Life history, natural enemies, and management of Disholcaspis quercusvirens (Hymenoptera)

Citation for published version:

Digital Object Identifier (DOI):
10.1603/EC12206

Link:
Link to publication record in Edinburgh Research Explorer

Document Version:
Publisher's PDF, also known as Version of record

Published In:
Journal of economic entomology

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Species of Disholcaspis, or bullet gall wasps, attack several oak species (Fagaceae: Quercus) in North and Central America (Eckberg and Cranshaw 1994, Melika and Abrahamson 2002). These cynipids have alternating generations (Melika and Abrahamson 2002), in which the asexual wasps develop singly or in clusters of detachable, spherical branch, or root galls. The sexual-generation eggs are laid in leaf or flower buds, which become small thin-walled galls (Morgan et al. 1953, Melika and Abrahamson 2002). Clusters of bullet galls are referred to as “compound galls”. An occasional cluster of bullet galls is considered harmless (Gilman and Watson 2011), but severe infestations may partially or completely girdle branches, thus reducing tree quality and value. Succulent bullet galls also produce a sticky exudate that attracts stinging insects, which creates a hazard for sensitive and allergic individuals (Felt 1940, Seibert 1993; J.P. Bird and E.A. Buss, personal observations).

Disholcaspis quercusvirens (Ashmead) infests southern live oak (Quercus virginiana Mill.), sand live oak (Quercus geminata Small), and running oak [Quercus minima (Sarg.) Small] (Price et al. 2004) in South Carolina, Georgia, Florida, Mississippi, Louisiana, and Texas (Krombein et al. 1979). Only wasps from its asexual, stem, or “bullet” gall generation have been taxonomically described (Ashmead 1881), and little of the wasp’s biology is known. As part of a gall maker complex in Florida, D. quercusvirens has contributed to the decreased production and sale of live oak clones, the return to seedling tree production, and has increased the number of treatments needed to produce marketable trees (E.A. Buss, personal observations). Knowledge of the life cycle and natural enemy complex of D. quercusvirens may help minimize labor-intensive corrective pruning (Elison and Potter 2000c) and help synchronize insecticide applications with adult or larval susceptibility to prevent full gall formation.

In this study, we identified the sexual generation of D. quercusvirens, described gall development of both asexual and sexual generations, determined adult cynipid and parasitoid activity periods, and evaluated the efficacy of three contact insecticides to suppress the gall makers and prevent additional gall formation. The oviposition period for asexual females occurred from late November to January in both years of the caging study. Eggs laid into dormant buds resulted in small bud galls in which the sexual generation developed for 4–5 mo. Sexual adults emerged and laid eggs in young elongating shoots in April. Bullet galls began protruding from branches in June, and asexual wasps emerged 5–7 mo later. Cynipids that emerged from the bullet (asexual generation) and bud (sexual generation) galls were genetically identical. Both generations were heavily parasitized. Targeting asexual females with an early December treatment of bifenthrin or acephate significantly reduced the number of bud galls, but control did not extend to the next generation of bullet galls, possibly because of reinvasion from neighboring infested trees.

KEY WORDS  gall wasp, bullet gall, alternating generation, Quercus virginiana
Materials and Methods

Study Sites. The biology, development, and management of *D. quercusvirens* were studied primarily at a tree nursery in Penney Farms (Putnam County), FL, from 2007 to 2008. Blocks of similarly sized (≈5 m in height, ≈6 cm diameter at breast height), field grown, clonal Cathedral live oak trees that were heavily infested with *D. quercusvirens* stem galls were selected for use. A secondary site with Cathedral live oak trees (University of Florida Horticultural Tree Unit, Gainesville, FL) was used to augment bud gall collections in 2008 (trees were ≈5 m in height, ≈11 cm diameter at breast height).

Asexual Bullet Gall Development and Inhabitants. Four to 10 compound bullet galls of the asexual generation assumed to be actively growing were periodically cut from randomly selected live oak trees at the tree nursery from August to December 2007 and from July to November 2008 to monitor mean gall growth. Bullet galls were then frozen and later examined under a binocular dissecting scope. To rear and identify arthropods inhabiting the bullet galls, another 5–10 compound galls were collected every 2–3 wk from early July through mid-December 2007. The number of bullet galls per compound gall was counted, and the mean maximum compound gall length, individual bullet gall diameter (two measurements taken perpendicular to each other and averaged) and height (from base of stem gall after removal from branch to the highest point), and mean diameter of the branch apical to the compound gall were measured with a digital caliper. Dimensions of individual and compound galls were associated with the insects reared from them to determine if the inhabitant altered bullet gall morphology.

To rear specimens, bullet galls were placed individually into scintillation vials, held in the laboratory (a photoperiod of 13:11 [L:D] h; 23.7°C, 48% relative humidity [RH]), and checked weekly. All specimens collected were preserved in 95% ethyl alcohol (EtOH) and identified by G.M. The effects of bullet gall inhabitant (i.e., gall maker, parasitoid, or inquiline) on mean maximum gall diameter and height, and number of galls per compound gall, were compared by using multivariate analysis of variance (ANOVA) (SAS Institute, version 9.2, 2008). In addition, the abdomens of 20 asexual female *D. quercusvirens* stored in 95% EtOH were dissected under a binocular dissecting scope (20× magnification) and the number of mature eggs were counted (potential fecundity). Previtellogenic eggs were not present.

Asexual Female Longevity. Ten branches containing one to four compound bullet galls were collected weekly from the tree nursery on 2–3-yr-old unpruned trees starting ≈20 October 2008. Compound galls were held in 0.12-liter plastic Solo cups with vented lids (≈2.5-cm, in diameter, hole covered with white chiffon mesh) to allow air flow. Galls in cups were held under partial shade in a shelf (0.3 by 0.3 by 0.6 m) with a solid roof to better approximate outdoor conditions. Cups were checked daily for *D. quercusvirens* emergence.

To determine the longevity of individual *D. quercusvirens* females, a no-choice test was conducted during early (21 November to 2 December 2008) and late (4–9 January 2009) wasp emergence from bullet galls (n = 30 wasps per treatment at each time interval). Within 24 h of emergence from a bullet gall, a female was placed into a clear plastic deli container (0.47 liter; 7 cm in height, 8.5 cm bottom diameter) with a plastic Solo cup (29.6 ml) secured to the bottom. Each Solo cup contained water, a 10% honey solution, or nothing (control). The water and honey solutions were soaked into a 5.1-cm-long dental wick projecting through each lid, and solutions were replaced every 2 d. Because a variable number of *D. quercusvirens* emerged daily, single replicates were prepared over time. Containers were held in a rearing chamber (a photoperiod of 13:11 [L:D] h; 26.4°C, ≈40% RH). The effects of diet on gall maker longevity were compared by using PROC LIFETEST (SAS Institute, version 9.2, 2008).

Confirmation of the Alternate Generation. To minimize parasitism, maximize gall wasp survival, and prevent invasion by other gall makers, we caged young compound galls of the asexual generation on their branches in the lower crown (≈2.2 m) (n = 30) and upper crown (≈2.2–4.3 m) (n = 20) of infested live oaks in June and July 2007. Cages consisted of a fine white organza mesh. Asexual *D. quercusvirens* females eventually emerged and oviposited within the cages. Because dormant buds were anticipated to be the targets for oviposition, the initial sizes of 30 buds present during the asexual wasp emergence period were recorded. Caged branches were removed in late March 2008. Any new galls found on those branches were presumed to contain the alternating sexual generation.

In April 2008, three male and two female cynipids recovered from bud galls within the cages were preserved in 95% EtOH and, based on their morphology, were confirmed as a species of *Disholcaspis*. That the cynipids from the bullet and bud galls comprised a single species lifecycle was then tested by using a DNA barcoding approach using mitochondrial and nuclear sequence data for the three male sexual *Disholcaspis* specimens and three female asexual *D. quercusvirens* specimens (for an example of this approach see Stone et al. 2008). Molecular work was carried out at the NERC Genepool facility at the University of Edinburgh, United Kingdom.

Bud Gall and Gall Maker Development. On 9, 11, and 16 March 2009, 10 branches were selected randomly from trees at the nursery near Penney Farms. Twenty bud galls of the sexual generation per date were dissected to determine gall maker survival and development, and to document gall morphology. The length and width of galled and ungaUld bud galls while bud scales completely enclosed the galls were compared by using a t-test (SAS Institute, version 9.2, 2008). To minimize variability, bud galls that were visible because leaves already had started to expand and bud scales were falling off were excluded. The bud
stage (i.e., dormant, swelling, green tip, or expanded leaves [Eliaison and Potter 2000b]), bud length and width, gall length and width (after bud scales were removed), and gall maker life stage were recorded.

**Bud Gall Inhabitants.** To determine the survival of the sexual generation of *D. quercusvirens* in bud galls and describe its natural enemy complex, bud galls were collected weekly from 4 April to 14 May 2008 from the lower crowns of live oak trees at the nursery in Penney Farms and the UF Horticultural Tree Teaching Unit. Bud galls (n = 450) were carefully removed from branches and placed individually into clear gelatin capsules (size 0) (Shorthouse 1972) in the laboratory (a photoperiod of 13:11 [LD] h; 23.7°C; and 48% RH). Cynipid and natural enemy emergence was monitored daily. All specimens were preserved in 95% EtOH and identified by G.M.

The activity period of the sexual generation emerging from bud galls in 2009 was also monitored. Ten branches (~10 cm long) were collected on 9 March 2009, before bud galls were visible to the collector, from live oaks at the nursery in Penney Farms, and each bud was placed individually into a gelatin capsule (size 0), held at room temperature, and insect emergence dates and species were recorded. In addition, at least 20 bud galls were collected on 16, 19, and 24 March; 17 and 21 April; and 4 and 11 May 2009, and reared in a similar manner. Twenty sexual-genera-
tion females reared from bud galls collected in mid-May were later dissected under a binocular dissecting scope to count the number of eggs per female abdomen.

**Insecticide Trial.** Three contact insecticides were evaluated for their ability to either kill asexual *D. quercusvirens* during emergence or before ovipositing, thus preventing bud gall formation. Treatments included acephate (1.17 liters/ha, Orthene TTO 97, Valen-
t USA Corporation, Walnut Creek, CA), bifenthrin (1 ml/liter, Bifen XTS, Control Solutions, Inc., Pasa-
dena, TX), carbaryl (292.3 ml/ha, Sevin SL, Bayer Environmental Science, Research Triangle Park, NC), and an untreated control.

Heavily galled trees within a block of field-grown live oaks at the nursery in Penny Farms were selected for this test. Three untreated tree rows separated each treatment row to minimize insecticide drift and contamina-
tion. Within a treatment row (n = 6 replicates/rows), four sets of five to seven heavily galled trees were selected and each was randomly assigned one treatment (e.g., acephate, bifenthrin, carbaryl, or control). Insecticides were applied to the foliage of two trees in a set either once (24 November 2008, after most field-collected bullet galls contained adult *D. quercusvirens*, based on monitoring) or twice (24 No-
vember and 15 December 2008, before peak asexual adult emergence was expected) to the other two trees. Within each set, one to three buffer trees separated each pair of treatment trees. Treatments were sprayed to runoff with a two-nozzle (XR Tee Jet 110/2 VS) 1-m boom by using a CO₂ backpack sprayer at 0.22 MPa (Weed Systems Inc., Hawthorne, FL).

To evaluate efficacy, two branches (~23–25 cm in length) from each cardinal direction in the upper and lower canopy of each tree were cut, bagged, brought to the laboratory on 24 March 2009, and all bud galls on each branch were counted. Intact bud galls of the sexual generation with no emergence holes were placed individually into clear gelatin capsules and held in the laboratory (a photoperiod of 13:11 [LD] h; 23.7°C; and 48% RH) to record the percentage of cynipid and parasitoid emergence. Proportion of in-
sect emergence data were arcsine-transformed, treat-
ment differences were analyzed by ANOVA, and, when significant, means were separated by Tukey’s honestly significant difference test (PROC GLM, SAS Institute, 2008). Data were presented as a percentage of the insects that emerged.

On 12 August 2009, subsequent bullet gall develop-
ment was estimated on trees that had been treated only once in November 2008, during early asexual *D. quercusvirens* emergence (trees treated twice were excluded because results from the bud gall generation did not statistically vary with single-treatment trees). The approximate total number of young red-colored bullet galls per tree was counted for 60 s by J.B. and E.B. (one on the east half and the other on the west half of each tree). Results were analyzed with an ANOVA (PROC GLM, SAS Institute, 2008), and means were separated by Tukey’s honestly significant difference test.

**Results and Discussion**

**Stem Gall Development and Inhabitants.** Bullet galls were initiated by the sexual generation of *D. quercusvirens* during early shoot expansion in the spring. Slight stem swellings were visible by June, and small red-colored bullet galls with soft and spongy tissue (Fig. 1A) emerged through the bark by August. A sweet and sticky exudate occurred on the bullet galls from August to mid-October, when only larvae were present inside the galls. Similarly, stem gall secretions on branches of *Quercus gambelli* Nutt. coincided with *Diaporthe perníciosa* Bassett larval development (Seibert 1993). This exudate attracted red imported fire ants (*Solenopsis invicta* Buren), velvet ants (*Hy-
menoptera: Mutillidae*) (Fig. 1B), and paper wasps (*Hymenoptera: Vespidae*), and supported sooty mold growth (Fig. 1C). Despite oaks not having extraoral nectaries, certain gall makers (e.g., *Disholcaspsis ełloðraedensis* Beutenmüller) can cause their host trees to secrete a honeydew-like substance. This can result in a mutualism with ants that protect the gall makers from parasitism (Inouye and Agrawal 2004). *D. quercusvirens* bullet galls were mature and woody (Fig. 1D) in the fall, reaching their maximum diameter by November (Fig. 2).

Mean maximum bullet gall diameter was 7.7 ± 0.1 mm in 2007 and 4.8 ± 0.1 mm in 2008 on the trees at the nursery (Fig. 2). Compound galls were 30.4 ± 1.6 mm in length with 9.3 ± 0.6 bullet galls per compound gall (range: 1–44 bullet galls) in 2007 and 25.5 ± 1.4
mm in length with 11.0 ± 0.9 bullet galls per compound gall (range: 1–52 bullet galls) in 2008.

Asexual females (Fig. 3A) emerged during cold weather periods from late-November through January and oviposited into dormant buds (1.3 ± 0.07 mm in length, 1.2 ± 0.09 mm in width at base). Females (n = 20) carried 64.0 ± 3.0 eggs (range: 43–88 eggs). All eggs within female abdomens appeared to be of similar size, but dimensions were not measured.

In total, 1,650 bullet galls were collected from 1 August to 18 December 2007, and 91.3% were parasitized (Table 1). The natural enemy complex included species of Synergus (Cynipidae), Eupelmus (Eupelmidae), Eurytoma and Sycophila (Eurytomidae), Ormyrus (Ormyridae), and Acaenacis (Pteromalidae) (Fig. 4). Of the insects reared from bullet galls, 8.7% were D. quercusvirens, 27.3% were inquilines (species of Synergus, Fig. 4A and B), and 63.9% were parasitoids (Hymenoptera: Eupelmidae, Eurytomidae, Ormyridae, and Pteromalidae) (Table 1). The activity period of the different parasitoid species was not determined.

Of the 1,470 bullet galls collected from the tree nursery in 2008, many did not develop properly, had smaller diameters than bullet galls measured in 2007, and gall inhabitants were unable to complete development on the trees. Only five inquiline specimens were reared (three of an undescribed Synergus species and two Synergus ficigerae Ashmead). Seibert (1993) similarly reported that a tree had aborted all 24 compound galls of D. perniciosa after it had supported many viable compound galls the previous year. Why this occurred is unknown, but we speculate that tree susceptibility or ability to support galls may vary from year to year (Morgan et al. 1983, Frankie et al. 1992). Other mortality factors could include the introduction
of pathogens that could contaminate the gall tissue or infect the gall maker (Seibert 1993), or parasitoids could have inserted their ovipositors and injured the gall maker, but not deposited an egg, which would have stopped gall growth at that time.

Bullet gall height, diameter, and number of bullet galls per compound stem gall were influenced by the gall inhabitant (Table 2; multivariate ANOVA, Wilks’ $\lambda = 0.8032; df = 6, 316; P < 0.0001$). Bullet galls containing \textit{D. quercusvirens} and parasitoids in 2007 were significantly wider than those containing inquilines. Bullet galls with live \textit{D. quercusvirens} were significantly taller than those containing parasitoids or inquilines. Furthermore, compound galls with more than five bullet galls in a cluster produced more \textit{D. quercusvirens} than compound galls having five or fewer bullet galls (data not shown).

Bullet galls from which gall makers successfully emerged did not produce any parasitoids or inquilines. However, two or more parasitoids and/or inquilines emerged from many \textit{D. quercusvirens} bullet galls and presumably killed or out-competed the gall maker. Although inquilines are phytophagous, their development may cause gall maker death because of overcrowding or competition (Askew 1984, Csóka et al. 2005). These results are similar to those found in other studies on cynipids, in which all gall makers died because of the occupation of the gall by a \textit{Synergus} inquiline (Washburn and Cornell 1981, Frankie et al. 1992, Seibert 1993, Plantard et al. 1996). Frankie et al. (1992) documented that species of \textit{Synergus} actually changed the morphology of the galls they inhabited by creating denser galls with fewer empty air chambers compared with those galls occupied by parasitoids or gall makers. Similar responses have been observed in rose cynipid galls colonized by \textit{Periclistus} inquilines (Brooks and Shorthouse 1998). This phenomenon of

\begin{figure}
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\includegraphics[width=0.9\textwidth]{fig2.png}
\caption{Mean diameter (millimeters) of bullet galls collected from live oak trees (Clay Co., FL) from July to December 2007 and July to November 2008. *Means ± SEM within columns followed by a different letter are significantly different at $\alpha = 0.05$ (ANOVA; $F = 260.25; df = 12, 3264; P < 0.0001$). Letters only represent differences comparing diameters within a month for each year (2007 and 2008), not diameters across months within the same year. (Online figure in color.)}
\end{figure}

\begin{figure}
\centering
\includegraphics[width=0.9\textwidth]{fig3.png}
\caption{Life stages of \textit{D. quercusvirens}. (A) Adult asexual female. (B) Pupa of sexual female. (C) Adult sexual female (photo credit: H. Ferrand). (D) Adult sexual male (photo credit: H. Ferrand). (Online figure in color.)}
\end{figure}
altered gall morphology because of Synergus inhabitants was not observed in this study.

**Asexual Female Longevity.** Female survival in the laboratory ranged from an average 2.5 to 8.1 d (Table 3). For both trials, *D. quercusvirens* provided with water or honey–water lived significantly longer than unfed (control) wasps (Table 3; Trial 1: $\chi^2 = 16.843$, df = 2, $P < 0.0002$; Trial 2: $\chi^2 = 17.443$, df = 2, $P < 0.0002$).

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**Table 1.** Total number of gall makers, inquilines, and parasitoids reared from bullet and bud galls collected from live oak trees, 2007–2009

![Fig. 4. Parasitoids and inquilines reared from *D. quercusvirens* galls. (A) *Synergus ficigerae* male, (B) *Synergus succinipes* female, (C) *Acaenacis lausus* male, (D) *Eurytoma* spp. male, (E) *Eurytoma* spp. female, (F) *Brasema auratus* male, (G) *Sycophila* spp. female, (H) *Ormyrus hegeli* female, and (I) *Brasema gemmarii* female. (Photo credits: H. Ferrand.) (Online figure in color.)](https://academic.oup.com/jee/article-abstract/106/4/1747/805904)
survived when provided different diets in the laboratory. To extend their survival in the feeding by adults may not be necessary for egg deposition (P. Morgan et al. 1983), although we did not directly observe this production of hemipterans in the oak trees (Eliason and Potter 2001), although we did not directly observe this behavior. Other environmental conditions (e.g., ambient temperature, relative humidity, or photoperiod) also may affect adult wasp longevity. Because cynipid eggs are generally ready to be laid on asexual adult emergence (Seibert 1993, Eliason and Potter 2000a), feeding by adults may not be necessary for egg development or maturation.

Confirmation of the Alternate Generation. DNA sequencing of the mitochondrial cytochrome b gene and the nuclear ITS2 gene revealed identical sequences for the sexual and asexual wasps. This DNA barcode testing, along with production of sexual offspring by caged asexual D. quercusvirens and morphological examination, confirmed that the asexual generation developing in the bud galls and the sexual generation from the bud galls were of the same species. Details of the morphological and DNA analyses demonstrating the alternate generations of D. quercusvirens have been described by Melka et al. (2013).

Bud Gall and Sexual Generation Development. Small bud galls developed beneath bud scales (Fig. 1E) and were located on the branches at the base of a petiole, hidden between young leaves (Fig. 1F). Bud galls were 2.5 ± 0.02 mm in length and 1.1 ± 0.01 mm in width. They likely were initiated by asexual wasp oviposition (i.e., November–January), and remained on trees until May. Bud gall tissue was soft and spongy and continued to grow and harden until the bud scales fell off the galls. The tips of young bud galls, sometimes red in color (Fig. 1E), were visible in early- to mid-March. Emergence of D. quercusvirens ended by mid-April, and parasitoid emergence continued until all bud galls fell off the trees in May. Bud galls were fragile after hardening, and, similar to the D. cinerosa system (Frankie et al. 1992), could be easily dislodged after adult emergence or inclement weather.

Sexual generation pupae (Fig. 3B) were present in bud galls by 11 March 2009, and 65% of the bud galls collected on 16 March contained white pupae. First adult emergence (Fig. 3C and D) occurred on 21–23 March 2009 from laboratory rearings, and emergence continued through ~21 April 2009. Although longevity of the sexual generation of D. quercusvirens was not evaluated, mated females of D. cinerosa were reported to live for ~1 wk (Frankie et al. 1992). Sexual-generation D. quercusvirens females had 87.2 ± 3.0 eggs within their abdomens. All eggs appeared to be of the same size and maturity, although measurements were not taken. The ratio of males to females was ~2:3.

Bud Gall Arthropod Inhabitants. In total, 480 bud galls were collected from 4 April to 14 May 2008, at the end of the sexual D. quercusvirens emergence period. Parasitoids comprised 99.1% and D. quercusvirens comprised 0.9% of the specimens reared (Table 1). Parasitoids reared from these bud galls included a species of Aprostocetus, Baryscapus, and Pediobius (Eulophidae); Brausena auratus (Ashmead) (Eupelmidae); and Acacenia lausus (Walker) (Pteromalidae). The most abundant species was B. auratus. No inquilines were reared from the bud galls; a feature shared with some other small and structurally simple oak cynipid galls (Stone et al. 1995, Schönrogge et al. 2000).

From 19 March to 14 May 2009, 3,884 bud galls were collected. These bud galls had a more diverse parasitoid complex (Table 1), which included a species of Aprostocetus, Baryscapus, and Pediobius (Eulophidae); B. auratus and Brausena gemmarii (Ashmead) (Eupelmidae); Syzophila and an unknown species (Eurytomidae); Ormyrus hegeli (Girault) (Ormyridae); and A. lausus (Pteromalidae). Only one specimen was reared from each bud gall (e.g., bud galls were monothalamous), as observed in other small sexual-generation oak cynipid galls (e.g., Stone et al. 1995, Schönrogge et al. 2000). The most abundant parasitoid in 2009 was A. lausus, and it was reared also from bullet galls in 2007. Pteromalid wasps in cynipid galls are

<table>
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<th>Treatment</th>
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<th>Trial 2</th>
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<td>5.1 ± 0.4a</td>
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<td>Water</td>
<td>8.1 ± 0.6b</td>
<td>3.3 ± 2.6b</td>
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<tr>
<td>Honey</td>
<td>7.3 ± 0.7b</td>
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* Trials 1 and 2 were significantly different at α = 0.05 (TTEST; F = 9.56; df = 177; P < 0.0001).

b Means ± SEM within columns with different letters represent significant differences at α = 0.05 (Trial 1: LIFETEST; χ² = 16.843; df = 2; P < 0.0002).

c Means ± SEM within columns with different letters represent significant differences at α = 0.05 (Trial 2: LIFETEST; χ² = 17.443; df = 2; P < 0.0002).
D. quercusvirens and bullet gall formation when entire field-grown trees were treated in a nursery

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Trade name</th>
<th>Rate</th>
<th>Total no. bud galls per branch per tree</th>
<th>Total no. bullet galls per tree</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acephate</td>
<td>Orthene TTO 97</td>
<td>1.17 liters/ha</td>
<td>11.5 ± 4.7b</td>
<td>98.3 ± 21.9a</td>
</tr>
<tr>
<td>Bifenthrin</td>
<td>Bifen XTS</td>
<td>1 ml/L</td>
<td>1.8 ± 2.7a</td>
<td>90.5 ± 19.5a</td>
</tr>
<tr>
<td>Carbaryl</td>
<td>Sevin SL</td>
<td>292.3 ml/ha</td>
<td>15.4 ± 9.7c</td>
<td>96.0 ± 35.6a</td>
</tr>
<tr>
<td>Control</td>
<td>—</td>
<td>—</td>
<td>16.7 ± 12.5c</td>
<td>87.8 ± 12.1a</td>
</tr>
</tbody>
</table>

a ANOVA for means ± SEM within columns followed by a different letter were significantly different at α = 0.05 (F = 8.87; df = 27, 347; P < 0.0001).

b ANOVA for comparison of mean ± SEM bullet galls counted (F = 0.05; df = 3, 20; P = 0.987).

Typically parasitoids, and their larvae feed on the eggs or larvae of their hosts (Krombein et al. 1979, Askew 1984). O. hegeli (Girault) (Ormyridae) was also reared from both bud galls in 2009 and bullet galls in 2007.

The parasitoid species Brasema ficigerae (Ashmead), Euelpinus quercus Ashmead, Eurytoma querciglobuli rubra Bugbee, Eurytoma studiosa Say, Tormyus elegantiissimus (Ashmead), Tormyus fagospirum (Provancher), and Tormyus lissimus (Walker) were previously reared from the egg of D. quercusvirens (Krombein et al. 1979), but were not found in the current study. Eurytoma and Tetrastichus (Eulophidae) can feed on developing larvae of D. perniciosa in bullet galls (Seibert 1993). It is assumed that the parasitoids reared from the D. quercusvirens bud galls also fed on the gall maker larvae or pupae.

**Insecticide Trial.** We demonstrated that one well-timed application of a contact insecticide before asexual adult emergence can sufficiently reduce the next generation by preventing eggs from being laid. Applications of bifenthrin and acephate at the beginning of asexual D. quercusvirens emergence significantly reduced the number of bud galls on sampled branches when compared with carbaryl and the control, with bifenthrin having the greatest reduction of bud galls (Table 4; F = 8.87; df = 27, 347; P < 0.0001). Number of applications (one or two) was initially included in the model, but did not significantly affect the number of bud galls formed. Treatments in November did not reduce the number of new bud galls per tree in the following year when compared with the control (Table 4; ANOVA, F = 0.05; df = 3, 20; P = 0.987).

These results suggest that one application of bifenthrin or acephate applied in late November or December can significantly reduce bud gall formation compared with the control, without reducing parasitism in the following bullet gall generation. This knowledge could minimize the number of applications made by live oak tree growers during the winter months. However, one winter application did not reduce bullet gall formation the following summer, which could justify the addition of a spring application aimed against the sexual-generation adults or their offspring. In this test, gall makers from surrounding buffer (untreated) trees may have immigrated to treated trees in the spring, after residues broke down, increasing the numbers of bullet galls that developed in the subsequent generation. Under commercial conditions, entire blocks of trees would be treated, which could minimize tree reinestation. Targeted applications at or after bud break or repeated applications over two or more years may be necessary to significantly increase tree quality by minimizing gall makers and their galls. Future trials should assess gall maker control in blocks or fields to minimize the potential of reinestation from neighboring trees.

**Effect on Natural Enemy Complex.** Treatments had no significant effect on the percentage of parasitoids (F = 0.58; df = 3, 43; P = 0.632) or D. quercusvirens (F = 2.33; df = 3, 43; P = 0.0875) that emerged from bud galls. Overall, more parasitoids emerged from bud galls than did D. quercusvirens (F = 98.07; df = 1, 22; P < 0.0001). Unfortunately, gall maker emergence is underestimated in this test because branches were harvested during the period in which gall makers emerged from galls. All D. quercusvirens adults that emerged in the bags before bud galls were placed into rearing containers were excluded from the data set.

Of all parasitoids reared from intact galls, Acaenacis lausus (Hymenoptera: Pteromalidae) was the most abundant, followed by B. auratus (Hymenop-
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tera: Eupelmidae), Baryscapsus (Hymenoptera: Eu-
lophiidae), Sycophila sp. (Hymenoptera: Eurytomi-
da), O hegeli (Ormyridae), and Eurytoma sp. (Eurytoma-
da) (Table 5).

The authors, outside the study site at the tree nur-
sery in Penney Farms (Putnam Co.), reared another
two species from the bud galls of D. quercusvirens: 
Brasema flavovarigatus (Ashmead) and a Eurytoma sp. 
different from a Eurytoma sp. reared from bullet 
galls.

When insecticide applications coincided with 
asesexual adult emergence and were applied to whole 
trees, results were similar to those of Eliason and 
Potter (2000c), who showed that treating the stem 
gall (asesexual) generation of Callirhitis cornigera 
(Osten Sacken) reduced the leaf gall (sexual) gen-
eration. The numbers of sexual-generation galls for 
both C. cornigera and D. quercusvirens were signifi-
cantly reduced on sampled shoots and branches for 
treated tree canopies when compared with the control 
(untreated) trees, whereas the percentages of para-
sitized galls and the number of bullet galls formed the 
following year did not differ between treatments.

This is the first study to associate the asexual- and 
sexual-generation galls and adults of D. quercusvirens, 
identify their natural enemy complex, and describe 
gall development and the emergence periods of the 
alternating generations. This information will help live 
red oak nurserymen and landscape managers better for-
mulate a management program to minimize tree 
infestations and improve tree quality.

Acknowledgments

We are grateful for the technical assistance provided by P. 
Ruppert, J. Cash, A. Washuta, K. Cho, M. Gilbert, and H. 
Ferrand. E. Gilman, V. Lietze (University of Florida), and P. 
Stiling (University of South Florida) kindly reviewed an 
earlier draft of this manuscript. We thank Gary A. P. Gibson 
(Canadian National Collection of Insects, Ottawa, Canada), 
E. E. Grissell (Sonoita, AZ), and P. Hanson (Escuela de 
Biología, Universidad de Costa Rica, San Jose, Costa Rica) for 
early identifications of chalcidoid parasitoid specimens, 
which served as voucher specimens for us during the iden-
tification of the D. quercusvirens parasitoid complex. Partial 
funding and many trees were kindly provided by Shadow-
lawn Nursery.

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Received 20 May 2012; accepted 25 May 2013.