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Diversity of Soil Inhabiting Arthropods in Intercropping of Chili and Chinese Mustard Green Applied With *Bacillus thuringiensis* Based Bio-Insecticides and Synthetic Insecticides Treatment

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Article history

Received	Received in revised form	Accepted	Available online
07 April 2020	30 August 2020	31 August 2020	31 August 2020

Abstract: Bio-insecticide is one of alternative ways in controlling plant pests with many advantages compared to synthetic insecticides, especially in environmental aspects. *Bacillus thuringiensis* is a microbial agent which most widely used as bio-insecticide to control insect pests belongs to Filum Arthropods. Biodiversity of arthropods in intercropping plants with application of B. Thuringiensis based bio-insecticide was investigated. Chili and mustard chinese green intercropped in which chili was one season and the other was two seasons. *B. thuringiensis*-based bio-insecticide and chemical insecticide were applicated on those plants. Sampling of arthropods were conducted by *pitfall trap* and *yellow pan trap* The result showed relative abundance of arthropods was higher in Bt *B. Thuringiensis*-based bio-insecticide treatments compared with chemical one. Dominance Index of second season was higher than in first season. Index of evenness resulted no difference (E < 0.5) between first season and second season shows that population level of each species was not different.
Key words : Arthropods, biodiversity, bio-insecticide, Bacillus thuringiensis

Abstrak: Bio-insektisida merupakan salah satu alternatif dalam pengendalian organisme pengganggu tanaman (OPT) dengan berbagai kelebihannya dibandingkan dengan insektisida kimia terutama terhadap aspek lingkungan. *Bacillus thuringiensis* (Bt) merupakan agen mikroba yang sampai saat ini paling banyak dimanfaatkan sebagai bahan bioinsektisida dalam mengendalikan serangga hama. Tujuan penelitian adalah untuk mempelajari keanekaragaman speseies arthropoda yang hidup pada tanaman tumpang sari cabai dan caisim. Pengamatan dilakukan dengan metode perangkap serangga yakni perangkap *pitfall trap* dan *yellow trap* pada perlakuan aplikasi bioinsektisida BT dan insektisida kimia pada lahan dengan sistem pola tanam tumpang sari cabe dan 2 rotasi penanaman caisin. Hasil penelitian menunjukkan bahwa kelimpahan relatif individu arthropoda lebih tinggi pada perlakuan bioinsektisida berbahan aktif *B. thuringiensis* dibandingkan perlakuan insektisida kimia. Indeks dominasi memiliki tren tersendiri dimana terdapat dominasi tinggi pada rotasi 2 dan spesies yang mendominasi signifikan yaitu arthropoda yang berperan sebagai predator pada perlakuan bioinsektisida. Indeks keanekaragaman dan kemerataan mempunyai angka yang tidak berbeda signifikan pada masing-masing perlakuan maupun rotasi dimana indeks keanekaragaman pada kriteria sedang dan indeks kemerataan yang relatif merata (E < 0,5) menunjukkan bahwa kekayaan individu yang dimiliki masing-masing spesies tidak jauh berbeda.
Kata kunci : Arthropoda, keanekaragaman, bio-insektisida, Bacillus thuringiensis

1. Introduction

The increasing trend of limiting-use of chemical agents in pest management has generated considerable interest in the use of natural alternatives for urban, agriculture and forestry control. Several kind of bio-insecticides have been studied and applied commercially for pest control [1]. One alternative for good and environmental friendly pest control is by using bio-insecticides [2].

Bio-insecticides have some advantages i.e. they are shorter shelf lives, required in less quantity for higher efficacy, and harmless for animals and human beings in comparison to the synthetic insecticides. They possess specific targets with specific modes of action, often affecting only a single species of insect, slow in action, and the timing of their applications is relatively critical [3].

For microbial biopesticides, the best strain used is *Bacillus thuringiensis* (Bt), and almost 90% of the biopesticides are derived from this bacterium.

Recently, only 1-2 % of the total crop protection market is occupied by bio-alternatives and most of them are *B. thuringiensis.* Researchers all over the world are being engaged in developing bio-products which easier to use, can overcome the diseases caused by dephyto-pathogens, and cover more target crops[4].

B. thuringiensis produces parasporal body and crystallin protein in its propagation with suitable media. Crystallin proteins known as delta-endotoxins were toxic to some target insects; however, it is not toxic to useful and beneficial insects, crops animals and human being [5].

Some insects belong to orders of Lepidoptera, Diptera and Coleoptera are often found to play a role as pests of vegetable crops. The level of diversity of soil insects in some places can be different. Species diversity tends to be low in the ecosystem which are physically restrained, namely that have limiting factors of physical and chemical. Soil insects present in an ecosystem is limited by geological and ecological factors resulted in different insect species diversity [6, 7].

During their growth, plants play a role as host of many herbivores and natural enemies of herbivores. For this reason, changes in the relative abundance of different plant components may produce great impacts in the spontaneous communities of arthropods [8, 9]. This study aimed to investigate diversity of arthropods found in vegetable crops with intercropping patterns applied with *B. thuringiensis*-based bio-insecticides and chemical insecticide.

2. Materials and Methods

The research was conducted in vegetable culture land owned by farmer at HM Noerdin Pandji Street, Kebun Sayur District alley, Palembang, South Sumatera Province. *B. thuringiensis*-based bio-insecticide was produced in Laboratory of Phytopathology, Department of Plant Protection, and Faculty of Agriculture Sriwijaya University.

This research was conducted from November 2018 until February 2019. Extensive field trials was equal to 4 m x 9 m meters consisted of 6 plots measuring 4.25 m x 1 m. Distance between plots was 0.5m

B. thuringiensis-based bio-insecticide was prepared in laboratory, with main media were tofu-liquid waste and molasses [10]. Chemical insecticide with active ingredient of Monosulphate was used. Bio and chemical insecticides solution were sprayed to crop once a week every afternoon at 4.00-5.00 PM.

Spraying of insecticide was done when mustard Chinese green in the age of 2 weeks after planting in each season. There were 3 times of spraying until the crops must be harvested.

Arthropods Observation

In order to collect arthropods, pitfall traps and yellow pan trays were installed in each plot. The collection of arthropods was done the day after spraying of insecticides.

Pitfall traps were made from a plastic cup with a surface circle (d=10 cm) and height of 20 cm, while yellow pan traps were made from a rectangular plastic (50 cm x 5 cm). Each trap was filled with a solution of detergent by one-third higher of the trap container.

Insects trapped were rinsed with sterile water and collected into a vial bottle containing alcohol of 70%. Furthermore, they were identified in laboratory and counted each number of species in the laboratory.

Data Analysis

Data composition of species and number of individuals of arthropods was used to determine the relative abundance (KR), Shannon Wiener diversity index, Simpson dominance index and evenness index of Pielou.

Variables observed were number of individuals of each phylum arthropods obtained, and then they were used to calculate some of the following quantities:

(i) The relative abundance [11] by the formula:

$$P_i = \frac{n_i}{N} x\ 100\ \%$$

with:

Pi = relative abundance
ni = the number of individuals
N = the total number of all individuals

(ii) Shannon Wiener diversity index [12] calculated using the formula:

$$H' = -\sum_{i=1}^{s}\left(\frac{ni}{N}\right)\ln\left(\frac{ni}{N}\right)$$

with:

H '= Shannon diversity index.
s = the number of species
ni = Number of individuals of all species i
N = Number of individuals of all species

In which:

H' < 1 Biodiversity was low (Number of individual species and low, one is no dominant species).
1 ≤ H'≤ 3. Biodiversity moderate (Number of species and individuals were moderates; number of individuals is not varied).
H' > 3 High diversity (amount individual species and higher, there is no dominant species).

(iii) Simpson dominance index [13,14] calculated using the formula:

$$D = \sum\left(\frac{ni}{N}\right)^2$$

With:

D = Dominance Index

ni = Number of individuals of all species-i

N= Total individuals of all species

In which:

D < 0.5 there are no species dominate other species or community structure in a stable condition.

D > 0.5 there are species dominated to other species or community structure unstable.

(iv) Evenness index Pielou [15] be calculated by the formula:

$$E = \frac{H'}{H_{max}}$$

E = Index of evenness

H ' = Diversity index

H*max* = the maximum diversity index (ln s)

s = Number of species

in which :

E < 0.5 Evenness among species low, meaning that the individual richness of each species is very different.

E > 0.5 Evenness relative interspecies flattening or the number of individuals of each species are relatively the same.

3. Results and Discussions

This study was conducted during two rotations planting of mustard chinese green. There were 2 types of traps with 3 times of observations on each rotation of the 2 treatments, *B. thuringiensis*–based bio-insecticides and chemical insecticides (Figure 1).

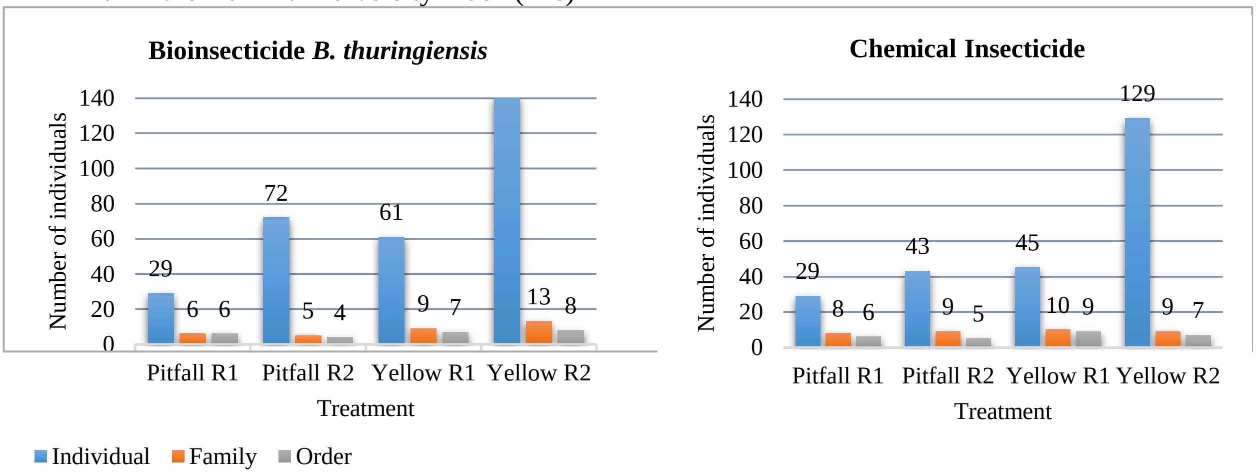

Figure 1. Comparison of the number of individuals of species, families and orders of each treatment R1: First rotation, R2 : Second Rotatio

Total arthropods captured in application of B. thuringiensis were 303 individuals while in chemical insecticide were 246 individuals. Arthropods captured mostly were on the yellow pan compared with pitfall trap. It was reasonable because the coverage of yellow pan trap was wider than pitfall trap.

Basically, ecosystem balance occurs because of interconnected each components. The components have recesses (way of life), different functions and related to each other. During these components of ecosystems performed, both the regularity of the ecosystem will remain intact [16, 17].

The presence of soil arthropods was in particular dependent on the availability of energy sources. Development and activity of soil arthropods will be prefect if environmental factors were well supported. Thus, it will have a positive impact on soil fertility reciprocally. Interaction between soil animals must occur and become part of the compiler of the food chain in the soil[18, 19].

Table 1. The relative abundance of soil inhabiting arthropods at planting patterns of intercropping.

Species	*B. thuringiensis* Bioinsecticides (%)				Chemical Insektickides (%)				Average
	PT R1	PT R2	YT R1	YT R2	PT R1	PT R2	YT R1	YT R2	
Gryllotalpa brachyptera	0	0	0	2.13	3.45	2.33	6.67	0.78	1.92
Anaxipha longipennis	13.79	6.94	1.64	4.96	13.79	2.33	15.56	4.65	7.96
Valanga nigricornis	0	2.78	14.75	0.71	0	2.33	0	0	2.57
Epilachna vigintioctopunctata	0	0	0	0.71	0	4.65	0	1.55	0.86
Aulocophora similis oliver	6.90	0	3.28	2.13	3.45	2.33	0	0	2.26
hydrophilus piceus	0	0	0	0	3.45	0	2.22	0	0.71
Necrodes littoralis	0	0	1.64	0.71	0	0	0	0	0.29
Paederus littoralis	0	0	0	0	0	2.33	0	1.55	0.48
Paraponera clavata	0	0	9.84	0.71	13.79	0	4.44	1.55	3.79
Solenopsis sp	3.45	0	0	1.42	3.45	0	4.44	0	1.59

Aleiodes indiscretus	0	0	0	0.71	0	0	0	0	0.09
Spodoptera Litura	0	0	0	0	0	0	2.22	0	0.28
Forfucula auricularia	0	0	0	2.13	0	0	0	0	0.27
Musca domestica	3.45	1.39	1.64	9.22	3.45	0	6.67	5.43	3.9
Helopeltis sp	0	0	3.28	0.71	0	0	4.44	0	1.05
Orthomorpha coartata	37.93	19.44	19.67	9.93	17.24	58.14	8.89	34.11	25.67
Trigoniulus coralinus	0	0	0	0	0	4.65	2.22	1.55	1.05
Lycosa sp	34.48	69.44	44.26	63.83	37.93	20.93	42.22	48.84	45.24
Total	100	100	100	100	100	100	100	100	

PT R1 = Pitfall Trap Rotation 1 PT R2 = Pitfall Trap Rotation 2
YT R1 = Yellow Pan Trap Rotation 1 YTR2= Yellow Rotation Tray 2

Table 1 showed arthropods abundance were dominated by spiders *Lycosa sp.* and millipedes *Orthomorpha coartata*. In all terrestrial environments, spiders occupied virtually every conceivable habitat, including the shelters and artifacts of a host of other animal[20, 21]. The effect of a spider species on a pest population may be enhanced when pray population is increasing. This was a response to a rich supply of nutritious alternative prey [21, 22].

An abundance of spiders acted as predators because of the prey abundance. Spiders were among the dominant predators of any terrestrial community. When fauna of the soil and plant cover were trapped, they will come to light in vast numbers. It was the evident that they play a significant part in the life of every habitat [21, 23].

Table 2. The relative abundance of soil inhabiting arthropod on the pattern of intercropping.

Community Characteristics	*Bt.* Bioinsecticides				Chemical Insecticide			
	PT R1	PT R2	YT R1	YT R2	PT R1	PT R2	YT R1	YT R2
Diversity Index	1.42	0.92	1.62	1.40	1.80	1.37	1.90	1.31
Dominance Index	0.38	0.69	0.44	0.64	0.38	0.58	0.42	0.49
Evenness Index	0.80	0.57	0.74	0.53	0.82	0.62	0.82	0.60

PT R1 = Pitfall Trap Rotation 1 PT R2 = Pitfall Trap Rotation 2
YT R1 = Yellow Pan Trap Rotation 1 YT R2 = Yellow pan Trap Rotation 2

Diversity index obtained in the intercropping with both application of chemical insecticide and bio-insecticide was high in yellow pan trap rotation 1 and 2. It was showed using pan trap will let more arthropods come to the traps. The surface of pitfall traps was smaller than that on pan trap therefore more arthropods were trapped. Pitfall traps were trapped. In accordance with the Shannon-Wiener diversity index stated diversity in the population belonged to the medium category (1.0 < H' < 3) if arthropods species were quite diverse and well-balanced ecosystem conditions.

Insect diversity index in this study explained that arthropods were quite diverse.
This indicated establishment of agro-ecosystem in intercropping. The higher diversity of macro fauna soil in one place, the more stable the ecosystem [11]. In general, diversity of species tends to be lower in agro-ecosystems, because it was disturbed by human activities. Comparing to natural vegetation which was still maintained and there was no human intervention [13, 15, 24].

In first rotation, index of dominance was lower (D<0.5). It was found that there was no species dominate other species or community structure in a stable condition. In second rotation of chemical and bio-insecticide application showed higher index of dominance in yellow pan trap (D>0.5). There were species dominated to other species or community structure unstable.

Dominance index showed its own trends which are almost always in rotation 2 occurs enhancement of dominance index number (D>0.5), which means that high dominance index criteria where there is a species that dominates other species or community structure in a state of instability.

The dominance index on rotation 1 is entirely on the low dominance index criteria (D <5), which means that there are species that dominate other species or community structure in a stable condition.

Trends of dominance index showed effect of the treatment against arthropods dominance remained stable in the first growing season of mustard Chinese green. There was no dominancy between pests and predator. In rotation 2, spiders dominated ecosystem and acts as a predator. The application of *B. thuringiensis*-based bio-insecticide did not lead to the dominance of insect pests but are still dominated by predators [25].

B. thuringiensis- based bio-insecticide possessed a specific target. They did not kill any natural enemies such as predators and parasitoids. *B. thuringiensis* tend to be specific attacking insect pests of the Lepidoptera order [26]. In some target insects, Cry proteins merely were sufficient to intoxicate larvae by destroying enough midgut epithelial cells [27].

Evenness index based on Table 2 showed all treatments and rotation had relatively evenly evenness index criteria with a value of E > 0.5. It means that wealth of individuals of each species did not much different. The evenness of species on ecosystems crop vegetables monoculture was lower than the ecosystem vegetable polyculture [28].

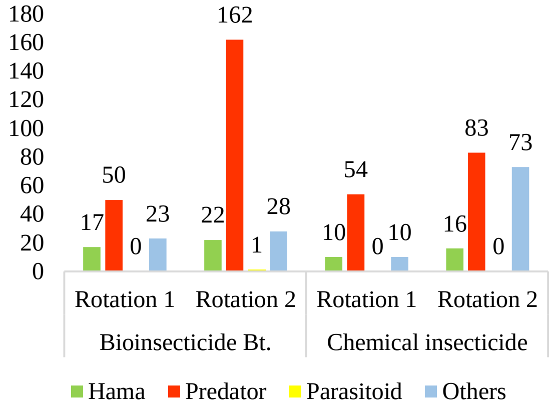

Figure 2. Number of Arthropods based on their roles in ecosystem

Figure 2 showed application of the *B. thuringiensis*-based bio-insecticide tend to increase arthropods with their role as a predator. It was predicted that the effect of treatment could suppress pests without affect individual arthropod predators. Therefore, predator population increased inversely proportionally to the arthropod pests. However, some agro-ecosystem has no favorable habitat for natural enemies due to high levels of interference. Management habitat and conservation was one of biological control approach, in which it could be supporting the existence of natural enemies in improving the biological control in agricultural systems [29].

Chemical insecticides treatment was able to suppress pest arthropods population in the first rotation, however there was an increased in second rotation. Population of predator was not different from first rotation to the next. Habitat disruption in the event of the components of habitat was subject to change. Changes in habitat could be no longer appropriate for the organism because it was different from the previous condition [19]. The excessive use of synthetic pesticides could ruin the natural balance of the ecosystem. Choosing non-selective pesticide may resulted increasing pest population due to the resistance and reduced the population of natural enemies to control pest populations [30].

4. Conclusion

Relative abundance of arthropods was higher in Bt *B. thuringiensis*-based bio-insecticide treatments compared with chemical one. Dominance Index of second season was higher than in first season. Index of evenness resulted no difference (E < 0.5) between first season and second season shows that population level of each species was not different.

Acknowledgments

The authors were grateful for the financial support provided by Professional Grant in the year 2018.

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