

LABORATORY PARASITISM BY *PHYMASTICHUS COFFEA* (HYMENOPTERA: EULOPHIDAE) UPON NON-TARGET BARK BEETLES ASSOCIATED WITH COFFEE PLANTATIONS

ALFREDO CASTILLO¹, FRANCISCO INFANTE¹, GUILLERMO LÓPEZ¹, JAVIER TRUJILLO²,
LAWRENCE R. KIRKENDALL³ AND FERNANDO E. VEGA⁴

¹El Colegio de la Frontera Sur (ECOSUR), Carretera Antiguo Aeropuerto km 2.5.
Tapachula 30700 Chiapas, Mexico

²Colegio de Posgraduados, Instituto de Fitosanidad, Montecillo, 56230 Edo. de México, Mexico

³Institute of Zoology, University of Bergen, Allégaten 41, N-5007 Bergen, Norway

⁴Insect Biocontrol Laboratory, U. S. Department of Agriculture, Agricultural Research Service
Beltsville, Maryland, 20705-2350, USA

ABSTRACT

Phymastichus coffea (LaSalle) is an African parasitoid of adults of the coffee berry borer *Hypothenemus hampei* (Ferrari) that has been introduced to Mexico and other Central and South American countries for the biological control of this important pest. The present study assessed the host specificity of this parasitoid in the laboratory. We tested the acceptance and parasitism of *P. coffea* on five species of bark beetle adults commonly found in coffee plantations of Mexico: *Hypothenemus crudiae*, *H. plumeriae*, *H. eruditus*, *Scolytodes borealis* and *Araptus fossifrons*. As a control, we used adults of *H. hampei*, the natural host. *P. coffea* parasitized and successfully completed its life cycle in *H. crudiae* and *H. eruditus*, as well as in *H. hampei*. The degree to which bark beetles were attacked by *P. coffea* was estimated by percent of parasitism, which was 64% for *H. hampei*, 14% for *H. crudiae*, and 6% for *H. eruditus*. The risk of potential deleterious effects of the parasitoid on non-target organisms in coffee agroecosystems is discussed.

Key Words: Host specificity, *Phymastichus*, *Hypothenemus*, *Scolytodes*, *Araptus*, Mexico.

RESUMEN

Phymastichus coffea (LaSalle) es un parasitoide africano de adultos de la broca del café *Hypothenemus hampei* (Ferrari) que ha sido introducido a México y otros países de Centro y Sudamérica para el control biológico de esta importante plaga. El presente trabajo se llevó a cabo con la finalidad de evaluar la especificidad de huéspedes de este parasitoide en el laboratorio. Se probó la aceptación y parasitismo de *P. coffea* sobre adultos de cinco especies de descortezadores comúnmente encontrados en plantaciones de café de México: *Hypothenemus crudiae*, *H. plumeriae*, *H. eruditus*, *Scolytodes borealis* y *Araptus fossifrons*. Como control se usaron adultos de *H. hampei* (hospedero natural). *P. coffea* parasitó y completó exitosamente su ciclo biológico en sólo dos especies de escolítidos, *H. crudiae* y *H. eruditus*, además de *H. hampei*. El grado en el cual los descortezadores fueron atacados por *P. coffea* fue estimado por el porcentaje de parasitismo el cual fue de 64% para *H. hampei*, 14% para *H. crudiae*, y 6% para *H. eruditus*. Es discutido el riesgo de los efectos negativos potenciales de este parasitoide sobre organismos no blanco en agroecosistemas de café.

Translation provided by the authors.

Phymastichus coffea LaSalle (Hymenoptera: Eulophidae) is a primary parasitoid of the coffee berry borer (CBB), *Hypothenemus hampei* (Ferrari) (Coleoptera: Scolytidae), the most devastating pest of coffee throughout the world (Baker 1999). This parasitoid, indigenous to Africa, was first recorded in 1987 from Togo parasitizing the CBB (Borbon-Martínez 1989), and subsequently was described as a new species (LaSalle 1990). *Phymastichus coffea* has a pan-African distribu-

tion, having been collected from east Africa (Kenya) and west Africa (Togo, Benin, Cameroon, Burundi, and Ivory Coast) (Infante et al. 1992). So far, this parasitoid has been introduced to coffee producing countries, such as Colombia, Guatemala, Honduras, Jamaica, El Salvador, Ecuador, India, Brazil and Mexico for biological control purposes (Castillo, unpublished data). This species is the only known parasitoid of adults of CBB, and is considered to be a potentially useful tool in

integrated pest management programs against *H. hampei* (López-Vaamonde & Moore 1998; Baker 1999).

Eulophidae is one of the largest families in Hymenoptera with nearly 4000 described species (Noyes 1998; Gauthier et al. 2000). The subfamily Tetrastichinae, to which *P. coffea* belongs, has an extraordinarily wide host range and exhibits a great variety of life styles. Members of this subfamily attack over 100 families of insects, as well as mites, spider eggs, and even nematodes (J. LaSalle, pers. comm.). Despite the fact that *P. coffea* has already been imported and released in various countries of the Americas, it is important to determine whether this parasitoid can attack non-target scolytids.

The objective of this study was to test the host specificity of *P. coffea* in the laboratory with six bark beetles species commonly found in coffee plantations of Mexico, with *H. hampei* serving as the control. A previous study in Colombia reported that *P. coffea* was able to parasitize *H. obscurus*, *H. seriatus*, and *Araptus* sp. in the laboratory (López-Vaamonde & Moore 1998). We included different species than those examined in Colombia; thus, this study serves to further elucidate the potential risk of *P. coffea* for non-target scolytids.

MATERIALS AND METHODS

Parasitoids

We used mated females of *P. coffea*, which were less than 1h old from the time they emerged from CBBs. The parasitoids were obtained from a colony established in the laboratory in March of 2000. The colony was initiated with insects imported from Guatemala with methodology described by Infante et al. (1994). Adult CBB females obtained from the field were used for parasitoid rearing. The insect colony is normally maintained at $26 \pm 2^\circ\text{C}$, $75 \pm 10\%$ RH and 8:16 (L:D) photoperiod.

Hosts

We offered six species of bark beetles collected as adults in coffee plantations near Tapachula, Chiapas to adults of *P. coffea* in the laboratory. The parasitoid was never released in the plantations where we collected the bark beetles. To minimize the risk, the CBBs were obtained by dissecting infested coffee fruits from the field, while we collected *Hypothenemus eruditus* Westwood, *Hypothenemus crudiae* (Panzer), *Hypothenemus plumeriae* (Nordlinger), and *Scolytodes borealis* Jordal from petioles of *Cecropia* sp. leaves. The three *Hypothenemus* species we used are widespread and co-occur with *Coffea* spp. where the latter are native (Wood 1982; Wood & Bright 1992). They are extremely polyphagous, and have

been collected from dozens of host species. The *Scolytodes* breeds only in fallen *Cecropia* leaves (Jordal 1998). *Araptus fossifrons* Wood, a Mesoamerican species previously collected from various seed pods and lianas (Wood & Bright 1992), was captured using CBB traps (Dufour 2002).

Experimental Procedure

The host specificity of *P. coffea* was evaluated in the laboratory in a non-choice test. Fifty specimens of each species were placed individually in 40×10 mm glass vials and immediately afterwards, a *P. coffea* female was introduced. The insects were observed for five hours under a 20W fluorescent lamp. We considered oviposition to have occurred when the female parasitoid adopted a characteristic ovipositing position on the elytra of the scolytids (López-Vaamonde & Moore 1998). Hosts attacked by *P. coffea* were transferred individually into vials containing CBB diet (Villacorta & Barrera 1996) and parasitization was assessed based on the emergence of the progeny, or by dissecting the hosts that did not yield parasitoids. We also recorded the time required for the encounter (defined as the time elapsed between the release of the parasitoid in the arena until it assumed the characteristic ovipositing position on the beetle), and the handling time (defined as the time elapsed between the encounter and the end of parasitization). Environmental conditions of the laboratory during the development of parasitoid's progeny were $26 \pm 2^\circ\text{C}$ and 70-80% RH.

Taxonomy

Samples of the species used were compared by Kirkendall with type material, or with specimens in his collection or in the S. L. Wood collection which had been compared with type material. Vouchers for all species are in Kirkendall's collection at University of Bergen, Norway.

RESULTS

Oviposition attempts by *P. coffea* were observed on all scolytid species tested, but parasitization and development of progeny was completed on only two species of bark beetles, in addition to *H. hampei* (Fig. 1). The percentage parasitism for *H. hampei*, *H. crudiae*, and *H. eruditus* was 64%, 14% and 6%, respectively (Table 1). We did not detect any oviposition by *P. coffea* in *S. borealis*, *H. plumeriae*, and *A. fossifrons*, nor did we find adult or immature stages of the parasitoid after hosts were dissected. Sex ratio of progeny produced by *P. coffea* in the three species was 1:1. The shortest encounter time was between *P. coffea* and *H. hampei* (mean = 23 min). Encounter time was 5-6 times longer when para-

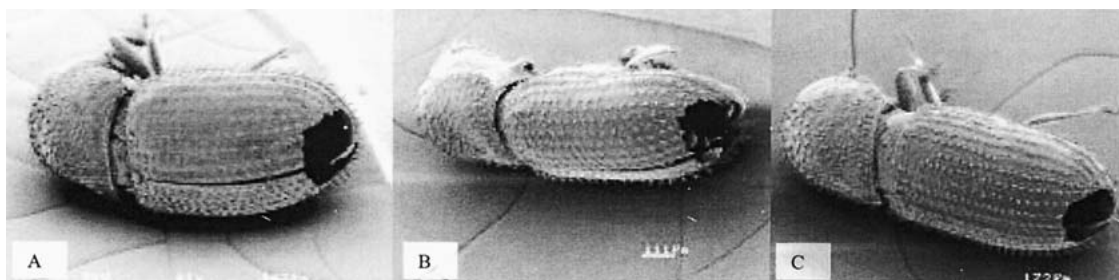


Fig. 1. Adults of three species of scolytids parasitized by *Phymastichus coffea* in the laboratory, showing the hole made by the emerging wasp adult: (A) *H. hampei* (B) *H. eruditus* and (C) *H. crudiae*.

sitization was on *H. crudiae* or *H. eruditus*. However, the handling time was shorter (1.5 min) in *H. eruditus* than in the other hosts. In most cases *P. coffea* allocated more than two eggs per host after a single attack. Development of immature stages of the parasitoid ranged from 39 to 42.6 days (Table 1).

DISCUSSION

Phymastichus coffea parasitized two species of *Hypothenemus* in addition to its natural host, *H. hampei*. Our findings confirm the oligophagic behavior of *P. coffea* reported by López-Vaamonde & Moore (1998). Five species of beetles are now known to serve as hosts for *P. coffea*: *H. obscurus*, *H. seriatus*, *Araptus* sp (Lopez-Vaamonde & Moore 1998), *H. crudiae* and *H. eruditus* (this study), thus indicating that this parasitoid is not specific to the CBB. Host specificity tests under laboratory conditions are the first step in assessing the potential host range of a given species (Orr et al. 2000; Babendreier et al. 2003a, 2003b). Although laboratory tests can overestimate the range of hosts under field conditions (Sands 1997), they are necessary to give an idea of the number of hosts at risk of being attacked in the field. Based on laboratory trials, specificity can be

demonstrated in the field later on (Barron et al. 2003; Babendreier et al. 2003b). Accordingly, parasitism by *P. coffea* on *H. crudiae* and *H. eruditus* in the laboratory does not necessarily mean that these beetles will be attacked by this parasitoid in the field. Careful observations under field conditions should be carried out before concluding that these beetles are alternative hosts of *P. coffea*.

There are no reports of parasitism by *P. coffea* on other hosts under field conditions. However, entomologists are not specifically collecting other bark beetles in or around coffee plantations, so parasitism of other hosts is unlikely to be recorded. Encounters between the parasitoid and scolytids in the field are likely, since the five species of polyphagous beetles parasitized by *P. coffea* in the laboratory are common in coffee agroecosystems and the disturbed habitats which often surround them (Atkinson & Equihua-Martínez 1985).

Our results could have important implications in the biological control of CBB in Latin America if further studies demonstrate that *P. coffea* attacks these scolytids in the field. First, with several host species available, there would be a risk of dilution of the parasitism exerted on CBB in the field. Second, one or more of these species might be more easily reared in large numbers in

TABLE 1. PARASITISM BY *PHYMASTICHUS COFFEA* ON SEVERAL BARK BEETLES SPECIES UNDER LABORATORY CONDITIONS. FIFTY SPECIMENS OF EACH SPECIES WERE EXPOSED TO FIFTY PARASITOID FEMALES INDIVIDUALLY.

Scolytidae species	Parasitism attempts (%)	Time required for the encounter with host (min \pm SE) ¹	Handling time (min \pm SE) ²	Parasitism (%)	Progeny production of <i>P. coffea</i>	Development of parasitoids (days)
<i>Hypothenemus hampei</i>	78	23.0 \pm 4.4	5.8 \pm 0.7	64	54	42.6
<i>Hypothenemus crudiae</i>	58	136.2 \pm 26.0	4.2 \pm 0.8	14	14	40.4
<i>Hypothenemus eruditus</i>	50	123.0 \pm 23.0	1.5 \pm 0.4	6	4	39
<i>Hypothenemus plumeriae</i>	36	0	—	0	0	0
<i>Scolytodes borealis</i>	8	0	—	0	0	0
<i>Araptus fossifrons</i>	46	0	—	0	0	0

¹Defined as the time elapsed between the release of the parasitoid in the arena and when it assumed the characteristic oviposition position on the beetle.

²Defined as the time elapsed between the encounter and the end of the parasitization.

the laboratory than the CBB, which has more stringent food requirements, for mass rearing of *P. coffea* for augmentative biological control programs against CBB. Third, these scolytids could be important as alternative hosts in the field, increasing survival of the parasitoid during the intercropping season, when the CBB population is at its lowest. Intercropping season in coffee interferes severely with the establishment of natural enemies of CBB (Barrera et al. 1990).

Finally, taking into consideration that parasitism of non-target hosts is usually much higher in the laboratory than in the field (Orr et al. 2000), it is possible that the levels of laboratory parasitism of *H. crudiae* and *H. eruditus* do not reflect a potential risk of this parasitoid for populations of non-target bark beetles in the field. Only careful field observations can answer this important question.

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REFERENCES CITED

- ATKINSON, T. H., AND A. EQUIHUA-MARTÍNEZ. 1985. Notes on the biology and distribution of Mexican and Central American Scolytidae (Coleoptera). II. Cryphalini and Cortlyli. Coleopterists Bull. 39: 355-363.
- BABENDREIER, D., S. KUSKE, AND F. BIGLER. 2003a. Non-target host acceptance and parasitism of *Trichogramma brassicae* Bezdenko (Hymenoptera: Trichogrammatidae) in the laboratory. Biol. Control 26: 128-138.
- BABENDREIER, D., S. KUSKE, AND F. BIGLER. 2003b. Parasitism on non-target butterflies by *Trichogramma brassicae* Bezdenko (Hymenoptera: Trichogrammatidae) under field cage and field conditions. Biol. Control 26: 139-145.
- BAKER, P. S. 1999. La broca del café en Colombia. Informe final del proyecto MIP para el café. DFID-CENICAFE-CABI Bioscience. 148 pp.
- BARRERA, J. F., D. MOORE, Y. J. ABRAHAM, S. T. MURPHY, AND C. PRIOR. 1990. Biological control of the coffee berry borer, *Hypothenemus hampei*, in Mexico and possibilities for further action. Brighton Crop Protection Conference-Pest and Diseases 4: 391-396.
- BARRON, M. C., N. D. BARLOW, AND S. D. WRATTEN. 2003. Non-target parasitism of the endemic New Zealand red admiral butterfly (*Bassaris gonerilla*) by the introduced biological control agent *Pteromalus puparum*. Biol. Control 27: 329-335.
- BOURBON-MARTÍNEZ, O. 1989. Bioecologie d'un ravageur des baies de cafeier, *Hypothenemus hampei* Ferr. (Coleoptera; Scolytidae) et de ses parasitoides au Togo. Ph. D. Dissertation. Université Paul Sabatier. Toulouse, France 185 pp.
- DUFOUR, B. P. 2002. Validación de la trampa BROCAP para el control de la broca del café. Boletín de Divulgación. IICA-PROMECAFE 93: 14-20. Guatemala.
- GAUTHIER, N., J. LASALLE, D. L. QUICKE, AND H. C. J. GODFRAY. 2000. Phylogeny of Eulophidae (Hymenoptera: Chalcidoidea), with a reclassification of Eulophinae and the recognition that Elasmidae are derived eulophids. Systematic Entomol. 25: 521-539.
- INFANTE, F., J. F. BARRERA, S. T. MURPHY, J. GÓMEZ, AND A. CASTILLO. 1992. Cría y cuarentena de *Phymastichus coffea* LaSalle (Hymenoptera: Eulophidae) un parasitoides de la broca del café introducido a Mexico. XV Simposio Latinoamericano de Cafecultura, Xalapa, Veracruz, 80 pp.
- INFANTE, F., S. T. MURPHY, J. F. BARRERA, J. GÓMEZ, W. DE LA ROSA, AND A. DAMON. 1994. Cría de *Phymastichus coffea* parasitoides de la broca del café, y algunas notas sobre su historia de vida. Southwestern Entomol. 19: 313-315.
- JORDAL, B. H. 1998. A review of *Scolytodes* Ferrari (Coleoptera: Scolytidae) associated with *Cecropia* (Cecropiaceae) in the northern Neotropics. J. Natural Hist. 32: 31-84.
- LASALLE, J. 1990. A new genus and species of Tetrastichinae (Hymenoptera: Eulophidae) parasitic on the coffee berry borer, *Hypothenemus hampei* (Ferrari) (Coleoptera: Scolytidae). Bull. Entomol. Res. 80: 7-10.
- LÓPEZ-VAAMONDE, C., AND D. MOORE. 1998. Developing methods for testing host specificity of *Phymastichus coffea* LaSalle (Hym.: Tetrastichinae), a potential biological control agent of *Hypothenemus hampei* (Ferrari) (Col.: Scolytidae) in Colombia. Biocontrol Sci.Tech. 8: 397-411.
- NOYES, J. F. 1998. Catalogue of the Chalcidoidea of the world. Electronic publication (CD-ROM). ETI, Amsterdam.
- ORR, D. B., C. GARCIA-SALAZAR, AND D. A. LANDIS. 2000. *Trichogramma* non-target impacts: a method for biological control risk assessment, pp. 111-125 In P. A. Follet and J. J. Duan [eds.]. Non-target Effects of Biological Control. Kluwer Academic Publishers, USA.
- SANDS, D. P. A. 1997. The "safety" of biological control agents: assessing their impact on beneficial and other non-target hosts. Mem. Museum of Victoria 56: 611-616.
- VILLACORTA, A., AND J. F. BARRERA. 1996. Techniques for mass rearing of the parasitoid *Cephalonomia stephanoderis* (Hymenoptera: Bethyilidae) on *Hypothenemus hampei* (Coleoptera: Scolytidae) using an artificial diet. Vedalia. 3: 45-48.
- WOOD, S. L. 1982. The Bark and Ambrosia Beetles of North and Central America (Coleoptera: Scolytidae), A Taxonomic Monograph. Great Basin Naturalist Memoirs 6: 1-1359.
- WOOD, S. L., AND D. E. BRIGHT. 1992. A Catalog of Scolytidae and Platypodidae (Coleoptera), Part 2: Taxonomic Index, Volume B. Great Basin Naturalist Memoirs 13: 833-1553.