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Source: Florida Entomologist, 89(2) : 263-265

Published By: Florida Entomological Society

URL: [https://doi.org/10.1653/0015-4040\(2006\)89\[263:RDTCCO\]2.0.CO;2](https://doi.org/10.1653/0015-4040(2006)89[263:RDTCCO]2.0.CO;2)

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# REARING *DIOMUS TERMINATUS* (COLEOPTERA: COCCINELLIDAE) ON THE CORN LEAF APHID, *RHOPALOSIPHUM MAIDIS* (HOMOPTERA: APHIDIDAE)

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*Diomus terminatus* (Say) (Coleoptera: Coccinellidae), a native species, has demonstrated potential as an augmentation biological control agent for pest aphids (White et al. 2001). It occurs from Texas to Vermont, including the entire state of Florida (Gordon 1976), and also has been found in Bermuda (Hilburn & Gordon, 1989). *Diomus terminatus* survives in a wide range of habitats, feeds on a variety of aphids and, because it is native species, poses far less environmental risk than exotic natural enemies (Lenteren et al. 2004). Hall (2001) and Hentz and Nuessly (2002) raised *D. terminatus* on the yellow sugarcane aphid, *Sipha flava* (Forbes), and (Osborne, unpublished) raised it on the cotton aphid, *Aphis gossypii* Glover, and green peach aphid, *Myzus persicae* (Sulzer). Our goal was to rear *D. terminatus* on the corn leaf aphid, *Rhopalosiphum maidis* (Fitch) and standardize the associated rearing methods.

Sorghum (*Sorghum bicolor* var. hybrid grain sorghum SS800) was grown during the summer in a greenhouse maintained at  $23 \pm 6^\circ\text{C}$  and 70% RH with ambient light. It was used 4-6 weeks after planting when it attained a height of at least 40 cm. The corn leaf aphid colony was maintained on the sorghum in three Florida Reach-In® growth chambers (Walker et al. 1993) at the University of Florida, Entomology and Nematology Department. The chambers were programmed to maintain a constant temperature of  $22^\circ\text{C}$ , 50% RH, and an 18 h photophase (16 h full light, 1399 Lux, followed by 2 h reduced light, 334 Lux) and 6 h scotophase. Two 20-watt white fluorescent lamps (Sylvania® Cool White) were placed in the back of each growth chamber behind a Plexiglas® panel and supplemented with two 20-watt white fluorescent lamps (Sylvania® Cool White) and two 20-watt blue/white fluorescent lamps (Sylvania® Gro-Lux Aquarium) overhead. Inoculation with aphids was accomplished by inserting aphid-infested leaves directly into the whorls of the plants. This succulent part of the plants stimulated rapid aphid reproduction. A colony could be used 1-3 weeks after inoculation.

The *D. terminatus* colony was maintained in the growth chambers. For each generation, 30 adults were established per plastic container (21 cm diam  $\times$  7.5 cm high) (Pioneer Packaging,

Dixon, KY). The beetles were not sexed due to their minute size. Each container was fitted with two (4 cm diam) or three (3 cm diam) ventilation holes in the lid. Tape was used to open or close the screened ventilation holes and maintain 70-90% RH. Low humidity could be raised by misting the lids of the containers with water or by providing moist cotton balls over the holes. Each container was fitted with a rack made of 1-cm mesh hardware cloth folded so that it stood approximately 4 cm above the bottom. Two rolled laboratory tissues (38.1  $\times$  42.6 cm) (Kimwipes® Ex-L, Kimberly-Clark, Roswell, GA) were placed under each rack to absorb excess moisture and provide pupation sites. Several wax paper strips (22 cm  $\times$  2 cm wide) were provided to separate females and serve as an extra oviposition substrate. The beetles were fed by placing aphid-infested sorghum leaves directly on the hardware cloth rack. After five days, the adults were removed from the container and the remaining leaves, Kimwipes®, and wax paper were held until larvae emerged. For routine rearing, about five aphids were provided per beetle larva in the first and second instars, and seven in the third and fourth. By day 10, most of the larvae pupated on the Kimwipes®, with only a few on the desiccated leaves or the lids of the rearing containers. The adults that began to emerge were collected by aspiration. The rearing procedure is summarized in Fig. 1.

The preoviposition period of *D. terminatus* fed corn leaf aphid nymphs was determined by removing pupae from colony cages on a single day and collecting the emerging adults within a 24-h period. Five beetles of undetermined sex were placed in each of 12 containers (10 cm wide  $\times$  10 cm long  $\times$  7.5 cm high). Each container was supplied with 10 aphids per beetle per day, a drop of water, and two strips of wax paper (10 cm  $\times$  2 cm) before being sealed with Parafilm®. The wax paper, leaves, and containers were thoroughly examined daily for eggs, and fresh aphids and water were added.

To determine the effect of adult nutrition on fecundity, *D. terminatus* pupae were collected from the colony and emerging adults were given an excess of corn leaf aphids on sorghum leaves for 10 d. This was sufficient time for mating and pre-ovi-

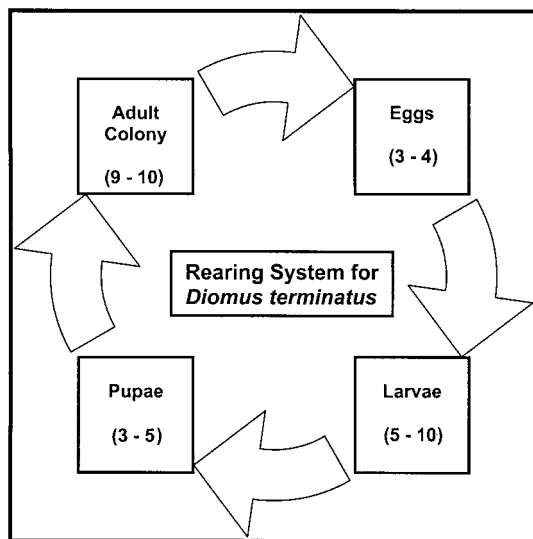


Fig. 1. Diagram of the standardized rearing system for *Diomus terminatus*. The adult colony is replenished by placing newly emerged beetles into mating/oviposition containers. The pre-oviposition period is 4-5 d and the females are held to produce eggs for 5 d. Eggs are collected daily and held for emergence of larvae in 3-4 d. Larvae are transferred to rearing containers and fed fresh corn leaf aphids daily until they pupate in 5-10 d. Pupae mature in 3-5 d and the adults emerge to complete the cycle.

position. Thereafter, individual beetles were isolated in Petri dishes (5 cm × 0.9 cm) and provided daily with 1, 4-6 or 7-10 large corn leaf aphid nymphs, a drop of water, and a small strip of wax paper (4 cm × 1 cm) for oviposition. The numbers of eggs deposited were recorded daily for 20 d. Every 3 d, the beetles were transferred to new Petri dishes because of the accumulation of frass. Comparisons were made of the number of eggs deposited/female/d with the General Linear Model (GLM) and LS means procedures in SAS (The SAS System for Windows v8, 2001).

These methods for rearing *D. terminatus* on corn leaf aphids produced results similar to those previously obtained with yellow sugarcane aphids (Hall 2001; Hentz & Nuessly 2002), and cotton aphids and green peach aphids (Osborne unpublished). Beetles held in large rearing containers and fed corn leaf aphids on sorghum leaves oviposited in 4 d. Females oviposited single eggs ( $0.68 \pm 0.001$  mm long by  $0.37 \pm 0.001$  mm wide) mainly on the leaves but also on the wax paper and Kimwipes®.

The survival and reproduction of *D. terminatus* can be optimized by providing an abundance of aphids and adequate space. When provided with one aphid, only 53% of the females oviposited, averaging  $0.5 \pm 0.1$  egg/15 females/d during the 5 d that the adult cages were maintained. After feed-

ing on 4-6 or 7-10 aphids, 100% of the females oviposited and produced  $7.8 \pm 0.5$  and  $14.1 \pm 1.0$  eggs/15 females/d, respectively. When Hall (2001) held 10 beetles in individual large tubes (15 cm × 2.2 cm), fecundity averaged 1.4 eggs/female/d. We maintained the beetles individually in Petri dishes (5 cm × 0.9 cm) and recovered about 1 egg/female/d. Osborne (unpublished) determined that each female produced a total of 86 eggs when fed cotton aphids and 37 on a diet of green peach aphids. Average daily and total consumption rates, respectively, were 13.5 and 425.5 cotton aphids, and 8.7 and 243.3 green peach aphids.

The results of *D. terminatus* rearing have been highly variable regardless of prey aphid or handling methods. When *D. terminatus* was fed yellow sugarcane aphids, both Hall (2001) and Hentz & Nuessly (2002) reported 3-4 d for embryogenesis and 4-5 d as pupae but different times for larval development; 10 versus 5 d, respectively. According to Hall (2001), *D. terminatus* females that consumed 5-10 yellow sugarcane aphids lived for 17-18 d, and produced a total of 43 eggs. Osborne (unpublished) determined that development of the egg, larva and pupa averaged 6.3, 9.4, and 6.4 d, respectively, on cotton aphids and 6.2, 7.4, and 4.1 d on yellow sugarcane aphids. In our rearing system, eggs were held for 3-4 d for larval eclosion, larvae developed in 5-10 d, and adults emerged from pupae within 3-5 d and lived for 30 d. The average egg to adult survival of  $38.8 \pm 0.1\%$  (range of 7-93%) was consistent with the 39.4% reported by Hall (2001). Our *D. terminatus* colony was maintained on corn leaf aphids for 10 generations.

We thank Ken Portier, Jeremy Jarabek, H. Alejandro Arevalo, Katherine Houben, and Judy Gillmore for technical assistance. We thank Michael Thomas and Susan Halbert for confirmation of insect identification. We are grateful to Drs. Donn Shiling and Jianjun Chen for providing a significant portion of the funding for this research.

#### SUMMARY

A standardized tritrophic rearing system was developed for *Diomus terminatus* (Say) (Coleoptera: Coccinellidae) based on the sorghum-raised corn leaf aphid, *Rhopalosiphum maidis* (Fitch) (Homoptera: Aphididae). With 30 beetles per adult container and an ample supply of aphids, *D. terminatus* females had a minimum 4-d pre-oviposition period and 100% produced eggs. After oviposition began, the beetles were maintained for 5 d and each container yielded an average of 14.1 eggs/d. Survival from egg to adult averaged 39%, yielding 20-40 beetles per adult container. Presentation of aphids on sorghum leaves and large rearing containers supported continuous production of *D. terminatus* for 10 generations.

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