# Development of alternative pheromone dispensing technologies for management of codling moth

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#### Introduction

Recent advances in pheromone mating disruption of codling moth have included newer pheromone delivery technologies: sprayable microencapsulated formulations and aerosol emitters ("Puffers"). Two companies, 3M and Consep, are currently developing sprayable formulations for suppressing codling moth. Both sprayable formulations are based on encapsulating small amounts of pheromone within tiny beads of various synthetic polymers. These beads are then applied using traditional spray application equipment. In contrast, aerosol emitters are being developed by several companies or groups including Paramount Farming, Consep Inc., Michigan State University Microsprayer, and researchers at Iowa State. The aerosol emitters rely on a few point sources that release high levels of pheromones, which is distributed downwind to other sections of the orchard.

Recent economic conditions continue to necessitate the development of management programs that help to minimize input costs and labor needs. While handapplied pheromone dispensers for codling moth have provided economically, and certainly biologically, effective management tools, hand applied dispensers require a relatively large labor supply at specific periods in the growing season. For some areas and in some locations, it is difficult to find an adequate labor pool. Similarly, the economics of some management systems using hand-applied dispensers have proven difficult to justify when specific conditions (e.g. excessive large tree canopies) or areas where codling moth does not require extensive pesticide treatments (e.g. 1-2 per season) make the dispensers unwieldy. Both of the newer technologies offer opportunities to potentially minimize labor needs and material costs, if reduced application rates are possible.

An emphasis was placed on 2 main research fronts for 2000: a) development and confirmation of our understanding of how pheromone plumes from aerosol emitters behave and b) determination of the potential of the sprayable formulations of codling moth pheromone for suppressing codling moth pheromone traps. Both studies were the initial steps necessary to justify more risky trials in which grower orchards will be placed at risk to damage from codling moth

In addition, both application techniques have the flexibility of fitting easily into multi-tactic programs or within the context of management decisions for other pests. For example, applications of sprayable pheromone formulations might overlap with other spray operations such as blight, thus helping to distribute application costs. Similarly, if coverage with sprayable formulations proves inadequate early in the growing season, then combination programs of the two techniques are possible given that the aerosol emitters can be turned off and on at will.

Finally, understanding how pheromone plumes behave and are structured in the field provides a way to better design and ultimately to optimize the placement of aerosol dispensers. If the plume from a single puffer can be defined in terms of minimal range (throw) or breadth under a variety of conditions, then puffer placement can be reduced to avoid excessive overlap in plumes and cost. Alternatively, understanding plume structure

also will allow us to understand how to place puffers such that here is enough redundancy in the pattern to account for any unexpected mechanical failures with the devices.

Studies from both pear and walnuts orchards are reported herein given that the same types of data are being collected for codling moth in both systems. While the studies shared similarities, the studies varied enough in experimental specifics or orchard conditions that the larger dataset greatly improves our ability to make more general conclusions. The complimentary nature of the studies allowed us to discover patterns that would not be discernable with only single commodity research. For example, differences in the apparent longevity of one sprayable formulation between pear and walnut orchards ultimately identified a potential problem that required changes in formulation strategies. Each set of trials was independently funded and supported by their respective commodity groups.

#### **Methods and Materials**

## I. Determination of pheromone plume shape and size under various orchard conditions from aerosol emitters: Paramount Puffers

#### General

Given that this research is in the early phases of development, indirect indices of program performance were used for program efficacy rather than place a grower's crop at risk. Thus, trap shutdown under a variety of conditions was used as the first step in assessing the efficacy of puffers, which will be followed by more direct damage assessments in 2001. Failure to provide high to absolute trap suppression is assumed to indicate program inadequacies, whereas complete trap shutdown is not assumed to be synonymous with damage suppression. Instead, successful trap suppression will result in moving to the next phases of program assessment (direct nut damage suppression). Sterilized codling moths were released in a uniform pattern into a grid of pheromone-baited traps. Using trap recapture of moths as an indicator of mating suppression, traps were checked every 1-3 days for each experiment period. Given that the sterile male releases also provide control of codling moth and thus confound any control assessment, no estimates of damage were made.

### Monitoring of codling moth

For all experiments Trece Delta VI traps, loaded with red septum 1 mg CM lures (Trece, 3111) were hung in the lower third of the tree. Traps placed in the lower portion of the tree canopy will normally be suppressed in pheromone treated orchards. Typically, when traps baited with 1 mg lures start to catch more than a few moths on a consistent basis, the grower will have to treat the orchard with a supplemental application of insecticide. Given that very high numbers of sterilized moths are being released throughout the orchard (up to 31,000 per week), the traps are not expected to necessarily be suppressed 100%. Instead the relative trap captures in the orchard are assumed to reflect the relative pheromone levels and indirectly, the relative level of suppression by the pheromone. Lures were changed out of each trap every two weeks.

#### **Aerosol Puffers**

Aerosol puffer devices used (Paramount Puffer CM, Paramount Farming Co.) were always hung in the uppermost third of the tree. Puffer devices were loaded with a formula of

23.50 % codling moth pheromone (8,10 dodecadien-1-ol) contained in an aerosol can. The aerosol emission of the devices were standardized according to the manufactures specifications, at 15 mg ai/puff and the device set to release 4 puffs per hour. The devices were programmed to run for either 12 or 24-hour cycles depending on the experiment.

#### Sterile moths

Sterile codling moths were obtained from the sterile insect release facility operated by Sterile Moth Release Program in Penticton, British Columbia. Moths were typically shipped on a Monday from Canada, received in Berkeley on Tuesday and taken to the field for release that same day. Unfortunate, on two occasions, moths were dead on arrival due to shipping errors. Moths, contained in sealed petri dishes were kept cool on blue ice until release in the field. Petri dishes were placed into a limb crotch in the lower third of each pear tree. Releases of ca. 800 moths per release point were made for all experiments, hereafter referred to as a sterile release. Sex ratios are approximately 50:50. Sterile moths are identified by a pink coloration of the gut contents, which is the result of a dye placed in to their diet by the rearing facility. Moths were verified as being sterile in the field by crushing and noting the presence of the pink color of their internal fluids.

## **Environmental Monitoring**

Data relating to the daily wind patterns was collected for this study. The data was collected from several sources for both the walnut and pear trials. For the walnut trials, data was collected using the PestCast weather network and database on the Internet. Three different weather station locations were used to cross verify the daily information. All stations were not more distant than 10 miles from the study site. The closest weather station was approximately 2 miles northwest of the study site. Each station utilizes a RM Young Wind Sentry, 3001 anemometer and vane. The anemometer is accurate to within 2 mph, according to the manufacture and the vane calculates an hourly average wind direction based on the settings programmed by the PestCast staff. Data was downloaded through the Internet at regular intervals throughout the study to monitor for any significant changes in the daily wind patterns.

For the pear trials, data loggers for wind speed and direction were established in the project using an OWL data logger system (Electronically Monitored Ecosystems, Berkeley, CA). The current, average and maximum wind speeds and directions were recorded every 10 minutes over the course of each day. Two sets of anemometers and wind vanes were installed in the orchard, one at the upper margin of the tree canopy (ca. 16 feet) and the other in the midcanopy (ca. 11 feet). The anemometers and vanes were located at the same location in the orchard. In addition, data was collected using the PestCast weather network and database on the Internet. A PestCast weather station was chosen to corroborate the daily information from the in-orchard OWL system. The PestCast station is located at Canal Ranch, approximately 2 miles from the study site. The PestCast station utilizes a RM Young Wind Sentry, 3001 anemometer and vane. The anemometer is accurate to with in 2 mph, according to the manufacture and the vane calculates an hourly average wind direction based on the settings programmed by the PestCast staff. Data was downloaded through the Internet at regular intervals throughout the study to monitor for any significant changes in the daily wind patterns.

### **Experiments and Grids – Walnut Trials**

#### **Orchard conditions**

The orchard used for this study was of Chandler variety walnuts planted in 1994. The orchard dimensions are approximately 1210' x 3180' which covers 74 acres and is orientated length wise in a west-east direction. Tree rows are planted in a north-south direction with 22' between trees with in each row and 19' between rows. The dominant wind direction at the site was estimated to be from the west, and would therefore travel perpendicular to the tree rows. The orchard is bordered on the south by cherries and apples, on the west by apples, on the north by an open field and on the east by a highway.

## **Experiments and Grids - Walnuts**

A series of three different trap grids were utilized during the course of this project (Figure 1). Puffer devices were always placed near the assumed upwind portion of the grid plot. The grids were established to examine the pheromone plumes behavior under particular conditions. Sterile moths were released into the grid and all release points were exactly two rows, 38' directly east (i.e. downwind) of each trap. Each grid was used for a series of experiments. Experiments were setup in one or two week intervals. Two-week experiments consisted of a puffer "on" period followed by a puffer "off" period. These periods were used to first establish a pheromone plume in the orchard, then to turn off the puffer device and examine the presence and effects of any "ghost" plume. The "ghost" plume was suggested from some research in New Zealand, in which codling moth pheromone was shown to bind to the surface of foliage, then be released over several days, If this is true, then we were interested in understanding if the pheromone plume from a puffer would likewise have a residual effect even if the puffers were turned off. One-week experiments were always puffer "on" periods. Unless other wise noted, all experiments received one moth release at the beginning of each "on" and "off" period.

## Grid Pattern 1- Single Puffer Placed Within an Orchard (Used for Experiments 1a, 1b, 2, 3a, and 3b) - Walnuts

Grid 1 was setup to confirm the presence of the pheromone plume and to investigate the possible existence of a "ghost" plume of residual pheromone after the puffer device had been turned off. The grid was composed of 30 traps (Trece Delta VI) and one puffer device. Traps were hung in 6 rows with 5 traps per row, each spaced 198' (9 trees) apart. The first row was 798' west of the puffer device; this was done to ensure an upwind control trap row. We expected the upwind row traps to be unsuppressed by the pheromone. The puffer device was hung, facing east, in the uppermost third of the center trap tree of the second row. Each of the remaining four rows of traps were spaced 456' from each other, with the final row east at a distance of 1824' from the puffer device. We used this grid pattern for a series of three experiments.

## **Experiments 1-3 - Walnuts**

Experiment 1a and 1b. Pre-puffer Trials. Before any pheromone application in the orchard, an attempt was made to verify a random distribution of moth recapture in the absence of any pheromone plume. Therefore, the puffer was not turned on for the duration of the both replicates of experiment 1. We assume that the lack of any discernable pattern in the distribution of trap recapture indicates no unexpected effects of orchard structure. For Experiments 1a and 1b, the 30 traps were checked on days 3, 6, and 7, following the sterile release. The recaptured moths were removed and verified as sterile (pink coloration of internal fluids) or wild individuals.

Experiment 2. Plume Structure – 24 hour. Experiment 2 was performed to determine the presence of a pheromone plume. However, due to time constraints experiment 2 was not repeated. A 24-hour emission cycle was programmed into the puffer device (15 mg ai/puff at 4 puffs/hour) that delivered a total of 1440 mg pheromone per each cycle for a grand total of 11.52 grams pheromone emitted over an 8-day period. The traps were checked, and recaptured moths removed and verified as sterile or wild on days 1 through 3 and again on day 8 following the sterile release.

Experiment 3a and 3b. Plume Structure - 12 hour and Ghost Imaging. The third set of experiments was designed to test 2 objectives: a) the size and shape of a pheromone plume emitted and b) how long does the effect of the pheromone plume last after the puffer device is turned off. As such, each experiment lasted 2 weeks; for the first week the puffer device was turned on, for a 12-hour cycle each day and then was turned off for the next week to look for any "ghost" or residual effects. The puffer device was programmed to emit on a 12-hour cycle, from 6 pm to 6 am, at the standard emission rate (15 mg ai/puff at 4 puffs/hour) for a total of 720 mg pheromone delivered per cycle. The timing of the emission was set to roughly coincide with assumed daily moth flight, occurring around the time of sunset and through the early morning hours. A total of 3.6 g of pheromone was emitted over a 5-day "on" period for the first replicate of this experiment. During the "on" period, traps were checked, moths removed and verified as sterile on days 1, 4 and 5 after the sterile release. A second sterile release was made the day the puffer was turned off. During the "off" period traps were checked and moths verified and removed on days 1 through 3, 6 and 7 after the sterile release. The second replicate, 3b, of this experiment was performed immediately following the "off" period of experiment 3a. A total of 5.04g of pheromone was emitted during the 7-day "on" period. The traps were checked; moths were removed and verified as sterile on days 1 through 3, 6 and 7 after the initial sterile release. As before, a second sterile release was made the same afternoon that the puffer was turned off. Traps were again checked on days 1, 2, 3 and 6 after the sterile release.

## Grid Pattern 2 – Single Puffer Placed on Orchard Border (Experiments 4a and 4b,) - Walnuts

The puffer device was moved from the interior of the orchard to the western, windward edge. This grid was designed to examine the effects of orchard edges and direct wind exposure on the pheromone plume in a series of 2 experiments. As was the protocol with grid 1, experiments in grid 2 were composed of alternate puffer "on" and puffer "off" periods. Grid 2 was composed of 39 traps (Trece Delta VI) and one puffer device. Traps were hung in five rows, with in each traps were spaced 132' (6 trees) apart. The first trap row was in the west edge row of the orchard. The remaining four rows of traps were spaced 190' apart with the

final row a distance of 760' from the puffer device. The puffer device was hung, facing east, in the uppermost third of the center tree in the first western orchard row.

## **Experiments 4a and 4b. Plume Structure on Edges - Walnuts**

Similar to experiments 3a and 3b, experiments 4a and 4b lasted for 2 weeks, consisting of puffer "on" and "off" periods to test for "ghost" plume effects at the orchard edge. Experiments 4a and 4b used a single puffer, programmed to emit on a 24-hour cycle at the standard rate of 15mg ai/puff, for a total of 1440 mg pheromone emitted each 24-hour cycle. A total of 10.08 g of pheromone was emitted over a 7-day "on" period for the first replicate of this experiment. During the "on" period, traps were checked, moths removed and verified as sterile on days 1 through 3, 6 and 7 after the sterile release. During the "off" period traps were checked and moths verified and removed on days 1 through 3, 6, 7 and 8 after the puffer was turned off. There was no sterile release made for this "off" period due to problems with the shipment form Canada, which resulted in the moths arriving in Berkeley dead. The second replicate, 4b, of this experiment was preformed immediately following the "off" period of experiment 4a. A total of 8.64 g of pheromone was emitted during the 6-day "on" period. The traps were checked; moths were removed and verified as sterile on days 1, 2, 5 and 6 after the sterile release. A second sterile release was made the same afternoon that the puffer was turned off. Traps were again checked on days 1 through 3, 6 and 7 after the sterile release.

## Grid Pattern 3 - Multiple Puffers Placed on Orchard Borders - Walnuts

Grid 3 represents an attempt to test the full-scale abilities of the puffer in whole orchard (ca. 74 acres) suppression of traps. This grid consisted of 36 traps (Delta VI) hung in 7 rows; each row contained 5 traps with the exception of row 2 which contained 6 traps. The first trap row was located in the western edge of the orchard. Each of the remaining rows was spaced 494' (26 rows) apart and the final row was no more than 513' from the eastern edge of the orchard. With in each row, traps were spaced 198' (9 trees) apart with the end traps of each row located 110' from the orchard edge. For this grid, 5 puffer devices were placed on the western edges of the orchard. Placement of the puffers was chosen to give the greatest coverage and least over lap of plume distribution. The use of only 5 puffers was used to aggressively push the program to try and identify "holes" in pheromone coverage. A higher level of redundancy will be needed for actual implementation efforts. Three puffers were hung in the first western row of the orchard. The first was hung in the same location as in grid 2, in the center tree, and the other two puffers were hung on the north and south edges respectively, each 132' (6 trees) from an edge. The remaining two puffers were hung in row 14 from the west orchard edge in the edge tree of each end of the row, one on the north and one on the south edge.

## **Experiment 5 – Whole Orchard Suppression - Walnuts**

Experiment 5 consisted of an "on" period of 7 days followed by an "off" period, which included trap reads for 10 days. As in experiments 2 and 4, experiment 5 utilized a 24-hour emission cycle at the standard emission rate of 15 mg ai/puff. A total of 1440 mg ai was delivered in to the orchard per puffer device, totaling 7.2 g pheromone during each 24-hour cycle. A grand total of 50.4g of pheromone was emitted over the seven-day "on" period into the entire orchard. This was the equivalent of approximately 681 mg of pheromone per acre applied. For the "on" period traps were read, moths removed and verified as sterile or wild on days 1 through 3 and days 6 and 7 after the initial sterile release. On day 7 following the initial

sterile release, puffers were turned off, removed from the orchard and a second sterile release was made. For the "off" period traps were read on days 1 through 3, and days 6 and 10 after the second sterile release. Due to seasonal time constraints and the lack of any more sterile moths for the season, this experiment could not be replicated.

## **Experiments and Grids - Pear Trials**

#### General - Pears

This project is a continuation of work we began in 1999 and utilized much of the same methods and materials that generated clear results in the previous year. Sterilized codling moths were released in a uniform pattern in to a grid of pheromone-baited traps. Using trap recapture of moths as an indirect indicator of mating suppression, traps were checked at intervals appropriate for each experiment.

#### **Orchard Conditions - Pears**

The project site was an established orchard of Bartlett variety pears, located outside Walnut Grove, California. The orientation of the orchard is long in the north-northeast/south-southwest directions and covers approximately 85 acres. A significant feature of this orchard was a cut-through road running diagonally through it from the northwest to southwest corner. This particular situation results in an orchard with two large triangular blocks of trees divided by an open swath approximately from 40-80 feet wide. The tree spacing is regular with 20' between trees in rows and 20' between rows. We defined rows as such using the grower's tractor run pattern, however there is no apparent row pattern in the tree structure. For the purpose of this study, row orientation was defined as running in a north-south direction.

#### **Experiments and Grids - Pears**

A series of three different trap grids were utilized during the course of this project (Figure 13). Puffer devices were always placed near the assumed upwind portion of the grid plot. The grids were established to examine the pheromone plumes behavior under particular conditions. Each grid was used for a series of experiments. Sterile moths were released into the grid of traps. All release points were exactly two trees, 40' away from each trap, but direction of the release point from each trap varied with changes made to the individual grid layouts. Experiments were setup in one or two week intervals. The two-week experiment consisted of a puffer "on" period followed by a puffer "off" period. These periods were used to first establish a pheromone plume in the orchard during the puffer on period, and then to examine the residual effects of the plume after the puffer had been turned off. One-week experiments were always puffer "on" periods. Unless otherwise noted, all experiments received one moth release at the beginning of each period.

## Grid Pattern 1, Single Puffer Placed Within an Orchard (Experiment 1a and 1b) - Pears

Grid 1 (Figure 13) was composed of 30 traps set in a grid of five rows with six traps per row. Row 1 was located on the west side of the orchard, approximately 160' from the edge. Rows 1 through 4 were each separated by 280' and row 5 located 240' from row 4. Traps were separated by 360' within rows with the exception of row 3 where the 3<sup>rd</sup> and 4<sup>th</sup> traps were offset by 140' (7 trees) to the north and south respectively. This offset placement was the necessitated by missing trees in the cut-through. A single puffer device was hung midway

between the 3<sup>rd</sup> and 4<sup>th</sup> traps in row 2 from the west edge of the orchard. One petri dish of sterile codling moths was released exactly at 40 feet (2 trees) directly east of every trap for each experiment.

## **Experiments 1a and 1b - Pears**

Experiments 1a and 1b represent replicated tests to establish a detectable pheromone plume in the orchard. The puffer device was programmed with a 24-hour emission cycle (15 mg ai/puff at 4 puffs/hour) that delivered a total of 1440 mg of pheromone each cycle for a grand total of 10.08 g pheromone emitted over each 7-day period. For both experiments traps were checked and moths removed and verified as sterile or wild on days 3, 6, and 7 after the sterile release.

## Grid Pattern 2, Single Puffer Device Placed Within an Orchard, High Density Trap Grid Nested in Low Density Grid (Experiments 2, 3 and 4) - Pears

Grid 2 (Figure 13) was composed of 40 traps hung in two density patterns. A high-density grid surrounding the puffer device was set within the lower density grid pattern used in grid 1 with the exception that the northern trap was removed from each row. The high-density grid was established by adding 17 additional traps half way between the grid 1 traps within rows as well as adding new rows of traps half way between the rows of the grid 1 traps. The result was a nested grid of 23 traps separated by 180' (9 trees) within rows and 140' (7 trees) between rows. The remaining 17 traps remained in the same locations as in grid 1. One petri dish of sterile moths was released 40' (2 trees) directly north of every trap for each experiment. The location of the release points was moved in response to information collected from the onsite data logger indicating the dominate wind direction to be from the south-southwest direction.

### **Experiments 2 and 3 - Pears**

Experiment 2. This experiment lasted for 2 weeks and consisted of a 7-day puffer "on" period followed by an 8-day puffer "off" period. The purpose of the puffer "off" period was to examine the presence and any potential effects of a residual pheromone "ghost" plume. A sterile release was made at the beginning of each experimental period. The puffer device was programmed with a 24-hour emission cycle (15 mg ai/puff at 4 puffs/hour) that delivered a total of 1440 mg of pheromone per day for a grand total of 10.08 g pheromone emitted over the 7-day period. During the "on" period, traps were checked, moths removed and verified as sterile on days 3, 6 and 7 after the initial sterile release. The second sterile release was made the same day that the puffer device was turned off. During the "off" period traps were checked and moths verified and removed on days 1 through 3, 6 and 7 after turning the puffer off.

Experiment 3. Experiment 3 was another attempt to establish a pheromone plume after the "off" period of experiment 2. The puffer device was programmed with a 24-hour emission cycle at the standard rate (15 mg ai/puff at 4 puffs/hour). A total of 11.52 g of pheromone was emitted over the 8-day duration of this experiment. A sterile release was made the same day the puffer was turned on. The traps were checked, moths removed and verified as sterile on days 3 and 8 following the sterile release.

## Grid Pattern 3, Multiple Puffers Placed near Orchard Borders - Pears

Grid 3 (Figure 13) represents an attempt to test the limitations of the puffer in whole orchard (ca. 85 acres) suppression. The grid consisted of 33 traps hung in five rows, row 1, 2 and 5 each contained seven traps, while rows 3 and 4 each contained 6 traps. The spacing of rows and traps within rows was similar to that of grid 1. Row 1 was located on the west side of the orchard, approximately 160' from the edge. Rows 1 through 4 were each separated by 280' and row 5 located 240' from row 4. Traps were separated within rows by 360' with two exceptions: in row 3 the 3<sup>rd</sup> trap from the north and in row 4 the 7<sup>th</sup> trap from the north were both missing because of the cut-through road. For this grid, 6 puffer devices were used. Five puffers were placed throughout the southern end of the orchard and one in the northern end. Placement of the puffers was chosen to give the greatest coverage and least over lap of plume distribution. The use of only 6 puffers was used to aggressively push the program to try and identify "holes" in pheromone coverage. A higher level of redundancy will be needed for actual implementation efforts. The placement of the six puffers is as follows: 6<sup>th</sup> trap tree from the north in rows 1, 3, and 5; 4<sup>th</sup> trap tree from the north in rows 1 and 5 and 1<sup>st</sup> trap tree from the north in row 3. One petri dish of sterile moths was released 40' (2 trees) directly north of every trap for the experiment. The location of the release points and the puffer devices was chosen in response to information collected previously from the on-site data logger indicating the dominate wind direction to be from the south-southwest direction.

## **Experiment 4 - Pears**

Six puffer devices were programmed with a 24-hour emission cycle (15 mg ai/puff at 4 puffs/hour) that delivered a total of 8640 mg of pheromone each cycle for a grand total of 103.7 g pheromone emitted in to the orchard over the 12-day duration of the experiment. This was the equivalent of approximately 1218 mg of pheromone per acre applied. The sterile release was made 24 hours following the puffers being turned on. During the experiment period, traps were checked; moths removed and verified as sterile on days 1, 4, 7 and 12 after the sterile release.

#### II. Attract and Kill - Pears

A single rate of Last Call<sup>TM</sup> CM (IPM Technologies, Inc., Portland OR) was compared to a grower standard (no pheromone) treatment. Three replicates of each treatment were established across two orchards of Bartlett variety pears. The Hood orchard is located in the Sacramento delta and the Comstock orchard is located about 10 miles NE of Stockton, CA. Two replicates located in the Comstock site measured 1.93 acres with each separated from adjacent treatments by 200 feet. Plots in the Hood orchard were 2 acres, again separated by a minimum of 200 feet. Last Call<sup>TM</sup> was applied to the upper 1/3 of the canopy at the label rate of 1200 droplets per acre on 6/12/00 (Hood) and 6/13/00 (Comstock).

We conducted three releases of sterile codling moths purchased from the Sterile Moth Release Program in Penticton, British Columbia. Two dishes of sterile moths (approximately 800 per dish) were released per plot by placing the petri dishes securely in a tree crotch or branch. Release points were centered between pairs of traps in each plot.

Four traps (Pherocon® Delta VI, Trece, Inc.) were set in a square pattern centered in each plot. Minimum distance between traps was 80 feet (Hood) or 100 feet (Comstock). Traps were baited with CM standard 1 mg lures (Trece 3111) for sterile

releases conducted June 13 (Comstock) or June 14 (Hood) and June 20 (both sites). Lures were changed to the plant volatile "DA" lure (Trece 8693) for the sterile release conducted July 18. In addition, two traps baited with a single droplet of Last Call<sup>TM</sup> were placed in the center of the grower treatments at Comstock. Traps were read daily following the first release, every three days following the second release, and daily following the third release.

## **III. Pheromone Application Trials - Sprayable Formulations**

#### **Pear Trials**

A single rate of a sprayable product under development by 3M was compared to Isomate and grower standard treatments. Three replicates of each treatment were established across two orchards of Bartlett variety pears. One replicate was located near Hood, CA in the Sacramento delta and two replicates were situated in the Dal Porto orchard near Linden, CA. Sites are characterized as follows:

Hood ranch in the Sacramento delta is a single planting on a 16-foot square. Rows run approximately east-west. Treatment plots were 2 acres (304 feet by 288 feet or 19 rows by 18 trees) with approximately 200 feet separation between plots. Isomate was applied to the upper 1/3 of the canopy on 5/12/00 at 400 dispensers per acre. The 3M product was applied on 5/16/00 at a rate of 50 g ai per acre using 400 gallons spray per acre with a speed sprayer.

The Dal Porto ranch east of Stockton is a single block with row orientation north-south. It is double planted on an 11 foot by 22 foot spacing and is pruned for very open canopy between rows. Treatment plots were 1.8 acres (198 by 396 feet or 9 rows by 36 trees) with approximately 200 feet separation between plots. Plots were long in the north-south orientation. Predominate wind direction was observed to be from the west. Two replicates of each of the three treatments were designated. Isomate was applied to the upper 1/3 of the canopy on 5/12/00 at the rate of 400 dispensers per acre. 3M spray was applied on 5/23/00 at a rate of 50 g ai / at 400 gallons / acre by 500 gal air blast sprayer.

Four traps (Pherocon® Delta VI) total per plot were hung following the spray applications to monitor codling moth. Two traps were hung low and baited with standard 1 mg CM lures (Trece 3111); two traps hung high were baited with 10x CM lures (Trece 3160). At the orchard in Hood, four traps were placed in a square pattern centered in each plot with minimum separation of 80 feet between traps. Lure loads were randomly assigned to one of the four traps. Because plots were long and narrow at Dal Porto, all four traps were hung in the middle row of each plot. Lure load was randomly assigned to the first trap in a plot. The second trap in the pair was baited with the alternate lure. The next two traps mirrored the assignment of the first pair. Thus, trap pattern was either 1-10x-10x-1 or 10x-1-1-10x within a plot. This was an attempt to equalized trap exposure relative to sterile moth releases. Traps within a row were separated by 77 feet.

Sterile moths purchased from the Sterile Moth Release Program in Penticton, British Columbia were released weekly beginning 5/17/00 (Hood) or 5/23/00 (Dal Porto) and continuing for 10 weeks. Two dishes of sterile moths (approximately 800 per dish) were released per plot. Releases were made into the row midway between trap rows at Hood, and one row east of the trap row midway between each pair of traps at the Dal Porto orchard. Traps were read on a predetermined schedule coinciding with other data being collected. Thus, moths were counted and identified as our sterile release moths on

days 1, 3, 5, 7, 9, 11, 14, 17, 21, 24, 28, 31, 35, 38, 43, and 50 following the spray application and weekly thereafter through 8/9/00.

#### **Walnut Trials**

Consep. A single rate of a sprayable product under development by Consep was compared to Isomate and grower standard (no pheromone) treatments. Three replicates of each treatment were established across two varieties of walnuts in the Dondero orchard approximately 10 miles east of Stockton, CA. The site is about 146 acres planted to Vina, Hartley, Serr, and Chandler varieties in solid blocks. Two replicates of all treatments were placed in the Vina block and one replicate was placed in the Hartley block. The Vina's were approximately 25-30 feet tall, while the Hartley block was more densely canopied with trees 30-35 feet high. Tree spacing was 44 feet within rows and 22 feet between rows in a diamond planting. Treatment plots were 2 acres (286 by 308 feet) with a 200-foot separation between plots. All treatments including the nonpheromone treated Grower plots, the Consep trial plots and 3M trial plots were interspersed. Isomate was applied to the upper 1/3 of the canopy at the rate of 400 dispensers / acre on 5/12-13/00. The Consep application was made 5/17/00 at a rate of 20 g ai /100 gal / acre with a 500 gallon speed sprayer. Four traps (Pherocon® Delta VI) total per plot were hung following the spray applications to monitor codling moth. Two traps were hung low and baited with CM standard lures (Trece 3111); two traps hung in the upper canopy were baited with CM 10X lures (Trece 3160). The four traps were placed in a square pattern centered in each plot with an 88-foot separation between traps. Lure loads were randomly assigned.

Sterile moths purchased from the Sterile Moth Release Program in Penticton, British Columbia were released weekly beginning 5/17/00 and continuing for 10 weeks. Two dishes of sterile moths (approximately 800 per dish) were released per plot. Sex ratios were approximately 50:50. Releases were made into each trap row of the plots, midway between the two traps. Traps were read on a predetermined schedule coinciding with other data being collected. Thus, moths were counted and identified as our sterile release moths at days 1, 3, 5, 7, 9, 11, 14, 17, 21, 24, 28, 31, 35, 38, 43, and 50 following the spray application and weekly thereafter through 8/2/00.

3M Sprayable. Six rates of a sprayable pheromone product under development by 3M were compared to Isomate and grower standard (no pheromone) treatments in the Dondero orchard described above. Because of the space requirements and broad range of rates in this trial, the treatments not replicated. However, three replicates of each control plot (Isomate and grower standard) were established as described above. Two replicates of each control were placed in the Vina block and one replicate was placed in the Hartley block. All 3M plots were located in the Vina block. Spray applications were made 5/17/00 with a 500 gallon speed sprayer at the following rates: 5, 10, 20, 40, 80, and 150 g ai / 100 gallons / acre. Protocols for sterile moth releases and monitoring codling moth were followed as described above.

#### Results

## I. Determination of pheromone plume shape and size under various orchard conditions from aerosol emitters: Paramount Puffers

Only representative figures are shown within this report given the size and number of potential figures that could be shown.

#### Walnuts

#### **Experiments 1-3**

Experiment 1a and 1b. Pre-puffer Trials.

No discernable pattern for moth capture was observed in either Experiments 1a or 1b. As shown in Figure 2 for Experiment 1b, moth recapture appeared sporadically throughout the orchard with high counts observed at 912 feet from the proposed placement of the puffer. Upwind edges also caught high numbers of codling moth as would be expected from conventional orchards. Therefore, the assumption that moth releases would not show any apparent patterns before turning on the puffer device was supported by these data.

## Experiment 2. Plume Structure – 24 hour.

The first experiment used a conservative 24-hour cycle to see if a puffer plume could be detected within walnut orchards (Figure 3). Despite the wide spacing of the walnut trees and open nature of the first 6-9 feet of the orchard height, almost all traps downwind of the puffer were suppressed to less than 20 moths per trap compared to almost 60 moths per trap along the upwind edge. Similar to experiences in pears, the leading eastern edge of the orchard also experienced some trap recapture. Therefore, the plume in the walnut orchard appears to be at least 1800 feet in length and 600-800 feet in width, as suggested by trap suppression.

### Experiment 3a and 3b. Plume Structure – 12 hour and Ghost Imaging.

Despite only using a 12-hour emission cycle, a similar pattern was observed in Figure 4 as in Figure 3. When a single puffer was placed within an orchard, almost all traps downwind were suppressed to less than 20 moths per trap compared to 100 moths per trap upwind. The leading northern edge had a few traps recapture some moths, whereas traps on the southern edge were suppressed. In Figure 5, a three-day pattern emerged after the puffers were turned off. Twenty-four hours after the puffer was turned off, the residual image, or "ghost image" held for almost the entire downwind areas of the orchard. On both the southern and northern edges, increased trap recaptures were noted but spotty. On day 2 after shutdown, the total number of moths was fairly low, but higher counts were observed in the center and downwind of the puffer at 1368 feet. By day 3, counts were highest at 912 feet and in the center again suggesting that the pheromone was continuing to dissipate by days 4-6. The pattern continued to dissolve with higher counts observed on the leading northern edge and downwind at 912 feet of the puffer (Figure 6).

#### **Experiments 4a and 4b. Plume Structure on Edges**

High trap counts were observed along the entire margin of the orchard despite placement of the puffer at the edge (Figure 7). However, by 190 feet, the traps were again effectively suppressed by the puffer thus suggesting a relative rapid spread of the

pheromone from the point source. Some moths were caught in the rows at 190' from the puffer, but they were sporadic in nature and fairly low (9 or less). These data also confirm data from pear trials that upwind edges of puffers are not adequately suppressed and mating would be expected along these margins. Since traps placed as close as 66 feet away were not effectively suppressed, it also suggests that a tighter placement of puffers would not be expected to resolve this problem. The residual effect of the plumes appears to last the next 3 days without any signs of diminishing (Figure 8), and this was also seen for days 4-6 (Figure 9). However, caution must be used when trap counts beyond three days are used in that moth mortality could be increasing over time as well as upwind movement of the moths may be functionally taking them out of the experimental plots. So, lack of recapture of moths after 3-6 days may not reflect pheromone levels accurately.

## **Experiment 5 – Whole Orchard Suppression**

Placement of five additional puffers up for the 74 acres did not dramatically improve the trap suppression compared to the single puffer trials. Instead the upwind edges remained unsuppressed, whereas the downwind areas of the orchard were effectively suppressed (Figure 10). A small recapture was made in the northeastern corner of the orchard as well, but again, this was located along the margin of the orchard. When the puffers were turned off, the residual image appeared to remain for the first 3 days (Figure 11) as well as for days 4-6 (Figure 12). These data suggest that the number of puffers can be dramatically reduced if trap suppression proves synonymous with damage suppression.

#### **Pears**

## **Plume Structure**

The results of Experiment 1a-Pears for the first 3 days are shown in Figure 14. The results are overall similar to previous results but the plume was less clear in that trap recaptures were observed at 560 feet in an eastern direction. We would have expected the plume's effect to spread more quickly and suppress this region, whereas the upwind corners of the plots were assumed to be non-suppressed. The counts were somewhat less for days 4-6 (Figure 15) with no real counts found in the northwest corner, but counts were detected along the eastern margin.

#### **Experiments 2 and 3 - Pears**

When we attempted to define a more precise plume using more traps within a specific area, the general region remained suppressed and a somewhat defined plume structure was observed in terms of clear borders (Figure 16). Again the upwind portions were not suppressed, and the eastern border remained exposed. Moths were able to find the traps along the eastern portion at much higher rates. The area of influence of the plume appeared to run with the wind as expected with the farthest NE corner being consistently suppressed.

## Residual, "Ghost", Plume Effects

For the residual, or "ghost" period, the same general pattern was observed at 24 hours, but the plume appeared to be shortened (Figure 17). Similarly, along the road, high counts were observed fairly close to the puffer which suggests that pheromone dispersion may be influenced by the open nature of the road way. By day 2 (48 hours), the region of suppressed traps had shrunk considerably with one anomaly being the southeastern corner that did not catch moths as well. By Day 3, any residual effect disappeared and only the area immediately downwind of the puffers was suppressed, but high counts were observed elsewhere throughout the plots. For days 4-6, all patterns were completely lost including near the puffer device (Figure 18).

## Whole Orchard Suppression – Experiment 4 Pears

Given the difficulties that the wide roadway appeared to be presenting, we had hoped that an increase in puffers per orchard to 6 puffers would improve our ability to suppress traps. At least one to two puffers on the eastern border are now assumed to have contributed little to the suppression given the average wind conditions. This would also be true for the more northern puffer since the wind would be being blowing the pheromone from the orchard. So, we are currently assuming that only 3 puffers contributed significantly to the trap suppression because of the prevailing wind direction.

The unevenness observed in the early trials was not seen when the number of puffers was increased. The upwind areas along the southern border continued to catch moths, but this number decreased dramatically for traps placed within the expected regions influenced by the puffers (Figure 19). Traps were effectively suppressed for the 4-day period throughout most of the orchard.

The patchy trap suppression seen in the trials early in the season may have resulted from the more open nature of this orchard compared to plots used in 1999. However, both types of orchard are representative of orchard conditions found in California and any management programs will need to be robust enough to address this variation in orchard types. While the effects of single puffers appeared less dramatic in 2000, the use of several puffers again provided the strong trap suppression over large areas. However, as stressed throughout this report, our ability to suppress traps is NOT the same as damage suppression and these data will need to be confirmed in 2001 with suppression of fruit damage.

#### II. Attract and Kill - Pears

The results were disappointing for the effort in that moth recapture rates during the critical periods following the initial application were insufficient to resolve any treatment effects (average counts of less than <3 moths per trap). The reasons for the minimal trap recapture are not known. Despite considerable effort in establishing these plots, no reliable conclusions can be drawn about general efficacy of the Last Call program from these data.

## III. Results - Sprayable Formulations

## Pears -3M Sprayable

The average codling moth recaptures of the three plots are shown in Figures 20 and Figure 21 for traps baited with either the 1 or 10 mg lures. The season was again divided into 3 periods with each period lasting approximately one month. For the standard 1 mg lure, trap suppression for the 3M Sprayable formulation was excellent for the first month and was significantly reduced compared to the untreated Grower standard. While the counts were lower in the 3M treated plots, the results were not statistically significant from Isomate or grower plots early in the season. As the season progressed, the plots continued to have high trap suppression, but less than in the Isomate treated plots. A similar pattern was observed using traps baited with the 10 mg lures. Early in the season, the counts were roughly the same between the Isomate and 3M treated plots. However as the season progressed, counts were still suppressed until early August, but less than in the Isomate treated plots.

The data are also presented as flight curves for each plot with weekly codling moth counts. In the Dal Porto plots, moth recapture was very low until June 21 for reasons that are not known. However in Figure 22, the counts in the 1 mg lures indicated very strong trap suppression throughout June and into August when counts were compared to the non-pheromone treated grower controls. For the 10 mg baited traps (Figure 23), the counts were again suppressed, but less heavily as would be expected from these types of traps. The traps appeared to be similar to Isomate, but slightly higher counts were obtained in the 3M treated plots.

In the Hood plot, the 3M treated materials were heavily suppressed for ca. 40 days, but by late June, counts were starting to increase to levels similar to the control plots (Figures 24 and 25). However, additional suppression was still observed as the season progressed. As such, activity from the application was observed, but proved somewhat erratic.

### Walnuts

Trap suppression was excellent for the Consep sprayable at 20 gm ai / acre compared to either the untreated control plots or the Isomate treated plots (Figure 26). The season was divided into 3 periods of ca. 1 month and the results analyzed in a preliminary manner for the total counts for each plot. Using 1 mg baited traps, the differences were not significant but were consistently less in both the Isomate and sprayable plots. However, the high inter-plot variability made discrimination between treatments difficult. Using the 10 mg baited traps, the grower plots that did not receive any pheromone treatment showed significantly higher counts than either the Isomate or sprayable plots (Figure 27). The trends were the same between the 1 mg and 10 mg lures when figures 26 and 27 are compared. The duration of trap suppression exceeded expectations with ca. 81 days of suppression observed despite the constant release of sterile moths into each plot.

In contrast to the replicated plots for the Consep treatments, the plots treated with the 3M product spanned a range of application rates from 5 to 150 gm ai per acre. We had expected the results would be analyzed in a regressional format. From these data, we had expected to see a rate response in trap suppression ability or a threshold below which trap suppression was not achieved. The flight results for the 1 mg baited traps are shown in Figure 28, whereas the results for the 10 mg baited traps are shown in Figure 29. Both

the traps baited with 1 mg or 10 mg lures showed the same suppression by the sprayable formulations almost independent of rate. While the best results were obtained for plots treated with the 80 or 150 gm ai per acre, the relative trap suppression levels were roughly equal for all treatments. The plot treated with the 10 gm ai per acre did start to show increased counts later in the season, but the trend was not seen in the plot treated with 5 gm ai per acre. Again, the same positive results were obtained over the 81 days of the trial as observed with the Consep product.

#### **Conclusions**

The sprayable formulations provided trap suppression in excess of expectations with traps being suppressed at levels equal to trap suppression levels of acreage treated with hand-applied Isomate dispensers. Effective trap suppression appears to last up to 81 days. Equally promising was the lack of any clear rate response between trap suppression and application rate. The lack of a strong rate response suggests that program costs might be minimized due to the potential reductions in pheromone needed per acre to achieve adequate suppression. However, as discussed earlier, despite the 200-foot separation of the 2-acre plots, inter-plot effects were suggested. As such, trials in 2001 will need to increase the distance between the plots to confirm these results.

Equally positive for 2000 are the data from the puffers. The extremely long apparent plume suggested that a sharp decrease in the number of puffers might be possible. The effective suppression of 60-80 acres with only 1-5 puffers also suggests that the cost of the program could be decreased significantly in terms of number of dispensers, amount of pheromone per acre and in monitoring requirements of the actual dispensers. However, these data should NOT be construed as proof that damage will follow similar patterns. Instead these data suggest that trials in 2001 will need to examine fruit damage in relation to distance from the emitters. If confirmed with actual damage data, then we will need to consider making recommendations for placement of the aerosol emitters based on inter-puffer distances and expected plume lengths rather than number of puffers per acre. Obviously, the physical configuration of an orchard may ultimately decide the number of dispensers required per orchard.

Finally, the facts that upwind edges continue to be unsuppressed in puffer treated areas combined with our failure to discern continuous plumes in less dense pear orchards suggests that different puffer placement patterns will be required for different conditions. The most conservative position may be to define a program expected to meet the needs of even the most difficult of conditions. Use of puffers on an orchard wide basis will most likely require some type of supplemental edge treatments for the first few rows (e.g. insecticide or sprayable pheromones). At this time, it is important not to constrain our thinking to a program using single tactic approaches (e.g. hand-applied versus sprayables), but rather to focus on the most cost effective combinations that still meet our management goals.

Research in 2001 will need to take the next hard step that allows crops to be placed at some measure of risk to damage from codling moth. Whereas these data are very encouraging, our optimism must be tempered by the clear recognition that we have yet to demonstrate effective field suppression of codling moth damage. We need to better understand how the moths are responding to these pheromone plumes, if they are altering their distribution, and how these changes may influence program efficacy. By the end of 2001, we hope to be able to determine if this trap suppression will be matched by equally impressive damage suppression.

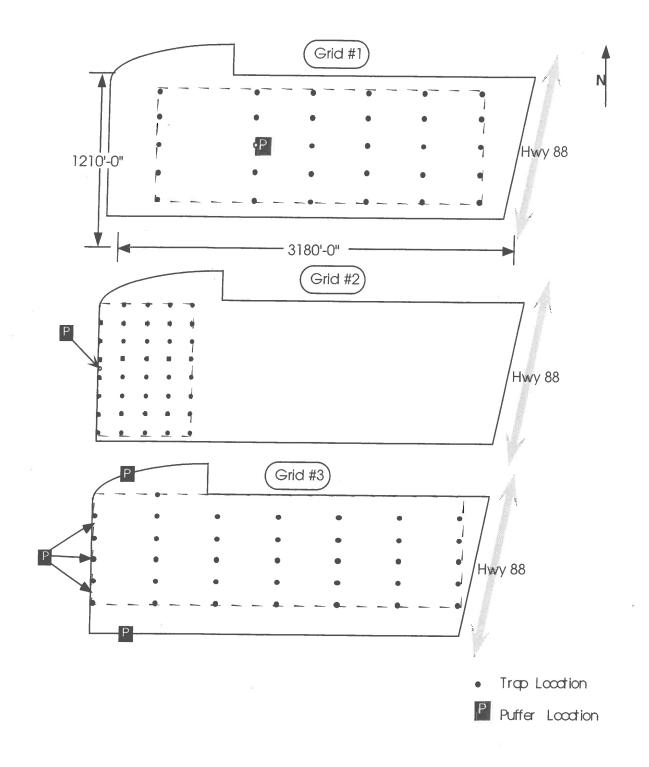


Figure 1. The layouts for the three experimental trapping grids within the walnut orchard.

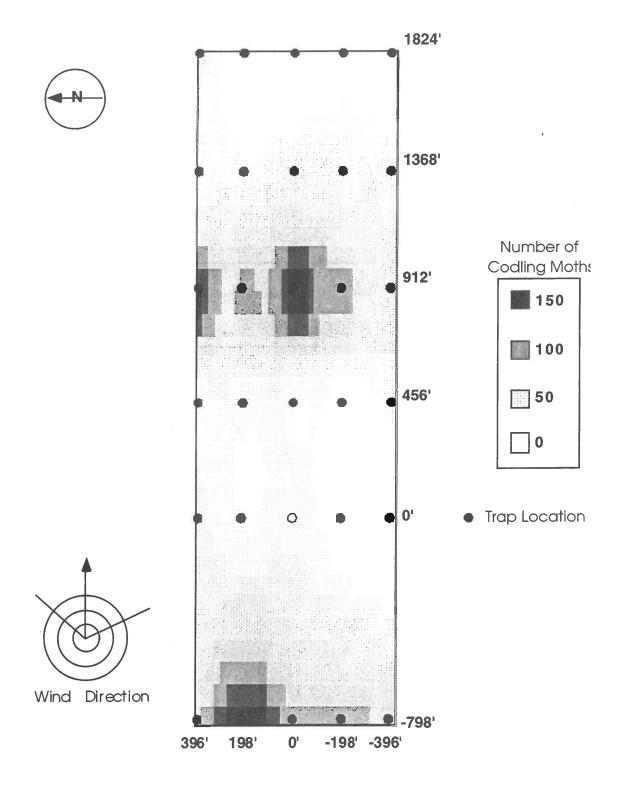


Figure 2. Experiment 1b. Pre-treatment codling moth recapture patterns. Trap catch totals for days 1-7.

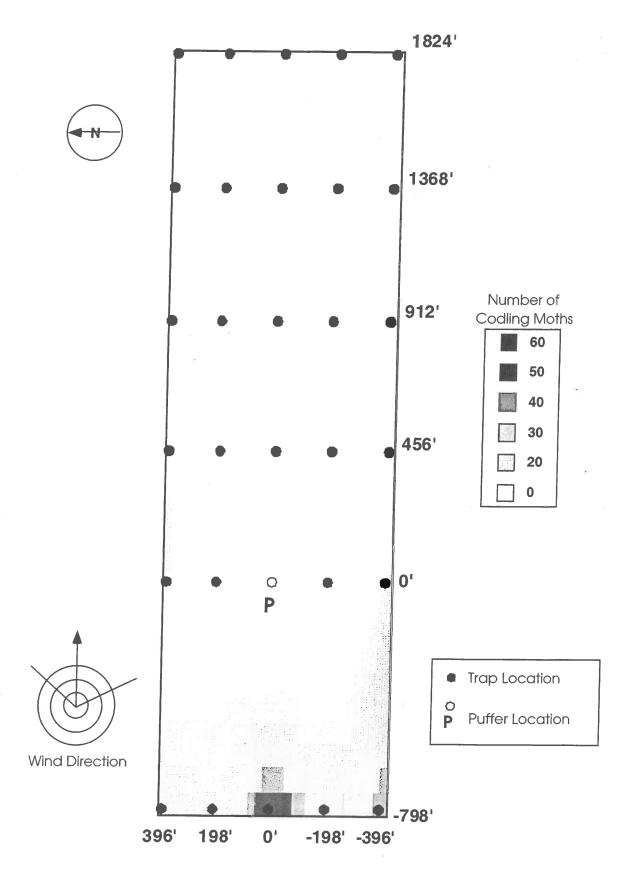


Figure 3. Experiment 2. Pheromone plume structure in walnuts - 24-hour puffer cycles. Cumulative trap catch totals for days 1-8.

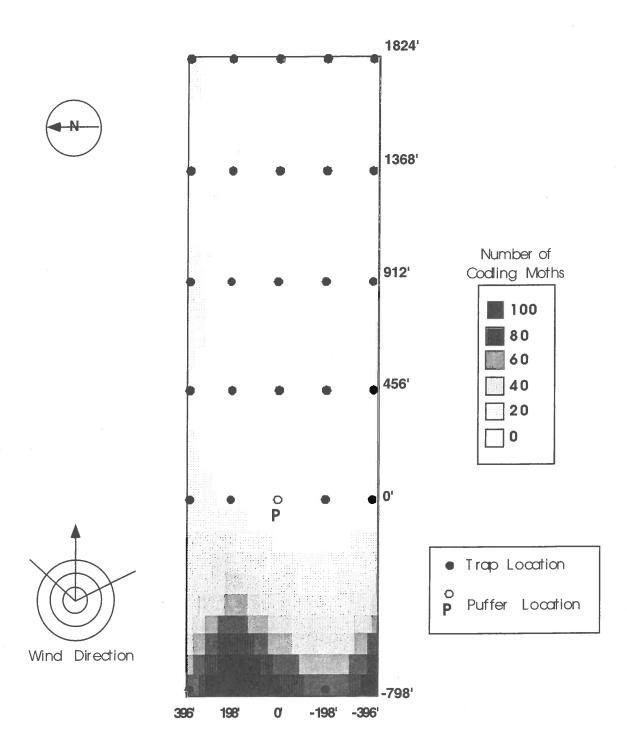


Figure 4. Experiment 3a, "on" period with single puffer device hung within the walnut orchard, 12-hour emission cycle. Five-day cumulative trap catch totals.

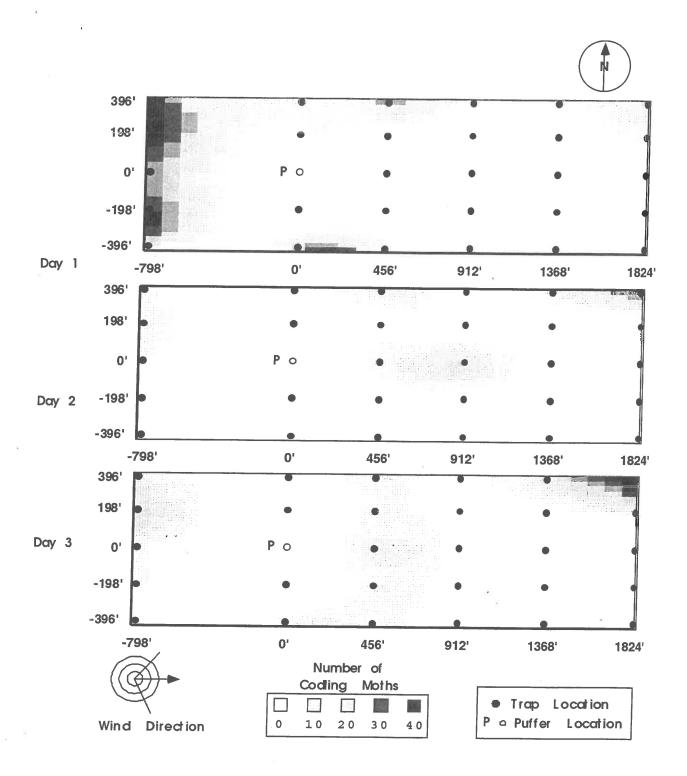


Figure 5. Experiment 3a, "off" period. Single puffer device hung within the walnut orchard, no emission cycle. Three-day comparison of trap recapture under "ghost" plume exposure.



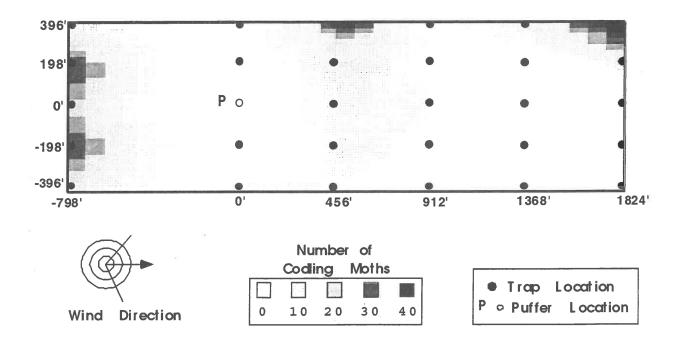


Figure 6. Experiment 3a, "off" period. Single puffer device hung within the walnut orchard, no pheromone emission. Cumulative trap catch totals for days 4 through 6 after last puffer emission cycle.

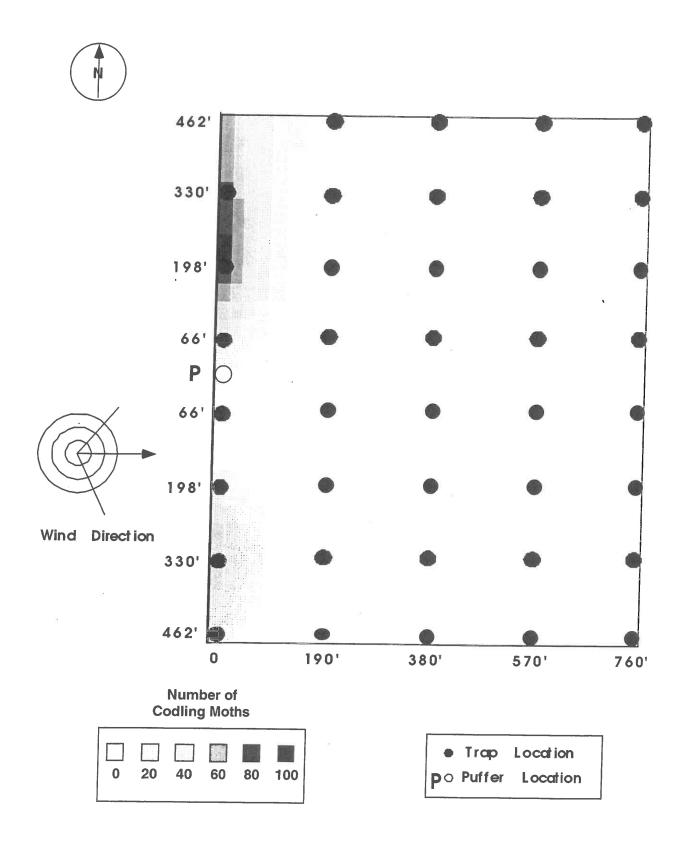


Figure 7. Experiment 4a, "on" period, single puffer device hung on west edge of the walnut orchard. 24-hour pheromone emission cycle. Cumulative 7-day trap catch totals.

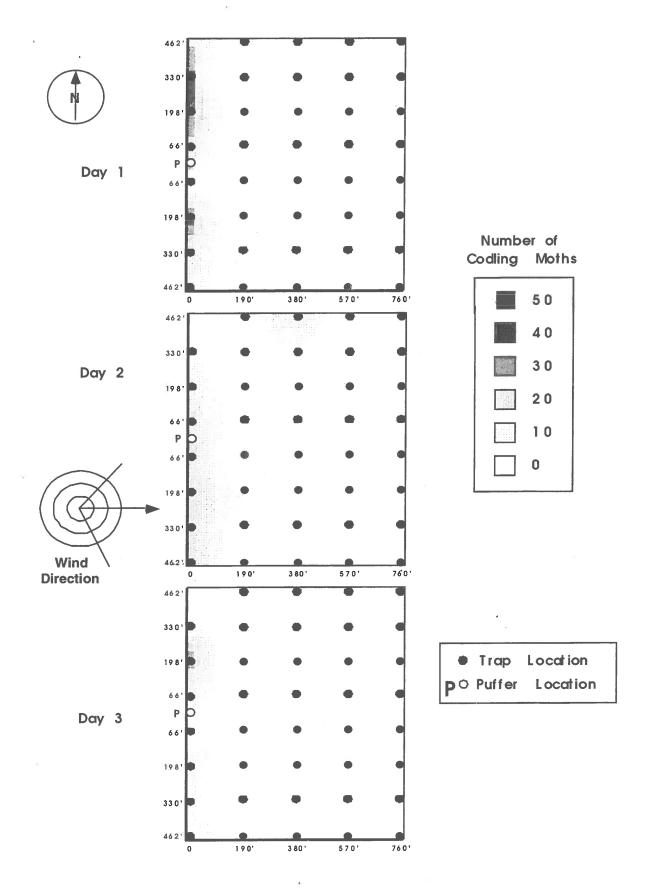


Figure 8. Experiment 4a, "off" period, single puffer hung on west edge of the walnut orchard. No pheromone emission. Three-day comparison of trap catches under the "ghost" pheromone plume.

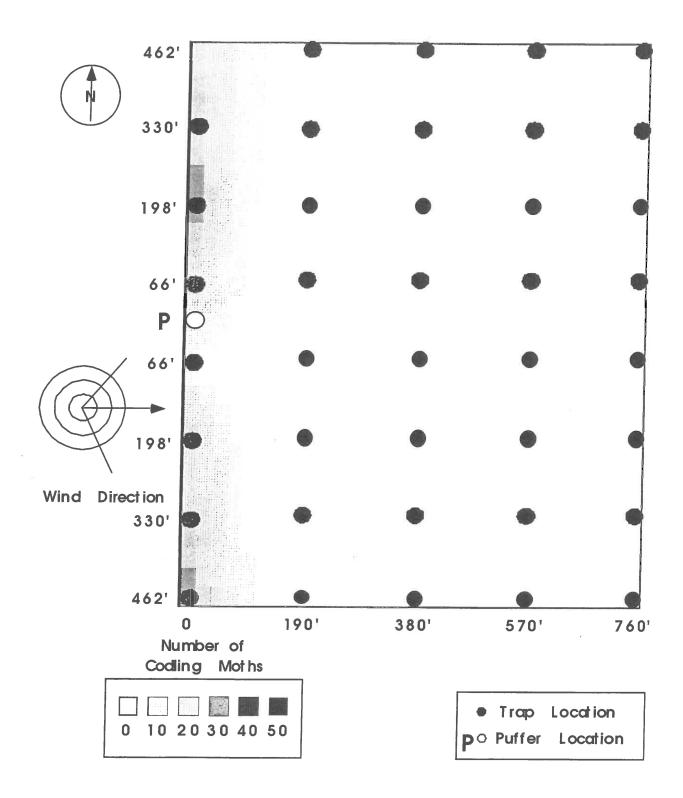


Figure 9. Experiment 4a, "off" period, single puffer hung on the west edge of the walnut orchard. No pheromone emission. Day 6 after last puffer emission cycle, 3-day cumulative trap catch totals.

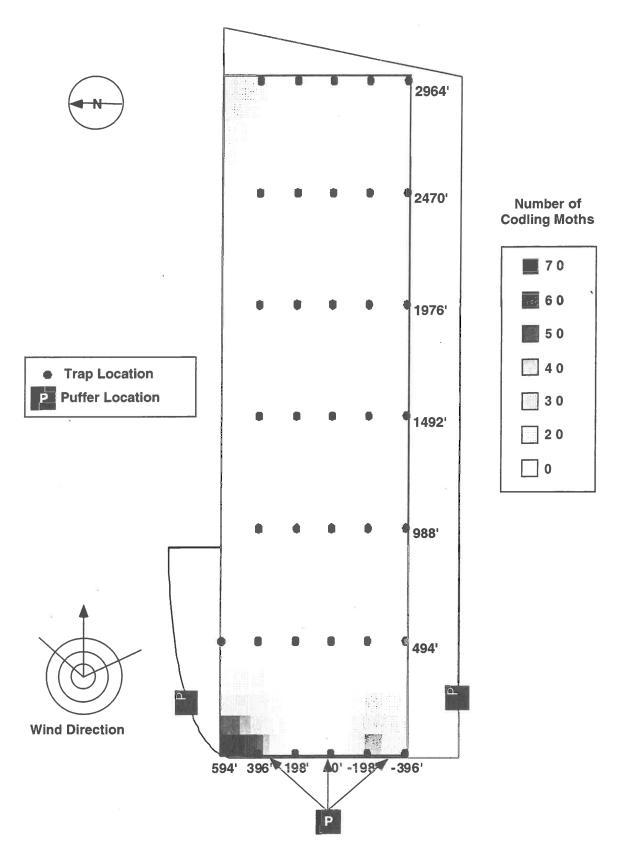


Figure 10. Experiment 5, "on" period, 5 puffer devices hung on the walnut orchard edges. 24-hour pheromone emission cycle. Seven-day cumulative trap catch totals.

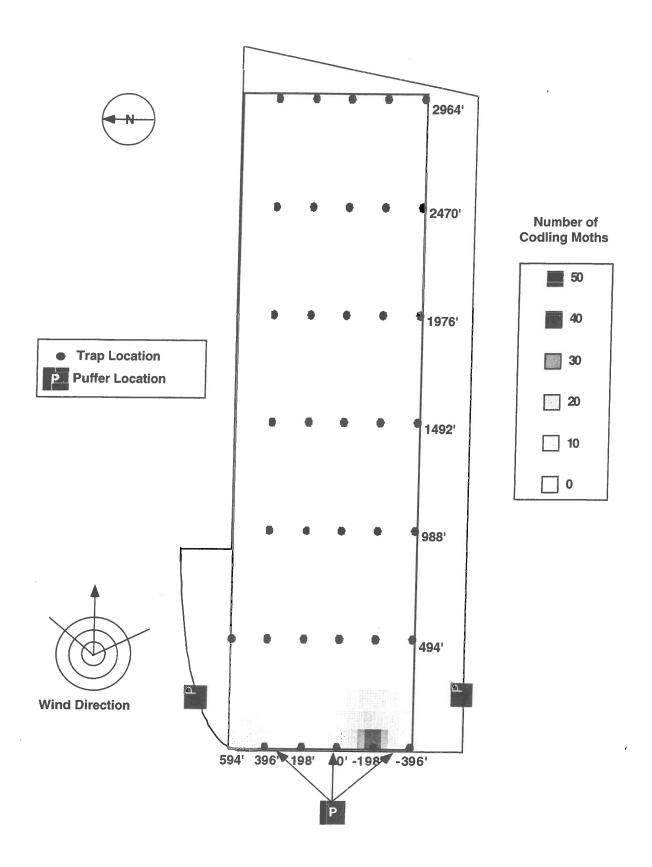


Figure 11. Experiment 5, "off" period, 5 puffer devices hung on the walnut orchard edges. No pheromone emission cycle. Cumulative trap catch totals for days 1-3 after the last previous puffer emission cycle.

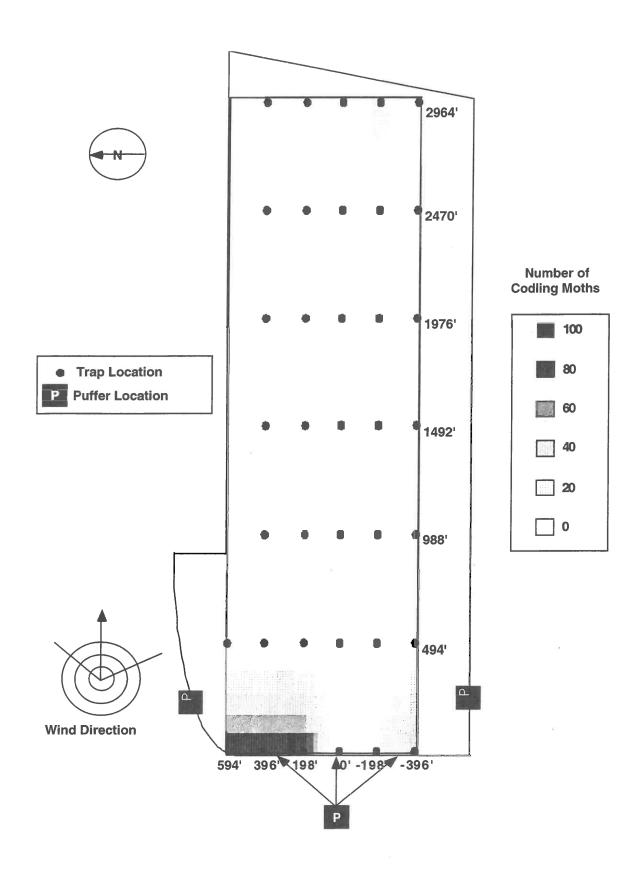


Figure 12. Experiment 5, "off" period, 5 puffer devices hung on the walnut orchard edges. No pheromone emission cycle. Cumulative trap catch totals for days 4-6 after the last previous puffer emission cycle.

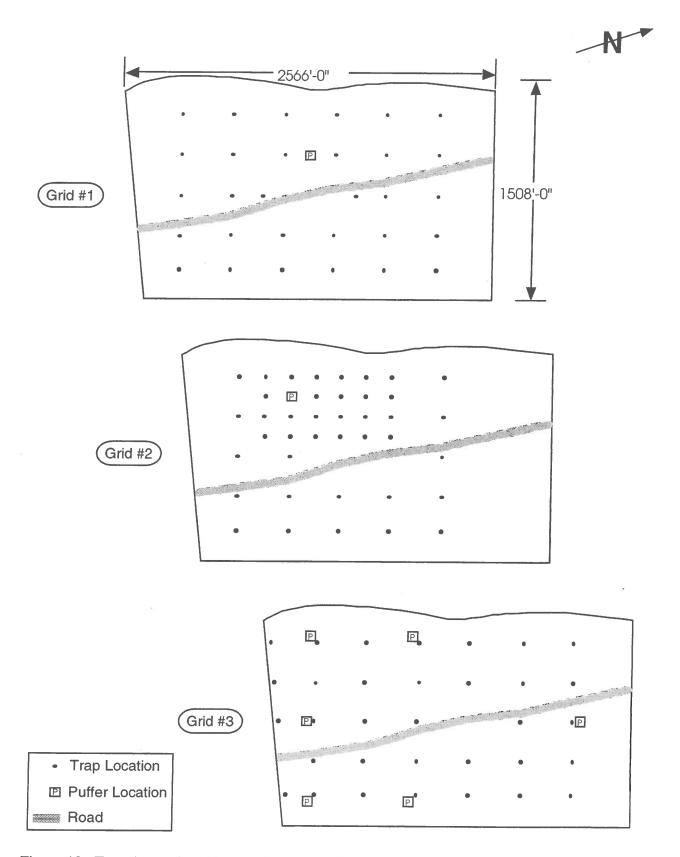


Figure 13. Experimental grid layouts for pear puffer trials.

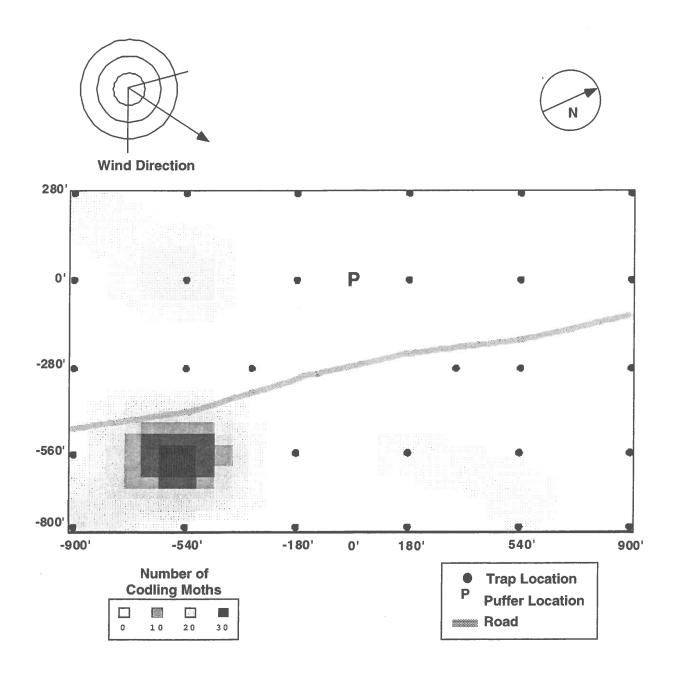


Figure 14. Experiment 1a, day 3 - three day cumulative trap catch

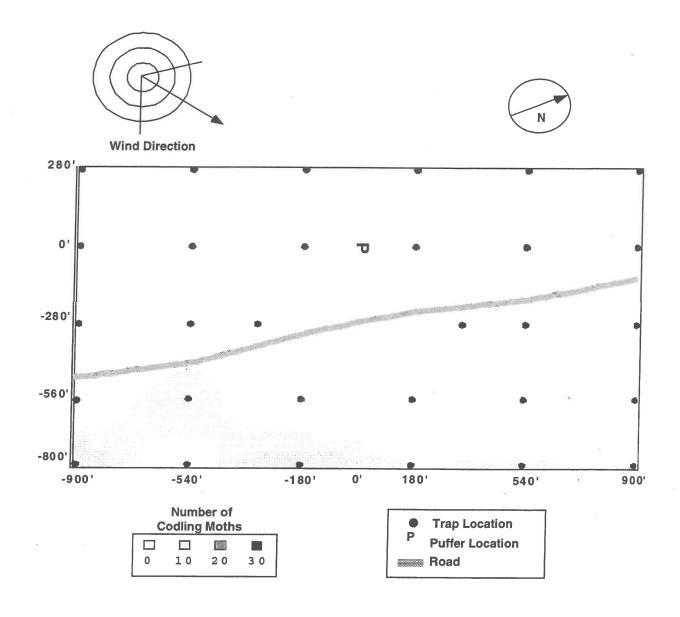


Figure 15. Experiment 1a, Day 6, three day cumulative trap catch totals

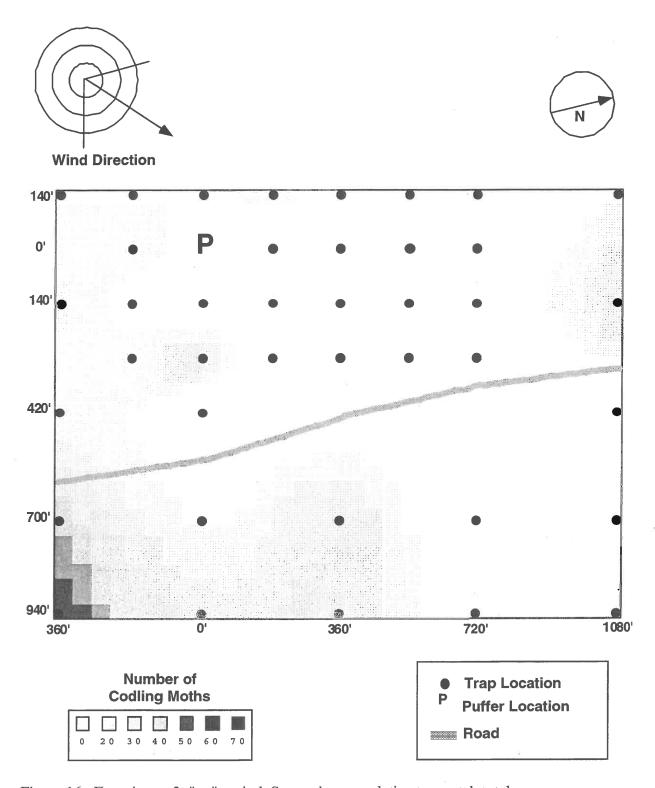


Figure 16. Experiment 2; "on" period, Seven-day cumulative trap catch totals

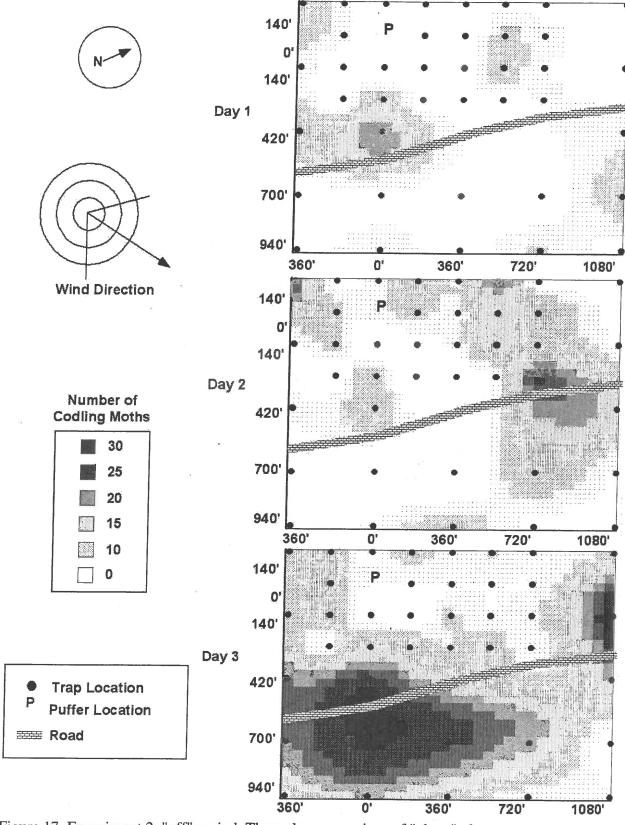


Figure 17. Experiment 2, "off" period, Three-day comparison of "ghost" plume

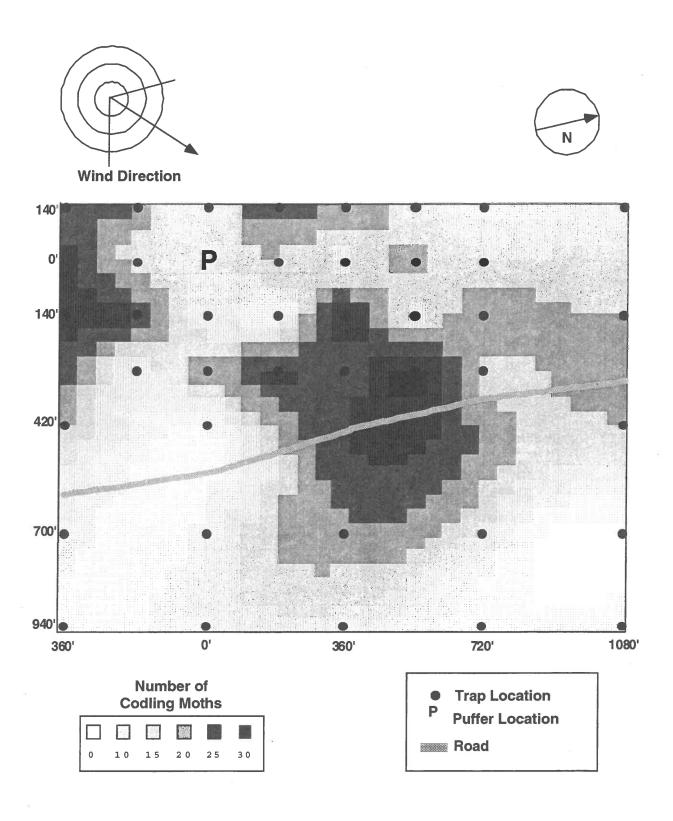


Figure 18. Experiment 2, "off" period. Day 6 after last emission cycle, three-day cumulative trap catch.

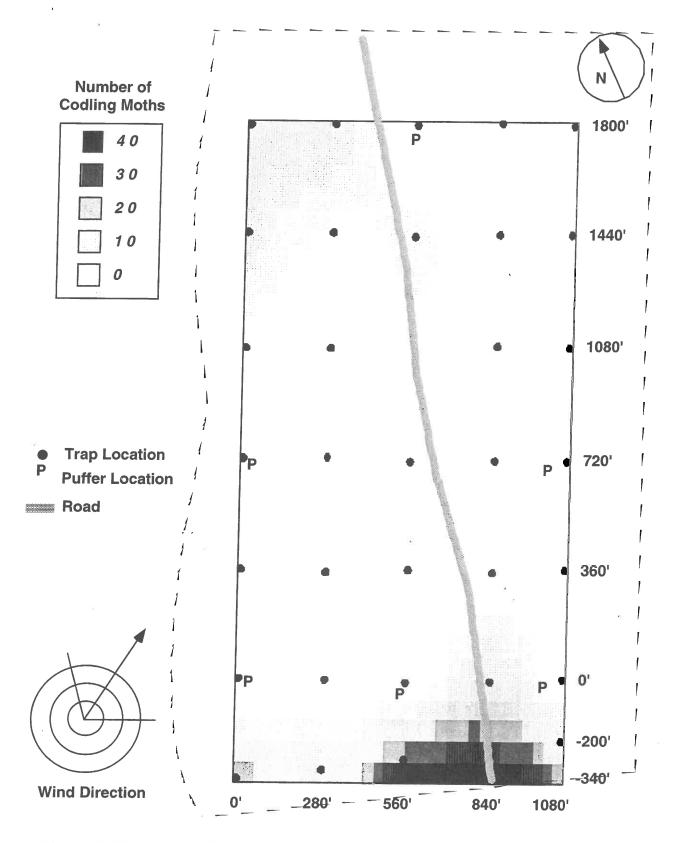


Figure 19. Experiment 4, Five puffer "shut down", multiple puffers on edges of the orchard. day 4, cumulative trap catch totals.

## PEARS: 3M Sprayable - 1 mg Lures

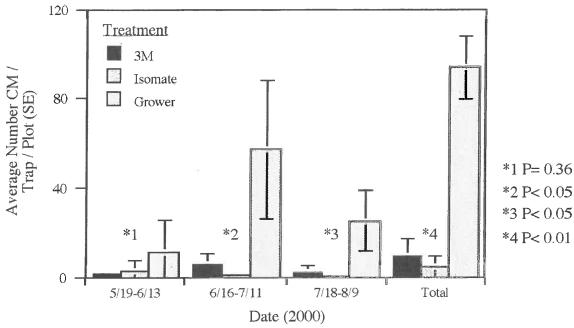


Figure 20. Average moth counts for 3 1-month periods from traps baited with 1 mg lures in pear plots treated with sprayable formulation, 3M.

## PEARS: 3M Sprayable - CM 10X Lures

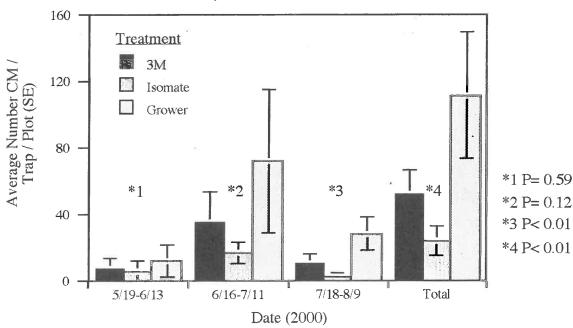


Figure 21. Average moth counts for 3 1-month periods from traps baited with 10 mg lures in pear plots treated with sprayable formulation, 3M

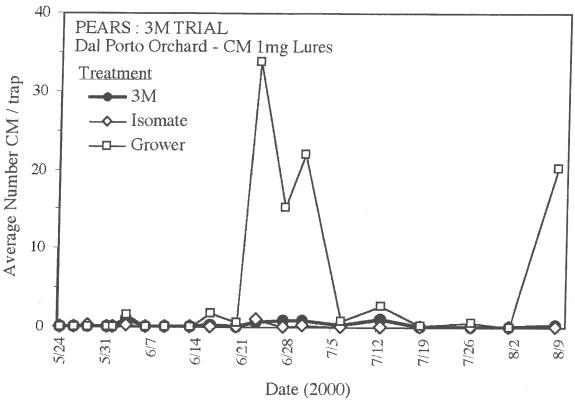


Figure 22. Moth flights from traps baited with 1 mg lures in pear Dal Porto plots treated with sprayable formulation, 3M.

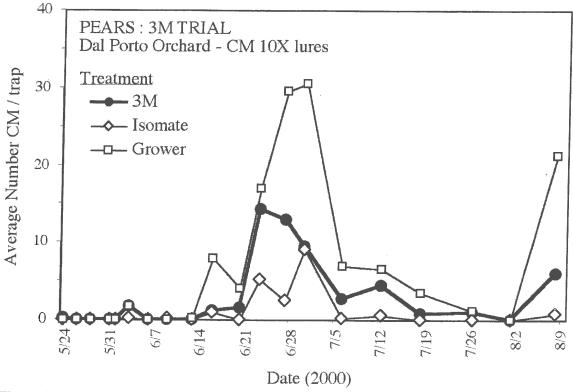


Figure 23. Moth flights from traps baited with 10 mg lures in pear Dal Porto plots treated with sprayable formulation, 3M.

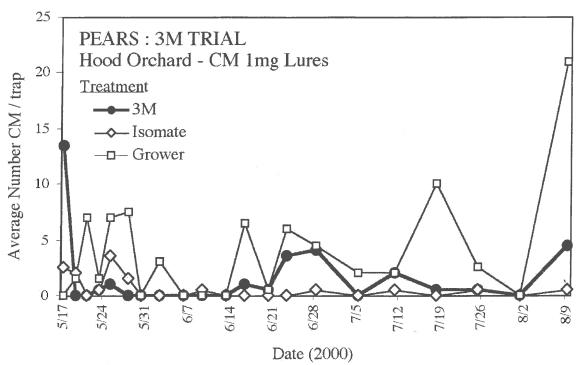


Figure 24. Moth flights from traps baited with 1 mg lures in pear Hood plots treated with sprayable formulation, 3M

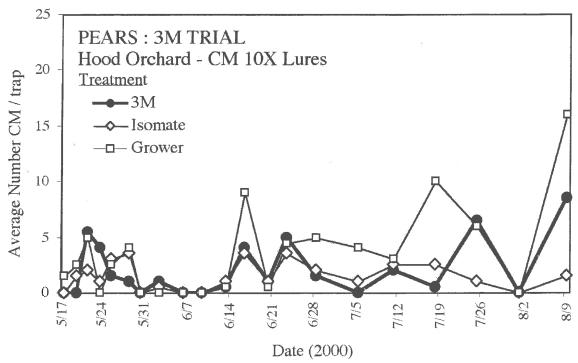


Figure 25. Moth flights from traps baited with 10 mg lures in pear Hood plots treated with sprayable formulation, 3M

## WALNUTS: Consep Sprayable - 1 mg Traps

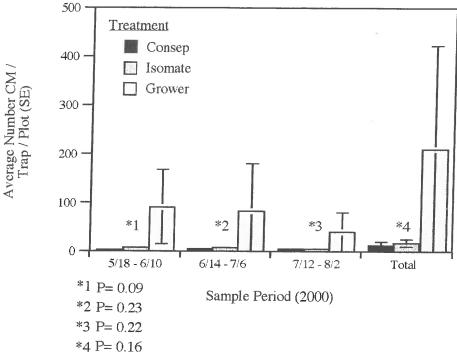


Figure 26. Moth flights from traps baited with 1 mg lures in Dondero walnut plots treated with sprayable formulation, Consep.

## WALNUTS: Consep Sprayable - 10 mg Traps

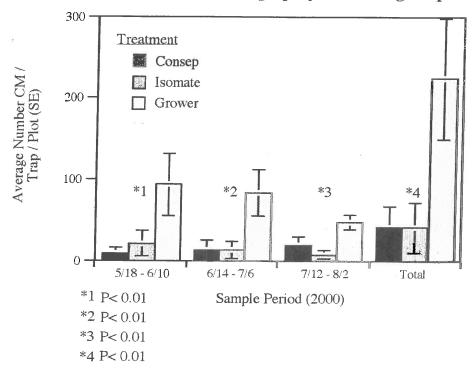


Figure 27. Moth flights from traps baited with 10 mg lures in Dondero walnut plots treated with sprayable formulation, Consep.

# **WALNUTS: 3M Sprays Dondero Orchard - 1mg Traps**

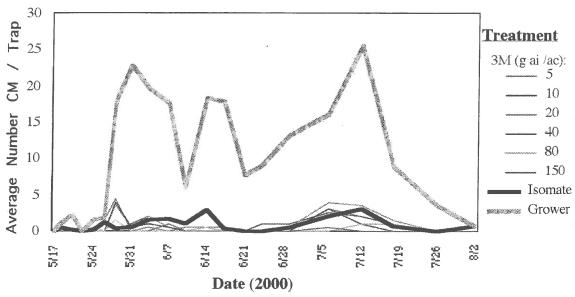


Figure 28. Moth flights from traps baited with 1 mg lures in Dondero walnut plots treated with sprayable formulation, 3M.

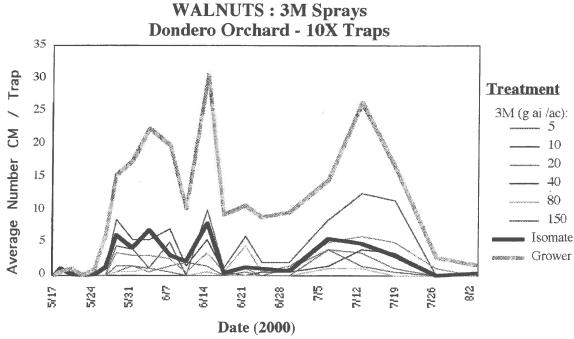


Figure 29. Moth flights from traps baited with 10 mg lures in Dondero walnut plots treated with sprayable formulation, 3M.

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