AES/CE MAR 84 Workgroup walnuts

University of California
Division of Agricultural Sciences

Project Plan/Research Grant Proposal

Project Year 2009-10 Anticipated Duration of the Project 1 year requested

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Project Title Why puffers work: Determining the effects of residual releases on control of codling moth

Keywords walnuts, codling moth, pheromone mating disruption

Commodity(s) walnuts Relevant AES/CE Project No.

Significance:

The mechanism by which traditional pheromone based programs work has been explained as-competition with females in the orchard by the synthetic pheromone devices. Presumably, the dispensers act as "false females" that compete for the time that a male has to look for true females. If mating is delayed for even a few days, then the reproductive output of the female declines severely. Thus, there has been an emphasis on deploying many pheromone dispensers per acre, which meant that pheromone mating disruption in walnuts becomes extremely difficult due to the logistic problems associated with the large canopies of mature walnuts.

In contrast, the puffer program that is currently used in walnuts deploys 1 pheromone dispensing unit (a "puffer") per 2 acres, thus competition through false trail following is a less likely mechanism. However, most of the previous studies that have looked at the number of point sources have used emission rates per unit that are quite low compared to the puffer units. Therefore, other mechanisms of mating disruption may be operating, e.g. habituation or arrestment of movement, or in fact secondary release from organic surfaces in the orchard may make the number of effective release points larger than expected. The crux of the grant is that if we are to improve the program over time or address problems that may arise with the puffer program, then we will need to understand how the puffer disrupts codling moth mating.

One of the unknowns with the puffer program is the impact of pheromone release from secondary sources such as the pheromone residue that accumulates on the surface of the puffer unit itself , or from organic material directly below the unit (e.g. leaves, bark, soil), or from downwind material (e.g. leaves). One researcher in New Zealand showed the release the pheromone from leaf surfaces was possible after exposure of the leaves to a pheromone plume, but again these studies were done with traditional Isomate type dispensers with much lower release rates.

If the puffer unit is releasing from the accumulated surface of the cabinet, then the unit is in fact a passive release device that effectively recharges itself with each "puff". This would mean that the time of day that puffing occurs may be largely irrelevant or possibly could be shorted if the recharging only occurs during night-time hours when UV breakdown is less likely. Similarly, if leaves or bark are acting as secondary release points, then the issue of point sources is largely negated as well. Theoretically, programs could be developed that space the puffers based on their ability to generate these secondary release points. The hope is that understanding these processes may also lead to changes that might reduce the amount of pheromone needed during the season, which would cut program costs.

One piece of data developed in our lab has suggested that residual effects of puffers can be important as follows: if puffer units are turned off and left in an orchard, codling moth will be effectively suppressed for 1-5 additional days. Using an approach that laid out a grid of traps, a map of the pheromone plume as indicated by trap suppression suggested that the pheromone plume was affecting at least 1800 feet downwind of the puffer unit.

Objectives:

1. Determine secondary pheromone release rates from a) the pheromone cabinet and b) surrounding organic material (bark and leaves) using head space volatile collections
2. Determine the correlation in space and time between secondary release rates and behavioral correlates of codling moth control: 1) trap suppression 2) female mating status 3) preliminary codling moth movement studies.

Plans and Procedures

Single puffer units will be placed into orchards and allowed to emit (puff) continuously for ca. 2 weeks. Within the same orchard but parallel to the wind, a second unit will be placed and allowed to puff for only 1 week. After the first week, the unit will be turned off and left in the orchard. A third portion of the orchard will not be treated with any pheromone. Replication will be achieved by repeating the treatments or new locations (or re-randomized locations) after the effective plumes are no longer detected.

Samples will be taken in a series of expanding grids with the puffer as the origin of the grid. Leaf, bark, and soil samples will be taken primarily in a downwind transect from the puffer. Leaf, bark, and soil samples will be taken back to the laboratory and head space volatile collections onto a solid matrix will be taken. The level of pheromone that binds to the solid matrix can be determined later with a gas chromatograph such that the relative levels of secondary emissions can be determined for both the type of sample and their relative position to the puffer. The contrast of the puffer "on" and puffer "off" treatments will provide an estimate to determine if and how long secondary emissions occur. The untreated area will provide an estimate of the direct effects of the puffer on

the various biological metrics. The appropriate time intervals will be tuned throughout the growing season as data is collected. For example, if residuals emissions are observed in the puffer "on" treatments and no residual emission in the puffer "off" plots after one week, then the time intervals will need to be shortened to daily samples.

A similar transect of virgin female codling moth will be placed in pheromone traps as lures. At peak codling moth flights, virgin females will be placed out on mating tables to determine mating status at different points in the orchard relative to the puffer placement. All samples will be taken from the three test areas. Suppression of virgin females that are used as lures in traps function as a proxy to estimate effective mate location by wild females. Using a grid of traps baited with virgin females contained in small wire mesh cages, suppression of traps will be monitored on a daily basis after the puffers are turned off in the puffer "off" program. Traps in areas with active puffers, inactive puffers, and non-treated areas will provide the appropriate contrasts.

Movement of codling moth in response to a pheromone plume can be evaluated using wild codling moth through the use of assays developed in Arizona for a variety of insects in general and in Washington for codling moth in particular. Dilute applications of a particular protein (egg, milk, or soy) can be applied to orchards using conventional spray equipment. Codling moths that are either directly sprayed or come in contact with sprayed foliage or bark are marked with the protein. A monoclonal antibody based assay can be used to detect the protein for 1-2 weeks after the application. Therefore, if a heavily infested site that is downwind of a puffer is treated with a dilute protein, then any moths found in the untreated area are presumed to have originated from the treated areas. The extent of movement or potentially sites of arrested movement can be determined with a series of graphs in the pheromone treated areas if combination lures are used to recapture the moths. Use of the combination lures will also allow movement of both males and females to be tracked in response to the pheromone plume. The movement portion of this grant is still very preliminary and the early part of the season will be to confirm the assay and associated parameters such as the duration of the protein marker.

BUDGET – Understanding behavioral mechanisms - walnuts

Salaries		
	Post doctoral researcher - Dani Casado (funded by national grant from Spain) – ca. $44,400 are being contributed to the project through his salary – see attached letter)	NA
	SRA II (15% time @ $5084 / mo)	$4,067
	Salary Subtotal	$4,067
Benefits (21.13%)		$813
	Subtotal	$4,881
Travel (Seasonal rental, mileage,...)		$3,000
Supplies	Miscellaneous (traps, lures, colony materials)	$1,000
	Monoclonal Assays	$2,500
	Residual Pheromone Analyses	$3,000
	Total	$14,381

Originator's Signature Date

COOPERATIVE EXTENSION County Director Date

Program Director Date

AGRICULTURAL EXPERIMENT STATION Department Chair Date

LIAISON OFFICER Date