## A MONOCLONAL ANTIBODY SPECIFIC TO GLASSY-WINGED SHARPSHOOTER EGG PROTEIN: A TOOL FOR PREDATOR GUT ANALYSIS

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# ABSTRACT

Egg-specific monoclonal antibodies (MAbs) have been developed to the glassy-winged sharpshooter (GWSS). Younger GWSS eggs yield stronger ELISA reactions than older eggs. The indirect ELISA format was not effective for detecting GWSS egg antigen in whole body or gut dissected earwig specimens. Further ELISA optimization tests are underway to increase the efficiency of the ELISA procedure. A survey of the predator complex inhabiting citrus in Riverside, CA indicates that the earwig, *Forficula auricularia* is the most common predator.

# INTRODUCTION

Effective control of the glassy-winged sharpshooter (GWSS) will require an areawide integrated pest management approach (AW-IPM). A major component of AW-IPM is the exploitation of the pest's natural enemies, which, when utilized to their greatest potential, can increase the effectiveness of other control tactics. Unfortunately, very little information exists on GWSS's predaceous natural enemies. Evidence of predation of GWSS eggs and adults has been observed in the field (JH pers. obs.); however, the composition of the predator complex, and the relative impact of each predator on GWSS mortality is unknown. A major obstacle is the difficulty of studying predators in their natural environment. Unlike parasitoids, predators rarely leave evidence of attack. Laboratory experiments can be used to evaluate the suitability of particular prey and the rates of predation. However, lab studies seldom translate to field situations. Direct field observations are sometimes used to identify predators of key pests, but the small size and cryptic nature of predators and GWSS make direct observations difficult. Furthermore, direct field observations are time consuming, labor intensive, and disruptive to the normal predator foraging process. Microscopic analysis of predator gut contents have been used but the process is not suitable for predators that liquefy prev contents for consumption. These difficulties have resulted in a deficiency of information on the impact that predators have on suppressing key insect pests. A proven "indirect" technique for measuring predation is use of proteinbased immunological assays employing pest-specific monoclonal antibodies (MAbs) (Hagler et al. 1992, Hagler and Naranjo 1994-a,b). To this end, we have developed egg-specific MAbs to GWSS (Hagler et al. 2002). The MAb assays provide an avenue to qualitatively assess the impact of predator species on populations of GWSS eggs.

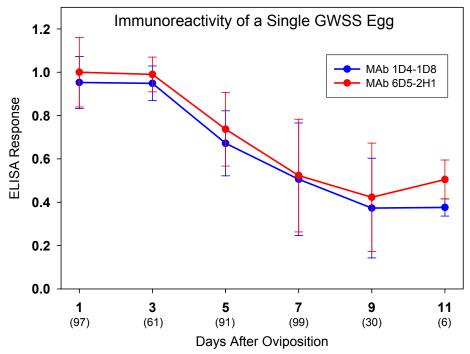
## **OBJECTIVES**

Our ultimate goal is to identify the composition of the GWSS predator complex and to identify the relative impact each predator species has on GWSS using a pest-specific ELISA. Prior to examining the gut contents field-collected predators for GWSS remains, a few ELISA optimization studies are needed. Here we report on tests conducted to determine GWSS-specific ELISA responses to: (1) individual GWSS eggs of known age, and (2) predators that have consumed GWSS eggs. These studies are needed prior to conducting ELISAs on field-collected predators. We also report on the abundance of potential GWSS predators found in citrus.

## RESULTS

# ELISA response to GWSS eggs of known age

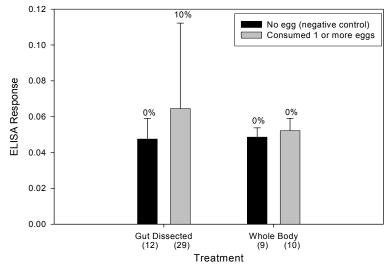
GWSS eggs were assayed by the egg-specific indirect ELISA described by Hagler et al. (2002). Data indicate that the ELISA reactions were greater for younger eggs than older eggs. (Figure 1).



**Figure 1.** Mean (±SD) ELISA reaction yielded by two GWSS egg-specific MAbs to a single egg of known age. The numbers in parenthesis below the x-axis are the sample sizes for each time interval.

#### ELISA response to earwigs that consumed GWSS eggs

Individual adult earwigs, *Forficula auricularia* (Dermaptera: Forficulidae) were placed in Petri dishes with a single GWSS egg mass. An individual earwig was allowed to feed continuously on the egg mass. Once an individual ceased feeding it was removed from the Petri dish and frozen. Each earwig was then analyzed by ELISA for the presence of GWSS egg antigen. Two groups of earwigs were examined by ELISA: (1) whole-body specimens, and (2) gut dissected specimens. Data indicate that none of the whole-body specimens and only 10% of the gut dissected specimens yielded a positive ELISA reaction (Figure 2). Previous work with other insect species has shown that the indirect ELISA format is less sensitive than the sandwich ELISA format for detecting prey remains in large predators (Hagler et al. 1997, Hagler and Naranjo 1997, Hagler 1998). We are developing a sandwich ELISA for use in detecting GWSS remains in earwig guts. Additionally, we are testing the indirect ELISA on other predator species (e.g., *Chrysoperla carnea, Collops vittatus, Hippodamia convergens*, etc.).



**Figure 2.** Mean  $(\pm SD)$  response of the GWSS-specific ELISA to earwigs that consumed one or more GWSS eggs. The number above each error bar is the percentage of individuals from each treatment that yielded a positive ELISA reaction for the presence of GWSS egg antigen. The numbers in parenthesis below the x-axis are the sample sizes for each treatment.

### Arthropods collected from citrus

Total counts of arthropod species inhabiting the citrus canopy in Riverside, CA were obtained from whole-tree samples collected by D. Akey (pers. comm.). Briefly, 3 entire trees were sampled every other week during GWSS outbreaks. The area under each tree was covered by ground cloth and the tree canopy was covered with a waterproof tent. Each tree was fogged with pyrethrum and PIP. After 3 h, the tent was removed, the tree was shaken for 5 min, and the arthropods on the ground cloth were counted. The densities of GWSS nymphs and adults are reported by Akey et al. in this volume. The abundance of the other arthropods collected from trees is given in Figure 3. By far, the most abundant predator species encountered was the earwig, *F. auricularia*. Laboratory feeding trials are underway to determine earwig feeding activity on GWSS eggs (egg consumption rate, handling time, etc.). Various species of beetles, ants, and spiders were also collected from the citrus trees. Gut content ELISAs will be performed on each predator as soon as the ELISA procedure has been optimized.

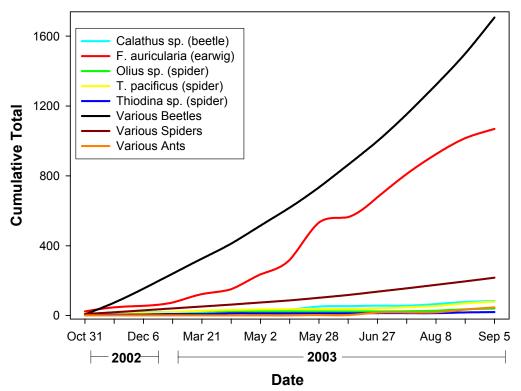


Figure 3. The cumulative population of arthropods collected from citrus trees in Riverside, CA.

## CONCLUSIONS

An understanding of the key natural enemies of GWSS will contribute to an areawide IPM approach for GWSS control. Once key predators are identified they can be better exploited for conservation and augmentative biological control programs.

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