

The Black Oak Gall Wasp on Cape Cod and Martha's Vineyard

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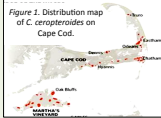
Abstract

Black oak, *Quercus velutina*, is the dominant deciduous tree on Cape Cod and Martha's Vineyard. In recent years, oaks on the Cape and the islands have experienced severe canopy loss due to the infestation of the black oak gall wasp (*Callirhytes ceropteroides*). Sandy soil conditions and previous winter moth exposure are two factors that may increase oak tree vulnerability. Little is known about the lifecycle of *C. ceropteroides* and the taxonomy of the species is still unclear. Our research aimed to investigate the lifecycle of *C. ceropteroides*, specifically emergence patterns, generation time and oviposition location. Using the CO1 gene, we determined that *C. ceropteroides* is the same species that also infests black oaks on Long Island and in Rhode Island. We found that both Cape Cod and Long Island have the same parasitoid (*Sycophilla* spp.). We also recovered an additional parasitoid (*Pteromalidae* spp.) on Long Island that we have not yet collected on Cape Cod. Our results showed that the density of *C. ceropteroides* on Long Island was significantly lower than that on Cape Cod, indicating that something is keeping its population in check. Our research may lay the foundation for future biological control efforts and will help arborists and landowners make management decisions regarding *C. ceropteroides* on Cape Cod and Martha's Vineyard.

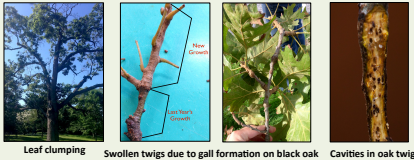
Introduction

Order: Hymenoptera
Family: Cynipidae
Species: *Callirhytes ceropteroides*

Host: Black Oak (*Quercus velutina*)
Distribution: Cape Cod, Martha's Vineyard, Long Island and Rhode Island. Found in residential, managed and forested areas.



Damage and Symptoms:



History of Damage:

Impact of *C. ceropteroides* was confused with damage by winter moth (*Operophtera brumata*) and fall cankerworm (*Alopiophila pomatoria*) when it began attacking trees on Martha's Vineyard and Cape Cod 4-6 years ago. The infestation is now widespread and has huge social, economic and environmental impacts on Cape Cod and Martha's Vineyard.

History of Infestation on Long Island in the 1990's:

Long Island experienced similar widespread damage by presumably the same gall wasp species in 1990, but the population crashed and damage subsided by 1995. These populations still exist today, but they no longer cause significant tree damage or oak mortality. Therefore, the Long Island population could hold a clue as to how to coexist with this pest.

Forms of Reproduction in Cynipidae Family:

Type of Parthenogenesis	Type of Generations	# of generations each year	Emergence Time
Obligate	asexual	1 per year	Spring
Facultative: 2 yrs	asexual, sexual	Alternate: 1 per year	Spring
Facultative: 1 yr	asexual, sexual	Alternate: both in year	Spring and Fall

Figure 2: Cynipid oak gall wasps can reproduce through facultative or obligate parthenogenesis. Their lifecycles are variable, especially the number of generations per year and generation length.

Methods

Gall Wasp Life Cycle and Adult Emergence Patterns

Emergence Patterns: We examined the timing of adult emergence of the black oak gall wasp. We used 3 x 3 inch organza bags secured with zip ties to cover new and last years growth on 15 tree branches in Dennis, MA. We checked the bags monthly and then weekly during crucial months for insect emergence. We will repeat this study on a larger scale in the spring of 2015.

Developmental Stages: We sampled bi-weekly to evaluate how many generations the gall wasp has per year and document it's stages of development. From each collection, we dissected five tips from 8 different black oak trees in Dennis, MA and recorded life stage data. Photographs of each life stage were taken under the microscope and the date was recorded.

Oviposition Preference: We aimed to resolve where females of *C. ceropteroides* prefer to oviposit their eggs. Post-oviposition, we randomly removed 3 branches from 20 infested trees in Dennis and West Harwich, MA. We dissected new and last year's growth to determine if there was larvae present in either or both of these tissues.

CO1 Gene Analysis and Taxonomy

We compared mitochondrial DNA from adult gall wasp samples from Cape Cod, Long Island and Rhode Island. We sent 96 adults from 8 different sites across New England to the University of Guelph for CO1 gene analysis. Through DNA extraction and PCR, they amplified the *cytochrome c oxidase I* gene and sequenced the nucleotides to retrieve a barcode for each individual. We blasted Barcode of Life and GenBank databases to look for possible matches to known cynipids. Then Rodger Gwiazdowski helped us create a maximum likelihood evolutionary tree using Fig Tree software. Our specimens were identified by Matt Buffington (*C. ceropteroides*) and Mike Gates (parasitoids) of the USDA, ARS Systematic Entomology Laboratory, Washington, DC.

Methods Continued

Population Comparison Study: Long Island and Cape Cod

We evaluated differences in gall wasp density on Long Island and Cape Cod. We used two sites: Dennis, MA and Riverhead, NY. The trees used in this study were infested for 3+ years.
Dissections: Every month since August 2014, we removed 5 tips from 8 trees at both our Cape Cod and Long Island sites. We dissected new and last years growth of each tip and recorded cavity density.
Bag Study: We placed 4.5 x 7 in organza bags on 5 tips of 10 different trees at each site and sealed the bags with zip ties. After spring emergence 2015, we will record emergence date/range, parasitoid count and gall wasp count per tip.

Results: Gall Wasp Lifecycle and Emergence

Emergence:

We concluded that May 7th – May 25th was the average emergence period of the spring generation of *C. ceropteroides* (2 weeks). We witnessed parasitoids flying until June 11, 2014.

Timeline and Stages of *C. Ceropteroides* Development



Figure 3. Photographs of the development of the current generation of *C. ceropteroides*, including two distinct larval stages and pupal and pharate adult stages.

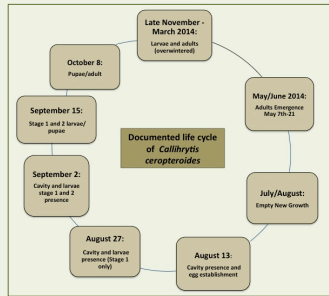


Figure 4. Lifecycle of *C. ceropteroides* created from emergence and development data.

Oviposition Preference:

We concluded that *C. ceropteroides* will lay eggs on both new and last years growth. Therefore, researchers must look at both new and last year's growth for an accurate representation of the black oak gall wasp population.

Results: Co1 Gene and Taxonomy

CO1 gene sequences for wasps from Rhode Island, Cape Cod, Long Island and Martha's Vineyard were 100% identical.

- There was no genetic variation among individuals
- There was no close relationship between our species and other *Callirhytes* spp. in the Genbank or Barcode of Life databases.

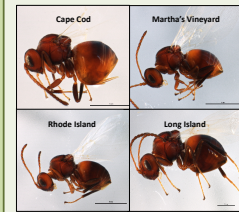


Figure 5. Microscopic images of individual gall wasps from different research sites and states.



Figure 6. Maximum likelihood tree created from CO1 gene sequences to compare gall wasps in different areas using Fig Tree software.

Results: Co1 Gene and Taxonomy

Parasitoid Results:

Sycophilla spp. (Hymenoptera: Eurytomidae) were found on Cape Cod and Long Island (Oakdale, NY)
Pteromalid parasitoid tentatively identified as *Acaenasis tacit* was found on Long Island, but not yet on Cape Cod.



Results: Population Study

Dissections were completed in August and September to compare the population size of *C. ceropteroides* on Long Island and Cape Cod. We found that both gall wasp cavity density and cavity count is significantly lower on Long Island. Our results support the idea that gall wasp densities are suppressed on Long Island and open the door for possible biological control agents.

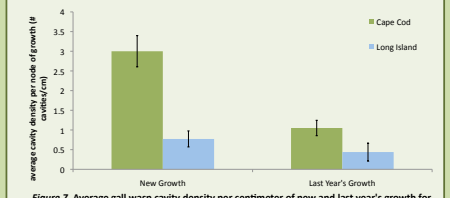


Figure 7. Average gall wasp cavity density per centimeter of new and last year's growth for sites on Long Island and Cape Cod (n=160).

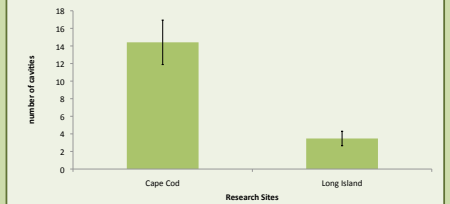


Figure 8. Average number of new gall wasp cavities per stem (including new and last years growth) on Long Island and Cape Cod (n=160).

Conclusions and Future Research

We have concluded that *C. ceropteroides* emerges in May and begins development again in August. At this moment, it appears that *C. ceropteroides* overwinters in the pharate adult stage and we have not yet discovered a sexual generation. When accounting for population size, researchers need to evaluate both new and last years growth, because *C. ceropteroides* will oviposit in both. Managers across state lines should work together to control this pest, because it is present in Long Island, Cape Cod, Martha's Vineyard and Rhode Island. We found a parasitoid that is present on Long Island, where gall wasp densities are much lower than Cape Cod. We have not yet recovered this species on Cape Cod. Future research should focus on why densities of *C. ceropteroides* are now much lower on Long Island than they were in the 1990s and present day Cape Cod. In addition, we will continue to monitor parasitoid populations on Cape Cod and Long Island and try to estimate percent parasitism. We will also continue our lifecycle analysis, look for galls representing a sexual generation and fill in the gaps we have in late fall and early summer.

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