Parasitoids of *Dasiops* (Diptera: Lonchaeidae) in sweet granadilla orchards in an Andean forest of Peru

Paolo Salazar Mendoza,* Yoseph Ojeda-Enriquez and Carmen Salcedo-Velarde

*Departamento de Entomologia e Acarologia, Universidade de São Paulo, Piracicaba, Brazil; 4Escuela de Formación Profesional de Agronomía, Universidad Nacional Daniel Alcides Carrión, Oxapampa, Pasco, Peru; 5Subdirección de Control Biológico, Servicio Nacional de Sanidad Agraria, Vitarte, Lima, Peru

**ABSTRACT**

We report hymenopteran larval and pupal parasitoids of the *Dasiops* species in sweet granadilla (*Passiflora ligularis*) orchards in a Peruvian Andean forest. Flower bud and fruit samples infested by *Dasiops* were collected in three orchards from Oxapampa (Pasco, Peru). In addition, we exposed non-parasitized *Dasiops* pupae at ground level to verify the presence of pupal parasitoids. We identified Utetes anastrephae (Viereck) and Opis sp. (Hymenoptera: Braconidae) on flower buds infested by *Dasiops inedulis* Steyskal. However, Aganaspis pelleronai (Brèthes) (Hymenoptera: Figitidae) emerged from *D. frieseni* pupae reared from unripe fruits. Also, the pupal parasitoids *Pachycrypeodeus vindemmiae* (Rondani) (Hymenoptera: Pteromalidae) and *Coptera* sp. (Hymenoptera: Diapriidae) emerged from pupae of both *D. inedulis* and *D. frieseni* exposed to parasitism at ground level. Our findings shed new light into interactions for species in the genus *Dasiops* and their larval and pupal parasitoids in sweet granadilla agroecosystems from the Neotropical region.

**RESUMEN**

El objetivo de esta investigación fue registrar las especies de hymenópteros parasitoides de larva y pupa de *Dasiops* en fincas de granadilla (*Passiflora ligularis*) localizadas en una selva andina de Perú. Muestras de botones florales y frutos infestados por *Dasiops* fueron colectados en tres fincas en la provincia de Oxapampa (Pasco, Perú). Asimismo, pupas de *Dasiops* no parasitadas fueron expuestas al suelo para la verificación de parasitoides de pupa. Identificamos a *Utetes anastrephae* (Viereck) y Opis sp. (Hymenoptera: Braconidae) en botones florales infestados por *Dasiops inedulis* Steyskal. Asimismo, Aganaspis pelleronai (Brèthes) (Hymenoptera: Figitidae) emergieron de pupas de *D. frieseni* Norrbom & McAlpine criados en frutos. Por otro lado, los parasitoides de pupa *Pachycrypeodeus vindemmiae* (Rondani) (Hymenoptera: Pteromalidae) y *Coptera* sp. (Hymenoptera: Diapriidae) emergieron de pupas de *D. inedulis* y *D. frieseni* expuestas al parasitismo en el suelo. Nuestros hallazgos brindan nuevas luces sobre nuevas interacciones de especies del género *Dasiops* y sus parasitoides de larva y pupa en agroecosistemas de granadilla para la región Neotropical.

In agroecosystems, cultivated plants are constantly damaged by key insect pests who promote significant losses in production for the farmers [1]. Among these insects, the lance flies (Diptera: Tephritoidea: Lonchaeidae) are important pests of several commercial and non-commercial passion flowers (*Passifloraceae*) in the Neotropical region. In Peru, two *Dasiops* species have recently been reported to cause damage on sweet granadilla. *Dasiops inedulis* Steyskal that feeds on ovaries and anthers of flower buds causing it to rot and drop to the ground, and *Dasiops frieseni* Norrbom & McAlpine that feeds on the pulp and seeds of young unripe fruits causing them to wrinkle [2,3]. These insects cause high losses in fruit production and infestation levels can reach up to 80% in commercial passion flowers [4,5].

Native parasitoids play an important function in the natural suppression of various pest species of Tephritoidea in agroecosystems in the Neotropical region [6]. In this regard, biological control may help suppress *Dasiops* population densities and provide a new tool to reduce environmental risks and improve public health. However, to date, there is limited information available on the tritrophic interactions among passion flowers, *Dasiops*, and their parasitoids. Thus, we aimed to identify the diversity of larval and pupal parasitoids associated with *Dasiops* species in sweet granadilla orchards in an Andean forest of Peru.

Field trials were done from January to August 2013 in three conventional sweet granadilla orchards (2–4 years old; 1.5–2 ha). Each orchard (named “Cantarizú,” “Grapanazú,” and “San Marcos”) was
located in three different districts in Oxapampa (Pasco, Peru) (Figure 1). Oxapampa is a province located on the eastern slope of the Andean mountains (central tropical forest) and recognized as the principal production area of sweet granadilla in Peru, with half of the total national Peruvian production [7]. All sweet granadilla orchards used in this study were surrounded by native forest associated with some cultivated plants such as lemon (Citrus limonia Osbeck) and guava (Psidium guajava L) in Cantarizu; coffee (Coffea arabica L), chili pepper (Capsicum annuum L), and guava in Grapanazú; and loquat (Eriobotrya japonica L), stuffing cucumber (Cyclanthera pedata Schrad.), and lucumo (Pouteria lucuma [Ruiz and Pav.]) in San Marcos.

We collected twelve flower buds (Figure 2(a)) and fruit (Figure 2(b)) samples (a sample = eight flower buds or four fruits) in six different weeks from each sweet granadilla orchard. Flower buds and fruits showing symptom of Dasiops damage were sampled to increase the probability of finding larvae. After field collection, samples were taken to the laboratory at Servicio Nacional de Sanidad Agraria (SENASA, Oxapampa) for further processing. In the laboratory, individual samples were weighed and placed in plastic cups (15 cm high x 8 cm diameter) for flower buds or in Styrofoam boxes (38 x 20 x 20 cm) for fruits. In each container, a layer (3 cm) of sterilized and moistened sand was placed in the bottom to allow larval development and pupal formation. Three weeks after field collection, containers were checked, and flower buds and fruits were dissected, for presence of fruit fly larvae or pupae. Pupae were subsequently transferred to plastic cups (250 ml) with sterilized and moistened sand and covered with a fine mesh on the top. Daily, for up to 45 days, plastic cups were checked for adult Dasiops emergence or emergence of larval parasitoids.

To detect pupal parasitoid species in the field, we exposed D. inedulis and D. fieseni pupae to parasitism using the sentinel pupal technique [8]. Pupae were obtained from fruits and flower buds that were infested in the laboratory (and were thus unparasitized). Transparent plastic containers (11 cm diameter x 7 cm height, 250 ml) with the upper part open were buried in the ground under the canopy of sweet granadilla plants and filled with organic substrate. Thereafter, 20 pupae (12–48 hours-old) of the two Dasiops species were exposed separately in different containers. In order to avoid damage by predators and abiotic factors while still allowing access to parasitoids, the containers with pupae were placed inside a nylon mesh (1.5 mm diameter holes) and remained covered under a thin layer of organic substrate (Figure 2(c,d)). Holes were made to the bottom of the containers to avoid water saturation. Monthly, from March to August 2013, pupae of both species were placed in each orchard for 72 hours. After field exposure, pupae were placed inside Petri dishes (10 cm diameter
x 1.5 cm height), and taken to the laboratory. Daily, for up to 35 days, Petri dishes were checked for adult Dasiops emergence or emergence of pupal parasitoids. Laboratory conditions were kept at 25 ± 3°C, 65 ± 10% RH, and 12:12 L:D.

Dasiops species identification were confirmed by Pedro Carlos Strikis (Universidade de São Paulo). Larval and pupal parasitoid species were identified based on taxonomical keys. Some parasitoid specimens were confirmed by Angelica Penteado Dias (Universidade Federal de São Carlos, Brazil) and Valmir Costa (Instituto Agronômico de Campinas, Brazil). Voucher specimens of Dasiops and their parasitoids were deposited at the Entomology Museum of the Universidad Nacional Daniel Alcides Carrión (Oxapampa, Pasco, Peru). The parasitism levels found in larvae and pupae of Dasiops were calculated according to the formula: percent parasitism = a/(a + b) × 100, where a = number of emerged adult parasitoids, and b = number of emerged adult Dasiops from each sample.

A total of 439 D. inedulis pupae and 29 adult parasitoids were obtained from flower buds, resulting in a mean parasitism of 7.3%. Utetes anastrephae (Viereck) (Hymenoptera: Braconidae) was the most abundant (78.9%) of all emerged parasitoids, followed by Opis sp. (21.1%) (Hymenoptera: Braconidae) (Table 1). While only two Aganaspis pelleranoi (Brèthes) (Hymenoptera: Figitidae) individuals emerged from 199 D. frieseni pupae collected from fruits, with an overall mean parasitism of 2.2% (Table 1).

Two parasitoid species were obtained from exposed D. inedulis and D. frieseni pupae in the field. The wasps Pachycrepoideus vindemmiae (Rondani) (Hymenoptera: Pteromalidae) and Coptera sp. (Hymenoptera: Diapriidae) (Table 2). For D. inedulis pupae, P. vindemmiae was the most abundant (71.4%), followed by Coptera sp. (28.6%); resulting in an overall mean parasitism of 5.3%. In contrast, for D. frieseni pupae, Coptera sp. (87.5%) was more abundant than P. vindemmiae (12.5%); resulting in an overall mean parasitism of 3.8% (Table 2).
Table 1. Parasitism rates of larval parasitoids recorded from *Dasiops* species collected in flower buds and fruits at three sweet granadilla orchards in Oxpampa, Pasco, Peru.

| Location    | Tissue infested | No. fruit/flower bud | Weight (kg) | No. Pupae | Dasiops Parasitoids | % Parasitism | % of total parasitoids |
|-------------|----------------|----------------------|-------------|-----------|---------------------|--------------|------------------------|
| Cantarízú  | Flower bud      | 96                   | 0.77        | 112       | 94                  | 4            | 4.1                    | 100 | 0 | 0 |
| Grananazú  | Flower bud      | 96                   | 0.81        | 149       | 113                 | 10           | 8.1                    | 50  | 0 | 0 |
| San Marcos  | Flower bud      | 96                   | 0.79        | 178       | 139                 | 15           | 9.7                    | 86.7| 13.3 | 0 |
| Cantarízú  | Fruit           | 48                   | 3.6         | 55        | 31                  | 0            | 0                      | 0   | 0 | 0 |
| Grananazú  | Fruit           | 48                   | 4.1         | 90        | 56                  | 0            | 0                      | 0   | 0 | 0 |
| San Marcos  | Fruit           | 48                   | 3.5         | 54        | 28                  | 2            | 6.7                    | 0   | 0 | 100 |
| Total       | Flower bud      | 288                  | 2.37        | 439       | 346                 | 29           | 7.3                    | 78.9| 21.0 | 0 |
| Total       | Fruit           | 144                  | 11.2        | 199       | 115                 | 2            | 2.2                    | 0   | 0 | 33.3 |

Table 2. Parasitism rates of pupal parasitoids recorded on *Dasiops* species at three sweet granadilla orchards in Oxpampa, Pasco, Peru.

| Location    | Pupae | No. pupae exposed | Adult emergence | % Parasitism | % of total parasitoids |
|-------------|-------|-------------------|-----------------|--------------|------------------------|
| Cantarízú  | *D. inedulis* | 120               | 45              | 0            | 0                      | 0             | 0                      |
| Grananazú  | *D. inedulis* | 120               | 55              | 5            | 12.5                   | 60            | 40                     |
| San Marcos  | *D. inedulis* | 120               | 58              | 2            | 3.3                    | 100           | 0                      |
| Cantarízú  | *D. friesi*   | 120               | 67              | 7            | 8.2                    | 16.7          | 83.3                   |
| Grananazú  | *D. inedulis* | 120               | 61              | 2            | 3.2                    | 0             | 100                    |
| San Marcos  | *D. inedulis* | 120               | 39              | 0            | 0.0                    | 0             | 0                      |
| Total       | *D. inedulis* | 360               | 138             | 7            | 5.3                    | 71.4          | 28.6                   |
| Total       | *D. friesi*   | 360               | 167             | 8            | 3.8                    | 12.5          | 87.5                   |

Larval parasitoids are the most commonly reported natural enemies attacking insects of the superfamily Tephritioidea in the Neotropical region, particularly on fruit flies (Diptera: Tephritidae) [6]. However, some surveys done on lance flies in Colombia [5,9–11], Peru [12] and Brazil [13] have recorded parasitoids attacking *Dasiops*

Table 3. Larval and pupal parasitoids recorded in association with *Dasiops* species-host reports in passion flowers from South America.

| Parasitoids | State/Country | Host | Tissue affected | Passion flowers | References |
|-------------|---------------|------|----------------|-----------------|------------|
| *Braconidae* |               |      |                |                 |            |
| *Aspilota sp.* | Huila/Colombia | *D. inedulis* | Flower bud | *P. ligularis* | [9]        |
| *Microcrasis sp.* | Cundinamarca/Colombia | *D. gracilis* | Flower bud | *P. edulis f. edulis; P. edulis* | [10]      |
| *Utetes sp.* | Valle del Cauca/Colombia | *D. inedulis* | Flower bud | *P. edulis f. flavicarpa* | [5] |
| *Utetes anastrephae* | Valle del Cauca/Colombia | *D. inedulis* | Flower bud | *P. edulis f. flavicarpa* | [11] |
| *Opus sp.* | Espírito Santo/Brazil | *D. inedulis* | Flower bud | *P. edulis f. flavicarpa* | [13] |
| *Figitidae* |               |      |                |                 |            |
| *Aganaspis sp.* | Valle del Cauca/Colombia | *D. inedulis* | Flower bud | *P. edulis f. flavicarpa* | [11] |
| *Aganaspis pelleranoi* | Cundinamarca/Colombia | *D. inedulis* | Flower bud | *P. ligularis* | [10] |
| *Cundinamarca/Colombia* | Cundinamarca/Colombia | *D. inedulis* | Flower bud | *P. edulis f. flavicarpa* | [10] |
| *Cundinamarca/Colombia* | Cundinamarca/Colombia | *D. yepezii* | Fruit | *P. ligularis* | [10] |
| *Pasco/Peru* | *D. friesi** | Fruit | *P. ligularis*** | [10] |
| *Pteromalidae* |               |      |                |                 |            |
| *Spaliasp. ev. vindemmiae* | Valle del Cauca/Colombia | *D. inedulis* | Flower bud | *P. edulis f. flavicarpa* | [11] |
| *Pachycyropeidea vindemmiae* | Huila/Colombia | *D. inedulis* | Flower bud | *P. ligularis* | [5,9] |
| *Pasco/Peru* | *D. inedulis* | Flower bud | *P. ligularis*** | [11] |
| *Diapriidae* |               |      |                |                 |            |
| *Pentrapia sp.* | Huila, Cundinamarca/Colombia | *D. inedulis* | Flower bud | *P. ligularis* | [9,10] |
| *Boyacá/Colombia* | *D. caustonae* | Fruit | *P. molliusma* | [10] |
| *Boyacá, Cundinamarca/Colombia* | *D. gracilis* | Fruit | *P. edulis f. edulis; P. edulis* | [10] |
| *Basalys sp.* | Huila/Colombia | *D. inedulis* | Flower bud | *P. ligularis* | [9] |
| *Trichopia sp.* | Cundinamarca/Colombia | *D. gracilis* | Fruit | *P. edulis f. edulis; P. edulis* | [10] |
| *Coptera sp.* | Pasco/Peru | *D. inedulis* | Flower bud | *P. edulis f. edulis* | [10] |

* First parasitoid reports; ** New *Dasiops* species-host reports ***New passion flower-host reports
larvae in passion flowers (Table 3). The koinobiont endoparasitoids *U. anastrephae* and *Opis* sp. (that emerged from *D. inedulis*) and *A. pelleranoi* (that emerged from *D. frieseni*) were three larval parasitoids identified in this study. These parasitoids have also been found frequently parasitizing fruit fly species in other fruit hosts in several countries of the Neotropical region [14]. Overall, *U. anastrephae* was the most abundant larval parasitoid that emerged from *D. inedulis* and was found in all sweet granadilla orchards sampled in our study. Because our studies were conducted in different landscape environments in terms of vegetation surrounding the orchards, we consider this parasitoid to be the most versatile and promising candidate for future biological control studies on *D. inedulis* in sweet granadilla. Moreover, despite the hardness of the sweet granadilla fruit epicarp, our study found *A. pelleranoi* as the only parasitoid emerging from *D. frieseni*. Aluja et al. [15] suggested that *A. pelleranoi* females commonly penetrate the fruit in search of *Anastrepha* spp. larvae through punctures. Likely, wrinkles on the fruit surface caused by *D. frieseni* could facilitate the access of *A. pelleranoi* in sweet granadilla. Additional studies are needed to better understand the behavior of this wasp when parasitizing *D. frieseni*.

Five species of pupal parasitoids belonging to the families Pteromalidae and Diapriidae have been associated with *Dasiops* in passion flowers cultivated in Colombia [5,9–11] (Table 3). No other pupal parasitoid survey has been done on *Dasiops* in South America. In our study, we found for the first time *Coptera* sp. emerging from both *D. inedulis* and *D. frieseni* pupae in sweet granadilla. There have been no reports of *Coptera* sp. parasitizing *Dasiops* pupae in other passion flower crops. Endoparasitoids in the genus *Coptera* have been widely associated with many fly families, such as Muscidae, Drosophilidae, Lonchaeidae and Tephritidae [16]. For example, *Coptera haywardi* Loiácono has been considered as a promising candidate for fruit fly biological control [17]. Likewise, our study found that *P. vindemianae* attacks pupae of the two *Dasiops* species in sweet granadilla. This generalist pupal ectoparasitoid usually attacks many cyclorrhaphous flies including various fruit fly species [18]. In recent years, *P. vindemianae* has received increasing attention as a potential candidate for biological control of the invasive pest *Drosophila suzukii* (Matsumura) [19].

In summary, this study reports three larval parasitoids and two pupal parasitoids of *Dasiops* species in sweet granadilla orchards in an Andean forest of Peru. *Utetes anastrephae*, *Opis* sp., and *Coptera* sp. are reported for the first time parasitizing *Dasiops* in sweet granadilla. This information will assist future research on the development of biological control programs for *Dasiops* pests in this cropping system and region. Future research on these parasitoids should aim at their conservation and augmentation in biological control programs for suppressing *Dasiops* populations in sweet granadilla and other passion flower crops.

**Acknowledgments**

The authors are grateful to Dr. Pedro Strikis, Dr. Angelica Penteado Dias, and Dr. Valmir Costa for insect identification, to SENASA for allowing us to rear the insects in their laboratory, and to two anonymous reviewers for helpful comments on an earlier draft of this manuscript. Also, we thank the sweet granadilla growers who allowed us access to their orchards.

**Disclosure statement**

No potential conflict of interest was reported by the author(s).

**Funding**

The present work was funded by the authors’ own resources.

**ORCID**

Paolo Salazar Mendoza http://orcid.org/0000-0001-6791-1616

**References**

[1] Schoonhoven LM, Van Loon B, and van Loon JJ, et al. Insect-plant biology. : Oxford University Press on Demand; 2005: 421. https://books.google.com.pe/books?id=es&lr=&id=zQRDAAAQBAJ&oi=fnd&pg=P%20R%207%20o%20t%20s%20=1%209%20d%20m%20T%20J%20p%20D%20Q%20Y%20&sig=hzRcESDLu9Y%20SN%20QX%20U%20v%20O%20m%20v%20C%20V%20=%20onepage&q=false
[2] Salazar-Mendoza PS, Peralta-Aragón IE, Misialidis ML, et al. Lance flies associated with sweet passion fruit and contributions to the knowledge on Lonchaeidae in Peru. Arq Inst Biol. 2019; 86: 1–4.
[3] Salazar-Mendoza P, Ninahumán-Calderón A, Girón Fernández C, et al. First record of *Anastrepha pseudoparaellela* Loew (Diptera: Tephritidae) infesting passion flowers in Peru. Rev Peru Biol. 2020;27(2):229–232.
[4] Wyckhuys KA, Acosta FL, Rojas M, et al. The relationship of farm surroundings and local infestation pressure to pest management in cultivated Passiflora species in Colombia? Int J Pest Manag. 2010;57(1):1–10.
[5] Wyckhuys KA, Korytkowski C, Martinez J, et al. Species composition and seasonal occurrence of *Diptera* associated with passionfruit crops in Colombia. Crop Prot. 2012;32:90–98.
[6] García FR, Ovruski SM, Suárez L, et al. Biological control of tephritid fruit flies in the Americas and Hawaii: a review of the use of parasitoids and predators. Insects. 2020;11 (10):662.
[7] Ministerio de Desarrollo Agrario y Riego. Compendio Estadístico Perú [Statistical Compendium Peru]. Lima (Peru); 2018. Chapter 13, Agrario (pp. 951–1038). Available from: https://www.inei.gob.pe/media/MenuRecursivo/publicaciones_digitales/Est/Lib1635/cap13/cap13.pdf
[8] Petersen J, Watson DW. Comparison of sentinel and naturally occurring fly pupae to measure field parasitism by pteromalid parasitoids (Hymenoptera). Biol Control. 1992;2(3):244–248.
[9] Amaya OS, Devia EHV, Salamanca J. Prueba de extractos vegetales para el control de Dasiops spp., en granadilla (Passiflora ligularis Juss.) en el Huila, Colombia [Test of plant extracts for the control of Dasiops spp., in sweet granadilla]. Cienc Tecnol Agropecuaria. 2009;10(2):141–151.

[10] Santamaría M, Ebratt E, Castro A, et al. Hymenopterous parasitoids of Dasiops (Diptera: Lonchaeidae) infesting cultivated Passiflora spp. (Passifloraceae) in Cundinamarca and Boyaca, Colombia. Agron Colomb. 2016;34(2):200–208.

[11] Quintero EM, Lopez IC, Kondo T. Manejo integrado de plagas como estrategia para el control de la mosca del botán floral del maracuyá Dasiops inedulis Steyskal (Diptera: Lonchaeidae) [Integrated pest management as a strategy for the control of the flower bud fly Dasiops inedulis Steyskal (Diptera: Lonchaeidae) in passion fruit]. Cienc Tecnol Agropecuaria. 2012;13(1):31–40.

[12] Salazar-Mendoza P, Romero-Rivas C. Eficacia de trampas y atrayentes para moscas de la granadilla (Passiflora ligularis Juss) [Efficacy of traps and lures to fly in sweet granadilla (Passiflora ligularis Juss)]. Rev Per Entomol. 2016;51(2):31–37.

[13] Aguiar-Menezes EL, Nascimento RJ, Menezes EB. Diversity of fly species (Diptera: tephritoidae) from Passiflora spp. and their hymenopterous parasitoids in two municipalities of the southeastern Brazil. Neotrop Entomol. 2004;33 (1):113–116.

[14] Schlsimer P, Ovruski SM, De Coll OR, et al. Diversity and abundance of hymenopterous parasitoids associated with Anastrepha fraterculus (Diptera: tephritidae) in native and exotic host plants in misiones, Northeastern Argentina. Fla Entomol. 2010;93(2):175–182.

[15] Aluja M, Ovruski SM, Guillén L, et al. Comparison of the host searching and oviposition behaviors of the tephritid (Diptera) parasitoids Aganaspis pelleranoi and Odontosema anastrephae (Hymenoptera: figitidae, Eucolilinae). J Insect Behav. 2009;22(6):423–451.

[16] Muesebeck CFW. The Nearctic parasitic wasps of the genera Psillus Panzer and Coptera Say (Hymenoptera, Proctotrupoidae, Diapriidae). Washington, U.S. Department of Agriculture; 1980. (Technical Bulletin, no. 1617).

[17] Aguiar-Menezes EL, Menezes EB, Loiácono MS. First record of Coptera haywardi Loiácono (Hymenoptera: Diapriidae) as a parasitoid of fruit-infecting Tephritidae (Diptera) in Brazil. Neotrop Entomol. 2003;32(2):355–358.

[18] López M, Aluja M, Sivinski J. Hymenopterous larval–pupal and pupal parasitoids of Anastrepha flies (Diptera: tephritidae) in Mexico. Biol Control. 1999;15 (2):119–129.

[19] Stacconi RMV, Buffington M, Daane KM, et al. Host stage preference, efficacy and fecundity of parasitoids attacking Drosophila suzukii in newly invaded areas. Biol Control. 2015;84:28–35.