BIOLOGICAL CONTROL OF ORIENTAL FRUIT MOTH

PROJECT LEADER: Walt Bentley

COOPERATORS: Dr. Marshall Johnson and Dr. James Hagler

OBJECTIVES

1. Develop a small scale-rearing program for *Macrocentrus ancylivorus,* the parasite of Oriental fruit moth
2. Determine movement distance of parasite from release site into peach orchard

RESULTS

Having been established on sunflower moth since 2003, *Macrocentrus ancylivorus* has maintained a presence at Kearney Agricultural Center in an orchard of Crimson Lady peaches. This orchard is adjacent to a field of sunflowers, planted annually to provide an alternate overwintering host for *Macrocentrus ancylivorus*. The parasitoid was established in 2003 and 2004 using augmentative releases but is now parasitizing both Oriental fruit moth (OFM) and the sunflower moth in the absence of controlled releases. In 2006 preliminary work was done on selection of a reliable marking technique for *Macrocentrus ancylivorus,* enabling us to proceed in 2007 with distance movement studies. It was established that *Macrocentrus ancylivorus* sprayed with whole milk and soy milk successfully picked up the milk protein marker. Dr. James Hagler of the Arizona Cotton Lab (USDA) performed the analysis (ELISA), with excellent results showing the protein detected on all treated *Macrocentrus ancylivorus*. The Colorado State Department of Agriculture generously provided us with additional *Macrocentrus ancylivorus* pupae, allowing us to maintain and augment our own production.

Macrocentrus ancylivorus are reared at KAC using a procedure adapted from that perfected by Glenn L. Finney et al. (Hilgardia, 1947). It requires just over one month per generation of *Macrocentrus ancylivorus*, using the potato tuber moth (*Gnorimoschema operculella*) as a host. The ideal temperature appears to be 78-80° F with a humidity of 60% though we have found the humidity difficult to maintain. The entire process is entirely dependent on synchronization of the two insect life cycles and requires a great deal of attention. Tuber moth pupae are placed in a shallow box modified with a window screen mesh over the top. Following emergence of the majority of the moths a damp cloth is placed over the mesh screen to collect eggs. This remains in place for approximately three days while eggs are being laid. The cloth is then removed and placed over 20-25 Russet potatoes in an insect cage or Bugdorm (BioQuip, Inc.), Inc uniformly punctured with holes 3 mm. deep and 1 cm. apart in an enclosed space. As larvae emerge they

migrate to the holes in the potatoes. This process requires approximately five days at which time the cloth can be removed. Adult *Macrocentrus ancylivorus* are then introduced into the cage.

While the above tuber moth activity is occurring *Macrocentrus ancylivorus* pupae are allowed to emerge in a separate cage. A wicked vial containing a honey and water dilution is provided as a food source. Following emergence and allowing for mating to occur, approximately 75 adult female *Macrocentrus ancylivorus* are introduced into the potato cage and left for 10 days. Potatoes are then removed from the cage and placed in an egg carton suspended above a shallow tray lined with waxed paper and filled with sand to allow the larvae to migrate from the potato and pupate in the sand. It may take about 5 days to allow pupation to take place in 75% of the population.

Pupae are then removed from the sand (a strainer may be used to sift as much sand as possible from the pupae) and placed in a 50/50 bleach and water solution for a few minutes only. Stirring gently will remove most of the remaining sand and the pupae of the *Macrocentrus ancylivorus* and the tuber moth can be clearly differentiated. After removing the pupae from the bleach solution they are placed in a solution of 3 parts alcohol/2 parts water. The *Macrocentrus ancylivorus*s will float and can be gently skimmed off the top.

Macrocentrus ancylivorus pupae are then placed in an insect cage to await emergence. Tuber moth pupae are once again placed in the shallow box and the process is repeated. This process is capable of producing approximately 500-600 *Macrocentrus ancylivorus* per 10 lbs. of potatoes.

Russet potatoes are used as a food source and ovipositing site in our *Macrocentrus ancylivorus* rearing. They are infested with potato tuber moth using an established protocol (see Rearing above). Using infested and non-infested potatoes a greenhouse trial was set up to determine whether infested potatoes attracted more *Macrocentrus ancylivorus* than non-infested. Three russet potatoes were exposed to potato tuber moths for egg-laying. All potatoes were punctured with approximately the same number of holes. In a greenhouse corridor 144 ft. long, three potatoes infested with tuber moth larvae were placed at one end and three non-infested potatoes at the opposite end. Potatoes were placed approximately 10 ft. high on a ledge. A slit was made in each potato and a sticky trap inserted, much like wings on either side of the potato. *Macrocentrus ancylivorus*, 40 females and 10 males, were released in the center of the corridor and left for a period of 72 hours. Ambient temperature was monitored with a HOBO (Onset Corp.) data logger and remained within the parameters for *Macrocentrus ancylivorus* activity.
The same experiment was performed similarly in a smaller enclosed room (19 x 21 feet) in the greenhouse. Again, 50 *Macrocentrus ancylivoruss*, approximately half male and half female, were released. Potatoes were placed on opposite sides of the room, prepared as above. There were three replications. Temperature was monitored as above. Traps were checked at 24 hour intervals for three days.

No *Macrocentrus ancylivoruss* were recovered from traps in the greenhouse corridor. *Macrocentrus ancylivorus* were recovered from sticky traps in the smaller room, but results were inconclusive though more *Macrocentrus ancylivoruss* were recovered from the non-infested potatoes. A total of 7 Macrocentrus ancylivoruss, all male, were recovered from the infested, 11

Macrocentrus ancylivoruss, 8 females and 3 males, were recovered from the non-infested potatoes.

FIELD TRIAL

In August, 2007, in a young mixed peach and cherry orchard infested with OFM, sticky cardboard sleeves were placed over branches to trap *Macrocentrus ancylivoruss*. Twelve inch cardboard tubes, 2 in. diameter were covered with a sticky yellow tape called Rollertrap (Western Farm Service). These were placed over the ends of branches with the shoot end protruding as an attractant for *Macrocentrus ancylivoruss*. Altogether 96 tubes were placed, 4 each on 24 trees. Trees were located across 12 rows, within which the first and third trees were used.

Macrocentrus ancylivorus were sprayed in each of two Bugdorms. One group of approximately 200 was sprayed with soy milk, the other with whole milk. These markers were tested in 2006 and found to be effective. Before release, 25 unsprayed *Macrocentrus ancylivoruss*, 25 soy-sprayed *Macrocentrus ancylivoruss*, and 25 whole milk-sprayed were aspirated, placed in micro-capsules, and placed in a freezer to be sent for ELISA, along with any recaptured *Macrocentrus ancylivoruss*.

Sprayed *Macrocentrus ancylivorus* were released from two points at the south side of the orchard. The *Macrocentrus ancylivoruss* sprayed with whole milk were released at the edge of the orchard adjacent to the first row of trees, the soy treated *Macrocentrus ancylivoruss* were released 100 ft. directly south of the first point, in the midst of an adjacent vacant field.

Following conclusion of the first experiment, large barrier traps, approximately 6 ft x 6 ft, were constructed of sheets of corrugated cardboard, covered with Rollertrap. These were attached at each end to PVC poles which were then planted in 5 gallon buckets filled with stones and placed in front of the first trees in each of eight rows. Traps faced south, the direction of the release. Releases were done as before but in the evening rather than the morning.

No *Macrocentrus ancylivoruss* were found in traps after 24 hours. At one week, traps were checked again but no *Macrocentrus ancylivoruss* were recovered.

An unseasonal rain and windstorm moved through the area about 24 hours after the experiment was set up and compromised many of the traps. However, after three days, seven *Macrocentrus ancylivorus* were recovered from traps and placed in micro-capsules, then in the freezer. Damaged traps were reset and checked again in 24 hours.

The ELISA results indicated that 4 of the 7 recaptured *Macrocentrus ancylivoruss* tested positive for the whole milk protein (edge of orchard) while only one tested positive for the soy milk (vacant field). Two *Macrocentrus ancylivoruss* tested negative for both. As all of the pre-release sprayed samples, 25 of each, tested positive for their respective milk proteins and the unsprayed controls tested negative for both, it is possible that some *Macrocentrus ancylivorus* were already present in the orchard and would account for the negative readings.

DISCUSSION

It was our intention to release at least 1000 *Macrocentrus ancylivoruss* with each protein marker but, despite receiving an excess of 3500 *Macrocentrus ancylivorus* pupae, our emergence percentages were unusually low. This occurred in two consecutive emergence samples and we have not established a cause. This has not been a problem in the four years that we have been performing the *Macrocentrus ancylivorus* studies. It was encouraging to discover that the recapture method does have potential, but due to the small number of *Macrocentrus ancylivoruss* that were recaptured, it seems that a larger sample population will be required to draw any definitive conclusions regarding movement and flight distance.