BIOCHEMICAL MECHANISMS OF INSECTICIDE RESISTANCE IN *HELICOVERPA ARMIGERA*

R.V. GUNNING[1], H.A. ROSE[2], G. BARDEN[2] AND M.E. BALFE[1]

[1] NSW Agriculture, Agricultural Research Centre, RMB 944, Tamworth, NSW.
2340.

[2] Department of Plant Pathology and Agricultural Entomology, University of Sydney, Sydney, NSW. 2006

Introduction

Approximately one third of cotton production costs in Australia is spent on pest control. Cotton is attacked by a complex of pests, none more serious than *H. armigera.*. *H. armigera.* resistance to pyrethroids and other insecticides is a major threat to cotton production in Australia.

At the onset of *H. armigera* pyrethroid resistance in 1983, three resistance mechanism were identified, namely; nerve insensitivity (*Super Kdr)*, reduced pyrethroid penetration through the cuticle (*Pen*) and a factor synergisable by piperonyl butoxide (*Pbo Factor*). *Super Kdr* was the most important cause of resistance, the other mechanisms having a minor role. However, by 1986, the *Pbo Factor* had replaced *Super Kdr* as the major mechanism responsible for pyrethroid resistance. The disappearance of the *Super Kdr* gene from field populations was probably caused by reduced pyrethroid use. It was assumed, but without any real evidence, that piperonyl butoxide acted as a monooxygenase enzyme inhibitor.

The suspected importance of pyrethroid metabolism in resistant *H.*

armigera, made the investigation the biochemical basis of pyrethroid resistance in *H. armigera* imperative. In this background paper, we summarise our findings.

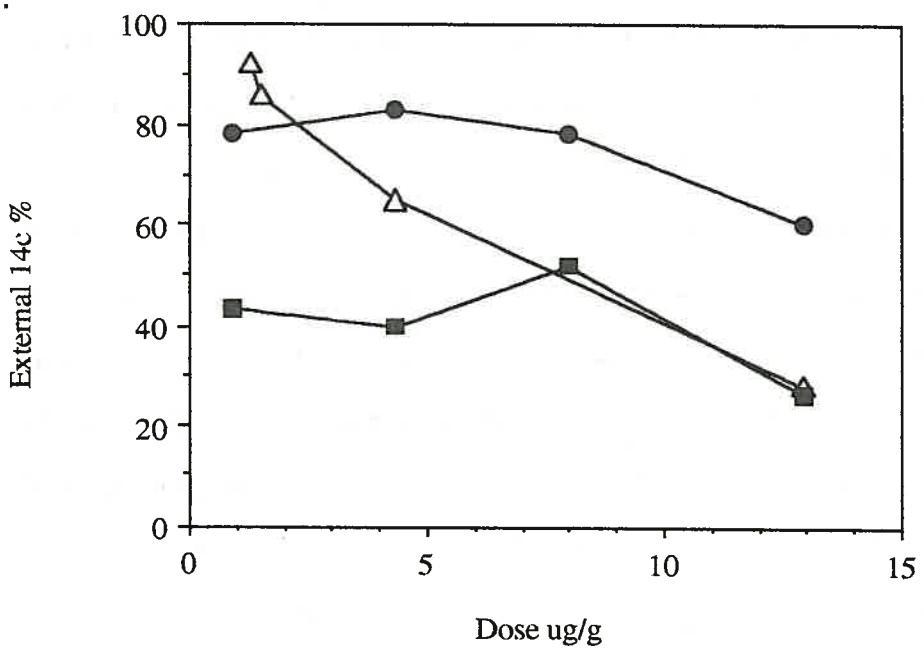
Results

1. Metabolism of fenvalerate by *H. armigera*

We studied metabolism of pyrethroids in resistant and susceptible *H. armigera* larvae using ^{14}C labelled fenvalerate. Fenvalerate and metabolites were extracted from the insect tissues by solvents. We found that while fenvalerate penetrated the cuticle of the susceptible strain much more rapidly than the resistant (Fig.1), there was little evidence of a greatly increased pyrethroid metabolism by the resistant strain.

Figure 1

The effects of insecticide dose and piperonyl butoxide on 3h penetration of fenvalerate through the cuticle of resistant and susceptible *H.armigera*. Results are expressed as the percentage of C^{14} fenvalerate dose remaining on the cuticle. Susceptible (■), resistant (●) and resistant strain pre-treated with piperonyl butoxide (△).



2. The mode of action of piperonyl butoxide / pyrethroid synergism.

We have found piperonyl butoxide facilitates the action of pyrethroids against resistant *H.armigera.* in two ways. Firstly, it aids pyrethroid penetration through the resistant cuticle (Fig. 1). Secondly, piperonyl butoxide may inhibit the metabolism of pyrethroids by monooxygenases. Penetration resistance and the Pbo *Factor* cannot be genetically separated (in collaboration with J. Daly, CSIRO Entomology).

3. Enzyme studies

Two enzyme systems, the mono-oxygenases and esterases, are known to metabolise pyrethroids in other insects. Our biochemical studies suggest that both are involved with *H.armigera* resistance.

We have successfully modified mono-oxygenase enzyme assay techniques for *H. armigera* , a new technique has been developed to use whole, small larvae for enzyme assays. Our results show that resistant *H.armigera* have slightly elevated monooxygenase activity compared to the susceptible strain, (~1.5 fold). Both resistant strain enzyme levels (cytochrome P450) and enzyme activity (as measured by the metabolism of aldrin to dieldrin) were a little higher than for the susceptible strain (Figs 2 and 3).

We have found an esterase which characterises all pyrethroid resistant *H.armigera.* and which is absent in susceptible individuals. The esterases were studied using electrophoretic techniques. The esterase frequency in field populations was very similar to pyrethroid resistance frequencies, as estimated by bioassay. We consider that the biochemical test to be a very reliable resistance detection method. Synergist studies also suggest that esterases are involved in the detoxification of pyrethroids by resistant *H. armigera*..

Figure 2.

Monooxygenase levels in larvae of pyrethroid resistant and susceptible strains of *H. armigera*

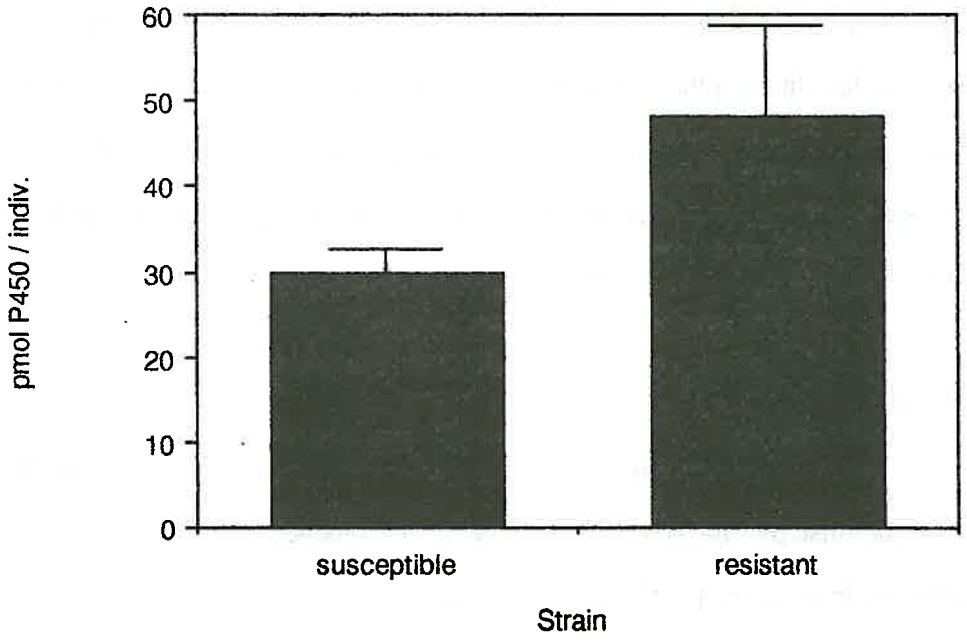
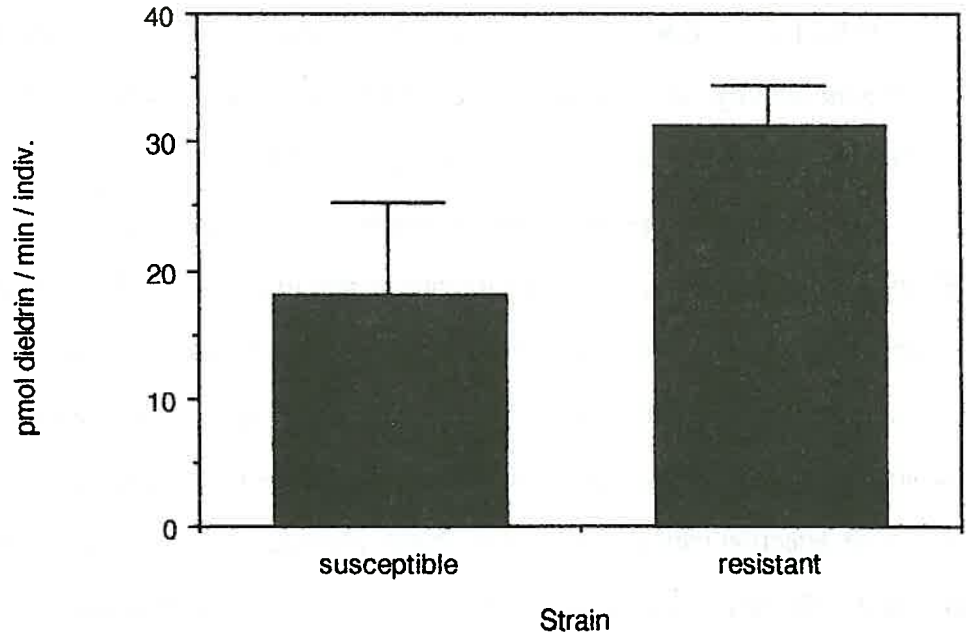


Figure 3.

Monooxygenase activity (as measured by epoxidation of aldrin to dieldrin) in larvae of pyrethroid resistant and susceptible strains of *H. armigera*.



4. Nerve Insensitivity (*Super Kdr*) Resistance Detection

We have developed electrophysiological methods to directly detect pyrethroid nerve insensitivity. *H.armigera* populations are monitored each season to determine *Super Kdr* resistance gene frequencies because it is essential to ensure that resistance management decisions do not favour the re-selection of *Super Kdr* . Since 1987, *Super Kdr* gene frequency has been at a virtually undetectable level in pyrethroid resistant *H.armigera* populations.

5. Acknowledgments

The support of the Australian Cotton Research Council and the Winston Churchill Memorial Trust of Australia is gratefully acknowledged. We are indebted to Miss N. A. Coleman, Mr R. V. Hall for technical assistance and to Dr A.L. Devonshire and Mr G. D. Moores, Insecticides Department, Rothamsted Experimental Station, U.K.

6. References

Gunning, R.V., Easton, C.S., Balfe, M.E. and Ferris, I.G. (1991).- Pyrethroid resistance mechanisms in Australian *Helicoverpa armigera.* *Pesticide Science* **33** : 473 - 490.