

## *Trichogramma minutum* Relationship to Codling Moth and Red-banded Leaf Roller Eggs

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### Abstract

Observations were made of the gross changes to embryos of codling moth, *Laspeyresia pomonella* (L.), and red-banded leaf roller, *Argyrotaenia velutinana* (Walker), at various ages and the relationship of the eggs of these species to the egg parasite, *Trichogramma minutum* Riley. Codling moth and red-banded leaf roller eggs were suitable hosts for complete parasite development within 2 days and 4 days of host oviposition, respectively. Older eggs could be killed by repeated parasite ovipositional "stings" but did not support parasite development.

Field experiments to suppress codling moth, *Laspeyresia pomonella* (L.), and red-banded leaf roller, *Argyrotaenia velutinana* (Walker), with inundative releases of a local strain of *Trichogramma minutum* Riley require that the host: parasite relationships be studied so that prospects for success be enhanced. Some of these relationships and the rearing method of *T. minutum* on codling moth and red-banded leaf roller eggs at the Humid Areas Deciduous Fruit Insects Investigations laboratory at Vincennes, Indiana, were described (2). The objectives of this experiment were to determine: 1) development of the parasitized host embryo; 2) relationship of age of the host egg to: a) attractiveness to the parasite and b) parasite development.

### Materials and Methods

Codling moth and red-banded leaf rollers were reared as described by Cleveland (1). Moths were allowed to oviposit on waxed paper (9x6 inches) for 24 hours in chambers with continuous light. The eggs on the waxed paper were then placed in a wooden box, 36x18x24 inches, with a glass top. The boxes were exposed to continuous fluorescent light, 78-84°F, and 70-73% relative humidity. Sections of paper containing 20 scattered codling moth eggs or about 100 red-banded leaf roller eggs in 1-2 masses, were snipped from the egg sheets and introduced into another wooden box containing about 4000 adult *Trichogramma minutum* for a 1-day exposure to the parasites. At 24-hour intervals, similar sections of eggs were removed from the original egg sheets and placed into the parasite chamber to replace the eggs that had been exposed to *T. minutum* for 24 hours. The latter were then returned to the first glass-topped box. This exchange and rotation of egg samples continued until eclosion of unparasitized host eggs on the original egg sheets was completed. Sections of egg sheets that were not exposed to the parasites were used as a check and for observations of the development pattern of the host embryos.

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Daily observations of parasite and host embryonic activity were made by viewing the sheets through the glass-topped boxes. All eggs were examined daily under a binocular microscope during the course of the experiment.

Host eggs that died between observations were detectable by the cessation of development and changes in shape and color due to desiccation and decomposition.

### Results and Discussion

The development pattern for the unparasitized eggs of both species of moths was similar and readily observed with a binocular microscope without disturbing the eggs. Newly laid eggs of codling moth are translucent white and those of red-banded leaf roller are translucent yellow. An opaque peripheral ring soon forms with a stalk that terminates at the center of the egg. The embryo is attached to the end of this stalk and becomes more conspicuous as it increases in size and recognizable features form. Eventually, dark eye-spots and then well-defined intersegmental lines of the abdomen appear. The mandibles, head capsule, and prothoracic shield become conspicuous and then darken in pigmentation. The mature embryo fills the egg shell cavity in a curled position and may be seen moving its mandibles side to side before it cuts the shell and emerges.

Under these conditions, development of the embryo was more rapid in codling moth, and hatching began in 4-5 days from oviposition and was completed by 6 days. The leaf roller eggs did not begin to hatch until 6-7 days old and 10 days lapsed before eclosion was terminated (Table 1).

The results of the codling moth eggs exposed to the parasites are shown in Table 2. Although the number of eggs available was low, there are indications that eggs in the 0 to 1 and 1 to 2-day-old classes are readily parasitized and that successful development and emergence of *T. minutum* follow. Eggs in the 2 to 3-day-old range may be parasitized in low numbers, particularly the youngest eggs of the group. In this test, the few older eggs with well-developed host embryos evidently did not support the development of *T. minutum*. This phenomena was observed by the senior author in the laboratory colony on numerous occasions, as well. Codling moth eggs with small embryos may be parasitized, but those with larger embryos at the pigmented eye-spot stage, or older, are apparently able to suppress parasite development. Peterson (5) reported similar findings when he observed that parasitism by *T. minutum* would stop host embryonic development in codling moth and oriental fruit moth, *Grapholitha molesta* (Busck), up to, but not beyond, the time that the host larva acquired a well-developed head. In his study, oriental fruit moth embryos reached this stage of development in 56-72 hours.

With a slower development rate, it is evident that the 3 to 4-day-old eggs of red-banded leaf roller may be successfully parasitized and

TABLE 1. *Development pattern of unparasitized codling moth and red-banded leaf roller eggs.*

Age of eggs in days	Condition of codling moth eggs
0-1	Translucent with an opaque peripheral ring with stalk leading into egg center
1-2	Small embryo at apex of stalk. 30% of eggs dead and desiccating.
2-3	As in preceding to larger embryos with eye spots, abdominal segments. 45% of eggs dead and desiccating.
3-4	Embryos with eye spots to well-developed embryos with dark head capsules and prothoracic shields in equal numbers of both types.
4-5	Well-developed embryos. Beginning of eclosion.
5-6	Eclosion completed. 50% of eggs dead and dried.

  

Condition of red-banded leaf roller eggs	
0-1	Translucent with scattered opaque bodies.
1-2	Formation of an opaque peripheral ring with stalk leading into egg center.
2-3	Similar with above structures better defined. Small embryo at apex of stalk.
3-4	Embryos larger with eye spots.
4-5	No obvious change.
5-6	Mandibles sclerotized. Some embryos can move head and mandibles. Sterile and dead eggs are semi-dehydrated.
6-7	All embryos have sclerotized mandibles. Embryos are inactive or are chewing chorion. Beginning of eclosion.
7-8	Eclosion about 80% completed.
8-9	Eclosion about 90% completed.
9-10	Eclosion about 100% completed. 25% of total eggs are dead and semi-dehydrated.

a few are susceptible 1 day later (Table 3). A few 5 to 6-day-old eggs were parasitized but *T. minutum* died before emerging. Unlike codling moth eggs at that stage, successful parasitization was effected in leaf roller eggs containing host embryos with eye spots.

In this experiment, 56.3% of the codling moth eggs and 52.7% of the red-banded leaf roller that were parasitized (as evidenced by darkened egg chorion) did not produce live adult *T. minutum*.

All eggs of both species in different stages were attractive to *T. minutum* females in some degree and elicited an ovipositional response. Females were attracted to newly-introduced red-banded leaf roller eggs and up to 3-4 females were observed simultaneously ovipositing on the same egg mass. In a typical oviposition behavioral pattern, the female moves back and forth (within a few body lengths) on an egg mass, while touching the egg with her antennae. Eventually, she stops, depresses her abdomen, and unsheathes the ovipositor, a hair-like structure. This takes about 10 seconds. The ovipositor is

TABLE 2. Results of exposing codling moth eggs<sup>1</sup> of different ages to *Trichogramma minutum* Riley.

Age of eggs exposed to parasites (days)	No. live eggs exposed	No. eggs parasitized (darkened egg chorion)	No. eggs with parasite emergence	Average no. parasites per parasitized egg	No. eggs hatched following parasite exposure	No. dead prehatch host embryos
0-1	20	12	8	3.9	0	16
1-2	14	10	8	2.1	0	0
2-3	11	1	0	—	0	2
3-4	14	0	0	—	2	12
4-5	9	0	0	—	3	16

<sup>1</sup> 10 of 20 eggs in the check (no parasite exposure) survived to a completed eclosion.

TABLE 3. Results of exposing red-banded leaf roller eggs<sup>1</sup> of different ages to *Trichogramma minutum* Riley.

Age of eggs exposed to parasites (days)	Approximate No. live eggs exposed	No. eggs parasitized (darkened egg chorion)	No. eggs with parasite emergence holes	Average no. parasites per parasitized egg	No. eggs hatched following parasite exposure	No. dead prehatch host embryos
0-1	100	50	20	1	0	0
1-2	100	80	52	1	0	0
2-3	100	60	28	1	0	0
3-4	90	60	24	1	30	1
4-5	90	16	4	1	40	7
5-6	70	5	0	1	60	10
6-7	50	0	0	0	25	15

<sup>1</sup> Approximately 75% of the check eggs (no parasite exposure) hatched.

inserted through the chorion until the abdomen is very close to the host egg. While the female is ovipositing, the sheath is held at about a 45° angle, her ovipositor is perpendicular and the abdomen rotates 360° as the ovipositor is drilled into the egg. The female then sits motionless for about 10 seconds as the eggs are deposited. After egg deposition is completed, she withdraws the ovipositor. It was observed that younger host eggs are more attractive to the female than older ones. When the parasites were exposed to 3 to 4-day-old codling moth eggs, they showed an immediate preference for eggs containing the younger embryos at the early eye-spot stage and initially ignored eggs containing embryos with dark head capsules and prothoracic shields. Male *T. minutum* seeking mates often disrupted the ovipositional behavior pattern of the females.

The older eggs introduced into the parasite chamber did not produce parasite progeny but the host survival to a successful eclosion was much lower than in the check. It is suspected that multiple oviposition punctures by the parasites resulted in a moisture loss that indirectly resulted in embryonic mortality. Fully-developed host embryos moving under the chorion were not adversely affected by these punctures when exposed to the parasites.

The presence of a well-developed embryo is not a deterrent to successful *T. minutum* parasitism in all lepidopterous species, however. More than half of the eggs of cabbage loopers, *Trichoplusia ni* (Hübner), exposed to egg parasites could be successfully parasitized in 63-hour-old eggs, the age when some eggs began to hatch (4). Parasitism of the eggs of the almond moth, *Cadra cautella* (Walker), by *T. evanescens* Westwood occurred even when the host embryo had formed head capsules (3), although only in numbers of 10% while the check had 93% parasitism.

From our studies, we conclude that for successful parasitization codling moth eggs must be "stung" within 2 days of oviposition and red-banded leaf roller eggs within 3-4 days. Older eggs with well-developed embryos may not be the subject of successful *T. minutum* parasitization, but the egg may be killed by repeated ovipositional punctures.

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