LYGUS BUGS IN ARIZONA REGAIN SUSCEPTIBILITY TO KEY INSECTICIDES J. E. Russell, and T. J. Dennehy Extension Arthropod Resistance Management Laboratory Dept. of Entomology, University of Arizona Tucson, AZ L. Antilla, M. Whitlow and R. Webb Arizona Cotton Research & Protection Council Tempe, AZ J. Pacheco Dupont Agricultural Products Phoenix, AZ

Abstract

Adult lygus bugs, *Lygus hesperus* (Knight), were collected from alfalfa fields in 22 different cotton-producing areas of Arizona. A standardized, glass vial method was used to estimate susceptibility of the collected populations to the pyrethroid insecticide Capture[®] (bifenthrin) and the organophosphate Orthene[®] (acephate). Large differences were recorded in susceptibility of Arizona populations to both insecticides. The most susceptible lygus populations continue to be found in the eastern areas of the state and the least susceptible in central Arizona. Lygus from throughout the state were substantially more susceptible to Capture and Orthene in 1996 than in 1995.

Lygus bioassayed repeatedly from the same locations in 1996 exhibited moderate-to-small seasonal variability in susceptibility to Capture. However, some of the same populations varied widely in susceptibility to Orthene in 1996. The large changes in susceptibility to Orthene were attributed to episodic movements of lygus from other hosts. Eleven insecticides were evaluated against populations most and least susceptible to Capture and Orthene to identify promising candidates for future lygus field trials. A Safford population was substantially more susceptible than a Maricopa population to Admire®, Curacron®, Cygon[®], Malathion[®] and Vydate[®]. Most surprising was the greatly reduced susceptibility to Admire of the Maricopa population. The older insecticides Bidrin[®], Lannate[®], Monitor[®], Naled[®], Ovasyn[®] and Thiodan[®] were quite similar in toxicity to both the Safford and Maricopa populations and therefore are good candidates for further field evaluations to judge their merit for inclusion in lygus insecticide rotations.

Bioassay results were related to field performance of four insecticides in a field trial conducted in Central Arizona. Findings showed that the two insecticides that caused the lowest mortality in vial bioassays, Orthene and Vydate, resulted in the greatest suppression of lygus in the field.

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These findings underscore that the absolute level of mortality observed in bioassays should not be assumed to reflect relative efficacy in the field. It also confirms that Orthene and Vydate continue to be good choices for lygus control, even in Central Arizona where populations are decidedly less susceptible to these insecticides.

We have demonstrated that the adult vial bioassay provides a reliable method for measuring differences between Arizona lygus populations in susceptibility to a broad range of conventional insecticides. We found the method sufficiently sensitive to repeatedly detect significant withinseason and regional differences in lygus susceptibility. These new insights will allow us to better manage the insecticides used in Arizona to control this important pest.

Introduction

Between May and September, lygus bugs, primarily Lygus hesperus, migrate from other hosts into Southwestern cotton fields where feeding can reduce yields due to shedding of immature squares and damage to bolls. In response to this threat, frequently insecticide treatments are necessary to protect cotton yields. Because insecticides are of such great importance for controlling this pest in Arizona, in 1994 we began a long-term effort to systematically investigate key parameters of the impact of insecticides on lygus bugs. An important finding from 1995 was that lygus bugs in many areas of Arizona had strikingly reduced levels of susceptibility to Capture[®] and Orthene[®], relative to the previous year. This worrisome reduction in susceptibility of lygus corresponded with the Arizona whitefly resistance crisis in 1995 that caused some Central Arizona producers to apply 8-12 applications of insecticides to control whiteflies.

In this paper we summarize the results of our statewide lygus program for 1996. Our main objectives were: to maintain statewide monitoring of lygus bug susceptibility to Capture and Orthene; to determine how much lygus susceptibility changes throughout the year at specific locations; to evaluate toxicity to potentially promising new and old insecticides for controlling lygus; and, to relate the control of lygus observed in field trials to bioassay results from the same populations.

Materials and Methods

Collection of Lygus

Using sweep nets, approximately 400-600 adult lygus bugs were collected from each field location. Bugs were emptied from the sweep nets into lunch-size paper bags in which a base of alfalfa cuttings had first been placed. These bags were then placed over ice within ice chests and transported to the laboratory in Tucson. In the laboratory, most lygus were tested within 24 hours. When necessary they were held in one-quart plastic containers, with hinged snap lids, for 24-48 hours at 15-20°C, prior to being tested. From

these containers, groups of five adult lygus were aspirated into each bioassay vial. Lygus populations were sampled from Buckeye (2 locations), Casa Grande (1 location), Cochise County (1 location), Coolidge (1 location), Eloy (1 location), Gila Bend (3 locations), Gila River Indian Reservation (1 location), Gilbert (1 location), Goodyear (1 location), Harquahala (1 location), Higley (1 location), Hyder (1 location), Laveen (1 location), Maricopa (2 locations), Marana (1 location), Mohave (1 location), Paloma (2 locations), Parker (2 locations), Peoria (1 location), Safford (1 location), Tonopah (1 location) and Yuma (4 locations), Arizona. All collections were made from alfalfa fields located adjacent to cotton.

Bioassay Method

We used the glass vial bioassay technique described by Knabke and Staetz (1991). Modifications we made to this technique included: drying treated vials on a commercial hot dog warmer, covering infested vials with dialysis membrane instead of screw caps, and the elimination of carbon dioxide for anesthetizing bugs to facilitate handling.

Standard 20 ml, screw-cap scintillation vials were used. These were treated with solutions of insecticide diluted in acetone or, for controls, acetone only. A volume of 0.5 ml of solution was placed in each vial. Vials were immediately placed on the hot dog warmer, operating at room temperature, and slowly rotated until the solvent evaporated. This provided thorough coverage of the insecticides on the inner surface of the vials.

Solutions were made using technical insecticide on the basis of weight of active ingredient insecticide to total volume of solution. Infested vials were closed with 1"x 1" squares of dialysis membrane secured with a #8 rubber band. Infested bioassay vials were then held for 3 hours in an incubator, maintained at 27°C, after which mortality was recorded. Individuals unable to exhibit repetitive movement of locomotory appendages were scored as dead. Those unable to walk one body length but exhibiting repetitive movement were scored as moribund. Live individuals walked at least one body length. Mortality values reported herein represent only the individuals scored as dead. Inclusion of moribund individuals in mortality estimates did not alter our results appreciably. Statistical significance of differences between the populations evaluated was determined by ANOVA of mean mortality values, transformed with $\arcsin\sqrt{x}$.

<u>Statewide</u> Surveys of Lygus Bug Susceptibility to Capture and Orthene

For the statewide monitoring, acephate (Orthene) concentrations used were: 0 (control), 1,000, and 10,000 μ g/ml. Bifenthrin (Capture) concentrations were: 0, 10, and 100 μ g/ml. Acephate and bifenthrin solutions were prepared each day that bioassays were conducted and were used within 24 hours.

Year-to-Year Changes in Lygus Susceptibility

Where possible, collections of lygus were made at the same locations from 1994 through 1996 to allow comparisons of year-to-year changes in susceptibility to Capture and Orthene. Susceptibility data was obtained all three years from a total of 7 locations (Fig.3a-b).

Within-Season Changes in Lygus Susceptibility

At four locations in Arizona lygus susceptibility to Capture and Orthene was repeatedly measured in the 1996 season. The locations were Casa Grande, Marana, Paloma, and Parker, Arizona. Each site was visited throughout the season on a monthly basis and, when present, lygus were collected and returned to the laboratory in Tucson as described previously for the statewide surveys. Each collected population was then assayed for susceptibility to Capture and Orthene using the standard procedure described above. Box plots of mean mortality observed for each population and concentration tested were constructed to depict within-season changes in lygus susceptibility at each of the four locations.

Potentially Promising New and Old Insecticides

Contrasts of susceptibility of Maricopa and Safford lygus bugs were conducted with the following formulated insecticides: Admire[®](imidacloprid), Bidrin[®] (dicrotophos), Curacron[®] (profenofos), Cygon[®] (dimethoate), Lannate[®] (methomyl), Malathion, Monitor[®] (methamidaphos), Naled[®] (dibrom), Ovasyn[®] (amitraz), Thiodan[®] (endosulfan), and Vydate® (oxamyl). Each insecticide was evaluated against both populations using 5-6 concentrations ranging from 0.1 to 10,000 µg/ml. Eight to ten replications were conducted of each concentration tested. All concentrations were computed on the basis of weight of active ingredient to total volume of solution. Imidacloprid solutions were prepared with distilled water, and imidacloprid-treated vials were rotated for 24 hours to dry. Acetone was used as the diluent for the other 10 insecticides and vials treated with these solutions were rotated for a minimum of 10 minutes but no more than one hour. Insecticide-treated vials were used for bioassays on the same day they were prepared, except with imidaclopridtreated vials. Vials treated with imidacloprid were stored in darkness for up to 2 weeks before being used.

Field Trial

Delta Pine 5415 cotton was dry planted on 4/2/96 and irrigated on 4/4/96. The row width was 40". The row direction was north-south. Each replicate comprised 13.33ft. x 50.0 ft. (4 rows x 40").

Foliar treatments of Curacron $8E^{\circ}$, Orthene $90S^{\circ}$, Thiodan $3E^{\circ}$ and Vydate C-LV $^{\circ}$ were each made on 7/12, 7/29, 8/20 and 9/5/96 for a total of four replications. An untreated plot was included in each replication. Treatments included Knack 0.86E $^{\circ}$ at 0.05 lb.a.i./acre, Applaud 70W $^{\circ}$ at 0.35 lb. a.i./acre, and Supracide 2E $^{\circ}$ at 1.00 lb. a.i./acre. In the first application only, all treatments included Knack

0.86E® at 8.0 fl. oz./acre. In the second application only, all treatments included Applaud 70W® at 0.5 lb. product/acre. Application equipment used was a JD 6000 HiCycle/Compressed Air sprayer with nine TJ60 11002 nozzles, 0.21 gallons per minute per nozzle, at 40 PSI, with a 20" spacing boom configuration. Treatments were applied at 22 gallons per acre at 2.8 MPH in 1st gear.

Results and Discussion

Statewide Surveys of Lygus Bug Susceptibility to Capture and Orthene

Susceptibility of lygus populations from throughout Arizona is illustrated in Fig. 1a-b (Capture) and Fig. 2a-b (Orthene). As in previous years, striking differences were recorded in susceptibility of Arizona lygus bugs to both of these insecticides. The least susceptible populations in 1996 were Gila Bend and Paloma. The most susceptible populations were Cochise County and Safford. We now have a strong foundation of data for interpreting the intensity of lygus resistance to these insecticides. To view the 1996 susceptibility data in the context of previous findings, these data were plotted with contrasting monitoring results from 1995. Whereas the highest levels of resistance to Capture detected in 1995 were not detected in 1996 (Fig. 3a), populations exhibited a wide range of intensities of resistance to Orthene in both 1995 and 1996 (Fig. 3b).

Year-to-Year Changes In Lygus Susceptibility

Figures 4a-b contrast the same locations from which we repeatedly obtained data regarding lygus susceptibility in 1994, 1995, and 1996. In 1995 a significant decrease in susceptibility to Capture and Orthene was detected in Arizona lygus bug populations statewide. This corresponded with severe problems with whitefly resistance to pyrethroids in Arizona and the related high levels of insecticide use in the 1995 season. We are pleased to report that at all but one location lygus bugs were more susceptible to Capture (Fig. 4a) and Orthene (Fig. 4b) in 1996 than they were in 1995. The 1996 Arizona Whitefly Resistance Management Program resulted in substantial reductions in use of insecticides, and especially synergized pyrethroids, in Arizona cotton, . We hypothesize that these changes in insecticide use in cotton contributed substantively to bringing about the observed reductions in resistance in lygus bugs.

Within-Season Changes In Lygus Susceptibility

There is substantial movement of lygus bugs throughout the year as crops are harvested and weeds and desert vegetation dry up. In addition to this movement of the pest other factors such as prior exposure to pesticides, heat-related stress and seasonal difference in lygus physiology could cause our monitoring results to change from one collection to the next. Therefore, we conducted repeated measurements of lygus susceptibility at four locations to determine the magnitude of such changes.

Changes in susceptibility to Capture were moderate to small, depending on the location (Fig. 5a,b). For example, at Casa Grande and Parker mortality estimates from different sampling dates varied less than 20% at the two concentrations of Capture tested (Fig.5a). Susceptibility to Orthene, on the other hand, varied widely throughout the season at many, but not all, of the same locations (Fig.5b). We believe that the large within-season changes in estimates of susceptibility to Orthene result from movement of lygus from refuge crops or wild hosts. Such movement had less impact on within-season susceptibility to Capture because there was less difference between Arizona lygus in susceptibility to this insecticide (Fig. 3a). Importantly, these findings illustrate how pest susceptibility can change remarkably with highly mobile pests, even over the course of one month.

Potentially Promising New and Old Insecticides

To identify insecticides most promising for controlling lygus, we tested a broad range of insecticides against lygus representing populations most (Safford) and least (Maricopa) affected by Capture and Orthene.

We found substantial differences between these two populations in susceptibility to Admire (Fig. 6a), Curacron (Fig. 6c), Cygon (Fig. 6d), Malathion (Fig. 6f), and Vydate (Fig. 6k). In all these cases the Safford population was significantly more susceptible than the Maricopa population.

The relatively new insecticide Admire produced surprisingly large differences in mortality to Maricopa and Safford lygus bugs in both 1995 and 1996 (Fig. 6a). The older insecticides Bidrin (Fig. 6b), Lannate (Fig. 6e), Monitor (Fig. 6g), Naled (Fig. 6h), Ovasyn (Fig. 6i) and Thiodan (Fig. 6j) varied little in toxicity to Safford versus Maricopa lygus but differed widely in their relative potency. These compounds comprise good candidates for field evaluations in the coming year. The objective therein will be to identify new and old chemicals that offer promise for inclusion in out rotations of insecticides for lygus control.

<u>Relating Bioassays to the Field Performance of</u> <u>Insecticides</u>

Bioassay data ultimately need to be coupled with field efficacy trials so that the specific levels of mortality observed in bioassays can be related to the relative efficacy of insecticides in field trials. This objective was furthered in 1996 in a lygus trial conducted at the Maricopa Agricultural Center (MAC). Conventional field applications were made with four insecticides--Curacron, Thiodan, Orthene and Vydate. Bioassays of adult lygus were conducted concomitantly and yielded very large differences in mortality by the four insecticides. The order of toxicity in bioassays, from greatest to least toxic, was Thiodan, Curacron, Vydate , and Orthene (Fig. 7a). Whereas bioassays of 10,000 μ g/ml of Thiodan and Curacron yielded no survivors, assays with this concentration resulted in substantially lower mortality with Orthene and Vydate (Fig. 7a).

Orthene and Vydate treatments provided significantly better control of lygus nymphs than either Curacron or Thiodan (Fig. 7b,c). Significantly fewer adults were present in the Orthene and Vydate treatments than in the Curacron treatments and Vydate had significantly fewer adults than Thiodan (Fig 7d,e). Therefore, the two insecticides that yielded the lowest mortality in vial bioassays resulted in the best performance in the field. This result underscores that laboratory-based bioassays alone should not be used to predict the relative efficacy of insecticides in the field. Bioassay data for specific insecticides must be correlated with field observations in order to be able to relate given levels of mortality in bioassays with general levels of control (e.g., poor, satisfactory or excellent) of lygus populations in the field. The critical contribution of vial bioassays of lygus, as shown herein, is that of detecting previously undescribed resistances in populations, and describing regional, year-toyear and within-season differences in the response of pest to insecticides.

Summary

Susceptibility of lygus bugs to key insecticides varied widely between regions of Arizona. With the information we have collected over the past three years, such differences in lygus bug susceptibility to Capture and Orthene can be interpreted reliably. Additionally, new insights have been gained regarding susceptibility of Arizona lygus to a broad range of insecticides.

We showed that lygus resistance levels can change rather quickly, for better or for worse. The much-reduced use of synergized pyrethroids in 1996 for control of whiteflies was correlated with a significant increase in *susceptibility* of lygus bugs to Capture and Orthene. These data demonstrate that a resistance crisis with other insect pests of cotton can result in collateral problems, in this case making lygus bugs substantially more difficult to control.

We have demonstrated that the adult vial bioassay is a reliable yardstick for measuring differences in susceptibility of Arizona lygus populations to a broad range of conventional insecticides. Also, we found the method sufficiently sensitive to detect significant within-season and regional differences in lygus susceptibility. This new insight into effects of insecticides on lygus will allow us to increase the sophistication of our control recommendations for this pest. Additionally, it explains why growers have experienced regional and seasonal differences in the performance of specific insecticides.

We have found that Central Arizona lygus are some of the most refractory to insecticides in the State. They are significantly less susceptible to a broad range of chemicals, including Orthene and Vydate. Yet, of the insecticides routinely used for this purpose in Central Arizona, Vydate and Orthene appear to continue to provide some of the best lygus control currently available. A resistance management objective will be to incorporate these and other effective insecticides into rotations of products that will reduce the selection for resistance to specific insecticides or crossresistance groups. It is for this reason that we have reported herein the efficacy of promising new and old insecticides for controlling this pest and will continue to evaluate promising products.

Lygus management programs in Arizona will in the coming year place renewed emphasis on effective use of monitoring and thresholds to combat this pest. Additionally, our continued statewide monitoring of lygus resistance will be coupled with field trials whenever possible so that we can provide cotton growers with region-specific recommendations for controlling lygus most efficiently and economically. It is our hope that doing so will allow Central Arizona lygus bugs to continue the trend toward increased levels of susceptibility observed from 1995 to 1996.

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Figure 1a. Mortality of Arizona lygus bugs in 1996 vial bioassay treatments of $10 \mu g/ml$ bifenthrin (Capture®).



Figure 1b. Mortality of Arizona lygus bugs in 1996 vial bioassay treatments of 100 µg/ml bifenthrin (Capture®).



Figure 2a. Mortality of Arizona lygus bugs in 1996 vial bioassay treatments of $1,000 \ \mu g/ml$ acephate (Orthene®).



Figure 2b. Mortality of Arizona lygus bugs in 1996 vial bioassay treatments of $10,000 \mu g/ml$ acephate (Orthene®).



Figure 3a. Contrasts of 1996 lygus susceptibility to Capture with selected results from 1995. Note that each line represents a different location in Arizona at which lygus were collected and bioassayed with Capture using the adult vial bioassay.



Figure 3b. Contrasts of 1996 lygus susceptibility to Orthene with selected results from 1995. Note that each line represents a different location in Arizona at which lygus were collected and bioassayed with Orthene using the adult vial bioassay.



Figure 4a. Changes in Arizona lygus bug susceptibility from 1994 to 1996 as depicted by vial bioassay mortality in treatments of $100 \,\mu\text{g/ml}$ bifenthrin (Capture®).



Figure 4b. Changes in Arizona lygus bug susceptibility from 1994 to 1996 as depicted by vial bioassay mortality in treatments of 10,000 μ g/ml acephate (Orthene®).



Figure 5a. Within-season variation in mortality observed in adult vial bioassays of 10 and 100 μ g a.i./ml bifenthrin (Capture®) of lygus bugs tested repeatedly in 1995 and 1996 from specific locations in four cotton producing regions of Arizona. Each data point denotes the results of a single collection.



Figure 5b. Within-season variation in mortality observed in adult vial bioassays of 1000 and $10,000 \mu g$ a.i./ml acephate (Orthene®) of lygus bugs tested repeatedly in 1995 and 1996 from specific locations in four cotton producing regions of Arizona. Each data point denotes the results of a single collection.



Figure 6a. Susceptibility of the Maricopa and Safford populations of lygus bugs to imidacloprid, as depicted by mortality observed in vial bioassays



Figure 6b. Susceptibility of the Maricopa and Safford populations of lygus bugs to dicrotophos, as depicted by mortality observed in vial bioassays.



Figure 6c. Susceptibility of the Maricopa and Safford populations of lygus bugs to profenofos, as depicted by mortality observed in vial bioassays.



Figure 6d. Susceptibility of the Maricopa and Safford populations of lygus bugs to dimethoate, as depicted by mortality observed in vial bioassays.



Figure 6e. Susceptibility of the Maricopa and Safford populations oflygus bugs to methomyl, as depicted by mortality observed in vial bioassays.



Figure 6f. Susceptibility of the Maricopa and Safford populations of lygus bugs to malathion, as depicted by mortality observed in vial bioassays.



Figure 6g. Susceptibility of the Maricopa and Safford populations of lygus bugs to methamidaphos, as depicted by mortality observed in vial bioassays.



Figure 6h. Susceptibility of the Maricopa and Safford populations of lygus bugs to dibrom, as depicted by mortality observed in vial bioassays.



Figure 6i. Susceptibility of the Maricopa and Safford populations of lygus bugs to amitraz, as depicted by mortality observed in vial bioassays.



Figure 6j. Susceptibility of the Maricopa and Safford populations of lygus bugs to endosulfan, as depicted by mortality observed in vial bioassays.



Figure 6k. Susceptibility of the Maricopa and Safford populations of lygus bugs to oxamyl, as depicted by mortality observed in vial bioassays.



Figure 7a. Mortality of adult lygus bugs from the Maricopa Agricultural Center in vial bioassays of four insecticides concomitantly evaluated in field efficacy trials at the same location.



Figure 7b. Lygus nymphs per 100 sweeps in a lygus control trial conducted at the Maricopa Agricultural Center. Vydate, Orthene, Curacron and Thiodan were each applied four times as noted.



Figure 7c. Confidence intervals (95%) of season-long averages (means) of lygus nymphs per 100 sweeps in a lygus control trial conducted at the Maricopa Agricultural Center. Vydate, Orthene, Curacron and Thiodan were each applied four times.



Figure 7d. Lygus adults per 100 sweeps in a lygus control trial conducted at the Maricopa Agricultural Center. Vydate, Orthene, Curacron and Thiodan were each applied four times as noted.



Figure 7e. Confidence intervals (95%) of season-long averages (means) of lygus adults per 100 sweeps in a lygus control trial conducted at the Maricopa Agricultural Center. Vydate, Orthene, Curacron and Thiodan were each applied four times.