

EMERALD ASH BORER BIOLOGICAL CONTROL

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ABSTRACT

Emerald ash borer (EAB) (*Agrilus planipennis*), an invasive buprestid from northeast Asia, was identified in 2002 as the cause of ash (*Fraxinus*) tree mortality in southeast Michigan and adjacent areas of Ontario, Canada. This destructive beetle apparently arrived in North America via infested solid wood packaging materials from China in the early 1990s near Detroit, MI. This infestation has killed tens of millions of ash trees in Michigan, and infestations are now known in Illinois, Indiana, Kentucky, Maryland, Minnesota, Missouri, New York, Ohio, Pennsylvania, Virginia, West Virginia, and Wisconsin and in areas of Ontario and Quebec in Canada. Eradication of EAB is no longer possible, but management tactics to lessen its negative impact are being developed and evaluated for efficacy. Researchers studying populations of EAB since 2002 in the U.S. have determined natural enemies are scarce here, whereas in China, EAB control agents are prevalent and effective. After extensive research on the biology and host specificity of three EAB parasitoids from China, a USDA interagency classical biological control program is expected to be launched in 2010 (USDA 2010).

EAB natural enemies in the U.S. Research on EAB natural enemies started in 2003 throughout southeastern Michigan (Bauer et al. 2004). Less than 1 percent of EAB larvae were parasitized by five species of parasitic Hymenoptera, and no egg parasitoids were found (Bauer et al. 2008). The larval parasitoids included four native species [*Atanycolus hicorae*, *A. simplex*, *Spathius simillimus* (Braconidae),

Phasgonophora sulcata (Chalcididae)] and one exotic species [*Balcha indica* (Eupelmidae)] (Bauer et al. 2008). In a more recent study of EAB in western Pennsylvania, two species of eupelmid, *B. indica* and *Eupelmus pini*; two braconids, *Atanycolus nigropyga* and *Spathius laflammeii*; and one ichneumonid, *Dolichomitus vitticrus*, parasitized ~3.7 percent of EAB larvae (Duan et al. 2009). In an Ohio woodlot, ~2.8 percent EAB larvae were parasitized by *Leluthia astigma*, another native braconid species (Kula et al. in press). Except for the two eupelmid species (*B. indica* and *E. pini*), these wasps are reported in the literature as parasitoids of native *Agrilus* spp. Moreover, except for *P. sulcata*, which is an endoparasitoid, all are ectoparasitoids of late-instar larvae. Although the diversity of EAB parasitoids is relatively high and variable from site to site and from year to year, reports of prevalence remain substantially lower than for EAB in northeast China (Liu et al. 2007) and for native *Agrilus* spp. in North America. One exception, however, was the discovery in 2007-2008 of EAB larval parasitism ranging from 9 to 71 percent by *Atanycolus cappaerti* at two sites in southeast Michigan (Cappaert and McCullough 2009). This confirms that some native parasitoids have the potential to switch from native hosts to EAB.

EAB natural enemies in China. There are three important natural enemies of EAB in China: *Tetrastichus planipennis* (Eulophidae), *Spathius agrili* (Braconidae), and *Oobius agrili* (Encyrtidae) (Liu et al. 2003, Zhang et al.

2005). *T. planipennisi*, a gregarious koinobiont endoparasitoid of EAB larvae, overwinters as mature larvae in EAB galleries and produces an average of 68 adults per host larva. In areas north of Beijing, *T. planipennisi* is the most prevalent and widespread parasitoid of EAB. *O. agrili* is a parthenogenic, solitary parasitoid of EAB eggs up to ~13 days after they are laid; it diapauses as a mature larva inside EAB egg and likely completes two generations per year. At one field site in Jilin province, where the distribution of *T. planipennisi* and *O. agrili* overlap, parasitism suppressed EAB populations by ~75 percent on infested green ash trees (*F. pennsylvanica*) (Liu et al. 2007). *S. agrili*, a gregarious idiobiont ectoparasitoid of late-instar EAB larvae, produces an average of 8 adults per host larva and is found more commonly in ash trees south of Beijing.

EAB biological control starts in Michigan. By 2007, research findings on parasitoid biology, laboratory rearing, host specificity, and risk assessment were completed, and results were submitted to APHIS with requests for permits to release the three EAB parasitoids from China in Michigan. Our results were compiled into an environmental assessment and posted on the Federal Register for public comment. After review by researchers, land managers, and the public, APHIS issued a finding of no significant impact (Federal Register 2007) and granted release permits for the three parasitoid species in Michigan in late July 2007.

Evaluating parasitoid reproduction and overwintering. The objective of the 2007 parasitoid releases was to assess reproduction and overwintering capability of these parasitoid species in central Lower Michigan. Parasitoid release sites with high ash densities were selected on state and township lands. The ash trees showed characteristics of building EAB populations (some canopy decline and woodpecker feeding on mid to upper trunks), but had low larval densities in the lower trunks based on sampling using bark windows. From Forest Service laboratory colonies, *O. agrili* (~700 females) were released

in July and August and *T. planipennisi* (~600 mated females + some males) were released in July through September at two different sites in Ingham County. Reared by the APHIS Otis CPHST laboratory, *S. agrili* (~100 mated females + some males), were released in September at one site each in Gratiot, Oakland, and Saginaw Counties. The parasitoids were released as adults on GPS-marked epicenter trees at each site. In winter 2008, four release trees were felled and cut into logs for parasitoid recovery. For the two larval parasitoids, every other log section was peeled, and immature EAB and parasitoids were reared in the laboratory for parasitoid emergence. The other log section was placed in a cardboard rearing tube for emergence of adult EAB and parasitoids. For the egg parasitoid, *O. agrili*, release-tree logs were either (1) inspected for EAB eggs, which were then removed and held for parasitoid emergence in Petri dishes, or (2) placed in cardboard tubes for emergence. We successfully recovered *O. agrili* from one of the Ingham County sites and *S. agrili* from the Oakland County site, confirming field reproduction and overwintering in Michigan.

Establishing release sites to study parasitoid establishment, dispersal, and impact. In 2008, we selected EAB-infested sites in Michigan, Indiana, and Ohio where the three parasitoid species were released for long-term monitoring of parasitoid establishment and efficacy; three of the Michigan parasitoid-release sites are being monitored with cohort life tables and other methods. Due to successful research on *O. agrili* diapause induction, ~2,100 were available for release during EAB's egg laying period; these were released in June at Indiana and Ohio sites and in July through August at Michigan sites. However, fewer than anticipated larval parasitoids were available for release because of unexpected rearing problems at the two laboratories; thus ~600 mated *T. planipennisi* females (+ some males) were released from June through October at Michigan sites, and ~300 mated *S. agrili* females (+ some males) were released in August at Michigan and Ohio sites. For recovery of larval parasitoids in 2008, trees were sampled and peeled during either the fall or winter after release;

egg parasitoid recovery was done by sampling EAB eggs from ash bark and rearing them in the laboratory. At the three life table parasitoid-release sites, trees were randomly selected and dissected in July 2009, resulting in the recovery of *O. agrili* and *T. planipennisi*. We also recovered *T. planipennisi* ~800 m away from the release epicenters at two of our release sites by dissection of heavily infested ash trees.

In 2009, we were able to increase the number of parasitoids released per site, due to better rearing methods, and construction and staffing of the new APHIS EAB Biocontrol Laboratory in Brighton, MI, completed in January. After technology transfer of the rearing methods, transfer of parasitoid stock cultures, periodic training, and trouble shooting, the personnel at the EAB Biocontrol Laboratory successfully reared ~7,000 *S. agrili* females (+some males) and ~1,000 *T. planipennisi* females (+some males) for field release this summer. The combined total from the three laboratories resulted in release of ~8,000 mated *S. agrili* females (+some males), ~20,000 mated *T. planipennisi* females (+some males), and ~5,200 *O. agrili* at research sites in Michigan, Ohio, Indiana, Illinois, and Maryland.

At release sites, we are monitoring for parasitoid reproduction, overwintering success, establishment, spread, and prevalence. This is done by dissecting infested trees for EAB and parasitoids; checking emergence traps stapled to tree trunks; setting out sentinel logs with EAB eggs or EAB larvae; and caging the larval parasitoids on tree trunks with EAB larval insertions. We are also collecting data to evaluate the impact or efficacy of biocontrol by comparing changes in ash condition and EAB densities in parasitoid-release vs. control (non-release) sites. Using hand-held computers with high resolution GPS, parasitoid-release and recovery data are being recorded for each tree. For 50 ash trees (<25 cm d.b.h.) at each release and control site in Michigan and Ohio, the following data were collected in 2008 and 2009: species, d.b.h., crown class, epicormic shoots, woodpecker feeding, and EAB exit holes.

Life table study. In 2008, we started cohort life table studies to determine stage-specific mortality of EAB by each parasitoid species. This 3-year study is being replicated at three parasitoid-release and control sites in Ingham County, Michigan. For the last 2 years, parasitoids were released on ash trees with EAB egg and larval cohorts. In 2008 and 2009, *O. agrili* was recovered at low prevalence (<1 percent parasitism) from eggs sampled at the three release sites; none were detected at control sites. To assess establishment of the larval parasitoids at our life table sites by 2009, trees were randomly selected and peeled, and larvae were reared to determine parasitoid prevalence. In fall 2009, *T. planipennisi* was recovered from ~10 percent of EAB at the three release sites. At two of the sites, EAB larvae parasitized by *T. planipennisi* were found about 800 m from their initial release points, suggesting good dispersal potential for this species.

In 2009, we also started field research to observe and detect parasitism in the field and determine overwintering survival of the two larval parasitoids, *T. planipennisi* and *S. agrili*.

Literature Cited

- Bauer, L.S.; Liu, H-P; Haack, R.A.; Petrice, T.R.; Miller, D.L. 2004. **Natural enemies of emerald ash borer in southeastern Michigan.** In: Proceedings of the emerald ash borer research and technology meeting, Port Huron, MI. FHTET 2004-02. U.S. Department of Agriculture, Forest Service, Forest Health Technology Enterprise Team: 33-34. <http://www.nrs.fs.fed.us/pubs/4571>
- Bauer, L.S.; Liu, H-P; Haack, R.A.; Gao, R-T; Zhao, T-H.; Miller, D.L.; Petrice, T.R. 2005. **Update on emerald ash borer natural enemies in Michigan and China.** In: Proceedings of the emerald ash borer research and technology meeting, Romulus, MI. FHTET-2004-15. U.S. Department of Agriculture, Forest Service, Forest Health Technology Enterprise Team: 71-72. <http://www.nrs.fs.fed.us/pubs/9610>
- Bauer, L.S.; Liu, H-P; Miller, D.L.; Gould, J.R. 2008. **Developing a classical biological control program for *Agrilus planipennis***

- (Coleoptera: Buprestidae), an invasive ash pest in North America. Newsletter of the Michigan Entomological Society. 53: 38-39.<http://www.nrs.fs.fed.us/pubs/1439>
- Cappaert, D.; McCullough, D.G. 2009. **Occurrence and seasonal abundance of *Atanycolus cappaerti* (Hymenoptera: Braconidae), a native parasitoid of emerald ash borer, *Agrilus planipennis* (Coleoptera: Buprestidae).** The Great Lakes Entomologist. 42: 16-29.
- Duan, J.; Fuester, R.W.; Wildonger, J.; Taylor, P.B.; Barth, S.; Spichiger, S.E. 2009. **Parasitoids attacking the emerald ash borer (Coleoptera: Buprestidae) in western Pennsylvania.** Florida Entomologist. 92: 588-592.
- Federal Register. 2007. **Emerald ash borer; availability of an environmental assessment.** <http://www.regulations.gov/fdmspublic/component/main?main=DocketDetail&d=APHIS-2007-0060>
- Kula, R.R.; Knight, K.S.; Rebbeck, J.; Cappaert, D.L.; Bauer, L.S.; Gandhi, K.J.K. In press. ***Leluthia astigma* (Ashmead) (Hymenoptera: Braconidae: Doryctinae) as a parasitoid of *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae: Agrilinae) in Ohio, with a review of *Leluthia Cameron*.** Proceedings of the Entomological Society of Washington.
- Liu, H-P; Bauer, L.S.; Gao, R-T.; Zhao, T.H.; Petrice, T.R.; Haack, R.A. 2003. **Exploratory survey for the emerald ash borer, *Agrilus planipennis* (Coleoptera: Buprestidae), and its natural enemies in China.** The Great Lakes Entomologist. 36: 191-204
- Liu, H-P.; Bauer, L.S.; Zhao, T.H.; Gao, R-T.; Song, L-W.; Luan, Q-S.; Jin, R-Z.; Gao, C-Q. 2007. **Seasonal abundance and population dynamics of *Agrilus planipennis* (Coleoptera: Buprestidae) and its natural enemies *Oobius agrili* (Hymenoptera: Encyrtidae) and *Tetrastichus planipennis* (Hymenoptera: Eulophidae) in China.** Biological Control. 42: 61-71.
- Ulyshen, M.D.; Duan, J-J.; Bauer, L.S. 2010. **Interactions between *Tetrastichus planipennis* and *Spathius agrili*, biocontrol agents for the emerald ash borer.** Biological Control. 52: 188-193.
- USDA. 2010. **USDA emerald ash borer biological control: 5-year implementation strategy.** http://nrs.fs.fed.us/disturbance/invasive_species/eab/local-resources/downloads/eab-biocontrol-5yr-strategy_plan.pdf
- Zhang, Y-Z.; Huang, D-W.; Zhao, T-H.; Liu, H-P.; Bauer, L.S. 2005. **Two new species of egg parasitoids (Hymenoptera: Encyrtidae) of wood-boring beetle pests from China.** Phytoparasitica. 53: 253-260.