Spodoptera litura Fabr. Control Using Beauveria bassiana to Reduce Pest Damage Intensity and Maintain the Yield of Soybean

Yati Setiati Rachmawati*, Dina Gustiana, Ayu Rosmiati, Cecep Hidayat, Efrin Firmansyah

Department of Agrotechnology, Faculty of Sains and Technology UIN Sunan Gunung Djati, Bandung, Indonesia

*email: yatisetiati@uinsgd.ac.id

Abstract. Soybean production has not been able to meet Indonesian needed. The production decreases every year due to the Spodoptera litura attack. The purpose of this study was to determine the effectiveness of Beauveria bassiana as a biological agent in controlling S. litura attacks in the field. The research was conducted at the Pest Laboratory of the Department of Agrotechnology, UIN Sunan Gunung Djati, Bandung, Indonesia from January to March 2017. The experiment used Completely Randomized Design (CRD) with eight B. bassiana spores density treatments with three replications. These treatments, i.e., negative control, positive control, 10^5, 10^6, 10^7, 10^8, and 10^9/ ml aquades applied to S. litura instar II larva. The post-hoc test used was Duncan's Multiple Range Test of 5%. The results showed that the density of B. bassiana spores in 10^10/ ml aquades caused mortality of S. litura larva at 76.67%. B. bassiana spores in 10^10/ ml aquades was able to maintain the growth of soybean plants with pest damage intensity of 3.81%, maintained photosynthates to seeds with the harvest index of 0.42 and the seed weight per plant reached 31.64 g. B. bassiana can be used as an entomopathogen in controlling attacks by Spodoptera litura.

Keywords: Beauveria bassiana, Spodoptera litura, soybean, yield

1. Introduction

Spodoptera litura is one of the main pests in soybean crop due to its polyphagous which can feed on almost all kinds of produce. This pest spreads to 22 provinces in Indonesia. It lives on various crops and hosts more than a hundred crop species, such as tobacco, groundnut, soybean, sweet potato, chili, shallot, mung bean, and corn [1], [2]. S. litura is polyphagous and feeds almost every kind of crop. S. litura larva attacks leaves and young pods that causes leaf damage as holes of bite marks and disrupts photosynthesis in soybean [3].

In farmer level, S. litura control is depend on insecticide of synthetic chemical compound which able to kill non-target organism, pest resistance, pest resurgence, and increase residual effect in plant and environment [4]. For these reasons, using chemical insecticide continuously brings out the concept of “Back to nature”, which propose non-chemical pest control using biological agents such as entomopathogenic fungi as bio-insecticide. Utilization of this entomopathogenic fungi is one of components of Integrated Pest Management.

One of entomopathogenic fungi which potential in controlling some pest species is Beauveria bassiana. This entomopathogen is reported as a biological agent that effectively controls some species from Coleoptera,
Lepidoptera, Hemiptera, Homoptera, Orthoptera, and Diptera. B. bassiana is disease-causing white muscardine agent in pest with white mycelium and conidium (spore), oval in shape, and grow in zig zag pattern on its conidiophore [5].

Entomopathogens infect pests characterized by the presence of white hyphae on the cuticle of the body and leading to the hemocoel in the infected pest's body. In hemocoel, hyphae will form “yeastlike hyphal bodies” (blastospore), which reproduce by forming buds. Blastospore grows and thrives in hemocoel by absorp haemolimphs fluid. Besides, this fungal infection produces toxic destruction enzim and causes tissue damage in insect [6].

B. bassiana in 100 gl⁻¹ concentration towards S. litura instar II larva has 85.71% mortality in 21 days after application [7]. Otherwise, in 150 gl⁻¹, larva instar IV increases 93.10% mortality in 21 days after application. This research aimed to control S. litura using B. bassiana to maintain damage intensity and yield of soybean.

2. Methode

This research was conducted in Pest Laboratory, Department of Agrotechnology, Faculty of Science and Technology, Universitas Islam Negeri Sunan Gunung Djati Bandung and in Cileunyi Wetan, Cileunyi, Bandung from January to May 2017. The material used B. bassiana isolate, S. litura larva, aquades, and soybean crop. The experimental method used a Completely Randomized Block Design with 8 treatments i.e. positive control, negative control, B. bassiana spores density of 10⁵, 10⁶, 10⁷, 10⁸, dan 10⁹ ml⁻¹ aquades with three replications.

The first step was S. litura propagation and culture. B. bassiana isolate collected from BALITSA (Balai Penelitian Tanaman Sayuran) Lembang, Kabupaten Bandung Barat. Reisolation of B. bassiana from S. litura. PDA media with 1 L potatoes extraction was cultured consisting of 200 g potatoes, 20 g sugar, and 20 g plain agarose and incubated in room temperature for 7 – 14 days.

Second step was making B. bassiana suspension liquid with positive control density, negative control (0 spores ml⁻¹), density of B. bassiana spores for 10⁵, 10⁶, 10⁷, 10⁸ and 10⁹ ml⁻¹, each was replicated three times. Afterhand, application of S. litura larva instar II to soybean plant for 10 larvae per plant. B. bassiana suspension applied directly to soybean plant in vegetative phase or 3 weeks after planting. B. bassiana suspension sprayed to S. litura larvae.

The parameters observed were (a) Damage Intensity (%), which observed in 1L days after B. bassiana suspension application and calculated using the formula [8] as below:

\[ DI = \frac{\sum (n \times v)}{(Z \times N)} \times 100\% \]  

(b) Plant Height (cm), which observed by measuring main stem from surface growing media to plant bud. This parameter observed in 4 weeks after application of S. litura infestation. (c) Harvest Index it is defined as the ratio of grain yield to total aboveground biomass. (d) Yield (g) was weighed in 90 days after planting or in harvest time.

3. Result and Discussion

3.1 Damage Intensity (%)

The results showed that B. bassiana spores effected to damage density (Table 1).

| B. bassiana spores density/ml aquades | Damage Intensity (%) |
|--------------------------------------|----------------------|
| A = 0 (control-)                      | 0.00    a            |
| B = 0 (control+)                      | 20.93   c            |

Note :
DI : Damage Intensity (%)
n : Number of samples with certain score
v : Score from each attack category
N : Number of observed sample unit
Z : Highest score used

(b) Plant Height (cm), which observed by measuring main stem from surface growing media to plant bud. This parameter observed in 4 weeks after application of S. litura infestation. (c) Harvest Index it is defined as the ratio of grain yield to total aboveground biomass. (d) Yield (g) was weighed in 90 days after planting or in harvest time.
Damage intensity observed 6 weeks after planting as *S. litura* larvae were 4 weeks after infestation and applied by *B. bassiana* suspension for 10 days observation. According to the results, *B. bassiana* affected to reduce *S. litura* larva attacks in soybean plant. The variance analysis showed that *B. bassiana* application has a significant effect on the damage intensity of soybean plant (Table 1).

Table 1 showed concentration of spore density significantly affected to percentage of damage intensity. The higher density of spores, higher percentage of damage intensity. Spores density of $10^{10}$ spores/ml aquades has percentage of damage intensity for 3.81% significantly different from positive control (20.93%) and not significantly different from spore density of $10^{9}$, $10^{8}$, $10^{7}$, and $10^{6}$/ml aquades.

Concentration of spores density for $10^{10}$, $10^{9}$, $10^{8}$, $10^{7}$, dan $10^{6}$/ml aquades obtained damage intensity lower than positive control, thus it is effective enough to reduce damage intensity in soybean caused by *S. litura*. The higher concentration of spore density, the lower damage intensity caused by *S. litura*. It can be claimed that there were more spores infected larva so the enzymes (protease, lipolitic, amylase, and chitinase) and toxin (Beauveriacin) infected *S. litura* by showing the symptom and died (Figure 1). Damage intensity related to larva mortality and higher density of *B. bassiana* obtained high mortality.

![Figure 1 Larva of *S. litura* infected by *B. bassiana* and covered by white mycelium](imagelink)

### 3.2 Plant Height (cm)

Plant height measurement conducted by measuring main stem from upper ground/surface to plant bud after applying *S. litura* larva to the plant on 4 weeks after planting. The results showed that application *B. bassiana* application was not significantly different to the plant height (Table 2).

Table 2. The Effect of *B. bassiana* application to the Plant height of Soybean on 6 weeks after planting

| *B. bassiana* spores density / ml aquades | Plant Height (cm) |
|------------------------------------------|-------------------|
| A = 0 (Control-)                          | 68.67 a           |
| B = 0 (Control+)                          | 68.00 a           |
| C = $10^5$                                | 68.00 a           |
| D = $10^6$                                | 67.00 a           |
| E = $10^7$                                | 67.67 a           |
| F = $10^8$                                | 67.63 a           |
| G = $10^9$                                | 67.67 a           |
| H = $10^{10}$                             | 68.00 a           |

Note: Numbers in the same column followed by different letters showed significantly different results as Duncan's Test in 5%.

All treatments showed no significant different to the plant height, thus *B. bassiana* application was able to maintain soybean growth by reducing damage intensity caused by *S. litura* larva. Damage intensity appears to
include the low category because the percentage was < 40% [9]. It showed that the damage was not harm, only the symptom appeared as bite holes on leaves. *S. litura* attack on leaves is not obstructing photosynthesis because it does not damage all plant organs; thus, leaves still have chlorophyll to process the photosynthesis.

Photosynthesis produces assimilate which is used for plant growth and development. Assimilate is utilized on vegetative and generative phases. In vegetative phase shows increasing height, stem diameter and number of leaves, while in the generative phase, it shows fruit formation [10].

### 3.3 Harvest Index

Harvest index is plant ability to transfer assimilate, non-unit. Harvest index measured one time during harvest by calculate dry seed weight and dry stover weight (whole plant). Almost 90% dry materials of plant are from photosynthesis, thus the parameter was measured by plant dry weight. Drying the parts of plant usinh oven on tempreature of 80°C. The constant weights are absolute from plant organs without water content. The results showed that hasvest index was not significantly different for each treatment (Table 3).

**Table 3. Effect of *B. bassiana* to The Harvest index of soybean**

| B. *bassiana* spores density / ml aquades | Harvest Index |
|------------------------------------------|---------------|
| A = 0 (Control-)                         | 0.43 a        |
| B = 0 (Control+)                         | 0.26 a        |
| C = 10⁵                                  | 0.29 a        |
| D = 10⁶                                  | 0.29 a        |
| E = 10⁷                                  | 0.32 a        |
| F = 10⁸                                  | 0.34 a        |
| G = 10⁹                                  | 0.36 a        |
| H = 10¹⁰                                 | 0.42 a        |

Note: Numbers in the same column followed by different letters showed significantly different results as Duncan's Test in 5%.

According to the Table 3, harvest index were not significantly different to all treatments. This is because the photosynthate produced by plants is distributed to the seeds. *B. bassiana* maintains the harvest index in soybean plants by suppressing the attack of *spodoptera litura*.

### 3.4 Seed weight per plant (g)

The dry seed weighed on 90 days after planting or during harvest and drying the seeds. The seed drying to eliminate the water content inside the seeds so the weight is the absolute numbers. The seeds weighted are the full seed (Figure 2.a).

![Figure 2. Seed characteristics](image)

(a) full seeds of soybean, and (b) empty seeds of soybean

The full seeds has a soft testa (seed coat), yellow in color, circle shape, and hard in texture, otherwise, the empty seed (Figure 2.b) has a wrinkle testa, oval in shape, various colors from green to brown. The variance analysis showed the *B. bassiana* significantly affected the dry seed weight of soybean (Table 4).

**Table 4. Effect of *B. bassiana* to Dry Seed Weight of Soybean**
### B. bassiana spores density / ml aquades

| B. bassiana spores density / ml aquades | Dry Seed Weight (g) |
|----------------------------------------|---------------------|
| A = 0 (control-)                        | 34.25 e             |
| B = 0 (control+)                        | 11.06 a             |
| C = 10⁵                                 | 14.05 ab            |
| D = 10⁶                                 | 16.28 ab            |
| E = 10⁷                                 | 18.99 ab            |
| F = 10⁸                                 | 21.04 cd            |
| G = 10⁹                                 | 25.04 cd            |
| H = 10¹⁰                                | 31.64 de            |

Note: Numbers in the same column followed by different letters showed significantly different results as Duncan's Test in 5%.

*B. bassiana* could effect the numbers of soybeans seeds, dry seed weight. Application of *B. bassiana* $10^{10}$ spores / ml aquades resulted in soybean seed weight of 31.64 g. This treatment was not significantly different from the negative control (34.25 g) but significantly different from the positive control (11.06 g). Based on these data, with the application of b.bassiana as an entomopathogen, soybean production can be maintained.
4. Conclusion

*B. bassiana* is useful as a biological agent that can infect a disease, even causing death in the host insect. The lowest percentage of damage intensity showed by spore density of $10^{10}$ ml$^{-1}$ for 49% damage. It can also maintain soybean growth by not affecting plant height and keeping the photosynthate into the seed with the highest harvest index value is 0.42 and dry seed weight of 31.64 g plant$^{-1}$.

5. Acknowledgments

Our big thanks to the head of the agrotechnology department, the dean of the science and technology faculty of UIN Sunan Gunung Djati for supporting and financing the cost of publishing this research.

References

[1] Muhtarudin *et al.*, “Effect of Grass Variety and Shade under Palm Oil Plantation on Production and Proportion of Stems, Leaves and Nutrition Content of Grass,” *J. Biol. Sci.*, vol. 20, no. 3, pp. 116–122, 2020, doi: 10.3923/jbs.2020.116.122.

[2] C. Bragard *et al.*, “Pest categorisation of Spodoptera litura,” *EFSA J.*, vol. 17, no. June, pp. 1–35, 2019, doi: 10.2903/j.efsa.2019.5765.

[3] A. Krisnawati, M. Santi, Y. Ika, and M. M. Adie, “Identification of soybean genotypes based on antixenosis and antibiosis to the armyworm (Spodoptera litura),” vol. 9, no. 2, pp. 164–169, 2017, doi: 10.13057/nusbiosci/n090210.

[4] Shakti Singh Bhati, “We are IntechOpen, the world ’s leading publisher of Open Access books Built by scientists, for scientists TOP 1% Management of Spodoptera litura (Fab.) in Green Gram,” *Trends Integr. Insect Pest Manag.*, 2019, doi: http://dx.doi.org/10.5772/intechopen.85645 armigera).

[5] V. Shanmugam and P. Seethapathy, “Isolation and characterization of white muscardine fungi Beauveria bassiana (Bals.) Vuill. - A causative of mulberry silkworm,” vol. 5, no. 3, pp. 512–515, 2017.

[6] M. Alejandra, E. Mora, A. Marcelo, C. Castilho, and M. E. Fraga, “Classification and infection mechanism of entomopathogenic fungi,” *Agric. Microbiol.*, vol. 84, pp. 1–10, 2017, doi: 10.1590/1808-1657000552015.

[7] D. R. Indriyanti, S. Mahmuda, and M. Slamet, “Effect Of Beauveria Bassiana Doses On Spodoptera Litura Mortality,” vol. 6, no. 09, 2017.

[8] D. Norman, R. J. Henny, and J. M. F. Yuen, “Disease Resistance in TwentyDieffenbachia Cultivars,” *Hot Sci.*, vol. 32, no. 4, 1997.

[9] A. Otuka, M. Matsumura, and M. Tokuda, “Dispersal of the Common Cutworm, Spodoptera litura, Monitored by Searchlight Trap and Relationship with Occurrence of Soybean Leaf Damage,” *Insects*, vol. 11, no. 7, p. 427, Jul. 2020, doi: 10.3390/insects11070427.

[10] N. I. Zakaria, M. R. Ismail, Y. Awang, P. E. Megat Wahab, and Z. Berahim, “Effect of Root Restriction on the Growth, Photosynthesis Rate, and Source and Sink Relationship of Chilli (*Capsicum annuum* L.) Grown in Soiless Culture,” *Biomed Res. Int.*, vol. 2020, p. 2706937, 2020, doi: 10.1155/2020/2706937.