

The Growth Response of Anjasmoro Soybean (glycine max(1.)Merr.) On combination Inoculated Ultisol Soil Of Azotobacter SP., Mycorrhizal, and Organic Fertilizer

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Abstract- Background: Indonesia has not been able to meet demand of soybean from the national production, because fertile land for soybean cultivation has decreased. Land for soybean cultivation in Indonesia is dominated by ultisol which poor of nutrition. This study aims to investigate the growth response of Anjasmoro varieties of soybean on combination inoculated ultisol soil of Azotobacter sp., Mycorrhizal, and organic fertilizers.

Method: The study was conducted with a factorial randomized block design with two factors. The first factor is the combination of Azotobacter sp. and Mycorrhiza with 3 treatments, namely : the combination of Azotobacter sp. with a density of 10^3 CFU ml⁻¹ and Mycorrhiza 30 grams (K1); the combination of Azotobacter sp. with a density of 10^4 CFU ml⁻¹ and Mycorrhiza 25 grams (K2); the combination of Azotobacter sp. with a density of 10^5 CFU ml⁻¹ and Mycorrhiza 35 grams (K3). The second factor is Organic Fertilizer with 4 treatments, namely : without organic fertilizer (P0); organic fertilizer 2 tons ha⁻¹(P1); organic fertilizer 4 tons ha⁻¹(P3) organic fertilizer 6 tons ha⁻¹(P4). These experiments were 12 combinations of treatments, 6 replications. Parameter as measured in this research was high concentration plant (cm, number of leaves (sheets), number of branches, biomass wet, biomass dry, number of productive segments, measured at the age of plants 30,45,and 60 days after planting. Dry biomass done with draining to an oven with the temperature 80⁰C for 96 hours. Data analysis applied analysis of average and anova with significance 0.05, by Duncan test.

Result: Growth effect (plant height, number of leaves, number of branches, wet biomass, dry biomass, and number of productive segments) from treatment of organic fertilizers factors had significant difference, while treatment of factors combination of Azotobacter sp. and Mycorrhizal had not significant difference. Combination of Azotobacter and mycorrhizal shows average growth response was different, but statistical analysis did not show the significant difference. The higher doses of organic fertilizers, the higher density of Azotobacter (CFU) and dosage of mycorrhizal (gram), thus response growth varieties of Anjasmoro soybean on the ultisol soil was higher. The best response growth was giving organic fertilizers 6 ton⁻¹ and inoculation of Azotobacter 10^5 CFU ml⁻¹ and Mycorrhizal 35 grams propagul (P3K3). Treatment by the lowest response growth was without organic fertilizers and of Azotobacter application 10^4 CFU ml⁻¹ and mycorrhizal 25 grams propagul (P0K2).

Conclusion: The growth Response (plant height, number of leaves, number of branches, wet biomass, dry biomass and the number of productive segments) The varieties of Anjasmoro soybean plant increased by application of organic fertilizers and inoculation combination of Azotobacter sp. and mycorrhizal.

Key words: Soybean Growth Response, Azotobacter sp. and Mycorrhizal, organic fertilizer.

I. INTRODUCTION

SSoybean production in Indonesia in 2013 is about 867.16 thousand tons of dry beans, national demand reached 2.95 million tons, and 2.128 million tons imported [1]. Indonesia has not been able to meet the demand of soybeans from the national production, because fertile land for soybean cultivation has decreased. Soybean growth in ultisol can be increased through the physical, chemical and biological improvement of soil. Mycorrhizal is one of boletus which living in the ground, associated with plants and symbiotic [2], which will raise the in absorbing of nutrient element, and water [3], as element phosphate [4] and its sour in acid soil and limited high of pH, so that P become major parapet element in plants productivity, including soybean. Status of soil nutrient on the ultisol soil is very low, the womb organic matter is low, soil unfriendliness is high,, poor of macro nutrient especially N, P, K [5].

The increased of production and growth of very hanging from chemical fertilizers, but the use of chemical fertilizer for a long time can damage the environment [6]. The use of chemical fertilizer can increase pollution of the environment, water pollution , and can be lowered human health [7].

Solutions to lower the risk of accumulation of chemical fertilizers in the environment is combining chemical fertilizers with biofertilizers [8]. The beneficial microorganisms used to able to improve the plant growth but also maintain the environmental health and productivity of the soil. Recent studies confirm that a number of species of bacteria mainly associated with the rhizosphere of plants are beneficial to the growth, yield

and crop quality [6]. These are called as Plant Growth Promoting rhizobacteria (PGPR). The PGPR involving free living (asymbiotic) growth promoting rhizobacteria, symbiotic rhizobia and phosphate solubilizers [9] have been used for enhancing the production of different crops (Mohite, 2013; Viruel *et al.*, 2014) including legumes [10]. The PGPR include of the genus *Azospirillum*, *Azotobacter* and others [11]. Previous studies showed that PGPR stimulates growth and increases yield of different fruits such as apple, cherry and peach [12][13]. Genus *Azotobacter* is a free-living bacteria that fix atmospheric nitrogen, can grow well in the media without nitrogen. These bacteria use atmospheric nitrogen for the synthesis of the cellular proteins, and cellular protein is mineralized after the death of the cell, thus contributing to the availability of nitrogen for wild plants and crops [14]. *Azotobacter* synthesizes and secretes considerable amounts of biologically active substances like B vitamins, nicotinic acid, pantothenic acid, biotin, heteroxins, gibberelins etc. which enhance root growth of plants [15].

Mycorrhiza is the association of fungi with roots, which are involved in the absorption of nutrients from soil, these are found between hyphal fungi and the underground organs of the roots of seed plants and the sporophytes of most pteridophytes. Mycorrhiza are known to assist the plants to absorb mineral nutrients from the soil, particularly low available elements like phosphorus, and increasing of plant growth on conditions of low available phosphorus [16]. Seed inoculation with *Azotobacter chroococcum* also significantly increased the growth parameters of mungbean [17]. Using of *Mycorrhiza* symbiotic is an effective alternative method instead of applying chemical Phosphorus fertilizer because *Mycorrhiza* can increase the ability of plants to absorb Phosphorus even in unavailable Phosphorus resources [18].

Based on the reference for research of *Azotobacter* and Mycorrhizal application for soybean especially to increase Anjasmoro varieties growth has rarely been implemented. Hence investigation about soybean growth effect in *Azotobacter* sp. , Mycorrhizal and Compost on the ultisol soil to the development of ultisol soil use in indonesia for appropriate soybean varieties cultivation.

II. MATERIAL DAN METHOD

The material used in this research was the soybean seed varieties anjasmoro, *Azotobacter* sp. Mycorrhizal, and organic fertilizer. *Azotobacter* sp. was isolated from ultisol soil samples [19]. Indigenous Mycorrhizal collected by halim [20]. The research applied random design group factorials with two factors. First factor was the combination of *Azotobacter* sp. and Mycorrhizal (K) with 3 treatment, namely: a combination *Azotobacter* sp. with the density 10^3 CFU ml⁻¹ and 30 grams propagul Mycorrhizal (K1), a combination of *Azotobacter* sp. with the density 10^4 CFU ml⁻¹ and 25 grams propagul Mycorrhizal (K2), and a combination of *Azotobacter* sp. with the density 10^5 CFU ml⁻¹ and 35 grams propagul Mycorrhizal (K3). Second Factors was organic fertilizer (P) with 4 treatment, namely: without organic fertilizer (P0); the provision of organic fertilizer 0.8 kg plot⁻¹ equivalent to two tons ha⁻¹ (P1); the provision of organic fertilizer 1.6 kg plot⁻¹ equivalent to four tons ha⁻¹ (P2); the provision of organic fertilizer 2.4 kg plot⁻¹ equal to six tons ha⁻¹ (P3). There were 12 combination treatment six repeated, so the total were 72 experiment plot with each size of 2 x 2 m. Planting land was in ultisol soil in the Kendari city of Southeast Sulawesi. Parameter in this research was plant height (cm), number of leaves (sheets), number of branches, wet biomass, dry biomass, number of productive segments, measured at the age of plants 30, 45, and 60 days after planting. Dry biomass was conducted with draining to an oven with the temperature 80°C for 96 hours. Data analysis with using average and ANAVA analysis with significance 0.05 and Duncan test.

III. RESULT AND DISCUSSION

A. Result

Organic fertilizers factors (compost) has significant difference, while combination *azotobacter* and mycorrhizal has not significant difference on the plant height parameter, number of leaves, number of branches, wet biomass, dry biomass of Anjasmoro soybean varieties at the age of 30 days (table 1), age was 45 days (tabel2), and the age of 60 days (tabel3) and the number of productive segments (table 3). Treatment of organic fertilizers shows different growth response. Treatment combination of *Azotobacter* and Mycorrhizal shows growth response on average different, but statistical analysis do not show significant difference. The higher doses of organic fertilizers and density of *azotobacter* (CFU), the higher growth response of Anjasmoro soybean varieties on the ultisol soil. Treatment of the best growth response was giving of organic fertilizers 6 ton⁻¹ and application of *Azotobacter* 10^5 CFU ml⁻¹ and Mycorrhizal 35 grams propagul (P3K3). Treatment by lowest growth response was without organic fertilizers and application of *Azotobacter* 10^4 CFU ml⁻¹ and Mycorrhizal 25 grams propagul (P0K2).

Table 1 .The results of the variance of plant height (PH), number of leaves (NL), the number of branches (NB), wet biomass (WB), dry biomass (DB) the Anjasmoro soybean plant varieties age 30 days

Treatment	PH (cm.)	NL	NB	WB (gr.)	DB (gr.)
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P0	36.08a	7.44a	2.00a	12.28a	3.58a
P1	38.14a	9.44a	3.11b	18.72a	5.12ab
P2	39.95a	12.66b	4.00c	27.40b	6.73b
P3	53.55b	15.44c	4.55c	44.12c	11.44c
P (Sig.)	0.000*	0.000*	0.000*	0.000*	0.000*
K1	38.20a	10.83a	3.16a	24.04a	6.14a
K2	41.82ab	11.08a	3.41a	24.42a	6.43a
K3	45.78b	11.83a	3.66a	28.42a	7.59a
K (Sig.)	0.007*	0.549ns	0.341ns	0.115ns	0.142ns
PxK (Sig.)	0.804ns	0.621ns	0.509ns	0.353ns	0.768ns
P3xK3	59.83	17.33	4.33	46.86	12.86
P0xK2	36.50	7.66	2.00	10.83	3.13

Table 2. The results of the variance of plant height (PH), number of leaves (NL), the number of branches (NB), wet biomass (WB), dry biomass (DB) the Anjasmoro soybean plant varieties age 45 days

Treatment	PH (cm.)	NL	NB	WB (gr.)	DB (gr.)
P0	49,33a	25.22a	4.88a	49,96a	17.45a
P1	52.44a	34.22b	5.44a	79,37a	27.52b
P2	63.55b	36.77b	6.22ab	108,37b	33.07b
P3	77.66c	42.55c	6.88b	143.37c	42.01c
P (Sig.)	0.000*	0.000*	0.000*	0.000*	0.000*
K1	61.01a	34.66ab	5.66a	88.12a	29.75a
K2	61.25a	31.00a	5.75a	87.37a	26.73a
K3	59.08a	38,41b	6.16a	111.7b	33.55a
K (Sig.)	0.648ns	0.012*	0.626ns	0.032*	0.078ns
PxK (sig.)	0.586ns	0.606ns	0.124ns	0.730ns	0.894ns
P3xK3	79.33	46,66	7.00	177.20	46.76
P0xK2	47.33	17.00	4.33	31.83	12.13

Table3. The results of the variance of plant height (PH), number of leaves (NL), the number of branches (NB), wet biomass (WB), dry biomass (DB), and number of productive segments (NPS) the Anjasmoro soybean plant varieties age 60 days

Treatment	PH (cm.)	NL	NB	WB (gr.)	DB (gr.)	NPS
P0	60,33a	29.77a	5.66a	68.07a	25.97a	20.25a
P1	64.00a	36.11ab	6.11a	100.47ab	37.05ab	27.00b
P2	71.55b	39.88b	6.66a	143.15bc	46.65b	30.57bc
P3	82.72b	44.00b	7.88b	166.91c	50.28b	31.59c
P (Sig.)	0.000*	0.000*	0.000*	0.000*	0.000*	0.000*
K1	63.37a	35.66a	6.58a	113.49a	38.63a	26.65a
K2	69.66ab	37.83a	6.58a	129.65a	43.13a	27.24a
K3	73.08b	38.83a	6.58a	115.81a	38.21a	28.18a
K (Sig.)	0.040*	0.678ns	1.000ns	0.628ns	0.655ns	0.115ns
PxK (Sig.)	0.192ns	0,611ns	0.078ns	0.892ns	0.953ns	0.764ns
P3xK3	84.66	52.00	7.66	177.80	50.66	43.44
P0xK2	62.00	30.66	6.00	75.63	28.13	19.91

Means in a column followed by the same letter are not significantly different at $P \leq 0.05$.

Figures that followed by the same letter at the same column was not significant different at the duncan test with standard 5%; ns = non significant; the (*) = significant at $P \leq 0.05$.

B. Discussion

The analysis variance results of Anjasmoro soybean varieties growth which given organic fertilizers treatment (P) and combinations of *Azotobacter* sp. + Mycorrhizal (Table 1, 2, 3). Compost Treatment with the different situation (compost 0 ton ha⁻¹, 2 tons ha⁻¹, 4 tons ha⁻¹ and 6 tons ha⁻¹) on the ultisol soil had have real impact on plant height, number of leaves, number of branches, wet biomass, dry biomass of Anjasmoro soybean varieties plant on 30, 45, and 60 days after planting and had have real impact on the number of productive segments of Anjasmoro soybean varieties plant at 60 days after planting.

The combination treatment of *Azotobacter* + mycorrhizal contribute the effect of growth increases with the increasing of *Azotobacter* and mycorrhizal density, but statistically do not show significant difference to plant height, number of leaves, number of branches, wet biomass, dry biomass. The number of productive segments of Anjasmoro soybean varieties plant on 30, 45 and 60 days after planting. Plant height on 30 and 60 days after planting, number of leaves on 45 days after planting, wet biomass on 45 days after planting, combination treatment of *Azotobacter* + mycorrhizal have an significant difference. The difference of low growth because contributions of nitrogen from *azotobacter* was low, this is in accordance with opinion [21], that contributions of free-living rizobakterium like *Azotobacter* to low nitrogen soil, so the difference of density population of *Azotobacter* sp. and mycorrhizal is not significantly difference.

The results of the study in line with the results of a study [6] on the use of *Azotobacter* sp., shows better results on growth parameters: height of plant, content of chlorophyll, fresh and dry root weight of strawberry (*Fragaria vesca*) in hydroponic system. nocolation of *Azotobacter* sp. Can increase the growth and the results of tomato (*Lycopersicon esculentum* Mill.): plant height, number of leaves, the number of fruit, heavy wetness of fruit, the womb of chlorophyll and protein [22]. The results of study, shows the effects of *Azotobacter* and *Mycorrhiza* is significant on percentage of root colonization, percentage seed of protein content, percentage seed nitrogen content, percentage seed phosphorus content, and percentage potassium content [23]. Interaction of *Azotobacter* × *Mycorrhiza* had significant effects on percentage seed phosphorus content, and percentage potassium content. The results indicates an effective function of biofertilizers in improving plant growth and increasing products quality of wheat cultivars under dry land conditions. The application of *Azotobacter* of and *Mycorrhiza* has a positive effect on absorption nutrient elements by plant. In Cotton, inoculation of *A. chroococcum* and AM fungi has a synergistic effect on boll plant height, number of flower, boll weight and yield [24]

Combination of *Azotobacter* sp. + mycorrhizal by the provision of compost, the interaction do not occur in influencing the growth of anjasmoro soybean varieties, the result in accordance with research Hasanuddin (2003) reported that inoculation of mycorrhizal and *azotobacter* and provision of organic matter do not indicate the significant difference to absorption N and P as well as the results of pipilan corn, but it is independently significant difference [25]. The results of the study of Dachlan et al. (2012), that type of compost, treatment concentration *Azotobacter* and interaction the two are not significantly difference on the number of rice saplings per a thicket, the number of productive segments per a thicket, the number of vacuum grain per panicles, and heavy grain 1000 grains [26].

Table 1, 2, and 3 above show the highest growth happened to treatment combination *Azotobacter* 10⁵ + mycorrhizal 35g and compost 6 tons ha⁻¹ because the *Azotobacter* and mycorrhizal activities will continue to increase with increasing the addition of compost in the soil, because compost is a source of nutrition and energy for microbes and provides a source of nutrient to plants. The lowest Growth in treatment combination of *Azotobacter* 10⁴ + mycorrhizal 25g and without compost are on the parameter plant height, number of leaves, number of branches, and the number of productive segments because the increasing number of *Azotobacter* and *Mycorrhizal* can increase competition among microbes with the limited organic matter. Microbes competition increases with the limited supply of organic matter cause stunting .

Treatment of *Azotobacter* + mycorrhizal and compost statistically do not indicate interaction two factors, but combination *Azotobacter* 10⁵ CFU ml⁻¹ + *Mycorrhizal* 35g and compost 6 tons ha⁻¹ gave the highest average on plant height, number of leaves, number of branches, wet biomass, dry biomass, and the number of productive segments of anjasmoro soybean varieties plant on 30, 45, and 60 days after planting. Activity of *Azotobacter* and *Mycorrhizal* and soybean growth on the ultisol soil can distimuli by the presence of compost the provision organic matter can increase pH of soil, nutrient P, soil KTK, nutrient cycle, improve the ability of water reserve, reduce soil erosion, sent down the levels of Al, Fe, and M [27][28]. Yaseen, Burni and Hussain (2011), reported that inoculation of *Mycorrhizal* can increase plant height, the amount of flower, and increase the absorption of N, P, K, Ca, and Mg in *Vigna unguiculata* [29].

Hasanuddin (2003) reported that independently inoculation of *Mycorrhizal* and *Azotobacter* and provision of organic matter had have significant impact on absorption N, P, and the results of pipilan corn [25]. Inoculation of *Azotobacter* can increase the germination of seeds, growth, and improve the result up to 30% on wheat [30]. *Azotobacter* can menstimuli for soybean growth, because *Azotobacter* can provide nitrogen, vitamin, amino acid, auxin and giberelin that can be stimulated the growth of plants and *Mycorrhizal* can be stimulated soybean growth, because mycorrhizal can increase the absorption of P by soybean. The results of the

study on basil (*Ocimum Basilicum L.*) shows that the combination use of mycorrhiza, azotobacter and vermicompost significantly improve shoot fresh weight, shoot dry weight and essential oil in compared with control. The increase relationship between mycorrhiza, azotobacter and vermicompost show that they are supplementary for each other [31].

Dry weight of soybean plant shows the growth of plants and the amount of hara element employed per unit weight biomasa produced. The dry biomass shows the activity of assimilation (photosynthesis). An increase of dry biomass in line with an increase in compost measurement that given and the provision of combination of *Azotobacter* and Mycorrhizal. The increase was caused by increasing in element P and N compost and the availability of N and P that can be absorbed plants because the results of the activity of *Azotobacter* sp and mycorrhizal. Element P had a role in spur the photosynthesis, while element N spur the growth of plants. In plants the P is essential phospholipid membrane components, ATP, PEP, and NADPH that important in the use and the storage of energy [32]. Soybean which deficiencies of phosphorus is inefficient in absorption of radiation so unfavorable to plant [33]. An increase in the availability of P through the compost and mycorrhizal activity can increase growth leaves, so this can increase the rate of photosynthesis plants.

Result study on *Carthamus tinctorius* with inokulasi *G. intraradices* and *Azotobacter chroococcum*, Increased root dry weight 8.47% and grain yield 5.20% over control [34]. Inoculations with plant growth promoting rhizobacteria and Arbuscular Mycorrhizal Fungus consistently increased the growth and yield, N and P concentrations, and quality of wheat grains [35]. Inoculation *Azotobacter chroococcum* + *Glomus fasciculatum* can increase biomass roots, biomass shoot, and biomass total wheat exceeds biomass by inoculation single azotobacter chroococcum or *glomus adenostoma* and without inoculation [35].

Study on wheat (*Triticum aestivum L.*) indicates that increasing plant height, grains number per ear and biological yield have been affected significantly by inoculation with *Azotobacter*, because this biofertilizer can enhance absorb of nitrogen by plant. According to calculated, inoculation wheat grains in planting date with *Azotobacter* to cause increase yield about 6 percentage and also reduce using of about 50 percentage chemical nitrogen [36].

IV CONCLUSION

The provision of organic fertilizers, *Azotobacter* sp. and Mycorrhizal on ultisol soil can increase plant height, number of leaves, number of branches, wet biomass, dry biomass, and the number of productive segments of Anjasmoro soybean varieties. Biological fertilizer can improve the availability of nutrition element on ultisol soil to favor the growth of soybean .

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