



DAQ 122C

# Development of Novel Pest Management Options for Cotton in Central Queensland

Report prepared for  
The Cotton Research and Development Corporation

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September 2005



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## Executive Summary

The primary purpose of DAQ 122C was to investigate alternative pest management options under central Queensland conditions with a view to supporting and expanding the role of area wide management. The project accomplished a broad suite of research spanning the use of assassin bugs as applied biological control agents through to investigating the potential of using attract and kill technology (Magnet®) for the management of *Helicoverpa* and *Bt* resistance.

The threat of ascochyta leaf blight disease gaining a foot-hold within the CQ chickpea industry provided an impetus to re-investigate alternative legume options for spring trap cropping. Field peas (cvs Alma or Glenroy) were found to be the most effective legume for *Helicoverpa* trap cropping and were unique in that the majority of eggs laid on it perished due to dislodgement from the waxy foliage making it a partially self-maintaining trap crop. The adoption of field peas for spring trap cropping circumvents any problems associated with chickpea leaf blight disease.

The use of refuges to augment natural enemies in cotton was investigated, with niger identified as being attractive to the broadest range of natural enemies. However, the use of refuges was found to be an inconsistent method for augmenting natural enemies commonly associated with cotton systems. The patterns of natural enemy abundance observed in the refuge treatments were probably more closely associated with variations within the surrounding environment than any in-field modifications to vegetative biodiversity via the provision of refuges. These results suggest that the key to reliably predicting and augmenting endemic populations of natural enemies within cotton farming systems may potentially exist in developing a more refined understanding of the interactions that occur between beneficial species and the broader natural environment.

The assassin bug, *Pristhesancus plagipennis* was demonstrated to be an effective biological control agent for *Helicoverpa* and mirids in conventional cotton. Assassin bugs were found to be innately tolerant of Steward®, Admiral®, Tracer®, Regent® and Affirm®, as well as NPV and *Bt* biopesticides. The compatibility of these products with assassin bugs in the field is highly desirable and our experiments demonstrated that the strategic integration of these insecticides with releases of assassin bugs provided effective pest management with 50% less insecticide whilst maximising crop yield. Should assassin bugs become commercially available in the future, significant potential exists to utilise them within a low spray IPM program for conventional cotton.

Population dynamic studies demonstrated that Silver Leaf Whitefly, *Bemisia tabaci* Biotype (B) has become a regular pest of cotton within the Dawson Valley, infesting crops each year during late November and December and peaking in abundance during February. Sampling has also determined that native *Eretmocerus* parasitoids have become well established with their increased abundance correlating with a reduction in regional whitefly populations on cotton. Mean parasitism rates recorded in cotton have risen from 15% in 2003 to 87% in 2005 whilst applications of Insect Growth Regulators (IGRs) for whitefly control have decreased from 40% to <5% of fields over the same period. The reduction in IGR use constitutes savings of \$100 per hectare not treated. Our data suggest that it is imperative to ensure the compatibility of pest management practices with whitefly parasitoid conservation and that this consideration will continue to influence future pest management research.

DAQ122C identified the potential for using Magnet® as a regional moth busting tool for targeting last generation *Helicoverpa* emerging from Bollgard® fields. Such an approach could supersede the requirement for summer trap cropping as part of the CQ *Bt* resistance management strategy and will be the focus for future research. Refinement of the current *Bt* resistance management strategy is of critical importance to ensuring the prolonged viability of pesticide reducing transgenic technologies.

Best pest management practices and novel research outcomes were promoted to CQ cotton growers throughout DAQ122C via several interactive field days each season and the provision of significant technical support and input for three IPM short courses conducted in the Dawson Valley and Central Highlands regions of CQ.

## Summary Details

CRDC Project Number: **DAQ122C**

Annual Report: ☒ Due 30-September

Progress Report: ☐ Due 31-January

Final Report: ☐ Due 30-September  
(or within 3 months of completion of project)

**Project Title: Development of Novel Pest Management Options for Cotton in Central Queensland**

Project Commencement Date: 1/7/2002

Project Completion Date: 30/6/2005

Research Program: Crop Protection

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## Background for DAQ 122C

At the commencement of DAQ 122C, previous research conducted in central Queensland had broadly supported the philosophy of Area Wide Management and highlighted the need to develop new novel tools and techniques to further manipulate pest populations at both the local and regional scale. Under this backdrop DAQ 122C commenced with a charter to develop novel in field control techniques such as the use of assassin bugs for biological control of *Helicoverpa* and mirids through to the development of more effective trap crops for regional *Helicoverpa* resistance management.

However, the circumstances in which the central Queensland industry found itself changed considerably during the early phases of DAQ 122C. Just as the project commenced, silver leaf whitefly became a major pest of cotton. The outbreak of this pest was to have direct consequences on the pest management practices conducted for it and other pest species. Drought was also a companion during the first two years of DAQ122C with resultant irrigation water shortages placing renewed emphasis on water use efficiency and gaining more “crop per drop”. The need for water use efficiency resulted in area-wide management strategies such as trap cropping being increasingly questioned by growers in terms of resistance management efficacy. Thirdly the introduction of Bollgard® II varieties has significantly shifted management emphasis away from *Helicoverpa* spp. to other pest insects previously considered to be of a secondary nature. The widespread adoption of Bollgard® varieties has also placed renewed emphasis on ensuring that Bt resistance management strategies are adequate for the longer term. Each of these events caused DAQ122C to take a different research path to what was initially proposed in 2001 and resulted the substitution of several key initiatives that were commenced (better trap crops, beneficial insect refuges and assassin bugs for biological control) for research targeted towards secondary pest management in relation to silver leaf whitefly and better tailoring Bt resistance management strategies to CQ conditions. This report is a culmination of the results from a broad range of research initiatives some of which will be continued under the new CRDC funded Central Queensland Farming Systems Research project.

## DAQ 122C Project Objectives and Achievements

Project Objective	Achievements
Evaluate recruitment and origin patterns of <i>Helicoverpa</i> populations within the Dawson Callide in collaboration with UQ Centre for Diagnostics.	<i>Helicoverpa</i> were extensively sampled from throughout the Dawson Callide during the 02/03 and 03/04 seasons and sent for analysis to the Centre for Diagnostics at University of Queensland. The results of this research will be reported by Dr Kirsten Scott from the Centre for Diagnostics. A paper detailing the first year of sampling results has been published in the Australian Journal for Agricultural Research 2003.
Implement and assess field pea as an alternative spring trap crop in central Queensland.	This project identified field peas as a more effective spring trap crop for central Queensland conditions. Compared to chickpeas, field pea attracted five fold higher <i>Helicoverpa</i> egg densities, was natural enemy friendly and ascochyta leaf blight disease resistant. Field peas were adopted by some growers within the Dawson (not Emerald due to water shortages) during the 03/04 season. However ongoing dry conditions for the 04/05 season combined with an inability to substantiate the relative impact of trap cropping has led to a complete lapse in spring trap cropping in central Queensland.
Evaluate the potential for using natural enemy refuges in central Queensland cotton crops.	The potential of several refuges to attract and breed natural enemies was tested under central Queensland conditions. Of the refuges investigated, the data suggested that Niger had the greatest potential to attract and generate beneficial insects. Further experiments were conducted using Niger as a companion refuge plant in cotton to test whether this plant could attract and impart beneficial insects to neighbouring cotton. In this experiment Niger was found to impart few additional beneficial insects when used as a companion plant compared to plots of cotton alone.
Evaluate the use of assassin bugs as a biological control tool for IPM in cotton.	Field trials conducted during 02/03 and 03/04 demonstrated that releases of assassin bugs caused significant reductions in <i>Helicoverpa</i> and mirid populations in conventional cotton. The integration of this predator with selected soft option insecticides (i.e. NPV's) also provided additional impacts with the integrated treatment yielding as highly as conventionally managed cotton plots whilst using 50% less insecticide. Compatibility testing with a range of pesticides demonstrated that assassin bugs could be combined with a range of new generation insecticides if needed. The products, Indoxacarb, Fiprinol, Spynosad, Pyriproxifen and Buprofezin were found to be of a low toxicity to assassin bug nymphs. A novel method for releasing assassin bug eggs onto cotton crops was developed and tested. This technique utilised a commonly available agricultural foaming agent in which eggs could be mixed and applied to a cotton crop resulting in comparable establishments rates with the more labour intensive and expensive release of assassin bug nymphs.



## Project Objective

## Achievements continued

Survey for silver leaf whitefly and parasitoids in Dawson Valley.

Following the initial outbreaks of Silver Leaf Whitefly on the Central Highlands during the 2001-02 season, Silver leaf whitefly also became prominent throughout the Dawson valley. A sampling program for silver leaf whitefly and their parasitoids was conducted during the last three years and identified whitefly as being a regularly abundant cotton pest each season. This abundance has been partially offset by the establishment of high populations of *Eretmocerus* parasitoids which were largely absent 3 years ago. The increase in parasitoid activity has coincided with a decrease in IGR applications for the control of whitefly across the Dawson Valley. In the 02/03 season parasitism peaked at 15% followed by 56% 03/04 and 87% 04/05 season. The proportion of IGR's applied over the same three seasons decreased from 40% of fields treated to just 3-4 fields during the 2004-05 season.

Preliminary investigation using Magnet® as an end of season moth busting tool for *Bt* Resistance management in central Queensland.

A large scale pilot trial was conducted in the Theodore irrigation area during the 2004-05 season to examine the potential for using area-wide applications of the attract and kill product Magnet® as an alternative for end of season trap cropping. Pilot trial results suggest that the area-wide application of Magnet® caused significant reductions in *Helicoverpa* moth populations as well as other key cotton pests like *Spodoptera litura*. More extensive experiments to examine the potential use of Magnet® for *Bt* resistance management will be conducted in the coming season under the newly funded CQ Cotton Farming Systems research project.

Promote best management practices amongst CQ cotton growers.

Several field days have been held with central Queensland growers concerning the use of field peas as a spring trap crop, silver leaf whitefly management and the use of Magnet® as a moth busting tool for resistance management. Significant support was also provided to the IPM Short course program conducted at Emerald during the 02-03 and 03-04 seasons as well as Theodore during 2004-05. Numerous topical and educational articles have also been circulated amongst the CQ industry via the Cotton Tales series and other industry mediums during DAQ122C.

## Overview of Research Background, Methodology, Results and Conclusions for DAQ 122C

A broad suite of research was conducted during DAQ122C. For the purpose of this report each set of experiments will be presented in separate sections detailing the background, methodology, results and conclusions. These sections cover the below range of topics

- **Field Peas – A Better Spring Trap Crop of CQ.** This section describes our research investigating alternative spring trap crops to chickpeas for CQ conditions
- **Vegetative Refuges for Natural Enemies in CQ.** This chapter details our research investigating the potential for utilising refuges grown for augmenting beneficial insects in cotton.
- **Assassin Bugs for Cotton IPM.** This section covers a range of experiments conducted to determine the compatibility of assassin bugs with new generation insecticides through to augmentation with soft options in cotton for the biological control of *Helicoverpa* and mirids.
- **Silver Leaf Whitefly – A Dawson Valley Perspective.** This chapter details our sampling program for silver leaf whitefly and their parasitoids in the Dawson Valley from 2002-2005.
- **Moth Busting for Bt Resistance Management.** This final research section describes our pilot experiment to examine the potential use of the attract and kill product Magnet® as a replacement tool for summer trap cropping.

## Field Peas – A Better Spring Trap Crop for CQ

### Summary

Mounting levels of insecticide resistance within Australian *Helicoverpa* populations have resulted in the adoption of non-chemical IPM control practices such as spring trap cropping with chickpea, *Cicer arietinum* (L.). However, a new leaf blight disease affecting chickpea in Australia has the potential to limit its use as a trap crop. Therefore we evaluated the potential of a variety of winter-active legume crops to be used as an alternative spring trap crop to chickpea as part of an effort to improve the area wide management strategy for *Helicoverpa* in CQ. The densities of *Helicoverpa* eggs and larvae were compared over three seasons on replicated plantings of chickpea, field pea *Pisium sativum* (L), vetch, *Vicia sativa* (L.) and faba bean, *Vicia faba* (L.). Of these treatments, field pea was found to harbour the highest densities of eggs. A partial life table study on the fate of eggs oviposited on field pea and chickpea suggested that large proportions of the eggs laid on field pea suffered mortality due to dislodgment from the plants after oviposition. Plantings of field pea as a replacement trap crop for chickpea under commercial conditions confirmed the high level of attractiveness of this crop to ovipositing moths. It is now our recommendation that growers in CQ use field peas (cvs Alma or Glenroy) for their spring trap crop.

### Background

Increasing levels of insecticide resistance and rising costs of field control in cotton crops during the 1990s prompted a significant shift in the *Helicoverpa* control strategy, away from individual field-based insecticide applications to a season long area wide basis. Two studies on the population dynamics of *Helicoverpa* spanning a period from 1996-2001 in central Queensland suggested that *Helicoverpa* abundance is largely driven by patterns of crop succession and resultant population exchanges that occur at key times between cotton and grain crops within the cropping region (Sequeira 2001; Sequeira & Playford 2001). The pattern of *Helicoverpa* recruitment strongly suggested that large populations of *Helicoverpa* were cycling between winter rain fed and summer irrigated components of the central Queensland cropping system. The proposed pattern of cyclical population dynamics is also supported by analyses of inter-seasonal genetic shifts at the regional level in central Queensland (Scott *et al.* 2003).

An Area Wide Management strategy was proposed for central Queensland in an effort to limit the rate of in-crop *Helicoverpa* recruitment and exchange between

cropping systems (Sequeira 2001; Sequeira & Playford 2001; 2002). Part of the Area Wide Management strategy for central Queensland has involved the implementation of a trap cropping program that aims to divert and capture *Helicoverpa* during early spring and late summer when these populations would typically experience bottle necks in host-plant availability associated with the transition between crops, seasons or both (Sequeira 2001). Cotton growers plant approximately 1-2% of their total crop area to a trap crop of chickpea in late winter and pigeon pea, *Cajanus cajan* (L.) in summer, both timed to coincide with key periods during which significant population exchange has been observed to occur between cotton and grain crops (Sequeira 2001). After attracting large populations of *Helicoverpa* larvae and pupae, the trap crops are destroyed by slashing and cultivation

A problem since encountered with using chickpea as a spring trap crop is its susceptibility to leaf blight disease caused by the pathogen *Ascochyta rabiei* (Pass.) (teleomorph *Didymella rabiei*). This seed borne pathogen has recently spread throughout southern Australia and caused serious disease epidemics in commercial chickpeas (Khan *et al.* 1999). To date central Queensland is one of the few remaining regions still free of this disease, however the growing of chickpeas as trap crops presents a potential risk in terms of disease introduction or providing additional point sources for infection.

To address this problem, a number of winter active legumes were evaluated for their attractiveness to *Helicoverpa* in an attempt to identify an alternative to chickpea that could be used for spring trap cropping in central Queensland. An alternative legume may also have application in southern Australian regions where *A. rabiei* is established and poses a problem with the use of chickpea for trap cropping.

## Materials and Methods

**Host choice assessment:** Two experiments were conducted within a 20 ha field of wheat, *Triticum aestivum* (L.) cv Kennedy near the township of Biloela, central Queensland (24°22'S, 150°06'E) during the winter and spring of 2001 and 2002. In each experiment, treatment plots with dimensions 20m x 20m and 1m row spacing were arranged in a randomised block design with four replicates of each treatment. The plots were separated by 15 m buffer strips sown to wheat on all sides. In the 2001 experiment, legume treatments of chickpea cv Amerthyst, field peas cv Alma and two varieties of vetch cvs Namoi and Popani were compared. In the 2002 experiment, comparisons were made between treatments of chickpea cv

Amerthyst, field pea cv Alma and faba beans cv Fiord. The plots and buffers were planted on 26 June 2001 and 4 July 2002.

In the early crop stages, sampling for *Helicoverpa* was done at approximate 10 day intervals. The sampling frequency was increased once *Helicoverpa* activity was observed to increase in the treatment plots. The data were expressed as numbers of insects m<sup>-1</sup> for each treatment.

*Helicoverpa armigera* was the dominant species, with only low numbers (<30%) of *H. punctigera* observed each season. Visual counts of *Helicoverpa* eggs and larvae were made on two separate sets of randomly selected 1-m lengths of crop foliage in each treatment replicate. When sampling for eggs, four 1-m lengths of foliage was cut from each plot and returned to a field laboratory for close inspection. A beat sheet sampling method was used to assess the densities of *Helicoverpa* larvae on four 1-m lengths of foliage. The sheet was 1.5 m wide by 2 m long and made from yellow canvas. A 25 mm diameter piece of timber dowel (1.5 m long) was fixed to each end of the sheet to prevent the ends lifting in the wind. Samples were taken by placing the sheet behind the legume plants to be sampled, along the inter-row and up over the adjacent row of foliage to create a 'wall' to catch insects. A one metre long stick was then used to shake 1 m of row onto the sheet for assessment. The legume foliage was shaken several times from the base of the plants to the top. The number of larvae were then assessed before being returned to the foliage from which they were sampled.

The treatment plots were destroyed by cultivation on 3 and 9 October respectively for the 2001 and 2002 experiments.

**Impact of host choice on survival:** The fate of eggs laid on chickpea and field pea (these two treatments were chosen because eggs were the most abundant) was investigated during each experiment using methods similar to those described by Titmarsh (1992). White eggs were individually tagged and revisited each day over a period of one week to determine survivorship through the egg and early larval stages. A total of 960 white eggs (60 in each treatment replicate in each experiment) were monitored. The positions of individual eggs were recorded by marking the adjacent leaf surface with a fine tipped, non-toxic pilot felt pen. The corresponding leaf node or branch was also flagged with coloured tape to allow ease of location each day.





**Photo 1.** ODPI&F Technical Officer, Mrs Sherree Short tagging *Helicoverpa* eggs on field pea to examine their fate.



**Photo 2.** Recently laid *Helicoverpa* eggs on field peas marked for later examination.

**On-farm evaluation of trap crops:** Following the 2001 experiment, field pea were substituted for chickpea as the trap crop in several locations in central Queensland to compare *Helicoverpa* activity under commercial conditions.

In the 2002 season, two trap crops of field peas were planted in the last week of June in the cotton irrigation area surrounding the township of Theodore (24°55'S, 149°58'E). In 2003, three trap crops of field peas were planted during the last week of June and first week of July. The trap crops were planted in an area of 2-3 hectares at each site. *Helicoverpa* abundance on these field pea trap crops was compared with nearby chickpea trap crops also planted on 2-3 hectare fields in the same region, all within a radius of 15 km.

Visual counts of *Helicoverpa* eggs and larvae were made on four randomly selected 1-m lengths of foliage in each trap crop using the methods described above. Beneficial insects were also surveyed at the same time. Samples were taken every 4-8 days. Data were expressed as larvae and eggs m<sup>-1</sup>.

**Analysis of data:** The count data from each experiment and farm evaluation studies for *Helicoverpa* eggs and larvae at each sampling date were analysed using a repeated measurements analysis using the method of residual maximum likelihood (REML) with ante dependence covariate structure of order 1 with the Gens tat computer program (Payne *et al.* 1989). This model was used to assess treatment by time interactions. The egg survival data was subject to ANOVA using the Gens tat program, and least significant differences were calculated to determine treatment differences at  $P < 0.05$ .

## Results

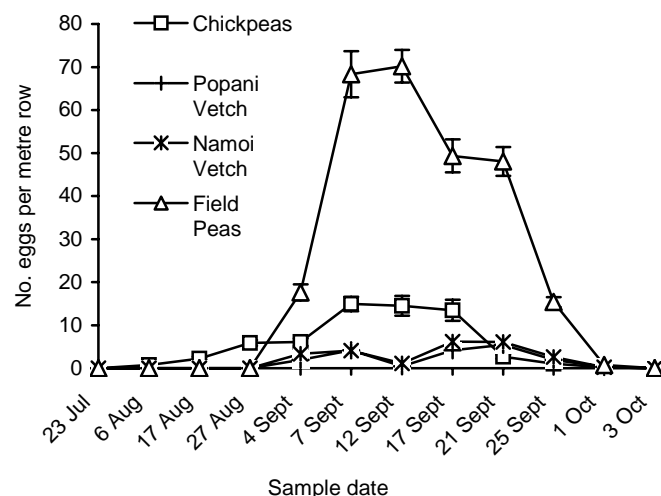
**Host choice assessment:** Field peas attracted significantly higher ( $P < 0.01$ ) numbers of *Helicoverpa* eggs than all other treatments including chickpea in the 2001 assessment (fig. 1) whilst there were no significant differences in the density of eggs between the two vetch treatments. This trend was repeated in the 2002 assessment wherein field peas attracted significantly higher ( $P < 0.01$ ) numbers of eggs than the chickpea and faba bean treatments (fig. 2).

In contrast, *Helicoverpa* larvae densities were significantly higher ( $P < 0.01$ ) in chickpea compared to the other treatment legumes in 2001 with the same significant trend ( $P < 0.01$ ) repeated in 2002 (figs 3 & 4). The density of *Helicoverpa* larvae was also significantly higher in field pea compared to the two vetch varieties in 2001 and significantly higher than the densities recorded in faba bean treatments during 2002 (figs 3 & 4).

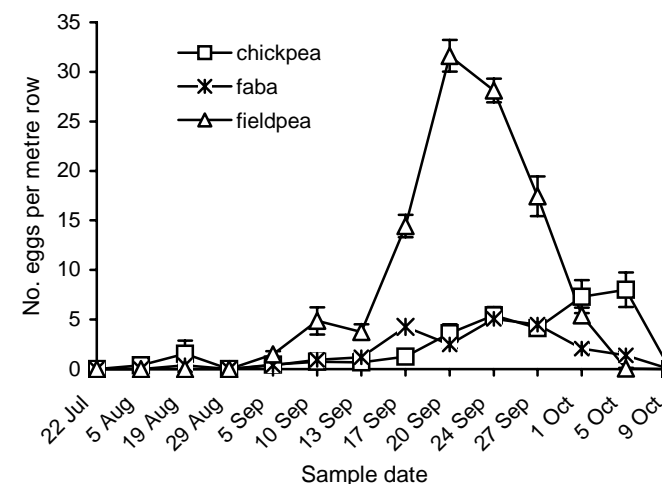
In both experiments few *Helicoverpa* eggs were observed on field pea plants prior to anthesis. In contrast, eggs were observed on chickpea plants prior to flowering.

**Impact of host choice on survival:** A large number of the tagged eggs disappeared from the plants, particularly in the field pea treatment. The eggs or resultant neonates that disappeared from the plants could not be accounted for either as cadavers or by the appearance of other individuals. The percentage of eggs unaccounted for and therefore presumed dead in the field peas was significantly ( $P < 0.01$ ) greater than in chickpeas in both assessments (fig 5).

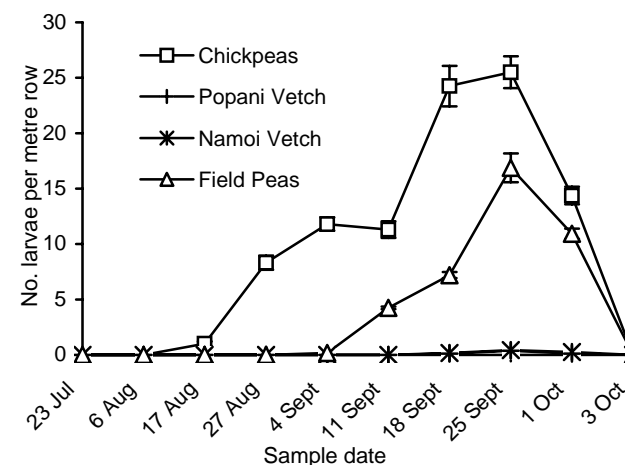
**On-farm evaluation of trap crops:** When planted as a trap crop under commercial conditions field pea attracted significantly higher numbers ( $P < 0.01$ ) of *Helicoverpa* eggs compared with chickpeas during the 2002 and 2003 seasons (figs 6 & 7). *Helicoverpa* oviposition in the field pea plots was observed primarily after the onset of anthesis. Field peas were also observed to host various Coccinellid and Neuroptera species unlike chickpeas which did not host any beneficial insect species.



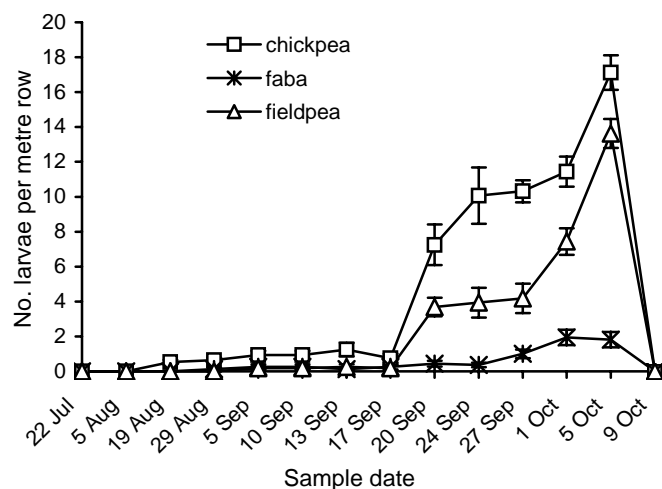
**Fig 1.** *Helicoverpa* eggs per m<sup>-1</sup> of crop foliage in the treatment plots of chickpea, popani vetch, namoi vetch and field pea in the 2001 legume assessment. Error bars denote s.e.m



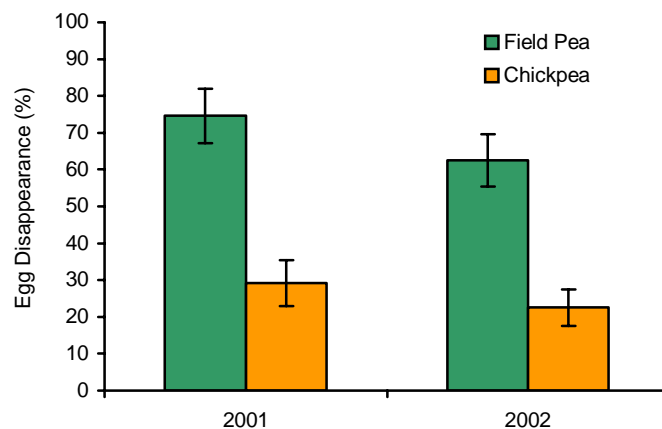
**Fig 2.** *Helicoverpa* eggs per m<sup>-1</sup> of crop foliage in the treatment plots of chickpea, faba bean and field pea for the 2002 legume assessment. Error bars denote s.e.m



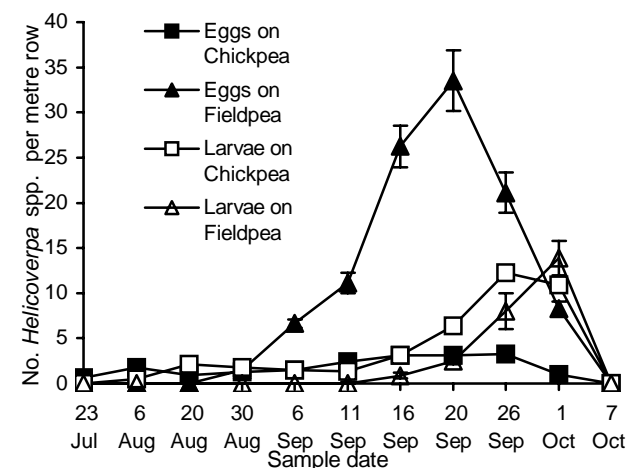
**Fig 3.** *Helicoverpa* larvae per m<sup>-1</sup> of crop foliage in the treatment plots of chickpea, popani vetch, namoi vetch and field pea for the 2001 legume assessment. Error bars denote s.e.m.



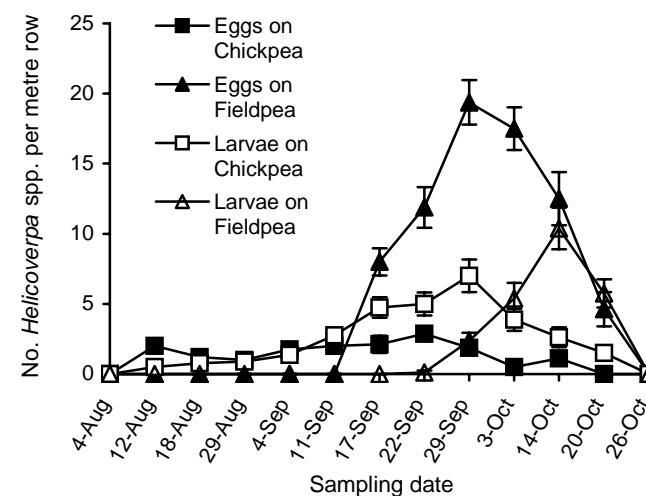
**Fig 4.** *Helicoverpa* larvae per m<sup>-1</sup> of foliage in the treatment plots of chickpea, faba bean and field pea for the 2002 legume assessment. Error bars denote s.e.m.



**Fig 5.** Disappearance (presumed mortality) of marked *Helicoverpa* eggs from the chickpea and field pea plots during the 2001 and 2002 legume assessments. Error bars denote s.e.m.



**Fig 6.** *Helicoverpa* eggs and larvae per m<sup>-1</sup> of foliage in the chickpea and field pea trap crop fields at Theodore 2002. Error bars denote s.e.m.



**Fig 7.** *Helicoverpa* eggs and larvae per m<sup>-1</sup> of crop foliage chickpea and field pea trap crop fields at Theodore 2003. Error bars denote s.e.m.



## Conclusions

A major proportion of *Helicoverpa* immatures that develop within field crops die, with much of the mortality believed to occur during the early life stages (Fitt 1989). These suspected losses were investigated and verified by Titmarsh (1992) who found that most of the mortality affecting *Helicoverpa* in field crops occurs prior to the third-instar stage.

During each experiment, field pea attracted higher levels of egg laying activity by *Helicoverpa* than the other legume treatments, although this did not correspond with significantly higher larvae numbers. Upon hatching *Helicoverpa* spp. larvae typically consume part or all of the eggshell except for the base which remains adhered to the foliage surface (Waterhouse & Norris 1987; Reed 1989). The presence of this residual shell was used during our tagging observations to determine whether or not the eggs had hatched. In the absence of residual shell or neonate larvae within close proximity to the marked site, the eggs were assumed to have been dislodged prior to hatching.

A much greater proportion of eggs were observed to have been dislodged from the field peas compared to chickpeas during the 2001 and 2002 assessments (fig 5). The higher levels of egg retention observed in chickpea may partly explain why this legume carried higher densities of larvae compared to field pea.

Dislodgement of eggs from field pea may be largely due to the waxy nature of the leaves. During the collection and handling of field pea foliage for egg sampling, many of the *Helicoverpa* eggs were observed to readily dislodge, something that was not observed in chickpeas.

In these experiments field pea acted as a superior sink in terms of capturing a greater proportion of moth progeny than chickpea. This trend was replicable when field pea was substituted for chickpea as the trap crop within the Theodore cotton irrigation area, again demonstrating the attractiveness of this crop to ovipositing *Helicoverpa* moths.

The substitution of field pea for chickpea as a spring trap crop in central Queensland is advantageous in that it circumvents potential problems with leaf blight caused by *A. rabiei* that continues to threaten the disease-free status of central Queensland's commercial chickpea industry. Field pea is also advantageous compared to chickpea which can frequently serve as a *Helicoverpa* spp. nursery by hosting substantial populations during the early vegetative stages under central Queensland conditions. These early populations in chickpea often require chemical control to prevent dispersal.

These results show that field pea is highly attractive to *Helicoverpa* moths in spring after the onset of anthesis. Unlike chickpea, many of the eggs laid on field pea perish which in part makes it a self sustaining trap crop during the first weeks of becoming attractive to *Helicoverpa*. Field pea was also observed to host various predatory arthropods which could potentially disperse into surrounding cotton crops upon trap crop destruction.

The use of field pea in southern regions for spring trap cropping may also have merit and warrants investigation. If successful under cooler conditions, the use of field pea would also ease the management of trap crops in these regions by eliminating the current need to apply several fungicides for the control of *A. rabiei*.



*"Field pea was the most effective Helicoverpa spring trap crop. The majority of eggs laid on field pea perish due to dislodgement from the waxy foliage making it a partially self- sustaining trap crop. The use of field peas for trap cropping in CQ circumvents any problems with the chickpea disease ascochyta leaf blight".*

## Vegetative Refuges for Natural Enemies in CQ

### Summary

The growing of dedicated refuges to encourage natural enemy abundance has been a regularly touted strategy for use in cotton IPM. In particular, the use of lucerne as a refuge has been promoted in southern regions to augment beneficial insects as well as to provide a sink for mirid pests. However, lucerne is not entirely suited to CQ conditions and therefore we evaluated the potential of a range of alternative plant species for their ability to harbour and generate natural enemies. The densities of various natural enemies were compared on replicated plantings of niger, *Guizotia abyssinica* (L. f. Cass), peanuts, *Arachis hypogaea* (L), sorghum, *Sorghum bicolor* (L.), lablab, *Dolichos purpureus* (L.) and cotton. Of these treatments, niger attracted the most natural enemies and was the only refuge in which predatory bug breeding was observed. During a second experiment niger was utilised as a companion plant in cotton where it was found to have little impact on natural enemy numbers compared to cotton planted alone. Potential hypotheses for our inability to predictably influence natural enemy abundance through the provision of refuges are discussed with a view to how future natural enemy augmentative research should perhaps be considered at a landscape level.

### Background

Beneficial insects represent an important component within cotton IPM programs. In particular natural enemies have been found to play a key role in the regulation of many secondary cotton pests such as mites and whitefly (Deutscher *et al.* 2004). The hidden influence of these natural enemies is frequently exposed when disruptive insecticides such as organophosphates or pyrethroids are used, causing secondary pest flares. With the advent and widespread adoption of *Bt* crops, insecticide usage has been greatly reduced thus creating an environment that is perhaps less hostile to predators and parasitoids. Within the low spray environment of *Bt* crops, potential exists to develop strategies that further enhance natural enemy abundance beyond what might have been considered possible in earlier conventional production systems.

In order to prosper, natural enemies require adequate food and a suitable environment. Within a monoculture environment such as cotton, the provision of plant refuges may assist in better meeting these needs by creating a more diversified habitat which encourages the continued presence and activity of

beneficial insects by providing shelter, alternative prey insects, pollen and nectar (Hickman & Wratten 1996, Landis *et al.* 2000). However, benefits gained from increasing habitat diversity are largely dependent on the properties of the plants used and the herbivorous inhabitants or prey species present (Letourneau & Altieri 1983, Barbosa & Wratten 1998). Therefore, the selection of refuge plant species and their strategic placement in both space and time within the farming system is important and should aim to create functional diversity that provides resources for the continued survival of beneficial insects that aid in pest management (Bowie *et al.* 1995, Landis *et al.* 2000).

A range of techniques can be used to incorporate refuges into agricultural environments. These have included the creation of "island habitats" within farmland (Thomas *et al.* 1992) or in Australian cotton by growing spaced strips of vegetation throughout the cropping area (Mensah & Khan 1997, Mensah 1999). Cotton offers limited food and shelter for natural enemies during the first months after planting. Therefore the provision of quick to establish refuge plants may act to reduce potential resource limitations by providing earlier alternate prey and floral resources and thus may encourage more rapid in-field predator population establishment.

Research conducted with lucerne as a refuge for cotton suggested that such a strategy has potential to attract and generate beneficial insects that may disperse into cotton production areas and thus enhance biological control (Mensah 1999). However, lucerne does not thrive under CQ conditions and growers are reluctant to dedicate production land to permanent areas of lucerne as a perennial refuge.

During the 2002 and 2003 seasons we examined a range of plants for their potential to be used as a natural enemy refuge under CQ conditions and looked at incorporating them as a companion planting option (opposed to a dedicated refuge strip) as a less expensive technique for increasing in-field biodiversity compared to the use of permanent refuge beds.

### Materials & Methods

#### 2002/03 Comparison of early season refuges

A replicated experiment was conducted on the Biloela Research Station during the 2002/03 season. Plots of sorghum, niger, peanuts, lab lab and cotton (control) were planted within a cotton field during the first week of October 2002 on 1m rows. The treatments were arranged in a randomised block design with four replicates. Treatment plots were 320m<sup>2</sup> (20 rows x 16m) with each plot being surrounded with 10 m of buffer sown to cotton on all sides.



Beneficial and pest arthropods were monitored in each of the refuges during the first half of the season until the end of December. Weekly counts were made using a beat sheet on 4 randomly selected 1m lengths of foliage row.

Count data for beneficial insects were pooled according to type and subject to ANOVA to compare the density of insects between treatments for each sampling interval during the experiment with the GenStat computer program (GenStat 2000 for Windows. Release 6.2. Fifth Edition. VSN International Ltd., Oxford). Differences at each sample date were determined by comparing the treatment predicted means using the standard error of differences.

### 2003/04 Companion Planting Cotton with Niger

A replicated experiment to examine the potential for using niger as a companion plant to augment beneficial numbers was conducted at Theodore during the 2003/04 season. To overcome seed mixing and sowing difficulties the smaller niger seed was processed by Selected Seeds in Biloela who used a clay based coating material to build up the niger seeds to a similar size to cotton seed.

Four replicated plots of cotton mixed with niger seed at 2% (100m x 25m) were established together with cotton only plots within a larger field (10ha) of cotton. All of the cotton sown was Bollgard® II non roundup ready.

Beneficial and pest insects were monitored in the two treatments during the first half of the season up until the end of December at which time the niger had flowered and was being shaded out by the actively growing cotton crop. Counts were made using a beat sheet on randomly selected 1 m lengths of foliage row in each treatment plot.

## Results

### 2002/03 Comparison of early season refuges

Peanuts were a disappointing refuge treatment, being very slow to establish and largely devoid of insects that were abundant in the other treatments. The remaining refuges harboured a diverse range of beneficial species.

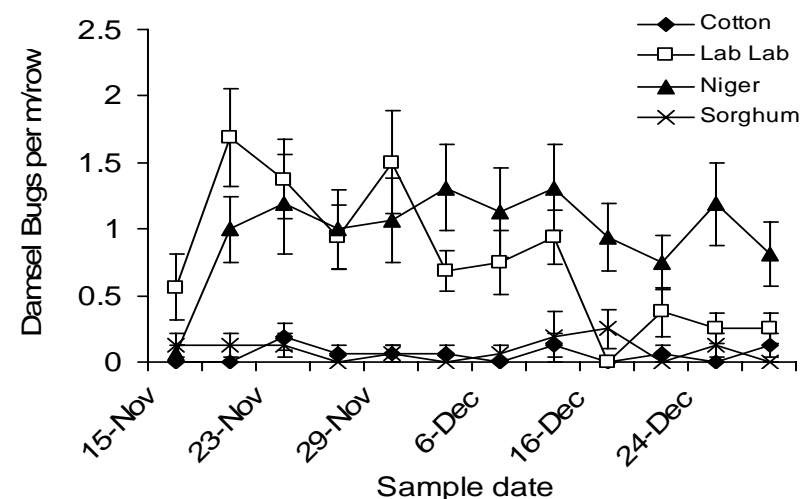
Amongst the predatory bug species recorded, damsel bugs, *Nabis kinbergii* were more abundant ( $P<0.05$ ) in both the niger and lab lab treatments compared to cotton and sorghum (Fig 8). Similarly big eyed bugs, *Geocoris lubra* were recorded at the highest densities in the niger refuge ( $P<0.05$ ) compared to the other treatments (Fig 9). Nymphs of big eyed bug were also regularly found in the niger treatment indicating that reproduction was occurring in this refuge.

When the various predatory bug species were combined niger was found to host the highest levels of activity ( $P<0.05$ ) compared to the other treatments (Fig 10).

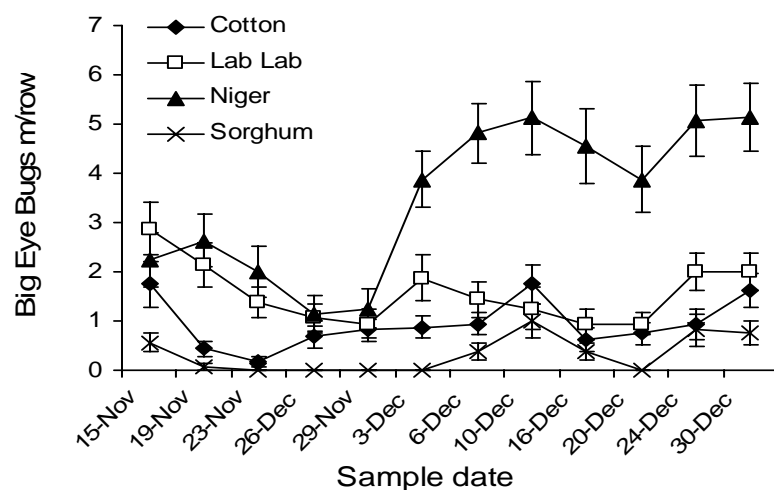
Spiders were prevalent within each of the refuges although no significant differences were observed between treatments (Fig 11).

Adult ladybirds appeared to be more transient than other beneficial species with their numbers observed to peak in different treatments at different stages during the trial (Fig 12). The sorghum was the only refuge to support ladybird reproduction with juveniles of the three banded ladybird, *Harmonia octomaculata* observed.

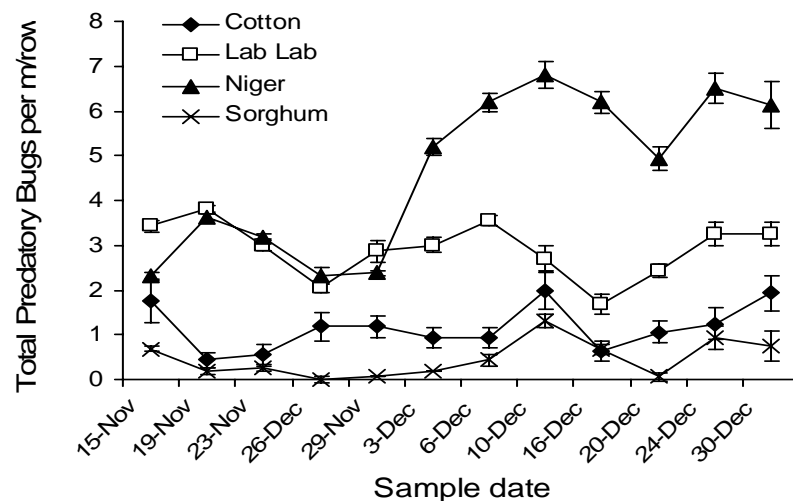
Jassids were the primary prey species recorded on each of the refuge treatments throughout the experiment. Count data show that jassids were significantly higher on niger than all other treatments ( $P<0.05$ ) (Fig 13).



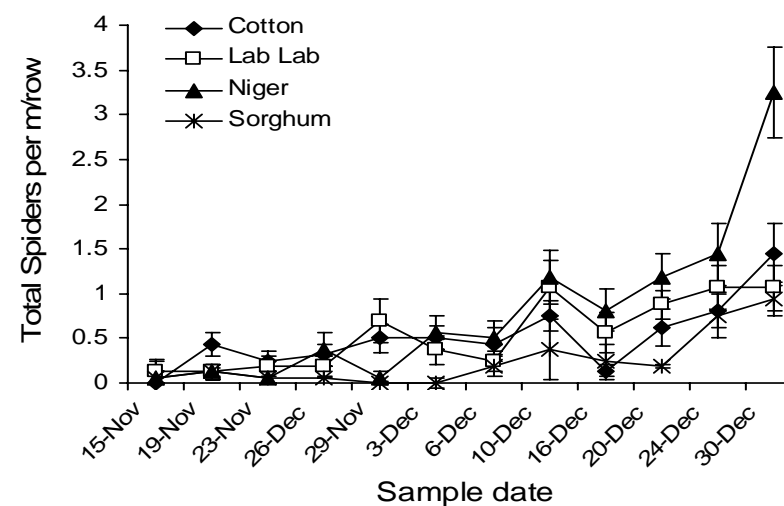
**Figure 8.** Mean densities of damsel bugs per m row of foliage for the four different refuge treatments. Error bars denote s.e.m



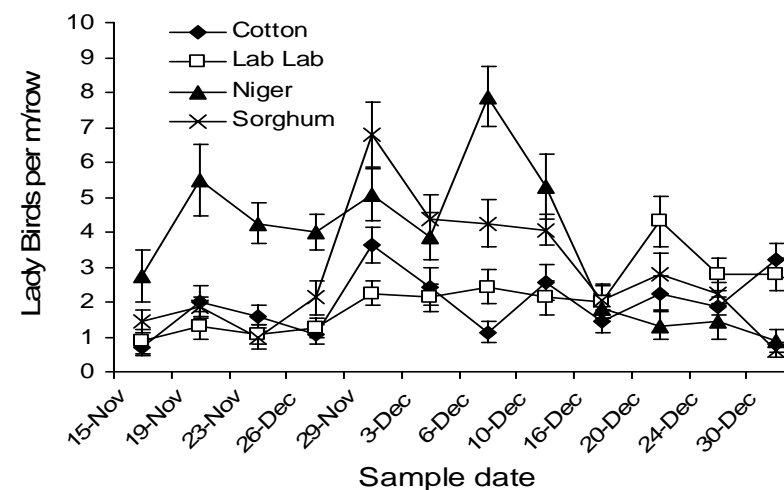
**Figure 9.** Mean densities of big eyed bugs per m row of foliage for the four different refuge treatments. Error bars denote s.e.m



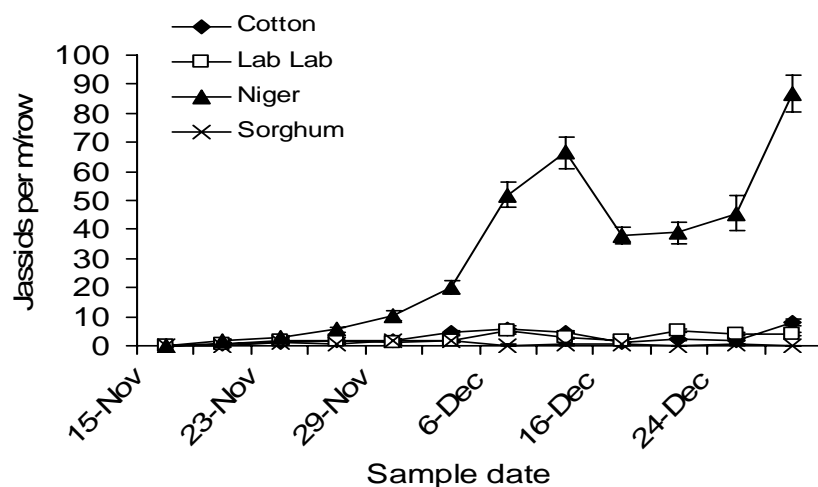
**Figure 10.** The mean density of all predatory bug species pooled per m row of foliage for the four different refuge treatments. Error bars denote s.e.m



**Figure 11.** The mean density of all spider species pooled per m row of foliage for the four different refuge treatments. Error bars denote s.e.m.



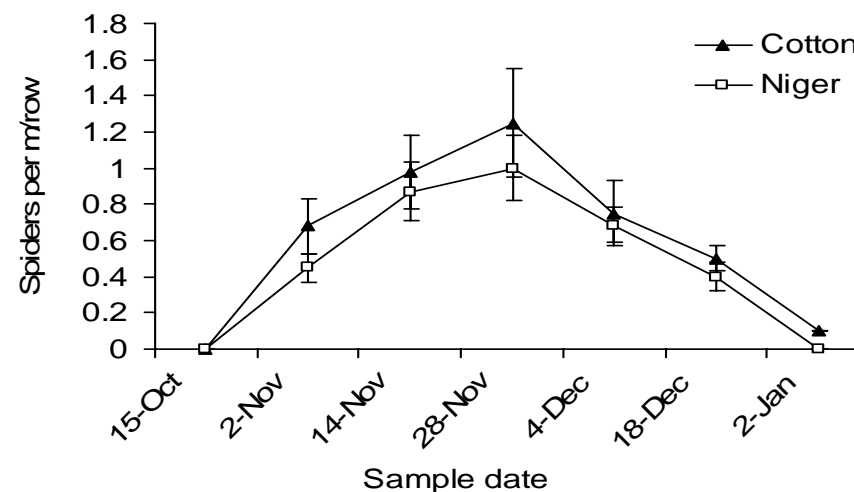
**Figure 12.** The mean density of all coccinellid species pooled per m row of foliage for the four different refuge treatments. Error bars denote s.e.m



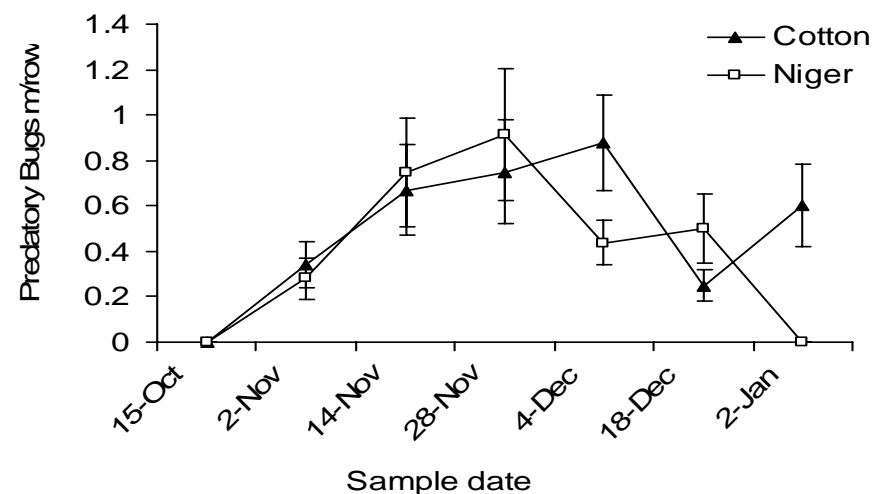
**Figure 13.** The mean density of jassids per m row of foliage for the four different refuge treatments. Error bars denote s.e.m

### 2003/04 Intersowing cotton with Niger

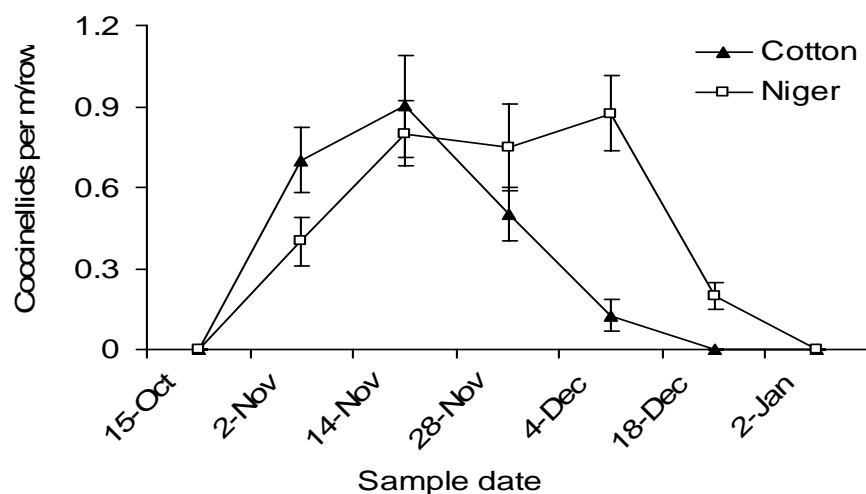
Predatory insects recorded in the plots were pooled into three groups: predatory bugs, spiders and ladybirds. No significant differences were recorded between spider and predatory bug densities in the pots with and without niger (Fig 14 & 15). Ladybirds were significantly higher in the niger/cotton plots during December (Fig 16). This increase in adult ladybird abundance coincided with the flowering of the niger plants in the plots.



**Figure 14.** The mean density of spider species pooled per m row for the plots of cotton with and without niger. Error bars denote s.e.m



**Figure 15.** The mean density of predatory bugs species pooled per m row for the plots of cotton with and without niger. Error bars denote s.e.m



**Figure 16.** The mean density of coccinellid species pooled per m row for the plots of cotton with and without niger. Error bars denote s.e.m

## Conclusions

As in previous refuge studies for natural enemies, the differences observed between the treatments in our experiments may have been due in part to prey availability, refuge canopy architecture and phenology (Hickman & Wratten 1996; Barbosa & Wratten 1998). Indeed, the late increase in ladybird adults in the niger companion planting experiment co-incided with niger flowering which may have provided attractive additional floral food resources.

In terms of prey availability, niger had an abundance of jassids on which a range of natural enemies are known to feed. Niger also provided a prolonged abundance of pollen and nectar compared to the other treatments.

With the exception of spiders which mostly colonised the plots as wind-borne immatures, our sampling data suggested that the refuges did little other than attract and retain the adult stages of various species for differing portions of time during the experiment. The only natural enemies to exhibit reproductive activity observed in the plots were big eyed bugs and three banded ladybirds in the niger and sorghum refuges respectively.

An alternative hypothesis for the trends observed during each experiment may be that the natural enemy populations recorded were merely a reflection of larger spatial and temporal movements of communities of beneficial arthropods as a whole throughout the greater farming eco-system. With the exception of big eyed bugs and three banded ladybirds there was limited evidence to suggest that any of the refuges were colonised for the purpose of reproduction by populations of beneficial insects that may have then imparted additional individuals to adjacent crops.

Our knowledge of even the most commonly documented predatory species in cotton crops is at best limited. Anecdotal reports from researchers and consultants alike suggest that most species are highly variable in terms of their spatial and temporal abundance. Our inability to better predict or describe these patterns are perhaps limited by a lack of understanding regarding species biology and ecology particularly with regard to what occurs outside of cotton fields. It is plausible that much of the research conducted with natural enemies in cotton agro-ecosystems is deficient for this reason. The majority of biological control research conducted within the Australian cotton industry has focused on manipulating natural enemies within or directly adjacent to cotton fields. Of the range of research conducted, the most successful has been the development of insecticide use strategies that protect and conserve natural enemy populations already present within cotton fields. Alternatively, most research attempting to encourage natural enemy colonisation of cotton (refuges, crop succession & food sprays) has proven difficult to reliably implement. This could be considered further evidence to suggest that factors beyond the boundaries of cotton farms are responsible for the broader temporal and spatial patterns in natural enemy population dynamics observed.

The differing abundance of the natural enemy taxa recorded in the refuge treatments may have simply been varying degrees of amplification of broader population flows through the environment with treatment differences being a reflection of relative refuge attractiveness compared to the surrounding environment at each sampling point in time. As the nature of the broader environment (of which cotton is only a small part) is continually changing and its impact on endemic populations of natural enemies and hosts is not well understood, it could be argued that a refuge consisting of a single vegetation type would be limited for the dynamic purpose of generating natural enemies on farm. This may explain the variation in the performance of niger in our experiments as well as in earlier studies (Grundy & Maelzer 2003) and the use of lucerne for augmenting natural enemies in adjacent crops that has also been found to be variably effective (Pearce & Zalucki 2005). Although difficult, the effectiveness of the refuge treatments tested in our experiments may have been better gauged by

comparing the relative abundance of natural enemies recorded within the refuges with those in the surrounding environment to make more informed conclusions about refuge suitability than within refuge treatment comparisons.

It stands to reason that climate and surrounding habitat interactions are a more likely driver for natural enemy populations frequently recorded in cotton fields. These habitats, whether they are grassy roadsides or remanent vegetation all have biodiversity as a common feature. A diverse refuge of perennial plants or shrubs specifically selected to ensure prey and shelter continuity may perhaps serve as a more successful refuge than the annual crop plants tested here, although the length of establishment and sacrifice of agricultural cropland to permanent refuge areas presents some experimental and economic limitations.

Future research that serves to gain a better understanding of the linkages that exist between the natural enemies considered to be the most functional from an IPM perspective and surrounding habitat areas would perhaps provide the information that is required to successfully and repeatedly augment endemic populations and potentially allow for the future establishment of permanent refuges that are valued as a contributing component of functional biodiversity within the farming system.



*"Niger was found to be attractive to various natural enemies. However, when used as a companion refuge planting with cotton it did not increase natural enemy abundance compared with cotton alone. Evidence of natural enemy reproduction was limited in the refuges tested".*



## Assassin Bugs for Cotton IPM

### Summary

Previous research has shown the potential for utilising the large Australian assassin bug, *Pristhesancus plagipennis* as a biological control agent for insect pests in cotton. *P. plagipennis* like many assassin bug species, has until recently been ignored because of low abundance in cotton coupled with a poor understanding of its predatory behaviour. The mass-release of assassin bugs into cotton can overcome the problem of low predator abundance, although a significant challenge exists between achieving a viable balance between predator release rates and biological control efficacy.

Our research with assassin bugs during DAQ 122C focussed on identifying whether a viable balance between release rates and control efficacy could be achieved by combining their biological control impacts with compatible insecticides as an integrated strategy. This approach reduced pesticide inputs by 50% whilst maintaining yields comparable to insecticide grown cotton.

### Background

Arthropod predators and parasitoids are considered to be important mortality agents in Australian cotton production systems although they are rarely capable of controlling *Helicoverpa* when unassisted (Fitt 2000). In an effort to assist the biological control afforded by natural enemies in cotton crops there has been a shift towards the adoption of Integrated Pest Management. The increasing adoption of IPM programs within the Australian cotton industry over the last 10 years has seen a shift away from largely broad-spectrum insecticide dependent programs (Forester *et al.* 1993; Fitt 1994) to strategies that utilise selective narrow spectrum insecticides (Holloway & Forrester 1998), *Helicoverpa* nuclear polyhedrosis virus biopesticides (Mensah & Liang 2002), sacrificial trap crops grown to divert pest species from cropping areas (Sequeira 2001; Grundy *et al.* 2004) and genetically engineered crop varieties that express endotoxin genes from *Bacillus thuringiensis* subsp. *kurstaki* (Bt) (Fitt 2000). However, even with these beneficial insect compatible changes the biological control afforded by natural enemies in cotton is typically inadequate to prevent economic loss.

Augmentation via mass release is another method that can be used to increase the effectiveness of predators and parasitoids within intensive cropping systems (New 2002). In this regard generalist predators, particularly predatory bugs, have been

largely ignored for their augmentation potential in cotton production systems (King & Powell 1992). However, in a monoculture environment where the main pests, *Helicoverpa* and mirids, *Creontiades* spp. are characterised by migratory behaviour and a multi-voltine lifecycle (Zalucki *et al.* 1986; Miles 1995), generalist predators may have a survival advantage as their population dynamics are not solely dependent on any one pest species (Murdoch *et al.* 1985; Nyffeler *et al.* 1992).

The assassin bug, *Pristhesancus plagipennis* (Walker) (Hemiptera: Reduviidae) is a natural enemy of bug and larval insects in both orchard and field crops (Pyke & Brown 1996; Smith *et al.* 1997).

Several studies have suggested that *P. plagipennis* may be suited for augmentation against pest insects such as *Helicoverpa* and mirids with inundative releases resulting in reduced populations of these pests in cotton (Grundy & Maelzer 2002; Grundy 2004). Results from these studies suggested that effective densities of 1 *P. plagipennis* nymph per metre row were sufficient to reduce *Helicoverpa* larvae densities on cotton (Grundy & Maelzer 2002; Grundy 2004). However, during these studies it was evident that a release rate of 10,000 nymphs per hectare were insufficient to prevent economic loss during periods of intense *Helicoverpa* population pressure that can occur during some Australian production seasons. An integrated approach that combines compatible insecticides during incidences of peak pest activity with assassin bug release maybe a more robust and cost effective augmentation strategy (Grundy 2004). Such integrated approaches have been beneficial in the augmentation of other natural enemies in field crops (Hough-Goldstein & Keil 1991).

Previous insecticide compatibility studies suggested that *P. plagipennis* were tolerant of some organochlorine and carbamate insecticides (Grundy *et al.* 2000). However, these products are considered to be disruptive to other natural enemies within the context of IPM programs and since this earlier research, several new generation insecticides (e.g. spinosans, mectins, nicotinoids) have entered the Australian market place. A number of these insecticides are touted to be soft on various assemblages of natural enemies. The effects on *P. plagipennis* were unknown.

The following chapter focuses on identifying the impacts of new generation insecticides on *P. plagipennis* and then testing the potential for combining soft option insecticides with augmentative releases of *P. plagipennis* as an alternative to conventional insecticide based management strategies.

## Materials and Methods

### Insecticide Compatibility

Four-day old first instar *P. plagiipennis* were used in each experiment as earlier studies indicated that this life-stage was the most sensitive and therefore provided a “worst case” test result (Grundy *et al.* 2000). Pesticides that are found to be non-toxic using the assumptions of a “worst case” test generally require no further testing (Hassan *et al.* 1994).

A series of experiments were done with each insecticide during August and September 2002. The active ingredient, formulation and manufacturer for each insecticide are listed (Table 1). Each product was tested at its maximum registered rate for the control of insect pests on cotton within Australia as well as at three dilutions (75, 50 & 25% of recommended rate) as the application of insecticides at below label rates for the improved conservation of natural enemies is becoming more common place within the Australian cotton. Agral® non-ionic wetter (Nonyl Phenol Ethylene Oxide Condensate) (Crop Care, Australia) was added at the rate of 0.1 ml/L to each insecticide suspension before application because wetting agents are commonly mixed with pesticides to enhance spray coverage in Australia. Agral® was also mixed with distilled water at the same rate and used as a control in each experiment.

For the laboratory tests, disposable 200mm diameter Petri dishes were used as a standardized application target. The Petri dishes were modified by punching four 30 mm diameter holes into the lid of each container and gluing a piece of muslin gauze over the opening for ventilation. Three replicates of twenty nymphs were topically treated on the Petrie dish plates using a Potter precision spray tower to apply 2 ml aliquots of insecticide as described by Holland and Chapman (1995) and Herron *et al.* (1998). Before being treated the nymphs within each Petri dish were temporarily immobilized with carbon dioxide (CO<sub>2</sub>) gas to allow easy handling and to slow the nymphs and prevent their escape during application. After treatment the Petrie dishes containing the sprayed nymphs were placed in a constant climate laboratory under conditions used for rearing for 24 hours. The nymphs were then transferred to clean Petri dishes and provided with *T. molitor* larvae which were killed by immersion in hot water (70°C) and those that successfully moulted to the second instar were recorded as having survived the treatment. The provision of *T. molitor* prey minimized nymphal desiccation and cannibalism.

**Table 1.** Active ingredient (AI), formulation and recommended application rates of insecticides or plant growth regulator compared for their activity against *P. plagiipennis*.

Active Ingredient	gAI/L & Formulation	Manufacturer	Application rate	
			mL/L	L/ ha
<i>B. thuringiensis</i>	Biological	Valent	20	2
Nucleopolyhedrovirus	Biological	Bayer Crop Science	5	0.5
Buprofezin	200g/L EC	Syngenta	10	1
Pyriproxifen	500g/L EC	Sumitomo	5	0.5
Indoxacarb	200g/L SC	Du Pont	8.5	850
Spinosad	480g/L SC	Dow AgroSciences	2	0.2
Fiprinol	200g/L SC	Bayer Crop Science	1.25	0.125
Emamectin benzoate	17g/L EC	Syngenta	5.5	0.55
Abamectin	18g/L EC	Syngenta	6	0.6
Diafenthiuron	500g/L SC	Syngenta	6	0.6
Imidacloprid	200g/L SC	Bayer Crop Science	2.5	0.25
Omethoate	800g/L SL	Bayer Crop Science	1.4	0.14
Mepiquat	38g/L AC	Bayer Crop Science	10	1

SC= Suspension Concentrate, SL = Soluble Liquid, EC = Emulsifiable Concentrate, AC = Aqueous Concentrate

A second experiment was also conducted with emamectin benzoate, spinosyn and indoxacarb applied at the full recommended rate to three replicates of each nymphal instar using the same methods outlined to investigate tolerance differences between instars.

All treatment results were corrected for control mortality using Abbott's formula (Abbott 1925).

### Integrated Field Studies

Two experiments were conducted within a 2.5-ha field of irrigated cotton (cv Sicot 71) during the summer of 2002-03 and 2003-04 near the township of Biloela, central Queensland (24°22'S, 150°06'E). In each experiment, treatment plots with dimensions 25m x 10m and 1m row spacing were arranged in a randomised block design with five replicates of each treatment. The plots were separated by 6 m buffers which comprise 2 metres of bare earth adjacent to a 2 metre strip of cotton all sides.

Five treatments were compared in the 2002 experiment: Third instar *P. plagiipennis* released at 1.0 nymphs per m row (10000 nymphs per hectare); the same

predator release treatment again but combined with selected compatible insecticides; a soft option sprayed treatment to which the same compatible insecticides were applied at the same time as those applied to the predator treatments; a conventionally sprayed treatment; and a *P. plagipennis* nymph and insecticide free control. The same treatment regime was repeated during the 2003 experiment.



**Photo 3.** Plots used for assassin bug field testing at Biloela Research Station 2001-02

*P. plagipennis* nymphs were released in each experiment within a week of the first flowers appearing on the crop on 17 and 20 December 2002 and 2003 respectively. Nymphs for each treatment were released singularly onto the terminal shoots of the crop foliage using a camel hair brush late in the afternoon after 17:00h during each experiment.

The sprayed treatments were managed with insecticides chosen in accordance to the Insecticide Resistance Management Strategy set by the Australian cotton

industry for each season. Application decisions were based on commercially accepted density thresholds for *Helicoverpa* and mirids as well as crop damage models for bud and fruit retention (Schulze & Tomkins 2002; Johnson & Farrell 2003). Insecticide applications on the sprayed plots were made at daybreak whilst wind was minimal to avoid insecticide drift into adjacent plots. A record of the insecticides applied to the conventional, soft only and soft and assassin bug treatments is given in Table 2. No pesticides were used on the crop area except for those sprayed treatment plots.

**Table 2.** The insecticides applied to the conventionally sprayed (CS), soft option only (SO) and soft options with assassin bug (SO&AB) treatments during the 2003 and 2004 experiments.

Pest	Active	Rate	Treatments Sprayed	Application Date
<i>Helicoverpa</i>	NPV	500mL/Ha	CS, SO, SO&AB	13 Dec 2002
<i>Helicoverpa</i>	NPV	250mL/Ha	SO, SO&AB	18 Dec 2002
<i>Helicoverpa</i>	Synosad	200mL/Ha	CS	20 Dec 2002
<i>Helicoverpa</i> & Mirids	Fiprinol/ NPV	40mL/Ha & 250mL/Ha	CS, SO & SO&AB	9 Jan 2003
<i>Helicoverpa</i>	Synosad	200mL/Ha	CS	9 Jan 2003
<i>Helicoverpa</i>	NPV	250mL/Ha	CS, SO, SO&AB	14 Jan 2003
<i>Helicoverpa</i>	Indoxacarb	750mL/Ha	CS, SO, SO&AB	20 Jan 2003
<i>Helicoverpa</i>	NPV	500mL/Ha	CS, SO, SO&AB	30 Dec 2003
<i>Helicoverpa</i>	NPV	250mL/Ha	SO, SO&AB	5 Jan 2004
<i>Helicoverpa</i>	Spynosad	200mL/Ha	CS	5 Jan 2004

In each experiment, pre-release insect counts were made prior to predator release and then every 3-7 days until the end of the experiment. The data were expressed as numbers of insects per metre row for each treatment.

Visual counts of *Helicoverpa* eggs and larvae on the cotton plants were made on 4 randomly selected 1 m row lengths of cotton plants in each treatment replicate. The growing points and squares of the upper two thirds of the plants canopy were searched for eggs and small larvae because these instars are frequently found in those plant regions (Farrer & Bradley 1985). Flowers and bolls throughout the plants were also inspected for larger larvae. Larvae were recorded as small 2-10mm, medium 11-20mm and large >20mm. Numbers of *P. plagipennis* nymphs were recorded at the same time.



A beat sheet sampling method was used to assess the presence of mirids and other insects. The sheet used was 1.5m wide by 2m long and made from yellow canvas. A one metre long stick was then used to shake 1m of crop foliage onto the sheet for assessment. Insects were then aspirated off the sheets with a domestic battery operated vacuum cleaner and returned to the laboratory for assessment. Beat sheet samples were made on 4-6 randomly selected 1m row lengths of cotton plants in each treatment replicate

Each crop was grown through to harvest and assessed for yield. The cotton was picked from the six central rows of each treatment replicate with an experimental two-row picker. Heavy rain due to a cyclone depression delayed the harvest of the 2002 crop and resulted in significant yield losses due to boll rot, tight lock and weather damage. The 2002 crop was picked on 10 April 2003 and the 2003 crop was picked on 11 March 2004.

The cotton picked from each plot was weighed and a sub-sample taken for ginning to determine the relative proportions of lint and seed. The yield from each plot was divided by the sub-sample gin turnouts for the proportion of lint and seed and expressed as kg/plot. From this data, yields in terms of bales/hectare were estimated.

Count data for *Helicoverpa* and mirids at each sampling date were analysed using a repeated measurements analysis using the method of residual maximum likelihood (REML) with ante dependence covariate structure of order 1 with the Gens tat computer program (GenStat 2000 for Windows. Release 6.2. Fifth Edition. VSN International Ltd., Oxford). This model was used to assess treatment by time interactions. Differences at each sample date were determined by comparing the treatment predicted means using the standard error of differences.

### Mass Release Techniques

Some basic experiments were conducted to examine the potential for mass releasing the eggs of *P. plagipennis* instead of nymphs. Two treatments of either twenty eggs or first instars were released into replicated plots in early squaring cotton in three different fields on the Biloela Research Station. The plots consisted of a single row of cotton 20 metres in length with a 2 metre bare earth buffer surrounding each plot. The nymphs were released individually onto the plants throughout the plots within a day of hatching using a camel hair brush. For the eggs release technique, eggs that were 48 hours from hatching were chosen and mixed with long lasting high expansion agricultural line marking foam (anionic wetting agent), agitated to create foam and applied to the plants with a modified pressure pack sprayer. The foam served to stick the eggs to the leaves of the

plant. A replicated series of *P. plagipennis* free plots were also implemented to check whether any dispersal between plots had occurred.

Nymph densities were then assessed 3 weeks post treatment to determine the relative rates of establishment. By this stage surviving nymphs had developed into third instars and could be considered to be established and effective in terms of providing biological control (Grundy & Maelzer 2000). The plants in the plots were extensively searched visually and nymph densities recorded.

This experiment was repeated on three occasions commencing on the 3, 10 and 25 of November 2003.



**Photo 4** Assassin bug eggs applied in foam to cotton foliage.

## Results

### Insecticide Compatibility

Pyriproxyfen, Buprofezin, *Bacillus thuringiensis* and Nucleopolyhedrovirus were found to be non-toxic to *P. plagipennis* nymphs whilst Indoxacarb was of very low toxicity (Table 3). Spynosyn and fiprinol were of low to moderate toxicity which decreased markedly with reduced application rates whilst emamectin and abamectin were of moderate to high toxicity. Diafenthiuron, imidacloprid and omethoate were highly toxic to *P. plagipennis* nymphs even when applied at reduced rates (Table 3).

The susceptibility of *P. plagipennis* nymphs to insecticide decreased with older instars with fourth and fifth instars remaining relatively un-affected by direct exposure to indoxacarb, spynosad, fiprinol and emamectin benzoate (Table 4).

**Table 3.** The mean percentage mortality and s.e. of first instar *P. plagipennis* treated with various insecticides and plant growth regulator in laboratory bioassays. Products are listed in increasing order of toxicity.

Product	Percentage of Recommended Field Rate Tested			
	100	0.75	0.5	0.25
B. thurengiensis	0	0	0	0
Nucleopolyhedrovirus	0	0	0	0
Mepiquat	0	0	0	0
Buprofezin	0	0	0	0
Pyriproxyfen	2.2 ± 0.1	0	0	0
Indoxacarb	7 ± 2.8	2 ± 0.1	0	0
Spynosad	27 ± 1.9	11 ± 0.1	12 ± 1.93	7 ± 3.4
Fiprinol	43 ± 2.8	25 ± 3.0	18 ± 1.1	14 ± 0.1
Emamectin benzoate	69 ± 8.4	47 ± 2.8	42 ± 8.1	16 ± 2.3
Abamectin	84 ± 1.1	61 ± 2.0	51 ± 1.9	41 ± 8.5
Diafenthiuron	100	100	91 ± 2.8	84 ± 1.1
Imidacloprid	100	100	96 ± 0.9	94 ± 1.0
Omethoate	100	100	100	100

**Table 4.** The mean percentage mortality and s.e. of each *P. plagipennis* instar treated with various insecticides at the full recommended rate in laboratory bioassays

Insecticide	Percentage Mortality of Each <i>P. plagipennis</i> Instar				
	I	II	III	IV	V
Indoxacarb	6 ± 2.7	4 ± 2.7	0	0	0
Spynosad	28 ± 2.9	11 ± 2.2	4 ± 2.2	0	0
Fiprinol	39 ± 5.5	29 ± 2.2	18 ± 4.4	9 ± 2.2	4 ± 2.7
Emamectin benzoate	65 ± 9.4	33 ± 3.8	11 ± 2.2	8 ± 1.9	4 ± 2.2

## Field Studies

### 2002-03 Experiment

Loopers were sampled whilst beat sheeting for mirids in the plots and Figure 17 shows that conventional chemistry caused the largest reductions in looper numbers. The soft options only, soft options and assassin bugs and assassin bug only treatments also had a significant ( $P < 0.05$ ) impact in reducing looper numbers in the plots during the later half of January compared to the control.

The conventional, soft option only and soft option and assassin bug treatments resulted in significantly ( $P < 0.05$ ) reduced mirid populations compared to the control. Although a delay in control was recorded, the assassin bug only treatment also resulted in significant ( $P < 0.05$ ) reduction in mirid numbers compared to the control during the latter half of January (Fig 18).

*Helicoverpa* larvae were significantly ( $P < 0.05$ ) higher in the control compared to all other treatments on several occasions during the experiment (Fig 19). The conventional and assassin bug only treatments had the lowest densities of larvae on the 21 January. The soft options only and assassin bug and soft option treatments had higher numbers of larvae on this occasion due to flaring caused by an earlier fiprinol application for mirid on 9 January 2003 which resulted in much higher egg and neonate survival in these two treatments.

Treatment impacts on yield during this experiment were difficult to determine due to adverse weather conditions in February. The onset of boll opening in the plots coincided with an extended period of wet weather from ex-tropical cyclone "Beni" which crossed the Queensland coastline north of Rockhampton. A total of 380 mm fell on the crop over a period of three weeks with very little in the way of clear weather occurring between rainy periods. As a result all of the treatments suffered

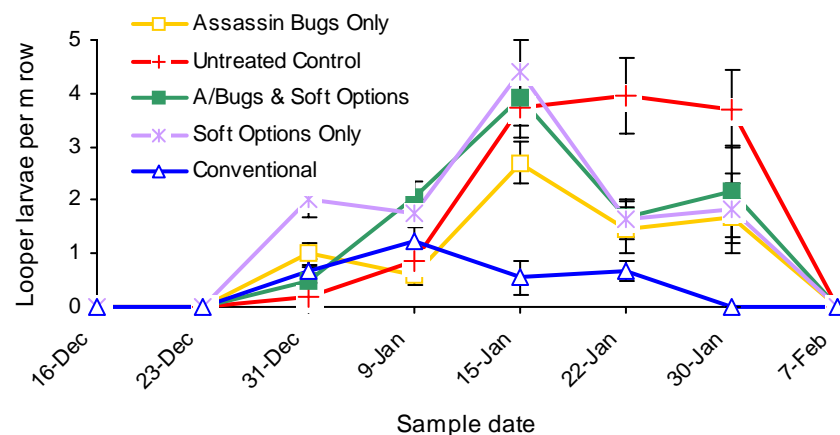


extensive boll rot and tight lock (>20%) damage except the control, which was significantly less affected due to a later pattern of fruit set. All treatments yielded significantly ( $P<0.05$ ) more lint than the control despite the weather damage and compensatory later yield in the control replicates (Fig 20).

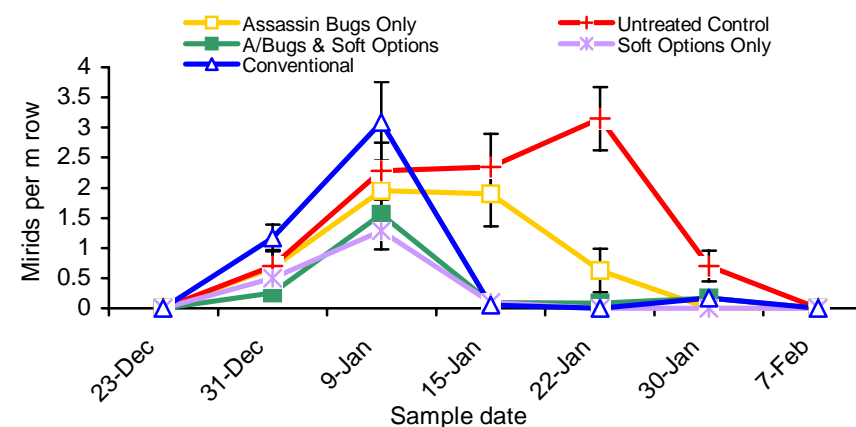
### 2003-04 Experiment

The 2003-04 experiment was subject to very low levels of pest pressure with little in the way of mirid or *Helicoverpa* activity recorded. Each of the treatments had significantly lower densities of *Helicoverpa* than the control. The soft options and assassin bug treatment recorded the lowest density of larvae at some sample intervals although this was not statistically evident due to the overall low pest densities.

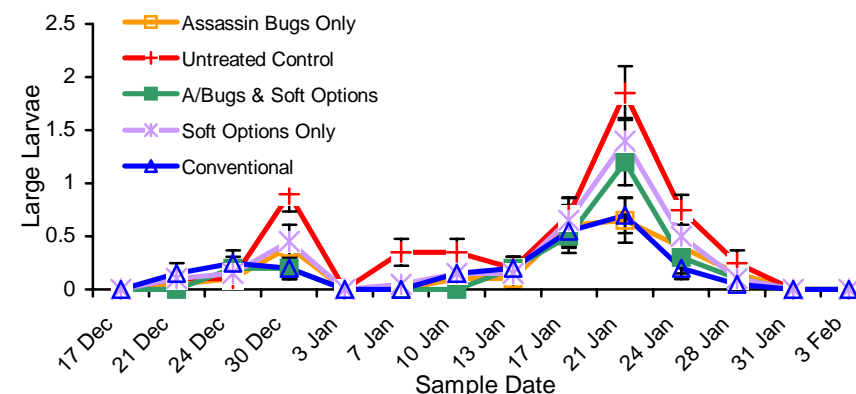
The conventional and assassin bug with soft option treatments yielded the same amount of lint both of which were significantly more than the unsprayed control (Fig 21). The unsprayed treatment yielded in excess of 9 bales/hectare which reflect the low pest densities recorded in the plots throughout the experiment.



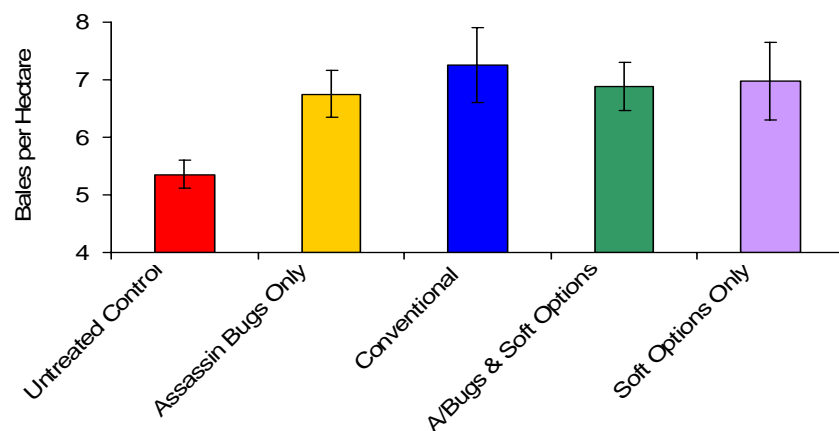
**Figure 17.** Time series showing numbers per m row of Looper larvae in cotton plots for the two assassin bug releases (with and without soft options), a conventionally sprayed treatment, soft option only treatment and untreated control. Assassin bugs were released on 17 December 2002. The bars denote  $\pm$  s.e.



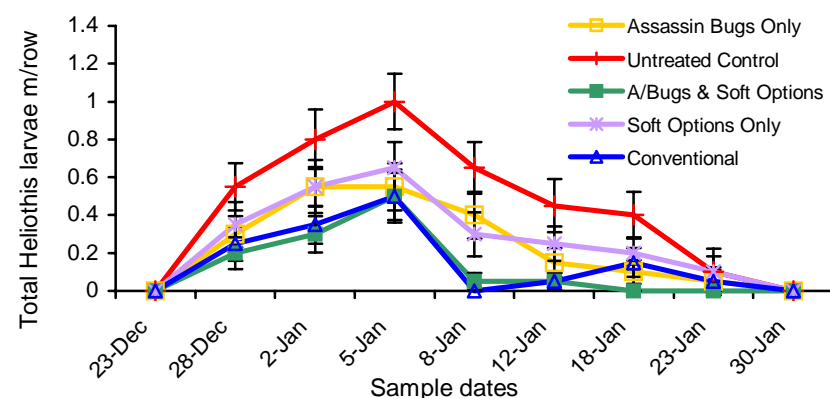
**Figure 18.** Time series showing numbers per m row of mirids in cotton plots for the two assassin bug releases (with and without soft options), a conventionally sprayed treatment, soft option only treatment and untreated control. Assassin bugs were released on 17 December 2002. The bars denote  $\pm$  s.e.



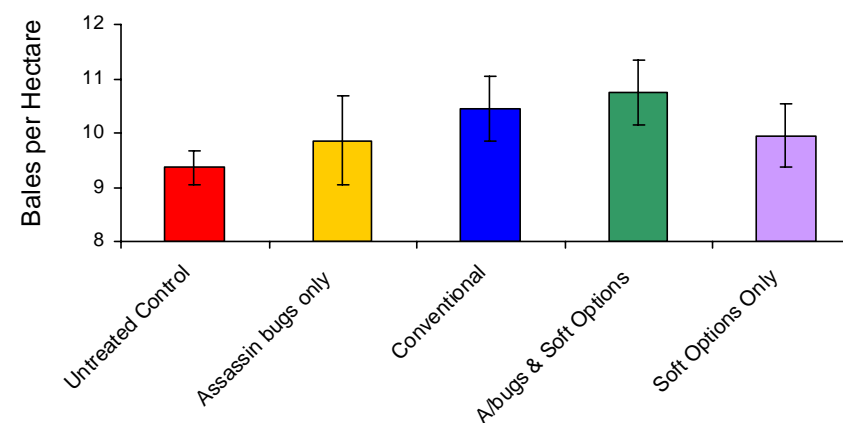
**Figure 19.** Time series showing numbers per m row of large *Heliothis* larvae in cotton plots for the two assassin bug releases (with and without soft options), a conventionally sprayed, soft option only treatments and unsprayed control. Assassin bugs were released on 17 December 2002. The bars denote  $\pm$  s.e.



**Figure 20.** The picked and ginned yields from the two assassin bug releases (with and without soft options), a conventionally sprayed treatment, soft option only treatment and unsprayed control during the 2002-03 experiment. The bars denote  $\pm$  s.e



**Figure 21.** Time series showing the total numbers of *Heliothis* per m row in the cotton plots for the two assassin bug releases (with and without soft options), a conventionally sprayed treatment, soft option only treatment and unsprayed control during the 2003-04 season. Assassin bugs were released on 20 December 2003. The bars denote  $\pm$  s.e.



**Figure 22.** The picked and ginned yields from the two assassin bug releases (with and without soft options), a conventionally sprayed treatment, soft option only treatment and unsprayed control during the 2003-04 experiment. The bars denote  $\pm$  s.e.

### Mass Release Techniques

The release of eggs onto cotton gave comparable predator establishment rates as the release of nymphs (Table 5). Predator losses from the plots due to dispersal were considered to be minimal as no nymphs were found in the controls.

**Table 5.** The mean percentage (out of 20 released) and s. e. of *P. plagipennis* nymphs recovered from the plots three weeks post-release for the three release experiments. The release treatments were eggs and first instar *P. plagipennis*. No nymphs were recorded in the control plots.

Treatment	Recovery of Third instars Post release (% $\pm$ SE)			
	Test 1	Test 2	Test 3	Mean
Control	0	0	0	0
<i>P. plagipennis</i> eggs	48 $\pm$ 8.8	51 $\pm$ 5.4	55 $\pm$ 6.5	51.3 $\pm$ 6.9
<i>P. plagipennis</i> nymphs	53 $\pm$ 7.3	56 $\pm$ 13	66 $\pm$ 9.3	58.3 $\pm$ 9.8

## Conclusions

### Insecticide Compatibility

The results from testing insecticides on assassin bugs demonstrate the robustness of these predators. The tests indicated that *P. plagipennis* are compatible with Steward®, Admiral®, Applaud®, Tracer®, Regent® and potentially Affirm® as well as all currently registered NPV and Bt biopesticides (Table 3). The flexibility afforded by being able to use these products with assassin bugs in the field is highly desirable from an IPM program perspective. Our tests also show that as *P. plagipennis* nymphs grow and develop, they become more tolerant to insecticide exposure (Table 4). This is of advantage as it allows the use of a broader suite of products post release as time progresses and insecticide choices become more limited.

### Integrated Control

The 2002 and 2003 mass release experiments were conducted for the purpose of validating the use of assassin bugs as a pest management tool within an integrated program that reduces pesticide inputs whilst maximising crop yield. Both experiments demonstrated that the combination of *P. plagipennis* with selected soft options could result in significant reductions in *Helicoverpa* and mirid populations at levels not significantly different to conventionally managed plots. The assassin bug only plots also exhibited significant reductions in pest densities although delays in control were recorded.

The full potential of the treatments in each experiment were not fully realised due to adverse climatic conditions or low pest densities. The 2002/03 trials were severely effected by an extended period of wet weather during February 2003 from ex-tropical cyclone Beni. This wet period caused considerable yield losses which masked potential cumulative treatment impacts. The control plots were least affected by the wet weather in this experiment as early to mid season damage had resulted in later fruit set which was less susceptible to boll rot and tight loch. The compensatory growth and late pick of the plots due to the weather reduced the relative yield differences between treatments and the control compared to more typical pre-picking conditions.

The impact *P. plagipennis* on *Helicoverpa* and mirids in the 2002/03 experiment may have also been diluted due to high densities of loopers that served as substitute prey. Indeed significant looper reductions ( $P < 0.05$ ) associated with *P. plagipennis* release were recorded during the experiment (Fig 17). In hindsight, the use of a soft option such as Steward® instead of *Helicoverpa* specific NPV's

during this period may have enhanced the later levels of biological control afforded by *P. plagipennis* by removing the alternate, largely un-economic prey species from the crop canopy.

Low pest densities during the 2003/04 season limited the extent to which treatment differences could be compared. This is evidenced by the high yields recorded in the untreated control. Only the conventional and soft options with assassin bug treatments yielded significantly more than the control suggesting that an integrated approach using *P. plagipennis* and soft options combined is just as an effective strategy as insecticide dependant conventional management. Excluding the use of NPV's the combined use of *P. plagipennis* and soft options resulted in a greater than 50% reduction in conventional insecticide application during each season. This reduction is significant considering that there were no differences in yields between conventional and soft option combined assassin bug managed plots in each experiment.

### Mass Release

A significant challenge for using augmentative releases of a predator like *P. plagipennis* in cotton is the logistics of evenly dispersing large numbers of insects over broad-acre areas (every hectare contains an average of 10km of crop row). Assassin bug nymphs are flightless and have limited dispersal ability. It is therefore essential that any prospective release technique ensures the even distribution of insects throughout the target field.

Our first solution to this problem was to mix nymphs with a bulking agent such as vermiculite and then spread the mixture over crop areas with a fertiliser spreader. This method resulted in significant nymph establishment and with calibration provides uniform distribution (Grundy & Maelzer 2002). However, the main constraints for this method entail the physical challenges of handling large quantities of nymphs and loading them into fertiliser spreading equipment. To be successful such a method would need to be fully integrated with a future commercial supplier from a packaging perspective to minimise handling difficulties.

An alternative release strategy is to use *P. plagipennis* eggs. In comparison to nymphs, eggs are less expensive to produce and easier to handle but are more likely to suffer post-release mortality losses compared to nymphs. The problem of dispersing eggs was overcome by mixing them with foam and spraying them onto the crop. The foam served the purpose of being a carrier that adhered the eggs to the foliage of the crop and prevented them from dropping onto the soil where they might be exposed to climatic extremes or predators. The experiments that were conducted with this method suggested that the release of eggs using foam may be

a successful alternative to releasing third instars. The establishment rate of eggs to the third instar stage was greater than 50% in each experiment. This rate of establishment was similar to the release of first instars which suggests that the release of eggs compared to recently hatched nymphs had minimal impact on survival and that the majority of losses occurred during the early nymph stages.

The advantages of an egg release method include ease of handling, packing and shipping as well as the ability to store in the fridge for several days until required. Eggs are also significantly cheaper to produce and handle from a mass-production perspective. Disadvantages associated with egg release include susceptibility to being washed off the plant by rain and the biological control time delay requiring earlier release than would be necessary with the use of nymphs. Eggs are also less robust and therefore compensatory numbers need to be released compared to the release of third instar nymphs. The cost benefit for releasing additional eggs is difficult to assess until such time as assassin bugs are commercially produced although laboratory culture estimates suggest that the release of twice as many eggs would be far less costly than releasing third instars.

## The Future

The results presented here along with our earlier experiments collectively demonstrate that assassin bugs could play a valuable role within cotton IPM programs for the control of *Helicoverpa* and mirids. Our results show that the control afforded by these predators on cotton when combined with soft options is sufficient to provide similar yields to conventionally sprayed cotton. The ability to combine these predators with a range of selected insecticide options make them a more flexible biological control agent compared to many other natural enemies that are highly sensitive to most registered insecticides.

To take advantage of these predators would require augmentation on a massive scale. Assassin bugs are generally scarce within cotton agro-ecosystems and therefore the most reliable augmentation strategy is to mass-rear and release them into cotton crops. Our earlier research suggests that *P. plagipennis* can be easily mass-reared for release onto broad-acre crops (Grundy *et al.* 2000), although viability would depend on commercial insectaries being able to produce large quantities of assassin bugs cost effectively.

Presently the rearing of these predators is achieved using mealworms as a food source, meaning a commercial insectary would have to breed two insects for the sale of one. The cost of production for mealworms is around \$30/kg not including the capital infrastructure investment required to house a mealworm colony.

A potential cost reducing solution is to use an artificial diet to mass rear assassin bugs. An artificial diet would have significant advantages in that it supersedes the need for expensive mealworms, would allow greater production consistency and reliability, is capital less intensive, more labour efficient and hence more cost-effective. During the last 2 years we have been investigating the potential to use artificial diets for rearing *P. plagipennis*. These diets have been developed by researchers in the United States and Europe for the production of other predatory bug species. Of these diets, a meat-based formulation that is composed primarily of a mix of beef liver, mince and hens eggs published by Cohen & Smith (1998) has shown some potential for rearing assassin bugs. This diet costs around \$4/kg to manufacture.



**Photo 5** Third instar assassin bugs being reared on artificial diet in the laboratory.



When formulated, we found *P. plagipennis* nymphs readily accept this diet and it is sufficient to support their growth through until the adult stage without any obvious impediment compared to nymphs reared on mealworms. However, the fecundity from the *P. plagipennis* reared on the diet was very low compared to those reared on mealworms and therefore was not adequate.

The acceptance of and development of *P. plagipennis* nymphs through to adulthood on artificial diet as well as the limited production of eggs strongly suggests significant potential exists for utilising some sort of artificial diet-based rearing system. Such a diet would obviously need to be refined to overcome fecundity issues and to ensure the field fitness of predators reared artificially. Many overseas research organisations are currently investigating artificial diet improvements and it is likely that future developments in diet technology will pave the way for the mass-production of a range of natural enemies including assassin bugs. It is probable that improved natural enemy production capabilities due to cost reducing artificial diets and other technological developments will precipitate the greater use of mass-released natural enemies.

Our research with assassin bugs demonstrates that these predators have significant field potential for the control of *Helicoverpa* and mirids in cotton. If these predators become commercially available in the future due to advances in mass-rearing technology they may well play an important role in the production of cleaner and greener conventional cotton.



*"Assassin bugs are effective biological control agents for Helicoverpa and mirids in conventional cotton. Key advantages include their tolerance to a range of insecticides making them ideal for inclusion into a multi-pest IPM program that can reduce conventional insecticide applications by 50%. However, a lack of commercial availability prevents the use of assassin bugs in cotton and with the current dominance of transgenic technologies within the market place, private sector investment in developing this biocontrol seems unlikely. Pending breakthroughs in revolutionary rearing technologies such as the advent of better artificial diets may change this scenario in the future".*



## Silver Leaf Whitefly – A Dawson Valley Perspective

### Summary

Following an initial minor outbreak on the Central Highlands during 2000-01 season, Silver Leaf Whitefly, *Bemisia tabaci* has become a regular pest of CQ cotton that require considerable crop checking inputs and control action each season. During the last three years we have monitored whitefly populations in the Dawson Valley and found that whitefly colonise crops by December with peak population densities observed by February each season. The endemic complex of parasitoids comprising predominantly of *Eretmocerus* spp. have increased in abundance each season with parasitism peaking at 87% during the 2004-05 season. The judicious in crop threshold sampling of whitefly and utilisation of IPM practices that best conserve *Eretmocerus* parasitoids are likely to remain predominant drivers for pest management activities and research in CQ for the foreseeable future.

### Background

The past three seasons have seen the CQ cotton industry face one of its largest challenges, Silverleaf Whitefly (SLW) *Bemisia tabaci* B Biotype. An introduced, sap-sucking insect, SLW has the potential to put major constraints on producing cotton in this region.

Silver leaf whitefly was first discovered in Australia in 1994, and was thought to be introduced on live poinsettia cuttings imported from the USA. The insect has been identified as a major pest on nearly every continent in the world and has a host range of at least 500 crops and ornamental plants.

When SLW was inadvertently introduced into Australia, it brought with it resistance to most organophosphate, carbamate and synthetic pyrethroid insecticides. Since then, SLW has also developed resistance to additional compounds including imidacloprid, endosulfan, bifenthrin, insect growth regulators and amitraz in some areas of Australia.

SLW was known to be present in irrigated crops around CQ for the previous decade. The warm temperatures of the area and the year-round abundance of suitable host plants for the pest highlighted CQ as a region prone to SLW outbreak. Despite this, SLW populations didn't reach problematic levels in crops until autumn of 2000-01 (April) when high numbers were found in a very late maturing cotton crop close to the town of Emerald.

In response to this minor outbreak, monitoring began on the Central Highlands in August, and through to November, it appeared that there would not be a problem in the 2001-02 cotton season. At this stage there were a small number of fields with low populations, and these were predominantly adjacent to alternative hosts such as melons and pumpkins. By December populations had increased significantly, particularly in the fields where the pest had been initially found. In addition, the SLW populations had spread to the point where they were present in almost every field throughout the Central Highlands by the end of the month.



**Photo 6** Silver Leaf Whitefly adults and eggs on cotton leaf (Photo courtesy Dave Kelly).

Through January and early February populations continued to escalate and crops were being treated regularly for the pest. Pegasus® was used quite widely (60% of the area) achieving adequate results, although given the nature of the pest, the

effects of the product were short-lived. Pegasus® use was also limited by a 35 day withholding period and a label requirement of six weeks between applications. Some knock-down products such as pyrethroid mixes (with PBO or organophosphates), used to control *Helicoverpa* offered limited control of SLW adults but provided very little residual or activity on nymphs.

Defoliation began in early February causing major migration from defoliated crops to later planted crops or those delayed by hail damage sustained in November. By mid to late February, these non-defoliated crops were receiving extreme pressure and in many cases attempts at control seemed pointless given the lack of residual control and the immediate reinfestation.

In 2001-02 it was estimated that cotton growers spent in the order of \$AUS 110 per hectare on additional insect control for SLW. This equated to approximately \$AUS 2.4 million across the Central Highlands. The widespread use of broad-spectrum insecticides for SLW late in the season decimated natural predator and parasitoid populations, which are very important for integrated pest management of all pests.

During the same season there were also lesser populations of SLW recorded in the Dawson and Callide cotton growing areas. In the Dawson valley, around the Theodore irrigation area, several crops developed considerable SLW populations very late in the season. In the Callide, some rock melon and cotton crops developed populations during March and April.

It was evident after this season that a concerted effort would be required to develop comprehensive management strategies for SLW and to modify existing control strategies for other cotton pests to ensure that *Helicoverpa* and mirid management practices were not adding to the outbreak of SLW. A basic monitoring program for SLW and their parasitoids was conducted during the 2002-05 seasons within the Dawson valley to identify potential problems and gauge the extent of local parasitoid populations.

### SLW and Parasitoid Sampling In The Dawson Valley

Sampling for SLW and their parasitoids was conducted within Dawson Valley cotton fields during the 2002-03, 2003-04 and 2004-05 seasons. SLW populations were assessed in randomly selected fields over the course of each season and the 5<sup>th</sup> node leaf of 60 plants examined in each field for nymphs and adults. At least 6 fields were sampled on each occasion. Sampling for SLW was conducted on a monthly basis throughout each season. Parasitoids were assessed at the same time as the SLW surveys were conducted. At least 200 leaves between 5 and 7

nodes were collected from each field and returned to the laboratory upon which nymphs were assessed for parasitism. SLW were recorded as nymphs and adults per 5<sup>th</sup> node leaf and parasitism as the percentage total nymphs recorded on sampled leaves.

### Results

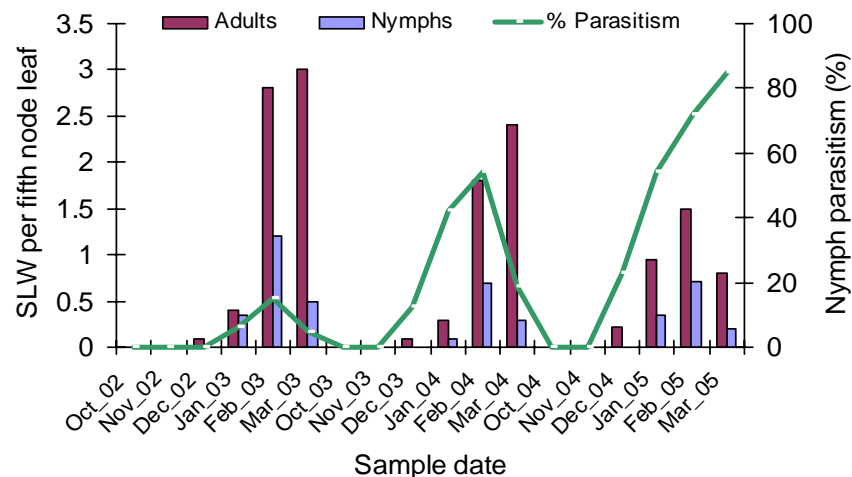
The sampling data suggest that SLW did not reach the outbreak levels reported during the previous 2001/02 season at Emerald. The lower population densities recorded during the 2002/03 season were due in part to the use of IGR's on approximately 40% of the crop combined with a near complete lapse in the use of organophosphate and pyrethroid insecticides on Dawson valley crops. Parasitism was recorded at low levels during this season (Fig 22). A further reduction in SLW densities were recorded during the 03/04 season. This decline coincided with an increase in *Eretmocerus* spp. parasitism. Pyriproxifen usage in the Dawson also declined to approximately 30% of fields treated during this season.

Parasitism by native *Eretmocerus* spp was observed to increase significantly during the 2004/05 season. This increase coincided with the full scale introduction of Bollgard® cotton varieties which comprised approximately 85% of the Dawson Valley crop area. This season was also characterised by a reduction in IGR usage with only 3-4 fields reported to be treated with Pyriproxifen.

### Conclusions

SLW were abundant throughout the Dawson Valley and reached IGR threshold levels in a number of fields. Sampling during the last three seasons suggest that SLW colonise cotton crops in very low numbers during late November and December and rapidly increase in number by January and February. Sampling has also shown that *Eretmocerus* spp. parasitoids have become increasingly abundant in each successive season. Whether the belated increase in parasitoid densities are indicative of a biological control lag phase dynamic or a direct consequence of improved in-crop pest management practices (or a combination of the two) is unclear. A classical biological control lag phase phenomena is plausible given the initial rapid establishment of SLW throughout CQ coupled with possible delays in native parasitoids adapting to take advantage of a newly abundant host source. The second scenario of grower pest management practice change is also likely as pesticide use survey data from CQ reported by Kelly *et al.* (2004) suggest radical reductions in organophosphate and pyrethroid usage after the 2001-02 SLW outbreak year. The very high adoption rate of Bollgard® cotton varieties by

growers during 2004-05 season resulted in a further reduction in broad spectrum insecticide usage would also support this hypothesis and perhaps account for the peaks of parasitism in excess of 80% observed during the same season.



**Figure 22.** The population dynamics of adult and nymph SLW on fifth node leaves and the rate of nymph parasitism by *Eretmocerus* spp. recorded during the 02/03, 03/04 and 04/05 cotton seasons.

It is evident from the data that SLW are likely to be a frequently occurring pest within the CQ environment and that future pest management strategies will need to account for the conservation of *Eretmocerus* spp. parasitoids that appear to be an important source of SLW mortality within cotton fields.



*"Sampling has identified that native species of Eretmocerus parasitoids are well established within the Dawson Valley and if conserved can act as an effective biological control agents within a SLW IPM program. It is imperative that all other pest management practices are compatible with the conservation of this important group of natural enemies".*



## Moth Busting for Resistance Management

### Summary

End of season trap cropping has been an integral component of the *Bt* resistance management strategy for CQ where pupae busting is potentially ineffective due to low *Helicoverpa armigera* diapause rates. However, the effectiveness of the trap crop strategy is unknown and recent drought years have seen a reduction in the quality of trap crops grown throughout CQ thus casting further doubt on resistance management efficacy. The area wide use of the attract and kill product Magnet<sup>®</sup> offers an alternative option to trap cropping for targeting resistant escapes from Bollgard<sup>®</sup> fields. A large scale pilot study conducted during the 2004-05 season suggested that area wide applications of Magnet<sup>®</sup> had significant impacts on *Helicoverpa* and *Spodoptera litura* moth populations and that potential exists to use regional applications of Magnet<sup>®</sup> as a quantifiable and more effective alternative to trap cropping.

### Background

Growers of Bollgard<sup>®</sup> cotton varieties in central Queensland are required to undertake a number of preventative resistance management actions. Typically this involves the growing of an unsprayed refuge of pigeon pea as well as an additional later sowing of pigeon pea as a trap crop. The refuge serves the purpose of generating additional susceptible *Helicoverpa* to dilute potentially resistant individuals emerging from Bollgard<sup>®</sup> crops whilst the trap crop serves to attract the offspring of the last *Helicoverpa* generation to emerge from cotton so that any insecticide resistance developed over the season can be confined and minimised (Sequeira 2001).

The trap cropping strategy has been implemented in CQ since 1997. Patches were required to comprise the greater of 1% or 2 hectares of total farm area. These patches are sown with pigeon pea after the main cotton crop is established and are ideally managed so that the trap crop is at peak attractiveness after the main cotton crop has cut out and ceases to be as attractive to *Helicoverpa* moths (Sequeira 2001). In the 8 years since the introduction of this strategy, several problems have tended to emerge. Firstly the efficacy of trap crops for attracting *Helicoverpa* moths emerging from adjacent cotton fields and capturing their progeny has not been well quantified. Secondly the last 4 years have been impacted by drought conditions which have resulted in some growers not irrigating pigeon pea trap crops in a manner that ensures that they are at peak

attractiveness after cotton crop cut out. The efficacy of poorly managed pigeon pea trap crops is questionable. Thirdly with the release of stacked Bollgard<sup>®</sup> cotton varieties it has become even more important that CQ has an effective strategy for targeting and destroying the final generation of *Helicoverpa* that emerge from cotton fields as the reduction in Cry1ac expression in Bollgard<sup>®</sup> II varieties is known to decrease after cutout allowing for a greater frequency of escapes.

With the industries dependence on transgenic *Bt* varieties likely to remain in the foreseeable future, the need to seek improvements to the current resistance strategy will remain a research priority. Trap crops represent a potential weak link within the *Bt* resistance management strategy for CQ. However, alternatives to this strategy have not existed until recently.

A new opportunity to refine the CQ strategy presented itself in the form of Magnet<sup>®</sup>, a Cotton CRC developed product that is attractive to foraging *Helicoverpa* spp. moths. Magnet<sup>®</sup> contains select volatiles that simulate native flora known to be attractive to feeding *Helicoverpa* moths. Typical use involves lacing Magnet<sup>®</sup> with an insecticide and applying it to crops in widely spaced bands to attract and kill *Heliothis* moths that may be active in the area.

With the imminent commercialisation and registration of Magnet<sup>®</sup>, potential exists to develop an alternative strategy for targeting last generation *Helicoverpa* spp moth populations than trap cropping. Compared to the current regional trap cropping program, the use of Magnet<sup>®</sup> on a regional scale could offer the following advantages:

**Strategic** – Can be better timed to coincide with last generation moths.

**Direct** – Kills female and male resistance gene carrying moths directly and it takes the trap to the source crop rather than being remotely located.

**Measurable** – Unlike trap cropping, the impact of Magnet<sup>®</sup> on local *Heliothis* populations can be measured

**Economic** – Magnet<sup>®</sup> cost is offset by savings from not trap cropping

**Uniform** – The whole region would be treated at the same time in the same way.

**Easy** – The product can be applied aerially.

**Proactive** – The CQ industry will have a refined unique strategy and be seen as taking proactive responsibility in preserving *Bt* technology.

A large scale pilot study was conducted in the Dawson Valley to assess the potential for using Magnet<sup>®</sup> as an alternative tool for trap cropping in CQ.

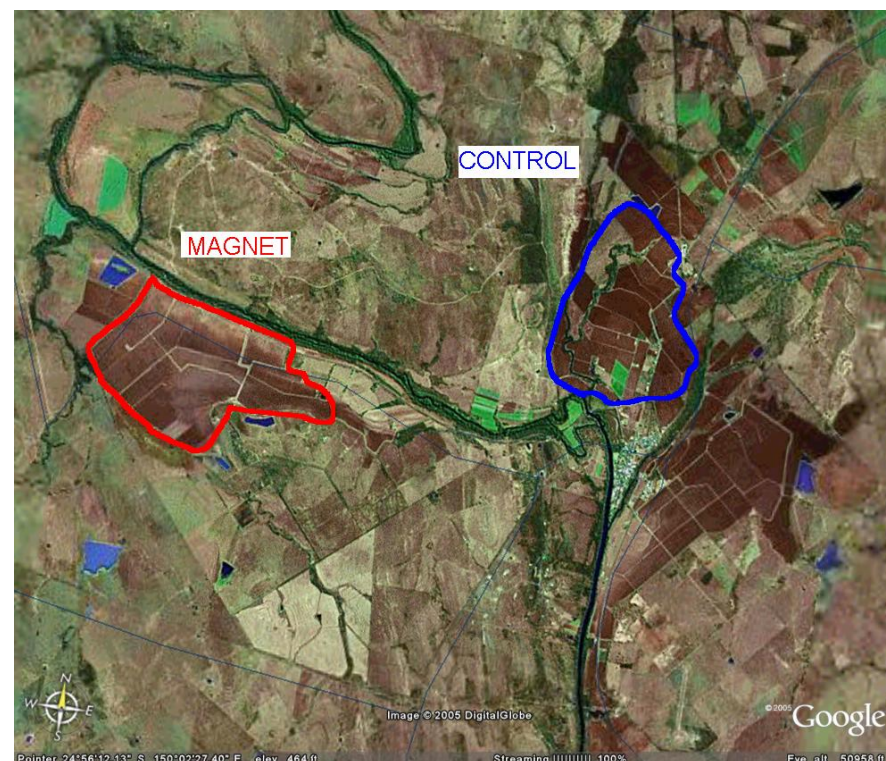


## Materials and Methods

Two 800 hectare patches of Bollgard® II cotton were chosen in the Theodore region for the pilot study. These patches were approximately 5 km apart and were separated by mixed cropping and native vegetation (Fig 23). The patch on the western side of the Dawson irrigation area was chosen for treatment as it was considered to be predominantly downwind of the control.

*Helicoverpa* spp. moth populations were assessed pre and post treatment using light traps and flush counts. Four light traps were placed throughout each of the two treatment patches (8 traps in total) and cleared of insects every day. The contents of each light trap were stored in ethanol and returned to the laboratory for examination at a later date. Flush counts were made on randomly selected fields throughout the two patches either every day or second day to estimate moth densities per hectare of cotton. Flush counts were conducted by gathering a 5 litre bucket of soil at the outside of each field and then walking a 100m transect into the field throwing handfuls of soil at the preceding crop and counting *Helicoverpa* moths as they emerged. We calibrated our flush count technique during the initial stages by following disturbed moths to the point where they could be identified as *Helicoverpa* or some other species. This “calibration” suggested that at least 70% of the flushed moths being counted were likely to be *Helicoverpa* species. Eight transects were conducted in each of the two patches at each sampling occasion.

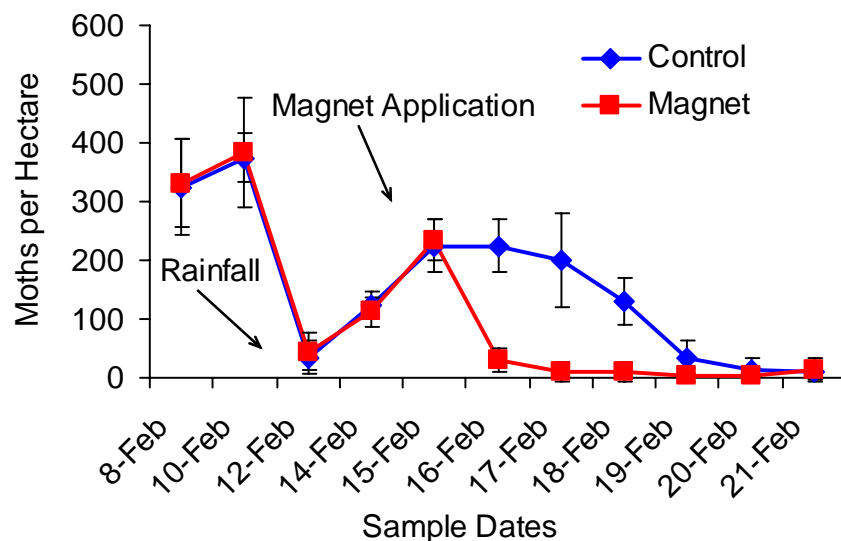
The Magnet® treatment was applied by aircraft on the afternoon of the 15 February®. The Magnet® was mixed with methomyl insecticide according to label directions and applied in 1 metre wide bands approximately 72m apart over the entire Gibber Gonyah cotton area.



**Figure 23.** An aerial photo showing the Theodore channel irrigation area. The area outlined in red represents the Gibber Gonyah area that was treated with Magnet®/methomyl mix. The untreated area used as a control comparison is outlined in blue in the cotton area north of Theodore (Photo from www.Google Earth.com).

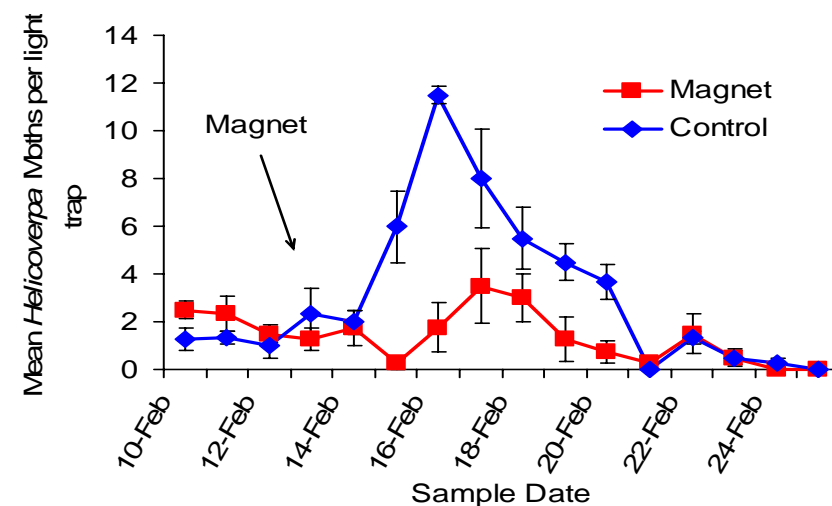
## Results

The Magnet<sup>®</sup> treatment had an immediate impact on the Gibber Gonyah moth population with a reduction of 97% recorded during the first 48 hours post-treatment (Fig 24).

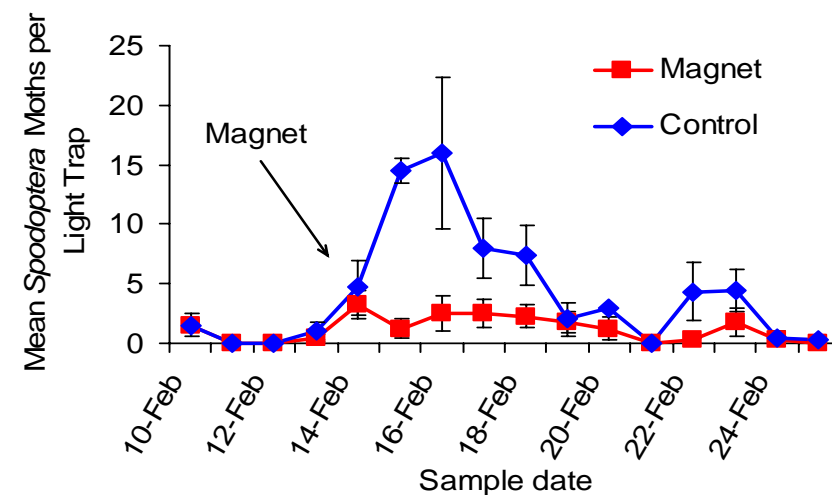


**Figure 24.** The mean estimated number of *Helicoverpa* moths recorded per hectare using flush counts in the control and Magnet<sup>®</sup> treatment areas.

Light trap catches during the experiment also suggested a reduction in *Helicoverpa* numbers post-treatment compared to the control (Fig 25). A similar impact was also recorded for *Spodoptera litura* a secondary cotton pest of which the larvae were observed abundantly throughout Bollgard<sup>®</sup> II crops during the season (fig 26).



**Figure 25.** Mean number of *Helicoverpa* moths caught per trap in the control and Magnet<sup>®</sup> treatment areas.



**Figure 26.** Mean number of *Spodoptera litura* moths caught per trap in the control and Magnet<sup>®</sup> treatment areas.

## Discussion

The results suggest that Magnet<sup>®</sup> mixed with Methomyl caused an immediate and significant reduction in *Helicoverpa* moth densities within the treatment area. The impact of the treatment was also significant for *Spodoptera litura* which suggests that the use of Magnet<sup>®</sup> for moth busting as a replacement for trap cropping may have additional benefits for secondary pest management.

A notable trend within the results was the decline in *Helicoverpa* moths recorded in the control several days post-treatment. The causes for this are unknown but it is probable that the treatment impacted the control area by either influencing patterns of random moth exchange between the two areas or actually attracting moths away from the control area. Light traps set up 7 days after application approximately 15km from the treated and control areas suggested that moths were more abundant in the wider region, thus suggesting a localised impact may have occurred. Another possibility was that local moth populations were already in decline and that the Magnet<sup>®</sup> treatment hastened that process.

Although preliminary, the results suggest that Magnet<sup>®</sup> had a significant impact on *Helicoverpa* moth densities and that the targeting of populations at a regional level may be a possible application for this technology. The ability to manipulate *Helicoverpa* populations at a regional scale has not previously existed and the potential applications for such a tool could be many and varied. For the purposes of resistance management, the targeted use of Magnet<sup>®</sup> at seasons end could provide a more effective alternative to trap cropping and may also have other unexplored resistance management applications.

As these results are preliminary, further research will be needed to ascertain the full impacts for using Magnet<sup>®</sup> on a regional scale. These results need to be replicated together with more accurate assessments being made on treatment impacts. Future experiments will need to consider potential use patterns in terms of Magnet<sup>®</sup> application technique as well as non-target implications.

The use of Magnet<sup>®</sup> for regional management of *Helicoverpa* and other lepidopterous pests appears to be highly promising. Future research will consolidate these findings and focus on scientific and technical questions.



*"The pilot study suggests that Magnet<sup>®</sup> has significant potential to be used as a moth busting tool on an area-wide basis and that this may have significant implications for Bt resistance management in CQ and the broader industry. A likely application for this technology is for the replacement of trap cropping. However, other resistance management applications may exist and will be the focus of future research" (Photo courtesy Anthony Hawes)*



## Economic, Environmental and Social Outcomes from DAQ 122C

Our research fulfils the CRDC's triple bottom line expectations. Improved pest management through the implementation of better whitefly, *Helicoverpa* and *Bt* resistance management strategies contribute uniquely to each of these objectives.

From an economic perspective the development of effective management strategies for silver leaf whitefly is essential to the economic viability of cotton production in central Queensland as well as the broader industry. Should Australia's image for producing quality cotton be tarnished by stickiness from whitefly, the economic impacts on CQ growers and the wider industry would be significant. This project has contributed to the industries ability to manage silver leaf whitefly through the extension of best management practices to local growers as well as the collection of population dynamics data that has identified the importance of selecting soft option insecticides that conserve native *Eretmocerus* spp. parasitoids. These parasitoids have been shown to be effective mortality agents of silver leaf whitefly and if conserved can reduce the need for expensive IGR product usage thus saving \$100 per hectare of cotton grown. The development of better resistance management tools such as the regional use of Magnet® for moth busting will also serve to prolong the serviceable life of transgenic technologies such as BollGard® and thus constrain insect related production costs.

Each of the projects research activities have focussed on developing more environmentally sustainable pest management methods for growing cotton. The use of assassin bugs for control of *Helicoverpa* and mirids was shown to reduce pesticide inputs by over 50% without compromising yield. The use of Magnet® and better trap crops for area wide management that serve to minimise the development of *Bt* resistance will support pesticide reducing technologies such as BollGard® whilst the conservation of *Eretmocerus* parasitoids will limit insecticide usage for whitefly control. Each of these outcomes reduce insecticide usage, therefore lessening the environmental impact of cotton.

The reduction in insecticide usage and increased industry profitability that is supported through the development of pest management technologies investigated during this project have social benefits in terms of creating a better living environment which is conducive to greater harmony between cotton and non cotton residents. The effective management of whitefly has largely reduced pest influxes that were affecting local town residents and thus had beneficial social

implications. More profitable cotton production also serves to support the local economies of these communities of which cotton is a significant economic driver.



*"The research and adoption of more sustainable and effective management strategies for silver leaf whitefly (facilitated in part by this project) have succeeded in alleviating the social impacts that this pest via the cotton industry was having on local communities in central Queensland. With ongoing refinements to the whitefly management strategy, scenes like the 2001-02 plagues that greatly diminished air quality are unlikely to be repeated". (Photo courtesy Greg Jensen).*



## Future Research & Further Results Exploitation

Some of the research conducted under DAQ122C will be further exploited and consolidated within the new CQ Farming System Project funded by the CRDC. In particular the potential applications for using Magnet® for *Bt* resistance management will undergo more rigorous testing with a view (if successful) to implementing a modified *Bt* resistance strategy for CQ regarding end of season moth busting. Consideration will also be given to identifying if other potential applications for this technology exist with regard to area-wide management.

The whitefly research conducted during this project, has highlighted the importance of *Eretmocerus* spp. conservation within CQ cotton crops. With the advent of Bollgard® varieties that virtually eliminate the use *Helicoverpa* targeted insecticides, it is probable that the most disruptive management practices to affect resident whitefly parasitoids in cotton will be insecticide applications for mirid control. Insecticide use strategies have already changed for mirid management with the industry abandoning organophosphates such as dimethoate and omethoate in favor of low rates of Fiprinol. However, this has lead to a second potentially undesirable situation where the industry is completely reliant on one product (fiprinol) for mirid management.

Recent research by Dr Moazhem Khan (QDPI&F) has suggested that organophosphate chemistry can be used more safely with natural enemies by reducing the applied rate and adding salt with no loss of control efficacy. To investigate the potential to use of low rate organophosphates for mirid control a pilot study was conducted during the 2004-05 season (not reported here) that suggested such an approach may be compatible with whitefly management and alleviate the current situation whereby the CQ industry is largely reliant on one insecticide. This approach will be fully explored during the new Farming Systems project with a view (if successful) to exploiting the use of low rate organophosphates to re-expand the mirid management tool kit for CQ.

The Dawson Valley whitefly data gathered during DAQ122C will also be incorporated into a whitefly booklet being prepared by Dr Richard Sequeira (QDPI&F) for later publication and circulation within the industry.

## DAQ 122C Publications

The trap crop research has been published during this project, details of which are provided below. A manuscript encompassing the assassin bug field trials and insecticide testing has been formatted into a manuscript and will be submitted for publication in the coming months. The whitefly survey data will be included into a manuscript being prepared in collaboration with Dr Richard Sequeira for submission also in the near future. Publications from DAQ122C are listed below.

### Refereed Journals

Grundy PR, Sequeira R & Short S. 2004. Suitability of legume species as trap crops for management of *Helicoverpa* spp. (Lepidoptera: Noctuidae) in central Queensland cotton cropping systems. ***Bulletin of Entomological Research*** 94: 481-486.

Scott KD, Wilkinson KS, Merritt MA, Scott LJ, Lange CL, Schutze MK, Ketn JK, Merritt DJ, Grundy PR & Graham GC. 2003. Genetic shifts in *Helicoverpa armigera* Hübner (Lepidoptera: Noctuidae) over a year in the Dawson/Callide valleys. ***Australian Journal of Agricultural Research*** 54: 739-744.

### Industry Publications

Kelly D, Sequeira R & Grundy P. 2003. Managing whitefly in central Queensland. ***Australian Cottongrower*** 24(5): 32-36.

Grundy P & Short S. 2003. Field Peas- A potential alternative to chickpea for trap cropping. ***Australian Cotton Grower*** 24(3): 14-17

### Conference Proceedings

Grundy, PR, Kelly D & Sequeira RV. 2004. The Silver Leaf Whitefly Management Challenge: A New Pest in Central Queensland. 12<sup>th</sup> Australian Cotton Conference Proceedings, Gold Coast.

Grundy PR & Short KS. 2004. Field Peas for Trap Cropping In Central Queensland. 12<sup>th</sup> Australian Cotton Conference Proceedings, Gold Coast.

Grundy PR and Short KS. 2004. Assassin Bugs and Cotton IPM, Prospects and Limitations. 12<sup>th</sup> Australian Cotton Conference Proceedings, Gold Coast.

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## Acknowledgements

We would like to acknowledge and thank the following people and organisations for their contributions to this project.

Mr Dave Kelly (formerly QDPI&F) for his assistance with several grower field days, preparation of industry publications and technical advice.

The QDPI&F Biloela Research Station staff for their assistance in maintaining various field trial experiments throughout the project.

Ms Nicole Purvis-Smith and Mr Les Redman for their technical assistance in conducting the spring trap crop experiments on the Biloela Research Station.

Mr Anthony Hawes (Ag Biotech) for his assistance with the Magnet<sup>®</sup> moth busting trial work and Ag Biotech for providing product.

Assoc Prof Peter Gregg for his advice regarding the implementation of the moth busting trials at Theodore.

The growers of the Dawson Valley, who have provided us with access to their properties on different occasions for sampling or trial work. In particular we would like to thank the Anderson, French and Gee families for accommodating the refuge and trap crop experiments on their properties.