

# Distribution of *Caenocholax fenyesei* (Strepsiptera: Myrmecolacidae) and the Habitats Most Likely To Contain Its Stylopized Host, *Solenopsis invicta* (Hymenoptera: Formicidae)

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**ABSTRACT** *Caenocholax fenyesei* Pierce has been collected sporadically throughout the Neotropics and southeastern United States. We present the known distribution of *C. fenyesei*, including the addition of 5 new distributional records. The distribution of *C. fenyesei* was evaluated in Brazos County, Texas, as an example of a concise geographic region saturated with Myrmecolacidae. *Caenocholax fenyesei* Pierce is known from numerous locality reports, where adult males were collected in traps. The only exception to this was with the discovery that a host of male *C. fenyesei* in Texas is *Solenopsis invicta* Buren (Kathirithamby and Johnston 1992). *Caenocholax fenyesei* was collected from 7 southern states in the United States. The first report of *C. fenyesei* in the United States was from Florida (Frost 1962, 1963), collected in a study at Archbold Biological Station, Highlands County. Meadows (1967) collected *C. fenyesei* in light traps from 15 additional counties in Florida. One additional Florida county record was added during a study conducted from 1986 to 1992 (Kathirithamby and Peck 1994). Khalaf (1968) collected *C. fenyesei* in Louisiana from light traps in the vicinity of New Orleans and other areas of southern Louisiana and later from central Louisiana and 2 locations in Hancock County, Mississippi (Khalaf 1969). Johnson and Morrison (1979) reported *C. fenyesei* from Georgia and Arizona. A single male, collected in 1960 from Madera Canyon in the Santa Rita Mountains, AZ, in the University of Arizona collection is actually the earliest known collection of *C. fenyesei* in the United States, but the specimen was not identified and reported until several of the above reports were made. Jones et al. (1980) collected *C. fenyesei* in Baldwin County, Alabama, at light traps. Kathirithamby and Johnston (1992) reported male *C. fenyesei* with its host, *S. invicta*, from Brazos County, Texas. Although most reports are currently from the United States, *C. fenyesei* appears to have a wide-

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spread Neotropical distribution including Cordoba, Mexico (Pierce 1909); Tabasco, Mexico (Kifune 1979); Peten, Guatemala (Kifune 1979); Matagalpa, Nicaragua (Maes and Kathirithamby 1993); Costa Rica (Kathirithamby 1992); Panama (Bohart 1941); Ecuador and Chile (Kathirithamby 1992); Missions, Argentina (Bohart 1941); Andros Island, Bahamas (Kathirithamby and Peck 1994); and Cuba (Genaro and Peck 1995).

These collections begin to establish a large geographic range for *C. fenyesei*. Available collection data establishes a distribution that appears disjunct, but may be a result of sporadic collections. An examination of the distribution of a strepsipteran species within a concise geographic region has never been attempted. A study of this type can lead to better understanding of the distributional pattern and possible separation of populations within the species. The objective of this study was to provide a better understanding and prediction of the range of *C. fenyesei* using information obtained throughout this study, along with additional collection records. This information could be useful in determining the potential of using *C. fenyesei* as a biological control agent of *S. invicta*. We determine if stylopized *S. invicta* have a continuous or disjunct distribution within an area where hosts are not a limiting factor, and also determined the prevalence of stylopized *S. invicta* in this area.

**Materials and Methods**

**Distribution.** We examined the distribution of stylopized *S. invicta* colonies within Brazos County, Texas, from February 1994 to October 1995. We

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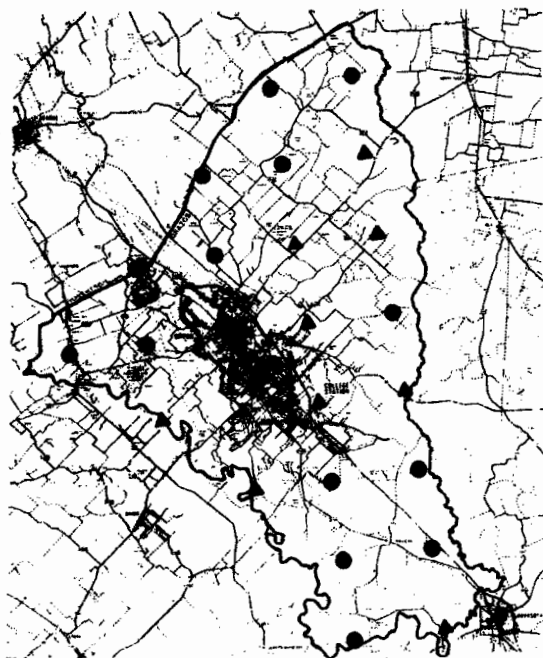


Fig. 1. Distribution of *S. invicta* colonies stylopedized by *C. fenyesei*. ▲, transect sites contained stylopedized *S. invicta*; ●, transect sites had no *C. fenyesei*.

chose this concise area because of its abundance of *S. invicta* and because *C. fenyesei* occurs in this region.

The county was divided into 25 equally spaced collection sites, each  $\approx 8$  km apart (Fig. 1). Collections were taken as close to the sites designated by the grid as possible. Three colonies from each site were taken to the laboratory for observation and random sampling of ants. We collected each grid site twice, at different times of the year, to give a total of 6 colonies evaluated from each location.

Colonies were collected by shoveling ant mounds into 18.9-liter (5-gal) buckets that were coated on the inside surface with talcum powder to prevent ants from escaping. Colonies were taken to the laboratory and water was slowly dripped into the bucket until the submersion of the soil caused ants to raft together at the water surface. From this raft of ants we took a random sample of at  $\geq 200$  ants and preserved them in 80% ethanol. Random sampling was conducted by removing a portion of rafting ants with forceps and submerging it into a vial of ethanol. Remaining ants were placed into a plastic petri dish (150 by 15 mm) that had  $\approx 5$  mm of Labstone (Miles Dental Products, Rockville Center, NY) in the base. Two holes (5 cm diameter) in the upper portion of the dish allowed ants to go in and out of the dish. Dishes were kept in plastic shoe boxes (37 by 27 by 9 cm) that had the inside walls coated with Fluon AD-1 (Northern Products, Woonsocket, RI). An dowel (eight cm long), kept upright by insertion into a rubber stopper, was added to the shoe box to collect possible stylopedized ants. Ants that are

stylopedized tend to climb to a high perch and remain in a posture resembling gaster flagging before emergence of the strepsipteran (unpublished data). Colonies were provided with water, mealworms, and 5% honey water daily.

Ants suspected of being stylopedized with male strepsipterans were isolated in petri dishes (35 by 10 mm) for strepsipteran emergence. Any emergence from the colony or from isolated ants was recorded. If no emergence occurred from isolated ants within 48 h, they were dissected or cleared using 10% KOH to check for the triungulin or its exuvium, which is not eliminated by the ant host. The presence of a triungulin or its exuvium in a cleared ant was used to indicate stylopedization.

The sample of ants previously placed in EtOH was partially cleared in 10% KOH for 7 d at 22 C° to check for parasitization. A record was kept of all stylopedized ants, and results of the grid-site analysis were plotted on a Brazos County map. Sites that contained colonies with stylopedized ants were evaluated for significant differences in stylopedization percentages using a 1-way ANOVA (evaluated at  $\alpha = 0.05$ ) using Minitab Release 10 (Minitab, Inc.)

**Habitat.** To discover the habitats most likely to contain colonies with stylopedized ants, we chose an area at the edge of Raintree Subdivision, at the southeast edge of College Station, TX. This area contains 3 distinctive habitat types, each covering  $\approx 300$  m in a straight line from one side to the other. The 1st habitat type was post oak-savanna and consisted of grassy areas along with stands of trees, primarily post oak, *Quercus stellata* Wang. The 2nd habitat type was dense woodland with little or no grass. A trail was the only disturbed area in the woodland habitat region. The 3rd habitat type was grassland, consisting of open pasture with no trees or brush. Two equally spaced sampling sites were established in each of the habitats. We collected 1 colony at each transect site and replicated the experiment 3 times, using other colonies from the same area, over the course of several months. Collections were made in July 1994, March 1995, and July 1995. Colonies were handled as previously described. Results were analyzed using a chi-square test Minitab Release 10 (evaluated at  $\alpha = 0.05$ ) to determine if the habitats contained significantly different percentages of stylopedized ant colonies.

To test further for significant differences between the 3 habitats listed above, additional site collection information for part of a larger study (Cook 1996) was used to produce a larger data set. Each of the 244 collection's habitat type and level of stylopedization was recorded. The percentage of stylopedized colonies was compared using a chi-square test to determine if there were significant differences between the habitats. A 1-way analysis of variance (ANOVA) was used to determine if there were significant differences in the percentage of stylopedized ants within colonies from the different habitats.

Representative voucher specimens were deposited in the Texas A&M University Insect Collection

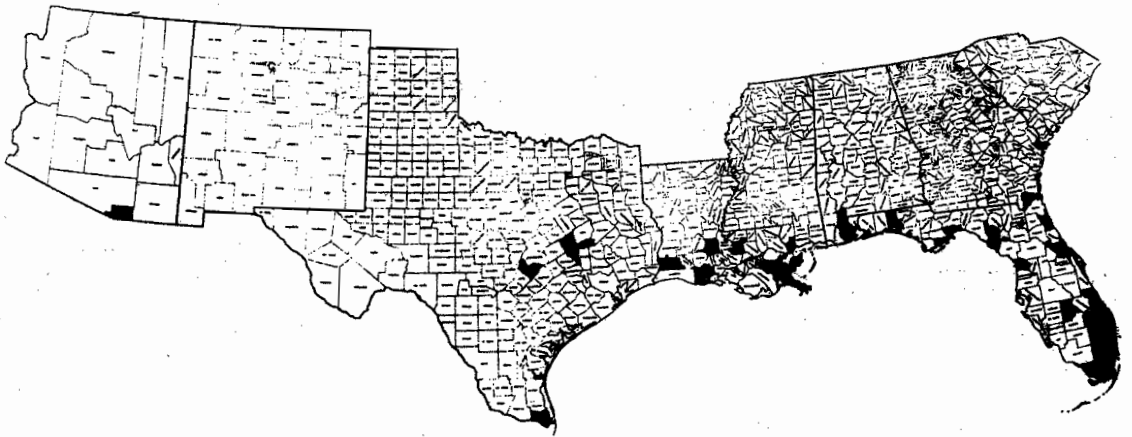


Fig. 2. Distribution of *C. fenyesei* in the southern United States. Shaded areas are counties where collections of *C. fenyesei* have been made.

(TAMUIC), Texas A&M University, College Station, TX (accession no. 619). The remaining specimens from this study are in the collection of J.L.C.

### Results

**Geographic Distribution of *C. fenyesei*.** The current known distribution of *C. fenyesei* in the United States is shown in Fig. 2. The current known world distribution is shown in Fig. 3. We collected *C. fenyesei* from 3 additional Texas counties and report on 1 other Texas county where *C. fenyesei* was col-

lected and reported to us. Additionally, we add a new record from Mexico.

We collected *C. fenyesei* from stylopized ants with a colony brought back to the laboratory from Cook's Point (Robertson County), TX, on 10 October 1993. Will Godwin collected a male *C. fenyesei* at lights from Democrat Crossing (Madison County), TX, on 3 September 1995. We identified this specimen which is now deposited in TAMUIC. A 3rd Texas county now included in the distribution of *C. fenyesei* is Travis County, from specimens found by Ed Vargo in dissections. Ed Riley, Mike Quinn, and H. Blackman collected a male *C. fenyesei* at UV light on 16 October 1993 from Cameron County, Texas. This specimen is now in TAMUIC. James Woolley collected a single male strepsipteran in a Malaise trap (27–30 July 1993) at Gomez Farias, Tamaulipas, Mexico. The specimen was found in the TAMUIC where we later identified it as *C. fenyesei*. The reported host ant of *C. fenyesei*, *S. invicta*, is not found in this area. However, a closely related fire ant, *Solenopsis geminata* (F.), is common.

**Distribution of *Caenocholax fenyesei* in Brazos County, Texas.** Ten of the 25 Brazos County sites contained stylopized *S. invicta*. However, not all colonies from these 10 transect sites contained stylopized ants.

Twenty-eight of the 60 colonies collected from the 10 sites contained ants stylopized by *C. fenyesei*. There was no site from which all 6 of the sampled colonies contained stylopized ants. Frequency of stylopized colonies, of 6 colonies collected at each site, are as follows: 5 stylopized colonies at 1 site; 4 stylopized colonies at 2 sites; 3 stylopized colonies at 3 sites; 2 stylopized colonies at 3 sites; 1 stylopized colony at 1 site; and no stylopized ants at 15 sites. The mean stylopization rate of all colonies was 0.4 percent (range, 0–2.5%). The mean stylopization rate of infected colonies was 0.9 percent (range, 0.5–2.5%). There was no significant difference be-



Fig. 3. World distribution of *C. fenyesei*. ▲, recorded collections.

Table 1. Comparison of pooled data for all colonies collected in Brazos County, Texas, 1993-1996

Habitat type	No. colonies		% Infected when styloplized
	Styloplized	Not styloplized	
Dense woodland	35	48	1.2
Post oak-savanna	31	57	1.1
Grassland	9	64	0.8

tween percentages of styloplization in the sites that contained *C. fenyesei* ( $F = 0.39$ ; d.f. = 8, 45;  $P = 0.919$ ).

The distribution pattern of styloplized transect sites appears to be in a zone across the center of the county, except for 1 site at the county's southern edge (Fig. 1). No common factor is known among these sites containing styloplized ants.

**Habitat of Styloplized *S. invicta*.** Five of 7 colonies collected in the woodland habitat contained styloplized ants. One woodland habitat site was collected on only the 1st of the 3 collection dates. On other dates, this site did not have detectable fire ant colonies for collecting. Four of 6 savanna habitat sites and 2 of 6 grassland habitat sites (sites 2 and 3) contained colonies with styloplized ants. Thus, styloplized ants were in all habitat types, but were more common in woodland and savanna habitats.

The mean percentage of styloplized ants in affected colonies was 1.2% for woodlands, 1.0% for savanna, and 0.5% for grasslands. Ants in the grassland habitat had the lowest level of styloplization (0.5%), which was found in each of 2 colonies that contained styloplized ants. Styloplization levels in the savanna and woodland habitats ranged from 0.5 to 3.3% and 0.5 to 2.0% of styloplized colonies, respectively. The percentage of styloplized ants found in colonies were not significantly different between habitats in this experiment ( $F = 1.61$ ; d.f. = 2, 16;  $P = 0.231$ ). There was also no significant difference between collection dates ( $F = 1.41$ ;  $P = 0.274$ ).

Additional habitat information from another study (Cook 1996) is given in Table 1. These collection sites also were from Brazos County but were not randomly chosen as in the previous study. However, the records of all styloplized colonies provide a larger number of colonies for comparison of habitats and percentages of styloplized ants present. It was not possible to obtain this number of colonies from the habitat experiment site alone. Pooled information, from all sources, is shown in Table 1. Fewer colonies collected from the grassland habitat are styloplized, 12.3% (9 of 73), than those found in woodland and savanna habitats, 42.2% (35 of 83) and 35.2% (31 of 88), respectively. These results indicate that the number of styloplized colonies in grassland habitats is significantly different from the other habitats ( $\chi^2 = 17.548$ , d.f. = 2,  $P = 0.000$ ). The styloplization level of each styloplized colony was 1.2% in woodland habitats, 1.1% in savanna, and 0.8% in grasslands.

## Discussion

The distribution of *C. fenyesei* appears to be Neotropical, with the southern United States forming its northern limit. More reports have been made from the southern United States than from Neotropical areas, but this phenomenon is likely a result of more intensive collecting in this part of the world. With more collections from the Neotropics, the range of *C. fenyesei* will likely become more continuous. Strepsipterans also often occur in small numbers. Small numbers, at any given time, may be a result of very short adult male life spans and may not necessarily reflect a scarcity of styloplized hosts. Having very small males and parasitic adult females also cause them to be overlooked. However, the disjunct distribution found in Brazos County shows that the distribution may not be continuous throughout its range. The habitat preference of hosts of *C. fenyesei* may play an important role in determining the complete distribution pattern.

The geographic distribution of *C. fenyesei* does not coincide with the distribution of its only known host, *S. invicta*. The distribution of *S. invicta* and *C. fenyesei* overlap in the United States, except for a collection from southern Arizona. *S. invicta* also does not occur in the regions of Mexico, Central America, and parts of South America where *C. fenyesei* occurs. Thus, *C. fenyesei* must have a different host in these regions because *C. fenyesei* is a relatively weak flier and has a very short adult life (Cook 1996). This conclusion is noteworthy because most myrmecolacid strepsipterans are currently known from only 1 host, although most species have an unknown host association. It remains to be proven whether *C. fenyesei* has always had >1 male host, or if there has been a host switch in the United States. We feel that the most likely scenario is that a host switch occurred in the United States after the introduction of *S. invicta* (also noted by Kathirithamby and Hamilton 1995).

Several factors support the hypothesis of a recent host switch in the United States. First, for *C. fenyesei* to have been introduced with *S. invicta*, its females and males would have to have been introduced at the same time for them to establish and spread with *S. invicta*. This problem is further complicated by the dual hosts of *C. fenyesei* and the rapid spread of *S. invicta*. Given the dual host relationship of *C. fenyesei*, it is unlikely that the strepsipteran parasite of *S. invicta* spread in all directions away from the point of introduction in such a short time. Finally, the western boundary of the range of *C. fenyesei* in North America is in an area of the United States (Arizona) where *S. invicta* is not known to occur.

*Solenopsis geminata* and *S. xyloni* McCook are likely candidates for the previous host of *C. fenyesei* in the United States because they have a biology that is relatively similar to *S. invicta* and are found within the known range of *C. fenyesei*. However, *C. fenyesei* has yet to be associated with *S. geminata* or *S. xyloni*. Therefore, we propose that a likely scenario is that

a host switch of male *C. fenyesei* has occurred from a closely related native fire ant.

An alternative hypothesis to the above argument is that *C. fenyesei* has a wider host range than is supposed. If this 2nd hypothesis is true, it is plausible that male *C. fenyesei* might be, or is capable of, styloping other species of fire ants, including several *Solenopsis* species found in South America.

Strepsipteran habitats are largely a function of their host habitat preference. Females of the family Myrmecolacidae are parasitic throughout their development and adult lives. Male myrmecolacids are parasitic throughout their development, but emerge for a very short free-living adult life. The adult life span is very short, ranging from a few hours to a few days (Kathirithamby 1989). The only other free-living stage is the triungulin, which leaves the female via a brood canal to the outside of its host. The triungulin is the stage that searches for its new host. Because of this relationship, strepsipterans have a distribution coinciding with that of their host. There is insufficient time for the adult male to establish and inhabit its own unique habitat, and a triungulin that is not in its future host's habitat will not be successful in finding a host and will perish. The heteronomy of *C. fenyesei* and other myrmecolacids suggests that habitats with which they are associated should be an intersection of its dual host's habitats.

We found no apparent distribution pattern within Brazos County. Fire ant colonies are abundant throughout the county, but the distribution of the host of the female, *Hapithus agitator* Uhler (Cook 1996), is unknown. The spotty distribution of *C. fenyesei* found in Brazos County is likely a reflection of female host distribution but could be the result of a recent host switch as proposed above. *C. fenyesei* might still be expanding through its new host population.

*Solenopsis invicta* was found in all habitats surveyed in this study. The large *S. invicta* populations found at these collection sites should not be a limiting factor for levels of *C. fenyesei*. Styloping *S. invicta* appear to be less common in grassland habitats. A chi-square test on pooled habitat data statistically supports this view. Significant differences between the grassland habitat and other habitats could be a result of the female host habitat or a different, unforeseen, limiting factor. This study shows that styloping *S. invicta* are not excluded from any of these habitat types.

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