AN ABSTRACT OF THE THESIS OF

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Title: Monitoring Pesticide Resistance in Psylla pyricola Foerster from Western Oregon Pear Orchards

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A modified slide-dip bioassay was used to survey and establish baseline resistance levels in Psylla pyricola Foerster from western Oregon orchards to four pesticides. Orchards selected in the Hood River, Rogue River and Willamette Valleys spanned a wide range of environmental and management conditions. During the study period of 1982-1983, management conditions present at these sites included intensively sprayed, minimally sprayed (IPM), recently abandoned, and long abandoned orchards.

Lethal concentration (LC_{50}) values determined for psylla populations ranged from 0.31-12.64 g AI/l for azinphosmethyl, 0.14-1.73 g AI/l for endosulfan, 0.02-0.10 ml AI/l for fenvalerate, and 0.41-1.52 ml AI/l for Perthane. Pear psylla from all orchards showed some level of resistance to azinphosmethyl (12- to 41-fold) and endosulfan (2- to 12-fold) when compared to the susceptible OSU Entomology Farm strain. Comparative differences in susceptibility

among populations to fenvalerate and Perthane, calculated by comparing each strain to the most suceptible strain, were between 1- and 5-fold.

Resistance was not correlated with insecticide usage in individual orchards in the Hood River and Rogue River Valleys, where commercial pear production is intensive. Psylla from unmanaged orchards had similar or slightly higher LC₅₀ values than managed trees for each compound. In contrast, in the Willamette Valley, where pear production is much less intensive and orchards are widely scattered, psylla resistance levels were better correlated with local insecticide usage patterns.

Pairwise comparisons of mean LC_{50} values for a region indicated that psylla resistance levels were significantly different at the regional level for azinphosmethyl and endosulfan. The mean LC_{50} value for Rogue River Valley populations to azinphosmethyl of 9.00 g AI/l was significantly higher (p<.05) than Hood River Valley and Willamette Valley psylla, which had mean values of 4.93 and 4.23 g AI/l, respectively. The mean LC_{50} value of 1.29 g AI/l for Hood River Valley psylla to endosulfan was significantly higher (p<.05) than mean values determined for Rogue River Valley and Willamette Valley psylla, which were 0.64 and 0.36 g AI/l, respectively. These regional trends in resistance accurately reflect differences in the intensity of use of the two compounds in these regions. Regional differences in mean LC_{50} values for fenvalerate and Perthane were not statistically significant.

A regional hypothesis of resistance development and population movement of pear psylla was proposed to explain the observation that

managed and unmanaged populations were equally resistant to azinphosmethyl and endosulfan in regions of intense pear production (i.e. Rogue River and Hood River Valleys). An overwhelming proportion of the psylla in a region develop in well-managed trees in commercial orchards and thus experience selective pressure from insecticides. In unsprayed orchards, which are few in these regions, tree vigor is low and natural enemies are abundant, which limits psylla reproduction and development during the season. During fall migration by overwintering psylla, populations from managed orchards and the limited populations from unmanaged orchards probably mix. The abundance of psylla produced in managed orchards and the psylla's dispersal behavior appear to have combined to produce populations possessing regional resistance characteristics in these regions.

In areas of less intense pear production (i.e. Willamette Valley), orchards are more scattered and population mixing is probably less extensive. Levels of azinphosmethyl and endosulfan resistance better reflected orchard-specific management in the Willamette Valley.

Monitoring Pesticide Resistance in Psylla pyricola Foerster from Western Oregon Pear Orchards

by

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Monitoring pesticide resistance in <u>Psylla</u> <u>pyricola</u> Foerster from western Oregon pear orchards

Introduction

With high demands on world agriculture to produce more food and fiber, man has turned to plant breeding, increased use of fertilizers, improved cultural practices, and pest management for innovative crop production. Agricultural pests (insects and acarines, pathogens, and weeds) take 1/3 to 1/2 of world crops each year (Pimentel 1977). Consequently, much effort is made to reduce the deleterious effects of these pests. Before the 1940's, man relied on simple inorganic compounds, such as mercurials and arsenicals for chemical pest control. The introduction of DDT and its nearly ubiquitous use for insect control ushered in the present era of intensive use of synthetic organic pesticides.

The first case of recognized resistance dates back to 1908, when Melander (1914) observed decreased effectiveness of lime sulfur to control San Jose scale (Quadrapediatus perniciosus (Comst.)). Loss of effectiveness due to resistance has been the fate of many other pesticides since that time. A recent complilation lists 432 species of

insects and acarines that have developed resistance to pesticides worldwide -- 262 of these are agricultural pests (Patton et al. 1982).

Considering the time and costs for developing and registering new compounds, it is useful to consider ways to prolong their effective use-life. The pesticide industry targets product development toward lucrative markets such as pests of cotton, wheat, corn, soybeans (these crops account for over 70% of total pesticide sales) (Conway 1982). Pests of less important crops, such as pear, must be controlled with compounds developed in these more profitable markets. As a result, resistance in pests of these minor crops may be of greater consequence if remedial compounds are not available. Pesticide susceptibility can be viewed as an exhaustable resource and pesticide use (which may cause resistance) has long term costs to the user (Hueth and Regev 1974; Comins 1979b).

The threat that resistance poses to modern agriculture has stimulated worldwide efforts to better understand resistance mechanisms and identify strategies of pesticide use that avoid, delay or revert resistance. This strategy has been referred to as "resistance management" (Georghiou 1972). There are two basic tenets underlying this type of approach, 1) reducing the selective pressure by a pesticide, and 2) anticipating resistance development (Georghiou 1972). In theory, reducing the selective pressure is the simplest and most effective method of resistance management, however this is not always feasible within the constraints of current agriculture. In addition to chemical management tactics such as compound alternations,

mixtures, and synergists, which act to reduce selective pressure to a single compound, many integrated pest management (IPM) concepts such as use of economic thresholds, biological control, intercropping, pest and environmental monitoring, and host-plant and predator resistance, act to reduce pesticide use, as well.

For a smooth transition from intensive chemical use to IPM programs including resistance management tactics, effective monitoring systems are essential to anticipate resistance problems (Croft 1982). In the past, resistance in a particular pest has been discovered by failure on particular crops rather than by pesticide susceptibility monitoring (Brown 1976). Determining baseline levels of resistance in the field and further resistance monitoring will furnish more accurate and realistic information on the rate and degree of evolving resistance.

The goal of this project was to better understand the patterns of resistance in pear psylla, <u>Psylla pyricola</u> Foerster, a serious pest of pear. This pest is notable for its ability to quickly develop field resistance to insecticides. This project developed a bioassay technique to accurately evaluate insecticide resistance in field populations of pear psylla. Field populations were chosen for the bioassay because rearing psylla in the laboratory is difficult and time consuming (Fye 1981). The specific bioassay selected was a slide-dip method, modified from a standardized method used in testing resistance in spider mites (Busvine 1980). In this study, the bioassay was used to survey and establish baseline resistance levels in Psylla pyricola Foerster from western Oregon pear orchards to four

pesticides. Once resistance baselines are established, populations can be compared and resistance evolution followed by routine monitoring. Ideally, accurate baselines can be used to develop a single-dosage diagnostic test to simplify the monitoring effort. This test would be invaluable in area-wide mapping to follow resistance development, progression and movement. The slide-dip test met the criteria of being simple, inexpensive and relatively quick. To my knowledge, this project represents the first attempt to develop a resistance bioassay for pear psylla.

Literature Review

Pear psylla bionomics

The Pacific Coastal states of California, Oregon and Washington produce over 95% of the pears grown in the United States. Oregon has approximately 7900 ha. in pear production. The pear psylla, <u>Psylla pyricola</u> Foerster (Homoptera:Psyllidae), is the most costly pest of pear to control in the three state region. Pear psylla was introduced from the East Coast into Washington in 1939, and subsequently spread through Oregon (1949-1950) and northern California (1953-1958) (Westigard and Zwick 1972).

Two forms of the psylla are produced each year. Overwintering adults are produced late in the summer and disperse from orchards in late summer and early fall seeking protected habitats to overwinter. Psylla occur on other hosts throughout the year, but require pear on which to complete their development. In late January or early February, the majority of the overwintering adult psylla return to pear trees to mate and reproduce (Westigard and Zwick 1972). Mated females lay an average of 664 eggs (Burts and Fischer 1967). Eggs are layed singly on or near leaf and fruit buds prior to budbreak, and on leaves after budbreak. Egg hatch coincides with budbreak and immatures feed on lush green foliage where they suck cellular fluids from many types of leaf tissues (Ullman, unpublished data). In the Pacific Northwest, 2-4 summer generations of psylla are produced each season (Westigard and Zwick 1972).

In the absence of pesticides, pear psylla populations can be regulated by a complex of natural enemies (Madsen et. al. 1963; Westigard and Zwick 1972; Fields and Zwick 1977; Keimer 1983). The influence of natural enemies on psylla seems to be related to the kinds of predators, their density and the time they appear in the orchard (Gut et al. 1982) Commercial orchards receive preventative chemical sprays for the codling moth, Cydia pomonella L., a direct pest of pear fruit, several times per season. The broad spectrum pesticides now in use destroy natural enemies of the pear psylla during the period they are immigrating into orchards in the spring and early summer.

Feeding by unchecked psylla populations can produce several types of damage. Honeydew exuded during feeding causes necrosis of foliage, serves as a site for fungal infection by a sooty mold, and causes russeting on the fruit (Burts 1968). Fruit blemishes result in downgrading of fruit intended for the fresh market. Heavy psylla infestations produce phtyotoxicity causing reduced photosynthesis, stunting of vegetative growth, and leaf and fruit drop (Burts 1968). Pear psylla also vectors the mycoplasma that causes pear decline. Rapid and slow decline in varieties with susceptible rootstocks results from damage to phloem tissue at the union of stock and scion wood. Infected trees on more tolerant rootstocks develop leaf curl symptoms prematurely in late summer and show reduced leaf and shoot growth, poor fruit set and size, and dieback of branches (Williams et al. 1978). In 1962, it was estimated that Californian pear production had fallen to 40-50% of its previous level because of pear decline

(Westigard and Zwick 1972).

Sprays applied against psylla fall into two general categories: pre-bloom or dormant sprays and post-bloom or foliar sprays. Nymphs and summer adults feeding on the foliage receive foliar sprays. Dormant sprays destroy overwintering forms and limit egg laying in the spring (Burts 1968). Effective dormant sprays must be accurately timed so that psylla overwintering outside the orchard have returned or have left protected sites and are active on the tree (temperatures above 7°C).

Burts (1983) compared a soft (selective) pesticide program with a hard (non-selective) pesticide program typical of commercial pear orchards in the Pacific Northwest. The soft pesticide regime, consisting of two prebloom sprays of petroleum oils followed by four postbloom washes, failed to prevent psylla damage (russeting) on the fruit. The standard (non-selective) program of fenvalerate and oxythioquinox sprays prebloom and foliar sprays with amitraz provided better control than the soft program. Amitraz is very effective against nymphs but not adults. Fenvalerate appears to be effective against most stages but is restricted to dormant season use to reduce the probability of resistance development (see later discussion of factors of resistance development). Pre- and post-bloom sprays of effective materials are currently the only means to reliably control pear psylla to subeconomic levels (Westigard and Zwick 1972, Burts 1983).

History of resistance in pear psylla

The development of resistance in populations of pear psylla, based on control failures in the field, is summarized here from Westigard and Zwick (1972). The chemicals to which psylla has developed resistance are shown in table 1. A number of organophosphates, including malathion, parathion and EPN, effectively controlled psylla in Oregon until 1956. These compounds were replaced by the cyclodienes aldrin, dieldrin and toxaphene, but within 2-4 years these materials were also ineffective. Azinphosmethyl became the widely accepted compound in 1960, but field resistance had developed by 1965. Combination of azinphosmethyl with petroleum oils extended its useful life to 10 years. Other organophosphates, cyclodienes and Perthane (a DDT relative) were in use until the early 1970's when they too were abandoned because of resistance.

Concepts in resistance development

A "resistant" strain has a biochemical, physiological or behavioral mechanism which increases its survivorship compared to a susceptible strain when exposed to a level of pesticide.

Intensive spray use creates a strong selective pressure for pest insects. Resistant individuals survive spray applications to mate and reproduce. The genetic traits conferring resistance to the parent are passed on to their offspring and further mating and reproduction by

Table 1. Pear psylla control agents abandoned after losing effectiveness in the field (1950-1984).

Group	Compounds		
Organophosphates	azinphosmethyl, malathion, parathion, EPN		
Organochlorines	BHC, dieldrin, endrin, toxaphene, DDT, Perthane, endosulfan		

resistant individuals in subsequent generations also under selective pressure produces a population with a higher proportion of individuals resistant to the chemical (Craig and Patton 1982). With repeated use, chemical compounds and groups of compounds have become ineffective in controlling pear psylla (Harries and Burts 1965; Westigard and Zwick 1972). Typically, as resistance develops in the field, the number of spray applications are increased and/or made at higher rates in an attempt to control the pest at comparable subeconomic levels. As application rates or the number of applications increase, selective pressures produce an increasingly resistant population.

It has been proposed that a resistance scenario has several phases. Initially, the population may exhibit a period of slightly increasing susceptibility as the genes associated with resistance are often associated with reduced fitness (survivorship). Resistance eventually appears and slowly increases before accelerating through a log phase increase to high levels under intensive selection pressure (Croft 1979). When an organism develops resistance to a pesticide causing abandonment of the chemical, the genotypes conferring resistance may return to low frequencies in the population if the genotypes are associated with lower fitness (Brown 1976). Reversion to susceptibility from high levels is usually slow, but reversion during the early stages of resistance development can be rapid (Abedi and Brown 1960; Zilbermints 1977; Devonshire and Sawicki 1979; Sawicki et al. 1980). The phenomenon of slow reversion may be due to multiple resistance factors that incorporate fitness traits into the resistance genome (Brown 1976). Thus, stabilization of resistance requires the

accumulation of certain ancillary alleles to constitute a favorable geneotype for a given condition (Milani 1958).

Genes conferring resistance are believed to arise from mutations present at very low frequencies in the population, before insecticidal selection occurs (Patton et al. 1982). Most all mutations, if they produce a recognizable effect, are harmful to an individual well adapted to a particular environment. The beneficial mutations necessary for evolution are expressed in response to severe changes in the environment (Crow 1983; MacDonald 1983). An intensively sprayed crop system is an example of a severe environmental stress that creates a strong selective force. Mutations are known to be variable in the time and constancy of expression, and mutation rates vary greatly among loci and among individuals (Crow 1983). Recent evidence from molecular genetics suggests there may be a significant environmental component to mutation rates - specifically, mutation rates may significantly increase in response to environmental stress (Macdonald 1983).

Three primary mechanisms of resistance in insects are reduced penetration, modification of the target site, and increased detoxification (Georghiou 1972). Penetration may be reduced at the level of the cuticle or the nervous system. Decreased sensitivity at the target site may involve the alteration of a target enzyme, such as synaptic acetylcholinesterase, or lowered nerve or nerve membrane sensitivity. Enhanced metabolic detoxification capabilities may result from changes in the complement of mixed function oxidases, esterases, hydrolases, and glutathione s-transferases. Increased detoxification

may also result from altered gene amplification or enzyme induction systems. Resistance to organophosphate, chlorinated organic, carbamate and synthetic pyrethroid insecticides is a result of one or more of these factors (see reviews by Plapp 1976, and Georghiou and Saito 1983).

Certain behavioral resistance mechanisms, such as duration of exposure and host habitat preference, are fortuitous properties that confer selective advantage to an organism although they are not direct causes of resistance. However, since all biological processes are affected by genes, behavioral responses can also be modified by selection and changes in the genome. Some have speculated that behavioral modifications are hypothetically as likely to arise as physiological or biochemical ones (Pluthero and Singh 1983).

Georghiou and Taylor (1977a,b) discuss the operational, biological and genetic factors that influence the rate of resistance development. Genetically, the rate at which an arthropod will develop resistance is determined by the degree of dominance, the monofactorial or polyfactorial nature, and the initial frequency of the genes conferring resistance. A single gene is usually responsible for each mechanism, but modifier genes may reinforce the primary resistance mechanism. Resistance arises much quicker when the gene is dominant than recessive when the resistance factor is monofactorial. The expected frequency of alleles conferring resistance is 10^{-3} to 10^{-4} in arthropods (Georghiou and Taylor 1976). The lower the inital gene frequency the longer it may take for resistance to appear.

Biological factors affecting the rate of resistance include the

number of generations per year, reproductive potential, migration, host specificity and the presence or absence of refugia (Georghiou and Taylor 1977a). In general, the rate of resistance increases as the number of potentially reproductive individuals continuously exposed to the selective force increases. Hence, multivoltine species generally develop resistance faster than univoltine species, highly reproductive species faster than species with low reproductive potential, and so forth. The pear psylla has several biological attributes that pre-adapt it to rapid resistance development. The psylla is sedentary during the growing season, has a high fecundity, is multivoltine and feeds and matures only on pear (Riedl et al. 1982).

Key operational (management) factors are dosage and frequency of application, type and sequence of pesticide application, application thresholds, and life stages selected (Georghiou 1977b). Here, increasing the selective pressure of the compound or a group of related compounds over time, will accelerate resistance development in the pest. In pests that have been exposed to a number of materials, multiple resistance factors and cross resistance may be expressed during a resistance episode (Georghiou and Lagunes 1983).

Computer simulation studies (Taylor and Headley 1973; Comins 1977; Georghiou and Taylor 1977a,b; Plapp et al. 1979; Tabashnik and Croft 1983) have shown migration, functional dominance and dose to be especially influential in a resistance episode. Tabashnik and Croft (1983) point out that the effects of a given factor may differ

depending on the biological and operational conditions under which it is evaluated.

Concepts in resistance management

Obviously, the surest means to delay or avoid resistance development is to reduce the selective pressure. Four in-field strategies involving the manipulation of pesticides to reduce this selective force are 1) alternating compounds with different modes of action, 2) the use of synergists, 3) well timed spray applications, and 4) accurate economic thresholds (Riedl et al. 1981). Developing accurate economic thresholds will minimize the number of necessary pesticide exposures. Well timed sprays maximize the sprays effect and reduce the need for later control. Alternating compounds will, in theory, create opposing selective forces, maintaining resistance genes for a single mechanism at low levels in the population (Georghiou 1982). Synergists have been combined with pyrethrins to inhibit oxidative mechanisms of detoxification (Plapp 1976).

Cross resistance between related and unrelated compounds may arise from selective pressures exerted by alternations and mixtures. Georghiou and Taylor (1976) explain that when alternating or mixing compounds, "each compound seems to improve the residual inheritance of the supporting genome in favor of the development of resistance in the other". Several researchers have observed negatively correlated cross resistance (Ogita 1958; Georghiou et al. 1978; Chapman and Penman 1979; Lagunes-Tejeda 1980). In the case of negatively correlated cross

resistance, pest resistance to one compound causes reversion toward susceptibility to another compound to which resistance had previously developed.

Encouraging resistance development in natural enemies may help regulate pest populations to subeconomic levels and reduce the number of necessary spray applications (Hoy 1979a, Croft and Strickler 1983). Strip, alternate row or patch spraying can control economic pest outbreaks while minimizing selective pressure on the population. Creating refugia in a crop system serves the same function.

Susceptible individuals are left untreated and mate with resistant survivors. In the case where genes conferring resistance are recessive or functionally recessive (Tabashnik and Croft 1983), the mixing of susceptible individuals with the resistant population reduces the chance of resistant individuals mating and slows resistance development in the field. The presence of refugia was a major factor contributing to slowed resistance development in models tested by Comins(1977), Kable and Jeffery (1980), Tabashnik and Croft (1983), and Taylor and Georghiou (1979).

It was pointed out earlier that pesticide resistance management tactics are most likely to succeed when resistance can be detected early in its genesis. A test to determine the proportion of individuals in a population that possess resistance traits at an early stage would be a valuable supplement to present LC_{50} determinations used to characterize resistance and resistance development (Zilbermints 1977). This would be a more discriptive index of population changes. Genotypic rather than phenotypic tests would

describe the monofactorial or polyfactorial nature of resistance. An example of a descriptive test for resistance monitoring is the starch gel electrophoresis test used by Georghiou and Pasteur (1978) to detect esterase properties in OP-resistant or susceptible $\underline{\text{Culex}}$ mosquitos.

Materials and Methods

As noted earlier, the objectives of this study were: 1) to develop a slide-dip technique for bioassaying susceptibilities of pear psylla populations to insecticides, and 2) to test and characterize the differential susceptibilities of psylla populations from diverse orchard environments to four insecticides.

Orchard site selection

To sample resistance in <u>P. pyricola</u>, 13 pear orchards across western Oregon were selected which differed in geographical location, climatic conditions, surrounding vegetation, and management practices. The orchards chosen spanned the environmental and management diversity experienced by this pest in the three principal pear growing regions of the state - the Hood River Valley (central Cascades), the Corvallis and Salem areas (central Willamette Valley), and the Medford area (Rogue River Valley)(figure 1). During the study period of 1982-1983, the range of management conditions present at these sites included intensively sprayed, minimally sprayed (IPM), recently abandoned and long (40 years) abandoned orchards (table 2).

Describing specific characteristics of each orchard in each of the three pear growing regions illustrates the approach to experimental design. Each region is described in terms of it's geographical location, climatic patterns and vegetation types (Franklin and Dyrness 1973). Each orchard is described in terms of its

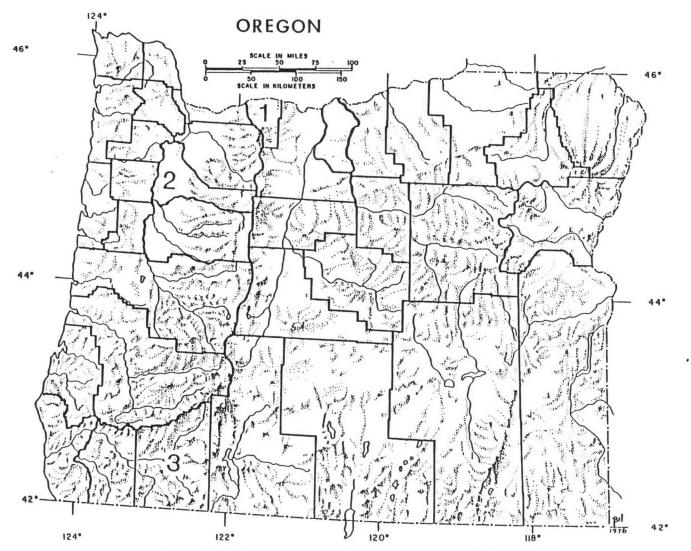


Figure 1. Three principal pear growing regions of Oregon. 1. Hood River Valley 2. Willamette Valley 3. Rogue River Valley.

Table 2. Orchard sites sampled during the 1982 and 1983 growing seasons.

Region	Orchard	Management
Willamette Valley	Corvallis	commercial
	Wiley	commercial
	OSU Ent. Farm	unmanaged
	Botany Farm	experimental/commercial
Hood River Valley	McCarty	commercial
	Facteau	unmanaged/shade trees
	Merz	IPM/commercial
	Logan	unmanaged
	Tamiyasu	commercial
Rogue River Valley	Rogue	unmanaged/abandoned
	Cherry Ln.	unmanaged/abandoned
	Carpenter Hill	unmanaged
	Hanley	experimental/commercial

management practices, surrounding environment and location within the region.

Hood River Valley - This valley, located in the central Cascades of northern Oregon has two distinct climatic regions. The upper valley receives relatively heavy rainfall (as high as 2300mm), substantial snowfall and experiences cool summer temperatures. The lower valley, moderated by the marine air of the Columbia River Gorge, receives less rainfall (ca. 650-800mm), much less snowfall and experiences warm summer temperatures. The Hood River Valley is in the transition zone of the Cascades where orographic rainfall produces a wet western slope and a drier eastern slope. The drier eastern slope of the lower valley characteristically supports Pinus ponderosa (Ponderosa pine) and the remainder of the valley supports Pseudotsuga menziesii (Douglas-fir) and Abies grandis (Grand fir). The valley has approximately 3970 ha. in pear production.

McCarty orchard - prebloom and foliar sprays are applied on a calendar basis in this commercial orchard which is located in an area of concentrated orchards in Rockford in the lower valley.

There are mixed conifers and hardwoods bordering the orchard on the north.

Logan orchard - formerly a commercial orchard, this orchard was unmanaged during the 1983 growing season. Located in Pine Grove on the dry eastern slope of the lower valley, this orchard is

surrounded by commercially sprayed orchards on three sides and an open grassy field to the north. This area has a high proportion of managed pear orchards.

Tamiyasu orchard - is a commercial orchard situated in Oak
Grove on the moist western ridge, which is on the edge of a
concentrated pear growing area of the lower valley. The orchard
consists of mixed tree fruits and is surrounded by conifers.

Merz orchard - is located in Parkdale in an area of
concentrated orchards in the upper valley and is bordered by
coniferous forest, a lava flow and a tree nursery. This orchard
seldom requires sprays against the codling moth. As a result,
natural enemies often control pear psylla to sub-economic levels,
which further reduces the number of insecticide sprays each year.
I characterize this orchard as a minimally sprayed or "IPM"
orchard.

<u>Facteau orchard</u> - this site consists of a dozen pear shade trees at a residence in Hood River in the lower valley. To the resident's knowledge, these trees have never received a spray for insect control. The residence is in an area of concentrated commercial orchards.

<u>Willamette Valley</u> - This valley lies in western Oregon between the Cascade and the Coastal Ranges. Vegetation is mixed agriculture on the valley floor and <u>Quercus garryana</u> (Garry oak)/grassland in the foothills of the Coastal Range (including the area around Corvallis). Temperatures are moderate, average annual precipitation is ca.

1000-1150mm, and average snowfall annually is 200mm in the valley. Approximately 115 ha. of pear production are scattered throughout the valley. The single largest concentration of orchards occurs north of Salem.

<u>Wiley orchard</u> - is situated northwest of Salem in Grand Island on the Willamette River. This orchard is managed commercially and is distinct from any other orchard in that it has a sod ground cover. The surrounding area is mixed agriculture and no managed pear orchards occur wthin eight kilometers.

OSU Entomology Farm orchard - pear trees on this research farm in Corvallis have not been sprayed for more than 30 years. Within a three kilometer radius, there is only one small commercial block of pears and scattered abandoned trees. Orchard trees are surrounded by other deciduous fruit trees (apple, cherry, peach) which are experimentally treated with insecticides from time to time.

Botany/Plant Pathology Farm - is located near Corvallis. Psylla were collected from a block used for experimental fungicide trials. This block receives insecticide applications only when insect pressure is observed, which is less frequent than in most commercial orchards. A wide variety of fruit, nut, grain and forage crops surround this orchard.

Corvallis orchard - is a small commercially managed orchard.

This pear orchard is located in Corvallis and is bordered by a christmas tree farm and residences. There are very few pear

orchards in this area.

Rogue River Valley - The Rogue River Valley runs between the Siskiyou Mountains and the lower Cascades in the southwestern corner of Oregon. Pear growing is concentrated in the southeastern end of the valley near Medford and includes approx. 3770 ha. This region is warmer than the other two valleys and receives less rainfall (ca. 500mm). Dry sites in this area are dominated by oak woodland, including Quercus garryana and Q. kelloggi (Oregon white oak and California black oak), and where the sites are even more xeric, grass coverage increases and oak coverage decreases. Mesic sites are composed primarily of Pseudotsuga menziesii, Pinus ponderosa and Libocedrus decurrens (Incense cedar).

Rogue orchard - this overgrown orchard has been abandoned and unsprayed for 40 years or more. The site is very dry and dominated by oaks and sage. Located in the northwest corner of the valley, this orchard is isolated from the main area of pear production and is 15 kilometers from the nearest commercial orchard.

<u>Carpenter Hill orchard</u> - until the 1983 season, this orchard had been managed commercially. The orchard was left unmanaged during the 1983 season and harbored a large population of pear psylla. It is located in Phoenix near the center of the region's commercial pear culture.

Cherry Lane orchard - two dozen trees form a fence row along

this road on the outskirts of Medford. The surrounding area was at one time planted entirely to pear tree, but now only a few scattered trees remain. These trees have not been sprayed with insecticides for over 40 years.

Southern Oregon Agricultural Experiment Station (Hanley)

orchard - this block of densely planted trees is intensively sprayed with experimental and commercial compounds. It is in an area of mixed agriculture outside of Medford. There is a low density of commercial orchards in this area.

Compound selection

The insecticides choosen for resistance tests represent three classes of compounds whose use in past and current chemical pest management programs in pear differs markedly. These differences occur both within and between regions. The four compounds tested were fenvalerate (Pydrin 2.4EC), azinphosmethyl (Guthion 50WP), endosulfan (Thiodan 50WP) and Perthane (4EC). Azinphosmethyl is an organophosphate, endosulfan and Perthane are chlorinated organics, and fenvalerate is a synthetic pyrethroid insecticide.

The degree to which these four compounds were used in each region is summarized in table 3. Since 1960, azinphosmethyl has been a commonly used compound to control the codling moth in all regions.

Because codling moth control is preventative, this compound is applied to an orchard several times every season. Orchards in the Rogue River

Table 3. Patterns of insecticide use in the Hood River (HRV), Rogue River (RRV) and Willamette (WV) Valleys.

Compound	Region	Used in pear orchards	Average number of applications per year	Timing of applications	Maximum recommended field rate	Field resistance detected in psylla	Target pests
	HR♥	1960-present	2-3	summer	0.45 g AI/1	1965	cm, pį
azinphosmethyl	RRV	1960-present	3-4	summe r	0.45 g AI/1	1965	cm, p
	WV	1960-1980	1	summer	0.45 g AI/1		cm, s
	HRV	1971-1977	1-2	dormant & summer	0.75 g AI/1		PP
endosul fan	RRV	rarely	-	summer	0.75 g AI/1		рр, в
	NV	rarely	•	8 umme r	0.75 g AI/1		8p
	HRV	1978-present	2	dormant	0.12 ml AI/1		PP
fenvalerate	RRV	1978-present	2	dormant	0.12 ml AI/1		PP PP
	wv	rarely	_	do rma nt	0.12 ml AI/1		PP
	HRV	1965-1977	3-4	dormant & summer	1.13 ml AI/1	1970	pp
Perthane	RRV	1965-1978	1	dormant	1.13 ml AI/1		PP
	· WV	rarely	-	dormant	1.13 ml AI/1		PP

¹ pp = pear psylla, cm = codling moth, sp = sporadic pests.

Valley experience greater codling moth pressure and so require more frequent sprays for this pest (3-4 sprays per season) than the other two valleys (2-3 sprays per season). The codling moth develops more rapidly in the warmer Rogue River Valley and experiences lower 1st instar mortality because of less rainfall in this region (Westigard et al. 1976). When it was first introduced in 1960, azinphosmethyl was also used to control psylla, but after five years of use resistance was detected in the field.

Endosulfan has never been particularly effective against pear psylla. It has been used infrequently in the Rogue River and Willamette Valleys in combination with other compounds for psylla control. Endosulfan's most extensive use has been in the Hood River Valley where it was used in combination with Perthane for dormant and summer control of pear psylla from 1971-1977.

Present psylla management emphasizes a well-timed dormant spray with fenvalerate to kill overwintering forms as they return to orchards to mate and oviposit in late January or early February. Resistance to fenvalerate has not appeared in Hood River but has been reported from a single site in the Rogue River Valley (Riedl et al. 1981). It is still not used widely in the Willamette Valley.

Perthane is an interesting example of a compound with a different pattern of state-wide use. In the Hood River Valley, it was applied against psylla as a dormant and foliar spray, whereas it was used only as a dormant spray in the Rogue River Valley. The extent of its use in the Willamette Valley is uncertain.

Psylla populations have experienced varying degrees of selective

pressure to these and related compounds in the past, depending primarily on, 1) the orchardist's philosophy toward pest management and pesticide use, 2) the commercial/non-commercial status of the orchard, and 3) the pattern of regional use of the compound. The regional use pattern is a function of many variables, the most important of which are the length of time a chemical was recommended for use against pear psylla and the effectiveness of the compound to economically control pear pests.

Slide-dip technique

Adult pear psylla were tested for susceptibility to insecticides using a slide-dip method adapted from a standard FAO method used to test resistance in spider mites (Busvine 1971, 1980). Several aspects of this method were evaluated to determine their relative importance in the mortality response of tested pear psylla. These evaluations are described in a later section. Many adjustments were made during the developmental stage of the method until a standardized method could be defined. This standardized method is explained in detail below.

Summer adult psylla are collected in the field by jarring pear foliage with a tapping bar (Burts`1973). Populations from managed orchards were not collected until at least one week after the latest field treatment of pesticides. After jarring, psylla fall to a hand-held catching frame from which they are aspirated into a padded vial. Collections are made from the lower branches while walking along orchard rows. After 150-200 pear psylla have been aspirated, the vial

is capped and placed in an ice cooler (at ca. 4° C). Psylla are kept cool during transportation and storage prior to their treatment in the laboratory.

In the laboratory, psylla are first anesthetized with CO₂. Carbon dioxide is blown into the vial through a hypodermic needle which is connected to a CO₂ tank by surgical tubing. The needle is inserted through a soft vial cap. Anesthetized psylla are placed with a moistened #1 camel hair brush on their dorsum on a 3cm strip of nylon sticky tape (Permacel brand) that has been stuck to a slide with a strip of double adhesive tape (3M brand). The two-sided adhesive tape is used to secure the single-sided sticky tape because the former is not sticky enough to hold psylla in place for the length of the test evaluation period. Under a stereomicroscope, wings are parted posteriorly and pressed back against the adhesive tape surface to secure adult psylla to the slide for the duration of the test.

Commonly, 25-30 psylla are mounted per slide. Males and females are tested indiscriminately. The slide is then dipped and stirred for five seconds in the diluted toxicant.

A dilution series is performed to obtain the range of dosages for a particular compound. The highest concentration is measured out and then diluted to fractional concentrations to complete the series. Formulated compounds should be stored in a refrigerator and re-prepared every three days. Specimen jars (264ml = 8 oz.) are ideal mixing containers that allow easy and complete dipping of the slides. Shaking the jar immediately before dipping produces a uniform suspension of active ingredient.

The slide-dip procedure is replicated four times (n=100) for each concentration in the serial dilution for a compound. For every 400 psylla dipped in a toxicant, 50 psylla are dipped in water to serve as a control. Slides are air-dried for five minutes (tapping the slide on edge on a paper towel will blot away large water droplets) and placed in a holding chamber for 48 hours. The chamber has high humidity (95-100%) and is stored at between $16-24^{\circ}$ C.

The holding chamber for slides with mounted and treated psylla is a cafeteria tray covered by a thin sheet of plexiglas. Wet paper towels covering the bottom of the tray maintain high humidity in the chamber and adhere to the undersides of the slides to prevent movement of slides during handling. Thin plexiglas (3mm) will bend upward at the corners allowing venting of evaporative moisture. A sheet of glass, on the other hand, will form an airtight seal causing water droplets to form on it's under surface. The droplets may drip onto the mounted psylla below, causing mortality.

Mortality readings are made after 48 hours using the set of mortality criteria given below. Psylla are prodded using a fine camel hair brush. The following responses indicate survival:

1) a "jump" response - hind legs shoot (flex) forward toward the head; this is a very quick reflex that would under normal conditions catapult the psylla into flight.

- 2) activity in all six legs (or all legs that are not disabled or adhering to the tape).
- 3) psylla upright (wings not stuck or poorly stuck) and responds to prodding by flexing or extending it's legs.
 - 4) rapid "boxing" response from front legs.

Dead psylla are characterized as:

- 1) legs often fully flexed and close to the body a "fetal-type" position.
- does not respond to prodding, or may slowly flex one or several legs.

The criteria presented above were chosen in order to distinguish a psylla that would be an able participant in reproduction, from an individual that would obviously no longer be an active member of a reproducing population. The mortality decision is to some extent a subjective interpretation, but these criteria provided the most accurate index of susceptibility for these studies.

Analysis of slide-dip data

The mortality figures from each slide-dip test were evaluated using a log dosage-probit (ld-p) analysis program. Abbott's formula was used to adjust values for check mortality. The LC_{50} values

generated from this analysis were used to compare the degree of susceptibility of an orchard population to those of other orchards.

Regional LC₅₀'s for each compound were grouped and a mean computed. F-values from ANOVA were calculated from the pooled variances and then a pairwise comparison was performed using the Wilcoxon rank sum test (Tashman and Lamborn 1979) to reveal inter-regional differences in susceptibility for a compound. (The Wilcoxon rank sum test is also called the Wilcoxon two-sample test and is equivalent to the Mann-Whitney test.) This non-parametric test was deemed appropriate for the data because of the small sample size and the unknown distribution of insecticide tolerance/resistance traits in the population.

Evaluation of potentially important variables in testing susceptibility in psylla

The objective was to develop a test that used field collected psylla because laboratory rearing is extremely difficult and expensive. Throughout the development of the slide-dip test, I observed and speculated on potential sources of variability. The bioassay proposed herein incorporates the inherent variability in natural field populations (i.e. nutritional, environmental, physiological), but attempts to standardize the variability of controllable factors. Intrinsic variables, such as age and stage, and extrinsic variables, including temperature, humidity, photoperiod, formulation, holding period and handling technique, are standardized

to within reasonable limits. Several manipulable variables in the testing procedure or related to the bioassay, or factors of suspected importance, were evaluated more closely to determine their potential influence on variability.

1)Effect of multiple carbon dioxide anesthetizations:

Carbon dioxide was applied in treatments of 1, 5, 10 and 15 exposures, to 100 pear psylla. Of the 100 in each treatment, 50 were dipped in water and 50 in a 0.012 ml AI/l concentration of fenvalerate by the standard slide-dip method. Psylla mortality was observed after 48 hours.

The manner of CO_2 treatment mimicked the procedure commonly followed in a slide-dip test. First, pear psylla were exposed to 60 seconds of CO_2 in the vial, the vial was then uncapped and left open to the ambient air for 60 seconds, and finally recapped for another 60 seconds before the next CO_2 exposure. This was repeated 1, 5, 10 and 15 times for the four different treatments.

2)Holding period:

During preliminary testing to determine chemical dosage ranges, mortality readings were taken after 48 and 72 hours. Results from these readings were used to select a standard holding period.

3) Fluctuations in susceptibility measures:

Preliminary data from 1982 indicated that, as the summer progressed, adult psylla became increasingly susceptible to several of the test compounds. To document this phenomenon, three times during the summer of 1983, 200 psylla were collected from an orchard in Medford and tested for their susceptibility to two dosages of fenvalerate. Fluctuations in mortality resonses were recorded over this period.

Results and Discussion

Observations on sources of variability in the slide-dip technique

The precise use of the slide-dip technique is largely dependent on controlling four sets of variables: temperature and humidity, mounting technique, holding period, and mortality criteria.

1. Temperature and humidity conditions

Maintaining proper temperature and humidity conditions appeared to be the most important extrinsic variables. When exposed to direct sunlight, a "greenhouse effect" is created inside a collection vial or holding chamber producing high temperatures. For example, when the ambient air temperature is 31° C in the sun, air temperatures in a collection vial reach 35° C after only five minutes in direct sunlight.

Desiccation can cause high mortality in psylla during the 48-hour holding period if humidity is not kept high. The importance of this variable was demonstrated when mounted psylla in a holding chamber were placed in an environment chamber to control temperature. Without humidity control, the rapidly circulating air in an environment chamber dried the wet paper towels in the holding chamber, producing low relative humidity and causing excessively high mortality.

2. Mounting technique

Psylla must be handled delicately during mounting. Also, minimizing handling time will reduce the chance of injury which can

contribute to mortality. A fine camel hair brush and Permacel brand nylon packing tape are important items. Wetting the brush forms a bead of water on the hairs allowing quick and gentle transferal of psylla. Wings adhere easily to Permacel brand nylon tape, whereas other brands may require injurious handling to stick wings securely.

Multiple anesthetizations of psylla with carbon dioxide before mounting did not appear to affect subsequent treatment mortality. As the number of exposures increased, mortality readings did not increase in a regular fashion for either the water or fenvalerate treatment. After 1, 5, 10, and 15 exposures mortality readings were 0.0%, 7.8%, 4.2%, and 6.0% for psylla dipped in water (control), and 25.0%, 12.7%, 18.9%, and 8.2% for psylla dipped in a 0.012 ml AI/l solution of fenvalerate. This result is consistent with Busvine's (1971) experience that many species survive very long (hours) exposures to carbon dioxide.

3. Holding period

Preliminary tests showed that check mortality was generally low (0.0-3.8%) after 48 hours in the holding chamber, but often increased to considerably higher levels (0.0-20.0%) after 72 hours or more. Therefore, 48 hours became the standard holding period. The average check mortality for all 48 hour readings over the two year testing period was 8.7%.

4. Mortality criteria

In initial tests, psylla were judged alive if they showed any appendage movement when prodded. The modification of this criteria to

differentiate between movements of a healthy psylla and slowed movements of a marginal survivor (see discription in <u>Materials and Methods</u> section) is a more accurate index of survivorship in the field, and with practice is a relatively objective method.

Fluctuations in susceptibility measures

An additional variable was observed to be significant. Pear psylla susceptibility to insecticides appeared to change during the course of the season. While running preliminary slide-dip tests during the summer of 1982, psylla tested in late August showed higher mortality to Perthane, and substantially higher check mortality, than psylla tested earlier in August. This difference in mortality was attributed to the higher proportion of "old age", summer adults late in August.

Susceptibility levels to fenvalerate were measured in summer adult psylla collected from the Hanley orchard in Medford over a four month period at four sampling dates (May 29, July 5, July 18, August 15). Using a slide-dip test and a 0.2 lb. AI/A rate of fenvalerate, mortality rates were 60%, 20%, 30%, and 40%, respectively; the same flucuating pattern of mortality readings appeared at a lower rate (0.1 lb AI/A). This suggests that there is either considerable variability in this test procedure or, alternatively, that physiological or biochemical changes may occur in psylla throughout the year which affect their tolerance to fenvalerate. More study is needed to identify the cause of this variability.

In light of this observation, it is recommended that 1) orchard populations be tested early in the season, when a uniform age distribution of adults is present and before environmental stress factors (e.g.,leaf tissue hardness, temperature) become important, and 2) tests be run at all sites in a region over as short a time period as possible. This will reduce the variability in mortality responses among orchards that may possibly result from a population's fluctuating susceptibility.

A susceptible strain of pear psylla

An important prerequisite to documenting suspected resistance is to obtain a susceptible strain against which other strains can be compared. An unsprayed or unmanaged orchard, where selective pressure from insecticides is low or non-existent, would be expected to contain susceptible psylla. In reality, most abandoned or unmanaged orchards did not contain susceptible psylla in this study. The only population that appeared to be inherently susceptible was the OSU Entomology Farm strain in the Willamette Valley, which was sampled extensively in the first summer of the study (1982). Collecting in other isolated, unmanaged orchards in this valley produced very few psylla. Over the study period (1982-83), it is estimated that over ten abandoned orchards in the Willamette Valley were inspected in hopes of finding psylla without success. In contrast, abandoned and unmanaged orchards in the Hood River and Rogue River Valleys supported more dense populations of psylla and these strains were as resistant to

pesticides as psylla from commercially sprayed orchards. The implications of this discovery will be discussed later.

Unfortunately, although individuals were collected from the OSU Entomology Farm psylla population during the second summer of sampling (1983), their numbers were not sufficient for testing. Consequently, baselines of susceptibility using this strain were determined only for azinphosmethyl and endosulfan, which were the two compounds tested in 1982. The least susceptible strain as measured herein served as a susceptibility baseline for comparisons among all populations tested for fenvalerate and Perthane.

Levels of susceptibility in pear psylla from western Oregon orchards

Table 4 summarizes lethal response values of psylla populations to azinphosmethyl, endosulfan, fenvalerate and Perthane. Complete data analysis using log-probit for each compound is given in appended tables A1-4.

Comparing LC₅₀ values of psylla populations within and between regions revealed several interesting features of resistance. Firstly, psylla from all orchards showed some level of resistance to azinphosmethyl (12- to 41-fold) and endosulfan (2- to 12-fold) when compared to the OSU Entomology Farm Strain (table 5). Resistance factors for fenvalerate and Perthane were calculated by comparing each strain to the most susceptible strain (McCarty in the case of fenvalerate, Logan in the case of Perthane)(table 6). Generally,

Table 4. Summary of lethal concentration (LC $_{50}$) values for \underline{P} . $\underline{pyricola}$ collected from western Oregon pear orchards.

			COMI	POUNDS	
Region	Orchard	azinphosmethyl (gAI/ℓ)	endosulfan (gAI/Ł)	fenvalerate (mlAI/l)	Perthane (mlAI/l)
Maximum recommended field rate	-	0.45	0.75	0.12	1.13
Willamette Valley	Corvallis Wiley OSU Ent Farm	6.90* 4.52 0.31a	0.56 0.27 0.14a	0.05	
	Botany Farm	5.18	0.38,0.54	0.06,0.07	
Hood River Valley	McCarty Facteau	4.21 6.42	0.63 1.73	0.02 0.03	0.42 1.52*
	Merz Logan	5.47	1.26*	0.05* 0.10	0.41* 0.29
	Tamiyasu	3.60	1.55	0.03	0.61
Rogue River Valley	Rogue Cherry Ln.	6.30* 10.20*	0.66* 0.54*		
	Carpenter Hill Hanley	12.64 6.84	0.71 0.64	0.08 0.03,0.04	1.06* 0.36

^{*} Entries followed by an asterisk have been estimated based on a single diagnostic dosage and an estimated slope.

a LC₅₀ values based on accumulated data collected over several time periods.

Table 5. Comparison of LC50s of psylla populations from western Oregon to azinphosmethyl and endosulfan.

		<u>az</u> inp	hosmethyl	<u>endosulfan</u>	
Region	Orchard	LC ₅₀ (gAI/L)	Resistance Factor	LC ₅₀ (gAI/L)	Resistance Factor
illamette Valley	OSU Ent. Farm	0.31b	-	0.14b	_
_	Corvallis	6.90*	22.3	0.56	4.0
	Wiley	4.52	14.6	0.27	1.9
	Botany Farm	5.18	16.7	0.38,0.54	2.7,3.9
Hood River Valley	McCarty	4.21	13.6	0.63	4.5
•	Facteau	6.42	20.7	1.73	12.4
	Merz Logan	5.47	17.6	1.26*	9.0
	Tamiyasu	3.60	11.6	1.55	11.1
ogue River Valley	Rogue	6.30*	20.3	0.66*	4.7
•	Cherry Lane	10.20*	32.9	0.54*	3.9
	Carpenter Hill	12.64	40.8	0.71	5.1
	Hanley	6.84	22.1	0.64	4.6

 $^{^{\}rm a}$ Entries followed by an asterisk have been estimated based on a single diagnostic dosage and an estimated slope.

 $^{^{\}rm b}$ LC50 estimate based on accumulated data collected over several time periods.

^C Susceptible strain.

Table 6. Comparison of LC₅₀ values for psylla populations from western Oregon to fenvalerate and Perthane.

	:	fenva	lerate	Perthane	
Region	Orchard	LC ₅₀ (mlAI/l)	Resistance Factor	LC50 (mlAI/l)	Resistance Factor
Hood River Valley	McCarty	0.02b	-	0.42	1.4
	Facteau	0.03	1.5	1.52*	5.2
•	Me rz	0.05*	2.5	0.41*	1.4
	Logan	0.10	5.0	0.29	-
	Tamiyasu	0.03	1.5	0.61	2.1
Willamette Valley	Corvallis				
· ·	Wiley OSU Ent. Farm	0.05	2.5		
	Botany Farm	0.065	3.3		
Dan a Di W 33					
Rogue River Valley	Rogue				
	Cherry Lane	0.00	4 0	1 00+	2.7
	Carpenter Hill	0.08	4.0	1.06*	3.7
	Hanley	0.035	1.8	0.36	1.2

Entries followed by an asterisk have been estimated based on a single diagnostic dosage and an estimated slope.

b Susceptible strain.

comparative differences in susceptibility were between 1- and 5-fold for these two compounds.

The relatively high levels of resistance to azinphosmethyl and endosulfan are not surprising. As measured by field efficacy trials, psylla developed resistance to both compounds in the Hood River and Rogue River Valleys almost simultaneously (1965, azinphosmethyl; 1972, endosulfan). In addition, these compounds are effective against other pests and their use in pear orchard pest management has continued since the time resistance appeared in pear psylla.

Differences in susceptibility to fenvalerate were only 1.5- to 5-fold (table 6). This level of tolerance is generally considered to be within the boundaries of experimental variability and indicates that the populations are as yet susceptible. Fenvalerate is still effective against psylla and is used in the field as a dormant spray against overwintering adults.

Unfortunately, data collection on psylla for their response to Perthane was incomplete. Psylla were collected for this test late in the season when populations were beginning to decline. "Resistance" levels were between 1.5- and 5-fold (table 6). Perthane was used extensively in the Hood River Valley where resistance developed, and to a lesser degree in the Rogue River Valley and Willamette Valley, where it remained effective until it was abandoned because of mounting resistance elsewhere. Potentially, Perthane might have shown the widest range of lethal concentration values because of this marked difference in regional use patterns and observed field resistance.

A second observation revealed that resistance was not correlated with insecticide usage in individual orchards in the Hood River and Rogue River Valleys. However, in the Willamette Valley, psylla resistance was better correlated with local insecticide usage patterns.

Figure 3 shows the averaged LC_{50} values for psylla populations from managed and unmanaged orchards in the Hood River and Rogue River Valleys for each compound. It is notable that psylla from unmanaged orchards in these regions commonly had higher values than those from managed trees for each compound (table 4). For example, the Logan and Carpenter Hill orchards were both managed orchards up until the 1983 season when they were left untreated. The Carpenter Hill strain showed the highest level of resistance to azinphosmethyl (12.64 g AI/l) and the Logan strain was the most tolerant of fenvalerate (0.10 ml AI/1). The Facteau strain, residing in unsprayed trees surrounding a residence, had the highest level of resistance to endosulfan (1.73 g AI/1) and was most tolerant of Perthane (1.52 ml AI/1). Conversely, the McCarty and Tamiyasu populations, both of which reside in intensively sprayed orchards, were the least resistant to azinphosmethyl (4.21 and 3.60 g AI/l, respectively). Further, the McCarty strain was the most susceptible to fenvalerate (0.02 ml AI/1). Differences in average LC_{50} values between managed and unmanaged orchard populations were significant (p = 0.1) for azinphosmethyl, but not for endosulfan, fenvalerate and Perthane for these two regions.

It is difficult to explain the tendency of psylla from unmanaged orchards to exhibit higher resistant than those from managed orchards.

A possible explanation for this pattern is that psylla in a sprayed environment experience some degree of sublethal effect from previously sprayed pesticide residues (e.g. reduced levels of active acetylcholinesterase) that lowers their tolerance to a new exposure below that of an unaffected psylla from an unsprayed environment. Although psylla were always collected from sprayed orchards at least two weeks after treatment, some residual, sublethal amounts of compound in the organism may have rendered them more susceptible to a subsequent chemical exposure. This interpretation of the data, if correct, would indicate that resitance values for commercial orchard populations probably are conservative and would be higher if the orchard were left totally untreated before testing occured.

In the Willamette Valley, a different trend in resistance between managed and unmanaged orchard populations was observed. The OSU Entomology Farm population, the only sizable unmanaged population located in this valley, was highly susceptible to azinphosmethyl ($LC_{50} = 0.31$ g AI/1) and endosulfan ($LC_{50} = 0.14$ g AI/1) as compared to the three managed orchard populations sampled from this region (table 4). Possible explanations for this observation will be discussed in an upcoming section.

On a statistical basis, psylla from unsprayed (unmanaged) orchards and commercially sprayed (managed) orchards show similar levels of susceptibility to all compounds when all three regions are analyzed collectively (figure 2). Recall that McCarthy, Merz, Tamiyasu, Corvallis, Wiley, Botany Farm and Hanley were commercially managed orchards, while Facteau, Logan, OSU Entomology Farm, Rouge,

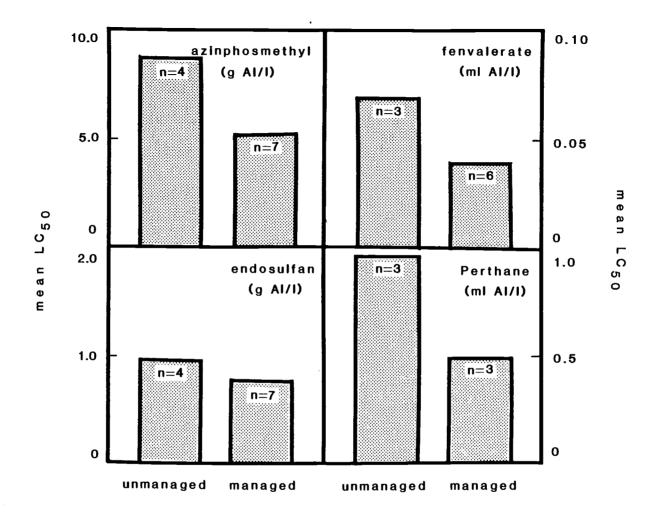


Figure 2. Comparison of mean LC50 values for psylla populations from managed and unmanaged orchards in the Hood River, Rogue River and Willamette Valleys, Oregon (1982-83).

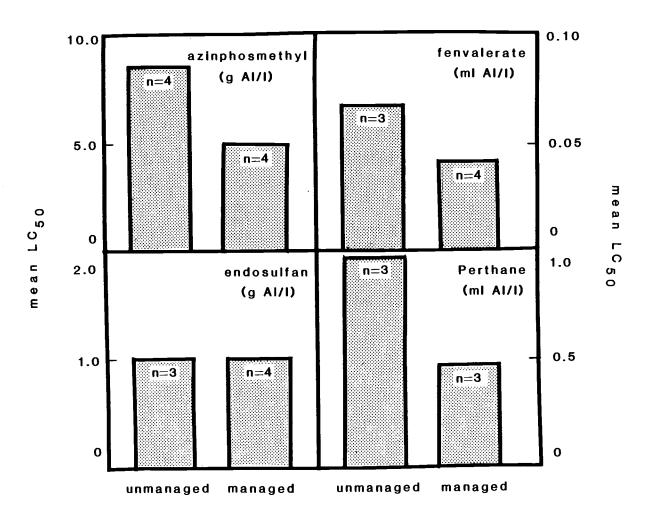


Figure 3. Comparison of mean LC50 values for psylla populations from managed and unmanaged orchards in the Hood River and Rogue River, Oregon (1982-83).

Cherry Lane and Carpenter Hill were unmanaged in the 1983 season (table 2). The mean LC_{50} value for psylla from managed orchards for azinphosmethyl was 5.25 g AI/l (range = 3.60-6.84 g AI/l) and 8.89 gAI/1 (range = 0.31-12.64 g AI/1) from unmanaged orchards (table 4). Lethal concentration values for endosulfan showed a similar trend. Populations from managed orchards had an mean value of 0.77 g AI/l (range = 0.27-1.26 g AI/1), while unmanaged orchard populations averaged 0.91 g AI/1 (range = 0.14-1.73 g AI/1). Lethal concentration values were less variable for fenvalerate and Perthane but showed the same trend, as well. Lethal concentration values for fenvalerate averaged 0.04 ml AI/l (range = 0.02-0.07 ml AI/l) in managed orchard populations, and 0.07 ml AI/l (range = 0.03-0.10 ml AI/l) in unmanaged orchard populations. Lethal concentration values for Perthane averaged $0.48 \, \text{ml} \, \text{AI/I}$ (range = from $0.41-0.61 \, \text{ml} \, \text{AI/I}$) in managed orchards, and 0.98 ml AI/l (range = 0.36-1.52 ml AI/l) in unmanaged orchard populations. None of the differences between managed and unmanaged orchard population means was found to be significantly different (at the p = 0.1 level) using the Wilcoxon rank sum test.

A third trend in psylla resistance was that differences in resistance appear to be significant at the regional level. Table 7 shows that significant differences in mean lethal concentration values exist among regions for azinphosmethyl and endosulfan but not for fenvalerate and Perthane

Pairwise comparisons were performed on mean LC_{50} values between regions for azinphosmethyl and endosulfan. The differences in azinphosmethyl and endosulfan resistance correlated well with

Table 7. Mean LC₅₀ values (\pm S.E.) for regional pear psylla populations. \pm 1/

		COMP	OUNDS	
Region	azinphosmethy1 $\frac{2}{gAI/\ell}$	endosul fan <u>3</u> / (gAI/Ł)	fenvalerate <u>4</u> / (mlAI/l)	Perthane4/ (mlAI/l)
Willamette Valley	4.23 (<u>+</u> 1.40)a	0.36 (<u>+</u> 0.10)c	0.06 (+0.01)	-
Hood River Valley	4.93 (<u>+</u> 0.63)a	1.29 (<u>+</u> 0.24)a	0.05 (<u>+</u> 0.01)	0.65 (+0.22)
Rogue River Valley	9.00 (<u>+</u> 1.89)b	0.64 (<u>+</u> 0.04)b	0.06 (+0.02)	0.71 (+0.22)

 $[\]frac{1}{}$ Means followed by different letters in these vertical columns are significantly different at p<.10 using the Wilcoxon rank sum test.

²/ ANOVA f-ratio = 5.81 is significant at p<.05.

³/ ANOVA f-ratio = 13.58 is significant at p<.01.

^{4/} ANOVA f-ratio is not significant.

differences in regional use patterns (table 4). Azinphosmethyl was used against pear psylla in all three regions from 1960 until 1965, when resistance appeared in the Hood River Valley. Psylla have been exposed to azinphosmethyl since then however, because it was commonly applied as a cover spray for preventative codling moth control. Codling moth pressure is greatest in the Rogue River Valley where growers must apply 3-4 cover sprays per season for this pest. Less intense codling moth pressure in the Hood River and Willamette Valley require only 2-3 sprays per year. The mean lethal concentration value for Rogue River Valley psylla of 9.00 g AI/l was significantly higher (p<.05) than Hood River and Willamette Valley psylla, which had mean values of 4.93 and 4.23 g AI/l, respectively.

Endosulfan has been used most extensively in the Hood River Valley. From 1971-1977, endosulfan was applied in combination with Perthane as a dormant spray for psylla in this region. (Endosulfan alone is not used for psylla control.) This compound has been applied infrequently against sporadic pests (e.g. lygus bug, rust mites) in the Rogue River and Willamette Valleys. The average lethal concentration value of 1.29 g AI/l for psylla from the Hood River Valley was significantly higher (p < .05) than values determined for Rogue River and Willamette Valley psylla, which were 0.64 and 0.36 g AI/l, respectively.

A regional hypothesis

Five factors relating to regional pear culture and pear psylla biology form a foundation on which to develop a regional hypothesis explaining the resistance patterns observed in pear psylla in this study.

- (1) pear psylla develop and reproduce only on pear (Westigard and Zwick 1972).
- (2) psylla populations have the greatest capacity for increase and attain highest densities when feeding on vigorous pear foliage and when natural enemies are absent or low in numbers (Madsen et al. 1963; Gut et al. 1982).
- (3) because of pruning, fertilization and irrigation practices, managed trees have more, suitable foliage for psylla development throughout the season than unmanaged trees.
- (4) managed pear trees far outnumber unmanaged trees in the Hood River and Rogue River Valleys, and to a lesser degree in the Willamette Valley.
- (5) overwintering pear psylla can disperse widely (Westigard and Zwick 1972).

(6) - in late summer and fall, many psylla emigrate from pear orchards seeking protective sites to overwinter (Westigard and Zwick 1972).

It is inferred from factors 1-4 that an overwhelming proportion of the pear psylla in a region develop in managed pear orchards, and therefore experience selective pressure from insecticides. In all three regions, managed orchards may support large populations of psylla. Abandoned trees support moderate to small populations in the Hood River and Rogue River areas and very few or no psylla in the Willamette Valley. In any case, because relatively few psylla are left untreated, the immigration of susceptible or less resistant individuals into orchards probably has a negligible effect in diluting existing population resistance.

Pear psylla leave orchards in the fall and disperse widely (factors 5 and 6). The rapid rate at which psylla colonized orchards during its spread north in the 1950's is evidence for its dispersal capabilities (Westigard and Zwick 1972). In the Hood River and Rogue River Valleys, where commercial pear orchards are nearly continuous, a large amount of population mixing among orchards likely occurs during this dispersal. Among the scattered orchards in the Willamette Valley, dispersal may not result in such extensive population mixing.

The abundance of pear psylla in managed pear orchards and the psylla's dispersal behavior have combined to produce populations possessing regional resistance characteristics in the Hood River and Roque River Valleys. Psylla from abandoned, isolated orchards show

high levels of resistance, suggesting that they originated in managed orchards. This finding, and the closeness with which resistance values accurately reflect historical patterns of insecticide use, support this regional hypothesis for pear psylla resistance.

Resistance in the Willamette Valley, where pear orchards are widely scattered and unsprayed trees are in greater proportion to sprayed trees, did not show a regional trend. Lethal concentration values determined for azinphosmethyl and endosulfan in Willamette Valley orchard populations suggest that localized rather than regional resistance developed in this region. Except for the OSU Entomology Farm orchard, unmanaged orchards in this region supported very few psylla. The unmanaged OSU Entomology Farm, where psylla were as yet susceptible to azinphosmethyl and endosulfan, probably represents a unique situation allowing susceptible psylla to perpetuate. As noted, psylla are generally rare on unmanaged trees because natural enemies are abundant and the amount of vigorous foliage on which psylla thrive is limited. At the OSU Entomology Farm, pear trees are unsprayed but are occasionally pruned and regularly fertilized, which stimulates vigorous leaf growth, and surrounding cherry and apple trees are sprayed, effectively reducing the natural enemy load on nearby trees and also on the pear trees because of insecticide drift. In addition, this orchard is isolated from any commercially sprayed orchards. Hence, a low population has survived without appreciable insecticide pressure and with relatively little exposure to immigration from outside areas.

The slide-dip test as a measure of field resistance

Maximum field rate values presented in the first row of table 4 provide a yardstick to compare lethal concentration dosages determined with the slide-dip with dosages psylla experience in the field. If psylla experience similar exposure to a toxicant during the slide-dip as they do during a field spray, then the lethal concentration values in table 4 reflect approximate levels of resistance in field populations to equivalent rates. In theory, psylla would develop resistance up to some level below the maximum field rate where effective control in the field would be lost and the compound abandoned.

The lethal concentration values for fenvalerate are generally below the maximum field rate, but in some cases (e.g., Logan) close to the point where effective control in the field may be in jeopardy (table 4). Values for Perthane average 0.65 and 0.71 ml AI/l in the Hood River and Rogue River Valleys, respectively (table 4). When compared to the maximum field rate of 1.13 ml AI/l, it seems possible this level of resistance could have reduced the effectivness of this compound before it was abandoned in 1978. In the case of azinphosmethyl, high levels of resistance (8-28X the maximum field rate of 0.45 g AI/l) in psylla may have been attained because of continued pressure from cover sprays for codling moth. Endosulfan resistance shows a similar pattern but to a lesser degree (0.3-2.3X the maximum field rate of 0.75 g AI/l), probably for the same reasons.

Conclusions

There is a high demand and premium placed on unblemished pear fruit, hence, growers attempt to control pear psylla at a "zero tolerance" level. Although recently developed threshold tolerance levels are now being promoted, most growers still routinely spray by the calendar or at first sighting of psylla in the orchard. If we are to reduce the selection pressures leading to resistance, the frequency of insecticide exposures must be reduced and integrated pest management schemes implemented (Croft and Hoyt 1978).

A foundation for developing resistance management schemes is gathering baseline susceptibility information. Upon determination of these baselines, developing resistance can be detected by pesticide susceptibility surveillance procedures. This project describes a slide-dip technique that can be used to track resistance development in pear psylla.

Susceptibility data collected over the 1982 and 1983 seasons revealed several interesting patterns of psylla resistance. Foremost among these is the regional pattern of resistance. Whether future pest management takes the form of simple substitution of a more effective compound, combining compounds with synergists, or manipulating the genetic, biological, ecological and operational factors acting on the pest-crop ecosystem, the proposed regional hypothesis suggests that pear psylla resistance management must take on a regional focus. If psylla populations within a region are mixing during fall migrations, an orchardist will not realize the benefits of resistance management

practices in his own orchard unless all growers in the region have collectively adopted the strategy. This becomes particularly urgent as the number of registered and effective compounds for psylla control diminishes.

Pear psylla management relies on two compounds - amitraz and fenvalerate. Amitraz (BAAM) is the only insecticide currently registered and effective for control of summer forms of psylla (Burts 1983). Its conditional registration is being reviewed in 1984. Several orchardists reported poor control from dormant sprays of fenvalerate last season (1983)(E.C. Burts and R.W. Zwick, pers. comm.). The effectiveness of this compound is sensitive to temperature and this or poor coverage may explain the control failure. Yet, even suggestions of fenvalerate resistance highlight the need to develop an alternative resistance management program. We should proceed forthwith to implement such a program based on intensive monitoring of resistance using the slide-dip technique while seeking to develop improved methods of pesticide resistance surveillance for this species.

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APPENDIX

Table A-1. Lethal concentration values of pear psylla from western Oregon pear orchards to azinphosmethyl (g AI/ ℓ) \underline{b} .

Region	Orchard	LC ₅₀	95% CI	LC95	95% CI	S1 ope	r ²
	OSU Ent Farm	0.31a	0.28-0.33	1.31	1.02-1.69	2.59	.98
Valley	Corvallis	6.90*					
variey	Wiley	4.52	3.47-5.88	103.13	57.63-184.55	1.21	.95
	Botany Farm	5.18	4.61-5.81	23.16	20.14-26.65	2.53	.81
Hood River	McCarty	4.21	3.13-5.66	>1000			
Valley	Facteau	6.42	3.57-11.55	50.30	21.49-118.91	1.84	.54
, · -J	Merz	5.47	4.27-6.98	96.58	61.73-151.12	1.32	.99
	Logan	_					.57
	Tamiyasu	3.60	3.19-4.07	13.76	12.01-15.76	2.82	.88
Rogue River	Rogue	6.30*					
Valley	Cherry Ln. Carpenter	10.20*					
	Hill	12.64	11.68-13.69	67.80	59.39-77.49	2.26	.88
	Hanley	6.84	6.35-7.36	38.01	34.32-42.16	2.21	.89

Entries followed by an astrisk have been estimated based on a single diagnostic dosage and an assumed slope.

a LC50 estimate based on data accumulated over several time periods.

b Determined by log-probit analysis.

Table A-2. Lethal concentration values of pear psylla from western Oregon pear orchards to endosulfan $(gAI/\ell)^b$.

Region	Orchard	LC ₅₀	95% CI	LC95	95% CI	S1 ope	r ²
Willamette	OSU Ent Fam	0.14a	0.14-0.16	1.86	1.37-2.54	1.49	.65
Valley	Corvallis	0.56	0.39-0.82	2.86	0.91-11.81	2.34	.89
	Wiley	0.27*	0.26-0.28	1.06	0.94-1.18	2.78	.95
	Botany Farm	0.38	0.37-0.39	0.97	0.90-1.05	4.00	.95
	y	0.54	0.49-0.61	2.46	1.69-3.54	2.50	.78
Hood River	McCarty	0.63	0.59-0.67	4.24	2.84-6.68	1.98	. 87
Val ley	Facteau Merz Logan	1.73 1.26*	1.52-1.95	6.17	3.88-9.83	2.97	.91
	Tamiyasu	1.55	1.38-1.75	6.53	3.79-11.26	2.64	.88
Rogue River	Rogue	0.66*					
Val ley	Cherry Ln. Carpenter	0.54*					
	Hill	0.71	0.61-0.82	2.71	1.49-4.90	2.82	.99
	Hanley	0.67	0.61-0.66	1.99	1.80-2.20	3.32	.9 9

 $^{^{\}star}$ Entries followed by an asterisk have been estimated based on a single diagnostic dosage and assumed slope.

 $^{^{\}rm a}$ LC50 estimate based on data accumulated over several time periods.

 $^{^{\}rm b}$ Determined by log-probit analysis.

Table A-3. Lethal concentration values of pear psylla from western Oregon pear orchards to fenvalerate ($m\ell AI/\ell$)a.

Orchard	LC ₅₀	95% CI	LC95	95% CI	S1 ope	r ²
OSU Ent Farm		•				
Corvallis						
	0.05	0.05-0.06	0.32	0.27-0.38	10.16	.98
•	0.06	0.05-0.08	0.22	0.15-0.32	3.00	.88
2004.9	0.07	0.06-0.08	0.43	0.34-0.55	2.07	.95
McCarty	0.02	0.01-0.02	0.05	0.05-0.06	3.04	.92
Facteau	0.03	0.02-0.03	0.16	0.15-0.18	2.04	.98
Merz	0.05*					
Logan	0.10	0.07-0.14	22.46	-	0.70	.89
Tamiyasu	0.03	0.02-0.03	0.07	0.06-0.08	4.28	.85
Roque						
•		•				
•	0.08	0.04-0.05	0.17	0.15-0.19		
•			0.09	0.08-0.10	3.08	.98
Hanley	0.04	0.03-0.04	0.14	0.14-0.15	3.42	.99
	OSU Ent Farm Corvallis Wiley Botany Farm McCarty Facteau Merz Logan Tamiyasu Rogue Cherry Ln. Carpenter Hill	OSU Ent Farm Corvallis Wiley 0.05 Botany Farm 0.06 0.07 McCarty 0.02 Facteau 0.03 Merz 0.05* Logan 0.10 Tamiyasu 0.03 Rogue Cherry Ln. Carpenter 0.08 Hill 0.03	OSU Ent Farm Corvallis Wiley 0.05 0.05-0.06 Botany Farm 0.06 0.05-0.08 0.07 0.06-0.08 McCarty 0.02 0.01-0.02 Facteau 0.03 0.02-0.03 Merz 0.05* Logan 0.10 0.07-0.14 Tamiyasu 0.03 0.02-0.03 Rogue Cherry Ln. Carpenter 0.08 0.04-0.05 Hill 0.03 0.02-0.03	OSU Ent Farm Corvallis Wiley 0.05 0.05-0.06 0.32 Botany Farm 0.06 0.05-0.08 0.22 0.07 0.06-0.08 0.43 McCarty 0.02 0.01-0.02 0.05 Facteau 0.03 0.02-0.03 0.16 Merz 0.05* Logan 0.10 0.07-0.14 22.46 Tamiyasu 0.03 0.02-0.03 0.07 Rogue Cherry Ln. Carpenter 0.08 0.04-0.05 0.17 Hill 0.03 0.02-0.03 0.09	OSU Ent Farm Corvallis Wiley 0.05 0.05-0.06 0.32 0.27-0.38 Botany Farm 0.06 0.05-0.08 0.22 0.15-0.32 0.07 0.06-0.08 0.43 0.34-0.55 McCarty 0.02 0.01-0.02 0.05 0.05-0.06 Facteau 0.03 0.02-0.03 0.16 0.15-0.18 Merz 0.05* Logan 0.10 0.07-0.14 22.46 - Tamiyasu 0.03 0.02-0.03 0.07 0.06-0.08 Rogue Cherry Ln. Carpenter 0.08 0.04-0.05 0.17 0.15-0.19 Hill 0.03 0.02-0.03 0.09 0.08-0.10	OSU Ent Farm Corvallis Wiley 0.05 0.05-0.06 0.32 0.27-0.38 10.16 Botany Farm 0.06 0.05-0.08 0.22 0.15-0.32 3.00 0.07 0.06-0.08 0.43 0.34-0.55 2.07 McCarty 0.02 0.01-0.02 0.05 0.05-0.06 3.04 Facteau 0.03 0.02-0.03 0.16 0.15-0.18 2.04 Merz 0.05* Logan 0.10 0.07-0.14 22.46 - 0.70 Tamiyasu 0.03 0.02-0.03 0.07 0.06-0.08 4.28 Rogue Cherry Ln. Carpenter 0.08 0.04-0.05 0.17 0.15-0.19 Hill 0.03 0.02-0.03 0.09 0.08-0.10 3.08

^{*} Entries followed by an asterisk has been estimated based on a single diagnostic dosage and assumed slope.

a Determined by log-probit analysis.

Table A-4. Lethal concentration values of pear psylla from western Oregon pear orchards to Perthane $(m\ell AI/\ell)^a$.

Region	Orchard	LC ₅₀	95% CI	LC95	95% CI	S1 ope	r ²
Willamette Valley	OSU Ent Famm Corvallis Wiley Botany Farm						
Hood River Valley	McCarty Facteau Merz	0.42 1.52* 0.41*	0.39-0.44	8.24	3.81-17.81	1.27	.88
	Logan Tamiyasu	0.29	0.29-0.30 0.59-0.63	1.94 7.55	1.76-2.14 4.62-12.30	2.59	.89
Rogue River	Rogue						
Valley	Cherry Ln. Carpenter	1.06*					
	Hill Hanley	0.36	0.30-0.42	49.34	-		.85

^{*} Entries followed by an asterisk have been estimated based on a single diagnostic dosage and assumed slope.

a Determined by log-probit analysis.