CURACRON RESISTANCE MONITORING - 1995 SURVEY RESULTS

H. W. Ray, D. V. Allemann,
N. D. Ngo, and S. T. Moore, Sr.
Research Specialist, Insect Control Manager, Sr. Research Specialist, and Station Manager, respectively
Ciba Crop Protection
Vero Beach, FL
Greensboro, NC
Greenville, MS, respectively

Abstract

Ciba Crop Protections' Curacron® resistance monitoring program was begun in 1993 and has now completed the third cotton growing season. In 1995 field collections of tobacco budworm adults (*Heliothis virescens* F.) from several areas throughout the Cotton Belt were made. Eggs from these moths were shipped to Ciba's Vero Beach Research Center in Vero Beach, Florida, by overnight air where IRAC Method #7 was used to conduct discriminating dose and multi-dose bioassays with Curacron 8E against late 2nd instar larvae. In discriminating dose bioassays field strains of larvae from McGehee, Arkansas, and Cheneyville, Louisiana, showed slight increases in resistance gene frequency from 1994 to 1995, with a field strain of larvae from Tifton, Georgia, showing the highest increase in resistance gene frequency. However, this occurred during the two year period from 1993 to 1995, with no data obtained in 1994. In contrast to these increases in resistance gene frequency, a strain of larvae from Friars Point, Mississippi, showed a marked decrease. Multi-dose bioassays showed slight to moderate increases in tolerence to Curacron® with larvae from McGehee, Arkansas, (1.2-fold) from 1994 to 1995 and Tifton, Georgia, (3.2-fold) from 1993 to 1995. Increases in sensitivity were found in field strains of larvae from Cheneyville, Louisiana, and Greenville, Mississippi.

Introduction

In 1982, Curacron® brand profenofos was introduced into the US cotton market and has since become Ciba Crop Protections' most important tool for controlling the tobacco budworm, *Heliothis virescens* (F.) Therefore, in 1990, there was some concern generated when control failures were reported with several of the organophosphates in Louisiana (Leonard et al. 1991). This, plus reports of control failures in subsequent years from other locations, some of which were blamed on suspected Curacron® resistance prompted Ciba Crop Protection to began a Curacron® resistance monitoring program during the cotton growing season of 1993. When we started this program we realized that we had no baselines from pre-Curacron® years and didn't know exactly what to expect.

The objectives of this program were:
1. Set baselines so that sensitivity or lack of it could be monitored in subsequent years.
2. Look at seasonal variation within a population.
3. Assess variability among populations.
4. Become better stewards of Curacron®.

Testing in 1993 consisted of adult vial tests as described by Kanga and Plapp (1992) at Ciba's Delta Research Station (DRS) in Greenville, Mississippi, and larval bioassays at Ciba's Research Center in Vero Beach, Florida, (VBRC) and also at the Delta Research Station. Subsequently, in 1994 and 1995 the adult vial tests were scaled back, and larval testing was expanded with the majority of the bioassays conducted at the Vero Beach Research Center with some supplemental tests conducted at the Delta Research Station.

Materials and Methods

Nine of the ten participants in the 1994 resistance monitoring program were in the 1995 program and two new ones were added - one in Texas and one in Louisiana. In 1994 the original ten cooperators had been provided with either battery powered or 110 volt walk-in blacklight traps. During the 1995 season all of the traps remained in the same locations with the exception of the one at College Station, Texas, which was relocated to nearby Snook, Texas.

The light traps were used to collect tobacco budworm moths which were then placed in one-gallon cardboard containers and allowed to oviposit on cheesecloth sheets. The egg sheets were collected and shipped in insulated containers by overnight air to the Vero Beach Research Center where the eggs were hatched and bioassays conducted against late 2nd instar larvae.

In general, the quantity of eggs obtained this season was somewhat higher than in 1993 or 1994, but there were still some diasppointments. Droughts, beet armyworms, and the lack of tobacco budworm pressure caused inadequate eggs samples to be collected from four of the eleven collection sites.

Bioassays

If the quantity of eggs and resulting larvae was low then discriminating dose bioassays as described by Roush and Miller (1986) which show the resistance gene frequency in a population were conducted. The discriminating doses, 15 ppm and 30 ppm, were based on previous laboratory experiments which showed that the 15 ppm dose would kill about 50% of a susceptible strain and the 30 ppm dose

Reprinted from the *Proceedings of the Beltwide Cotton Conference*
Volume 2:1067-1069 (1996)
National Cotton Council, Memphis TN

about 90% of that same strain and only a small percentage of a resistant strain.

Multi-dose bioassays which provided data on the average response of entire populations were conducted when sufficient larvae were available. Each multi-dose bioassay required 300 larvae as there were 5 doses, 3.75, 7.5, 15.0, 30.0, and 60.0 ppm plus an untreated control with each treatment requiring 50 larvae.. Laboratory colonies from the USDA laboratory at Stoneville, Mississippi, and the Vero Beach Research Center were bioassayed as untreated reference strains.

All bioassays, both discriminating dose and multi-dose were conducted using the IRAC #7 method which is a leaf-dip bioassay designed to be simple and suitable for all types of resistance. Leaf disks, 1.9 cm in diameter, were punched from cotton leaves taken from potted cotton plants (var. Stoneville 132). grown in a greenhouse. These disks were placed in 400 ml. beakers containing the various concentrations of Curacron 8E mixed in distilled water and stirred for 45 seconds using a magnetic stirrer. The leaf disks were then placed on wire racks and allowed to dry for a minimum of 1 hour. After drying they were placed into 2.5 cm. x 10 mm. plastic petri dishes which contained single sheets of 2.3 cm. Whatman #3 filter paper wetted with 55 microliters of distilled water. One late 2nd instar larva was introduced into each dish using a camel's hair brush and the dishes sealed. The dishes were then placed on fiberglass trays and held at room temperature in darkness for 48 hours at which time they were opened and mortality evaluations made. Larvae unable to sustain coordinated movement when probed were also counted as dead, and when necessary, mortality was adjusted according to Abbott's Formula (Abbott 1925).

Results

Previous research had shown that there was wide variability among tobacco budworm strains in response to various classes of pesticides. Response variations up to 270-fold to methyl parathion, an organophosphate, had been demonstrated by Wolfenbarger et al. (1984). Roush and Luttrell (1989) had shown up to 9.6-fold differences in response to fenvalerate, a synthetic pyrethroid (Table 1), and Stone and Sims (1993) had shown up to 3.5-fold differences in response to a biological, *Bacillus thuringiensis* (Table 2). Therefore, it was expected that there would also be variability in the response of different field strains of tobacco budworms to Curacron®. This proved to be true as shown in both the discriminating dose and multi-dose bioassays.

Discriminating Dose Bioassays

In the discriminating dose bioassays the two field strains of tobacco budworm from McGehee, Arkansas, and Cheneyville, Louisiana, showed slight increases in the resistance frequency gene, and a strain from Tifton, Georgia, showed a substantial increase (Table 3). In the McGehee, Arkansas, strain the mortality remained fairly steady at the 15 ppm dose, 6.0% in 1993, 8.0% in 1994, and 8.0% in 1995. However, at the 30 ppm dose, there was a greater fluctuation, 35.0% mortality in 1993, 43.0% in 1994, and down to 38.0% in 1995. Mortality in the Cheneyville, Louisiana, strain at 15 ppm was 42.0% in 1993, 21.7% in 1994, and 18.6% in 1995; at 30 ppm mortality was 44.8%, 66.7% , and 52.0%, respectively. The Friars Point, Mississippi, strain showed a marked decrease in the resistance frequency gene. Mortality at 15 ppm was 0.0% in 1993, 26.1% in 1994, and 96.7% in 1995; at 30 ppm it was 2.5%, 75.0%, and 100.0%, respectively. First year data from the Winnsboro, Louisiana, and Snook, Texas, field strains showed that at 15 ppm mortality was 2.0%, and 59.9%, respectively. At the 30 ppm dose the Winnsboro, Louisiana, strain had 24.0% mortality and the Snook, Texas, strain 72.9% mortality.

Multi-dose Bioassays

Multi-dose bioassays showed that LC_{50} values ranged from 3.3 ppm in the Vero Beach Research Center laboratory colony to 59.3 ppm in the Greenville, Mississippi, field strain (Table 4). Data also show that the LC_{50} value increased from 1994 to 1995 in the tobacco budworm field strain from McGehee, Arkansas. This increase was from 30.0 ppm to 36.4 ppm and indicated an increase in tolerance to Curacron®. At the same time the field strain from Greenville, Mississippi, showed a declining LC_{50} value from 74.9 ppm in 1994 to 59.3 ppm in 1995. This indicated an increase in sensitivity to Curacron®.

Only the McGehee, Arkansas, collection site provided early- (June), mid- (August) and late- (end of September/early October) season egg collections which allowed the seasonal variation to be observed (Table 5). From these three collections the LC50 values were determined to be 21.3 ppm, 37.5 ppm and 33.3 ppm, respectively, which showed an increase in tolerance to Curacron® by midseason followed by an increase in sensitivity at the end of the season.

Summary

Discriminating dose bioassays showed slight increases in the frequency of the resistance gene(s) in two field strains of tobacco budworms from 1994 to 1995, and a substantial increase in another from 1993 to 1995. One strain also showed a decrease in the frequency of the resistance gene(s).

Multi-dose bioassays showed that one field strain had an increased LC50 value, and one field strain had a decreased LC50 value from 1994 to 1995. The LC_{50} value of the Tifton, Georgia strain increased from 1993 to 1995. All other strains had values that remained stable, or there were no previous values with which to compare.

Conclusions

There appears to be some upward shifting in the tolerance to Curacron® in some field strains of tobacco budworms as demonstrated by both discriminating dose and multi-dose bioassays. The shifts have been less than 10-fold, but are of some concern and must continue to be monitored. Therefore, Ciba Crop Protection remains fully committed to Curacron® resistance monitoring and will continue the program in 1996. Our goal is to keep Curacron® as a valuable tool for tobacco budworm control many years to come.

References

Abbott, W. S. 1925. A method for computing the effectiveness of an insecticide. J. Econ. Entomology. 18:265-276.

Kanga, L. B., and F. W. Plapp, Jr. 1992. Development of a glass vial technique for monitoring resistance to organophosphate and carbamate insecticides in the tobacco budworm and the boll weevil. In Proceedings 1992, Beltwide Cotton Production and Research Conferences. Nashville, TN.

Leonard, B. R., et al. 1991. Tobacco Budworm: Insecticide resistance and field control in the Macon Ridge region of Louisiana, 1990. In Proceedings 1991, Beltwide Cotton Production and Research Conferences. San Antonio, TX.

Roush, R. T., and R. G. Luttrell. 1989. Expression to pyrethroid insecticides in adults and larvae of tobacco budworm (Lepidoptera: Noctuidae): Implications for resistance monitoring. J. of Econ. Entomol. 82(5): 1305-1310.

Roush, R. T., and G. L. Miller. 1986. Considerations for design of insecticide resistance monitoring programs. J. Econ. Entomol. 79:293-299.

Stone, T. B., and S. R. Sims. 1993. Geographic susceptibility of *Heliothis virescens* and *Heliocoverpa zea* (Lepidoptera: Noctuidae) to *Bacillus thuringiensis*. J. Econ. Entomol. 86: 989-994.

Wolfenbarger, D. A., J. A. Harding, and S. H. Robinson. 1984. Tobacco budworm (Lepidoptera: Noctuidae): variations in response to methyl parathion and permethrin in the subtropics. J. of Econ. Entomol. 77(3):701-705.

Table 1. Susceptibility of several strains of Heliothis virescens to fenvalerate.

Strain	LC_{50}
MSU (Laboratory)	14.8
Uvalde, TX	49.5
Casa Grande, AZ	30.2
Midland, TX	141.0

Roush and Luttrell (1989)

Table 2. Susceptibility of several strains of Heliothis virescens to Dipel® (diet bioassay).

Strain	LC_{50} (ug/ml of diet)
South Carolina	9.3
Arkansas	10.0
PEG (ICI)	10.2
Georgia	12.2
Texas	13.5
Monsanto	14.1
Mississippi	16.5
Arizona	18.6
Louisiana	21.5
Alabama	22.2
Virgin Islands	23.6
North Carolina	33.7

Stone and Sims (1992)

Table 3. Discriminating dose leaf dip bioassays with Curacron® against several strains of *Heliothis virescens*.

Strain	Rate	Percent Mortality at 48 Hours		
		1993	1994	1995
Friars Point, MS	15 ppm	0.0	26.1	96.7
	30 ppm	2.5	75.0	100.0
McGehee, AR	15 ppm	6.0	8.0	8.0
	30 ppm	35.0	43.0	38.0
Cheneyville, LA	15 ppm	42.0	21.7	18.6
	30 ppm	44.8	66.7	52.0
Winnsboro, LA	15 ppm	NA	NA	2.0
	30 ppm	NA	NA	24.0
Tifton, GA	15 ppm	90.0	NA	10.0
	30 ppm	94.0	NA	76.0
Vero Beach, FL (L)	15 ppm	NA	72.0	96.0
	30 ppm	NA	100.0	100.0
Snook, TX	15 ppm	NA	NA	59.9
	30 ppm	NA	NA	72.9
Raymondville, TX	15 ppm	0.0	22.7	NA
	30 ppm	8.3	66.7	NA

(L) = Laboratory Strain

Table 4. Susceptibility of several strains of *Heliothis virescens* to multiple dose leaf dip bioassays with Curacron®.

Strain	LC_{50} (ppm)		
	1993	1994	1995
McGehee, AR	31.8	30.0	36.4
Cheneyville, LA	8.1	28.7	28.6
Winnsboro, LA	NA	NA	41.0
Friars Point, MS	16.2	NA	NA
Greenville, MS (L)	61.3	74.9	59.3
Stoneville, MS (L)	NA	7.2	8.2
Tifton, GA	7.1	NA	22.8
Vero Beach, FL (L)	3.6	NA	3.3
Vero Beach, FL	NA	9.3	9.5
Snook, TX	NA	NA	26.0

(L) = Laboratory Strain

Table 5. Seasonal variation in LC_{50} values with a field strain of *Heliothis virescens* from McGehee, Arkansas.

Strain	LC_{50} (ppm)		
McGehee, AR	21.3 (June)	37.5 (August)	33.3 (Sept./Oct.)