SELECTIVE PESTICIDES AND BIOLOGICAL CONTROL IN WALNUT PEST MANAGEMENT

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ABSTRACT

The walnut aphid, *Chromaphis juglandicola* is a pest known in California for more than 100 years. The introduction of the parasitic wasp *Trioxys pallidus* from Iran in 1969 led to a dramatic success in the biological control of walnut aphid populations in California, and has provided sustained control of this devastating pest for the past 39 years. However, in recent years we have seen renewed outbreaks of walnut aphids in some orchards, and we discovered a new white color morph of *C. juglandicola* for the first time in 2003. This white morph of *C. juglandicola* a slight advantage over the normal yellow color morph through early reproduction, but otherwise its significance in terms of pest management remains largely unknown. Accompanying these recent changes with respect to walnut aphid has been a shift away from the traditional broad spectrum organophosphate insecticides for the control of primary pests such as codling moth and walnut husk fly to newer reduced risk products. As a result, the impacts of these newer products on the stability of integrated pest management in walnuts has become an important concern. Evidence suggests that the newer reduced risk pesicides are not always compatible with the natural enemies that contribute very effectively to the management of secondary pests, and in some cases can be equally disruptive as the more traditional organophosphates. For many of these secondary pests, the consequences of changing pesticide use can be complex with differential selectivity among the three trophic levels represented.

From bioassays with the walnut aphid, it appears that Guthion is not as effective as Lorsban, particularly for the new white color morph of the aphid. In addition, the pyrethroids Asana and Warrior are both less effective than the organophosphates, particularly with respect to the yellow color morph of the aphid. Of the other pest management products tested this year, both Success and Omite had a significant although weak impact on the walnut aphid, while there was no observable impact of Intrepid. Similarly, from bioassays of the impact of oral and residual exposure of adult parasitoids to these same pesticides, it is clear that *T. pallidus* adults are far less tolerant of Guthion than was the case for adult emergence from treated aphid mummies. While bioassays for adults of the hyperparasitoid *S. aphidivorus* are still ongoing, those for *T. pallidus* adults show that there was minimal tolerance of the organophosphates (Guthion and Lorsban), but greater tolerance of the pyrethroids (Asana and Warrior). In addition, there was surprisingly low tolerance to products such as Success and Omite, although there was minimal impact of Intrepid.

INTRODUCTION

The walnut aphid, *Chromaphis juglandicola* (Kaltenbach) has been known in California for more than 100 years. When present in large number in the spring, aphid feeding reduces tree vigor, nut size and quality. During the summer, it induces a shriveling of the kernels before harvest. Extremely high populations of aphids may lead to leaf drop, exposing nuts to sunburn. The introduction of the parasitic wasp *Trioxys pallidus* (Halliday) from Iran in 1969 led to a dramatic success in the biological control of walnut aphid populations in California, and has provided sustained control of this devastating pest for the past 39 years. However, both growers and PCAs have noted that aphid outbreaks, though not consistent, are of increasing concern in the Central Valley requiring in season spray treatments required in some cases. As pest management practices in walnuts change to both reduce costs and make use of new pesticides, it is important to retain the benefits of the long-term biological control of walnut aphid provided by the introduction of *T. pallidus*.

In addition, we discovered a new white morph of the walnut aphid in 2003 which has since spread to many parts of the eastern side of the Sacramento Valley and seems likely to spread southward into the San Joaquin Valley over the next few years. Our recent research indicates that this white morph differs from the typical yellow morph in two important ways; it has an early reproductive advantage, and it is less susceptible to attack in its later instars (Mills et al., 2005). These two characteristics indicate that the white morph is more likely to develop outbreak populations than the typical yellow morph, and that the importance of effective aphid management is likely to become of greater importance in the future. In addition, it is possible that the new white morph is less susceptible to insecticides than the yellow morph, and could thus present an even more serious threat to walnut pest management in the future.

Pronounced changes are currently occurring in walnut pest management programs in California. The increasing success of pheromone-based mating disruption for the control of codling moth has the potential to lead to substantial reductions in the use of broad spectrum insecticides, and in response to Food Quality Protection Act-mandated reductions in organophosphate (OP) insecticides use a new range of insecticides have become available. The new “reduced risk” pesticides are replacing the OPs, but their impacts on natural enemies are poorly understood and potentially more disruptive than originally thought. Preliminary observations indicate that we can create unstable management programs with repeated applications of the newer insecticides, as these products substantially disrupt natural enemy populations and lead to outbreaks of secondary pests. To remain competitive, stable IPM programs that effectively conserve natural enemies must be developed as even a small increase in natural enemy induced-mortality significantly lowers pest pressure.

The goal of this project is to screen new pesticides to identify selective products that will enhance the level of biological control of walnut aphid in orchards. Pesticides that are currently or likely to be registered for use in walnuts in the near future will be bioassayed using a standard tiered assay approach to determine their impact on walnut aphid, *T. pallidus*, and its dominant hyperparasitoid *Syrphophagus aphidivorus*.

OBJECTIVES

The general objective is to better understand the impact of new pesticides used for the management of primary pests in walnuts on the walnut aphid system, one of the best examples of an effective biological control in western orchards. The specific objectives are:

1. To compare the susceptibility of white and yellow walnut aphids to walnut pest management products using simple laboratory bioassays.
2. To assess the differential susceptibility of *T. pallidus* and *S. aphidivorus* to walnut pest management products using simple laboratory bioassays.
3. To use a demographic bioassay to test a subset of pest management products for their compatibility with and enhancement of biological control of walnut aphid.

PROCEDURES

Objective 1. To compare the susceptibility of white and yellow walnut aphids to walnut pest management products using simple laboratory bioassays

The most effective way to compare the susceptibility of aphids to a variety of pest management products is to screen them through standard laboratory bioassays. From an earlier comparative study of bioassay techniques it was found that rearing 3rd instars on treated leaf disks provided the most accurate and useful information on susceptibility of tree dwelling aphids to a variety of insecticides. Initial trails using walnut aphids on leaf disks showed that this particular aphid is too active and would not settle on the disks for a long enough period to be assayed in this way. As an alternative, we used whole walnut leaflets with clip cages to encourage settlement of the aphids. Using this approach, walnut leaflets were treated by being dipped for 3 secs into either a full strength solution (100% field rate) or a dilute solution (25% field rate) of each pesticide or into water as a control. To ensure effective coating of the leaf surfaces a surfactant (Agral 90) was added to both the insecticide solutions and the water control at a uniform concentration of 0.25ml/liter. Initial trails showed that this concentration was sufficient to give a uniform coating without having any impact of the survivorship of the walnut aphid.

For each pesticide and aphid color morph a set of 8 treated leaflets were used for each of the three treatments (100%, 25%, control). After drying, each leaflet was inserted into a vial of water with a cotton wool stopper to maintain the turgor of the leaflet, and inoculated with 10 3rd instar aphids of one of the two color morphs. The aphids were then enclosed in a ventilated clip cage prepared from a 3cm diameter diet cup and the leaflet placed in a plastic sandwich box in an incubator at 22°C and 16h daylength. The survival of each color morph of the aphid after 48h was used as the endpoint for the assay and differences between color morphs and treatments were determined by 2-way ANOVA.

Isofemale lines of individual clones of both color morphs of the walnut aphid were collected in 2008 from a single orchard in Durham in the Sacramento Valley and were maintained in a glasshouse at UC Berkeley on potted walnut seedlings. Initial products to be tested this year included those currently registered in walnuts either for codling moth, husk fly or aphids including Guthion (as a historical baseline), Lorsban, Asana, Warrior, Intrepid, Success, and Omite. Subsequently we will include newer and organic products such as Altacor, Delegate, Brigade, and Omni oil.

The resulting survivorship data was analyzed separately for each pesticide using two-way ANOVA with aphid color morph and pesticide treatment as the two factors. Percent survival at

48h was first transformed using arcsine square root. While this transformation was successful in equalizing the variance between treatments it did not always normalize the error distribution.

Objective 2. To assess the differential susceptibility of T. pallidus and S. aphidivorus to walnut pest management products using simple laboratory bioassays

Laboratory assays were used to determine the effect of pest management products on the survivorship of adult *Trioxys pallidus* and *Syrphophagus aphidivorus* exposed orally and to surface residues. The same three treatments for each pesticide, as used in the walnut aphid assays, were used to assay the adult parasitoids (Table 1). Glass vials (8 x 2.5 cm) were filled with pesticide solutions and then drained and dried to provide a consistent residue on the inner surface. Similarly, small droplets of a honey-sugar-agar mixture (10-1-1 ratio) were used as food for the adult parasitoids and treated by dipping in insecticide solutions to gain a surface coating of residue. Three 1-2 day old adult female parasitoids were placed into a treated glass vial, provided with treated food that was changed every 2 days, and held at 22°C and 16h daylength. Ten replicate vials were used for each parasitoid species and each pesticide treatment, and adult longevity was monitored daily.

The average longevity of the adults in each replicate vial was analyzed separately for the two parasitoid species and each pesticide using a one-way ANOVA with pesticide treatment as the factor. The data were first log transformed to equalize the variances and normalize the error distribution.

Objective 3. To use a demographic bioassay to test a subset of pest management products for their compatibility with and enhancement of biological control of walnut aphid.

This component of the project was not addressed in this first year of the project and will be reproted on in the future once the tier I assays have been completed.

RESULTS

Objective 1. To compare the susceptibility of white and yellow walnut aphids to walnut pest management products using simple laboratory bioassays

The 48h survivorship of walnut aphids in the leaflet bioassays are presented in Fig. 1. For the two organophosphate insecticides, Guthion and Lorsban, the standard 100% field rate killed all 3rd instar nymphs in all replicates. The low concentration treatment (25% field rate) was also very effective in the case of Lorsban, but some aphids survived in the case of Guthion. The differences between treatments were significant with each treatment distinct from the others, with the exception of the 25% and 100% rates for Lorsban.

For the two pyrethroid insecticides, Asana and Warrior, there was a significant difference between aphid color morphs as well as between pesticide treatments. In both cases, the yellow aphids showed some tolerance of the 25% field rate while this low concentration killed all the white aphids. Both concentrations of the two pesticides were significantly different from the survivorship in the control treatment.

Perhaps surprisingly, both the insecticide Success and the acaricide Omite also showed a significant although small effect of treatment, even though neither is considered to be an aphid material. The effect was a little stronger for Success than for Omite where the control treatments were significantly different from both the 25% and 100% field rates. However, in the case of Omite, the yellow color morph is significantly more susceptible than the white color morph, with only the 100% field rate being significantly different from the control treatment.

Then finally, as would be expected, there were no significant impacts of the insect growth regulator Intrepid on either color morph of the walnut aphid.

Objective 2. To assess the differential susceptibility of T. pallidus and S. aphidivorus to walnut pest management products using simple laboratory bioassays

The longevity of the adult parasitoids exposed orally and to surface residues of the same range of pesticides is presented in Fig. 2. The bioassays of the hyperparasitoid are still in progress and currently are available only for Lorsban, Success and Intrepid, while those for *T. pallidus* are complete.

For the two organophosphates, Guthion and Lorsban, the longevity of *T. pallidus* was severely reduced even for the 25% field rate concentration, with both concentrations being significantly different from the control. Lorsban also had a very similar influence on the longevity of *S. aphidivorus*, which is typically much longer lived, as shown in the control treatment.

In comparison to the organophosphates, the two pyrethroid insecticides had rather less influence on the longevity of *T. pallidus* adults. In the case of Warrior, the influence was not statistically significant, whereas for Asana the influence was significant but not as marked. This result is consistent with field observations made in 2004 that *T. pallidus* could be seen actively searching walnut foliage treated one week earlier with Warrior.

Consistent with the patterns observed above from the aphid bioassays, both Success and Omite had significant impacts on the longevity of *T. pallidus*, while there was no apparent influence of Intrepid on either *T. pallidus* or *S. aphidivorus*. In this case though, the impact of Omite was as strong as that of Success and equivalent to that of the organophosphates.

DISCUSSION

As the management of primary pests in walnuts moves away from the traditional broad spectrum organophosphate insecticides to newer reduced risk products, the impacts of these newer products on the stability of integrated pest management becomes an important concern. Evidence suggests that the newer reduced risk pesicides are not always compatible with the natural enemies that contribute very effectively to the management of secondary pests, and in some cases can be equally disruptive as the more traditional organophosphates. For many of these secondary pests, the consequences of changing pesticide use can be complex, as there can be differential influences of these products at the different trophic levels (pest, primary natural enemy, secondary natural enemy). This is particularly evident in the case of the walnut aphid, a secondary pest which has been very successfully controlled through the importation of an effective primary parasitoid (*Trioxys pallidus*) from Iran in 1969 which has subsequently established throughout the Central Valley. This primary parasitoid is known to be heavily attacked by indigenous secondary or hyperparasitoids which have the potential to prevent it from providing effective biological control of walnut aphid.

During the 1980s as codling moth pressure on walnut increased, efforts were made to isolate and release *T. pallidus* biotypes resistant to azinphos-methyl (Guthion) the standard spray treatment for codling moth at the time. Apparently some of these resistant parasitoids successfully established in walnut orchards and interbred with native field biotypes to provide some level of tolerance to Guthion in field populations. From our earlier bioassays on the success of parasitoid adult emergence from aphid mummies we were able to show that Guthion had no significant influence on *T. pallidus*, but totally prevented emergence of one of the dominant hyperparasitoids *Syrphophagus aphidivorus*. This suggested that Guthion may be compatible with biological control by favoring the survivorship of *T. pallidus* over its hyperparasitoid within aphid mummies. A differential effect of pesticides on primary and hyperparasitoid emergence from mummies was also seen for Warrior, Asana and Success, indicating the importance of pesticide selectivity at the different trophic levels.

From our bioassays this year, it appears that Guthion is not as effective against walnut aphid as Lorsban, particularly for the new white color morph of the aphid. In addition, the pyrethroids Asana and Warrior are both less effective than the organophosphates against aphids, and in contrast to the case of the organophosphates, it is the yellow color morph of the walnut aphid that shows the greater tolerance to these insecticides. Of the other pest management products tested this year, both Success and Omite had a significant although weak impact on the walnut aphid, while there was no observable impact of Intrepid.

From our bioassays of the impact of oral and residual exposure of adult parasitoids to these same pesticides, it is clear that *T. pallidus* adults are far less tolerant of Guthion than was the case for adult emergence from treated aphid mummies. In contrast, the hyperparasitoid *S. syrphophagus* showed equal susceptibility to Guthion for both emergence from mummies and as an adult. While bioassays for the hyperparasitoid adults are still ongoing, those for *T. pallidus* adults show that there was minimal tolerance of the organophosphates (Guthion and Lorsban), but greater tolerance of the pyrethroids (Asana and Warrior). In addition, there was surprisingly low tolerance to products such as Success and Omite, although there was minimal impact of Intrepid.

The patterns arising from this work point to important differences between pest management products in terms of their selectivity with regard to walnut aphid, its primary parasitoid and hyperparasitoids. Further testing will be necessary to complete these initial lab-based bioassays, and subsequent microcosm and field tests will be used to verify these initial observations. However, these initial results highlight the complexity of the effects of these pesticides and indicate the importance of understanding these effects for stabilizing IPM in walnuts.

Table 1. Commercial name, active ingredient and field rate of products tested in Objectives 1 and 2

Product	Active ingredient	Field rate (100%)
Lorsban®	Chlorpyrifos (OP)	16 fl oz/acre
Penncap-M®	Methyl parathion (OP)	6 pt/acre
Guthion®	Azinphos-methyl (OP)	0.75 lb/acre
Warrior®	Cyalothrin lambda (pyrethroid)	2.5 fl oz/acre
Asana®	Esfenvalerate (pyrethroid)	8 fl oz/acre
Success®	Spinosad (spinosyns)	3.2 fl oz/acre
Intrepid®	Methoxyfenozide (IGR)	18 fl oz/acre
Omite®	Propargite (miticide)	10 lb/acre

Fig 1. Percentage survival of 3rd instar walnut aphids after 48 h enclosed in clip cages on walnut leaflets that were dipped in pesticide solutions at 25% and 100% of the field rates (see Table 1) in comparison to a control treatment.

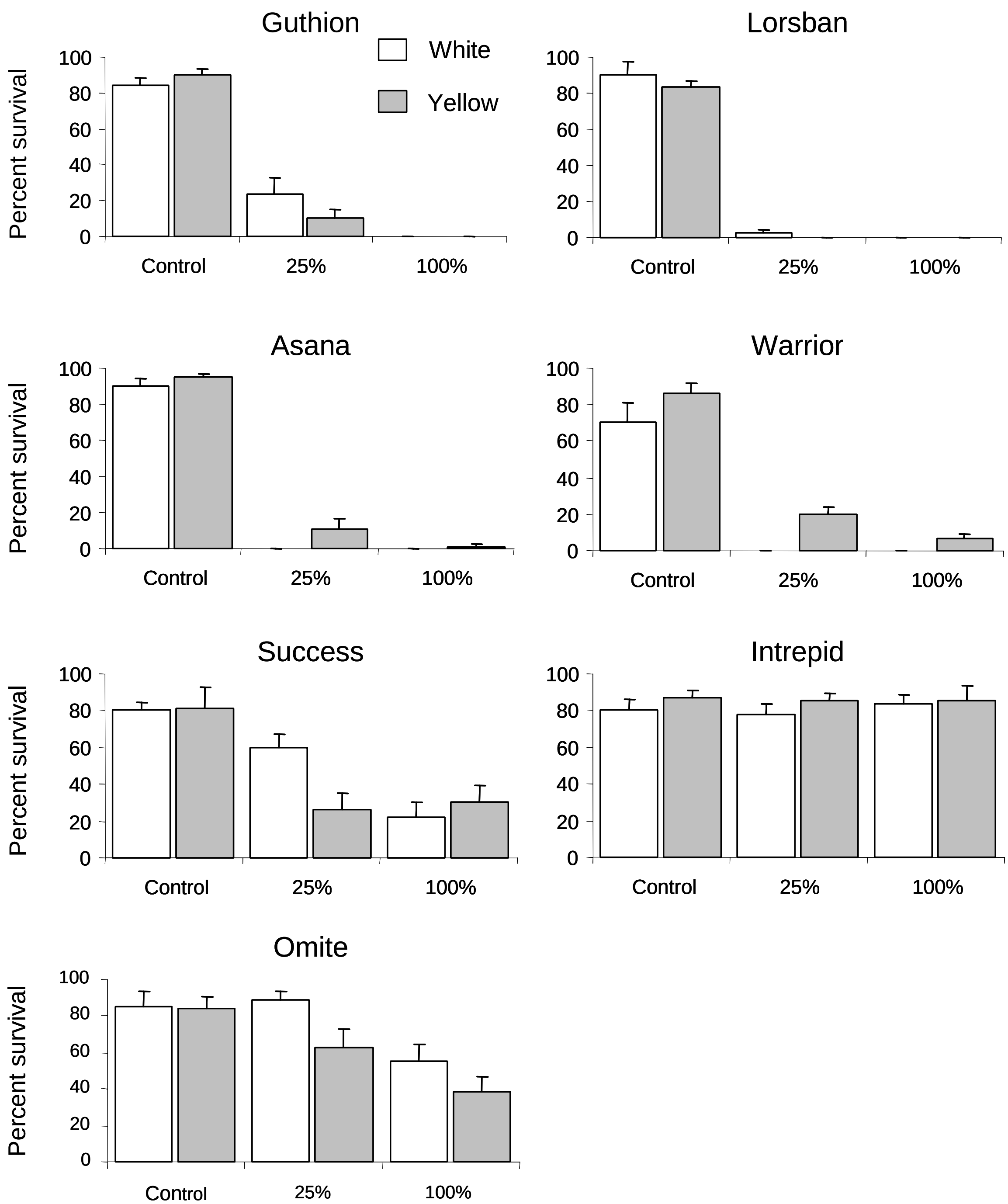

Fig. 2. The longevity in days of adult *Trioxys pallidus* and *Syrphophagus aphidivorus* exposed orally and to surface residues of pesticides at 25% and 100% of the field rates (see Table 1) in comparison to a control treatment..