State Horticultural Association of Pennsylvania		Research Grant Proposal
Title:	Evaluating the use of mating disruption for management of codlin moth in small acreage apple orchard blocks	
Personnel:		
Principle Investigator	Dr. Tracy C. Leskey, Research Ento Appalachian Fruit Research Station Kearneysville, WV 25430-2771. En Phone: (304) 725-3451 ext. 329	mologist, USDA-ARS (AFRS), 2271 Wiltshire Rd., nail: <u>tracy.leskey@usda.gov</u> .
Co-Principle Investigators	Dr. Laura J. Nixon, Post-Doctoral R ARS Appalachian Fruit Research St Rd., Kearneysville, WV 25430-2771	esearch Associate, USDA- ation (AFRS), 2217 Wiltshire l
	Dr. Johanna E. Elsensohn, Post-Doc USDA-ARS Appalachian Fruit Rese Wiltshire Rd., Kearneysville, WV 23	toral Research Associate, earch Station (AFRS), 2217 5430-2771
	Dr. James Hepler, Post-Doctoral Res Appalachian Fruit Research Station Kearneysville, WV 25430-2771	search Associate, USDA-ARS (AFRS), 2217 Wiltshire Rd.,
Duration of Project:	2 years (April 20 th , 2022 – October 3	31 st , 2024)

Justification:

Pest status and management of codling moth. Codling moth *Cydia pomonella* L.(CM) is a global pest of pome fruits and is widely considered one of the most destructive apple pests in the world; it is one of four key pests of apples in the eastern USA (Pedigo et al. 2021). CM overwinters as a fully developed larva in a thick cocoon, usually found under loose bark and in soil. Adults emerge in the spring, begin mating, and deposit eggs on host plants. The larval form of CM directly damages fruit as they bore into the fruit core and feed until larvae complete development. The resulting tunnel fills with red/brown frass, decreasing fruit quality for fresh market. Apples sent for processing are randomly sampled for larval infestation, with a single finding resulting in the rejection of the whole shipment.

CM can be managed with season-long spray programs utilizing monitoring traps and degree day models (Krawczyk 2017a, Pfeiffer et al. 2021). Biological control agents such as *Trichogramma sp.* parasitoids have not been compatible with commercial orchards. While spraying selective viruses is commonly recommended, the development of resistance has reduced their effectiveness (Lacey et al. 2008). Mating disruption is one of the most commonly used control measures for management of CM in larger orchards and involves deploying dispensers or puffers containing the CM sex pheromone, codlemone, in the upper canopy of trees throughout the orchard. Codlemone competes with the pheromones released by females (known as competitive attraction) and then deactivates males for the remainder of the night (Miller et al. 2010).

Oriental fruit moth. Oriental fruit moth (*Grapholita molesta* Busck) (OFM) is a common pest of stone and pome fruits in the Eastern US. Similar to CM, OFM larvae cause internal fruit damage, with the early generation larvae additionally causing shoot flagging. OFM are most problematic in peaches but have become a more serious pest in apples in recent years. Monitoring traps for OFM are typically deployed in April to establish a biofix, and this pest is managed using degree day models and season-long spray programs (Krawczyk 2017b, Pfeiffer et al. 2021). This pest is also commonly managed using mating disruption strategies, and both insecticide programs and mating disruption products tend to simultaneously target CM. Therefore, while OFM is not the main focus of this proposal, we will include this pest as part of the mating disruption portions of the project. CM

Mating disruption in small scale acreage. The generally accepted guidelines for successful use of CM mating disruption in apple orchards are blocks 1-2 ha in size, squareshaped, with flat topography, isolated to avoid immigration of gravid females, and containing small trees (Witzgall et al. 2008). Thus, it is rare for orchards to fit all of these generalized criteria, limiting its usefulness to growers. Currently, larger orchard block size recommendations are primarily based on only two studies from the 1980s-1990s. The first was in western Switzerland in the 1980s, over 35 years ago (Charmillot 1990), in which mating disruption failure was documented in smaller scale blocks but few details beyond heavy damage at orchard borders were provided. The second study was conducted in WA, USA (Knight et al. 1995) with the main focus on establishing pheromone dispenser density required for disruption using 0.4 ha blocks. Additionally, a female dispersal distance of 275-425 m was estimated using a post-hoc analysis of CM injury in 0.4 ha blocks and assumed a location for the source population CM females. This calculated dispersal distance was then extrapolated to infer that acreage needed to be greater than 2 ha for mating disruption (Knight et al. 1995). Neither Charmillot (1990) nor Knight et al. (1995) directly measured female dispersal or conducted the studies necessary to establish optimal acreage size for mating disruption. Moreover, mating disruption products and orchard architecture have advanced since these studies were conducted, and abiotic conditions found in the mid-Atlantic are markedly different from their study environments.

CM dispersal. The dispersal patterns of male CM are well understood, as their behavior has been studied extensively in the development of both sterile insect technique and mating disruption. However, there is minimal literature on the dispersal patterns and capacity of female CM. Male moths tend to fly upwind into orchards as they follow the sex pheromone plume of females, zig-zagging across and within the plume. Female CM locate oviposition hosts by orienting upwind in response to olfactory stimuli such as fruit volatiles (e.g., Yan et al. 1999). A study from Germany reported that male CM are mostly sedentary, but a small proportion can disperse distances up to 11 km (Mani and Wildbolz 1977). Similarly, a flight mill study under laboratory conditions found that females are relatively sedentary and are able to disperse up to 11 km (Schumacher et al. 1997). Kinship analyses of offspring from a female mark-release-recapture trial in Europe indicated that most females only dispersed 40 – 80 m, with a maximum distance of around 200 m (Margaritopoulos et al. 2012). These results confirm the generally sedentary nature of CM but fail to demonstrate that female moths routinely disperse long distances under field conditions. *Ultimately, it is poorly understood how female CM disperse and how dispersal is affected by physiological state and abiotic factors.*

Preliminary Data. In 2021, we conducted preliminary studies using Isomate CM/OFM twin tubes dispensers at a rate of 80 dispensers/ha in blocks of 0.10-0.40 ha. We compared fruit at harvest from three disrupted and three control blocks (200 fruit per block evaluated). While

injury was low, there were significant differences between disrupted and control blocks (χ^2 =4.4, df=1, P=0.04), with no injury detected in disrupted blocks and 1.5% in control blocks. Despite low overall injury, it still points to the capacity of mating disruption to potentially disrupt CM and OFM in smaller scale acreage.

Here, we propose to: 1) determine if CM/OFM mating disruption can be effective in apple blocks <1 ha; 2) the dispersal capabilities of female CM and how that may affect mating disruption efficacy as this is often pointed to as the reason for failures.

Objective(s):

Years 1-2

1. Evaluate the efficacy of CM mating disruption in apple blocks <1 ha through fruit damage assessments at harvest.

Year 1

2. Establish conditions under which female CM will disperse and quantify dispersal capacity.

Procedures:

Objective 1. Evaluate the efficacy of CM mating disruption in apple blocks <1 ha through fruit damage assessments at harvest.

In 2022 and 2023, twelve research apple blocks ranging 0.25 – 1 acre in size at the Appalachian Fruit Research Station, Kearneysville, WV will be used for this trial. Four replicates of three management strategies will be assigned among these blocks: 1) grower standard insecticide program for CM and OFM using pheromone traps and degree day models, 2) Isomate-CM/OFM mating disruption pheromone dispensers only, supported by a grower standard reduced pesticide schedule, 3) unmanaged control. Mating disruption dispensers will be deployed in respective blocks and cardboard trunk traps and pheromone-baited monitoring traps will be deployed in all blocks to monitor adult and larval population. Isomate- CM/OFM pheromone dispensers are designed to be deployed only once in the season and provide mating disruption for CM and OFM. As insecticide programs for CM and OFM are also complimentary, data will be collected for OFM as well. Monitoring traps will be checked weekly for both CM and OFM.

At harvest of each year, 200 fruit per block will be taken from each experimental block for CM and OFM damage assessment; four external and sixteen internal trees will be randomly selected per block and ten fruit will be randomly sampled per tree.

Outcomes: The results obtained from this trial will help determine if acreage less than reported minimums necessary for successful mating disruption can potentially be disrupted for CM/OFM. Subsequently, these data will be used to support a larger extramural grant application.

Objective 2. Quantify the dispersal capacity of gravid and virgin female CM under laboratory and field conditions.

The capacity for female CM dispersal and orchard colonization will be assessed in the field between June – August 2022 using a mark-release-recapture method in three apple blocks at the Appalachian Fruit Research Station. Colony-reared (Benzon Research, PA) gravid and virgin female CM (500 moths per release point) will be marked with UV-fluorescent dye and released on potted apple trees (stripped of fruit) in an open field at five distances (0, 50, 100, 200, 400 m)

from the orchard border rows to simulate potential female dispersal from wild host habitat. Moths will be released at each distance pre-dawn, and each distance cohort will be marked with a different color. Beginning at dusk the same day, 20-minute timed searches of orchards for marked moths will be performed for three consecutive nights using UV-flashlights. The color and location within the orchard of each observed moth will be recorded. We will also deploy passive flight interception traps between trees throughout the planting at a rate of one trap per three trees (~15m) at upper canopy height (Knight 2010), and these will also be checked each evening. Pear ester-baited traps will not be deployed for this experiment as we want to track natural female CM dispersal in the absence of additional stimuli. This experiment will be replicated over time and space, with one trial (three blocks) per week for ten weeks, for a total of 30 replicates. Weather data will be recorded during each trial. This will allow us to estimate the distance from which female CM disperse into an orchard when moving from wild host habitat.

In fall/winter 2022, laboratory-based flight mill assays will be used to establish flight capacity of female CM. Gravid and virgin female moths from both wild sources and colony will be placed on a flight mill (constructed as per Lee and Leskey 2015 for 24h and their flight patterns measured using the DASYLab program (Measuring Computing, Norton, MA) (Lee and Leskey 2015). Wild sourced adults will be reared from pupae collected in the cardboard trunk traps in Objective 1. This will generate data directly comparing the flight capacity of gravid and virgin females, and wild and colony-sourced CMs. We will compare these results to those from the field dispersal experiment to determine the applicability of flight mill assays to CM dispersal within orchard systems. While flight mill data and field dispersal data for CM often do not align, no studies to our knowledge have used both measures of quantifying dispersal capacity within the same study.

Outcomes: This study will establish the propensity of female CM to disperse into orchards from wild host habitat and provide baseline to determine the extent to which female CM dispersal may lead to mating disruption failures in small scale acreage.

Total= \$17.329.00

Overall Budget:

<u>Year 1</u> Salaries	N/A	Supplies (trans. lures. CM/OFM MD	3980.00
	1 1/1 1	dispensers. CM egg masses)	0/00.00
Hourly Wages	6100.00	Travel	N/A
(Summer Intern x 12 weeks)			
Fringe Benefits	2562.00	Misc	N/A
Year 2			
Salaries	N/A	Supplies (traps, lures, CM/OFM MD	1800.00
		dispensers)	
Hourly Wages (Summer	2033.00	Travel	
Intern x 4 weeks)			
Fringe Benefits	854.00	Misc	

Other Support: *Post-doctoral salary and labor associated with orchard maintenance will be absorbed by other sources of in-house and extramural funding.*

Literature Cited

- Charmillot, P. 1990. Mating disruption technique to control codling moth in western Switzerland, Dekker, New York.
- Knight, A. L. 2010. Effect of sex pheromone and kairomone lures on catches of codling moth. Journal of the Entomological Society of British Columbia 107: 67-74.
- Knight, A. L., J. Howell, L. M. McDonough, and M. Weiss. 1995. Mating disruption of codling moth (Lepidoptera: Tortricidae) with polyethylene tube dispensers: determining emission rates and the distribution of fruit injuries. Journal of Agricultural Entomology 12: 58-100.
- Krawczyk, G. 2017a. Tree Fruit Insect Pest Codling Moth. PennState Extention https://extension.psu.edu/tree-fruit-insect-pest-codling-moth.
- Krawczyk, G. 2017b. Tree Fruit Insect Pest Oriental Fruit Moth. PennState Extention <u>https://extension.psu.edu/tree-fruit-insect-pest-oriental-fruit-moth</u>.
- Lacey, L.A., D. Thomson, C. Vincent, and S.P. Arthurs. 2008. Codling moth granulovirus: a comprehensive review. Biocontrol Science and Technology. 18: 639-663.
- Lee, D.-H., and T. Leskey. 2015. Flight behavior of foraging and overwintering brown marmorated stink bug, *Halyomorpha halys* (Hemiptera: Pentatomidae). Bulletin of Entomological Research 105: 566-573.
- Mani, E., and T. Wildbolz. 1977. The dispersal of male codling moths (*Laspeyresia pomonella* L.) in the Upper Rhine Valley. Zeitschrift für angewandte Entomologie 83: 161-168.
- Margaritopoulos, J. T., C. C. Voudouris, J. Olivares, B. Sauphanor, Z. Mamuris, J. A. Tsitsipis, and P. Franck. 2012. Dispersal ability in codling moth: mark-releaserecapture experiments and kinship analysis. Agricultural and Forest Entomology 14: 399-407.
- Miller, J. R., P. S. McGhee, P. Y. Siegert, C. G. Adams, J. Huang, M. J. Grieshop, and L. J. Gut. 2010. General principles of attraction and competitive attraction as revealed by large-cage studies of moths responding to sex pheromone. Proceedings of the National Academy of Sciences 107: 22-27.
- Pedigo, L. P., M. E. Rice, and R. K. Krell. 2021. Entomology and Pest Management, 7th Edition ed. Waveland Press.
- Pfeiffer, D. G., J. C. Bergh, C. Quesada, C. R. R. Hooks, S. Sherif, C. S. Walsh, K. S. Yoder, M. Rahan, J. B. Kotcon, J. F. Derr, R. S. Chandran, D. L. Frank. 2021. 2021 Spray Bulletin for Commercial Tree Fruit Growers. Virginia, West Virginia, and University of Maryland Extension https://www.pubs.ext.vt.edu/content/dam/pubs_ext_vt_edu/456/456-419/ENTO-391.pdf.
- Schumacher, P., A. Weyeneth, D. C. Weber, and S. Dorn. 1997. Long flights in *Cydia pomonella* L.(Lepidoptera: Tortricidae) measured by a flight mill: influence of sex, mated status and age. Physiological Entomology 22: 149-160.
- Witzgall, P., L. Stelinski, L. Gut, and D. Thomson. 2008. Codling moth management and chemical ecology. Annual Review of Entomology 53: 503-522.
- Yan, F., M. Bengtsson, and P. Witzgall. 1999. Behavioral response of female codling moths, *Cydia pomonella*, to apple volatiles. Joural of Chemical Ecology 25: 1343-1351.