Final Report, Development of a Monitoring and Mass Trapping Tool for Wood-destroying Beetles in Structures.

By

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Background

The project award period was for two years starting September 15, 1997. After delays in final approval and setting up an account number, two extensions were requested and granted. The ending date of the project was December 31, 2001.

Sources of Infested Wood

During the first year of the project, several hundred pieces of wood were collected from the Bay Area revealing signs of woodboring beetles. The most promising source of beetles came from Daly City, California. In September 1998, a large, fifteen-year-old Douglas-fir deck (15 foot by 20 foot) was dissembled and brought back to the Berkeley campus. Visual inspection of the wood suggested that California deathwatch beetles had infested the deck. A local pest control company also provided infested boards.

Rearing Boxes

Two large wooden boxes (4 foot by 4 foot by 4 foot, 64 cubic feet) were constructed. The boxes had screened bottoms, black lights, collection funnels, and interior walls painted white. Humidity in the boxes was maintained at 14-17% by using water sprayers set timers (three to four sprays per day). An additional box (4 foot by 4 foot by 8 foot, 128 cubic feet) was acquired from University of Nevada at Reno. Approximately 200 pieces of wood (mostly 3-foot sections) were put into rearing boxes.

Two woodpiles each containing over one hundred boards were maintained at the Berkeley campus and Forest Products Laboratory. To prevent beetles from threatening wooden structures, colonies were maintained in a glass greenhouse or maintained in an outdoor woodpile far removed from structures.

Beetles

Boards collected from the woodpile were dissected in March and May 1999 and revealed live beetle larvae. The larvae appeared to be anobiid, a family of beetles that contains the California deathwatch beetle. These structural infesting beetles are ranked third in economic importance in the State in the damage they cause. Only decay and termites cause more damage to structures.

Adult beetles began emerging from the boards in the rearing boxes during the last week of May 1999. Samples were sent to the State beetle expert, Dr. Fred Andrews (Pest Diagnostic Laboratory, California Department of Food and Agriculture, Sacramento) for species

determination. Dr. Andrews determined the samples to the California deathwatch, *Hemicoelus gibbicollis*.

During summer 2000, the California deathwatch beetle, *H. gibbicollis* was difficult to collect from Bay Area structures. Instead, a related deathwatch beetle, *Ptilinus basalis* LeConte, was collected from an infested California laurel (bay) tree (*Umbellularia californica*) in Morgan Hill, California. Although *Ptilinus basalis* is a different species from *H. gibbicollis* (structure infesting species), both beetles are in the same family, and we assumed that by working with *P. basalis* we would gain insight into the pheromone biology of *H. gibbicollis*. From April through June, over one thousand beetles were collected from rearing boxes on the UC-Berkeley campus.

Pheromone Chemistry

To provide a crude extract of female deathwatch attractants, the ovipositors (egg-laying tubes) of 25 female beetles were removed and placed in methylene chloride. Female ovipositors are used for egg laying and also are reportedly the site of pheromone production in this beetle species and in others that occur overseas. Pheromones are chemicals that deathwatch beetles use to communicate their location for mating. Air samples containing pheromones from adult virgin females have also been collected. Both samples have been shipped to Dr. Steven Seybold at the University of Minnesota for chemical analysis. Ovipositors and air samples were frozen to -80 ^{0}C and analyzed using gas chromatography and mass spectrometry to identify attractants and pheromones. Stegobinone was found in the ovipositors of female California deathwatch beetles, *H. gibbicollis.* Stegobinone is a frequently reported sexual attractant (pheromone) used by a number of wood boring and stored product beetles in the family Anobiidae.

Bioassay Method

Using dissecting microscopes, we removed the ovipositor (egg-laying organ) from several hundred female beetles and placed the tissue into an ether/hexane solution. A small amount of this solution was injected into a gas chromatograph (GC), which is an instrument used in the laboratory to analyze the chemical composition of biological and environmental samples. Gas chromatography can be coupled with another technique called mass spectrometry (MS) to definitively determine chemical structures. For example, this technique is widely used in drug testing. By GC-MS analysis we detected stegobinone in the female *Ptilinus* tissue extract. A year earlier we reported this chemical from the California deathwatch beetle, *H. gibbicollis*. Next we sent our samples and live beetles to a cooperating scientist, Dr. Allard Cossé, located in Peoria, Illinois at the United States Department of Agriculture Center for Agriculture Utilization Research. Dr. Cossé is an internationally recognized expert at determining whether or not prospective pheromone chemicals cause electrical stimulation in the antennae of insects. His GC instrument had an adapter to blow volatile emissions from the female ovipositor extract over the antenna of a male beetle. A very tiny electrode was inserted into the antenna of the male beetle to register its response to the female extract. This bioassay technique generates an output called an electroantennagram (EAG), which is a measure of how males respond to sex pheromones produced by females.

Results

Gas chromatography results show the presence of stegobinone in the crude extracts prepared from female ovipositors (Figure 1). Results from the electroantennagraph (EAG), show male antennae had a spike of activity (recognized) the presence of stegobinone in volatile

extracts from female ovipositors (Figure 2). The red trace shows the time it takes for different chemicals (represented as spikes) in the female extract to pass through the GC machine. The arrow in Figure 2 is the location where stegobinone appears in the trace for females. The black line trace is the simultaneous response of male antennae to crude female extract containing stegobinone when blown over the male's antennae. Notice the male's response lag occurs just after the first appear of stegobinone in the female extract.

Natural Enemies/ Biological Control

Another insect emerged in large numbers from the wood in the rearing boxes. Since the first week of May 1999, 150 tiny wasps belonging to the family Braconidae, have emerged. Samples have been collected and sent to a parasitic insect specialist, Dr. Paul March, Kansas State University, for species determination. Dr. Marsh has identified the wasp as a new species, *Heterospilus luridostigmus*. This is the first report of a natural enemy for the California deathwatch beetle. We believe that the wasp has caused significant mortality in our laboratory colonies.

Outcome of Study

This study has shown for the first time the production and importance of stegobinone in two species of deathwatch beetles. The reporting of a new species of wasp (*Heterospilus luridostigmus*) that may be a parasitic of deathwatch beetles was also an exciting discovery. Future work should focus on determining the exact chemical structure of the stegobinone used by different species of deathwatch beetles. Additional bioassay work must also be conducted to confirm male and possible female response to varying levels of stegobinone or other unreported pheromone components. Lastly, the impact made by parasitic wasps on deathwatch beetles needs to be assessed in the lab and field. Although still in the distant future, this work may lead to improved methods of deathwatch beetle detection and biological control.

Technology Transfer

Educational Presentation. During the last three years, at least 10 presentations on the results of this project have been given around the State. Hundreds of pest control operators (PCOs) in California have heard findings from this study by attending various seminars (e.g., Target Specialty Products Seminars).

Scientific Presentations. One presentation was given to a scientific audience at Cloquet, Minnesota in October 2000, on the response of male deathwatch beetles (*Ptilinus basalis*) to female sex pheromone using electroanntenagraphs. The complete citation of the abstract is listed below.

Cabrera, B., a. Cosse, S. Seybold, and V. Lewis. 2000. Isolation and identification of the sex pheromone stegobinone from the deathwatch beetle, *Ptilinus basalis* LeConte. Presented paper at Biorational Methods for Insect Pest Management: Bio-organic and molecular approaches, Cloquet, MN, Oct. 2000.

Trade Magazine Articles. Two articles updating the research appeared in a statewide pest control magazine. Complete citation is listed below.

Cabrera, B. J., V. R. Lewis, and S. J. Seybold. 2000. Berkeley research on deathwatch beetle finds potential traps, enemies. The Voice PCOC,

Winter 2000, pages 11-12.

Lewis, V. R., B. J. Cabrera, and S. J. Seybold. 2001. Update on deathwatch beetle study: New discoveries. The Voice of PCOC, Spring 2001, in press.

Scientific Publications. One scientific article on the results of the study is currently in review. The citation is listed below.

Cabrera, B. J., P. M. Marsh, V. R. Lewis, and S. J. Seybold. 2001. A new species of *Heterospilus* (Hymemoptera: Braconidae) associated with the deathwatch beetle *Hemicoelus gibbicollis* (LeConte) (Coleoptera: Anobiidae). Submitted to Pan-Pacific Entomologist.

Publications in preparation. At least one paper on the results of male beetle bioassays using electroantennagraphs is being prepared for submission to a scientific journal.

Budget Information

The accounting unit at the Forest Products Laboratory is currently tabulating all project expenditures. Fiscal closing and final billing for the project will be completed by March 31, 2001.

Addenda. Copies of all published papers, abstracts, and submitted manuscripts are included as addenda.

Acknowledgments. The authors wish to thanks the efforts of the laboratory crew, Salvador Gracia-Rubio, Gee Chow, Leon Daniels, and Robyn Raban for their efforts. Special thanks goes to **LANA???** for help in GL analysis and **??????(guy at U of M who helped me)** for NMR analysis, both at the University of Minnesota. Mr. Samuel Kala in Daly City, CA City, and Doug Carver, Live Oak Structural Termite Control in Berkeley, CA provided boards. The Oxford Tract, College of Natural Resources, University of California, Berkeley, CA, provided rearing facilities and laboratory space for wood processing. The Structural Pest Control Board (Project No. #84SA901707) graciously provided funding.