DRAFT PROTOCOL
PILOT PROJECT FOR METHYL EUGENOL/NALED AMBIENT AIR MONITORING

I. INTRODUCTION

The California Department of Food and Agriculture Division of Plant Industry uses methyl eugenol and naled as a fruit fly attractant and pesticide, respectively, during trapping and eradication programs. Methyl eugenol has been chosen for further study by the federal government because of its similarity to other oncogenic chemicals. Naled is a Category I material. In response to the expressed perception that many people can be exposed to methyl eugenol/naled mixtures during eradication programs, CDFA has decided to conduct a pilot study to determine the feasibility of monitoring ambient air levels of methyl eugenol and naled (including DDVP, a naled degradation product) during eradication programs.

Questions have been raised about fruit from trees in which methyl eugenol/naled traps have been placed. Traps may be positioned in one tree for up to 13 weeks. Fruit from these trees may be affected by the compounds used in the traps. Several varieties will be analyzed for total residues of both compounds.

II. OBJECTIVE

Our objective is to determine the feasibility of monitoring ambient air levels of methyl eugenol and naled (Dibrom®)/DDVP during eradication programs. The results of the pilot study will be used to design and implement a monitoring program for future eradication projects.

Additionally, fruits from trees which contain fruit fly traps will be tested for residues of the above products.

III. PERSONNEL

The pilot study and ambient air monitoring will be conducted by the Calfornia Department of Food and Agriculture's (CDFA) Environmental Hazard Assessment Program (EHAP). Key EHAP personnel are listed below:

Bonnie Turner - Project leader

Sally Powell - Experimental design, statistical analysis

Karen Wiese - Field group coordinator

Nancy Miller - Laboratory liaison/quality control

Duc Tran - Chemistry lab coordinator

IV. EXPERIMENTAL DESIGN

Ambient Air Monitoring

The study will be performed at the Folsom C&E facility. In order to simulate ambient air concentrations that might be found in an eradication area, 19 bait stations will be set up in two concentric circles with high volume air samplers at the center of the circles (Figure 2). These air samplers will be set up within two meters of the center station, at least one meter apart from each other and downwind of the center station. Another set of samplers will be placed 25 meters from the center station (downwind, if feasible) and will be used to collect samples on the same schedule as the central set. The distance between bait stations will be the same as it will be in the eradication area, approximately 45-50 meters. Each bait station will be constructed by nailing a one-foot square of plywood (the target) to the end of a 8-foot two-by-four, which will be supported vertically. The lure and pesticide will be sprayed onto the plywood square from a truck in the same manner as it is applied during an eradication.

plus background

Since the time required to trap detectable amounts of methyl eugenol is unknown, different air samplers will take 4-hour, 8-hour and 24-hour samples. A total of 17 samples for each chemical at each distance will be collected. Table 1 gives the sampling intervals for each sampler. Because the concentrations are expected to be very low, sampling will be done most intensively during the eight hours following application. However, since it is also unknown (and depends in part on the weather) how long detectable concentrations may remain in the area,

samples will be taken one, two and seven days after application. Bait will be applied to the stations from a truck in the same manner performed during an eradication project. Sampling will begin when the application begins (this will define Hour 0). Air temperature and wind speed at the site will be recorded during each sampling interval. Samples collected on Day 7 will not be analyzed unless samples collected on Day 1 or 2 are positive.

The results of the pilot study will be used to determine the experimental design used during monitoring of future eradication programs which use methyl eugenol and naled.

Fruit

Fruits will be sampled from four trees used as actual trapping stations. Up to four varieties of fruit will be tested. The trees selected must have had a trap in place for at least four weeks prior to sampling. On each tree, fruit will be sampled at two distances from the trap: approximately 1 meter and at 3-5 meters (depending on the size of the tree). One composite sample will be taken at each distance. Enough fruit will be picked at each distance to make a composite sample approximately one pound in wet weight. The individual fruits making up one composite sample will be picked, as nearly as possible, from the surface of a sphere 1 m (or 3-5 m) in diameter whose center is the trap. The sampling scheme for a tree is diagrammed in Figure 1. A total of eight samples will be collected (four trees X two distances). Samples will be split into two aliquots for separate methyl eugenol and naled/DDVP analysis. Background fruit samples will be collected from adjacent trees (same fruit variety) if they are available.

V. SAMPLING METHODS

All sampling will be performed using standard EHAP procedures (Sava, 1986). Replicate air samples will be collected using high volume and possibly low volume impinger air samplers calibrated at appropriate flowrates. XAD-2 resin and ethylene glycol will serve as the trapping medium. Background and post application sampling periods will be a minimum of 4 hours.

Fruit from trees containing traps will be collected using fruitpicking equipment and stored in 1-liter jars. Air and fruit samples will be frozen at -70°F until delivered to the laboratory for analysis. Chain of custody records will be kept to document sample handling from generation to analysis.

VI. STATISTICAL ANALYSIS

The data collected from the pilot project will be presented in graphical form showing mean and standard deviations for ambient levels of both compounds. Fruit residue values will be presented in tabular form.

VII. ANALYTICAL METHODS/QUALITY CONTROL

Analytical method development and sample analysis will be performed by the CDFA laboratory. Fruit will be analyzed for total residues only. Air and fruit samples will be analyzed for methyl eugenol, naled, and DDVP (degradation product of naled). One solvent blank, 1 matrix spike sample and 2 replicate injections for 1 positive sample will be analyzed with each extraction set for each matrix.

VIII. BUDGET

Personnel Expenses:	$4000
Operating Expenses:	11000
Total Expenses:	$15000

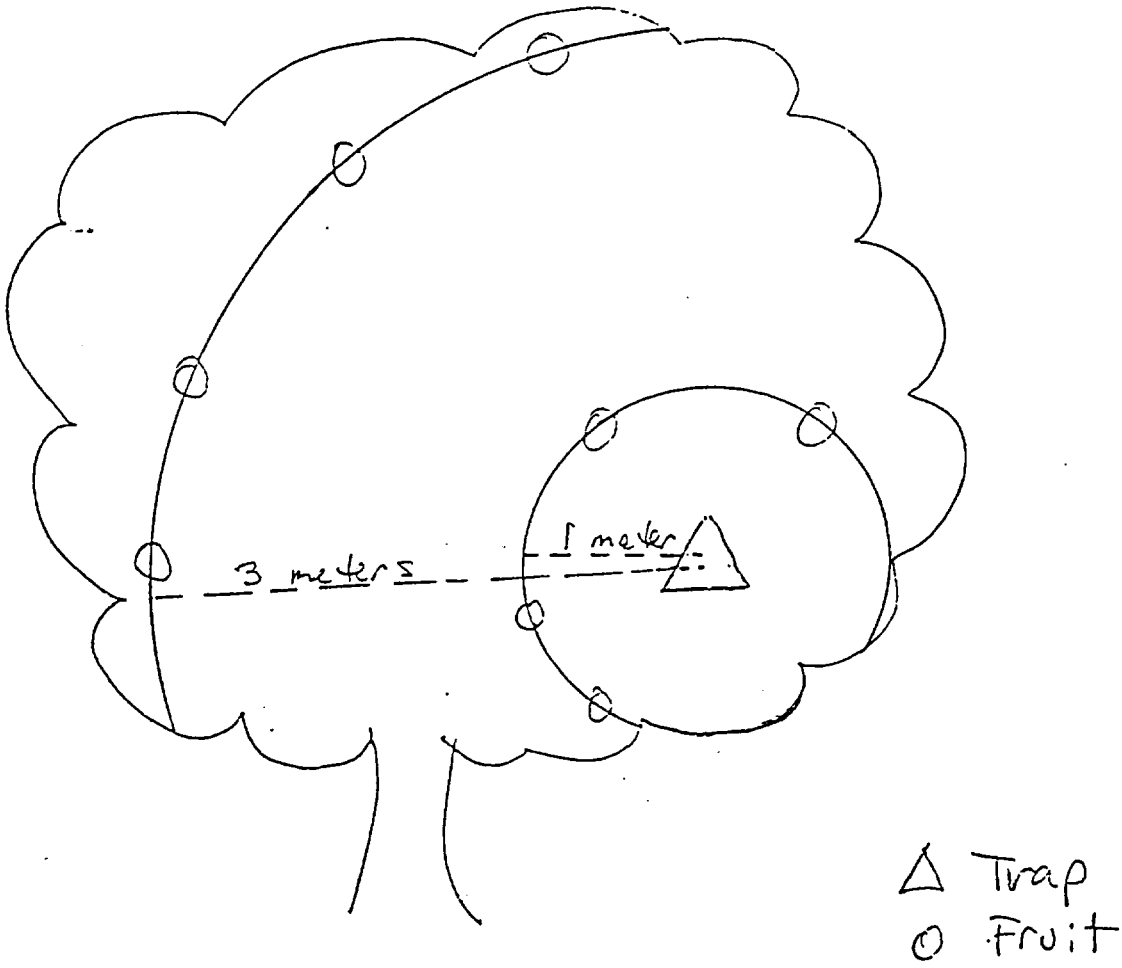
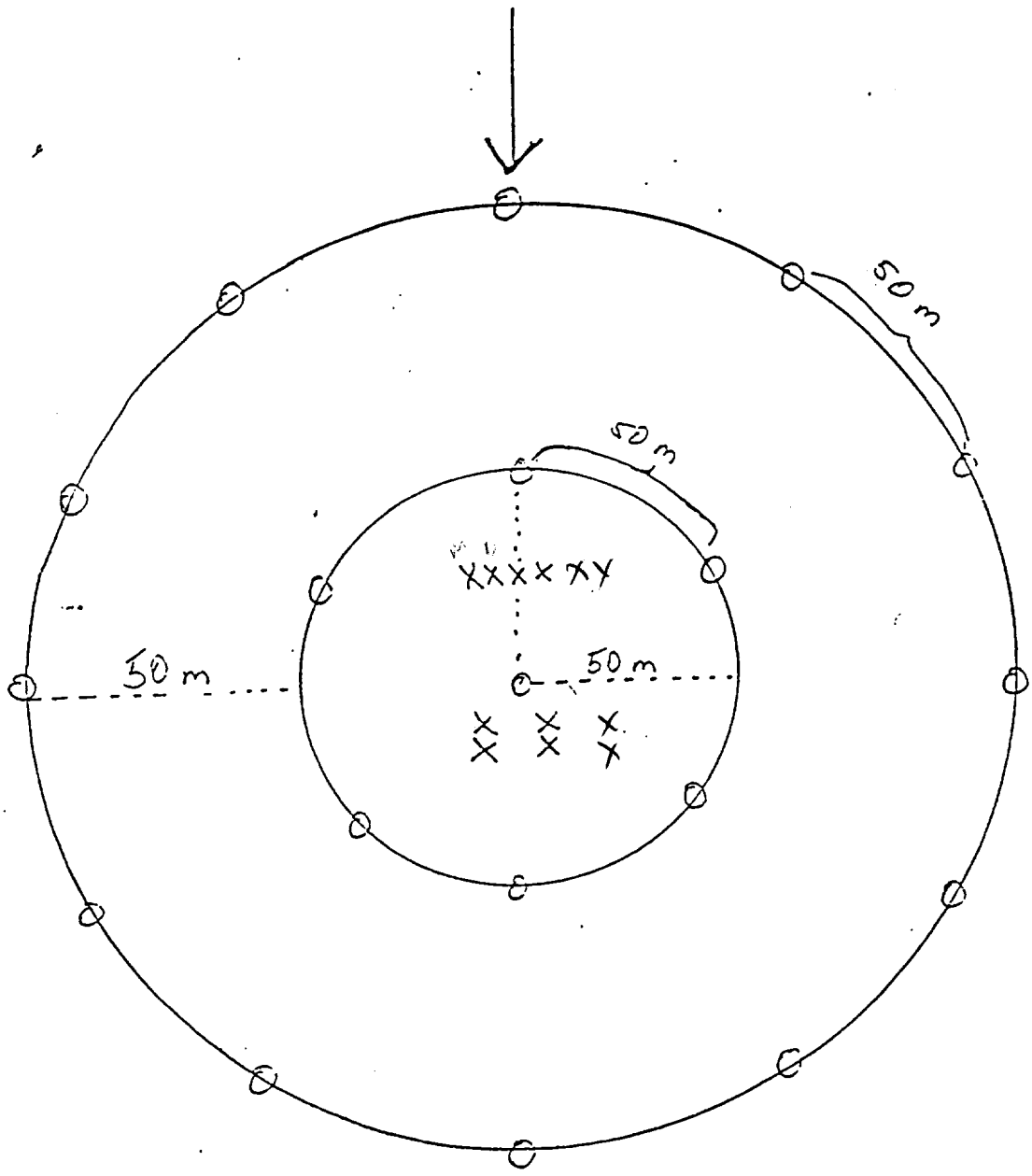


Figure 1. Cross-sectional diagram of sampling scheme for one fruit tree.



○ Bait station
× Air sampler
↓ Direction of prevailing wind

Figure 2. Top view of experimental set-up for measuring air concentrations.

Fruit

DRAFT PROTOCOL

METHYL EUGENOL/NALED MONITORING AT FRUIT FLY TRAPPING LOCATIONS

March 31, 1989

I. INTRODUCTION

In 1988 the California Department of Food and Agriculture conducted a monitoring program which included collecting fruit samples from trees which contained fruit fly traps. Eight samples were analyzed for methyl eugenol, naled and DDVP residues, chemicals used as bait for the traps. One fruit sample contained both methyl eugenol and DDVP residues (confirmed by two analytical methods); therefore, CDFA undertook additional monitoring in the winter of 1988. Results from samples collected were negative but due to seasonal temperature differences between the first and second monitoring periods, it was proposed that additional monitoring be undertaken during the late spring and summer of 1989 to determine whether or not fruit absorbs these chemicals.

II. OBJECTIVE

Our objective is to determine if residues can be found in fruits collected from trees containing fruit fly traps. If analysis confirms the presence of methyl eugenol, naled or DDVP, further sampling for dissipation rate determination may be conducted.

III. PERSONNEL

Fruit sampling will be conducted by the Environmental Hazard Assessment Program field group. Key personnel are:

Bonnie Turner - Project Leader

Sally Powell - Experimental design and statistical analysis

Karen Wiese - Field group coordinator

Nancy Miller - Laboratory liaison/quality control

Duc Tran - Chemical analysis

Public/Agency Contact - Madeline Ames, (916) 324-8916

IV. MONITORING DESIGN/STATISTICAL ANALYSIS

Fruit samples will be collected from five fruit tree species commonly found in the Sacramento area. Fourteen replicates for each species will be collected from separate trees in which baited traps have been placed for a minimum of six weeks. Initial samples (0.50 kg wet weight) will be collected four hours after traps have been re-baited. An additional sample from each tree will be collected 24 hours after re-baiting. All fruit will be collected no less than 31 cm or greater than 61 cm from the trap unless sufficient fruit is not available within this area. In the case of citrus fruit, an additional sample will be collected so that separate analysis of outer skin and inner fruit is possible. Ambient temperature (°C) will be recorded at the time of rebaiting and sample collection.

It will be necessary to collect a large number of samples of each type of fruit in order to establish with statistical certainty whether residues exist. Therefore, 14 samples will be collected and if all are found to contain no detectable residue, it can be concluded with 95% statistical confidence that the percentage of all possible samples containing residue is less than 20%. (To conclude with the same degree of

confidence that the percentage is less than 10% would require 25 samples, all found to be negative.) Sampling will be conducted as follows:

Species	Sampling Period	Replicates
Apricot	May-August	14
Peach	May-August	14
Apple	July-September	14
Fig	June, August-November	14
Citrus	All Year	14+14
Total (84 samples x 2 days x 2 analyses):		336

V. SAMPLING METHODS

Fruit will be collected from trees using fruitpickers, placed in 2-liter glass jars, chilled on ice and delivered to the laboratory for immediate extraction. The fruitpickers and trapsetting poles will be cleaned with alcohol and dionized water between samples.

VI. ANALYTICAL METHODS/QUALITY CONTROL

Analysis will be performed by the CDFA laboratory using methods developed from earlier monitoring programs. Separate analyses for methyl eugenol and naled/DDVP are required. Confirmation of positive finds will be made using GC/MS. One solvent blank, 1 matrix spike and 2 replicate injections for 1 positive sample will be analyzed with each extraction set.

VII. TIMETABLE

Sampling Period	May - October, 1989
Extraction/Chemical Analysis	May - November, 1989
Data Analysis/Report	November-December, 1989

VIII. BUDGET

Personnel Expenses:	$5,000
Operating Expenses:	51,000
Total Cost:	$56,000

DRAFT

PROTOCOL FOR PILOT PROJECT AND ENVIRONMENTAL MONITORING FOR METHYL EUGENOL/DIBROM

I. OBJECTIVE

To determine the feasibility of monitoring ambient air levels of methyl eugenol (a fruit fly attractant) and naled (Dibrom®), a pesticide used for several eradication projects. In addition, fruits from trees which contain fruit fly traps will be tested for residues of the above products. The results of the pilot study will be used to design and implement a monitoring program for future eradication projects.

II. PERSONNEL

The pilot study and ambient air monitoring will be conducted by the Calfornia Department of Food and Agriculture's (CDFA) Environmental Hazards Assessment Program (EHAP). Key EHAP personnel are listed below:

Bonnie Turner - Project leader
Sally Powell - Experimental design, statistical analysis
Karen Wiese - Field group coordinator
Nancy Miller - Laboratory liaison/quality control
Duc Tran - Chemistry lab coordinator

III. EXPERIMENTAL DESIGN

Fruit

Fruits will be sampled from four trees used as actual trapping stations. Up to four varieties of fruit will be tested. The trees selected must have had a trap in place for at least four weeks prior to sampling. On each tree, fruit will be sampled at two distances from the trap: approximately 1 meter and at 3-5 meters (depending on the size of the tree). One composite sample will be taken at each distance. Enough fruit will be picked at each distance to make a composite sample approximately one pound in wet weight. The individual fruits making up one composite sample will be picked, as nearly as possible, from the surface of a sphere 1 m (or 3-5 m) in diameter whose center is the trap. The sampling scheme for a tree is diagrammed in

Figure 1. A total of eight samples will be collected (four trees X two distances). Background fruit samples will be collected from adjacent trees (same fruit variety) if they are available.

Pilot Study for Ambient Air Monitoring

In order to simulate ambient air concentrations that might be found in an eradication area, 19 bait stations will be set up in two concentric circles with the air samplers at the center of the circles (Figure 2). The distance between stations will be the same as it will be in the eradication area, approximately 45-50 meters. Two air samplers will be set up within two meters of the center station, at least one meter apart from each other. If it is possible to determine the direction of the prevailing winds, the samplers will be placed downwind of the center station. The experiment will take place in an open field (Folsom field site). Each bait station will be constructed by nailing a one-foot square of plywood (the target) to the end of a 8-foot two-by-four, which will be supported vertically. The lure and pesticide will be sprayed onto the plywood square from a truck in the same manner as it is applied during an eradication.

Since the time required to trap detectable amounts of methyl eugenol is unknown, one air sampler will take one-hour and the other will take two-hour samples. A total of 22 samples will be collected. Table 1 gives the sampling intervals for each sampler. Because the concentrations are expected to be very low, sampling will be done most intensively during the eight hours following application. However, since it is also unknown (and depends in part on the weather) how long detectable concentrations may remain in the area, samples will be taken one, two and seven days after application. On these days, sampling will be done in the morning (the stillest part of the day) and afternoon (the hottest part of the day), when air concentrations are likely to be highest. Bait will be applied to the stations from a truck in the same way it will be done during an eradication project. Sampling will begin when the application begins (this will define Hour 0). Air temperature and wind speed at the site will be recorded at the beginning of each sampling interval.

It would be desirable also to measure air concentration at a point equidistant between bait stations. If resources permit, a second pair of samplers will be placed 25 meters downwind from the center station and will be used to collect samples on the same schedule as the first pair.

Monitoring of Eradication Project

Monitoring will begin whenever EHAP is notified of an eradication program starting up within the State of California after the pilot program has been completed. Ambient air samples in the vicinity of the eradication bait stations will be collected over the eradication program or until proof of non-detectable levels of pesticides occurs at the treatment sites. If several eradication programs are underway in the state at the same time, only one will be monitored.

Six bait stations will be randomly selected from the eradication area. Two air samplers, one for sampling methyl eugenol and one for sampling Dibrom, will be set up near each selected station. Air samples will be collected at three times following each application of bait, tentatively at 0, 1 and 7 days after the application. It is anticipated that four or five applications will be made. The total number of samples of each compound will be 6 stations X 4 applications X 3 days = 72 samples (90 if 5 applications are monitored).

The results of the pilot study will be used to determine whether six stations is an adequate sample size or whether more will be needed. The pilot results will also be used to decide the distance of the samplers from the stations, the time of day for sampling, the duration of the sampling interval, and how many days post application to continue sampling. Since it expected that it will be very difficult to detect the methyl eugenol, these choices will be made to maximize the possibility of detection.

IV. SAMPLING METHODS

All sampling will be performed using standard EHAP procedures (Sava, 1986). Replicate air samples will be collected using high volume and

possibly low volume impinger air samplers calibrated at appropriate flowrates. XAD-2 resin and ethylene glycol will serve as the trapping medium. All air background and post application sampling periods will be a minimum of 1 hour. The application sampling period will start when the naled/methyl eugenol mixture is applied and continue for 60 minutes. Available food crops (primarily fruits) from local areas near hanging traps will be collected and stored in 1-liter jars. Air and fruit samples will be frozen at -70°F until delivered to the laboratory for analysis. Chain of custody records will be kept to document sample handling from generation to analysis.

V. STATISTICAL ANALYSIS

The data collected from the pilot project will be presented in graphical form showing mean and standard deviations for ambient levels of both compounds. Fruit residue values will be presented in tabular form.

Air concentrations measured during monitoring of an eradication project will be analyzed as a two-factor repeated measures Analysis of Variance (ANOVA) with Application and Days-Post as the repeated factors, and the six stations as replicates. The ANOVA will show whether there is a significant accumulation of material over applications (main effect of Application), whether there is a significant build-up or decline of material over the days following application (main effect of Days-Post), or whether the behavior of the material over days is different after different applications (interaction of Application and Days-Post). The best-fitting model will be used to estimate mean concentration (and confidence limits) at each time point.

VI. ANALYTICAL METHODS/QUALITY CONTROL

Analytical method development and sample analysis will be performed by the CDFA laboratory. Fruit will be analyzed for total residues only. Air and fruit samples will be analyzed for methyl eugenol, naled, and DDVP (degradation product of naled). One solvent blank, 1 matrix spike sample and 2 replicate injections for 1 positive sample will be analyzed with each extraction set for each matrix.

VII. BUDGET

Personnel Expenses:	
Pilot Project	$4000
Eradication Monitoring	9500
Operating Expenses:	
Pilot Project	11000
Eradication Monitoring	22000
Total Costs:	$46500

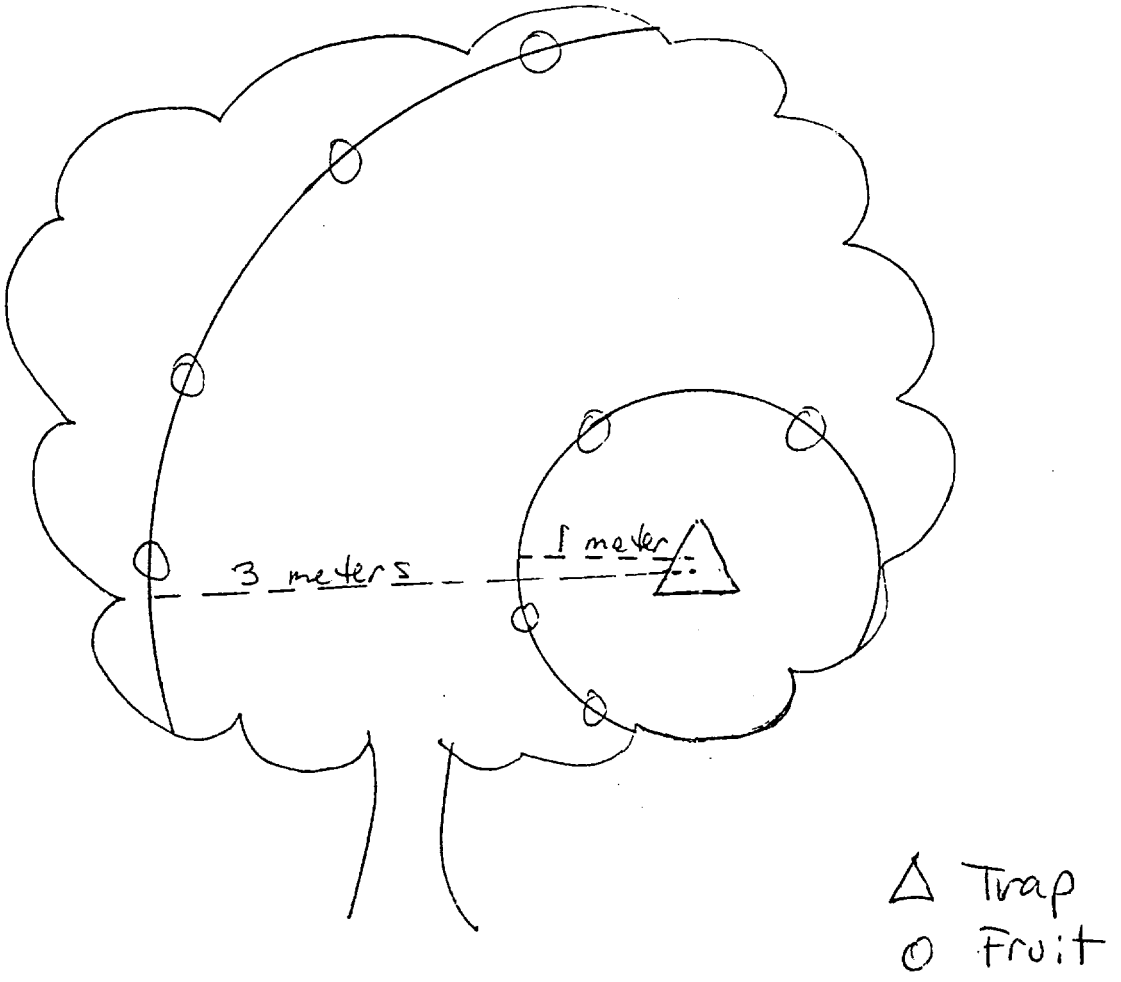
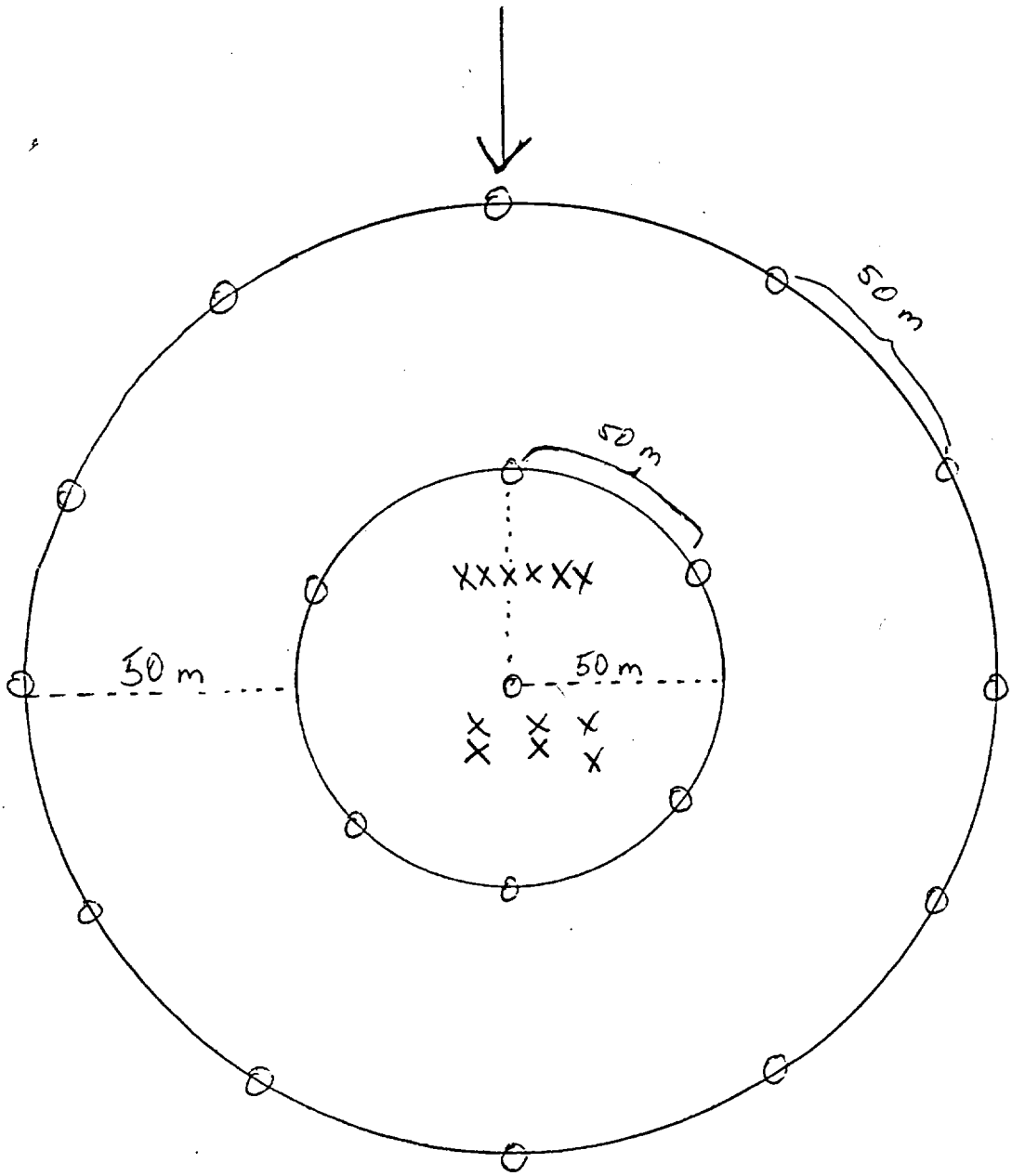


Figure 1. Cross-sectional diagram of sampling scheme for one fruit tree.



○ Bait station
× Air sampler
↓ Direction of prevailing wind

Figure 2. Top view of experimental set-up for measuring air concentrations.

Table 1. Sampling Intervals for Pilot Study of Methyl Eugenol and Naled Concentrations in Ambient Air near Bait Stations

Days after Application	Sampling Interval: 1-hour sampler (hours post application)	Sampling Interval: 2-hour sampler (hours post application)
Background	~~-4 0~~ ~~-1 -- 0~~	-2 -- 0
Application (Day 0)	0 -- 1 ~~2 -- 3~~ ~~4 -- 5~~ ~~6 -- 7~~ ~~7-8~~	0 -- 2 2 -- 4 4 -- 6 6 -- 8
1	early morn 1 hr mid-day 1 hr	early morn 2 hr mid-day 2 hr
2	early morn 1 hr mid-day 1 hr	early morn 2 hr mid-day 2 hr
7	early morn 1 hr mid-day 1 hr	early morn 2 hr mid-day 2 hr
Totals:	~~11~~ samples	11 samples

NC

DRAFT PROTOCOL
ENVIRONMENTAL MONITORING OF ERADICATION PROJECTS
WHICH USE METHYL EUGENOL

I. INTRODUCTION

The California Department of Food and Agriculture Division of Plant Industry uses methyl eugenol as a fruit fly attractant during trapping and eradication programs. This chemical has been chosen for further study by the federal government because of its similarity to other oncogenic chemicals. In response to the expressed perception that many people may be exposed to methyl eugenol during eradication programs, CDFA has decided to monitor an eradication program to determine the ambient air level of the chemical, if measurable, during and after typical applications.

II. OBJECTIVE

Our objective is to determine the ambient air level of methyl eugenol in neighborhoods where eradication programs are ongoing during 1988.

III. PERSONNEL

Ambient air monitoring will be conducted by the Calfornia Department of Food and Agriculture's (CDFA) Environmental Hazard Assessment Program (EHAP). Key EHAP personnel are listed below:

Bonnie Turner - Project leader
Sally Powell - Experimental design, statistical analysis
Karen Wiese - Field group coordinator
Nancy Miller - Laboratory liaison/quality control
Duc Tran - Chemistry lab coordinator

IV. MONITORING DESIGN

Monitoring will begin whenever EHAP is notified of an eradication program starting up within the State of California after the pilot program has been completed. Ambient air samples in the vicinity of the eradication bait stations will be collected over the eradication program or until proof of non-detectable levels of pesticides occurs at the treatment sites. If several eradication programs are underway in the state at the same time, only one will be monitored.

A maximum of four bait stations will be randomly selected from the eradication area. A high volume air sampler will be set up near each selected station. Air samples will be collected at three times following the first and last application of bait, tentatively at 0, 1 and 5 days after the application. The second and third applications will not be monitored. A total of 28 samples will be collected for analysis.

Distance of the samplers from the stations, the time of day for sampling, the duration of the sampling interval, and how many days post application to continue sampling will be determined from the results of the pilot study conducted earlier in 1988. Since it expected that it will be difficult to detect the methyl eugenol, these choices will be made to maximize the possibility of detection.

V. SAMPLING METHODS

All sampling will be performed using standard EHAP procedures (Sava, 1986). Replicate air samples will be collected using high volume air samplers calibrated at appropriate flowrates. XAD-2 resin will serve as the trapping medium. All air background and post application sampling periods will be a minimum of 4 hours. Air samples will be frozen at -70°F until delivered to the laboratory for analysis. Chain of custody records will be kept to document sample handling from sample container preparation through chemical analysis.

VI. STATISTICAL ANALYSIS

Air concentrations measured during monitoring will be analyzed as a two-factor repeated measures Analysis of Variance (ANOVA) with Application and Days-Post as the repeated factors, and the four stations as replicates. The ANOVA will show whether there is a significant accumulation of material over applications (main effect of Application), whether there is a significant build-up or decline of material over the days following application (main effect of Days-Post), or whether the behavior of the material over days is different after different applications (interaction of Application and Days-Post). The best-fitting model will be used to estimate mean concentration (and confidence limits) at each time point.

VII. ANALYTICAL METHODS/QUALITY CONTROL

Analytical method development and sample analysis will be performed by the CDFA laboratory. Air samples will be analyzed for methyl eugenol. One solvent blank, 1 matrix spike sample and 2 replicate injections for 1 positive sample will be analyzed with each extraction set.

VIII. BUDGET

The following expenses have been calculated based upon an eradication program which would take place in southern California. The budget does not include the expenses incurred by two employees to be furnished by the local Pest Detection field office.

Personnel Expenses:	
1 Seasonal employee at $8/hr for 12 days	$ 768
Operating Expenses:	
Per Diem --	
2 employees x 12 days x $82/day	1968
Airfare (LA) --	
2 employees x 3 trips x $150/trip	900
1 employee x 1 trip x $150/trip	150
Vehicles --	
2 vehicles x 1000 mi/ea x $0.25/mi	500
Chemistry --	
2 applications x 4 stations x 4 days (background, 0, +1, +5 for 1st application and 0, +1, and +5 for last) = 28 samples total x $150 each	4200
Total Costs:	$ 8486