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Source: Florida Entomologist, 92(3) : 497-499

Published By: Florida Entomological Society

URL: https://doi.org/10.1653/024.092.0312

PARASITISM OF THE BROWN CITRUS APHID IN DOMINICA BY *LYSIPHLEBUS TESTACEIPES* AND *LIPOLEXIS OREGMAE* (HYMENOPTERA: APHIDIINAE)

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The brown citrus aphid *Toxoptera citricidus* (= *citricida*, Nieto Nafría et al. 2005) (Kirkaldy) (Hemiptera: Aphididae) is an efficient vector of the citrus tristeza virus (CTV), and an economically important pest in areas where citrus species are grafted on rootstocks susceptible to CTV (i.e., sour orange) (Rocha-Pena et al. 1995). *Toxoptera citricidus* originated in Asia and invaded Florida and the Caribbean basin during the 1990s, causing serious economic losses (Hoy et al. 2007). The parasitoid *Lysiphlebus testaceipes* Cresson (Hymenoptera: Aphidiinae) has been recorded parasitizing the brown citrus aphid in Florida, Jamaica, and Puerto Rico (Yokomi & Tang 1996; Persad et al. 2004; Hoy et al. 2007), but control of the brown citrus aphid was poor. As a part of a classical biological control program directed against *T. citricidus*, the endoparasitoid *Lipolexis oregmae* Gahan (Hymenoptera: Aphidiinae) was evaluated for introduction into Dominica because it established in Florida and was detected in Jamaica in 2004, where it was fortuitously introduced (Hoy et al. 2007; Persad et al. 2007).

Before importation and release of *L. oregmae* in Dominica, a survey was conducted during Feb and Apr 2007 at 6 parishes to evaluate the distribution and parasitism rates of brown citrus aphids. Brown citrus aphids and aphids of unknown species were collected in 95% ethyl alcohol and DNA was analyzed for parasitoid DNA with the High-fidelity PCR protocol developed by Persad et al. (2004). Genomic DNA from pooled brown citrus aphids was extracted with Puregene reagents according to the method suggested by the manufacturer (Gentra Systems, Minneapolis, MN) and resuspended in 10 µL of sterile water. The nuclear rRNA ITS2 sequences of *L. oregmae* were amplified with the specific forward primer LO-ITSF 5'-GGCCAGTTGTCGAGTCC-3' in combination with the 28 S-R reverse primer (5'-ATGCTTAAATTTAGGGGGTA-3'), while the rRNA ITS2 partial sequences of *L. testaceipes* were amplified with the forward primer LT-ITSF 5'-CTAGCGATAAATGAATGTTC-3' in combination with the 28 S-R reverse primer (Persad et al. 2004). PCR products were separated by electrophoresis on 2% agarose gel, stained with ethidium bromide, and photographed. The *L. oregmae*-specific primers produced a 270-bp PCR product, while the ITS2 sequences amplified from *L. testaceipes* produced a 520-bp PCR product.

Although *L. oregmae* was not purposefully released in Dominica, the survey indicated that both parasitoids are present throughout the island (Table 1), with *L. oregmae* found in all 6 parishes at 76% of the locations sampled, and *L. testaceipes* found in 5 parishes at 53% of the sampling sites. Both *L. oregmae* and *L. testaceipes* were detected from aphids of unknown species on weeds within citrus groves in St. George parish at 3 locations. This indicated that both parasitoids parasitize alternative aphid hosts in the presence of the brown citrus aphid. When and by which mechanisms *L. oregmae* was introduced to Dominica are unknown. The fortuitous introduction of parasitoids of citrus pests into different Caribbean islands indicates that, as in Florida, it is difficult to prevent invasive insect introductions.

Although evaluation of the effectiveness of parasitoids of the brown citrus aphid was beyond the goal of this study, the rate of parasitism on a single date was assessed by selecting randomly 5 aphids from each of 4 parishes and testing them individually with both *L. testaceipes*- and *L. oregmae*-specific primers following the protocol described above (Table 2). The percentage of parasitized aphids was remarkably high, ranging from 80 to 100%. None of the brown citrus aphids tested was positive for *L. oregmae* only, while 20, 20, and 40% of the samples from St. Mark, St. David, and St. Peter, respectively, were positive for *L. testaceipes* only. Most of samples were positive for both *L. oregmae* and *L. testaceipes*, indicating that both parasitoids had parasitized the brown citrus aphid. The oviposition sequence, age of larvae, and larval development time are key factors that affect the development of parasitoids within parasitized brown citrus aphids, so it is impossible to resolve which species would emerge from the brown citrus aphid in these cases (Persad & Hoy 2003). However, the parasitism level estimated for *L. oregmae* and *L. testaceipes* ranged from 80 to 100% overall in this limited sample, suggesting that they are common parasitoids of the brown citrus aphid in Dominica. De-

TABLE 1. SURVEY ON DISTRIBUTION AND ABUNDANCE OF BROWN CITRUS APHID PARASITOIDS IN CITRUS-GROWING AREAS OF DOMINICA DURING 2007 WITH A HIGH-FIDELITY PCR PROTOCOL AND SPECIES-SPECIFIC PRIMERS.

Sample date	Parish	Location	Host	No. of pooled aphids tested	*L. oregmae*[1]	*L. testaceipes*[1]
7 Feb	St. Peter	Syndicate	Citrus	3	+	+
7 Feb	St. Peter	Syndicate	Citrus	22	+	–
7 Feb	St. Peter	Syndicate	Citrus	15[2]	+	+
2 Feb	St. Patrick	Grand Bay	Citrus	50	+	–
6 Feb	Unknown	Sundalae	Citrus	200	+	+
16 Feb	St. Andrew	Melville Hall	Citrus	100	+	–
15 Feb	St. Andrew	Wesley	Sweet pepper	50	–	–
20 Apr	St. Mark	Soufrière	Citrus	50	+	+
20 Apr	St. Andrew	Marigot	Citrus	50	+	+
20 Apr	St. Andrew	Hatton Garden	Citrus	50	+	+
20 Apr	St. Andrew	Woodford Hill	Citrus	50	+	–
20 Apr	St. Andrew	Woodford Hill	Citrus	50	+	+
20 Apr	St. David	Castle Bruce	Citrus	50	+	+
23 Apr	St. George	Giraudel	Composite weed	100[3]	–	–
23 Apr	St. George	Morne Prosper	Composite weed	50[3]	+	+
20 Apr	St. George	Roseau	Citrus	50[3]	–	–
23 Apr	St. George	Giraudel	*Colocasia* sp.	50[3]	–	–

[1]+ = positive detection; – = negative detection.
[2]Adult wasps collected from a citrus grove.
[3]Aphids of unknown species collected from citrus or composite weeds within a citrus grove.

TABLE 2. PARASITISM ASSESSMENT OF 5 INDIVIDUAL BROWN CITRUS APHIDS AT EACH SITE BY *L. OREGMAE* AND *L. TESTACEIPES* IN DOMINICA DURING 2007 WITH SPECIES-SPECIFIC PRIMERS AND A HIGH-FIDELITY PCR PROTOCOL.

				Percentage positive by PCR		
Sample date	Parish	Location	Host	*L. oregmae* only	*L. testaceipes* only	*L. oregmae* and *L. testaceipes*
19 Apr	St. Peter	Syndicate	Citrus	0	40	60
20 Apr	St. Andrew	Marigot	Citrus	0	0	100
23 Apr	St. Mark	Soufrière	Citrus	0	20	60
20 Apr	St. David	Castle Bruce	Citrus	0	20	80

spite this apparent abundance, natural enemies cannot prevent transmission of diseases such as CTV by aphids, so replanting with citrus on CTV-resistant rootstocks should be considered.

SUMMARY

The brown citrus aphid parasitoids *Lipolexis oregmae* and *Lysiphlebus testaceipes* are present and widely distributed in Dominica. *Lipolexis oregmae* was not purposefully released and it is not clear when and by which pathway the parasitoid was introduced to Dominica. The brown citrus aphid samples tested were parasitized by both parasitoids (80-100%), suggesting that both *L. oregmae* and *L. testaceipes* might be effective parasitoids of *T. citricidus* in Dominica.

ACKNOWLEDGMENTS

The authors thank Peter Hill, Naomi Commodore, Ryan Anselm, and Bernitta Serrant of the Ministry of Agriculture, Fisheries and the Environment, Commonwealth of Dominica, for assistance in collecting parasitized aphids. This research was funded by the Autonomous Region of Sardinia (A.C.) and the Davies, Fischer and Eckes Endowment in Biological Control (M.A.H.).

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