Project Concluding: Summary Report

Development of Novel Repellents for the Honeybee, *Apis mellifera*

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This project's goal has been to develop a novel repellent for the honeybee, *Apis mellifera.* Our approach to insect control involves nontoxic solutions that manipulate the insect's chemosensory system – specifically, the olfactory and gustatory systems. Since insects use tastes, odors, and other chemical cues from the environment to control important behaviors, products that manipulate the way the insect responds to these cues can alter insect behavior without negatively affecting existing integrated pest management (IPM) strategies or introducing toxins.

Although bees are essential for pollination in many plant species, consumers are increasingly interested in seedless citrus. Mandarins exemplify this trend; seeded mandarins are currently harder to market than seedless varieties, and honeybees cannot be controlled with conventional insecticides without harming apiculture and potentially interfering with existing IPM schemes.

The citrus industry would benefit from a honeybee repellent that is easy to use, effective, and harmless to beneficial species so that it could be incorporated into existing IPM plans.

We are therefore targeting the components of the honeybee chemosensory system in order to develop a species-specific, nontoxic bee repellent for use on blooming mandarins. Our method (Figure 1) is to identify proteins present in the bee's chemosensory organs, and then isolate compounds or molecules that alter bee behavior by disrupting the function those proteins necessary for the bee to recognize cues from the environment. In this case, the cues are scents and tastes that attract bees to citrus.

During the past two years we used a bioinformatic approach to identify many key *Apis* chemosensory genes, and the expression patterns of these genes have been analyzed to identify potential targets (Figure 2A). Several of these potential target genes have been expressed in the laboratory (Figure 2B).

This past year, we have made a major breakthrough in our platform technologies. Our new high throughput system, called *Attenu*, dramatically improves the speed of our screening methods and allows us to test thousands of compounds or molecules simultaneously for binding to a honeybee chemosensory protein (Figure 2C). By binding to honeybee chemosensory proteins, these molecules may disrupt protein function and alter the chemosensory system, thus altering honeybee behavior. Thus far, we have identified 30 genes encoding potential target proteins and we are screening a combinatorial chemical library – a large collection of compounds – to isolate molecules that bind to *Apis* chemosensory proteins. These molecules will be tested with live bees to see if they have repellent properties.

Our efforts to date and the progress made in 2005 have generated sufficient data for us to seek additional support for this project from the USDA. This external funding, the CRB's support in 2004 and 2005, and our successes in 2005 will allow us to test novel molecules for behavioral effects soon. Our new assay system will allow us to complete the small molecule screens over the next several months, and we anticipate testing potential repellent molecules this coming year on schedule to begin field trials by the end of year four.

Contact Citrus Research Board for figures.