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Compatibility of biofresh and agrisoy biological agents in various types of organic materials against increased resistance to mosaic virus and soy productivity in ultisol land

A Khaeruni¹, Sulqifly¹, T Wijayanto², G A K Sutariati², T Hemon³, V N Satrah¹, Asniah¹ and H Hamdayanty²

¹Department of Plant Protection, Faculty of Agriculture, Halu Oleo University Jalan HEA Mokodompit, Kendari

²Department of Agrotechnology, Faculty of Agriculture, Halu Oleo University Jalan HEA Mokodompit, Kendari

³Department of Soil Science, Faculty of Agriculture, Halu Oleo University Jalan HEA Mokodompit, Kendari

⁴Hasanuddin University

Email: andikhaeruni.uho@gmail.com

Abstract. The subject of the research was aimed to assess the compatibility of organic matter and bio-fertilizers type in improving plant resistance to disease mosaic virus (SMV) and the production of soybean on Ultisol. This study was conducted by two phases; the first phase was compatibility testing of Bokashi types and biological fertilizers to improve plant resistance to disease mosaic virus (*Soybean mosaic virus*) and Production of Soybean (*Glycine max* L. Merrill), organized by Split plot Design in a randomized block design consisting of 16 treatment repeated three replications. Therefore, there were 48 experimental units. Data were analyzed using analysis of variance followed by Duncan's Multiple Range Test if the treatment significantly. The results showed that both of the biological agents Biofresh and Agrisoy that applied with soybean litter compost is able to improve the resistance to disease mosaic virus (*Soybean mosaic virus*) and productivity of soybean in Ultisol.

1. Introduction

Indonesia is a country with abundant natural resources, mainly from agricultural commodities; one of them is soybeans. According to statistical data from the Ministry of Agriculture, soybean production in Indonesia reached 954,000 in 2014 [1]. Southeast Sulawesi is one of the potential areas for developing soybean plants because it has quite extensive dry land. This land potential has not been appropriately utilized due to the condition of the land, which is dominated by Ultisol land. Ultisol soil has infertile characteristics. Average pH <4.50, high Al saturation, poor macronutrient content, and low organic matter content. As a result, the growth of plant roots is inhibited because the translucency of roots into the soil is reduced [2].

In addition to the constraints of infertile land, soybean disease also becomes an obstacle in increasing crop productivity. One such disease is a mosaic virus, which can cause yield loss in some susceptible plants [3,4], which is caused by the Soybean mosaic virus (SMV) [5]. SMV disease can result in a 50-90% reduction in soybean yields [6]. Furthermore, Li et al. [7] reported that SMV infection could reduce production by 35% to 50% under natural conditions in the field.



In the cultivation of soybean needed the right technology to overcome the problems in Ultisol soil and plant diseases. One appropriate alternative technology that can be applied is the use of biological agents and organic materials (bokashi). At this time in the Faculty of Agriculture, University of Halu Oleo has developed a biological agent-based on three local rhizobacterial isolates that are antagonistic against various soil-borne diseases [8,9] and increase the yield and resistance of soybean plants to Soybean Mosaic Virus [10]. Meanwhile, Malang Research Institute for Beans and Tubers also developed Agrisoy biological agents containing effective and tolerant *N Bradyrhizobium japonicum*-inhibiting bacteria in acid to pH four so as to increase soybean production [11]. This study aims to examine the compatibility of Biofresh and Agrisoy biological agents in various types of organic material to increase plant resistance to Soybean mosaic virus and soybean production in Ultisol soil.

2. Material and methods

2.1. Material

Anjasmoro soybean seeds and Agrisoy biological agents were obtained from the Malang Peanut and Tuber Various Research Institute, Biofresh biological agents were obtained from the FP-UHO Plant Protection Laboratory, cow manure, soybean litter, and rice straw.

2.2. Experimental design

This study uses a Divided Plot Design in a Randomized Block Design (RBD). The main plot is the type of bokashi consisting of 4 levels, namely without bokashi (B_0), soybean litter bokashi (B_1), rice straw bokashi (B_2), soybean litter bokashi + rice straw (B_3). Plots are types of biological agents consisting of 4 levels, namely, without biological agents (A_0), biological agents Biofresh (A_1), biological agents Agrisoy (A_2), biological agents Biofresh + Agrisoy biological agents (A_3). In total, 16 treatment combinations were obtained. Each treatment combination was repeated three times so that there were 48 experimental units.

2.3. Land preparation

The land is processed until loose with a tractor, then made beds measuring 2 m x 3.5 m, 30 cm high as many as 48 beds. The distance between the beds in groups is 30 cm, while the distance between groups is 50 cm. Two weeks before planting, the dolomite (CaCO_3) lime was evenly distributed at a dose of 1.3 tons ha^{-1} .

2.4. Bokashi application

Bokashi application is made two weeks before planting by sowing bokashi evenly over the surface of the plot in accordance with the respective treatment plot. The dosage of the bokashi application is 10 tons. ha^{-1} . Then the map is processed so that Bokashi is immersed in the ground.

2.5. Application of Biological Agents

Biofresh biological agent application is carried out twice, each with a dose of 10 grams per clump. The first application coincides with the time of planting as a planting hole cover, and the second application is carried out at the age of the plant four weeks after planting by sprinkling near the plant roots. Application of Agrisoy biological agents is carried out on soybean seeds by means of soybean seeds moistened with sufficient water, then Agrisoy biological agents are mixed with soybean seeds at a dose of 10 grams Agrisoy / 2 kg of seeds and stirred evenly. Application of Biofresh + a biological agent is made by combining the two biological Agents in accordance with the treatment of each biological agent singly.

2.6. Planting

The soybean seeds used are Anjasmoro varieties. In the treatment of Agrisoy biological agents, seeds are mixed with Agrisoy biological agents according to the recommended dosage just before planting. Planting is done as much as 3 per planting hole with a spacing of 30 cm x 25 cm.

2.7. Inorganic fertilization

Inorganic fertilizers used are Urea, SP36 and KCl fertilizer with recommended dosages of Urea 100 kg.ha⁻¹, SP36 125 kg.ha⁻¹ and KCl 85 kg.ha⁻¹ respectively. Giving inorganic fertilizer is done at a dose of 50% of the recommended dose at the age of 2 weeks.

2.8. Plant maintenance

Plant maintenance includes replanting, weeding, and watering. Stitching is done at one week after planting, by replacing dead plants or growth that is not good. Weeding is done by pulling or cutting weeds that grow around the plantations at the age of 4 weeks after planting. Watering is done every two days in the afternoon if it does not rain.

2.9. Observation

2.9.1. The severity of virus mosaic disease. The severity of SMV disease was calculated using a scoring method through the assessment of diseased plant leaf scores based on the mosaic symptoms formed in sample plants [12]. The SMV attack category scores on leaves are 0, healthy leaves (no symptoms); 1, mosaic symptoms <50% of leaf area; 2, mosaic symptoms > 50% of leaf area; 3, mosaic symptoms and leaf size shrink; 4, the symptoms of mosaics with shrinking and wrinkled leaves; 5, mosaic symptoms with leaf size shrinking and wrinkled and leaf curling.

The results of the scoring assessment are then used to calculate the severity of the disease using a formula as proposed by Gultom [13], as follows:

$$DS = \sum_{i=0}^n \left(\frac{n.V}{Z.N} \right) \times 100 \% \quad (1)$$

Note DS, disease severity (%); n, number of leaves in each attack category; N, number of leaves observed; V, The scale value of each attack category; and Z, the highest scale value of the attack category.

2.9.2. Plant growth. Observation of plant growth was carried out on sample plants at the age of 10 WAP, including plant height (cm) measured from ground level to leaf tips and the number of productive branches.

2.9.3. Crops. Harvesting is done by cutting the base of the soybean stem that has matured physiology. Five plants were observed for each treatment and repetition. Observations include: (a) dry pod weight per plant (gram), (b) number of pods per plant, (c) number of seeds per plant, (d) seed weight per plant (gram), (e) weight 100 seeds per plant (grams), (f) crop production (ton ha⁻¹).

3. Results and discussion

3.1. The severity of virus mosaic disease

At the time of the research, the dominant disease in the field was a mosaic virus caused by a naturally developing soybean mosaic virus (SMV). Symptoms of SMV on the leaves observed were: mosaic, malformation, cupping, wrinkled, and yellowing. The symptoms found are the same as reported by Rahim et al., [14], who stated that the symptoms of Soybean mosaic virus found in soybean plantations in Java generally are mosaics, the leaf surface is uneven/blistered, thickening of leaf bones, leaf edges curved upward and yellowing. Analysis of variance showed that the interaction of treatments of different types of biological agents and organic matter had no significant effect on the

severity of SMV disease. Significant influence occurs on the independent treatment of biological agents and the independent treatment of organic matter (Table 1).

Biofresh (A_1), AgriSoy (A_2), and Biofresh + Agrisoy biological agents independently treated, each had an SMV disease severity of 16%, 15.67%, and 17.62%, these values were not significantly different from each other, but all are significantly different from controls that have a disease severity of 26.82%, this indicates that the application of biological fertilizers is able to induce a system of plant resistance to SMV infection. Khalimi and Suprpta [15] reported that the rhizobacteria of *Pseudomonas aeruginosa* could reduce the incidence of diseases caused by soybean stunt virus (SSV) in soybean plants by 10% to 75%.

Table 1. The effect of biological agents and organic matter on the severity of soybean mosaic virus in soybeans.

Independent Treatment	Disease Severity (%)	Control Effectiveness (%)
Main Plot: Type of Organic Material		
B0. Without Organic Material	15.28 b	-
B1. Straw Compost	21.93 a	(-) 43.52
B2. Soy Litter Compost	21.82 a	(-) 42.80
B3. Straw Compost+Soy Litter	17.07 ab	(-) 15.31
Subplot: Type of Organic Material		
A0. Without Biological Agent	26.82 a	-
A1. Biofresh Biological Agent	16.00 b	40.34
A2. Agrisoy Biological Agent	15.67 b	41.57
A3. Biofresh and Agrisoy Biological Agent	17.62 b	30.83

Note: The numbers in the same column followed by the same letters are not significantly different according to the DMRT test at the 95% confidence level

In the treatment of organic matter independently the lowest disease severity of 15.28% was found in the treatment without organic matter (B_0) not significantly different from the treatment of soybean + rice straw (B_3) litter compost mixture with disease severity of 17.07%, but significantly different from the treatment B_1 and B_2 . This shows that the treatment of organic material independently does not have an effect on the severity of SMV disease. The low incidence of disease in control because the plants in the treatment-experienced growth inhibition so that it looks stunted. Giesler [3] and Tilmon et al. [16], stated that *Aphis glycines* and *Empoasca terminalis* are the main vector insects of the Soybean mosaic virus in soybean plants. Stunted plants are not attractive for these vector insects to carry out feeding activities, this causes very low virus transmission which has implications for low disease events as well, but this resistance is the only apparent resistance

3.2. Plant growth

Analysis of the various interactions of the treatment types of biological agents and types of organic matter had no significant effect on plant growth, especially on plant height and stem diameter. Significant influence occurs on the self-treatment of organic matter and the self-treatment of biological agents. In the treatment of main independent plot, the highest plants obtained in the treatment of straw compost + soybean litter were not significantly different from the treatment of straw compost with values of 49.08 cm and 46.05 cm respectively, but significantly different from the control treatment and compost of rice straw each with a value of 38.31 cm and 43.73 cm. On the observation variable of the number of productive branches, the highest productive branch was obtained in the compost treatment of soybean litter with the number of productive branches of 2.90, which was significantly different from the control (Table 2).

In the independent treatment of biological agents, the best plant height was obtained in the treatment of biological agents with different Biofresh that were not significantly biological agents

Biofresh + Agrisoy biological agents. In the productive branch variable, the highest yield was obtained for the treatment of biological agents Biofresh + Agrisoy with the number of productive branches being 3.08, which was not significantly different from the treatment treatments and biological agents Agrisoy. These results support the results of previous studies that the application of Biofresh and Bokashi bio-fertilizer solid formulations seemed to give soybean yields the highest average plant height and number of leaves [10].

Table 2. The effect of biological agents and organic matter on soybean growth.

Independent Treatment	Plant Height (cm)	Productive Branch
Main Plot: Type of Organic Material		
B0. Without Organic Material	38.31 b	2.08 bc
B1. Straw Compost	46.05 a	2.88 a
B2. Soy Litter Compost	43.73 b	2.90 a
B3. Straw Compost+Soy Litter	49.08 a	2.80 ab
Subplot: Type of Organic Material		
A0. Without Biological Agents	39.22 b	2.53 a
A1. Biofresh Biological Agents	50.67 a	2.80 b
A2. Agrisoy Biological Agents	37.64 b	2.34 a
A3. Biofresh dan Agrisoy Biological Agents	49.65 a	3.08 b

Note: The numbers in the same column followed by the same letters are not significantly different according to the DMRT test at the 95% confidence level

3.3. Crops and productivity

The results showed that the interaction of different types of biological agents and organic matter had no significant effect on production variables, the real effect was seen on the independent treatment of organic substances and the independent treatment of biological agents (table 3).

Table 3. The average value of variable yields and production (tons/ha) soybean treated types of biological agents and the different organic materials.

Independent Treatment	amount Pod (fruit)	Weight Pod (g/tan)	amount Pod (fruit)	weight seed (G/tonne)	Productivity (ton / ha)	Efektivititas (%)
Main Plot: BO						
B0.Without BO	34.32 b	12.81 b	61.28 b	8.15 b	0.92 c	0
B1.Soy Compost	51.83 a	21.93 a	94.30 a	13.90 a	1.63 a	43.56
B2.Straw Compost	49.58 a	19.63 b	84.85 a	12.59 a	1.31 b	38.16
B3. Mixed BO	52.02 a	19.25 a	84.67 a	12.82 a	1.55 ab	40.64
Subplot: AH						
A0.Without AH	38.22 b	15.36 b	69.73 b	9.89 b	1.15 b	0
A1.Biofresh Agents	54.28 a	21.45 a	96,02 a	13.85 a	1.53 a	24.83
A2 Agrisoy Agent s	39.17 b	15.05 b	68.68 b	9.87 b	1.12 b	-2.67
A3.Mixed AH	56.08 a	21.76 a	90.67 a	13.84 a	160 a	28.13

Note: The numbers in the same column followed by the same letters are not significantly different according to the DMRT test at the 95% confidence level

The results in table 3 show that in the organic matter treatment independently, the soybean litter compost treatment showed a better yield variable than the other treatments, although it was not significantly different from B2 and B3 treatments, but significantly different from B0. The highest productivity was found in the treatment of soybean litter organic material that is 1.63 tons/ha with an effective increase in production of 43.56%.

While observing the biological agent treatment independently, the Biofresh (A₁) biological agent treatment and the Biofresh+AgriSoy mixture are two treatments that show higher yield values than the other two treatments. The two treatments did not differ from each other in all observational variables. In this independent treatment, the application of a mixture of Biofresh and Agrisoy Biological Agents is able to provide productivity of 1.60 tons/ha with an effective value of increasing production by 28.13%, not much different from the productivity of Biofresh biological agents treatment alone at 1.53 tons/ha with the effectiveness of increasing production by 24.83%.

The application of biological fertilizers also plays a role in the supply of nutrients for plants through the activities of basic microbial ingredients of biological fertilizers. Khaeruni et al. [8] reported that the biofresh making compost microbial consisting of rhizobacteria *Bacillus subtilis* ST21b, *Bacillus cereus* ST21B, and *Serratia* SS29a are a group of microbes capable of producing growth hormone (IAA), dissolving phosphate and fixing nitrogen from the air. Furthermore, Thakuria et al. [17], that the ability of rhizobacteria as a plant growth promoter is demonstrated by the ability to provide and mobilize the absorption of various nutrients in the soil and synthesize and change the concentration of various phytohormones. The supply of nutrients from bokashi and biological fertilizers is sufficient and balanced to support the process of plant photosynthesis and produce photosynthates that will be utilized by plants to form productive branches. Therefore the provision of bokashi and biological fertilizers can increase the availability of nutrients to support the growth and production of soybean plants in marginal lands.

4. Conclusion

The treatment of organic material and biological fertilizer does not influence the interaction of the incidence of viral mosaic disease (SMV), growth, and soybean production in marginal land. The independent treatment of organic matter and independent biological fertilizer affect the severity of SMV disease, plant height, number of productive branches, and soybean crop production. Biofresh and Agrisoy biofertilizers show compatibility in increasing resistance to SMV and production by 28.13% of plants in Ultisol soil.

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