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THE PATHOGENICITY OF ENTOMOPATHOGENIC NEMATODES AGAINST *Spodopteraexigua*

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ABSTRACT

Spodopteraexigua is an insect pest that threat to a wide range of agricultural crops. Entomopatho genicnematodes are the parasitic nematodes that have the ability to attack the insects pest. The study to analyzed the pathogenicity of the entomopathogenic nematodes (EPN) from Steinernematidaein controlling the third instar larvae of *S. exigua.* The study's design was completely randomized design (CRD) with six treatment groups and four times of repetition (10 larvae at each repetition). *S. Exigua* were taken from the field, andmass breedingwas doneat Laboratoryfor two generation.EPN was isolated from organic soil. EPN breeding carried out using a *Tenebriomolitor* caterpillar.Third larvae of *S. exigua* were used for Bioassay test. The number of larvae killed by EPN was recorded for 96 hours.The data analysis used probit analysis from Minitab 1.5 software. The results showed that EPN gave positive response against larvae of *S. exigua* within 96 hours. The conclusion was LD_{90} of EPN against the larvae of *S. exigua*was 772 IJ/2 ml.

Keywords: pathogenicity, LD_{90} -96hours, Entomopathogenic Nematodes, *Spodopteraexigua.*

INTRODUCTION

The red onion (*Allium cepa*) is known as ingredients to make spices taste in food. One of the main production areas on *A. cepa* in Indonesia is Brebes, Central Java. The harvested area is 30954 Ha with production of onion reached 3759742Kwintal and in average was 121.46 Kw/Ha (Brebes District's Central Bureau of Statistic, 2017). *A. cepa* from Brebesare well known for their good quality. They are better than *A. cepa* from other places in Indonesia and even from other countriessuch as Thailand and China. High economic value of *A. cepa* liven up the Farmers interest on cultivating *A. cepa.* However, there werestill many problems oncultivating*A. cepa* such as the viral diseases (Gunaeni *et al*., 2011), and pest attacks (Basuki, 2014), including attack from *Spodopteraexigua* (Marhaen *et al.* 2016; Herwanto *et al*., 2012). *S.exigua* is aninsect pest that usually attacksgrowingplant in Asia, Europe, Africa, Australia, and America (Agata *et al.,* 2005). *S. exigua* larvae actively attack the *A. cepa* leaves, especially the young leaves. The damage can reach 100 percent if not controlled (Abdi, 2003).

The explotionof *S. exigua* population at field drives thefarmers to use chemical pesticide excessively (Djojosumarto, 2008; Suhartono, 2010). Using chemical pesticide excesssively lead to the distruction of environment and health problems.*S. exigua* population in Brebeshas been exposed some pesticides including hydrochloride, deltramethrin, methoxy fenozide and phyraclotose (Moekasan and Basuki, 2007;Wibisono *et al*., 2007).

Continuous use of pesticides can cause health problems, Fikri (2010) found that the there was an arsenic content in the urine of Farmer in Brebes who sprayed pesticide everyday, indicating that it was harmful to human being.

To overcome the negative effect of using chemical pesticide, it is important to conduct a study on controlling *S. exigua*pest using an effective and environmentally friendly biocontrol agent. One of biocontrol agent that can be used is Entomopathogenicnematode (EPN). EPN is endoparasiticnematodeespecialy for the insects. EPN used as biocontrolcome from the family of Steinerematidae and Heterorhabditidae (Boemare *et al,* 2002; Raucha *et al*., 2017; Indriyanti *et al*., 2017).

Steinernematidae and Heterorhabditidae are potentialbiocontrol agents for various pest insects (Ehlers, 1996). Both of them are effective on contolling pest insect from Lepidoptera, Coleoptera, and Diptera for 24 to 48 hours (Chaerani, 1996) and it is also safe for non target organisms (Grewal and Richardson, 1993). However, there is no information about controlling *S. exigua* with EPN. So, it is importantt to conducted a study in order to determine the effective dosage of EPN (LD_{90}) to control S. *exigua* in Laboratory.

MATERIAL AND METHODS

Mass breedingof *S. exigua*

Mass breeding of *S. exigua*was conducted at Biology Laboratory of Universitas Negeri Semarang. The larvae were obtained from onion farmingin Brebes District. They werereared for two generations at laboratory. Third instars larvae were used to Bioassay test.

Isolating and breeding EPN

EPN was isolated from organic soil, it was isolated by the following step: digging the organic soil in the field as deep as 20 cm, took 250 g of organic soil, and put into container (6 cm in height and 13 cm in diameter).

Tenebriomolitor caterpillar was used to trap the EPN from soil for 5-7 days. After *T. molitor* was infected

by EPN. EPN were reared using *T. molitor* caterpillar as a host in white trap method. After the process of rearing, EPN were harvested, then used for pathogenicity test (Indriyanti&Muharromah, 2016). The introduction test was conducted to determine the range of doses.The range doses of LD_{90} for 96 hours EPN was on 800 JI-1000JI/2 ml.

EPN pathogenicity test

Effectiveness test was conducted to determine the LD_{90} for 96hours. Research design was Complete Random Design (CDR), consisting of 6 treatments (P0-P5) by 4 times repetition (the number of larva in each repetition were 10). There were 24 unit experiments, total larvae were 240 (Kamariah *et al.*, 2013). The treatmentstest arrangement wereas follow:

P0 = EPN population density0 JI (Juvenile infective)/2ml aquadest (Control)
P1 = EPN population density 750 JI/2 ml aquadest
P2 = EPN population density 800/2 ml aquadest
P3 = EPN population density 850/2 ml aquadest
P4 = EPN population density 900/2 ml aquadest
P5 = EPN population density 950/2 ml aquadest

Data analyzed

The *S. exigua* mortality data for 96 hours was analyzed probit test using Minitab 1.5 program.

RESULT AND DISCUSSIONS

The death of *S. Exigua*larvae caused by EPN was characterized by changingin the body color from green to dark brown. The body texture of larvae was also became mushy.The use of third instar of *S. exigua* larvae was because it is the most active larvae (Hadi and Soviana, 2000). The result of pathogenicity test is presented on Table-1.

Table-1. *S. exigua* mortality for 96 hours.

Repetition	Treatment					
	P0 (0 JI/2 ml)	P1 (750 JI/2 ml)	P2 (800 JI/2 ml)	P3 (850 JI/2 ml)	P4 (900 JI/2 ml)	P5 (950 JI/2 ml)
1	0	7	10	10	10	10
2	0	8	8	10	10	10
3	0	8	10	10	10	10
4	0	10	10	10	10	10
Total	0	33	38	40	40	40
Average	0	8.25	9.5	10	10	10
Percentage	0	82.5	95	100	100	100

Table-1 shows that the rate of death larvae of*S. Exigua* was diversed based on EPN population density. More density of EPN given wasresulting in higher rate of larvae mortality. This result is suitable with Iskandar *et al.* (2003) & Kamariah *et al.* (2013) statement that the more density of EPN population speed up the rate of death larvae.

Ninety hours after theapplication, the larvae mortality formtreatment P3, P4, and P5 had been reached 100%. EPN (pathogenicity) effectiveness test for 96 hours showed that values of LD_{90} on P1 and P2 were 82.5% and 95% respectively. The data of *S. exigua* mortality was counted and EPN population density for each treatment was analyzed using probit test (software Minitab 1.5) to determine the value of LD_{90} and the result obtainedwas 772 JI/2 ml. The result was different with the result of study on *S.litura* by Uhan (2006). EPN population density to kill *S.litura* samples using 95% (pathogenicity) effectiveness reached 800 JI/ml. It is because *S. Exigua* and *S.litura* are two different species. So, it is possible if the effective EPN population density needed was different. Increasing mortality of *S. exigua* was showedsignificantly, it indicated that EPN have an ability to kill in a short time. It was suitable with Wagiman *et al.* (2001) who stated that the superiority of EPN as a biocontrolcan kill insects through hemolymphquickly (24-48 hours).That ability is caused by symbiont bacteria which come out from EPN after penetrate into the insect (Kaya, 1993). Steinernematidae family has a symbiosiswith bacteria of *Xenorhabdus*spp. Meanwhile, Heterorhabditidae has symbiosis with bacteria of Photorhabdussp (Boemare *et al.*, 2002). That bacteria release sometoxins (eksitoxin and endotoxin) like protease, lipase, pectinase. That combination of bacterial toxins cause the insect die faster (Dowds, 1998). However, the identification of EPN symbiont bacteria was not performed in this research.

The symptoms of *S. exigua* larval mortality were the changing in color of body from light green to be dark yellow, brownish or blackish, then themushy body texture, production of liquid and no respond of any touch. This condition indicated that the larvaehad been infected by EPN from Steinernematidae family. It was in accordance with the study of Simoes and Rosa (1996) which showed that the larvae that were attacked by Steinernematidae had a mushy dark brown body and produced a little liquid.

Figure-1. Larvae of *S. exigua* after 96 hours of application EPN.

The EPN killing ability was not only determined by symbiosis between EPN bacteria simbion but also S. exigua ability onself-defence. Ehler (1996) stated that EPN killing ability to host insects is not only determined from acomplex of EPN bacteriasimbion but also the level of immunity of host insect.

CONCLUSIONS

Based on the result of the study, it can be concluded that EPN was effective on controlling *S. exigua* pest in *A. cepa* with the value of $LD_{90}$for 96 hours was 772 Jl/2ml.

REFERENCES

Abdi N. 2003. Penggunaan Analisis Probit Untuk Pendugaan Tingkat Populasi *Spodopteraexigua* Terhadap Deltametrin Di Daerah Istimewa Yogyakarta. Jurnal Informatika Pertanian.1(2): 1-9.

Agata J. Just M V. Jadwiga Z. 2005.Characterization of A Nucleopolyhedrovirus Isolated From TheLabolatory Rearing of The Beet Armyworm *Spodopteraexigua* (Hbn.) In Poland. Journal of Plant Protection Research.44 (4): 279-286.

Basuki R. 2014. Problems Identification and Shallots Farming Analyze in the Highland at Rainy Season in Majalengka District. J. Hort. 24(3): 266-275.

Brebes District's Central Bureau of Statistic. 2017. Kabupaten Brebes Dalam Angka Tahun 2008. Brebes : Badan Pusat Statistik Kabupaten Brebes.

Boemare N E. Lanmond and MauleonH. 2002. The entomopathogenic nematodes Bacterium complex, biology, life cycle and vertebrate safety. Journal of Biocontrol Science and Technology. 6(1): 333-346.

Chaerani M. 1996. Nematoda Patogen Serangga. Bogor: Balai Penelitian Bioteknologi Tanaman Pangan Bogor.

Djojosumarto P. 2008. Pestisida dan Aplikasinya. Jakarta : Agromedia Pustaka.

Dowds B C. 1998. Bacterial Virulence Mechanisms. European Cooperation in the Field of Scientific and Technical Research.COST. 819. pp. 9-16.

Ehlers R U. 1996. Current and future use of nematodes in biocontrol: practice and commercial aspects with regard to regulatory policy issues. Journal of Biocontrol Science and Technology. 6(1): 303-316.

Fikri E. 2010. Hubungan Paparan Pestisida Dengan Kandungan Arsen (As) dalam Urin dan Kejadian Anemia. Jurnal Kesehatan Lingkungan Indonesia. 11(1): 29-37

Grewal P S and Richardson P N. 1993. Effect of application rates of Steinernema feltiae on biological control of the mushroom fly Lyccoriella auripila (Diptera: Sciaridae). Journal of Biocontrol Science and Technol. 8(1): 29-40.

Gunaeni, N.1, A.W. Wulandari, A.S. Duriat& A. M. 2011. Insiden Penyakit Virus Tular Umbi pada Tigabelas Varietas Bawang Merah Asal Jawa Barat dan Jawa Tengah. J. Hort. 21(2): 164-172.

Hadi K U dan Soviana S. 2000. Ektoparasit:Pengenalan, Diagnosis dan Pengendaliannya. Bandung : IPB.

Herwanto, E. Martono, A. Trisyono & Wahyono. 2012. Pengaruh Ekstrak Limbah Daun Tembakau Madura terhadap Aktivitas makan Larva *Spodoptera exigua*. Biosaintifika: Journal Biologi & Biology Education. 4(1):1-9

Iskandar E R. Djumali M. Yose D. 2003. Uji Coba Penggunaan Nematoda Entomopatogen Terhadap Penanggulangan Hama Penggerek Batang Gmelina. Jurnal RIMBA Kalimantan Fakultas Kehutanan Unmul. 11(1): 36-42.

Indriyanti D R. & Muharromah N. 2016. Mass Cultivation of Entomopathogenic Nematode In Artificial Media. Biosaintifika: Journal of Biology & Biology Education. 8(1): 113-120.

Indriyanti D R., Widiyaningrum P., Haryuni, Slamet M & Maretta YA. 2017. Effectiveness of *Metarhizium anisopliae* and Entomopathogenic Nematodes to Control *Oryctes rhinoceros* Larvae in the Rainy Season. Pakistan Journal Biological Sciences, 20(7): 320-327.

Kamariah R. Burhanuddin N dan Johanis P. 2013. Efektivitas Berbagai Macam Konsentrasi Nematoda Entomopatogen (*Steinernema* sp) terhadap Mortalitas Larva Spodoptera exigua Hubner. Jurnal Agrotekbisnis. 1(1): 17-22.

Kaya M G. 1993. Efficacy Against Soil-Inhibiting Insect Pests. In: Gaugler Kaya H K. (Ed) Entomopathogenic Nematodes in Biological Control. Florida: CRC Press.

Moekasan K T & Basuki R S. 2007. Status resistensi Spodoptera exigua Hubn. Pada tanaman bawang merah asal Kabupaten Cirebon, Brebes, dan Tegal terhadap insektisida yang umum digunakan petani di daerah tersebut. Jurnal Hortikultura. 17(4): 21-24.

Marhaen LS, Aprianto F , Hasyim A. 2016. Potential Mixtures Between SeNPV with Botanical Insecticides to Increase Larvae Mortality of *Spodoptera*. J. Hort. 26(1): 103-112.

Raucha H., B. Steinwenderc, J. Mayerhoferd, L. Sigsgaardc, J.Eilenbergc, J.Enkerlid, R. Zelgera, H. S. 2017. Field efficacy of Heterorhabditis bacteriophora (Nematoda: Heterorhabditidae), *Metarhizium brunneum* (Hypocreales: Clavicipitaceae), and chemical insecticide combinations for Diabrotica virgifera virgifera larval management. In Biological Control. 107: 1-10.

Simoes N and Rosa J S. 1996. Pathogenicity and Host Specificity of Enthomopatogic Nematodes. J. Biocontrol Sci and Technol. 6(1): 403-4011.

Suhartono. 2010. Keracunan Pestisida dan Hipotiroidisme Pada Wanita Usia Subur di Daerah Pertanian. Jurnal Kesehatan Masyarakat Nasional. 9 (5): 217-222.

Uhan T S. 2006. Bioefikasi *Steinernema carpocapsae* (Rhabditidae : Steinernematidae)StrainLembang terhadap Larva Spodoptera litura di Rumah Kaca. Jurnal Agric. 17(3):225-229.

Wagiman F X. Triman B. Uhan T danMoekasan K T. 2001.EvaluasiPenggunaanNematoda*SteinernemaCarpocapsae*dalamPengendalianHayati Hama SpodopterasppPadatanamanBawang.LembagaPenelitianUniversitasGadjah Mada. 40 Hlm.

Wibisono I I, Y. Andi Trisyono, Edhi Martono, A. P. 2007.EvaluasiResistensiterhadapMetoksifenozidapada*Spodopteraexiqua* di Jawa. Jurnal Perlindungan Tanaan Indonesia. 13(2): 127-135.

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