
THE EFFECT OF SEED COATING WITH *Trichoderma* sp. AND APPLICATION OF BOKASHI FERTILIZER TO THE QUALITY OF SOYBEAN (*Glycine max.* L) SEED

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Abstract. The decrease of soybean productivity was caused by low quality of seed. To improve the quality of seed, soybean seed were coated with *Trichoderma* sp. and adding bokashi organic fertilizer. This research aimed at finding the best dose combination of *Trichoderma* sp. and bokashi fertilizer to improve the quality of soybeans. The research was conducted in the experiment field and Laboratory of Seed Technology at Padjadjaran University in April - August 2017. The experimental design in this research was Randomized Block Design (RBD). The treatments were the combination of four dose values of coating the seed by *Trichoderma* sp. (0g/100 seeds, 1g/100 seeds, 2g/100 seeds, and 3g/100 seeds) and three dose of bokashi (0g/polybag, 300g/polybag, and 600g/polybag) and each treatment was replicated three times. The experiment result showed that all treatments on soybean seeds did not affect the germination percentange, vigor index, and conductivity value, but affected the seed quantity of 600g/polybag of bokashi and without seed coating with *Trichoderma* sp. on 100 seed mass and seed weight per plant.

Keywords: Bokashi, Soybean seeds, *Trichoderma* sp.

Citation

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INTRODUCTION

In Indonesia, soybean consumption increases every year along with increasing population growth and community needs. According to BPS data of 2015, Indonesian's needs for soybeans have reached 2.23 million tons of dried beans. However, these needs are not balanced with soybean production which only reaches 1.5 tons/ha. One of the ways to increase soybean production is by using quality seeds. Quality seeds are determined shown

by viability, vigor of seeds, and high yield. These quality seeds can be maintained and enhanced by seed coating and organic fertilizer application.

The treatment of seed by placing chemical on the seed coat is a way to enhance seed performance without altering the shape of the seed (Ilyas, 2012). If seed coating uses insecticides, it can protect the seeds from pests and diseases that attacks at the beginning of the growth phase, so that plant growth is not disturbed and can last until the final phase

(Cox et al, 2007). Seed coating with insecticides can be antagonistic microorganism as seed anti pathogens called biological seed treatment (Agustiansyah, et al, 2010; Ilyas, 2012). One of the microorganisms which can be used is *Trichoderma*. *Trichoderma* has an important effect on plant growth, productivity, germination, and seed vigor, and can also control soil borne diseases (Harman, 2006; Tančić et al, 2013).

Sumadi et al. (2015) who has tested and compared several types of seed coatings on detached soybean seeds concluded that seed coating with *Trichoderma* sp. with a dose of 2 g/100 seeds was better than that of *thiamektosam* and *Rhizobium* sp. Results indicated that the seed coating with *Trichoderma* sp. also produced a better number of pods, seeds, and also seed weight than other seed coatings.

The provision of seed coating has a good effect on the viability and vigor of seeds during growth but does not affect the yield and the yield components of soybean. Therefore, the treatment of soybean seed coating and application of bokashi affected both the initial vegetative phase and the reproductive phase (Sumadi et al, 2011). In the next study, Sumadi et al. (2012) concluded that the coating seed with *thiamektosam* insecticide at 2 ml kg⁻¹ accompanied by bokashi of 15 tons ha⁻¹ decreases the effects of fly pest attack, and increased the number of effective root nodules, growth, yield components, and the yield of soybean crops. For this, a combination of seed coating to support the initial growth of seeds with application bokashi to provide of nutrition to the reproductive phase is needed.

Bokashi has a role in improving plant growth and protein content so that it can produce good quality seeds (Saro, 2007). Beside being a source of nutrition, bokashi contains a lot of microorganisms that are beneficial for soil fertility because it can improve the physi-

cal, chemical and biological properties of soil so as to provide nutrients in increasing the growth and production of plant (Binardi, 2014). Bokashi is made by processing the organic matter with EM (Effective Microorganism). There are a lot of microorganisms in EM4. Here are 4 main EM4 microorganisms: (1) photosynthetic bacteria (phototropic bacteria), (2) lactic acid bacteria (*Lactobacillus* sp.), (3) yeast, and (4) *Actinomycetes* sp. (Musnamar, 2003).

Research conducted by Rahman et al. (2014) show that 300 grams of bokashi per polybag could increase growth, yield components and the quality of soybean seed with 99% germination and 9.44% vigor index. However, when combined with *P* fertilizer, it showed that the best result was at 600 grams dose of bokashi per polybag without adding *P* fertilizer. The use of biological agent of *Trichoderma* sp. as seed coating and the provision of organic fertilizer in the form of Bokashi fertilizer can support green agriculture. Green agriculture is currently a trend for consumption patterns in the community, because nowadays more and more people care about their health. In addition, the balance of ecosystems in the soil can be maintained. Based on this framework, it is expected that the combination of seed coating treatment with *Trichoderma* sp. and application of bokashi can improving the quality of seed yields, so the results of this study will provide information about the effect of the treatment and determinate the appropriate dose of the combination of coating the seed using *Trichoderma* sp. and bokashi application.

MATERIALS AND METHODS

The study was conducted with pot experiments carried out in the experimental field of

Faculty of Agriculture, Padjadjaran University, Jatinangor, West Java, with an altitude of \pm 750m above sea level. This study was conducted from April to July 2017. The seed quality testing was carried out at the Seed Technology Laboratory Department of Agriculture, Faculty of Agriculture, UNPAD. Soil analysis and microbial population on planting media and the content analysis of bokashi were carried out at the Soil Fertility Laboratory and Plant Nutrition, Faculty of Agriculture, UNPAD.

The materials used in this experiment were Anjasmoro varieties of soybean obtained from the Research Institute of Assorted Peanut and Tuber Crops (*read* UPBS-BALITKABI), Malang, East Java. Tricho-G (inoculum *Trichoderma* sp. 10^{12} CFU/g) as a coating for seeds was produced by PT. Primasid Andalan Utama Jakarta. The ingredients for making bokashi were as follow: cow dung, EM4, bran, liquefied sugar, husks, water; urea fertilizer; SP-36 fertilizer; KCl fertilizer; 500 EC Curacron insecticide; and aquades.

The experiment was arranged in a Randomized Block Design (RBD) consisting of seed coating with *Trichoderma* spp (0g/100white, 1g/100boss, and 3g/100boss) and bokashi (300g/polybag and 600g/polybag) with three replications, there were 36 experimental units. The treatment arrangements carried out in this experiment included:

- A: Seed coating with *Trichoderma* sp. 0g/100 seeds + bokashi 0g/polybag
- B: Seed coating with *Trichoderma* sp. 1g/100 seeds + bokashi 0g/polybag
- C: Seed coating with *Trichoderma* sp. 2g/100 seeds + bokashi 0g/polybag
- D: Seed coating with *Trichoderma* sp. 3g/100 seeds + bokashi 0g/polybag
- E: Seed coating with *Trichoderma* sp. 0g/100 seeds + bokashi 300g/polybag

- F: Seed coating with *Trichoderma* sp. 1g/100 seeds + bokashi 300g/polybag
- G: Coating of seeds with *Trichoderma* sp. 2g/100 seeds + bokashi 300g/polybag
- H: Coating the seeds with *Trichoderma* sp. 3g/100 seeds + bokashi 300g/polybag
- I: Coating of seeds with *Trichoderma* sp. 0g/100 seeds + bokashi 600g/polybag
- A: Seed coating with *Trichoderma* sp. 1g/100 seeds + bokashi 600g/polybag
- K: Seed coating with *Trichoderma* sp. 2g/100 seeds + bokashi 600g/polybag
- L: Seed coating with *Trichoderma* sp. 3g/100 seeds + bokashi 600g/polybag

The experiment was initiated by making bokashi made from cow dung. The application of cooked bokashi was done a week before planting in accordance with the dosage. Before planting, seed coating was done by mixing *Trichoderma* sp. with seeds, adding a little water in a plastic bag, and letting it stand for 30 minutes. Planting (three seeds per polybag) was done by applying N, P, and K. Fertilizers. The maintenance was done by thinning the plants carried out at 2 weeks after planting, watering, controlling pest and disease. The seed testing was done before planting and a month after harvesting. Harvesting was done when harvesting maturity, physiologically which was characterized by 95% of pods turning brownish in colour, dry stems, and around 5-10% senescence of leaves form the plants.

The experimental parameters were divided into secondary observation and main observation. Secondary observations included initial soil analysis, bokashi analysis, initial weight of 100 seeds, initial germination, rainfall data, pests and diseases, and harvest age. The main observations included the seed weight per plant, the weight of 100 seeds, seed

mination, vigor index, and electrical conductivity.

The data analysis of seed germination, vigor index, and electrical conductivity was done using the F test at 5% level of significance in which the significant effect was tested by Duncan's Multiple Range Test at 5%. Whereas in the observation of the weight of 100 seeds and the weight of seed per plant Scott-Knott Test we used at the level of 5%. The data analysis was done using SPSS and Microsoft Excel programs.

RESULTS AND DISCUSSION

The results of the initial soil analysis showed that the physical properties of the soil used in the experiment (dusty clay) was in accordance with the requirements of growing soybean, while the chemical properties in the soil fertility was lacking some nutrients, low content of N (0.2%), so it requires additional fertilization. *Azotobacterium* bacteria ($1.9,00 \times 10^7$ CFU/g) and *Trichoderma* sp. (2.00×10^3 CFU/g) were found in the field before the experiment. The content analysis of bokashi had N (1.18%), P (1.18%), and K (0.57%) were in accordance with the maturity of bokashi according to SNI 19-7030-2004, but the content of C-Organic and C/N ratios exceeded the standard of maturity so that the provision of bokashi was done a week before planting. In observing the temperature, humidity, and rainfall it was in accordance with the requirements of growing soybeans so that it did not have an effect. Before planting, seeds we tested 100 seeds weight and germination percentage with 15.8g and 84% as the initial data.

In general, the plants in the experiment did not get significant effects due to abiotic or biotic interferences. The main insect pests observed include grasshoppers (*Valanga nigricornis*), armyworm (*Spodoptera litura*), and

long-haired caterpillars (*Chrysodeixis chalcites Esper*) which damaged the leaves and ladybug pests (*Riptortus linearis* Fabricius) which sucked the pods. They were observed during the reproductive phase of the plant. The intensity of the attack was low ($\leq 5\%$). Pests attack could be controlled by applied pesticides regularly. Soybean in the experiment were harvested when they reach physiological maturity which was characterized by 95% yellow pod at 85 days after planted.

The weight of 100 seeds and the weight of seed per plant

The weight of 100 seeds and the weight of seed per plant seeds were carried out to determine the size and number of seeds produced. Seed size was one of the things that determined the quality of seeds. The results of statistical analysis (Table 1) show significant difference on weight of 100 seed and the seed weight per plant.

Observation on the weight of 100 seeds, the treatment combination generally had greater results than the weight of 100 seeds in the description of Anjasmoro varieties of soybean plants (14.8-15.3g/100 seeds). The weight of 100 seeds in this experiment rang from 14.53 to 17.23 g/100 seeds which meant that the size of the seed was large (>14 g/100 seeds). The large seeds contain more food reserves and could affect the seed germination. The best effect on the weight of 100 seeds was indicated by treatment H (3g *Trichoderma* + 300g Bokashi), I (0g *Trichoderma* + 600g Bokashi), J (1g *Trichoderma* + 600g Bokashi), K (2g *Trichoderma* + 600g Bokashi), and L (3g *Trichoderma* + 600g Bokashi). Whereas in the observation of seed weight the treatment crop had the best effect, namely in treatment I (0gram *Trichoderma* + 600gram plant⁻¹ Bokashi), J (1 gram *Trichoderma* + 600gram plant⁻¹ Bokashi) and L (3gram *Trichoderma* + 600gram plant⁻¹

Bokashi). However, when viewed from the economic value, treatment I (0gram *Trichoderma* + 600gram plant⁻¹) bokashi was more effective to be used in the field.

Secondly, observation on the weight of 100 seeds and the seed weight per plant, it was found that the dose given by bokashi had an effect. The higher the dose given, the better the results realised. However, on seed coating with *Trichoderma* sp. it had no effect. This was observed in the analysis of the initial experimental soil which contained *Trichoderma endogenus* with a population density of 2×10^3 CFU/g.

Trichoderma could increase the weight of plant seeds because *Trichoderma* had a role in increasing soil fertility (indirect effects) which had a good effect on the weight of plant seeds (Chamzurni et al, 2011). In addition, *Trichoderma* produced auxin hormones in the form of IAA (*Indole Asetic Acid*) which affected the root growth and can improve the plant productivity through hormone stimulation (Lestari et al, 2007). However, if auxin

hormones were produced with high concentrations, it would inhibit the cell lengthening, because auxin hormones with high concentrations would stimulate the formation of ethylene hormones with high concentrations so that it could inhibit the process of growth and elongation of root cells (Chamzurni et al, 2011).

According to Birnadi (2014) Bokashi contain *Micrococcus* and *Azopirillum* which are able to increase P and N nutrients in the soil and increased the root growth which resulted in the increase of P and N absorption. In addition, the presence of EM4 as a bokashi element was very useful since EM4 in the soil could synergistically improve soil fertility both physically, chemically and biologically so that it can increase the productivity of soil and plants (Wididana and Higa, 1993 in Rahman et al, 2012). This resulted in an increased weight of seeds produced according to Syafaat et al. (2014) and Rahman et al. (2014) bokashi cow dung could increase the percentage of the weight of 100 seeds and the seed weight per plant.

Table 1. The effect of combining the seed coating with *Trichoderma* sp. and applying bokashi to the weight of 100 seeds and the seed weight per plant.

No	Treatment	Weight of 100 Seeds (g)	Seed Weight per Plant (g)
1	A (0g Tricho + 0g Bokashi)	15.13b	14.43c
2	B (1g Tricho + 0g Bokashi)	15.93b	14.18c
3	C (2g Tricho + 0g Bokashi)	14.53b	14.72c
4	D (3g Tricho + 0g Bokashi)	15.00b	14.32c
5	E (0g Tricho + 300g Bokashi)	16.33b	16.12c
6	F (1g Tricho + 300g Bokashi)	15.83b	17.43b
7	G (2g Tricho + 300g Bokashi)	15.60b	16.27c
8	H (3g Tricho + 300g Bokashi)	16.17a	16.65c
9	I (0g Tricho + 600g Bokashi)	16.97a	20.86a
10	J (1g Tricho + 600g Bokashi)	17.23a	20.43a
11	K (2g Tricho + 600g Bokashi)	16.83a	17.93b
12	L (3g Tricho + 600g Bokashi)	16.53a	19.50a
	cv	5.27	9.49

Description: The average value followed by significantly different letters according to the *Scott Knott* Test at the 5% level.

Germination Percentage, Vigor Index, and Electrical Conductivity

The observation on germination, vigor index, and electrical conductivity was carried out to determine the quality of the seeds produced. The seed quality can be determined from the viability and vigor of seeds. The seed viability shows the life force of metabolically active seeds that are needed in the germination process while the seed vigor shows the speed and simultaneity of the germinating seeds. According to Ilyas (2012) the germination percentage is one of the benchmarks to determine the seed viability and the seed vigor which can be determined by vigor index and electrical conductivity. The observation of germination percentage, vigor index, and electrical conductivity showed that they were not significantly different but had good results (Table 2). The results on germination showed 87.33-96.67% exceeding the minimum standard of certified seed of 80%, the vigor index (8.7-9.46) was also said to be good because it approached the maximum vigor index value of 10, and the observation of electrical conductivity approached a value of 0 (0.12-0.17) which indicated that there was no leakage of seeds.

The fact that there were no difference caused by all treatments that were met to the standards of quality seeds. The seeds tested were new so that the seeds were able to germinate well and the food reserves for the germination process were sufficient (Sumadi et al, 2005). This was also supported by the large amount of seed produced from this experiment so that the food reserve was high so and could increase the seed vigor. The provision of bokashi fertilizer and seed coating with *Trichoderma* sp. also had an effect, because bokashi fertilizer had elements of nitrogen, phosphate, and potassium needed by soybean in a balanced and sufficient quantity so that the production and quality of seeds increase. Whereas the coating of *Trichoderma* sp., which could spur the growth of sprouts, was related to the productive formation of lateral roots, had an important role as a producer of auxin, and stimulated the growth (Hexon et al, 2009 in Nurahmi et al, 2012). In addition, it was known that the initial soil content had a very high P and K content with the provision of Bokashi which increased the soil nutrients.

Table 2. The effect of combining the seed coating with *Trichoderma* sp. and applying bokashi to germination Percentage (%), vigor index, and electrical conductivity ($\mu\text{S/g}$).

No	Treatment	Germination Power (%)	Vigor Index	Electrical Conductivity ($\mu\text{S/g}$)
1	A (0g Tricho + 0g Bokashi)	89.33a	8.71a	0.17a
2	B (1g Tricho + 0g Bokashi)	92.00a	8.96a	0.13a
3	C (2g Tricho + 0g Bokashi)	95.33a	9.16a	0.13a
4	D (3g Tricho + 0g Bokashi)	93.33a	9.08a	0.17a
5	E (0g Tricho + 300g Bokashi)	96.00a	9.30a	0.15a
6	F (1g Tricho + 300g Bokashi)	96.67a	9.37a	0.12a
7	G (2g Tricho + 300g Bokashi)	96.67a	9.46a	0.14a
8	H (3g Tricho + 300g Bokashi)	95.33a	9.36a	0.13a
9	I (0g Tricho + 600g Bokashi)	94.00a	9.12a	0.13a
10	J (1g Tricho + 600g Bokashi)	89.33a	8.70a	0.16a
11	K (2g Tricho + 600g Bokashi)	87.33a	8.47a	0.17a
12	L (3g Tricho + 600g Bokashi)	94.67a	9.25a	0.15a
	<i>cv</i>	5.59	5.90	21.68

Description: the average values followed by letters that are not significantly different according to *Duncan's Multiple Range Test* at the 5% level.

In conclusion the combination of seed coating using *Trichoderma* sp. with bokashi only had a significant effect on the weight of 100 seeds and seed weight per plant, while the germination, vigor index, and electrical conductivity did not give a real effect but gave good results. The combination of without coating the seed and 600g per polybag of bokashi provided the most effective and efficient results on the weight of 100 seeds and the seed weight per plant.

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