FINAL REPORT FOR CDFA AGREEMENT Number 11-0147-SA

8 March 2013

PROJECT TITLE

VOUCHERING SPECIMENS OF EGG PARASITOIDS OF THE GLASSY-WINGED SHARPSHOOTER COLLECTED BY THE CDFA PIERCE'S DISEASE BIOLOGICAL CONTROL PROGRAM IN CALIFORNIA AND TEXAS A&M IN TEXAS

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Reporting Period: The results reported here are from work conducted from 1 July 2011 to 8 March 2013.

INTRODUCTION

Recently (since 1997), major efforts have been undertaken by the CDFA GWSS Biological Control Program to survey for egg parasitoids of glassy-winged sharpshooter, *Homalodisca vitripennis* (Germar) (GWSS), in California and to release several egg parasitoid species (*Anagrus epos* Girault, *Gonatocerus* spp.) from other states in the USA and also northeastern Mexico as part of the classical biological control effort (CDFA 2012).

It is well known that the taxonomic impediment in identification of natural enemies may adversely affect the biological control efforts against agricultural pests. In the case of the GWSS, early misidentifications (due to objective reasons, such as partially inaccurate existing keys) of one of the species of the California native egg parasitoids of GWSS, such as *Gonatocerus morrilli* (Howard), resulted in the inability of biological control practitioners to distinguish them from the introduced "real" *G. morrilli* from Texas and northwestern Mexico. Therefore, contamination of the colonies in the mass-rearing program was noticed only after the molecular methods distinguished them as two genetically different entities. The "California *G. morrilli*" was later described taxonomically as a new species, *Gonatocerus walkerjonesi* Triapitsyn, based on the combination of molecular evidence and some morphological differences that are difficult to observe without special preparation of the specimens (Triapitsyn 2006). Another native species, *Gonatocerus morgani* Triapitsyn, was also described from Orange Co., CA (Triapitsyn 2006); it is now being mass-produced and released in other parts of California infested with GWSS (Son et al. 2011). As proper part of the ongoing biological control program against GWSS, the CDFA GWSS Biological Control Program has conducted extensive pre- and post-surveys of the egg parasitoids of GWSS in California from 2001. These surveys, which also included egg parasitoids of the native sharpshooter in California, the smoke-tree sharpshooter *Homalodisca liturata* Ball, have resulted in collection of more than 10,000 specimens of egg parasitoids (Mymaridae and Trichogrammatidae) which are stored, along with voucher specimens of the numerous colonies of GWSS egg parasitoids maintained by the CDFA, in several thousand vials at the CDFA Mt. Rubidoux Field Station in Riverside, CA. Also, Dr. Forrest L. Mitchell has kindly donated to the CDFA

GWSS Biological Control Program 930 vials of GWSS parasitoids collected in Fredricksberg Co., TX, by Texas A&M staff. These insects were collected since 2005, each vial containing parasitoids that have emerged from a single egg mass, with many individuals per vial. Date and location from which each mass emerged have been recorded.

SUMMARY OF MAJOR ACTIVITIES AND ACCOMPLISHMENTS AND RESULTS FOR EACH OBJECTIVE

(Objective 1) Check the taxonomic identities of all the specimens of GWSS egg parasitoids from California; pull out specimens of taxonomic and voucher interest.

Activities and accomplishments: The PI has identified thousands of specimens from California and checked their taxonomic identities in more than 7,500 vials (arranged in boxes in trays, Fig. 1) and pulled out the voucher specimens of taxonomic interest for drying from alcohol and point-mounting, followed by their labeling and databasing. Particularly, all the specimens of *Ufens* (Trichogrammatidae) were identified to species: two are known in North America as GWSS egg parasitoids (Triapitsyn 2003; Al-Wahaibi et al. 2005). In addition, 10 specimens of each species (both sexes if available) were pulled out by the Jessica Nichols (CDFA) from the collections in California; these were dried out from ethanol and then mounted on both points and slides by Vladimir Berezovskiy (a technician employed by the project) and identified to genus and species by the PI. A written report on those identities was submitted by the PI to Dr. David Morgan and Jessica Nichols (Integrated Pest Control Branch, Biological Control Program, CDFA).

The following species of GWSS egg parasitoids were identified from California (Triapitsyn 2012): *Gonatocerus* (*Cosmocomoidea*) *ashmeadi* Girault, *G.* (*Cosmocomoidea*) *fasciatus* Girault (intentionally introduced, from a few release sites only), *G.* (*Cosmocomoidea*) *incomptus* Huber, *G.* (*Cosmocomoidea*) *morgani* Triapitsyn, *G.* (*Cosmocomoidea*) *morrilli* (Howard) (intentionally introduced), *G.* (*Cosmocomoidea*) *novifasciatus* Girault, *G.* (*Cosmocomoidea*) *triguttatus* Girault (intentionally introduced), and *G.* (*Cosmocomoidea*) *walkerjonesi* Triapitsyn (Mymaridae), as well as *Ufens ceratus* Owen and *U. principalis* Owen (Trichogrammatidae).

Newly discovered and reported in California (four females from Riverside) is an apparently undescribed species of the genus *Pseudoligosita* Girault (Trichogrammatidae), which is morphologically different from *P. plebeia* (Perkins) from Sonora, Mexico, reported as *Pseudoligosita* sp. by Triapitsyn & Bernal (2009) and later identified as such by the PI. Also of taxonomic interest is an undescribed species of *Polynema* Haliday (subgenus *Doriclytus* Foerster) which was quite rare among the reared egg parasitoids of GWSS in California (only one female and two males from Pomona and two females from Riverside). The latter species was quite commonly collected in southern California by other (non-reared) methods. Most likely GWSS is not a primary host for both aforementioned species, and they attack its eggs only occasionally.

(**Objective 2)** Transfer of the bulk of the voucher specimens from Texas into leak-proof vials for long-term storage; label properly (using archival, acid-free paper) and database all the vials using barcodes with unique numbers. Fully identify and catalog approximately 7,000 GWSS parasitoids collected by Texas A&M in the native range of GWSS.

Activities and accomplishments: All 930 vials with GWSS egg parasitoids received from Texas AgriLife Research and Extension Center at Stephenville were completely curated by the two technicians employed by the project: these were transferred from the original poor vials into leak-proof vials for long-term storage; labeled properly (with the data label inside and the identification label and the database number and a bar code outside of each vial), and databased using barcodes with unique UCRC ENT numbers, and arranged into boxes for storage (Figs 2, 3). At the end of this project, these have been turned over to Dr. David Morgan (CDFA Mt. Rubidoux Field Station in Riverside, Integrated Pest Control Branch, Biological Control Program, CDFA), for the eventual deposition in the California State Collection of Arthropods in Sacramento.

The following species of GWSS egg parasitoids were identified from Texas (Triapitsyn 2012): *Gonatocerus* (*Cosmocomoidea*) *ashmeadi* Girault (very common), *G.* (*Cosmocomoidea*) *incomptus* Huber (quite rare), *G.* (*Cosmocomoidea*) *morrilli* (Howard) (less common), *G.* (*Cosmocomoidea*) *novifasciatus* Girault (quite rare), *G.* (*Cosmocomoidea*) *triguttatus* Girault (common) (Mymaridae), as well as *Burksiella spirita* (Girault) and *Ufens ceratus* Owen (Trichogrammatidae, both quite common).

(Objective 3) Prepare valuable representatives of each species that will be dried from ethanol using a critical point dryer, point-mounted as museum quality vouchers, labeled, and databased.

Activities and accomplishments: Representatives of each species (at least 50 specimens of the common species from Texas, as many as possible of the less common species, all specimens of the rare species, and also 10 specimens of each species pulled out by Jessica Nichols from the collections in California) were critically point dried from ethanol and point-mounted. Then representatives of both sexes of each species (both sexes) were selected and slide-mounted in Canada balsam (Fig. 4). All the mounted specimens were properly labeled using acid-free archival paper and databased; this database was made available online among the UCRC records submitted to Discover Life (www.discoverlife.org). Most voucher specimens from Texas have been submitted the CDFA Mt. Rubidoux Field Station in Riverside (Dr. David Morgan) to be eventually transferred for storage at the California State Collection of Arthropods in Sacramento, its permanent depository; a few duplicate representatives and taxonomically important specimens (such as specimens of the new species to be described by the PI eventually) have been deposited in UCRC.

PUBLICATIONS OR REPORTS RESULTING FROM THE PROJECT

Triapitsyn, S.V. 2011. Vouchering specimens of egg parasitoids of the glassy-winged sharpshooter collected by the CDFA Pierce's Disease Biological Control Program in California and Texas A&M in Texas 2011. In: *Proceedings of the 2011 Pierce's Disease Research Symposium, December 13–15, 2011, Sheraton Grand Sacramento Hotel, Sacramento, California, organized by California Department of Food and Agriculture* (Esser, T., Chief Ed., West, D., Associate Ed.). Time Printing, Inc., Sacramento, California, pp. 17-18. Available online at http://www.cdfa.ca.gov/pdcp/Research_Symposium_Index.html.

Triapitsyn, S. V. 2012. Vouchering specimens of egg parasitoids of the glassy-winged sharpshooter collected by the CDFA Pierce's Disease Biological Control Program in California and Texas A&M in Texas. In: *Pierce's Disease Research Progress Reports, December 2012, Pierce's Disease Control Program, California Department of Food and Agriculture*, *Sacramento, California* (Esser, T., Chief Ed., Randhawa, R., Associate Ed.), pp. 8-11. Available online at http://www.cdfa.ca.gov/pdcp/Research.html.

PRESENTATION ON RESEARCH

The PI made a presentation, as a speaker, at the 2011 Pierce's Disease Research Symposium, December 13–15, 2011, Sheraton Grand Sacramento Hotel, Sacramento, California, organized by California Department of Food and Agriculture, entitled: "Egg parasitoids of sharpshooters in the New World".

RESEARCH RELEVANCE STATEMENT

Specimens from these collections have been curated to preserve the invaluable voucher specimens for further analyses (including molecular, distributional, taxonomic, biological, etc.), thus making them available. The specimens and information on them will be useful for the California Department of Agriculture GWSS/PD Biological Control Program and biological control research practitioners in this state and beyond.

LAY SUMMARY OF PROJECT ACCOMPLISHMENTS

This project is now complete; its main objectives were to label (using archival paper and unique plastic database numbers with barcodes), identify, database (including georeferencing), preserve, partially dry from ethanol, and point- and slide-mount specimens among at least 17,000 voucher specimens of mymarid and trichogrammatid egg parasitoids of GWSS. These were either collected (reared) by the CDFA Pierce's Disease Biological Control Program personnel in California since 2001 in the course of pre- and post-release surveys, are irreplaceable vouchers of the colonies of the biological control agents that were released in California (both exotic and native), or were collected by staff of Texas A&M in Texas since 2005. Taxonomic identifications were checked and, when necessary, specimens were identified by the PI for the California material (in more than 7,500 vials); particularly, all Trichogrammatidae were identified to genera and species. Specimens from Texas (in 930 vials) were all identified to genera and species and were transferred into leak-proof vials with good caps to prevent alcohol leakage, labeled properly, and databased using University of California, Riverside Entomology Research Museum (UCRC) numbering system which has been made available online. Valuable representatives of each species were dried from ethanol using a critical point dryer and point-mounted (and

representatives of both sexes slide-mounted) as museum quality voucher specimens, and also were labeled using acid-free archival paper and databased. Most voucher specimens from Texas have been submitted to the CDFA Mt. Rubidoux Field Station in Riverside to be eventually transferred for storage at the California State Collection of Arthropods (CDFA) in Sacramento, its permanent depository; some duplicate representatives and a few taxonomically important specimens have been deposited in UCRC to be used for further taxonomic investigations.

STATUS OF FUNDS

A no-cost extension has been granted for this project until 6/30/2013. As of 3/8/ 2013, all funds allocated to this project have been spent, and all work on it has been completed. Therefore, this final report, rather than a progress report due 3/15/2013, is being submitted.

SUMMARY AND STATUS OF INTELLECTUAL PROPERTY PRODUCED DURING THIS RESEARCH PROJECT

Not applicable.

REFERENCES CITED

Al-Wahaibi, A. K., A. K. Owen & J. G. Morse. 2005. Description and behavioural biology of two *Ufens* species (Hymenoptera: Trichogrammatidae), egg parasitoids of *Homalodisca* species (Hemiptera: Cicadellidae) in southern California. *Bulletin of Entomological Research* 95 (3): 275-288.

CDFA. 2012. Biological Control. http://www.cdfa.ca.gov/pdcp/Biological_Control.html. Last accessed 2 October 2012.

Son, Y., H. Nadel, S. Baek, M. W. Johnson & D. J. W. Morgan. 2012. Estimation of developmental parameters for adult emergence of *Gonatocerus morgani*, a novel egg parasitoid of the glassy-winged sharpshooter, and development of a degree-day model. *Biological Control* 60 (3): 233-240.

Triapitsyn, S. V. 2003. Taxonomic notes on the genera and species of Trichogrammatidae (Hymenoptera) - egg parasitoids of the proconiine sharpshooters (Hemiptera: Clypeorrhyncha: Cicadellidae: Proconiini) in southeastern USA. *Transactions of the American Entomological Society* 129 (2): 245-265.

Triapitsyn S. V. 2006. A key to the Mymaridae (Hymenoptera) egg parasitoids of proconiine sharpshooters (Hemiptera: Cicadellidae) in the Nearctic region, with description of two new species of *Gonatocerus*. *Zootaxa* 1203: 1-38.

Triapitsyn, S. V. 2012. Vouchering specimens of egg parasitoids of the glassy-winged sharpshooter collected by the CDFA Pierce's Disease Biological Control Program in California and Texas A&M in Texas. In: *Pierce's Disease Research Progress Reports, December 2012, Pierce's Disease Control Program, California Department of Food and Agriculture*, *Sacramento, California* (Esser, T., Chief Ed., Randhawa, R., Associate Ed.), pp. 8-11. Available online at http://www.cdfa.ca.gov/pdcp/Research.html.

Triapitsyn, S. V. & J. S. Bernal. 2009. Egg parasitoids of Proconiini (Hemiptera: Cicadellidae) in northwestern Mexico, with description of a new species of *Gonatocerus* (Hymenoptera: Mymaridae). *Journal of Insect Science* 9 (5), 1-9. Available online at http://www.insectscience.org/9.05/.

ILLUSTRATIONS

Fig. 1. A tray with CDFA vials with identified GWSS egg parasitoids from California.

Fig. 2. Boxes with curated, labeled, and databased vials with identified GWSS egg parasitoids from Texas.

Fig. 3. Typical curated vials containing GWSS egg parasitoids from Texas.

Fig. 4. Typical slides of the identified GWSS egg parasitoids from California and Texas.