogen fixed and subsequently more chlorophyll and enzyme synthesised. The increase of chlorophyll and enzyme synthesis resulted in the increase of photosynthesis and vegetative, generative growth (yield) (Ramdana and Retno, 2015). Mycorrhiza is known

to increase water availability in the rhizosphere, which will result in the effective nodule and increased weight of nodule (Nike-Triwahyuningsih, 2004). The result showed that the formation of nodule in coastal sandy soil was longer than in fertile soil. Mycorrhiza is also known to increase diameter and effectiveness of nodule. The larger nodule diameter, the higher nodule effectivity as the larger diameter gives indication that the development of *Rhizobium* sp. inside nodule is good. Small nodules indicate slow growing tissue bacteroid, which subsequently reduced the effectiveness of nitrogen fixation (Ramdana and Retno 2015).

Effect of treatment on the effectivity of mycorrhizal inoculation

The results of this study demonstrated that inoculum and cultivar treatment have the same effect on mycorrhizal-infected root percentage. *Rhizobium* sp.-mycorrhiza association, however, demonstrated higher mycorrhizal-infected root percentage during 9 week. *Rhizobium* sp. produces Nod Factor as growth regulator for mycorrhiza, therefore Nod factor induces colonization and development of mycorrhiza through nodogen induction (van Brussel *et al.*, 1986 cit Xie *et al.*, 1995). Table 2 showed the number of spores as the indicator of mycorrhizal growth, while the average of mycorrhizal-infected root percentage is presented in Table 3.

Table 2. Average number of mycorrhizal spore (spore/ml) x104 in the ninth week

Treatment	<i>Rhizobium</i> sp.-mycorrhiza	<i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	<i>Rhizobium</i> sp.-mycorrhiza- <i>Rhizobacteria</i>	Without inoculum	Average
Grobogan	616,67 a	166,67 cd	283,33 bc	533,33 a	400,00
Detam-1	66,67 d	358,33 b	275,00 bc	616,67 a	329,17
Petek	233,33 bc	375,00 b	166,67 cd	325,00 b	275,00
Average	305,56	300,00	241,67	491,67	(+)

Notes: numbers followed by the same letter showed no significant difference based on the F test α 5% and DMRT.

(+) There was interaction between treatments

Table 3. Average of mycorrhizal-infected root percentage in the ninth week

Treatment	Mycorrhiza infection percentage (%)**
Inoculum:	
<i>Rhizobium</i> sp.- mycorrhiza	98,89 a
<i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	95,56 a
<i>Rhizobium</i> sp.- mycorrhiza- <i>Rhizobacteria</i>	97,78 a
Without inoculum	94,44 a
Soybean cultivar:	
Grobogan	95,00 p
Detam-1	97,50 p
Petek	97,50 p
Interaction	(-)

Notes: numbers followed by the same letter showed that no significantly difference based on the F test α 5% and DMRT.

(-) No interaction between treatments

** arc-sin data transformation

The different root exudates of each cultivar may contribute to the difference of microorganisms thrive in the rhizosphere, which may also affect the mycorrhizal growth. The presence of microorganism compatible with each cultivar may also increase plant growth. It was suggested that the factors which affect plant host may also affect to mycorrhizal growth (Tutik *et al.*, 2016).

The effect of treatment on activity of Merapi-*indigenus Rhizobacteria*

After inoculation was observed Merapi-*indigenus Rhizobacteria* population dynamics population. Microbial population dynamics for 9 weeks was presented in Figure 1.

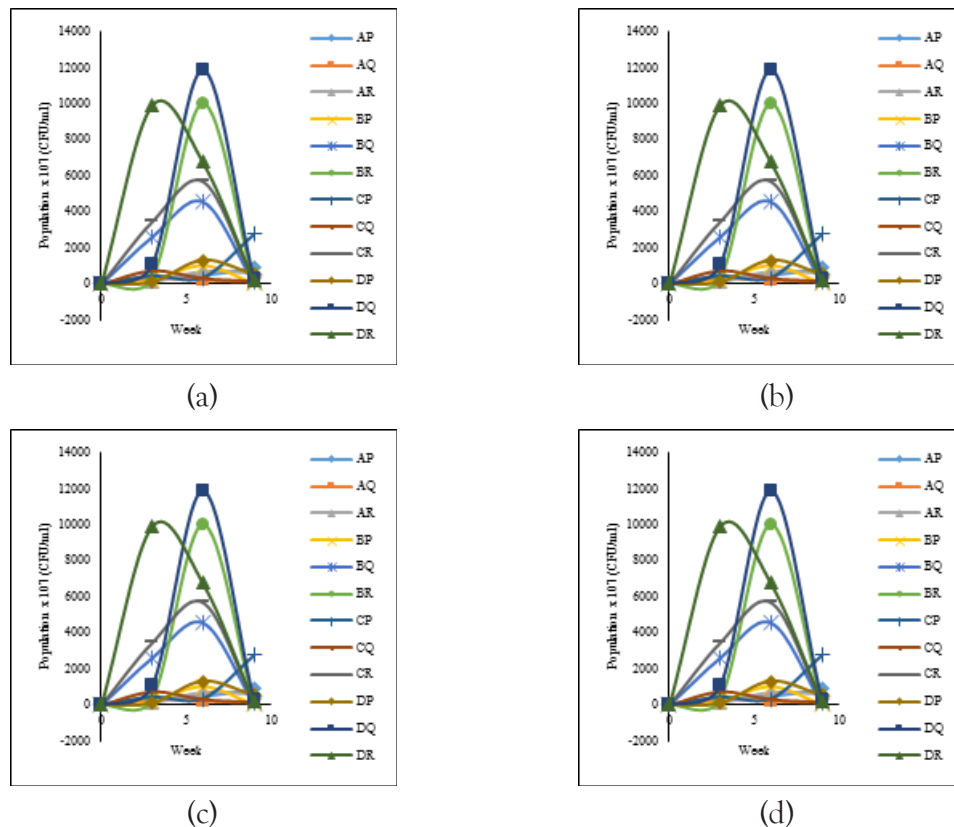


Figure 1. Population dynamics: (a) total bacteria, (b) other bacteria, (c) Merapi *indigenus Rhizobacteria*, MB isolate and (d) Merapi *indigenus Rhizobacteria*, MD isolate on three soybean cultivars

Remark:

- | | |
|--|--------------|
| A = <i>Rhizobium</i> sp.-mycorrhiza | P = Grobogan |
| B = <i>Rhizobium</i> sp.- <i>Rhizobacteria</i> | Q = Detam-1 |
| C = <i>Rhizobium</i> sp.-mycorrhiza <i>Rhizobacteria</i> | R = Petek |
| D = Without inoculum | |

Figure 1 showed that group of *Rhizobacteria* which compatible to each cultivar. MB and MD isolates in combination with mycorrhiza and *Rhizobium* sp. demonstrated the ability to adapt well as evidenced by the increase of population in the sixth week. Figure 1 (c) showed that Grobogan cultivar inoculated with MB isolate in combination with *Rhizobium* sp.-mycorrhiza demonstrated good adaption as shown by entering the log phase in the 0 – 3rd week, while other treatment still in the adaptation phase. On the other hand, the population of MD isolate (Figure 1.d) on all of treatment showed tendency to decrease, with the exception of Detam-1 cultivar inoculated with *Rhizobium* sp.-mycorrhiza-*Rhizobacteria* that the *Rhizobacteria* reached the highest level of log phase until the sixth week, and reached the death phase after nine weeks. It was also observed that inoculation with mycorrhiza increased the Merapi-indigenous *Rhizobacteria* growth. Marcia *et al.* (2011) suggested that the positive effect of one microbe to another is attributed by the metabolite secretion.

Root proliferation of three soybean cultivars cultivated on coastal sandy soil

Root proliferation illustrates the spread of the roots on the media. It was observed that inoculum and cultivar treatments resulted in a similar effect on root proliferation due to the presence of indigenous mycorrhizal spore and the facts that all treatments were infected by mycorrhiza including treatment without inoculum. Mycorrhiza-legumes association may increase the availability of phosphorus, as well as the increase of nitrogenase activity which promoted root proliferation (Sylvia *et al.*, 2005).

Root system is influenced by both genetic factors and growth media. Most of soil nutrients are absorbed from soil solution through the root. It

was found that inoculum and cultivar treatments showed interacting effect on root length. The uninoculated Detam-1 cultivar showed tendency to have longer root as mycorrhiza-legume association may increase root proliferation (Sylvia *et al.*, 2005). The average length of root, fresh and dry weight of root are presented in Table 4.

Table 4. Average length of root, fresh and dry weight of root after nine weeks

Treatment	Length of root (cm)	Fresh root weight (g)	Dry root weight (g)
Grobogan- <i>Rhizobium</i> sp.- mycorrhiza	60,83 ab	15,90 bcde	2,08 bcde
Grobogan- <i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	40,83 bc	10,08 e	1,48 de
Grobogan- <i>Rhizobium</i> sp.- mycorrhiza- <i>Rhizobacteria</i>	55,00 abc	11,12 e	1,33 de
Grobogan-without inoculum	54,83 abc	12,11 de	1,12 e
Detam-1- <i>Rhizobium</i> sp.- mycorrhiza	59,00 abc	21,65 ab	2,22 bcde
Detam-1- <i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	59,33 abc	11,31 e	1,68 cde
Detam-1- <i>Rhizobium</i> sp.- mycorrhiza- <i>Rhizobacteria</i>	66,30 a	23,88 ab	2,80 bc
Detam-1- without inoculum	72,17 a	19,82 abcd	2,38 bcd
Petek- <i>Rhizobium</i> sp.- mycorrhiza	67,67 a	24,47 a	4,13 a
Petek- <i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	65,67 a	21,05 abc	2,39 bcd
Petek- <i>Rhizobium</i> sp.- mycorrhiza- <i>Rhizobacteria</i>	43,50 bc	12,81 cde	1,83 bcde
Petek- without inoculum	57,67 abc	18,37 abcde	2,96 b
Interaction	(+)	(+)	(+)

Notes: numbers followed by the same letter showed that no significantly difference based on the F test α 5% and DMRT.
(+) There was interaction between treatments

Rhizobium sp.-mycorrhiza-Petek interaction resulted in nutrient absorption and high photosynthesis accumulation on the root, which increased fresh and dry roots. Such increased was supported by the high number of leaf and leaf area on Petek. It was suggested that factors affecting photosynthesis were genetic and the absorption of nutrients from soil (Gardner *et al.*, 1991).

Vegetative growth of three soybean cultivars cultivated on Coastal Sandy Soil

Table 6 lists growth parameter analysed. It is demonstrated that almost all growth parameters were affected by cultivar except fresh and dry

canopy weight. Several studies also suggest that cultivar affects plant height (Wayan *et al.*, 2011; Rosi and Santi, 2012), number of leaf (Wayan *et al.*, 2011) and flowering date (Anna, 2015; Rosi and Santi, 2012). Suhartina (2005) showed that Detam-1 height was higher than Grobogan and Petek suggesting that every cultivar brings out different character. Every cultivar has different ability to absorb nutrients resulting in different number of leaf and leaf area (Yutono, 1988 *in*: Farida, 2004). Gardner *et al.* (1991) also suggested that every cultivar showed different photosynthetic rates, while flowering date is affected by genetic factors, length of days and temperature (Suyanto and Musalamah, 2010).

Table 5. Average plant height, number of leaf, fresh and dry canopy weight, after nine weeks and the flowering date

Treatment	Plant height (cm)	Number of leaf	Fresh canopy weight (g)*	Dry canopy weight (g)*	Flowering date
Inoculum:					
<i>Rhizobium sp.- mycorrhiza</i>	45,51 a	35,31 a	66,97 a	14,83 a	33,30 a
<i>Rhizobium sp.-Rhizobacteria</i>	43,34 a	22,56 a	49,02 a	10,27 a	35,89 a
<i>Rhizobium sp.- mycorrhiza-Rhizobacteria</i>	41,64 a	23,19 a	47,87 a	10,15 a	35,81 a
Without inoculum	42,31 a	31,33 a	68,23 a	15,34 a	35,11 a
Soybean cultivar:					
Grobogan	36,38 q	15,86 q	44,90 p	10,84 p	28,92 r
Detam-1	46,58 p	26,88 q	56,33 p	11,01 p	33,11 q
Petek	46,64 p	41,56 p	72,84 p	16,08 p	46,06 p
Interaction	(-)	(-)	(-)	(-)	(-)

Note: numbers followed by the same letter showed no significant difference based on the F test α 5% and DMRT.

(-) No interaction between treatments
* square root data transformation

Leaf area was found affected by cultivar and inoculum. Average leaf area in the ninth week is presented in Table 7. Petek, in combination with *Rhizobium sp.-mycorrhiza* resulted in significantly higher leaf area. Every cultivar has different ability to absorb nutrients resulting in different leaf area (Yutono, 1988 *in*: Farida, 2004), while the

inoculum increased nutrient availability (*note: there is no data supporting this notion*). Mycorrhiza inoculation on soybean cultivated on coastal sandy soil increased nitrogen and kept the humidity surrounding the rhizosphere (Gunawan, 2014).

Table 6. Average leaf area in the ninth week (cm²)*

Treatment	<i>Rhizobium sp.- mycorrhiza</i>	<i>Rhizobium sp.- Rhizobacteria</i>	<i>Rhizobium sp.- mycorrhiza-Rhizobacteria</i>	Without inoculum	Average
Grobogan	744,00 d	699,33 d	720,33 d	465,33 d	657,25
Detam-1	837,00 cd	1347,00 bcd	854,33 cd	1938,67 abc	1244,25
Petek	2779,67 a	1066,67 cd	1190,33 cd	2383,33 ab	1855,00
Average	1453,56	1037,67	921,67	1595,78	(+)

Notes: numbers followed by the same letter showed no significant difference based on the F test α 5% and DMRT.

(-) No interaction between treatments
* square root data transformation

Yield Components of Three Cultivars Soybean Cultivated on Coastal Sandy Soil

The productivity of plant was the ultimate goal of cultivation, including the cultivation of soybeans. Table 7 shows the average soybean yield components.

Table 7. Average number of pod, filled-pod percentage, pod dry weight and weight of 100 grains

Treatment	Number of pod*	Filled-pod percentage (%)*	Pod dry weight (gram)*	Weight of 100 grains (gram)
Inoculum:				
<i>Rhizobium sp.- mycorrhiza</i>	128,83 a	24,93 a	11,64 a	4,75 a
<i>Rhizobium sp.-Rhizobacteria</i>	142,75 a	26,97 a	12,59 a	5,19 a
<i>Rhizobium sp.- mycorrhiza-Rhizobacteria</i>	115,50 a	15,51 a	10,94 a	4,57 a
Without inoculum	128,44 a	18,49 a	10,14 a	4,51 a
Soybean cultivar:				
Grobogan	45,62 r	6,72 q	7,06 r	5,51 p
Detam-1	127,40 q	19,27 pq	15,78 p	4,49 p
Petek	213,63 p	38,43 p	11,14 q	4,26 p
Interaction	(-)	(-)	(-)	(-)

Note: numbers followed by the same letter showed no significant difference based on the F test α 5% and DMRT.

(-) No interaction between treatments
* square root data transformation

Cultivar was found to affect the number of total pods (Yudhy and Inorah, 2009), filled-pod percentage and pod dry weight. *Rhizobium sp.-*

Rhizobacteria association also showed tendency to have the best result on number of pod, pod dry weight, grain weight and weight of 100 grains. Some of PGPR bacteria have been demonstrated to develop association which resulted in the increase of nodulation and nitrogen absorption of legume (Figueiredo *et al.*, 2011). Petek was the cultivar with the highest number of pod and filled-pod percentage, while Detam-1 and Grobogan were the cultivars with highest pod dry weight, and the highest weight of 100 grains, respectively.

Table 8. Average grain weight and yield

Treatment	Grain weight (g)*	Yield (ton/ha)*
Grobogan- <i>Rhizobium</i> sp.- mycorrhiza	0,74 b	0,33 b
Grobogan- <i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	2,01 b	0,89 b
Grobogan- <i>Rhizobium</i> sp.- mycorrhiza- <i>Rhizobacteria</i>	1,86 b	0,83 b
Grobogan-without inoculum	0,44 b	0,19 b
Detam-1- <i>Rhizobium</i> sp.- mycorrhiza	1,26 b	0,56 b
Detam-1- <i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	2,91 b	1,29 b
Detam-1- <i>Rhizobium</i> sp.- mycorrhiza- <i>Rhizobacteria</i>	1,41 b	0,63 b
Detam-1- without inoculum	2,95 b	1,31 b
Petek- <i>Rhizobium</i> sp.- mycorrhiza	13,43 a	5,97 a
Petek- <i>Rhizobium</i> sp.- <i>Rhizobacteria</i>	10,47 a	4,65 a
Petek- <i>Rhizobium</i> sp.- mycorrhiza- <i>Rhizobacteria</i>	2,91 b	1,29 b
Petek- without inoculum	4,41 b	1,96 b
Interaction	(+)	(+)

Notes: numbers followed by the same letter showed that no significantly difference based on the F test α 5% and DMRT.

(-) No interaction between treatments
* square root data transformation

Grain weight and yield on Petek in combination with *Rhizobium* sp.-mycorrhiza-*Rhizobacteria* were significantly higher than other treatments. *Rhizobium* sp.- mycorrhiza or *Rhizobium* sp.-*Rhizobacteria* inoculation were better than third inoculation. It is known that grain weight and yield is supported by length root, root fresh and dry weight as a result of inoculation. The longer and the more complex roots, the greater σ water and nutrient absorption (Lakitan, 2007). The availability of nutrients, combined with dif-

ference number of leaves and leaf area of each cultivar required for photosynthesis, will affect the filling of seed which leads to the increase of grain weight and yield. It should also be noted that each cultivar has different adaptation to the environment (Yudhy and Inorlah, 2009), which means that Petek cultivar is more adaptive to coastal sandy soil as it is regarded as quite drought resistant cultivar (Sri *et al.*, 2015).

CONCLUSION

The present study demonstrated that inoculation of Petek cultivar using *Rhizobium* sp.-mycorrhiza resulted in significantly increased root growth, leaf area, and yield (5.97 ton/ha) and diameter of nodule. Therefore, it is concluded that association between *Rhizobium* sp.-mycorrhiza and *Rhizobium* sp.-Merapi-indigenous *Rhizobacteria* with Petek is suitable for soybean cultivation on coastal sandy soil.

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