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# **COVERING LETTER**

# Dear Editor-in-Chief,

I herewith enclosed a research article,

# Title:

Herbivore insects and predatory arthropods in freshwater swamp rice field in South Sumatra sprayed with bioinsecticides of entomopathogenic fungi and abamectin

# Author(s) name:

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# Novelty:

(state your claimed novelty of the findings versus current knowledge)

This study highlights the finding that the bioinsecticide from *Beauveria bassiana*, *Metarizhium anisopliae*, and *Cordycep militaris* did not decrease the abundance and species diversity of predatory arthropod (non-target arthropods), but could decrease the herbivore insects (target pests) population.

# Statements:

This manuscript has not been published and is not under consideration for publication to any other journal or any other type of publication (including web hosting) either by me or any of my co-authors. Author(s) has been read and agree to the Ethical Guidelines.

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### Place and date:

Palembang, 6 June 2020

# Sincerely yours,

(fill in your name, no need scanned autograph) Siti Herlinda

#### Herbivore insects and predatory arthropods in freshwater swamp rice 1 field in South Sumatra sprayed with bioinsecticides of 2 entomopathogenic fungi and abamectin 3

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14 Abstract. Herbivore insect population and predatory arthropod in rice field may be effected by the application entomopathogenic fungi 15 or synthetic insectide. The objective of this research was to analyze individual quantity of herbivore insects and predatory arthropod 16 inhabiting freshwater swamp rice field treated with bioinsecticides and abamectin (commercial insecticide). The research used the 17 bioinsecticides made from fungi (Beauveria bassiana, Metarhizium anisopliae, Cordyceps military), and the abamectin. This research 18 found 12 families of herbivore insects with 22 species dominated by Nilaparvata lugens and Leptocorisa acuta. This study found 32 19 species of spider belonged to 8 families dominated by Tetragnatha virescens and Oxyopes matiensis. The species diversity of spider was 20 higher in the plots of the bioinsecticide compared to that of the abamectin. Predatory insects found belonged to 14 species belonged to 21 8 families dominated by species of Ophionea nigrofasciata, Verania discolor, and Paedorus fuscipe. The abundance of predatory 22 insects in plots sprayed with the bioinsecticides was higher compared to that of the abamectin plot. It concluded that the abundance and 23 species diversity of predatory arthropod sprayed with bioinsecticide of B. bassiana, M. anisopliae, and C. militaris did not decrease, 24 while the population of herbivore insect tent to decrease.

25 Key words: Beauveria bassiana; Metarhizium anisopliae; Cordyceps militaris; neutral insect; parasitoid

26 Abbreviations (if any): -

27 Running title: Arthropods in freshwater swamp rice field

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### **INTRODUCTION**

29 Rice cultivation in freshwater swamps of South Sumatra has specific characteristics making it different from rice 30 cultivation in other ecosystem in Indonesia. Rice cultivation has been depended on climate conditions, especially rainfall and tidal flush. In the period of November to April, freshwater swamps are generally inundated so that local farmers 31 cultivate rice only in the period of May to September (Herlinda et al. 2018; Herlinda et al. 2019a). Due to the difficulty in 32 33 managing the water, the farmers grow rice once a year using transplanting method. In maintaining their rice cultivations, 34 the local farmers scarcely apply synthetic insecticide (Herlinda et al. 2019b).

35 The population of pest insect in freshwater swamp rice has been increasing and starting to cause yield losses, especially 36 rice bug (Leptocorixa acuta), while the previously disappeared brown planthopper (Nilavarpata lugens) started appearing 37 (Hanif et al. 2020). The pest insects of rice have been controlled using entomopathogenic fungi, Beauveria bassiana (Li et 38 al. 2012; Li et al. 2014; Lee et al. 2015) and Metarizhium anisopliae (Girish and Balikai, 2015; Chinniah et al. 2016). There 39 has been the report on the use of Cordycep militaris to control pest insect of rice (Prabawati et al. 2019). The C. militaris 40 could produce beauvericin of toxin insects (Rachmawati et al. 2018). B. bassiana had been proven to be effective against homopteran (Li et al. 2012; Li et al. 2014; Lopez et al. 2014; Lee et al. 2015; Sumikarsih et al. 2019), coleopteran 41 42 (Kavallieratos et al. 2015), hemipteran (Girish and Balikai 2015), and lepidopteran insects (Ayudya et al. 2019; Ma et al. 43 2019; Gustianingtyas et al. 2020). M. anisopliae had also been demonstrated to be effective against homopteran (Mweke 44 et al. 2019), lepidopteran (Gustianingtyas et al. 2020), and coleopteran insects (Kavallieratos et al. 2015). M. anisopliae 45 could be used in multiple roles, ranging from controlling the insect pests to promoting plant growth (Liu et al. 2017).

46 Being effective against various orders of insects, including the coleopteran insects (Kavallieratos et al. 2015), the rice 47 field ecosystem which is dominated by species of predatory insects, it is important to be very careful in the application of 48 entomopathogenic fungi in the ecosystem to avoid bad impact on non-target arthropods (Scorsetti et al. 2017). The high 49 species diversity and abundance of arthropod in the freshwater swamp rice field (Herlinda et al. 2018) might be disturbed by the application of the entomopathogenic fungi. However, there have been some reports suggested that entomopathogenic 50 fungi did not affect abundance and species diversity of arthropods. For example, predatory insect in paddy field (Andrallus 51 spinidens) was reported to be resistant to *B. bassiana* (Firouzbakht et al. 2015; Gholamzadeh-Chitgar et al. 2017; 52 53 Scorsetti et al. 2017). Parasitoid tends to avoid host insect infected by entomopathogenic fungi (Emami et al. 2013). 54 Coccinellidae, the family of generalist predatory insect, was reported to be resistant to *M. anisopliae* (Bayissa et al. 2016). 55 This research aimed to analyze individual quantity of herbivore insects and predatory arthropod inhabiting freshwater 56 swamp rice field treated with bioinsecticides contaning Beauveria bassiana, Metarhizium anisopliae, and Cordyceps 57 *militaris* and the abamectin.

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#### MATERIALS AND METHODS

#### 59 Study area

The research was conducted from April to August 2018 in freshwater swamp rice field located in Village Pelabuhan Dalam, Sub District Pemulutan, District Ogan Ilir, and South Sumatra. The experiment was arranged in a Completely Randomized Block Design consisted of four plots of treatment. The first three plots were treated with bioinsecticide containing *B. bassiana*, *M. anisopliae*, *C. military*, and the forth plot was treated with the abamectin (the commercial insecticide) as control. The plot area was  $120 \text{ m}^2$  for a treatment and each the treatment was replicated five times.

# 65 Land preparation, transplanting, and crop maintenance

Land preparation was conducted using moldboard plow and was continued by using harrow plow. Before being 66 67 plowed, the land was cleared using hoe and long knife. The excessive water was pumped out until the soil was slightly watery, and dung compost was added at a dosage of 1 ton ha<sup>-1</sup> Rice seed used were certified purple labeled seed of 68 Mekongga variety at a dosage of 50 kg/ha. Rice was cultivated using transplanting system from which, the seedlings were 69 prepared in 34 x 26 x 5 cm<sup>3</sup> plastic trays filled with 3 cm high soil. The seeds were soaked in water for 24 hours before 70 being planted in the prepared plastic trays. The soaked seeds were spread evenly in each tray at a dosage of 150 g per tray. 71 The trays were then covered with thick plastic for 7 days. The 7 day old seedlings were then transferred to places 72 73 receiving enough sun light. Fourteen day old seedlings were transplanted to all prepared plots at a density of 2 seedlings 74 per hole in a 2:1 (12.5 x 25 x 50 cm<sup>3</sup>) "jajar legowo" planting system (Figure 1).



Figure 1. Two day old rice seedling in a plastic tray (A), 7 day old seedlings (B), rice transplanting (D) rice planted in "jajar legowo" planting system (E)

Crop maintenance was conducted in the form of replanting, irrigating, weeding, and fertilizing. Replanting was conducted 7 days after transplanting to replace the dead seedlings. Irrigating was conducted by making use of water flush from river under high tide and pumping system. Weeding was conducted both at vegetative and generative stages using sickle. Fertilizer used was shrimp shell extract compost prepared according to method by Suwandi et al. (2012). The compost was applied every two weeks started from 14 days after planting until harvesting (84 days) at a dosage of 2 L ha<sup>-1</sup>. The extract compost was a fermentation result of shrimp shell meal containing chitinolitic bacteria, selulotic bacteria and sulphate diluter. Nitrogen fertilizer was applied 30 days after planting at a dosage of 100 kg ha<sup>-1</sup>.

# 87 Formulation and application of bioinsecticides

All species of fungi have been explored from soil according to Kin et al. (2017). *B. bassiana* and *M. anisopliae* used in
this research were explored from freshwater swamp soil of South Sumatra, while *C. militaris* was explored from soil in
Central Kalimantan. *B. bassiana*, *M. anisopliae*, and *C. militaris* were cultured in solid media of Sabouraud Dextrose
Agar (SDA, Merck) made from 16.2 g SDA in 250 mL aquadest. The cultures were incubated for 10 x 24 hours. The 10
days old fungal solid cultures were then cultured in liquid media (Figure 2) of Sabouraud Dextrose Broth (SDB, *Merck*)
made from 30 g SDB in 1000 ml aquadest and were incubated in a 120 rpm shaker for 7 x 24 hours. The liquid culture of
the entomopathogenic fungi were used to make bioinsecticide. Seven days after incubation, the density of fungal spores of

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95 the bioinsecticide was counted, the counting was stopped when the spore density reached 1 x 10<sup>9</sup> conidia mL<sup>-1</sup>. The 96 bioinsecticide was made by adding shrimp shell meal extract compost as carrier, while active ingredients used were 97 entomopathogenic fungi and vegetable oil. One liter of bioinsecticide comprised of 400 mL shrimp shell meal extract 98 compost, 600 mL liquid culture of entomopathogenic fungi and 10 mL vegetable oil. Compost extract used to make 99 bioinsecticide was previously autoclaved under 1 atm for 2 hours.

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**Figure 2.** Cultures of *Beauveria bassiana* (A), *Metarhizium anisopliae* (B), and *Cordycep militaris* (C) on SDB; liquid bioinsecticide of *Beauveria bassiana* (D), *Metarhizium anisopliae* (E), and *Cordycep militaris* (F)

The bioinsecticides were applied at dosage of  $2 \text{ L} \text{ ha}^{-1}$  per application. The applications were conducted at 13, 27, 41, 55, 69, and 83 days after transplanting (DAT). The bioinsecticide applications were conducted in the evening (at 5 pm) to avoid spores damage by ultra violet. Control plot was sprayed with abamectin at a dosage of 0.5 L ha<sup>-1</sup>. The insecticide contained abamectin active ingredient and was sprayed at the same time as bioinsecticide applications. The control plot was 50 m away from treatments plots to avoid contamination in the treatment plots, and there was 10 m distance between bioinsecticide treatment plots. A day after application the bioinsecticides and the abamectin, arthropod samplings were carried out to collect arthropods inhabiting rice canopy.

# 112 Arthropod sampling in rice canopy

Sampling to collect arthropods inhabiting rice canopy was conducted using an entomological net according to Lami et al. (2016) and Ivantsova et al. (2017). The net had 75 cm length and 30 cm diameter of net and 100 cm length of handle. Sampling was made in every treatment subplot and was repeated five times. Samplings were conducted at 14, 28, 42, 56, 70, and 84 DAT. Arthropod samplings were conducted in the morning, at 6-7 am. During sampling, the sweep net was swept 30 cm depth into rice canopy to collect representative samples.

Sampled arthropods were cleaned and placed in plastic bottles (330 mL) containing 100 mL absolute ethanol, the bottles were labeled. The samples were taken into the Laboratory of Entomology, Department of Plant Pest and Disease, Faculty of Agriculture, Universitas Sriwijaya for identification. Spider identification was conducted using reference of Barrion and Litsinger (1995) while for insect identification we used reference of Gullan and Cranston (2014) and Heinrichs et al. (2017).

# 118 Data analysis

Data of arthropods species were grouped based on their guild, comprised of herbivore insects, predators, parasitoids, 119 120 and neutral insects, and was presented in the form of graphic. The difference of individual quantity of the herbivore insects, spiders, and predatory insects amongst treatments was analyzed using Analysis of Variance (ANOVA). If there 121 significant difference among treatments, the analyses was continued with Tukey's HSD (Honesty Significant 122 was Difference) test at 5% degree of significance. Analyses data was conducted using Software of SAS University Edition 2.7 123 124 9.4 M5. Data of abundance were used to analyze species diversity by using Shannon index (H'). Degree of diversity was 125 counted using the Evenness index (J') derived from the Shannon function, and Berger-Parker dominance biodiversity 126 indices. The coefficient of Sorensen was counted to measure degree of similarity of spider or predatory insect among 127 treatments (Magurran 2004).

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### **RESULTS AND DISCUSSION**

### 129 Abundance of herbivore insects in one rice cropping season

130 In one rice cropping season, it was found 22 species of herbivore insects belonged to 12 families. Dominant herbivore 131 species were *Nilaparvata lugens*, *Leptocorisa acuta*, *Acrida turita*, *Valanga nigricornis*, *Dicladispa armigera* (Figure 3).

Population of the herbivore was not significantly different among treatments, except for Family Cecidomyidae (Table 1).

133 Therefore, the capacity of bioinsecticide in suppressing the population of the herbivore insects has generally almost similar 134 to the capacity of the abamectin.



**Figure 3.** Herbivore insects found in rice canopy: *Spodoptera* sp. (A), *Pelopidas* sp. (B), *Amata nigriceps* (C), *Oxya chinensis* (D), *Scirpophaga* sp. (E), *Dicladispa armigera* (F), *Chrysolina coerulans* (G), *Acrida turita* (H), *Acrida turita* (I), *Nilaparvata lugens* (J)

**Table 1.** Population of herbivore insects in rice canopy sprayed with bioinsecticide containing *B. bassiana*, *M. anisopliae* and *C. militaris* and abamectin

No		Mean of herbivore abundance (individuals/nets)			F	D voluo	Tukov'e	
•	Ordo / Family/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	value	(0.05)	HSD test
	COLEOPTERA	0.2	0.13	0.17	0.23	0.26	0.85	
	Chrysomelidae	0.2	0.13	0.17	0.23	0.26	0.85	
1	Chrysolina coerulans	0.07	0	0.07	0.2	2.11	0.15	
2	Dicladispa armigera	0.13	0.13	0.1	0.03	0.53	0.67	
	DIPTERA	$0.4^{b}$	0.03 <sup>ab</sup>	$0.07^{ab}$	$0.00^{a}$	4.40*	0.03	0.20
	Cecidomyiidae	$0.4^{b}$	0.03 <sup>ab</sup>	$0.07^{ab}$	$0.00^{a}$	4.40*	0.03	0.20
3	Unknown sp.	0.33	0	0	0	2.55	0.1	
4	<i>Orseolia</i> sp.	0.07	0.03	0.07	0	0.81	0.51	
	HEMIPTERA	1.67	1.1	1.7	0.97	2.42	0.12	
	Alydidae	0.57	0.4	0.73	0.5	2.02	0.17	
5	Leptocorisa acuta	0.57	0.4	0.73	0.5	2.02	0.17	
	Cicadellidae	0.17	0.1	0	0.1	0.42	0.74	
6	Nephotettix cincticeps	0	0	0	0.03	1.00	0.43	
7	Nephotettix virescen	0	0.1	0	0.07	1.04	0.41	
8	Recilia dorsalis	0.17	0	0	0	1.00	0.43	
	Delphacidae	0.93	0.6	0.97	0.37	2.59	0.1	
9	Nilaparvata lugens	0.77	0.53	0.9	0.27	2.23	0.14	
10	Sogatella furcifera	0.17	0.07	0.07	0.1	0.50	0.69	
	LEPIDOPTERA	0.37	0.3	0.5	0.17	2.61	0.1	
	Pyralidae	0.33	0.23	0.33	0.17	0.99	0.43	
11	Cnaplocrosis medinalis	0	0	0	0.03	1.00	0.43	
12	Scirpophaga incertulas	0.17	0.13	0.17	0.07	1.31	0.32	
13	<i>Scirpophaga</i> sp.	0.17	0.1	0.17	0.07	0.66	0.59	
	Erebidae	0	0	0.07	0	1.00	0.43	
14	Amata nigriceps	0	0	0.07	0	1.00	0.43	
	Hesperiidae	0	0	0.03	0	1.00	0.43	
15	Pelopidas mathias	0	0	0.03	0	1.00	0.43	
	Noctudae	0.03	0.07	0.07	0	0.47	0.71	
16	Spodoptera sp.	0.03	0.07	0.07	0	0.47	0.71	
	ORTHOPTERA	0.63	0.83	0.67	0.8	0.58	0.64	
	Acrididae	0.63	0.83	0.67	0.77	0.48	0.7	
17	Acrida turrita	0.2	0.17	0.1	0.4	2.68	0.09	
18	Gesonula mundata	0.03	0.1	0.03	0.1	0.35	0.79	
19	Oxya chinensis	0.07	0.1	0.1	0.1	0.07	0.97	
20	Valanga nigricornis	0.33	0.47	0.43	0.17	2.02	0.17	
	Pyrgomorphidae	0	0	0	0.03	1.00	0.43	
21	Atractomorpha crenulata	0	0	0	0.03	1.00	0.43	
	THYSANOPTERA	0.1	0.1	0.1	0.07	0.11	0.95	
	Thripidae	0.1	0.1	0.1	0.07	0.11	0.95	
22	Liothrips sp.	0.1	0.1	0.1	0.07	0.11	0.95	
	TOTAL	3.37	2.5	3.2	2.23	2.77	0.09	

141 Note: \*: significantly different; values within a row followed by the same letters were not significantly different at P<0.05 according to</li>
 142 Tukey's HSD test

During field observations, insect died due to infection by entomopathogenic fungi were documented and showed specific symptoms of each fungus. Larva of *S. litura* found infected by *B. bassiana* in the rice field showed white mycelia covering its body, dried and not smelly (Figure 4). Larvae of *S. litura* infected by *M. anisopliae* showed symptoms of dried body covered by greenish white mycelia and was not smelly, while larvae of *S. litura* infected by *C. militaris* showed symptoms similar to those of *B. bassiana* infection, larval body drying, shriveled, covered by white mycelia, and not smelly. Entomopathogenic fungi infected sampled insects were isolated and purified in the laboratory. Color, structure, and spore of the isolated fungi were as presented in Figure 5.

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**Figure 4.** Herbivore insects infected by the entomopathogenic fungal of bioinsecticide in rice field: healthy larva of *Spodoptera* sp. (A), infected by *B. bassiana* (B), infected by *M. anisopliae* (C), and infected by *C. militaris* (D)



Figure 5. Spores of *Beauveria bassiana* (A), *Metarhizium anisopliae* (B), and *Cordycep militaris* (C); colonies of *Beauveria bassiana* (d), *Metarhizium anisopliae* (E), and *Cordycep militaris* (F)

# 159 Abundance of spider and predatory insect in one rice cropping season

This research found 32 species of spider belonged to 8 families. The dominant species of them were *Tetragnatha virescens, T. virescens, T. maxilosa, Argiope catenulata,* and *Oxyopes matiensis* (Figure 6). Total abundance of spider in the plot treated with the abamectin was significantly lower than that of plots treated with bioinsecticide (Table 2). The abundance of *Bathyphantes* sp. in abamectin plot was decreasing. The highest abundance of *Araneus inustus* was found in plot treated with bioinsecticide *C. militaris* and was different from that of other treatments. The abundance of Tetragnathidae was significantly decreasing in plot treated with insecticide compared to that of plots treated with bioinsecticide.

Predatory insects found in this research belonged to 14 species and 8 families. The dominant species were
 *Formicomus* sp., *Ophionea nigrofasciata, Verania discolor, Verania lineata, Menochilus sexmaculatus, Micrapis inop, Paedorus fuscipe, Cyrtorhinus lividipennis*, and *Orthotylus* sp. (Figure 7). The abundance of predatory insects was not
 significantly different among treatments (Table 3).

# 171 Characteristic of spider and predatory insect community in one rice cropping season

Number of spiders were increasing with the increase of rice age (Table 4). The highest number of spiders occurred when the rice were at mature grain ripening stage (84 DAT). The abundance of spider in the plot treated with abamectin tent to be lower compared to that of plots treated with the bioinsecticides. Total number of spiders in one rice cropping season were higher in the plots treated with the bioinsecticide than that in plot treated with the abamectin. The species diversity of spiders was also higher in the plots treated with entomopathogenic fungal bioinsecticide was also followed by the high species evenness and low species dominance.

Number of predatory insects were increasing with the increase of rice stage, but at approaching to harvest (84 DAT),
the number of predatory insects drastically decreased (Table 5). The highest abundance of predatory insect occurred at
milk grain ripening stage (70 DAT). The phenomenon of abundance fluctuation of predatory insect was slightly different
from that of spider which its peak occurred at mature grain ripening stage (84 DAT). The abundance of predatory insect in

183 plots treated with abamectin was generally lower than that of plots treated with the entomopathogenic fungal 184 bioinsecticide. Total of abundance of predatory insects in one rice cropping season of plots sprayed with 185 entomopathogenic fungal bioinsecticide was higher compared to that of abamectin plot, however, species diversity, 186 evenness and species dominance tent to be similar among treatments.

Spider community in plot sprayed with the bioinsecticide of *B. bassiana* tent to be more similar to that of plots sprayed with bioinsecticide (*M. anisopliae* and *C. militaris*) than that of abamectin plot. Total of one rice cropping season, spider community in plot sprayed with bioinsecticide *B. bassiana* was more similar to that of plot of *C. militaris* (0.77), followed by plot *M. anisopliae* (0.76), and least similar to that of abamectin plot (0.49) (Table 6).

Predatory insect community in plot sprayed with bioinsecticide *B. bassiana* tent to be more similar to that of plots sprayed with bioinsecticide *M. anisopliae* and *C. militaris* than that of abamectin plot (Table 7). Total of one rice cropping season, predatory insect community in plots sprayed with bioinsecticide *B. bassiana* was more similar to that of plot *M. anisopliae* (0.91), followed by plot of *C. militaris* (0.85), and least similar to that of insecticide plot (0.66).



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**Figure 6.** Spiders found in rice canopy: Linyphiidae (A), *Tetragnatha virescens* (B), *Tetragnatha maxilosa* (C), *Argiope catenulate* (D), *Oxyopes matiensis* (E), Lycosidae (F), Salticidae A (G), Salticidae b (H), *Oxyopes javanus* (I), *Pardosa* sp. (J)



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**Figure 7.** Predatory insect in rice field: Verania lineata (A), Menochilus sexmaculatus (B), Micrapis inops (C), Formica sp. (D), Odontoponera transversa (E), Argia sp. (F), Agriocnemis sp. (G), Ophionea nigrofasciata (H), Paederus fuscipes (I), and Conocephalus longipennis (J)

# 207 The abundance of arthropod guilds in one rice cropping season

The abundance of arthropod of all guilds tent to increase with the increase of rice growth stage (Figure 8). In all observations, the abundance of predatory insect was always higher compared to the abundance of other guilds (neutral insect, herbivore, and parasitoid). The abundance of predator was generally higher in plots sprayed entomopathogenic fungal bioinsecticide compared to that of abamectin plot. The abundance of all guilds of arthropod in plot of abamectin was always lower compared to that of bioinsecticide plots. When rice was 70 days old, the abundance of herbivore reached its peak, then decreased at mature grain stage. The abundance of predator reached its peak when rice was 84 days old.

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Figure 8. Arthropod abundance on rice field treated with bioinsectides and abamectin (insecticide)



No		Mean	<u>of spider abunda</u>	<u>nce (individual</u>	s/nets)	-	P voluo	Tukey's
110	Ordo / Family/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	F-value	(0.05)	HSD test
	ARACHINIDA							
	Araneidae	0.03	0.07	0.27	0.13	2.02	0.17	
1	Araneus inustus	$0.00^{a}$	0.03 <sup>ab</sup>	$0.2^{b}$	0.03 <sup>ab</sup>	4.82*	0.02	0.11
2	Unknown	0.03	0.03	0.07	0.07	0.35	0.79	
3	Gea subarmata	0.00	0.00	0.00	0.03	1.00	0.43	
	Linyphiidae	0.47	0.30	0.20	0.10	2.76	0.09	
4	Bathyphantes tagalogensis	0.00	0.00	0.07	0.00	2.67	0.10	
5	Bathyphantes sp. A	0.07	0.00	0.00	0.07	0.89	0.48	
6	Bathyphantes sp. B	0.13 <sup>ab</sup>	$0.2^{b}$	$0.07^{ab}$	$0^{\mathrm{a}}$	3.74*	0.04	0.12
7	Atypena adelinae	0.03	0.03	0.03	0.03	0.00	1.00	
8	Erigone bifurca	0.03	0.00	0.00	0.00	1.00	0.43	
9	Linyphiidae Sp. A	0.20	0.03	0.03	0.00	2.46	0.11	
10	Atypena formosana	0.00	0.03	0.00	0.00	0.98	0.44	
	Lycosidae	0.10	0.13	0.37	0.10	1.90	0.18	
11	Arctosa sp.	0.00	0.03	0.03	0.00	0.60	0.63	
12	Pardosa birmanica	0.03	0.00	0.00	0.00	1.00	0.43	

13	Pardosa pseudoannulata	0.03	0.00	0.27	0.07	2.10	0.15	
14	Pardosa apostoli	0.00	0.03	0.00	0.00	1.00	0.43	
15	Pardosa pullata	0.03	0.03	0.03	0.00	0.37	0.77	
15	Hogna rizali	0.00	0.00	0.03	0.00	1.00	0.43	
16	Pirata luzonensis	0.00	0.03	0.00	0.03	0.62	0.62	
	Oxyopidae	0.03	0.03	0.10	0.03	0.51	0.68	
17	Oxyopes javanus	0.00	0.03	0.07	0.03	0.76	0.54	
18	Oxyopes matiensis	0.00	0.00	0.03	0.00	1.00	0.43	
19	Oxyopes pingasus	0.03	0.00	0.00	0.00	1.00	0.43	
	Salticidae	0.07	0.07	0.07	0.07	0.00	1.00	
20	Unknown A	0.03	0.00	0.03	0.03	0.38	0.77	
21	Unknown B	0.03	0.07	0.03	0.03	0.37	0.77	
	Theridiidae	0.07	0.03	0.03	0.00	0.62	0.62	
22	Unknown	0.07	0.03	0.03	0.00	0.62	0.62	
	Tetragnathidae	1.33 <sup>b</sup>	1.2 <sup>ab</sup>	$1.2^{ab}$	0.63 <sup>a</sup>	4.16*	0.03	0.28
23	Tetragnatha javana	0.33	0.40	0.43	0.20	2.40	0.12	
24	Tetragnatha maxillosa	0.07	0.07	0.03	0.03	0.24	0.87	
25	Tetragnatha montana	0.07	0.00	0.03	0.03	0.37	0.77	
26	Tetragnatha virescens	0.47	0.33	0.37	0.23	0.76	0.54	
27	Tetragnatha nitens	0.00	0.03	0.03	0.03	0.38	0.77	
28	Tetragnatha okumae	0.33	0.13	0.10	0.10	1.88	0.19	
29	Tetragnatha mandibulata	0.03	0.07	0.10	0.00	0.84	0.50	
30	Tetragnatha vermiformis	0.00	0.10	0.07	0.00	1.27	0.33	
31	Tetragnatha iwahigensis	0.03	0.07	0.07	0.00	0.76	0.54	
	Theridiosomatidae	0.00	0.00	0.07	0.00	1.00	0.43	
32	<i>Wendilgarda</i> sp.	0.00	0.00	0.07	0.00	1.00	0.43	
	Total Abundance (N)	2.1 <sup>b</sup>	1.83 <sup>ab</sup>	2.3 <sup>b</sup>	$1.07^{a}$	7.29*	0.00	0.30

 Note: \*: significantly different; values within a row followed by the same letters were not significantly different at P<0.05 according to Tukey's HSD test</th>

**Table 3.** The abundance of predatory insect in rice field sprayed with bioinsecticide containing *B. bassiana*, *M. anisopliae*, and *C. militaris* and abamectin

Na	Onde / Fermile/ Species	Mean of p	redatory insect ab	undance (individ	luals/nets)	F value	P value
INO.	Ordo / Family/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	0.05	(0.05)
	COLEOPTERA	0.47	0.63	0.40	0.63	0.64	0.60
	Anthicidae	0.07	0.07	0.07	0.00	0.86	0.49
1	Formicomus sp.	0.07	0.07	0.07	0.00	0.86	0.49
	Carabidae	0.10	0.17	0.17	0.07	1.07	0.40
2	Ophionea nigrofasciata	0.10	0.17	0.17	0.07	1.07	0.40
	Coccinellidae	0.67	0.57	0.57	0.40	1.41	0.29
3	Verania discolor	0.03	0.07	0.03	0.10	0.19	0.90
4	Verania lineata	0.00	0.00	0.00	0.07	1.00	0.44
5	Menochilus sexmaculatus	0.13	0.04	0.07	0.07	0.24	0.86
6	Micrapis inops	0.50	0.47	0.47	0.17	3.14	0.07
	Staphylidae	0.27	0.30	0.33	0.27	0.36	0.78
7	Paedorus fuscipes	0.27	0.30	0.33	0.27	0.36	0.78
	HEMIPTERA	0.13	0.10	0.00	0.03	1.49	0.27
	Miridae	0.13	0.10	0.00	0.03	1.49	0.27
8	Cyrtorhinus lividipennis	0.13	0.03	0.00	0.00	2.81	0.09
9	Orthotylus sp.	0.00	0.07	0.00	0.03	0.74	0.58
	HYMENOPTERA	0.30	0.30	0.20	0.10	0.87	0.48
	Formicidae	0.30	0.30	0.20	0.00	0.87	0.48
10	Formica sp.	0.07	0.13	0.03	0.00	0.61	0.66
11	Odontoponera transversa	0.23	0.17	0.17	0.10	0.27	0.85
	ODONATA	0.33	0.20	0.20	0.20	0.34	0.80
	Coenagrionidae	0.33	0.20	0.20	0.20	0.34	0.80
12	Pyrrhosoma sp.	0.20	0.07	0.10	0.10	1.00	0.43
13	Agriocnemis pygmaea	0.20	0.07	0.10	0.10	0.10	0.96
	ORTHOPTERA	0.33	0.27	0.27	0.17	0.56	0.65
	Tettigonidae	0.33	0.27	0.27	0.17	0.56	0.65
14	Conocephalus longipennis	0.33	0.27	0.27	0.17	0.56	0.65
	Total Abundance (N)	2.20	1.97	1.80	1.23	1.25	0.34

230 Note: values within a row followed by the same letters were not significantly different at P<0.05 according to Tukey's HSD test

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Table 4. Characteristic of spider community in rice canopy sprayed with bioinsecticide containing *B. bassiana*, *M. anisopliae*, and *C. militaris* and abamectin

Characteristic of spider community	B. bassiana	M. anisopliae	C. militaris	Abamectin
14 days				
Abundance (individual/hill)	0.80	0.40	0.80	0.80
Biodiversity index (H')	1.03	0.69	1.38	1.38
Dominance index (D)	0.50	0.50	0.25	0.25
Evenness index (E)	0.94	1.00	1.00	1.00
28 days				
Abundance (individual/hill)	1.00	1.20	1.20	0.40
Biodiversity index (H')	1.33	1.24	1.79	0.69
Dominance index (D)	0.40	0.50	0.16	0.50
Evenness index (E)	0.96	0.89	1.00	1.00
42 days				
Abundance (individual/hill)	2.2	1.80	2.00	0.80
Biodiversity index (H')	1.03	1.67	1.74	1.03
Dominance index (D)	0.45	0.33	0.40	0.50
Evenness index (E)	0.94	0.93	0.89	0.94
56 days				
Abundance (individual/hill)	3.00	1.80	2.20	1.00
Biodiversity index (H')	1.70	1.30	1.12	1.33
Dominance index (D)	0.33	0.55	0.54	0.40
Evenness index (E)	0.87	0.80	0.80	0.96
70 days				
Abundance (individual/hill)	2.40	2.40	2.60	1.00
Biodiversity index (H')	1.97	1.81	2.35	1.79
Dominance index (D)	0.25	0.41	0.15	0.16
Evenness index (E)	0.95	0.87	0.98	1.00
84 days				
Abundance (individual/hill)	3.20	3.40	5.00	2.20
Biodiversity index (H')	1.53	1.39	2.07	1.66
Dominance index (D)	0.56	0.58	0.40	0.45
Evenness index (E)	0.73	0.71	0.80	0.85
TOTAL				
Abundance (individual/hill)	12.6	11	13.8	6.2
Biodiversity index (H')	2.48	2.59	2.75	2.45
Dominance index (D)	0.22	0.21	0.18	0.22
Evenness index (E)	0.82	0.83	0.85	0.88

**Table 5.** Characteristic of predatory insect community in rice field sprayed with bioinsecticide containing *B.bassiana*, *M. anisopliae*, and *C. militaris* and abamectin

Characteristic of spider community	B. bassiana	M. anisopliae	C. militaris	Abamectin
14 days old rice				
Abundance (individual/hill)	1.00	0.60	0.20	0.80
Biodiversity index (H')	1.33	1.09	0.00	0.00
Dominance index (D)	0.40	0.33	1.00	0.25
Evenness index (E)	0.96	1.00	0.00	0.00
28 days old rice				
Abundance (individual/hill)	1.60	1.20	1.40	0.80
Biodiversity index (H')	1.21	1.56	0.55	1.38
Dominance index (D)	0.25	0.33	0.28	0.25
Evenness index (E)	0.67	0.96	0.34	1.00
42 days old rice				
Abundance (individual/hill)	2.60	1.80	1.60	1.00
Biodiversity index (H')	1.94	1.88	1.66	1.66
Dominance index (D)	0.15	0.22	0.37	0.40
Evenness index (E)	0.88	0.97	0.93	1.20
56 days old rice				
Abundance (individual/hill)	2.80	2.60	2.40	1.80
Biodiversity index (H')	1.94	1.81	1.86	1.83
Dominance index (D)	0.28	0.30	0.25	0.33
Evenness index (E)	0.93	0.93	0.95	0.94
70 days old rice				
Abundance (individual/hill)	2.60	2.80	2.20	1.60
Biodiversity index (H')	1.60	1.90	1.67	1.49
Dominance index (D)	0.30	0.28	0.27	0.37
Evenness index (E)	0.89	0.91	0.93	0.92
84 days old rice				

Abundance (individual/hill)	0.20	2.80	3.00	1.40
Biodiversity index (H')	1.95	2.06	1.69	1.54
Dominance index (D)	0.23	0.21	0.33	0.28
Evenness index (E)	0.93	0.94	0.87	0.96
TOTAL				
Abundance (individual/hill)	13.2	11.8	9.8	7.4
Biodiversity index (H')	2.25	2.27	2.11	2.25
Dominance index (D)	0.91	0.89	0.88	0.94
Evenness index (E)	0.22	0.23	0.28	0.21

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Table 6. Matrix of similarity (Index Sorensen) of spider community in rice field sprayed with bioinsecticide containing *B. bassiana*, *M. anisopliae*, and *C. militaris* and abameetin

Rice ages	Treatments	B. bassiana	M. anisopliae	C. militaris.	Abamectin
	B. bassiana	1	-		
14 days	M. anisopliae	0.33	1		
14 days	C. militaris.	0.25	0.00	1	
	Insecticide	0.25	0.00	0.50	1
	B. bassiana	1			
20.1	M. anisopliae	0.18	1		
28 days	C. militaris.	0.55	0.17	1	
	Insecticide	0.29	0.25	0.25	1
	B. bassiana	1			
40.1	M. anisopliae	0.40	1		
42 days	C. militaris.	0.48	0.42	1	
	Insecticide	0.40	0.31	0.43	1
	B. bassiana	1			
561	M. anisopliae	0.58	1		
56 days	C. militaris.	0.31	0.40	1	
	Insecticide	0.40	0.43	0.50	1
	B. bassiana	1			
70.1	M. anisopliae	0.67	1		
/0 days	C. militaris.	0.32	0.64	1	
	Insecticide	0.22	0.33	0.32	1
	B. bassiana	1			
04.1	M. anisopliae	0.73	1		
84 days	C. militaris.	0.54	0.67	1	
	Insecticide	0.59	0.50	0.44	1
	B. bassiana	1			
TOTAL	M. anisopliae	0.76	1		
TOTAL	C. militaris.	0.77	0.84	1	
	Insecticide	0.49	0.60	0.56	1

**Table 7.** Similarity of predatory insect community in rice field sprayed with bioinsecticide containing *B. bassiana*, *M. anisopliae*, and *C. militaris* and abamectin

Rice ages	Treatments	B. bassiana	M. anisopliae	C. militaris.	Abamectin
	B. bassiana	1			
14.1	M. anisopliae	0.50	1		
14 days	C. militaris.	0.33	0.50	1	
	Insecticide	0.22	0.00	0.00	1
	B. bassiana	1			
20 1	M. anisopliae	0.57	1		
28 days	C. militaris.	0.53	0.31	1	
	Insecticide	0.33	0.20	0.18	1
	B. bassiana	1			
10.1	M. anisopliae	0.73	1		
42 days	C. militaris.	0.76	0.82	1	
	Insecticide	0.44	0.57	0.46	1
	B. bassiana	1			
56.1	M. anisopliae	0.89	1		
56 days	C. militaris.	0.69	0.88	1	
	Insecticide	0.70	0.64	0.76	1
	B. bassiana	1			
70.1	M. anisopliae	0.74	1		
/U days	C. militaris.	0.75	0.72	1	
	Insecticide	0.57	0.55	0.74	1
84 days	B. bassiana	1			

	M. anisopliae	0.89	1			
	C. militaris.	0.71	0.76	1		
	Insecticide	0.50	0.67	0.64	1	
	B. bassiana	1				
Total	M. anisopliae	0.91	1			
Total	C. militaris.	0.85	0.91	1		
	Insecticide	0.66	0.77	0.79	1	

# 244 Discussion

245 From the research result, it was found that population of herbivore insects in plots sprayed with the entomopathogenic 246 fungal bioinsecticide was not significantly different from population of the herbivore in abamectin plot. This because the entomopathogenic fungi applied in this research was effective and had capacity equal to the abamectin in reducing 247 248 population of herbivore insects. B. bassiana was proved to be effective in killing pest insect of rice, such as brown 249 planthopper (Lee et al. 2015) and rice bug (Girish and Balikai 2015). In this research, S. litura found was infected by B. 250 bassiana, M. anisopliae, and C. militaris. Gustianingty as et al. (2020) also reported that S. litura subjected to infection 251 by M. anisopliae. C. militaris could suppress population of lepidopteran (Shrestha et al., 2016) and coleopteran pest 252 insects (Kryukov et al. 2014)

Based on this study, symptoms developed on S. litura larvae infected by B. bassiana in rice field were similar to 253 symptom reported by Gustianingty as et al. (2019) that host insect infected by *B. bassiana* was covered by white mycelia 254 255 and its body was shriveled. Morphological characteristics of B. bassiana colony was in accordance to the result of isolation in the laboratory, white mycelia with hyaline 1-celled cylindrical and septate conidia (Safitri et al. 2018). M. 256 anisopliae infecting S. litura in this research showed symptom as reported by Humber (2012) that the integument of 257 infected insect turned to white to dark green, green to vellow conidia, 1-celled and cylindrical conidia, and septate hypha. 258 259 Larvae of S. litura infected by C. militaris in the rice field showed symptoms similar to those of B. bassiana infection, 260 and in accordance to symptoms characteristics reported by Kryukov et al. (2014) that infected insect was white in color with fruiting bodies appear from the insect, the colony was yellowish white in color, and conidia was globular in shape 261 262 (Zheng et al. 2011).

263 In this research, the abundance of hunting spider, such as species of Lycosidae, did not decrease after being sprayed with the entomopathogenic fungal bioinsecticides well as after being sprayed with abamectin. The abundance of web 264 spider, such as Bathyphantes, A. inustus and Family of Tetragnathidae decreased after application of the abamectin, while 265 application of the bioinsecticide could only decrease the population of A. inustus. Therefore, the entomopathogenic fungal 266 bioinsecticides were relatively safer than the abamectin. Furthermore, spiders generally prey on healthy herbivore insects, 267 268 while the herbivore insects infected by entomopathogen or the unhealthy ones would not be preyed (Chaubey and Yadav 269 2017). Web spider abundance decreased after application of the abamectin and the bioinsecticides because the spiders 270 were move less in rice canopy, making the insecticide as well as the bioinsecticide easy to reach the spider body. Therefore, it would better to stop application of bioinsecticide when the abundance of web spider is high. 271

272 The abundance of spider increased followed the increase of rice stage and the highest abundance occurred just before 273 harvesting. The abundance of spider continued to high until harvesting because spider likes complex habitat (Amzah et al. 274 2018) and inhabited by various insect species. Spider is also generalist predator, prey on various insect species, include 275 neutral insects which are generally abundant before harvesting (Karenina et al. 2019). The species diversity of spiders in 276 this reasearch was also higher in plots sprayed with the entomopathogenic fungal bioinsecticide compared to that of the 277 abamectin plot. The higher abundance and species diversity of spiders indicated that the entomopathogenic fungal bioinsecticide was not dangerous to their life. Many reports showed that entomopathogenic fungi did not kill spiders, 278 especially hunting spiders (Prabawati et al. 2019; Hanif et al. 2020). 279

The abundance of predatory insects did not decrease after the application of the entomopathogenic fungal bioinsecticide or abamectin. Predatory insect has high mobility which differ them from web spiders, so that predatory insect could escape from being exposed to bioinsecticide and abamectin. Furthermore, there is evident that the avoidance behavior in predatory insects from *B. bassiana*-infected insect preys (Seiedy et al. 2015). Therefore, application of entomopathogenic fungi was relatively safe for predatory insects.

The abundance of predatory insect was increasing until 70 DAT, and was decreased at 84 DAT. The predatory insects were generally more specialist in term of prey species compared to spiders. Therefore, at approaching to harvest when the number of herbivore insects decreased, the predatory insects also decreased. Such phenomenon frequently happened in specialist predatory arthropod because population of the predator depended on the fluctuated population of their preys which are called as functional response (Karenina et al. 2019).

The community of spider and predatory insects in plot of *B. bassiana* tent to more similar to the community in plots of bioinsectiside *M. anisopliae* and *C. militaris* compared to that in the abamectin plot. This showed that the effect of application of three entomopathogenic fungal bioinsecticide on the predator communities was not significantly different. The low community similarity between the bioinsecticide and the abamectin because the abamectin could reduce the abundance of spiders and predatory insects. There have been a lot of reports of the decrease of abundance and species diversity of spiders (Preetha et al. 2010) and predatory insects (Salachna et al. 2020) due to application of abamectin containing organophosphate (Bai and Ogbourne 2016). In this research, abamectin as active ingredient is a stomach 297 poison and toxic against insect species of *Menochilus sexmaculatus* (Azod et al. 2016) and spider species of P. 298 pseudoannulata (Baehaki et al. 2017).

299 Guild group made based on function corespondence of arthropods i.e. herbivore insects, neural insects, predators and 300 parasitoids. For one rice cropping season, there was a tendency that the abundance of all guilds group were lower in the abamectin plot. Interestingly, the abundance of predator was always more dominant in plots sprayed with the 301 entomopathogenic fungal bioinsecticide compared to those of herbivore insects, neutral insects, and parasitoid. Neutral 302 303 insects were not found in the synhetic insecticie plot. Thus phenomenon showed that predator guild was more tolerant to 304 the entomopathogenic fungal bioinsecticide (Bavissa et al. 2016; Firouzbakht et al. 2015; Gholamzadeh-Chitgar et al. 305 2017), while abamectin tent to reduce the abundance of predators, herbivore insects, neutral insects and parasitoid 306 (Prabawati et al. 2019; Hanif et al. 2020). Among the three entompathogenic fungi, there was a tendency that the 307 abundance of the predators in plot sprayed with the bioinsecticide of C. militaris was higher than that in plots of B. 308 bassiana and M. anisopliae. Therefore, predator arthropods tent to be more tolerant to C. militaris than to B. bassiana 309 and *M. anisopliae*.

310 It could be concluded that the abundance and diversity of arthropod predator species inhabiting freshwater swamp 311 sprayed with the bioinsecticides of B. bassiana, M. anisopliae, and C. militaris did not decrease, while the population of 312 herbivore insects tent to decrease to the same level as its decrease in the abametin plot. So that, the bioinsecticide of B. 313 bassiana, M. anisopliae, and C. militaris did not decrease the abundance and species diversity of predatory arthropod 314 (non-target arthropods), but could decrease the herbivore insect population.

#### 315

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# Herbivore insects and predatory arthropods in freshwater swamp rice field in South Sumatra sprayed with bioinsecticides of entomopathogenic fungi and abamectin

**Abstract.** Herbivore insect population and predatory arthropod in rice field may be effected by the application entomopathogenic fungi or synthetic insectide. The objective of this research was to analyze individual quantity of herbivore insects and predatory arthropod inhabiting freshwater swamp rice field treated with bioinsecticides and abamectin (commercial insecticide). The research used the bioinsecticides made from fungi (*Beauveria bassiana, Metarhizium anisopliae, Cordyceps military*), and the abamectin. This research found 12 families of herbivore insects with 22 species dominated by *Nilaparvata lugens* and *Leptocorisa acuta*. This study found 32 species of spider belonged to 8 families dominated by *Tetragnatha virescens* and *Oxyopes matiensis*. The species diversity of spider was higher in the plots of the bioinsecticide compared to that of the abamectin. Predatory insects found belonged to 14 species belonged to 8 families dominated by *species of Ophionea nigrofasciata, Verania discolor*, and *Paedorus fuscipe*. The abundance of predatory insects in plots sprayed with the bioinsecticides was higher compared to that of the abamectin plot. It concluded that the abundance and species diversity of predatory arthropod sprayed with bioinsecticide of *B. bassiana*, *M. anisopliae*, and *C. militaris* did not decrease, while the population of herbivore insect tent to decrease.

Key words: Beauveria bassiana; Metarhizium anisopliae; Cordyceps militaris; neutral insect; parasitoid

Abbreviations (if any): -

Running title: Arthropods in freshwater swamp rice field

#### **INTRODUCTION**

Rice cultivation in freshwater swamps in the South Sumatra region has specific characteristics making it different from rice cultivation in other ecosystem in Indonesia (reff...?) what characters you mean in this sentence?. Rice cultivation is affected by climate conditions, especially rainfall and tidal flush (reff...). In the period of November to April, freshwater swamps are generally inundated so that local farmers cultivate rice only in the period of May to September (Herlinda et al. 2018; Herlinda et al. 2019a). Due to difficulties in managing water, farmers grow rice once a year using the transplanting method. In maintaining their rice cultivations, the local farmers scarcely apply synthetic insecticide (Herlinda et al. 2019b).

The population of insect pests attacking freshwater swamp rice plants is increasing and is starting to result in yield loss, especially rice bug (*Leptocorixa acuta*), while brown planthoppers (*Nilavarpata lugens*) that have not appeared for a long time start attacking (Hanif et al. 2020). Pests that attack rice plants have been controlled using various entomopathogenic fungi including *Beauveria bassiana* (Li et al. 2012; Li et al. 2014; Lee et al. 2015), *Metarizhium anisopliae* (Girish and Balikai, 2015; Chinniah et al. 2016) and *Cordycep militaris* (Prabawati et al. 2019). *C. militaris* is reported to produce beauvericin that are toxic to insects (Rachmawati et al. 2018) and *B. bassiana* has been reported to be effective against homopteran (Li et al. 2012; Li et al. 2014; Lopez et al. 2014; Lee et al. 2015; Sumikarsih et al. 2019), coleopteran (Kavallieratos et al. 2015), hemipteran (Girish and Balikai 2015), and lepidopteran insects (Ayudya et al. 2019; Ma et al. 2019; Gustianingtyas et al. 2020). *M. anisopliae* has also been demonstrated to be effective against homopteran (Mweke et al. 2019), lepidopteran (Gustianingtyas et al. 2020), and coleopteran insects (Kavallieratos et al. 2015). *M. anisopliae* could be used in multiple roles, ranging from controlling insect pests and promoting plant growth (Liu et al. 2017).

Being effective against various orders of insects, including the coleopteran insects (Kavallieratos et al. 2015), the rice field ecosystem which is dominated by species of predatory insects, it is important to be very careful in the application of entomopathogenic fungi in ecosystem to avoid negative impact on non-target arthropods (Scorsetti et al. 2017). High species diversity and abundance of arthropod in freshwater swamp rice field (Herlinda et al. 2018) might be disturbed by the application of the entomopathogenic fungi. However, there have been some reports suggested that entomopathogenic fungi did not affect abundance and species diversity of arthropods...references..?. For example, predatory insect in paddy field (*Andrallus spinidens*) was reported to be resistant to *B. bassiana* (Firouzbakht et al. 2015; Gholamzadeh-Chitgar et al. 2017; Scorsetti et al. 2017). Parasitoid tends to avoid host insect infected by entomopathogenic fungi (Emami *et al.* 2013). Coccinellidae, the family of generalist predatory insect, was reported to be resistant to *M. anisopliae* (Bayissa et al. 2016). This research aimed to analyze individual quantity of herbivore insects and predatory arthropod inhabiting freshwater swamp rice field treated with bioinsecticides contaning *Beauveria bassiana*, *Metarhizium anisopliae*, and *Cordyceps militaris* and the abamectin.

#### MATERIALS AND METHODS

#### Study area

The research was conducted from April to August 2018 in freshwater swamp rice field located in Village Pelabuhan Dalam, Sub District Pemulutan, District Ogan Ilir, and South Sumatra. The experiment was arranged in a Completely Randomized Block Design consisted of four plots of treatment. The first three plots were treated with bioinsecticide containing *B. bassiana*, *M. anisopliae*, *C. military*, and the forth plot was treated with the abamectin (the commercial insecticide) as control. The plot area was 120 m<sup>2</sup> for a treatment and each treatment was replicated five times. Distance between plots....?, the distance will give an overview that your treatment on each plot does not affect other plots. Please explain why you do not provide plots without abamectin and bioinsecticides for control?

#### Land preparation, transplanting, and crop maintenance

Land preparation was conducted using moldboard plow and was continued by using harrow plow. Before being plowed, the land was cleared using hoe and long knife. The excessive water was pumped out until the soil was slightly watery, and dung compost was added at a dosage of 1 ton ha<sup>-1</sup> Rice seed used were certified purple labeled seed of Mekongga variety at a dosage of 50 kg/ha. Rice was cultivated using transplanting system from which, the seedlings were prepared in 34 x 26 x 5 cm<sup>3</sup> plastic trays filled with 3 cm high soil. The seeds were soaked in water for 24 hours before being planted in the prepared plastic trays. The soaked seeds were spread evenly in each tray at a dosage of 150 g per tray. The trays were then covered with thick plastic for 7 days. The 7 day old seedlings were then transferred to places receiving enough sun light. Fourteen day old seedlings were transplanted to all prepared plots at a density of 2 seedlings per hole in a 2:1 (12.5 x 25 x 50 cm<sup>3</sup>) "jajar legowo" planting system (Figure 1).



Figure 1. Two day old rice seedling in a plastic tray (A), 7 day old seedlings (B), rice transplanting (D) rice planted in "jajar legowo" planting system (E)

Crop maintenance was conducted in the form of replanting, irrigating, weeding, and fertilizing. Replanting was conducted 7 days after transplanting to replace the dead seedlings. Irrigating was conducted by making use of water flush from river under high tide and pumping system. Weeding was conducted both at vegetative and generative stages using sickle. Fertilizer used was shrimp shell extract compost prepared according to method by Suwandi et al. (2012). The compost was applied every two weeks started from 14 days after planting until harvesting (84 days) at a dosage of 2 L ha<sup>-1</sup>. The extract compost was a fermentation result of shrimp shell meal containing chitinolitic bacteria, selulotic bacteria and sulphate diluter. Nitrogen fertilizer was applied 30 days after planting at a dosage of 100 kg ha<sup>-1</sup>.

# Formulation and application of bioinsecticides

All species of fungi have been explored from soil according to Kin et al. (2017). *B. bassiana* and *M. anisopliae* used in this research were explored from freshwater swamp soil of South Sumatra, while *C. militaris* was explored from soil in Central Kalimantan. *B. bassiana*, *M. anisopliae*, and *C. militaris* were cultured in solid media of Sabouraud Dextrose Agar (SDA, Merck) made from 16.2 g SDA in 250 mL aquadest. The cultures were incubated for 10 x 24 hours. The 10 days old fungal solid cultures were then cultured in liquid media (Figure 2) of Sabouraud Dextrose Broth (SDB, *Merck*) made from 30 g SDB in 1000 ml aquadest and were incubated in a 120 rpm shaker for 7 x 24 hours. The liquid culture of the entomopathogenic fungi were used to make bioinsecticide. Seven days after incubation, the density of fungal spores of the bioinsecticide was made by adding shrimp shell meal extract compost as carrier, while active ingredients used were entomopathogenic fungi and vegetable oil. One liter of bioinsecticide comprised of 400 mL shrimp shell meal extract compost, 600 mL liquid culture of entomopathogenic fungi and 10 mL vegetable oil. Compost extract used to make bioinsecticide was previously autoclaved under 1 atm for 2 hours.



**Figure 2.** Cultures of *Beauveria bassiana* (A), *Metarhizium anisopliae* (B), and *Cordycep militaris* (C) on SDB; liquid bioinsecticide of *Beauveria bassiana* (D), *Metarhizium anisopliae* (E), and *Cordycep militaris* (F)

The bioinsecticides were applied at dosage of 2 L ha<sup>-1</sup> per application. The applications were conducted at 13, 27, 41, 55, 69, and 83 days after transplanting (DAT). The bioinsecticide applications were conducted in the evening (at 5 pm) to avoid spores damage by ultra violet. Control plot was sprayed with abamectin at a dosage of 0.5 L ha<sup>-1</sup>. The insecticide contained abamectin active ingredient and was sprayed at the same time as bioinsecticide applications. The control plot was 50 m away from treatments plots to avoid contamination in the treatment plots, and there was 10 m distance between bioinsecticide treatment plots. A day after application the bioinsecticides and the abamectin, arthropod samplings were carried out to collect arthropods inhabiting rice canopy.

#### Arthropod sampling in rice canopy

Sampling to collect arthropods inhabiting rice canopy was conducted using an entomological net according to Lami et al. (2016) and Ivantsova et al. (2017). The net has 75 cm length and 30 cm diameter of net and 100 cm length of handle. Samplings were conducted in every treatment subplot and was repeated five times. Samplings were conducted at 14, 28, 42, 56, 70, and 84 DAT. Arthropod samplings were conducted in the morning, at 6-7 am. During sampling, the sweep net

was swept 30 cm depth into rice canopy to collect representative samples.

Sampled arthropods were cleaned and placed in plastic bottles (330 mL) containing 100 mL absolute ethanol, the bottles were labeled. The samples were brought into the Laboratory of Entomology, Department of Plant Pest and Disease, Faculty of Agriculture, Universitas Sriwijaya for identification. Spider identification was conducted using reference of Barrion and Litsinger (1995) while for insect identification we used reference of Gullan and Cranston (2014) and Heinrichs et al. (2017).

#### Data analysis

Data of arthropods species were grouped based on their guild, comprised of herbivore insects, predators, parasitoids, and neutral insects, and was presented in the form of graphic. The difference of individual quantity of the herbivore insects, spiders, and predatory insects amongst treatments was analyzed using Analysis of Variance (ANOVA). If there was significant difference among treatments, the analyses was continued with Tukey's HSD (Honesty Significant Difference) test at 5% degree of significance. Analyses data was conducted using Software of SAS University Edition 2.7 9.4 M5. Data of abundance were used to analyze species diversity by using Shannon index (H'). Degree of diversity was counted using the Evenness index (J') derived from the Shannon function, and Berger-Parker dominance biodiversity indices. The coefficient of Sorensen was counted to measure degree of similarity of spider or predatory insect among treatments (Magurran 2004).

### **RESULTS AND DISCUSSION**

#### Abundance of herbivore insects in one rice cropping season

In one rice cropping season, it was found 22 species of herbivore insects belonging to 12 families. Dominant herbivore species were *Nilaparvata lugens, Leptocorisa acuta, Acrida turita, Valanga nigricornis, Dicladispa armigera* (Figure 3). Population of the herbivore was not significantly different among treatments, except for Family Cecidomyiidae (Table 1). Therefore, the capacity of bioinsecticide in suppressing the population of the herbivore insects has generally almost similar to the capacity of the abamectin.



**Figure 3.** Herbivore insects found in rice canopy: *Spodoptera* sp. (A), *Pelopidas* sp. (B), *Amata nigriceps* (C), *Oxya chinensis* (D), *Scirpophaga* sp. (E), *Dicladispa armigera* (F), *Chrysolina coerulans* (G), *Acrida turita* (H), *Acrida turita* (I), *Nilaparvata lugens* (J)

**Table 1.** Population of herbivore insects in rice canopy sprayed with bioinsecticide containing *B. bassiana, M. anisopliae* and *C. militaris* and abamectin

Ordo / Family/ Spacing	Mean o	Mean of herbivore abundance (individuals/nets)					Tukey's HSD
Oruo / Fanny/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	r-value	(0.05)	test
COLEOPTERA	0.2	0.13	0.17	0.23	0.26	0.85	
Chrysomelidae	0.2	0.13	0.17	0.23	0.26	0.85	
Chrysolina coerulans	0.07	0	0.07	0.2	2.11	0.15	
Dicladispa armigera	0.13	0.13	0.1	0.03	0.53	0.67	
DIPTERA	$0.4^{b}$	0.03 <sup>ab</sup>	$0.07^{ab}$	$0.00^{a}$	4.40*	0.03	0.20
Cecidomyiidae	$0.4^{b}$	0.03 <sup>ab</sup>	$0.07^{ab}$	$0.00^{a}$	4.40*	0.03	0.20
Unknown sp.	0.33	0	0	0	2.55	0.1	
Orseolia sp.	0.07	0.03	0.07	0	0.81	0.51	
HEMIPTERA	1.67	1.1	1.7	0.97	2.42	0.12	
Alydidae	0.57	0.4	0.73	0.5	2.02	0.17	
Leptocorisa acuta	0.57	0.4	0.73	0.5	2.02	0.17	

Cicadellidae	0.17	0.1	0	0.1	0.42	0.74
Nephotettix cincticeps	0	0	0	0.03	1.00	0.43
Nephotettix virescen	0	0.1	0	0.07	1.04	0.41
Recilia dorsalis	0.17	0	0	0	1.00	0.43
Delphacidae	0.93	0.6	0.97	0.37	2.59	0.1
Nilaparvata lugens	0.77	0.53	0.9	0.27	2.23	0.14
Sogatella furcifera	0.17	0.07	0.07	0.1	0.50	0.69
LEPIDOPTERA	0.37	0.3	0.5	0.17	2.61	0.1
Pyralidae	0.33	0.23	0.33	0.17	0.99	0.43
Cnaplocrosis medinalis	0	0	0	0.03	1.00	0.43
Scirpophaga incertulas	0.17	0.13	0.17	0.07	1.31	0.32
Scirpophaga sp.	0.17	0.1	0.17	0.07	0.66	0.59
Erebidae	0	0	0.07	0	1.00	0.43
Amata nigriceps	0	0	0.07	0	1.00	0.43
Hesperiidae	0	0	0.03	0	1.00	0.43
Pelopidas mathias	0	0	0.03	0	1.00	0.43
Noctudae	0.03	0.07	0.07	0	0.47	0.71
Spodoptera sp.	0.03	0.07	0.07	0	0.47	0.71
ORTHOPTERA	0.63	0.83	0.67	0.8	0.58	0.64
Acrididae	0.63	0.83	0.67	0.77	0.48	0.7
Acrida turrita	0.2	0.17	0.1	0.4	2.68	0.09
Gesonula mundata	0.03	0.1	0.03	0.1	0.35	0.79
Oxya chinensis	0.07	0.1	0.1	0.1	0.07	0.97
Valanga nigricornis	0.33	0.47	0.43	0.17	2.02	0.17
Pyrgomorphidae	0	0	0	0.03	1.00	0.43
Atractomorpha crenulata	0	0	0	0.03	1.00	0.43
THYSANOPTERA	0.1	0.1	0.1	0.07	0.11	0.95
Thripidae	0.1	0.1	0.1	0.07	0.11	0.95
Liothrips sp.	0.1	0.1	0.1	0.07	0.11	0.95
TOTAL	3.37	2.5	3.2	2.23	2.77	0.09

Note: \*: significantly different; values within a row followed by the same letters were not significantly different at P<0.05 according to Tukey's HSD test

During field observations, insect died due to infection by entomopathogenic fungi were documented and showed specific symptoms of each fungus. Larva of *S. litura* found were infected by *B. bassiana* in the rice field showed white mycelia covering its body, dried and not smelly (Figure 4). Larvae of *S. litura* infected by *M. anisopliae* showed symptoms of dried body covered by greenish white mycelia and was not smelly, while larvae of *S. litura* infected by *C. militaris* showed similar symptoms to those of *B. bassiana* infection, larval body drying, shriveled, covered by white mycelia, and not smelly. Entomopathogenic fungi infected sampled insects were isolated and purified in the laboratory. Color, structure, and spore of the isolated fungi were presented in Figure 5.







**Figure 5.** Spores of *Beauveria bassiana* (A), *Metarhizium anisopliae* (B), and *Cordycep militaris* (C); colonies of *Beauveria bassiana* (d), *Metarhizium anisopliae* (E), and *Cordycep militaris* (F)

#### Abundance of spider and predatory insect in one rice cropping season

This research found 32 species of spider belonged to 8 families. The dominant species of them were *Tetragnatha virescens*, *T. virescens*, *T. maxilosa*, *Argiope catenulata*, and *Oxyopes matiensis* (Figure 6). Total abundance of spider in the plot treated with the abamectin was significantly lower than that of plots treated with bioinsecticide (Table 2). The abundance of *Bathyphantes* sp. in abamectin plot decreased. The highest abundance of *Araneus inustus* was found in plot treated with bioinsecticide *C. militaris* and was different from other treatments. The abundance of Tetragnathidae in plots treated with insecticide was found to be lower compared to plots treated with bioinsecticide.

Predatory insects found in this research belonged to 14 species and 8 families. The dominant species were *Formicomus* sp., *Ophionea nigrofasciata, Verania discolor, Verania lineata, Menochilus sexmaculatus, Micrapis inop, Paedorus fuscipe, Cyrtorhinus lividipennis, and Orthotylus sp.* (Figure 7). The abundance of predatory insects was not significantly different among treatments (Table 3).

#### Characteristic of spider and predatory insect community in one rice cropping season

Number of spiders decreased with increasing age of rice (Table 4). The highest number of spiders occurred when the rice were at mature grain ripening stage (84 DAT). The abundance of spider in the plot treated with abamectin tent to be lower compared to that of plots treated with the bioinsecticides. Total number of spiders in one rice cropping season were higher in the plots treated with the bioinsecticide than that in plot treated with the abamectin. The species diversity of spiders was also higher in the plots treated with entomopathogenic fungal bioinsecticide was also followed by the high species diversity in plots treated with entomopathogenic fungal bioinsecticide was also followed by the high species evenness and low species dominance.

Number of predatory insects decreased with increasing stages of rice, but at approaching to harvest (84 DAT), the number of predatory insects drastically decreased (Table 5). The highest abundance of predatory insect occurred at milk grain ripening stage (70 DAT). The phenomenon of abundance fluctuation of predatory insect was slightly different from that of spider which its peak occurred at mature grain ripening stage (84 DAT). The abundance of predatory insect in plots treated with abamectin was generally lower than that of plots treated with the entomopathogenic fungal bioinsecticide. Total of abundance of predatory insects in one rice cropping season of plots sprayed with entomopathogenic fungal bioinsecticide was higher compared to that of abamectin plot, however, species diversity, evenness and species dominance tent to be similar among treatments.

Spider community in plot sprayed with the bioinsecticide of *B. bassiana* tent to be more similar to that of plots sprayed with bioinsecticide (*M. anisopliae* and *C. militaris*) than that of abamectin plot. Total of one rice cropping season, spider community in plot sprayed with bioinsecticide *B. bassiana* was more similar to that of plot of *C. militaris* (0.77), followed by plot *M. anisopliae* (0.76), and least similar to that of abamectin plot (0.49) (Table 6).

Predatory insect community in plot sprayed with bioinsecticide *B. bassiana* tent to be more similar to that of plots sprayed with bioinsecticide *M. anisopliae* and *C. militaris* than that of abamectin plot (Table 7). Total of one rice cropping season, predatory insect community in plots sprayed with bioinsecticide *B. bassiana* was more similar to that of plot *M. anisopliae* (0.91), followed by plot of *C. militaris* (0.85), and least similar to that of insecticide plot (0.66).



**Figure 6.** Spiders found in rice canopy: Linyphildae (A), *Tetragnatha virescens* (B), *Tetragnatha maxilosa* (C), *Argiope catenulate* (D), *Oxyopes matiensis* (E), Lycosidae (F), Salticidae A (G), Salticidae b (H), *Oxyopes javanus* (I), *Pardosa* sp. (J)



**Figure 7.** Predatory insect in rice field: Verania lineata (A), Menochilus sexmaculatus (B), Micrapis inops (C), Formica sp. (D), Odontoponera transversa (E), Argia sp. (F), Agriocnemis sp. (G), Ophionea nigrofasciata (H), Paederus fuscipes (I), and Conocephalus longipennis (J)

# The abundance of arthropod guilds in one rice cropping season the utilization of guild is not appropriate. You should use another term such as associated arthropod thar refer to associated arthropod with rice.

The abundance of associated arthropods tent to increase with increasing rice growth stage (Figure 8). In all observations, the abundance of predatory insect was found to be higher compared to the abundance of other guilds (neutral insect, herbivore, and parasitoid). The abundance of predator was generally higher in plots sprayed entomopathogenic fungal bioinsecticide compared to that of abamectin plot. The abundance of all guilds of arthropod in plot of abamectin was always lower compared to that of bioinsecticide plots. When rice was 70 days old, the abundance of herbivore reached its peak, then decreased at mature grain stage. The abundance of predator reached its peak when rice was 84 days old.





Figure 8. Arthropod abundance on rice field treated with bioinsectides and abamectin (insecticide)

**Table 2.** The abundance of spider inhabiting rice canopy sprayed with bioinsecticide containing *B.bassiana*, *M. anisopliae*, and *C. militaris* and abamectin

Orde / Ferrile/ Section	Mean	of spider abunda	nce (individua	ls/nets)	El	P value	Telessia HCD 4ast
Ordo / Family/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	F-value	(0.05)	Tukey's HSD test
ARACHINIDA							
Araneidae	0.03	0.07	0.27	0.13	2.02	0.17	
Araneus inustus	$0.00^{a}$	0.03 <sup>ab</sup>	$0.2^{b}$	0.03 <sup>ab</sup>	4.82*	0.02	0.11
Unknown	0.03	0.03	0.07	0.07	0.35	0.79	
Gea subarmata	0.00	0.00	0.00	0.03	1.00	0.43	
Linyphiidae	0.47	0.30	0.20	0.10	2.76	0.09	
Bathyphantes tagalogensis	0.00	0.00	0.07	0.00	2.67	0.10	
Bathyphantes sp. A	0.07	0.00	0.00	0.07	0.89	0.48	
Bathyphantes sp. B	0.13 <sup>ab</sup>	$0.2^{b}$	$0.07^{\mathrm{ab}}$	$0^{\mathrm{a}}$	3.74*	0.04	0.12
Atypena adelinae	0.03	0.03	0.03	0.03	0.00	1.00	
Erigone bifurca	0.03	0.00	0.00	0.00	1.00	0.43	
Linyphiidae Sp. A	0.20	0.03	0.03	0.00	2.46	0.11	
Atypena formosana	0.00	0.03	0.00	0.00	0.98	0.44	
Lycosidae	0.10	0.13	0.37	0.10	1.90	0.18	
Arctosa sp.	0.00	0.03	0.03	0.00	0.60	0.63	
Pardosa birmanica	0.03	0.00	0.00	0.00	1.00	0.43	
Pardosa pseudoannulata	0.03	0.00	0.27	0.07	2.10	0.15	
Pardosa apostoli	0.00	0.03	0.00	0.00	1.00	0.43	
Pardosa pullata	0.03	0.03	0.03	0.00	0.37	0.77	
Hogna rizali	0.00	0.00	0.03	0.00	1.00	0.43	
Pirata luzonensis	0.00	0.03	0.00	0.03	0.62	0.62	
Oxyopidae	0.03	0.03	0.10	0.03	0.51	0.68	
Oxyopes javanus	0.00	0.03	0.07	0.03	0.76	0.54	
Oxyopes matiensis	0.00	0.00	0.03	0.00	1.00	0.43	
Oxyopes pingasus	0.03	0.00	0.00	0.00	1.00	0.43	
Salticidae	0.07	0.07	0.07	0.07	0.00	1.00	
Unknown A	0.03	0.00	0.03	0.03	0.38	0.77	
Unknown B	0.03	0.07	0.03	0.03	0.37	0.77	
Theridiidae	0.07	0.03	0.03	0.00	0.62	0.62	
Unknown	0.07	0.03	0.03	0.00	0.62	0.62	
Tetragnathidae	1.33 <sup>b</sup>	$1.2^{ab}$	$1.2^{ab}$	0.63 <sup>a</sup>	4.16*	0.03	0.28
Tetragnatha javana	0.33	0.40	0.43	0.20	2.40	0.12	
Tetragnatha maxillosa	0.07	0.07	0.03	0.03	0.24	0.87	
Tetragnatha montana	0.07	0.00	0.03	0.03	0.37	0.77	
Tetragnatha virescens	0.47	0.33	0.37	0.23	0.76	0.54	
Tetragnatha nitens	0.00	0.03	0.03	0.03	0.38	0.77	
Tetragnatha okumae	0.33	0.13	0.10	0.10	1.88	0.19	
Tetragnatha mandibulata	0.03	0.07	0.10	0.00	0.84	0.50	
Tetragnatha vermiformis	0.00	0.10	0.07	0.00	1.27	0.33	
Tetragnatha iwahigensis	0.03	0.07	0.07	0.00	0.76	0.54	
Theridiosomatidae	0.00	0.00	0.07	0.00	1.00	0.43	
Wendilgarda sp.	0.00	0.00	0.07	0.00	1.00	0.43	
Total Abundance (N)	2.1 <sup>b</sup>	1.83 <sup>ab</sup>	2.3 <sup>b</sup>	1.07 <sup>a</sup>	7.29*	0.00	0.30

Note: \*: significantly different; values within a row followed by the same letters were not significantly different at P<0.05 according to Tukey's HSD test

Table 3.	The abundance	of predatory	insect in	rice field	d sprayed	l with	bioinsecticide	containing	В.	bassiana,	М.	anisopliae,	and	С.
militaris a	and abamectin													

Orde / Family/Sussian	Mean of p	redatory insect ab	undance (individ	luals/nets)	F value	$\mathbf{D}$ and $\mathbf{D} = (0, 0.5)$
Ordo / Family/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	0.05	P value (0.05)
COLEOPTERA	0.47	0.63	0.40	0.63	0.64	0.60
Anthicidae	0.07	0.07	0.07	0.00	0.86	0.49
Formicomus sp.	0.07	0.07	0.07	0.00	0.86	0.49
Carabidae	0.10	0.17	0.17	0.07	1.07	0.40
Ophionea nigrofasciata	0.10	0.17	0.17	0.07	1.07	0.40
Coccinellidae	0.67	0.57	0.57	0.40	1.41	0.29
Verania discolor	0.03	0.07	0.03	0.10	0.19	0.90
Verania lineata	0.00	0.00	0.00	0.07	1.00	0.44
Menochilus sexmaculatus	0.13	0.04	0.07	0.07	0.24	0.86
Micrapis inops	0.50	0.47	0.47	0.17	3.14	0.07
Staphylidae	0.27	0.30	0.33	0.27	0.36	0.78
Paedorus fuscipes	0.27	0.30	0.33	0.27	0.36	0.78
HEMIPTERA	0.13	0.10	0.00	0.03	1.49	0.27
Miridae	0.13	0.10	0.00	0.03	1.49	0.27
Cyrtorhinus lividipennis	0.13	0.03	0.00	0.00	2.81	0.09
Orthotylus sp.	0.00	0.07	0.00	0.03	0.74	0.58
HYMENOPTERA	0.30	0.30	0.20	0.10	0.87	0.48
Formicidae	0.30	0.30	0.20	0.00	0.87	0.48
Formica sp.	0.07	0.13	0.03	0.00	0.61	0.66
Odontoponera transversa	0.23	0.17	0.17	0.10	0.27	0.85
ODONATA	0.33	0.20	0.20	0.20	0.34	0.80
Coenagrionidae	0.33	0.20	0.20	0.20	0.34	0.80
<i>Pyrrhosoma</i> sp.	0.20	0.07	0.10	0.10	1.00	0.43
Agriocnemis pygmaea	0.20	0.07	0.10	0.10	0.10	0.96
ORTHOPTERA	0.33	0.27	0.27	0.17	0.56	0.65
Tettigonidae	0.33	0.27	0.27	0.17	0.56	0.65
Conocephalus longipennis	0.33	0.27	0.27	0.17	0.56	0.65
Total Abundance (N)	2.20	1.97	1.80	1.23	1.25	0.34

Note: values within a row followed by the same letters were not significantly different at P<0.05 according to Tukey's HSD test

**Table 4.** Characteristic of spider community in rice canopy sprayed with bioinsecticide containing *B. bassiana*, *M. anisopliae*, and *C. militaris* and abamectin

Characteristic of spider community	B. bassiana	M. anisopliae	C. militaris	Abamectin
14 days				
Abundance (individual/hill)	0.80	0.40	0.80	0.80
Biodiversity index (H')	1.03	0.69	1.38	1.38
Dominance index (D)	0.50	0.50	0.25	0.25
Evenness index (E)	0.94	1.00	1.00	1.00
28 days				
Abundance (individual/hill)	1.00	1.20	1.20	0.40
Biodiversity index (H')	1.33	1.24	1.79	0.69
Dominance index (D)	0.40	0.50	0.16	0.50
Evenness index (E)	0.96	0.89	1.00	1.00
42 days				
Abundance (individual/hill)	2.2	1.80	2.00	0.80
Biodiversity index (H')	1.03	1.67	1.74	1.03
Dominance index (D)	0.45	0.33	0.40	0.50
Evenness index (E)	0.94	0.93	0.89	0.94
56 days				
Abundance (individual/hill)	3.00	1.80	2.20	1.00
Biodiversity index (H')	1.70	1.30	1.12	1.33
Dominance index (D)	0.33	0.55	0.54	0.40
Evenness index (E)	0.87	0.80	0.80	0.96
70 days				
Abundance (individual/hill)	2.40	2.40	2.60	1.00
Biodiversity index (H')	1.97	1.81	2.35	1.79
Dominance index (D)	0.25	0.41	0.15	0.16
Evenness index (E)	0.95	0.87	0.98	1.00
84 days				
Abundance (individual/hill)	3.20	3.40	5.00	2.20

Biodiversity index (H')	1.53	1.39	2.07	1.66	
Dominance index (D)	0.56	0.58	0.40	0.45	
Evenness index (E)	0.73	0.71	0.80	0.85	
TOTAL					
Abundance (individual/hill)	12.6	11	13.8	6.2	
Biodiversity index (H')	2.48	2.59	2.75	2.45	
Dominance index (D)	0.22	0.21	0.18	0.22	
Evenness index (E)	0.82	0.83	0.85	0.88	

**Table 5.** Characteristic of predatory insect community in rice field sprayed with bioinsecticide containing *B.bassiana*, *M. anisopliae*, and *C. militaris* and abamectin

Characteristic of spider community	B. bassiana	M. anisopliae	C. militaris	Abamectin
14 days old rice				
Abundance (individual/hill)	1.00	0.60	0.20	0.80
Biodiversity index (H')	1.33	1.09	0.00	0.00
Dominance index (D)	0.40	0.33	1.00	0.25
Evenness index (E)	0.96	1.00	0.00	0.00
28 days old rice				
Abundance (individual/hill)	1.60	1.20	1.40	0.80
Biodiversity index (H')	1.21	1.56	0.55	1.38
Dominance index (D)	0.25	0.33	0.28	0.25
Evenness index (E)	0.67	0.96	0.34	1.00
42 days old rice				
Abundance (individual/hill)	2.60	1.80	1.60	1.00
Biodiversity index (H')	1.94	1.88	1.66	1.66
Dominance index (D)	0.15	0.22	0.37	0.40
Evenness index (E)	0.88	0.97	0.93	1.20
56 days old rice				
Abundance (individual/hill)	2.80	2.60	2.40	1.80
Biodiversity index (H')	1.94	1.81	1.86	1.83
Dominance index (D)	0.28	0.30	0.25	0.33
Evenness index (E)	0.93	0.93	0.95	0.94
70 days old rice				
Abundance (individual/hill)	2.60	2.80	2.20	1.60
Biodiversity index (H')	1.60	1.90	1.67	1.49
Dominance index (D)	0.30	0.28	0.27	0.37
Evenness index (E)	0.89	0.91	0.93	0.92
84 days old rice				
Abundance (individual/hill)	0.20	2.80	3.00	1.40
Biodiversity index (H')	1.95	2.06	1.69	1.54
Dominance index (D)	0.23	0.21	0.33	0.28
Evenness index (E)	0.93	0.94	0.87	0.96
TOTAL				
Abundance (individual/hill)	13.2	11.8	9.8	7.4
Biodiversity index (H')	2.25	2.27	2.11	2.25
Dominance index (D)	0.91	0.89	0.88	0.94
Evenness index (E)	0.22	0.23	0.28	0.21

<b>Rice ages</b>	Treatments	B. bassiana	M. anisopliae	C. militaris.	Abamectin
	B. bassiana	1			
14 days	M. anisopliae	0.33	1		
14 days	C. militaris.	0.25	0.00	1	
	Insecticide	0.25	0.00	0.50	1
	B. bassiana	1			
20 1	M. anisopliae	0.18	1		
28 days	C. militaris.	0.55	0.17	1	
	Insecticide	0.29	0.25	0.25	1
	B. bassiana	1			
42 Jan	M. anisopliae	0.40	1		
42 days	C. militaris.	0.48	0.42	1	
	Insecticide	0.40	0.31	0.43	1
	B. bassiana	1			
FC James	M. anisopliae	0.58	1		
56 days	C. militaris.	0.31	0.40	1	
	Insecticide	0.40	0.43	0.50	1
	B. bassiana	1			
70 1	M. anisopliae	0.67	1		
70 days	C. militaris.	0.32	0.64	1	
	Insecticide	0.22	0.33	0.32	1
	B. bassiana	1			
94 J	M. anisopliae	0.73	1		
84 days	C. militaris.	0.54	0.67	1	
	Insecticide	0.59	0.50	0.44	1
	B. bassiana	1			
TOTAL	M. anisopliae	0.76	1		
IUIAL	C. militaris.	0.77	0.84	1	
	Insecticide	0.49	0.60	0.56	1

**Table 6.** Matrix of similarity (Index Sorensen) of spider community in rice field sprayed with bioinsecticide containing *B. bassiana*, *M. anisopliae*, and *C. militaris* and abameetin

**Table 7.** Similarity of predatory insect community in rice field sprayed with bioinsecticide containing *B. bassiana, M. anisopliae*, and *C. militaris* and abamectin

Rice ages	Treatments	B. bassiana	M. anisopliae	C. militaris.	Abamectin
	B. bassiana	1			
14 days	M. anisopliae	0.50	1		
	C. militaris.	0.33	0.50	1	
	Insecticide	0.22	0.00	0.00	1
	B. bassiana	1			
28 days	M. anisopliae	0.57	1		
	C. militaris.	0.53	0.31	1	
	Insecticide	0.33	0.20	0.18	1
	B. bassiana	1			
12 dava	M. anisopliae	0.73	1		
42 days	C. militaris.	0.76	0.82	1	
	Insecticide	0.44	0.57	0.46	1
	B. bassiana	1			
56 dama	M. anisopliae	0.89	1		
56 days	C. militaris.	0.69	0.88	1	
	Insecticide	0.70	0.64	0.76	1
	B. bassiana	1			
70 dava	M. anisopliae	0.74	1		
70 days	C. militaris.	0.75	0.72	1	
	Insecticide	0.57	0.55	0.74	1
	B. bassiana	1			
04 1	M. anisopliae	0.89	1		
84 days	C. militaris.	0.71	0.76	1	
	Insecticide	0.50	0.67	0.64	1
	B. bassiana	1			
T-4-1	M. anisopliae	0.91	1		
Total	C. militaris.	0.85	0.91	1	
	Insecticide	0.66	0.77	0.79	1

#### Discussion

From the research result, it was found that population of herbivore insects in plots sprayed with the entomopathogenic fungal bioinsecticide was not significantly different from population of the herbivore in abamectin plot. This because the entomopathogenic fungi applied in this research was effective and had capacity equal to the abamectin in reducing population of herbivore insects. How do you ensure that this population decline is because of abamectin and fungi, if you don't have plots for control (without any treatment)? *B. bassiana* was proved to be effective in killing pest insect of rice, such as brown planthopper (Lee et al. 2015) and rice bug (Girish and Balikai 2015). In this research, *S. litura* found was infected by *B. bassiana*, *M. anisopliae*, and *C. militaris*. Gustianingtyas et al. (2020) also reported that *S. litura* subjected to infection by *M. anisopliae*. *C. militaris* could suppress population of lepidopteran (Shrestha et al., 2016) and coleopteran pest insects (Kryukov et al. 2014)

Based on this study, symptoms developed on *S. litura* larvae infected by *B. bassiana* in rice field were similar to symptom reported by Gustianingtyas et al. (2019) that host insect infected by *B. bassiana* was covered by white mycelia and its body was shriveled. Morphological characteristics of *B. bassiana* colony was in accordance to the result of isolation in the laboratory, white mycelia with hyaline 1-celled cylindrical and septate conidia (Safitri et al. 2018). *M. anisopliae* infecting *S. litura* in this research showed symptom as reported by Humber (2012) that the integument of infected insect turned to white to dark green, green to yellow conidia, 1-celled and cylindrical conidia, and septate hypha. Larvae of *S. litura* infected by *C. militaris* in the rice field showed symptoms similar to those of *B. bassiana* infection, and in accordance to symptoms characteristics reported by Kryukov et al. (2014) that infected insect was white in color with fruiting bodies appear from the insect, the colony was yellowish white in color, and conidia was globular in shape (Zheng et al. 2011).

In this research, the abundance of hunting spider, such as species of Lycosidae, did not decrease after sprayed with the entomopathogenic fungal bioinsecticides well as after being sprayed with abamectin. The abundance of web spider, such as *Bathyphantes*, *A. inustus* and Family of Tetragnathidae decreased after application of the abamectin, while application of the bioinsecticide could only decrease the population of *A. inustus*. Therefore, the entomopathogenic fungal bioinsecticides were relatively safer than the abamectin. Furthermore, spiders generally prey on healthy herbivore insects, while the herbivore insects infected by entomopathogen or the unhealthy ones would not be preyed (Chaubey and Yadav 2017). Web spider abundance decreased after application of the abamectin and the bioinsecticides because the spiders were move less in rice canopy, making the insecticide as well as the bioinsecticide easy to reach the spider body. Therefore, it would better to stop application of bioinsecticide when the abundance of web spider is high.

The abundance of spider increased followed the increase of rice stage and the highest abundance occurred just before harvesting. The abundance of spider continued to high until harvesting because spider likes complex habitat (Amzah et al. 2018) and inhabited by various insect species. Spider is also generalist predator, prey on various insect species, include neutral insects which are generally abundant before harvesting (Karenina et al. 2019). The species diversity of spiders in this reasearch was also higher in plots sprayed with the entomopathogenic fungal bioinsecticide compared to that of the abamectin plot. The higher abundance and species diversity of spiders indicated that the entomopathogenic fungal bioinsecticide was not dangerous to their life. Many reports showed that entomopathogenic fungi did not kill spiders, especially hunting spiders (Prabawati et al. 2019; Hanif et al. 2020).

The abundance of predatory insects did not decrease after the application of the entomopathogenic fungal bioinsecticide or abamectin. Predatory insect has high mobility which differ them from web spiders, so that predatory insect could escape from being exposed to bioinsecticide and abamectin. Furthermore, there is evident that the avoidance behavior in predatory insects from *B. bassiana*-infected insect preys (Seiedy et al. 2015). Therefore, application of entomopathogenic fungi was relatively safe for predatory insects.

The abundance of predatory insect increased until 70 DAT, and decreased at 84 DAT. The predatory insects were generally more specialist in term of prey species compared to spiders. Therefore, at approaching to harvest when the number of herbivore insects decreased, the predatory insects also decreased. Such phenomenon frequently happened in specialist predatory arthropod because population of the predator depended on the fluctuated population of their preys which are called as functional response (Karenina et al. 2019).

The community of spider and predatory insects in plot of *B. bassiana* tent to more similar to the community in plots of bioinsectiside *M. anisopliae* and *C. militaris* compared to that in the abamectin plot. This showed that the effect of application of three entomopathogenic fungal bioinsecticide on the predator communities was not significantly different. The low community similarity between the bioinsecticide and the abamectin because the abamectin could reduce the abundance of spiders and predatory insects. There have been a lot of reports of the decrease of abundance and species diversity of spiders (Preetha et al. 2010) and predatory insects (Salachna et al. 2020) due to application of abamectin containing organophosphate (Bai and Ogbourne 2016). In this research, abamectin as active ingredient is a stomach poison and toxic against insect species of *Menochilus sexmaculatus* (Azod et al. 2016) and spider species of *P. pseudoannulata* (Baehaki et al. 2017).

Guild group made based on function corespondence of arthropods i.e. herbivore insects, neural insects, predators and parasitoids. For one rice cropping season, there was a tendency that the abundance of all guilds group were lower in the abamectin plot. Interestingly, the abundance of predator was always more dominant in plots sprayed with the entomopathogenic fungal bioinsecticide compared to those of herbivore insects, neutral insects, and parasitoid. Neutral

insects were not found in the synhetic insecticie plot. Thus phenomenon showed that predator guild was more tolerant to the entomopathogenic fungal bioinsecticide (Bayissa et al. 2016; Firouzbakht et al. 2015; Gholamzadeh-Chitgar et al. 2017), while abamectin tent to reduce the abundance of predators, herbivore insects, neutral insects and parasitoid (Prabawati et al. 2019; Hanif et al. 2020). Among the three entompathogenic fungi, there was a tendency that the abundance of the predators in plot sprayed with the bioinsecticide of *C. militaris* was higher than that in plots of *B. bassiana* and *M. anisopliae*. Therefore, predator arthropods tent to be more tolerant to *C. militaris* than to *B. bassiana* and *M. anisopliae*.

It could be concluded that the abundance and diversity of arthropod predator species inhabiting freshwater swamp sprayed with the bioinsecticides of *B. bassiana*, *M. anisopliae*, and *C. militaris* did not decrease, while the population of herbivore insects tent to decrease to the same level as its decrease in the abamectin plot. So that, the bioinsecticide of *B. bassiana*, *M. anisopliae*, and *c. militaris* did not decrease the abundance and species diversity of predatory arthropod (non-target arthropods), but could decrease the herbivore insect population.

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# Herbivore insects and predatory arthropods in freshwater swamp rice field in South Sumatra sprayed with bioinsecticides of entomopathogenic fungi and abamectin

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Abstract. Herlinda S, Prabawati G, Pujiastuti Y, Susilawati, Karenia T, Hasbi, Irsan C. 2020. Herbivore insects and predatory arthropods in freshwater swamp rice field in South Sumatra sprayed with bioinsecticides of entomopathogenic fungi and abamectin. Biodiversitas 21: xxxx. Herbivore insect population and predatory arthropods in rice field may be effected by the application entomopathogenic fungi or synthetic insectide. The objective of this research was to analyze individual quantity of herbivore insects and predatory arthropod inhabiting freshwater swamp rice field treated with bioinsecticides and abamectin (commercial insecticide). This research was conducted in the freshwater swamp rice field located in Village Pelabuhan Dalam, Sub District Pemulutan, District Ogan Ilir, and South Sumatra. The experiment was arranged in a Completely Randomized Block Design consisted of four plots of treatment. The research used the bioinsecticides made from entomopathogenic fungi (Beauveria bassiana, Metarhizium anisopliae, and Cordyceps military), and abamectin. Sampling to collect arthropods inhabiting rice canopy was also conducted using an entomological net in the study locations. This research found 12 families of herbivore insects with 22 species dominated by Nilaparvata lugens and Leptocorisa acuta and 32 species of spider belonged to 8 families dominated by Tetragnatha virescens and Oxyopes matiensis. The species diversity of spider was higher in the plots of the bioinsecticide compared to that of the abamectin. Predatory insects found belonged to 14 species belonged to 8 families dominated by species of Ophionea nigrofasciata, Verania discolor, and Paedorus fuscipe. The abundance of predatory insects in plots sprayed with the bioinsecticides was higher compared to that of the abamectin plot. The abundance and species diversity of predatory arthropod sprayed with bioinsecticide of B. bassiana, M. anisopliae, and C. militaris did not decrease, while the population of herbivore insect tended to decrease.

Key words: Beauveria bassiana; Metarhizium anisopliae; Cordyceps militaris; neutral insect; parasitoid

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### **INTRODUCTION**

Rice cultivation in freshwater swamps of South Sumatra has specific characteristics making it different from rice cultivation in other ecosystems in Indonesia (Karenina et al. 2020). Rice cultivation is affected by climate conditions, especially rainfall and tidal flush (Herlinda et al. 2020). From November to April, freshwater swamps is generally inundated so that local farmers cultivate rice only in the period of May to September (Herlinda et al. 2018; Herlinda et al. 2019a). Due to difficulties in managing water, farmers grow rice once a year using the transplanting method. In maintaining their rice cultivations, the local farmers scarcely apply synthetic insecticide (Herlinda et al. 2019b).

The population of insect pests attacking freshwater swamp rice plants is increasing and starting to result in yield loss, especially rice bug (Leptocorixa acuta), while brown planthoppers (Nilavarpata lugens) that has not appeared for a long time start attacking (Hanif et al. 2020). Pests that attack rice plants have been controlled using various entomopathogenic fungi including Beauveria bassiana (Li et al. 2012; Li et al. 2014; Lee et al. 2015), Metarizhium anisopliae (Girish and Balikai, 2015; Chinniah et al. 2016) and Cordycep militaris (Prabawati et al. 2019). C. militaris is reported to produce beauvericin that is toxic to insects (Rachmawati et al. 2018) and B. bassiana has been reported to be effective against homopteran (Li et al. 2012; Li et al. 2014; Lopez et al. 2014; Lee et al. 2015; Sumikarsih et al. 2019), coleopteran (Kavallieratos et al. 2015), hemipteran (Girish and Balikai 2015), and lepidopteran insects (Ayudya et al. 2019; Ma et al. 2019; Gustianingtyas et al. 2020). M. anisopliae has also been demonstrated to be effective against homopteran (Mweke et al. 2019), lepidopteran (Gustianingtyas et al. 2020), and coleopteran insects (Kavallieratos et al. 2015). M. anisopliae could be used in multiple roles, ranging from controlling insect pests and promoting plant growth (Liu et al. 2017).

Being effective against various orders of insects, including the coleopteran insects (Kavallieratos et al. 2015), the rice field ecosystem which is dominated by species of predatory insects, it is important to be very careful in the application of entomopathogenic fungi in ecosystem to avoid negative impact on non-target arthropods (Scorsetti et al. 2017). High species diversity and abundance of arthropod in freshwater swamp rice field (Herlinda et al. 2018) might be disturbed by the application of the entomopathogenic fungi. However, there have been some reports suggested that entomopathogenic fungi did not affect abundance and species diversity of arthropods (Prabawati et al. 2019; Hanif et al. 2020). For example, predatory insect in paddy field (Andrallus spinidens) was reported to be resistant to B. bassiana (Firouzbakht et al. 2015; Gholamzadeh-Chitgar et al. 2017; Scorsetti et al. 2017). Parasitoid tends to avoid host insect infected by entomopathogenic fungi (Emami et al. 2013). ISSN: 1412-033X

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Coccinellidae, the family of generalist predatory insect, was reported to be resistant to *M. anisopliae* (Bayissa et al. 2016). This research aimed to analyze individual quantity of herbivore insects and predatory arthropod inhabiting freshwater swamp rice field treated with bioinsecticides containing *Beauveria bassiana*, *Metarhizium anisopliae*, and *Cordyceps militaris* and the abamectin.

#### MATERIALS AND METHODS

#### Study area

The research was conducted from April to August 2018 in the freshwater swamp rice field located in Village Pelabuhan Dalam, Sub District Pemulutan, District Ogan Ilir, and South Sumatra, Indonesia. The experiment was arranged in a Completely Randomized Block Design consisted of four plots of treatment. The first three plots were treated with bioinsecticide containing *B. bassiana*, *M. anisopliae*, and *C. military*, and the forth plot was treated with the abamectin (the commercial insecticide) as control. The plot area was 120 m<sup>2</sup> for treatment and each treatment was replicated five times. Distance between plots was 10 m.

#### Land preparation, transplanting, and crop maintenance

Land preparation was conducted using a moldboard plow and was continued by using harrow plow. Before being plowed, the land was cleared using a hoe and a long knife. The excessive water was pumped out until the soil was slightly watery, and the dung compost was added at a dosage of 1 ton ha<sup>-1</sup>. Rice seed used were certified purple labeled seed of Mekongga variety at a dosage of 50 kg/ha. Rice was cultivated using a transplanting system from which the seedlings were prepared in 34 x 26 x 5  $cm^3$ plastic trays filled with 3 cm high soil. The seeds were soaked in water for 24 hours before being planted in the prepared plastic trays. The soaked seeds were spread evenly in each tray at a dosage of 150 g per tray. The trays were then covered with thick plastic for seven days. The seven-day-old seedlings were then transferred to places receiving enough sunlight. Fourteen day old seedlings were transplanted to all prepared plots at a density of 2 seedlings per hole in a 2:1 (12.5 x 25 x 50 cm<sup>3</sup>) "jajar legowo" planting system (Figure 1).

Crop maintenance was conducted in the form of replanting, irrigating, weeding, and fertilizing. Replanting was conducted seven days after transplanting to replace the dead seedlings. Irrigating was conducted by making use of water flush from the river under high tide and pumping system. Weeding was conducted both at vegetative and generative stages using a sickle. Fertilizer used was shrimp shell extract compost prepared according to the method by Suwandi et al. (2012). The compost was applied every two weeks started from 14 days after planting until harvesting (84 days) at a dosage of  $2 L ha^{-1}$ . The extract compost was a fermentation result of shrimp shell meal containing

chitinolytic bacteria, cellulolytic bacteria, and sulfate diluter. Nitrogen fertilizer was applied 30 days after

planting at a dosage of 100 kg ha<sup>-1</sup>.



**Figure 1.** Two day old rice seedling in a plastic tray (A), 7 day old seedlings (B), rice transplanting (D) rice planted in "jajar legowo" planting system (E)

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#### Formulation and application of bioinsecticides

All species of fungi have been explored from soil following the modified method of Kin et al. (2017). B. bassiana and M. anisopliae used in this research were explored from freshwater swamp soil of South Sumatra, while C. militaris was explored from the soil in Central Kalimantan, Indonesia. B. bassiana, M. anisopliae, and C. militaris were cultured in solid media of Sabouraud Dextrose Agar (SDA, Merck) made from 16.2 g SDA in 250 mL aquadest. The cultures were incubated for 10 x 24 hours. The 10 days old fungal solid cultures were then cultured in liquid media (Figure 2) of Sabouraud Dextrose Broth (SDB, Merck) made from 30 g SDB in 1000 ml aquadest and were incubated in a 120 rpm shaker for 7 x 24 hours. The liquid culture of the entomopathogenic fungi was used to make bioinsecticide. Seven days after incubation, the density of fungal spores of the bioinsecticide was counted, the counting was stopped when the spore density reached  $1 \times 10^9$  conidia mL<sup>-1</sup>. One liter of bioinsecticide was made by adding 400 mL shrimp shell meal extract compost as a carrier and active ingredient consisting of 600 mL liquid culture of entomopathogenic fungi and 10 mL vegetable oil. Compost extract used to make bioinsecticide was previously autoclaved under 1 atm for 2 hours.

The bioinsecticides were applied at a dosage of 2 L ha<sup>-1</sup> per application. The applications were conducted at 13, 27, 41, 55, 69, and 83 days after transplanting (DAT). The bioinsecticide applications were conducted in the evening (at 5 pm) to avoid spores damaged by ultraviolet. The control plot was sprayed with abamectin at a dosage of 0.5 L ha<sup>-1</sup>. The insecticide contained abamectin active ingredient and was sprayed at the same time as bioinsecticide applications. The control plot was 50 m away from treatment plots to avoid contamination in the treatment plots, and there was 10 m distance between bioinsecticide treatment plots. A day after application, arthropod samplings were carried out to collect arthropods inhabiting rice canopy.

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#### Arthropod sampling in rice canopy

Sampling to collect arthropods inhabiting rice canopy was conducted using an entomological net (75 cm length, 30 cm diameter, and 100 cm length of handle) (Lami et al. 2016; Ivantsova et al. 2017). Sampling was conducted in every treatment subplot and was repeated five times. Samplings were conducted at 14, 28, 42, 56, 70, and 84 DAT. Arthropod samplings were conducted in the morning, at 06.00-07.00 am. During sampling, the sweep net was swept 30 cm depth into rice canopy to collect representative samples.

Sampled arthropods were cleaned and placed in the labeled plastic bottles (330 mL) containing 100 mL absolute ethanol. The samples were brought into the Laboratory of Entomology, Department of Plant Pest and Disease, Faculty of Agriculture, Universitas Sriwijaya for identification. Spider identification was conducted using the reference of Barrion and Litsinger (1995), while for insect identification we used reference of Gullan and Cranston (2014) and Heinrichs et al. (2017).

#### Data analysis

Data of arthropods species were grouped based on their guild, comprised of herbivore insects, predators, parasitoids, and neutral insects, and was presented in the form of a graphic. The difference of individual quantity of the herbivore insects, spiders, and predatory insects amongst treatments was analyzed using Analysis of Variance (ANOVA). If there was a significant difference among treatments, the analysis was continued with Tukey's HSD (Honesty Significant Difference) test at 5% degree of significance. Analyses data was conducted using Software of SAS University Edition 2.7 9.4 M5. Data of abundance were used to analyze species diversity by using the Shannon index (H'). Degree of diversity was counted using the Evenness index (J') derived from the Shannon function, and Berger-Parker dominance biodiversity indices. The coefficient of Sorensen was counted to measure the degree of similarity of the spider or predatory insect among treatments (Magurran 2004).



**Figure 2.** Cultures of *Beauveria bassiana* (A), *Metarhizium anisopliae* (B), and *Cordycep militaris* (C) on SDB; liquid bioinsecticide of *Beauveria bassiana* (D), *Metarhizium anisopliae* (E), and *Cordycep militaris* (F)

### **RESULTS AND DISCUSSION**

# Abundance of herbivore insects in one rice cropping season

In one rice cropping season, it was found 22 species of herbivore insects belonged to 12 families. Dominant herbivore species were *Nilaparvata lugens, Leptocorisa acuta, Acrida turrita, Valanga nigricornis,* and *Dicladispa armigera* (Figure 3). The population of the herbivore was not significantly different among treatments, except for Family Cecidomyiidae (Table 1). Therefore, the capacity of bioinsecticide in suppressing the population of the herbivore insects has generally almost similar to the capacity of the abamectin.

During field observations, insect died due to infection by entomopathogenic fungi were documented and showed specific symptoms of each fungus. The larvae of S. litura found infected by B. bassiana in the rice field showed white mycelia covering its body, the dried, and not smelly (Figure 4). Larvae of S. litura infected by M. anisopliae showed symptoms of the dried body covered by greenish white mycelia and was not smelly, while larvae of S. litura infected by C. militaris showed similar symptoms to those of B. bassiana infection, larval body drying, shriveled, covered by white mycelia, and not smelly. Entomopathogenic fungi infected sampled insects were isolated and purified in the laboratory. Color, structure, and spore of the isolated fungi were as presented in Figure 5.

# Abundance of spider and predatory insect in one rice cropping season

This research found 32 species of spiders belonged to eight families. The dominant species of them were *Tetragnatha virescens*, *T. virescens*, *T. maxilosa*, *Argiope catenulata*, and *Oxyopes matiensis* (Figure 6). The total abundance of spiders in the plot treated with the abamectin was significantly lower than that of plots treated with bioinsecticide (Table 2). The abundance of *Bathyphantes* sp. in abamectin plot decreasedThe highest abundance of *Araneus inustus* was found in the plot treated with bioinsecticide *C. militaris* and was different from that of other treatments. The abundance of Tetragnathidae was significantly decreasing in the plot treated with insecticide was found to be lower compared to plots treated with bioinsecticide.

Predatory insects found in this research belonged to 14 species and eight families. The dominant species were *Formicomus* sp., *Ophionea nigrofasciata*, *Verania discolor*, *Verania lineata*, *Menochilus sexmaculatus*, *Micrapis inop*, *Paedorus fuscipe*, *Cyrtorhinus lividipennis*, and *Orthotylus* sp. (Figure 7). The abundance of predatory insects was not significantly different among treatments (Table 3).

# Characteristic of spider and predatory insect community in one rice cropping season

The number of spiders increased with increasing of rice age (Table 4). The highest number of spiders occurred when the rice were at a mature grain ripening stage (84 DAT). The abundance of spider in the plot treated with abamectin tend to be lower compared to that of plots treated with the bioinsecticides. The total number of spiders in one rice cropping season were higher in the plots treated with the bioinsecticide than that in the plot treated with the abamectin. The species diversity of spiders was also higher in the plots treated with entomopathogenic fungal bioinsecticide compared to that in the abamectin plot. The high species diversity in plots treated with entomopathogenic fungal bioinsecticide was also followed by the high species evenness and low species dominance.

Number of predatory insects decreased with increasing stages of rice, but at approaching harvest (84 DAT), the number of predatory insects drastically decreased (Table 5). The highest abundance of predatory insects occurred at the milk grain ripening stage (70 DAT). The phenomenon of abundance fluctuation of predatory insect was slightly different from that of spider, which its peak occurred at a mature grain ripening stage (84 DAT). The abundance of the predatory insects in plots treated with abamectin was generally lower than that of plots treated with the entomopathogenic fungal bioinsecticide. Total of abundance of predatory insects in one rice cropping season sprayed with entomopathogenic fungal of plots bioinsecticide was higher compared to that of abamectin plot. However, species diversity, evenness, and species dominance tend to be similar among treatments.

Spider community in plot sprayed with the bioinsecticide of *B. bassiana* tent to be more similar to that of plots sprayed with bioinsecticide (*M. anisopliae* and *C. militaris*) than that of abamectin plot. The total of one rice cropping season, spider community in plot sprayed with bioinsecticide *B. bassiana* was more similar to that of plot of *C. militaris* (0.77), followed by plot *M. anisopliae* (0.76), and least similar to that of the abamectin plot (0.49) (Table 6).

Predatory insect community in plot sprayed with bioinsecticide *B. bassiana* tent to be more similar to that of plots sprayed with bioinsecticide *M. anisopliae* and *C. militaris* than that of the abamectin plot (Table 7). The total of one rice cropping season, predatory insect community in plots sprayed with bioinsecticide *B. bassiana* was more similar to that of plot *M. anisopliae* (0.91), followed by plot of *C. militaris* (0.85), and least similar to that of insecticide plot (0.66).



**Figure 3.** Herbivore insects found in rice canopy: *Spodoptera* sp. (A), *Pelopidas* sp. (B), *Amata nigriceps* (C), *Oxya chinensis* (D), *Scirpophaga* sp. (E), *Dicladispa armigera* (F), *Chrysolina coerulans* (G), *Leptocorisa acuta* (H), *Acrida turita* (I), *Nilaparvata lugens* (J)

Table 1. Population of herbivore insects in rice canopy sprayed with bioinsecticides (B. bassiana, M. anisopliae, C. militaris) and abamectin

Onde / Fernile/ Secondar	Mean of herbivore abundance (individuals/nets)				Employe	P value	Tukey's HSD
Ordo / Family/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	<b>F</b> -value	(0.05)	test
COLEOPTERA	0.2	0.13	0.17	0.23	0.26	0.85	
Chrysomelidae	0.2	0.13	0.17	0.23	0.26	0.85	
Chrysolina coerulans	0.07	0	0.07	0.2	2.11	0.15	
Dicladispa armigera	0.13	0.13	0.1	0.03	0.53	0.67	
DIPTERA	$0.4^{b}$	0.03 <sup>ab</sup>	$0.07^{ab}$	$0.00^{a}$	4.40*	0.03	0.20
Cecidomyiidae	$0.4^{b}$	0.03 <sup>ab</sup>	$0.07^{ab}$	$0.00^{a}$	4.40*	0.03	0.20
Cecidomyiidae sp.	0.33	0	0	0	2.55	0.1	
Orseolia sp.	0.07	0.03	0.07	0	0.81	0.51	
HEMIPTERA	1.67	1.1	1.7	0.97	2.42	0.12	
Alydidae	0.57	0.4	0.73	0.5	2.02	0.17	
Leptocorisa acuta	0.57	0.4	0.73	0.5	2.02	0.17	
Cicadellidae	0.17	0.1	0	0.1	0.42	0.74	
Nephotettix cincticeps	0	0	0	0.03	1.00	0.43	
Nephotettix virescen	0	0.1	0	0.07	1.04	0.41	
Recilia dorsalis	0.17	0	0	0	1.00	0.43	
Delphacidae	0.93	0.6	0.97	0.37	2.59	0.1	
Nilaparvata lugens	0.77	0.53	0.9	0.27	2.23	0.14	
Sogatella furcifera	0.17	0.07	0.07	0.1	0.50	0.69	
LEPIDOPTERA	0.37	0.3	0.5	0.17	2.61	0.1	
Pyralidae	0.33	0.23	0.33	0.17	0.99	0.43	
Cnaplocrosis medinalis	0	0	0	0.03	1.00	0.43	
Scirpophaga incertulas	0.17	0.13	0.17	0.07	1.31	0.32	
Scirpophaga sp.	0.17	0.1	0.17	0.07	0.66	0.59	
Erebidae	0	0	0.07	0	1.00	0.43	
Amata nigriceps	0	0	0.07	0	1.00	0.43	
Hesperiidae	0	0	0.03	0	1.00	0.43	
Pelopidas mathias	0	0	0.03	0	1.00	0.43	
Noctudae	0.03	0.07	0.07	0	0.47	0.71	
Spodoptera sp.	0.03	0.07	0.07	0	0.47	0.71	
ORTHOPTERA	0.63	0.83	0.67	0.8	0.58	0.64	
Acrididae	0.63	0.83	0.67	0.77	0.48	0.7	
Acrida turrita	0.2	0.17	0.1	0.4	2.68	0.09	
Gesonula mundata	0.03	0.1	0.03	0.1	0.35	0.79	
Oxya chinensis	0.07	0.1	0.1	0.1	0.07	0.97	
Valanga nigricornis	0.33	0.47	0.43	0.17	2.02	0.17	
Pyrgomorphidae	0	0	0	0.03	1.00	0.43	
Atractomorpha crenulata	0	0	0	0.03	1.00	0.43	
THYSANOPTERA	0.1	0.1	0.1	0.07	0.11	0.95	
Thripidae	0.1	0.1	0.1	0.07	0.11	0.95	
Liothrips sp.	0.1	0.1	0.1	0.07	0.11	0.95	
TOTAL	3.37	2.5	3.2	2.23	2.77	0.09	

Note: \*: significantly different; values within a row followed by the same letters were not significantly different at P<0.05 according to Tukey's HSD test



**Figure 4.** Herbivore insects infected by the entomopathogenic fungal of bioinsecticide in rice field: healthy larvae of *Spodoptera* sp. (A), infected by *B. bassiana* (B), infected by *M. anisopliae* (C), and infected by *C. militaris* (D)



**Figure 5.** Spores of *Beauveria bassiana* (A), *Metarhizium anisopliae* (B), and *Cordycep militaris* (C); colonies of *Beauveria bassiana* (d), *Metarhizium anisopliae* (E), and *Cordycep militaris* (F)



**Figure 6.** Spiders found in rice canopy: Linyphiidae (A), *Tetragnatha virescens* (B), *Tetragnatha maxilosa* (C), *Argiope catenulate* (D), *Oxyopes matiensis* (E), Lycosidae (F), Salticidae sp. A (G), Salticidae sp. B (H), *Oxyopes javanus* (I), *Pardosa* sp. (J)



Figure 7. Predatory insect in rice field: Verania lineata (A), Menochilus sexmaculatus (B), Micrapis inops (C), Formica sp. (D), Odontoponera transversa (E), Argia sp. (F), Agriocnemis sp. (G), Ophionea nigrofasciata (H), Paederus fuscipes (I), and Conocephalus longipennis (J)



Figure 8. Arthropod abundance on rice field treated with bioinsecticides and abamectin (insecticide)

<b>Table 2.</b> The abundance of spider inhabiting rice canopy sprayed with bioinsecticides	(B.bassiana, M. anisopliae, and C. militaris) and
abamectin	

	Mean of spider abundance (individuals/nets)					P value	Tukey's
Ordo / Family/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	- F-value	(0.05)	HSD test
ARACHINIDA							
Araneidae	0.03	0.07	0.27	0.13	2.02	0.17	
Araneus inustus	$0.00^{a}$	$0.03^{ab}$	$0.2^{b}$	0.03 <sup>ab</sup>	4.82*	0.02	0.11
Araneidae sp.	0.03	0.03	0.07	0.07	0.35	0.79	
Gea subarmata	0.00	0.00	0.00	0.03	1.00	0.43	
Linyphiidae	0.47	0.30	0.20	0.10	2.76	0.09	
Bathyphantes tagalogensis	0.00	0.00	0.07	0.00	2.67	0.10	
Bathyphantes sp. A	0.07	0.00	0.00	0.07	0.89	0.48	
Bathyphantes sp. B	0.13 <sup>ab</sup>	0.2 <sup>b</sup>	$0.07^{ab}$	$0^{a}$	3.74*	0.04	0.12
Atypena adelinae	0.03	0.03	0.03	0.03	0.00	1.00	
Erigone bifurca	0.03	0.00	0.00	0.00	1.00	0.43	
Linyphiidae sp. A	0.20	0.03	0.03	0.00	2.46	0.11	
Atypena formosana	0.00	0.03	0.00	0.00	0.98	0.44	
Lycosidae	0.10	0.13	0.37	0.10	1.90	0.18	
Arctosa sp.	0.00	0.03	0.03	0.00	0.60	0.63	
Pardosa birmanica	0.03	0.00	0.00	0.00	1.00	0.43	
Pardosa pseudoannulata	0.03	0.00	0.27	0.07	2.10	0.15	
Pardosa apostoli	0.00	0.03	0.00	0.00	1.00	0.43	
Pardosa pullata	0.03	0.03	0.03	0.00	0.37	0.77	
Hogna rizali	0.00	0.00	0.03	0.00	1.00	0.43	
Pirata luzonensis	0.00	0.03	0.00	0.03	0.62	0.62	
Oxvopidae	0.03	0.03	0.10	0.03	0.51	0.68	
Oxvopes javanus	0.00	0.03	0.07	0.03	0.76	0.54	
Oxvopes matiensis	0.00	0.00	0.03	0.00	1.00	0.43	
Oxyopes pingasus	0.03	0.00	0.00	0.00	1.00	0.43	
Salticidae	0.07	0.07	0.07	0.07	0.00	1.00	
Salticidae sp. A	0.03	0.00	0.03	0.03	0.38	0.77	
Salticidae sp. B	0.03	0.07	0.03	0.03	0.37	0.77	
Theridiidae	0.07	0.03	0.03	0.00	0.62	0.62	
Theridiidae sp.	0.07	0.03	0.03	0.00	0.62	0.62	
Tetragnathidae	1.33 <sup>b</sup>	$1.2^{ab}$	$1.2^{ab}$	0.63 <sup>a</sup>	4.16*	0.03	0.28
Tetragnatha javana	0.33	0.40	0.43	0.20	2.40	0.12	
Tetragnatha maxillosa	0.07	0.07	0.03	0.03	0.24	0.87	
Tetragnatha montana	0.07	0.00	0.03	0.03	0.37	0.77	
Tetragnatha virescens	0.47	0.33	0.37	0.23	0.76	0.54	
Tetragnatha nitens	0.00	0.03	0.03	0.03	0.38	0.77	
Tetragnatha okumae	0.33	0.13	0.10	0.10	1.88	0.19	
Tetragnatha mandibulata	0.03	0.07	0.10	0.00	0.84	0.50	
Tetragnatha vermiformis	0.00	0.10	0.07	0.00	1.27	0.33	
Tetragnatha iwahigensis	0.03	0.07	0.07	0.00	0.76	0.54	
Theridiosomatidae	0.00	0.00	0.07	0.00	1.00	0.43	
Wendilgarda sp.	0.00	0.00	0.07	0.00	1.00	0.43	
Total Abundance (N)	2.1 <sup>b</sup>	1.83 <sup>ab</sup>	2.3 <sup>b</sup>	1.07 <sup>a</sup>	7.29*	0.00	0.30

Note: A submatrice (14) 2.1 1.65 2.5 1.07  $7.29^{-6}$  0.00 0.30Note: \*: significantly different; values within a row followed by the same letters were not significantly different at P<0.05 according to Tukey's HSD test Table 3. The abundance of predatory insect in rice field sprayed with bioinsecticides (*B. bassiana, M. anisopliae*, and *C. militaris*) and abamectin

Onde / Femile/ Seesies	Mean of	F value	P value			
Ordo / Family/ Spesies	B. bassiana	M. anisopliae	C. militaris	Abamectin	0.05	(0.05)
COLEOPTERA	0.47	0.63	0.40	0.63	0.64	0.60
Anthicidae	0.07	0.07	0.07	0.00	0.86	0.49
Formicomus sp.	0.07	0.07	0.07	0.00	0.86	0.49
Carabidae	0.10	0.17	0.17	0.07	1.07	0.40
Ophionea nigrofasciata	0.10	0.17	0.17	0.07	1.07	0.40
Coccinellidae	0.67	0.57	0.57	0.40	1.41	0.29
Verania discolor	0.03	0.07	0.03	0.10	0.19	0.90
Verania lineata	0.00	0.00	0.00	0.07	1.00	0.44
Menochilus sexmaculatus	0.13	0.04	0.07	0.07	0.24	0.86
Micraspis inops	0.50	0.47	0.47	0.17	3.14	0.07
Staphylidae	0.27	0.30	0.33	0.27	0.36	0.78
Paederus fuscipes	0.27	0.30	0.33	0.27	0.36	0.78
HEMIPTERA	0.13	0.10	0.00	0.03	1.49	0.27
Miridae	0.13	0.10	0.00	0.03	1.49	0.27
Cyrtorhinus lividipennis	0.13	0.03	0.00	0.00	2.81	0.09
Orthotylus sp.	0.00	0.07	0.00	0.03	0.74	0.58
HYMENOPTERA	0.30	0.30	0.20	0.10	0.87	0.48
Formicidae	0.30	0.30	0.20	0.00	0.87	0.48
Formica sp.	0.07	0.13	0.03	0.00	0.61	0.66
Odontoponera transversa	0.23	0.17	0.17	0.10	0.27	0.85
ODONATA	0.33	0.20	0.20	0.20	0.34	0.80
Coenagrionidae	0.33	0.20	0.20	0.20	0.34	0.80
Pyrrhosoma sp.	0.20	0.07	0.10	0.10	1.00	0.43
Agriocnemis pygmaea	0.20	0.07	0.10	0.10	0.10	0.96
ORTHOPTERA	0.33	0.27	0.27	0.17	0.56	0.65
Tettigonidae	0.33	0.27	0.27	0.17	0.56	0.65
Conocephalus longipennis	0.33	0.27	0.27	0.17	0.56	0.65
Total Abundance (N)	2.20	1.97	1.80	1.23	1.25	0.34

Note: values within a row followed by the same letters were not significantly different at P<0.05 according to Tukey's HSD test

# The abundance of associated arthropod with rice in one cropping season

The abundance of associated arthropods tent to increase with increasing rice growth stage (Figure 8). In all observations, the abundance of predatory insect was found to be higher compared to the abundance of other guilds (neutral insect, herbivore, and parasitoid). The abundance of predator was generally higher in plots sprayed entomopathogenic fungal bioinsecticide compared to that of abamectin plot. The abundance of all guilds of arthropod in the plot of abamectin was always lower compared to that of bioinsecticide plots. When rice was 70 days old, the abundance of herbivore reached its peak, then decreased at a mature grain stage. The abundance of predators reached its peak when rice was 84 days old.

### Discussion

From this study, the population of herbivore insects in plots sprayed with the entomopathogenic fungal bioinsecticides was not significantly different from the population of the herbivore in the abamectin plot. This was because the entomopathogenic fungi applied in this research was effective and had capacity equal to the abamectin in reducing the population of herbivore insects. *B. bassiana* was proved to be effective in killing pest insects of rice, such as brown planthopper (Lee et al. 2015) and rice bug (Girish and Balikai 2015). In this research, *S. litura* found was infected by *B. bassiana*, *M. anisopliae*, and *C. militaris*. Gustianingtyas et al. (2020) also reported that *S. litura* subjected to infection by *M. anisopliae*. *C. militaris* could suppress the population of lepidopteran (Shrestha et al. 2016) and coleopteran pest insects (Kryukov et al. 2014).

Based on this study, symptoms developed on S. litura larvae infected by B. bassiana in the rice field were similar to the symptoms reported by Gustianingtyas et al. (2019) that host insect infected by B. bassiana was covered by white mycelia and its body was shriveled. Morphological characteristics of B. bassiana colony were in accordance with the result of isolation in the laboratory, white mycelia with hyaline 1-celled cylindrical and septate conidia (Safitri et al. 2018). M. anisopliae infecting S. litura in this research showed symptom as reported by Humber (2012) that the integument of infected insect turned to white to dark green, green to yellow conidia, 1-celled and cylindrical conidia, and septate hypha. Larvae of S. litura infected by *C. militaris* in the rice field showed symptoms similar to those of *B. bassiana* infection, and in accordance to symptoms characteristics reported by Kryukov et al. (2014) that infected insect was white in color with fruiting bodies appear from the insect, the colony was yellowishwhite in color, and conidia was globular in shape (Zheng et al. 2011).

Table 4. Characteristic of spider community in rice canopy sprayed with	bioinsecticides (B.	bassiana, M.	anisopliae,	and C. militaris)
and abamectin				

Characteristic of spider community	B. bassiana	M. anisopliae	C. militaris	Abamectin
14 days				
Abundance (individual/hill)	0.80	0.40	0.80	0.80
Biodiversity index (H')	1.03	0.69	1.38	1.38
Dominance index (D)	0.50	0.50	0.25	0.25
Evenness index (E)	0.94	1.00	1.00	1.00
28 days				
Abundance (individual/hill)	1.00	1.20	1.20	0.40
Biodiversity index (H')	1.33	1.24	1.79	0.69
Dominance index (D)	0.40	0.50	0.16	0.50
Evenness index (E)	0.96	0.89	1.00	1.00
42 days				
Abundance (individual/hill)	2.2	1.80	2.00	0.80
Biodiversity index (H')	1.03	1.67	1.74	1.03
Dominance index (D)	0.45	0.33	0.40	0.50
Evenness index (E)	0.94	0.93	0.89	0.94
56 days				
Abundance (individual/hill)	3.00	1.80	2.20	1.00
Biodiversity index (H')	1.70	1.30	1.12	1.33
Dominance index (D)	0.33	0.55	0.54	0.40
Evenness index (E)	0.87	0.80	0.80	0.96
70 days				
Abundance (individual/hill)	2.40	2.40	2.60	1.00
Biodiversity index (H')	1.97	1.81	2.35	1.79
Dominance index (D)	0.25	0.41	0.15	0.16
Evenness index (E)	0.95	0.87	0.98	1.00
84 days				
Abundance (individual/hill)	3.20	3.40	5.00	2.20
Biodiversity index (H')	1.53	1.39	2.07	1.66
Dominance index (D)	0.56	0.58	0.40	0.45
Evenness index (E)	0.73	0.71	0.80	0.85
TOTAL				
Abundance (individual/hill)	12.6	11	13.8	6.2
Biodiversity index (H')	2.48	2.59	2.75	2.45
Dominance index (D)	0.22	0.21	0.18	0.22
Evenness index (E)	0.82	0.83	0.85	0.88

In this research, the abundance of hunting spider, such as species of Lycosidae did not decrease after being sprayed with the entomopathogenic fungal bioinsecticides well as after being sprayed with abamectin. The abundance of a web spider, such as Bathyphantes, A. inustus, and Family of Tetragnathidae, decreased after application of the abamectin, while application of the bioinsecticide could only decrease the population of A. inustus. Therefore, the entomopathogenic fungal bioinsecticides were relatively safer than the abamectin. Furthermore, spiders generally prey on healthy herbivore insects, while the herbivore insects infected by entomopathogen or the unhealthy ones would not be preyed (Chaubey and Yadav 2017). Web spider abundance decreased after the application of the abamectin and the bioinsecticides because the spiders were moving less in rice canopy, making the insecticide as well as the bioinsecticide easy to reach the spider body.

Therefore, it would better to stop the application of bioinsecticide when the abundance of web spiders is high.

The abundance of spiders increased, followed by the increase of rice stage, and the highest abundance occurred just before harvesting. The abundance of spiders continued to high until harvesting because spider likes complex habitat (Amzah et al. 2018) and inhabited by various insect species. Spider is also a generalist predator, prey on various insect species, include neutral insects, which are generally abundant before harvesting (Karenina et al. 2019). The species diversity of spiders in this research was also higher in plots sprayed with the entomopathogenic fungal bioinsecticide compared to that of the abamectin plot. The higher abundance and species diversity of spiders indicated that the entomopathogenic fungal bioinsecticide was not dangerous to their life. Many reports showed that entomopathogenic fungi did not kill spiders, especially hunting spiders (Prabawati et al. 2019; Hanif et al. 2020).

Table 5. Characteristic of pro-	edatory insect community	y in rice field sprayed	with bioinsecticides (. B	. bassiana, M.	anisopliae, and C.
militaris) and abamectin					

Characteristic of spider community	<b>B</b> bassiana	M. anisonliae	C. militaris	Abamectin
14 days old rice	Droussiana	nii anisopnae	0	Tibuliceun
Abundance (individual/hill)	1.00	0.60	0.20	0.80
Riodiversity index (H <sup>2</sup> )	1.00	1.00	0.20	0.80
Dominance index (D)	0.40	0.33	1.00	0.00
Evenness index (E)	0.40	1.00	0.00	0.23
28 days old rice	0.90	1.00	0.00	0.00
Abundance (individual/hill)	1.60	1.20	1.40	0.80
Riodiversity index (H <sup>2</sup> )	1.00	1.20	0.55	1.38
Dominance index (D)	0.25	0.22	0.35	0.25
Evenness index (E)	0.23	0.33	0.28	1.00
42 days old rice	0.07	0.90	0.34	1.00
Abundance (individual/hill)	2.60	1.80	1.60	1.00
Biodiversity index (H')	1.94	1.88	1.66	1.66
Dominance index (D)	0.15	0.22	0.37	0.40
Evenness index (E)	0.88	0.97	0.93	1.20
56 days old rice	0100	0177	0170	1.20
Abundance (individual/hill)	2.80	2.60	2.40	1.80
Biodiversity index (H')	1.94	1.81	1.86	1.83
Dominance index (D)	0.28	0.30	0.25	0.33
Evenness index (E)	0.93	0.93	0.95	0.94
70 days old rice				
Abundance (individual/hill)	2.60	2.80	2.20	1.60
Biodiversity index (H')	1.60	1.90	1.67	1.49
Dominance index (D)	0.30	0.28	0.27	0.37
Evenness index (E)	0.89	0.91	0.93	0.92
84 days old rice				
Abundance (individual/hill)	0.20	2.80	3.00	1.40
Biodiversity index (H')	1.95	2.06	1.69	1.54
Dominance index (D)	0.23	0.21	0.33	0.28
Evenness index (E)	0.93	0.94	0.87	0.96
TOTAL				
Abundance (individual/hill)	13.2	11.8	9.8	7.4
Biodiversity index (H')	2.25	2.27	2.11	2.25
Dominance index (D)	0.91	0.89	0.88	0.94
Evenness index (E)	0.22	0.23	0.28	0.21

The abundance of predatory insects did not decrease after the application of the entomopathogenic fungal bioinsecticide or abamectin. The predatory insect has high mobility which differs them from web spiders so that predatory insect could escape from being exposed to bioinsecticide and abamectin. Furthermore, there is evidence that the avoidance behavior in predatory insects from *B. bassiana*-infected insect preys (Seiedy et al. 2015). Therefore, the application of entomopathogenic fungi was relatively safe for predatory insects.

The abundance of the predatory insect was increasing until 70 DAT and was decreased at 84 DAT. The predatory insects were generally more specialists in terms of prey species compared to spiders. Therefore, at approaching to harvest when the number of herbivore insects decreased, the predatory insects also decreased. Such a phenomenon frequently happened in specialist predatory arthropod because the population of the predator depended on the fluctuated population of their prey which are called as a functional response (Karenina et al. 2019).

The community of spider and predatory insects in the plot of *B. bassiana* tent to more similar to the community in plots of bioinsecticide M. anisopliae and C. militaris compared to that in the abamectin plot. This showed that the effect of the application of three entomopathogenic fungal bioinsecticides on the predator communities was not significantly different. The low community similarity between the bioinsecticide and the abamectin because the abamectin could reduce the abundance of spiders and predatory insects. There have been a lot of reports of the decrease of abundance and species diversity of spiders (Preetha et al. 2010) and predatory insects (Salachna et al. 2020) due to the application of abamectin containing organophosphate (Bai and Ogbourne 2016). In this research, abamectin as an active ingredient is a stomach poison and toxic against insect species of Menochilus sexmaculatus (Azod et al. 2016) and spider species of P. pseudoannulata (Baehaki et al. 2017).

<b>Rice ages</b>	Treatments	B. bassiana	M. anisopliae	C. militaris.	Abamectin
	B. bassiana	1			
14 days	M. anisopliae	0.33	1		
14 days	C. militaris.	0.25	0.00	1	
	Abamectin	0.25	0.00	0.50	1
	B. bassiana	1			
20.1	M. anisopliae	0.18	1		
28 days	C. militaris.	0.55	0.17	1	
	Abamectin	0.29	0.25	0.25	1
	B. bassiana	1			
12 days	M. anisopliae	0.40	1		
42 days	C. militaris.	0.48	0.42	1	
	Abamectin	0.40	0.31	0.43	1
	B. bassiana	1			
56 dava	M. anisopliae	0.58	1		
Jo uays	C. militaris.	0.31	0.40	1	
	Abamectin	0.40	0.43	0.50	1
	B. bassiana	1			
70 dava	M. anisopliae	0.67	1		
70 days	C. militaris.	0.32	0.64	1	
	Abamectin	0.22	0.33	0.32	1
	B. bassiana	1			
84 days	M. anisopliae	0.73	1		
64 uays	C. militaris.	0.54	0.67	1	
	Abamectin	0.59	0.50	0.44	1
	B. bassiana	1			
TOTAL	M. anisopliae	0.76	1		
IUIAL	C. militaris.	0.77	0.84	1	
	Abamectin	0.49	0.60	0.56	1

**Table 6.** Matrix of similarity (Index Sorensen) of spider community in rice field sprayed with bioinsecticides (*B. bassiana, M. anisopliae*, and *C. militaris*) and abamectin

Table 7. Similarity of predatory in	nsect community in rice field spra	yed with bioinsecticides	(B. bassiana, M	1. anisopliae, a	and C.
militaris) and abamectin					

Rice ages	Treatments	B. bassiana	M. anisopliae	C. militaris.	Abamectin
	B. bassiana	1			
14 days	M. anisopliae	0.50	1		
	C. militaris.	0.33	0.50	1	
	Abamectin	0.22	0.00	0.00	1
	B. bassiana	1			
28 days	M. anisopliae	0.57	1		
	C. militaris.	0.53	0.31	1	
	Abamectin	0.33	0.20	0.18	1
	B. bassiana	1			
42 days	M. anisopliae	0.73	1		
	C. militaris.	0.76	0.82	1	
	Abamectin	0.44	0.57	0.46	1
	B. bassiana	1			
5C dama	M. anisopliae	0.89	1		
56 days	C. militaris.	0.69	0.88	1	
	Abamectin	0.70	0.64	0.76	1
	B. bassiana	1			
70.1	M. anisopliae	0.74	1		
/0 days	C. militaris.	0.75	0.72	1	
	Insecticide	0.57	0.55	0.74	1
	B. bassiana	1			
04 1	M. anisopliae	0.89	1		
84 days	C. militaris.	0.71	0.76	1	
	Abamectin	0.50	0.67	0.64	1
	B. bassiana	1			
Total	M. anisopliae	0.91	1		
	C. militaris.	0.85	0.91	1	

Abamectin	0.66	0.77	0.79	1

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Guild group made based on function correspondence of arthropods i.e. herbivore insects, neural insects, predators, and parasitoids. For one rice cropping season, there was a tendency that the abundance of all guilds group were lower in the abamectin plot. Interestingly, the abundance of predator was always more dominant in plots sprayed with the entomopathogenic fungal bioinsecticide compared to those of herbivore insects, neutral insects, and parasitoid. Neutral insects were not found in the synthetic insecticide plot. Thus phenomenon showed that predator guild was more tolerant to the entomopathogenic fungal bioinsecticide (Bayissa et al. 2016; Firouzbakht et al. 2015; Gholamzadeh-Chitgar et al. 2017), while abamectin tent to reduce the abundance of predators, herbivore insects, neutral insects and parasitoids (Prabawati et al. 2019; Hanif et al. 2020). Among the three entomopathogenic fungi, there was a tendency that the abundance of the predators in plot sprayed with the bioinsecticide of *C. militaris* was higher than that in plots of *B. bassiana* and *M. anisopliae*. Therefore, predator arthropods tend to be more tolerant of *C. militaris* than of *B. bassiana* and *M. anisopliae*.

It could be concluded that the abundance and species diversity of arthropod predators inhabiting freshwater swamp sprayed with the bioinsecticides of *B. bassiana*, *M. anisopliae*, and *C. militaris* did not decrease, while the population of herbivore insects tent to decrease to the same level as its decrease in the abamectin plot. So that, the bioinsecticides of *B. bassiana*, *M. anisopliae*, and *C. militaris* did not decrease the abundance and species diversity of predatory arthropods (non-target arthropods) but could decrease the herbivore insect population.

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