

Sustainable Management of the Invasive Asian Chestnut Gall Wasp? Interactions between native and introduced natural enemies

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Fig. 1. Fully developed gall caused by the Asian chestnut gall wasp, *D. kuriphilus*.

The Asian chestnut gall wasp, *Dryocosmus kuriphilus*, forms globular galls on actively growing shoots and causes significant damage to susceptible chestnut (8). Galling prevents infested shoots from producing normal shoot growth and reproductive flowers, thereby reducing or eliminat-

ing nut production. Galling also reduces tree vigor and can cause tree mortality (5, 8). Chestnut production and chestnut restoration efforts throughout eastern North America are threatened by the persistent spread of this exotic, invasive insect (1). The gall wasp was first observed in the USA in 1974 in-

festing Chinese chestnut near Bryon, GA. It has since spread as far north as Pennsylvania and Maryland, and westward to central Kentucky.

The gall wasp has one generation per year and reproduces asexually. Adult females lay eggs inside buds in early summer. Eggs hatch soon after, but larvae remain inactive. Larval growth begins at budbreak the following spring, when one or more larvae induce rapid formation of conspicuous stem, petiole, or leaf galls (Fig. 1). These galls provide the developing wasp protection throughout the larval and pupal stages. Adults emerge from galls in the early summer and fly to new shoots to lay eggs for the next generation (8, 9). Once the wasps emerge, galls become woody and dry, and can remain on the tree for several years. Small leaves remain attached to the galls during the winter; these are highly visible and are useful in detecting infestations (Fig. 2).

In its native range natural enemies effectively keep gall wasp populations in check, and some of these natural enemies have been successfully introduced to other areas (6). In North America, *Torymus sinensis* (Hymenoptera: Torymidae) and other parasitoids, which feed internally on their host insects, were introduced for Asian chestnut gall wasp control in the mid-1970's (9). *T. sinensis* is now well established in the eastern USA. In addition, several native parasites have been found in association with the Asian chestnut



Fig. 2. Galls retain their leaves during winter months and are more easily detected.

gall wasp. One species in particular, *Ormyrus labotus* (Hymenoptera: Ormyridae), occurs in numbers that suggest it may contribute to gall wasp population suppression (2).

We've been working to characterize associates of the Asian chestnut gall wasp in eastern North America to more fully understand what factors regulate gall wasp populations. Our ultimate goal is to gain an understanding of the ecological interactions, dispersal patterns, and mechanisms regulating gall wasp populations in eastern North America. Here we report on interactions between the introduced parasitoid, *T. sinensis*, and the native parasitoid, *O. labotus*, on the exotic Asian chestnut gall wasp in North America, and evaluate how associated vegetation may affect natural enemy occurrence and activity.

We established four geographically distinct study sites, including two orchard settings (Meadowview, VA and Hiram, OH), ornamental Chinese chestnuts in a suburban setting (Broadview Heights, OH), and naturally occurring American chestnuts in a forest (Bowling Green, KY). To relate gall wasp and natural enemy occurrence to surrounding vegetation, woody plant

vegetation and site characteristics were surveyed, focusing especially on chestnut and oaks.

We collected galls monthly and measured fresh weight and volume before dissecting. The number of chambers per gall was counted and the thickness of the protective schlerenchyma layer surrounding the larval chambers was measured. The contents of each gall chamber were identified as gall former, parasitoid, fungus, or empty chamber.

Since larval parasitoids are morphologically indistinguishable we used a molecular approach to focus on identification of the two most numerous parasitoids of the gall wasp in North America, the introduced *T. sinensis*, and the native *O. labotus* (2). DNA was extracted from parasitoid larvae and amplified. Species specific size variation in ribosomal ITS2 PCR products was determined by direct sequencing of DNA extracted from known *D. kuriphilus*, *T. sinensis*, and *O. labotus* adults collected and identified previously (2). Each unknown larval parasitoid was identified by molecular comparison with known adult specimens.

Our collections confirmed that parasitism accounts for the greatest amount of gall wasp mortality (2). Fungi within the gall interior also caused significant mortality, as well as a failure of adults to emerge for indeterminate reasons. Additionally, black lesions on the gall exteriors caused by an unidentified

fungus and feeding by the lesser chestnut weevil, *Curculio sayi* (Coleoptera: Curculionidae), were positively correlated with gall wasp mortality, but we are not certain whether this relationship is causative or simply correlative.

Our molecular approach was able to effectively identify parasitoids of the Asian chestnut gall wasp that were otherwise indistinguishable. PCR products from *T. sinensis* and *O. labotus* differed enough from *D. kuriphilus* and from each other to adequately distinguish unknown larvae (Fig. 3). Using this molecular tool we found that the introduced parasitoid *T. sinensis* was present at each collection site, but was most prevalent at orchard settings with the greatest relative density of chestnut. This suggests that this parasitoid is moving with expanding gall wasp populations in eastern North America, perhaps in infested plant material. *T. sinensis* is univoltine; adults emerge in early spring and parasitize newly developing galls. It occurs in galls throughout the year and overwinters in dead galls as pupae and larvae (8, 9). *T. sinensis* is a generalist parasitoid of cynipid gall wasps in China, but acts as a specialist on Asian chestnut gall wasp in Japan, and possibly North America, where *D. kuriphilus* galls are abundant where chestnut persists (10). This synchrony with the *D. kuriphilus* life cycle contributes to the efficacy of *T. sinensis* as a parasitoid of *D. kuriphilus*, and

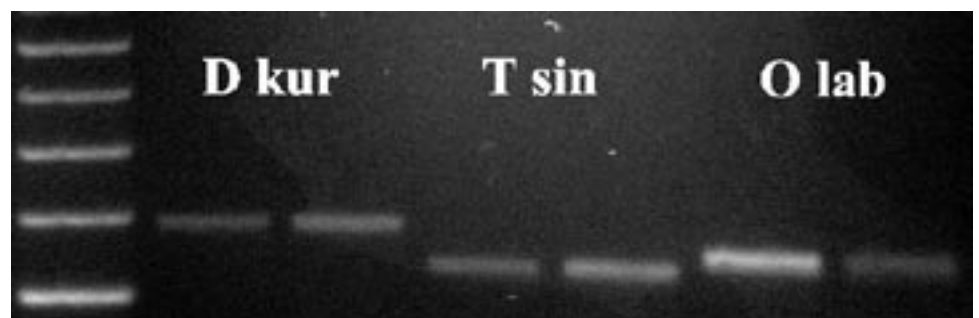


Fig. 3. The ribosomal DNA sequence ITS2 was PCR amplified from DNA isolated from adult *D. kuriphilus* (Dkur), the introduced parasitoid, *T. sinensis* (Tsin), and the native parasitoid, *O. labotus* (Olab) collected and identified from previous studies (2). The left lane is a 1000kb ladder.

may explain why *T. sinensis* appears to act as a specialist on the Asian chestnut gall wasp in North America.

The native parasitoid *O. labotus* utilized the gall wasp and the introduced parasitoid, *T. sinensis*, as hosts within chestnut galls. *O. labotus* produces multiple generations per year and exploits more hosts, including cynipid gall wasps and cecidomyiid gall flies, than other known *Ormyrus* species (3). *O. labotus* was only present in chestnut galls in late spring and early summer and was absent for the remainder of the year, indicating that this native parasitoid utilizes additional gallmaking hosts for its subsequent generations.

O. labotus also parasitized larger galls with thicker schlerenchyma layers relative to *T. sinensis*, in spite of having a shorter ovipositor. This may be attributable to differences in parasitoid phenology. *O. labotus* appeared in chestnut galls prior to *T. sinensis*, and may parasitize gall wasp larvae before the development of protective schlerenchyma layers (4). Subsequent generations of *O. labotus* then appear to utilize *T. sinensis* as a host. Thus the overlap in parasitoid phenologies appears to provide *O. labotus* with the opportunity to utilize both the chestnut gallmaker and the introduced parasitoid as its host. *O. labotus* may actually suppress *T. sinensis* populations in some locations, depending on habitat and associated vegetation. *T. sinensis* was the primary parasitoid where chestnut is numerically dominant, whereas *T. sinensis* and *O. labotus* were both common in the more vegetatively diverse study sites. *O. labotus* was most commonly collected from our forested site containing the highest oak component and greatest woody plant diversity, which would provide a source population of *O. labotus* from gallmaking hosts on oak and woody plant species.

The Asian chestnut gall wasp is a relatively new host for *O. labotus* (2). Our results suggest that competitive interactions are evolving between these

native and introduced parasitoids, and that these interactions may be influenced by variations in habitat. The native *O. labotus* appears to be making a host shift from oak-gallers to the chestnut-galler and its introduced parasitoid, and is consistent with our previous work suggesting potential antagonistic interactions between the native and introduced parasitoids (2).

Our work is providing important insight into the ecological interactions between two important parasitoids of *D. kuriphilus*, and provides evidence for *O. labotus* host shift to an invasive gall wasp and its introduced parasitoid. Collectively, our work is improving our understanding of interactions between native and introduced gall parasitoids and could lead to improved management of the Asian chestnut gall wasp in cultivated chestnut and forest settings.

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