LABORATORY EVALUATION OF SELECTED INSECTICIDES ON FIELD-COLLECTED POPULATIONS OF BOLLWORM AND TOBACCO BUDWORM LARVAE

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Abstract

Bollworm (CEW) and tobacco budworm (TBW) larvae and adults were collected from a variety of host crops and evaluated for susceptibility to MVP II®, cyperemthrin and spinosad (Tracer®) during the 2009 season. Results were compared to historical data collected throughout a fifteen-year study period beginning in 1995. As expected, CEW larvae were less susceptible to the effect(s) of MVP II® than were TBW larvae; however, the susceptibilities of both CEW and TBW have remained relatively stable throughout the study period with annual average fluctuations in LC_{50} values of only 2-3-fold. Although cypermethrin remains an effective insecticide to control CEW larvae, average LC_{50} values were highest during the 2009 season and ca. 6-fold higher than LC_{50} values recorded in the mid-1990s. The effectiveness of cypermethrin for the control of TBW larvae has declined throughout the fifteen-year study period also. The average LC_{50} values for cypermethrin against TBW larvae collected during the 2009 season were ca. 37-fold higher than the LC_{50} value obtained for a pyrethroid-susceptible laboratory strain and ca. 13-fold higher than the average LC_{50} values obtained during the mid-1990s. Decreases in the susceptibilities of CEW and TBW populations were confirmed by the use of topical application bioassays and adult vial tests. Spinosad (Tracer®) has remained highly effective against CEW and TBW larvae throughout the study period; however, the highest average LC_{50} values to date were obtained during the 2009 season.

Introduction

The bollworm (CEW; *Helicoverpa zea*) and the tobacco budworm (TBW; *Heliothis virescens*) are two of the more economically important pests of cotton in the United States. Because CEW and TBW populations have developed resistance to many of the insecticides used for their control, it is essential that research efforts and agricultural practices be devoted to the preservation of those insecticides that are still effective and to the development of new replacement compounds and technologies. Programs to monitor insecticide susceptibilities of field-collected populations of CEW and TBW are critical to the development of those effective management strategies. Samples of CEW and TBW populations were collected from cotton, tobacco, peanut and corn fields throughout Georgia during the summer of 2009. Larvae from those field-collected samples were assayed for susceptibility to a variety of insecticides using treated-diet and topical application bioassays; adults were evaluated using an adult vial test bioassay. Results were compared to baseline data collected between 1995-1999 and 2003-2005.

Materials and Methods

The counties from which bollworm (CEW) and tobacco budworm (TBW) have been collected throughout the study period are shown in Figure 1. During the 2009 season, CEW and TBW were collected from 11 counties including Burke, Colquitt, Decatur, Dooly, Early, Miller, Mitchell, Sumter, Taylor, Terrell, and Tift. Field-collected CEW and TBW moths or larvae were transported to facilities at the University of West Georgia. Larvae were transferred to a pinto bean/wheat germ, agar-based diet, and adults were placed in mating cages to produce adequate numbers of larvae for testing. Larvae and adults were maintained at 27˚C, LD 14:10 and ca. 40% RH. The insecticides used were MVP II® (19.1% A.I., Monsanto Corporation, St. Louis, MO); cypermethrin (94.3% A.I., FMC Corporation, Princeton, NJ); and spinosad (91.3% A.I., Dow AgroSciences, Indianapolis, IN).

Larvae were evaluated using a modified insecticide-treated diet bioassay or by topical application; adults were evaluated using an adult vial test (AVT) protocol. For the insecticide-treated diet assay, an insecticide test solution (100 µl) was added to 50 ml of liquefied pinto bean/wheat germ, agar-based diet at ca. 57˚C while mixing with a variable speed stirrer. The insecticide-treated diet (ca. 2.5 ml) was distributed into 1 oz. clear plastic medicine cups. The treated diets were allowed to cool and gel. One neonate or one late 2^{nd} instar larva (depending upon the insecticide being evaluated) was added to each cup, and mortality was monitored over a 4 day period. For the topical application bioassay, a 1 µl droplet an insecticide solution or acetone (control) was applied to the dorsal thorax of a 4th instar larva (ca. 35 mg). Mortality was assessed after a 48 h exposure period. For the adult vial test, a single moth was placed in an insecticide-treated or acetone-treated (control) vial. Mortality was assessed after a

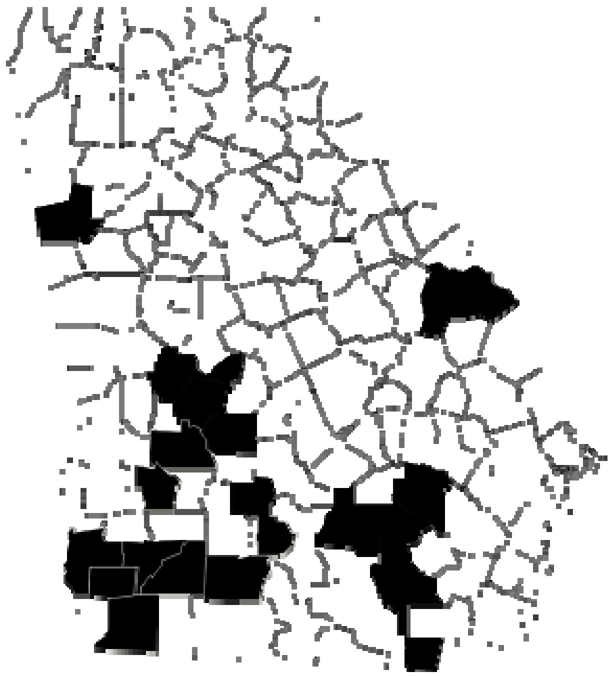

Figure 1. Bollworm and tobacco budworm collection sites.

24 h exposure period. Mortality was defined as the inability of the larva to move across the diet surface when probed or for a moth to fly a distance of 1 meter when dropped from a 2 meter height. During the treatment period, the larvae and adults were held in an environmental chamber at 27˚C, LD 14:10 and ca. 40% RH.

Results and Discussion

As expected, MVP II® was less effective against CEW larvae as compared to TBW larvae (Tables 1-4; Figures 2, 3, 4 and 5). The average CEW LC_{50} values were ca. 20-fold greater than the average TBW LC_{50} values. Although isolated CEW and TBW populations exhibited high levels of survival following exposure to MVP II ®, the average CEW and TBW LC_{50} values for the 2009 season were comparable to the average CEW and TBW LC_{50} values obtained during the mid-1990s, and TBW LC_{50} values for the field-collected strains have remained comparable to the LC_{50} values obtained for the laboratory-maintained HRV, OPS, OPR and PYR reference strains. The highest CEW LC_{50} values (> 250 ppm) were recorded during the 2004-2005 seasons and were more than 75-fold higher than the LC_{50} values obtained for the most susceptible CEW field-strain.

Although decreases in the susceptibilities of CEW populations to pyrethroid insecticides were noted throughout the study period and are of considerable concern, data indicated that CEW populations in Georgia remained relatively susceptible to cypermethrin. LC_{50} values for field populations collected during the 2003-2005 seasons were only two-fold to three-fold greater than LC_{50} values obtained for field populations collected during the 1996 and 1997 seasons (Table 3; Figures 2 and 6). However, 2009 LC_{50} values (for both CEW and TBW) were the highest LC_{50}s recorded to date (Tables 1-4; Figures 6 and 7). The average 2009 CEW LC_{50} value was ca. 6-fold higher than the average 1996-1997 CEW LC_{50} value. The data also indicated that pyrethroid resistance remains an issue for the control of TBW populations in Georgia. The average 2009 TBW LC_{50} value was ca. 13-fold higher than the average

1995-1996 TBW LC_{50} value; more than 37-fold higher than the LC_{50} value obtained for the pyrethroid-susceptible HRV laboratory strain; and 1.5-fold higher than the LC_{50} value obtained for a laboratory-selected, pyrethroid-resistant strain (PYR) (Table 4).

Topical application (Tables 1 and 2) and AVT (Table 1 and 2; Figure 8) data also indicated decreases in the susceptibilities of CEW and TBW populations to cypermethrin over time. Compared to a pyrethroid-susceptible laboratory strain, topical LD_{50} values for the 2009 CEW populations were 3- to 12-fold higher, and the percent survival of CEW adults using the AVT has steadily risen since 1998. In field-collected TBW populations, pyrethroid resistance was confirmed by topical application in the Tay2 05 strain (LD_{50} = 49.3 µg/g larva; RR = 31.4; data not shown), the Cam 09 strain (LD_{50} = 93.8 µg/g larva; RR = 59.7) and the Tif C 09 strain (LD_{50} = 80.5 µg/g larva; RR = 51.3) (Tables 2). Furthermore, an evaluation of LC_{95} values for cypermethrin against TBW larvae have indicated an annual and sharp increase since the monitoring project began (Figure 9).

To date, spinosad (Tracer®) has remained effective against all strains tested (Tables 1-4; Figures 2, 3, 9, 10, and 11). Mean LC_{50} values for CEW larvae (0.49 ± 0.07) and TBW larvae (0.46 ± 0.05 ppm) were comparable and have remained stable throughout the fifteen-year study period.

Summary

Throughout the fifteen year study period bollworm (CEW) and tobacco budworm (TBW) populations in Georgia have remained relatively susceptible to MVP II®. As expected, the data have indicated that CEW larvae were more tolerant to the effects of MVP II® than TBW larvae. CEW and TBW populations have become more resistant to cypermethrin. In 2009, CEW populations were 6 times more resistant to cypermethrin than CEW populations sampled during the mid-1990s and 3 times more resistant to cypermethrin than CEW populations sampled during the mid-2000s. TBW populations collected during the 2009 season were on average 12 times more resistant than TBW populations sampled during the mid-1990s, 5 times more resistant than TBW populations sampled during the late 1990s, and ca. 1.5 times more resistant than TBW populations sampled during the mid-2000s. The data indicated that spinosad (Tracer®) has remained effective in the control of CEW and TBW populations in Georgia. There have been no substantial fluctuations in the activity of spinosad against CEW and TBW larvae throughout the study period. In general, the treated diet-96 h activity spectrum for the insecticides tested were as follows: CEW: Spinosad (Tracer®) > Cypermethrin > MVP II®; TBW: Spinosad (Tracer®) > MVP II® > Cypermethrin.

Acknowledgements

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Table 1. Susceptibilities of field-collected bollworm populations to MVP II®, spinosad (Tracer®), and cypermethrin using treated diet, topical application, and adult vial test (AVT) bioassays--2009.

Colony	Treatment	Diet LC_{50} (C.I.; Slope), ppm	Topical* LC_{50} (C.I.; Slope), µg/g	AVT** LC_{50} (C.I.; Slope), µg/vial
BUR 09	MVPII®	15.4 (10.7-21.8; 0.89)		
COL A 09		87.1 (61.2-127; 1.23)		
COL B 09		19.5 (9.03-41.9; 0.84)		
DEC 09		5.01 (3.18-7.72; 0.72)		
MIL 09		3.33 (1.66-6.30; 1.51)		
SUM 09		161 (75.5-498; 0.63)		
TAY 09		25.0 (14.4-44.5; 0.77)		
TIF 09		4.50 (2.35-8.73; 1.03)		
BUR 09	Spinosad	0.38(0.23-0.64; 2.75)		
COL B 09		0.46 (0.34-0.65; 4.54)		
DEC 09		0.55 (0.34-0.89; 3.52)		
EAR 09		0.80 (0.47-1.32; 2.75)		
BUR 09	Cypermethrin	9.11 (7.91-10.5; 3.61)	----------	3.10 (2.29-4.03; 3.46)
COL A 09		7.10 (5.74-8.79; 2.13)	1.25 (0.85-1.75; 4.17)	2.68 (1.60-3.84; 1.83)
COL B 09		----------	----------	2.42 (1.70-3.18; 4.40)
DEC 09		4.70 (3.27-6.77; 3.08)	----------	3.81 (2.73-5.37; 5.03)
EAR 09		11.7 (9.04-15.9; 4.43)	----------	4.06 (2.39-8.25;2.55)
SUM 09		10.9 (8.65-13.6; 3.27)	----------	3.22 (2.09-4.74; 2.84)
TIF 09		---------	4.93 (0.58-24.7; 0.66)	----------

* larval weight: ~35 mg **Adult Vial Test (males and female adults evaluated)

Table 2. Susceptibilities of field-collected tobacco budworm populations to MVP II®, spinosad (Tracer®), and cypermethrin using treated diet, topical application, and adult vial test (AVT) bioassays—2009.

Colony	Treatment	Diet LC_{50} (C.I.; Slope), ppm	Topical* LC_{50} (C.I.; Slope), µg/g	AVT** LC_{50} (C.I.; Slope), µg/vial
CAM 09	MVPII®	0.57 (0.26-1.05; 0.85)		
DEC 09		1.65 (0.58-3.82; 1.26)		
TIF 09		10.4 (5.82-19.0; 1.02)		
DOO 09		0.78 (0.03-3.66; 0.44)		
TIFC 09		1.95 (1.14-2.69; 1.44)		
COL 09		0.34 (0.19-0.62; 2.09)		
CAM 09	Spinosad	0.68(0.43-1.07; 2.02)		
DEC 09		0.57 (0.36-0.91; 1.28)		
TIF 09		0.37 (0.23-0.58; 1.99)		
DOO 09		1.08 (0.42-2.92; 2.08)		
TIFC 09		0.35 (0.12-0.92; 1.77)		
CAM 09	Cypermethrin	96.6 (71.7-175; 2.71)	93.8 (44.0-380; 1.05)	5.89 (3.24-11.8; 1.93)
DEC 09		25.7 (21.5-30.9; 2.24)	10.3 (7.01-15.3; 2.32)	4.33 (2.47-6.98; 3.33)
TIF 09		17.2 (11.2-27.6; 2.10)	6.50 (4.50-9.25; 1.43)	5.05 (3.76-6.38; 2.57)
DOO 09		78.2 (51.1-132; 4.47)	----------	----------
TIFC 09		----------	80.5 (12.5-1570; 0.76)	----------

* larval weight: ~35 mg **Adult Vial Test (males and female adults evaluated)

Table 3. Mean susceptibilities of bollworm larvae to MVPII®, cypermethrin and spinosad (Tracer®) following a 96 h exposure period using an insecticide-treated diet bioassay--1996-2009.

Year	LC$_{50}$, ppm (Slope)		
	MVPII®	Cypermethrin	Spinosad
1996	38.9 (1.7)	1.40 (2.1)	0.30 (1.6)
1997	68.3 (1.6)	1.31 (2.2)	ND
2003	110 (0.6)*	4.49 (1.8)	0.51 (1.5)
2004	128 (1.1)*	2.63 (3.4)	0.30 (2.1)
2005	122 (0.3)*	1.13 (0.8)	ND
2009	40.1 (0.9)*	8.72 (3.3)	0.54 (3.4)

ND = Not Determined * Data based on tests using neonate larvae

Table 4. Mean susceptibilities of tobacco budworm larvae to MVPII®, cypermethrin and spinosad (Tracer®) following a 96 h exposure period using an insecticide-treated diet bioassay--1995-2009.

Strain	LC$_{50}$, ppm (Slope)		
	MVPII®	Cypermethrin	Spinosad
HRV	ND	1.42 (5.2)	0.38 (1.4)
OPS	0.75 (0.7)	5.01 (3.2)	0.14 (3.3)
OPR	ND	5.48 (2.7)	0.37 (2.2)
PYR	1.23 (1.9)	36.5 (2.1)	0.40 (3.4)
1995	0.95 (1.0)	0.46 (1.1)	0.84 (1.7)
1996	9.63 (1.0)	4.32 (3.0)	0.48 (3.1)
1997	8.68 (1.2)	7.55 (2.5)	0.35 (1.8)
1998	ND	12.1 (1.7)	ND
1999	ND	11.5 (0.9)	0.20 (1.9)
2003	1.00 (0.5)*	33.1 (1.4)	0.52 (1.1)
2004	1.20 (1.6)*	33.1 (1.3)	0.40 (1.6)
2005	3.33 (0.5)*	27.6 (1.2)	0.32 (1.2)
2009	2.66 (1.2)*	52.7 (2.2)	0.61 (1.8)

ND = Not Determined * Data based on tests using neonate larvae

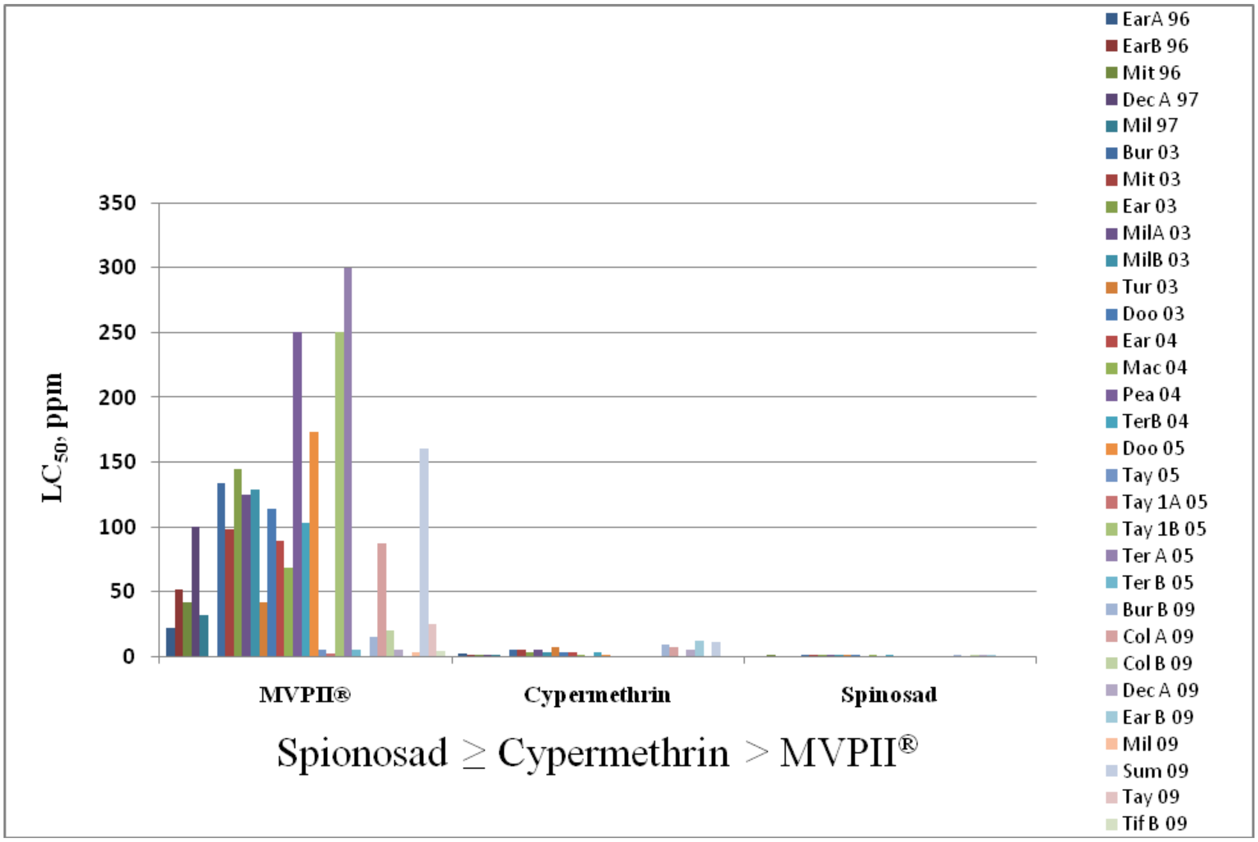

Figure 2. Susceptibilities of field-collected bollworm larvae to MVP II®, spinosad (Tracer®), and cypermethrin using a treated diet bioassay—1996-2009.

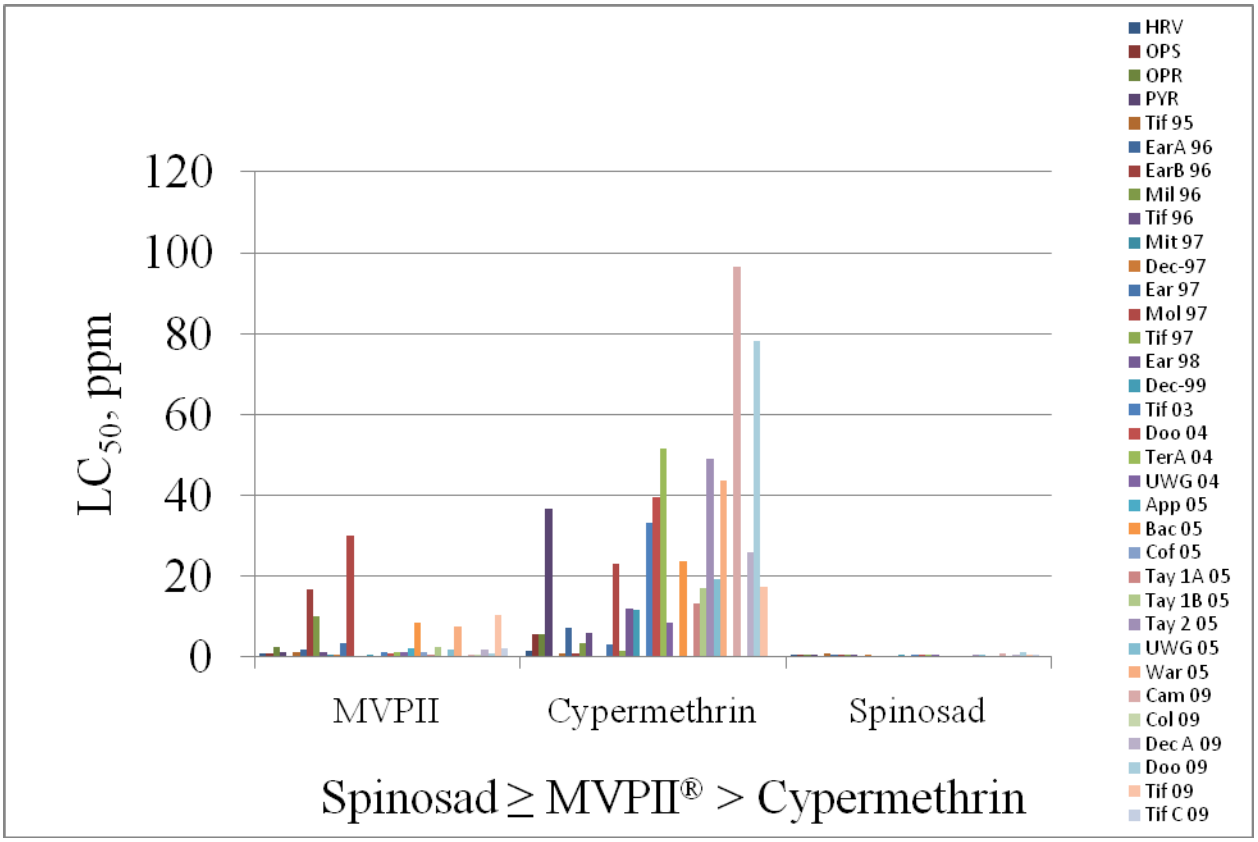

Figure 3. Susceptibilities of field-collected tobacco budworm larvae to MVP II®, spinosad (Tracer®), and cypermethrin using a treated diet bioassay—1996-2009.

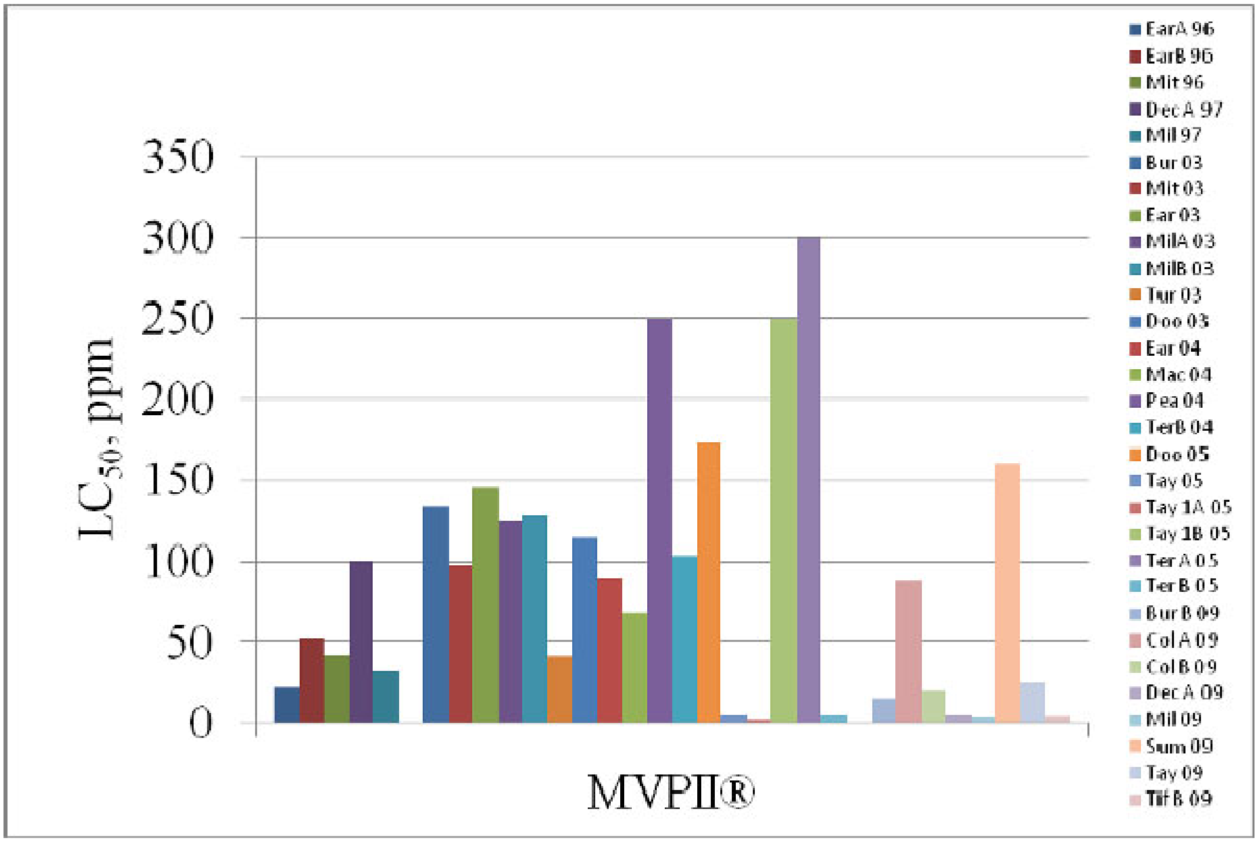

Figure 4. Susceptibilities of field-collected bollworm larvae to MVP II® using a treated diet bioassay—1996-2009.

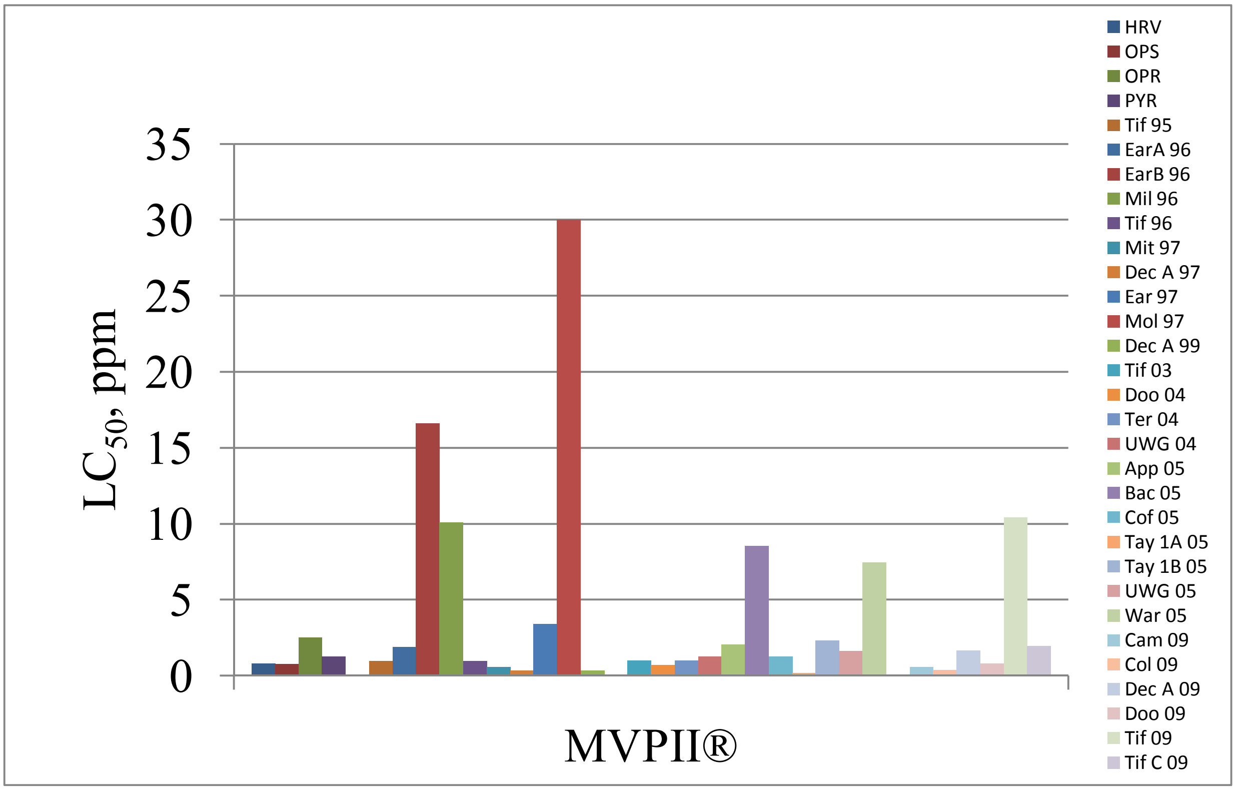

Figure 5. Susceptibilities of field-collected tobacco budworm larvae to MVP II® using a treated diet bioassay—1995-2009.

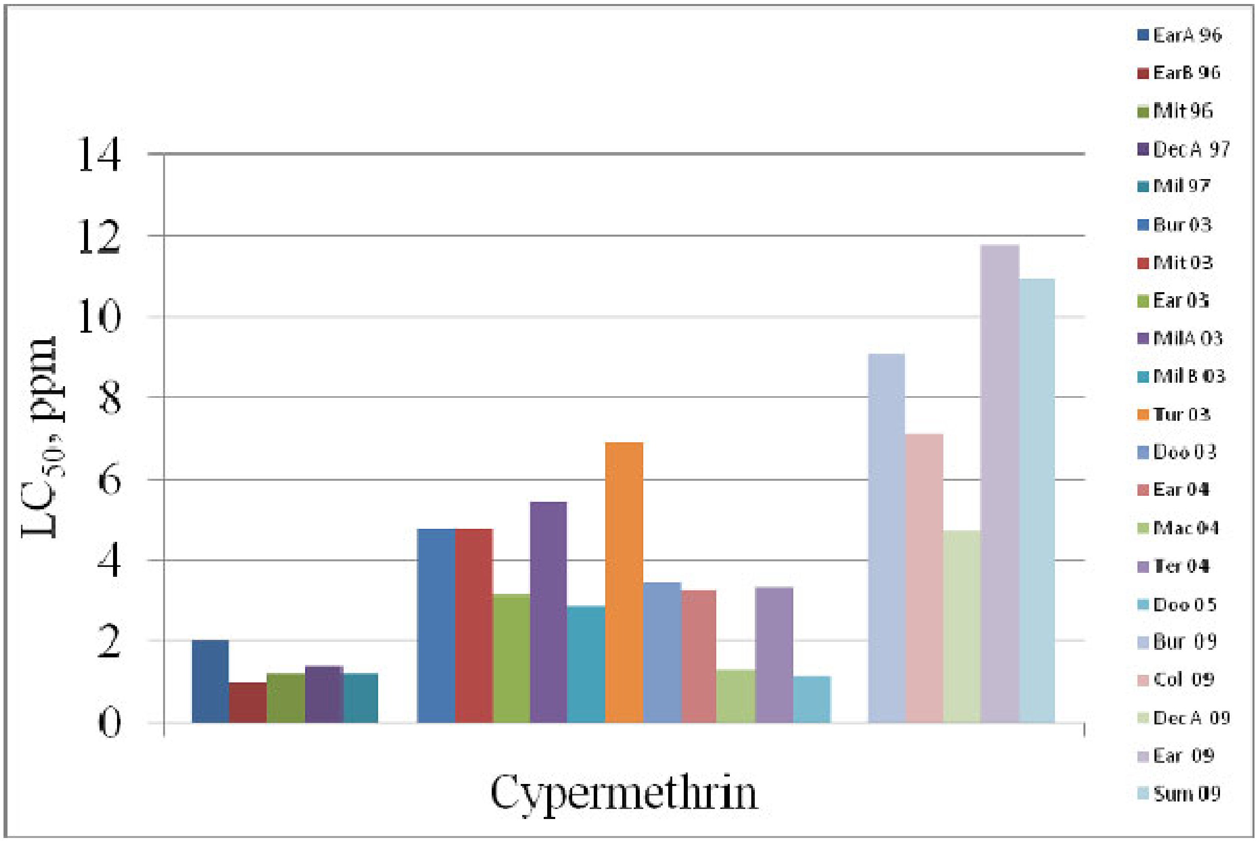

Figure 6. Susceptibilities of field-collected bollworm larvae to cypermethrin using a treated diet bioassay—1996-2009.

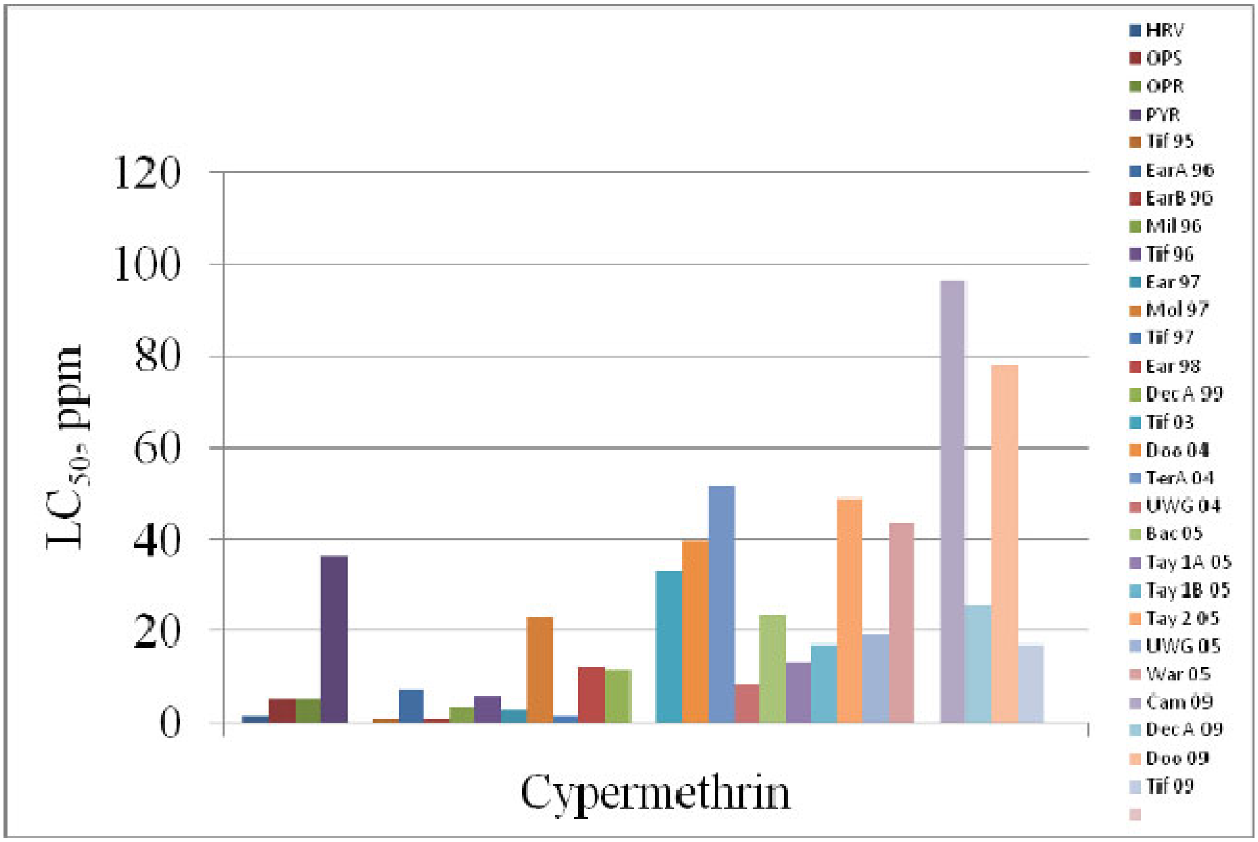

Figure 7. Susceptibilities of field-collected tobacco budworm larvae to cypermethrin using a treated diet bioassay—1995-2009.

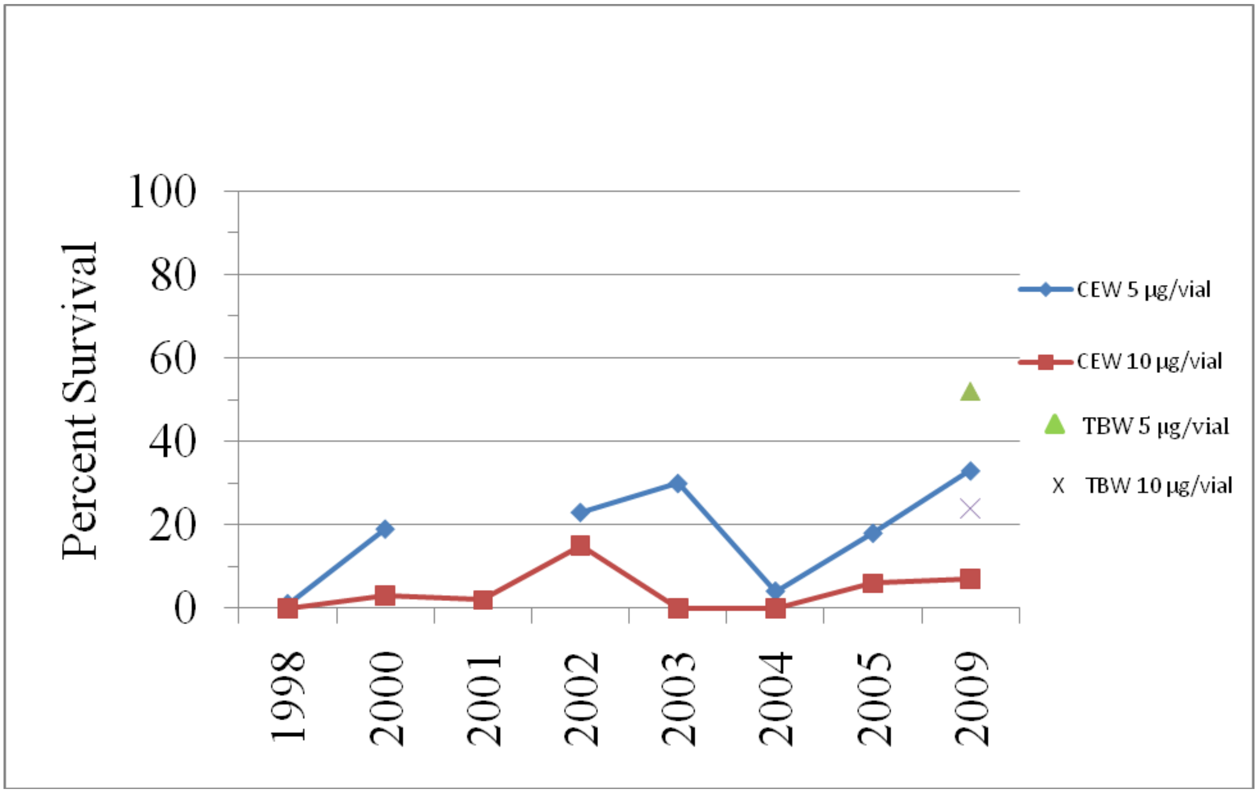

Figure 8. Susceptibilities of bollworm and tobacco budworm adults to cypermethrin using an adult vial test (AVT) bioassay—1998-2009.

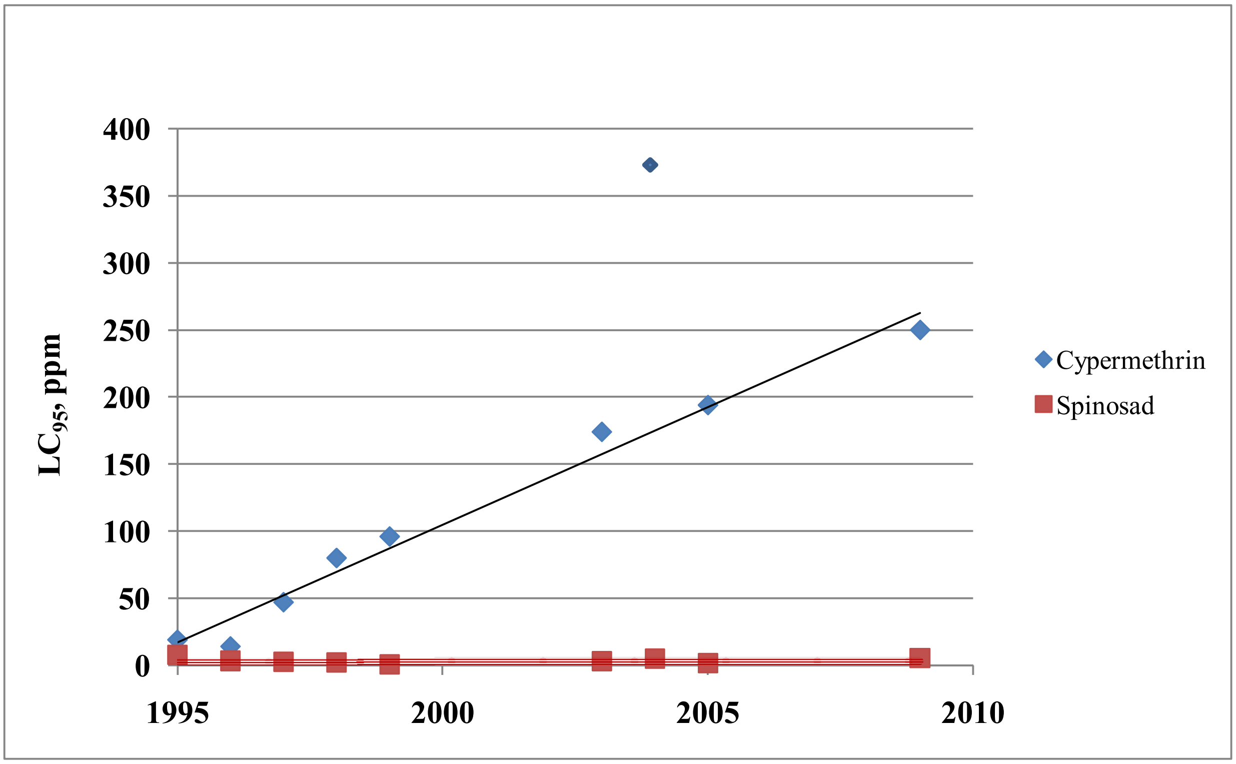

Figure 9. Susceptibilities of bollworm larvae to cypermethrin and spinosad (Tracer®) expressed as the LC_{95} using a treated diet bioassay—1995-2009.

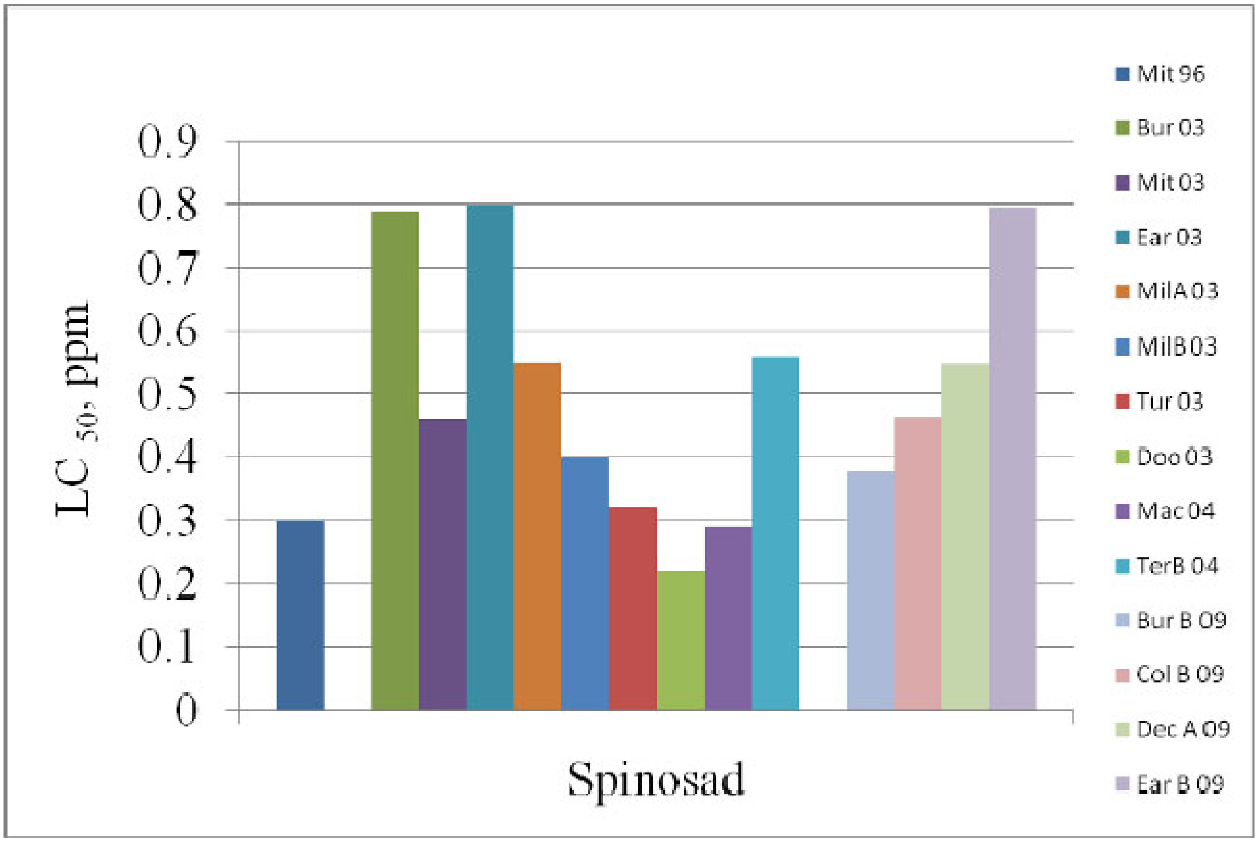

Figure 10. Susceptibilities of field-collected bollworm larvae to spinosad (Tracer®) using a treated diet bioassay—1996-2009.

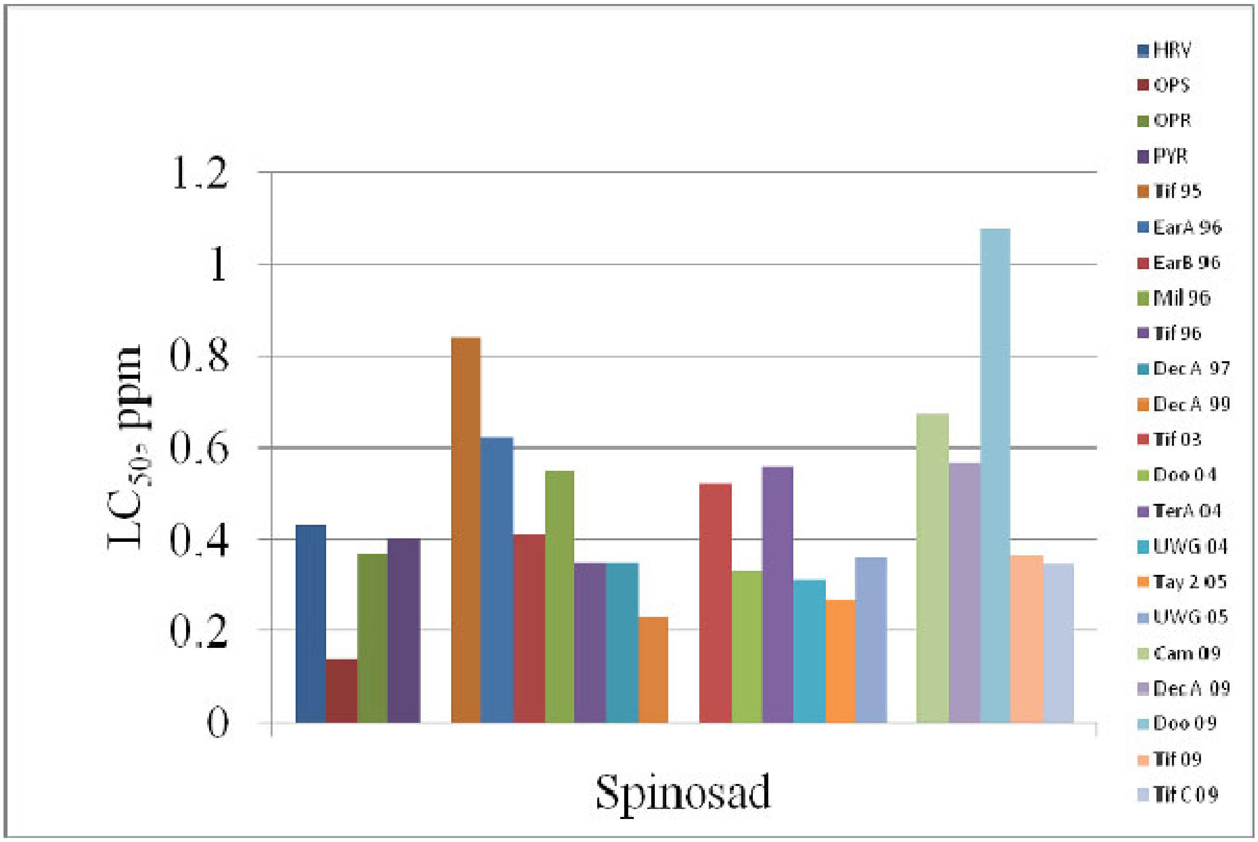

Figure 11. Susceptibilities of field-collected tobacco budworm larvae to spinosad (Tracer®) using a treated diet bioassay—1996-2009.