Establishment of a Whitefly Resistance Documentation and Management Program in Arizona

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Abstract

Adult whiteflies were collected from six regions of Arizona and evaluated for susceptibility to fenpropathrin (Danitol®), acephate (Orthene®) and endosulfan (Thiodan®), and mixtures of fenpropathrin+acephate and fenpropathrin+endosulfan. Strong indications of resistance to fenpropathrin, acephate and the fenpropathrin+acephate mixture were documented in some areas of the state. With all populations evaluated, endosulfan was consistently the most toxic of the insecticides evaluated (singly) and was highly toxic in mixtures with fenpropathrin. Whitefly resistance to pyrethroid insecticides and especially mixtures of pyrethroid+organophosphate insecticides could have serious ramifications for the prevention of sticky cotton in Arizona. To combat further development of pyrethroid resistance cotton growers will need to reduce the number of pyrethroid treatments made per season.

Introduction

Insecticides play a critical role in the management of whitefly populations in cotton, vegetables, melons and ornamental crops in Arizona (Byrne et al. 1990, Dittrich et al. 1990). For this reason, insecticide resistance management is an important aspect of whitefly control programs. The first principle of resistance management is to ensure that pesticides are being used as sparingly as possible. Therein, careful monitoring of whitefly densities, coupled with the use of appropriate action thresholds, promotes resistance management by limiting insecticide treatments to the lowest levels compatible with the high yield and quality demands of production agriculture (Denholm and Rowland 1992). In the case of whiteflies and cotton, the ultimate objective is avoiding losses in quality resulting from stickiness caused by honeydew produced by the pest (Watson et al. 1992).

Accurate and timely information from resistance monitoring is an essential starting point for building resistance management programs. Monitoring information discloses resistances before they become widespread, identifies products that continue to provide acceptable results and helps growers to avoid using ineffective products (Dennehy and Omoto 1994). Additionally, routine monitoring of resistance provides a mechanism for evaluating novel pesticide use regimes and testing the value of resistance management strategies, such as rotations or alternations of control measures.

In 1994 the University of Arizona and the USDA Western Cotton Research Laboratory established collaborative programs to enhance documentation and management of whitefly resistance to insecticides in Arizona. These multi-year efforts include: 1) establishment of programs to monitor resistance in Arizona whitefly populations, 2) validation and implementation of a sticky-trap method for detecting resistance, 3) evaluation of the impact on resistance of novel rotations of insecticides, and 4) collection of baseline data on susceptibility of whitefly populations to the new insecticide, imidacloprid (Admire®). In this paper we report on susceptibility of populations from six regions of Arizona to fenpropathrin (Danitol®), acephate (Orthene®) and endosulfan (Thiodan®), and mixtures of fenpropathrin + acephate and fenpropathrin + endosulfan. These findings represent the initiation of a long-term data base on whitefly resistance, to be maintained by the UA Extension Arthropod Resistance Management Laboratory, for the purpose of assisting Arizona growers with management of resistance in this important pest.

Materials and Methods

Collection of Whiteflies

Whiteflies were collected from cotton fields near Buckeye, Casa Grande, Marana, Maricopa, Safford, and Yuma, Arizona, during August and September, 1994. A Makita® cordless vacuum (4071D) was used as a vacuum source. Plastic vials (13 dram) were placed snugly into the hose connection aperture of the vacuum. These vials, into which the whiteflies were collected, had been previously fitted with screens on the vial bases.

Collections of adult whiteflies were made directly from cotton foliage throughout the fields sampled. Collections were made from 20-200 plants per field, depending on the whitefly density and distribution. In this manner 1000 to 2000 adults were collected into each of 6-8 plastic vials. These vials were placed within ice chests containing ice and were transported to the Resistance Laboratory within 4-12 hours. Bioassays were conducted within 24 hours of arrival at the Resistance Laboratory.

Bioassays of Whiteflies

A leaf disk method (Rowland et al. 1991) was used to estimate whitefly susceptibility to insecticides. This method exposed whiteflies to insecticide-treated leaf disks held in glass vials. The leaf disks, 2.5 cm in diameter, were excised from mature leaves of *Phaseolus vulgaris L.* They were dipped into 100 ml of the desired solutions of insecticide (or water controls) for 10 seconds and allowed to dry. Once dry they were placed individually over 1.5 ml layers of agar (1.3% by weight) held in the bottom of standard 20 ml scintillation vials. The agar prevented the leaf disks from wilting during the 48 hour duration of the bioassay. The top surface of the leaf disks was placed on the agar, leaving the lower leaf surface exposed to the whiteflies.

Dilution series of insecticide were made using formulated insecticide and water, on the basis of μg active ingredient per ml of solution. The insecticides evaluated were fenpropathrin (Danitol® 2.4 EC), acephate (Orthene® 90S), and endosulfan (Thiodan® 3EC). Fenpropathrin and acephate concentrations evaluated were 0, 10, 100, 1000 and 10000 μg/ml. Endosulfan concentrations were 0, 1, 10, 100 and 1000 μg/ml. Two mixtures were evaluated: fenpropathrin + acephate and fenpropathrin + endosulfan. To measure the efficacy of these mixtures we held the concentration of acephate (1000 μg/ml) and endosulfan (100 μg/ml) constant within the mixture and varied the concentration of fenpropathrin. Results presented in Figure 1d, therefore, are from bioassays in which 1000 μg/ml of acephate was added to solutions of concentration 0.1, 1, 10, and 100 mg fenpropathrin/ml. In the mixtures of fenpropathrin+endosulfan (Figure 1e), 100 μg/ml of endosulfan was added to solutions of concentration 0.1, 1, 10 and 100 μg fenpropathrin/ml. A non-ionic surfactant, Triton X-100®, was added to each solution (including controls) at the rate of 0.01%. Dilution series of insecticide were prepared within 48 hours of dipping leaves.

Twenty-five adult whiteflies were aspirated directly from field collection vials into each glass bioassay. Bioassays containing whiteflies were then held for 48 hours in an incubator maintained at 27°C (±1°C), and with a 16 hour photophase, after which mortality was scored. Individuals exhibiting any repetitive (non-reflex) movement were scored as alive. Susceptibility of each population sampled was estimated from a total of 6-8 replications of each of the concentrations noted above. Statistical significance of differences between the populations was determined by ANOVA of mean mortality values, transformed with arcsin x and by *t*-tests of mean mortality observed with specific concentrations of insecticide evaluated.

Results and Discussion

Variation in Response to Specific Insecticides

Fenpropathrin (Danitol®)

We documented significant reductions in sensitivity of whiteflies to fenpropathrin at some locations, most notably populations from Buckeye and the Maricopa Agricultural Center (Figure 1a). The Yuma collection was the most susceptible to fenpropathrin. Whereas concentrations of 10 µg/ml caused high levels of mortality to Yuma whiteflies, treatments of 1000 µg/ml resulted in less than 50 % mortality of the Buckeye population. This reflects at least a 100-fold reduction in susceptibility (i.e., resistance) to fenpropathrin. Such populations should be monitored closely in the future to determine whether resistance to this pyrethroid insecticide is increasing and to measure the impact on whitefly control in the field. Fenpropathrin should be used in rotation with other insecticides in order to reduce selection for the mechanisms conferring this resistance. Studies must now be conducted to determine if whiteflies with reduced susceptibility to fenpropathrin are commensurately less susceptible to other registered pyrethroid insecticides.

Acephate (Orthene®).

All but one population evaluated, Yuma, had relatively low sensitivity to acephate (Figure 1b). Bioassay treatments of 1000 and 10000 µg/ml resulted in only 20-60% mortality for all but the Yuma whitefly population. Given the comparatively long duration of the bioassay we used (48 hours), these data indicated that resistance to acephate was widespread and very intense. However, the fact that the Yuma population was significantly more susceptible to acephate than the other populations emphasizes that resistance scenarios can differ widely from region to region.

Directed efforts should be made to reduce reliance on acephate in cotton, in attempt to manage resistance to this important insecticide. Future studies will focus on determining whether acephate resistance declines in populations, once its use has been suspended and on obtaining a more detailed understanding of the susceptibility to acephate of Arizona populations of whiteflies.

Endosulfan (Thiodan®)

Endosulfan was the most toxic insecticide, of the three insecticides evaluated singly (Figure 1c) and it had the least variability in susceptibility between populations. Unlike bioassays with 100 µg/ml of fenpropathrin or acephate, which had greater than 70% survivorship in the least susceptible populations (Figures 1a,b), 100 µg/ml of endosulfan resulted in 0-5% survivorship (Figure 1c). It merits reminder that our bioassays test adults only and may not fully reflect the impact that insecticides have on populations in the field. Nonetheless, there is every indication from these results that endosulfan is very toxic to adult whiteflies, even against those populations that were resistant to acephate or fenpropathrin. There are indications that use of endosulfan in Arizona cotton has declined sharply in recent years. Our results indicate that it continues to offer utility for reducing some of the over-reliance on acephate and pyrethroid insecticide mixtures.

Fenpropathrin + Acephate (Danitol®+Orthene®)

We found clear evidence that resistance is developing to the insecticide mixture most commonly-used in Arizona cotton, fenpropathrin+acephate (Figure 1d). Once again, the Buckeye population was the least susceptible and the Yuma collection the most susceptible. As with the evaluations of fenpropathrin (used singly), there was an over 100-fold difference in response to the mixture. Bioassays of 10 µg/ml fenpropathrin +1000 µg/ml acephate killed approximately 50% of adults from Buckeye. In contrast, bioassays of 0.1 µg/ml fenpropathrin + 1000 µg/ml acephate killed over 70% of the Yuma and Safford populations. These data indicate that whiteflies can and have become resistant to the most common organophosphate + pyrethroid mixture. Given the extreme reliance by growers on this mixture, these results should serve as further warning of the need to diversify the insecticides used to control whiteflies.

Fenpropathrin + Endosulfan (Danitol®+Thiodan®)

As expected from the high toxicity of endosulfan alone, the combination of endosulfan and fenpropathrin was very toxic to all populations that we tested. No whiteflies tested survived concentrations of 1µg/ml fenpropathrin+100µg/ml endosulfan.

Regional Differences in Susceptibility of Whiteflies

Substantial differences were found between regions of Arizona in the susceptibility of whiteflies. Contrasts of results from Buckeye (Figure 2a), Casa Grande (Figure 2b) and Yuma (Figure 2f) illustrate this point. Yuma whiteflies were the most susceptible, Buckeye whiteflies were the least susceptible, for all insecticides tested except fenpropathrin+endosulfan, for which differences between locations were very small.

Synergism of Fenpropathrin by Acephate

Though of low toxicity to whiteflies, acephate caused greatly enhanced activity (synergism) of fenpropathrin. This was true even with the populations least susceptible to fenpropathrin. The Safford results (Figure 2e) illustrate this phenomenon nicely. Tests of 1000 µg/ml acephate (tested singly) resulted in less than 20% mortality (Figure 2e). Similarly, tests of 1000 µg/ml fenpropathrin (tested singly) produced approximately 80% mortality of the Safford whiteflies. Therefore, the multiplicative activity of these two insecticides would be expected to result in 20% mortality from acephate (at 1000 µg/ml) and 80% mortality from fenpropathrin (at 1000 µg/ml), yielding a total mortality of 84%. Figure 2e shows far greater mortality was observed from the mixture of 1000µg/ml rates of fenpropathrin+acephate than predicted from the multiplicative action of both compounds. Indeed, strong synergism of this mixture is indicated by the few survivors of fenpropathrin treatments of 1.0 µg/ml (+1000 µg/ml acephate).

To the cotton grower this synergism means that acephate greatly enhances the activity of fenpropathrin. A concern, however, is that whiteflies are demonstrating clear evidence of developing a strong resistance to this mixture (Figure 1d). Synergism of fenpropathrin by endosulfan appears to be equally impressive, though our data do not illustrate this point as clearly.

Practical Conclusions

Our findings indicate that resistance to fenpropathrin, acephate and mixtures of fenpropathrin+acephate are a reality in Arizona cotton. This is a warning of potentially greater problems in the future, should control of whiteflies continue to hinge on a single class of chemicals, the pyrethroids. We cannot estimate how intensively pyrethroid resistance will be expressed in homogeneous resistant populations from Arizona cotton. Also unknown is the degree to which reductions in susceptibility to fenpropathrin are linked to reductions in susceptibility of whiteflies to other pyrethroid insecticides used in cotton. These questions will have to be addressed by future field and laboratory investigations.

The endosulfan results indicate that this insecticide has an important place in chemical rotations used to control whiteflies. There was no indication of strong resistance to endosulfan, such as was observed with fenpropathrin and acephate. However, bioassays of adult whiteflies do not provide the entire picture, since control of eggs and immature stages are critical to the efficacy of insecticides in the field. To determine the most suitable chemicals to mix with endosulfan, refer to the recommendations in *Insect Pest Management for Cotton* for results of efficacy trials.

Any steps that reduce reliance on pyrethroids and acephate in cotton appear to be well advised. Additionally, our data illustrate the urgency for registration of novel insecticide products, such as pyriproxyfen and buprofezin, to increase the diversity of physiological targets being selected by insecticides used against whiteflies.

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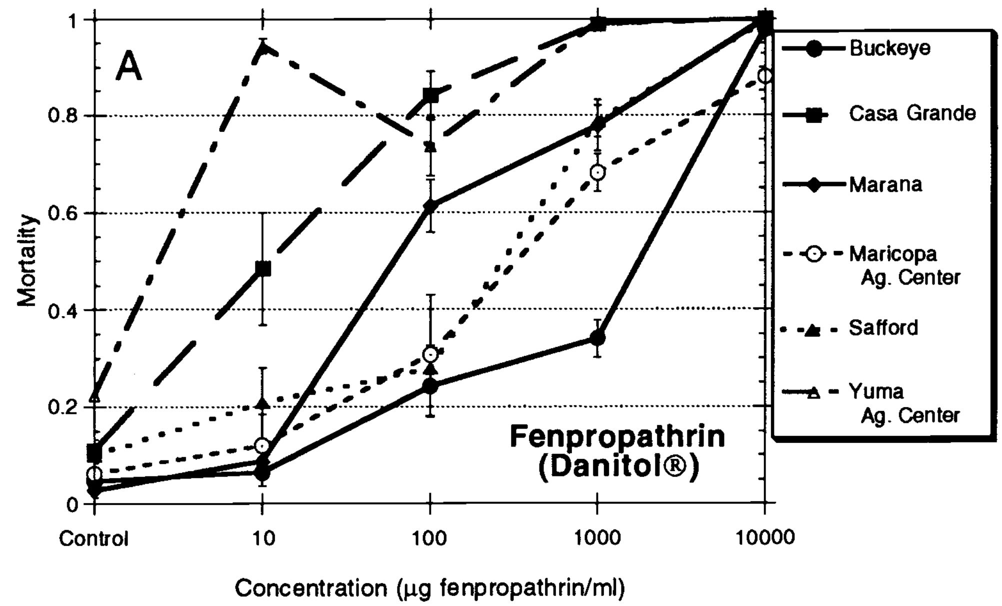
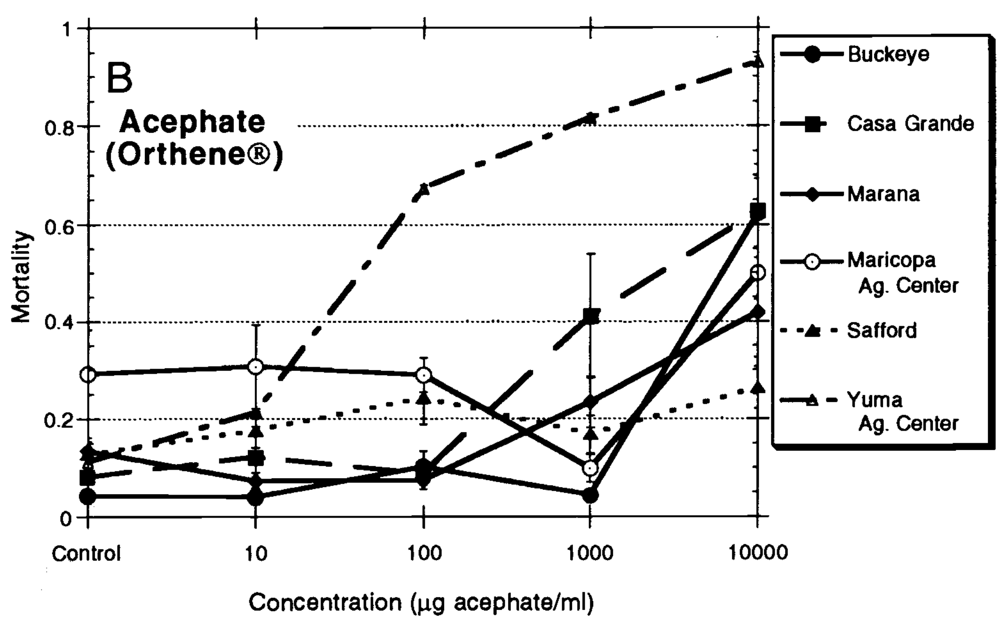
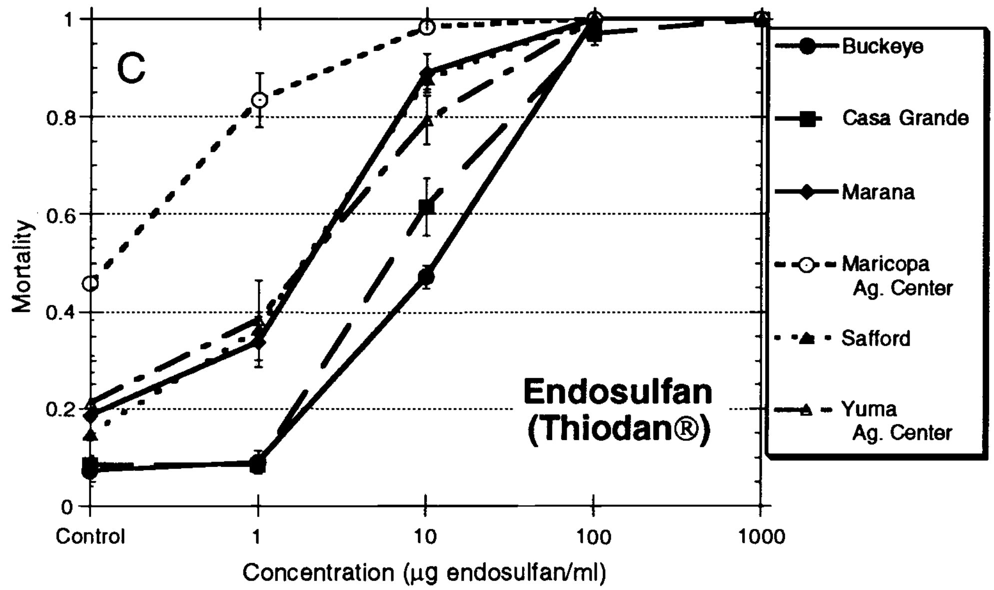
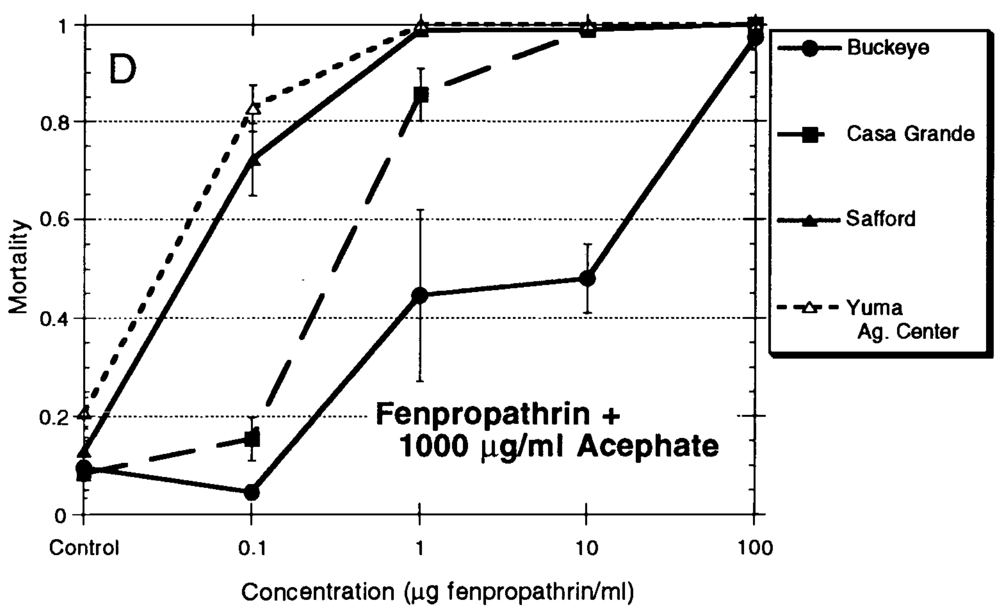
Figure 1. Susceptibility of Arizona populations of whiteflies to: a) fenpropathrin (Danitol®), b) acephate (Orthene®), c) endosulfan (Thiodan®) and d) mixtures of fenpropathrin + 1000 µg/ml acephate and mixtures of fenpropathrin + 100 µg/ml endosulfan.







C
Endosulfan (Thiodan®)
Mortality
Concentration (µg endosulfan/ml)
Control
1
10
100
1000
0
0.2
0.4
0.6
0.8
1
Buckeye
Casa Grande
Marana
Maricopa Ag. Center
Safford
Yuma Ag. Center



D
Fenpropathrin + 1000 µg/ml Acephate
Mortality
Concentration (µg fenpropathrin/ml)
Control
0.1
1
10
100
0
0.2
0.4
0.6
0.8
1
Buckeye
Casa Grande
Safford
Yuma Ag. Center

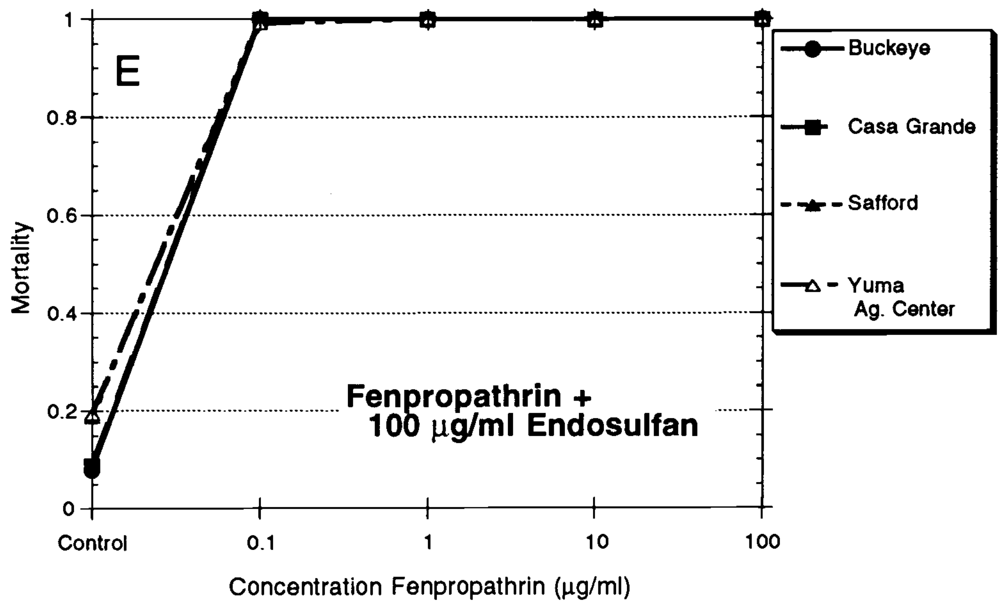
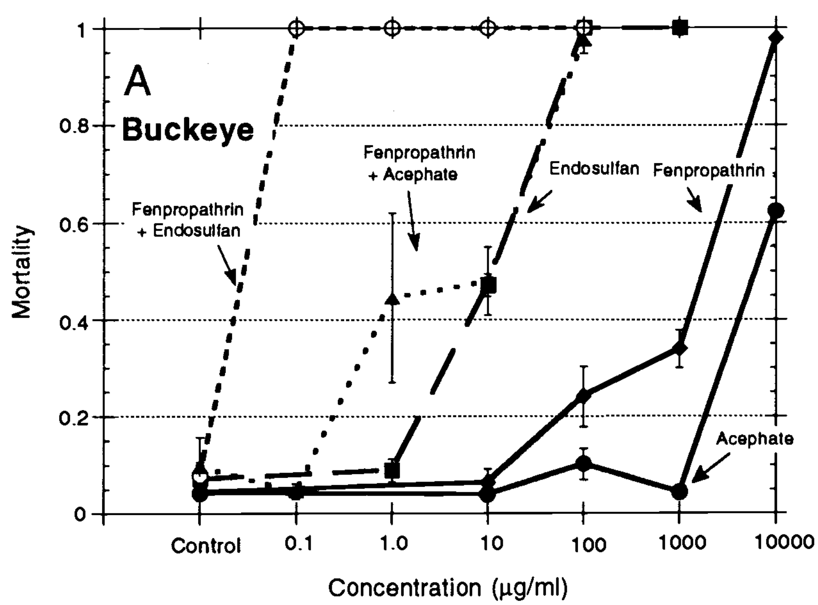
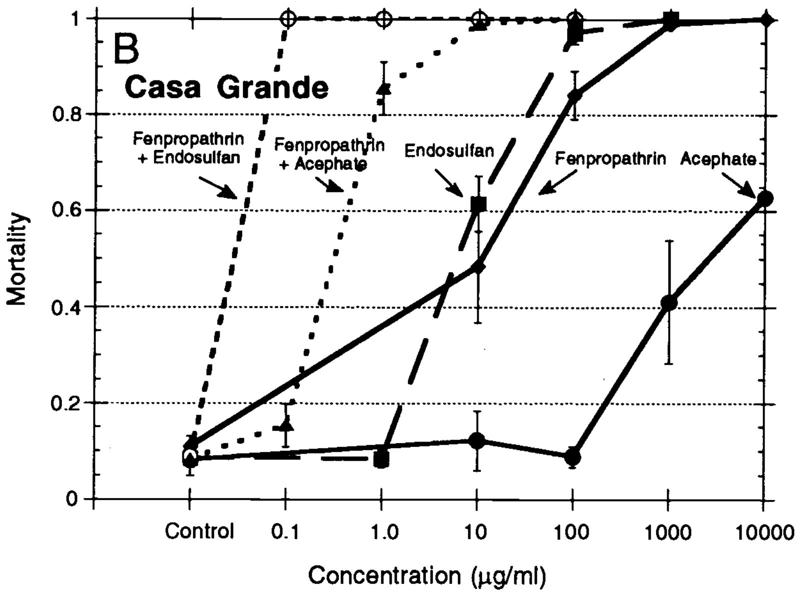
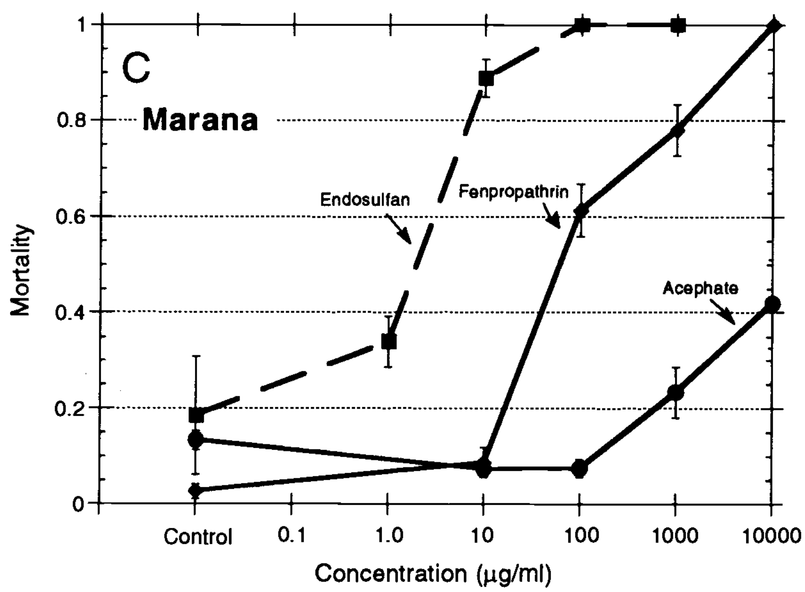
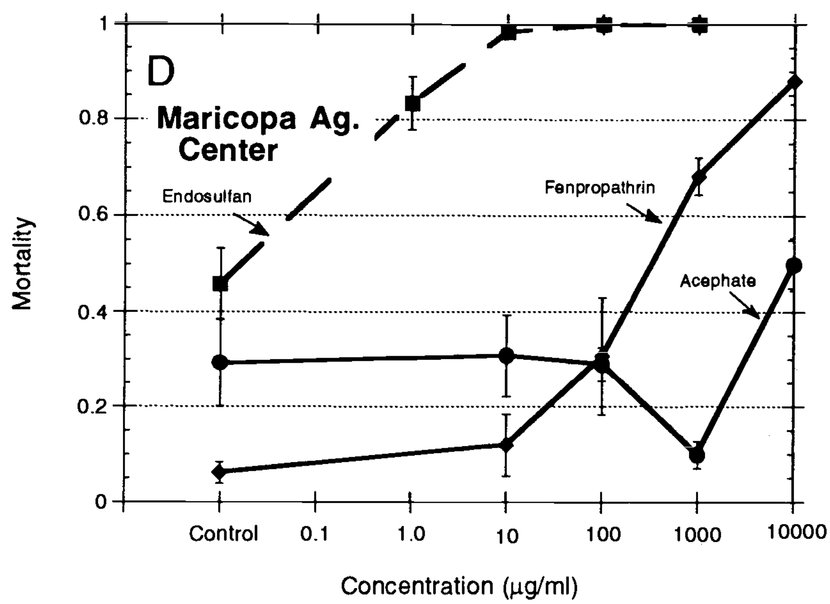


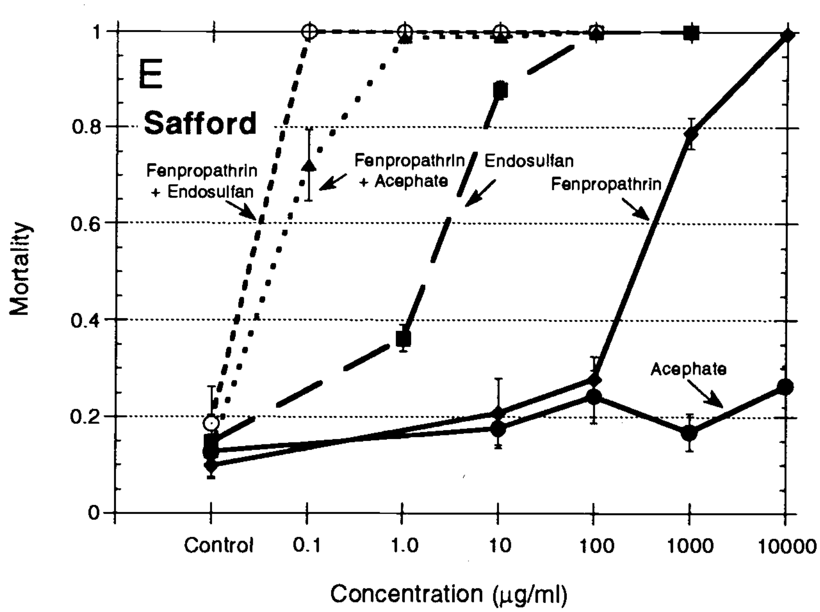
Figure 2. Comparative toxicity of five insecticide treatments to whitefly populations from six cotton-producing regions of Arizona. Fenpropathrin, acephate and endosulfan were tested singly. The line for fenpropathrin+acephate represents varying amounts of fenpropathrin (x-axis) tested in mixtures with a fixed amount (1000 µg/ml) of acephate. The line for fenpropathrin+endosulfan represents varying amounts of fenpropathrin (x-axis) tested in a mixture with a fixed amount (100 µg/ml) of endosulfan. See text for details.

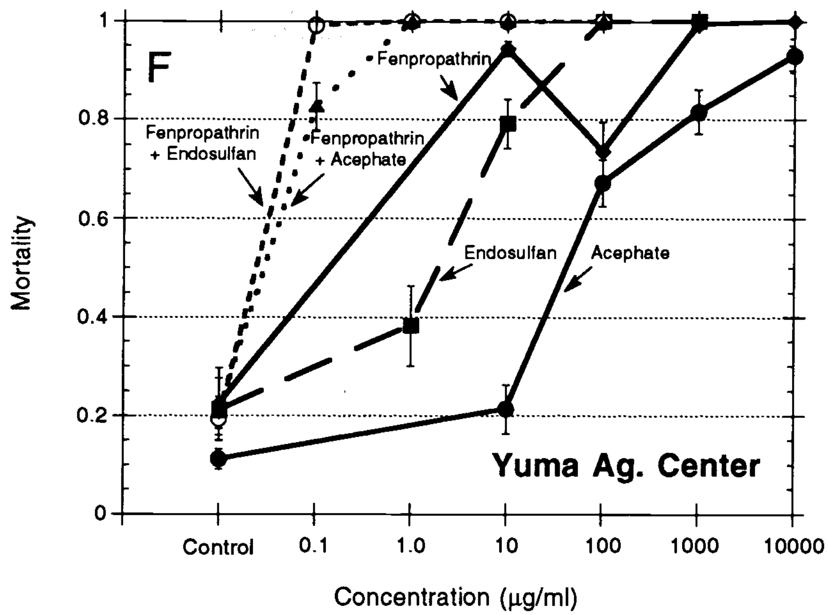



B
Casa Grande
Mortality
1
0.8
0.6
0.4
0.2
0
Fenpropathrin + Endosulfan
Fenpropathrin + Acephate
Endosulfan
Fenpropathrin
Acephate
Control
0.1
1.0
10
100
1000
10000
Concentration (μg/ml)


C
Marana
Mortality
1
0.8
0.6
0.4
0.2
0
Endosulfan
Fenpropathrin
Acephate
Control
0.1
1.0
10
100
1000
10000
Concentration (μg/ml)


D
Maricopa Ag. Center
Endosulfan
Fenpropathrin
Acephate
Mortality
1
0.8
0.6
0.4
0.2
0
Control
0.1
1.0
10
100
1000
10000
Concentration (µg/ml)


E
Safford
Fenpropathrin + Endosulfan
Fenpropathrin + Acephate
Endosulfan
Fenpropathrin
Acephate
Mortality
1
0.8
0.6
0.4
0.2
0
Control
0.1
1.0
10
100
1000
10000
Concentration (µg/ml)


F
Fenpropathrin
Fenpropathrin
+ Endosulfan
Fenpropathrin
+ Acephate
Endosulfan
Acephate
Yuma Ag. Center
Mortality
1
0.8
0.6
0.4
0.2
0
Control
0.1
1.0
10
100
1000
10000
Concentration (µg/ml)