

**TOXICITY OF APPLAUD® AND KNACK®
AGAINST SILVERLEAF WHITEFLIES
FROM SOUTHERN CALIFORNIA:
IMPLICATIONS FOR**

SUSCEPTIBILITY MONITORING

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Abstract

A survey of silverleaf whitefly populations from different cotton growing regions in California was conducted in 1997 to establish baseline toxicological responses to Applaud® and Knack®. Both compounds proved to be highly toxic even in minute quantities. Geographical and temporal variation in responses to each product was observed, but generally was within the range of normal fluctuation. One possible exception was the consistently higher LC₅₀s observed in San Joaquin Valley whiteflies compared to either Palo Verde Valley or Imperial Valley whiteflies. Future surveys will help to establish whether variation in responses to Applaud® and Knack® is due principally to inherent differences among geographical populations or to agro-environmental conditions.

Introduction

The annual cost to control infestations of silverleaf whiteflies, *Bemisia argentifolii* Bellows and Perring, an important pest on a number of agricultural and floricultural crops, is estimated to be approximately \$1 billion in the USA. Insecticides have proven essential in suppressing high numbers of *B. argentifolii* populations on various crops. However, high selection pressure resulting from intensive pesticide use to control whiteflies may lead to insecticide resistance problems. Therefore, there is a need to integrate new chemicals with different modes of action in resistance management programs that may help to avoid developing resistance to insecticides.

Two new chemistries that have recently been introduced into California agriculture are the insect growth regulators (IGRs), buprofezin (Applaud® 70 WP) and pyriproxyfen (Knack® 0.86 EC). Applaud®, an inhibitor of chitin synthesis, is active against the immature stages of whiteflies. It is also known to cause egg sterility and suppression of oviposition, but otherwise is not acutely toxic to adults (Ishaaya et al. 1988). Knack® is toxic to whitefly eggs by suppressing embryogenesis. Knack® also mimics juvenile hormone activity by inhibiting metamorphosis from the pupal to adult stage (Ishaaya and Horowitz 1992). Both Knack® and Applaud® are extremely

effective in suppression of whitefly numbers when applied at an appropriate stage during the development of a crop infestation.

To maximize the effective lives of these two products, the development of an effective resistance management strategy is essential to help minimize selection pressure by the IGRs as well as other conventional insecticides. One of the first steps in this process is to quantify the responses of whitefly populations from various geographic regions of California to the novel chemistries represented in Applaud® and Knack® prior to their widespread use. Baseline susceptibility data of this species will aid in formulating resistance monitoring programs to detect changes in responses of whiteflies. Quantitative information on differential toxicity obtained in a monitoring program is crucial for validating and fine-tuning a particular resistance management strategy.

Materials and Methods

Insecticides

A commercial grade sample of Applaud® was obtained from AgrEvo and Knack® from Valent. Serial dilutions of these two materials to the desired concentrations were made in water on the same day of application. Units for these two compounds are presented in µg (AI)/ml.

Bioassay Techniques

Applaud®

Cotton (*Gossypium hirsutum* 'Deltapine 5) plants in the two true-leaf stage were used for the bioassays. Forty adult whiteflies were confined on individual attached cotton leaves in clip cages of 8 cm² for 24 h to allow for oviposition. After a 24 h oviposition period, adults from the infested leaves were removed. The plants infested with the eggs were maintained in whitefly-free cages within a growth chamber at 27° C, 30-40% RH with a 12:12 photoperiod. After 7 days of egg deposition, the first instar nymphs that hatched and settled were counted and sprayed with various concentrations of Applaud® till run-off. Control plants were sprayed with water alone. Immature mortality was assessed by counting the third and fourth stage nymphs that were alive on day 16 after oviposition. The numbers alive were deducted from the total number of first stage nymphs that were recorded on day 7 before treatment.

Knack®

Eggs were obtained in a similar manner on cotton plants as for tests of Applaud® by confining forty adults per leaf in the clip cages for 24 h. The total number of eggs on each attached infested leaf were counted. The egg-infested leaves were sprayed till run-off with various concentrations of Knack®. To avoid reinfestation by loose flying adults the treated plants were maintained in whitefly-free cages in a growth chamber to allow development of the immatures. Mortality of the eggs was assessed 7 days after treatment by counting the number of live first stage nymphs as well as

the number of eggs that were dead. Total mortality was calculated by subtracting the number of first stage nymphs alive from the total number of eggs that were laid.

Insects

For determining the baseline data for Knack® and Applaud® in silverleaf whiteflies, a number of field collections were made from commercial melons, *Cucumis melo* L. and from cotton crops at 2 locations in Imperial Valley, 2 sites in Palo Verde Valley and 3 sites in San Joaquin Valley. Collections were made at various times in summer 1997 during mid- and late season cotton and early season melons. Adults were collected by vacuuming on the foliage of the two crops. The adults were transported to the laboratory in wooden cages on cotton plants for testing.

Data Analysis

Bioassay data were analysed using the probit model by POLO program (LeOra Software, 1987).

Results and Discussion

Applaud® Toxicity

Although all the whitefly populations tested appear to be highly susceptible to Applaud®, there were regional differences in susceptibility. LC₅₀s ranged from 0.0023 to 0.065 µg (AI)/ml for the whiteflies from the three valleys. No striking differences were found in whitefly responses from Imperial and Palo Verde Valleys as seen by similar LC₅₀s ranging from 0.0023 to 0.008 µg (AI)/ml, approximately 3-fold difference. However, whiteflies from the three San Joaquin Valley field sites showed lower sensitivities to Applaud® than the populations from Imperial and Palo Verde, the LC₅₀s ranging from 0.026 to 0.039 µg (AI)/ml, showing a difference in toxicity of upto 28-fold.

Knack® Toxicity

Results showed a fairly wide range in sensitivity to Knack® by whiteflies from the three locations. LC₅₀s ranged from 0.00003 to 0.010 µg (AI)/ml for Knack®. In general, Knack® was more toxic to whiteflies than Applaud® as seen by the lower LC₅₀s, but with one exception. The first field site in Palo Verde Valley showed the highest LC₅₀ of 0.010mg (AI)/ml among the whitefly populations tested. This was an 8-fold difference in toxicity compared to the whiteflies from the second field site. Similarly an 8-fold difference in susceptibility was observed between whiteflies from field sites 1 and 3 in Imperial Valley. Whitefly populations from San Joaquin Valley were the most susceptible to Knack® as indicated by the lowest LC₅₀s recorded at 0.00003 to 0.0001 µg (AI)/ml.

Preliminary results from our study provide a limited baseline information on the responses of whiteflies from three regions in California to the two IGR's. Considerable variability in toxicity to both Knack® and Applaud® was observed among the geographically diverse populations from California. Further testing during subsequent cotton

seasons is necessary to determine what level of variation is due to inherent differences among populations. Other sources of variation in bioassays include prior environmental exposure of the test populations as well as inconsistencies in technique.

The bioassay we developed to test the susceptibility of the two IGR's using attached cotton leaves was sensitive to detect the differences in whitefly responses. All of the California populations tested appeared to be more sensitive to Knack® in general. Whiteflies from San Joaquin Valley were the most susceptible to Knack® and the least to Applaud®. The results presented here can serve as baseline data to Applaud® and Knack® for California populations before their widespread use. This information will be useful to formulate resistance monitoring programs to detect changes in whitefly responses to the two IGR's.

References

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Table 1. Toxicity of Applaud® to *B. argentifolii* from three regions of California.

Location	Field Collection		<i>n</i>	LC50	
	No.	Date		($\mu\text{g AI/ml}$)	Slope \pm S.E.
Imperial	1	July '97	1148	0.0080	1.5 \pm 0.11
Valley	2	"	792	0.0023	1.1 \pm 0.04
	3	Sept. '97	574	0.0076	2.2 \pm 0.58
Palo Verde	1	Aug. '97	670	0.0047	1.1 \pm 0.20
Valley	2	"	698	0.0033	1.2 \pm 0.17
San Joaquin	1	Aug. '97	378	0.039	3.0 \pm 0.13
Valley	2	"	280	0.065	2.8 \pm 0.63
	3	Sept. '97	360	0.026	2.4 \pm 0.57

Table 2. Toxicity of Knack® to *B. argentifolii* from three regions of California.

Location	Field Collection		<i>n</i>	LC50	
	No.	Date		($\mu\text{g AI/ml}$)	Slope \pm S.E.
Imperial	1	July '97	1178	0.005	1.9 \pm 0.24
Valley	2	"	752	0.0015	2.1 \pm 0.07
	3	Sept. '97	566	0.0006	1.5 \pm 0.46
Palo Verde	1	Aug. '97	414	0.010	5.9 \pm 0.57
Valley	2	"	575	0.0012	2.3 \pm 0.45
San Joaquin	1	Aug. '97	585	0.00006	1.9 \pm 0.77
Valley	2	"	390	0.0001	1.8 \pm 0.29
	3	Sept. '97	620	0.00003	2.3 \pm 0.43