



Australian Government

**Cotton Research and
Development Corporation**

FINAL REPORT

**Insecticide Resistance in *Helicoverpa* spp. and the role
of IPM/Area Wide Management in Resistance
Management**

**CRDC Project No. DAN 173C
(incorporating CRDC 228C)**

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EXECUTIVE SUMMARY

Insecticide resistance in the cotton bollworm *Helicoverpa armigera* has been one of the greatest limitations to successful pest control and economic cotton production in Australia. The introduction of transgenic cotton has reduced the dependence on insecticides for control of this pest, however sprayed conventional (non Bt) cotton is still a viable cropping option in its own right as well as acting as a refuge for transgenic cotton. It is imperative therefore that resistance is monitored and managed to ensure insecticides remain an effective option now and in the future.

This research project focussed on the incidence and cause of insecticide resistance by *Helicoverpa armigera* and to a lesser extent *H. punctigera*. The objectives included continuing the insecticide resistance monitoring program for *Helicoverpa* spp., investigating resistance development within a *Helicoverpa* spp. population, assessing the impact of AWM/IPM on resistance management, and formulating and promoting improved resistance management guidelines. Key findings include:

- Detection of low frequency resistance to the newer IPM compatible chemistries used against *H. armigera* including indoxacarb (Steward[®]) and emamectin benzoate (Affirm[®]).
- Indication that resistance to spinosad (Tracer[®]) and chlorfenapyr (Intrepid[®]) which had been increasing in previous years has decreased to low frequencies.
- Resistance is still present at variable frequencies in field populations to those chemistries that *H. armigera* are known to have developed resistance to, including endosulfan, pyrethroids, methomyl and organophosphates (profenofos).
- Very low frequency detection of resistance to endosulfan, pyrethroids and abamectin (Agrimec[®]) by *H. punctigera* in some valleys.
- Information recorded on species complex across time and space within cotton growing regions which has implications for the monitoring project and also for resistance management.

This information was used in assessment of current strategies and formulation of new strategies for managing insecticide resistance by the TIMS committee. The results and general resistance management tactics and information were promoted to the industry both verbally and written.

Various external factors inhibited effective analysis of the effects of AWM and IPM practises on resistance management. These included drought effects, low *H. armigera* pressure and high *H. punctigera* pressure resulting in several farms in the Macintyre Trial in 2003/04 and 2004/05 exiting the trial to use harder chemistry on this pressure.

Attempts to study resistance development to indoxacarb and emamectin benzoate, key IPM compatible chemistries about which little is known in regard to resistance, were unsuccessful. Problems encountered however have implications for resistance management, with further research to be undertaken.

FULL REPORT

1. Background

Helicoverpa armigera, and to a lesser extent *H. punctigera*, are the primary insect pests of the cotton industry in Australia. Insecticides have until recently provided the key management tool in the control of these pests. The introduction of transgenic cotton, firstly Ingard® and now Bollgard II® have provided an opportunity for growers to reduce their insecticidal inputs. Insecticides however remain the primary method of pest control in conventional non-transgenic cotton crops grown as either refuges for Bollgard II® or as crops in their own right. The biggest threat to their continued efficacy is the development of resistance, hence the importance of resistance related research incorporating monitoring and management.

Resistance monitoring and active management has been conducted in the Australian Cotton Industry for over twenty years, since pyrethroid resistance was observed at the field level in 1983. Such monitoring and management has ensured these chemicals are still viable options when used appropriately today. *Helicoverpa* spp. monitoring and management has evolved to include twelve chemicals in ten groups, encompassing the older less selective chemistries as well as the newer less disruptive insecticides utilised as part of an integrated pest management approach. Resistance monitoring and the formulation of management tactics is conducted for all cotton growing valleys and areas across Eastern Australia.

The expansion of Area Wide Management (AWM) groups and the adoption of IPM guidelines in many districts have raised the question, 'how effective are AWM strategies in limiting the development of resistance?' In the Macintyre Valley growers have suggested that if AWM and IPM strategies can be used to significantly reduce insecticide use, does this provide the opportunity for a more flexible resistance management strategy. Such a strategy was trialled by four AWM groups in the Macintyre Valley over three seasons from 2002/03 with resistance monitoring data collected as part of assessing and answering this question.

In adopting IPM strategies, growers may delay the use of insecticides until crop damage thresholds are reached. This may result in insecticides being applied to *Helicoverpa* populations that contain a higher proportion of small and medium larvae than wouldn't be present if lower thresholds were used. While growers may reduce overall insecticide use, one concern is that spraying larger larvae may reduce the effectiveness of insecticides and thereby increase the risk of resistance developing. To further assess the merit of AWM and the use of IPM strategies in resistance management it would be of use to determine the impact of using higher insect thresholds on the mortality of susceptible and both homozygous and heterozygous resistant populations.

This research project was designed to monitor resistance across all cotton growing regions as an indicator of emerging resistance problems and as assessment of the strategy taken to manage resistance. The monitoring program also allowed for assessment of the impact of AWM/IPM practises in the Macintyre Valley. Additional research investigating the effects of higher thresholds on insects resistant to some of the IPM compatible chemistries was included utilising field collected insect material.

evolved. Abamectin (Agrimec[®]) was also included as a chemistry that specifically targets *H. punctigera*.

Impact of AWM / IPM practices on resistance management

A trial was conducted in the Macintyre Valley involving four AWM groups incorporating IPM principles. A less restrictive IRMS was followed in this trial to allow for maximum effectiveness of beneficial preservation. Intensive insecticide resistance monitoring was undertaken in this trial using egg collections from dedicated egg collectors. Collections were taken from farms across the four area wide management groups as well as from non trial farms. Insecticide resistance results were used to investigate if the less restrictive IRMS had any impact on resistance development when incorporated with beneficial preservation.

It was originally anticipated that this study would include another AWM group in the Lower Namoi, the TRAMS group. This area was omitted as inadequate collections were obtained from this area, particularly in 2003/04 when effects of the drought resulted in greatly reduced planting in this area.

In addition to this trial, insect samples supplied from across all cotton growing regions for resistance monitoring were sent to a research group at the University of Queensland for use in moth dispersal studies (Project UQ35c). These samples were dead larvae after they had been tested for resistance and would have otherwise been disposed.

Laboratory Resistance Studies

Two approaches were taken to initiate resistance studies in *H. armigera*:

1. Insects found to be resistant to key chemistries through the monitoring program were reared through to adults and single pair mated with lab reared insects to establish a resistant colony, with further outcrossing to establish some genetic diversity within a resistant colony.
2. *H. armigera* colonies reared from field collections were selected with varying doses of specific insecticides to establish resistant colonies for further studies.

Both approaches resulted in limited success, as discussed in the results and discussion (Section 4), with further development and threshold studies not able to be conducted.

4. Results and discussion

4.1 Insecticide Resistance monitoring

Insecticide resistance monitoring was successfully conducted for the 2002/03, 2003/04 and 2004/05 seasons. Total data collected from field samples for monitoring purposes includes information on overall species complex at the time of sampling. This information is used in the monitoring program and also has implications for resistance management, with *H. punctigera* easily controlled due to its susceptible status. Over the last three seasons *H. punctigera* pressure has been in most areas typically high during late spring/early summer, unusually continuing through to January in some areas in 2003/04 and 2004/05 (Tables 1a-c.). While *H. armigera* eggs sampled in 2002/03 indicated this to be the dominant species yielding moderate pressure in the mid-late part of the season, overall pressure on cotton throughout the season in 2003/04 and 2004/05 was low. The exception to this high *H. punctigera* and low *H. armigera* pressure was observed in the Darling Downs, where individual samples were observed to vary greatly in their species complex. This variation can be attributed to the diversity of cropping systems in this area, and individual field proximity to these other crops.

The implications for the resistance monitoring program of the species complex observed were that the low numbers of *H. armigera* made accurate analysis and determination of resistance incidence and trends difficult. Reasonable numbers of *H. armigera* were collected in some areas 2002/03 (10,500) to give an idea of the incidence of resistance for some of the key insecticides. Despite extensive collecting in 2003/04 and 2004/05 however, with over 500 and 390 samples processed respectively, the numbers of *H. armigera* tested across all areas across the season was greatly reduced, totally only 7500 in 2003/04 and 3600 in 2004/05, with particularly low *H. armigera* pressure in the latter season. Conversely, total numbers of *H. punctigera* collected were 2800 in 2002/03, 12,000 in 2003/04 and 19300 in 2004/05. Analysis of the resistance status of *H. armigera* within the cotton industry therefore has focussed on detecting any resistant individuals in the testing and reporting results in terms of whole numbers tested, with trend observations and statistical analysis between areas, years and within the season almost impossible and moreover meaningless.

Overall resistance monitoring results for *H. armigera* for the three seasons are summarised in Appendix 1: Tables 2a-c.

Methomyl

Methomyl was not included in monitoring in 2002/03 because resistance was known to be established and widespread throughout all cotton growing regions from previous resistance monitoring projects, with insect material utilised to concentrate on the newer softer chemistries most used. It was re-introduced in 2003/04 following requests to continue to monitor where possible all chemistries in use. The monitoring results in 2003/04 confirmed that methomyl resistance was still present in *H. armigera* populations at variable but high frequencies (Figure 8a), with limited insect material resulting in little monitoring in 2004/05.

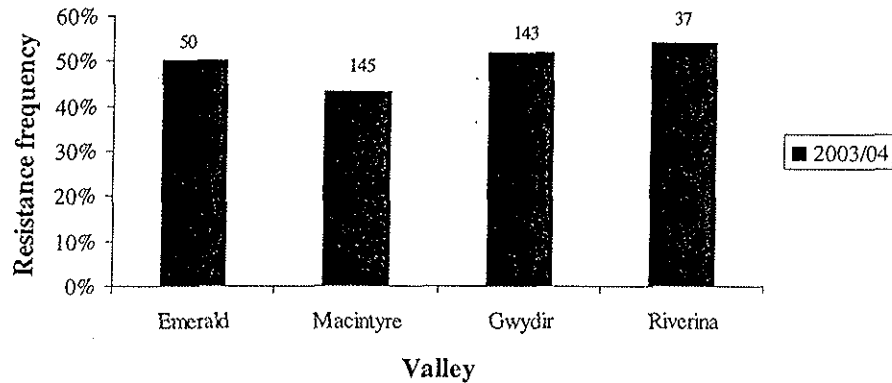


Figure 8a: Methomyl resistance frequencies for several valleys in 2003/04. Numbers on the graph indicate total larvae tested.

Ovicides – Amitraz and Methomyl

No ovicide resistance has yet been detected for either amitraz or methomyl (ovicidal rate) in *H. armigera* in cotton growing regions of Australia.

4.2 Assessment of AWM/IPM practises on resistance management

Assessment of the impact of AWM and IPM practises on resistance management was made difficult over the last three seasons for a variety of factors. In 2003/04 drought effects resulted in one area that was to be used, the TRAMS area wide management group in the Lower Namoi being omitted due to very little cotton being grown that year. In the Macintyre Valley where four active area wide management groups were following a trial IRMS, several farms in both 2003/04 and 2004/05 broke the strategy and reverted to the standard because very high *H. punctigera* pressure resulted in more sprays than anticipated, with the broad spectrum pyrethroids and organophosphates providing a cheap and effective control measure. In addition, in both years, while there was high *H. punctigera* pressure, there was low *H. armigera* pressure, particularly in 2004/05. Despite extensive egg collections, there were insufficient *H. armigera* available for testing differences in resistance between different groups and those areas/farms not in the trial, and coupled with farms breaking the strategy, any benefit of AWM or IPM practises could not be assessed.

Figure 11 compares insecticide resistance frequencies in trial areas with non trial areas for 2003/04. This graph show the low numbers of *H. armigera* tested, and the inability to distinguish a clear difference between trial and non trial areas. Data for 2004/05 are not provided due to the exceptionally low numbers of *H. armigera* tested (approx. 330 compared with 1260 in 2003/04).

Part of the difficulty in assessing the impact of AWM/IPM practises within a small area is moth movement within valleys and between valleys. As part of investigating the extent of moth movement in 2003/04 and 2004/05, larvae that had been utilised in resistance monitoring were supplied to a group at the University of Queensland for movement studies, CRDC Project UQ35c. This project has investigated moth movement between valleys and also within valleys between different crops, showing that moths can move very large distances, certainly beyond the confines of an AWM group or valley. Contribution to this project has resulted in one publication currently undergoing review (see 9.1 Publications).

5.4 Formulate and promote improved insecticide resistance management strategies incorporating secondary pests and other broad acre crops.

Formulation and promotion of improved insecticide resistance management strategies is vital in ensuring appropriate resistance measures are applicable in an ever evolving industry. This objective was completed through involvement with the TIMS committee in formulating the annual IRMS, with further extension to the industry via the Resistance Roadshow, grower and consultant meetings, and written extension articles (see Section 9).

6. Cotton R&D Corporation outputs

Resistance monitoring and formulation of management strategies, including the role of AWM/IPM strategies in resistance management, link strongly into the CRDC output of sustainability. This project has provided information on *Helicoverpa* spp. insecticide resistance in the field for use in directly assessing current insecticide resistance management strategies as well as formulating new ones to minimise the development of resistance. Insecticide resistance is one of the greatest threats to successful insect control and sustainability of the cotton industry in which insect control is a major cost.

This project also contributes to the CRDC output of profitability & competitiveness. Chemical control of insect pests is a major cost to cotton growers. Resistance can result in increased sprays, and therefore decreased profits, and overall decreased competitiveness on world markets. In addition, there are great costs involved in developing new chemistries often required if current ones lose their efficacy because of resistance. This monitoring project allows for resistance to be detected before it is observed at the field level, enabling management strategies to be modified and implemented if necessary with a view to retain all chemical options available for effective insect control.

8. Further research and development

As long as insecticides with action against *Helicoverpa* spp. are utilised in cotton, resistance will remain a threat to their future efficacy. With the widespread adoption of Bollgard II in 2004/05 and anticipated for the 2005/06 season, insecticides are no longer the primary defence against *Helicoverpa* spp. for the majority of cotton being grown. They do however remain the key defence in conventional cotton and it is important that their use continues to be managed in a way that minimises the development of resistance. Further, future use of Bollgard II is unknown, and may overall or in some seasons or particular areas not be the dominant cropping system, so for the foreseeable future conventional sprayed cotton will continue to have a place in cotton production in Australia. It is important therefore that resistance monitoring and management continue for the industry to have confidence in the chemical options available.

Resistance management is benefited by information on the mechanisms conferring resistance to insecticides, and with the exception of chlorfenapyr and spinosad, little is known about this for the key IPM compatible chemistries, including indoxacarb and emamectin benzoate. Future research shall include investigation of these resistance mechanisms and their inheritance for use in developing effective resistance management strategies. Incorporated in these studies shall be investigation of cross resistance to other insecticides as well as to the Bt toxins in Bollgard II, which if present is a major threat to resistance management for both insecticides and Bollgard II.

In addition to future research and formulation of resistance management techniques, promotion of resistance management strategies and dissemination of resistance results will remain a key objective. It is important that all involved in the cotton industry are aware of the resistance issues that affect successful pest control and the strategies that ensure best management of these pests. It is also important that the basis for resistance management techniques are understood, particularly where multiple pest complexes exist, and these shall continue to be explained and promoted through both verbal and written communication.

10. Likely impact of research outcomes (cost/benefit)

The insecticide resistance monitoring results have been utilised by the industry for over twenty years in detecting resistance and implementing strategies to prevent or delay its development. The benefit of such a project is evident through the preservation of the older chemistries such as endosulfan and the pyrethroids which have a history of resistance in *H. armigera* dating back over 20 years, however they are still used today. In more recent times, the monitoring program has detected increasing resistance to spinosad, with measures taken following this detection to restrict its use to ensure its continued efficacy. Such a project coupled with formulation and promotion of resistance management strategies has proved the benefits of such research outweigh the associated costs.

The results of the resistance monitoring project in the last three years have not provided strongly conclusive results regarding the exact frequencies of resistance to all the chemistries used against *Helicoverpa* spp. The research has however satisfied the requirement of detecting the presence of resistance, particularly to the newer IPM compatible chemistries which have established themselves as an integral part of chemical pest control in cotton, namely indoxacarb, emamectin benzoate, abamectin and spinosad. This has enabled the management strategies in place for these chemistries to be assessed and modified where necessary, impacting directly on the industry.

Additional species complex information has also been gained from the monitoring program in the last three seasons which has major implications for both resistance and pest management. A shift in the species present in the field may influence chemical use in the future, particularly if *H. punctigera* becomes the dominant species, with the annual migration of this species making it a lower risk for resistance development. Collection of this information is therefore vital for ensuring resistance management tactics are relevant and effective.

