Influence of the Physiological Condition of the Host, *Papilio xuthus* L. (Lepidoptera: Papilionidae), on the Development of the Parasite, *Trogus mactator* TOSQUINET (Hymenoptera: Ichneumonidae)

Keiko OMATA

Laboratory of Forest Zoology, Faculty of Agriculture, University of Tokyo,
Bunkyo-ku, Tokyo 113, Japan

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Observations on the development of the parasite *Trogus mactator* TOSQUINET revealed that the first instar larvae did not develop to the second instar until their host *Papilio xuthus* L. pupated. To learn the effect of the physiological condition of the host on the development of its parasite, the following experiments were carried out. The parasitized fifth instar larva was ligatured and the development of the parasite was examined. When the abdomen of the ligatured host pupated, the parasite developed to the second instar, but did not if it remained as a larva. When the diapausing host pupa was exposed to a long day photoperiod after pupation, the parasite left the host, while it entered into diapause under a short day condition in the host pupa. These results indicate that the process to the pupation of the host is an important factor in the revival of moulting of the first instar parasite, but the development of the parasite will be further influenced by other factors, possibly photoperiodic condition.

INTRODUCTION

Trogus mactator TOSQUINET is a solitary endoparasitic wasp which oviposits in the larva of the swallowtail, *Papilio xuthus* L. and some other *Papilio* species (YASUMATSU and WATANABE, 1964).

Several studies on the relationship between endoparasites and their hosts have shown that the parasite may be arrested in the 1st larval instar until its host attains a certain particular stage (FISHER, 1971). It is assumed that the development of the parasite larva is affected not only by the external environment, such as temperature and photoperiod, but also by the internal environment which changes as the host grows. SCHNEIDER (1950, 1951), CORBET (1968), CALS-USCIATI (1969, 1975) and BARONIO and SEHNAL (1980) have shown that the host's physiological condition influences the development of the parasite. However, in most cases, the mechanism which controls the parasite development has not yet been fully elucidated.

This study was carried out to examine the development of the parasite when parasitized at various larval stages of the host, and to investigate the influence of the host's physiological condition on the development of the parasite by ligature experiment. The effects of the photoperiod and the physiological state of the host (dia-

pausing or nondiapausing) on diapause initiation of the parasite were also investigated.

MATERIALS AND METHODS

Adults of the parasite used in this study were captured directly or obtained from the host, *Papilio xuthus* in the fields of Kanagawa and Chiba Prefectures, Japan; they were fed on a 50 % honey solution at room temperature. The parasites were given host larvae in a 9.3×12.9 cm glass vial where oviposition took place. Each parasitized host larva was transferred into a 10×4.5 cm plastic cup and reared on an artificial diet (KAMANO, 1965, slightly modified) at about 25°C under a long day (15L-9D) photoperiod.

The duration of the time from oviposition to adult emergence was recorded. Also, hosts were dissected at various time intervals after parasitization, and width of the parasite head capsule was measured to identify the developmental stages of the parasite. To determine the effect of host development on the moulting of the parasite, a ligature experiment was performed. The parasitized host larvae were ligatured between the thorax and abdomen with cotton thread 4 or 6 days after the last larval ecdysis to check the parasite's development.

Most experiments were carried out under a long day photoperiod (15L-9D) which led to nondiapausing host development. The larvae were reared under a short day condition (10L-14D) throughout the larval development to induce pupal diapause as described by HIDAOKA and HIRAI (1970). They were parasitized on the 2nd day after the last larval ecdysis. After pupation, half of them were held under the same photoperiodic condition, and the rest were transferred to a long day condition to see

Fig. 1. Relationship between the developmental duration and the timing of parasitization of parasitic Ichneumonid, *Trogus mactator*. Closed circles show the duration of time from oviposition to adult emergence of the parasite. Open circles show the periods from host pupation to parasite adults emergence. The host animals were reared at ca. 25°C under a long day condition (15L-9D).

Fig. 2. Development of the parasite oviposited in the first (A), fourth (B) and fifth (C) instar host larva. Host stage: I–V, first to fifth larval instars; PP, pharate pupa; P, pupa. Host ages are indicated as days after respective ecdysis or maturation. Parasite stage: E, egg; I, II and III– indicate, first, second and third or fourth larval instars, respectively. The hosts were reared at ca. 25°C under a long day condition (15L–9D). Each circle corresponds to one sample.

the effect of photoperiod on the parasite's development.

RESULTS

Papilio larvae were parasitized at various developmental stages, and the duration of time from parasite oviposition to adult emergence was examined (Fig. 1). When the parasites oviposited in the 1st instar host larvae, the developmental period was 40 ± 3.52 days (mean $\pm$ S.D.) at about 25°C, but the duration shortened to 26 ± 1.31 days when they oviposited on the 5th instar larvae. Thus, the older the host was when parasitized, the faster the parasite developed, suggesting that young host larvae might cause a delay in the development of the parasite. On the other hand, the period from host pupation to adult emergence of the parasite was almost constant, being 19 ± 1.57 days, irrespective of parasitized stages of the host (Fig. 1).

As shown in Fig. 2, all the parasite eggs hatched within 48 hr after oviposition in every case examined. However, these parasites remained in the 1st larval instar until the host pupated. These results suggest that the 1st larval moult of the parasite may coincide with the larval–pupal transformation of the host. This possibility was further supported by the following experiment. Parasitized hosts were ligatured between the

Table 1. Development of the parasite in the abdomen of the ligatured host

Form of abdomen of the ligatured host	No. of parasites in each larval instar		
	I	II	III-IV
Larval	10	0	0
Larval-pupal intermediate	0	1	2
Pupal	1	3	7

Fifth instar host larvae were parasitized on the day of the 4th ecdysis and ligatured between the thorax and abdomen 4 or 6 days later. The abdomens of these animals were dissected 10 days after the last larval ecdysis and the stage of the parasite was determined based on the width of head capsule.

Table 2. Effect of photoperiod on the development and diapause of the host and its parasite^a

Photoperiodic condition of the host		No. of hosts tested	Developmental state ^b of	
Larval stage	Pupal stage		Host ^c	Parasite ^d
13L-11D	13L-11D	23	ND	ND
10L-14D	13L-11D	30	D	ND
10L-14D	10L-14D	30	D	D
13L-11D	10L-14D	5	ND	D (or died)

^a Hosts reared at ca. 25°C were parasitized on the second day after the last larval ecdysis.

^b N: nondiapause development, D: diapause.

^c Result of nonparasitized pupal host.

^d A parasite which remained in the second or third larval instar 40 days after host pupation was judged to be in diapause.

thorax and abdomen just around the critical time of ecdysone secretion by the prothoracic glands, and the age of the parasite larvae inside the host abdomens was examined 7 days after the ligation. Some of the isolated abdomens remained as larvae, but the others moulted into pupae or larval-pupal intermediates. As shown in Table 1, all of the parasite larvae taken out from the larval forms were identified as 1st instar larvae. By contrast, those parasites contained in the larval-pupal intermediates or pupal abdomens were found to be 2nd or much advanced instar larvae except one individual. These results clearly show that the parasite cues in on the pupal moult of the host to undergo its 2nd larval moult and then subsequently develops in the host pupae.

Next, the effect of photoperiod on the diapause induction of the parasite was investigated (Table 2). The parasitized host larvae, as well as nonparasitized normal larvae, entered pupal diapause when they were reared under a short day photoperiod (10L-14D) throughout larval development. After pupation, they were subjected to a short day (10L-14D) or a long day condition. The parasites exposed to a long day condition inside the diapausing pupal host developed to adults in 20 days after the host pupation, while those exposed to a short day did not emerge from the hosts even after 40 days of host pupation. No exceptions were observed. Dissection of the parasitized host showed that the parasite larvae remained in the 3rd larval instar, indicating that

they were diapausing. These results suggest that the parasite inside the host pupa is sensitive to photoperiod and its diapause determination is also under photoperiodic control.

DISCUSSION

As this study clearly shows, adults of *Trogus mactator* can lay their eggs in all the larval instars of the host, *Papilio xuthus*, and the eggs laid hatch 2 days after oviposition. However, the larva hatched remains in the 1st instar until the host ecdysis into a pupa. Thus, the moulting of the 1st instar larva of the parasite into the 2nd instar is closely correlated with larval-pupal transformation of the host.

Many endoparasitic species of Hymenoptera and Diptera are known to stay in the 1st larval instar until the host reaches certain particular stages of development (FISHER, 1971). For example, *Diplazon fissorius* initiates development during the larval-pupal transformation of the host (SCHNEIDER, 1950, 1951). The 1st instar larvae of *Nemeritis canescens* also initiate a rapid development after moulting to the final instar of the host (CORBET, 1968). Recently, BARONIO and SEHNAL (1980) suggested that the development of the 1st instar larvae of *Gonia cinerascens* is triggered by the release of the host's ecdysone.

The last larval instar of insects can be characterized by a decrease of juvenile hormone titer and two subsequent successive surges of ecdysone (RIDDIFORD, 1976, 1980). In the parasites which initiate moulting in the final larval instar of the host larvae, their development may be directly affected by these hormones or indirectly by physiological and biochemical changes caused by the hormones. The ligature experiment performed as part of the present experiment showed that only the parasite in the host abdomen transformed to larval-pupal intermediate or pupal form can moult into a 2nd instar larva. This strongly suggests that the larval moult of the 1st instar larvae of the parasite is induced by the action of ecdysone released by the host's prothoracic glands. However, juvenile hormone might be involved as well, because the parasite larvae do not moult before the host pupation in spite of the periodic surge of ecdysone in each larval moult. Studies are now under way to test the possibility.

First instar larvae of the parasite moult into the 2nd instar within 2 to 4 days after the host pupation. This occurs irrespective of the diapause-nondiapause state of the pupal host. However, as this study demonstrated, these 2nd (and/or 3rd) instar larvae are sensitive to photoperiod. They develop to the adult in the diapausing host pupae when exposed to a long day photoperiod. By contrast, they enter larval diapause in the host when exposed to a short day photocondition. Similar phenomena have been observed in *Apanteles sprius* (host: *Pieris brassicae*) (GEYSPITS and KYAO, 1953), *A. congregatus* (host: *Manduca sexta*) (RABB and THURSTON, 1969) and *A. melanoscelus* (host: *Lymantria dispar*) (WESELOH, 1973).

In conclusion, the development of the 1st instar larvae of *T. mactator* is influenced by certain physiological changes of the host during larval-pupal transformation of the latter. Once the parasites moult into the 2nd and the 3rd instar, they begin adult development or enter diapause depending on the surrounding photocondition.

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