

Developing a Classical Biological Control Program for *Agrilus planipennis* (Coleoptera: Buprestidae), an Invasive Ash Pest in North America

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The emerald ash borer (EAB), *Agrilus planipennis*, a buprestid beetle native to northeast Asia, was determined as the cause of ash tree (*Fraxinus* spp.) mortality in areas of southern Michigan and Ontario in 2002. Infestations have since been found in Ohio, Indiana, Illinois, Maryland, Virginia, Pennsylvania, West Virginia, Wisconsin, Missouri, and Quebec. Regulatory agencies recently shifted their policy from eradication to management for this pest in North America. Classical biological control with three EAB natural enemies from China is being evaluated for use in the sustained control of EAB in the U.S.

Since 2002, considerable progress has been made in developing an EAB biological control program. During 2002-2004, we evaluated infested ash trees in southeastern Michigan woodlots and parks for EAB parasitoids. Parasitoids confirmed from EAB larvae included native wasps known to attack native *Agrilus* spp: *Atanycolus hicorae*, *A. simplex*, and *Spathius simillimus* (Braconidae), *Phasgonophora sulcata* (Chalcididae), and the exotic wasp *Balcha indica* (Eupelmidae). However, we found that less than 1% of immature EAB were parasitized, and no egg parasitoids were found. This rate of parasitism detected for EAB in the U.S. is much lower compared to parasitism of EAB observed in China (Liu et al. 2007), as well as those reported in the literature for our native *A. anxius* (Loerch & Cameron 1983). The lack of native natural enemies attacking EAB in the U.S. supported the need for biological control of EAB in North America (Federal Register 2007).

In 2003, we began studying EAB and its natural enemies in ash stands in China. We found three hymenopteran parasitoids for possible use as EAB biocontrol agents in North America: a



Oobius agrili ovipositing in EAB egg. (H-P Liu).

gregarious larval ectoparasitoid *Spathius agrili* (Liu and Liu 2002; Liu et al. 2003; Yang et al. 2005), a gregarious larval endoparasitoid *Tetrastichus planipennisi* (Liu et al. 2003; Yang et al. 2006; Liu et

al. 2007), and a solitary, parthenogenic egg parasitoid *Oobius agrili* (Zhang et al. 2005; Liu et al. 2007).

Oobius agrili (Encyrtidae) was discovered in 2004 in Jilin Province, China (Zhang et al. 2005). In China, *O. agrili* is a solitary and parthenogenic egg parasitoid with at least two generations per year; it spends the winter and spring as a mature larva in EAB eggs, and adult emergence is synchronized with the EAB oviposition period during July and August in the field. We developed laboratory rearing methods and determined the life cycle of *O. agrili* parasitizing EAB eggs at 25°C (Bauer and Liu 2007). We performed no-choice assays with eggs of six *Agrilus* spp., two cerambycid beetles, and four lepidopterans. Overlap in physiological host range was found for three native *Agrilus* spp. with eggs of similar size to EAB. For these three species, paired choice assays revealed *O. agrili* strongly preferred to oviposit in EAB eggs laid on ash than in eggs of other *Agrilus* spp. on their respective host plants (Bauer and Liu 2007).

Tetrastichus planipennisi (Eulophidae) was discovered in 2003 in Jilin and

Liaoning Provinces of China (Liu et al. 2003) and later in Heilongjiang Province (Yang et al. 2006). *T.*



T. planipennisi ovipositing in bark. (H-P Liu)

planipennisi oviposits into the haemocoel of actively-feeding third- and fourth-instar EAB larvae. In China, it completes at least four generations per year and overwinters as mature larvae inside the host gallery. Adults emerge the following spring, with an average of 35 parasitoids (range: 5 - 122) emerging from a single host larva. We developed laboratory rearing methods and determined the life cycle of *T. planipennisi* parasitizing EAB larvae at 25°C (Liu and Bauer 2007). Using no-choice assays, groups of female and male *T. planipennisi* were exposed to actively-feeding larvae of eight buprestid species, five cerambycid species, and a wood-boring sawfly, all implanted in small branches of their respective host plants. We also assayed larvae of a tenebrionid beetle and two lepidopteran species by implantation in small ash branches, and sphingid larvae by exposure on host leaves. *T. planipennisi* rejected all species except actively-feeding EAB larvae implanted in ash branches (Liu & Bauer 2007).

Spathius agrili (Braconidae) was first reported in Tianjin, China (Liu and Liu 2002) where it is a prevalent parasitoid of EAB in stands of *Fraxinus velutina*, an ash species native to Mexico and southwestern U.S. In Tianjin, the emergence of *S. agrili* adults is well synchronized with the availability of third- and fourth-instar EAB larvae, its preferred host stages and completes three generations per year (Yang et al. 2005). Females oviposit through the tree bark, paralyzing the larva and laying a clutch of eggs on the integument. At maturity, larvae of *S. agrili* spin a cocoon and pupate within the host gallery. No-choice laboratory assays of larval wood-boring insects from China and North America showed some overlap in the physiological host



Spathius agrili female (H-P Liu)

range of *S. agrili*, although successful parasitism was significantly lower in non-hosts than in EAB; no borers in genera other than *Agrilus* were attacked. Therefore, we evaluated the ecological host range of *S. agrili* using an olfactometer to determine the attractiveness of certain host plants. We found *S. agrili* was only attracted to *F. pennsylvanica*, *F. velutina*, and willow (*Salix babylonica*) in Y-tube olfactometer tests. In nature, if parasitoids are not attracted to the host tree they will be unlikely to encounter and parasitize the non-target larvae. In China, no *S. agrili* or *T. planipennis* were reared from six species of field-collected *Agrilus* larvae (n = 2074). Considering the combination of evidence from no-choice and olfactometer tests, the lack of *S. agrili* reared from other *Agrilus* spp. in China and that native *Spathius* spp. were rarely reared from EAB in North America, we predict only incidental parasitism on non-EAB *Agrilus* in North America (Gould et al. 2007).

Given the known risk of EAB to North American ash resources, the high potential benefit of these parasitoids in suppressing EAB populations, and the relatively low potential risk to native *Agrilus* spp., permit requests were submitted in January 2007 to USDA APHIS for release of each species in Michigan. An Environmental Assessment was prepared and posted in the Federal Register (2007). After review by researchers, land managers, and the public during a 60-day public comment period, APHIS determined that the potential benefits outweighed the potential risks and issued a "Finding of No Significant Impact" (FONSI) (Federal Register 2007). Release permits were issued at the end of July, and field releases began in central Lower Michigan. In 2007, adult *O. agrili* (female n = 1406) were released in July and August at two sites in Ingham Co., Michigan; adult *T. planipennis* (female n = 1360) were released from July through October at two sites in Ingham Co., MI; adult *S. agrili* (female n = 311) were released in August and September at one site each in Gratiot, Shiawassee, and Oakland Co., Michigan. In 2008, we determined that *S. agrili* was established at one site in Oakland Co. and *O. agrili* was established at two sites in Ingham Co., MI.

In 2007, USDA FS and APHIS, agreed to collaborate on developing an EAB Biological Control Program (EABCL 2008). APHIS completed construction of the EAB Biocontrol Laboratory (EABCL) in Brighton, MI in fall 2008 where all three EAB parasitoids will be reared. Additional parasitoid-release sites will be added annually, and we plan to track release sites for parasitoid establishment, dispersal, and efficacy over the next five years or more.

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Poster presentation