

Parasitoid (Hym., Braconidae, Aphidiinae) Complex of the Black Citrus Aphid, *Toxoptera citricidus* (Kirkaldy) (Hem., Aphididae) in Costa Rica and Its Relationships to Nearby Areas

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ABSTRACT

A survey was conducted to determine the parasitoid (Hym., Braconidae) complex of *Toxoptera citricidus* (Kirkaldy) in Costa Rica. The parasitoids were obtained by collecting the host aphid on its host plants, throughout various locations in the country. In total three species consisting, *Aphidius colemani* Viereck, *Lipolexis oregmae* (Gahan) and *Lysiphlebus testaceipes* (Cresson) were determined as parasitoids of *Toxoptera citricidus* (Kirkaldy) in Costa Rica. Faunal interrelationships with the other Central American-Caribbean areas were analysed. *L. oregmae* was detected to be derivable from two origins, from an unknown incidentally introduced one (hypothetically through the Panama strait from southeastern Asia) and from a purposely introduced population from Guam (via Florida).

Key words: Aphids, parasitoids, Costa Rica, *Lipolexis oregmae*, *Toxoptera citricidus*.

INTRODUCTION

The black citrus aphid, *Toxoptera citricidus* (Kirkaldy) is believed to be native to Southeastern Asia, now widely distributed in Asia, India, New Zealand, Australia, Pacific Islands, subsaharan Africa, Madagascar, Indian Oceanic Islands, South America, Central America, the Caribbean Basin, and in parts of the western Mediterranean (Invasive species compendium, 2011). Also, it is the most important aphid vector of Citrus tristeza disease in most of the distribution area (Batista *et al.*, 1995; Lastra *et al.*, 1991, 1992; Rocha *et al.*, 1995). The aphid was found in 1991 in Guadeloupe (Aubert *et al.*, 1992) and in 1992, in Dominican Republic and Puerto Rico (Lastra *et al.*, 1992). *T. citricidus* had spread into Central America including Costa Rica by 1989 (Lastra *et al.*, 1991; Voegtlin and Villalobos, 1992; Voegtlin *et al.*, 2003). It is now distributed also

in Cuba and Jamaica (Batista *et al.*, 1995; Yokomi *et al.*, 1994). Few parasitoids were recorded in association with *T. citricidus* (Starý, 1967a, Batista *et al.*, 1995). A single sub-effective parasitoid, *Lysiphlebia japonica* (Ashmead) is reported from its original area of distribution (Kato, 1970; Takanashi, 1991). On the other hand, the aphid has been targeted within the framework of biological control using the hymenopterous parasitoids in the area of invasion (Hoy and Nguyen, 2000; Hoy *et al.*, 2007; Persad *et al.*, 2007; Walker and Hoy, 2003). The objective of this study is to provide an initial insight of the parasitoids of *T. citricidus* in Costa Rica and to evaluate the geographic affinities, determination of the origin and distribution of the newly recorded species.

MATERIAL AND METHODS

Aphidiine parasitoids (Hym., Braconidae) were obtained by collecting the host aphid, *T. citricidus* from their host plants, since August 2008 to October 2009, throughout various locations in Costa Rica (Fig. 1). *T. citricidus* populations sampled in the field varied in size, and approximately 25 to 200 aphids were taken per sample. When the identity of the plant was unknown, samples were dried in a plant press for later identification. ArcMap ver. 9.2 software was used to create the sampling location and distribution maps (Figs. 1 and 10).

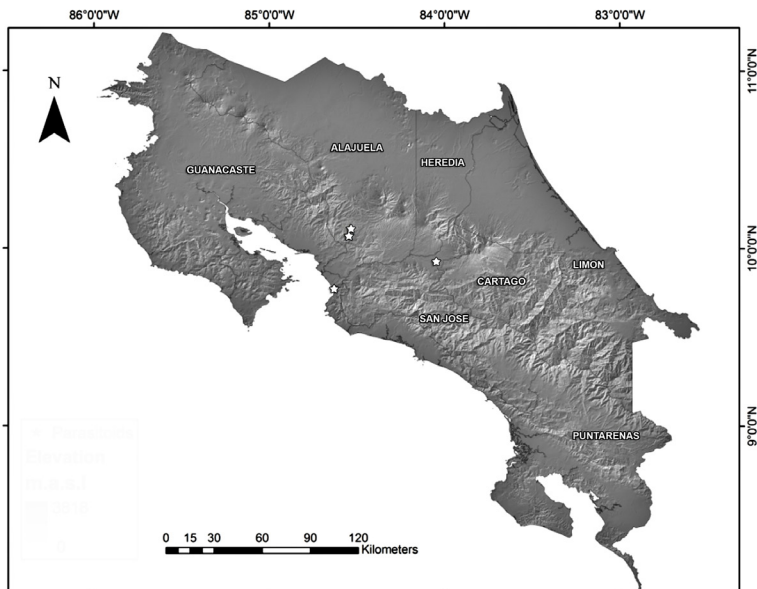


Fig. 1. Map of the sampling localities in Costa Rica. The localities, at which we found *L. oregmae* marked with white asterisk.

T. citricidus was the sole species present on the host plants. Each part of plant sampled with *T. citricidus*, was placed in square plastic containers of 10 cm in length and 10 cm height. The containers were maintained for 25-30 days under a temperature

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range of 24-28°C with constant ventilation and light. The samples were checked daily for emerged parasitoids. After emergence, the aphidiine parasitoids were placed in 70% ethanol for identification. The parasitoids were identified by P. Stary and E. Rakhshani within a framework of a broader revisional work. Carl Zeiss, Jena SM-XX and Olympus™ BH-2 phase contrast microscope were used for identification of parasitoids. The specimens are deposited in the Museum of Zoology at the University of Costa Rica and in P. Stary's collection (České Budějovice). All the material was sampled by the first author who also identified the aphids under supervision of second author, whom was his advisor during the thesis research. For the aphid identification a dissecting scope ZM-160 AT and a microscope Optima 6-303 were used. The relative abundance of each parasitoid species was also calculated based on the number of emerged wasps from all the samples.

RESULTS

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In total, 323 aphidiine specimens, comprising 3 species, were reared from *Toxoptera citricidus*, from a total of three localities. Below, the parasitoid species are listed in alphabetical order, along with their host plants, collecting locality, elevation, date of collection, number of specimens (spns.), sex proportion and (in parenthesis) lot number (Museum of Zoology, University of Costa Rica).

Aphidius colemani Viereck

Citrus sinensis: Puntarenas, Tárcoles, 60 m.a.s.l, 21-V- 09, 1♀ spn., (S-56).

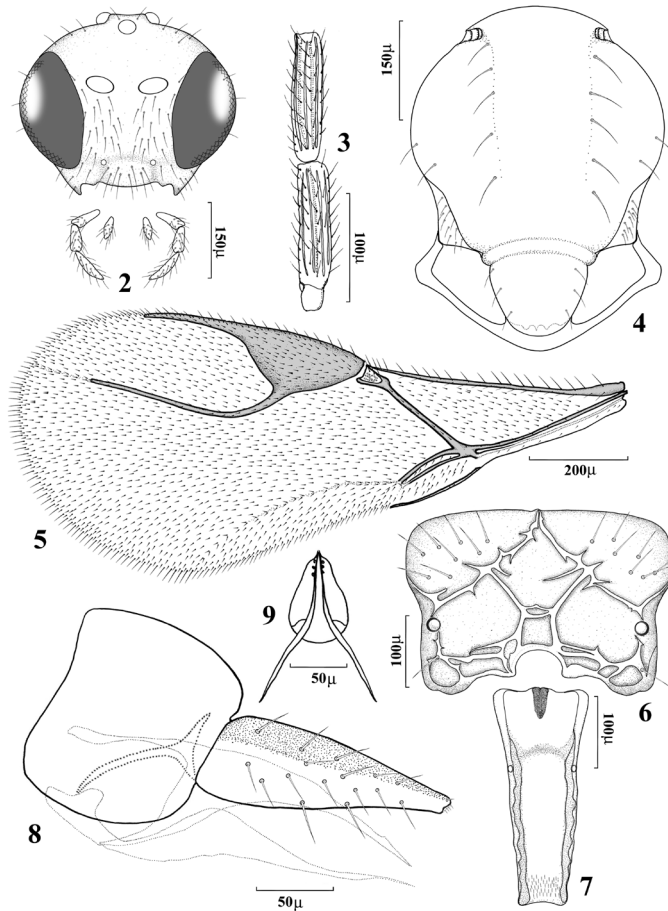
Lipolexis oregmae (Gahan)

Citrus aurantium: Alajuela, San Ramón, 916 m.a.s.l, 24-V-09, 1♀ spn. (S-58); on *Zanthoxylum* sp., Alajuela, San Ramón, 885 m.a.s.l, 13-VII-09, 132 (7♀♀, 5♂♂) spns., (S-68).

Lysiphlebus testaceipes (Cresson)

Citrus aurantium: Alajuela, San Ramón, 916 m.a.s.l, 10-IV-09, 59 (42♀♀, 17♂♂) spns. (S-33); 24-V-09, 6 (3♀♀, 3♂♂) spns., (S-58); on *Citrus sinensis*: San José, Montes de Oca, 1200 m.a.s.l, 17-IX-08, 17 (14♀♀, 3♂♂) spns. (S-9); Puntarenas, Tárcoles, 60 m.a.s.l, 21-V-09, 1♂ spn., (S-56); Alajuela, San Ramón, 916 m.a.s.l, 25-V-09, 59 (48♀♀, 11♂♂) spns., (S-59); on *Zanthoxylum* sp.: Alajuela, San Ramón, 885 m.a.s.l, 13-VI-09, 47 (36♀♀, 11♂♂) spns., (S-68).

The relative abundance of parasitoid species derived from the samples favors *L. testaceipes* (58.5%), followed by *L. oregmae* (41%), whereas *A. colemani* was apparently rare (0.5%). The morphological characters of the specimens of *L. oregmae* from Costa Rica (Figs. 2-9) compared with material from Florida (Konroe, Co. Sugarleaf, Hammok mal. Tr. VI 1986, S. J. Peck) and also from laboratory culture (Marjorie A. Hoy), Guam (Miller *et al.*, 2002) and India (*Greenidea* sp., *Psidium guajava*, 06 IV 2008, Aligarh, Z. Ahmed) and the results showed they are conspecific.



Figs. 2-9. Morphological characters of *Lipolexis oregmae* (Gahan): 2. frontal view of head and mouth parts. 3. First and second antennal flagellomeres. 4. Dorsal aspect of mesonotum. 5. Forewing. 6. Dorsal aspect of propodeum. 7. Dorsal aspect of metasomal tergum I. 8. Lateral aspect of female genitalia. 9. Aedeagus.

CONCLUSIONS AND DISCUSSION

Faunal analysis and relationships

The aphid parasitoid fauna of Costa Rica has been recently reviewed by Zamora Mejías *et al.* (2010). *Aphidius colemani* is a South-American species of presumably oriental origin (Starý, 1975) extending from the South to some areas of Central America. Its host range in Costa Rica is better illustrated if compared with its relatively complete range in Chile (Starý, 1995), Venezuela (Starý and Cermeli, 1989) and Brazil (Starý *et al.*, 2007). In general terms, it is associated prevalingly with the warmer areas (Starý, 1975).

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Lysiphlebus testaceipes is originally a North American species, expanding over the Central America to the most of South America. Its host range patterns from the North over the Central America to South America can be illustrated by several up-dated papers derived from tritrophic associations of this parasitoid species: Pacific Northwest USA (Pike *et al.*, 2000), Mexico (Starý and Remaudiere, 1982), Florida (Evans and Stange, 1997), Cuba (Starý 1968a, Batista *et al.*, 1995), Guadeloupe (Starý *et al.*, 1987), Trinidad (Bennett, 1985), Puerto Rico (Yokomi and Tang, 1996), Dominica (Cocco *et al.*, 2009), Chile (Starý, 1995), Costa Rica (Hanson and Gauld, 1995; Zamora Mejías *et al.*, 2010) and Brazil (Starý *et al.*, 2007). The host range patterns of *L. testaceipes* in Costa Rica may be expected to reflect the available aphid fauna, up to now, ten species of aphids were detected as hosts.

Both distribution and interaction of the two above mentioned species have varied in the Central America (Bennett, 1985; Batista *et al.*, 1995; Starý 1968a, 1972b; Starý *et al.*, 1987; Yokomi *et al.*, 1994; Cocco *et al.*, 2009).

Lipolexis oregmae has been newly determined in Costa Rica but its origin is worth of a special attention, related also to the situation as detected in Florida and in Central America. P. Starý has determined an older material of *L. oregmae* sampled in Florida (Konroe, Co. USA Florida, Sugarleaf, Hammok mal. Tr. VI 86, S. J. Peck), but the true origin of this population sounds unclear. Furthermore, no host evidence was known for those specimens.

Research on local aphid parasitoids of *T. citricidus* realized after its detection in Florida but brought merely *L. testaceipes* (Evans and Stange, 1997; Persad and Hoy, 2003). Release and subsequent establishment of *L. oregmae* (as *L. scutellaris* Mackauer), was undertaken in Florida, the original material sourcing from Guam (Guam biotype, Hoy and Nguyen, 2007). It was also recommended to be introduced into Bermuda and Jamaica (Hoy, 2005) and Dominica (Cocco *et al.*, 2009). The host range of the original population in Guam was listed by Miller *et al.* (2002).

However, the release programme realized in Dominica but attributed a rather peculiar and important situation as both *Lysiphlebus testaceipes* and *Lipolexis oregmae* were detected (from aphids of unknown species on weeds within citrus groves) prior to defined release of *L. oregmae* into this state. These data brought an evidence of both parasitoids to parasitize alternative hosts even prior to the presence of the brown citrus aphid. Secondly, of unknown mechanisms how *L. oregmae* was introduced to Dominica (Cocco *et al.*, 2009). The authors summarized this situation as a difficulty of preventing invasive insect introductions in the Caribbean islands as in Florida. Also, local information in Costa Rica sounds adversely for whatever purposeful introduction of aphid parasitoids into this country.

There are thus obvious interactions between the occurrence of *L. oregmae* and its purposeful introduction in the Caribbean and Central America. As there is also an interference of the research level on the fauna of parasitoids in the area, we may merely hypothesize and derive the probable origin of the target. *L. oregmae* is doubtlessly a tropical species, apparently easily transferrable with the aphids on nursery seedlings

and fruits, most probably through the Panama strait (Fig. 10). No evidence has been known from this area, but the up-dated evidence from Costa Rica presents two different hosts, including the common *T. citricidus* throughout the country and *Aphis illinoisensis*, from Coto Brus, Puntarenas, close to Panama border of Panama. Similarly, the parasitoid might be present earlier - although not evidenced - in Dominica and related Jamaica. For sure, not in Cuba until about 1965 (Starý, 1967b, 1972b) and Trinidad (Bennett, 1985). Also, another important phenomenon is the oligophagy of *L. oregmae* which does not make it primarily dependent on *T. citricidus*.

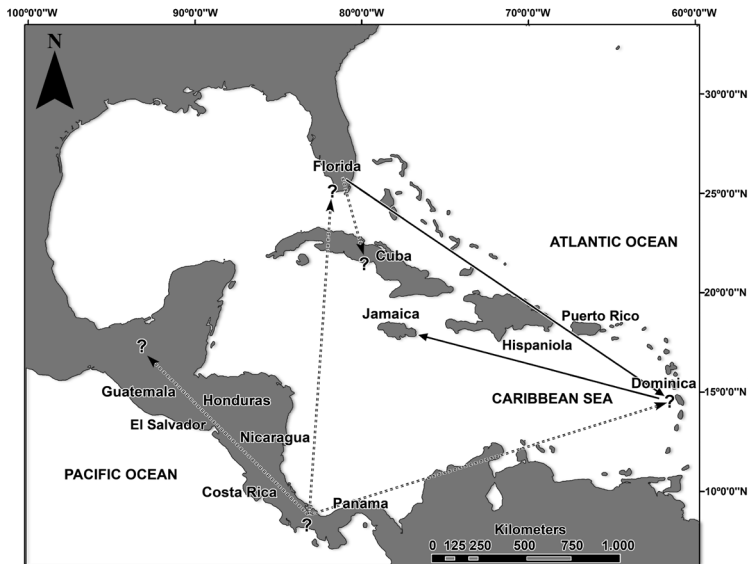


Fig. 10. Pattern of distribution for *Lipolexis oregmae* at Central America and in Caribbean area. The solid arrows showing the purposeful introduction of the parasitoid, while dashed arrows showing the accidental invasion, hypothetically through Panama strait.

Refugiums and ecosystem relationships

Aphid parasitoids manifest a wide range of ecosystem relationships, if their host range is followed. They follow, as a parasitoid group, rather closely the occurrence of aphids, but their respective ecosystem relationships may be almost identical or rather different (Starý, 1972a). This phenomenon may be also easily exemplified at least in some cases in Costa Rica. For example, *Aphis nerii* Boyer de Fonscolombe is specific to *Nerium oleander* and some other plants, whereas *Toxoptera citricidus* is mainly associated with *Citrus* and other Rutaceae, both of them are parasitised by *A. colemani* and *L. testaceipes*. Thus, parasitoids may alternate between *Aphis nerii*/*Nerium* and *Toxoptera aurantii*/*Citrus*. There are thus different parasitoid refugiums which - through host alternation (switching from one to another host aphid) may more or less interact in the course of the season. Similar relationships may be derived after more information from field on both the groups is obtained. Such research approaches

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were also described earlier, for example, in Cuba (Starý, 1967b; 1968a,b,c,d) or recently emphasized in Florida (Hoy, 2005; Hoy and Nguyen, 2007; Persad *et al.*, 2007). In the later case, the alternate hosts of *L. oregmae*/*T. citricidus* are *Aphis spiraecola* Patch, *A. gossypii* Glover, *A. craccivora* Koch and *Toxoptera aurantii* Boyer de Fonscolombe. In our opinion, the parasitoid complex (=guild), aphids host range and derived ecosystem relationships in Florida might finally evolve to the situation known from Guam (Miller *et al.*, 2002) or Central and South American countries.

Among the three parasitoid species of the black citrus aphid, *L. testaceipes* was the most frequently collected species, while *L. oregmae* occupied the second place in Costa Rica. The origin of *L. oregmae* has two sources in the Central America and in the Caribbean: One is accidental, hypothetically from the south-eastern Asia (over the Panama strait), the another one - the purposeful introduction - is originally from Guam via Florida. In Florida, both populations might also have interacted. Both the accidental (Costa Rica and Guam - Florida populations have been found conspecific (Hoy *et al.*, 2007 and our present results). Further studies using molecular markers on larger samples, both from original area of distribution and invaded area is necessary to confirm the presence of identical species with two purposefull and accidentally introduced population of *L. oregmae* in Caribbean and Central America.

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