FUNCTIONAL RESPONSE OF *OPIUS CHROMATOMYIAE* BELOKOBYLSKIJ & WHARTHON (HYMENOPTERA: EULOPIDAE) PARASITOID ON LEAF MINER, *LIRIOMYZA SATIVAE* Blanchard (DIPTERA: AGROMYZIDAE)

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A B S T R A C T

Opius chromatomyiae Belokobylskij & Wharthon (Hymenoptera: Eulopidae) is one type of parasitoids with its initial existence in Indonesia is dominant in the highlands and parasitizes the leafminer, *Liriomyza huidobrensis*. Currently, the presence of *O. chromatomyiae* has been reported to be able to parasitize *Liriomyza sativae* which is the dominant pest of leafminer in the lowlands. The phenomenon indicates that *O. chromatomyiae* has potential to control the population of *Liriomyza* spp. in the highlands and lowlands. One of the potential parameters of parasitoid in regulating the host population in the nature is by determining the functional response of parasitoid on host density. The study was conducted in a greenhouse with an average temperature of 29,8ºC and humidity of 78%. Parasitoids were maintained in a cage (20 cm x 20 cm x 20 cm) which already contained the 14 days old red kidney bean plants and invested by of *L. sativae* larvae instar-3with a density of 4, 8, 12, 16, 20 and 24 leafminers. Logistic regression was applied to examine the type of functional response. Non-linear regression was used to estimate the searching time (a) and the handling time of host (*Th*). Results of the study reveal that the functional response of *O. chromatomyiae* parasitoid is included in type II. Holling's disc equation shows that the value of R^2 = 0.862 which would increase the handling of *O. chromatomyiae* host capabilities of 0,862 any time there is an increase of the level of host density. The instantaneous search rate (a) and handling time of host (*Th*) are 0,862 hours^{-1}and 1,920 hours respectively. Maximum number of larvae that can be parasitized by Holling'sdisc equation as much as 12 larvae/day. The results of this study become the basic information to prepare the control programs of leaf miner, *Liriomyza sativae* by utilizing *O. chromatomyiae* as a biological control agent.

Keywords: *Liriomyza sativae*, *Opius chromatomyiae*, functional response.

INTRODUCTION

Liriomyza sativae (Diptera: Agromyzidae) is one type of leafminers with its initial existence in Indonesia is dominant on the vegetable crops in the lowlands*. L. sativae* has polyphagia type with the host plants including plants of the family of Chenopodiaceae, Compositae, Cucurbitaceae, Solanaceae, Leguminoceae, Liliaceae, Tropaeolaceae, Umbelifereae and Violaceae (Spencer, 1973). *L. sativae* attacks are able to reduce crop production by 70% (Rauf & Shepard, 2001). The control of the use of synthetic pesticides so far have not

been able to reduce the *L. sativae* population in nature (Georghiou & Saito, 2012). Moreover, there are strict requirements on pesticide residues on the market, thus the biological control is now gaining attention as an environmentally friendly control technique (Abd-Rabou, 2006; Chow & Heinz, 2004). *Opius chromatomyiae* is one of *Liriomyza* spp., parasitoids which is dominant in the highlands and only parasitizes the *L. huidobrensis* Blanchard (Diptera: Agromyzidae). However, Wahyuni *et al*, (2016) finds that the *O. chromatomyiae* now is able to parasitize *L. sativae* in the lowlands, especially in the islands of Nusa Tenggara and in the island of Sumatra. The phenomenon indicates that *O. chromatomyiae* has

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potential as a biological control agent with broad adaptability to the location and type of host. Parasitoid potential as a biological control agentis shown from the ability of natural enemies to manage the host populations in the nature, one of which is determined by the functional response (Solomon, 1994). Functional response of a parasitoid is defined as the relationship between the number of host parasitized and prey density (Holling, 1959). Type I functional response of the host parasitized is constant so the relationship is linear. On type II, the proportion of host parasitized declines sharply with the increase in host density and the relationship is exponential. While on the type III, the proportion of host parasitized initially improved but then gradually declines with the increase in host density (Hassell, 2000). An important parameter of a functional response is searching time *(a)* and handling time of host (*Th*). Parasitoid which has a good potential as a biological control agent is a parasitoid with high *a*value and low *Th* value (Hassel, 2000). Information regardingboth parameters may provide fundamental information about the relationship between parasitoid - host, which is expected to provide the basis for the preparation of biological control strategies to reduce the population of *L. sativae* using *O. chromatomyiae* parasitoid.

MATERIALS AND METHODS

Time and place: The study was conducted from October 2015 - June 2016 at the greenhouse of Faculty of Agriculture, University of Flores.

Host Plant Propagation: The growth medium used a mixture of soil and bokashi with a ratio of 1: 0.25. It was put in a polybag diameter of 7 cm. Red kidney beans were soaked for ± 12 hours and then drained and planted in polybags. Each polybag contained 3 grains of red kidney beans, thinning was performed when the plant shad passed 7 days by leaving 2 healthy plants. Planting was performed every three days, after the plants had passed 10-14 days the leaf thinning was performed until only 3-4 first leaves remained which would be used as a host plant.

***Liriomyza sativae* Propagation:** *Liriomyza sativae* imago collected from the host plants which were taken from the field and then preserved in hatching jars. *L. sativae* and *O. chromatomyiae* imagos that appeared would be used as the parents (F1) for the next propagation. Inside the cages which already contained 12 polybags of red kidney bean plants, they were infested with 20 *L. sativae* imagos. After

24 hours, the plants infested were transferred into another cage to be maintained until *L. sativae* larvae become pupae which was about 4-7 days. The leaves of plants that have been contained of late-instar larvae were harvested then inserted into the hatching glass to produce a new individual. These activities were carried out repeatedly until the supply of host insects was met.

***Opius chromatomyiae* Propagation:** *Opius croamtomyiae* propagation was performed by inserting two pairs of *O. chromatomyiae* which were ready to copulate (3 days old) into the cage containing plants infested by *L. sativae* instar-3 larvae. Infestation was carried out for 24 hours, then the host plants that had been infested by *L. sativae* and *O. chromatomyiae* were transferred into another cage and maintained. Leaves of plants which already contained host larvae parasitized were cut at three days after infestation (dai) and transferred into a hatching glass. Parasitoid imago began to appear on the dai 14th day. This activity was performed repeatedly until the supply of parasitoids wasmet. The test insects used were the parasitoid of the second offspring (F2).

Functional response of *O. Chromatomyiae* on Host Density of *L. Sativae:* Red kidney bean leaves were infested by *L. sativae* instar-3 larvae with a density of 4, 8, 12, 16, 20 and 24 larvae, inserted separately into each cage. Further, inside the cage, each one pair of three days old *O. chromatomyiae* was removed for 24 hours. Parasitoids were fed by 10% honey smeared on the walls of the jar. After 24 hours, the leaf bend was hatched to count the number of parasitoids which were successfully laid eggs. The experiments were performed with three replications on each host density.

Data Analysis: The estimation of *a* and *Th* value was performed by logistic regression analysis between the number of host parasitized with the amount of prey available (Juliano, 1993). Logistic equation used was defined as follows:

$$\frac{Na}{N} = \frac{\exp\ (P_0 + P_1 N + P_2 N^2)}{1 + \exp(P_0 + P_1 N + P_2 N_2)} \ \dots\dots\dots\dots\dots\dots\dots\dots\dots\ 1$$

Where Na was the number of host parasitized by a parasite in 24 hours, N is the initial host density and P0, P1, P2 were the model parameters that must be estimated by maximum likelihood method. Determination of the functional response model type (Type II or III) was based on the characteristic of the relationship curve between the number of host parasitized on the amount of prey available (Na/N) and N. On the Type III functional

response, Na/N decreased with the increase in N value. This condition would be seen in a logistic regression equation when the coefficient of negative (P1) linear component. Type III functional response, Na/N increased when N was still low and further decreased when N reached the medium to high value and would be visible in the logistics equation with coefficients of positive P1. Therefore, when the results of logistic regression analysis provided negative P1 value and significantly different from 0, then the model was included in the type II functional response. Conversely, if the results of the analysis provide positive P1value and significantly different from 0 then the model was included in the type III functional response. The determination of the estimator value for the two main parameters of functional response, namely *a* and *Th* used the method of non-linear least squares regression. In this experiment the number of host was not maintained constantly during the exposure time (T = 24 hours), then in the analysis using a functional response of disc model (Holling, 1959) as follows:

$$Na = \frac{a.T.N}{(1+a.Th.N)} \dots \dots \dots \dots 2 \text{ (type II equation model)}$$

Na = N {1 – exp[(d + bN)(Th-T) / (1 + cN)]} .....3 (type III equation model).

With b, c and d were constant parameters that became the part of the instantaneous search rate (*a*).

RESULTS AND DISCUSSION

The ability to parasitize *O. chromatomyiae* parasitoid on the increase in the host density of *L. sativae* shows a significant result of the increased amount of host density that is F = 13.57; db = 4; P <0.021 (Table 1). The condition shows that the greater the number of the host that is exposed, the more host parasitized. At low host density (4 larvae) *O. chromatomyiae* is able to parasitize by 83% - 100%, one of the effective parasitoid characteristics is capable to search and parasitize the host when the host population is low (Pervez &Omkar, 2005). The maximum number of hosts parasitized occurs in host density of 20 leafminers however, the decrease in the number of host parasitized occurs on the number of host density of 24 leafminers (Table 1).

Table 1. Mean host of *L. sativae* parasitized (x ± SD) by *O. chromatomyiae* parasitoid.

Density of host larvae	Mean Host parasitized
	L.sativae
4	3,678 a ± 0,577
8	4,333 a ± 0,577
12	5,678 b ± 0,577
16	6 ,000 b ± 1,000
20	7,667 c ± 0,577
24	7,333 c ± 0,577

Description

- Numbers in the column followed by different letters show significant difference in Duncan's test of 5% level.
- Data has been transformed by Arc sin $\sqrt{x}$

The results of the analysis of non-linear logistic regression were conducted to determine the model of functional response to the effects of host density with many host parasitized. Table 2. shows the value of the coefficient in the linear equation is negative at each treatment indicating that the functional response models, including type II model (Hassell, 2000). Value of the model suitability could also be seen from high R value and low standard error value (Juliano, 1993). Estimator value for P0, P1 and P2 are presented in Table 2.

Based on the estimator value on the parameters above, the curve of functional response relationship between the number of hosts available and the number of host parasitized can be described. Type II functional response is generally obtained in the experiments in the laboratory or a cage in the field with one host species provided (Schenk & Baker, 2002; Pervez &Omkar, 2005). This is related to the ease of parasitoids in finding a host in a limited area. Curve of the relationship between the host density and the number of hosts parasitized shows that the number of host parasitized increased along with the increasing number of hosts. The number of host parasitized by *O. chromatomyiae* on *L. sativae* host is maximum at the host density of 20 leafminers but the number of host parasitized shows a decrease in the density of 24 leafminers.

Table 2. The results of logistic regression analysis of the proportion of *L. sativae* larvae parasitized by *O. Chromatomyiae.*

Parameter	Standard Error	R^2	P	Type of functional response
Linear : Y = -0,025x +0,861	0,303	0,977	0,003	
Quadratic: $Y = 0,917e^{0,04x}$	0,230	0,988	0,228	Type II
Logarithmic: Y = 0,32 ln(x) + 1,297	0,283	0,961	0,095	
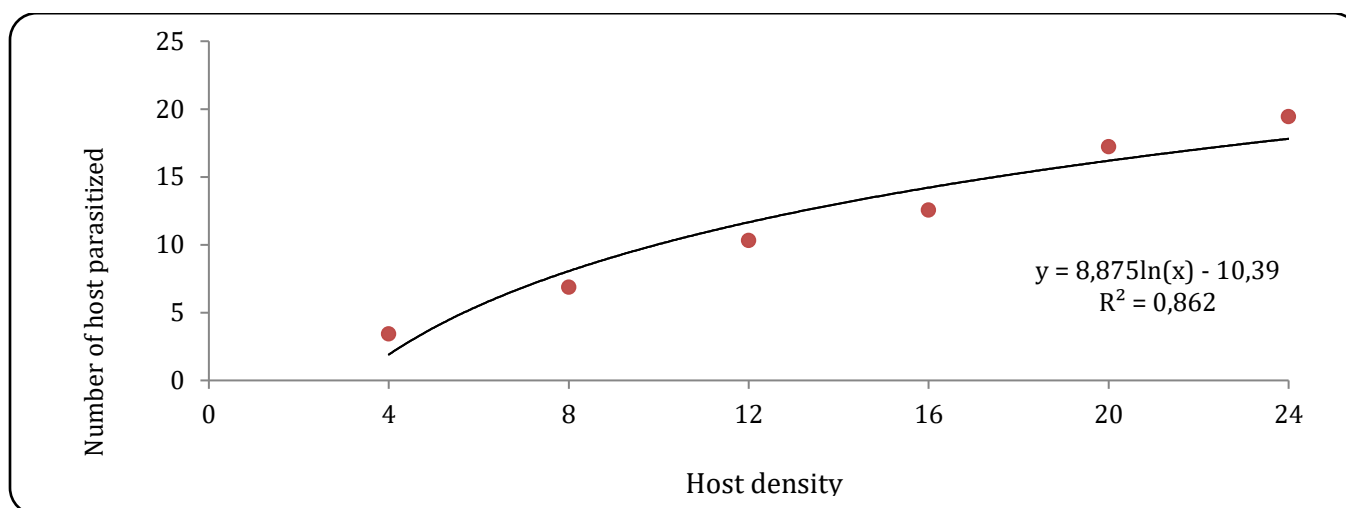
Figure 1. Functional Response of *O. chromatomyiae* on *L. sativae* density.

Functional response curve in Figure 1 shows that the number of host parasitized increases with the increasing of the host density. In addition to handling time of host, an important thing to determine the level of host parasitized is egg load (Heimpel& Rosenheim, 1998). Based on the model of functional response equation in Table 2 above, the value of the estimator value of searching time (*a*) and handling time of host (*Th*) is performed through Holling's disc equation approach. Selection of appropriate functional response model for the parasitoids depends on the behavior of female insects in finding the host. Equation disc is more appropriate to be used on a parasitoid that is able to distinguish the status of the host that is whether the host has been parasitized or not (Wang &Warsito, 1998). In this study, superparasit event by *O. chromatomyiae* is not found. This is due to the *O. chromatomyiae* ability to distinguish the host status (Wahyuni, 2006). The searching time (*a*) of *O. chromatomyiae* (0.862 hours[-1]± 0.110) is faster and the handling time of host (1.920 ± 0.007 hours) is shorter than the several other species of *Liriomyza* spp. parasitoid. The searching time *(a)* on *O. chromatomyiae* is 0.05 hours[-1] and the handling time of host(*Th*) is shorter

3.155 hours than *Hemiptarsenus varicornis* (Rustam *et al.,* 2007) while a value of *O. chromatomyiae* is faster 0,017 hours[-1] and 0,018 hours shorter than *O. dissitus*. The high value of handling time (*Th*) indicates the speed of parasitoids to be able to handle a host. Different host size allows the occurrence of speed difference in the host discovery. A quite large larvae host shareable to produce a stimulus to the parasitoid in the form of vibration caused by the movement and lead the parasitoid larvae to find a host quicker. This is also supported by the opinion of Pervez &Omkar (2005) who state that the difference in the value of the functional response parameter which is searching time (*a*) and the handling time of host (Th) may be caused by differences in the host size and parasitoid moving speed. The equation on the functional response curve (Figure 1) shows that every change of one unit of the host density will improve the handling capabilities of the host on the *O. chromatomyiae* amounted to 0,862 on *L. sativae.* Nevertheless, these results should be equipped with a numeric response through the release of parasitoid inundation in nature so *O. chromatomyiae* is able to regulate the *L. sativae* population in the nature.

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