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The Persea Mite Invasion into California: History, Biology, Management, and Current Status

The persea mite, *Oligonychus perseae* Tuttle, Baker, and Abbatiello (Acari: Tetranychidae) (Fig. 1), was first discovered in southern California in 1990 when unusual spot-like damage symptoms (Fig. 2) were noticed on leaves of backyard avocados growing in and around Coronado and La Jolla in San Diego County (Bender 1993).

Figure 1. Silk nest removed to expose persea mites.

Figure 2. Persea mite feeding damage.

This mite was recognized as new pest for California avocados but was initially mis-identified as *Oligonychus peruvianus* (McGregor) (Aponte and McMurtry 1997a). Persea mite was originally described from specimens collected in 1975 from infested leaves on avocado plants that were intercepted from Mexico at an El Paso Texas border security facility (Aponte and McMurtry 1997a, Hoddle 1998) and described in 1976 (Tuttle et al. 1976). This mite is native to México and has a wide distribution, being readily found on avocados in the states of Baja California, Chiapas, Guanajuato, México, Michoacán, Morelos, Oaxaca, and Puebla. After its establishment in San Diego in 1990, persea mite was found in Ventura, Santa Barbara, and San Luis Obispo Counties in 1993, 1994, and 1996, respectively (Faber 1997). Persea mite joined a resident leaf feeding guild of mites infesting avocado leaves that included *Oligonychus punicae* (Hirst) (avocado brown mite) and *Eotetranychus sexmaculatus* (Riley) (six spotted mite), which had been successfully suppressed over the years by indigenous natural enemies (Fleschner 1954, Fleschner et al. 1955). Persea mite severely disrupted this equilibrium and has greatly destabilized pest management practices for mites in California avocados. Once persea mite invaded commercial orchards, picking bins with infested avocado leaves or unclean equipment moving between groves likely helped to spread persea mite quickly throughout California's major avocado production areas. There are no reports of this pest having established in San Joaquin Valley avocado orchards. In addition to California, persea mite has successfully invaded Costa Rica (1974), Israel (2001), Spain (2004), and the Canary Islands (2006). Interestingly, this record for Costa Rica is two years before the published description of persea mite (Ochoa et al. 1994). This pest is not known from Guatemala which lies between México and Costa Rica, suggesting that persea mite is not native to Costa Rica but is likely an introduced pest from México.

Biology of Persea Mite

Persea mites are tiny, and in the field they are difficult to see without magnifying equipment such as a hand lens or optivisor. Colonies of mites are found on the undersides of avocado leaves, typically within nests made of fine strands of silk (hence the name "spider" mite for members of the Tetranychidae) that are made along the midrib, major leaf veins, or sometimes in depressions away from veins (Aponte and McMurtry 1997b). Adult and immature mites extrude silk from spe-

Figure 3. "Silvery" persea mite nests on underside of leaf.

cial glands in their palps, appendages found near the mouth. To build a nest, mites walk back and forth anchoring silk strands to the midrib, leaf veins, or trichomes. Nests are constructed with several semi-circular openings to allow mites to enter and leave. When populations within nests are small, mites leave the nest to defecate. As the population increases fecal pellets may be deposited on the "roof", or onto the floor (the leaf) of the nest. Several layers of silk may be laid over each other to build a nest, and layers tend to increase with elevated temperatures. When leaves infested with persea mite are tilted toward the sun, the silk nests provide a whitish or silvery reflection, which imparts the Spanish common name, the crystalline mite, "El ácaro or araña cristalina del aguacate" (Fig. 3.) Nests most likely protect mites from the desiccating effects of low humidity as well as from natural enemies. Depending on temperature, individual female persea mites may make between 6-12 nests each with 2-5 eggs over her life time (Aponte and McMurtry 1997b). It is within these nests that major life history events unfold – feeding, mating, egg laying, molting, and growth.

Mites damage leaves by using piercing-sucking mouthparts (called chelicerae) to extract the contents of cells located within the lower epidermal, spongy, and palisade parenchyma layers (Aponte and McMurtry 1997b). Subsequent feeding within nests causes the characteristic circular necrotic spots (sometimes referred to as "measles") which result from the death of plant cells that have been fed upon. These spots are readily apparent on the upper leaf surface even though the damage oc-

Figure 4. Sun burnt avocado fruit.

curred on the bottom side of the leaf. Feeding damage is initially concentrated along leaf veins because nests are typically made there first, but as populations grow, mites migrate to less damaged areas and colonies form to occupy areas between leaf veins. When leaves are heavily infested, necrotic spots tend to coalesce and leaves may be prematurely shed from trees as a result. Feeding damage can be measured with computer software (Kerguelen and Hoddle 1999a), and once about ~7.5% of lower leaf surface is damaged, trees may begin to prematurely shed leaves (Kerguelen and Hoddle 1999b). Defoliation may open the canopy exposing fruit to direct sunlight which can result in sunburn damage (Fig. 4).

Mating often occurs within nests, and males will wait beside females that are in the quiescent phase of their final developmental stage, the deutonymph. Males will sometimes aggressively compete to be the first to mate with these newly emerged and receptive females. To mate, a male will approach a female from behind and grasp her two posterior legs with his front legs. In this position, copulation may last from 1 minute to 19 minutes (Aponte and McMurtry 1997a). Following mating, egg laying and developmental rates for eggs and immature mites are strongly influenced by temperature, which also affects survivorship rates. Spider mites have four immature stages; egg, larva (first instar nymph), protonymph (second instar nymph), and deutonymph (third instar

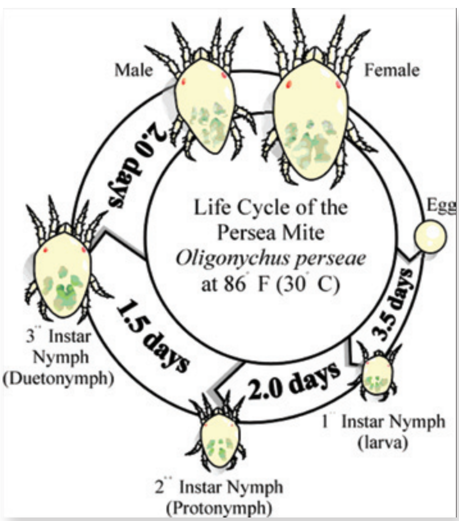


Figure 5. A general life cycle schematic for persea mite developing at a constant 86°F (30°C) in the laboratory. Note that males tend to be smaller than females and are characterized by pointed posteriors. Eggs are large relative to the size of the female and perfectly spherical.

nymph); before reaching the adult stage. Between each of these nymphal stages there is a quiescent period, where the immature mite is inactive as it transforms into the next developmental stage. A general life cycle is shown in (Fig. 5).

Laboratory studies at constant temperatures investigating developmental and reproductive biology have been completed and can be used to estimate the developmental times and reproductive output for persea mite. Developmental rates increase with increasing temperatures. This occurs because mites, like insects, are ectothermic or poikliothermic, and lack a complex physiology that enables them to regulate their body temperature independent of external temperatures. Because of this, temperature-development studies can be completed to determine how many thermal units persea mite needs to accumulate above a critical minimum threshold to develop. Below this threshold it is too cold for mites to develop, but above it, temperatures are warm enough to develop, and mites accumulate "degree-days." Mites and insects need to accumulate a specific number of degree-days to complete their development, therefore, the warmer the temperature, the faster the development. However, mite development doesn't accelerate linearly with increasing temperatures indefinitely. There is an optimal temperature for development, and above this there is an upper lethal temperature at which mites are unable to develop and they die of heat stress. Knowing the degree-days for specific insects or mites allows pest managers and researchers to predict

Table 1. Developmental statistics in days (d) for persea mite reared at four different constant temperatures (i.e., 24 hr per day) in the laboratory (Aponte and McMurtry 1997a).

Temperature °C (°F)	Egg	Larva (quiescent phase)	Protonymph (quiescent phase)	Deutonymph (quiescent phase)	All Stages	Sex ratio (♀: ♂)
15°C (59°F)	11.5 d	6.5 d (3.1 d)	4.1 d (2.9 d)	3.7 d (3.1 d)	35 d	2.4:1
20°C (68°F)	6.9 d	2.0 d (1.1 d)	2.1 d (1.2 d)	2.5 d (1.1 d)	17 d	2.1:1
25°C (77°F)	5.9 d	1.7 d (1.3 d)	1.3 d (1.2 d)	1.6 d (1.1 d)	14 d	1.8:1
30°C (86°F)	3.7 d	1.3 d (0.8 d)	1.0 d (0.5 d)	1.1 d (1.1 d)	9 d	2.1:1

the number of generations a pest will have and what the approximate generation times will be at different times of the year. This information may be useful for developing pesticide application schedules, especially if a biofix point can be identified (e.g., an important calendar date pertaining to pest biology [i.e., first detection of adults on leaves] or perhaps some phenological stage of the host plant). Degree-day calculators are available online that can be set up to utilize weather date from weather stations neighboring avocado orchards (http://www.ipm.ucdavis.edu/WEATHER/index.html). Developmental times for persea mite are presented in Table 1 and reproductive statistics are presented in Table 2 (all data are from Aponte and McMurtry 1997a).

The minimum temperature for development is about 8°C (46°F) and above this minimum temperature, male and female persea mites need to accumulate approximately 166 and 200 degree-days, respectively, to complete development. At a constant 30°C, ~37% of mites fail to reach adulthood, while at 20°C only 2% die prematurely. This suggests that at constant temperatures of 30°C or higher, mites start to experience heat stress which increases mortality.

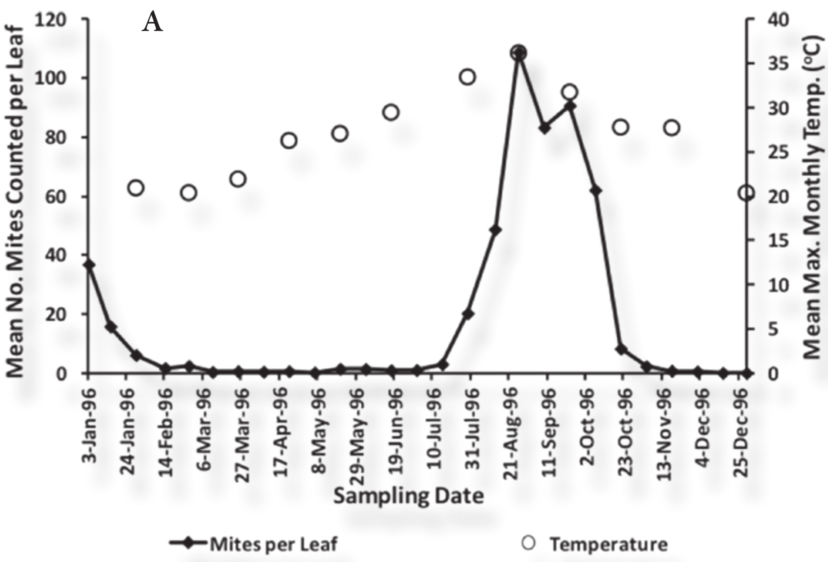
Table 2. Reproductive and longevity statistics for persea mite held at four constant temperatures (i.e., 24 hr per day) in the laboratory (Aponte and McMurtry 1997a).

Temperature °C (°F)	Preoviposition (days)[1]	Oviposition (days)[2]	Total Fecundity[3]	Daily Fecundity[4]	Longevity (days)[5]
15°C (59°F)	6.5	31.1	18	0.6	50
20°C (68°F)	2.6	30.3	37	1.2	40
25°C (77°F)	1.9	21.3	46	1.6	27
30°C (86°F)	1.4	11.7	21	1.8	15

[1]Preovipositon is the mean number of days before mature females commence egg laying.
[2]Oviposition is the mean number of days that females lay eggs over before egg laying ceases. Females may continue to live for 2-10 days after they finish laying eggs.
[3]Total fecundity is the average number of eggs laid by a female over her lifetime.
[4]Daily fecundity is the average number of eggs laid per female per day.
[5]Longevity is the mean number of days a female mite lives for before dying of old age.

Persea Mite Phenology and Cultivar Susceptibility to Persea Mite Infestation

Persea mites tend to be present year round in avocado orchards, but their populations can be very low and hard to detect during the winter and early spring. Populations begin to build in response to increasing temperatures, becoming obvious around July, then peak around August – September, and decline to almost undetectable densities again in November. (Fig. 6) shows persea mite population trends for two sites monitored in Escondido and Pala Mesa in San Diego County.



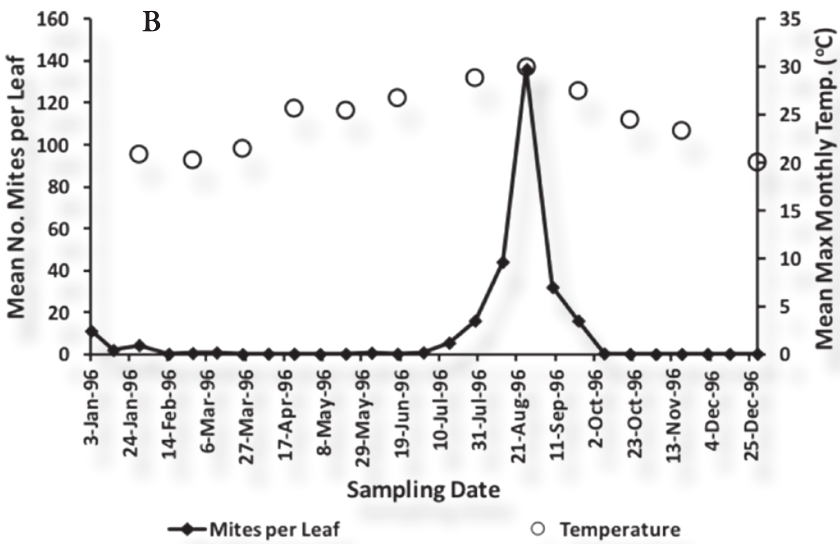


Figure 6. Persea mite population trends on Hass avocados and mean monthly maximum temperatures (ºC) for (A) Escondido and (B) Pala Mesa in San Diego County.

As populations increase, it is common for close to 100% of leaves to become infested with persea mites (Fig. 7A). Population declines, tend to be associated with declining food quality, and not natural enemy activity (see the section below on biological control for more on this). It is also over this time, as populations are peaking, that persea mites commence "ballooning" to disperse from resource poor leaves (Fig. 7B). To balloon, mites exude silk strands, the wind catches these strands, which carries mites away from the substrate upon which they were perched, often from the margins or tips of leaves.

Avocado cultivars vary in their susceptibility to persea mite infestation. This is reflected in two ways: (1) percentage leaf damage caused by feeding, and (2) how well persea mites can breed on the leaves of different types of avocados at different times of the year. A survey of seven avocado varieties using leaf damage measurements revealed three distinct classes: (A) susceptible to persea mite (Hass and Gwen avocados; (B) intermediate (Esther and Pinkerton), and (C) resistant (Lamb Hass, Fuerte, and Reed) (Kerguelen and Hoddle 2000). Life table stud-

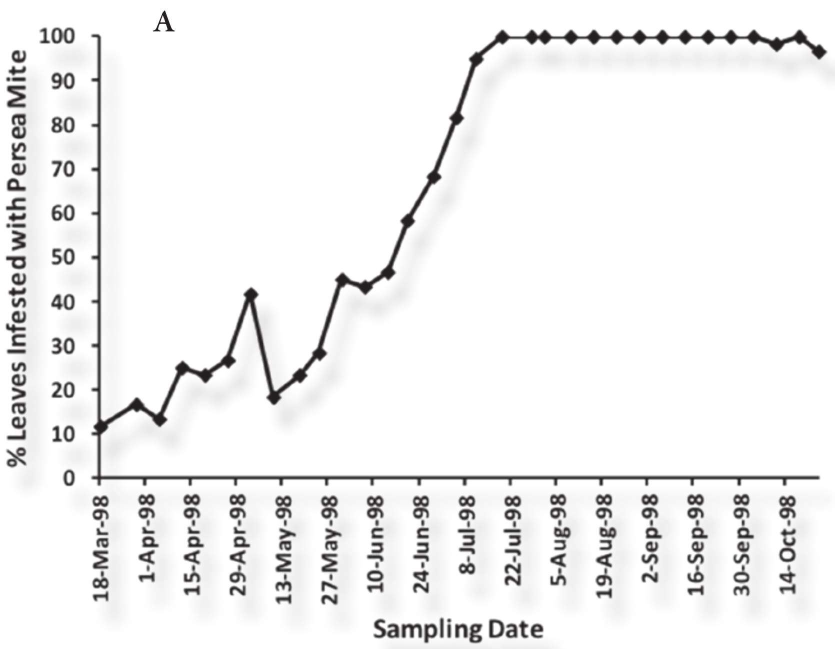


Figure 7. Percentage of leaves infested with persea mite in an orchard in Camarillo Ventura County in 1998 (A) and the mean density of mites per leaf and the ballooning activity of mites (monitored by captures on white sticky cards placed in the orchard)

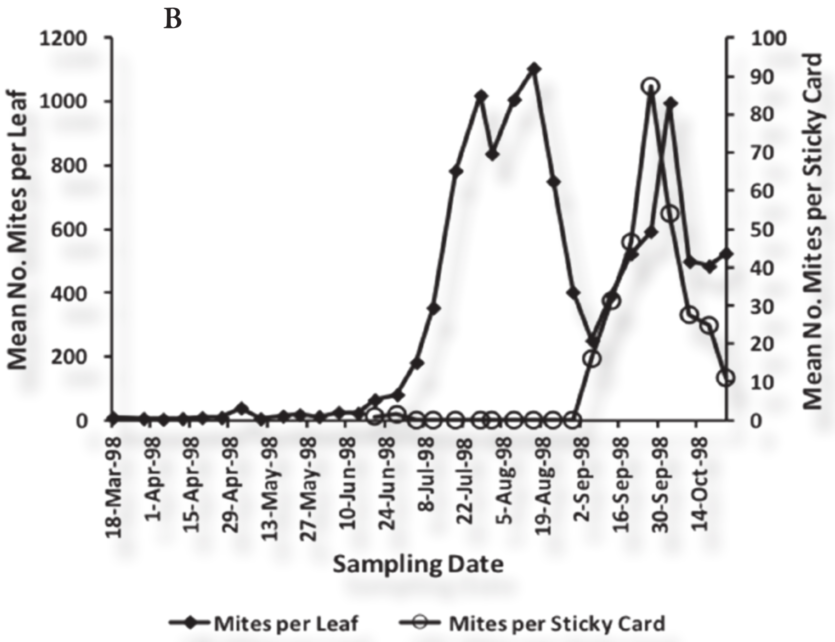


Figure 7. (B) in the same orchard and year.

ies in the laboratory quantifying life history traits that drive population growth indicated that leaf quality at certain times of the year greatly favored persea mite reproduction. This was most pronounced for Hass when mites were reared on leaves in July, the time of year when pest populations start to build in the field (Kerguelen and Hoddle 2000). It is possible that seasonal changes in the nutritional quality of leaves may be a major factor determining the susceptibility of avocado cultivars to persea mite. This hypothesis should be investigated further as it could be an important trait for breeding new avocado varieties that are resistant to leaf feeding pests like persea mite. Other factors may also be involved in resistance to persea mite, such as trichome densities on the undersides of leaves. These are noticeably greater on Lamb Hass when compared to Hass, and trichomes may deter pest feeding or enhance the activity of generalist natural enemies (Kerguelen and Hoddle 2000). It is worth noting that the persea mite problem, especially on Hass avocados, may be almost entirely due the growing of a cultivar that is either unusually sensitive to persea mite feeding, or is an extremely good, perhaps the best, type of avocado for this pest to breed on. If this is true, then management programs for this pest that relying either on pesticides or biological control are going to be constantly disadvantaged because there is little or no host plant resistance to support control measures.

Biological and Cultural Control of Persea Mite

Inoculative and Augmentative Biological Control

Considerable effort has been expended in evaluating the efficacy of commercially available natural enemies, in particular, predatory mites (Acari: Phytoseiidae) for use in inoculative or augmentative biological control programs. Here, we use "inoculative" to mean releases of natural enemies into orchards where they did not previously exist or were present at extremely low levels. These releases establish populations which build in response to the target pest, then die off after the pest population has collapsed. Augmentative biological control is the release of natural enemies to "augment" or bolster existing populations of the same species already present in orchards. These augmentative releases boost the numbers or increase the distribution of these resident natural enemies so they may be able to suppress pest populations.

Initial work focused on six species of commercially available phytoseiid; *Galendromus annectens* (DeLeon), *Galendromus helveolus* (Chant), *Galendromus occidentalis* (Nesbitt), *Galendromus pilosus* (Chant), *Neoseiulus californicus* (McGregor), and *Typhlodromus rickeri* Chant (Hoddle et al. 1999). Releases of 2,000 predators were made at two different times, early and late in the persea mite season, at an experimental Hass avocado orchard in Camarillo in Ventura County. The objective was to determine the effect of prey density on predator establishment rates and population growth, and subsequent control of persea mite. Early releases were made when 25% of sampled leaves were infested with one or more persea mite. No predators established at this time. A second release was made when 75% of leaves were infested with persea mite; here five of the six predators established (*G. pilosus* failed to do so). Densities of *G. helveolus* and *N. californicus* increased after establishment, but failed to control persea mite. In Late releases made when 95% of leaves were infested with persea mite. *Galendromus helveolus* and *N. californicus* were recovered, but failed to show increasing population growth as pest populations were declining when releases were made (Hoddle et al. 1999). It was concluded from these studies that *G. helveolus* and *N. californicus* were the best predators available for persea mite biological control in avocados and should be studied further.

Follow up studies assessed three release strategies with *G. helveolus* and *N. californicus* in Ventura County: (1) three consecutive releases of 2,000 *G. helveolus* per tree, (2) three releases of 2,000 *N. californicus*, and (3) three releases of *G. helveolus* (1,000) and *N. californicus* (1,000) combined. Releases were made when 50%, 75%, and 100% of sampled leaves were infested with persea mite. All three predator release treatments suppressed persea mite populations in comparison the control trees that received no persea mite suppression treatments, and all were more effective than oil sprays (5% NR 415 oil applied twice to individual trees using a motorized backpack sprayer at a rate of ~1.5 liters per tree [when these studies were run this was the industry's preferred pesticide for persea mite suppression]) because resurgence was observed on these trees (Kerguelen and Hoddle 1999b). Resurgence is recognized when pest densities reach even higher densities than they were before the application of pesticides. This may occur because natural enemies are killed by sprays or pesticides increase the reproductive output of females (this is known as hormoligosis). An important conclusion from this study was to conduct future research with *N. californicus* because it was just as effective as *G. helveolus* for controlling persea mite, but it was 33% less expensive to purchase. Also, average leaf damage measurements on trees treated with *N. californicus* were lower (6% of the surface area damaged for *N. californicus* alone and in combination with G. helveolus vs. 10% for *G. helveolus*). Leaves start to drop once 7.5% of the leaf surface is damaged by persea mite feeding (Kerguelen and Hoddle 1999). From a plant protection point of view, in order to minimize premature dropping of summer leaves, control measures need to keep leaf damage caused by persea mite below 8%.

An important question that needed to be answered next was how many, and how often, should *N. californicus* be released onto avocado trees to control persea mite? Studies conducted in a commercial Hass orchard in Orange County evaluated whether one, two, or three releases of 500, 1,000, or 2,000 *N. californicus* or two applications of 5% NR 415 oil applied twice to individual trees using a motorized backpack sprayer at a rate of ~2 liters per tree would be most effective at suppressing persea mite. Predator releases were made when 50% (release one), 75% (release

2), or 95% (release three) of leaves were infested with persea mite. It was concluded that a minimum of 2,000 N. californicus per tree was needed to successfully control persea mite in comparison to the oil treated trees and the control trees that received no treatments. Releasing 1,000 *N. californicus* twice (at 50 and 75% leaf infestation) or 2,000 once (50% leaf infestation) per tree provided control equal to that of the oil treatments and trees receiving more than 2,000 *N. californicus*. There was no added benefit to releasing more than 2,000 predators per tree. We concluded that the economics of this biological control program, even though it was effective, were not cost effective. The cost of the predators alone was 13-14 times more expensive than oil applications (Hoddle et al. 2000). Further, the releases were made by hand pouring predators into paper cups attached to trees, and this simple economic analysis didn't account for the time and labor to hand-distribute predators onto trees. Such an approach for releasing natural enemies in orchards is simply not feasible. Additionally, paper cups require predators to move from a point source (the cup) and quickly disperse all over the canopy searching for persea mites (Hoddle et al. 2000). A potential solution to these two problems could be the use of mechanized applicators, like a backpack sprayer. Such a device could greatly increase the efficacy of predator releases, especially if fewer predators per tree were needed because of better dispersal over the canopy, and releases would be quicker than hand releases.

A modified Stihl SR400 backpack mistblower successfully delivered *N. californicus* mixed with corn grits onto avocado trees when predators were sprayed with a fine mist of water. Predators were delivered up to 4 m (~12 ft) into the canopy and were recovered on sprayed trees 16 days after application (Takano-Lee and Hoddle 2001). However, using hand-releases of predators using paper cups attached to branches resulted in up to five times as many predators being recovered when compared to mistblower applications. This difference was likely caused by the accidental killing of predators because of hitting leaves and branches too hard (the airspeed of the mistblower couldn't be regulated to avoid this problem), or perhaps predators were falling out of the airstream before reaching the tree because of their very small size and lightness. The amount of material blowing through the canopy was unlikely the cause of poor mite establishment because experimental trees had full thick canopies; however, "spray-through" could be an issue for trees with thin or open canopies (Takano-Lee and Hoddle 2001). Although the modified mistblower demonstrated the capacity to deliver predator mites to trees, its efficacy was never field tested against hand releases of *N. californicus* or pesticide applications for control of persea mite.

The predatory behaviors of *N. californicus* and *G. helveolus* have also been studied in the laboratory (Takano-Lee and Hoddle 2002a). *Galendromus helveolus* is only able to attack persea mites once it has invaded the nest. It is classified as a nest-invading specialist predator and has long setae on the dorsal surface to hold webbing off the body as it pushes into nests. *Neoseiulus californicus* on the other hand, lacks these long setae and is not a nest invading specialist. However, it is more aggressive than *G. helveolus* and can attack persea mites in three different ways; (1) it will intercept and kill persea mites wandering outside of nests, (2) it will attack prey through the silk walls of the nest, and (3) it will rip open and invade nests and attack the mites hiding inside (Takano-Lee and Hoddle 2002). The nest invasion behavior of *N. californicus* has been confirmed by workers in Spain (Montserrat et al. 2008). Takano-Lee and Hoddle (2002a) also documented that both species predator mites held in cold storage (12°C or 54°F) for two weeks were much less effective at attacking persea mite when compared to “fresh” predators that were used in experiments <48 hr after arrival from the insectary. Therefore it is highly recommended that releases of natural enemies, such as predator mites, be made as soon as possible into avocado orchards after arrival from insectaries.

Natural Biological Control

Natural biological control is pest suppression that results from populations of naturally occurring biological control agents living in orchards that are not manipulated in any significant way. Several species of generalist predator exist naturally in California avocado orchards and these have the capacity to feed on persea mites. These predators include *Euseius hibisci* (Chant) (Acari: Phytoseiidae); stigmatid and anystid mites; six spotted thrips, *Scolothrips sexmaculatus* (Pergande) (Thysanoptera: Thripidae); *Aeolothrips kuwanaii* Moulton (Thysanoptera: Aeolothripidae); and cecidomyiid fly larvae (Diptera: Cecidomyiidae) (Hoddle et al. 1999, Kergeulen and Hoddle 1999b, Yee et al. 2001). Extensive surveys over multiple years and sites have consistently demonstrated that these natural enemies, especially *E. hibisci* which is a very common predatory mite in avocado orchards, fail to respond in a significant density dependent manner such that persea mite population control is achieved. This means that as persea mite populations increase in avocado orchards, these generalist natural enemies either don't respond with increased population growth to an increasing prey base, or the increase in predator population densities is too weak to reduce the pest population. The probable reason why *E. hibisci* does not provide adequate or consistent control of persea mite (or avocado thrips) is because this phytoseiid is a pollen-eating specialist (all *Euseius* species are specialist pollen feeding phytoseiids) and its population growth is consistently greatest at times of the year when avocados are producing pollen and perseae mite populations are very low (Kerguelen and Hoddle 1999b). Additionally, unpublished studies completed by Hoddle (UCR), Phillips (retired), and Faber (latter two are both UC Cooperative Extension advisors in Ventura County) in 1998 assessing the efficacy of releasing lacewing larvae for control of persea mite in Camarillo avocado orchards demonstrated that releases of these voracious predators were ineffective against this pest and couldn't be recommended for use. This result also suggests that naturally occurring lacewing populations probably provide little control of persea mite. It is curious to note that a well known spider mite predator, *Stethorus picipes* Casey (Coleoptera: Coccinellidae), has never been recorded attacking persea mite even though it is considered a very important predator of avocado brown mite, *O. punicae* (Tanigoshi and McMurtry 1977).

Conservation Biological Control

Conservation biological control is an approach that uses habitat modification to enhance the efficacy of resident natural enemy populations. Well known examples of conservation biological control include efforts to provide resources for natural enemies, often food, in the form of flowering plants that provide pollen and nectar, or shelter, such as beetle banks in European fields that provide hiding and overwintering places for predatory carabid beetles. Field and laboratory experiments in Israel (Maoz et al. 2011a) and Spain (González-Fernández et al. 2009) have demonstrated that conservation biological control may increase biological control of persea mite. In these studies, a food resource was provided in orchards, specifically windborne pollen. In Israel, the planting of Rhodes grass, *Chloris gayana* Kunth (Poales: Poaceae), between rows of avocados resulted in increased densities of pollen feeding *Euseius* *scutalis* (Athias-Henriot) and lower densities of persea mite on trees adjacent to this grass. The Israelis also confirmed that releases of *N. californicus* were very effective at reducing persea mite populations (Moaz et al. 2011a). Similarly in Spain, researchers noted that *Euseius stipulatas* populations increased in avocado orchards when trees were producing pollen or when pollen from neighboring olive trees was being released and blowing into orchards. Populations of *E. stipulatus* in avocado orchards appeared to benefit from maize pollen when maize was inter-planted between rows of avocado trees and persea mite populations were lower on trees close to maize (González-Fernández et al. 2009). Curiously, *E. stipulatus* can't enter persea mite nests, but *N. californicus* which was also present in Spanish avocado orchards when experiments were run, can attack this mite inside its nests. It is not clear which predator was actually responsible for biological control of persea mite in these studies, or whether the combined actions of both were needed for pest suppression (González-Fernández et al. 2009).

There are several shortcomings with conservation biological control for persea mite management. One issue pertains to the regular provisioning of pollen to keep predators abundant in orchards for the entire time they are needed. González-Fernández et al. (2009) indicated that maize was not able to do this and artificial applications of stored pollen to orchard trees would likely be necessary. Alternatively, sequential plantings of several different plant species that release pollen at different times might be needed (Irvin and Hoddle, unpublished). Studies by Irvin and Hoddle (unpublished) assessing conservation biological control in organic vineyards in southern California noted significant shortcomings with using flowering plants to enhance natural enemies. These included increased water needed to keep cover crops alive, which in turn lowered the quality of the crop at harvest, and caused the grower to exceed his water allocation during a period of water restrictions because of drought. Cover crops provided resources not only for natural enemies but also other pests and some diseases which spilled over onto grapes. Cover crops also made routine management practices difficult as access to vines was more difficult and additional irrigation equipment had to be set up, monitored for accidental leaks, and maintained. Finally, vertebrate pests, like rabbits, benefited from cover crops and required management because they destroyed these plants. Similar problems could be anticipated should cover crops be used in California avocado orchards for enhancing biological control of persea mite.

Classical Biological Control

Classical biological control, or introduction biological control, is the introduction and establishment of natural enemies from the pest's home range to control populations in an area that has been invaded. One reason that exotic organisms become pests when they establish in a new area is because they escape the control of their co-evolved natural enemies that use them for food. In "enemy-free" space, pest populations can grow and spread rapidly because their population growth is largely unchecked. A classical biological control project against persea mite was re-started in 2012, 22 years after the introduction of persea mite into California (Hoddle and Lara-Artiga, unpublished). Foreign exploration for natural enemies of persea mite has been conducted in major avocado growing regions of México. The biological control agents of most interest are predatory mites, possibly phytoseiids, which could be established in California to provide better levels of control than what is experienced presently. The first step of this project has been completed; predator mites from México have been collected and preserved with locality data. The second step of this project is now underway, i.e. the identification of these predators using molecular and morphological techniques. Once this inventory of Mexican predators is completed, it will be compared to a similarly prepared list for predatory mites already present in California's avocado orchards. Following the completion of these identification studies, the objective will then be to identify common predators associated with persea mites on avocado in México that are missing in California. These predators may be considered for future introduction into California for the classical biological control of persea mite.

Cultural Control

Cultural control results from efforts that attempt to retard pest populations by manipulating management techniques to disadvantage the pest. Examples of cultural control include altering planting and harvesting dates to reduce exposure to pest populations, the use of organic mulches to enhance the activity of soil borne natural enemies, or the deliberate removal and destruction of infestation sources. This later approach, removal of infested plant materials, was evaluated for persea mite control. When leaves defoliate due to mite feeding damage, it had been observed that colonies of mites would drop to the ground on these leaves, and up to 27% of dropped leaves would have live persea mites (Takano-Lee and Hoddle 2002b). It was hypothesized that mites could re-colonize trees by abandoning dropped leaves on the ground and walking up tree trunks or support stakes into the canopy. This idea was investigated experimentally via three different treatments: (1) removal of all fallen leaves under experimental trees, (2) application of tanglefoot barriers to tree trunks, and (3) removal of fallen leaves and application of tanglefoot to tree trunks (Takano-Lee and Hoddle 2002b). Persea mite populations on these trees were compared to control trees that did not receive these cultural management practices. Collected data revealed that these cultural practices had no impact on persea mite populations. One reason for this was likely due, in part, to the ballooning of mites from surrounding untreated trees onto trees that had received the cultural treatments (Takano-Lee and Hoddle 2002b). Even if these cultural controls were effective, the labor and cost of implementing such a program would be infeasible, especially on steep hillsides where mechanical equipment could not be used to clean up dropped leaves infested with persea mites.

Sampling for Persea Mite in Orchards

All management plans rely on sampling strategies to determine the density of pests and their distribution in an orchard before control measures can be recommended. Management plans rely on knowing two basic things: (1) the action threshold, and (2) the economic injury level. The action threshold concept is the idea that treatments need to be initiated once a critical density threshold is reached because pest populations are moving towards densities that will cause economic injury. Once the action threshold has been crossed, the impending losses incurred from pest feeding if no action is taken then outweigh the costs of treatment. As this time, it makes sense to apply a control treatment. When pest densities are below the action threshold, there is no need to initiate treatments because economic losses are not going to occur, and the cost of treatment is not warranted. Consequently, orchards need to be monitored by professionals who can accurately assess persea mite densities on leaves.

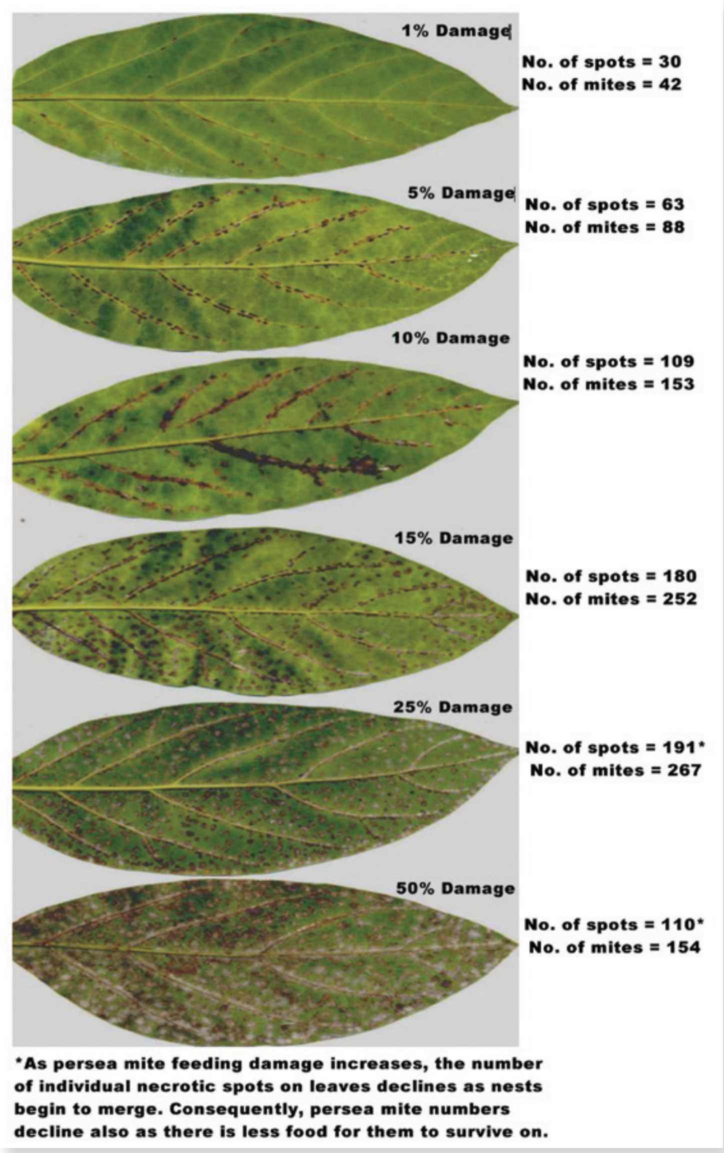


Figure 8. Avocado leaves showing varying levels of persea mite feeding damage. Photos can be used to estimate damage levels in orchards.

Currently, there is no industry standard for monitoring persea mites in California which makes uniform assessments and comparisons of pest severity within and between orchards difficult. Currently, monitoring for persea mite in California involves: (1) walking an orchard, looking at leaves, and using historical experience to gauge pest severity (the most common approach), (2) using photos of damaged leaves to estimate mite numbers in the orchard (Hoddle 2009) (never used [Fig. 8]), or (3) counting mites along the half second leaf vein with a hand lens and using mathematical formulae to estimate average mite densities on an entire leaf (Machlitt 1998) (seldom used [Fig. 9]).

In California, pest control advisors tend to use an action threshold of an average of 100 mites per leaf to initiate treatment recommendations. There is no scientific justification for this action threshold density; it was a number that was chosen during the initial crisis years of the persea mite invasion for determining when pesticides should be applied. Work by Israeli scientists have set an action threshold for managing persea mites when there is an average of 50-100 mites per leaf (Maoz et at. 2011b). Once mites cross this threshold, and average numbers approach 250 mites per leaf, yields can be reduced by 20% (Maoz et al. 2011b). A major shortcoming with this work in Israel was the use of the half second leaf vein method to estimate mite densities (we now realize this method is not accurate although we can certainly understand why it was developed because at the time, there was no good alternative).

To estimate persea mite numbers using the second half leaf vein method, pest monitoring scouts move through a section of an orchard and pick 10 leaves of mixed age at random. Using a 10-14x hand lens they count the number of motile mites that are within the viewing area of the lens along the upper side of the half second vein of each leaf. The half second vein is located on the left side of the upturned leaf and it is the second complete vein that extends from the midrib to the leaf margin. The scout tallies the total number of persea mites on all 10 leaves, divides this by 10 to obtain the average across all sampled leaves. The average is multiplied by 12 (this is the correlation factor used to estimate the total number of persea mites per leaf) and the resulting number is an estimate of the number of mites per leaf (see Machlitt 1998 for more details on this technique [Fig. 9]).

The reliability of this partial count and multiplication method was evaluated in six different avocado orchards in 2009. In comparison to whole leaf counts using a stereomicroscope in the lab, the leaf vein method consistently underestimated mite densities in a range of 8% to 61%, and the severity of the underestimate was related to the density of mites on the leaf. The higher the mite population being estimated on a leaf, the greater the underestimation, which is problematic for accurately estimating an action threshold. Further, counting mites in the

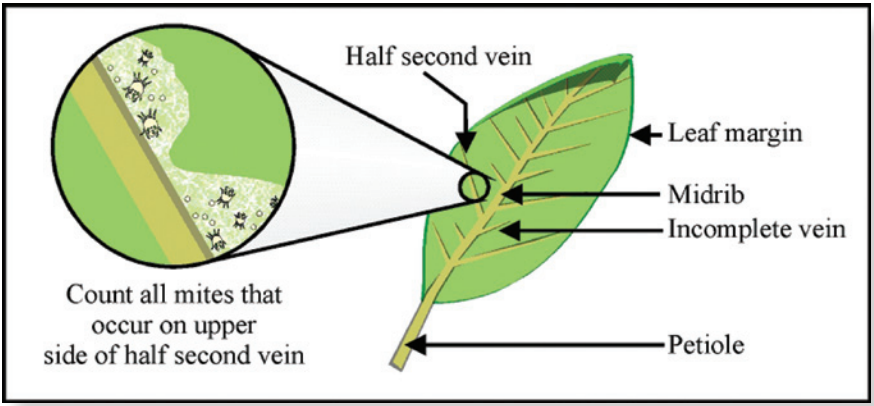


Figure 9. A schematic showing how to use the half second vein method to estimate the numbers of persea mites on a leaf.

field with a hand lens is a tedious task, and inaccurate for determining pest densities because mites are small, and their ability to develop very high population densities can rapidly overwhelm the counter. Additionally, existing sampling methods do not account for the spatial distribution of infested trees in orchards or leaves on trees. Consequently, there is no protocol for selecting trees and leaves in orchards for inspection other than selecting several convenient trees and picking a few leaves at "random".

New sampling strategies are under development for estimating persea mite densities in avocado orchards. The most significant of these is a "no count" method that relies on pest managers keeping a record of the numbers of leaves sampled that are either "infested" with persea mite or are "clean" and lack persea mites. This ratio of infested vs. clean leaves is used to estimate the average number of mites on an avocado leaf (DePalma et al. 2012, Li et al. 2012.) These methods have also developed sampling "rules", which is how to select leaves and trees so that the numbers of mites being counted on leaves are not influenced by the last tree that was sampled. This is important, as trees need to be adequately spaced between samples to ensure that the estimates of mite densities are independent of each other. If trees are too close together (perhaps they are immediate neighbors), the numbers of mites on these neighboring trees will be correlated. By moving about six or more trees away from the last sampled tree and taking the next sample, this spatial correlation or influence is eliminated (DePalma et al. 2012). Work by Ph.D. student

Jesús Lara-Artiga in Hoddle's lab has developed simple sampling methodologies using presence-absence sampling for monitoring perseae mite densities for use by California avocado growers. The application of these techniques is discussed in the following article in this yearbook. One other sampling strategy for persea mite monitoring is the recent development of "co-clustering" analyses to assess persea mite densities in large blocks of trees. Currently, if persea mite densities are considered high enough, entire blocks or orchards are treated with pesticides even though this may not be necessary because damaging pest populations may have been limited to parts of a block or a small section of an orchard. Co-clustering analyses aims to identify smaller or localized regions, often referred to as "hotspots" within an orchard for treatment, and avocados planted in orchards that are in rows and columns may be well suited to this emerging sampling methodology (Zhang et al. 2012).

Managing Persea Mite with Pesticides

As mentioned above, pest control advisors and growers use a variety of sampling methods and decision thresholds to decide whether or not to control persea mite chemically and to determine when to best time a treatment if it is needed. Table 3 provides an update (over the table provided in Morse and Hoddle 2012; note corrections in this table – Zeal is translaminar and Envidor is not) regarding pesticides that are registered in California for use against persea mite. Growers have relied heavily on abamectin for control of both avocado thrips and persea mite for many years but there are now several products in other classes of chemistry that are available for use and are quite effective against persea mite. In order to avoid pesticide resistance in populations of both avocado thrips and persea mite (see the next section), we believe it is critical that we start to learn how to best use Danitol, Envidor, FujiMite (once it is registered), and Zeal against persea mite.

Growers should also consider the economics of applying persea mite controls with helicopter applications in relation to their crop load in a particular year, the expected value of the crop, and anticipated persea mite population levels. Helicopter applications are typically applied using 50-100 gallons of water per acre (gpa) and are expensive (ca. $1.25 per gallon of water exclusive of the cost of the material), regardless of how they are applied. Growers often decide to minimize costs by applying treatments at 50 gpa, which is the minimum allowed for helicopter application on most labels. If persea mite levels are not high and/or trees are not large and/or dense, it makes sense that adequate control can be achieved using 50 gpa by helicopter. However, if the trees are large and/or dense, or one is dealing with a high persea mite population and/or a valuable crop, the grower and PCA may want to consider using more than 50 gallons of water per acre by air despite the higher cost. Table 4 lists the total cost of helicopter pesticide application at 50 vs. 75 vs. 100 gpa. If the pesticide + surfactant (if any is added) cost $15 per acre, then 50 and 75 gpa are 55% and 78% of the cost of 100 gpa; if the cost of the pesticide + surfactant is $45 per acre, then 50 and 75 gpa are 63% and 82% of the cost of 100 gpa. However, what many people do not realize is how dramatically more effective higher gallonage helicopter applications can be.

Working with pest control advisor Matt Hand, in commercial avocado groves in San Diego Co. during fall 2011, we ran a field trial comparing 50 vs. 100 gpa by helicopter using abamectin or Envidor for persea mite control. Four adjacent groves with similar size trees were used for this study. Trees were large and dense (20-30' in height) in all 4 groves and treatments were applied 25 August 2011. Fig. 10 and 11 show the mean number of motile persea mites (all life stages except eggs) per leaf at various dates before and after treatment. Notice that initial persea mite levels were significantly higher in both groves treated using 100 gpa than in the groves treated with the same material at 50 gpa.

Table 4. Costs of persea mite pesticide applications at 50, 75, or 100 gpa assuming application costs by helicopter are $1.25 per gallon of water per acre.

Assumed cost of pesticide + surfactant	Total per acre cost at 50 gpa (material + application)	Total per acre cost at 75 gpa (material + application)	Total per acre cost at 100 gpa (material + application)
$15 per acre	$77.50	$108.75	$140.00
$45 per acre	$107.50	$138.75	$170.00

We calculated cumulative mite days over the duration of the trial in each field (the product of average mite levels times the number of days they were present). Because persea mite impact is cumulative over time, cumulative mite days is a good proxy for the impact of persea mite populations on tree health. The results were quite dramatic. Despite higher

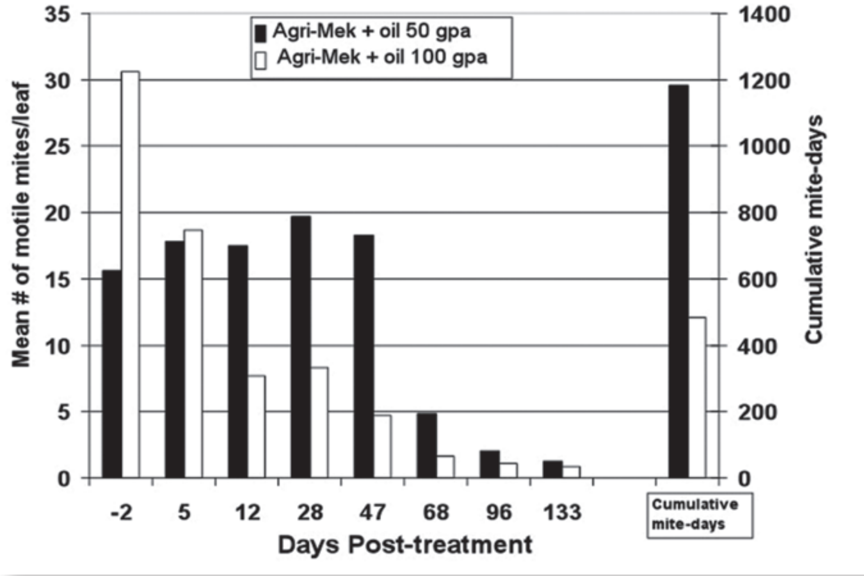


Figure 10. Agri-Mek 0.7 SC at 4.25 fl oz/acre + 4% Omni 6E Oil applied by helicopter at 50 vs. 100 gpa

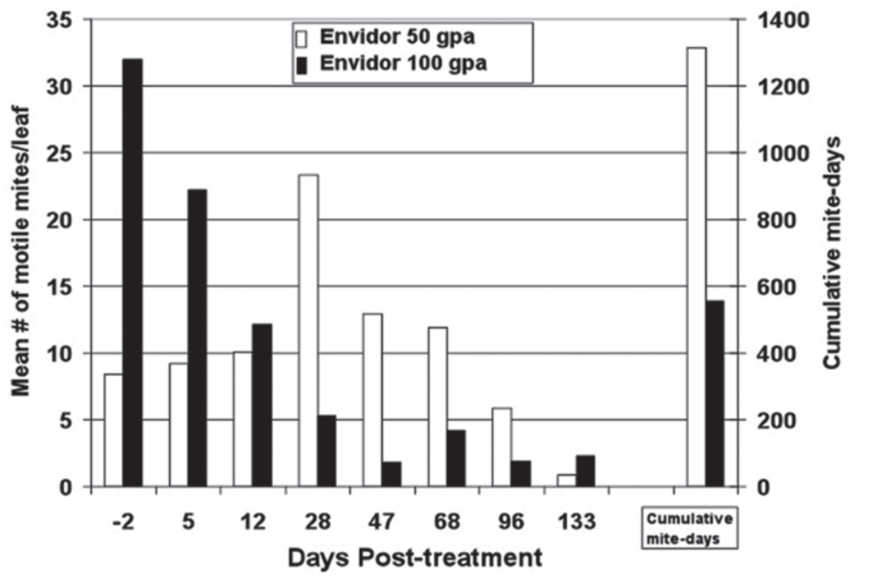


Figure 11. Envidor 2SC at 20 fl oz/acre (no oil) applied by helicopter at 50 vs. 100 gpa

initial levels in the grove treated with abamectin at 100 gpa (vs. 50 gpa), cumulative mite-days in the 100 gpa grove after treatment were 40.8%

of the level in the 50 gpa abamectin grove (Fig. 10). Comparative results in the two groves treated with Envidor were remarkably similar to results with abamectin and highlight the similar efficacy of these two pesticides. Despite much higher mite levels in the 100 gpa Envidor grove, cumulative mite-days in the 100 gpa block were 42.3% of the levels in the 50 gpa block (Fig. 11). Our conclusion is that if a grower has large and/or dense trees and a valuable crop and/or high persea mite levels and they are going to treat by helicopter, the grower should consider seriously applying the treatment higher using higher per acre gallonage (e.g., 75 - 100 gpa).

Managing Pesticide Resistance in Persea Mite

The article on avocado thrips by Morse and Hoddle (2012 in the 2011 CAS Yearbook) discusses in detail our concerns about how heavily the avocado industry in California has been relying on abamectin for **BOTH** persea mite and avocado thrips control ever since this material was made available for use in 1999 (i.e., over the past 14 years). Growers and PCAs are comfortable with the use of abamectin and how it performs and because generic formulations have driven the price down, it is considerably less expensive than the alternatives registered for persea mite control (Table 3). We strongly suggest that growers and PCAs start to use these alternatives and learn the strengths and weaknesses of each product. We also suggest that abamectin should not be used more often than once every 2-3 years in a particular grove (persea mite and avocado thrips treatments combined; no more than once every 3 years would be best). If California avocado producers continue to overuse abamectin, this material will be lost to resistance. Growers would then start to use the more popular of one of the other products, putting pressure on populations of persea mite and avocado thrips to develop resistance to that product. If such a trend were to continue, we could progressively lose one effective class of chemistry after another. Generally when resistance develops to a pesticide in a particular class of chemistry, the other products in that class are ineffective against the resistant pest. Because it takes a considerable period of time to register new products for use on avocados, we need to be careful of the products we have, i.e., we need to rotate between classes of chemistry and not overuse any one class of chemistry.

Table 3. Pesticides registered or nearing registration on avocados useful in control of persea mite that can be used in a proactive resistance management approach. Products are listed in the order they were registered on California avocados. This table is only for reference; make sure to check a current label to ensure that any planned application is consistent with a current label (e.g., language on different abamectin labels may vary; labels evolve over time). This table is based on the situation as of 7-5-13. Consult http://www.ipm.ucdavis.edu/ for Avocado Pest Management Guidelines kept current on the Statewide IPM Program website.

Trade name (formulation)	Common name	Class of chemistry[a]	Mode of action[a]	REI (hrs)[b]	PHI (days)	Air application language	Japanese MRL vs. U.S. tolerance	Notes
Many oils, e.g., PHT 440, Omni 6E, Leaf Life Gavicide Green 415, etc.	Narrow-range 415 or 440 oil	Paraffinic oils (Narrow range oils) – act via smothering / barrier effects	--	See label	See label	Check the label for specifics	Not applicable	Oil is an option for persea mite control (also used in some cases by ground against avocado thrips) either by ground (e.g., 100 gpa LV spray), application via hand-gun (100-500 gpa), or by air (100 gpa). OMRI approved oils are an organic option.
Envidor (2 lbs ai/gal)	spirodiclofen	Inhibitor of acetyl CoA carboxylase	23	12	2	18-20 fl oz/a in min 50 gpa	2.0 (U.S. 1.0)	Contact material (not translaminar or systemic). Limited evidence suggests addition of oil with ground treatments may reduce efficacy; this seems unlikely with air application given the relatively low gallonage used by air (so field experience will be needed to resolve this).
Zeal (72%)	etoxazole	Mite growth inhibitors	10B	12	1	2-3 oz/a in 20 gpa or more	Default 0.01 (U.S. 0.2)[c]	Maximum of 1 application per year. Material shows some translaminar activity and addition of a surfactant or oil (more work needed) may improve efficacy (stickers appear to limit translaminar activity and might be avoided).
Danitol (2.4 lbs ai/gal)[b]	fenpropathrin	Pyrethroids	3A	7 days[b]	1	16-21.3 fl oz/a in 50-100 gpa	2.0 (U.S. 1.0)	Contact material (not translaminar or systemic). Know of no evidence oil improves efficacy of fenpropathrin. We suggest limiting Danitol use to once every 3 years to reduce the potential for resistance evolution.
FujiMite 5EC (0.4 lb ai/gal)[f]	fenpyroximate	Mitochondrial complex I electron transport inhibitors	21A	12	1	2 pts/a in min 50 gpa	Default 0.02 (U.S. 0.15)[c]	Contact material (not translaminar or systemic). Know of no evidence oil improves the efficacy of fenpyroximate.

a Mode of action and class of chemistry based on IRAC (Insecticide Resistance Action Committee) classification; see http://www.irac-online.org/. See text for details on resistance management strategies.

b Restricted entry interval (REI) is the number of hours from treatment until the treated area can be entered without protective clothing. In some cases, the REI exceeds the PHI. The longer of the two intervals is the minimum time that must elapse before harvest. The Danitol PHI on avocado is 1 day but because of past worker exposure concerns, it is suggested that no more than 1% oil be added to Danitol applications on avocado and within 7 days of application, workers who re-enter for the purpose of harvest should wear coveralls, chemical resistant gloves, socks plus shoes, face protection, and protective eyewear.

c Based on the current MRL in Japan and because that is much less than the U.S. tolerance, the label PHI does not at all ensure MRL export compliance (i.e. contact that company's sales representative for suggestions regarding a safe PHI if the fruit is likely to be exported).

Persea mites do not disperse naturally over large distances. Thus, growers will largely determine their own fate regarding persea mite and avocado thrips pesticide resistance because resistant pest populations will persist in orchards and will not be diluted by the arrival of susceptible individuals from elsewhere. If growers have been using abamectin quite a bit over the years in a particular block for avocado thrips and/or persea mite control, and continue to use this product heavily, it is only a matter of time before abamectin resistance appears. A large part of the cost of a helicopter persea mite application is the application itself. Despite the somewhat higher cost of the alternatives to abamectin, we strongly suggest that growers start using Danitol, Envidor, FujiMite (once it is registered), and/or Zeal so as to reduce the exposure of both avocado thrips and persea mite to abamectin. We'd like to iterate this important point a final time, abamectin should not be used more than once every 2-3 years (3 years would be best).

Conclusions

The persea mite invasion into California in 1990 is an example of a much bigger threat facing the California avocado industry, i.e., invasive pest species. In particular, specialist pests that originate from the home range of the avocado, Mexico and Central and South America, are likely to cause severe problems for the California avocado industry in the future. At the present time, the industry is fortunate that its major pests are primarily leaf feeders, which typically cause modest levels of damage to fruit if managed correctly (this is especially true for avocado thrips, *Scirtothrips perseae* Nakahara [Thysanoptera: Thripidae]). The arrival of persea mite was the first major invasive pest crisis for California avocado growers to manage, and it has taken almost 15-20 years of research in the field and laboratory to identify management strategies that work, and to demonstrate conclusively those that don't and should not be used by growers. Yet despite these industry supported efforts on persea mite, there are still notable shortcomings in our knowledge of how to best manage persea mite. For example, California has not developed an action threshold or economic injury level for this pest. We don't know if the currently adopted 100 mite per leaf action threshold is correct, it is possible that it is not, and the action threshold could be lower. Additionally, we don't know what the economic injury level is. We are unable to state what yield reductions result as a consequence of varying pest densities on trees over time. We have assumed that there must be some cost to Hass avocados from defoliation and heavy disfigurement of leaves that are retained by trees, but what exactly this cost is we are unable to say. Growers have concluded that the persea mite problem is no longer an issue for California because unsprayed trees now seem better able to tolerate high density persea mite populations. Is this true, or are growers so used to seeing damaged leaves on trees and dropped leaves on the orchard floor that it is no longer considered unusual? We have very few data to support this assumption. Additionally, we have few data to support the idea that heat waves, that is temperatures > 100°F for several consecutive days, are responsible for the sometimes reported abrupt crashes in high density persea mite populations. Finally, the classical biological control program for persea mite is still in its infancy, and its completion is subject to the vagaries of annual funding cycles. We think that it is fair to state that there is still a lot of important work to do on persea mite despite it being a pest of California avocados for the past 22 years.

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