Understanding Mating Disruption by Aerosol Puffers: Trap Suppression, Mating Delay and Mating finding

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Puffers:

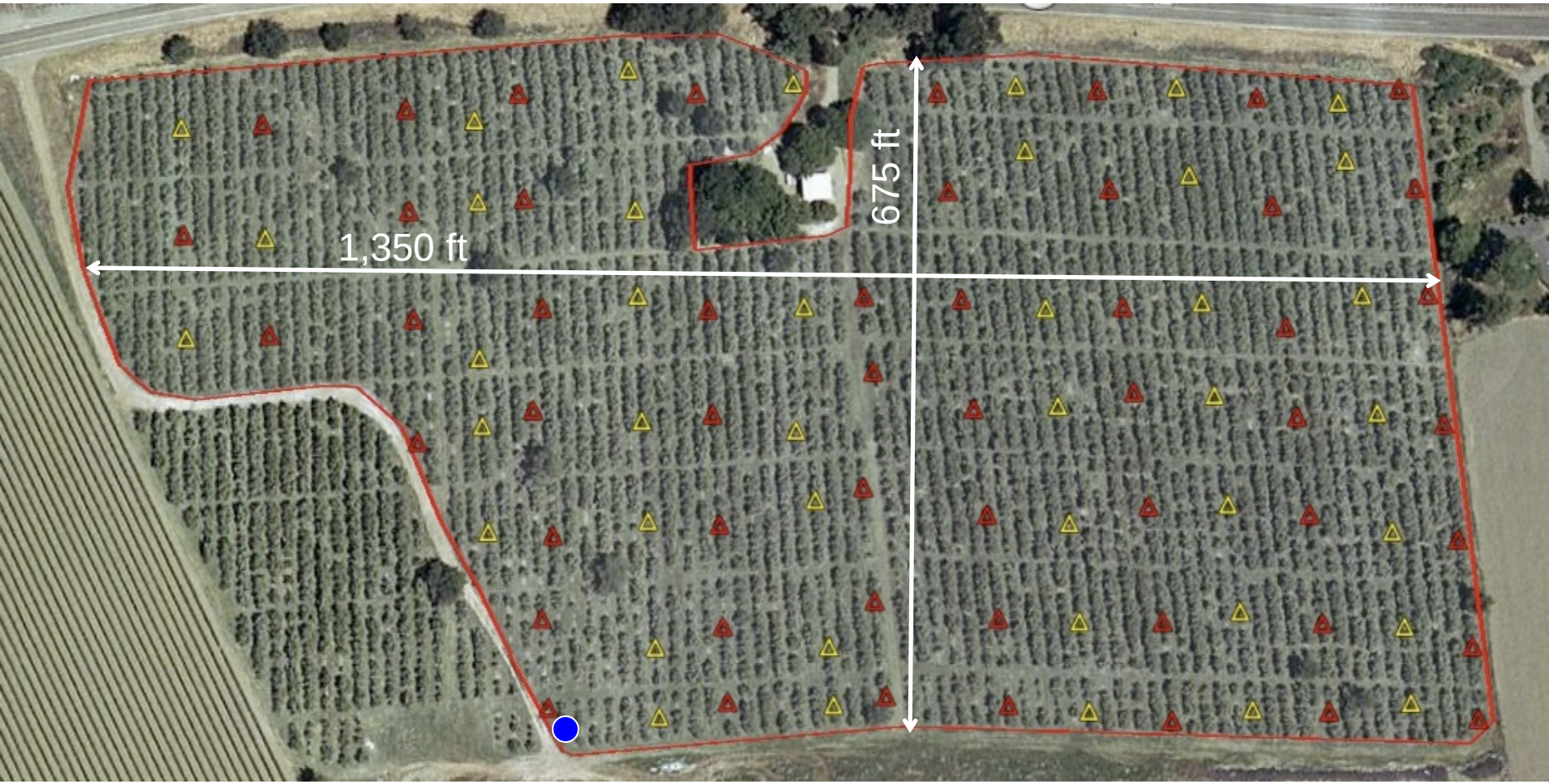
- Mechanical automatic devices

- Emit puffs of aerosol pheromone

- Mating disruption

- High emission rate

- Low density (1 units/2 acre)



- Pheromone puffer
- Pheromone-baited trap (53)
- Female-baited trap (40)

- Unfarmed (3 years) pear orchard
- 17 acres (7 ha) approx.
- Heavily infested by codling moth (20.4 ± 1.6 males/trap & night in early-June)

Baits

- Pheromone: 1 mg commercial lures
- Females:
  - 1 virgin female (< 24 h)
  - kept in a 4 x 4 x 4 cm^3 aluminum cages
  - sugar water supplied daily

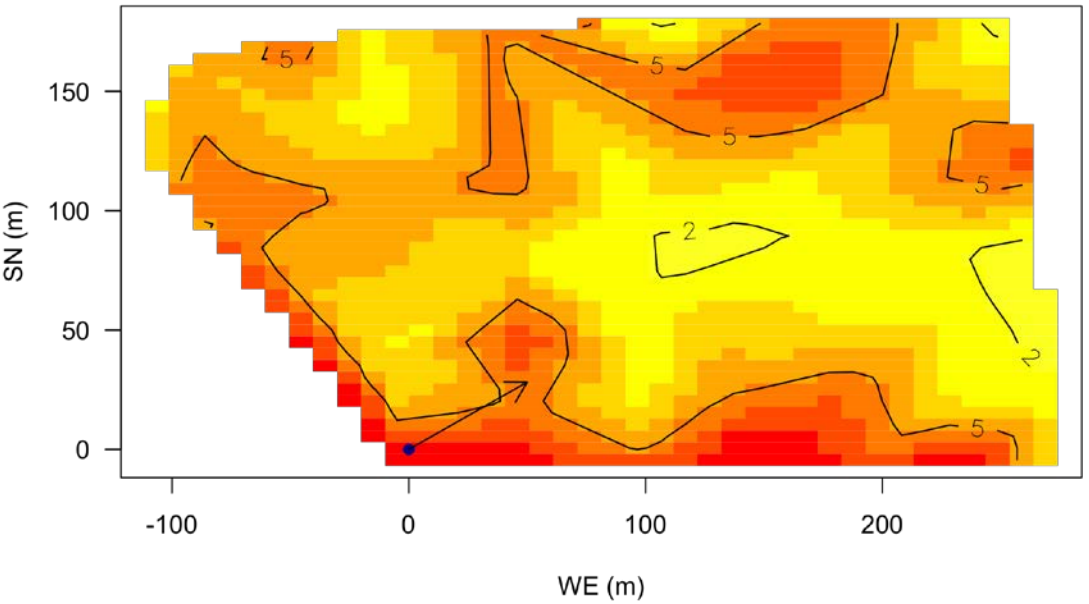
Assay

- 2 replicates: mid- and late-July; and 1 control: late-August
- Traps checked daily for 8 to 11 days (lifespan)

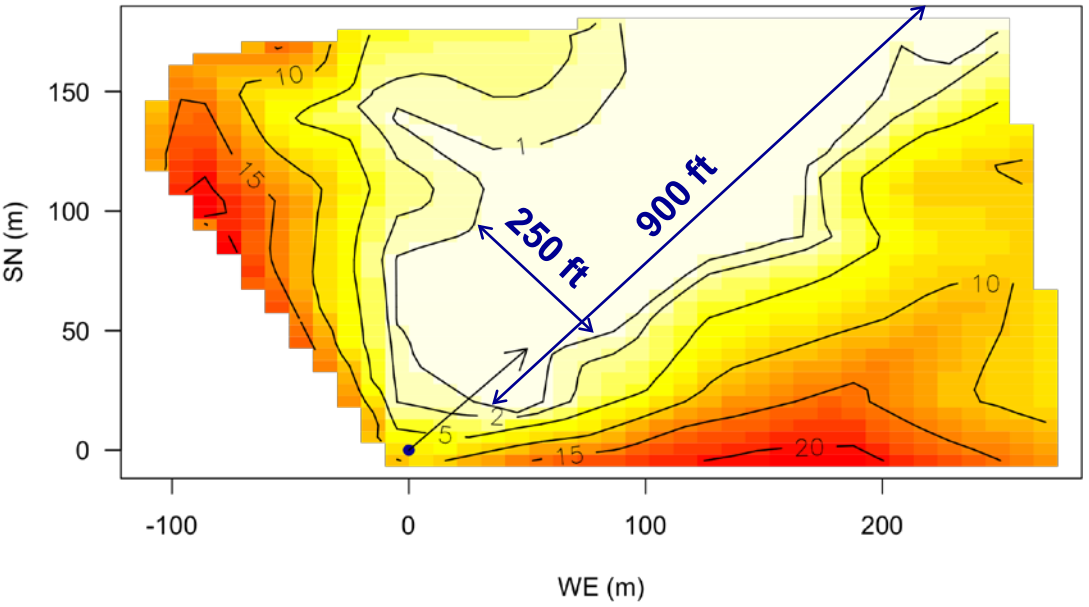
Data recorded

- Number of males/night
- Time to first capture (nights)
- Effectiveness (nights with captures/total nights)

No puffer



With puffer



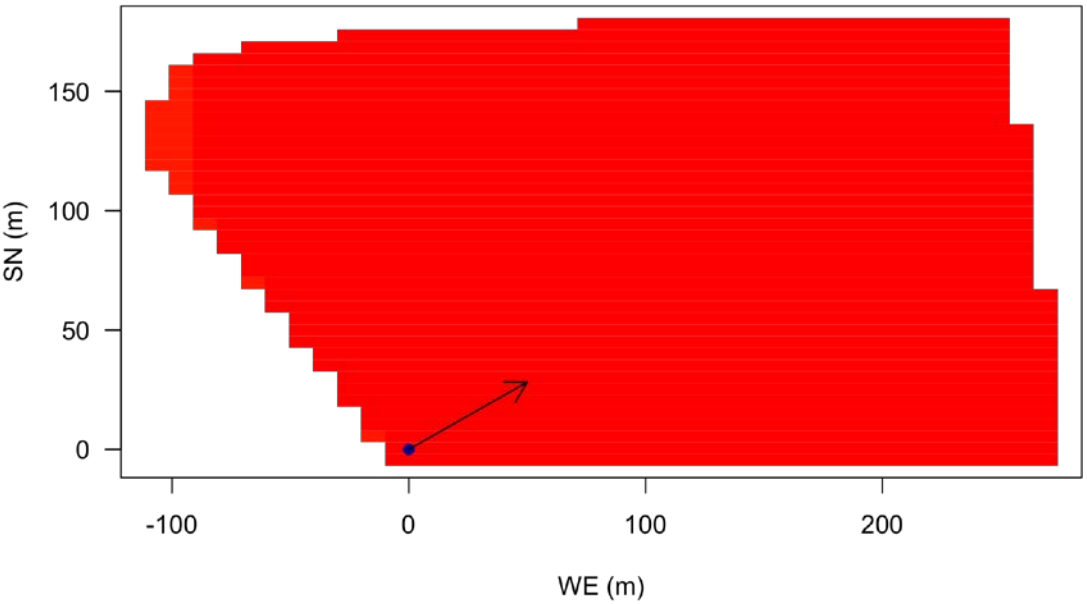
Captures/night (pheromone)

- Captures in absence of puffer were reasonably uniform across the orchard (despite some hot-spots)

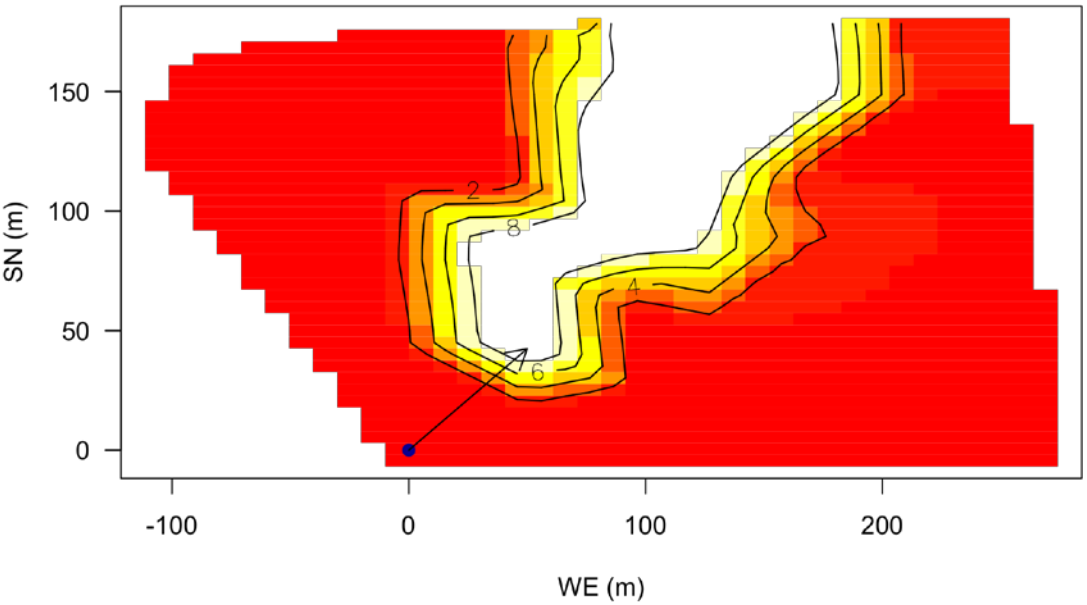
- In presence of the puffer a gradient of captures, perpendicular to the wind direction, was very apparent. Captures were totally suppressed up to 900 ft downwind.

- Smaller values without puffer due to timing of the control (late in season)

No puffer



With puffer



First capture (pheromone)

- In absence of the puffer all traps caught males from the first night (just one did not)

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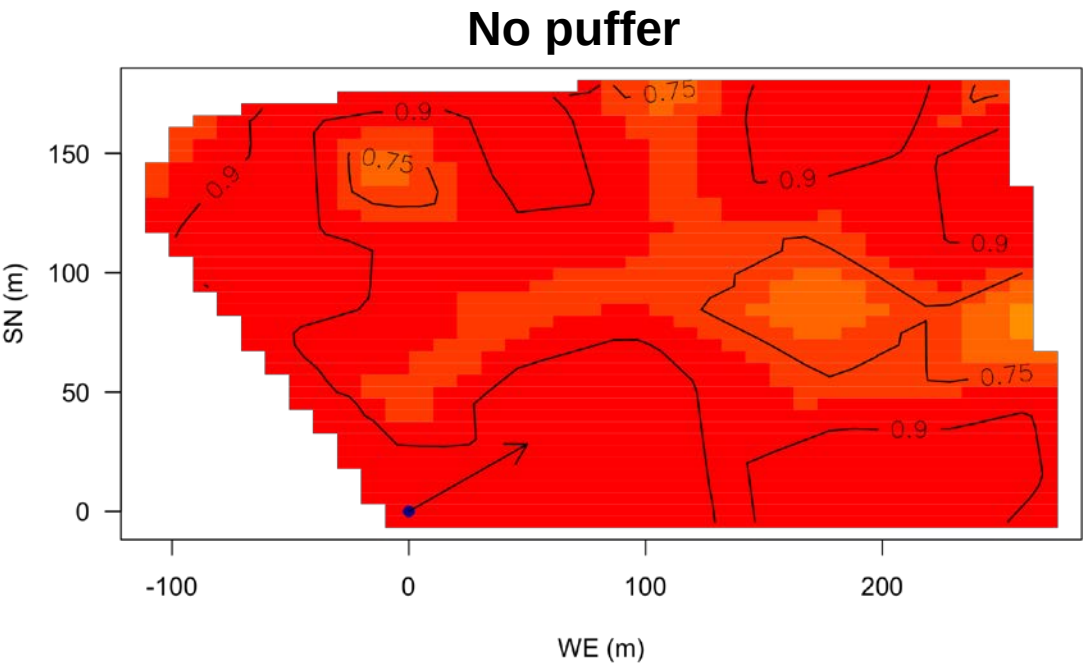
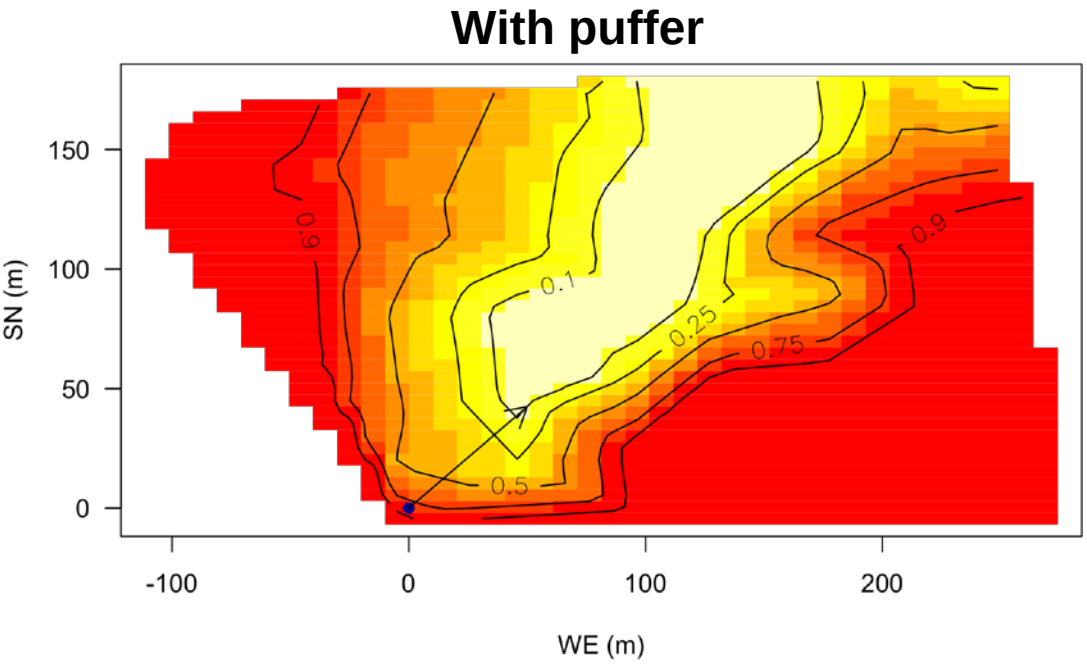
-When the puffer was present, the traps in the middle of the plume never caught, and a delay in captures was also present in the area surrounding the center

- Delay in mating reduces reproductive output in codling moth (other species too)

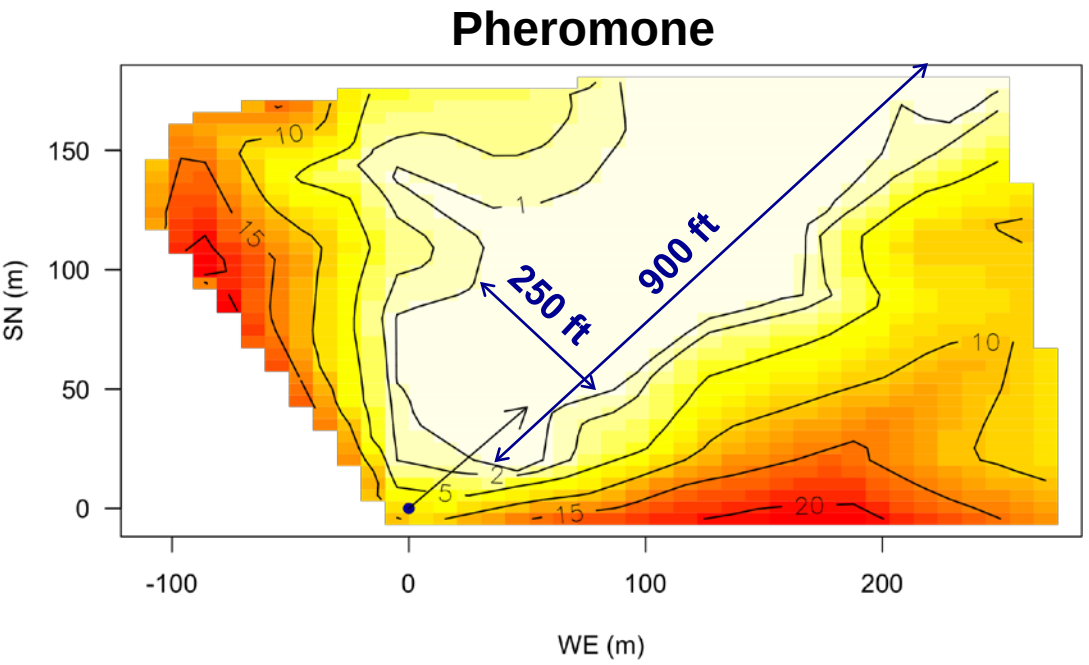


Effectiveness (pheromone)

- Traps caught almost every day when there was no puffer, regardless to their position in the orchard



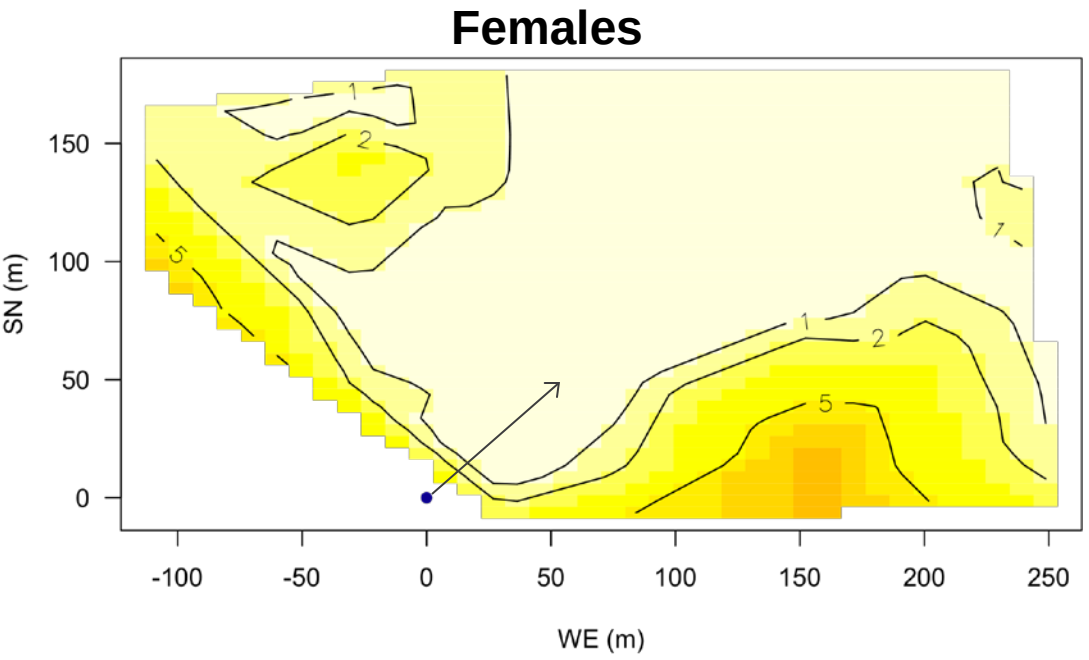
- When the puffer was working, the traps in the middle of the plume never caught, and a gradient of effectiveness was present from inside the plume to the outside



Females vs. pheromone (captures/night)

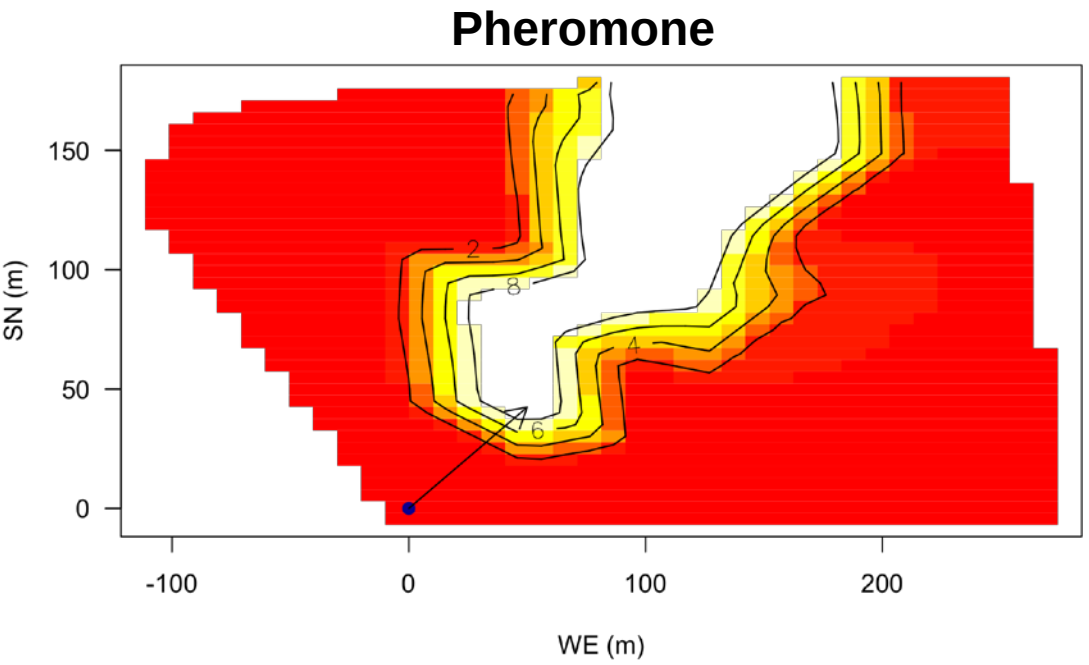
- Captures were lower in the female-baited traps than in their neighboring pheromone ones

- Why? Timing? Release rate? Lab females?

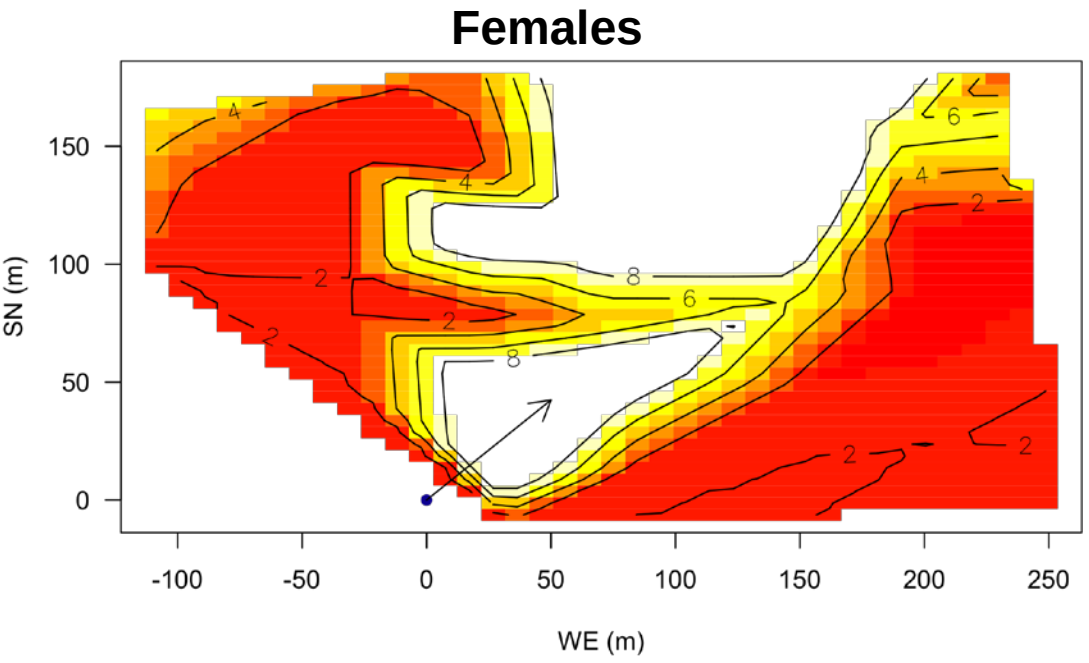


- The capture-free area was wider for female-baited traps

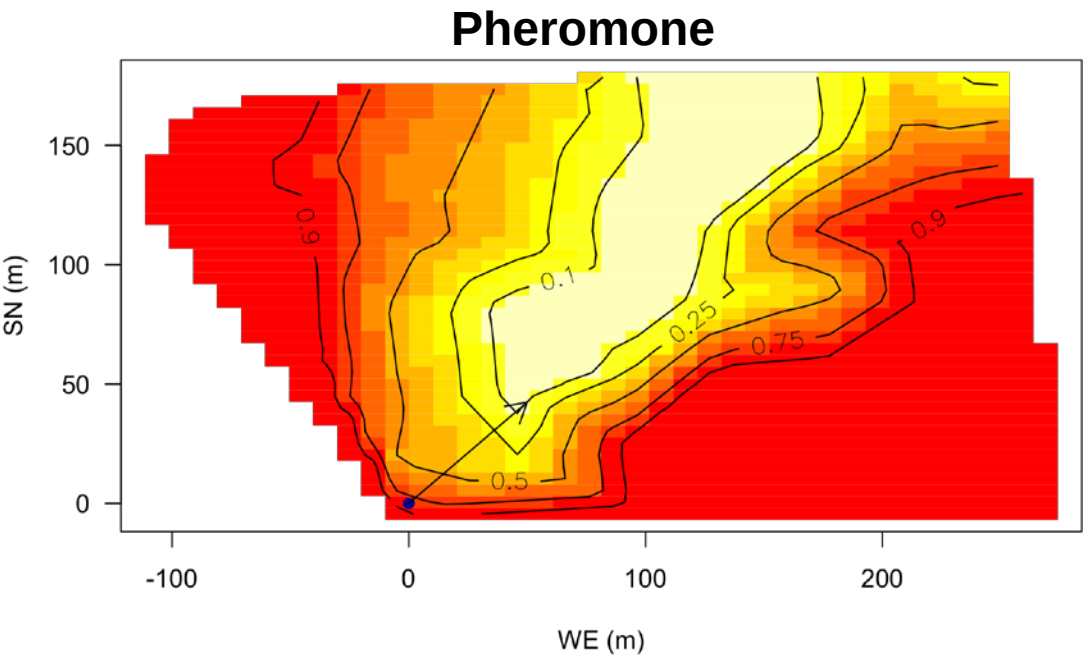
- There was a clear gradient of capture perpendicular to the wind direction for both kinds of traps



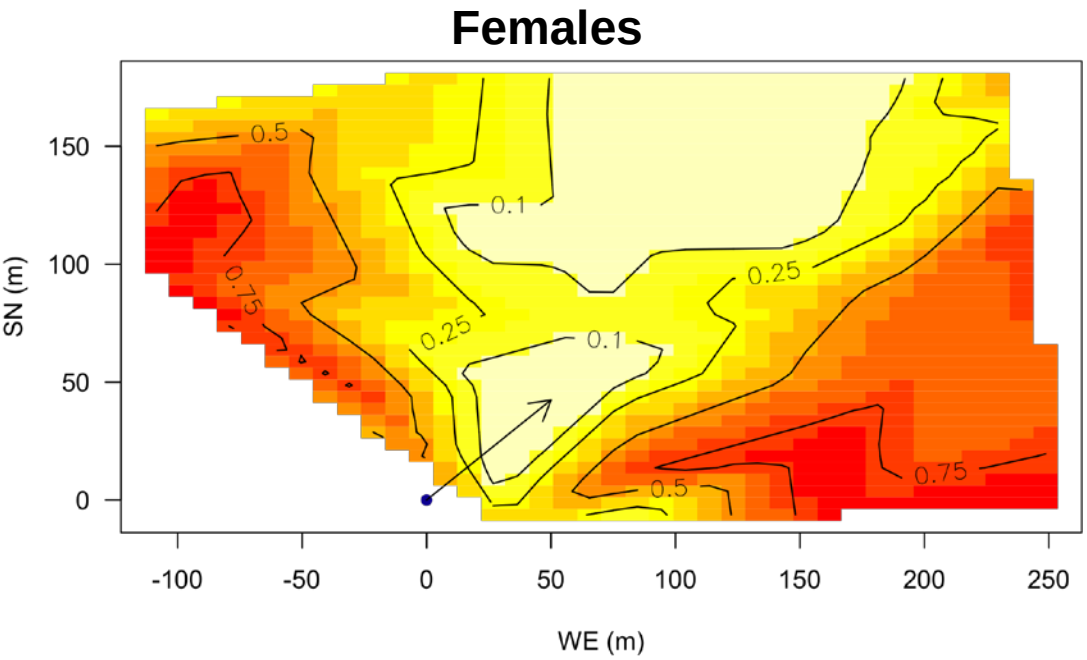
Females vs. pheromone (first capture)



-Captures in the female-baited traps were delayed longer than in their neighboring pheromone ones

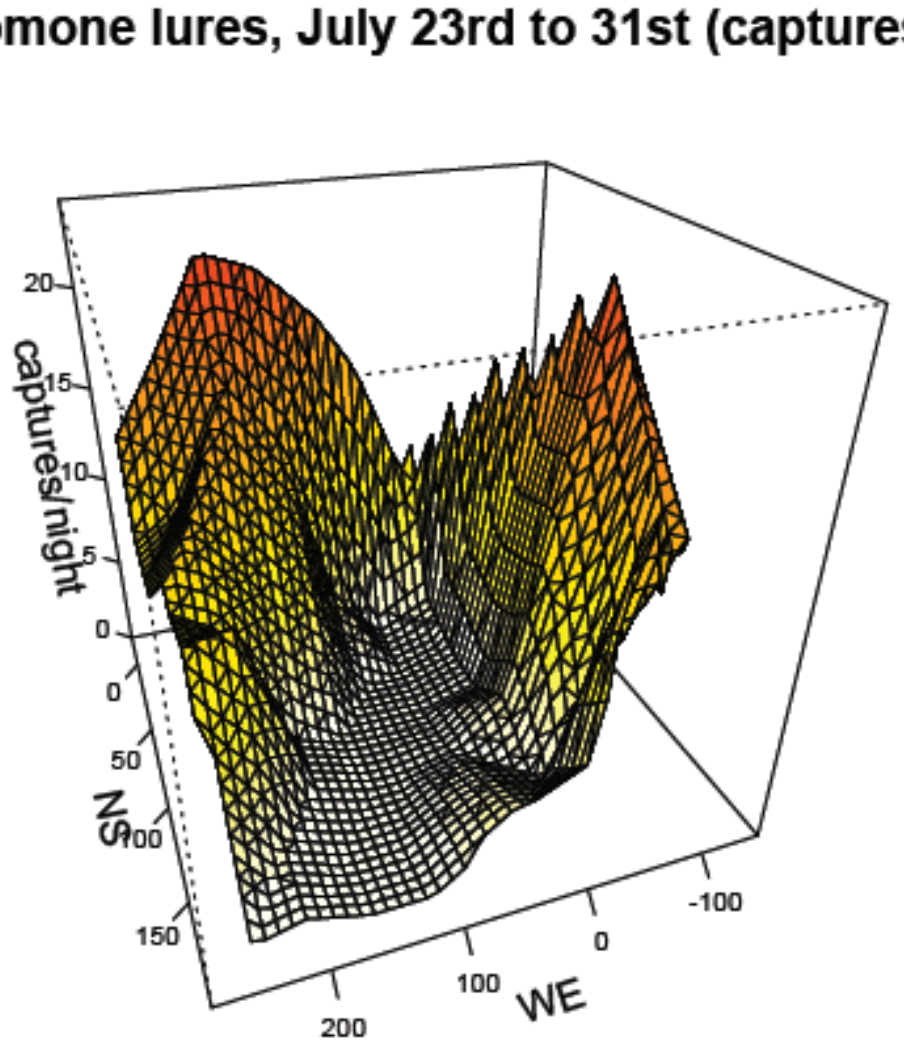


Females vs. pheromone (effectiveness)



- The same pattern was found again, with a stronger effect on female than in pheromone-baited traps

Plume cutting a swath through codling moth flight



Summary/conclusions:

- The plume of a single puffer totally suppressed male codling moth captures by pheromone traps in an area of 250 ft x 900 ft downwind. The end of the plume could not be reached

- The impact of the puffer appears to be stronger on females than on pheromone lures. However, laboratory females were used, and we are assuming that they behaved in the same way as wild ones would

- The puffer not only decreased captures of males per night, but also increased time to the first capture and decreased the number of nights that traps successfully caught. Mating delay negatively impacts reproductive output in codling moth as well as in many other pests (i.e. Torres-Vila *et al.* 2002, Jones *et al.* 2008), and can play a key role in mating disruption performance

SECONDARY EMISSION OF PHEROMONE FROM PUFFER-EXPOSED WALNUT LEAVES

- **Objective: test attraction of codling moth males to leaves exposed to a puffer plume**

  Pheromones have been shown to bind to surfaces (e.g. glass, leaves,…)

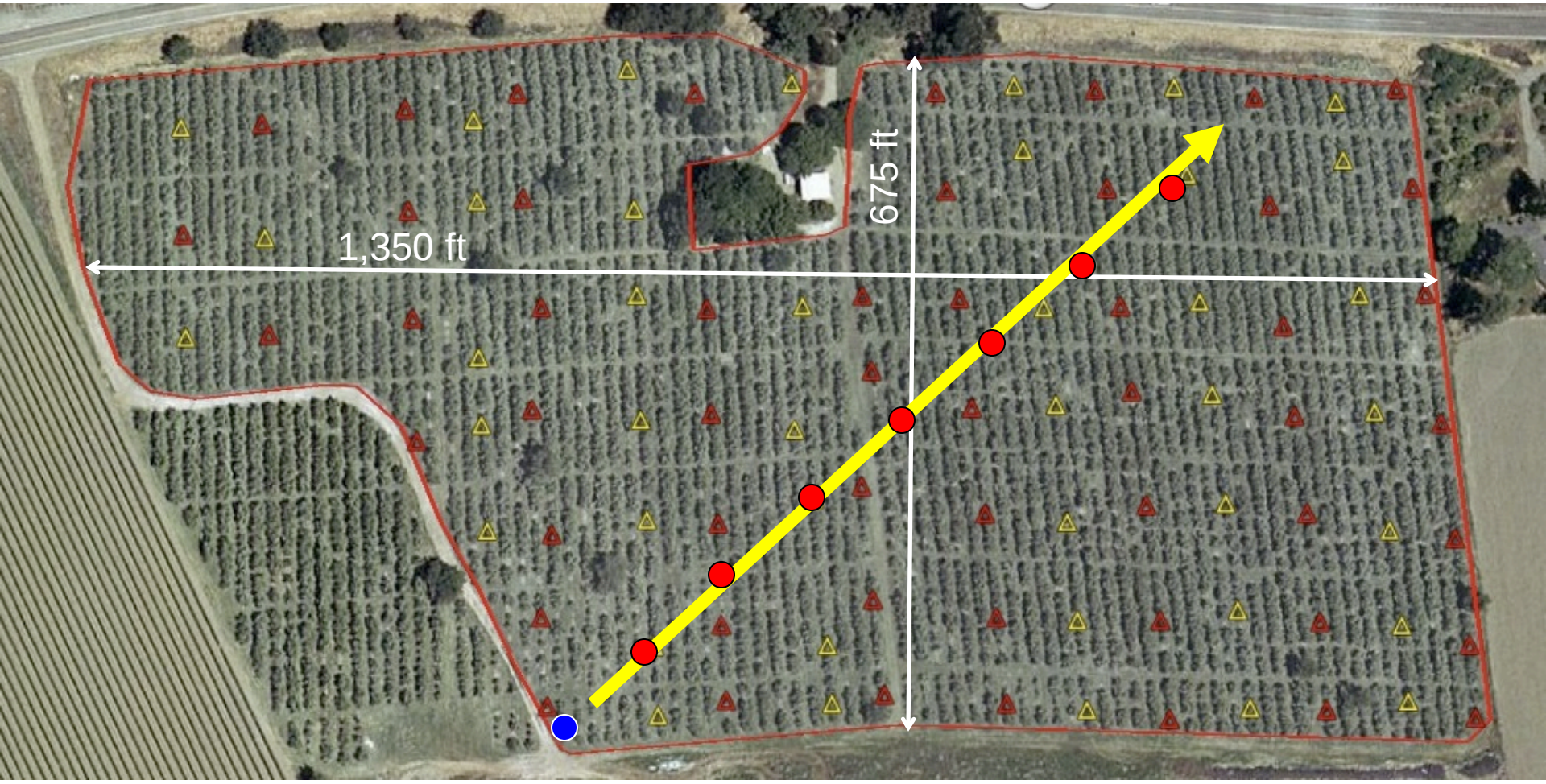
  For other insects, Lobesia, LBAM, pea moth, pheromones bind and are released later

  Residual plume images for codling moth suggested secondary release

  Do puffers impregnate the orchard such that the orchard becomes the emitter

Approach

- CM puffer run at standard settings in walnut and pear orchards
- Leaves collected after one week at different horizontal distances downwind from puffer and frozen
  - Distances of collection: <1, 17, 50, 100, 135 and 170 m
- In walnuts only, a vertical transect run at multiple distances from dispenser
- Replicates of 15 to 20 codling moth males were exposed to leaf samples in the wind tunnel
- Males were flown one-by-one, allowed a 3 minute response time



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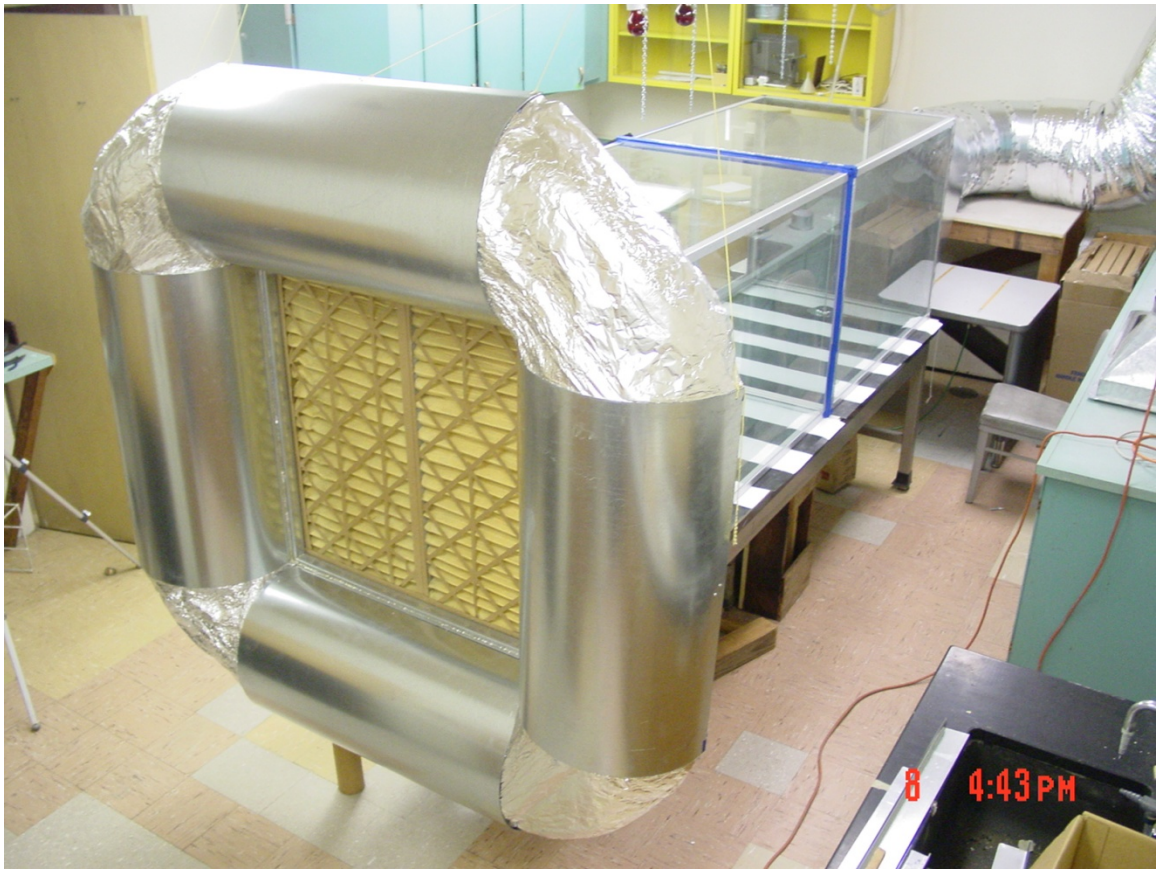
Moth Flight Wind Tunnel



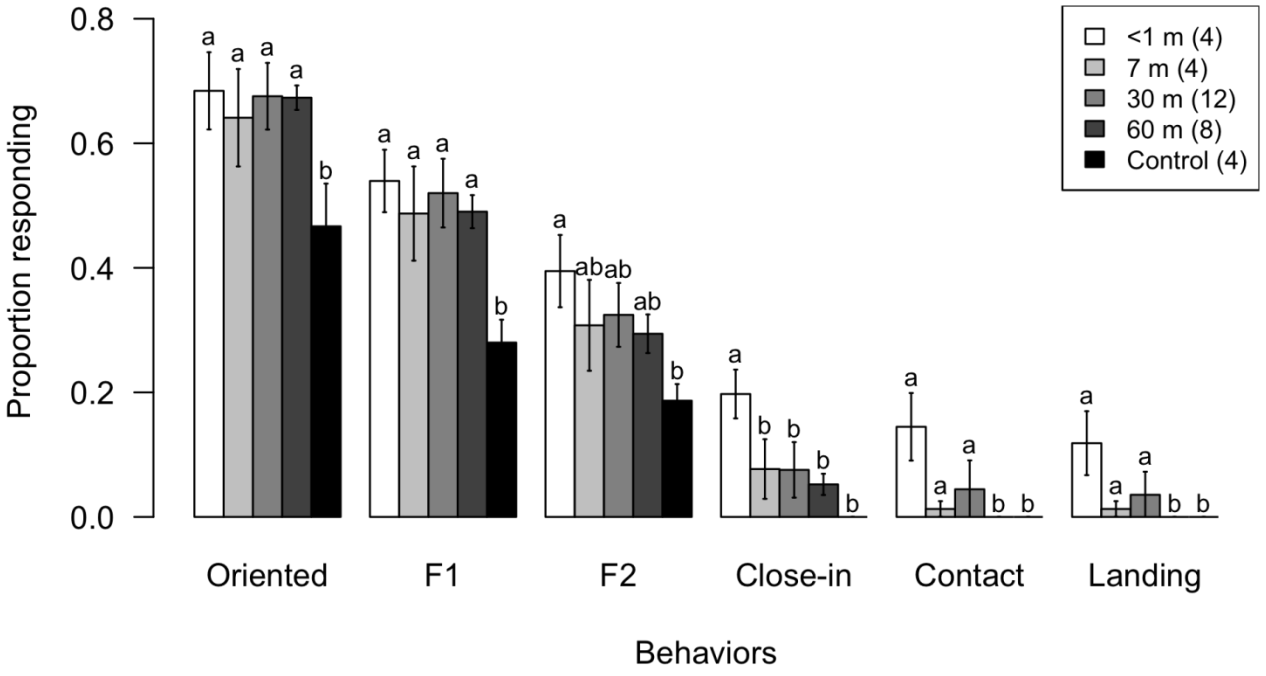
- Room air is filtered as it is pulled into the tunnel.
- Odor source placed at “upwind” position in tunnel
- Moths placed at downwind end of tunnel

Description of moth behaviors recorded in wind tunnel experiments

- **Oriented** - male finds the plume and starts oriented flight
- **F1** - upwind oriented flight 1/2 the length of the wind tunnel (around 1 m)
- **F2** - upwind flight up to $4/5^{th}$ of the wind tunnel length (around 1.7 m)
- **Close-in** - male approaches and zigzags closely (a few centimeters) to source
- **Contact** - male touches the source
- **Landing -** male lands on the source, walking and wing-fanning observed, excited

Attraction of Codling Moth Males to Pear Leaves Exposed to a Puffer Plume

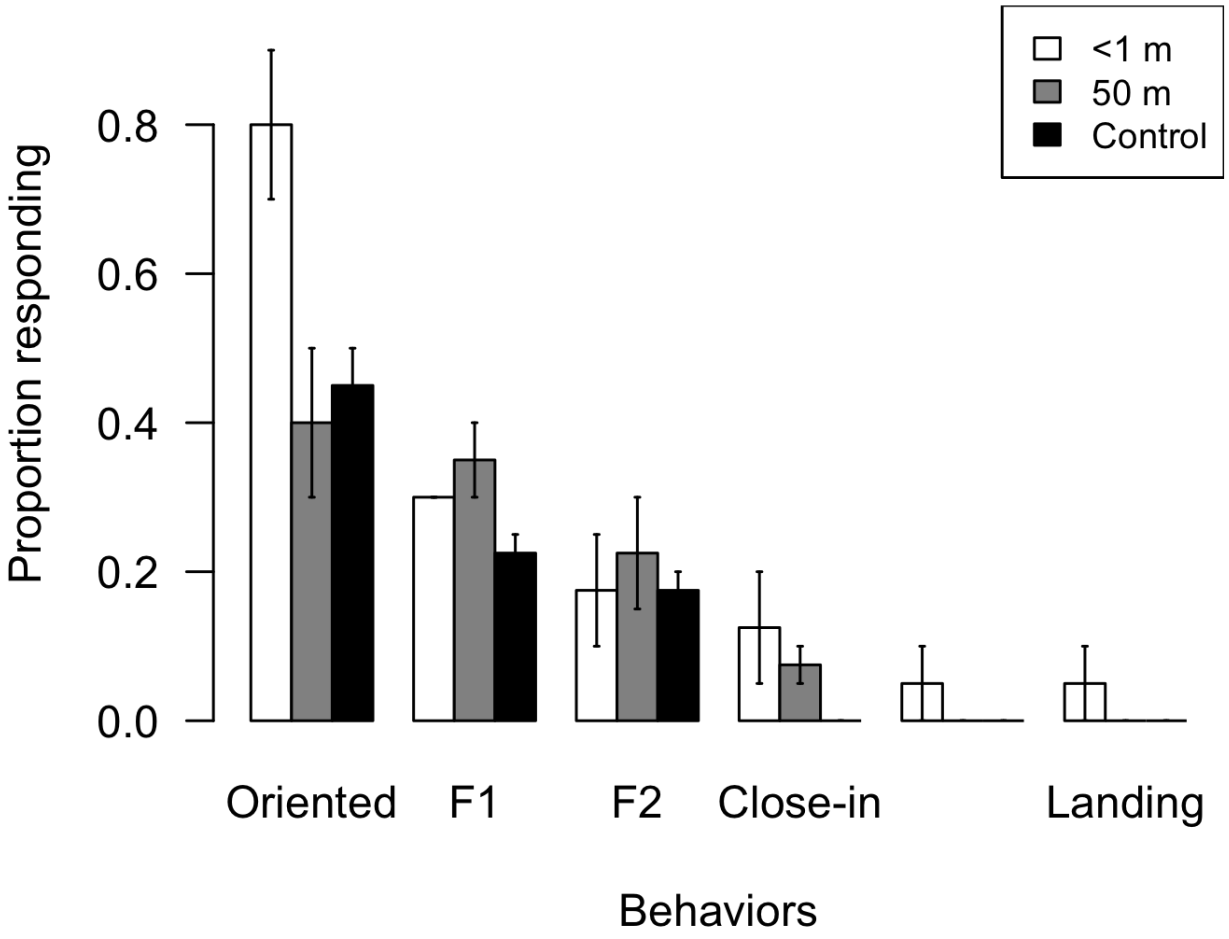
PEARS



- Controls stimulate upwind flight but no close approach
- Greatest response to leaf samples nearest puffer
- Complete flights observed in samples up to 30 m distant from puffer

Attraction of Codling Moth Males to Walnut Leaves Exposed to a Puffer Plume

WALNUTS



- Controls elicit upwind flight but no close approach
- Leaves at shortest distance elicit some complete flights (landing)
- At intermediate distance (50 m), some close-in behaviors observed
- Preliminary samples only

Changes in Codling Moth Movement in response to a puffer plume

- Do codling moths avoid, seek, or fail to move if exposed to high – dose plumes of puffers
- Are rates of movement affected, either positively or negatively?

ELISA Assays

1. Spray an area with a known protein

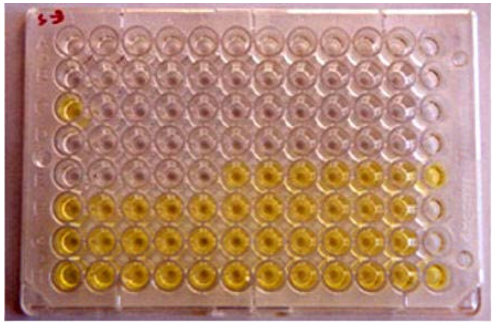
2. Capture insects in and around the area

3. Assay insects to isolate and bind protein to plate wells

4. Plate placed in a plate reader to measure absorbance of each well

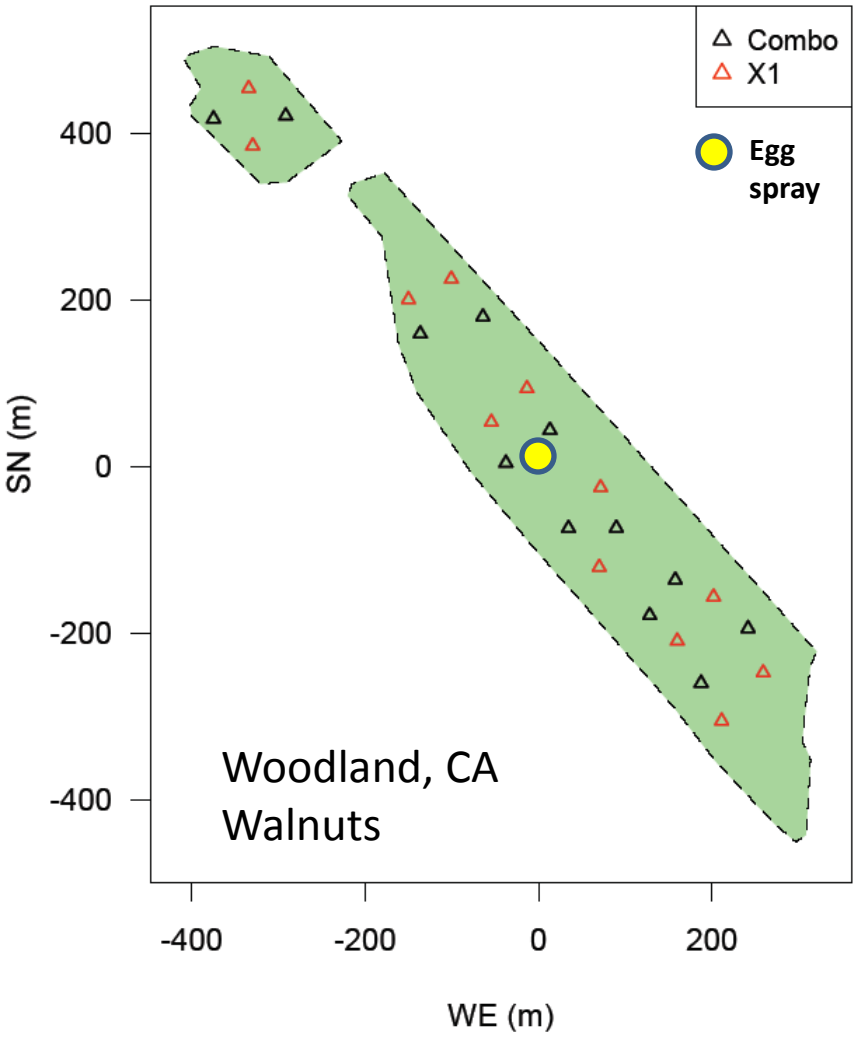
5 . Each moth is scored as either positive or negative for the protein

ELISA Assays



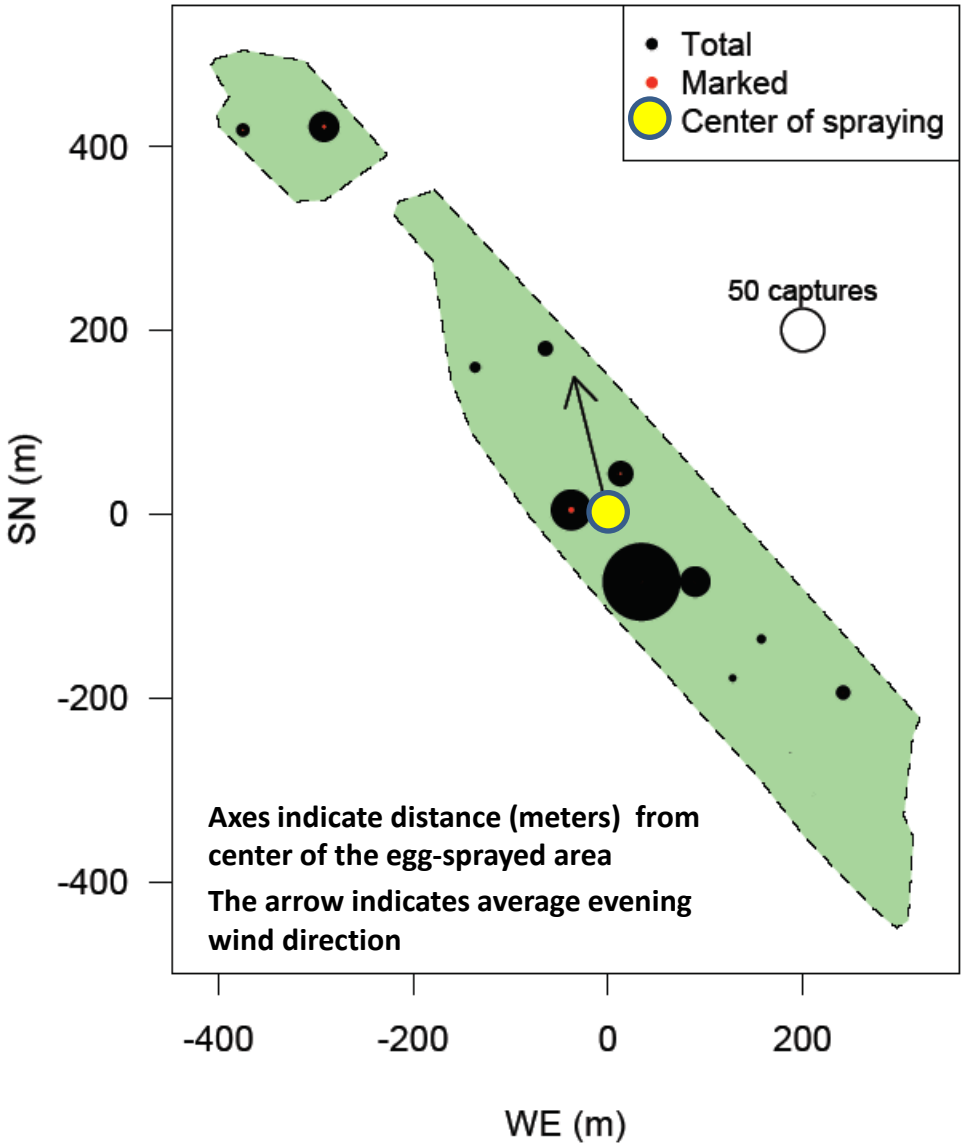
- **Enzyme-Linked Immunosorbant Assay**
  - **Antibody determined colormetric measurement determines presence of specific protein**
- **Cheap mass marking technique for insects**
- **Permits fine scale examination of movement**
- **Benefits over genetic techniques**

Methods - CM Movement Assays



- Site:
  - 30 acre Chandler + Hartley
- Treatment:
  - 1.1 acres (64 trees) sprayed with 10% solution of reconstituted egg white
- Evaluation:
  - Moths collected in 24 trap grid (12-1x, 12-Combo)
  - Elisa assays to detect egg protein marker
  - Anemometer recording of wind speed/direction

Total numbers of codling moth captured and marked insects at the trap locations.



- CM collection:
  - All combo and one 1x trap captured CM (366 moths)
- Elisa assays:
  - **10.7%** marked with egg protein
  - Most marked moths captured downwind of treatment
  - Orchard was a MD trial site with different pheromone treatments which may have impacted moth behavior
- Implication:
  - **Technique can demonstrate CM movement within or between orchards under different management strategies**

2010 Objectives

- Evaluate the effect of pheromone release rate on the functional size and strength of pheromone plumes
  - Determine the effects on trap suppression, female-baited trap suppression, and secondary release rates
  - Contrast the effect of pulsed vs. continuous release at the same release rates on plume size and efficacy
  - Evaluate if long-term adaptation of codling moth occurs when exposed to secondary release of pheromone from leaves.
  - Determine if codling moth movement is altered after exposure to high-dose pheromone emitters (puffers)

Objectives Continued

- Effect of reduced emission rates per puff on plume size and efficacy – large cost cutting potential
- Contrasting the plumes of single traditional emitters, meso emitters, and pheromones. Are they just part of a continuum or are do their effects (mechanisms) vary by emission rates?
- Continued evaluation of the meso-style emitters in field plots for suppression of codling moth damage
  - Registration of first meso already underway by Pacific Biocontrol
  - Potential near term registration of 2nd product (Suterra)
  - Need to have product challenged in more locations as well as within a final year of demonstration before grower adoption