

Original Article

Hyperparasitoids of *Xenostigmus bifasciatus* (Hymenoptera: Braconidae), parasitoid of *Cinara* spp. (Hemiptera: Aphididae), in *Pinus* plantations in Brazil

Hiperparasitoides de *Xenostigmus bifasciatus* (Hymenoptera: Braconidae), parasitoide de *Cinara* spp. (Hemiptera: Aphididae), em plantações de *Pinus* no Brasil

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Abstract

Hyperparasitoidism can reduce the biological control of *Cinara* spp. (Hemiptera: Aphididae) in *Pinus* (Gymnospermae: Pinaceae) plantations in Brazil. *Cinara atlantica* (Wilson, 1919) and *Cinara pinivora* (Wilson, 1919), introduced into Brazil in the 1990s, damage young *Pinus* plants. The primary parasitoid *Xenostigmus bifasciatus* (Ashmead, 1891) (Hymenoptera: Braconidae) was introduced and released between 2002 and 2004 in the states of Paraná, Santa Catarina, and São Paulo, Brazil. The dispersion of this biological control agent was extensive, with parasitoidism near 100%, stabilizing *Cinara* spp. populations for years, however, the population increase of these aphids in *Pinus taeda* (Linnaeus, 1753) plantations up to 2.5 years old between 2022 and 2025 suggests a reduction in the efficiency of biological control. Eight hundred and eighty-four mummies of *Cinara* spp. were collected from commercial plantations in Santa Catarina and reared in laboratory, from which 203 Hymenoptera specimens emerged, including 24 (11.82% of the total mummies collected) of *X. bifasciatus* and 179 (88.18%) of the hyperparasitoids *Asaphes californicus* Girault, 1917 (Hymenoptera: Chalcidoidea: incertae sedis) (55.17%); *Dendrocerus carpenteri* (Curtis, 1829) (Hymenoptera: Megaspilidae) (10.80%); *D. ramicornis* (Boheman, 1832) (Hymenoptera: Megaspilidae) (9.90%); and *Euneura sopolis* (Walker, 1844) (Hymenoptera: Pteromalidae) (12.31%). This is the first record of *D. ramicornis* and the four species associated with *Cinara* spp. in *P. taeda* in Brazil. The high incidence of hyperparasitoidism may be the main cause of the reduced efficiency of *Cinara* spp. biological control in *Pinus* plantations in southern Brazil by *X. bifasciatus*.

Keywords: *Asaphes californicus*, biological control, *Dendrocerus* spp., *Euneura sopolis*, hyperparasitoidism, *Pinus taeda*.

Resumo

O hiperparasitoidismo pode reduzir o controle biológico de *Cinara* spp. (Hemiptera: Aphididae) em plantações de *Pinus* (Gymnospermae: Pinaceae) no Brasil. *Cinara atlantica* (Wilson, 1919) e *Cinara pinivora* (Wilson, 1919), introduzidas no Brasil na década de 1990, causam danos a plantas jovens de *Pinus*. O principal parasitoide, *Xenostigmus bifasciatus* (Ashmead, 1891) (Hymenoptera: Braconidae), foi introduzido e liberado entre 2002 e 2004 nos estados do Paraná, Santa Catarina e São Paulo, Brasil. A dispersão desse agente de controle biológico foi extensa, com parasitoidismo próximo a 100%, estabilizando as populações de *Cinara* spp. por anos, entretanto, o aumento populacional desses pulgões em plantações de *Pinus taeda* (Linnaeus, 1753) com até 2,5 anos de idade entre 2022 e 2025 sugere uma redução na eficiência do controle biológico. Oitocentas e oitenta e quatro múmias de *Cinara* spp. foram coletadas em plantações comerciais em Santa Catarina e criadas em laboratório, das quais emergiram 203 espécimes de Hymenoptera, incluindo 24 (11,82% do total de múmias coletadas) de *X. bifasciatus* e 179 (88,18%) dos hiperparasitoides *Asaphes californicus* Girault, 1917 (Hymenoptera: Chalcidoidea: incertae sedis) (55,17%); *Dendrocerus carpenteri* (Curtis, 1829) (Hymenoptera: Megaspilidae) (10,80%); *D. ramicornis* (Boheman, 1832) (Hymenoptera: Megaspilidae) (9,90%); e *Euneura sopolis* (Walker, 1844) (Hymenoptera: Pteromalidae) (12,31%). Este é o primeiro registro de *D. ramicornis* e das quatro espécies associadas a *Cinara* spp. em *P. taeda* no Brasil. A alta incidência de hiperparasitoidismo pode ser a principal causa da reduzida eficiência do controle biológico de *Cinara* spp. em plantações de *Pinus* no sul do Brasil por *X. bifasciatus*.

Palavras-chave: *Asaphes californicus*, controle biológico, *Dendrocerus* spp., *Euneura sopolis*, hiperparasitoidismo, *Pinus taeda*.

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1. Introduction

The giant conifer aphids, *Cinara pinivora* (Wilson, 1919) and *Cinara atlantica* (Wilson, 1919) (Hemiptera: Aphididae), were first recorded in Brazil in 1996 and 1998, respectively, damaging *Pinus* spp. plants (Penteado et al., 2000, Lazzari et al., 2004; Oliveira et al., 2004, Oliveira et al., 2025). These aphids, native to the United States and Canada, are not considered pests in those countries (Lemes and Zanuncio, 2021).

The management of *C. atlantica* and *C. pinivora* in *Pinus* spp. plantations in Brazil was based on different strategies, mainly the introduction and release of the parasitoid *Xenostigmus bifasciatus* (Ashmead, 1891) (Hymenoptera: Braconidae). Between 2002 and 2004, from the southeastern United States in *Pinus* plantations in the states of Paraná, Santa Catarina, and São Paulo, Brazil (Reis Filho et al., 2004; Penteado, 2007; Cunha et al., 2016). One year after release, *X. bifasciatus* was detected in all areas of its introduction, with dispersal up to 80 km from the

release sites and parasitism rates of up to 100%, effectively reducing *Cinara* populations (Reis Filho et al., 2004).

Cinara spp. populations increased in *P. taeda* plantations up to 2.5 years old in the states of Santa Catarina and Paraná between 2022 and 2025, two decades after the release of *X. bifasciatus* (Personal communication: Klabin technical team).

The objective was to evaluate the parasitism rates of *C. atlantica* by *X. bifasciatus* and the occurrence of hyperparasitoids in *P. taeda* plantations in the state of Santa Catarina, Brazil.

2. Materials and Methods

The study was conducted in October and November 2025 in commercial *P. taeda* plantations in municipalities of the state of Santa Catarina, Brazil (Table 1).

Laboratory analyses and specimen rearing were carried out at the Forest Protection Laboratory of the Center

Table 1. Total emergence (TE) of the parasitoid *Xenostigmus bifasciatus* (Hymenoptera: Braconidae) (*Xb*) and hyperparasitoids *Asaphes californicus* (Hymenoptera: Chalcidoidea: incertae sedis) (*Ac*), *Dendrocercus carpenteri* (Hymenoptera: Megaspilidae) (*Dc*), *Dendrocercus ramicornis* (Hymenoptera: Megaspilidae) (*Dr*) and *Euneura sopolis* (*Es*) (Hymenoptera: Pteromalidae) from mummies of *Cinara* spp. (*Cspp*) collected in *Pinus taeda* plantations in different municipalities of the state of Santa Catarina, Brazil. Oct-Nov, 2025.

Municipalities	Age of plants (Months)	Coordinates UTM		Cspp	TE	Xb	Ac	Dc	Dr	Es
		Easting (m)	Northing (m)							
Alfredo Wagner (A1) ^a	8-12	665880.40	6945824.80	12	6	3	0	1	0	2
Canoinhas (A1)	24-30	537959.25	7069438.35	25	4	0	4	0	0	0
Canoinhas (A2) ^b	24-30	538045.49	7069680.54	34	2	0	0	2	0	0
Capão Alto (A1)	24-30	529573.27	6888891.51	18	1	0	1	0	0	0
Capão Alto (A2)	24-30	552520.71	6915757.90	22	3	0	1	0	0	2
Correia Pinto (A1)	12-18	557627.32	6935415.46	74	21	0	13	2	3	3
Correia Pinto (A2)	24-30	499260.66	6988171.52	18	4	0	4	0	0	0
Otacílio Costa (A1)	24-30	592879.05	6969407.85	32	4	1	3	0	0	0
Otacílio Costa (A2)	24-30	595272.83	6969610.30	62	7	0	1	2	4	0
Painel (A1)	24-30	585219.34	6916328.21	121	31	2	19	7	0	3
Painel (A2)	24-30	595710.99	6891250.71	22	1	0	1	0	0	0
Painel (A3) ^c	24-30	582866.83	6907488.54	154	50	4	34	5	7	0
Ponte Alta (A1)	24-30	576019.43	6960527.03	50	8	0	5	0	0	4
Ponte Alta (A2)	24-30	575513.33	6961310.80	84	12	0	9	0	0	2
Rio do Campo (A1)	24-30	589120.09	7012838.86	17	1	0	0	1	0	0
Rio do Campo (A2)	12-18	589408.02	7013181.72	16	1	0	1	0	0	0
São Joaquim (A1)	24-30	594435.71	6869224.67	22	18	5	6	1	3	3
Santa Cecília (A1)	24-30	553314.85	7040945.06	57	3	0	3	0	0	0
Urubici (A1)	24-30	618832.57	6892454.25	44	26	9	7	1	3	6
Total				884	203	24	112	22	20	25
Emergency Percentage (%)						11.82	55.17	10.80	9.90	12.31

Collection area 1^a, 2^b and 3^c.

for Agroveterinary Sciences (CAV), Santa Catarina State University (UDESC), in Lages, Santa Catarina, Brazil.

Eight hundred and eighty-four mummies of the giant conifer aphids, *Cinara* spp., were collected. Sampling was distributed at a ratio of one point per 300 hectares, covering plantations aged between eight and thirty months. Twenty plants were evaluated at each sampling point, with the complete removal of all mummies found. This age range was selected because it represents the period of greatest vulnerability of *Pinus* spp. plants to attack by *Cinara* spp. (Fox and Griffith, 1977; Penteado et al., 2000).

Needles bearing these mummies were placed in plastic containers covered with mesh/voil, kept in a climate-controlled chamber (BOD) at a constant temperature of $25 \pm 1^\circ\text{C}$, and checked daily for parasitoid emergence.

Live aphid specimens were preserved in 70% ethanol and identified (Blackman and Eastop, 1994).

Parasitoids, immediately after emergence, were preserved in 70% ethanol, mounted, and identified as *Asaphes californicus* Girault, 1917 (Hymenoptera: Chalcidoidea: incertae sedis) (Gibson and Vikberg, 1998; Li et al., 2024); *Dendrocerus carpenteri* (Curtis, 1829) and *D. ramicornis* (Boheman, 1832) (Hymenoptera: Megaspilidae) (Dessart, 1999; Pezzini et al., 2014); and *Euneura sopolis* (Walker, 1844) (Hymenoptera: Pteromalidae) (Ko et al., 2018; Lee et al., 2019).

Voucher specimens of aphids and parasitoids are deposited, respectively, in the Entomological Collection of Santa Catarina State University (Lages, SC) and in the

Entomological Collection of the Federal University of Espírito Santo (Vitória, ES).

3. Results

Two hundred and three Hymenoptera specimens emerged from aphid mummies (Table 1).

Twenty-four (11.82%) of the specimens emerged from the mummies were *X. bifasciatus*, a primary and solitary parasitoid of *Cinara* spp. The individuals emerged from the remaining 179 (88.18%) mummies belonged to four species of aphid Hymenoptera hyperparasitoids, namely 112 (55.17%) specimens of *A. californicus*; 22 (10.80%) of *D. carpenteri*; 20 (9.90%) *D. ramicornis*; and 25 (12.31%) of *E. sopolis* (Table 1).

4. Discussion

The maintenance temperature of *Cinara* spp. mummies ($25 \pm 1^\circ\text{C}$) may have affected the emergence of the different species because the development of *Asaphes* spp. and *Dendrocerus* spp. is better under milder thermal conditions (De Boer et al., 2019). Besides, thermal stress reduces the biological fitness of *Dendrocerus* spp., directly affecting fecundity (Buitenhuis et al., 2017; Mitsunaga et al., 2014), with reports of a 50% reduction in the progeny of *D. carpenteri* from different hosts between 19°C and 20°C (Walker and Cameron, 1981).



Figure 1. *Asaphes californicus*, female habitus.

Asaphes californicus (Figure 1) is a generalist hyperparasitoid of primary aphid parasitoids, such as Aphidiinae (Braconidae) and Aphelinidae species. The distribution of this species extends along the west coast of North America, South America, and China. Its females oviposit on the bodies of late-instar larvae, prepupae or pupae of primary aphid parasitoids and develop as ectophagous and solitary pseudohyperparasitoids (Gibson and Vikberg, 1998).

The sampled specimens treated as *A. californicus* sensu (Gibson and Vikberg, 1998) is because its diagnostic features are similar to those of *A. vulgaris* (Walker, 1834) and *A. suspensus* (Nees, 1834) (Hymenoptera: Chalcidoidea: incertae sedis), such as yellow hind trochanters and darkened tibiae in females, and the outer surface part of the antennal scape of males bears a subbasal, ovoid to elongate-lanceolate, microsetose sensory region (Gibson and Vikberg, 1998). Furthermore, all *Asaphes* specimens collected in South America (Argentina, Bolivia, Chile, Colombia, Ecuador, Uruguay, and Venezuela) are *A. californicus*; therefore, previous records of *A. vulgaris* for South America refer to the former species (Gibson and Vikberg, 1998). Records of *A. vulgaris* in Brazil (De Santis, 1960) should also be treated as *A. californicus*, indicating that this species was already present in Brazil before 1968.

The genus *Dendrocerus* is cosmopolitan, comprising about 120 species, some of which are ectophagous aphid hyperparasitoids, including *D. carpenteri*, (Figure 2 a, c, d)

recorded on all continents except Antarctica (Fergusson, 1980). This species has been recorded in Brazil and Chile (Dessart, 1975; Tavares, 1996) as a pseudohyperparasitoid of aphids (it lays eggs on the primary host after the aphid has been consumed) and has been associated with many aphid species and their primary and secondary parasitoids, including *Asaphes* and *Pachymeuron* (Hymenoptera: Pteromalidae) (Fergusson, 1980; Masner, 2006). The distribution of *D. ramicornis* (Figure 2 b, d and f) is Holarctic and associated with Lachninae aphids (Hemiptera: Aphididae), mainly species of *Cinara* and *Lachnus* (Fergusson, 1980). *Dendrocerus carpenteri* and *D. ramicornis* differ in the shape of antennae (mainly in males) and of the notaulices (both sexes). Males of *D. carpenteri* present the five basal flagellomeres triangular in profile (Figure 2 c), as in a serrate antennae. In *D. ramicornis*, males present each of the five basal flagellomeres with a long process dorsally (Figure 2 d) and all process of similar length. The notauli of the former species are distinctly convergent posteriorly and meet each other at the mesoscutal margin (Figure 2 e), but in *D. ramicornis* notauli are slightly convergent and reach the mesoscutal margin far from it other (Figure 2 f).

Euneura sopolis (Figure 3), with records from Europe, Korea, and Japan, appears to have an original Palearctic distribution (Lee et al., 2019). This parasitoid has been recorded in southeastern Brazil (Cunha et al., 2016), associated with species of *Pauesia* Quillis, 1931 (Hymenoptera: Aphidiidae) and *Xenostigmus* (Garrido Torres and Nieves-Aldrey, 1999), which are

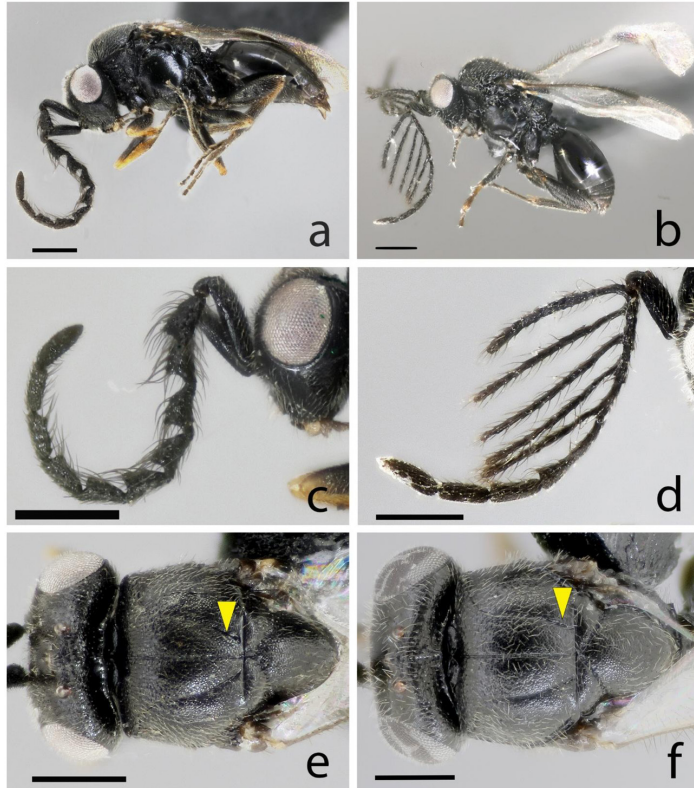


Figure 2. *Dendrocerus capenteri* (a, c, d) and *D. ramicornis* (b, d, f): habitus (a, b), antennae (c, d) and mesoscutum (e, f). The arrows indicate the notaulus.

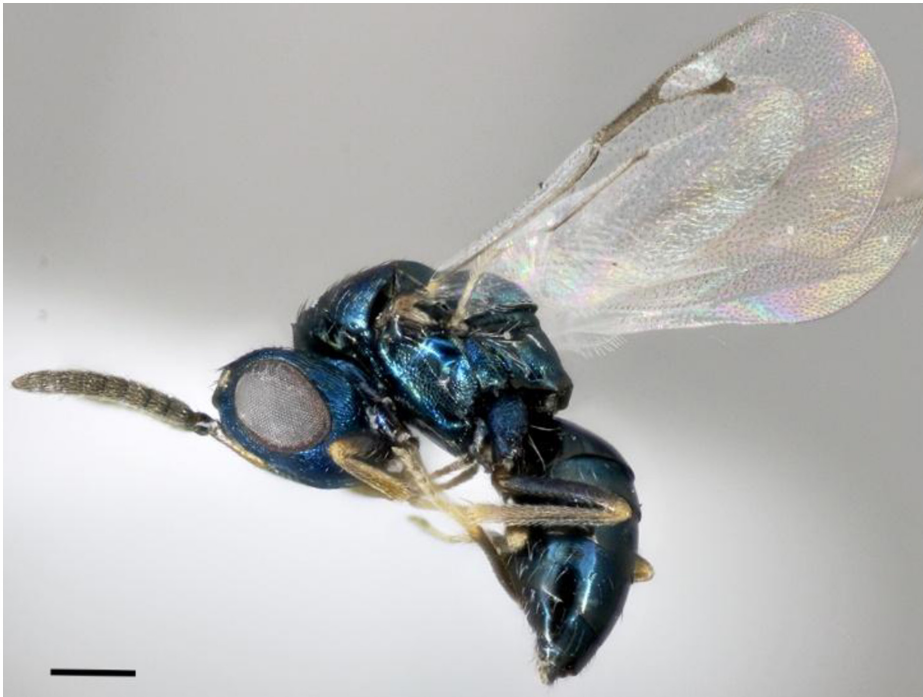


Figure 3. *Euneura sopolis*, female habitus.

primary parasitoids of aphids of the family Lachnidae (Cunha et al., 2016).

Several species of primary and secondary parasitoids of *C. atlantica* emerged during the quarantine process for the introduction of *X. bifasciatus* into Brazil, in 2002. The aphid mummies were shipped from the United States and the parasitoid emerged were: *X. bifasciatus* and *Diaeretus* sp. (Foerster, 1862) (Hymenoptera: Aphididae); *Alloxysta lachni* (Ashmead, 1885) (Hymenoptera: Figitidae), *Anastatus* sp. Motshulsky (Hymenoptera: Eupelmidae); *Asaphes suspensus* (Hymenoptera: Chalcidoidea: incertae sedis); *Dendrocercus* sp. (Megaspilidae); *Euneura lachni* and *E. sopolis* (Pteromalidae); *Syrphophagus* sp. Ashmead (Hymenoptera: Encyrtidae); and *Tetrastichus* sp. Haliday, 1844 (Hymenoptera: Eulophidae). Specimens of contaminant species were destroyed during quarantine, and only *X. bifasciatus* was released, between 2002 and 2004, for the control of *Cinara* spp. in the states of São Paulo, Paraná, and Santa Catarina (Penteado et al., 2004).

Among the hyperparasitoid species obtained here, *A. californicus* and *D. carpenteri* were present in Brazil before the introduction of *X. bifasciatus*; *E. sopolis* was reported in the country more than 10 years after this introduction. *D. ramicornis* is here recorded for the first time. This suggests that these latter two hyperparasitoid species, like *Cinara* spp., were introduced accidentally. On the other hand, these hyperparasitoids may have been introduced by natural dispersal, since their aphid hosts have been recorded in countries neighbouring Brazil, such as Argentina and Uruguay (Penteado et al., 2000), and these aphids and their hyperparasitoids are native to or have long been present in North America (Gibson and Vikberg, 1998).

The low emergence rate of *X. bifasciatus*, the main biological control agent of *Cinara* spp., and the high emergence rate of hyperparasitoids apparently limit the effectiveness of classical biological control of these aphids in *Pinus* plantations in Brazil. Surveys on the occurrence of these hyperparasitoids should be conducted in the states of Paraná and São Paulo, as well as in neighboring countries, since the resurgence of giant conifer aphids as major pests in *Pinus* plantations in the coming years is likely.

5. Conclusion

This is the first report of the hyperparasitoids *A. californicus*, *D. carpenteri*, *D. ramicornis*, and *E. sopolis* associated to *Cinara* species in *P. taeda* plantations in Brazil and of *D. ramicornis* in South America.

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Data Availability Statement

The entire data set that supports the results of this study was published in the article itself.

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