Host age preference of Microplitis mediator (Hymenoptera: Braconidae), an endoparasitoid of Mythimna separata (Lepidoptera: Noctuidae)

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Abstract

Microplitis mediator (Haliday) is a solitary endoparasitoid of larvae of the cotton bollworm, Helicoverpa armigera (Hübner) and the oriental armyworm, Mythimna = Leucania separata (Walker). The preference and suitability of different instars of M. separata for M. mediator were determined under laboratory conditions at a constant temperature of 26 ± 1 °C, 65 ± 5% RH and L14:D10 photoperiod. The selection coefficient revealed that M. mediator parasitized 1st to 4th instars, but preferred 2nd and 3rd instars. Seventy-one percent of parasitism was achieved within 24 h when the 2nd instars were used as hosts at a density of one parasitoid per 20 Larvae. Parasitoid egression and pupation were dependent on the host instar parasitized and occurred from the 1st through the 4th instar. The mean developmental time from egg to prepupae of M. mediator within 1st to 4th instars of the host was 8.27, 8.30, 8.30 and 9.20 days, respectively. Cocoon weights were lower when 1st and 2nd instars served as hosts rather than 3rd and 4th instars. The percentage of host larva that died before parasitoid egression declined as the age of the host increased, ranging from 26% to 2% for 1st–5th instars, respectively. The results of this study suggest that 2nd and 3rd instars of M. separata would be the best host stages for mass production of M. mediator in the laboratory and the best host instars to target for effective control in field releases.

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1. Introduction

Microplitis mediator (Haliday) (Hymenoptera: Braconidae), is a solitary endoparasitoid, widely distributed in the Palearctic region (Slovak, 1985; Arthur and Mason, 1986; Mason et al., 2001). It is polyphagous using about 40 lepidopteran species as hosts (Shenefelt, 1973). In field collections in 1982, M. mediator was the dominant parasitoid of the cotton bollworm and oriental armyworm in China, composing 58% of all larval parasitoids recovered from H. armigera (Wang et al., 1984). Therefore, these wasps have great potential as biological control agents of H. armigera and M. separata. Mass rearing would make augmentative release of M. mediator in cotton fields feasible and may increase its efficacy in suppressing H. armigera populations (Li et al., 2004).

Parasitoids have evolved an amazing array of mechanisms to manipulate host physiology and biochemistry in order to create an environment that is favorable for the development of the parasitoid but typically detrimental to that of the host insect (Lawrence, 1990; Coudron, 1991; Edwards and Weaver, 2001; Beckage and Gelman, 2004). Parasitoid preference for specific instar or larval stages can occur for a variety of reasons. Many parasitoids exhibit
a marked preference for a specific instar larva (Mackauer, 1990; Mattiacci and Dicke, 1995; McGregor, 1996). The preference may be based on increased survival of their offspring or ease of parasitism. There are differences in host quality associated with increasing age of the host, pertaining to the developmental performance for the larval parasitoid. As the developing hosts grow in size they increase their array of physical defense mechanisms, making the encounters more dangerous for the parasitoid (Pencacchio et al., 1992; Mattiacci and Dicke, 1995). Parasitoids, through their mechanism of host selection, can influence the timing of their own life history events (McGregor, 1996). However, before undertaking large-scale release of *M. mediator*, it is important to determine the host stage most effectively parasitized by *M. mediator*. With this information, *M. mediator* females could be released at the most effective time for management of *M. separata*. Therefore, the specific objectives of our study were to determine the effects of host stage on parasitism.

2. Materials and methods

2.1. Insects

A colony of *M. mediator* wasps was established from parasitized *H. armigera* larvae collected from cotton fields near Baoding, Hebei Province, late in the season during 1998. Cultures were maintained in the laboratory on host larvae of *Mythimna = Leucania separata* reared on either corn seedlings or artificial diet (Bi, 1981) at 26 ± 1 °C, 65 ± 5% RH and 14:10 (L:D) photoperiod. *M. separata* was used as a host for mass-rearing of the parasitoid *M. mediator* because *H. armigera* is cannibalistic and therefore uneconomical to mass rear. *M. separata* was obtained from a stock culture from Insect Natural Enemy Laboratory, Hebei Plant Protection Institute, Baoding, Hebei Province. Late-1st to 2nd instars were used to maintain the parasitoid culture. Adults were fed with a solution of 10% commercial clover honey impregnated into cotton wool. Parasitization was accomplished by placing 50 mated 2- to 4-day-old female parasitoids in a cage (40 cm × 30 cm × 25 cm) containing 1000 *M. separata* for 24 h. Parasitized larvae were held in bags (30 cm × 20 cm × 5 cm), containing corn plants with 1000 larvae per bag, until adult parasitoid emergence. All experiments were conducted under these conditions except where noted.

2.2. Influence of host stage on parasitism by *M. mediator* in non-choice tests

To determine the larval stages of *M. separata* that were most effectively parasitized by *M. mediator*, 4-day-old mated female parasitoids were exposed to host larvae in an oviposition cage (40 cm × 30 cm × 25 cm) for 24 h containing 10% honey water. Each cage contained one female parasitoid and 20 host larvae of a particular stage. Trials with each instar were replicated 10 times. After exposure, the host larvae were placed individually in 30 ml plastic cups containing host diet. Larvae were checked daily until they had pupated, died, or produced parasitoid cocoons. Larvae that died before pupation or parasitoid cocoons were dissected and checked for parasitoid eggs or larvae. The date and the number of parasitoid cocoons and unspun larvae were recorded.

2.3. Influence of host stage on parasitism by *M. mediator* in choice tests

To determine the stage of *M. separata* preferred by *M. mediator* for parasitization, 4-day-old mated female parasitoids were exposed to host larvae in the oviposition cage as described for the non-choice tests. Three replicates were performed. Each cage contained 10 parasitoids and 200 larvae (ca. 40 each of 1st–5th instars). After a 24 h exposure period, instars were separated and placed in feeding cages and reared on corn seedlings. Exposed host larvae were checked daily as described in the non-choice tests.

2.4. Effects of host stage on the development and survival of *M. mediator* offspring

To determine the effects of host stage on the development of *M. mediator* offspring, 4-day-old mated female parasitoids were exposed to host larvae as described in the non-choice tests. Each oviposition cage contained 200 host larvae of a particular stage and 10 female parasitoids. Trials with each instar (1st–5th) were replicated 3 times. After exposure, the host larvae were placed individually in 30 ml plastic cups containing host diet. Upon emergence of parasitoid larvae, host food was removed from the cups. When all parasitoid larvae pupated, host larvae were removed. When adult parasitoids emerged they were placed in a sleeve cage and provided with 10% honey water at 26 ± 1 °C, 65 ± 5% RH and 14:10 (L:D) photoperiod. The development time from egg to prepupa (spinning of cocoon) or pupae and from prepupa to adult, pupae weight, percentage adult eclosion and adult longevity were determined.

3. Data analysis

The following formulas were used to calculate the percentage of host mortality, parasitism, and the selection coefficient of *M. mediator* to host larvae (Cook, 1978; Vinson and Iwantsh, 1980).

Mortality (%) = (Number of larvae tested – Number of pupae – Number of parasitized larvae)/Number of larvae tested × 100.
Parasitism (%) = (Number of parasitized larvae/Number of larvae exposed) × 100. Selection Coefficient = Ri/∑m=1, m=1 Rm (Ri = the percentage parasitism during host instars i, m = number of host instars tested).

Where appropriate, data were subjected to analysis of variance to determine differences between means. Where significant differences occurred, Duncan’s multiple range tests was applied for mean separation. Data are presented
as Means ± SE, where means within the same column and followed by different letters are significantly different (P < 0.05; Duncan’s multiple range test).

4. Results

4.1. Influence of host stages on parasitism by M. mediator in non-choice tests

Second and third instars yield a significantly higher percentage parasitism than 1st and 4th instars (Table 1). This resulted in a significantly higher selection coefficient of M. mediator to 2nd and 3rd instar. Mortality, however, was significantly greater when parasitism occurred during the 1st and 2nd instars. No parasitism occurred in 5th instars.

4.2. Influence of host stage on the selectivity of parasitism in choice tests

When given a choice, M. mediator parasitized 1st to 4th instars of M. separata. The percentage parasitism was 32.67%, 61.67%, 57.33%, and 45.33% respectively (Table 1). The offspring of hymenopteran parasitoids depend on the host for all their nutritional needs. The nutrition quality may be different in different hosts or different host instars (Harvey et al., 2000; Harvey and Strand, 2002). Most parasitoids have the ability to determine host quality during the oviposition sequence and will often accept or reject hosts on this basis (Charnov and Skinner, 1985; Strand and Pech, 1995). Wang et al. (1984) tested 1st–4th instars and demonstrated that 1st–2nd instars of H. armigera were suitable for parasitization by M. mediator in cotton fields. Tanaka et al. (1984) examined the pattern of host growth after parasitism by M. mediator and showed no correlation between the reduction of the host weight gain and the developmental stage of the parasitoid. Liu et al. (2004) concluded that the size of H. armigera larvae had significant influence on oviposition by M. mediator, and showed that the parasitization rate decreased as the host size increased. From our field release investigation we showed that 1st and 2nd instars of H. armigera were parasitized by M. mediator in Xinjiang cotton fields (Li et al., 2004). Yet, from the study reported here, it appears that 1st and 2nd instars are less capable of withstanding parasitism, hence accounting for the greater percentage mortality in comparison with 3rd and 4th instars hosts. In addition, although parasitization by M. mediator is significantly greater when parasitism occurred during the 2nd and 3rd instar. Mortality, however, was significantly greater when parasitism occurred during the 1st and 2nd instars. No parasitism occurred in 5th instars.

4.3. Effects of host stage on the development of M. mediator offspring

It took an average of 9.2 days for M. mediator to develop from egg to cocoon in 4th instars of M. separata which was significantly longer than on larvae from earlier stadia. Mean parasitoid development times from cocoon to adult were not affected by the stage of the host that was parasitized. Mean weights of cocoons spun in host larvae exposed as 3rd and 4th instars were significantly higher than those of larvae exposed as 1st and 2nd instars. The stage of the host parasitized had no effect on the percentage of parasitoid emergence or adult longevity of the parasitoid offspring (Table 3).

5. Discussion

The offspring of hymenopteran parasitoids depend on the host for all their nutritional needs. The nutrition quality may be different in different hosts or different host instars (Harvey et al., 2000; Harvey and Strand, 2002). Most parasitoids have the ability to determine host quality during the oviposition sequence and will often accept or reject hosts on this basis (Charnov and Skinner, 1985; Strand and Pech, 1995). Wang et al. (1984) tested 1st–4th instars and demonstrated that 1st–2nd instars of H. armigera were suitable for parasitization by M. mediator in cotton fields. Tanaka et al. (1984) examined the pattern of host growth after parasitism by M. mediator and showed no correlation between the reduction of the host weight gain and the developmental stage of the parasitoid. Liu et al. (2004) concluded that the size of H. armigera larvae had significant influence on oviposition by M. mediator, and showed that the parasitization rate decreased as the host size increased. From our field release investigation we showed that 1st and 2nd instars of H. armigera were parasitized by M. mediator in Xinjiang cotton fields (Li et al., 2004). Yet, from the study reported here, it appears that 1st and 2nd instars are less capable of withstanding parasitism, hence accounting for the greater percentage mortality in comparison with 3rd and 4th instars hosts. In addition, although parasitization by M. mediator is significantly greater when parasitism occurred during the 2nd and 3rd instar. Mortality, however, was significantly greater when parasitism occurred during the 1st and 2nd instars. No parasitism occurred in 5th instars.

Table 1 Effects of host instar on % parasitism, selection coefficient and % mortality of M. separata hosts parasitized by M. mediator in non-choice tests

Table 2 Effects of host instar on % parasitism and % mortality of M. separata hosts parasitized by M. mediator in choice tests

Table 3 Means (± SE) development time from egg to cocoon formation, to adult emergence, cocoon weight, % emergence and adult longevity of M. mediator parasitizing M. separata hosts

Note: All the numbers of host larvae tested in this experiment is 200 with ca. 40 larvae in each instar. Means followed by same letter in columns (Duncan’s test) do not differ statistically (P≤0.05).
successful in 1st to 4th instars of *M. separata*, female parasitoids prefer 2nd and 3rd instars. The development time from egg to cocoon spinning (prepupae) was shorter when 2nd and 3rd instars were used as hosts but cocoon weight was greater when 4th instars served as hosts. Perhaps, the 4th instars provided a better nutritional quality than 2nd and 3rd instars. The selection coefficient of *M. mediator* for 2nd and 3rd instars was higher than that for 1st and 4th instars suggesting that 2nd and 3rd instars are the stages preferred by *M. mediator*.

During development, parasitoids not only regulate the host’s development, but also withstand attacks by the host’s defense system which increases in strength or effectiveness with the age of the host (Puttler, 1961; Vinson and Iwantsch, 1980; Harvey, 1996; Webb et al., 2001). Additionally, the host affects the development of the parasitoid (Lewis, 1970; Miles and King, 1975; Lawrence, 1990; Vinson et al., 1994; Harvey, 2000; Edwards and Weaver, 2001; Beckage and Gelman, 2004; Liu et al., 2004). Upon dissection, no *M. mediator* larvae were found in parasitized 5th instar which explains the absence of cocoons. The absence of parasitism of 5th instar *M. separata* by *M. mediator* supports the concept that the host’s immune system was strong enough by the 5th instar to prevent development of the parasitoid. This may also apply, in part, to 4th instars as well. In contrast, low parasitization success in 1st instars is due to high mortality.

In conclusion, these results indicate that the development of *M. mediator* in 2nd and 3rd instar hosts was faster than in 4th instar hosts. Although, cocoon weights were higher in 4th instar hosts, suggesting that 4th instars might be a better nutritional source, the host stage had no effect on either the percentage of parasitoid emergence or on the longevity of adult parasitoids developing from these hosts. Therefore, based on our current information, it is preferable to use 2nd and 3rd instars to mass rear *M. mediator* so as to optimize the percentage parasitism, parasitoid development time and survival. Similarly, when augmentative field releases of *M. mediator* for 2nd and 3rd instars should be released when 2nd and 3rd instars are present in the field so as to benefit from the corresponding high selection coefficient.

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