BEET ARMYWORM (*SPODOPTERA EXIGUA*) RESISTANCE MECHANISMS TO INSECTICIDES IN SOUTHERN TAMAULIPAS, MEXICO

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Abstract

In order to determine the main resistance mechanisms of the beet armyworm to insecticides, bioassays with insecticides mixed with sinergists inhibitors of the related enzymes in the detoxification of insecticides have been made. It was determined that the maximum sublethal dosis of the sinergists piperonyl butoxide (PBO) and butifos (DEF) on the larvae of *S. exigua* was 2.0 (μg/μl). The population of beet armyworm in southern Tamaulipas shows a strong tendency to non-metabolic mechanisms, such as insensitivity to cyclodienes, acetilcolinesterase insensitive to organophosphorus and carbamates and knock down resistance (kdr) to pirethroids. Therefore the management of resistance and the control of this pest with conventional insecticides would be very difficult. Then, the use of products with a different mode of action is needed, as long as their effectivity is determined in the field.

Introduction

The beet armyworm is an insect found in many wild and cultivated plants of the tropical and subtropical regions of the world (Metcalf and Metcalf, 1992).

In Mexico, this pest was found sporadically in cotton, but in 1992 and 1993 it emerged as a consistent pest in the North of Mexico (Obando et al. 1997) In southern Tamaulipas in 1994 more than 10,000 ha of cotton were severely defoliated by this pest, also causing severe damage in chile and onion. In the last two years it has been a serious problem in chiles, tomatoes and cotton. (Teran et al. 1997a).

The control of this pest has been very difficult due to the lack of effectivity of the currently registered products. This fact may be due to the high levels of resistance, found to be higher than 10,000 X to some of these insecticides. (Teran, 1997).

The development of resistance of pests to insecticides makes it necessary to find out its mechanisms. Determining these is of great value in the creation of pest management programs that include the appropriate use of insecticides to minimize the development of resistance. An approach to understand these resistance mechanisms implies the use of sinergists,whose main action is to inhibit enzymes involved in the detoxification of insecticides. Many types of resistance mechanisms can be identified using mortality differntials with a combination of a maximum sublethal concentration of each sinergists with insecticides in bioassays (Prabhaker et al. 1988).

In order to determine the main mechanisms of resistance of beet armyworm in southern Tamaulipas, bioassays with insecticides mixed sinergists were conducted.

Materials and Methods

This study was carried out in the entomology laboratory of the Campo Experimental Sur de Tamaulipas. During October and November of 1994 larvae were collected from cotton and kept on artificial diet (Southland Products Incorporated) until they reached pupal stage. They were transferred to three liter jars and the adults were fed with 10% sugar solution. Larvae, in groups of five, were placed in 1 oz plastic cups with 2 ml of artificial diet. Bioassays with third instar of the first and second generation (25± 3 mg.) larvae were conducted using the topical application method, depositing 1.0 μl of acetone solution in the dorsal part of the thorax with a microapplicator.

A susceptible strain, Zeneca-Dow (Wolfenbarger and Brewer 1993) was compared to the southern Tamaulipas strain.

Technical grade insecticide and sinergists were selected for this study: endosulfan (cyclodiene), chlorpyrifos and azinphos-methyl (organophosphorus), methomyl (carbamate) and permethrin and deltamethrin (pyrethroids). The sinergists were piperonyl butoxide (PBO) as an oxidase inhibitor (mixed-fuction oxidases) and Butifos (DEF) as an inhibitor of esterases and glutathione transferases.

Before conducting the bioassays with insecticides and synergists, the maximum sublethal dosis of each synergist was determined in order to estimate their enzymatic participation on resistance.

Mortality was determined 24 h after treatment. Results were modified by the Abbott formula and analyzed with Polo PC program.

Results And Discussion

Maximum Sublethal Dosis of Synergists on *S. Exigua* Larvae

Many resistance mechanisms can be identified with the combination of a maximum sublethal dosis of the synergist and the insecticide on bioassays (Prabhaker et al. 1988).

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In this study, the maximum concentration of PBO and DEF was 2.0 μg/μl, which was used with different insecticide concentrations, in order to inhibit the highest possible amount of enzymes necessary to estimate their participation on resistance.

Synergism Proportion to Insecticides on *S. Exigua* Larvae

By using synergists on bioassays, one can indirectly identify the participating resistance mechanisms on a population. They are bound to the enzymes that cause resistance, allowing the insecticides to "act freely" which is reflected on the resistance values on resistant insects. The synergism proportion (LD_{50} of the insecticide / LD_{50} of the insecticide plus the synergist) allows to estimate the proportion of the metabolism of an insecticide (Wilkinson 1976). In other words, it indicates how many times the LD_{50} of an insecticide by itself has diminished with respect to the LD_{50} of the insecticide plus the synergist.

Table 1 shows the resistance proportion and the synergism to insecticides on beet armyworm larvae from Southern Tamaulipas.

This population showed a synergism proportion of 1.9 X when endosulfan was mixed with PBO. In other words, the oxidases inhibitor reduced 1.9 times the LD_{50}, from 1.31 to 0.69 μg/μl, which shows a participation of the oxidative metabolism on the resistance to endosulfan. However, a non-metabolic mechanism (cyclodines insensitibity) can be an important factor as well, because the synergist did not eliminate resistance completely.

When using organophosphorus insecticides, chlorpyrifos obtained a synergism of 3.8 X with both PBO and DEF. The oxidases, esterases and glutathione transferases, have very similar participation on the resistance to this insecticide. When testing with azinfos methyl a greater synergism was observed when it was mixed with PBO than with DEF. This demonstrates that a contribution to these high levels of tolerance to this insecticide might be due to oxidases metabolism, without ruling out entirely the possible participation of esterases metabolism. With these insecticides a non-metabolic resistance factor (insensible acetilcolinesterase to organophosphorus) is the most important one on the resistance to this kind of insecticides, due to the fact that separated use of PBO and DEF, did not eliminate completely the tolerance.

Methomil showed a synergism proportion of 8.0 X when mixed with PBO 0.64 to 0.08 μg/μl which demonstrate the importance of the oxidative metabolism. However, a non-metabolic mechanism (Acetilcolinesterase insensitivity to carbamates) can be also an important factor on the resistance to this insecticide.

With the use of pirethroids (permethrin and deltamethrin) a greater synergistic proportion was observed when mixed with PBO than with DEF. These results show that the tolerance to this insecticide group could be mainly to the presence of the oxidative system and in greater proportion to esterases. However, the non-metabolic mechanism knock down resistance (Kdr) is of great importance, due to the fact that the synergists by themselves, could not supress the high levels of tolerance to this group of insecticides.

When comparing the susceptible population with the population from Southern Tamaulipas, high levels of resistance are observed (Table 1). These levels are lowered when the insecticides are mixed with synergists, which demostrates the importance of metabolic mechanisms of the resistance of this pest to conventional insecticides. However, a great tendency towards non-metabolic mechanisms is observed, considered of great importance, because of the lack of effectiveness in supressing high levels of resistance. As a result, the resistance management and the control of this pest with conventional insecticides is very difficult; therefore is necessary the use of insecticides belonging to different toxicologic groups and different mode-of-action, as long as their effectiveness could be demonstrated in the field. This is the case of tebufenozide, methoxyfenozide, hexaflumuron, Spinosad and chlorfenapyr (Teran et al. 1997b).

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Table 1. Resistance proportion and sinergism to insecticides in beet armyworm larvae from southern Tamaulipas, México.

Insecticide	Strain	LD_{50} ($\mu g/\mu l$)	95% confidence interval	PR	PS
Endosulfan	susc.	0.00004	$3(10^{-5})$-$6(10^{-5})$	-	-
	S. Tam.	1.31	0.40-3.02	32 750x	-
+ PBO	S. Tam.	1.69	0.56-0.87	17 250x	1.90x
Chlorpyrifos	Susc.	0.00002	$1(10^{-4})$-$2(10^{-4})$	-	-
	S. Tam.	0.38	0.25-0.55	19 000x	-
+ PBO	S . Tam.	0.10	0.08-0.13	5 000x	3.8x
+ DEF	S. Tam	0.10	0.08-0.13	5 000x	3.8x
Azinfos	susc.	0.002	0.001-0.003	-	-
Methyl	S. Tam	0.888	0.55-1.51	444x	-
+ PBO	S. Tam.	0.11	0.07-0.17	55x	8.0x
+ DEF	S. Tam.	0.26	0.19-0.36	130x	3.4x
Methomyl	Susc.	0.001	0.001-0.002	-	-
	S.Tam.	0.64	0.48-0.85	640x	-
+ PBO	S. Tam.	0.08	0.06-0.10	80x	8.0x
Permethrin	susc.	0.00028	$2(10^{-4})$-$4(10^{-4})$	-	-
	S. Tam	2.27	1.74-2.88	8 107x	-
+ PBO	S. Tam	0.22	0.15-0.33	785x	10.3x
+ DEF	S. Tam.	0.79	0.62-1.09	2821x	2.8x
Deltamethrin	susc.	$7(10^{-8})$	$5(10^{-8})$-$1(10^{-7})$	-	-
	S. Tam	0.17	0.12-0.26	$2.4(10^{-6})$	-
+ PBO	S. Tam	0.04	0.003-0.005	57 142x	42.5x
+ DEF	S. Tam	0.03	0.02-0.03	42 857x	5.6x

susc = susceptibility strain (Zeneca-Dow).
S. Tam = Southern Tamaulipas strain.
PS = resistance proportion.
PS = synergism proportion.