

THE BIOLOGY AND ECOLOGY OF *BELONOCNEMA TREATAE*
(HYMENOPTERA:CYNIPIDAE) ON ITS HOST PLANT, *QUERCUS FUSIFORMIS*

THESIS

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DEDICATION

In memory of Jeff Lund

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ABSTRACT

THE BIOLOGY AND ECOLOGY OF *BELONOCNEMA TREATAE*
(HYMENOPTERA: CYNIPIDAE) ON ITS HOST PLANT, *QUERCUS FUSIFORMIS*

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Through experimental rearing, heterogony (the alternation of unisexual and bisexual generations) was demonstrated in the cynipid species *Belonocnema treatae* Mayr 1881. Female *B. treatae* (bisexual generation) induce galls on the leaves of *Quercus fusiformis* Small (Fagaceae), from which unisexual females, previously described as *B. kinseyi* Weld 1921, emerge. Unisexual females induce galls on the roots of *Q. fusiformis*. The name *B. treatae* has priority, so *B. kinseyi* becomes a new synonym. Adults of each generation and the galls they induce are described. Timing events in the life cycle of *B. treatae* on its host plant are described for populations in central Texas. Hymenopteran inquiline and parasitoids of *B. treatae* are identified, and aspects of the impact of natural enemies on *B. treatae* success are described.

Introduction

A gall is a structure of modified plant growth that most probably represents an attempt by the plant to isolate the gall inducing agent. Galls may be induced by a variety of organisms including viruses, bacteria, fungi, nematodes, mites and insects. All organisms capable of gall-induction belong to the same guild based upon trophic similarities. The order Hymenoptera contains species having the most complex galls of the entire galling guild, based on the diversity in gall size, shape, and structure. Within this order, the family Cynipidae includes 2,000 species with 805 occurring in North America. Cynipid galls are induced by oviposition and subsequent larval feeding, providing the larvae with food, shelter, and a degree of protection.

The cynipids that gall oaks exhibit a complex reproductive cycle known as heterogony, wherein a bisexual generation alternates with a unisexual generation. This cycle involves two types of females that differ in mode of reproduction, morphology, and in the structure of galls they initiate. In Chapter 1, heterogony is demonstrated in Belonocnema treatae, including detailed descriptions of the unisexual and bisexual generations and the galls they induce on Quercus fusiformis.

The study of cynipid galls is one of the most interdisciplinary areas of natural science, involving interactions between plant tissues, the cynipid wasp gall-maker, and associated parasites. The host plant is central to all gall communities. It serves not only as the energy source for all gall inhabitants, but as the site to search for mates, to mate, select oviposition sites, and then overwinter. Each cynipid larva is confined for an extended period within plant tissue making it vulnerable to parasites. A gall may be inhabited by inquiline species (which consume the gall of the gall maker), parasitoid species (which feed on the larvae of the gall maker), hyperparasitoid species (feeding on parasitoid larvae), and/or the gall-

maker larva which induced its formation. These assemblages of insects form microcommunities and have been the subject of many studies regarding tri-trophic interactions. Chapter 2 elucidates details of the life cycle and ecology of B. treatae. The Hymenoptera commonly associated with B. treatae galls are identified, and relative abundance of community members and factors affecting B. treatae success are assessed. In addition, the Appendix provides a dichotomous key as well as family and species descriptions for the identification of associated Hymenoptera.

Many features make gall-inducing cynipids ideal for answering questions about the ecology of population dynamics, resource quality, community structure and development, and three trophic level interactions. Galls are easily located so that gall-makers, parasitoids and inquilines can be readily sampled. Gall wasps remain at one location on the plant throughout much of their development, so the effect of position and resource quality on growth and survival can be evaluated. Moreover, survivorship of eggs and larvae can be monitored since each gall contains the history of activity within it. In gall wasp communities, inquilines and parasitoids are usually abundant so that information on the third trophic level is easily obtained.

Recent recognition of the importance of plant-herbivore interactions gives a study of gall biology cogency. Such studies can contribute to our current understanding of ecological principles such as coevolution, host specificity, niche partitioning, competition, and community regulation and stability.

The chapters of this thesis are in the form of manuscripts appropriate for submission to scientific journals for publication. Chapter 1 has been accepted by the Proceedings of the Entomological Society of Washington and is in press at the time of thesis submission. Chapter 2 is being prepared for submission to the Southwestern Naturalist. The format of this thesis was approved by the Graduate School at Southwest Texas State University.

CHAPTER 1
HETEROGONY IN *BELONOCNEMA TREATAE* MAYR
(HYMENOPTERA: CYNIPIDAE)

Introduction

Phytophagous members of the family Cynipidae induce a spectacular diversity of plant galls that are often complex in structure. Worldwide, approximately 2,000 species of Cynipidae have been described, with 805 occurring in North America (Dreger-Jauffret and Shorthouse 1992). Typically, individual cynipid species induce galls on a single species of host plant or on a series of closely related plant species. In the case of phytophagous cynipids that induce galls on oaks, all host species are generally in the same subgenus. Knowledge of the biology and life history of known cynipid species is largely fragmentary, and there remain many undescribed species (Askew 1984, Meyer 1987, Dreger-Jauffret and Shorthouse 1992).

Cynipid wasps can exhibit an alternation of generations known as heterogony in which an all-female generation alternates with a bisexual generation (Lyon 1963, 1964; Felt 1965, Askew 1984, Rey 1992). The unisexual generation produces eggs parthenogenetically, and those eggs are usually inserted into a specific plant part. At the site of oviposition, galls are induced from which a bisexual generation develops and later emerges. Emergent males and females mate, and females in turn initiate galls from which the unisexual generation emerges. Females of the two generations may be morphologically dissimilar and may induce galls that differ greatly in morphology whether oviposition occurred in the same or different plant organs (Felt 1965, Lyon 1969, 1970, Meyer 1987, Rey 1992). Differences in the morphology of both female wasps and galls between

generations, coupled with incomplete knowledge of life cycles, has led to considerable taxonomic confusion within the Cynipidae. Alternating generations of numerous species have been described as separate species or even genera (Dreger-Jauffret and Shorthouse 1992, Rohfritsch 1992).

Here we demonstrate that two cynipid species, Belonocnema treatae Mayr 1881 and B. kinseyi Weld 1921, wasps that are generally similar in morphology but produce morphologically dissimilar galls on their host plant, plateau live oak Quercus fusiformis Small (formerly Q. virginiana var. fusiformis) represent alternate generations of a single species. This is the first demonstration of heterogony in North American Cynipidae outside the Pacific Slope region (Lyon 1996). The species name B. treatae has priority, thus B. kinseyi becomes a synonym, as is Dryorhizoxenus floridanus Ashmead 1881 (Ashmead 1886). We also provide a description of the life cycle of the species, a description of the galls induced by unisexual and bisexual females, and a list of the inquilines, parasitoids, and hyperparasitoids associated with galls produced by both generations. The observations and timing of events reported herein describe populations of B. treatae from the Southwest Texas State University Freeman Ranch located in Hays County, Texas. The experiments reported herein were conducted using wasps and plants from this same location.

Establishing Synonymy

Mayr (1881) described B. treatae, and Weld described B. kinseyi in 1921. Weld (1921) speculated that B. kinseyi and B. treatae could be alternate generations of a single species, but this relationship has only now been demonstrated. Belonocnema kinseyi emerges from pea-shaped galls on the leaves of Quercus fusiformis from mid October through early November. No males have been described for this species. Belonocnema treatae, described from both male and female specimens, has been obtained from root galls on Q. fusiformis from mid March through early April in both 1996 and 1997.

To link B. treatae emerging from root galls with the leaf galls from which B. kinseyi emerge, we performed a caging experiment in the field. Observations in the spring of 1995 and 1996 showed that leaf galls on Q. fusiformis developed from oviposition events that occurred during and shortly after bud break. In the spring of 1997, prior to bud break and prior to emergence of B. treatae, exclusion cages constructed of fine-mesh, polyester-fiber cloth were placed over eight branches of a Q. fusiformis tree that had exhibited a high density of B. kinseyi leaf galls during the 1996 growing season. Four additional bags were placed over branches of a second tree that exhibited a moderate density of leaf galls in 1996. Clusters of root galls were then collected from rootlets underneath both trees prior to bud break, returned to the laboratory, and placed in collection traps. Emergent wasps were provided with nectar sources and allowed to mate for 2 days. From 4 to 15 female wasps were then introduced into each of 5 cages on tree # 1, and into each of 2 cages on tree # 2. Five cages remained sealed as controls (tree #1; $n = 3$, tree #2; $n = 2$). Three months later all bagged branches were harvested and leaves were scored for the presence and number of oviposition scars and developing galls. In total, 2,378 oviposition scars and 207 developing B. kinseyi leaf galls were present among 1,356 leaves in cages in which B. treatae was introduced. No leaf galls were initiated in the 5 control cages. Thus, on plateau live oak, leaf galls that develop to produce the unisexual generation (i.e., B. kinseyi) are initiated by females of the bisexual generation (i.e., B. treatae) that emerge from root galls confirming the synonymy of the two species.

In the laboratory, we tested the oviposition preference of B. treatae reared from root galls of Q. fusiformis using both individual mated females and groups of mated females. Eight mated female B. treatae were placed separately in (3.2 x 9.6 cm) vials and presented with one shoot of Q. fusiformis and a second shoot from Q. shumardii Buckl. (southern red oak) for 24 hours. Four females oviposited exclusively on Q. fusiformis and four failed to oviposit. Three group cages, each containing 15 male and 15 female B. treatae, were established and wasps were provided with one shoot each of Q. fusiformis, Q.

shumardii, and Q. macrocarpa Michx. (bur oak). Leaves were scored for oviposition scars after 3 and then 4.5 hours. In all cages, oviposition scars were evident on Q. fusiformis after 3 hours, whereas no oviposition scars were visible on Q. shumardii or Q. macrocarpa at 4.5 hours. A clear preference for oviposition on Q. fusiformis was demonstrated by both individual and groups of B. treatae females.

Description of Galls

Leaf galls---Leaf galls produced by the bisexual generation develop following oviposition on buds and on newly unfurled leaves. Galls develop on the undersurface of leaves and are unilocular, smooth, and pea-shaped. At maturity, galls are fully lignified and range from 3 to 7 mm in diameter (Figs. 1, 2). Unisexual females emerged from lignified galls from mid October through early November in both 1996 and 1997. No males have been recorded from the 115 wasps that have emerged from leaf galls in the laboratory.

Root galls.---Root galls induced by unisexual females grow in irregularly shaped, multilocular clusters on small rootlets just below the soil surface (Figs. 3, 4). Clusters appear fleshy and yellow and detach easily from the root surface. Sampled galls were composed of from 1 to 28 chambers and measured 5 to 28 mm in length. Developing root galls have been found in early January and at this stage appear yellow-green in color. Mature galls (those from which the bisexual generation has emerged in the laboratory) have been collected in mid February. In the field, emergence holes in root galls were first observed in mid March 1997, coincident with the appearance of oviposition scars on leaves of the host plant.

Host Plant

Weld (1921) gave Quercus virginiana Miller (Fagaceae) as the host plant for both B. kinseyi and B. treatae at the type locality in Boerne, Texas. Recent analyses of Texas

live oak populations, however, indicate a complex pattern of hybridization between Q. virginiana (coast live oak) and Q. fusiformis (formerly listed as a variety of Q. virginiana) (Nixon 1984). Quercus fusiformis is common throughout the Edwards Plateau region of south-central Texas, whereas Q. virginiana reaches its western limit in coastal Texas east of the Brazos River. In the broad region bounded by the Brazos River and the Edwards Plateau, extensive hybridization between the two parent species followed by backcrossing has produced a complex mixture of phenotypes that tend to resemble Q. fusiformis more than Q. virginiana (Nixon 1984). We note that Boerne, Texas, is located within the geographic range of Q. fusiformis well to the west of the hybrid zone and the recognized range of Q. virginiana. Thus the host species recorded by Weld should be amended to Q. fusiformis.

Weld (1921) also noted the presence of leaf galls on live oaks in Houston, Wharton, Victoria, Cuero, Austin, Sabinal, and Kerrville, Texas, which he ascribed to B. kinseyi on the basis of gall morphology. We note that these sites span the entire range of the two parental oak species and their zone of hybridization in Texas. Thus, either B. treatae's host range includes both parental oak species and their hybrid, or Weld's site east of the Brazos River, that is, Houston, represents leaf galls induced by a congeneric species, B. quercusvirens (Osten Sacken) (Osten Sacken 1861, Krombein et al. 1979). At the present time, however, B. quercusvirens is known from only Florida (E. Grissell pers. comm.). All leaf galls and root galls in the present study were collected from the eastern edge of the Edwards Plateau and western edge of the hybrid zone. We have not examined leaf or root galls collected from pure Q. virginiana populations located east of the Brazos River. Thus, at the present, we list the host range of B. treatae as Q. fusiformis and the Q. fusiformis x Q. virginiana hybrid.

Associated Hymenopteran Species

Thirteen hymenopteran species belonging to 7 families have been commonly reared from mature leaf galls in addition to the gall former. Belonocnema treatae larvae developing within leaf galls are frequently attacked by 3 species of parasitoids: Acaenacis lausus (Walker) (Pteromalidae), Ormyrus labotus Walker (Ormyridae), and Torymus tubicola (Osten Sacken) (Torymidae). Four species described as being inquilines-- Synergus sp. (Cynipidae) Sycophila flava (Ashmead), Eurytoma furva Bugbee, and Eurytoma sp. (Eurytomidae)--and one hyperparasitoid Eurytoma bugbeeii Grissell have also been reared from leaf galls. The trophic level of 5 other species, Allorhogas sp. (Braconidae), 3 unidentified Brasema and Galeopsomyia sp. (Eulophidae), reared from galls is presently unknown. An additional 10 species have been reared rarely from leaf galls to date and await identification. Root galls held in collection traps in the laboratory yielded several specimens of Torymus tubicola (Torymidae) from mid to late April. Torymus tubicola is the only species that appears to parasitize larval B. treatae in both leaf and root galls.

Description of Adults

Belonocnema treatae Mayr

(Figs. 5-7)

Belonocnema treatae Mayr. 1881. Green Cove, Florida. 2 female wasps mounted on a single pin, labeled syntypes. Natural History Museum, Vienna.

Dryorhizoxenus floridanus Ashmead 1882(1881). Jacksonville, Florida. Holotype female #2813 and 5 paratypes. U. S. National Museum of Natural History, Washington, D.C. Describes male.

Dryorhizoxenus floridanus Ashmead. 1886. Acknowledges priority of Belonocnema over Dryorhizoxenus and places the latter in synonymy.

Belonocnema kinseyi Weld 1921. New Synonymy. Holotype female #22832 and 27 paratypes in the U. S. National Museum of Natural History, Washington, D.C. Examined by Lyon in 1961.

Type Data: Lewis Weld, in 1929, examined the 2 syntype females in the Natural History Museum, Vienna. The distinctive morphology was sketched and drawings were placed in Weld's personal notes now in the possession of R. Lyon. One female syntype was selected and labeled as the lectotype. The other female was mounted on a separate pin with the label paratype. There were 17 specimens sent to Mayr by Ashmead. These were labeled D. floridanus Det. Ashmead, and are synonyms of B. treatae. George Melika of the Systematic Parasitoid Laboratory, Hungary, examined the types at the Natural History Museum, Vienna in April, 1998.

Bisexual Generation

REDESCRIPTION (Lyon).---Female: Uniformly yellow brown, scutellum and propodeum dark brown, almost black; abdomen light brown (Fig. 5E). Length 3.5 to 3.75 mm (\bar{x} = 3.7 mm, n = 27 Weld specimens). Head as broad as mesosoma, transverse, coriaceous, gena not broadened behind eyes, malar space (Fig. 5C) 0.33x eyelength with groove; frons tumescent; interocular area broader than high. Antenna 14 segmented, filiform, segment 3 longer than 4, terminal segment 2x as long as 13. Scutum (Fig. 5B) slightly convex but flattened, smooth, shining, as broad as long; notauli percurrent, curving laterally at anterior portion with a few, scattered, setigerous punctures along lateral margins. Scutellum almost circular, nearly two-thirds length of scutum, coarsely rugose, roughly irregular along margin; foveae deep, separated with smooth, shining bottoms. Propodeum with irregular ridges, area between rugose. Mesopleuron bulging, slightly setose on lower portions. Forewing hyaline (Fig. 5A), short ciliate, longer on lower margins, pubescent; veins dark brown, R_{52} heavy, curving toward wing margin forming a

distinct club. Darkened areas around R_1 , R_{S1} , and R_{S2} , radial cell short. Legs bristly; foretibia with a distinctive spur (Fig. 5D), as long as furcula; tarsal claws edentate. Metasoma (Fig. 5E) longer than high with all terga visible along dorsal curvature. Hypopygeal spine short with bristles extending beyond apex.

Male: Similar to female but slightly smaller, length 3.4 to 3.75 mm (\bar{x} = 3.6 mm, n = 5). Color light brown but blackened along margins of pronotum. Antenna 15 segmented, filiform, 3rd longer than others and deeply excavated, terminal segment equal in length to 14th. Wing pubescent, ciliated, R_{S2} curving toward the wing margin, slender and only slightly enlarged at tip. Foretibial spur much shorter than in female and sometimes not elongated. Tergite 2 oval and occupying most of abdomen, remaining terga visible along dorsal curvature but short.

Unisexual Generation

REDESCRIPTION (Lyon).--- Female: Head and metasoma red brown; mesosoma very dark, almost black. Length 2.0 to 3.1 mm (\bar{x} = 2.7 mm, n = 62 Weld specimens). Head transverse (Fig. 6B), slightly broader than mesosoma, coriaceous, gena not broadened behind eyes, malar space (Fig. 6E) less than half length of eye with groove; interocular space broader than high; frons tumescent. Antenna 13 segmented, 3rd nearly 2x length of 4th segment, 4-12 gradually shorter, terminal segment 2x length of preceding one. Scutum (Fig. 6B) slightly convex but flattened, smooth, bare, shining; notauli percurrent, deep, narrow, curving slightly along upper lateral margins; a very short median posterior groove in some specimens. Scutellum sloping to a broad, rounded, roughened margin, slightly more than half length of scutum; foveal pits large, deep, separated with smooth, shining bottoms. Propodeum with two curved ridges, area between rugose. Mesopleuron bulging, shiny with setae along lower portion. Forewing hyaline (Fig. 6A), pubescent, short ciliate along anterior margin, longer on posterior margin; veins brown, R_{S1} with dark areas, R_{S2} curving toward wing margin with slightly flattened club at apex;

radial cell short, 2x as long as broad. Metasoma (Fig. 6C) longer than high with all terga showing along dorsal margin. Hypopygeal spine with bristles extending beyond apex. Foretibia with an extended spine almost as long as furcula (Fig. 6D).

Comparison of the unisexual and bisexual generation females---Average bisexual-generation females are larger than unisexual-generation females--3.6 mm versus 2.7 mm. This is unusual; unisexual-generation females are generally larger than those of the bisexual generation (R. Lyon in litt.). Bisexual females are mostly light yellow brown with dark scutellum and propodeum. Unisexual females are darker, with the mesosoma almost black and the head and metasoma red brown. The scutellum of bisexual females is broader and more rounded, appearing almost circular in outline, whereas the unisexual females have a narrower scutellum that slopes to a broad, more-rounded point. Bisexual females have 14 segmented antennae, unisexual females have 13 segmented antennae. Bisexual females have wing vein R_{S2} heavier, more sharply curving toward wing margin, and club at tip larger and more rounded than do unisexual females, for which R_{S2} is more slender and less sharply curved and the terminal club is smaller.

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Literature Cited

- Ashmead, W. H. 1882 (1881). American Entomological Society Transactions 9:Proceedings p. xxv.
- Ashmead, W. H. 1886. American Entomological Society Transactions 13:Proceeding 63.
- Askew, R. R. 1984. The biology of gall wasps, pp. 223-303. In T.N. Ananthakrishnan, ed., The Biology of Gall Insects. Oxford and IBH Publishing Company, New Delhi.
- Dreger-Jauffret, F., and J. D. Shorthouse. 1992. Diversity of gall-inducing insects and their galls, pp. 8-33. In J. D. Shorthouse and O. Rohfritsch, eds., Biology of Insect-Induced Galls. Oxford University Press, New York.
- Felt, E. P. 1965. Plant galls and gall makers. Hafner Publishing Company, New York, New York.
- Krombein, K. V., Hurd, P. D., Smith, D. R., and B. D. Burks. 1979. Catalog of Hymenoptera in America North of Mexico. Smithsonian Institution Press, Washington, D.C.
- Lyon, R. J. 1963. The alternate generation of Heteroecus pacificus (Ashmead) (Hymenoptera: Cynipoidea). Proceedings of the Entomological Society of Washington 65:250-254.
- Lyon, R. J. 1964. The alternate generation of Callirhytis agrifoliae (Ashmead) (Hymenoptera: Cynipoidea). Proceedings of the Entomological Society of Washington 66:193-196.
- Lyon, R. J. 1969. The alternate generation of Callirhytis quercussuttonii (Bassett) (Hymenoptera: Cynipidae). Proceedings of the Entomological Society of Washington 71:61-65.

- Lyon, R. J. 1970. Heterogony in Callirhytis serricornis (Kinsey) (Hymenoptera: Cynipidae). Proceedings of the Entomological Society of Washington 72:176-178.
- Lyon, R. J. 1996. New cynipid wasps from the southwestern United States (Hymenoptera: Cynipidae). Pan-Pacific Entomologist 72:181-192.
- Mayr, G. 1881. Die genera der gallenbewohnenden cynipiden. Zwanzigsten Jahresberichte der Communal Oberrealsch nie im. I. Bezirke, Wien, pp. 4, 16, 38.
- Meyer, J. 1987. Plant Galls and Gall Inducers. Gebrüder Borntraeger, Berlin.
- Nixon, K. C. 1984. A biosystematic study of Quercus series Virentes (the Live Oaks) with phylogenetic analyses of Fagales, Fagaceae and Quercus. Ph.D. dissertation, University of Texas, Austin.
- Osten Sacken, C. R. 1861. Proceedings Entomological Society of Philadelphia 1:59:241.
- Rey, L. A. 1992. Developmental morphology of two types of hymenopterous galls, pp. 87-101. In J. D. Shorthouse and O. Rohfritsch, eds., Biology of Insect-Induced Galls. Oxford University Press, New York.
- Rohfritsch, O. 1992. Patterns in gall development, pp. 60-86. In J. D. Shorthouse and O. Rohfritsch, eds., Biology of Insect-Induced Galls. Oxford University Press, New York.
- Weld, L. H. 1921. American gallflies of the family Cynipidae producing subterranean galls on oak. Proceedings US. National Museum 59:238-9, 241-2.



Figure 1. Unilocular leaf galls induced by the bisexual generation of *Belonocnema treatae*.

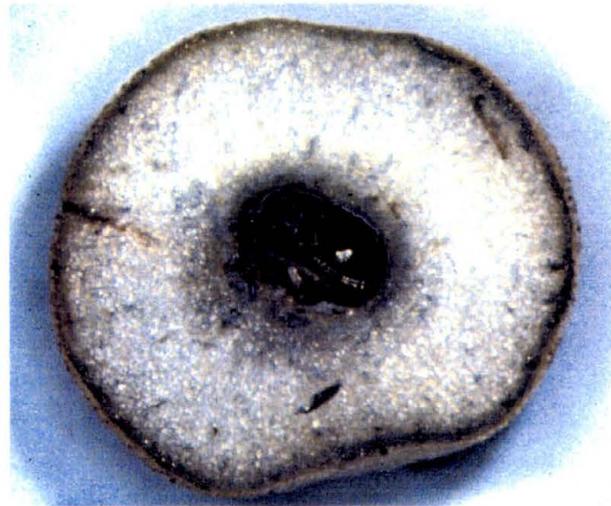


Figure 2. Sectioned leaf gall showing pre-emergent adult of the unisexual generation.



Figure 3. Multilocular root gall induced by the unisexual generation.



Figure 4. Multilocular root gall with pupae of the bisexual generation exposed in cross section.

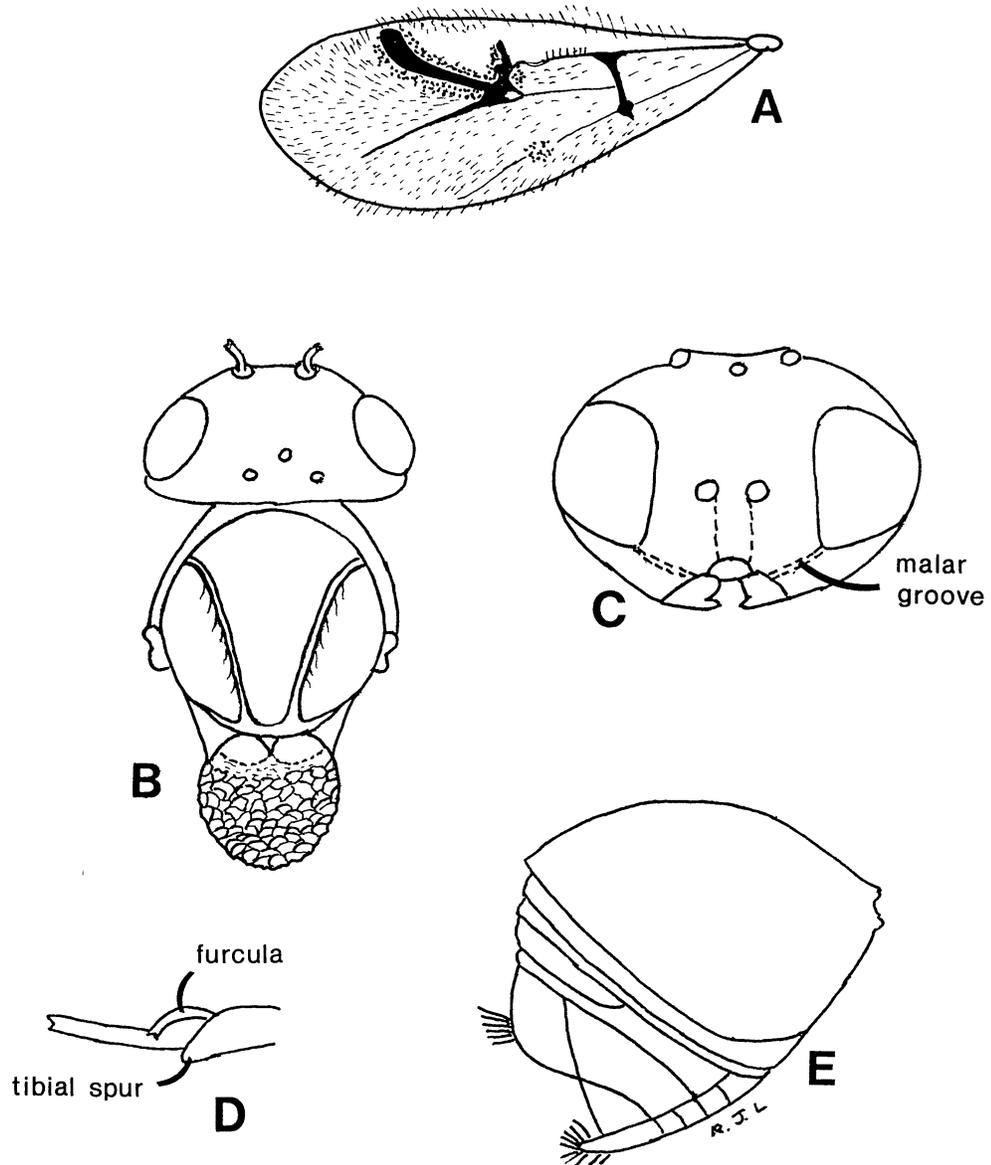


Fig. 5. Belonocnema treatae (bisexual female). A, forewing venation, lateral view (x 25); B, head and mesosoma showing detailed morphology of scutum and scutellum, dorsal view (x 40); C, frontal view of head showing malar furrow (x 60); D, foretibia showing elongated, characteristic spur and furcula; E, lateral view of metasoma showing shape of abdominal terga (x 60).

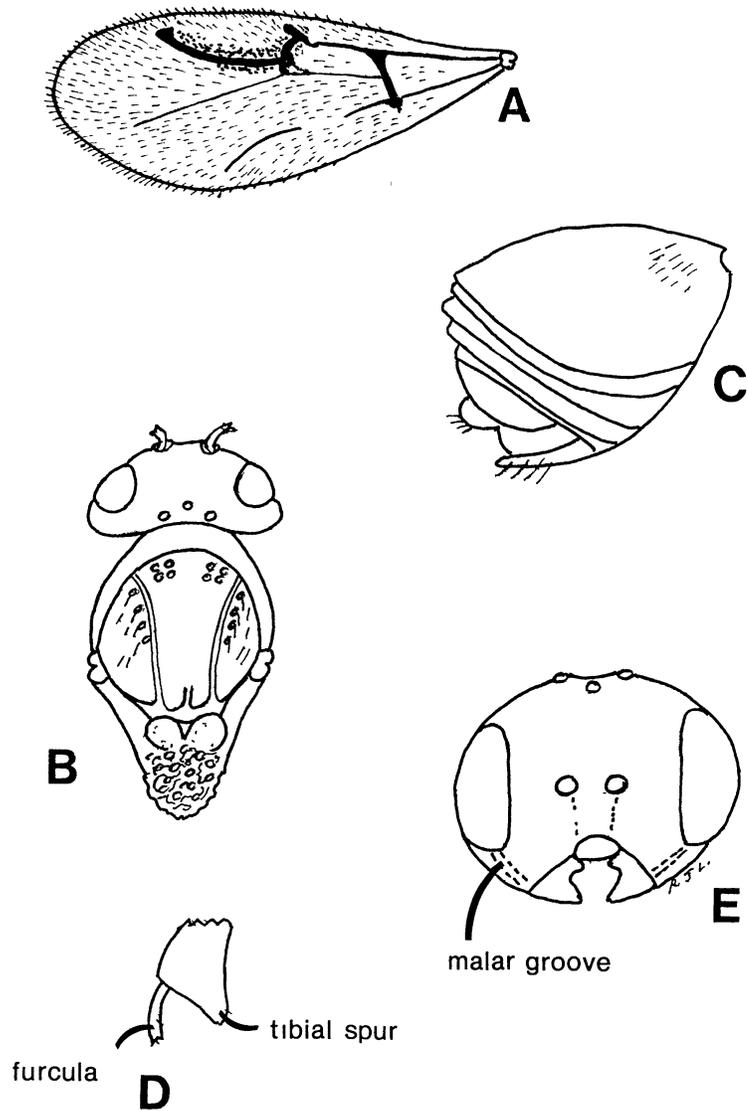


Fig. 6. *Belonocnema treatae* (unisexual generation female = *B. kinseyi*). A, forewing venation, lateral view (x 25); B, head and mesosoma showing detailed morphology of scutum and scutellum, dorsal view (x 40); C, lateral view of metasoma showing shape of abdominal terga (x 40); D, foretibia showing elongated, characteristic spur and furcula; E, frontal view of head showing malar furrow (x 60).



Figure 7. *Belonocnema treatae* adults; unisexual female (left) and bisexual female (right).

CHAPTER 2
LIFE CYCLE AND ECOLOGY OF THE GALL WASP
BELONOCNEMA TREATAE MAYR (HYMENOPTERA: CYNIPIDAE)

Introduction

An insect-induced gall is an atypical outgrowth on a plant induced by oviposition and subsequent larval feeding, the development of which is controlled by the insect (Askew 1984, Meyer 1987, Rey 1992). Gall insects are specialized herbivores whose larvae are sheltered and nourished by gall tissue throughout development to adulthood. Worldwide, there are 7 orders of gall-inducing insects, including 13,000 recorded species (Dreger-Jauffret and Shorthouse 1992). The order Hymenoptera includes 5 families of gall-inducers. Within the order Hymenoptera, the family Cynipidae includes 2000 species, 805 of which occur in North America alone. Gall inducers deposit eggs in or on tissues of the host plant, the eggs hatch and the larvae begin feeding, which promotes gall formation.

Many cynipids have a complex life cycle known as heterogony, whereby a bisexual generation comprised of males and females alternates with an unisexual female generation (Felt 1965; Askew 1984; Dreger-Jauffret and Shorthouse 1992; Rey 1992). Adults of each generation may attack the same or different plant organs usually on the same host plant, producing galls of very different structure. This reproductive cycle involves two types of females which differ in morphology and mode of reproduction (Felt 1965; Meyer 1987; Rey 1992). As a result, for many cynipids, the two generations have been incorrectly described as distinct species.

The primary inhabitants of cynipid galls are the larvae of the gall inducer, which are completely confined within host plant tissue and are thus easily located by natural enemies.

Associated with cynipid galls are numerous hymenopteran species that usurp the gall and/or attack gall maker larvae. Inquilines are common gall inhabitants that feed on gall tissue but are incapable of gall induction. Most are themselves cynipids; inquiline cynipids comprise 7 genera and about 175 species of the 2000 described cynipid species worldwide (Draeger-Jauffret and Shorthouse 1992). Inquilines commonly oviposit in newly initiated galls and become enclosed in discreet larval chambers within the gall as development proceeds, serving as an additional source of food for entomophagous species. Entomophagous inhabitants of cynipid galls include parasitoids, which feed on the larvae of the gall maker, and hyperparasitoids, which attack inquilines and parasitoids. Most belong to two Superfamilies, the Chalcidoidea and Ichneumonoidea, which currently include at least 38,000 described species (Grissell and Schauff 1990). Oviposition phenology of parasitoids associated with cynipid galls is delimited by development of the gall maker larva and development of the gall itself. Some entomophagous species are host specific, while others feed opportunistically on any larvae encountered in the gall (Wiebes-Rijks and Shorthouse 1992). Inquilines and parasitoids are a major cause of mortality in most cynipid populations (Askew 1984; Dreger-Jauffret and Shorthouse 1992; Wiebes-Rijks and Shorthouse 1992).

Belonocnema treatae induces galls on *Quercus fusiformis* (plateau live oak) and on *Q. fusiformis* x *Q. virginiana* (coast live oak) hybrids (Lund et al. 1998) (Figure 1). Live oaks are evergreen to sub-evergreen, with partial to complete leaf fall occurring from February-March (Correll and Johnston 1979, Lund personal observation). Flowering continues throughout March coincident with budbreak.

Belonocnema treatae exhibits heterogony. The alternate generations of *B. treatae* were previously described as two species (Mayr 1881, Weld 1921), but have recently been synonymized (Lund et al. 1998). *Belonocnema treatae* unisexual females develop within pea-shaped, unilocular leaf galls on *Quercus fusiformis* and *Q. fusiformis* x *Q. virginiana*

hybrids, while the bisexual generation matures within fleshy, multilocular galls on the roots of the same host plant (Lund et al. 1998) (Figure 2).

Leaf galls induced by *Belonocnema treatae* are common on live oak in central Texas, sometimes at extraordinary densities, and variation in gall density among closely adjacent trees may span 3 orders of magnitude (unpublished data). Yet knowledge of the life cycle and community associations of this species is largely incomplete. Aspects of the biology of *B. treatae* give an intensive study cogency. First, live oak is both popular and valuable as a shade tree in many parts of the southern United States. The impact of galling by *B. treatae* on tree fitness is therefore of interest. Moreover, *B. treatae* larvae develop in intimate association with host plant tissues including roots, making it a potential vector of oak wilt disease. The hymenopteran community associated with *B. treatae* galls is of interest due to the tremendous mortality exerted by parasitoids and the resultant selection pressure on the gall maker (Jeffries and Lawton 1984, Price et al. 1986). Indeed, parasitoids have been introduced successfully as biological control agents for gall inducing insects (Gagné 1984, Godfray 1994).

This study provides basic information on the life cycle of *Belonocnema treatae*, component species of the gall community, the relative abundance of each, and the ecological relationships among participants. The objectives of this project were, therefore, multifold. Aspects of the basic biology of *B. treatae* including timing events were studied in detail, the knowledge of which was prerequisite for further ecological study. Second, the Hymenoptera associated with *B. treatae* galls were identified in order to understand the nature of their interactions, and the relative effects of each on *B. treatae* success. Knowledge of the development of galls and their parasitoids establishes a firm foundation for future investigations into the evolutionary ecology of 4 trophic level interactions.

Methods

The major portion of this work was completed at Freeman Ranch, an area of 3600 acres owned by Southwest Texas State University and located approximately 5 miles west of San Marcos, Hays county, Texas. The study site is found along the western edge of the host plant hybrid zone.

Studies on the life cycle of *Belonocnema treatae*

Oviposition and development of leaf galls: To determine the timing and pattern of oviposition by *B. treatae* bisexual females in the field, shoots bearing buds and/or newly unfurled leaves were collected from March 10-April 28, 1997 at 1-3 day intervals from two live oak trees having high densities of *B. treatae* leaf galls the previous year. Shoots were fixed in 70% ethanol and buds and leaves were later microscopically examined for oviposition scars, defined as an area of tissue lysis on a leaf vein. For each date, number of oviposition scars per leaf was recorded. Percent leaves with oviposition scars by sample was also noted.

To observe oviposition behavior of bisexual females and the appearance of new oviposition scars, *B. treatae* were reared from root galls held in emergence traps in the laboratory. Eight mated female *B. treatae* were placed separately in (3.2 x 9.6 cm) vials, presented with one shoot of *Q. fusiformis*, and observed continuously for 3 hours.

To examine the pattern and timing of gall development and obtain an age-specific summary of mortality rates, a cohort of galls was monitored from initiation to maturity by conducting a biweekly census. In April of 1996, 60 shoots showing new growth were labeled on a live oak tree known to support a high gall density the previous year (tree #1). A total of 221 individual leaves were arbitrarily selected from labeled shoots and the number and position of oviposition scars were mapped. Gall diameter, number of exit holes, events of herbivore attack, general appearance, and gall color were monitored at 2 week intervals until all galls were fully lignified (October 9-11). Gall diameters were

measured in the field using a metric ruler. Evidence of emergence from galls through exit holes was noted by the presence of small spherical openings in the gall wall. Galls with a section of the gall wall removed were recorded as attacked by herbivores.

Emergence from leaf galls: To assess the relative abundance and timing of emergence of *B. treatae* unisexual females, galled leaves were collected from multiple moderate to heavily galled trees at two week intervals from August through November, 1996. Individual galls were detached from leaves and isolated in gelatin capsules. Number and date of emergence of *B. treatae* adults were recorded. Emerging wasps were fixed in 70% ethanol and identified.

Emergence from root galls: To document the timing of emergence and sex ratio of the bisexual generation, 18 root gall samples were collected from February 17-April 4, 1997, at 1-5 day intervals from live oaks known to bear a high density of *B. treatae* leaf galls by clipping a short section of root to which galls remained attached. These were placed in emergence traps in the laboratory and covered with moist soil. Date of emergence, number, and sex were recorded daily until emergence ceased, after which number of galls per sample and number of exit holes per gall were documented. Wasps collected from traps were fixed in 70% ethanol. A two-tailed chi-square goodness of fit test was performed to test the hypothesis that females and males emerge in equal numbers. To estimate life span of the bisexual generation, groups of wasps emerging on the same date were held in cages with pollen sources and monitored daily until none remained living.

To document emergence phenology and sex ratio of *B. treatae* from root galls in the field, a malaise trap was erected on March 4, 1997 under a tree having a high relative leaf gall density. This date preceded *B. treatae* emergence documented in 1996. Trapfall was collected daily and fixed in 70% ethanol, then scored with regard to date of emergence and number of individuals of each sex. A Spearman rank correlation analysis was conducted to compare the phenology of mean oviposition scars per leaf to that of bisexual females trapped in the field.

To document behavior of males and females in the field, daily observations on the activity of bisexual *B. treatae* adults were recorded under heavily galled trees during March 1997. Densities were estimated by delimiting an area of 1 square meter and counting individuals contained therein.

Hymenoptera associated with *B. treatae* galls

Identification, relative abundance, and emergence phenology: To identify the Hymenoptera associated with *B. treatae* leaf galls, their relative abundance and timing of emergence, galled leaves were collected bimonthly from two moderate to heavily galled trees from late July through early March (leaf fall). This time interval coincides with the availability of mature, lignified galls. Continuous sampling was necessary due to variation in developmental stage of individual galls throughout the growing season, unknown time of attack by parasitoids, and potential deleterious effects of timing of gall harvest on wasp emergence. Samples were collected for two seasons beginning summer 1995 through spring 1996, and again from summer 1996 through spring 1997. Galled leaves (range 190-1,305) from each sample were placed in emergence traps and emergent gall-associated Hymenoptera were fixed in 70% ethanol. Representatives of each "species" were sent for identification to the USDA Systematic Entomology Laboratory (SEL) in Beltsville, Maryland to establish a reference collection for identification of subsequent specimens. Number of emergents and date were documented for all species. Relative abundance of associated Hymenoptera was examined for both seasons by comparing the proportion of total emergents by species .

The emergence phenology of each species associated with *B. treatae* leaf galls was estimated by the number emerging per 10² galls, based on the cumulative number of galls collected to the sample date, computed weekly. The relative number of wasps emerging per sample date was expressed as wasps per gall to compensate for fluctuations in the number of galls per sample. Emergence phenology by guild was also examined each year.

To document the hymenopteran community associated with root galls induced by *B. treatae*, root galls were collected from mid February to early April from the same trees providing leaf gall samples, and placed in emergence traps covered with moistened soil. Hymenoptera were identified, and emergence phenology, sex, and relative abundance were documented for all species.

Currently no information is available on the nature of interactions among Hymenoptera associated with either leaf galls or root galls of *B. treatae*. To examine the nature of interactions among hymenopteran species, 2 complimentary studies were conducted. In the first, individual galls were isolated in gelatin capsules, and all incidents of multiple emergence and the species order of emergence were recorded. In the second, the frequency of interaction among *B. treatae* community members was estimated by the number of adult wasps that successfully emerged from individual leaf galls (gall productivity). Number of exit holes per gall was counted for 5 leaf gall samples from the 1995/96 season from which emergence of wasps had ceased. This method provides a minimum estimate of gall productivity because wasps may use existing holes in the gall wall through which to exit (JoAnne Lund personal observation). To relate this productivity to observable gall characteristics, galls were sorted into two categories, smooth and wrinkled, based on condition of the outer gall wall. From previous samples *B. treatae* emerged only from smooth galls, suggesting that smooth galls were successfully initiated and developed, while galls with a wrinkled texture were initiated but growth ceased at some point during development. Within each condition category, galls were sorted by number of exit holes per gall, used as a minimum estimate of gall productivity.

Success of unisexual *B. treatae*: To estimate success of the unisexual generation, the percent of mature leaf galls producing *B. treatae* was computed for two years. As emergence of *B. treatae* occurs only during a narrow window from mid October through early November, there is little probability of *B. treatae* emergence from galls collected after

mid November. To avoid erroneous deflation of success estimates, only gall samples collected prior to and during this window of emergence were included.

Gall size and *B. treatae* success: From leaf galls in gelatin capsules, gall diameter was examined with respect to species emerging. To test the hypothesis that *B. treatae* emerges from leaf galls non-randomly with respect to gall size, a Students' t test for unequal variances was performed.

Results

Oviposition and development of leaf galls: Oviposition scars were visible on samples collected March 21 and thereafter, which coincides with the appearance of bisexual females in trapfall samples (Figures 3, 9). Oviposition proceeded at a rapid rate during the following two weeks, reaching a value (mean \pm sd) of 15.5 ± 1.0 scars per leaf on tree # 1 and 32.1 ± 1.3 on tree # 2 by April 4. A graphic examination of percent of leaves bearing scars by date illustrates that bisexual females oviposit almost directly after emergence (Figure 4). On tree # 2, more than 80% of leaves had oviposition scars within 3 days, while tree # 1 exhibited over 60% scar-bearing leaves within one week. Within 2 weeks, 100% of leaves examined bore evidence of oviposition. This observation is corroborated by host plant preference studies for which bisexual females oviposited within minutes to hours of emergence in the laboratory (Lund et al. 1998).

Bisexual females deposit eggs almost exclusively into lateral veins on the leaf undersurface, rarely on the midrib (Figure 5). A single female performs numerous oviposition events on the same leaf, and may continue on additional leaves (Lund, personal observation). Within minutes, oviposition sites darken as lysis of leaf tissue begins.

Galls remained minute for about 2 months after oviposition only becoming visible to the unaided eye in early June. At 2-4 weeks after oviposition, darkened bulges are apparent within leaf veins (Lund, personal observation). Early galls appear as reddish, irregular growths covered with trichomes, later becoming white and globular (Figure 5). This

initiation phase was followed by a period of rapid growth in June and July, with galls reaching maturity by August and September (Figure 6). Galls may occur singly on a leaf, or in clusters that are so numerous as to completely cover the lower leaf surface. During development, galls undergo a series of color changes. The most common color sequence observed was white, pink, red, yellow, orange, and finally brown as galls became lignified at maturity. The earliest evidence of parasitoid emergence occurred in early July. A total of 2,822 oviposition scars was present on the sample of 221 leaves, yielding 181 galls initiated (6.4 %). Only 41 of the original 221 leaves remained by early October when galls were fully lignified, yielding 52 mature galls (Table 1).

Emergence from leaf galls: *B. treatae* unisexual females emerged during a narrow window of time from early October through mid November (Figure 7) and burrow in the soil, seeking oviposition sites on rootlets of the same host plant. Of 3,116 galls from which Hymenoptera emerged, only 3.1% (n = 99) produced *B. treatae* unisexual females.

Emergence from root galls: Bisexual *B. treatae* adults emerged from root galls in the laboratory from February 28 through April 20, with peak emergence occurring the last week in March (Figure 8). A total of 107 root galls were collected averaging 8.55 ± 3.54 (mean \pm sd) exit holes per gall. Males significantly outnumbered females ($\chi^2 = 12.3$; $P < 0.001$; $df = 1$). Mean life span of the bisexual generation in the laboratory was estimated at 4.9 ± 1.3 days (mean \pm sd) (range 3-7 days) from 13 group cages (n = 15-30 wasps/cage).

B. treatae was present in trapfall from March 17-April 28 (Figure 9). Females were most abundant in samples collected March 28-April 4, while males peaked in abundance on March 22. Number of females in malaise trapfall was strongly correlated with mean oviposition scars per leaf by sample date ($r = 0.9$, $P < 0.0001$).

From March 20-22, males were observed hovering low in the understory and at densities that increased markedly on a daily basis. On both March 23 and 24, the peak density of the "swarm" of *B. treatae* males was estimated at 30 males per square meter.

On March 22, the earliest observation of females was noted, coincident with the earliest appearance of oviposition scars on leaves.

As the bisexual generation develops, galls grow in irregularly-shaped, fleshy clusters just below the soil surface (Chapter 1, Figure 3). Developing root galls have been found in early January, and at this stage appear yellow-green in color. Galls at maturity range in size from 5 to 28 mm in length and are usually multilocular and yellow in color. They are easily detached from the root surface. Collections from spring 1997 included galls composed of from 1 to 28 chambers, each chamber containing a developing wasp (Chapter 1, Figure 4). Mature galls, those from which the bisexual generation emerged in the laboratory, have been collected in mid February. In the field, exit holes in root galls were first observed in mid March, coincident with the appearance of oviposition scars on leaves of the host plant. Males and females mate within minutes to hours of emergence.

Identification, relative abundance and emergence phenology: In addition to unisexual *B. treatae*, 13 hymenopteran species representing 7 families were reared from leaf galls (Table 2). Identified species include three parasitoids; *Acaenacis lausus*, (Pteromalidae); *Ormyrus labotus*, (Ormyridae); and *Torymus tubicola*, (Torymidae), four species described as inquilines; *Synergus* sp. (Cynipidae); *Sycophila flava*, *Eurytoma furva*, and *Eurytoma* sp. (Eurytomidae); and one hyperparasitoid, *Eurytoma bugbeeii*. The trophic level of five other identified Hymenoptera; *Allorhogas* sp. (Braconidae), *Galeopsomyia* sp. and 3 unidentified species in the genus *Brasema* (Eulophidae) reared from galls is presently unknown. The identity and trophic status of 10 additional morphotypes reared rarely (1.2% of emergents) from leaf galls remain unknown.

The inquiline Cynipid, *Synergus* sp., had the greatest relative abundance for both seasons (Figure 10). The gall-inducing cynipid wasp, *B. treatae*, was least abundant of all identified species. Unidentified species comprised 1.1% (1995/96) and 1.3% (1996/97) of emerging wasps.

The Hymenoptera associated with *B. treatae* leaf galls continue to emerge for several months following gall maturation, each species showing a characteristic pattern of emergence (Figures 11-14). All species except *Galeopsomyia* sp. show a peak of emergence just prior to *B. treatae* emergence from leaf galls from mid October to early November. Emergence phenology of most species exhibits a generally bimodal pattern, however the composite emergence pattern for all species appears trimodal (Figure 15). Leaf gall initiation by bisexual *B. treatae* females is followed by a peak of emergence in natural enemies from leaf galls initiated the previous season. *Sycophila flava* (1996), an inquiline Eurytomid, and *Galeopsomyia* sp. (1997), trophic status unknown, exhibited a period of emergence coincident with gall initiation. *Synergus* sp., an inquiline Cynipid, and *T. tubicola* (1997 only), a parasitoid, show an additional peak of emergence in May and June shortly after gall initiation and prior to the period of rapid gall growth.

In contrast to the diverse assemblage of wasps associated with leaf galls, root galls held in collection traps in the laboratory yielded 1 parasitoid species, *T. tubicola* (Torymidae), in addition to the gall former. Twenty female and 8 male specimens were collected from mid-late April. *Torymus tubicola* was the only parasitoid species collected from both leaf and root galls.

Interaction among parasitoids associated with *B. treatae* leaf galls: Incidents of multiple emergence of associated Hymenoptera from individual galls in gelatin capsules were not rare (7.6%) (Table 3). A single gall may produce multiple individuals of *Synergus* sp., *T. tubicola*, *Allorhogas* sp., or *Galeopsomyia* sp. *Synergus* sp. may exit after the initial emergence of 6 other species, including the gall maker. However, *B. treatae* does not emerge after the initial emergence of another species. No multiple emergence was observed among *Eurytoma* species, *A. lausus*, and *Brasema* sp. 3. Estimates of gall productivity indicate that smooth galls (mean \pm sd; 1.2 ± 1.1 holes/gall) are more productive than wrinkled galls (0.57 ± 0.9 holes/gall) ($t = 11.5$; $p < 0.001$; $df = 3,046$) (Figure 16). A total of 3,048 galls were sampled; 84% were smooth, while 16% had a wrinkled texture.

Among smooth galls, 29.9% bore no exit holes and 30.7% had more than one exit hole. By contrast, 62.8% of wrinkled galls had no exit holes, and only 13.7% bore 2 or more exits. Moreover, *B. treatae* emerged only from galls with a smooth texture.

Success of unisexual *B. treatae*: Emergence of unisexual females from leaf galls in emergence traps was infrequent in both 1995 (n = 12 of 2,658) and 1996 (n = 17 of 1,965) (Table 4). Survivorship was estimated at 0.45 and 0.86%, respectively. From 6,897 galls in gelatin capsules, 99 (1.4 %) produced *B. treatae* unisexual females from mid October to mid November.

Gall size and *B. treatae* success: The mean gall diameter (\pm sd) for all galls was 6.07 ± 0.76 mm, and 6.94 ± 0.63 mm for those from which *B. treatae* emerged (Figure 17). The difference in mean gall size for the two groups was statistically significant ($t = -12.9$; $p < 0.0001$; $df = 131.5$). Among galls producing *B. treatae*, 95% had a diameter larger than 6.1 mm. Percent of galls producing *B. treatae* was examined by size class (Figure 18). As leaf gall diameter increases, the probability of *B. treatae* emergence increases exponentially.

Summary and Discussion

This study describes the life history attributes of *Belonocnema treatae* on its host plant, *Quercus fusiformis*. *Belonocnema treatae* exhibits heterogony, or alternation of generations. From mid March through mid April, the bisexual generation emerges from root galls, mates, then females oviposit into buds or young leaves. Eggs hatch and larvae begin feeding which induces gall formation. Leaf galls mature by fall when the unisexual females emerge from early October through early November and oviposit into rootlets on the same host plant, inducing the development of root galls.

Oviposition by bisexual *B. treatae* females is closely synchronized with bud break and/or leaf flush of the host plant in spring. The host organ must be in a particular stage of development when oviposition occurs (Rohfritsch 1992), resulting in a synchronized cohort of leaf galls for the season. Thus predator species on the gall maker are presented a

restricted window of vulnerability during which to attack. The two heavily-galled trees at the study site exhibited bud break during the third week of March from 1996 to 1998, approximately 1-2 weeks later than adjacent live oak trees (J. Lund personal observation), suggesting that among tree variation in leaf flush phenology may be an important determinant of gall density.

Although the developmental morphology of *B. treatae* leaf galls was not investigated, observations were consistent with general patterns described for other cynipid species. Cynipid eggs are known to exhibit proteolytic, cellulolytic, and pectinolytic activity in vitro (Rey 1992). The resulting lysis of leaf tissue frequently produces a series of holes on either side of the midrib (Figure 5). In *Diplolepis* sp., lytic action combined with cell dedifferentiation and hypertrophy induces formation of a cavity into which the newly hatched larva moves (Rohfritsch 1992). Within 4-6 weeks, eggs and/or larvae are fully encapsulated and become visible as darkened bulges within leaf veins.

Although further study is needed, the data presented here may be used to begin to predict the content of leaf galls. Results demonstrate that 1) *B. treatae* emerges only from smooth galls, and 2) 95% of unisexual females emerge from galls with a diameter greater than 6.1 mm (Figure 17). Thus, *B. treatae* is not likely to emerge from galls smaller than 6.1 mm, which precludes emergence from nearly 50% of smooth galls in our measured samples. This knowledge may permit estimates of *B. treatae* success from leaf gall observations in the field. Furthermore, galls with different fates may differ in sequence of color change. Such color changes have been noted in galls during development, which may be related to developmental stage of the gall and/or the species contained within it.

Among bisexual Cynipids, emergence of males prior to females has commonly been reported (R. Lyon personal communication). Male *B. treatae* emerging from root galls were found in malaise trapfall earlier than females and observed to aggregate under host trees in low-lying swarms; the predominant movement pattern was lateral. Females were observed moving vertically through the male swarm; females bearing 3-4 males were

commonly observed on vegetation during this period (J. Lund personal observation). This suggests that emerging females must pass through the male swarm to oviposit, thus ensuring insemination. The male-skewed sex ratio observed in malaise trap samples may therefore be explained in part by behavioral differences between the sexes.

Among identified species, the hymenopteran community associated with *B. treatae* leaf galls is diverse, representing 7 families and 4 guilds (the gall maker, inquiline, parasitoids, and a hyperparasitoid). The relative abundance of identified species (Figure 10) illustrates a characteristic feature of communities; few species are very abundant, while the largest number of species are relatively rare (Krebs 1994). Relative abundance within guilds was not examined due to uncertain trophic status of several identified species and the extreme morphological similarity among the Eurytomids. This diverse assemblage is in contrast with the depauperate root gall community consisting of the gall maker and one parasitoid, *T. tubicola*. The presence and relative abundance of community members was relatively stable from year to year with the notable exception *Galeopsomyia* sp., which was absent in the 1995/96 season, and comprised 15% of emergents from leaf galls in the 1996/97 season. Whether *Galeopsomyia* sp. was present but rare in 1995/96 or a recent arrival to the *B. treatae* community in 1996/97 is unknown. Little is presently known about parasitoid dispersal because of technical difficulties involved with tracking movement of small insects, however indirect evidence suggests that parasitoids regularly move quite large distances (Askew 1968, Godfray 1994).

Incidence of multiple emergence from a single gall fosters questions on the nature of interactions between larvae contained therein. Although inquiline species are described as phytophagous, *B. treatae* did not emerge after 4 species listed as inquilines. The apparent death of the gall maker may occur due to competition for limiting resources. Alternatively, gall maker remains have been observed not infrequently in unilocular galls along with an active adult inquiline (R. Lyon personal communication), suggesting that at least some inquilines may be predatory or shift from phytophagy to entomophagy. Such shifts in diet

from host larva to gall tissue have been reported for parasitoid species as well (Roskam 1992, Wiebes-Rijks and Shorthouse 1992), which may explain successful development of gregarious parasitoid larvae (*T. tubicola*) and/or emergence of 2 parasitoid species (*O. labotus* and *T. tubicola*) from a single gall.

Based on studies of various gall-inducing species, about 90% of gall insects do not survive to the end of the growing season (Wiebes-Rijks and Shorthouse 1992). The predominant mortality factors associated with cynipid galls are host plant defense, abiotic factors, and natural enemies. Data from the leaf gall cohort analysis indicate that only 9.1% of oviposition events result in mature galls (Table 1). From gall productivity estimates based on 2,559 mature galls, 30% produced no insect (Figure 16). These mortalities may be attributable to host plant effects or environmental factors.

Nearly all cynipid gall inducers support parasitoid communities (Askew 1984; Dreger-Jauffret and Shorthouse 1992; Wiebes-Rijks and Shorthouse 1992). The impact of cynipid enemies ranges from consumption of gall tissue without causing harm to host extinction (Force 1974; Washburn and Cornell 1981). A total of 13 hymenopteran species emerging from leaf galls have been identified in addition to the gall maker, and data suggest these have a major impact on *B. treatae* success. Among identified species emerging from leaf gall samples, the gall maker was least abundant in both the 1995 and 1996 seasons (Figure 10). Moreover, *B. treatae* emerged from only 0.45 and 0.86% of galls for the same seasons (Table 4).

Few species of parasitoids are host specific, most attack a range of galls (Askew 1984, Wiebes-Rijks and Shorthouse 1992). Some species attack small galls, some only mature galls such that there is a temporal succession of attack by natural enemies throughout the season of gall growth. Host specific species often attack larvae of gall inducers within days of gall initiation (Wiebes-Rijks and Shorthouse 1992). Results illustrate the synchrony of leaf gall initiation in March and April with a peak in emergence of natural

enemies comprised largely of *Synergus* sp., an inquiline Cynipid (Figure 15). *Synergus* sp. also showed the greatest relative abundance in both 1995 and 1996 (Figure 10). Conversely, Hymenoptera that emerge throughout the year are likely generalists, attacking other gall inducers opportunistically in addition to *B. treatae*. For example, some *Eurytoma* and *Torymus* species are known to emerge from galls in the same season they are initiated, then search for galls induced by other species in which to oviposit and overwinter (Wiebes-Rijks and Shorthouse 1992).

Selection would favor protective mechanisms against such exploitation by natural enemies. The alternation of generations exhibited by Cynipids may allow escape from enemies by partitioning host space and/or providing enemy-free space (Price et al. 1980, Jeffries and Lawton 1984, Price and Clancy 1986, Denno et al. 1993). Due to heterogony, the primary host gall may not be available at the time of enemy emergence forcing a change in host galls. Askew (1961) found that the parasitoids attacking alternate generations of a given cynipid species usually differ. Among the 13 Hymenoptera associated with *B. treatae*, *T. tubicola* was the only species collected from both leaf and root galls.

Rapid gall development may also limit enemies of cynipids to a narrow window of opportunity during which galls are vulnerable to attack. Hardening of gall walls at maturity is an important deterrent to parasitoid attack (Washburn and Cornell 1979, Weis 1982). Leaf gall development in *B. treatae* requires approximately 6 months from initiation to emergence of the unisexual generation. A period of rapid gall growth was documented from June 1-July 15, with a 54% increase in mean gall diameter during the 2 week period from July 1-15 (Figure 6). By late July, 43% of galls surveyed were lignified. Moreover, the degree of gall parasitism may decline with increasing gall size (Jones 1983, Weis et al. 1985, Price and Clancy 1986). Species attacking early in gall development have shorter ovipositors than species attacking later, therefore rapid gall development may preclusively minimize gall parasitism (Weis et al. 1985, Wiebes-Rijks and Shorthouse 1992).

Results demonstrate that as leaf gall size increases, the probability of *B. treatae* emergence increases exponentially (Figure 18), suggesting that large galls may confer immunity to parasitoid attack. If, for the gall maker, there are strong penalties associated with small galls, selection would favor large gall size. Although heritability of gall size was not investigated, the potential remains for small changes in gall size to influence parasitoid populations, and in turn affect the population dynamics of the community consociation of *B. treatae*.

Literature cited

- Askew, R. R. 1961. On the biology of the inhabitants of oak galls of Cynipidae (Hymenoptera) in Britain. *Trans. Soc. Brit. Entomol.* 14:237-268.
- Askew, R. R. 1968. Considerations on speciation in Chalcidoidea (Hymenoptera). *Evolution* 22:642-645.
- Askew, R. R. 1984. The biology of gall wasps. Pages 223-303 *in* T.N. Ananthakrishnan, editor. *The biology of gall insects*. Oxford and IBH Publishing Company. New Delhi, India.
- Correll, D. S. and M. C. Johnston. 1979. *Manual of the vascular plants of Texas*. The University of Texas at Dallas. Richardson, Texas, USA.
- Denno, R. F., Larsson, S., and K. L. Olmstead. 1993. Role of enemy-free space and plant quality in host-plant selection by willow beetles. *Ecology* 71:124-137.
- Dreger-Jauffret, F. and J. D. Shorthouse. 1992. Diversity of gall-inducing insects and their galls. Pages 8-33 *in* Joseph D. Shorthouse and Odette Rohfritsch, editors. *Biology of insect-induced galls*. Oxford University Press. New York, New York, USA.
- Felt, E. Porter. 1940. *Plant galls and gall makers*. Hafner Publishing Company, New York, New York, USA.
- Force, D. C. 1974. Ecology of insect host-parasitoid communities. *Science* 184:624-632.
- Gagné, R. J. 1984. The geography of gall insects. Pages 305-322 *in* T.N. Ananthakrishnan, editor. *The biology of gall insects*. Oxford and IBH Publishing Company. New Delhi, India.
- Godfray, H. C. J. 1994. *Parasitoids: behavioral and evolutionary ecology*. Princeton University Press, Princeton, New Jersey, USA.

- Jeffries, M. J. and J. H. Lawton. 1984. Enemy free space and the structure of ecological communities. *Biological Journal of the Linnean Society* 23:269-286.
- Jones, D. 1983. The influence of host density and gall shape on the survivorship of *Diastrophus kincaidii* Gill (Hymenoptera:Cynipidae). *Canadian Journal of Zoology* 61:2138-2142.
- Krebs, C. J. 1994. *Ecology: The experimental analysis of distribution and abundance*. HarperCollins College Publishers, New York, New York, USA.
- Lund, J. N., Ott, J. R. and R. J. Lyon. 1998. Heterogony in *Belonocnema treatae* Mayr (Hymenoptera:Cynipidae). *Proceedings of the Entomological Society of Washington*. In Press.
- Mayr, G. 1881. Zwanzig. Jahresber. Comm.- Oberrealsch. I. Bez. Wien., pp. 4, 16, 38.
- Meyer, J. 1987. *Plant galls and gall inducers*. Gebruder Borntraeger. Berlin, Germany.
- Nixon, K. C. 1984. A biosystematic study of *Quercus* series *Virentes* (the live oaks) with phylogenetic analyses of Fagales, Fagaceae and *Quercus*. Dissertation. University of Texas, Austin, Texas, USA.
- Price, P. W., Westoby, M., Rice, B., Atsatt, P. R., Fritz, R. S., Thompson, J. N., and K. Mobley. 1986. Parasite mediation in ecological interactions. *Annual Review of Ecological Systems* 17:487-505.
- Price, P. W., and K. M. Clancy. 1986. Interactions among three trophic levels: Gall size and parasitoid attack. *Ecology* 67:1593-1600.
- Rey, L. A. 1992. Developmental morphology of two types of hymenopterous galls. Pages 87-101 in Joseph D. Shorthouse and Odette Rohfritsch, editors. *Biology of insect-induced galls*. Oxford University Press. New York, New York, USA.

- Rohfritsch, O. 1992. Patterns in gall development. Pages 60-86 in Joseph D. Shorthouse and Odette Rohfritsch, editors. Biology of insect-induced galls. Oxford University Press. New York, New York, USA.
- Roskam, J. C. 1992. Evolution of the gall-inducing guild. Pages 34-47 in Joseph D. Shorthouse and Odette Rohfritsch, editors. Biology of insect-induced galls. Oxford University Press. New York, New York, USA.
- Washburn, J. D. and H. V. Cornell. 1979. Chalcid parasitoid attack on a gall wasp population (*Acraspis hirta* (Hymenoptera:Cynipidae)) on *Quercus prinus* (Fagaceae). Canadian Entomologist 111:391-400.
- Washburn, J. O. and H. V. Cornell. 1981. Parasitoids, patches and phenology: their probable role in the extinction of a cynipid gall wasp population. Ecology 62:1597-1607.
- Weld, L. H. 1921. American gallflies of the family Cynipidae producing subterranean galls on oak. US. National Museum Proceedings 59:238-9, 241-2.
- Wiebes-Rijks, A. A. and J. D. Shorthouse. 1992. Ecological relationships of insects inhabiting cynipid galls. Pages 238-257 in Joseph D. Shorthouse and Odette Rohfritsch, editors. Biology of insect-induced galls. Oxford University Press. New York, New York, USA.
- Weis, A. E. 1982. Use of a symbiotic fungus by the gall maker *Asteromyia carbonifera* to inhibit attack by the parasitoid *Torymus capite*. Ecology 63:1602-1605.
- Weis, A. E., Abrahamson, W. G., and K. D. McCrea. 1985. Host gall size and oviposition success by the parasitoid *Eurytoma gigantea*. Ecological Entomology 10:341-348.

Table 1. Cohort analysis for leaf galls of *Belonocnema treatae*. Individual leaves were labeled and the outcome of oviposition by *B. treatae* bisexual females was monitored throughout the period of gall development, from March-October, 1996 (means given as mean + sd). Data on mature galls represents a subset (n = 41) of the original 221 leaves that remained on the tree until date of harvest, October 9-11, 1996.

		<u>Percent</u>	<u>Density</u>
Oviposition ↓ ↓ ↓ ↓	A) # <u>Oviposition scars (n = 2822)</u>		
	Percent leaves with oviposition scars	100	
	Mean oviposition scars per leaf		12.8 ± 4.8 (range 1-26)
Initiation ↓ ↓ ↓ ↓	B) # <u>Successfully initiated galls (n = 181)</u>		
	Percent leaves with galls initiated	41.2	
	Percent successfully established galls	6.4	
	Percent oviposition failure	39.8	
	Mean initiated galls per leaf		2.0 ± 1.6 (range 0-13)
Maturity ↓ ↓ ↓ ↓	C) # <u>mature galls (n = 52)</u>		
	Percent leaves with mature galls	73.2	
	Percent oviposition producing mature galls	9.1	
	Percent established galls that mature	69.3	
	Mean mature galls per leaf		1.3 ± 1.4 (range 0-8)

Table 2. Hymenoptera associated with leaf galls of the cynipid wasp *Belonocnema treatae*. A total of 13 species have been identified, representing 7 families. An additional 10 morphotypes that were collected only rarely from galls are not yet identified.

Inquilines

Synergus sp. (Cynipidae)
Eurytoma furva (Eurytomidae)
Eurytoma sp. (Eurytomidae)
Sycophila flava (Eurytomidae)

Parasitoids

Ormyrus labotus (Ormyridae)
Torymus tubicola (Torymidae)
Acaenacis lausus (Pteromalidae)

Hyperparasitoids

Eurytoma bugbeeii (Eurytomidae)

Association unknown

Allorhogas sp. (Braconidae)
Brasema sp. 1 (Eulophidae)
Brasema sp. 2 (Eulophidae)
Brasema sp. 3 (Eulophidae)
Galeopsomyia sp. (Eulophidae)

Unidentified

10 rare morphotypes

Table 3. Multiple emergence from individual galls (n = 237) of Hymenoptera associated with *Belonocnema treatae* leaf galls. Species listed horizontally emerged first followed by another species, listed vertically. Blanks are not missing data, but interactions that were not observed.

	<u>Synergus sp.</u>	<u>S. flava</u>	<u>Eurytoma</u>	<u>A. lausus</u>	<u>O. labotus</u>	<u>T. tubicola</u>	<u>Allorhogas sp.</u>	<u>Brasema sp. 1</u>	<u>Brasema sp. 2</u>	<u>Brasema sp. 3</u>	<u>Galeopsomyia</u>	<u>B. treatae</u>
Synergus	X	X				X	X	X			X	X
S. flava	X						X					
Eurytoma												
A. lausus												
O. labotus						X						
T. tubicola					X	X				X		
Allorhogas sp							X					
Brasema sp 1												
Brasema sp 2					X	X				X		
Brasema sp 3												
Galeopsomyia										X		
B. treatae												

Table 4. Survivorship of *Belonocnema treatae* from leaf galls. Survivorship was defined as percent of all leaf galls in samples from which unisexual *B. treatae* emerged.

	# <i>B. treatae</i>	# galls	Survivorship
<u>1995/96</u>	5	1305	
	0	220	
	7	1133	
Total	12	2658	0.45%
<u>1996/97</u>	1	245	
	1	204	
	2	162	
	1	266	
	1	244	
	1	192	
	4	222	
	1	172	
	5	258	
Total	17	1965	0.86%

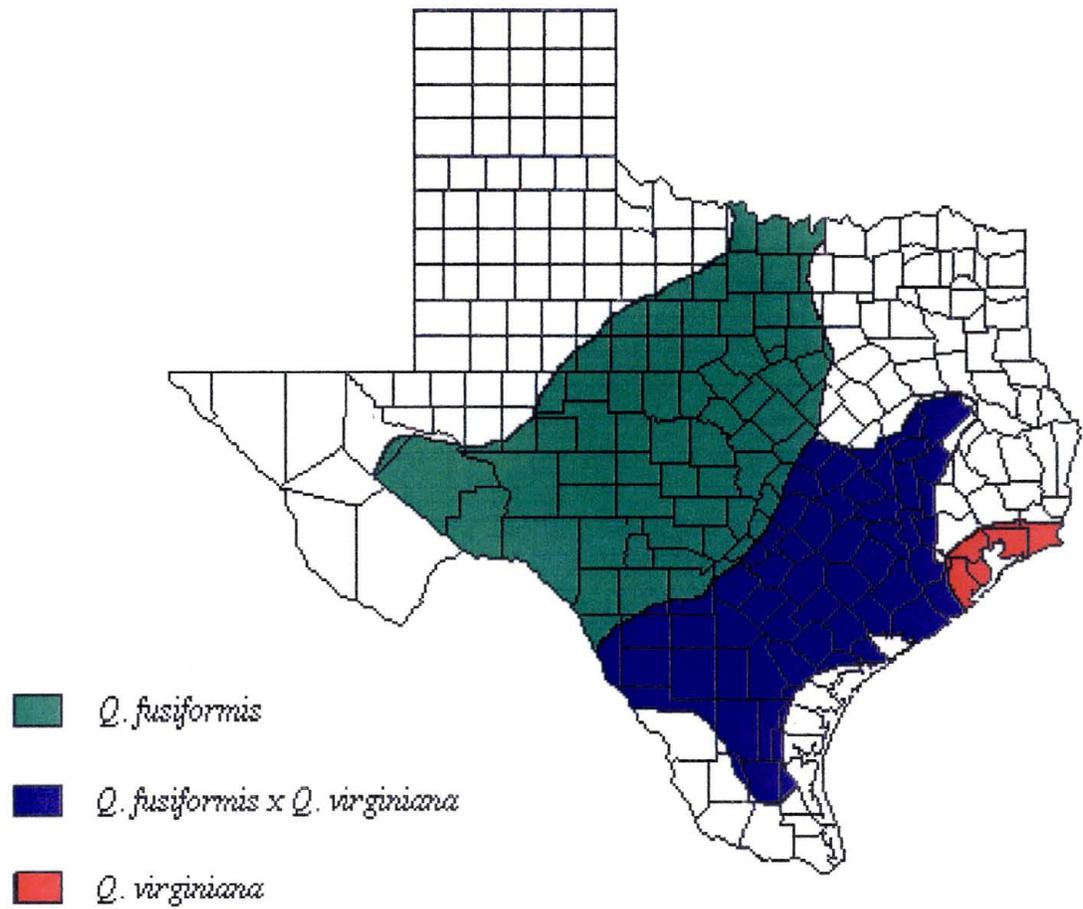


Figure 1. Distributions of *Quercus fusiformis*, *Q. virginiana* and their hybrids in Texas.

Unisexual generation

Bisexual generation

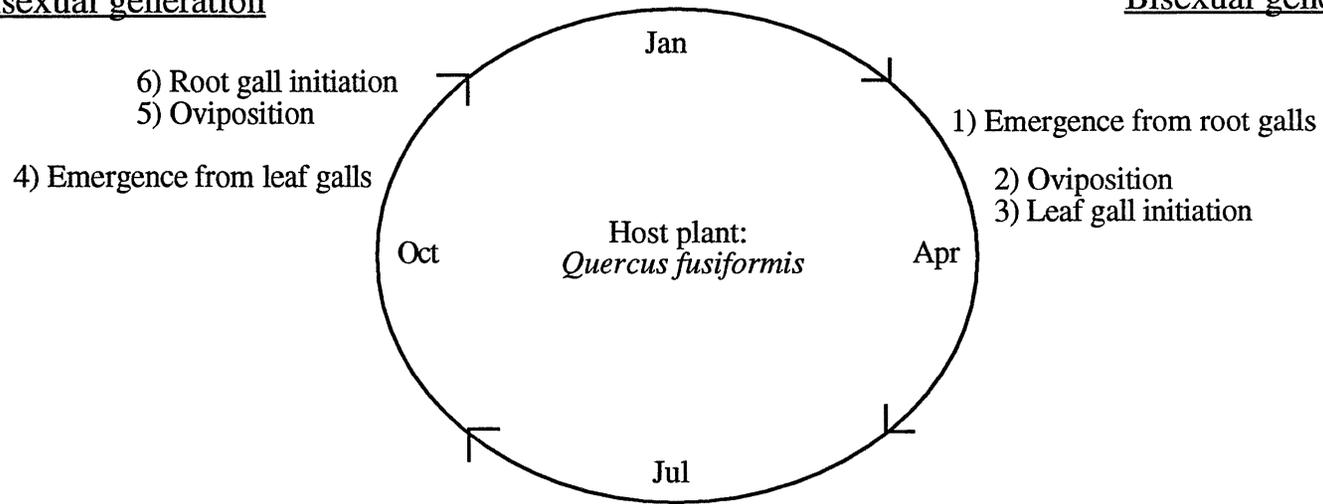


Figure 2. Life cycle of *Belonocnema treatae* Mayr, Freeman Ranch, Hays County, Texas.

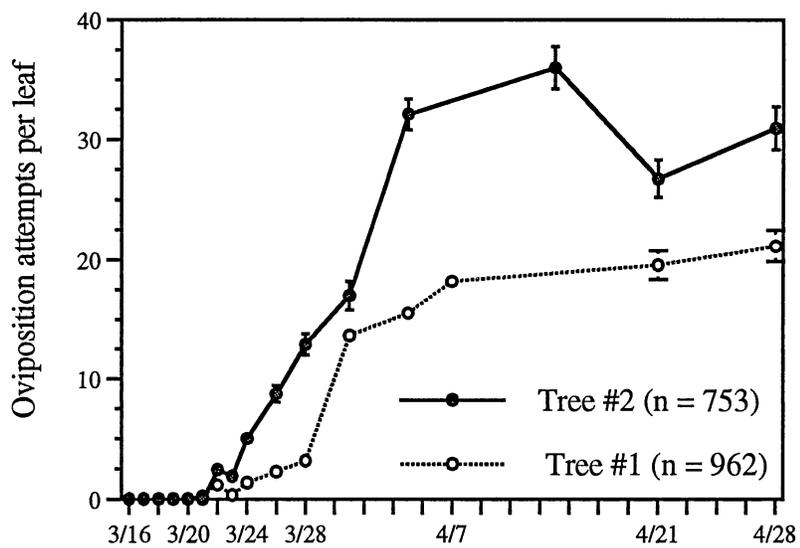


Figure 3. Phenology of oviposition attempts by *Belonocnema treatae* (bisexual generation) for two trees during spring 1997. Mean leaves per sample for tree #1 is 87.45 (range 47-120) and 75.3 for tree #2 (range 41-167).

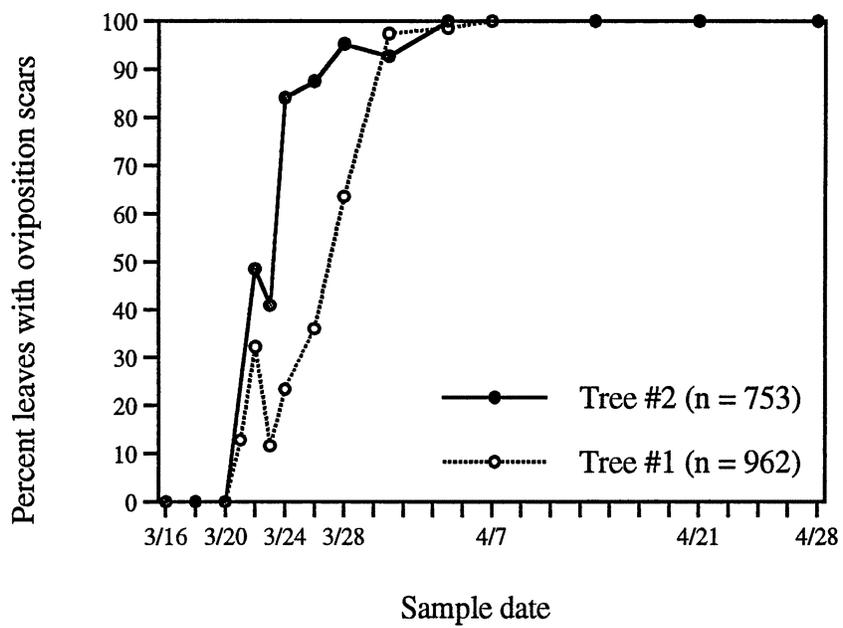


Figure 4. Percent leaves with oviposition scars by sample date for two heavily galled trees.



Figure 5. Leaf undersurface showing oviposition scars of bisexual females located almost exclusively on lateral veins. As development proceeds, galls become white and globular.

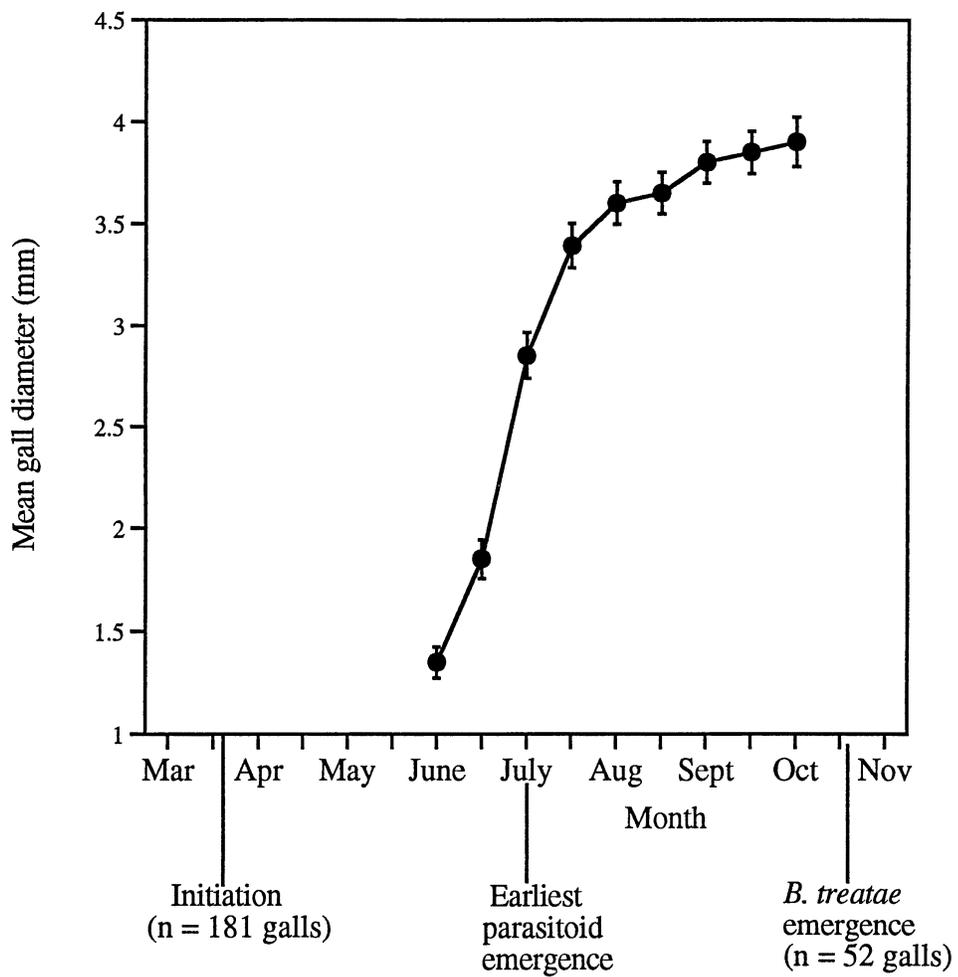


Figure 6. Phenology of gall development for *Belonocnema treatae* leaf galls documented in 1996 for tree #1. Standard errors are shown.

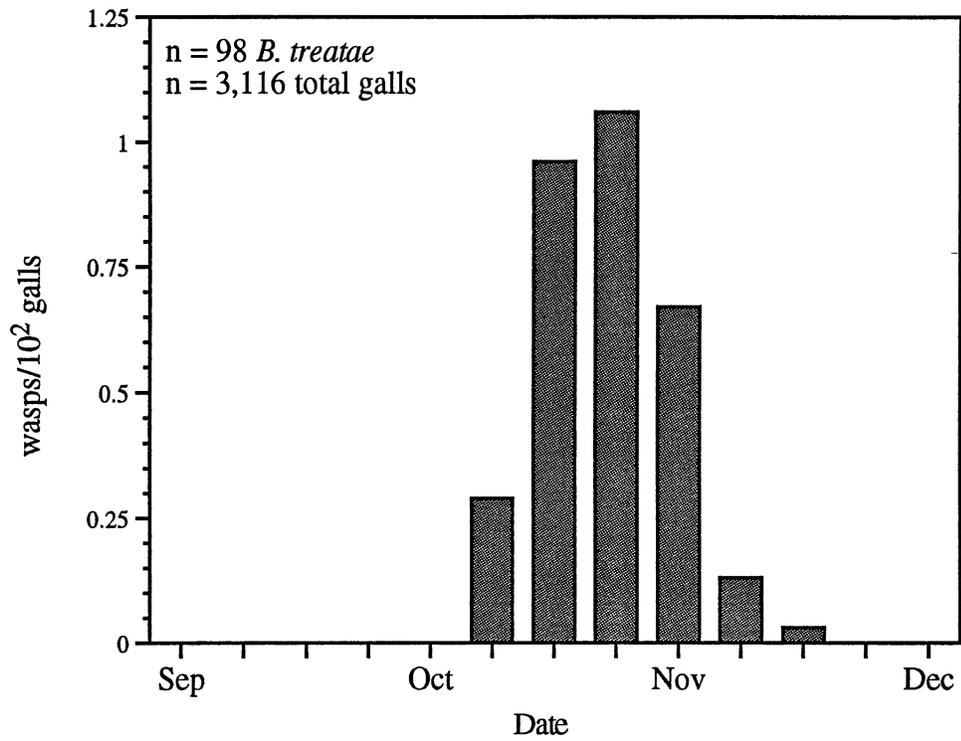


Figure 7. Emergence phenology of unisexual *Belonocnema treatae* in Fall, 1996. The proportion of leaf galls isolated in gelatin capsules from which *B. treatae* emerged (wasps/100 galls) is shown. Of 3,116 galls, 98 produced *B. treatae*.

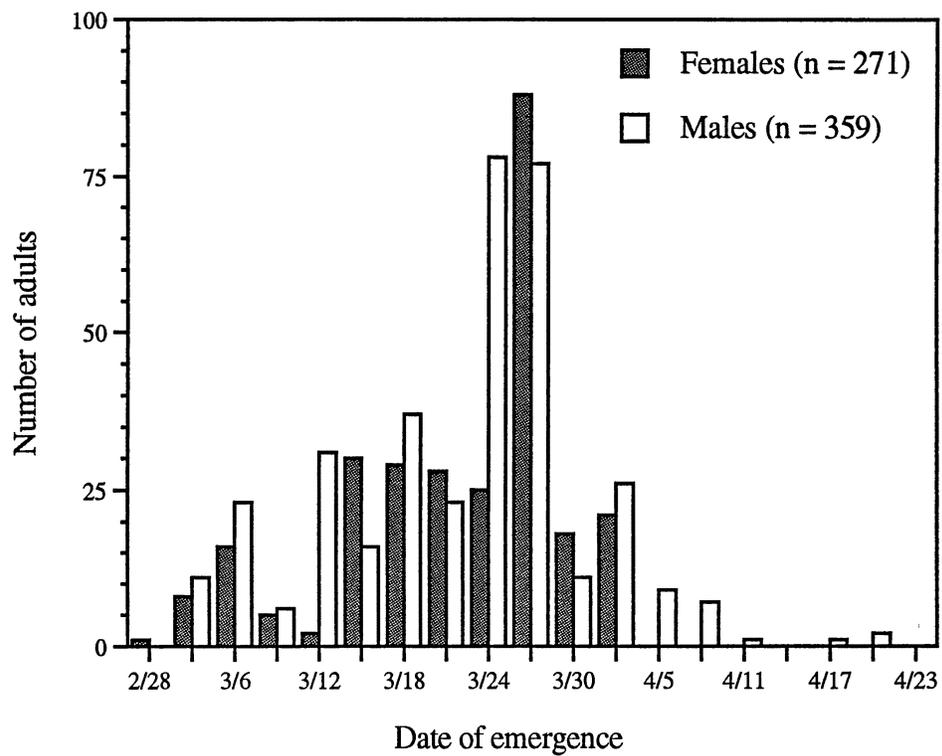


Figure 8. Emergence phenology of *Belonocnema treatae* bisexual generation collected from root galls in laboratory held collection traps, Spring 1997. Males emerged during a broader window of time than did females, and males greatly outnumbered females.

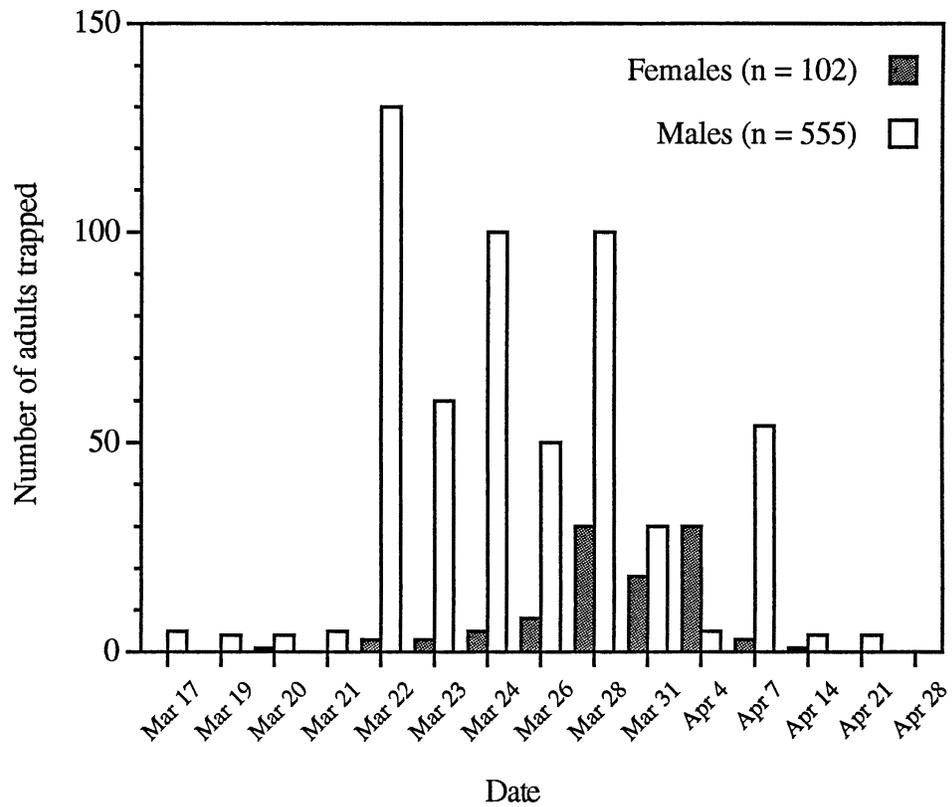


Figure 9. *Belonocnema treatae* bisexual generation collected via malaise trap (Spring 1997). Trap was placed under a tree that was heavily-galled the previous season. The gall maker was present in trapfall during a narrow window from mid March through mid April.

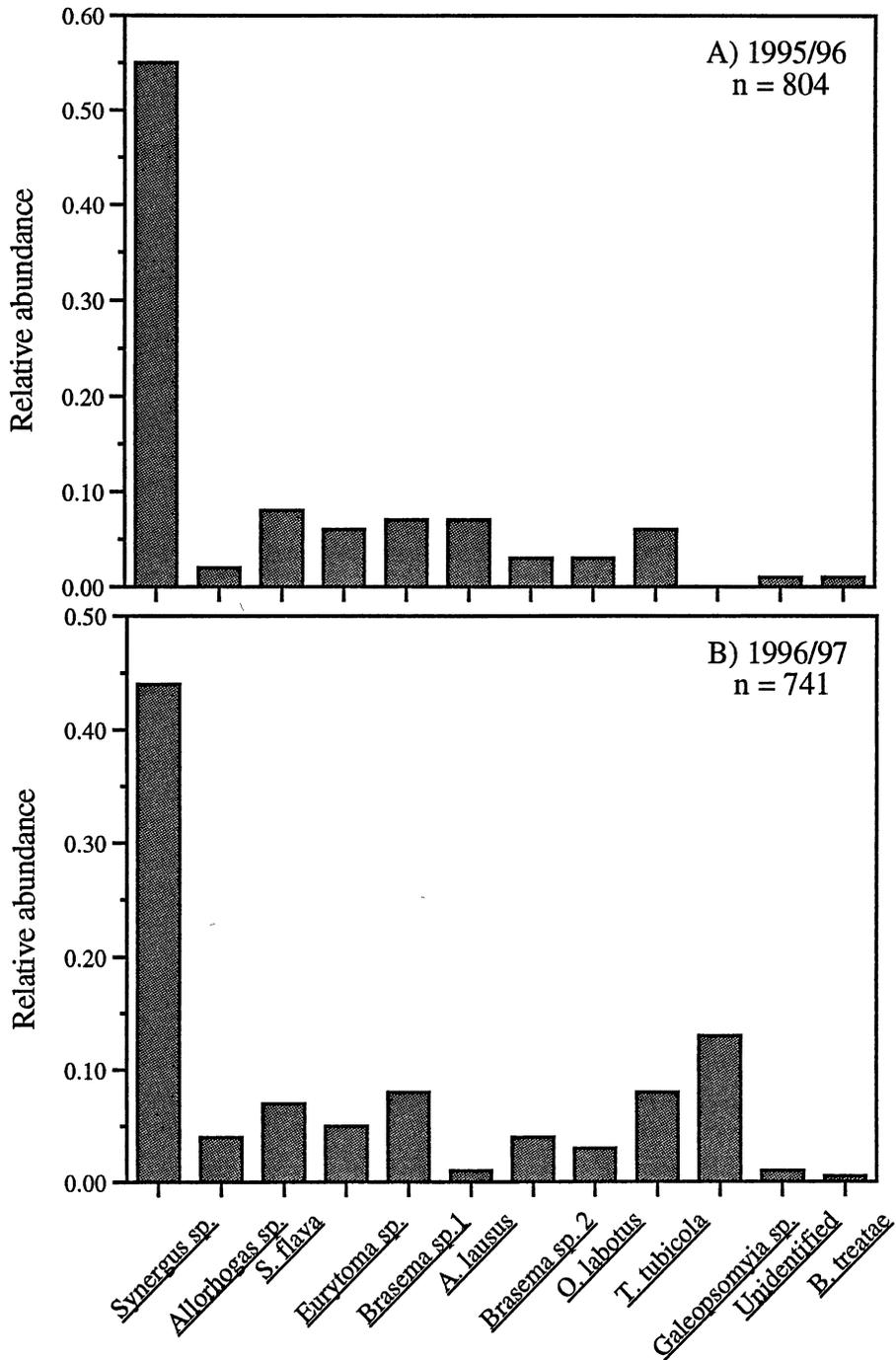


Figure 10. Relative abundance of identified Hymenoptera emerging from *Belonocnema treatae* leaf galls A) 1995/96 season and B) 1996/97 season. Relative abundance is defined as the percent of wasps emerging by species (n = total emergents).

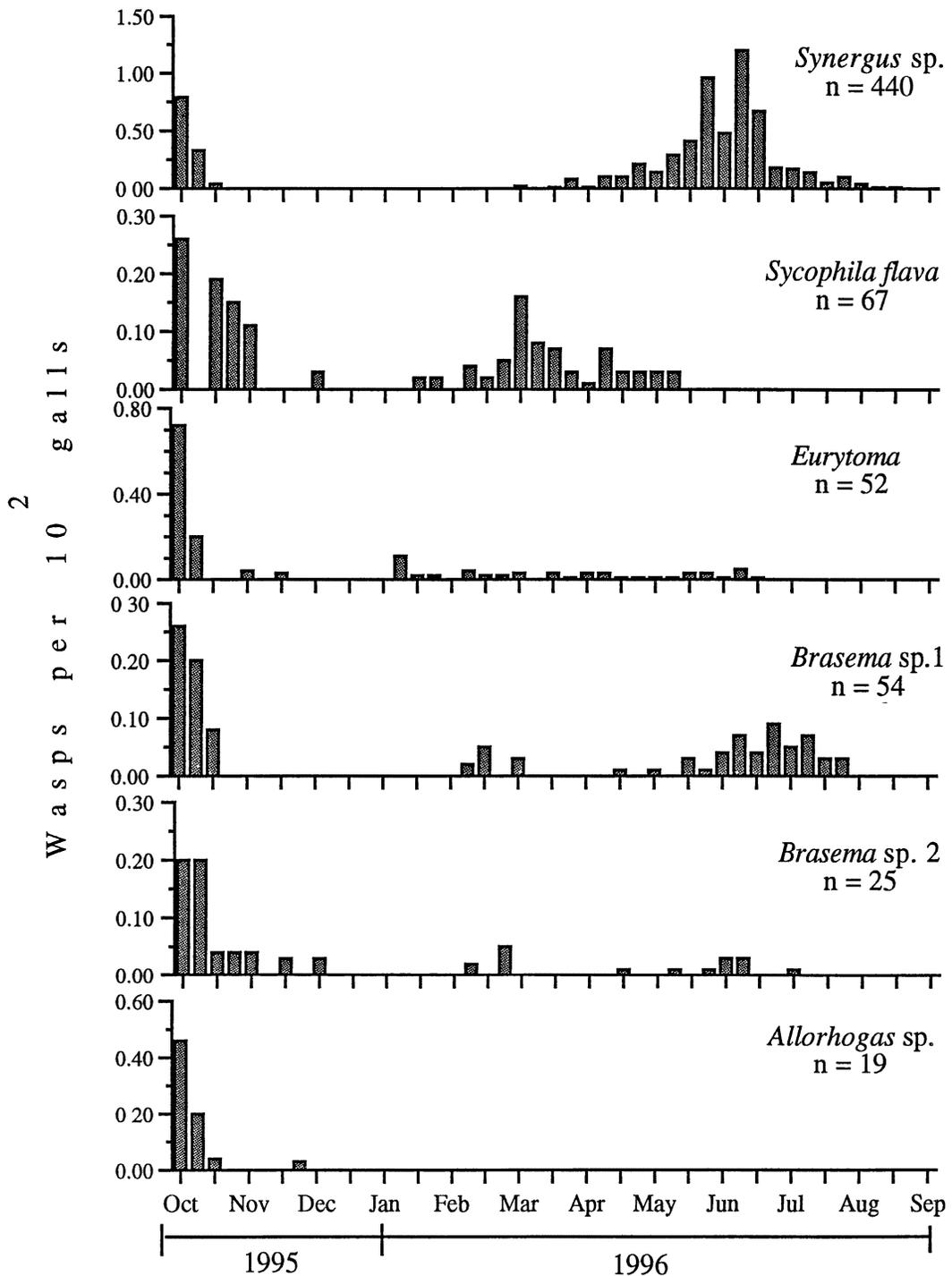


Figure 11. Emergence phenology for wasp species associated with *Belonocnema treatae* leaf galls in 1995/96. The Y axis shows relative abundance, computed weekly as wasps per 100 galls. *Synergus* sp. and *S. flava* are known inquiline. *Eurytoma* sp. includes *E. furva*, *Eurytoma* sp.(inquiline) and *E. bugbeeii* (hyperparasitoid). Trophic relationships of *Brasema* sp. 1 and 2 and *Allorhogas* sp. to *B. treatae* are unknown (n = total emergents).

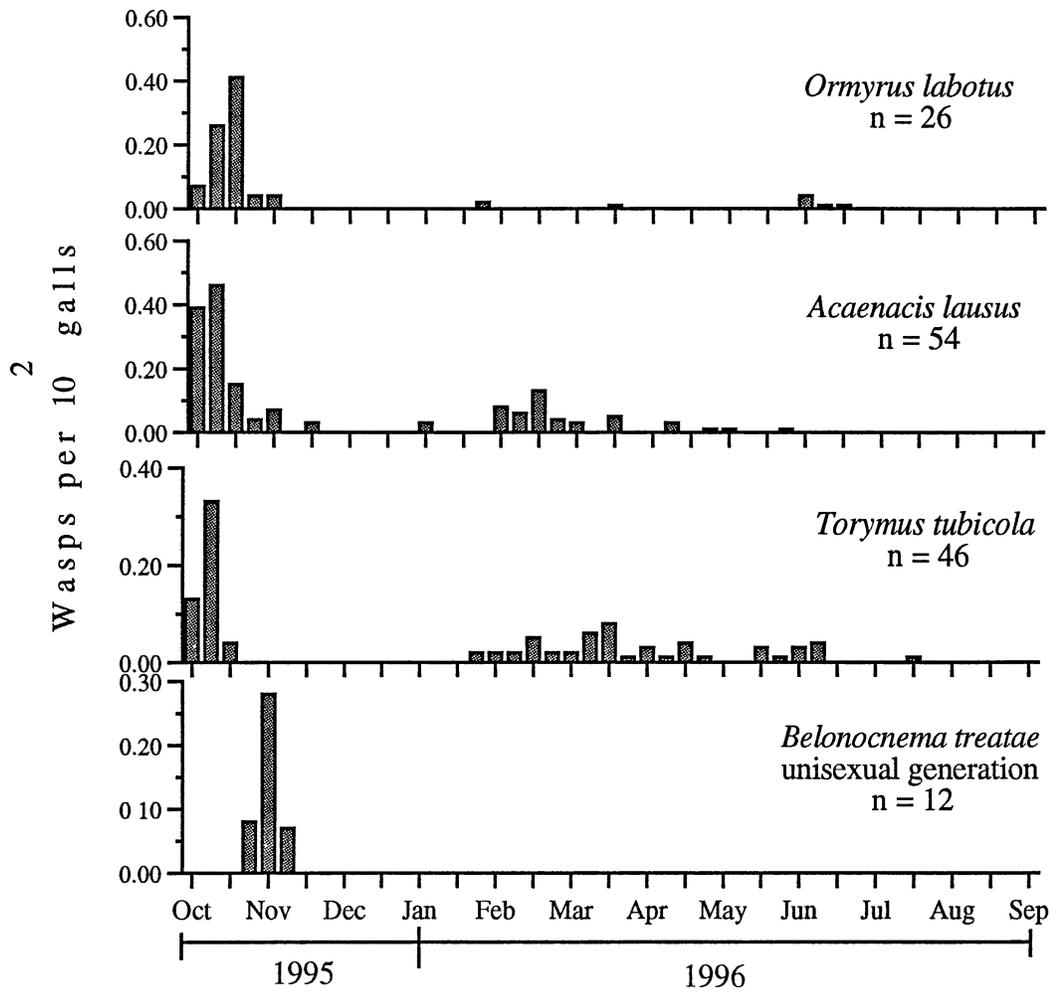


Figure 12. Emergence phenology for wasp species associated with *Belonocnema treatae* leaf galls in 1995/96. The Y axis shows relative abundance, computed weekly as wasps per 100 galls. *Ormyrus labotus*, *Acaenacis lausus*, and *Torymus tubicola* are known parasitoids, while *Belonocnema treatae* is the gall maker (n = total emergents).

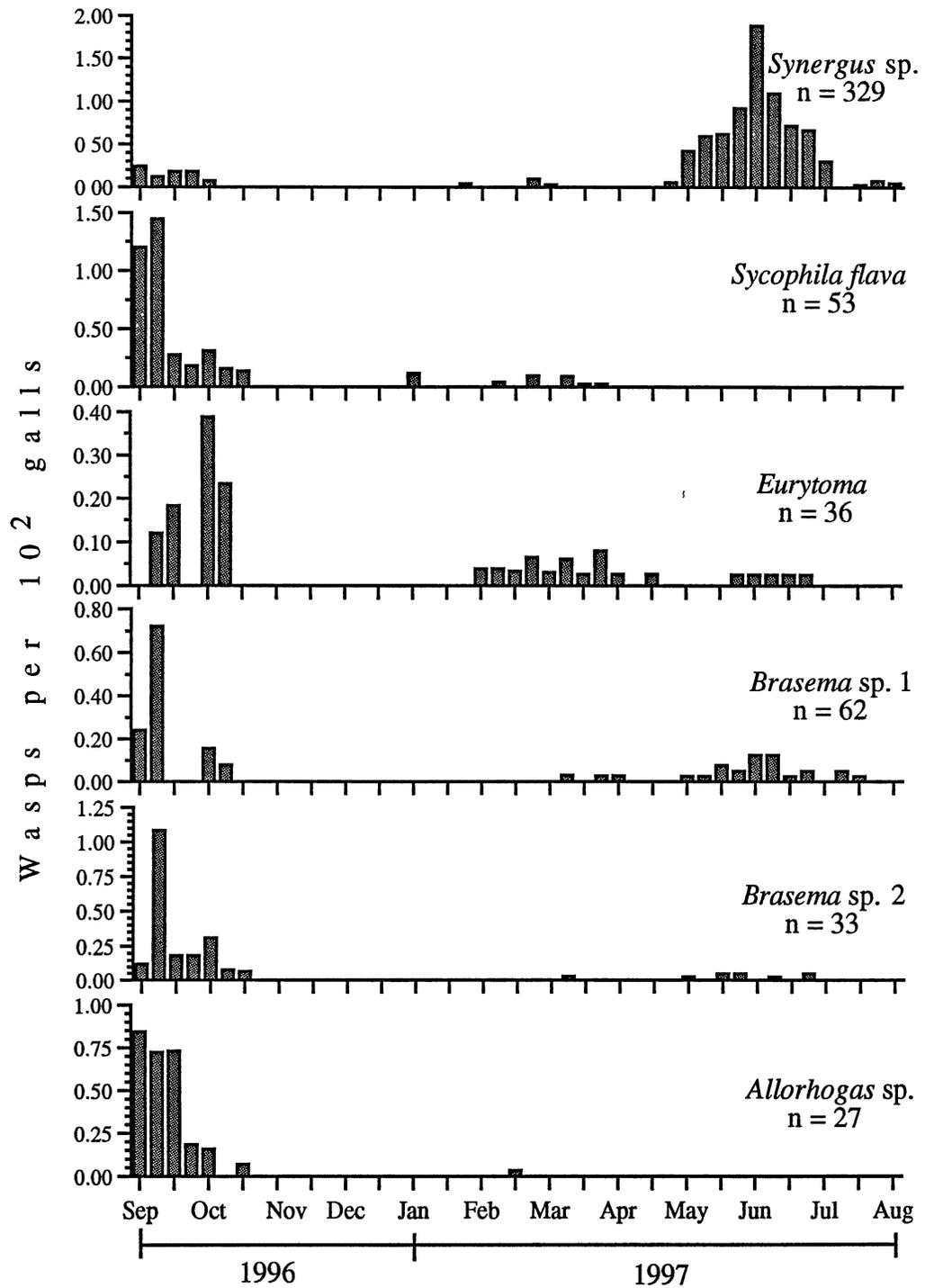


Figure 13. Emergence phenology for wasp species associated with *Belonocnema treatae* leaf galls in 1996/97. The Y axis shows relative abundance, computed weekly as wasps per 100 galls. *Synergus* sp. and *S. flava* are known inquiline. *Eurytoma* sp. includes *E. furva*, *Eurytoma* sp.(inquiline) and *E. bugbeeii* (hyperparasitoid). Trophic relationships of *Brasema* sp. 1 and 2 and *Allorhogas* sp. to *B. treatae* are unknown (n = total emergents).

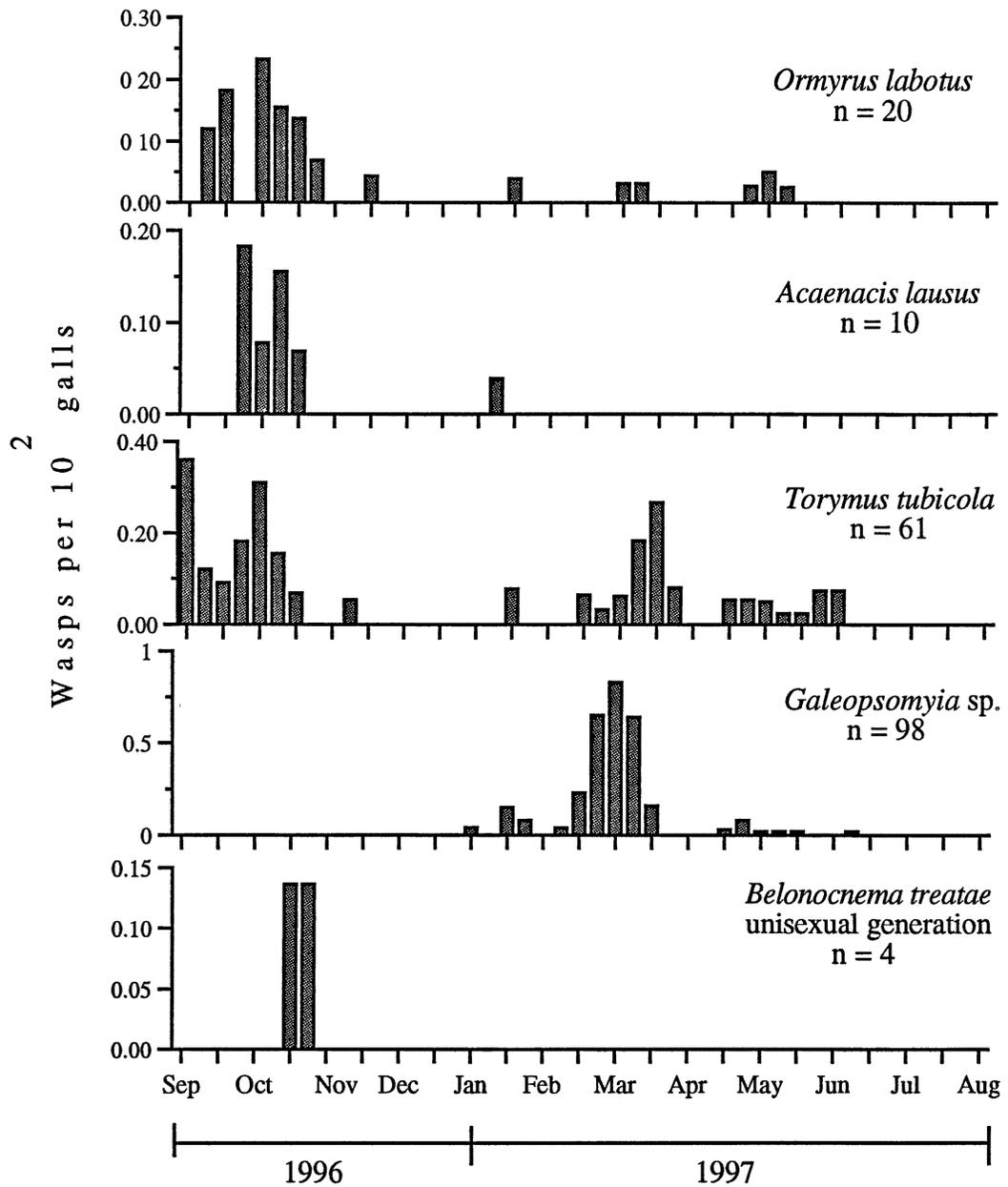


Figure 14. Emergence phenology for wasp species associated with *Belonocnema treatae* leaf galls in 1996/97. The Y axis shows relative abundance, computed weekly as wasps per 100 galls. *Ormyrus labotus*, *Acaenacis lausus*, and *Torymus tubicola* are known parasitoids, while *Belonocnema treatae* is the gall maker. The trophic status of *Galeopsomyia* sp. is unknown (n = total emergents).

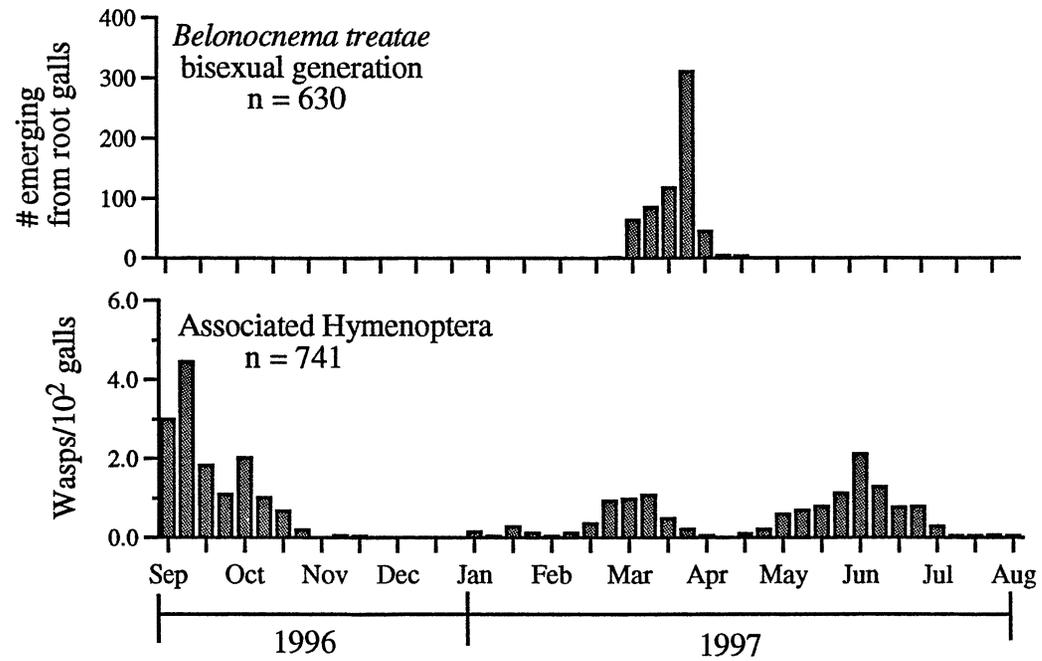


Figure 15. Emergence of the bisexual generation, immediately preceding leaf gall initiation, in relation to timing of emergence of natural enemies of *Belonocnema treatae* (n = total emergents).

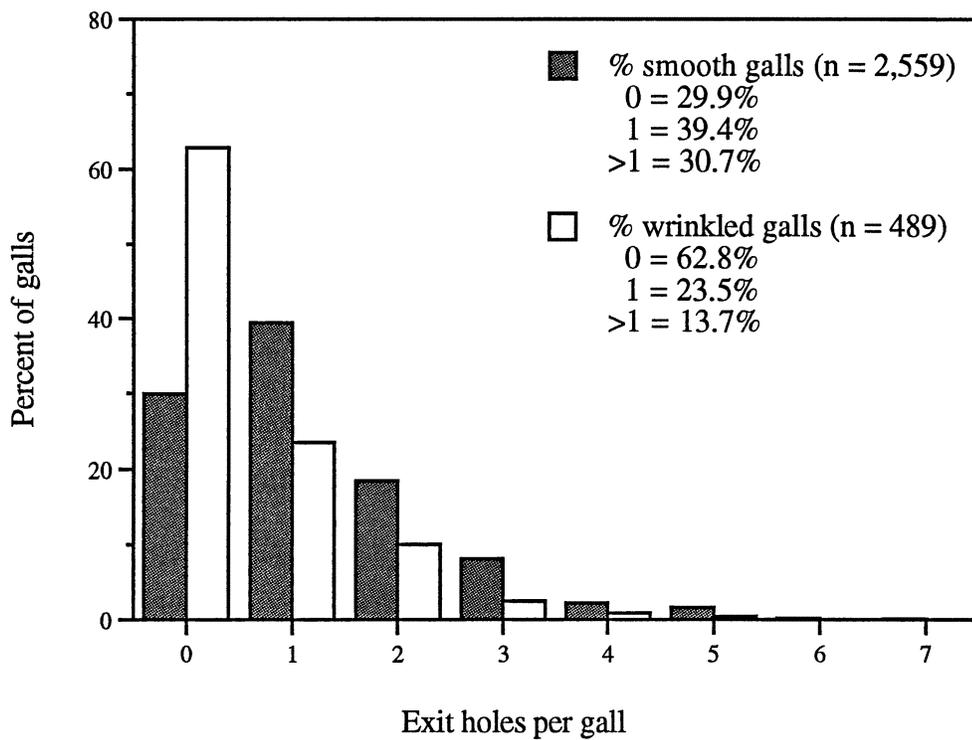


Figure 16. Frequency distribution of exit holes among smooth galls and wrinkled galls. Number of exit holes per gall was used to estimate gall productivity. Gall samples were collected from mixed tree sources from Fall 1995 through Spring 1996.

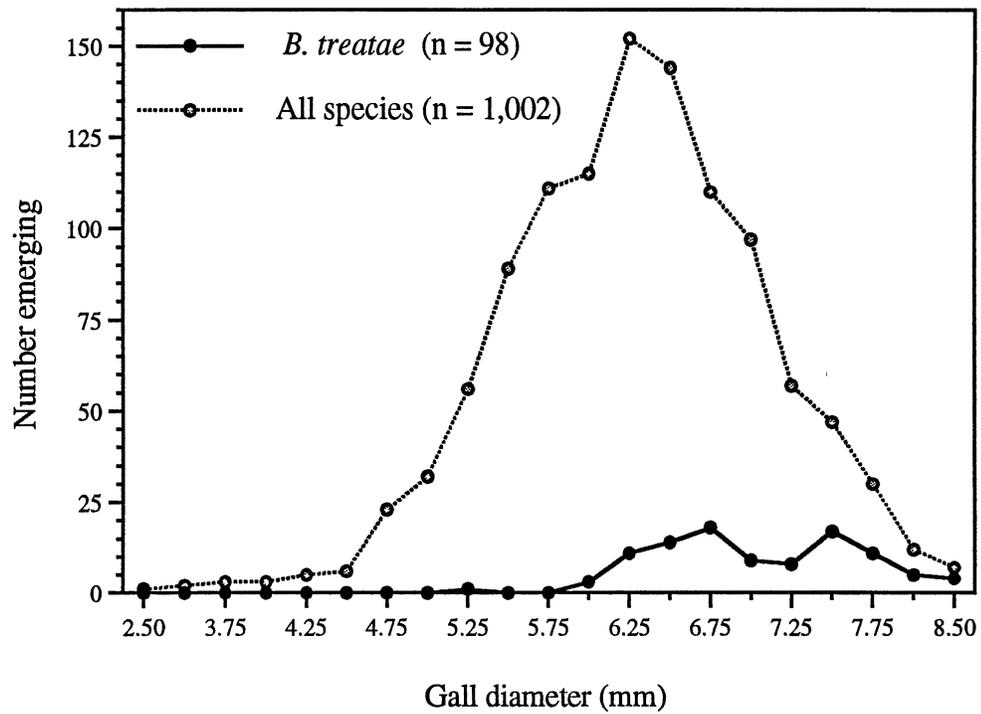


Figure 17. Size distribution for leaf galls from which all Hymenoptera emerged. Mean diameter (\pm se) is 6.07 ± 0.02 mm for all galls, and 6.94 ± 0.06 mm for those which produced *B. treatae*. Among galls producing the gall maker, 95% had a diameter larger than 6.1 mm.

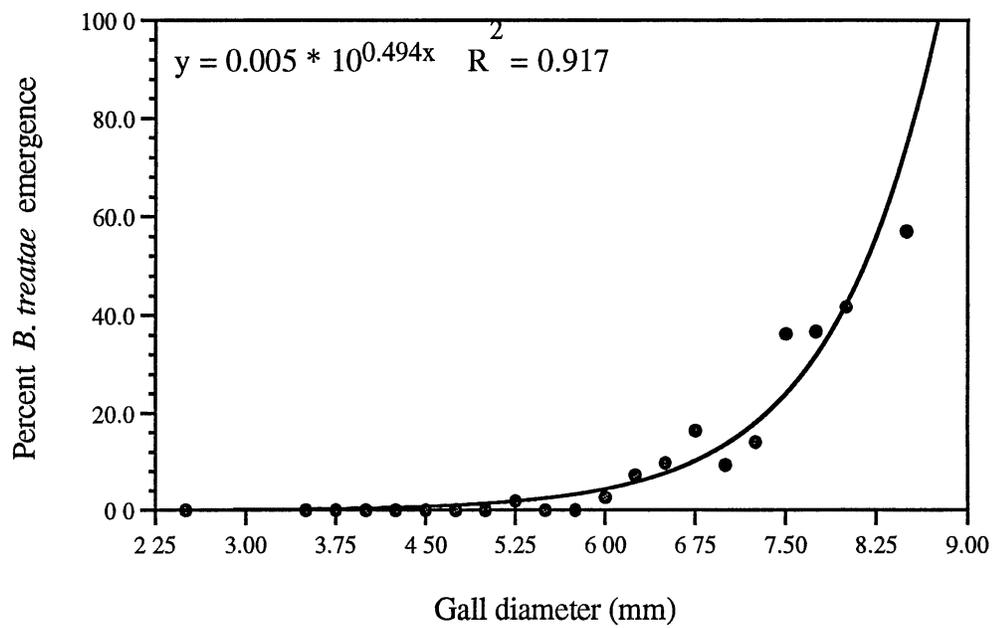


Figure 18. Percent emergence of *Belonocnema treatae* unisexual females by gall size class. As gall size increases, the probability of *B. treatae* emergence increases exponentially.

APPENDIX

The Hymenopteran Community Collection

Specimens collected from the leaf and root galls of *Belonocnema treatae* have been grouped into three collections:

1) USDA Systematic Entomology Laboratory (S. E. L.) reference collection: Includes specimens identified by S. E. L. staff. These vials are to remain sealed. No specimens may be removed or added to vials under any circumstances.

2) Southwest Texas State University (SWTSU) Laboratory reference collection: Includes a subset of specimens which were identified by S. E. L. staff. These vials may be used by local laboratory personnel as an aid to identification. No specimens may be removed or added without the consent of James R. Ott.

3) Working collection: Includes specimens that were identified by local laboratory personnel. Specimens in these vials may be added to and used as needed.

List of Hymenoptera Commonly Collected from the Leaf and
Root Galls of *Belonocnema treatae*

Cynipidae

Belonocnema treatae Mayr (unisexual generation)

Belonocnema treatae Mayr (bisexual generation)

Synergus sp.

Braconidae

Allorhogas sp.

Eulophidae

Brasema sp. 1

Brasema sp. 2

Brasema sp. 3

Galeopsomyia sp.

Ormyridae

Ormyrus labotus Walker

Torymidae

Torymus tubicola (Osten Sacken)

Pteromalidae

Acaenacis lausus (Walker)

Eurytomidae

Sycophila flava (Ashmead)

Eurytoma sp.

Eurytoma furva Bugbee

Eurytoma bugbeei Grissell

Key for the Identification of Hymenoptera
Associated with *Belonocnema treatae* Galls on Live Oak

- 1. Wing venation distinct; prepectus lacking.....2
- 1'. Wing venation greatly reduced; prepectus between base of forewing and pronotum.....5

- 2(1). Scutellum with medial oval or rounded keel dorsally; ovipositor not exerted.....3
- 2'. Scutellum without keel; ovipositor permanently exerted.....
.....*Allorhogas* sp., page 6

- 3(2). Antennae 14-segmented in female, 15-segmented in male.....4
- 3'. Antennae 13-segmented.....
.....*Belonocnema treatae* (unisexual generation), page 5

- 4(3). Body large, 3.4-3.75 mm in length, amber in color; legs brown.....
.....*Belonocnema treatae* (bisexual generation), page 6
- 4'. Body 2-3 mm in length, brown in color; legs yellowish.....
.....*Synergus* sp., page 6

- 5(1'). Hindcoxa much wider and longer than forecoxa.....6
- 5'. Hindcoxa nearly the same size as forecoxa.....7

- 6(5). Abdomen with rows of pits on dorsal surface; parapsidal sutures absent or indistinct; ovipositor not exerted.....*Ormyrus labotus*, page 9
- 6'. Abdomen without pits; distinct parapsidal sutures; ovipositor permanently exerted.....*Torymus tubicola*, page 9

7(5').	Tarsi 5-segmented.....	11
7'.	Tarsi 4-segmented.....	8
8(7').	Antennae 13-segmented.....	9
8'.	Antennae 8- to 11-segmented.....	10
9(8).	Body yellow.....	<i>Brasema</i> sp. 1, page 7
9'.	Body black.....	<i>Brasema</i> sp. 3, page 8
10(8').	Antennae 11-segmented; scutellum undivided; body small (≤ 1 mm in length).....	<i>Brasema</i> sp. 2, page 7
10'.	Antennae 8-segmented in female, 9-segmented in male; scutellum divided into 3 distinct regions; body length 2-3 mm	<i>Galeopsomyia</i> sp., page 8
11(7).	Pronotum visible as broad rectangle dorsally; body yellow or dark brown; abdomen round, shining.....	12
11'.	Pronotum weakly visible dorsally, body black, metallic; abdomen elongate, dusky.....	<i>Acaenacis lausus</i> , page 10
12(11).	Yellow body; orange eyes.....	<i>Sycophila flava</i> , page 11
12'.	Dark brown body, red eyes.....	13
13(12').	Antennae 8- to 9-segmented; two terminal segments fused.....	14
13'.	Antennae 10-segmented in female, 9-segmented in male; three terminal segments fused.....	<i>Eurytoma bugbeei</i> , page 12
14(13).	Legs yellowish; antennae 8-segmented.....	<i>Eurytoma furva</i> , page 11
14'.	Legs white; antennae 9-segmented.....	<i>Eurytoma</i> sp., page 11

Description of Hymenoptera Associated with
Belonocnema treatae Galls on Live Oak

Descriptions of the Families Cynipidae, Braconidae, and Eulophidae are after Borror, D. J., D. M. De Long, and C. A. Triplehorn. 1981. An introduction to the study of insects. Saunders College Publishing, New York, New York, USA. Descriptions of the Families Ormyridae, Torymidae, Pteromalidae, and Eurytomidae are after Grissell, E. E., and M. E. Schauff. 1990. A handbook of the families of nearctic Chalcidoidea (Hymenoptera). The Entomological Society of Washington, Washington, D.C., USA.

Family Cynipidae: Scutellum with medial oval or rounded keel dorsally; second abdominal tergum longer than third; antennae filiform, 11- to 16-segmented, usually 13-segmented in female and 15-segmented in male; wing venation distinct .

Subfamily Cynipinae: Small to minute; brown to black, never metallic; abdomen oval, somewhat compressed laterally, shining, second tergum covers half or more of the abdomen.

Belonocnema treatae Mayr (unisexual generation)

General appearance: Body brown, abdomen is larger than thorax

Description: Antennae 13-segmented; wings large, hyaline, venation pattern intricate; head medium brown with brown eyes; thorax medium brown with keel; abdomen amber and translucent, glassy, laterally compressed, eggs visible; ovipositor sheath extends length of abdomen, tip exserted; gall-inducer (root galls); common in fall.

Note: Females only. Wasps emerge from leaf galls from mid October-early November.

Belonocnema treatae Mayr(bisexual generation)

General appearance: Body large (3-4 mm long), amber

Description: Antennae 14-segmented in female, 15-segmented in male, as long as body length; wings large, wing venation pattern very pronounced; head amber with brown eyes, appears disproportionately small for body size; thorax amber, keeled, posterior end usually dark brown; abdomen dark brown, glassy, translucent, usually amber at base of propodeum, laterally compressed; female has very round abdomen with eggs visible, reduced and narrower in males; ovipositor sheath runs length of abdomen, tip sometimes exerted; gall-inducer (leaf galls); very common in March.

Note: Emerge from root galls from mid March-early April

Synergus sp.

General appearance: Body brown ; distal abdomen usually amber; legs yellow
Antennae 14-segmented in female, 15-segmented in male; wings setose, hyaline, wing venation discernible; head usually brown, sometimes amber, eyes brown; thorax usually brown, sometimes amber, keeled; abdomen brown, amber at distal tip, laterally compressed, glassy; ovipositor retracted into sheath; inquiline; very common.

Family Braconidae: Antennae filiform, usually at least 16-segmented; pronotum somewhat triangular laterally; wing venation distinct , one recurrent vein or none, costal cell absent; abdomen not greatly elongate, about as long as the head and thorax combined, not petiolate, or if petiolate, petiole is not curved or expanded apically; propodeum not prolonged behind hind coxae; hind trochanters 2-segmented; ovipositor arising anterior to the tip of the abdomen and permanently exerted; mostly small insects, rarely over 15 mm in length.

Allorhogas sp.

General appearance: Body yellow; legs yellow

Description: Antennae longer than body, 21-segmented in both female and male; wing vein pattern distinct, with stigma; head yellow; eyes brown, metallic; thorax yellow; abdomen yellow, curved, paler than head and thorax; ovipositor dark brown, same length as abdomen, permanently exerted; life habit unknown; common in fall.

Note: Identified by Paul Marsh, N. Newton, KS, USA.

Family Eulophidae: Antennae elbowed, generally 9-segmented or fewer, pectinate in males of many species; axillae extend forward beyond the tegulae; apical spur of front tibia small and straight; tarsi four-segmented; small insects, 1-3 mm in length; many have a brilliant metallic color; parasitoids and hyperparasitoids.

Brasema sp. 1

General appearance: Body yellow; head black

Description: Antennae 13-segmented, elbowed, dark, short, club-shaped, 3 terminal segments fused; wings have small dark spot near costal margin; head black with orange metallic sheen, eyes large, brown; thorax yellow, shorter than abdomen; abdomen yellow, elongate, with prominent spiracles on posteriolateral tip; midtarsal combs black; ovipositor robust, sheath runs length of abdomen, tip exerted; life habit unknown; very common.

Note: Collections contain females only

Brasema sp. 2

General appearance: Body black with orange metallic sheen, very small

Description: Antennae 11-segmented, club-shaped, slightly longer than head; wings have small dark spot near costal margin; head black, metallic, eyes brown; thorax black metallic, equal to length of abdomen; abdomen black metallic, translucent, segmentation apparent; midtibial spur long; hyperparasitoid; common.

Note: Collection contains males only

Brasema sp. 3

General appearance: Body black with orange metallic sheen

Description: Antennae 13-segmented (difficult to discern), elbowed, dark, short, club-shaped, 3 terminal segments fused; wings have dark anterior margin, small dark spot near costal margin; head black with orange metallic sheen, eyes large, brown; thorax black with orange metallic sheen, shorter than abdomen; abdomen black with orange metallic sheen, elongate, dorsoventrally compressed; midtarsal combs black; ovipositor robust, sheath runs length of abdomen, tip exerted; life habit unknown; not common.

Note: Collections contain females only

Galeopsomyia sp.

General appearance: Body black with slight metallic sheen, eyes red-orange, legs white

Description: Antennae 8-segmented in female, 9-segmented and pectinate in male, elbowed, 3 terminal segments fused; wings hyaline, small dark spot near costal margin (more prominent in males); head black with slight metallic sheen, slightly corrugated; thorax black with slight metallic sheen, slightly corrugated, scutellum divided into three distinct regions; abdomen black, oval, usually having apparent

segmentation, larger in female, slightly dorsoventrally flattened; ovipositor retracted; life habit unknown; uncommon in 1995-96 season, very common in 1996-97 season.

Family Ormyridae: Hindfemur not enlarged, hindtibia straight; antennae 5- to 7-segmented between pedicel and club; tarsi normally 5-segmented; midtibial spur thin and spinelike; head projecting downward; hindtibia at least as long as femur, usually longer, same in male and female; scutellum without submedian grooves; hindcoxa much longer and wider than forecoxa; in female, ovipositor not exerted; both sexes: abdomen with pits; hindtibial spurs thickened.

Ormyrus labotus Walker

General appearance: Body metallic, blue/black, legs yellow, eyes red-orange

Description: Antennae 11-segmented in female, 10-segmented in male, elbowed, club-shaped, short, first segment is four times the length of second segment; wings have small dark spot near costal margin; head metallic, blue/black, eyes red-orange; thorax metallic, blue/black; abdomen conical and elongate in female, oval in male, both sexes have rows of pits on dorsal surface; ovipositor robust, tip may be exerted; parasitoid; common.

Family Torymidae: Hindfemur not enlarged, hindtibia straight; antennae 5- to 7-segmented between pedicel and club; tarsae normally 5-segmented; midtibial spur thin and spinelike; head projecting downward; hindtibia at least as long as femur, usually longer, same in male and female; scutellum without submedian grooves; hindcoxa much longer and wider than forecoxa; in female, ovipositor strongly exerted; both sexes: abdomen without pits; hindtibial spurs thin.

Torymus tubicola (Osten Sacken)

General appearance: Body metallic, blue-black, eyes red-orange

Description: Antennae 12-segmented, dark, club-shaped, 3 terminal segments fused; wings have small dark spot near costal margin; head metallic, blue-black, eyes red-orange; thorax metallic, blue-black, parapsidal sutures distinct; abdomen metallic, blue-black, not pitted; ovipositor permanently exerted, as long as body length, black; parasitoid; very common.

Family Pteromalidae: Hindfemur not enlarged, hindtibia straight; antennae 5- to 7-segmented between pedicel and club; tarsae normally 5-segmented; midtibial spur thin and spinelike; head projecting downward; hindtibia at least as long as femur, usually longer, same in male and female; scutellum without submedian grooves; hindcoxa nearly the same size as forecoxa; body often metallic blue or green, rarely black; pronotum visible in dorsal view; abdomen usually elongate and petiole usually not visible; abdomen with 3 or more readily visible terga; body usually elongate.

Acaenacis lausus (Walker)

General appearance: Body black, eyes red-orange

Description: Antennae 9-segmented, terminal 2 segments fused, elbowed, antenna length is twice head length; wings hyaline, small spot near costal margin; head black with metallic sheen; thorax black with metallic sheen, slightly corrugated; abdomen black with metallic sheen, elongate, pointed and dark in female, oval and translucent with apparent segmentation in male, less than or equal to the length of the thorax; ovipositor retracted; parasitoid; common.

Family Eurytomidae: Hindfemur not enlarged, hindtibia straight; antennae 5- to 7-segmented between pedicel and club; tarsae normally 5-segmented; midtibial spur thin and

spinelike; head projecting downward; hindtibia at least as long as femur, usually longer, same in male and female; scutellum without submedian grooves; hindcoxa nearly the same size as forecoxa; female ovipositor not exerted; pronotum rectangular in dorsal view; body black, yellow, or mixed black and yellow, rarely metallic (and then only faintly); abdomen laterally compressed.

Note: *Eurytoma furva*, *Eurytoma bugbeei*, and *Eurytoma* sp. have been grouped together in the working collection due to extreme morphological similarity and ensuing confusion.

Sycophila flava (Ashmead)

General appearance: Body yellow, legs yellow

Description: Antennae 8-segmented in female, 9-segmented in male, elbowed, short, slightly longer than head; wings have two dark windows on each, anterior margin bears row of dark setae; head yellow, eyes iridescent orange; thorax yellow; abdomen yellow, translucent, glassy, propodeum long and narrow; ovipositor retracted; inquiline; common.

Note: In Fall 1996, many emerging individuals were darkly pigmented, ranging from streaks on legs to almost completely dark-bodied. Leaf galls from Aransas yielded dark-bodied individuals in March 1996.

Eurytoma sp.

General appearance: Body dark brown, eyes bright red-orange, legs white

Description: Antennae 9-segmented in female, 9-segmented and pectinate in male, 2 terminal segments fused, elbowed, slightly longer than head; wings hyaline, small dark dot near costal margin; head dark brown, eyes bright red-orange; thorax dark brown, corrugated; abdomen brown, smooth and glassy, propodeum pronounced; ovipositor sheath extends ventrally along length of abdomen, tip often exerted; inquiline; common.

Notes: Abdomen of female may be smaller than that of *E. furva* and *E. bugbeeii*

Eurytoma furva Bugbee

General appearance: Body dark brown, eyes bright red-orange, legs yellow

Description: Antennae 8-segmented in female, 2 terminal segments fused, elbowed, slightly longer than head; wings hyaline, small dark dot near costal margin; head dark brown, eyes bright red-orange; thorax dark brown, corrugated; abdomen large, brown, smooth and glassy, propodeum pronounced, ventral margin amber; ovipositor sheath extends ventrally along length of abdomen, tip often exerted; inquiline; common.

Note: There are no males of this species in the collections.

Eurytoma bugbeeii Grissell

General appearance: Body dark brown, eyes bright red-orange, legs yellow

Description: Antennae 10-segmented in female, 9-segmented and pectinate in male, 3 terminal segments fused, elbowed, slightly longer than head; wings hyaline, small dark dot near costal margin; head dark brown, eyes bright red-orange; thorax dark brown, corrugated; abdomen brown, smooth and glassy, propodeum pronounced ; ovipositor sheath extends ventrally along length of abdomen, tip often exerted; inquiline; common.

Note: Abdomen of female may be smaller than that of *Eurytoma furva*

VITA

JoAnne Nancy Lund was born in Milwaukee, Wisconsin, on August 28, 1956, the daughter of Anne Louise Lund and Allen Walter Lund. After graduation from Pewaukee High School, Pewaukee, Wisconsin, in 1974, she entered into the field of zoology at the University of Wisconsin-Madison. She earned the degree of Bachelor of Science in December, 1978. During the following year she was employed as a research assistant at the University of Wisconsin, then woke up one morning to a temperature of -37 ° F and decided to head south. She was employed as a research assistant at the University of Texas System Cancer Center in Smithville, Texas, from 1981-1984, then helped her supervisor move his laboratory to the University of California-Berkeley in January 1985. From June 1985-June 1986, she travelled by bicycle and donkey in Europe and northern Africa. She was employed as a research assistant (1986 and 1987) in the Department of Oncology Research at Rhode Island Hospital, Providence, Rhode Island, when the urge to see the world again overwhelmed her. She served as a Peace Corps Volunteer/Fish Culture Extension Agent in Burundi, central Africa, from 1988-1991. Returning once again to the USA, she worked as manager of the *Xiphophorus* Genetic Stock Center at Southwest Texas State University from 1992-1995, then entered the Graduate School of Southwest Texas State University, San Marcos, Texas in fall of 1995.

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