Systematics and Biology of *Agrilus planipennis* Fairmaire (Emerald Ash Borer) and its relatives: a new USDA ARS-FS international initiative

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Abstract We present an overview of a 3 year project between the International Programs of the USDA Forest Service (FS) and the Agriculture Research Service (ARS) with some of the preliminary research results of the first 4 months. The ultimate research goals of this project are to define the species group to which Emerald Ash Borer (EAB), *Agrilus planipennis* (Coleoptera: Buprestidae) belongs, provide an identification manual of all related species, and provide workshops in China and the United States to teach others how to recognize EAB and related species of metallic woodboring beetles.

Figure 5. Zeiss Discovery V.20 Stereomicroscope and automontage imaging station.

Figure 6. Clearing larvae for dissection and mounting after Alexeev technique.

Figure 7. Morphological study.

Figure 8. Museum of Natural History, London, UK. Main entrance. Houses important *Agrilus* material.

Figure 9. Buprestidae collection, Muséum national d'Histoire naturelle.

Figure 10. Box containing numerous *Agrilus* species. Red labels indicate type specimens. Natural History Collection, Prague, Czech Republic.

Figure 11. Natural History Museum, Prague, Czech Republic. Houses one of the most important *Agrilus* collections in the world.

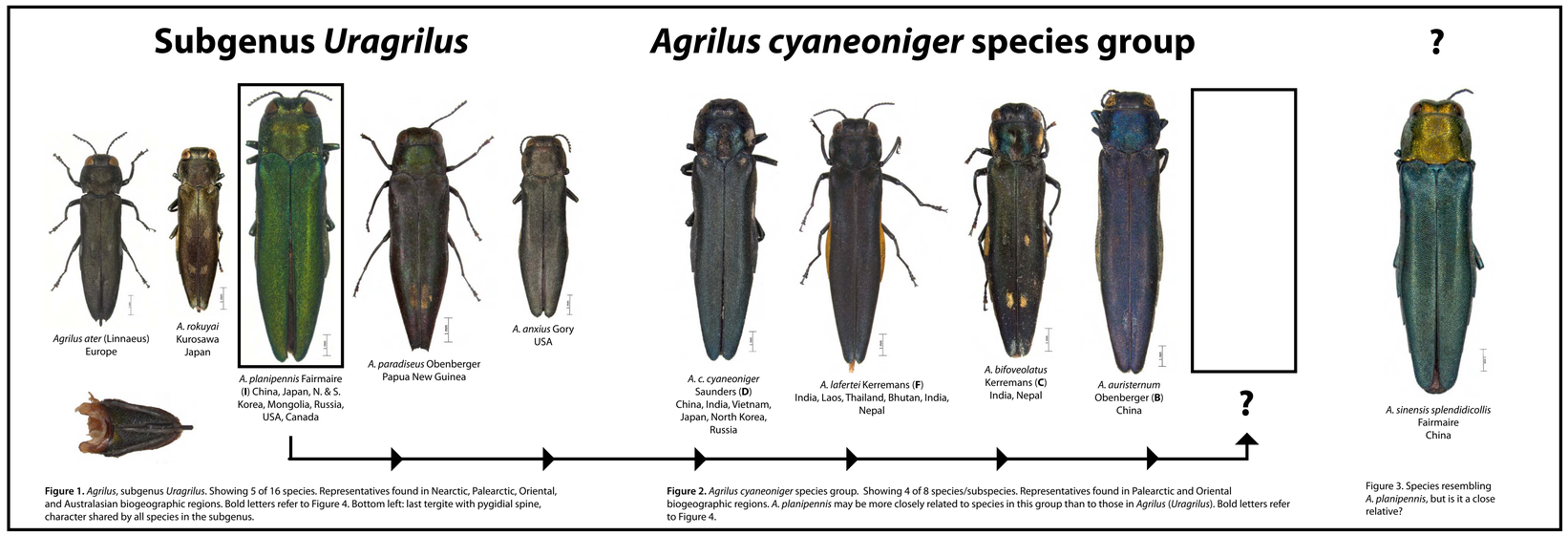

Figure 1. *Agrilus*, subgenus *Uragrilus*. Showing 5 of 16 species. Representatives found in Nearctic, Palearctic, Oriental, and Australasian biogeographic regions. Bold letters refer to Figure 4. Bottom left: last tergite with pygidial spine, character shared by all species in the subgenus.

Figure 2. *Agrilus cyaneoniger* species group. Showing 4 of 8 species/subspecies. Representatives found in Palearctic and Oriental biogeographic regions. *A. planipennis* may be more closely related to species in this group than to those in *Agrilus* (*Uragrilus*). Bold letters refer to Figure 4.

Figure 3. Species resembling *A. planipennis*, but is it a close relative?

Figure 12. *A. c. cyaneoniger*, dorsal view.

Figure 13. *A. c. cyaneoniger*, lateral view.

Figure 14. *A. planipennis*, dorsal view.

Figure 15. *A. planipennis*, lateral view.

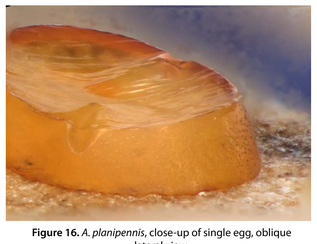

Figure 16. *A. planipennis*, close-up of single egg, oblique lateral view.

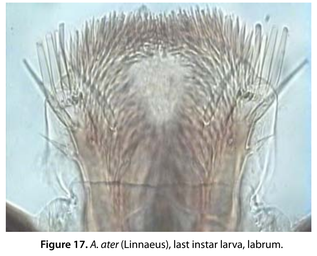

Figure 17. *A. ater* (Linnaeus), last instar larva, labrum.

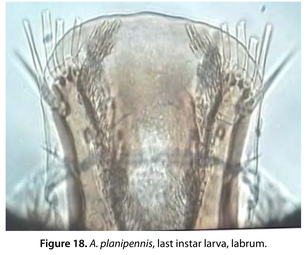

Figure 18. *A. planipennis*, last instar larva, labrum.

Introduction

Problem: lack of information about the potentially invasive species related to the Emerald Ash Borer.

Solution: develop an illustrated handbook to aid detection and identification of adults and larvae of the Emerald Ash Borer and its relatives rooted in cladistic principles.

Goals:
1. Determine, define, and illustrate the characters that enable identification of EAB and related species;
2. Organize workshops in China and USA to tranfer technology to make identifications;
3. Disseminate the information resulting from research.

This effort will mitigate future incursions of related species of these wood-boring beetles into the USA and build long-term collaborative relationships with Chinese colleagues and enhance expertise and collections in China.

Expected outcomes:
1. Ability to distinguish between native and invasive *Agrilus* species;
2. Create a biological reference to recognize invasive species;
3. Establish a Sino-American EAB biosystematic network;
4. Transfer information to Chinese biologists;
5. Educate the public and other scientists about the relatives of EAB;
6. Contribute knowledge needed to develop control strategies to manage outbreaks.

Background on *Agrilus*

- **2784** species, 99 subspecies (Bellamy 2008).
- Family Buprestidae, group commonly known as Jewel Beetles because of their metallic colors and striking patterns.
- Palearctic and Afrotropical *Agrilus* have been classified into 36 subgenera.
- However, these subgenera may not reflect evolutionary history and circumscription of the group focused on a narrow biogeographic area (Jendek 2006).
- Many of these species may be grouped together based on convergent features rather than ones inherited from a most recent common ancestor (Fig. 1).
- EAB is currently classified in the subgenus *Uragrilus* (Fig. 1).
- EAB may be closely related to species included in the *Agrilus cyaneoniger* species group (Haack *et al*. 2002; Eduard Jendek, pers. comm) (Figs 2, 4).

Figure 4. Distribution of species of *Agrilus* putatively related to the Emerald Ash Borer [the *A. cyaneoniger* group defined in Jendek (2000)]: **A**) *Agrilus agnatus* Kerremans; **B**) *Agrilus auristernum* Obenberger; **C**) *Agrilus bifoveolatus* Kerremans; **D**) *Agrilus cyaneoniger cyaneoniger* Saunders; **E**) *Agrilus cyaneoniger melanopterus* Solsky; **F**) *Agrilus lafertei* Kerremans; **G**) *Agrilus lubopetri* Jendek; **H**) *Agrilus qinling* Jendek; **I**) *Agrilus planipennis* Fairmaire.

Materials & methods

Phylogeny:
- Evolutionary relationships will be inferred based on morphology and DNA-sequence data. The following hypotheses will be tested:
  1. Species in *Uragrilus* are the closest relatives of EAB;
  2. Species in *Agrilus cyaneoniger* group are the closest relatives of EAB;
  - Representatives from other subgenera and species of uncertain placement from the Oriental, Neotropical and Nearctic biogeographic regions will also be included in the analysis.

Morphology:
- Detailed study of morphological characters of adults and immatures will form the basis of the identification manual and phylogenetic analysis (Figs 12-18).

Taxonomy:
- Study type material and authoritatively identified specimens (Figs 8-11).
- Capture high quality images to include in the identification manual (Fig. 5).

Workshops:
- Transfer information on EAB and relatives
  - 2012 Beijing, China; 2013 Washington, DC, USA.

Fieldwork:
- 2011 Yunnan, China; Japan; Vietnam; Laos; 2012 China; Thailand; India.

Progress and future directions

Phylogeny:
- Character matrix with more than 100 characters and 8 species constructed. Additional characters and exemplar species will be included.
- DNA sequencing will begin from material collected during 2011, 2012 fieldwork.

Morphology:
- Morphological characters of the adult, such as shape of the scutellum, pronotum, elytral apices, and submarginal carina, among others, are of particular diagnostic and phylogenetic importance (Figs 12-15).
- The first comprehensive study of the female genitalia in the genus is underway.
- A detailed description of the immature stages of EAB is currently underway (Figs 7, 16-18).

Taxonomy:
- Museums in Europe were visited and numerous types and species borrowed for study (Figs 8-11).

References cited

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