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Encapsulation and Hemocyte Numbers in *Crocidolomia pavonana* and *Spodoptera litura* Fabricius (Lepidoptera) Attacked by Parasitoid *Eriborus argenteopilosus* Cameron (Hymenoptera)

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***Eriborus argenteopilosus* is the most important parasitoid attacking cabbage pest *Crocidolomia pavonana* in Indonesia. Previous studies proved that parasitoid encapsulation was found to be an important factor limiting the effectiveness of the parasitoid in controlling pest population in the field. Since 1998, we have conducted series studies to investigate encapsulation mechanism developed by hosts against parasitoid, responses of parasitoid toward encapsulation ability and to determine factors that may help parasitoid avoid encapsulation. Parasitoid responses were examined on two different hosts *C. pavonana* and *Spodoptera litura*. Our findings showed that parasitization level was found to be high both on *C. pavonana* and *S. litura*. Encapsulation occurred to be high in all larva stages of *C. pavonana*, in contrast encapsulation was recorded very low in all larvae stages of *S. litura*. We recorded that encapsulation in the larval body of *C. pavonana* was completed in 72 hours and mostly occurred in higher larval stage. Melanization was only recorded in encapsulated parasitoid inside larva body of *C. pavonana*, not in *S. litura.* We recorded that encapsulation increased blood cell number of both larvae *C. pavonana* and *S. litura*. Encapsulation may affect development of immature parasitoid. Weight of *S. litura*'s pupae containing encapsulated parasitoid was found to be lower in *S. litura*, but not in *C. pavonana.* Our investigation also proved that superparasitism may help parasitoid avoid encapsulation.**

Key words: parasitoid, encapsulation, melanization, blood cell number, superparasitism

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INTRODUCTION

Encapsulation has been envisaged as one factor contributes to the failure of parasitoid's work in controlling pest population in the field (Hadi 1985; Goodfray 1994; Blumberg 1997; Sagarra 2000; Alleyne & Wiedenmann 2001). Encapsulation is widely defined as a common and an active physiological defense mechanism that is an immune response of the host against the intrusion of an external element, such as eggs of parasitoid and other foreign organism inserted into the host haemocol (Godfray 1994; Strand & Pech 1995; Pech & Strand 1996; Blumberg 1997; Quicke 1997; Chapman 1998; Sagarra *et al.* 2000). The successful development of an immature endoparasitoid is strongly influenced by host's defense system and the parasitoid's ability to evade the systems. Encapsulated parasitoid may be die by suffocation, starvation, or physical prevention of development (Blumberg 1997). Encapsulation involves host hemocytes that recognize the invader as non-self, subsequent of more hemocytes, and adherence of hemocytes to the invader's survace, eventually resulting in a multicellular capsule that kills the parasitoids (Godfray 1994; Pech & Strand 1996; Alleyne & Wiedenmann 2001). Partial encapsulated parasitoid may survive and continue to grow normally (van den Bosch 1964).

Encapsulation was reported to occur in many species of insect, such as in three families of Coccoidea (Homoptera), Coccidae (*Soft scale insects*), Diaspididae (*armored scale insect*), and Pseudococcidae (*mealybugs*) (Blumberg 1997; Blumberg & Driesche 2001), in *Hypera postica* (Coleoptera: Curculionidae) (Berberet *et al.* 2003), and in *Drosophila* (Nappi 1975; Rizki & Rizki 1984). Sagarra *et al.* (2000) reported that the hibiscus mealybug, *Maconellicoccus hirsutus* develops a cellular defense reaction that involves encapsulation and melanization of the endoparasitoid egg *Anagyrus kamali* (Encyrtidae). Encapsulation was also recorded in three Lepidopteran stemborers attacked by *Cotesia flavipes*-complex endoparasitoids (Alleyne & Wiedenmann 2001).

Eriborus argenteopilosus is the most important parasitoid attacking cabbage pest *Crocidolomia pavonana* in Indonesia. The parasitoid was also reported to attack several hosts such as *Helicoverpa armigera* (Kalshoven 1981), *Spodoptera litura,* and *Spodoptera exigua*. Hadi (1985) reported that encapsulation was found to be the most important factor limiting parasitoid's capability in controling the cabage pest in the field. Unfortunately, up till today, in Indonesia no study has ever been conducted to evaluate how encapsulation develops and works against parasitoid invasion and how is the response of parasitoid toward encapsulation ability. The objectives of the research were to explain encapsulation process and its impact on parasitoid immature development,

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to identify its effects on number of blood cells of parasitized hosts, and to study the parasitoid's counter measures to avoid encapsulation (e.g supperparasitism).

MATERIALS AND METHODS

Rearing of Hosts *C. pavonana* and *S. litura*. Larva *C. pavonana* was collected from cabbage plantations in Cianjur and *S. litura* was from soybean plantations in Bogor. Larvae were then brought into the laboratory for rearing. Larvae were maintained inside a plastic box (35 x 25 x 7 cm^3). In order to facilitate air circulation, plastic boxes were equipped with mesh window (25 mesh). One plastic box is only allowed to contain the same stage of larvae from the same species. Larvae which are getting pupation were removed into a plastic container containing sterilized wood dust and those containers were placed inside adult wood cages (40 x 40 x 40 cm^3). Emerged moths were than removed into adult cages and maintained with honey sollution (10%). Soybean leaf was placed inside the adult cages for oviposition. The same rearing treatment by using broccoli leaf was also applied for *C. pavonana*. Eggs laid by female moth were collected and incubated under room temperatur. Emerged larvae were used for experimental test and rearing.

Rearing of the Parasitoid *E. argenteopilosus*. Parasitoid *E. argenteopilosus* were collected by using sweep net from cabbage plantations in Cianjur, West Java. The adults were reared by exposing mated female on its host *C. pavonana* or *S. litura* inside the rearing cages for 24 hours. Parasitized hosts were then separated from female parasitoid and removed into a plastic box (35 x 25 x 7 cm^3). Parasitized hosts were maintained by using broccolli leaf until pupation. Emerged parasitoids were collected 8-9 days after pupation. Two-days old, mated females were used for experiment and for rearing purpose. Female parasitoids were considered experienced as they had been exposed upon emergence to different stages of host in the rearing cages for 24 hours.

Experimental Procedure. In prelimary test, two species of the parasitoid's host *C. pavonana* and *S. litura* were used. All immature stages of *C. pavonana* and *S. litura* were tested, however L4 and L5 of *S. litura* were not suitable for oviposition since their size are too big for parasitoid to attack. In this experiment, only active and normal parasitoid allowed to be used for oviposition. All tests were conducted under room temperature (27-30 ^oC).

Parasitization Tests. Parasitization test was conducted by exposing mated active female parasitoid on 20 larval of *C. pavonana* and *S. litura*. All larvae stages of *C. pavonana* were used for parasitization test (L1-L4), in the other side, only L1-L3. (1st instar) of *S. litura* were used. Higher stages of *S. litura* larvae were not suitable for parasitization due to higher size. Larvae were exposed on parasitoid for 24 hours, and then all larvae were dissected individually to count the parasitization level.

Encapsulation Ability of Two Hosts *C. pavonana* and *S. litura*. To evaluate encapsulation ability of two different hosts, approximatelly 25 larvae were directly exposed on active female parasitoid. Parasitized larvae were removed from the parasitoid and were maintained under laboratory condition for 72 hours. All parasitized larvae were dissected under binocular microscope and number of hosts containing encapsulated parasitoid were counted as a measurement host's capability in encapsulating parasitoid. All examination was set up for 10 replications.

Response of Host Immune Againsts Parasitoid: Chronology of Encapsulation Dissections. Twenty larvae of *S. litura* and *C. pavonana* were exposed individually on active female parasitoid under transparent glass tube (t = 20 cm, d = 3 cm) and after oviposition, hosts larvae were then removed and placed into plastic containers (t = 20 cm, d = 9 cm). The parasitized hosts were maintained with broccoli leaf for rearing until dissection. On each day that hosts were parasitized, randomly selected hosts were assigned to different time points at which they would be dissected. Both of host species were then dissected in a drop of ethanol (70%) at 0, 3, 6, 12, 18, 24, 48, 72, 96, 120, 144, and 198 h after removal of the parasitoids to observe the dynamics of the immature parasitoid encapsulation. However, different larvae stages or species, will be suitable for different dissection period since older hosts need shorter time to get pupation. Dissection will only be allowed to be conducted before hosts getting pupation. At each time interval, a total of 10 hosts of each stage and each species were dissected. Number of hosts containing encapsulated parasitoid were recorded and size of immature parasitoid were measured to determine the development of immature parasitoid.

Total Hemocyte Counts. Second Larvae (L2) of *S. litura* and *C. pavonana* were individually exposed to the active female parasitoid. After parasitization larvae were placed into plastic container with brocolli leaf and were observed daily until the scheduled assay time. One cohort consisted of 20 parasitized larvae and 20 control larva at the same age. Hemocyte numbers were determined at 3 and 5 days after parasitization. Hemolymph from a parasitized larvae was assayed at the same time as the hemolymph of unparasitized control larvae of exactly the same age. Hemolymph was squeezed from a cut proleg by using micropipette for 10 µl. Hemolymph was soluted with buffer chloride phosphat 1:10 (0.15 M NaCl, 5 mM KPO_4, pH 6.5) and was transferred with micropipettte to the Neubauer hemocytometer (Stoltz & Guzo 1986). The treatment was set up for 10 replications.

Effects of Encapsulated Parasitoid on the Weight of Survived Host's Pupae. First and second larvae of *C. pavonana* and *S. litura* were individually exposed to ovipositing *E. argenteopilosus* as previously described. All parasitized hosts then were maintained under laboratory condition. The parasitized larvae were observed daily and number of survived pupae were recorded. At least 10 survived pupae were balanced. In this test, weight of survived parasitized pupa and unparasitized pupae were measured.

Data Analyses. Statistical analyses of the data were performed using the software Statistica for Windows 6.0 (Statsoft 2001). Pearson's correlations were conducted when data proved to be normally distributed, otherwise Spearman rank correlations were used (Sokal & Rohfl 1995). Analysis of variance (ANOVA) was used to test for differences between

two or more groups. Differences between means were tested for significance by the Scheffe' test at (P = 0.05).

RESULTS

Parasitism Rate of Parasitoid *E. argenteopilosus* under Laboratory Condition. Parasitization represents the ability of parasitoid in controling its hosts under laboratory condition. Parasitization *E. argenteopilosus* on its host *C. pavonana* ranged between 27-50% (±67.70%) in L1, 50-100% (±78.96%) in L2, 5-77% (±52.91%) in L3, and 0-41% (±24.88%) in L4. Our analyses identified that parasitization was significantly affected by growth phase of host ($F_{3,36}$ = 12.34; P < 0.0001, n = 40). Parasitization level was recorded to be lower in the older stage compared with the younger one (Figure 1). This was also confirmed by conducting *Spearman rank correlation* between larval stages and parasitization. Significant negative correlation was found between those parameters (*Spearman rank correlation*: n = 40, R *Spearman* = -0.59, P < 0.0001), parasitization level decreases with increasing larval stages (not shown). In contrast, we did not find that growth phase affected the parasitization of the Parasitoid on *Spodoptera larva* ($F_{2,27}$ = 0.18; P = 0.835, n = 30).

Encapsulation Capability of *C. pavonana* and *S. litura*. Number of larvae containing encapsulated immature parasitoid was recorded to be higher in *C. pavonana* compared with *S. litura*. Encapsulation reached more than 80 percent in *C. pavonana*, conversely, encapsulation was recorded under 15 percent in all tested larval stages of *S. litura*. Number of larvae containing encapsulated immature parasitoid was significantly affected by host stage both in *C. pavonana* ($F_{2,27}$ = 26.90, P < 0.0001, n = 30) and in *S. litura* ($F_{2,27}$ = 10.25, P < 0.0005, n = 30). Older stages were recorded to have a higher capability in parasitoid encapsulation than the younger stages. Number of larvae *C. pavonana* containing encapsulated parasitoid was found to be lower in L1 compared with L2 and L3. Similar result was also documented for *S. litura*, number of larvae containing encapsulated parasitoid was found to be lower in L1 and L2 than the older stage (Figure 2).

Host Immune Response. Encapsulated immature parasitoid (larva or egg) can be clearly observed in the larvae body of *C.*

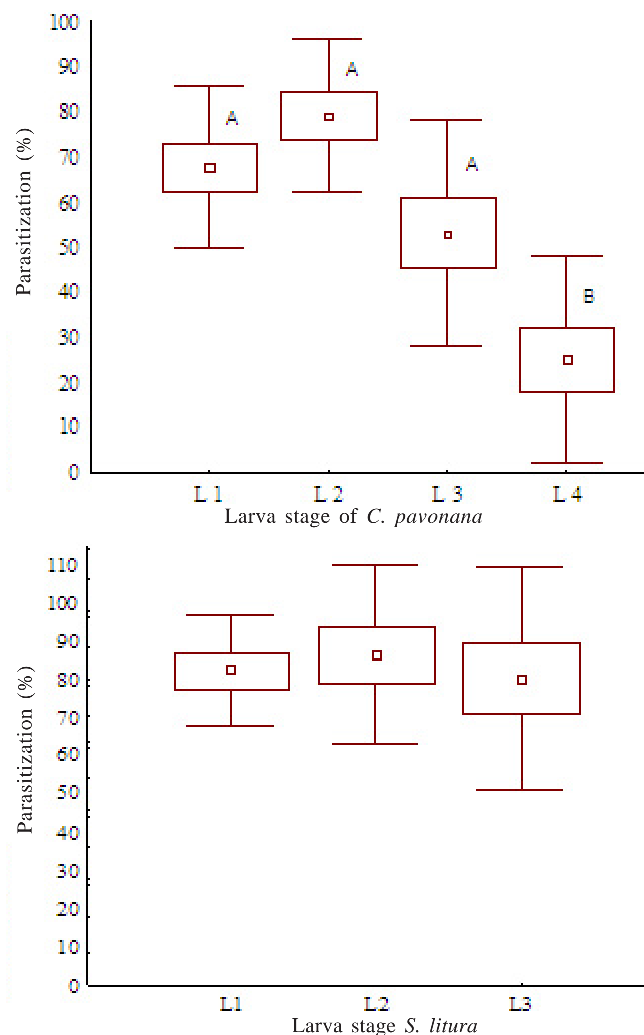

Figure 1. Parasitization level of Parasitoid *E. argenteopilosus* on its hosts *C. pavonana* and *S. litura* in different growth phases. : + Std. Dev, : + Std. Err, : Mean.

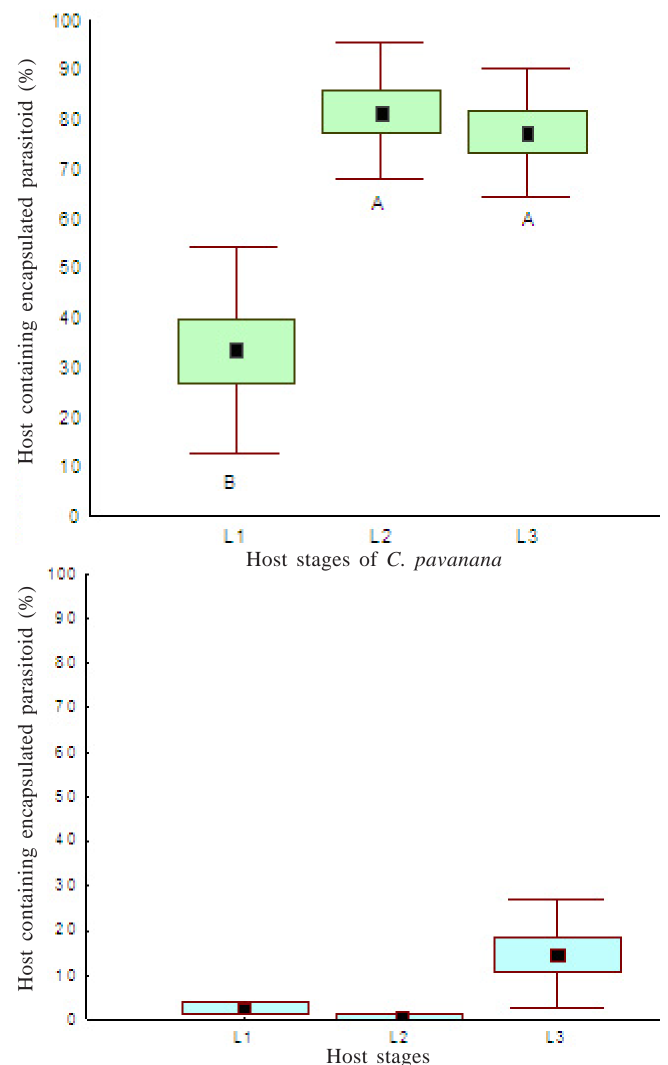

Figure 2. Number of larvae containing encapsulated parasitoid in *C. pavonana* and *S. litura*. : + Std. Dev, : + Std. Err, : Mean.

pavonana. The indication of immature parasitoid encapsulation was recorded from the formation of accumulated circle materials surrounding the object shaping a ring-like whose color changed gradually from transparent to yellowish-brown (amber). Encapsulation proccess continued by initiation of capsule shape from aggregates of melanized material which formed spots that gradually spread over the entire immature parasitoid surface. In this phase, capsule covering the object completely. Finally, multicellular capsule-like envelope coating the object was formed. The size of the melanized capsule increased with increasing periode of time.

The process of encapsulation inside the larva body of *C. pavonana* was started by the formation of circle materials spread over shaping a ring enclose the immature parasitoid. This initiation was started in different period of time for different larval stage. In L4, the initiation was started three hours after eggs oviposited by female parasitoid. In contrast, in L2/L3, the process can be observed about 18 hours after ovipostion. The number of circle materials increased with increasing periode of time, forming aggregate materials enveloping the immature parasitoid. The color of encapsulated material was gradually changed from transparent to amber. Melanization was recorded after the formation of multicelullar capsule-like envelope completed. The color of melanized material gradually changed from light-yellow into dark orange. Encapsulated parasitoid become a smaller material in the post-encapsulation. In the larvae of *S. litura*, melanization was not recorded and parasitoid encapsulated was rarely found.

Initiation of encapsulation process vary for different host species, larval stages, and individu of the same host. Initiation of encapsulation can be observed anytime wether at egg stage or larval stage of immature parasitoid. Partial encapsulation can obviously be found only when encapsulation was initiated at the larval stage. Number of encapsulated parasitoid increased with increasing period of time and reaching the peak 72 h after oviposition. Approximatelly, 90% imature parasitoid inside the larva body of L2 and L3 was encapsulated, and only 50% recorded for L1 in this dissection period (Figure 3). Encapsulated parasitoid decreased in size and become smaller or very smaller dead material. Number of encapsulated parasitoids decreased after dissection period for 72 h.

Effects of Encapsulation on the Development of Immature Parasitoid: Host Stage or Species? Body length of immature parasitoid developed inside L1's body was recorded to be higher than in older stages of host. This can be clearly seen after parasitoid eggs hatched and developed become young larvae (48 h after oviposition). Development of parasitoid egg size (before 24 h), did not prove to be significant between L1, L2, and L3. In contrast, larvae size was recorded to be higher in L1 than in older stages, at the dissection periods of 48 and 72 h after oviposition (Figure 4). The development of body length of immature parasitoid was found to be higher in L1 of *C. pavonana* than in *S. litura*, conversely, in L2, body length size of immature parasitoid was recorded to be higher in *S. litura* than in *C. pavonana* (Figure 5).

Hemocyte Numbers. Number of blood cell was recorded to be higher in parasitized larvae than unparasitized larvae of both *C. pavonana* (Day-3: $F_{1,18}$ = 19.97, P = 0.0003, n = 20; Day-5: $F_{1,18}$ = 18.09, P = 0.0005, n = 20), and *S. litura* (Day-3: $F_{1,18}$ = 7.62, P = 0.013, n = 20, Day-5: $F_{1,18}$ = 18.17, P = 0.0005, n = 20). Increasing period of time was followed by increasing number of blood cells. Number of blood cell involved in the response of host toward parasitoid invasion was found to be higher in larvae *C. pavonana* than *S. litura* (Figure 6).

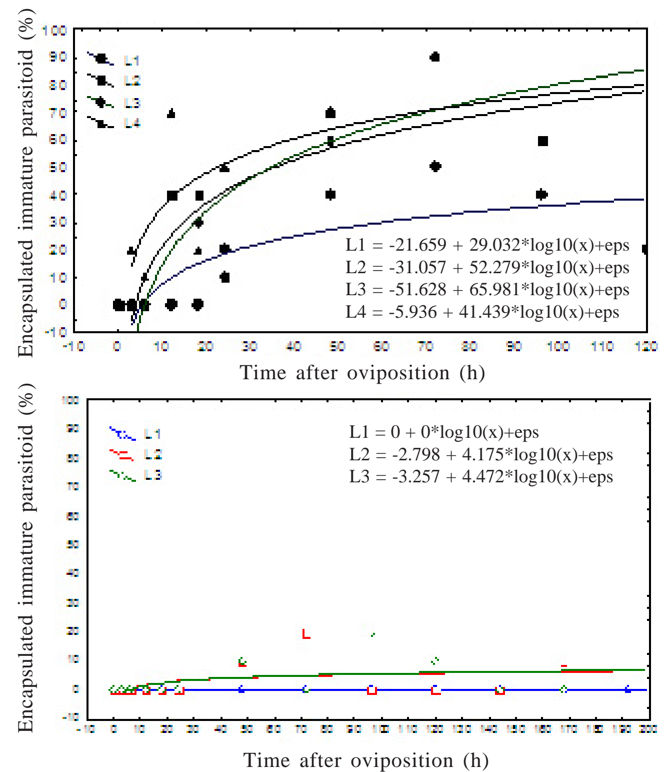

Figure 3. Percentage of encapsulation of immature stage of *E. argenteopilosus* as a function of time.

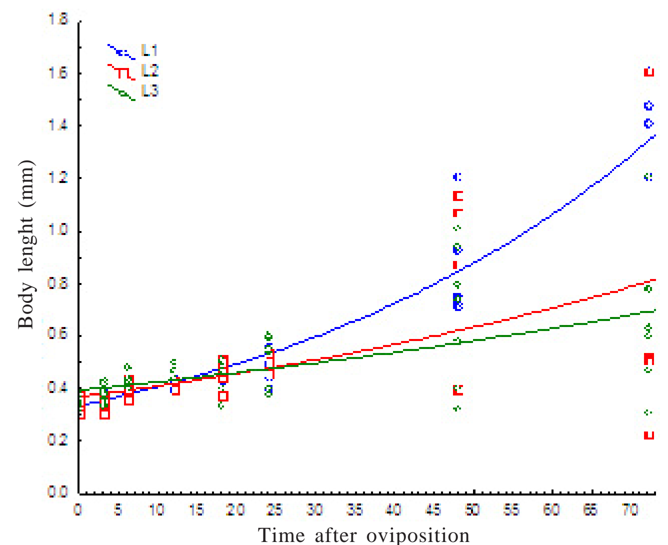

Figure 4. Body length (mm) of immature parasitoid inside larva body of L1, L2, and L3 *C. pavonana* in different period of time.

Effects of Encapsulated Parasitoid on the Weight (gr) of Survived Host's Pupae. Parasitized larvae which have can resist parasitism may survive to grow until pupae. Effects of encapsulated parasitoid on weight of survived pupae can be

observed by comparing the weight of pupae between unparasitized and parasitized pupae. There was no significant different found between weight of parasitized pupa and unparasitized pupae of *C. pavonana* (L1: $F_{1,18}$ = 1.22, P = 0.284, n = 20; L2:$F_{1,18}$ = 0.105, P = 0.749, n = 20). In contrast, significant different was recorded between parasitized pupae and unparasitized pupa of *S. litura* (L1:$F_{1,18}$ = 51.997, P < 0.0001, L2:$F_{1,18}$ = 21.56, P = 0.0002) (Table 1).

Does Superparasitism Contribute to Avoid Parasitoid Progeny from Encapsulation? Superparasitism was recorded to be occurred on female parasitoid *E. argenteopilosus* on its host *C. pavonana*. Superparasitism was occurred in a high rate between 62 and 72% in average. Statistical analyses did not find a significant different between L1, L2, and L3 with respect to superparasitim occurrences ($F_{2,27}$ = 2.22, P = 0.127, n = 30). Number of eggs observed per host individual ranged between 1 and 5 eggs, with average of about 2 eggs per host. Mean number of eggs laid by female parasitoid per host was not significant between larval stages. ($F_{2,27}$ = 1.167, P = 0.327, n = 30). In the superparasitized host, at least, we can find one egg escape from encapsulation mechanism developed by host. Mean number of survived immature parasitoid after 72 h was not recorded to be significant between larval stages ($F_{2,27}$ = 0.77, P = 0.47, n = 30) (Table 2).

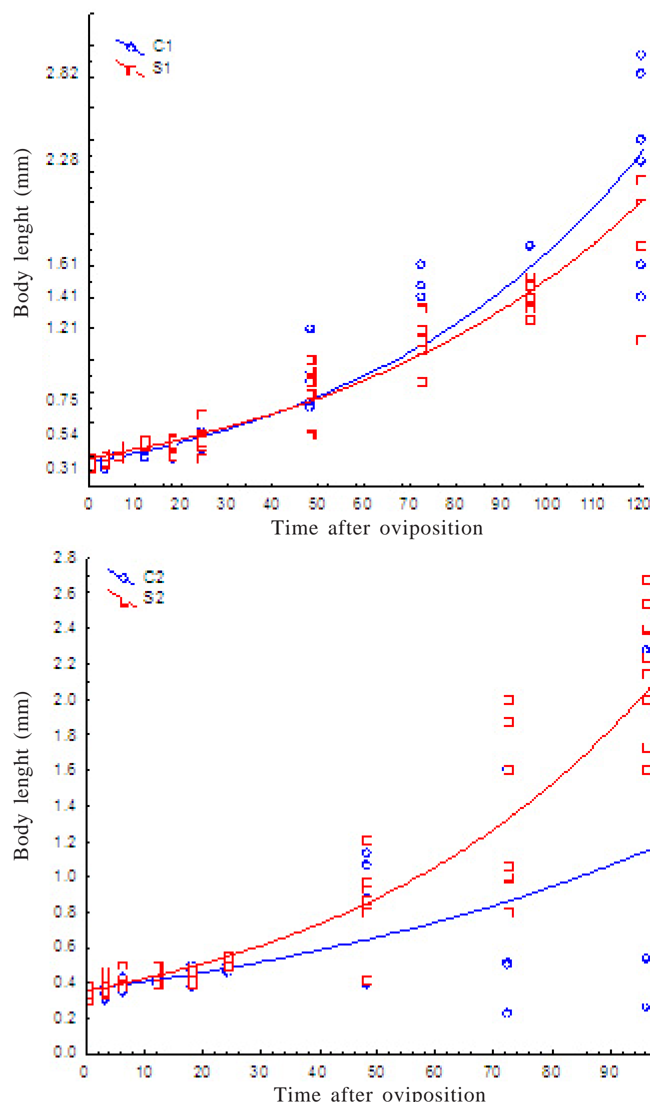

Figure 5. Body lenght of immature parasitoid inside L1 and L2 of *C. pavonana* (C) and *S. litura* (S).

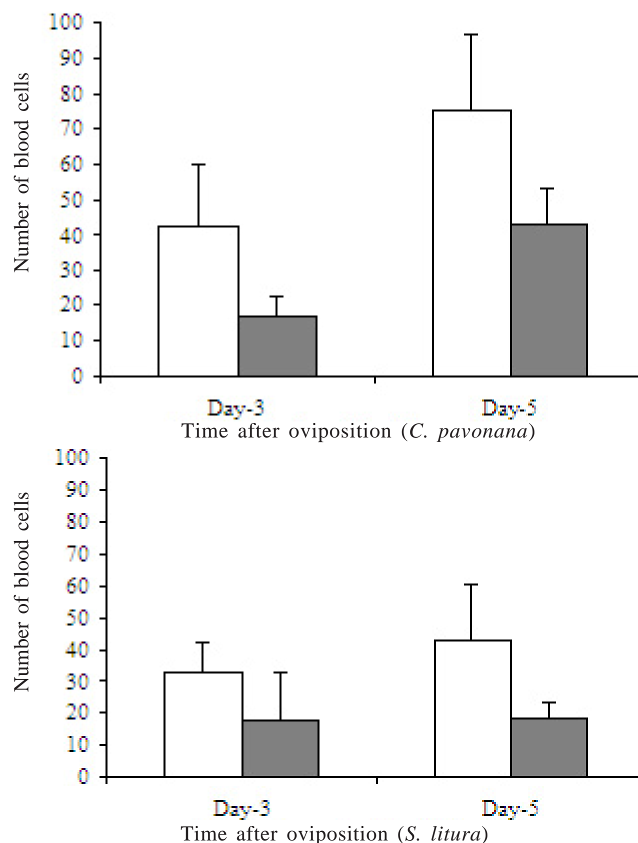

Figure 6. Number of blood cell of parasitized and unparasitized larvae three days and five days after oviposition. □: parasitized larvae, ■: unparasitized larvae.

Table 2. Number of eggs laid by female parasitoid in superparasitim incidence inside the larval body of *C. pavonana*

Stage	Superparasitism (%)	Mean number of eggs laid by female parasitoid per hosts	Mean number of survived immature parasitoid after 72 h
L1	71.79 ± 29.85a	2.11 ± 0.84a	0.77 ± 0.64a
L2	68.15 ± 19.17a	2.31 ± 0.88a	0.36 ± 0.53a
L3	62.38 ± 24.01a	2.13 ± 0.66a	0.64 ± 0.97a

Table 1. Weight (gr) of parasitized and unparasitized pupae of *C. pavonana* and *S. litura*

Hosts	L1		L2	
	Parasitized	Unparasitized	Parasitized	Unparasitized
Crocidolomia binotalis	0.0375 ± 0.004a	0.0346 ± 0.007a	0.0415 ± 0.004a	0.0408 ± 0.006a
Spodoptera litura	0.1001 ± 0.0662a	0.2106 ± 0.0359b	0.1087 ± 0.0384a	0.2256 ± 0.0341b

DISCUSSION

The capability of parasitoid in controling its hosts can be observed from the parasitization level. This research proved that parasitoid *E. argenteopilous* has a high capability in controling its hosts, this can be seen from a high percentage of parasitization on its two hosts *C. pavonana* and *S. litura*. Encapsulation as a host defense mechanism may confine the effectiveness of parasitoid in suppressing pest population in the field (Hadi 1985; Blumberg 1997; Sahari 1999; Sagarra 2000; Alleyne & Wiedenmann 2001). Previous studies identified that the failure of the parasitoid in controlling *C. pavonana* in the field may be related to encapsulation mechanism developed by the host (Hadi 1985). This study confirmed those findings that *C. pavonana* has a high capability in encapsulating immature parasitoid inserted by female parasitoid. Encapsulation of inserted immature parasitoid was recorded in all stages. However, with respect to encapsulation, the effectiveness of parasitoid may depend on host species. This study found that encapsulation was rarely found in other host species *S. litura*. Encapsulation was recorded to be very high in younger larvae and this mechanism can be observed earlier in the older stages. This may be related to the vigor of host both physiologically and physically. Older hosts may have a stronger immune response and better fitness than younger hosts (Godfray 1994). This was confirmed by Saggara *et al.* (2000) who reported that older stages of host have a mature immune system to encapsulate parasitoid.

Encapsulated parasitoid can be recognized by the formation of melanized material aggregates surrounding the immature parasitoid (Sagarra *et al*. 2000; Reed *et al*. 2007). The process of encapsulation involves many types of blood cell which work in recognizing and encapsulating invading objects (Gupta 1991; Chapman 1998). Encapsulation will start when granullar cells recognize and attached to the foreign target (Pech & Strand 1996). In the process of cellular encapsulation, the host's blood cell forming a multicellular capsule-like envelope around invading parasitoids (Blumberg 1997). In General, encapsulation is started 24 h after oviposition (Woodring 1985; Chapman 1998; Reed *et al.* 2007). In this study, initiation of encapsulation was related to host stage and host species. In L4 of *C. pavonana*, encapsulation can be observed only three hours after oviposition, conversely, in L2/L3 encapsulation can be detected between 18 and 24 hours after oviposition. In *S. litura*, encapsulated parasitoid can be recognized 48 hours after oviposition. Encapsulated parasitoid may die by suffocation, starvation, or physical prevention of development, however partial encapsulation may allow parasitoid continue to grow normally until pupa (Blumberg 1997).

Encapsulation may also has an effect on the development of immature parasitoid (Blumberg 1997). This was also confirmed by the result of the study. Body length of immature parasitoid developed inside L1's body was recorded to be higher than in older stages of host. This study also recorded that encapsulation in L1 was noted to be lower than older stages, and encapsulation process can only be observed after parasitoid egg hached into larva. This situation provided an advantage for immature parasitoid continue to grow. Previous finding documented that parasitoid may avoid encapsulation by active movement of encapsulated larvae (Woodring 1985; Chapman 1998). Sagarra (2000) reported that *Anagyrus kamali,* attacking the hibiscus mealybug, *Maconellicoccus hirsutus*, may evade encapsulation by ovipositing its eggs in early stages of hosts in which the immune system is not yet mature and unable to elaborate a significant defense. Older stages were documented to have a sufficiently developed immune system to effectivelly encapsulate the parasitoid eggs.

Haemocytes play an essential role in defending insect against parasites invading their haemocol (Pech & Strand 1996). Encapsulation is formed and performed by the blood cell of the host, therefore it is only effective against endoparasitoid which oviposit their eggs and develop within the host' body (Blumberg 1997). Effects of encapsulation on the host's haemosyte were documented by many studies (Strand & Noda 1991; Alleyne & Wiedenmann 2001). Number of blood cell was recorded to be higher in parasitized larvae than unparasitized larvae of both *C. pavonana* and *S. litura.* Similar result was also recorded for parasitized larvae of *Pseudoplusia includens* attacked by *Microplitis demolitor* (Strand & Noda 1991). Increasing period of time was followed by increasing number of blood cells. This finding suggests that number of blood cell increases at the older stage of host. Many studies identified that number and types of blood cell were found to be lower in young stage than in older stages (Gupta 1991; Chapman 1998). Number of blood cell involved in the response of host toward parasitoid invasion was found to be higher in larvae *C. pavonana* than *S. litura.*

Parasitized larvae which have a high capability of encapsulation may survive to grow until pupae. The question is can survived host's pupae grow normally? Effects of encapsulated parasitoid on weight of survived pupae can be observed by comparing the weight of pupae between unparasitized and parasitized pupae. Our findings indicated that weight of survived pupae of *S. litura* was lower than unparasitized pupae. This suggests that there is a possible relationship between encapsulated parasitoid and the survival of host. In contrast, we did not find a significant different between parasitized and unparasitized pupa of *C. pavonana* with respect to pupae's weight. Relationship between encapsulated parasitoid and the survival of host was confirmed by Fellowes *et al.* (1998) who found that pupae developing from larvae that have encapsulated the parasitoid *Asobara tabida* are smaller and have relatively thinner puparia. Thinner puparia are likely to be associated with a reduction in mechanical strength and possibly with a decrease in desiccation tolerance. They also mentioned that fecundity and size of adult *Drosophila melanogaster* emerged from pupa that have encapsulated its parasitoid was reduced.

Superparasitism was documented to be one factor may help parasitoid avoid encapsulation (Sagarra 2000). Superparasitism was also recorded to be occurred on female parasitoid *E. argenteopilosus* on its host. Superparasitism

may increase the probabilities of survival of parasitoid progeny with respect to encapsulation (Sagarra 2000). Our findings identified that at least there was one immature parasitoid survive from encapsulation in the superparasitism occurrence. This result was recorded to be similar with Sagarra (2000), who reported that supperparasitism helped *A. kamali* in avoiding encapsulation, immune system of host was exhausted by high level of superparasitism.

Further research are needed to look at the possibility to decrease encapsulation rate by hosts. This step is very important to develop strategy that can enhance the effectiveness of *E. argenteopilosus* in controling *C. pavonana*.

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