Australian Government
Cotton Research and Development Corporation

FINAL REPORT 2017

Choose an item.

Part 1 - Summary Details

Please use your TAB key to complete Parts 1 & 2.

CRDC Project Number: CSP1401

Project Title: Enhancing IPM in cotton systems

Project Commencement Date: 1 July 2013 **Project Completion Date:** 30 June 2018

CRDC Research Program: 1 Farmers

Part 2 – Contact Details

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Signature of Research Provider Representative: _____

Date Submitted: _____

Part 3 – Final Report

(The points below are to be used as a guideline when completing your final report.)

Background

1. Outline the background to the project.

Bt-cotton is challenged by pests including mirids, green vegetable bug (GVB), Silverleaf whitefly (SLW), thrips, mites and aphids. Emergent pests have developed including pale cotton stainer, cluster caterpillar, Solenopsis mealybug, spur-throated locust and broad mite. A previous project "IPM for silver leaf whitefly and emerging pests in central regions' emphasised development of knowledge and tools to support integrated pest management (IPM), including emerging pests.

That project developed methods and initiated studies to provide better understanding of SLW population dynamics and fate of honeydew on lint. It found that SLW use a range of hosts through-out the year, though research was hampered by rain so further effort was needed. Preliminary studies also showed that mortality of SLW from eggs to adults in cotton was often high but it was difficult to ascribe sources of mortality accurately. Techniques were developed to assess the level of honeydew contamination of cotton bolls and early observations suggested that rainfall can dramatically reduce honeydew levels.

The IPM fit of new insecticides and semiochemicals was investigated and reported in the Cotton Pest Management Guide, however, a range of new options needed testing. Techniques were developed to culture SLW and initiate field infestations. Experiments using these techniques showed that mirid management can dramatically increase the risk from SLW.

Research with Murray Sharman (DAFFQ) showed several Malvaceous weeds are CBT hosts and there is more than one strain of CBT.

The 5year project reported herein was proposed to follow on from previous research with a review of milestones after 3 years, which would coincide with the introduction of Bollgard III®. The project initiated several new research areas. It provided the people, skills and experience to be able to rapidly respond to emerging pest issues, including exotic species and supported the provision of mites and aphids for resistance testing by Dr Grant Herron.

Objectives

2. List the project objectives and the extent to which these have been achieved, with reference to the Milestones and Performance indicators.

A. To improve knowledge of and management of SLW by:

i. Identifying factors contributing to reductions in honeydew on cotton and implications for cotton fibre quality and defoliation.
ii. Identifying seasonal host use for SLW whitefly.
iii. Assessing mortality on cotton through the cotton season and identifying potential causes. There is potential to provide samples for a companion project 'Identifying predators of emerging pests in cotton systems' developed separately with Prof James Harwood (U. Kentucky). The companion project would use species specific primers for green vegetable bug and SLW to identify beneficial species that eat them. This will allow beneficial species to be targeted to consider consumption capacity, options to increase abundance and inclusion in IPM decision making.
iv. Undertaking sampling to understand the within plant and within field distribution of SLW adults and nymphs.

v. Working with Dr R. Sequeira (QDAFF), CRDC and the CCA to use data from (d.) to evaluate the suitability of current sampling recommendation and develop changes/additions to address industry concerns.*

B. To provide tools for IPM by:

i. Assessing the efficacy and non-target effects of new insecticides, biopesticides and semiochemicals.
ii. Testing options to manage mirids and GVB with reduced risk of flaring SLW or other secondary pests.
iii. Exploring options for alternative to the neonicotinoid seed treatments for control of thrips.
iv. Assessing the relationship between boll age and susceptibility to GVB damage
v. Improving understanding about insecticides used to manage whiteflies.

C. Manage early season damage by:

i. Assessing seed treatments and measure the effect of early season thrips damage on plant growth, yield and maturity in southern regions and/or assist NSW DPI (S. McDougall and J. Mo) in design and interpretation of research into thrips ecology and management in southern regions.

D. Understand Cotton Bunchy Top disease by:

i. Identifying alternative host species.
ii. Investigating the effectiveness of insecticide application to prevent spread of CBT.

E. Identifying and managing emerging pests by:

i. Providing flexibility to undertake research to manage emergent/exotic pests, including those arising due to changes in the farming system.

F. Investigating the effect of late season thrips damage to flowers on yield.*

G. Investigating the effect of early tip and fruit damage on yield and maturity of Bollgard. 3**

*This milestone was added after review of project directions in 2015 with CRDC.

** This milestone was added after heavy early season pest pressure in 2016/17 and followed on from * above.

Obj No.	Objective *See above re character limit*	NO.	Milestone *See above re character limit*	NO.	*Performance Indicator* *See above re character limit*	*Start Date (dd/mm/yy)*	*Finish Date (dd/mm/yy)*	*Comments*
1	Measure decline in honeydew	1.1	Experiments developed from previous project assessing effect of rain, UV, microbial degradation and effect on fibre quality and defoliation.	1.1	Data collected and analysed for 2013, 2014 and 2015	*1/7/2013*	*30/6/2016*	*Achieved in full*
		1.2	Reviewed honeydew research, revise milestones and establish options to package for industry with extension.	1.2	Review completed and new milestones developed.	*1/7/2016*	*30/9/2017*	*Achieved- ties in with CRDC Colour Projects*
		1.3	Finalise research and development of extension materials	1.3	Extension materials available and final report completed	*1/7/2016*	*30/6/2018*	*Achieved*
2	Assess SLW host use and SLW mortality factors	2.1	Review methodology and establish protocols for next 3 years based on previous project.	2.1	Data collected and analysed for 2013, 2014 and 2015	*1/7/2013*	*30/6/2016*	*Achieved in full*
		2.2	Assess progress and need for further research	2.2	Data collated, analysed and outcomes reviewed	*1/7/2016*	*30/9/2015*	*Achieved in full*
		2.3	Based on 2.2 finalise research and develop extension materials	2.3	Extension materials available and final report completed	*1/7/2016*	*30/6/2018*	*Achieved*
3	Assess IPM fit and efficacy of new control options,	3.1	Contact companies and Robert Mensah to define new chemistries, biopesticides or semiochemicals for evaluation as foliar sprays or seed treatments. Review annually.	3.1	Experiments completed, report written and cotton pest management guide updated	*1/7/2013*	*30/6/2018*	*Achieved in full*
4	Management options for mirids and GVB and efficacy of seed	4.1	Decide treatments for management of mirids and	4.1	Experiments completed and analysed each year.	*1/7/2013*	*30/6/2016*	*Achieved in full*

	treatments and manage early season damage (damage experiments)		**GVB and for managing early damage experiments and complete 2 experiments. Complete three experiment for seed treatments.**					
		4.2	**Review outcomes from 3.2 and decide need for and further research**		**Review completed and decision taken for directions of final two years**	***01/07/2016***	***30/06/2018***	***Achieved and research halted for final year of project***
5	**Manage CBT**	**5.1**	**Review current known alternative hosts and decide on further candidates to test, integrate with field collections of hosts to assess significance of hosts identified.**	**5.1**	**Hosts tested and field collections made and tested for presence of CBT**	***1/7/2013***	***30/6/2016***	***Achieved in full***
		5.2	**Experiments designed to assess effect of new chemistry on transmission of CBT by aphids.**	**5.2**	**Products tested and data reported.**	***1/7/2013***	***30/6/2016***	***Achieved in full***
		5.3	**Review directions of future research with CBT.**	**5.3**	**Review completed and final directions set. New milestones developed.**	***1/7/2016***	***30/6/2018***	***Achieved***
6	**Management of emergent/exotic pests and identification of potential farming system changes that could influence this.**	**6.1**	**Maintain contact with extension, CCA, other researchers and resellers to identify emerging issues as early as possible, including pests and changes in the cropping system.**	**6.1**	**Evidence of understanding emerging issues**	***1/7/2013***	***30/6/2018***	***Achieved***
		6.2	**Based on 6.1, assemble relevant information and assist extension in developing appropriate extension pathways and media**	**6.2**	**Evidence of working to develop extension media**	***1/7/2013***	***30/6/2018***	***Achieved***

		6.3	**Based on 6.1, review need for and if necessary design and complete research to allow for improved management of emerging pests**	**6.3**	**Experiments designed and completed**	***1/7/2013***	***30/6/2018***	***Achieved***
7	**Sampling for SLW**	**7.1**	**Review needs with Dr Sequeira, CRDC and CCA and design sampling strategy**	**7.1**	**Completed 3 seasons of sampling to obtain data on SLW within plant and within crop distribution and factors affecting these**	***1/7/2015***	***30/6/2018***	***Achieved in full***
		7.2	**Review outcomes from data with Dr Sequeira, CRDC and CCA.**	**7.2**	**Meetings with CRDC and CCA to review potential changes to sampling strategies**	***1/7/2017***	***30/6/2018***	***Achieved, led to new SLW Validation Project***

Note variations:
V1 - Milestone 4.2 – end of effort in 2016
V2 - Milestone 7 reflects the request from CRDC for diversion of effort and replaces Milestones 6.2 and 6.3 - 2017

3. Detail the methodology and justify the methodology used. Include any discoveries in methods that may benefit other related research.

Section A. To improve knowledge of and management of SLW by:

The Silverleaf whitefly (SLW) has been an important pest of cotton in central Queensland (CQ) since the initial outbreak of this pest in 2001. Over the last five years (2012-2017), SLW has grown in pest status in areas outside CQ, notably the Macintyre, Gwydir and Namoi valleys and is now considered a key pest of cotton in these areas. The incidence of SLW has gradually increased to the point where chemical control measures are now often required to prevent honey dew contamination of lint and to limit any adverse impacts on profitability and overseas marketing potential.

The impact of SLW on the cotton industry is two-fold. Firstly, stickiness of cotton lint contaminated with honeydew that results from SLW feeding activity has the potential to make Australian cotton unattractive to overseas buyers. Secondly, the accumulation of SLW honeydew encourages the growth of fungi that cause discolouration which can attract significant quality downgrades or, in extreme situations, even make the lint unmarketable. In this section we approached the issue of SLW from a number of different perspectives to increase knowledge of the pest and develop improved management techniques and/or systems to minimise its threat to clean and economically lucrative cotton production.

(i) Identifying factors contributing to reductions in honeydew on cotton and implications for cotton fibre quality and defoliation.

- **Rainfall and UV Radiation**

Honeydew research over the last two seasons was reviewed. We have conclusively shown that rainfall (10-20 mm) is sufficient to wash off honeydew contamination in the field. This has been presented to industry at several grower meetings, at the CCA AGM in May 2014 and at the Australian Cotton Research Conference (September 2013).

Looking forward, key questions remained including;

a. How effective are smaller rainfall events at removing honeydew – this is a gap in current data sets.

b. Clarify the effect of UV radiation on honeydew breakdown

c. Investigate causes of ‘mouldy’ bolls

d. Investigate methodology – number of puffs by recovery of honeydew

e. Determine the relative proportions of honeydew on the surface and inner lint of open cotton bolls

f. Analyse the sugar profile of mealybug honeydew

g. Determine which honeydew sugars are metabolised and degraded by alternatives to sooty mould fungi (baker's yeast)

In order to understand the processing methodology of cotton and quality issues better, Dr. Heimoana enrolled in the Cotton Fibre Field to Fabric Course in Geelong in August 2014. This was important with respect to the honeydew work, colouring issues from sooty mould, cotton stickiness and enabled her to collaborate with the Fibre Technology Group there to get samples tested for quality assessment.

Our long-term collaboration with Dr Michael O'Shea at BSES for analysis of sugars ceased as he was appointed to a senior position in Sugar Research Australia. His laboratory was no longer able to undertake work for non-sugar industry projects. Dr Anne Rae, CSIRO Agriculture and Food, agreed to undertake analyses in her laboratory at the Biosciences Precinct at the University of Queensland (UQ). Dr O'Shea has made available his methods and offered support in getting analysis up and running in Dr Rae's Lab. Dr Donna Glassop ran the analyses and liaised with Dr Heimoana. A request was lodged with CSIRO Capital Equipment funds and use to purchase additional equipment to expedite sugar analysis.

Honeydew bolls were generated by contamination with aphid, SLW or artificial honeydew. They were exposed to field situations, collected, washed and analysed for sugars (Dr O'Shea, BSES). The effects of rainfall, UV, sooty moulds and other microbes on honeydew were studied.

a. Honeydew removal by simulated small rainfall events - done at Andrew Watson's Kilmarnock, Boggabri

The aim of this work was to improve understanding of the relationship between honeydew removal and rainfall. A second experiment was carried out at Kilmarnock, Boggabri, in collaboration with Andrew Watson. Results for the 2012-13 season were reported in the Final Report for Project CRC1102. Those results showed a strong relationship between the reduction in concentration of honeydew on cotton and the amount of 'rainfall' (both natural rainfall and that applied by the sprinkler).

Methods

This experiment focussed more strongly on collecting data in the 0 – 15 mm range where a gap exists in current information. We contaminated bolls with artificial honeydew and placed them into the crop canopy – either above the canopy (top) or within the canopy (mid). Bolls were then exposed to consecutive and cumulative applications of water from an overhead irrigation system. After each run, a set of bolls was collected. Bolls were also placed in an adjacent furrow irrigated field to allow us to separate the effects of rainfall from those of the irrigator. Rain gauges were placed in both fields to estimate the amount of 'rainfall' the crop received from rain or overhead irrigation. After each pass of the irrigator, or each rainfall event a 'set' of bolls was collected, honeydew was extracted and its concentration analysed by Dr Donna Glassop at Dr Rae's laboratory at the CSIRO component of the Biosciences Precinct at UQLD.

Results & Discussion

Irrigation and rainfall both removed significant amounts of sugar (Fig. 1). More honeydew was removed from bolls placed above the canopy (0.26 mg sugar/ml, reduction of 78%) than those within the canopy (0.39 mg sugar/ml, reduction of 67%) ($P < 0.001$). This difference was

expected given that bolls within the canopy are probably protected slightly from rainfall. We combined the data for bolls in the mid canopy for this experiment with data from all other experiments and re-analysed using non-linear regression. There is a good fit through the data with over 70% of the variability explained (Fig. 2). Further data with 'real' rainfall at both higher and lower amounts would be useful though for now we have managed to set useful parameters for honeydew wash-off by rainfall.

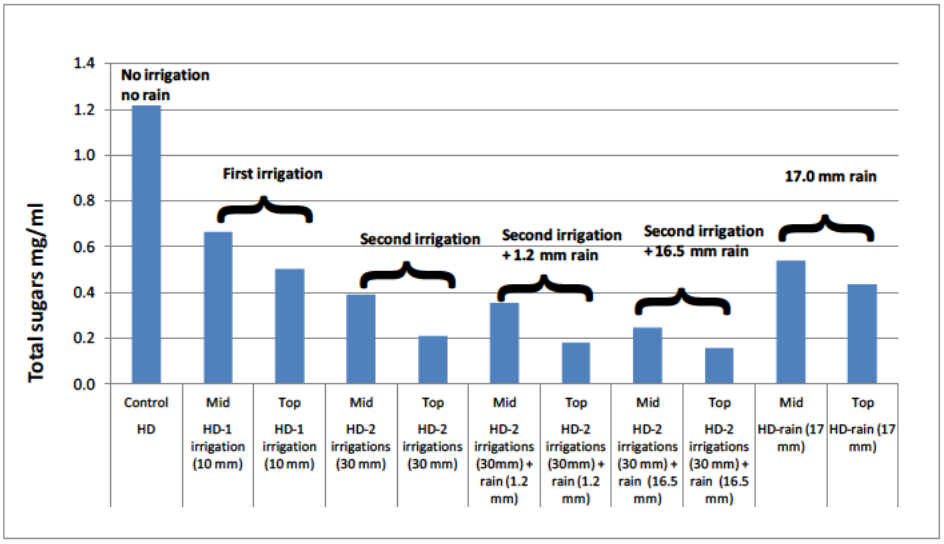


Figure 1: Effect of overhead irrigation and rainfall on total sugars remaining on contaminated bolls at Kilmarnock, 2013/14.

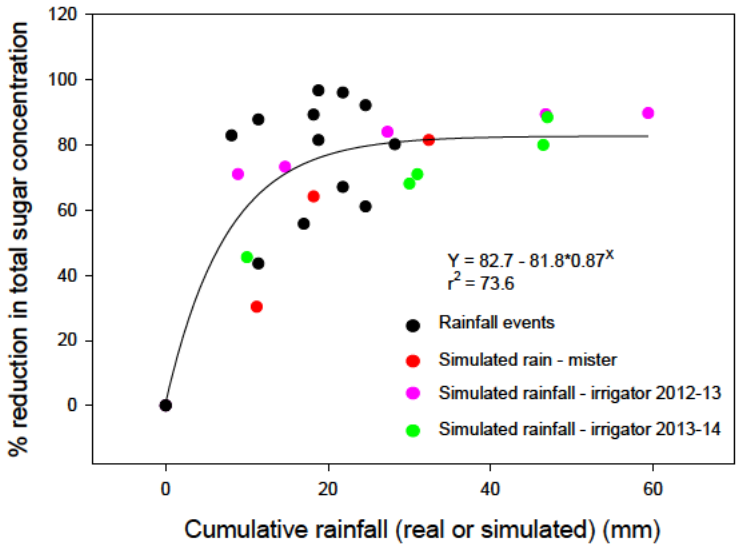


Figure 2: Relationship between cumulative 'rainfall' and the % reduction in total sugars on artificially contaminated bolls placed in the mid canopy for all years 2011-2014.

b. The effect of UV radiation on honeydew contaminated cotton bolls

Previous data suggested there was little direct breakdown of honeydew sugars by UV, but these experiments were often confounded by rainfall or were exposed to UV for relatively short periods (<7 days) as reported in the Final Report for CRC1102. The experiments conducted

within this project intended to add to and confirm existing data but over a longer time frame;

Methods

2013/14 UV Experiment 1: This experiment was an extension of a UV experiment conducted in 2011/12 UV which ran for 4 days but did not achieve significant honeydew breakdown during this period, and a UV experiment conducted in 2012/13 which was washed out by a storm. We attempted to compact this experiment spatially by pinning bolls onto a fence line (Fig. 3A) to expose them to UV radiation, rather than distributing them in a field since that would enable easier management of possible rain. Each treatment set consisted of 100 bolls with artificial honeydew and 100 bolls without honeydew, each set divided into 10 bolls x 10 reps. These sets were pinned to the fence line (Total of 400 bolls). Honeydew covered bolls received 5 puffs of artificial honeydew. Anticipating wet weather, we covered all the bolls with clear plastic to avoid water washing the honeydew off (Fig. 3B). Birds and ants feeding on the honeydew were initially a problem, but after the plastic went on, bolls were relatively protected. We included a tiny tag data logger under the plastic to estimate the microclimate there. While this approach did not allow us to manipulate UV degradation, as the plastic reduced this exposure, it allowed us to look at the long-term effects of the presence of the sugars on the fibres. Bolls covered in artificial honeydew and control bolls were exposed for 1 to 4 weeks to assess how much honeydew changed. Each week, one set of control and honeydew bolls was collected, sugar was washed out and samples were kept in the freezer for analysis. A set of bolls was kept immediately after treatment as controls (week 0) and placed in trays in the freezer to avoid loss of honeydew to plastic bags. This set allowed us to estimate the initial honeydew concentration of bolls.

Figure 3: A (left). Bolls exposed to UV radiation; B (right). Bolls protected from rain with clear plastic cover

2013/14 UV Experiment 2: In order to eliminate the possibility of rain and moisture, ants and sooty mould fungi interfering with the artificial honeydew on bolls, the experiment was further compacted. Sample size was halved to 5 bolls per rep and control and honeydew contaminated bolls (5 puffs per boll) were again exposed to UV radiation for extended time periods by being pinned to a portable clothes frame that was moved under cover when there was a threat of rain (Fig. 4). Clothes stands were exposed to direct sunlight from 8 am to 4 pm and were moved to a glasshouse overnight to avoid dew and rain. The feet of the stand were sprayed with surface

insecticide to prevent ants crawling up them. Because of the dry conditions, no sooty mould fungi developed over the 4-week period of exposure.

Figure 24: UVA3 Experiment with bolls pinned to clothes stand

2014/15 UV Experiment 3: We repeated Experiment 2 and this experiment was designed to confirm earlier findings about the contribution of sunlight on the breakdown of artificial honeydew on bolls. This experiment included an additional treatment to allow some bolls to also be exposed to dew as well as sunlight to see if this changed the rate of breakdown of the honeydew. The experiment used the method where we pinned bolls to a clothes stand (described above). Treatments included control bolls and honeydew bolls (5 puffs per boll). The stands were placed in a location exposed to full sunlight during the day. At night half of the racks were moved under cover, to prevent dew falling on them and half were left exposed to dew. If rainfall was forecast all racks were moved under cover. A set of bolls was collected after one week (week 1), another at three weeks (week 3) and a final set at five weeks (week 5). See Appendix 1 for 2015/16 UV Experiment 4 and 2016/17 UV Experiment 5.

Results

2013/14 UV Experiment 1: This experiment exposed artificial honeydew contaminated and control bolls to UV radiation for up to 4 weeks. Bolls were exposed for a minimum of 10 days (W1) and a maximum of 37 days (W4). During the first week, 112 mm of rainfall was received, however, bolls were protected from this by the plastic cover. No rainfall fell during Weeks 2 and 3 and 18.8 mm were received during Week 4. A Tiny Tag data logger recorded temperature and humidity under the plastic cover after the first week. Ambient temperatures ranged from 8.5 to 49.2°C over the 37 days and relative humidity ranged from 0 to 100% though temperature and humidity ranges under the plastic were 19.9-34.3°C and 30.3-70.2 %, respectively. Figure 5 shows daily averages for both parameters.

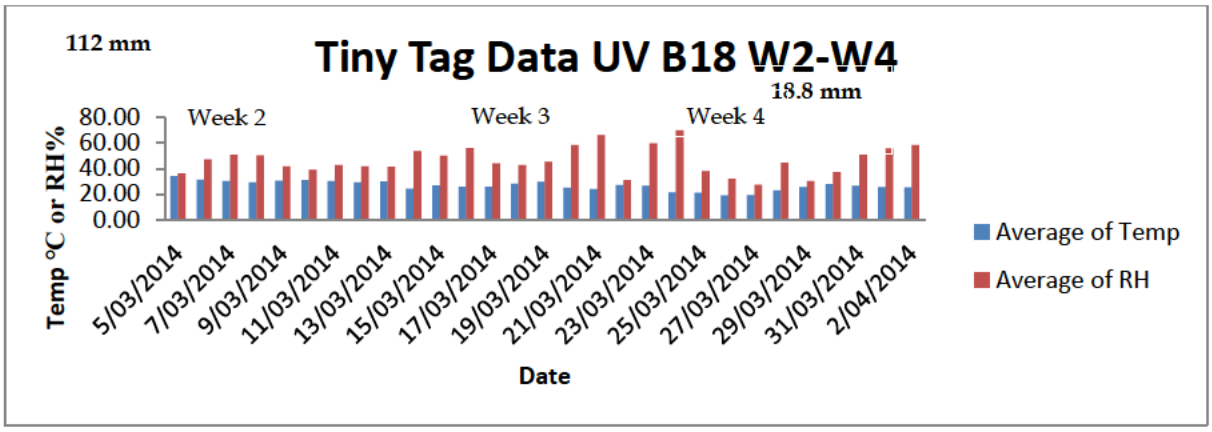


Figure 5: Average temperature and relative humidity under plastic cover UV B18 Experiment

Control bolls showed low levels of physiological sugars (glucose and fructose) indicating that the bolls were quite mature when picked. There were no significant changes in sugar levels between Week 0 and Week 1 hence bolls were not affected by rain or condensation moisture underneath the plastic cover (Fig. 6). Over the 4 weeks of exposure to UV radiation, there were no significant changes in sugar concentration for the control bolls. Any slight increases in concentration were possibly due to evaporation of moisture from the heat generated under the plastic cover.

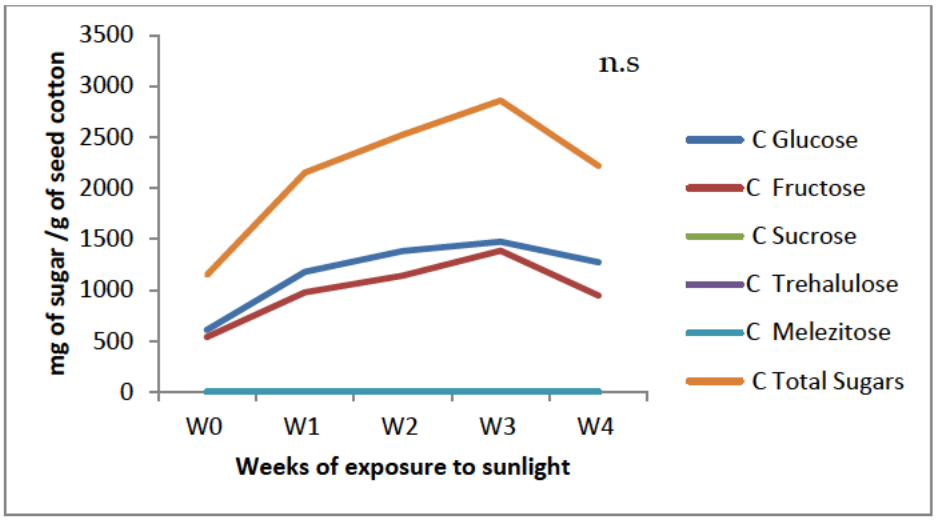


Figure 6: Sugar concentration of control bolls exposed to UV radiation (sunlight) for 4 weeks

In honeydew bolls, there was a significant decrease in the concentration of all sugars between Week 0 and Week 1 (Fig. 7). As control bolls were not affected by rain or moisture, it can be assumed that honeydew covered bolls were also not affected by these. Considering that the overall effect of UV exposure between Week 1 and Week 4 was relatively small in magnitude, honeydew during the 1st week must have been affected by other factors. It was noted that ants were crawling over the honeydew covered bolls presumably harvesting sugars just after bolls were pinned to the fence line (Fig. 9). This would explain the significant drop in sugar concentration during the first week of exposure. Again, any slight increases in concentration

between Week 1 and Week 2 were possibly due to evaporation of moisture from the heat generated under the plastic cover.

From Week 2 to Week 4, there was a significant drop in total sugar concentration, driven by significant drops in melezitose and sucrose. By Week 4, honeydew bolls were covered in sooty mould fungi (Fig. 9), which use sugars as substrate. In combination with adequate moisture (i.e. RH > 50%, Fig. 5) fungal growth would have been promoted and it is possible that sucrose and melezitose were the preferred substrate for sooty mould fungi. Again, any slight increases in concentration between Week 1 and Week 2 were possibly due to evaporation of moisture from the heat generated under the plastic cover.

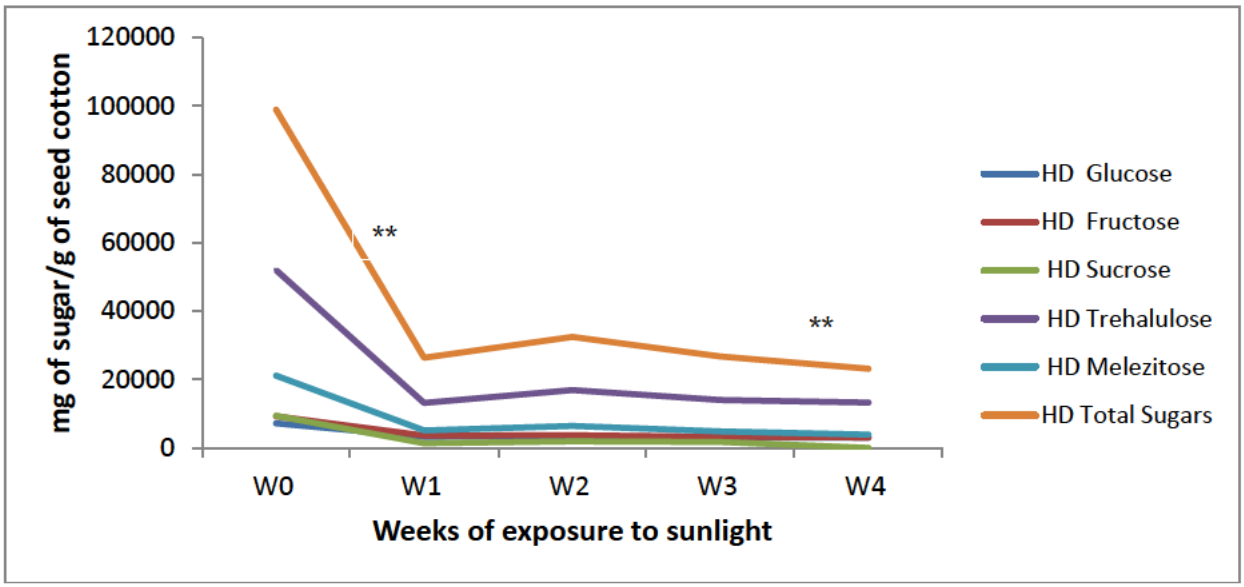


Figure 7: Sugar concentration of honeydew covered bolls exposed to UV radiation (sunlight) for 4 weeks

Figure 8: Ants harvesting sugar from honeydew covered bolls underneath plastic

Figure 9: Sooty mould fungal growth on honeydew covered bolls exposed to UV radiation for 4 weeks. Control bolls are free of sooty mould.

2013/14 UV Experiment 2: Artificial honeydew contaminated bolls were exposed to UV radiation for extended time periods by being pinned to a portable frame that could be brought under cover in case of rain. Control bolls for Week 0 to Week 2 (Fig. 10) showed very low levels of total sugars which comprised of the physiological sugars glucose and fructose, indicating that bolls were mature. Physiological sugars for Week 3 and Week 4 bolls were somewhat higher indicating that those bolls were slightly less mature when picked. The differences between total sugars were not significant.

For the honeydew bolls, total sugar levels for Weeks 1 to 4 were significantly different from the freezer Control bolls of Week 0 (Fig. 11). Week 0 bolls were half the amount of honeydew they were expected to be and this could not be explained by human error during honeydew application. We suspected that the freezer bags holding the bolls after spraying retained some of the honeydew on the plastic when the bolls were processed. Sugar levels between Weeks 1 and 3 did not change significantly, though during Week 4, total sugar levels fell significantly compared to levels in Week 3. However, these sugar levels were not significantly different from levels in Week 2.

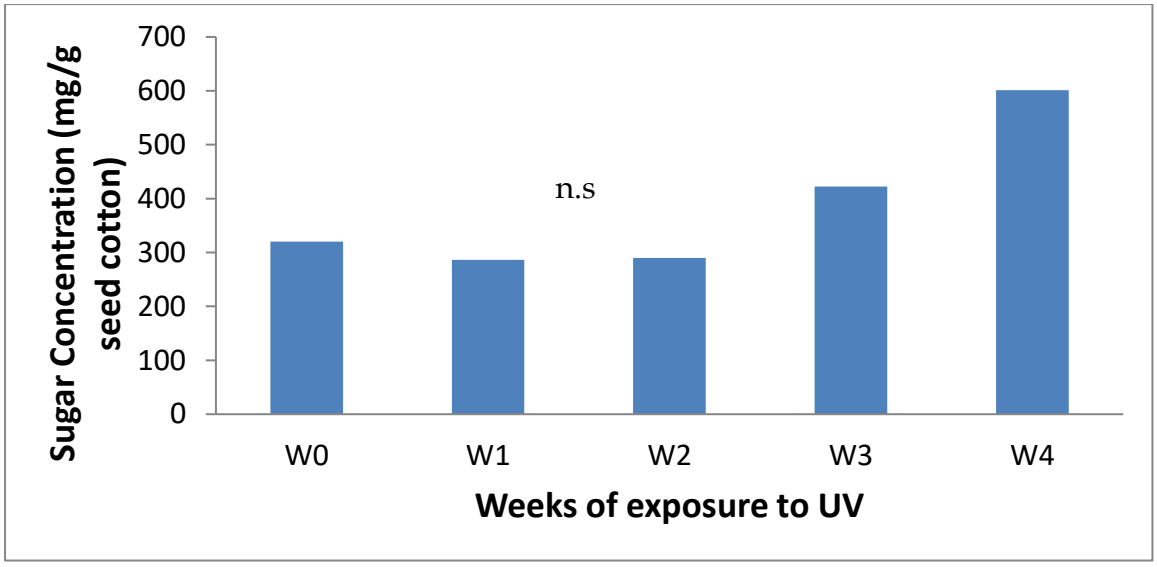


Figure 10: Sugar concentration (total sugars mg/g/S/C) on Control bolls for 4 weeks of exposure to UV radiation (sunlight). Bars with different letters are significantly different at p = 0.05 using ANOVA/LSD.

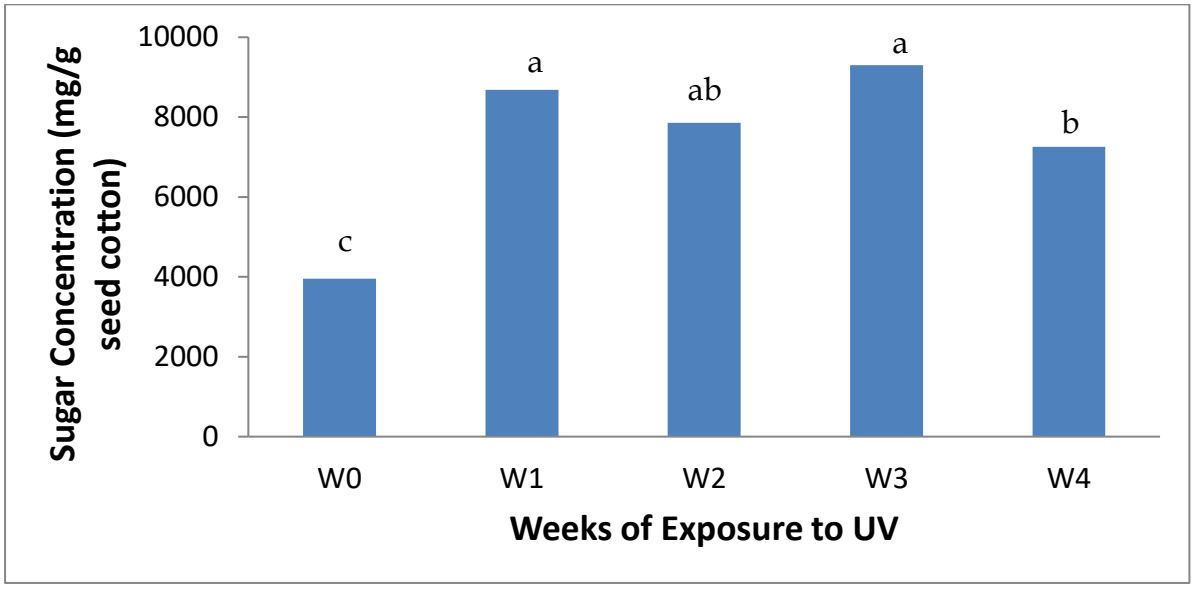


Figure 11: Sugar concentration (total sugars mg/g/S/C) on Honeydew covered bolls for 4 weeks of exposure to UV radiation (sunlight). Bars with different letters are significantly different at p = 0.05 using ANOVA/LSD.

2014-15 UV Experiment 3: This experiment was designed to confirm earlier findings about the contribution of sunlight on the breakdown of artificial honeydew on bolls. It was again confounded by control honeydew values. This time they were about 75% *higher* than the subsequently collected samples (last year they were significantly *lower*). We suspect that handling issues again played a role, possibly the sprayed control bolls not being handled as much as the bolls pinned on the wash stand and retaining more honeydew. ANOVA showed significance for Week, Dew, Honeydew and all their interactions (P=0.004, df (2, 47), LSD=2034.8). Exposure to night dew had a significant effect on honeydew bolls with a 22% reduction in total sugars (Fig. 12).

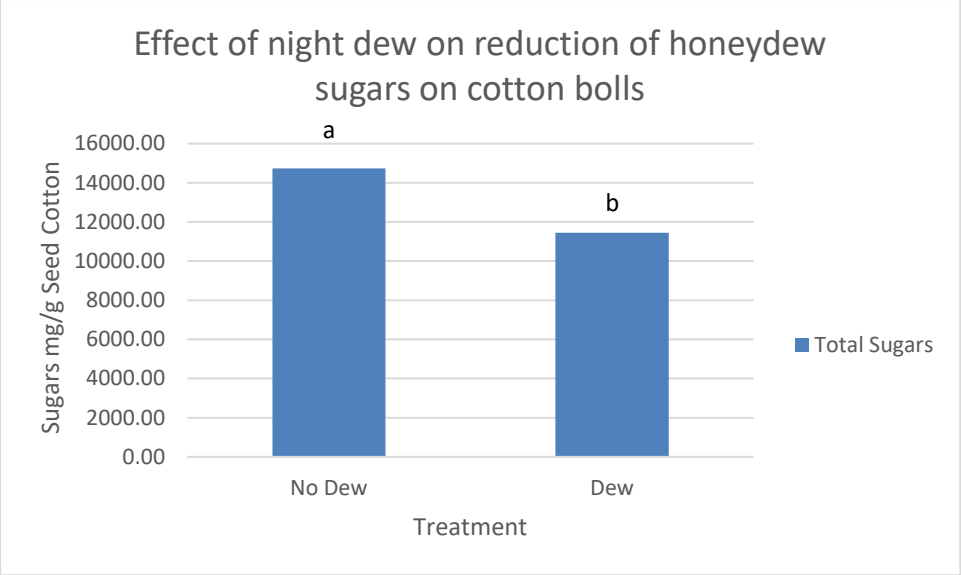


Figure 12: Sugar concentration (total sugars mg/g/S/C) on Honeydew covered bolls protected from or exposed to night dew and UV radiation (sunlight). Bars with different letters are significantly different at p = 0.05 using ANOVA/LSD.

Honeydew from samples collected after 1-week exposure was significantly lower than the control, irrespective of whether it was exposed to night dew (Figs 13&14). For samples not exposed to night dew, there was no difference between samples from Weeks 1, 3 and 5. For samples exposed to night dew, honeydew values for Week 3 were significantly higher than for Weeks 1 and 5. The nightly exposure to dew significantly reduced honeydew with the difference between the sheltered bolls and exposed bolls increasing over the 5 week period of the experiment (213, 1154 and 3748 mg HD per g seed cotton for Weeks 1, 3 and 5, respectively). Since the HD on controls was negligible (mean 246 mg HD per g seed cotton) the result is highly pertinent for the HD treated bolls. What is unusual is that the HD values for the No Dew samples increased significantly from week to week. The values are somewhat too high to attribute to dehydration of HD and therefore increased concentration alone.

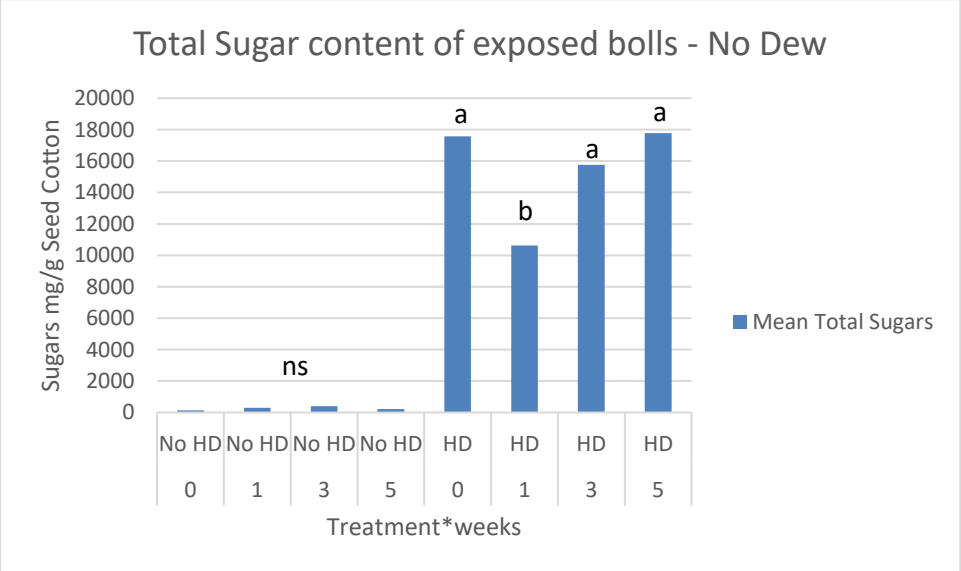


Figure 13: Sugar concentration (total sugars mg/g/S/C) on Control and Honeydew covered bolls protected from to night dew and exposed to UV radiation (sunlight). Bars with different letters are significantly different at p = 0.05 using ANOVA/LSD.

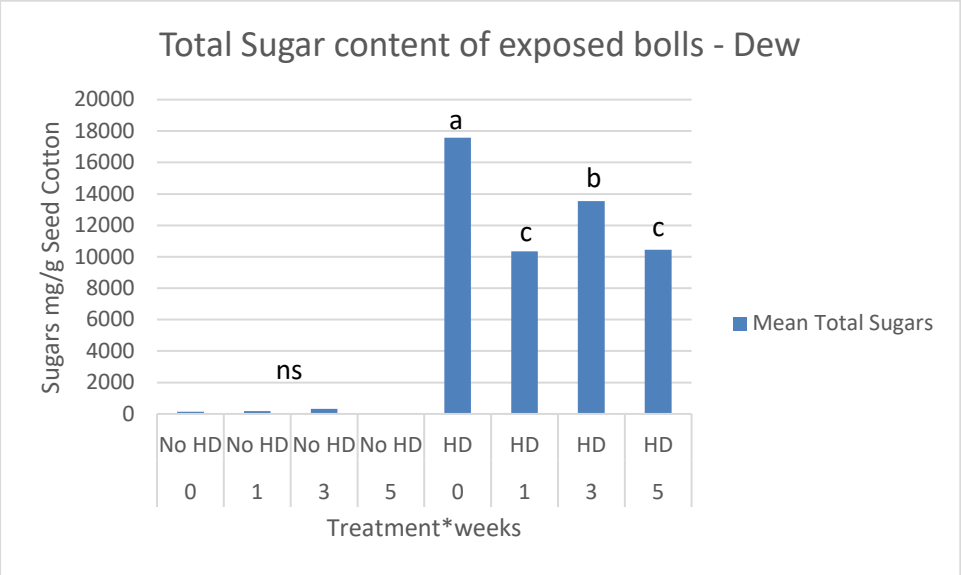


Figure 14: Sugar concentration (total sugars mg/g/S/C) on Honeydew covered bolls exposed to night dew and UV radiation (sunlight). Bars with different letters are significantly different at p = 0.05 using ANOVA/LSD.

Discussion

It is unlikely that extended exposure to UV radiation in the form of sunlight would have had any significant effects on the sugar concentration and stickiness of cotton bolls contaminated with artificial honeydew in Experiment 1. Two agents removing sugars from bolls were ants and sooty mould fungi, both well known for their preference for sugars as a carbohydrate source. In a practical sense, neither agent would be of any use to growers since ants also protect and promote the organisms producing honeydew – aphids and whiteflies. Sooty mould fungi may remove smaller amounts of sugar on bolls, however, they also produce black spores that discolour cotton lint and incur penalties. Confirmation of the ineffectiveness of UV radiation in the absence of ants and sooty mould could be achieved with a more compact follow up experiment, given below.

The compact design of the Experiment 2 allowed for the exclusion of ants and sooty mould as factors affecting honeydew. However, the reduction in replications (5 compared to the usual 10) has also reduced the reliability of the data which is evident in the fact that sugar levels for Week 4 were reduced significantly from those in Week 3 but not from those in Weeks 1 and/or 2. Hence, it could be expected that sugar levels did not change over 4 weeks of exposure, but that the higher sugar levels in Week 3 reflect variability in application of honeydew instead. To validate the indication that UV radiation does not affect honeydew levels, we repeated the experiment in similar format but again with 10 replications and an improved accuracy or quantity of honeydew application to bolls.

In Experiment 3 we considered reasons why the control HD bolls (from the freezer) would contain so much more honeydew than exposed HD bolls and believe that handling may play a role. Honeydew covered bolls are handled about 5 times between spraying HD on and washing the bolls compared to handling control bolls only twice at spraying and washing. Handling

experiments have shown that up to 25% of honeydew can be taken off a sprayed boll by a single handling event. Considering that the bolls in this experiment were handled up to 5 times, it is quite possible that the 23-40% losses seen in the No Dew bolls were due to repeated handling.

c. Mouldy Bolls

We were contacted by a grower, about the presence of mouldy bolls in his crop. We contacted his consultantand visited a typical field where the 'mouldy' bolls were present. This was a crop that had been planted early but had grown very tall. Bolls near the bottom of the plant were a grey/brown tinge and loads of brown/black spores puffed out of them when handled. Bolls in the upper canopy were brilliant white, though a small number did show signs of light recent honeydew contamination and very slight presence of sooty moulds (Fig. 15).

Methods

We collected bolls from the upper and lower canopy, photographed them and provided a sub-sample to Dr Stephen Allen to test for contamination with micro-organisms. We also noticed a picker in an adjacent field that was covered in a black 'dust'so we took wipe samples from the picker's exterior panels and provided them to Dr Allen.

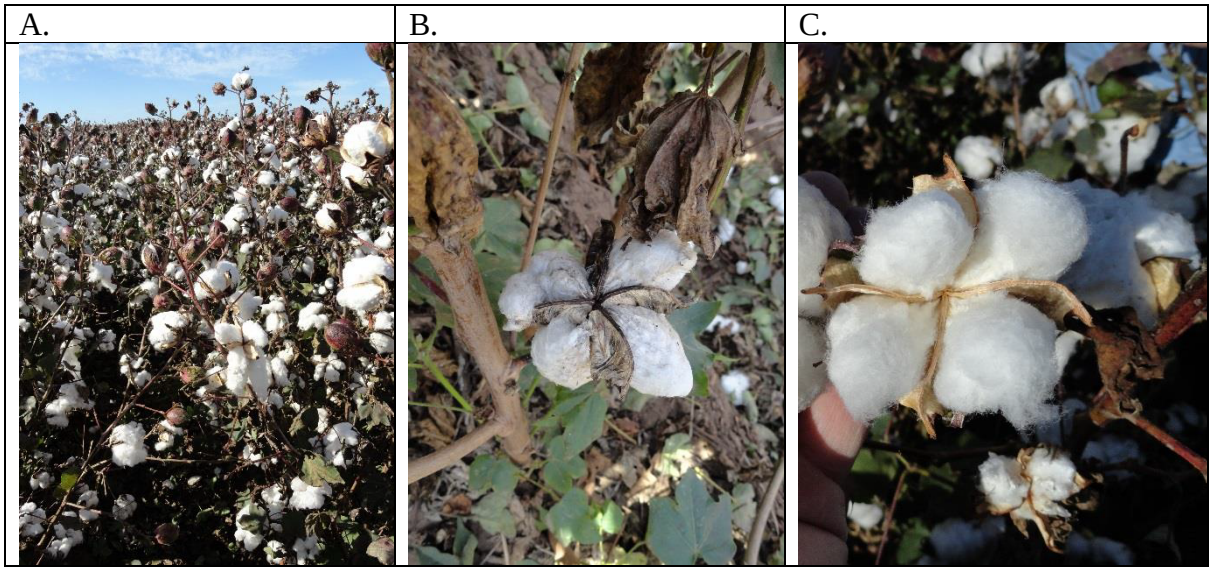


Figure 15: A. Tall crop with clean upper bolls B. lower bolls showing dull brown/grey appearance C. upper bolls showing brilliant white.

Results & Discussion

There was evidence of a very slight amount of sooty mould on some of the younger bolls near the top of the canopy – but this would not explain concerns about mouldy cotton. Dr Allen confirmed the presence of huge numbers of *Alternaria* spores in the lower bolls and from the picker. Regarding the history of the crop, the lower bolls had opened up relatively early and experienced about 80mm of rainfall. Conditions had then remained dry for the remainder of the boll opening. It is possible that the tall, dense canopy meant that lower bolls stayed damp for a long period – making them an ideal host for *Alternaria*. Bolls that were not open during the rainfall period escaped this problem – both because they were not open when it rained and because their higher position in the canopy would have exposed them to drying conditions.

Hence the 'mouldy bolls' may be due to a combination of boll opening time, crop height and density and rainfall. Dr Allen provided this telling comment "Some years ago I spent some time talking to pathologists in Israel (They were experts on *Alternaria*!). In one of their production areas they commonly had tall crops and as the crops matured they would typically get heavy dews. The combination of open bolls in a tall canopy with regular dews caused the development of significant quality problems. They found it necessary to 'bottom pick' the crops. If they waited for the top bolls to open then the fibre in the open bolls well down in the canopy deteriorated!"

d. Puff by wash

Previous data from spraying 1-6 puffs of artificial honeydew/boll were unclear but the wash experiment gave us a clear picture of how much honeydew we washed out in three consecutive washes at an application rate of 5 puffs of honeydew per boll (Table 1).

Table 1: Proportions of total sugars washed out of cotton bolls in three consecutive washes, 2012/13 (5 puffs of honeydew/boll)

Treatment	**% HD washed out**		
	No HD	Art. HD	Real HD
Wash 1	59.25	86.02	85.92
Wash 2	28.70	11.28	11.02
Wash 3	12.04	2.71	3.06
Total	100	100	100

Methods

Here we combined the experiments and applied 0, 1, 3 and 5 puffs of honeydew per boll and washed each boll 3 times. We used a washing methodology that used a water volume four times the weight of the seed cotton to wash off honeydew. We used stored bolls and applied artificial honeydew in 5 replications for each treatment combination (Puffs x Washes x Reps = 4 x 3 x 5 = 60 samples). Each boll was washed in warm (not above 36 °C) deionised water 4 times the weight of the seed cotton weight (e.g. S/C Wt = 25.63 g, Water = 102.52 g (ml) total sample weight with water = 128.15 g). Wash samples were agitated, then soaked for 15 minutes, agitated again, seed cotton was squeezed and a subsample was taken from the wash water and frozen for analysis. The wet seed cotton. Was re-weighed and this weight was subtracted from the total sample weight (128.15 g). The difference was the amount of water that needed to be added for the second wash (e.g. Wet seed cotton weight = 82.14 g, 128.15 g – 82.14 g = 46.01 g (ml) of water to be added). The same procedure for washing was used and a second subsample was taken. This procedure was repeated for the third wash (subtracted from the original total sample weight -128.15 g). All samples were frozen, and the squeezed seed cotton was placed into a paper bag and then in the dehydrator for dry weight determination.

Results

There was an indication that the more honeydew applied to bolls, the more honeydew was washed out in the first wash (Fig. 16). This trend could be seen for most sugars except sucrose which degrades rapidly into glucose and fructose in the presence of enzymes or microorganisms (Tables 2 & 3). At 1 and 3 puffs of HD per boll, the first wash generally yielded less sugar and the 2nd and 3rd wash gave roughly equal quantities. The Control bolls (0 puffs) had small amounts of glucose and fructose indicating physiological maturity, but also traces of melezitose and trehalulose, sugars typically found in aphid and whitefly honeydew. The field

that these bolls were collected from had a low level of infestation of these pests at the time of picking. The percentages of total sugars washed out in consecutive washes at 5 puffs per boll are very similar to the results from 2012/13 (Table 1) with around 80 % of total sugars washed out in two washes. Where sugar concentrations were higher (5 puffs/boll), approximately 80 % of total sugars were washed out in the first wash.

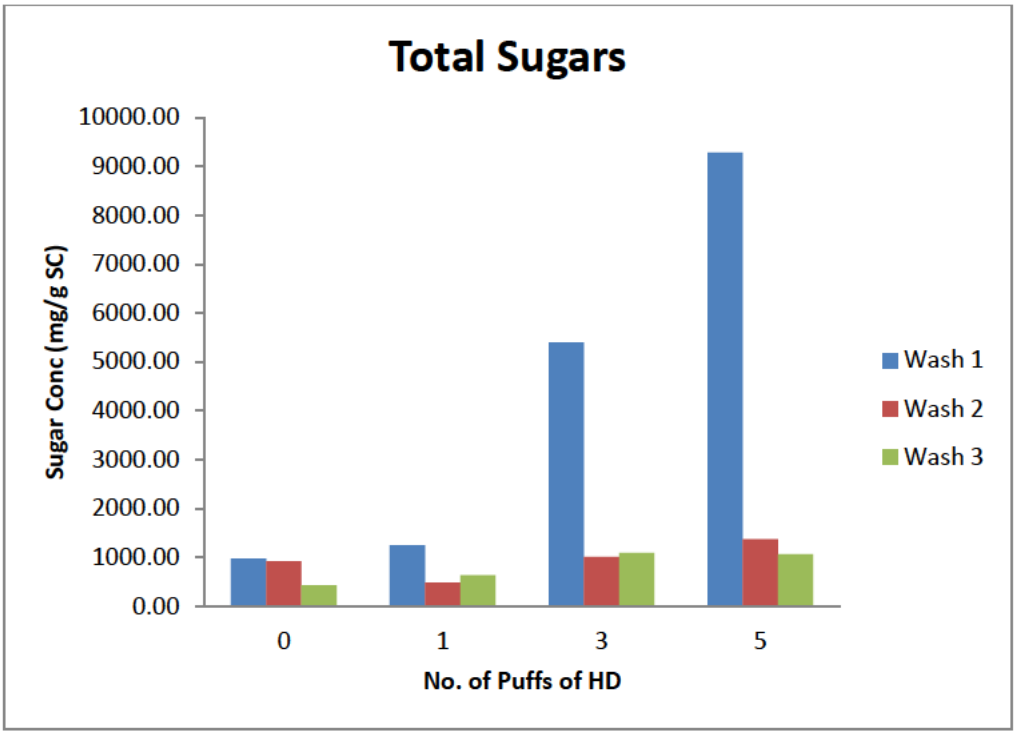


Figure 16: Total sugar concentration Puff x Wash Experiment 2013/14

Table 2: Percentages of different types of sugar washed out in three separate washes when applied to bolls at increasing amounts, ACRI 2013/14

%	Wash 1	Wash 2	Wash 3	Total Washed out
Puffs	**Total Sugars**			
0	41.90	39.51	18.59	100.00
1	52.48	20.65	26.87	100.00
3	71.95	13.43	14.63	100.00
5	79.26	11.69	9.06	100.00
	Glucose			
0	43.29	39.27	17.44	100.00
1	36.21	24.60	39.20	100.00
3	61.55	16.33	22.11	100.00
5	71.27	14.65	14.08	100.00
	Fructose			
0	40.83	40.33	18.84	100.00
1	40.66	22.96	36.38	100.00
3	66.45	15.43	18.13	100.00

Table 3: Percentages of different types of sugar washed out in three separate washes when applied to bolls at increasing amounts, ACRI 2013/14

%	Wash 1	Wash 2	Wash 3	Total Washed out
Puffs	**Sucrose**			
0	0.00	0.00	0.00	0.00
1	100.00	0.00	0.00	100.00
3	98.70	1.30	0.00	100.00
5	99.41	0.59	0.00	100.00
	Trehalulose			
0	11.59	45.98	42.43	100.00
1	75.28	16.25	8.47	100.00
3	77.97	10.81	11.23	100.00
5	81.02	11.47	7.51	100.00
	Melezitose			
0	40.52	35.84	23.64	100.00
1	62.38	16.48	21.14	100.00
3	73.93	16.74	9.32	100.00

Discussion

The value of this experiment was in giving us information about the optimum quantities of honeydew that we needed to apply in order to get the best results with the extraction technique that we were using. From this year's results we decided to return to applying 5 puffs of honeydew per boll to wash out a maximum amount of honeydew in the first wash since we routinely only washed samples once. This brought the scale of sugar extracted up and comparative values for the controls were virtually invisible on the same graph. The reason we initially reduced the amount of honeydew applied to two puffs per boll was to reduce the magnitude of the graph scale so that we could plot controls versus HD in a more meaningful way (and also to save on honeydew as cost of melezitose is $8.80/g). However, our data this year were much more variable – either the result of less accuracy on our part due to less honeydew applied or using a new lab for analysis, or a combination of both.

e. Within boll distribution of sugar

Methods

To assess the areas of the boll that may be contaminated with honeydew we separated lint from the outside of the boll (OUT) and the inner lint (IN) by cutting a thin layer of lint from the boll with a fine pair of snips (Fig. 17). We separated lint from control bolls (Cont), bolls with natural honeydew (NHD) obtained from glasshouse colonies of SLW on cotton and sprayed on artificial honeydew (AHD) (Fig. 18). To better see the sprayed-on honeydew we dyed it with blue food colour. Once separated, lint samples were washed individually and analysed by HPLC.

Figure 17: Preparing to separate sprayed on honeydew from the boll

Figure 18: Artificial honeydew layer (blue) and internal lint for sugar assessment

Results & Discussion

Results are shown clearly in Figure 19: no sugars were found on mature, uncontaminated control bolls and the low concentration of natural honeydew did not penetrate beyond the outer surface of the lint while artificial honeydew which was sprayed on in high concentration (5 puffs/boll) penetrated well into the subsurface of the outer lint (up to 5 mg/ml total sugars). Cotton lint is hydrophobic, hence the light droplets of the natural honeydew are unlikely to sink into the lint while the heavy concentrated and viscous droplets of the artificial honeydew are forced into the lint. It is therefore most probable that natural honeydew can be washed off nearly completely in a good shower of rain, but also, that the clean inner lint could dilute a light layer.

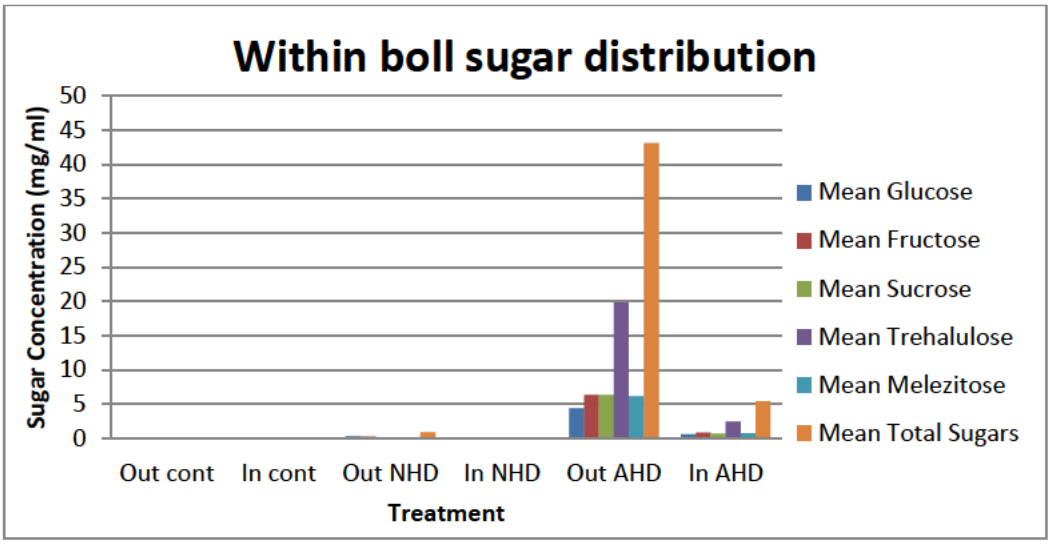


Figure 19: Within boll sugar distribution of control, natural honeydew and artificial honeydew bolls

f. Mealybug honeydew

Five samples of mealybug honeydew were supplied by Dr. Moazzem Khan from glasshouse colonies at QDAF, Toowoomba. To gain an understanding of the different sugars and their proportions in this type of honeydew, we included these samples in our HPLC testing.

Methods

Honeydew contaminated bolls were washed and processed in the manner described for boll washes above and frozen samples were sent to Donna Glassop.

Results & Discussion

The standout in the mealy bug honeydew is the high percentage of melezitose and the low percentage of trehalulose, which is so typical of whitefly honeydew (Table 4). Aphid honeydew also has a high proportion of melezitose but generally not as high as the mealy bug honeydew. It is likely that the glossy, stickiness of the mealy bug honeydew is due to the melezitose (which can be hydrolysed to glucose and sucrose isomers, i.e. turanose). In contrast, trehalulose is very stable and does not degrade readily into sucrose/glucose/fructose which may be why that honeydew is less glossy and sticky (it appears matte rather than glossy), especially when dry.

Sample no. 3 is quite different (Fig. 20). I cannot remember that there was anything special about it, whether it came from an older plant, whether there were more nymphs/adults on that particular leaf/plant or if it had degraded in warm/humid conditions but there is naturally much variability in the honeydew composition depending on the nutritional & water status of the plant, plant age, insect stage, etc.

Table 4: Mean mealy bug honeydew composition

	Glucose	Fructose	Sucrose	Trehalulose	Melezitose
Mean	3.53	15.30	23.66	0.62	56.88
without Sample 3	3.04	10.30	15.76	0.47	70.43

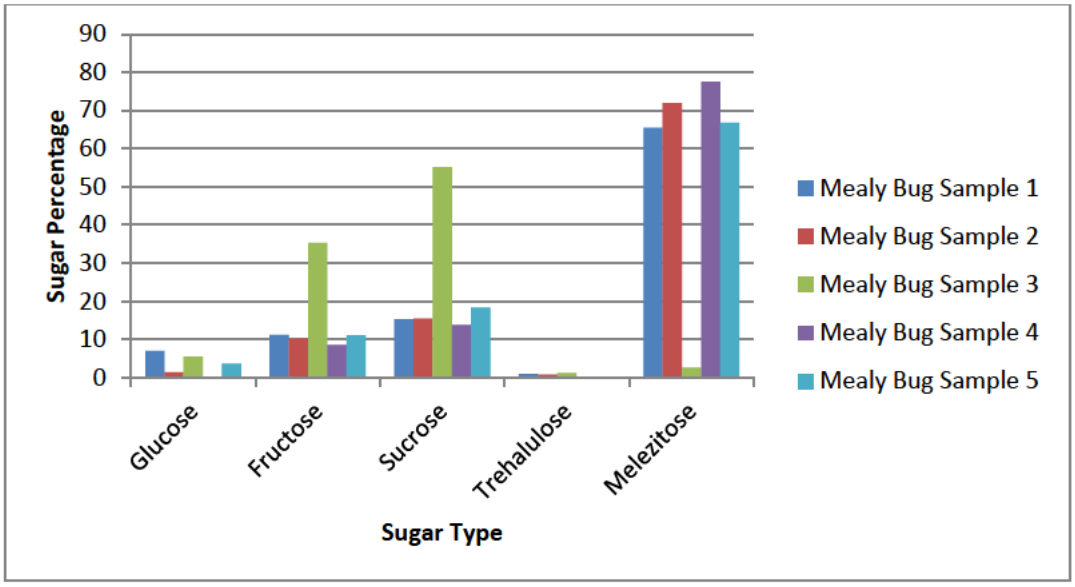


Figure 20: Composition of mealy bug honeydew, Toowoomba glasshouse, 2013

g. Yeast experiments

We have shown that rainfall will wash off honeydew but often with the risk of sooty mould formation on residual honeydew. Sooty mould fungi metabolise sugars but in this process they also produce cellulases and pectinases that weaken the cotton fibre. Their black fungi also add to the contamination of lint and a grey appearance may incur penalties. Rainfall may also discolour and weaken cotton fibres, especially if moist conditions prevail and other fungi, e.g. *Alternaria* spp., invade the lumen and the outside of the lint. We have been trying to find a fungus that – ideally – metabolises sugars, does not produce enzymes that attack the constituents of cotton fibres and produces only white spores. Our background searches have not been able to identify such an organism amongst the sugar fungi.

A different type of fungi, yeasts may be an alternative. The budding yeasts ("true yeasts") are classified in the Ascomycota, order Saccharomycetales, and are chemoorganotrophs, that use organic compounds as a source of energy and do not require sunlight to grow. Carbon is obtained mostly from hexose sugars, such as glucose and fructose, or disaccharides such as sucrose and maltose. Some species can metabolize pentose sugars such as ribose, alcohols, and organic acids. Honeydew consists of glucose, fructose and sucrose (which will split into the former) as well as melezitose and trehalulose. When exposed to high temperatures (200°C), melezitose degrades into mostly glucose and a small amount of turanose with about 10% of melezitose remaining. Trehalulose degrades into a small amount of glucose and non-

carbohydrate products, a reaction enhanced by catalysts. Hence, yeasts should be able to metabolise at least some sugars in honeydew.
To investigate the ability of common baker's yeast (*S. cerevisiae*) to metabolise all the sugars commonly found in insect honeydew we set up a preliminary experiment. All sugars were made into separate solutions (glucose, fructose, sucrose, melezitose and trehalulose) with water as the control.

Methods

We allowed for 4 treatment reactions: R1 = sugar solution only, no yeast (Control), R2 = Sugar + Yeast 10 min reaction, R3 = Sugar + Yeast 20 min reaction, R4 = Sugar + Yeast 50 min reaction (16 samples). The experiment was carried out using falcon tubes in a hot water bath at 37°C. 3 g of each sugar was dissolved in 30 ml of water and 1 g of baker's yeast was added to each sample. Each tube was shaken vigorously and allowed to froth for the determined time intervals. At 20 minutes R 4 was shaken again to remix the solution, then again at 30 and 40 minutes, assuming a reaction end point at 50 minutes. The degree of frothing for each sugar at each time was assessed as a sign of activity and scored (R = reaction, take photos). Froth Score: 0 = no froth, 1 = signs of froth, 2 = some froth, 3 = good froth, 4 significant froth. After scoring, each sample was immersed each sample into boiling water and heat to 62°C for 3 minutes to kill the yeast. Samples were poured through a filter funnel to separate the yeast, then pushed through a syringe nylon filter and divided into a falcon tube for sugar testing. (This however, did not occur due to the samples spoiling during the QBP power outage).

Results & Discussion

Fructose was rapidly metabolised by baker's yeast (Fig. 21), followed by glucose and sucrose (Table 5). The yeast metabolised trehalulose much slower than these, and never as vigorously as fructose or glucose. Melezitose was either not utilised by the yeast, or the reaction is so slow that it may take hours of incubation (Fig. 22). Considering that whitefly honeydew contains only a small amount of melezitose, yeast could potentially metabolise most of the sugars in that type of honeydew. The proportion of melezitose in aphid honeydew is usually less than 15% (except for Hendrix *et al.* (1992) – 38.3% melezitose) and the remainder consists of glucose, fructose and sucrose and a very small amount of trehalulose, all of which can be used by baker's yeast, hence most of the sugars in the honeydew could be used.

Table 5: Froth scores of individual sugars metabolised by baker's yeast.

Sugar	R1 (Control)	R2 (10 Min)	R3 (20 min)	R4 (50 min)
None	0	0	0	0
Glucose	0	3	4	2-3
Fructose	0	4	2-3	2-3
Sucrose	0	3	2-3	2-3
Melezitose	0	0-1	0-1	0-1
Trehalulose	0	2	2-3	2-3

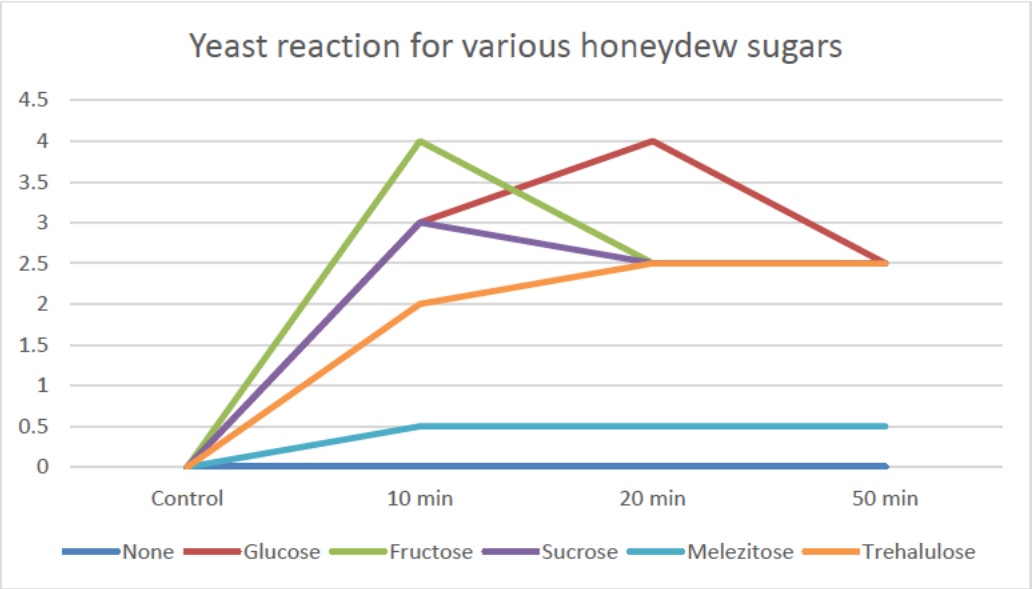
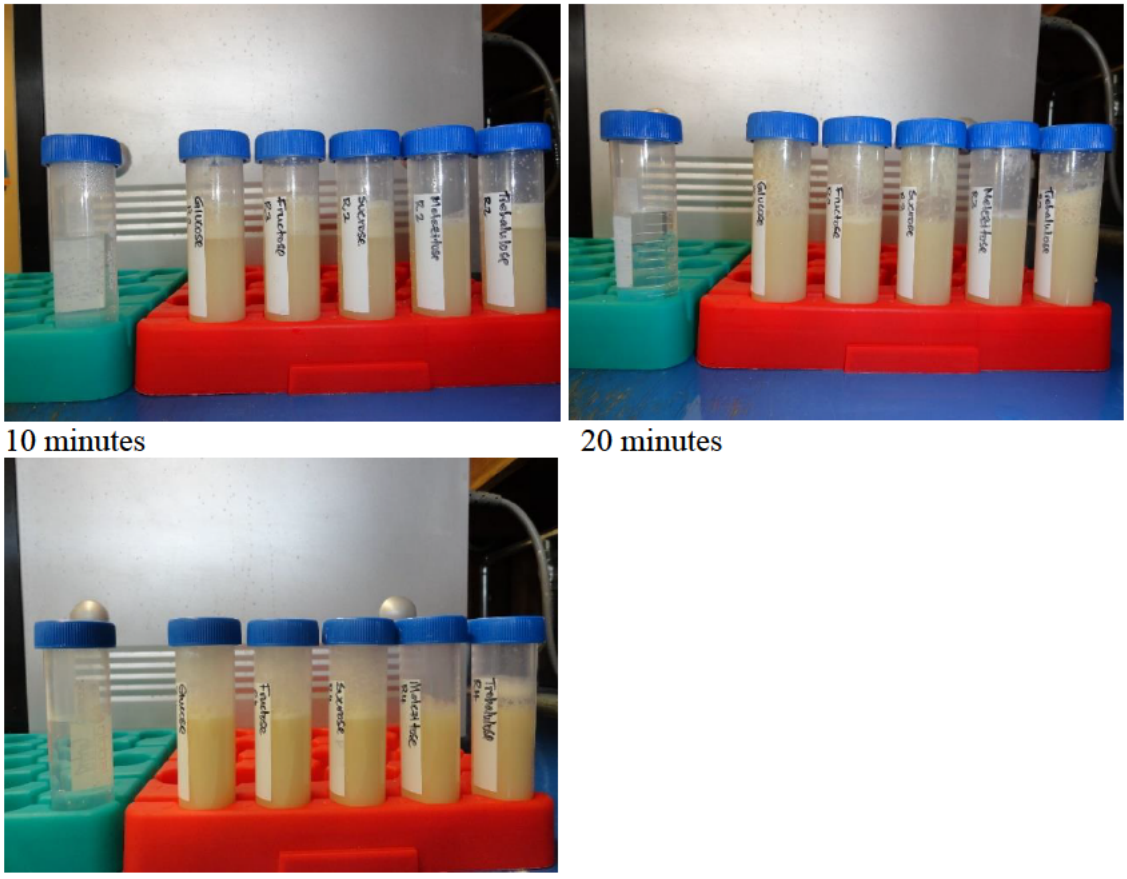


Figure 21: Froth score reactions of various honeydew sugars



10 minutes

20 minutes

50 minutes

Figure 22: Yeast reaction on various sugars (water Control on left)

- **Sooty Mould**

We have had considerable discussions with consultants and growers about the potential relationship between the presence of sooty moulds on bolls in crops and reductions in grades

for that crop when harvested. They believe that this occurs in fields where there have been significant SLW populations present after bolls started to open. These SLW have fed and produced honeydew, some of which has been deposited on open bolls. In some fields obvious sooty mould growth has occurred on these bolls. It is thought that these spores get mixed in with the lint during harvest and ginning resulting in penalties for colour. These penalties can be up to $50 per bale, or $500 per ha at 10 b/ha. However there have been instances of fields with sooty mould contaminated bolls incurring no penalties.

The link between sooty mould and grades is unproven and in most cases the presence of honeydew in the period between the start of bolls opening and harvest co-incides with rainfall during the same period. Rainfall alone can cause problems with grades, with the severity probably influenced by many factors including rainfall amount and duration, temperature and humidity and spore source.

To begin to tease this issue apart, we aimed to create treatments that included clean cotton, cotton contaminated with honeydew but no sooty mould and cotton with honeydew and sooty mould and have these ginned and graded. We also worked with consultants to identify situations where we could opportunistically collect honeydew contaminated cotton before and/or after rainfall events. CRDC (Susan Mass and Allan Williams) approached us to undertake research to understand factors affecting colour in cotton in collaboration with the CSIRO team at Geelong. We developed project ideas which led to the related Cotton Colour project CSP1703.

a. Degradation of honeydew by sooty moulds

Weathering Experiment 2015/16 - A pilot experiment was set up using bolls collected from the previous season and stored through winter. The bolls from this collection were either treated with artificial honeydew (+HD) or left untreated (-HD). We also set up small plastic glasshouses in the field to provide protection against rainfall. This allowed us to have non-rainfall (-RF) or natural rainfall (+RF) treatments. Combined with the honeydew treatments we had all four combinations (Fig. 23). The tents also enabled us to have a rain-free environment for the cotton colour work (CSP1703). As autumn was unusually wet with 200 mm of rainfall between March and July, using the tents was a good strategy to keep open bolls dry.

The purpose of the experiment was to test the effects of rainfall on levels of honeydew contamination (+HD-RF compared with +HD+RF) and to evaluate if sooty mould would develop in the absence of rainfall. We also wanted to test if this method would be useful for assessing the effects of rainfall on grade (e.g. comparing –HD-RF with –HD+RF) and to see if this was affected by the amount of rainfall. This last question was addressed by replicating the entire experiment and collecting bolls after a small sharp rainfall event (about 6mm) and after several events (totalling about 32mm). All the bolls were collected and processed, and samples taken to Linda Smith (QDAFF) to identify and quantify fungal contamination and to Donna Glassop at CSIRO Queensland Biosciences Precinct (QBP), St. Lucia, to quantify honeydew sugar levels. After ginning, remaining samples were graded by Andrew Baxter at Australian Cotton Classing Services in Wee Waa.

The experiment was set up in 11 replications of 10 cotton bolls. Each boll was individually identified and pinned into the mid-canopy on the 16/02/16. A total of 1080 bolls was used in the experiment (Table 6). Three reps were washed for sugar analysis by Donna Glassop,

CSIRO Brisbane, one rep was sent to Linda Smith QDPI, Brisbane, to analyse sooty moulds (2) and the remaining reps were pooled with the sugar analysis reps for ginning.

Table 6: Summary of treatments and intended effects: Weathering Experiment, ACRI F1, 2015/16

	- HD	+HD
Reference in freezer (-Rain)	-HD Control No sugar, no SM	+HD Control Sugar, no SM
-Rain (Controls)	Ultimate Control for Rain 1 – expect no SM Ultimate Control for Rain 2 – expect no SM	Dry HD effect for Rain 1 – Some SM development? Dry HD effect for Rain 2 – some SM development?
+Rain 1	Rain without HD – negligible SM effect?	Rain+ HD – will sugar be washed off therefore no SM?
+Rain 2	More rain without HD – possibly other MO effects - Alternaria?	Rain+ HD – will residual sugars cause SM & possibly other MO effect?
Total Bolls	1080 bolls	

The experiment was left in the field to be exposed to rainfall. The first rainfall events occurred mid-March and the first lot of bolls were collected after receiving 10.4 mm of rainfall. By mid-April two further rainfall events (2 x 5 mm and 22.4 mm) brought the total rainfall received to 44.8 mm and the second lot of bolls was harvested. Each boll in each replication was scored for honeydew presence based on a rating scale developed from variously contaminated bolls. Mean scores for each treatment were compared to sugar analyses. We were fortunate that these samples were not affected by the power outage at QBP.

Weathering Experiment 2016/17 – This experiment was set up with a dual purpose to firstly provide a humid environment for sooty mould production on honeydew covered bolls and secondly, to have rainfall-free control bolls for the cotton colour work (CSP1703). This was achieved by erecting plastic greenhouses in areas where whiteflies were either controlled or where the population was allowed to develop. This provided +/- rainfall (RF) and +/- honeydew (HD) treatments. Between the 13/03/17 and the 19/03/17 bolls received 106.4 mm of rainfall termed RF1 after which they were collected. During April, short showers (1.6-5.2 mm), which were termed RF2, occurred on 3 days totalling 10.4 mm and the second set of bolls was collected. Bolls were photographed and scored for sooty mould contamination based on a scale developed during the pilot experiment in 2015/16.

Results

Weathering Experiment 2015/16 - +HD bolls were severely contaminated (> 90 mg of total sugars/g of seed cotton) with artificicial honeydew sugars after they had been sprayed (Fig. 23, Control +HD treatment). Control bolls –HD contained minute amounts of natural sugars which indicated fibre maturity. In the 22 days between Rainfall event 1 and Rainfall event 2, sugar concentrations on all samples decreased between 40-60 %, except on the +RF – HD samples. At the same time, honeydew scores increased in the range of 185-633 %. There was no obvious relationship between these value changes though Figure 24 clearly shows that Honeydew contaminated bolls and bolls not exposed to rainfall had higher sooty mould scores. Figures 25 and 26 show the range of sooty mould contamination in each treatment. Samples sent to Linda Smith at QDAF have not been analysed for mould spore identification and quantification due to a staff shortage that made it impossible to take on external work.

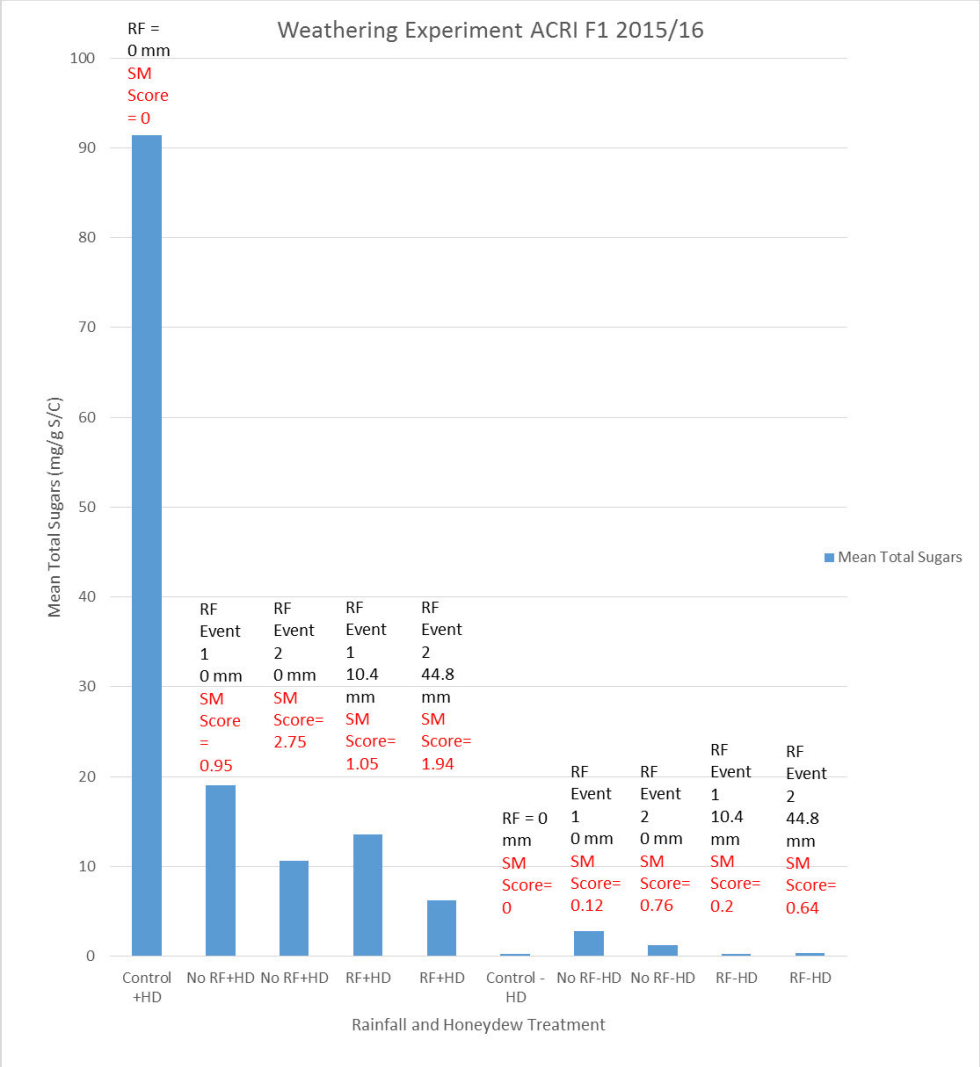


Figure 23: Reduction in honeydew sugars of contaminated lint exposed to rainfall and sooty mould activity. Weathering Experiment, ACRI F1, 2015/16

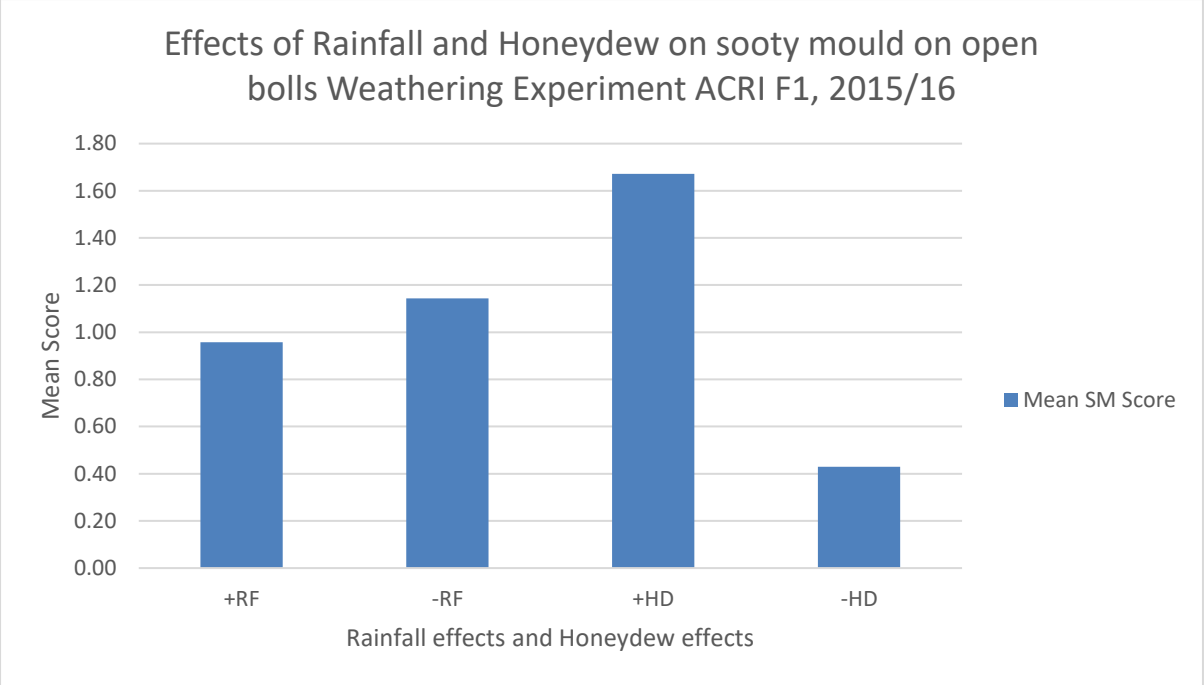


Figure 24: Effects of rainfall and honeydew on the sooty mould score of open bolls

Figure 25: Sooty mould contaminated bolls from different treatments – Rainfall 1= 10.4 mm
Treatments are from top to bottom: -RF-HD, +RF-HD, -RF+HD, +RF+HD

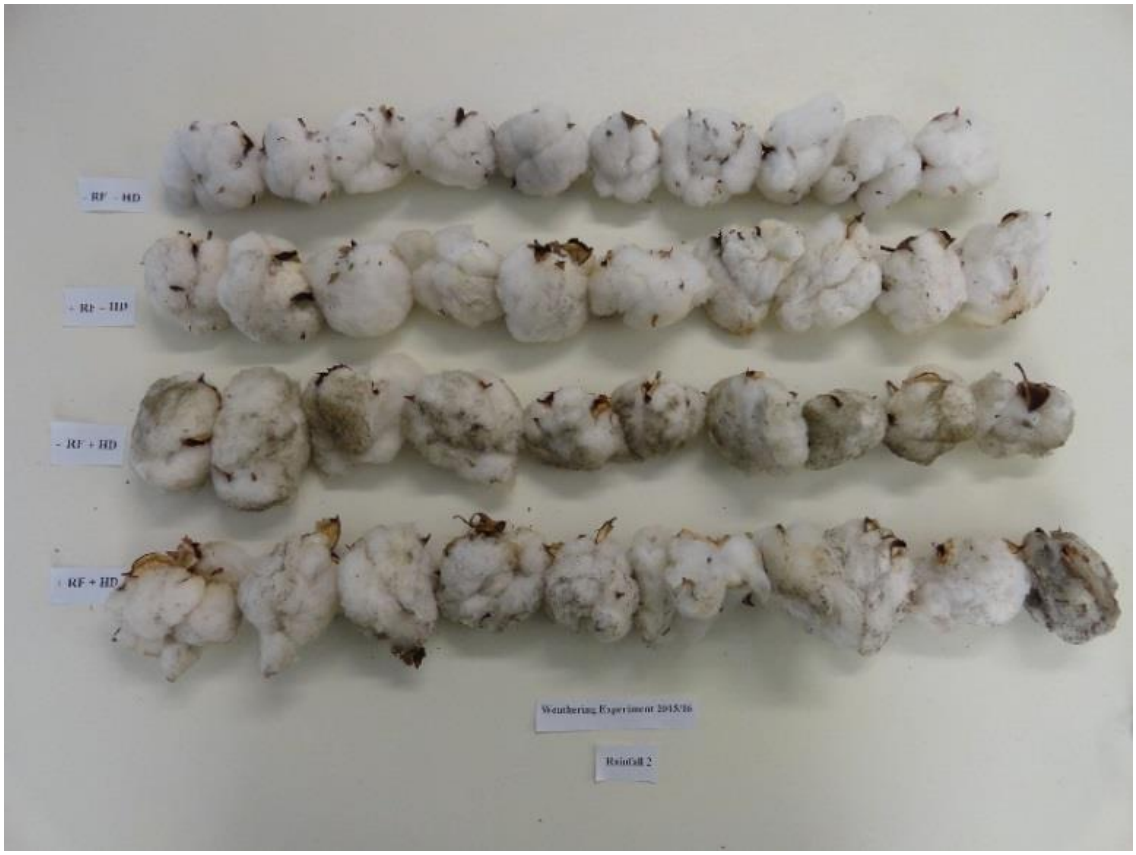
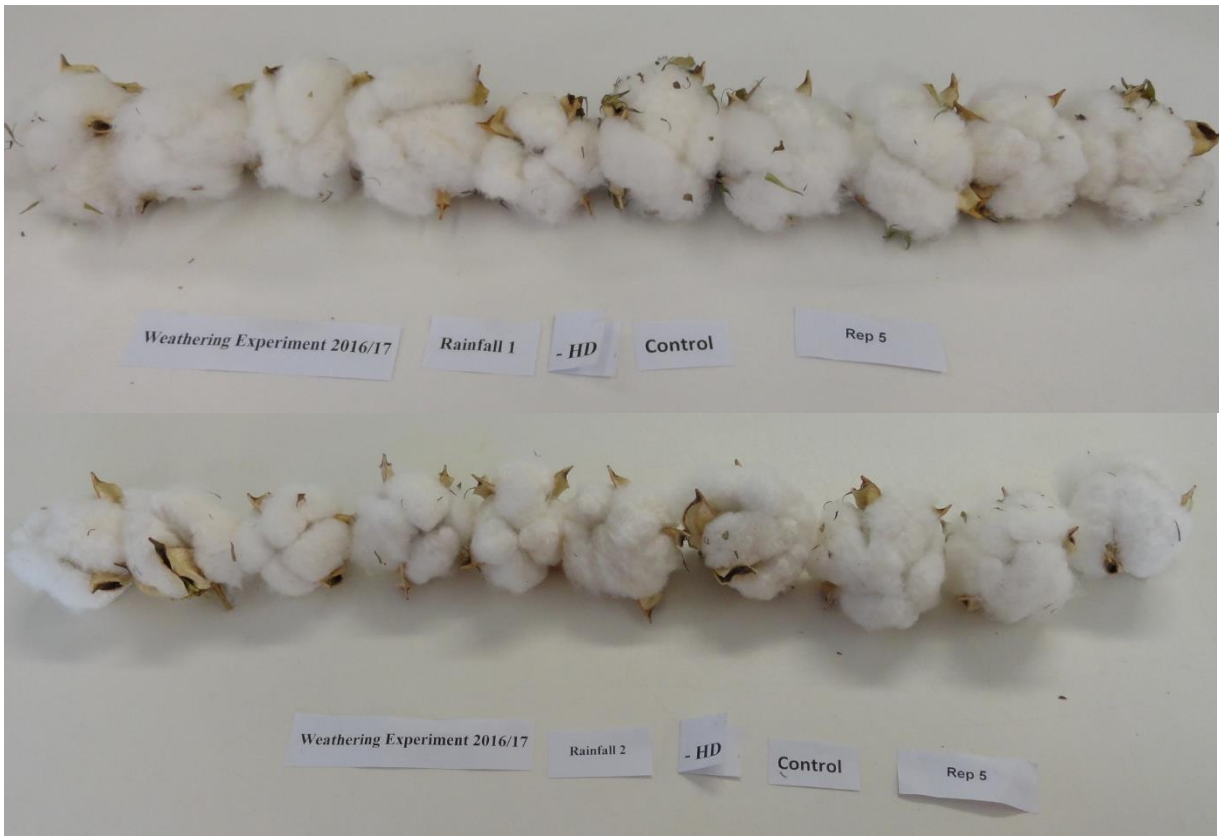


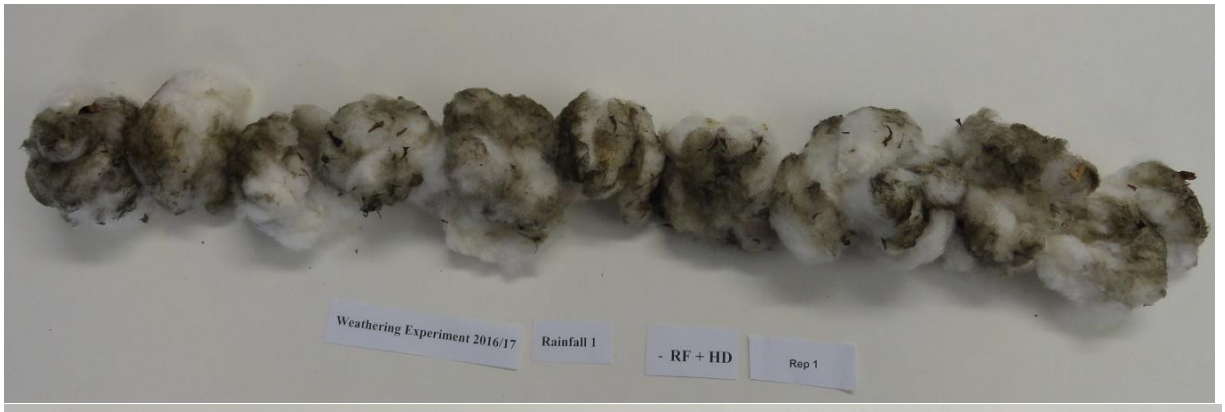
Figure 26: Sooty mould contaminated bolls from different treatments – Rainfall 2= 40.8 mm Treatments are from top to bottom: -RF-HD, +RF-HD, -RF+HD, +RF+HD

Weathering Experiment 2016/17 **-** We managed to generate cotton with severe sooty mould infections and were able to observe a different distribution of the mould over the boll surface. The strongest discolouration as seen in category 4 of the sooty mould scale (Fig 27a) shows spores sitting on the honeydew distributed over the *lint surface*. This season we noticed that sooty mould spread *through the lint* of the boll (Fig. 27b). This may be a result of sugars melting and seeping into the lint in hot plastic greenhouses. Sooty mould formation was strongest on honeydew contaminated bolls that had not experienced sugar wash-off from rainfall (Fig 28b). Less intense sooty mould was seen on contaminated bolls where some of the honeydew was washed off by rain (Fig 28d). Boll without honeydew that had been exposed to rainfall showed slight sooty mould contamination (Fig 28c) while control bolls which had not been exposed to either honeydew or rainfall, showed no sooty mould contamination and retained their creamy, fluffy appearance (Fig. 28a). Due to the high cost of refurbishing the HPLC at CSIRO in Brisbane ($35,000), and the fact that we have sufficient data on rainfall effects on honeydew, we decided not pursue measurement of sugar values in these samples but used them for sooty mould experiments in CSP1703.

Figure 27: Sooty mould distribution on (a) and in (b) lint



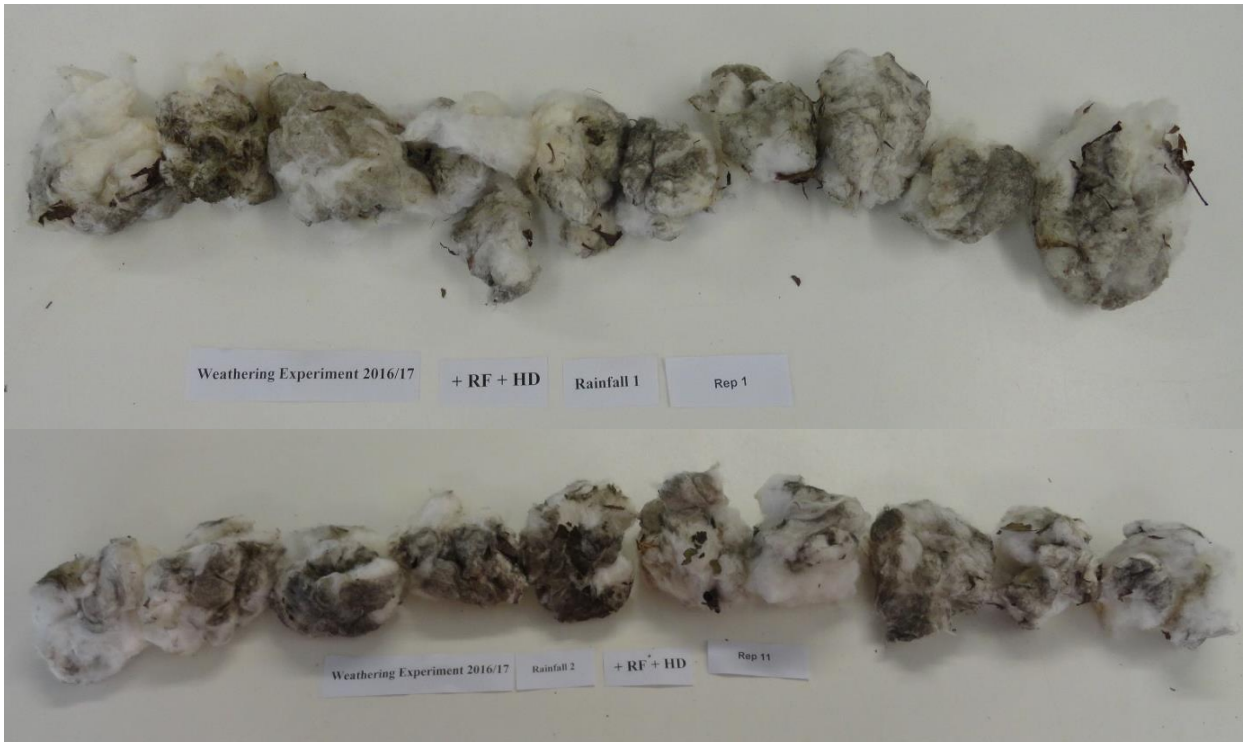
(a) Control bolls– HD –RF



(b) Sooty Mould bolls –RF +HD

Figure 28 a&b: Weathering Experiment 2016/17. Bolls from +/- Honeydew and +/- Rainfall treatments for both rainfall events

(c) Some sooty mould bolls +RF -HD



(d) Less intense sooty mould bolls +RF +HD

Figure 28 c&d: Weathering Experiment 2016/17. Bolls from +/- Honeydew and +/- Rainfall treatments for both rainfall events

b. Sooty Mould Scale

We also had the opportunity to collect sooty mould cotton from Croppa Creek which gave us a range of greyish-blackish lint contamination. We used this cotton to establish a scale that

helped us to rank sooty mould contamination which may be useful for the colour project to better describe sooty mould contamination. At present, descriptions of contamination levels are highly subjective and variable. In order to take future advantage of opportunities where honeydew has caused sooty mould growth on lint or where rainfall has caused discolouration, we made contact with consultants in different cotton growing areas to obtain their support and collaboration. This would save us travel to areas more than a few hours away, such as Griffith and Emerald, in order to collect suitable samples. It also helped spread the risk of receiving the required rainfall or natural sooty mould development that is critical to carrying out our experiments. Further, it kept us in touch with industry and gave assurance that science is working on solving the problems industry is concerned about.

Methods & Results

In the absence of any objective scale to describe sooty mould, we composed greyscale sooty mould boxes to have a standard when describing mould scores. The scale went from 0 to 4 with zero being clean cotton and four being the worst cotton we found. Bolls were presented with their mouldy side up and we tried to match them to paint cards (Fig. 29), however, this was highly subjective. The boxes were useful for three seasons after which we purchased a Hunterlab Miniscan EZ portable spectrophotometer (Fig. 30) to take objective cotton colour measurements. The instrument was part of a capital Expenditure Application (CSP1802) from savings made in the Cotton Colour Project CSP1703 and has been extremely useful in defining colour grades in the field.

Figure 29: Sooty mould rating scale

Figure 30: Hunterlab Miniscan EZ portable spectrophotometer

Conclusion

Between the research from the previous project and that done in CSP1401, we thoroughly investigated the issue of insect honeydew in cotton fields and established relationships between rainfall and sugar levels in contaminated lint. Many small technical experiments increased our knowledge of how honeydew behaves on open bolls and in the canopy. While rainfall plays an important role in washing honeydew out of open bolls, the location of the boll in the canopy, the manner in which rain falls (intensity and duration) and the weather conditions afterwards (cloudy or sunny) affect the quality of lint in the field. More honeydew washes off sticky bolls in the upper canopy during rainfall than off lower bolls. Short showers with subsequent sunshine are better at improving the condition of sticky lint than several overcast days which give opportunity for sooty moulds to grow on residual sugars on open cotton bolls. Price penalties for stickiness and lint-greying sooty mould are high. Hence, if growers find themselves in a situation where honeydew is a problem after boll opening, rain may only conditionally be the solution to cleaning the cotton up. Breakdown of honeydew by UV radiation (sunshine) does not occur in the short term and it is very unlikely that it does so in the long term. In dry warm conditions, whitefly honeydew just dries on the boll and on leaves and because it loses some of its stickiness, is more difficult to detect.

While most of the problem with discoloured lint is due to sooty mould fungi, there are situations where *Alternaria* plays a role. In a wet season there can be high spore loads in a field from *Alternaria* infected leaves and bolls and these can infect lint in open bolls to appear grey. The mechanism for this infection is unclear as *Alternaria* spores in lint are usually observed inside the lumen of the fibre but it is possible that the infection progresses through the pedicel, into seeds and then into fibres arising from the seed surface. This type of fungal quality problem requires separate study.

In general, honeydew is restricted to the outside surface of lint in open bolls, which means only a certain percentage of a boll is either sticky and/or sooty. If this percentage of the total pick is low, or the stickiness/sooty mould score of each boll is low, then the effect on the total pick may be small as effects are diluted during ginning. In this project, the generation of different degrees of sooty mould contamination depended on the presence or absence of sugars and moisture. While we learned much about the conditions conducive to mould growth, we did not investigate methods of prevention or remediation, hence sooty mould requires further study, which are being conducted through the new Cotton Colour Project CSP1901. The implications for growers with regards to honeydew are to prevent it from occurring by sound management of SLW using IPM principles.

ii) Identifying seasonal host use and ecology for SLW

This research built on earlier studies reported in the final report for CRC1102. The aim was to understand the hosts that SLW use throughout the years as this may provide allow (1) better targeting of specific weeds to reduce on-farm carry over of SLW and (2) better estimation of seasons where the risk of SLW populations carrying through winter and building in spring is higher, potentially enabling better planning of pest management strategies. Research in CRC1102 identified a range of potential hosts for *B. tabaci*. However, the next step in this process was to confirm the biotype of *B. tabaci* present on the potential host. This was important at the two biotypes of *B. tabaci* in Australia, AUS1 and MEAM1 are morphologically identical but quite different in host preferences and pest status.

Methods
We collected whitefly during survey work in the Namoi valley (2011-2016) to identify which plants SLW use in the local environment to survive and reproduce when they are not in cotton. Whitefly adults and nymphs were predominantly collected from cosmopolitan and introduced weed species. Commonly occurring natives plant species were included in surveys as well. We suspected that most of the collected whitefly were Silverleaf whitefly (*B. tabaci* MEAM 1), however the possibility of other biotypes or species needed to be tested. We sent a selection of whitefly adult and nymph samples (where we were able to collect them) from each host species to Susan van Brunschot (UQ). She used molecular techniques to confirm the species. The distinction between adult and nymph of different species is important because the presence of adults on a host indicates a possible feeding host for adults (though does not prove it) while the presence of nymphs on a host indicates it is both a host for feeding and reproduction.

Results & Discussion
Sharon has returned successful results for adults collected from 55 of the hosts found to most likely harbour whitefly, and nymphs from 40 (Appendix 2, Table 1). One of 10 samples sequenced from *Chamaesyce drummondii* (Caustic weed) turned out to be an unknown *Bemisia* nymph. This was a one-off finding, and all the other 145 samples sequenced by Sharon came back as *B. tabaci* MEAM 1. Interestingly we found no specimens of the Eastern Australian Native *B. tabac*i AUS1. Table 1 (Appendix 2) lists the more important breeding hosts in bold italics. Hosts at the bottom of the table tend to be either less common near cotton fields, or not well colonised by Silverleaf whitefly. Wheat, the last entry in the table, was the result of a single crop outbreak reported to Simone Heimoana and referred onto Tanya Smith for collections to be made. The crop concerned was planted early as animal feed and was located next to a cotton crop that had been defoliated. The surrounding area was in drought. It was inspected and monitored between May and October 2017. The initial adult infestation of SLW in the wheat was severe. Large numbers of eggs were deposited and many hatched, however the first instar crawlers didn't settle and very few first instars were found. No larger instars were found. Sharon recommended her colleague, Wanaporn Wongnikong to carry out confirmation IDs which confirm that the species was *B. tabaci* MEAM 1 for adults, but inconclusive for eggs. Based on these observations it was concluded that wheat was not a reproductive host for *B. tabaci* MEAM 1. Our results confirm that *B. tabaci* MEAM1 is the predominant species of whitefly utilising plants from the wide range of 178 hosts that we sampled and appears to have totally displaced the Eastern Australian Native *B. tabac*i AUS1.

iii) Assess mortality of SLW on cotton through the cotton season and identify potential causes.

In an associated project (CSP1303), primers for SLW were developed and used to identify the presence of SLW DNA in the gut of predators. Despite providing valuable results (see final report for 1303) there were some issues with the sensitivity of the primer. This problem emerged when we began studies to quantify how long SLW DNA remained in the gut of different predator species and stages. Such information is important for comparing the impact of the different predator species on SLW since their effect on prey populations which only have a short retention time in the gut may be underestimated. Conversely, their effect on prey with a long gut retention period may be overestimated. To test the retention period of SLW DNA in a predator species' gut, individuals of a species were each fed a single SLW adult. At varying

intervals after consuming the SLW adult, several of the predators were killed and processed to extract SLW DNA from their guts. Tests from successive sets of a given predator species over time provided information on the decline in SLW DNA in the gut of predators, and was indicated by the proportion of the predators still showing positive for the SLW DNA. We expected that the proportion of predators showing positive would be high, close to 100% at time interval zero (straight after they have fed), and then would gradually decline over subsequent intervals. However, we encountered problems due to low rates of positives for the SLW DNA, indicating poor reaction of primers with the DNA. This problem was most acute with spiders which were fed a single SLW under lab conditions and then processed straight away (time zero) to extract the whitefly's DNA. In this situation we would have expected 100% positive reaction at time zero, yet we were getting less than 10%. Similar problems occurred, though not as severe, with most other predator species tested. The implication was that predators collected from the field were probably consuming more SLW than our analyses were indicating.

Methods

The aim of the work was to improve our understanding of the current primer sensitivity and what it told us. We reasoned that if primer sensitivity was low then we may have to feed predators more than one SLW adult in order to get a clear 100% positive response to SLW DNA at time zero. We tested this using 'Detection tests' done by feeding predators either 1, 2, 3 or 4 SLW adults and then testing for positive reaction to SLW DNA. We made new collections of Lynx spiders (Lynx), Nightstalker spiders (NS), Red and blue beetles (R&B), Mite eating ladybeetles (MELB) and Apple dimpling bugs (ADB). We wanted them to be hungry and SLW free, hence they were starved to allow time for digestion and removal of gut contents, before they were fed with adult SLW. We fed them with either 1, 2, 3 or 4 adult SLW, then immediately preserved them in cooled alcohol and sent them to Dr Llewellyn's lab staff in Canberra who extracted the SLW DNA from each predator, without knowing if they had eaten SLW or not. This was important because the results required a level of interpretation and we wanted to avoid unconscious bias because of prior knowledge. Their results would enable us to determine if we could (i) achieve 100% detection from the predators, and (ii) estimate how many SLW each predator needed to consume to ensure 100% detection. The spiders tested were spiderlings between first and second moult and 1 in every 10 was adult, or pre-adult. The insect predators that we tested were adults. We chose to experiment with the same age groups used in retention trials in Project 1303, to reduce the potential for variability in our results. We repeated Nightstalker spider testing with only spiderlings, hatched in the lab, all of the same age and without exposure to SLW, to see if this made a difference to our findings. To get spiders of similar ages we collected females and held them in petri dishes for 7 days. We kept any resulting egg sacs until spiderlings hatched, then exposed them to SLW and found that they ate SLW readily.

Results

Apple Dimpling Bug

ADB adults tested struggled to eat more than 2 SLW in a 2-hour period, which was not surprising considering that the prey is as big as they are. They generally waited about an hour after consuming 1 adult SLW, before consuming another. This meant that we had limited data for 3 SLW eaten and no data for 4 SLW eaten after 3 hours. In our early work with retention times (Appendix 3, Fig. 1), 40% of ADB tested positive to SLW 30 minutes after consumption. Detection reduced beyond 30 minutes and was unlikely once 24 hours had passed. ADB returned similar results in our detection work for 1 SLW and after consuming three whitefly

all ADB tested positive (Appendix 3, Table 1). Based on these results, if ADB are consuming SLW at a rate of 3 insects inside a 3-hour period, 100% of those insects will be primer positive if preserved at the time of field collection.

Brown Smudge Bug

Previous retention tests on Brown smudge bugs (BSB) yielded satisfactory results when fed 1 adult SLW. In the original tests 70-75% of BSB tested positive 1 hour after consuming 1 whitefly. This percentage decreased exponentially with increasing time, and SLW was no longer detectable in BSB after 18 hours, indicating that BSB metabolism of SLW has similarities with that of Apple Dimpling bug. ADB detection reached a maximum in 30 minutes which was faster than BSB (1 hour), and SLW was detectable for longer in ADB (24 hours) than in BSB (18 hours) Appendix 3, (Figs 1 and 2). It would have been useful to do detection tests with BSB, including feeding varying numbers of prey but we were unable to collect sufficient BSBs to do this. The results indicate that if a BSB eats 1 SLW every hour, we will maintain a rate of detection of at least 70% and potentially greater than this.

Mite Eating Lady Beetles

Detection of SLW DNA in MELB ranged between 40 - 60% but showed no correlation with the number of prey eaten (Appendix 3, Table 2). Previous retention studies with MELB yielded disappointing results with only 10% positive detections for SLW DNA in beetles preserved 1 hour after being fed SLW. The variation in initial testing showed the primer can be unreliable for this predator. This is unfortunate as field observations of MELB when present with SLW indicated that it could be a voracious consumer of both nymphs and adults.

Red & Blue Beetles

Red and Blue beetles ate 2 SLW quickly, usually within 10 minutes, a third approximately 10 minutes later and a 4th approximately 15 minutes after that (Appendix 3, Table 3). Detection rate increased with the number of prey eaten with 100% positive after feeding on 3 or 4 SLW (Appendix 3, Table 3). In these detection studies, R&B beetles that ate 1 SLW were 70% positive for SLW while in earlier retention studies detection was slightly lower at 60% (Appendix 3, Table 3, Fig. 3). R&B consumption of 3 and 4 SLW was 100% detectable by the primer after maximum feeding periods of 55 minutes and 85 minutes, respectively. Red and Blue beetles consuming 3 or more SLW in a 1 to 1 ½ hour period will return a positive primer result. SLW DNA is likely to remain detectable for up to 18 hours (Appendix 3, Figure 3).

As in previous time trials, there was an early dip in detection rates (measured at 30 minutes after ingestion, i.e. later than the smaller predators) before recovery and a more uniform regression (Figure 3 – 30-minute data not shown). R&B beetles tested in earlier retention studies were positive in only 10% of samples when preserved 30 minutes after feeding. The difference in timing of the dip in detections between species of predators is probably a difference in the time taken to eat a SLW – R&B beetles took approximately 10 minutes to eat a SLW, whereas the MELB, BSB and ADB took up to an hour to complete a meal. The release of a chemical stimulated by initial feeding could be blocking the action of the primer and it could be at maximum effect (or beginning to subside) at the time small predators are first preserved after they finished eating. Based on the results for the other insects, it will take another 30 or more minutes to be fully active in the R&B beetles.

Lynx Spiders

Lynx spiders were slow to consume SLW adults, taking 75 minutes on average to consume 1 SLW adult. The time taken to consume 2, 3 and 4 SLW did not follow a consistent trend but took longer than 1.3 hours (Appendix 3, Table 4). At the extreme, individual spiders took as

little as 15 minutes to eat 1 SLW and up to 5 hours to eat 2 SLW. This extended time could have been distorted by the Lynx spiders' habit of often not consuming the prey's exoskeleton, instead carrying it around in their extended mouthparts for some time before dropping it. This made it difficult to tell whether they had completed a meal or not. According to protocol, Lynx spiders were deemed to be still feeding as long as they carried the exoskeleton in their mouthparts. On average Lynx spiders consumed 1 SLW every 20 to 30 minutes in lab conditions. Detection of SLW DNA broadly increased with the number of prey consumed up to 3 prey but at 4 prey detection declined slightly, though we do not understand why. There was also evidence of false positives with 10% of Lynx spiders showing positive for SLW DNA despite not having consumed it. In previous retention studies Lynx spiders preserved immediately after consumption tested less than 10% positive for SLW DNA after eating 1 SLW. Detection for the following 96 hours of the experiments varied inconsistently between 0% and 20%

Nightstalker spiders

Earlier retention studies showed that Nightstalker spiders (NS) fed 1 SLW and preserved for primer testing at and after eating, had low rates of detection, just under 10% for the first 2 hours. After this detection became erratic before dropping to zero, 36 hours after they completed eating (Appendix 3, Fig. 5). The spiders tested in those experiments were predominantly spiderlings (hatchlings or slightly older) but 10% were pre-adult or adult, evenly represented amongst tested groups for each time interval. Only spiderlings gave positive results for the presence of SLW DNA, indicating that they are more likely to score positive for SLW DNA when collected from the field.

In the more recent studies we completed two detection tests. The first test used a mixture of ages of spiders, the second only hatchling spiderlings. The spiders tested in one of the first detection test were chosen to be a similar mix of larger spiders and spiderlings as used in earlier retention studies. Total positive detections of 1 eaten SLW were 2.5 times greater in the first detection experiment than in the earlier retention work, despite the spider demographics being the same. For all spider sizes combined in the first detection test we found 30% of spiders scored positive for SLW DNA despite not being fed (Appendix 3, Table 5). Detection levels increased as the number of prey consumed increased, up to about 83% for spiders that had eaten 4 SLW adults, though these results were not as high as hoped for if the false positives are taken into account. Small spider results (Appendix 3, Table 5, numbers in brackets) were comparable with results generally, though false detections were higher (Appensix 3, Table 5). Large spiders were variable, and this was most likely due to the small numbers tested (28% false positives, no detections of 1 SLW, 33% of 2, 100% of 3 and 50% of 4 SLW consumed).

In the second test we used hatchlings (Appendix 3, Table 6). When compared with small spider results from the first test, primer detection findings were higher with essentially 100% detection for spiders that ate more than 1 SLW adult. The rate of false positives was lower at about 10%. Small spiders took longer to consume each SLW adult in the second test (30 minutes per SLW adult) compared to the first test (10-20 minutes per SLW adult). The longer prey consumption times and more promising results from the second test may reflect the uniformly small size of the hatchlings relative to the size of prey compared with the first experiment meaning each spiderling contains relatively more SLW DNA.

Discussion

ADB and R&B beetles registered similarly positive for consumption of 3 SLW in an hour and it is expected that lower consumption levels will be buffered similarly by previous consumption. Time reduced detections broadly in relation to Figures 1, 2 and 3. Without detection testing, we expect that BSB retention would fit closely to that of R&B beetles since we know that 75% of BSB that have just eaten SLW will be effectively detected using our primers. Using this information, we can make informed estimates as to the quantity of SLW being consumed at a point in time by each of the predators. We can monitor consumption changes by making collections over a time period, and population changes by sampling with with beatsheets, d-vacs or sweepnets, to inform us how much each of these generalist predators is impacting SLW populations and their development.

Results for Mite Eating Ladybeetles were disappointing, showing no consistency in detection with increasing levels of prey consumed. The long prey consumption period may have been a problem as the SLW DNA may have been digested and defecated as quickly as it was consumed, hence a positive response to increasing prey consumption may not have been detected. Nevertheless, with no false positives, a positive SLW detection for this species indicated that SLW were being consumed.

Results for Nightstalker spiders indicated that sampling should be based on a predetermined spider size to improve reliability. Retention results for both Lynx spiders and Nightstalker spiders were erratic though, and false detections for Nightstalkers in the 1st test (using all spiderlings) were unreasonably high (50%). For this reason, the primer was not suitable for use with SLW and Nightstalker spiders, and can be said to only indicate consumption in Lynx spiders where detection was greater than 10%.

This work has raised many questions and there is scope to develop or improve the primer. The size of the predators used in experiments should be considered and it would be prudent to explore whether the smaller insect predators returned poorer results because they were so small (ADB bodies approximately a slim 2mm, MELB 1-2mm) or because the primer was not as sensitive as it should have been. The laboratory work is highly labour intensive but it could be productive to increase the number of insects tested in each category in future experiments as this may refine results. It would be interesting to explore whether spider digestion varies and how that would impact detection. Another area of investigation would be the degradation of the primer during the external digestion process of spiders (Lynx and Nightstalkers) which means that the primer may not be entering the gut.

iv & v) Within plant and between plant distribution of SLW in central and southern regions.

Since 2007, the cotton industry has been using a SLW management system (sampling protocols and thresholds matrix) that was developed from research data and experiences gained from CQ. Over the last five years, an increasing number of cotton growers and agronomists have been reporting that current sampling and management recommendations for SLW give results that are not consistent from one season to the next and across different cotton growing areas. The central issue in managing whitefly across the Australian cotton industry is currently perceived to be lack of confidence in the ability of growers and crop managers to estimate SLW abundance and population growth rate within the crop accurately in relation to crop stage and link these parameters to the choice of insecticidal product(s) and the timing of spray decisions.

The overall objective of the research activities described in this section was to evaluate the accuracy and consistency over time of the existing SLW sampling recommendations and action thresholds (thresholds matrix as per the SLW section of the Cotton Pest Management Guide) in the main cotton growing areas outside Central Queensland (southern Qld and northern NSW). The research activities were conducted over four consecutive growing seasons (2014/15 – 2017/18). Analyses of the data have not been completed at the time of writing this report. An

overview of the main trends in the data are presented below. Detailed analytical results will be presented at a later date.

Methods, Results & Discussions

2014/15 - In the first year, the research objective was to determine whether or not the seasonal profile of adult SLW population density at main stem leaf node 5 (the current industry sampling recommendation) was more variable than other potential locations (sampling planes) within the crop canopy. Alternative benchmarking locations were selected at main stem leaf nodes 7, 9 and 12 (first fully unfurled leaf at the terminal = node 1) so as to represent the top and middle/bottom sections of the crop canopy. Two sampling activities were conducted in a cotton block planted on the 22/10/1014 at the ACRI experimental station at Myall Vale, Narrabri. First, sampling was directed towards quantifying SLW abundance at different nodes and was conducted from January to March 2015. The second set of activities was directed at determining whether or not adult movement and spatial distribution within the canopy were influenced by the time of day (TOD) when sampling was done. TOD sampling of leaves at nodes 5 and 9 was done at 0800, 1100 and 1400 hrs on the 23rd, 24th and 25th February 2015. The data from this first year were expected to provide the first basic insight into SLW population dynamics outside of CQ based on a formal (structured scientific) sampling protocol as opposed to data from commercial scouting of cotton crops that are typically inadequate and/or not sufficiently robust.

A summary of the 2014-15 abundance data for adult and nymph (instars 3-4, pupae) SLW in relation to crop age in days after planting and day degree (DD) accumulation {[(Temperature$_{max}$ + Temperature$_{min}$) – 24]/2} is shown in Figure 31. Mean log (base 10) density of adults increased with increasing node number, from node 5 – 12, while the coefficient of variation decreased. Adult density at node 5 was consistently more variable than the corresponding estimates on lower leaves. The predominance of adults and large nymphs in the lower half of the canopy, as evidenced by a relative increase in abundance at node 12, became evident from the start of the open boll stage at around 1800 DD (121 days after planting (DD)) (Fig. 31).

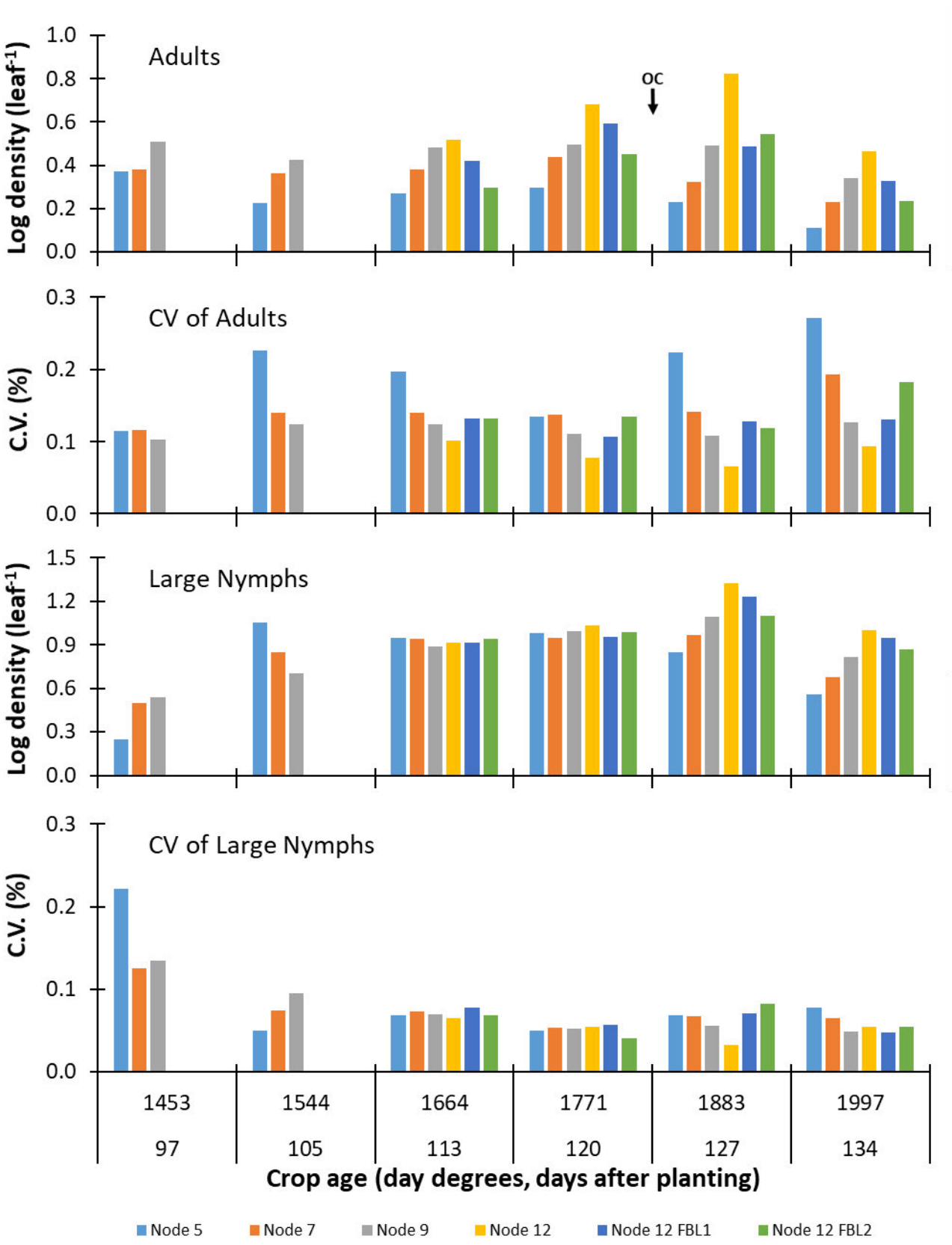


Figure 31: Seasonal profiles of mean density and coefficient of variation (C.V.) of SLW adults and large nymphs (instars 3-4 and pupae) on main stem leaves at nodes 5 – 12 and the two oldest leaves on the node 12 fruiting branch (FBL1, FBL2), at ACRI 2014/15. OC = median open cotton stage (50% of plant population at first cracked/open boll). See text for details.

The distribution of adults in the crop canopy varied among sampling dates, being higher in the morning than in the afternoon at nodes 5 and 9 on the first two and the reverse on the third occasion. A REML analysis (Genstat 19th Edition 2018) of log adult density with sampling

time and leaf node as factors nested within sampling date showed that the main effects and the sampling date x sampling time interaction effect (graphically shown in Fig. 32) were highly significant (P<0.001).

The trends in abundance and variability of adult SLW density estimates from the first year of sampling seemed to confirm industry reports of variable outcomes from sampling at the 5th node. Changes in temperature and relative humidity (RH) were suspected contributors to TOD effects and other seasonal changes in density profiles and were thus earmarked for inclusion in the research program for the second year.

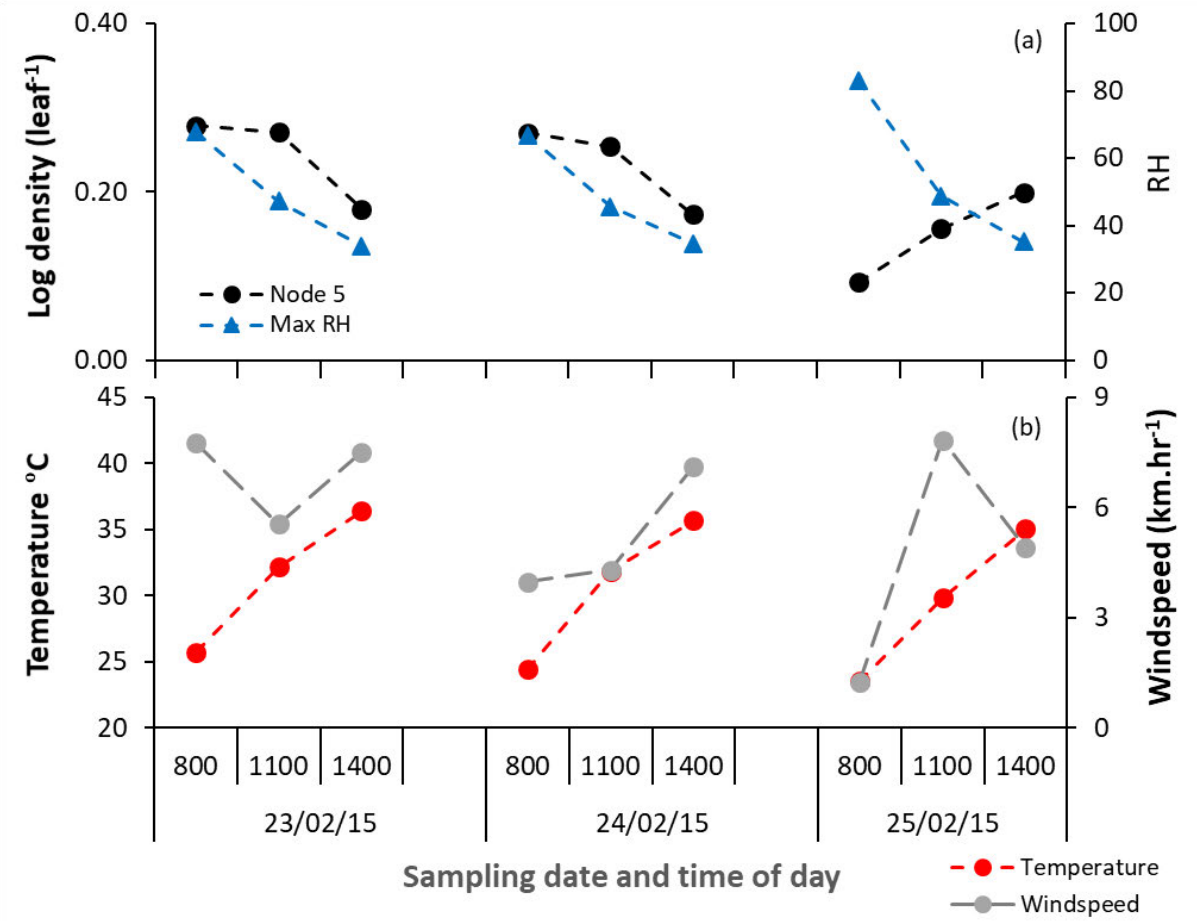


Figure 32: Changes in the mean density of SLW adults on cotton at the ACRI site (2016/17) on main stem leaves at node 5 in relation to (a) relative humidity, and (b) temperature and wind speed. Data from: see text for details.

2015/16 - The objectives of the second year of sampling were (1) to confirm the greater variability of abundance at the 5th node relative to lower sections of the canopy seen in the previous year, and (2) characterise the variability in terms of potential mechanisms and identify potential contributing factors. The experimental protocol was revised to include sampling at nodes 5 and 8 (discontinuation of nodes 7 and 9) to better align the outputs with previous SLW research in CQ and ease of sampling (R. Sequeira, unpublished data). Sampling to determine the seasonal abundance profile and variability was conducted at the ACRI and extended to four commercial cotton farms (Carson's Block, Merimbula, Retreat and Wangaree) in the Namoi/Gwydir valleys. The TOD assessment were replicated at the ACRI site with more appropriately defined sampling times (early = start time before 11AM, midday = start times between 1PM and 2PM, late = start times after 3PM).

Abundance of SLW at nodes 5 and 8 generally increased with crop age (Fig. 33). Overall abundance was lower than in the 2015/16 season. Population density at both sampling nodes varied significantly among sampling dates and time of day (unbalanced ANOVA on log_{10} transformed data with Start-time and Days after planting (DAP) as factors; $P<0.001$). Population density at node 5 was more variable than corresponding density estimates at node 8 in the first 120 days of the crop (Fig. 33). Sampling in the morning (before11AM) typically resulted in higher densities being found at both sampling nodes than sampling in the afternoon (after 3PM). This pattern became more accentuated when the crop reached the open cotton stage. There was some evidence of changes in the distribution of adults in the top third of the canopy in response to weather parameters, primarily relative humidity (RH). Figure 34 shows a significant, negative relationship between relative abundance (the difference in log density between nodes 5 and 8) and RH. The relationship is clearly stronger at midday ($R^2= 0.54$, $P < 0.001$) when temperature and RH approached their daily maxima and minima, than in the morning (early; $R2 = 0.29$, $P < 0.001$).

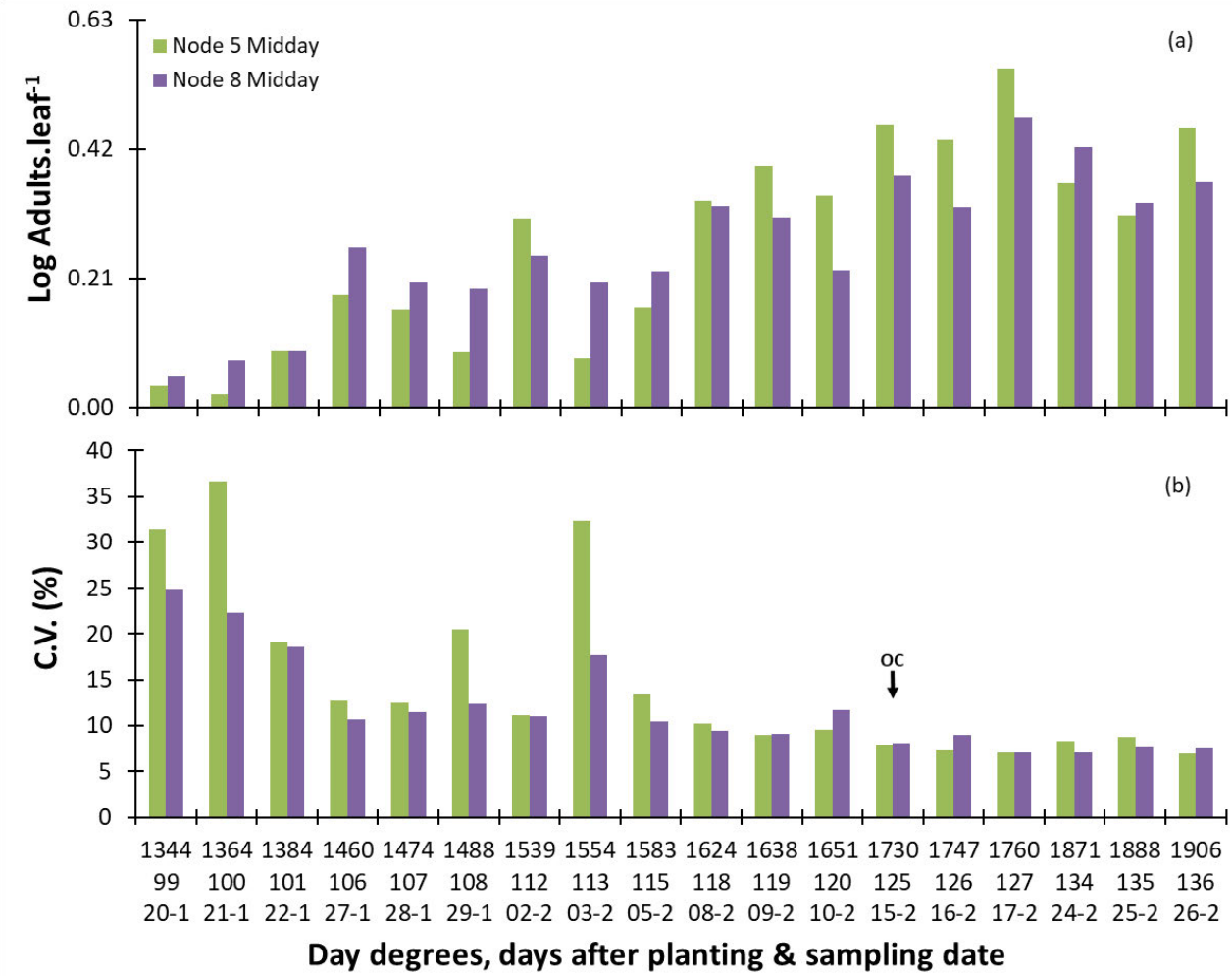


Figure 33: Seasonal profiles of mean density (a) and coefficient of variation (C.V.) (b) of SLW adults on main stem leaves at nodes 5 and 8 at ACRI 2015/16. OC = median open cotton stage (50% of plant population at first cracked/open boll). See text for details.

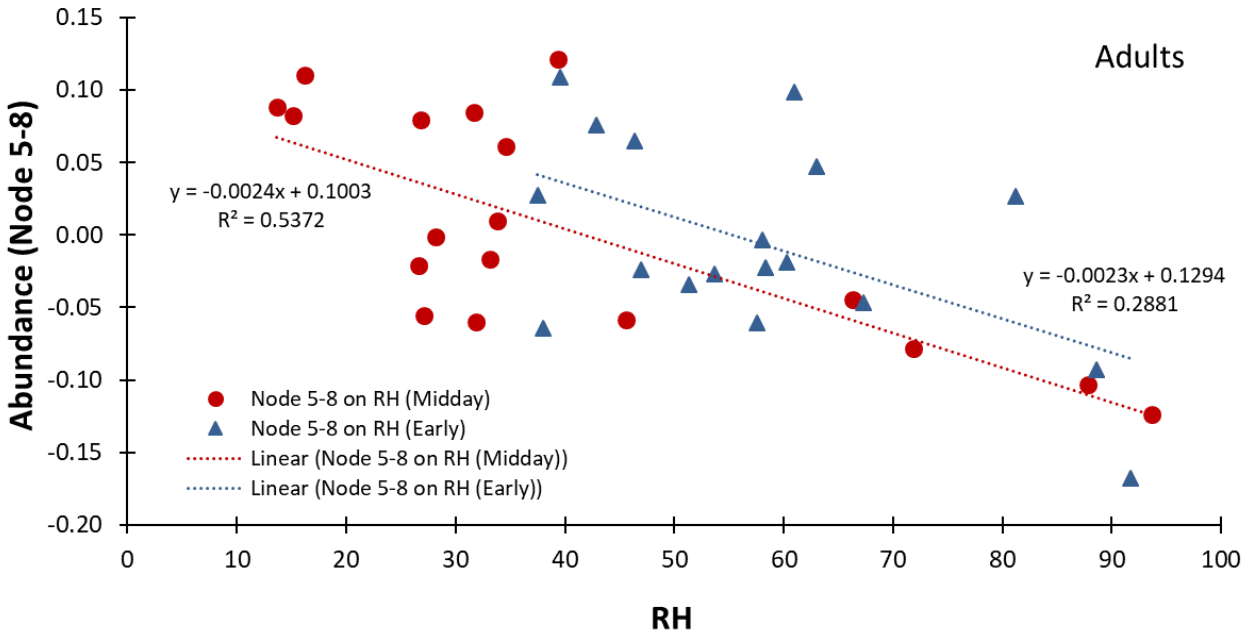


Figure 34: The change in relative abundance (log density, node 5 minus node 8) in response to the change in relative humidity (RH) in cotton at two sampling times (early and midday) at ACRI, 2015/16.

Adults seemed to be moving from the upper most to lower leaves (e.g. nodes 3-5 to node 8 and possibly lower) when RH increased to ≥70% and moved back up under more moderate humidity conditions. Estimates of the correlation between relative abundance and temperature proved to be statistically non-significant ($P>0.05$; $R^2 < 0.10$). The interaction between Start-time and DAP was highly significant ($P<0.001$), thereby indicating a dynamic shift in the vertical distribution of adults in the canopy upon the commencement of the open cotton stage (~125 DAP).

The data for large nymphs essentially mirrored the population growth and variability trends for the adults (Fig. 35). Population density (log transformed data) in the experimental population at ACRI grew exponentially leading up to the open cotton stage (~1730 DD). Density was more variable over time (crop age) at the 5th node than at the 8th. Although whitefly incidence (overall abundance) varied among sites, population density and variability profiles (linear and nonlinear trends) were similar across sites (Fig. 36).

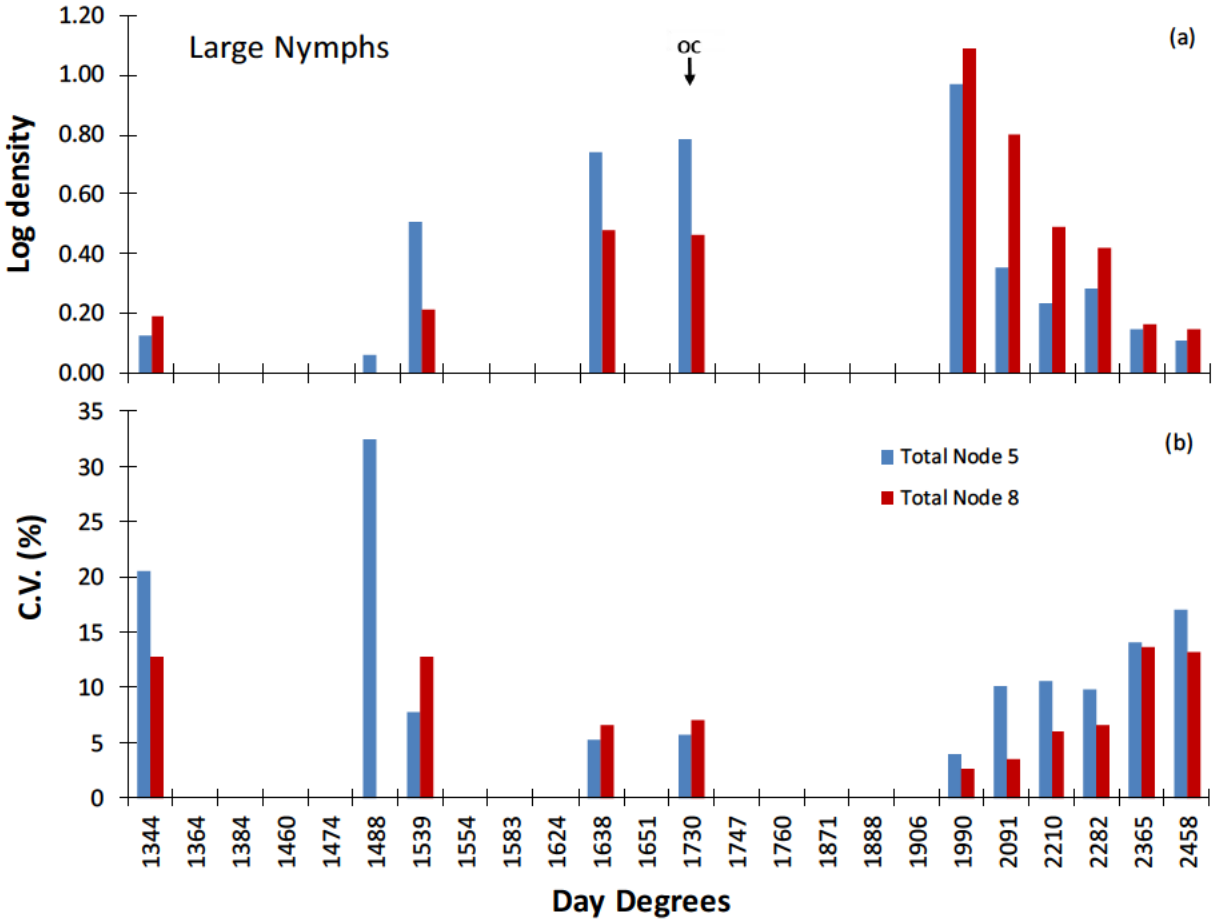


Figure 35: Seasonal profiles of mean density (a) and coefficient of variation (C.V.) (b) of SLW large nymphs on main stem leaves at nodes 5 and 8 at ACRI 2015/16. OC = median open cotton stage (50% of plant population at first cracked/open boll). See text for details.

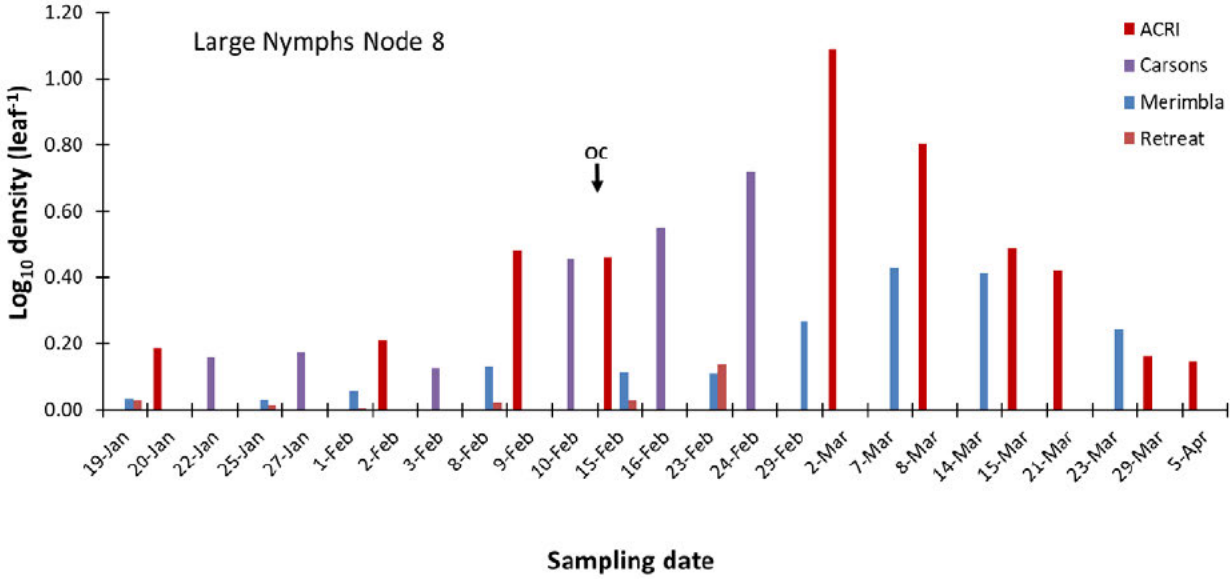


Figure 36: Seasonal profiles of mean density of SLW large nymphs on main stem leaves at nodes 5 and 8 at four different sites in 2015/16. OC = median open cotton stage (50% of plant population at first cracked/open boll). See text for details.

2016/17 - Confirmation of higher variability in sampling outcomes (and implicitly less reliability of population estimates) for adult SLW at the 5th node relative to lower nodal positions from the preceding two seasons prompted further revision of the objectives and protocols for the third season. Sampling within the canopy was extended to include a location in the bottom half of the canopy (node 14) and greater emphasis on the population dynamics of nymphs. The main research objective in this third season was to determine whether or not the population dynamics of large nymphs showed greater stability and predictability relative to their adult counterparts in the period (crop stage) from squaring to first cracked boll, which is critical from a whitefly management and insecticide application perspective. This objective was in line with industry demands for a new approach to whitefly management in cotton given the (often severe) limitations of the existing framework centred on the dynamics of adults.
Sampling for adults and large nymph was conducted at ACRI and five commercial cotton farms ([redacted]) using the 2015/16 protocol for estimating abundance/distribution and time of day effects. Below is a summary of the data for adults at the ACRI site and large nymphs at [redacted]. Whitefly densities at [redacted] were too low to warrant inclusion in this report.

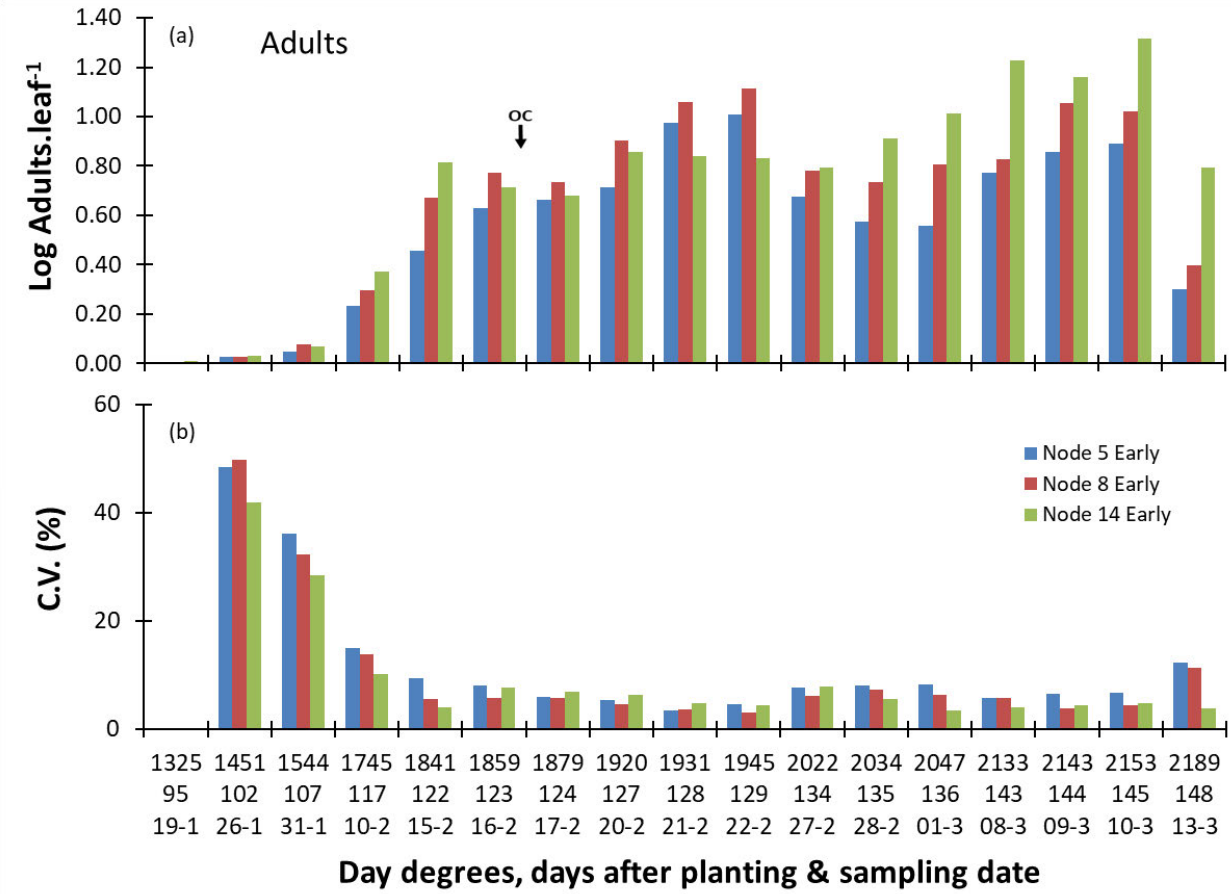


Figure 37: Seasonal profiles of mean density (a) and coefficient of variation (C.V.) (b) of SLW adults on main stem leaves at nodes 5, 8 and 14 at ACRI 2016/17. OC = median open cotton stage (50% of plant population at first cracked/open boll). See text for details.

The established pattern of greater variability of adult density at the 5th relative to lower nodes leading up to the cracked boll stage, as seen in the previous seasons, was again confirmed this season (Fig. 37). The profiles of relative abundance (difference in mean log density among nodes) indicated substantial changes in abundance of adult SLW between the upper and lower canopy during the course of the season (Fig. 38a). These were thought to be responses to changes in the weather parameters (RH and temperature).

Superimposed on the movement between upper and lower canopy sections was a more localised "cycling" of adults among leaves in the upper half of the canopy (e.g. between nodes 5 and 8), evidenced by a consistent (in 2015/16 and 2016/17) and strong negative relationship between relative abundance and RH (Fig. 38b). This mini cycling phenomenon appeared to be triggered by high RH (over 70%). From a TOD perspective, this phenomenon was identified more strongly in the midday sampling data in 2015/16 and in the early sampling data in 2016/17, i.e. in response to periods of unusually high RH. A REML analysis (Genstat linear models) showed highly significant effects of DAP, time of day and leaf node (P<0.001). All interaction effects were also highly significant (P<0.001).

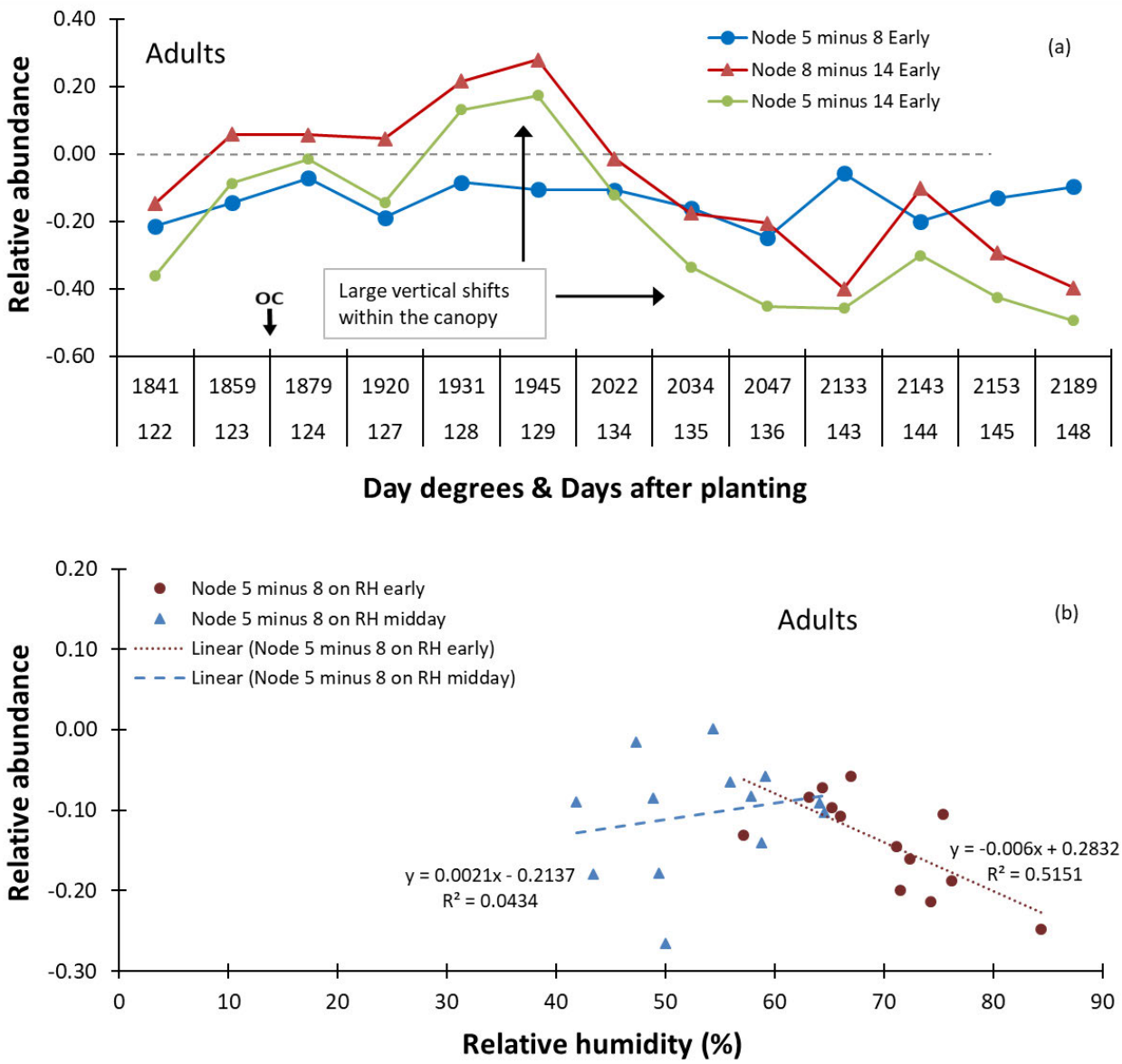


Figure 38: The change in relative abundance (difference in log density among nodes) in response to the change in relative humidity (RH) in cotton at two sampling times (early and midday) at ACRI, 2016/17.

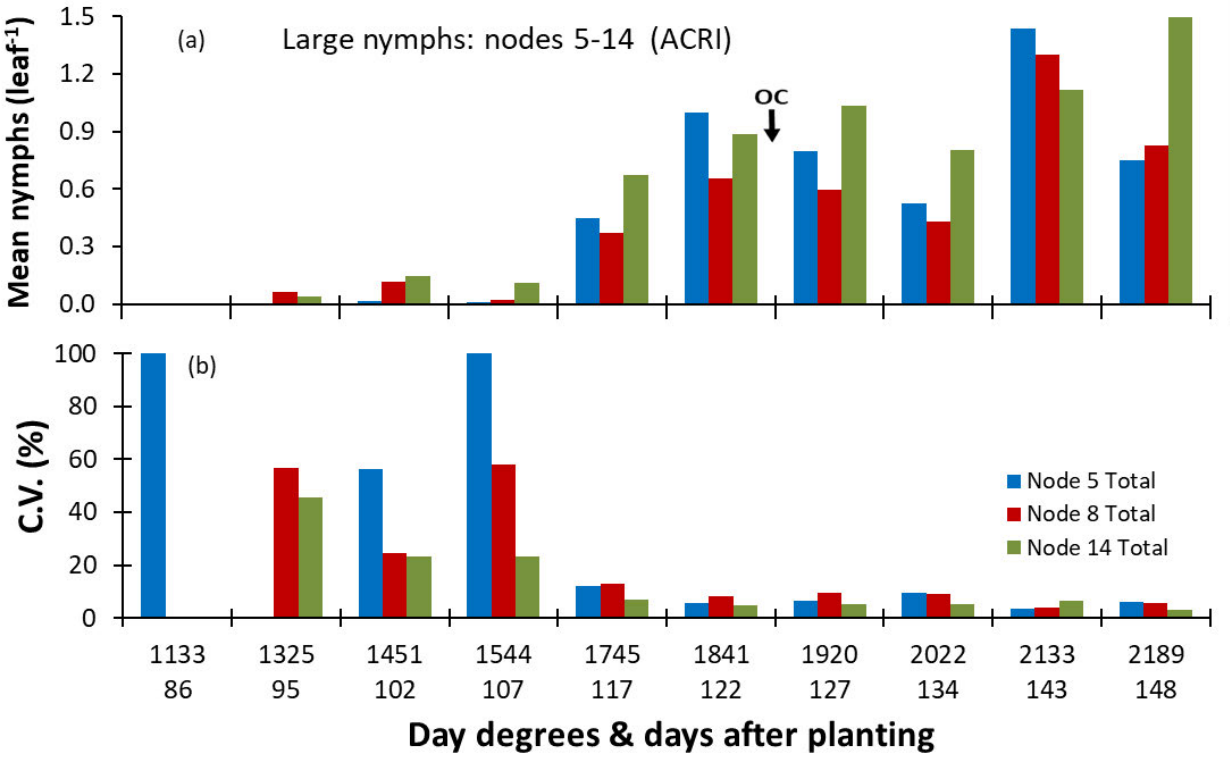


Figure 39: Seasonal profiles of mean density (a) and coefficient of variation (C.V.) (b) of SLW large nymphs on main stem leaves at nodes 5, 8 and 14 at ACRI 2016/17.

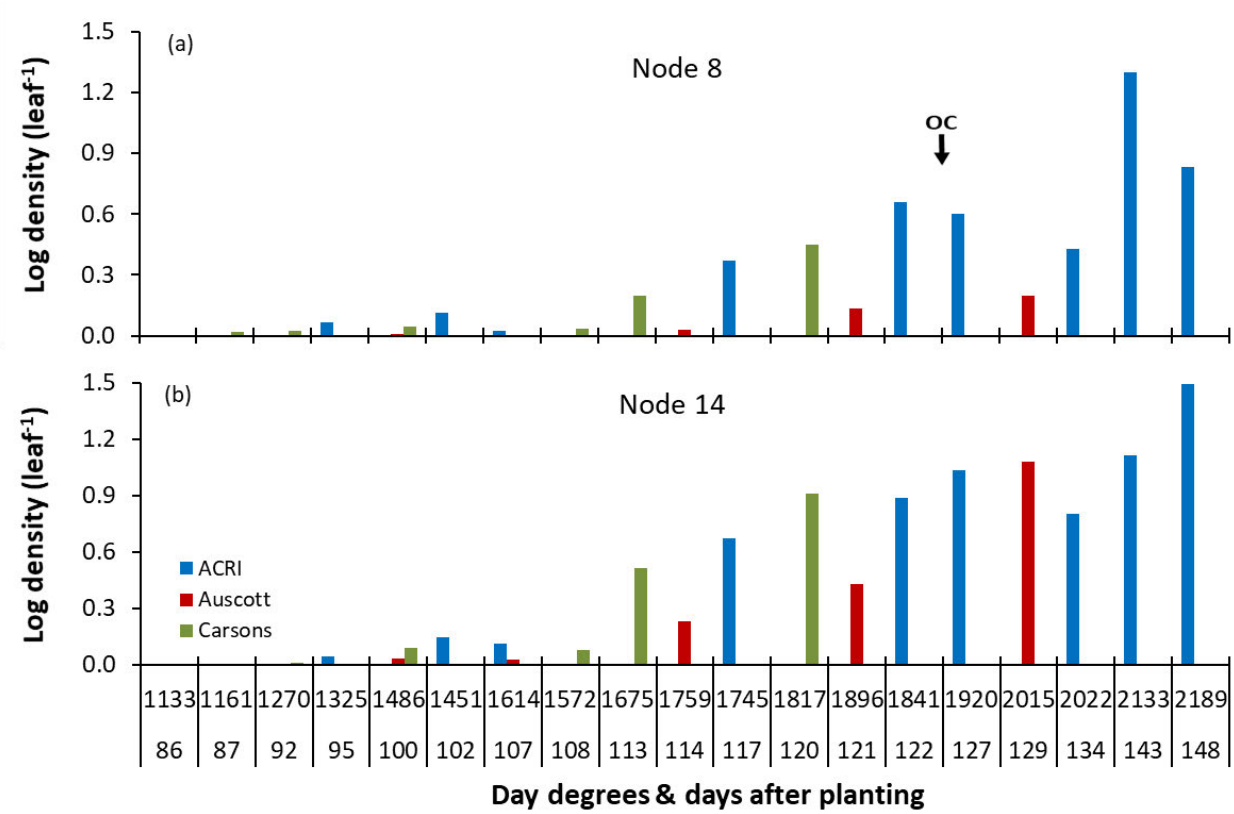


Figure 40: Seasonal mean density profiles of SLW large nymphs (includes healthy, parasitised, predated and dead) on main stem leaves at (a) node 8 and (b) node 14 at ACRI and two commercial cotton sites ([redacted]) 2016/17.

As with the adults, the population density of large nymphs at ACRI increased exponentially leading up to open cotton (cracked boll) stage and varied considerably after that (Fig. 39a). The 14th node offered the greatest consistency with the least variability in sampling estimates over

time compared to nodes 5 and 8 (Fig. 39b). An inter-site comparison of large nymph density profiles (Fig. 40) showed variability in SLW abundance among sites but similar exponential growth profiles. Density was generally higher at the 14th node than at the 8th node.

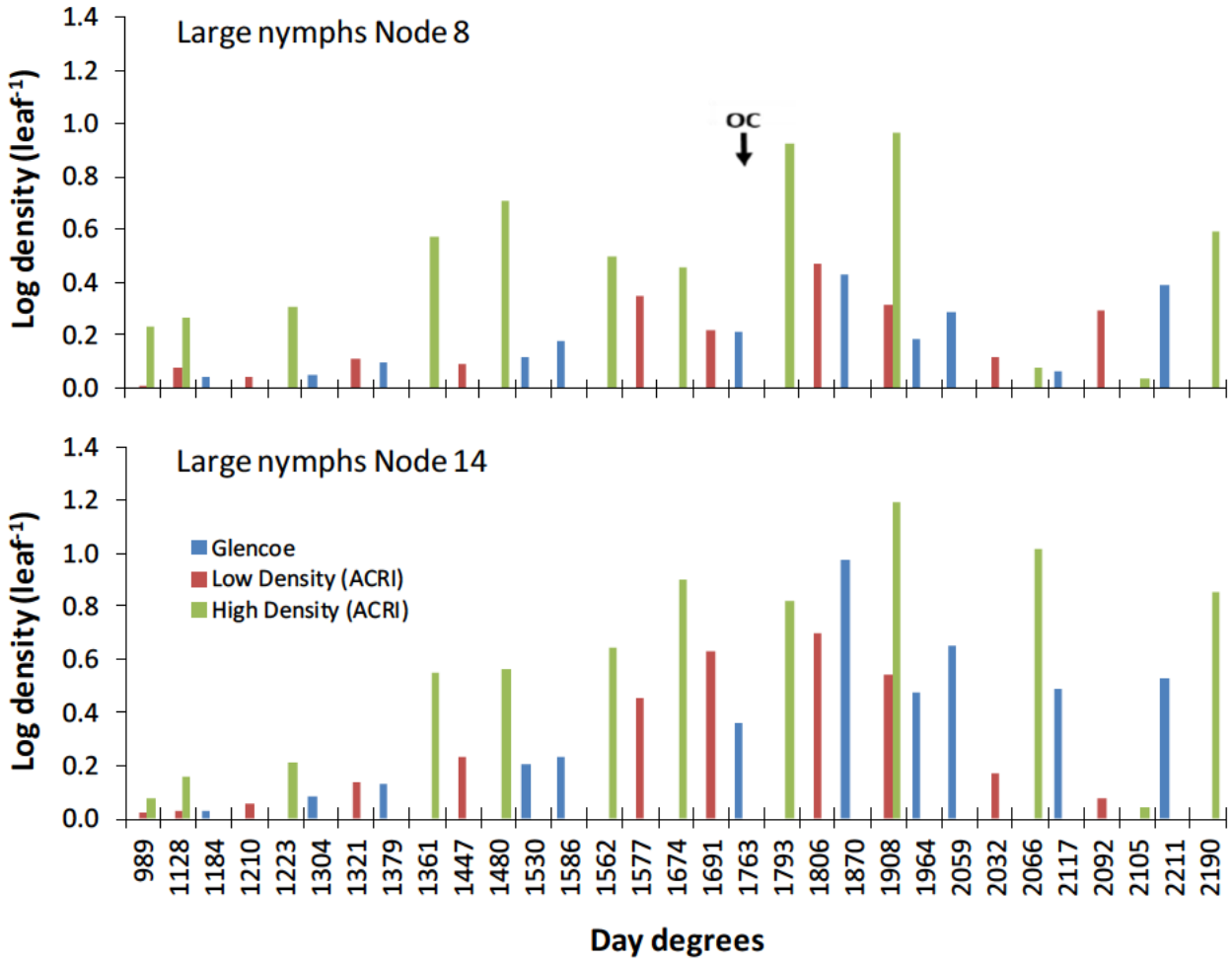


Figure 41: Seasonal mean density profiles of SLW large nymphs (includes healthy, parasitised, predated and dead) on main stem leaves at node 8 and node 14 at ACRI and a commercial cotton site (Glencoe) 2017/18.

2017/18 - The research objective in the fourth season was two-fold. Firstly, it was necessary to gather additional data to confirm the suitability of the lower half of the canopy (below node 10) as the target for a future SLW monitoring and sampling scheme based primarily on large nymphs, with the option to use adult density profiles as supporting information. Secondly, given that population growth rates can and usually do vary between regions and even among farms within regions, it was necessary to determine the potential upper limits of whitefly population growth to identify the range of densities at which crop damage (lint contamination) could be expected, as well as the minimum growth rate boundary below which the risk of crop damage is likely to be negligible. Determination of these upper and lower boundaries through sampling of commercial crops was deemed impractical and logistically impossible, primarily because such an objective would require many years of sampling. The use of experimentally manipulated "high density" and "low density" populations with intensive sampling and growth rate characterisation was deemed the most practical approach to the characterisation of population growth rate benchmarks, and implicitly the development of a virtual control decision support space (similar to the existing SLW thresholds matrix for adults) for a future SLW strategy.

The experimental protocol for assessing population growth over time was unchanged from the previous season and was applied to sampling two experimental populations (high density and

low density) created through seeding of whitefly in cotton blocks planted at ACRI. A commercial farm (Glencoe) was included in the assessment program to serve as an industry standard. Sampling to establish/verify TOD effects was dropped from the work program. Nymph sampling was extended to include the first and second fruiting branch leaves (closest to the main stem) at nodes 8 and 14.

Adult sampling at nodes 5, 8 and 14 revealed trends in population growth and variability of estimates over time similar to those observed in the previous three years. In the interest of brevity, these results are not discussed further. The data for large nymphs is shown in Figure 41 and are consistent with those from previous years in terms of population growth trend and variability over time.

Synthesis and summary

An inter-seasonal comparison of population growth over time and among sites for the large nymph data was undertaken to (a) identify the salient features of population dynamics within and among seasons, and (b) identify appropriate research questions, guide and focus the research program in the following project (DAQ1903).

Population growth profiles of large nymphs in the squaring, flowering and boll filling stages (1000 to approximately 1850 DD) were analysed for similarity using nonlinear regression methods in a two-stage process. First, an exponential growth model (Eq. 1) was fitted to the $\log_{10}$ transformed observed estimates of population density for individual sites within seasons:

$$Y = A + B*(X)^C \qquad \text{(equation 1)}$$

where A, B and C are regression parameters, Y is predicted population density and X is crop age in day degrees. The parameter A was assigned a fixed value of 4.34E-4 (equivalent to 0.001 nymphs (leaf^{-1}) in 100 metres of crop row) to constrain all growth curves to a common lower asymptote. This was necessary to model endogenous population growth of SLW in cotton and preclude computation of (unrealistic) separate intercepts for each growth curve.

In the second stage, predicted population densities for DD values ranging from 1300 to 1900 were compared among sites and seasons with respect to linear and nonlinear (shape) parameters using polynomial regression (Genstat 19th edition). The rationale for the selected range of DD values was the fact that the critical period for making whitefly spray decisions in cotton is from late flowering to the start of the open cotton stage (median cracked boll, defined as 50% of the plant population with at least one cracked boll). A third order (cubic) polynomial was fitted to predicted density for individual sites within seasons. A brief summary of the results is provided below.

The polynomial regression analysis showed that predicted growth curves for large nymphs at node 8 (Fig. 42) fall into 6 groups that are different from each other, based on the statistically significant differences among parameter estimates. The groups were ordered by the similarity (P >0.05) of linear and nonlinear (quadratic and cubic) terms that determined shape and acceleration of the growth rate: group 1 = 1415 ACRI; group 2 = 1718 ACRI high density; group 3 = 1516 (ACRI, [redacted] 1617 ACRI, 1718 ACRI low density and [redacted] group 4 = 1617 [redacted] group 5 = 1516 [redacted] [redacted] group 6 = 1617 [redacted] [redacted] The population curves at [redacted] [redacted] in both years were clearly different from each other and from all others by virtue of the uncharacteristic acceleration in population growth just prior to the cracked boll stage. Potential explanations for this anomaly at [redacted] [redacted] include practices aimed at managing other pests and additional influxes of adults from other fields/areas post colonization of the crop.

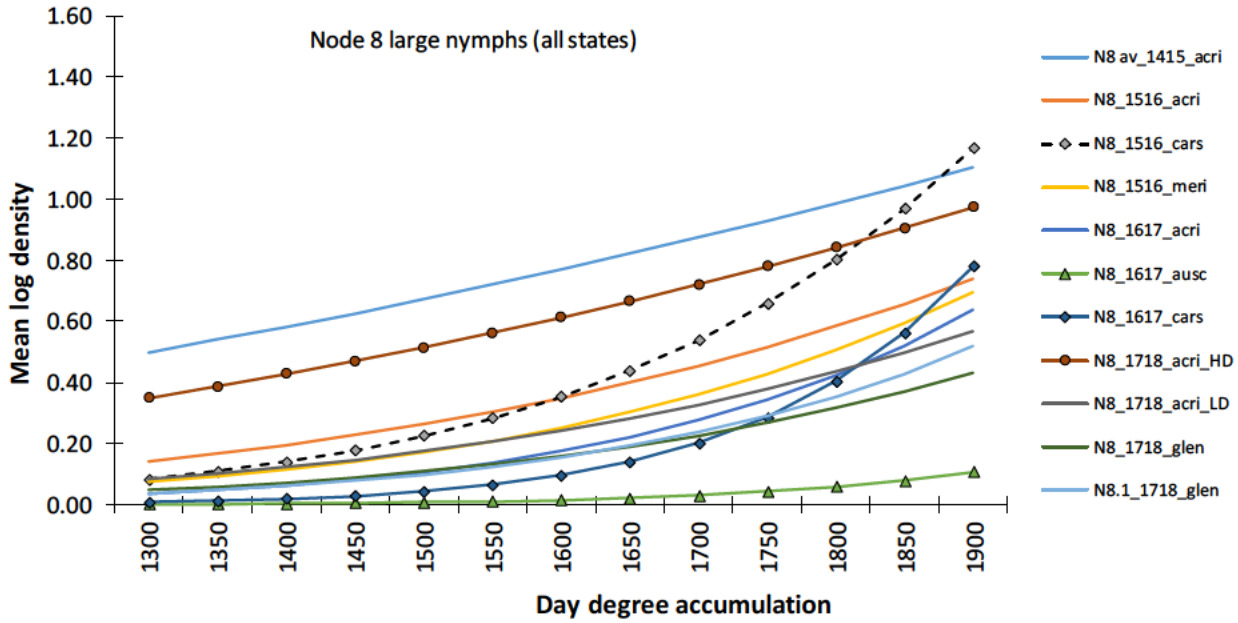


Figure 42: Predicted population growth curves for large nymphs at node 8 at multiple sites from 2014/15 (1415) to 2017/18 (1718). Site abbreviations: acri (ACRI), cars ([redacted] mere ([redacted] ausc ([redacted] acri_HD (ACRI high density), acri_LD (ACRI low density), glen ([redacted]

The corresponding analysis of node 14 curvilinear segments (Fig. 43) shows similar segregation of population growth profiles into groups differentiated on the basis of differences among parameter estimates. Of note is the visibly greater homogeneity of curves within groups and a lower level of separation among statistically distinct curves and/or groups of curves which is indicative of less variability among and within sites. The node 14 growth curves offer better discrimination between seasons, sites and the effect of extraneous influences relative to their node 8 counterparts. The distinction between natural population curves and experimentally manipulated curves (extraneous influence) is clearly apparent in Figure 43, as evidenced by the elevated curves for the high and low density population at ACRI in 2017/18 that were continually seeded with SLW to augment population growth. By comparison, natural SLW population growth curves that are typically the result of early, low level colonization of crops followed by endogenous population growth (home grown populations), as seen in the commercial crops, start at very low levels but experience higher growth rates as the crops approach the open cotton stage. The distinction between experimentally generated and natural population growth curves at node 8 is not clear.

Population growth curves for large nymphs, delineated at node 14 (Fig. 43), are an example of how a virtual decision support space, similar to the existing SLW matrix for adults, can be constructed based on knowledge of nymphal population growth characteristics in the lower half of the crop canopy. Although node 14 is an appropriate sampling location for a future SLW strategy, there may be some instances when sampling at this depth in the canopy may not be practical (e.g. sampling prior to the crop having achieved 14 fruiting branches). Future research needs to determine whether or not other nodes in the vicinity of node 14 (e.g. 11-13) afford similar pest density profile and variability results so as to offer sampling flexibility without affecting the accuracy of estimates and support for making effective spray decisions.

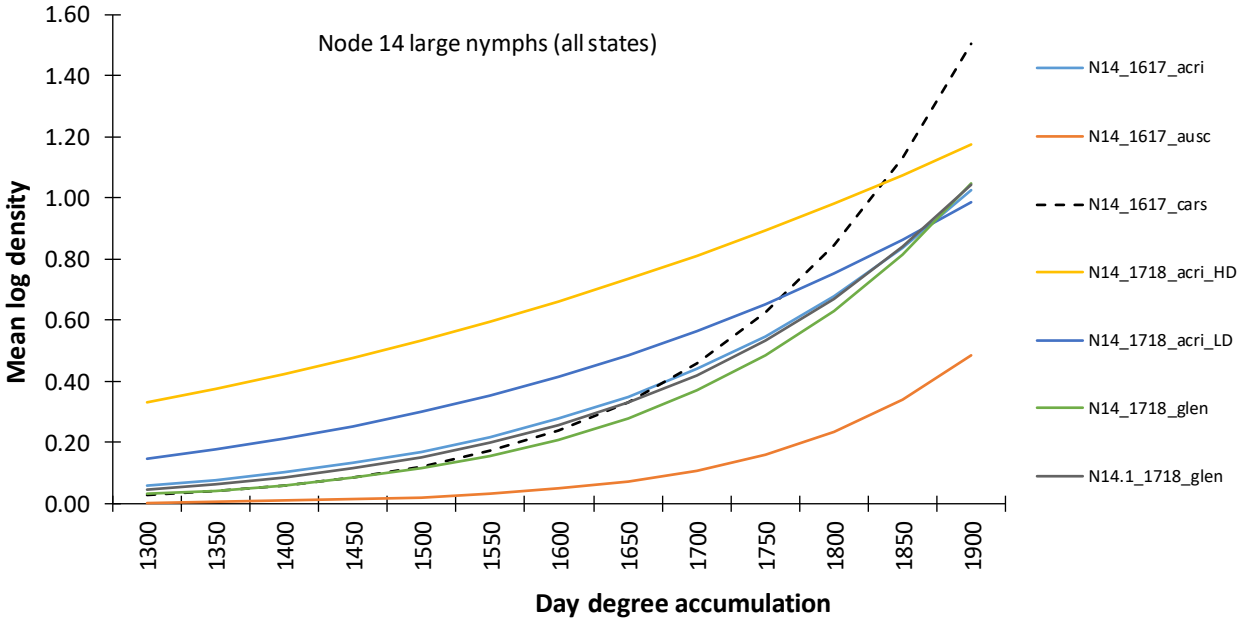


Figure 43: Predicted population growth curves for large nymphs at node 14 at multiple sites from 2016/17 (1617) to 2017/18 (1718). Site abbreviations: acri (ACRI), cars ([redacted] [redacted] ausc ([redacted] acri_HD (ACRI high density), acri_LD (ACRI low density), glen ([redacted]

Conclusion

The data from four seasons and a number of sampling sites collectively vindicate industry concerns about the lack of reliability of outcomes from the current SLW sampling and decision support recommendations. The implications are that a sampling strategy based only on changes in adult density in the upper half of the crop canopy (5th node) is no longer sufficiently robust to guide effective SLW management decisions. The weaknesses of an adult based, 5th node sampling strategy were recognised from the outset in the early 2000s when it was first formulated in CQ. However, the inherent weaknesses did not limit the effectiveness of the strategy, as evidenced by its continuing use in CQ. From an industry-wide perspective, the rapid evolution of the cotton crop in the last 15 years, as gauged by substantial increases in yield potential, changes in canopy structure and crop management, and the advent of new insect pests (e.g. cotton mealybug), has contributed to the demise of the current SLW management strategy.

The data and analyses presented here provide a strong foundation for the development of a robust and effective SLW thresholds and management strategy based on the dynamics of large nymphs in the lower half of the canopy.

Section B. IPM

i) IPM fit of new insecticides

These early season experiments are designed to evaluate the fit of new insecticides into the IPM systems in cotton and to provide this information to industry via the Cotton Pest Management Guide. This is done by evaluating the effect of new compounds on non-target species (predators and parasites) and ranking them according to a standardised system. Data is also obtained on the efficacy of the compounds against whatever pest species are present, though as the experiments are not designed specifically to test efficacy these results must be treated carefully. For instance, whiteflies usually do not build in cotton crops until mid- to late January when this experiment finishes.

Methods

Each season we contacted each of the agrochemical companies and Dr Mensah (representing the Centre for Biopesticides and Semiochemicals) to review options for testing. Our experiments accommodated up to 9 treatments – an unsprayed control and 8 spray options. The experiments required development of mite outbreaks on cotton seedlings in the glasshouse – which were then used to infest plots. Mites are a useful bio-indicator of the effects of sprays on beneficials. The experiments were sampled visually, with suction samplers and with plant/leaf samples. We used Decis Options @ 4.95 g a.i./ha (Industry Standard) and Control (no spray) as controls.

There were some slight anomalies that warrant explanation.

1. With dinotefuran we initially tested the two rates request by AGNOVA. However, even at the low rate this compound was highly efficacious against mirids so, after consultation with the company, we included it in two further years of testing at an even lower rate to see if selectivity against beneficials could be improved and efficacy against mirids maintained.
2. With flonicamid the company requested we test rates which were higher than those finally registered. This meant we had to include that product in two further years testing at a lower rate.
3. The formulation for SeroX changed after the first year of testing so we then included the final formulation for the next two years.
4. Skope at the lower rate flared mites in its first year of testing and after discussing this with the Adama we thought that the rate of emamectin benzoate in the formulation was too low to supress mites. We therefore included Skope at the higher rate in the following experiments.

Results and Discussion

The range of products and rates evaluated over the five years of the project is given in Table 7. Each year detailed reports were prepared for each compound, sent to CRDC for approval and provided to the companies. All reports are confidential to the companies and only include data for their compound(s), and the controls. As the products were registered the information in the reports was used to update Table 3 ‘Impact of insecticides and miticides on predators, parasitoids and bees in cotton.’ (Table 8). Depending on availability we also included additional information of the effect of a range of insecticides on bees, and *Eretmocerus hayati*, a parasitoid of Silverleaf whitefly, based on research completed by Dr Jamie Hopkinson (QDAF).

Table 7: Compound tested for their effects on target pests and non-target beneficials in the early season experiment 2016/17

Tradename & Compound	Company	Formulation (ai per kg or l)	Treatment rate (g ai / ha) tested	Product rate (ml or g formulation /ha)	Target Pests	2013/14 Testing	2014/15 Testing	2015/16 Testing	2016/17 Testing	2017/18 Testing
2GF-2032 SC Transform (Sulfoxaflor) Mirid Rate	Dow	240 g ai/l	72 g ai /ha	300 ml/ha	Green mirids	✓				
2GF-2032 SC Transform (Sulfoxaflor) Aphid Rate	Dow	240 g ai/l	48 g ai /ha	200 ml/ha	Cotton aphid, Green mirid	✓				
Fungus 1 (Metharizium anisopliae)	NSW DPI	100 g spores/l	50 g spores/ha	500 ml/ha	Dependent on strain	✓				
Flonicamid (Mainman) high rate	ISK / United Phosphorus	500 g ai/kg	100 g ai/ha	200 g/ha	Aphid, Mealybug, SLW, Mirids	✓				
Flonicamid (Mainman) low rate	ISK / United Phosphorus	500 g ai/kg	70 g ai/ha	140 g/ha	Aphid, Mealybug, SLW, Mirids	✓	✓			
Flonicamid (Mainman) Very low rate	ISK	500 g ai/kg	50 g ai/ha	100 g/ha	Aphids, Mirids			✓	✓	
Starkle 200SG (dinotefuran) High rate	Agnova	200 g ai /kg	75 g ai/ha	375 g/ha	SLW, Mirids	✓	✓			
Starkle 200SG (dinotefuran) Low rate	Agnova	200 g ai /kg	18 g ai/ha	90 g/ha	SLW, Mirids	✓	✓			
Starkle 200SG (dinotefuran) Very low rate	Agnova	200 g ai /kg	15 g ai/ha	90 g/ha	SLW, Mirids			✓	✓	
Cyclaniliprole (IKI 3106)	ISK/United Phosphorous	50 g ai/l	30 g ai/ha	600 ml/ha	SLW, Helicoverpa		✓	✓		
Pyriproxifen (Admiral Advance)	Sumitomo Chemical	100 g ai/l	50 g ai/ha	500 ml/ha	SLW, Helicoverpa		✓	✓		
Pyrifluquinazon		216 g ai/l	54 g ai/ha	250 ml/ha			✓	✓		
BAS440 00 I (afidopyropen)	BASF	100 g ai/l	10 g ai/ha	100 g/ha	Aphid, SLW, Scale, Jassid, Psyllid			✓	✓	
Skope (acetamiprid + emamectin) (Full rate)	Adama	218 g ai/L Acetamiprid 32.5 g ai/L Emamectin	76.3 g ai/ha Acetamiprid 11.36 g ai/ha Emamectin	175 ml/ha	Mirid, Aphid, Wfly, GVB, Heli					✓

		benzoate								
Skope (acetamiprid + emamectin) (Half rate)	Adama	218 g ai/L Acetamiprid 32.5 g ai/L Emamectin benzoate	38.15 g ai/ha Acetamiprid 5.68 g ai/ha Emamectin	175 ml/ha	Mirid, Aphid, Wfly, GVB, Heli				✓	✓
Voliam Flexi (chlorantraniliprole + thiamethoxam)	Syngenta	200 g ai/kg Chlorantraniliprole 200 g ai/kg thiamethoxam	40 g ai/ha Chlorantraniliprole 40 g ai/ha thiamethoxam	200 g/ha	Mirid, ADB, Aphid, Jassid, Heli				✓	✓
Success Neo (spinetoram)	Dow Agriculture	120 g ai/L + non-ionic wetter	48 g ai/ha	400 ml/ha	WFT, Heli				✓	✓
Sero X (FPO421A)	Growth Agriculture	380 g ai/l	760 g ai ha	2000 ml/ha	Mirid, ADB, GVB nymph, Heli		✓			
Sero X (SX151019)	Growth Agriculture	400 g ai/l	800 g ai ha	2000 ml/ha	Mirid, ADB, GVB nymph, Heli			✓	✓	
Biopest oil (Full rate)	Sacoa	815 g ai /L	1630 g ai/ha	2 L/ha	Aphids, Mites, Scale insects					✓
Biopest oil (Double rate)	Sacoa	815 g ai /L	3260 g ai/ha	4 L/ha	Aphids, mites, scale insects					✓
Buprofezin (Applaud)	Dow	440 g ai/L	440 g ai/ha	1 L/ha	SLW,					✓
Decis Options (deltamethrin) CONTROL	Bayer Crop Science	27.5g/L	4.95 g ai/ha	180 ml/ha	Mirid, GVB, Jassid, Rutherglen bug, Heli	✓	✓	✓	✓	✓

Table 8: Impact of insecticides and miticides on predators, parasitoids and bees in cotton. Note changes are marked in blue highlighter.

Insecticides (in increasing rank order of impact on beneficials)	Rate (g ai / ha)	Target Pest(s)							Beneficials																	Pest resurgence [12]			Toxicity to bees[14]
										Predatory beetles				Predatory bugs							Hymenoptera								
		Helicoverpa	Mites	Mirids	Aphids	Thrips	Silverleaf whitefly	Persistence[8]	Overall Ranking[10]	Total[1]	Red & Blue beetle	Minute 2-spt lady beetle	Other lady beetles	Total[2]	Damsel bugs	Big-eyed Bugs	Other Predatory bugs	Apple Dimpling	Lacewing adults	Spiders	Total (wasps)	Eretmocerus [19]	Trichogramma	Ants	Thrips[22]	Mite	Aphid	Helicoverpa	
Bt[11]		✓						very short	very low	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL				VL
NP Virus		✓						very short	very low	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL				VL
Pirimicarb	250				✓			short	very low	VL	VL	VL	VL	L	L	M	VL	VL	VL	VL	VL	M	M	VL	L				VL
PSO (Canopy)[16]	2%	✓			✓			short	very low	VL	L	L	VL	VL	VL	VL	VL	VL	VL	L	VL	-	VL	H	VL				VL
Methoxyfenozide	400	✓					✓	medium-long	very low	L	VL	VL	L	L	L	L	VL	VL	VL	VL	VL	-	VL	VL	VL				VL
Pyriproxyfen	50						✓	long	very low	M	-	M	M	VL	-	-	-	-	L	VL	VL	L	VL	VL	VL				L
Sero X (SX151019)	800	✓		✓			✓	short	low	VL	VL	M	L	VL	VL	VL	-	L	VL	VL	L	-	L	VH	VL				VL
Etoxazole	38.5		✓					short	low	VL	VL	-	L	VL	VL	VL	VL	VL	VL	M	L	-	VL	VL	L				VL
Indoxacarb (low)	60			✓				medium	low	L	L	H	M	VL	L	-	L	H	M	VL	VL	-	VL	H	VL		+ve		H
Indoxacarb (low+salt)	60			✓				medium	low	L	L	H	M	VL	L	-	L	H	M	VL	L	-	VL	H	VL				H
Indoxacarb (low+Canopy)	60			✓				medium	low	L	L	H	M	VL	L	-	L	H	M	VL	L	-	VL	H	VL		+ve		H
Chlorantraniliprole	52.5	✓						long	low	L	M	M	L	VL	VL	VL	L	VL	VH	VL	L	-	L	L	VL		+ve		VL
Dicofol[3]	960		✓					long	low	L	-	-	-	L	-	-	-	L	-	L	-	M	-	-	VL				VL
Sulfoxaflor (v. low)	24			✓	✓			medium	low	VL	L	VL	M	L	VL	L	L	H	L	VL	L	-	M	L	H	+ve		+ve	VH
Afidopyropen	10				✓			medium	low	L	L	H	M	VL	VL	VL	-	H	VL	VL	L	-	H	VH	L				VL
Amorphous silica[17]	2500	✓						short	low	L	L	-	M	M	-	VL	-	L	L	L	L	-	-	M	VL				-
Flonicamid very low	50			✓			✓	medium	low	VL	VL	L	L	M	VL	L	-	H	VL	L	L	-	H	VL	M				VL
Dinotefuran (very low)	15			✓				short	low	M	M	VH	M	VL	VL	L	-	H	VL	VL	L	-	VH	VH	L	+ve		+ve	VH
Spinosad	96	✓						medium	low	VL	M	L	VL	M	L	H	VL	L	VL	VL	M	H	H	H	H	+ve			H[15]
Spinetoram	48	✓				✓		medium	low	M	M	H	H	VL	VL	VL	VL	VL	VL	VL	M	-	H	VH	VH	+ve			VH
Diafenthiuron	350		✓		✓		✓	medium	low	M	H	VL	M	L	M	VL	L	H	VL	L	L	H	VL	H	L			+ve	M
Pymetrozine	150				✓			short	low	M	M	M	M	L	L	L	VL	H	M	L	L	L	L	L	M	VL				VL
Fipronil (very low)	8			✓				medium	low	L	L	L	VL	L	M	-	L	M	L	M	L	-	-	VH	L	+ve			VH
Fipronil (very low + salt)	8			✓				medium	low	L	L	L	VL	L	M	-	L	M	L	M	L	-	-	VH	L	+ve	+ve	+ve	VH
Indoxacarb	127.5	✓		✓				medium	low	H[13]	L	VH	VH	L	M	L	L	VH	M	VL	L	-	VL	VH	VL		+ve		H[15]

Cyantraniliprole	60	✓			✓[20]		✓	long	**moderate**	M	M	VL	L	M	M	M	H	L	VH	M	VL	-	VL	VL	H	+ve			VH[15]
Cyclaniliprole	30						✓	medium	**moderate**	M	M	M	VL	H	VL	H	VL	VL	VL	M	VL	-	VL	VH	L	+ve			H
Sulfoxaflor (low)	48			✓	✓			medium	**moderate**	L	L	L	M	L	VL	L	M	VH	H	VL	M	-	M	H	H	+ve		+ve	VH
Spirotetramat	96				✓		✓	medium	**moderate**	M	L	H	H	VL	VL	VL	VL	M	VH	M	M	-	M	M	M				VL
Flonicamid (high)	70			✓	✓			medium	**moderate**	VL	VL	VL	VL	H	H	VH	H	H	VL	M	M	L	H	VL	H				VL
Sulfoxaflor (mid)	72			✓	✓			medium	**moderate**	M	L	M	H	L	VL	L	M	VH	H	VL	M	M	H	H	H	+ve		+ve	VH
Dinotefuran (low)	18			✓			✓	medium	**moderate**	M	M	M	VL	M	VL	VH	M	H	VL	L	M	-	VH	H	M	+ve			VH
Abamectin	5.4	✓[6]	✓					medium	**moderate**	L	M	H	VL	M	L	M	M	H	VL	M	M	H	M	H	M				H
Emamectin	8.4	✓						medium	**moderate**	L	VL	M	VL	H	H	H	H	H	L	M	M	-	M	VL	M				H
Dimethoate (low)	80		✓[18]	✓	✓[18]	✓		short	**moderate**	M	L	H	H	M	L	-	H	M	M	L	M	-	M	H	M	+ve	+ve	+ve	H
Dimethoate (low + salt)	80		✓[18]	✓	✓[18]	✓		short	**moderate**	M	L	H	H	M	L	-	H	M	M	L	M	-	M	H	M	+ve	+ve	+ve	H
Propargite	1500		✓					medium	**moderate**	M	H	H	M	M	H	VL	VL	L	VL	M	M	L	H	H	M		+ve	+ve	L
Acetamiprid	22.5				✓			medium	**moderate**	M	M	VH	H	M	M	H	M	VH	L	VL	L	VH	H	VH	VH				M[15]
Clothianidin (low)	25			✓				medium	**moderate**	M	VL	-	H	L	M	VL	VL	H	H	M	M	H	M	VH	VL			+ve	VH
Voliam Flexi[23]	40& 40							medium	**moderate**	H	M	VH	H	L	VH	L	L	M	H	VL	M	-	VH	VH	M	+ve			VH
Amitraz	400	✓	✓[9]		✓[9]			medium	**moderate**	H	M	VH	H	L	-	-	-	H	VL	M	M	H	L	H	M				L
Skope (half)[24]	38.2 + 5.7	✓		✓	✓		✓	medium	**moderate**	H	H	VH	VH	M	-	-	-	VH	M	VL	VL	-	VH	VH	H	+ve			VH
Fipronil (low)	12.5			✓		✓		medium	**moderate**	L	L	H	L	L	H	L	L	VH	L	M	M	-	M	VH	VH	+ve	+ve	+ve	VH
Chlorfenapyr (low)	200	✓	✓					medium	**moderate**	M	L	VH	VL	M	VL	H	H	VH	L	L	M	-	VH	H	M				H
Thiamethoxam	100				✓			medium	**moderate**	H	H	H	H	M	M	M	H	H	M	VL	M	-	H	VH	H	+ve		+ve	H
Sulfoxaflor (full)	96			✓	✓		21	medium	**moderate**	H	L	M	H	L	VL	L	M	VH	H	L	M	-	H	VH	H	+ve		+ve	VH
Pyrifluquinazon	54			✓			✓	medium	**moderate**	M	M	VH	VH	H	H	H	-	VH	VH	VL	M	-	VH	VH	VL	+ve			L
Fipronil (high)	25			✓		✓		medium	**moderate**	L	VL	H	L	M	H	H	L	VH	L	M	M	M	M	VH	VH	+ve		+ve	VH
Imidacloprid	49			✓	✓			medium	**moderate**	H	L	VH	H	H	M	H	L	VH	L	L	L	VH	M	H	H	+ve		+ve	M
Clothianidin (high)	50			✓				medium	**moderate**	H	VL	-	VH	M	M	L	VL	H	H	M	M	VH	H	VH	VL	+ve		+ve	VH
Dinotefuran (high)	75			✓			✓	medium	**moderate**	H	H	H	VL	H	M	VH	M	H	VL	L	M	-	VH	VH	H	+ve		+ve	VH
Methomyl	169	✓						very short	**high**	H	L	VH	VH	M	L	VH	L	VH	M	M	M	VH	H	H	H	+ve			H[15]
Thiodicarb	750	✓						long	**high**	VH	M	VH	VH	M	M	L	L	VH	VL	M	M	-	M	M	H	+ve	+ve		M[15]
Dimethoate (high)	200		✓[18]	✓	✓[18]	✓		short	**high**	M	M	H	H	M	H	-	H	H	VH	M	H	H	H	VH	M	+ve		+ve	H
Chlorfenapyr (high)	400	✓	✓					medium	**high**	H	M	VH	L	H	H	H	H	VH	L	M	M	-	VH	VH	M		+ve		H
OP's[5]		✓	✓	✓	✓	✓		short-medium	**high**	H	M	H	H	H	M	H	H	VH	L	M	H	VH	H	VH	H	+ve			H
Carbaryl[3]								short	**high**	H	-	-	-	H	-	-	-	-	-	-	-	-	-	-	H	-	-	-	H
Pyrethroids[4]		✓	✓[7]	✓[7]			✓[7]	long	**very high**	VH	-	-	-	VH	-	-	-	VH	VH	VH	VH	VH	VH	VH	VH	+ve	+ve	+ve	H

1. Total predatory beetles – ladybeetles, red and blue beetles, other predatory beetles
2. Total predatory bugs – big-eyed bugs, minute pirate bugs, brown smudge bugs, glossy shield bug, predatory shield bug, damsel bug, assassin bug, apple dimpling bug
3. Information; Citrus pests and their natural enemies, edited by Dan Smith; University of California Statewide IPM project, Cotton, Selectivity and persistence of key cotton insecticides and miticides.
4. Pyrethroids; alpha-cypermethrin, cypermethrin,beta-cyfluthrin, cyfluthrin, bifenthrin, fenvalerate, esfenvalerate, deltamethrin, lambda-cyhalothrin,
5. Organophosphates; omethoate, monocrotophos, profenofos, chlorpyrifos, chlorpyrifos-methyl, azinophos ethyl, methidathion, parathion-methyl, thiometon
6. *Helicoverpa punctigera* only.
7. Bifenthrin is registered for mite and silverleaf whitefly control; alpha-cypermethrin, beta-cyfluthrin, bifenthrin, deltamethrin and lambda-cyhalothrin are registered for control of mirids
8. Persistence of pest control; short, less than 3 days; medium, 3-7 days, long, greater than 10 days.
9. Suppression of mites and aphids only.
10. Impact rating (% reduction in beneficials following application, based on scores for the major beneficial groups); VL (very low), less than 10%; L (low), 10-20%; M (moderate), 20-40%; H (high), 40-60%; VH (very high), > 60%. A '-' indicates no data available for specific local species.
11. *Bacillus thuringiensis*
12. Pest resurgence is +ve if repeated applications of a particular product are likely to increase the risk of pest outbreaks or resurgence. Similarly sequential applications of products with a high pest resurgence rating will increase the risk of outbreaks or resurgence of the particular pest species.
13. Very high impact on minute two-spotted ladybeetle and other ladybeetles for wet spray, moderate impact for dried spray.
14. Data Source: British Crop Protection Council. 2003. The Pesticide Manual: A World Compendium (Thirteenth Edition),. Where LD50 data is not available impacts are based on comments and descriptions. Where LD50 data is available impacts are based on the following scale: very low = LD50 (48h) > 100 ug/bee, low = LD50 (48h) < 100 ug/bee, moderate = LD50 (48h) < 10 ug/bee, high = LD50 (48h) < 1 ug/bee, very high = LD50 (48h) < 0.1 ug/bee. Refer to the Protecting Bees section in this booklet.
15. Wet residue of these products is toxic to bees, however, applying the products in the early evening when bees are not foraging will allow spray to dry, reducing risk to bees the following day.
16. May reduce survival of ladybeetle larvae – rating of moderate for this group.
17. May be detrimental to eggs and early stages of many insects, generally low toxicity to adults and later stages.
18. Will not control organophosphate resistant pests (e.g. mites, some cotton aphid (*Aphis gossypii*) populations
19. Rankings for *Eretmocerus* based on data from Jamie Hopkinson in semi-laboratory replicated experiments (QDAF) and on ranking for *E. mundus* (P. De Barro, CSIRO, unpublished) and for *E. eremicus* (Koppert B.V. , The Netherlands (http://side-effects.koppert.nl/#))
20. Suppression only
21. Transform is registered for control of greenhouse whitefly at the 96 g ai/ha rate.
22. Effects on thrips are for populations found on leaves. This is relevant to seedling crops, where thrips damage leaves, and to mid-late season when thrips adults and larvae help control mites by feeding on them as well as on leaf tissue. Note that flowers are a protected sites, so live adult thrips may be found in flowers even after crops have been treated with products that would control them on leaves.
23. Voliam Flexi is a mixture of clorantraniliprole at 200 g ai/kg and thiamethoxam at 200 g ai/kg. At the highest registered rate of 250 g formulated product/ha this is equivalent to 50 g ai/ha of each of the components
24. Skope is a mixture of acetamiprid and emamectin. At half rate (175 ml/ha) this is 38.2 g ai/ha acetamiprid and 5.7 g ai/ha emamectin. At the full rate (350 ml/ha) this is 76.3 g ai/ha acetamiprid and 11.4 g ai/ha emamectin

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ii) Options to manage mirids and GVB without flaring SLW, mites or aphids

Research reported in the Final Report for CRC1102 showed that insecticides applied to control green mirids could reduce the abundance of beneficial species and increase the risk of SLW outbreaks. We initiated further experiments to begin to understand if it is possible to effectively manage sucking pests such as green mirids and green vegetable bug without increasing the risk of inducing SLW outbreaks. The first two experiments (2012/13 and 2013/14) included a range of insecticides. However, at the conclusion of these experiments we realised that small plot sizes masked results due to plot to plot movement of insects so in Experiment 3 (2014/15) we moved to a simpler design with only three treatments and larger plots. We report here only the methods and data for the 2014/15 experiment, those for Experiments 1 and 2 can be found in Appendix 4.

Methods

***Experiment 3 (2014/15)* –** We extended plot size to 20 rows by 20 m and evaluated the effect of two insecticides, clothianidin and fipronil, both at lower rates (Table 9), which were applied against mirids and green vegetable bug, at the risk of SLW outbreaks. The method for these experiments were the same as in previous experiments but with improvements to stocking plots with GVB. Similar to the 2013/14 experiment we stocked plot sections with GVB adults and nymphs collected from nearby mungbeans or from the culture. We used beat sheets to sample the abundance of GVB and mirids in these areas.

A note on processes to develop sound experiments for pests that are challenging to work with:

GVB are sporadic with irregular distribution and shifting location in the canopy and therefore they are unreliable pest populations to work with. Their tendency to clump means that they may be missed which makes it difficult to evaluate chemical efficacy. Since they prefer the shade and coolness of the lower canopy, and sampling usually occurs in the upper canopy, they may be missed easily. Both mirids and GVB move fast. Suction sampling is noisy and leaf blockages occur frequently in taller cotton but the sampling covers 10-20 m of row which is better than a 1 m visual sample. Beat sheets still only sample 1 m but it captures most of the plant and works well for mirids and GVB and also enables release of insects.

We used a number of strategies to attract the pests to the crop. Since GVB prefer mung beans we planted mung bean strips between cotton plots, however, timing of the crops was critical so that GVB could breed up in podding mung bean while cotton was young, and then move into cotton as mung bean pods were maturing and drying off. Unfortunately GVB prefer dry mung bean pods to squaring and flowering cotton and they would not move over into the cotton in the first year of the experiment. So in the second year we decided to shift them by knocking them off with a metal bar behind the tractor and then slash the mung bean behind. We ended up with lots of dead bugs. So in the third year the objective was still to move the GVB out of the mung bean into the cotton – alive. Our breeding colonies in the lab were declining at that stage so we kept the mung bean strips as a backup resource of GVBs. However, levels of parasitism in the field were high. So in year 4 of the project (2014/15) we decided to artificially infest 2 m sections of larger cotton plots with ¾ instar GVB nymphs raised in extra rows of irrigated mung beans. We checked the following day to see how many nymphs were retained in the sections, then sprayed shortly after and re-sampled 2 and 4 days after the spray was applied. We repeated this cycle and finally had a successful experiment. In addition to managing the GVB infestations, we also had to manage the SLW infestations to evaluate effects

of GVB/mirid sprays on whitefly populations. Cotton plots were successfully infested with whitefly reared on kale in the glasshouse.

Table 9: Insecticide rates used for mirids and GVB in Experiment 3, ACRI 20114/15

Treatments	***Formulation ai/l or ai/kg***	***g ai/ha***	***Product Rate* (ml or g/ha)**
1. Control (untreated)	-		-
2. Fipronil + Salt (NaCl)	200 g/l	8.0	40 ml/ha + 1 kg NaCl/ha *
3. Clothianidin +MAXX	200 g/kg	50.0	250 ml/ha +0.02 l/l **

*One third full rate. One of the more selective options available, effective on sucking pests, short residual

** Higher rate targeting GVB. Broad spectrum, effective on sucking pests but also suppresses SLW

Results and Discussion

The terms "very low", "low", "moderate", "high" and "very high" used in this document have specific meaning, i.e. reductions compared with the untreated control of 0-10% - very low; 11-20% - low; 21-40% - moderate; 41-60% - high; >60% - very high. Where an insecticide had a negative effect on a beneficial group the magnitude is indicated in brackets.

Experiment 3 (2014/15) **–** Data in Table 10 shows that clothianidin and fipronil both effectively controlled GVB (Figs. 44 & 45), mirids (Fig. 46) and Apple dimpling bugs (*Campylomma liebknechti*). Clothianidin was slightly more effective against GVB nymphs while fipronil was slightly more effective against mirids. The mean level of parasitism of adults was similar across treatments (28.6 %, $F_{2,75} = 0.49$, $F = 0.62$).

Data for effects on beneficials are detailed in Appendix 4c. Clothianidin significantly reduced abundance of predatory coleopteran (high), red and blue beetles (*Dicranolaius bellulus*) (very high) and 'other predatory beetles' (high). Fipronil had no significant negative effects and had higher total abundance of Coccinellids than did the controls. However, the dominant species in this group were *Stethorus* spp (mite-eating ladybeetle), constituting about 80%. The high abundance of this species in the fipronil treatments, and to some extent in the clothianidin treatment probably reflect higher mite abundance in these treatments.

Both clothianidin (high) and fipronil (very high) significantly reduced the abundance of beneficials (Table 10) such as the big-eyed bug (*Geocoris lubra*). Clothianidin also significantly reduced the abundance of brown smudge bugs (*Deraeocoris signatus*). Clothianidin also significantly reduced abundance of lacewings (very high), especially larvae (very high). Fipronil reduced abundance of thrips (moderate).

Neither compound provided effective control of Rutherglen bug or jassids in this experiment. Mite abundance was significantly higher in plots treated with clothianidin or fipronil (Fig 47). Higher mite numbers probably reflect negative effects of both compounds, especially fipronil on key beneficial species, including *Stethorus* (Fig.48), thrips and Apple dimpling bugs (ADB).

Plots treated with clothianidin or fipronil had significantly higher yield than the untreated control, confirming the effect of mirids and GVB (mainly) on yield (Table 10). However in a

system it is important to also consider other effects of insecticide applications. We found that plots treated with either product also had higher mite numbers than the control. Plots treated with clothianidin had SLW abundance similar to the control but those treated with fipronil had slightly but significantly more, as well as higher honeydew contamination of leaves. The results broadly confirm those of experiments in 2012/13 and 2013/14. The higher abundance of SLW in plots treated with fipronil reflects negative effects on a range of beneficial species, while for clothianidin, negative effects on beneficials are probably offset by its suppression of SLW (see results for experiments in 2012/13 and 2013/14).

Table 10: Effect of different compounds targeting mirids and GVB on other spp.

Treatment	SLW[1,4] Adults & nymphs	Honeydew [1] contamination score (higher is worse)	Mites [2,4]	Mirids [3,4]	Total GVB[5]	Thrips [2,4]	Big-eyed bugs [3,4]	Total predatory beetles [3,4]	ADB [3,4]	Yield (b/ha)
Control	2.57	1.53	0.47	0.23	1.57	0.73	0.049	1.07	1.36	14.5
Clothianidin	2.67	1.44	0.87*	0.10*	0.90*	0.76	0.023*	0.64*	0.87*	15.2*
Fipronil + salt	2.69*	1.65*	0.76*	0.08*	0.97*	0.52*	0.009*	1.10	0.85*	16.0*
P	0.034	0.017	<0.001	<0.001	<0.001	<0.001	0.008	0.046	<0.001	<0.001
df	2,1236	2, 327	2, 83	2,83	2,112	2,83	2,83	2,83	2,83	2,17
LSD	0.09	0.14	0.133	0.053	0.19	0.10	0.025	0.02	0.09	0.71

[1]Leaf counts or scores
[2]Leaf washes
[3]Suction samples
[4]Values are ln(x+1)transformed.
[5]Beatsheet samples from artificially stocked sections of row.
*treatments significantly different from the control at 0.05 using ANOVA/LSD.

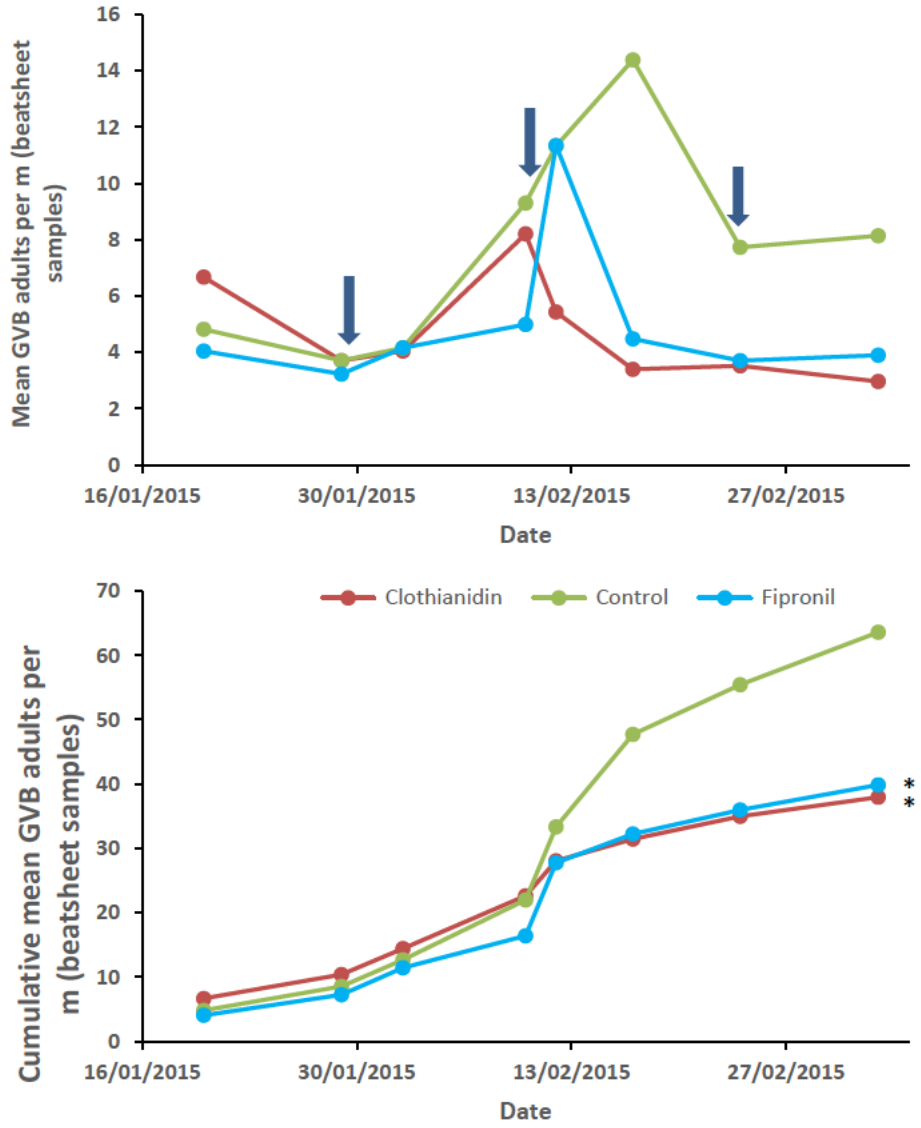


Figure 44: Adult populations of GVB (Top) mean adults from beat sheets and (Bottom) cumulative adults per beat sheet, Experiment 3, ACRI, 2014/15. Arrows indicate spray applications

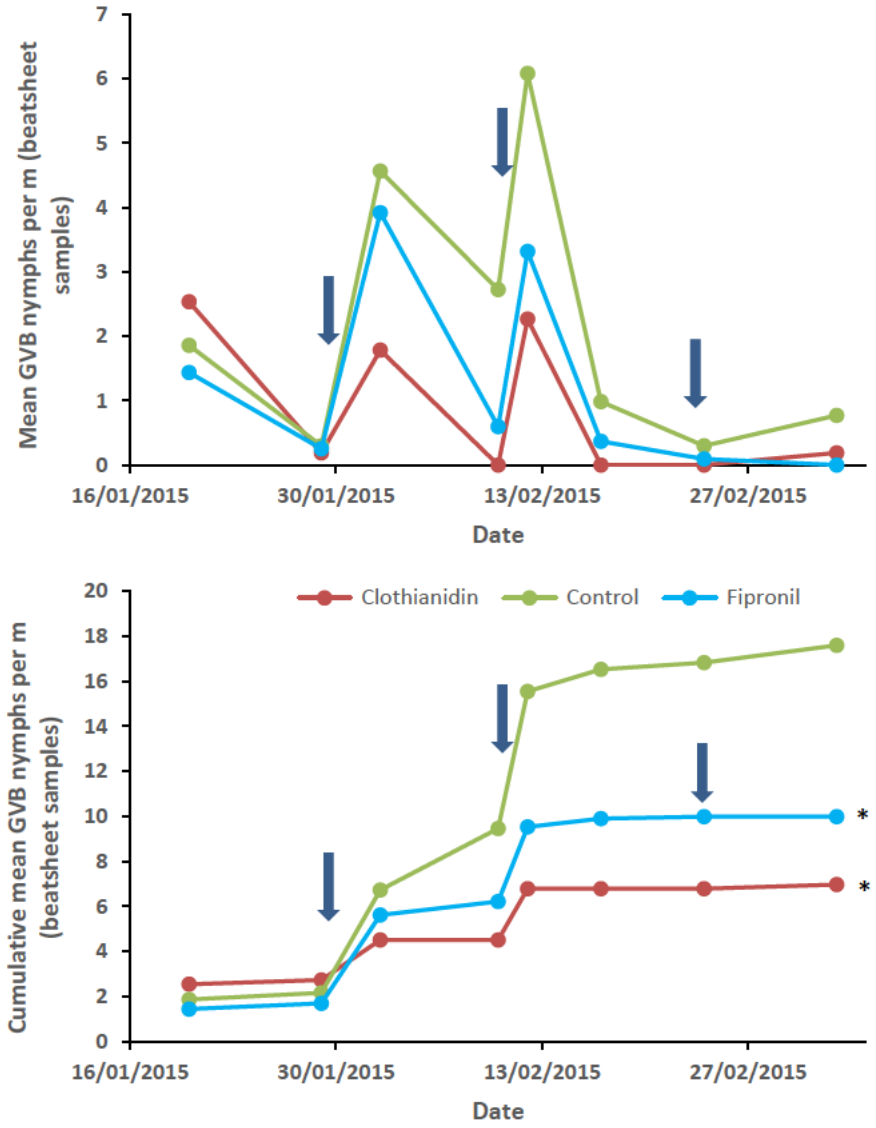


Figure 45: Nymph populations of GVB (Top) mean nymphs from beat sheets and (Bottom) cumulative nymphs per beat sheet, Experiment 3, ACRI, 2014/15. Arrows indicate spray applications

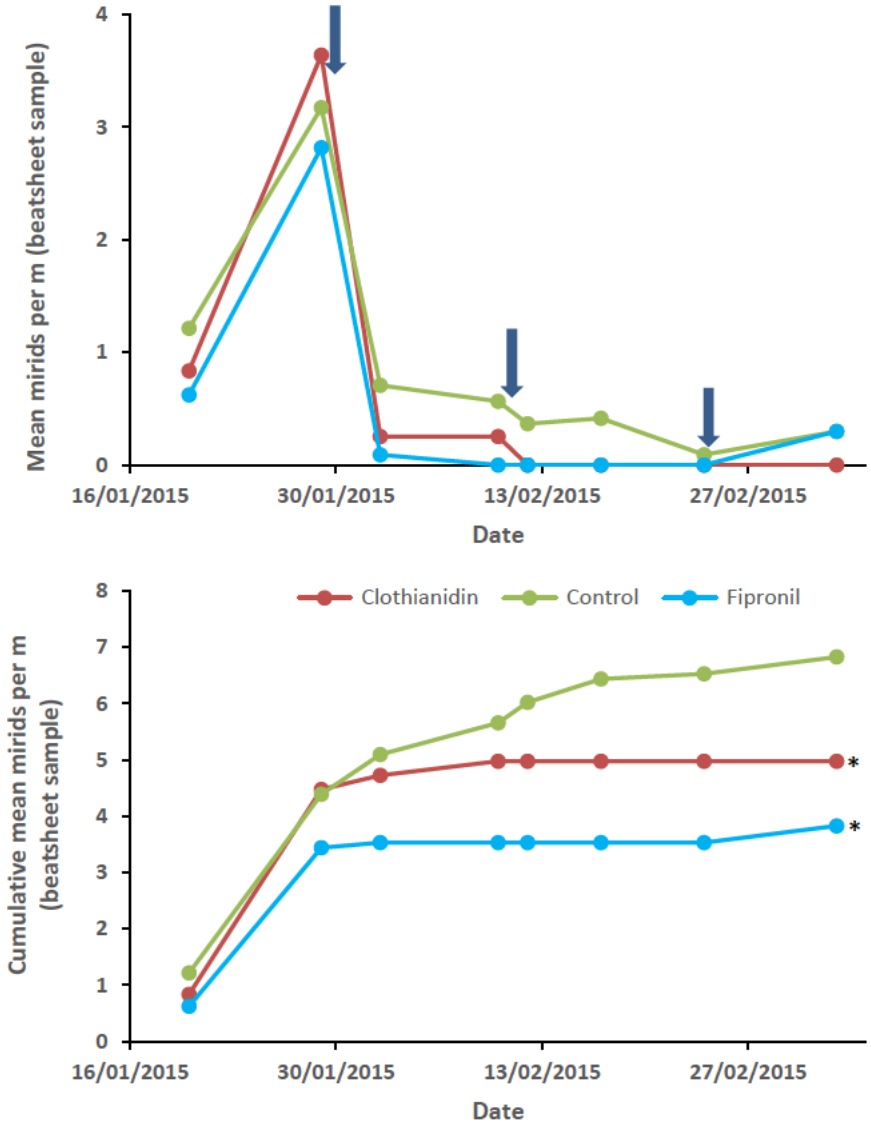


Figure 46: Populations of green mirids (Top) mean mirids from beat sheets and (Bottom) cumulative mirids per beat sheet, Experiment 3, ACRI, 2014/15. Arrows indicate spray applications

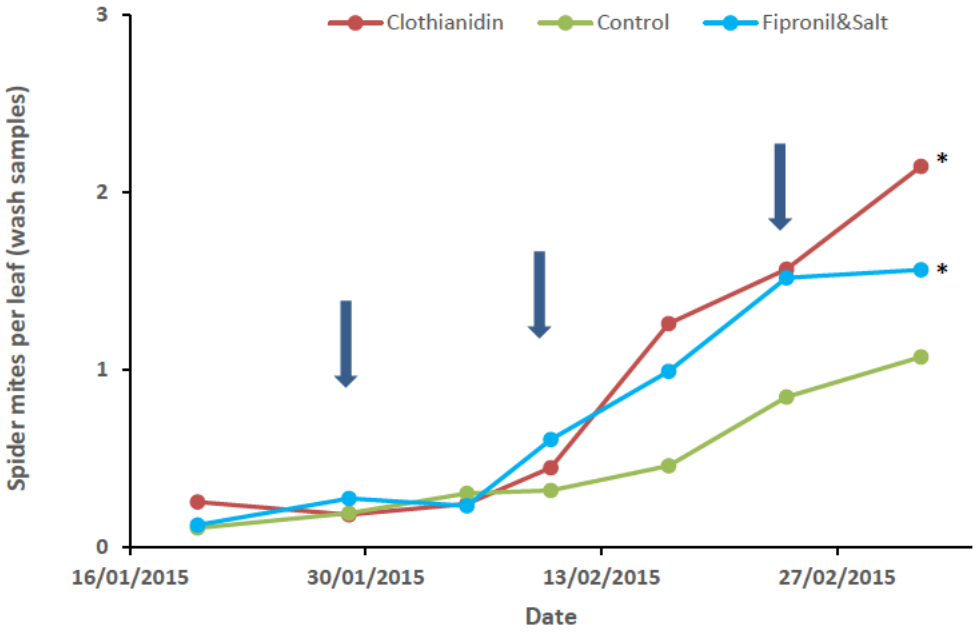


Figure 47: Populations of mites – mean mites per leaf from washes, Experiment 3, ACRI, 2014/15. Arrows indicate spray applications

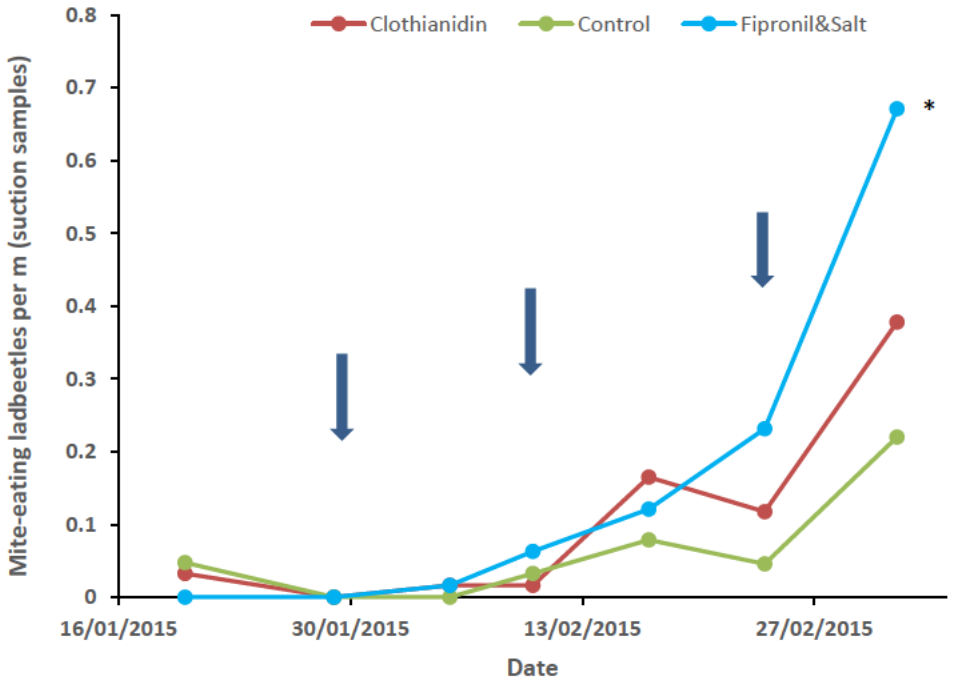


Figure 48: Populations of mite-eating ladybeetles - mean beetles from suction samples, Experiment 3, ACRI, 2014/15. Arrows indicate spray applications.

Conclusions (2012/13 to 2014/15)
These experiments showed the value of understanding insecticide, pest and beneficial interactions in a systems scenario. GVB were difficult to target but we were able to carry out experiments in these scenarios by assuring the abundance of pests and observing the interactions between the parameters. However, these experiments were difficult, time consuming, resource intensive (maintain cultures) and risky to complete. After discussion with CRDC about limited availability of water at ACRI for 2015/16 (hence area of cotton we could grow) and concerns about labour requirements we agreed to postpone these experiments to allow Tanya Smith to focus on new research developed with Dr Richard Sequeira (QDAF) and Susan Maas (CRDC) to re-evaluate sampling strategies and thresholds for SLW in central and southern regions, and to allow Dr Simone Heimoana to focus on research with whitefly honeydew and sooty moulds.

iii) To investigate options for seed treatments including several semiochemical options in collaboration with Dr Robert Mensah.

Control of seedling pests such as thrips relies heavily on use of seed treatments. These all include a neo-nicotinoid as the active ingredient targeting thrips and other seedling pests such as aphids and wireworm. In recent years there has been evidence of insecticide resistance in cotton aphid (*Aphis gossypii*) to the neonicotinoids, at least partially attributable to ongoing reliance on this group as a seed treatment. In addition, globally there are concerns about the use of neonicotinoids, even as seed treatments, and potential effects on bees, with implication for colony collapse. There is an opportunity to look for alternatives to the neonicotinoids, with a different mode of action, that provide effective management of seedling pests especially thrips and wireworm. We have begun to address this issue by comparing existing options for efficacy and by adding in novel treatments as they become available. This has included SeroX and a fungal treatment from Dr Mensah.

Methods

ACRI Experiment 1 (2013/14), ACRI Experiment 2 (2014/15), ACRI Experiment 3 (2015/16) and ACRI Experiment 4 (2016/17)

These experiments aimed to evaluate the effect of new seed treatment options on thrips, aphids, wireworm and beneficials and each year included a variety of seed treatments listed in Table 11. Treatments were developed in consultation with Robert Mensah (NSW DPI) and Rob Eveleigh (CSD). Experiments were planted at ACRI in a replicated design with plots each 8 rows by 15 m. The centre four rows of plots were sampled regularly for 6 weeks with samples collected for plant/leaf washing to assess thrips abundance. In 2015/16 and 2016/17 we also counted plant stand to assess emergence. We measured thrips numbers from leaf washes, subsamples were identified to species and dry weights of plant biomass were assessed. Non-target effects of the insecticide treatments on beneficial species were assessed from 2014/15 on by taking regular suction samples. Data was analysed by analysis of variance with treatment and date and main effects and their interaction. Crop maturity was assessed with sequential hand harvests and the centre row of each plot was machine harvested for an accurate yield assessment.

Table 11: Seed treatments tested at ACRI between 2013 and 2017

2013/14 Trts	**2014/15 Trts**
Sicot 74 BRF +Dynasty (Control)	Sicot 74 BRF +Dynasty (Control)
Sicot 74 BRF +Dynasty+ Cruiser	Sicot 74 BRF +Dynasty+ Cruiser
Sicot 74 BRF +Dynasty+ Cruiser Extreme	Sicot 74 BRF +Dynasty+ Cruiser Extreme
Sicot 74 BRF +Dynasty+ CBS 2 PXF5@ 1.0L/ha + Blood&Bone (new formulation)@0.5L/ha Sero X	Sicot74 BRF + Dynasty+ Genero
Sicot 74 BRF +Dynasty+ Blood&Bone @0.5L/ha	Sicot 74 BRF +Dynasty + Thimet (Control 2)
Sicot 74 BRF +Dynasty+ Fungus 1 @ 0.5L/ha *Metarhizium anisopliae* @ 50 g spores/ha	
Sicot 74 BRF +Dynasty + Thimet (Control 2)	
2015/16 Trts	**2016/17 Trts**
Sicot 74 BRF + Dynasty (Control)	Sicot 746 BRF + Dynasty (Control)
Sicot 74 BRF + Dynasty + Thimet	Sicot 746 BRF + Dynasty + Thimet
Sicot 74 BRF + Dynasty + Cruiser X	Sicot 746 BRF + Dynasty + Cruiser X
Sicot 74 BRF + Dynasty + Cruiser X + Thiodicarb	Sicot 746 BRF + Dynasty + Cruiser X + Thiodicarb + Fipronil
Sicot 74 BRF + Dynasty + Cruiser X + Thiodicarb + Fipronil	Sicot 746 BRF + Dynasty + Imidacloprid + Thiodicarb + Fipronil (A1)

Results and Discussion

ACRI Experiment 1 (2013/14)

The treatments produced significant differences in the abundance of thrips adults ($F_{30, 123} = 2.1$, $p = 0.002$), larvae ($F_{30, 123} = 2.1$, $p = 0.002$), total thrips ($F_{30, 123} = 8.3$, $p < 0.001$) and species composition ($F_{30, 123} = 7.1$, $p < 0.001$) (Table 12). Thrips adults are not usually a good indicator of product performance as even effective treatments are often swamped by the constant influx of them and this was reflected in inconsistent patterns of abundance between treatments (Table 12).

Thrips larvae are a better indicator as they cannot move away and they reflect both oviposition by adults as well as survival. On the first date, Cruiser had fewer larvae than the control (-77%) and Cruiser Extreme (-91%), and Thimet (-94%) provided even better control. For the next two dates, to mid-November, thrips larvae numbers in the Cruiser treatment were always lower than the control though this difference was not statistically significant (Table 12). Cruiser Extreme and Thimet showed consistently lower thrips larval abundance than the control for these two dates. Thereafter there were no consistent trends in treatment effects on thrips abundance. Dynasty with Blood and Bone, Fungus 1 and Sero X provided no significant control of thrips on any date. The results for total thrips were similar.

On the first sample date there were differences between treatments in the proportion of *Frankliniella occidentalis* in the population, with proportions higher in the Cruiser, Cruiser Extreme and Thimet treatments. This reflects poor control of this species but effective control of *Thrips tabaci* compared with the control and other treatments. Thereafter populations were generally dominated by *Thrips tabaci*, ranging between 52 – 100% of the total thrips population.

Table 12: Abundance and species composition of thrips at ACRI B17, 2013/14

Date	Treatment	Mean Thrips/plant			Mean Thrips Species as % of subsample			
		Thrips adults/plant	Thrips larvae/plant	Total thrips/plant	*F. occidentalis*	*F. schultzei*	*T. tabaci*	*Other thrips spp.*
01/11/13	Control	2.62 a	13.66 a	14.39 a	27.50 b	0.00	72.50	0.00
	Cruiser	2.54 a	3.06 b	3.81 b	61.20 a	0.00	35.00	3.75
	Cruiser X	2.90 a	1.11 c	1.59 c	53.20 a	0.00	44.30	2.50
	Dynasty	2.59 a	11.26 a	11.94 a	23.70 b	1.25	67.50	7.50
	BB	3.08 a	15.20 a	16.00 a	18.90 b	0.00	79.80	1.25
	Fungus 1	3.74 a	16.87 a	17.78 a	28.80 b	0.00	71.20	0.00
	Sero X	0.85 a	0.73 c	1.04 c	51.70 a	0.00	39.60	8.73
	Thimet							
08/11/13	Control	0.83 b	4.10 ab	4.72 ab	20.50 a	0.00	51.70	2.78
	Cruiser	1.66 a	1.76 bc	2.54 c	20.90 a	0.00	79.10	0.00
	Cruiser X	1.12 ab	0.88 c	1.57 c	24.20 a	0.00	75.80	0.00
	Dynasty	0.65 b	2.63 a	3.20 bc	11.40 a	0.00	87.00	1.56
	BB	0.87 b	4.88 a	5.48 a	11.60 a	0.00	79.30	9.09
	Fungus 1	0.77 b	5.99 a	6.63 a	16.90 a	0.00	83.10	0.00
	Sero X	1.08 ab	1.00 c	1.68 c	18.90 a	0.00	79.80	1.25
	Thimet							
14/11/13	Control	0.81 a	1.28 a	1.87 a	8.20 a	0.00	76.80	14.93
	Cruiser	0.64 a	0.96 ab	1.46 ab	17.50 a	0.00	74.80	7.74
	Cruiser X	0.43 a	0.44 b	0.75 b	26.70 a	0.00	73.30	0.00
	Dynasty	0.59 a	0.80 ab	1.25 ab	11.90 a	0.00	75.40	12.69
	BB	0.45 a	0.79 ab	1.08 ab	13.80 a	0.00	70.40	15.77
	Fungus 1	0.76 a	1.19 a	1.84 a	3.80 a	0.00	96.20	0.00
	Sero X	0.49 a	0.39 b	0.77 b	10.30 a	0.00	81.90	7.78
	Thimet							
20/11/13	Control	0.54 a	2.22 a	2.67 a	12.70 a	0.00	62.30	25.00
	Cruiser	0.57 a	2.40 a	2.90 a	12.00 a	0.00	60.20	2.78
	Cruiser X	0.38 ab	3.18 a	3.50 a	15.50 a	0.00	84.50	0.00
	Dynasty	0.49 a	3.28 a	3.73 a	16.70 a	0.00	83.30	0.00
	BB	0.55 a	3.50 a	3.99 a	14.00 a	0.00	86.00	0.00
	Fungus 1	0.11 b	2.52 a	2.93 a	1.00 a	0.00	90.00	10.00
	Sero X	0.30 ab	2.14 a	2.34 a	25.00 a	0.00	75.00	0.00
	Thimet							
27/11/13	Control	0.15 a	0.26 b	0.41 b	0.00 a	0.00	100.00	0.00
	Cruiser	0.42 a	0.97 a	1.34 a	11.20 a	0.00	88.70	0.00
	Cruiser X	0.29 a	0.69 ab	0.91 ab	8.30 a	0.00	58.30	33.33
	Dynasty	0.10 a	0.50 ab	0.59 ab	0.00 a	0.00	50.00	0.00
	BB	0.10 a	0.88 ab	0.93 ab	0.00 a	0.00	50.00	0.00
	Fungus 1	0.17 a	0.75 ab	0.94 ab	0.00 a	0.00	75.00	0.00
	Sero X	0.12 a	0.41 ab	0.53 b	0.00 a	0.00	50.00	0.00
	Thimet							
04/12/13	Control	0.34 a	0.71 a	0.92 a	0.00 c	0.00	50.00	0.00
	Cruiser	0.13 a	0.93 a	1.06 a	0.00 c	0.00	25.00	0.00
	Cruiser X	0.07 a	1.02 a	1.06 a	12.50 b	0.00	12.50	0.00
	Dynasty	0.24 a	1.06 a	1.10 a	50.00 a	0.00	25.00	0.00
	BB	0.26 a	1.17 a	1.39 a	0.00 c	0.00	50.00	0.00
	Fungus 1	0.10 a	0.71 a	0.75 a	25.00 b	0.00	25.00	0.00
	Sero X	0.15 a	0.61 a	0.75 a	2.00 b	0.00	75.00	0.00
	Thimet							

Values are back-transformed means from analysis of ln(x+1) transformed data.
Within each column and within each date treatments with different letters are significantly different from each other at $P<0.05$.

Analysing across all dates for thrips adults ($F_{6, 123} = 3.08$, $p = 0.008$), larvae ($F_{6, 123} = 24$, $p < 0.001$), total thrips ($F_{6, 123} = 3.08$, $p = 0.008$) and species composition ($F_{30, 123} = 20$, $p < 0.001$) (Table 13), Thimet significantly reduced the abundance of adults by about 40%. For larval and total thrips Cruiser reduced abundance by about 31%, Cruiser Extreme by about 52% and Thimet by about 65%.

Table 13: Abundance and species composition of thrips across all dates at ACRI B17, 2013/14

Treatment	Thrips A/plant	Thrips I/plant	Total Thrips/ plant	% F.o.	% F.s	% T.t	% Unidentified
Control	0.75[a]	2.26[bc]	2.69[ab]	11.50[bc]	0	68.90	7.12
Cruiser	0.84[a]	1.56[d]	2.04[c]	20.50[ab]	0	60.50	2.38
Cruiser X	0.69[a]	1.08[d]	1.43[cd]	23.40[a]	0	58.10	5.97
Dynasty BB	0.63[a]	2.19[c]	2.53[b]	18.90[abc]	0.21	64.70	3.63
Fungus 1	0.69 [a]	2.82[ab]	3.18[a]	9.70[c]	0	69.30	4.35
Sero X	0.75[a]	2.77[a]	3.18[a]	12.40[bc]	0	73.40	1.67
Thimet	0.46[b]	0.80[d]	1.10[d]	17.70[abc]	0	66.90	2.96

Values are back-transformed means from analysis of ln(x+1) transformed data.
Within each column treatments with different letters are significantly different from each other at P<0.05.

Yield

Hand picks

There was no significant difference in yield (bale/ha) between treatments at ginout percentages of 48-49%. There was also no difference for the number of bolls/m. For Cruiser and Cruiser Extreme both, 60% of bolls were open two and seven days earlier, respectively, than the control (Table 14).

Table 14: Yield at ACRI B17, 2013/14

Treatment	Yield (b/ha)	Bolls per m	60% open boll DAS*
Control	14.60	188.5	157.40[c]
Cruiser	15.21	196.0	149.65[a]
Cruiser X	16.85	211.2	145.71 [ab]
Dynasty BB	14.84	189.8	157.25[c]
Fungus 1	14.85	209.2	159.04[c]
Sero X	14.84	187.5	155.91[bc]
Thimet	16.85	213.5	157.25[c]
F	0.158	0.379	0.013
LSD (P=0.05)	n.s.	n.s.	7.584
df	27	27	27

*** Days after Sowing**

Disappointingly neither Dynasty BB, Fungus 1 nor Sero X provided control of thrips or increased yield.

Machine Picks

There were no significant treatment effects on yield from the machine picks (Table 15).

Table 15: Yield (machine picks) at ACRI B17, 2013/14

Treatment	Yield (bales/ha)
Control	9.68
Cruiser	10.17
Cruiser X	10.16
Dynasty BB	9.65
Fungus 1	9.54
Sero X	9.05
Thimet	9.75
F	0.664
LSD(P=0.05)	n.s.
df	6, 55

ACRI Experiment 2 (2014/15)

Thrips populations were predominantly *T. tabaci* (>85%) with a small percentage of *Frankliniella occidentalis*. There was generally low thrips pressure this season and although the seed treatments and in-furrow granular (Thimet) had slightly lower numbers of thrips than the untreated control, none provided significant control (Table 16). Similarly, there were no differences in plant dry weight, boll weight boll number, maturity date or machine harvested yield. The generally poor control of thrips across the treatments is surprising, and may reflect resistance but more likely poor uptake of insecticide because the crop was watered up rather than pre-irrigated. This is an issue that could be looked at in future.

Table 16: Comparison of neonicotinoid seed-treatment and Thimet effects on thrips abundance, plant growth, crop maturity and yield (machine picks). ACRI 2014/15

	Mean thrips/plant						
Treatment	**Adults**[1]	**Larvae**[1]	**Mean plant dry weight (g)**	**Mean boll weight (g)**	**Mean boll number**	**Maturity date (Days after sowing)**	**Yield (b/ha)**
Control	0.62	1.89	0.59	4.99	121.6	149.3	15.6
Cruiser	0.61	1.99	0.60	4.91	129.9	148.4	14.9
Cruiser X	0.61	1.26	0.57	5.07	131.9	151.3	15.5
Genero	0.58	1.66	0.60	4.87	128.6	150.1	15.0
Thimet	0.69	1.70	0.65	4.96	119.1	150.4	15.6
P value	0.79	0.15	0.51	0.76	0.66	0.53	0.87
LSD	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
df				4, 12			

[1]Values are back-transformed means from analysis of ln(x+1) transformed data.
Within each column treatments with different letters are significantly different from each other at P<0.05.

Impact of treatments on beneficial species

Overall the seed treatments and Thimet had low effects on beneficial species, possibly reflecting poor uptake of the product (Appendix 5a). However, since predatory insects are generally not herbivores, this would not be a factor of concern unless predators would be affected through the consumption of herbivorous prey. Systemic neonicotinoids are known to have lethal and sublethal effects on beneficials and the three neonicotinoid treatments caused significant reductions in the abundance of predatory Coccinellids ranging from high (Genero) to very high negative effects (Cruiser and Cruiser Extreme) (Table 17). This was reflected in

high significant negative effects on the overall predatory beetle fauna for Cruiser and Cruiser Extreme, and - though not statistically significant - Genero trended this way as well (Table 17). The abundance of spiders trended lower in all insecticide treatments compared with the untreated control in the suction samples and this was significant for Cruiser Extreme and Genero, and for Thimet in the leaf wash samples (Table 17).

Discussion 2013/14 & 2014/15

Although there were no differences in thrips abundance or yield between treatments the data indicate that the insecticide treatments can influence early season beneficial abundance. It is worthwhile to assess this further, especially in a situation where they provide very good thrips control compared with the untreated.

Table 17: Summary of mean abundance of key predatory or parasitic groups in each insecticide seed treatment, ACRI, 2014/15.

	Rate	Total Coccinellids (suction samples)		Total Coleoptera Beneficials (suction samples)		Total predatory Hemiptera (bugs) (suction samples)		Total wasps (Hymenoptera) (suction samples)		Total spiders Suction samples)		Total spiders (leaf washes)	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean[1]	%[2]	Mean[1]	%[2]	Mean[1]	%[2]	Mean[1]	%[2]	Mean[1]	%[2]
Cruiser		0.023*	-70.65	0.051*	-53.90	0.043	98.75	0.518	-12.63	0.893	-11.58	0.119	-27.52
Cruiser Extreme		0.026*	-66.22	0.055*	-50.03	0.011	-49.34	0.497	-17.08	0.835	-19.98	0.096*	-42.12
Genero		0.041*	-46.57	0.061	-44.54	0.030	38.05	0.515	-13.34	0.786	-26.78	0.098*	-41.34
Thimet		0.095	29.06	0.149	42.25	0.037	71.43	0.511	-14.16	0.855	-17.20	0.094*	-43.71
Control	---	0.075	0.00	0.107	0.00	0.022	0.00	0.575	0.00	0.968	0.00	0.161	0.00
P		<0.001		<0.001		0.322		0.646		0.162		0.046	
LSD (p = 0.05)		0.032		0.047		ns		ns		ns		0.042	
df		(4, 79 suction samples, 4, 72 leaf washes)											

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

ACRI Experiment 3 (2015/16)

Sampling began slightly later than usual (10/11/15) and continued weekly until the 03/12/15. The insecticide treatments had no significant effect on plant stand or dry weight, reflecting low pressure from wireworm and modest thrips abundance (Table 18).

Table 18: Effect of treatments on plant stand and plant dry weight, ACRI Field 1, 2015/16.

Treatment	Plant Stand 1	Plant Stand 2	Average plant dry weight (g)
Control	12.4	11.2	1.48
Cruiser X	12.1	11.9	1.61
Cruiser X + Thiodicarb	11.0	12.0	1.75
Cruiser X + Thiodicarb + Fipronil	13.7	12.6	1.71
Thimet	12.1	12.6	1.84
P	0.15	0.63	0.299
LSD (p=0.05)	n.s.	n.s.	n.s.
df	(4, 39)	(4, 39)	(4, 39)

The abundance of adult thrips was not affected by the treatments (Table 19).

Table 19: Effect of treatments on overall thrips abundance per plant, ACRI Field 1, 2015-16.

Treatment	**Adults/plant**	**Nymphs/plant**	**Tubulifera/plant**	**Total Thrips/plant**
Control	0.800	4.00^{ab}	0.0219 ^{b}	4.82^{a}
Cruiser X	0.931	5.26^{a}	0 ^{a}	6.19^{a}
Cruiser X + Thiodicarb	0.850	3.91^{b}	0 ^{a}	4.76^{a}
Cruiser X + Thiodicarb + Fipronil	0.756	4.67^{ab}	0.0062 ^{a}	5.44^{a}
Thimet	0.666	2.47^{c}	0 ^{a}	3.14^{b}
P	0.471	<0.001	<0.001	<0.001
LSD (p=0.05)	n.s.	1.280	0.0141	1.459
df	(4, 79)			

Figures are mean insect number per plant from plant washes

The abundance of larvae showed a significant treatment by date interaction, but only varied significantly between treatments on the first date (Table 20). On that date Thimet significantly reduced the abundance of thrips larvae compared with the untreated control. The species composition showed that in the untreated control *Thrips tabaci* (onion thrips) was the predominant species (>70%) which is similar to most years, though often the proportion is closer to 90% (Table 21). *Frankliniella occidentalis* (western flower thrips) was also present (16%) as well as *F. schultzei* (Tomato thrips) (9%). The proportion of WFT trended toward being higher in the treated plots (p = 0.1). This possibly reflects tolerance of WFT to many insecticides, so it survives in treated plots while other thrips species are suppressed. Maturity harvests indicated no effect of thrips on boll number or crop maturity date (60% bolls open) or yield (Table 22).

Table 20: Abundance of thrips larvae on each treatment for each sample date, ACRI Field 1, 2015/16.

Treatment * Date	10/11/15	17/11/15	24/11/15	03/12/15
Control	8.5[ab]	2.9 [a]	1.2[a]	3.3[a]
Cruiser X	11.1[a]	3.9[a]	1.4[a]	4.6[a]
Cruiser X + Thiodicarb	6.9[b]	3.0[a]	0.7[a]	4.9[a]
Cruiser X + Thiodicarb + Fipronil	8.5[ab]	4.1[a]	1.0[a]	5.0[a]
Thimet	3.7[c]	3.3[a]	0.7[a]	2.1[b]
P	0.001			
LSD (p=0.05)	2.6			
df	(12, 79)			

NB: letters of significance are applied within dates, not across dates

Table 21: Thrips species composition for different treatments, ACRI Field 1, 2015/16.

Treatment	% Fr*ankliniella occidentalis*	% *Frankliniella schultzei*	% *Thrips tabaci*	% *Thrips imaginis*
Control	16.6	8.6	73.8	0.9
Cruiser X	22.3	7.9	69.8	0
Cruiser X + Thiodicarb	33.1	9.6	57.3	0
Cruiser X + Thiodicarb + Fipronil	30.0	2.8	67.2	0
Thimet	33.5	5.6	59.9	1.1
P[1]	0.10	0.56	0.22	0.35
df	(4, 79)			

[1] Based on analysis of Arcsin transformed data

Table 22: Boll counts, yield and maturity date for the insecticide treatments, ACRI Field 1, 2015/16

Treatment	Bolls/m	Yield Bales/ha	60% Open Bolls DAS
Control	109.5	10.93	150.7
Cruiser X	119.2	11.22	151.5
Cruiser X + Thiodicarb	112.2	10.96	151.0
Cruiser X + Thiodicarb + Fipronil	115.0	10.61	150.3
Thimet	109.8	10.63	154.9
P	0.97	0.93	0.23
LSD (p=0.05)	n.s.	n.s.	n.s.
df	(4,19)		

Impact of treatments on beneficial species

Overall the seed treatments and Thimet had low effects on beneficial species (Table 23, details in Appendix 5b). The only beneficial group that was consistently negatively affected was the 'other predatory beetles' which was significantly lower in the Cruiser Extreme plus thiodicarb and Cruiser Extreme plus thiodicarb and fipronil treatments. Interestingly, spider abundance was higher than the untreated control in the three seed treatments.

Discussion

Overall low thrips abundance precluded strong conclusions about the efficacy of the seed treatments used in the study. On nymphs, Thimet was more effective than seed treatments for

the first two weeks, significantly reducing them. Though not significantly different from the control, Cruiser X + Thiodicarb was better than Cruiser X in supressing nymphs on the first date. This did not translate to differences in plant dry weight which likely reflects low levels of damage. Insecticide treatments and 'in-plant-furrow' insecticides generally have a low effect on beneficial species but it is worthwhile to assess this further.

Table 23: Summary of mean abundance of key predatory or parasitic groups in each insecticide seed treatment, ACRI, 2015/16.

	Rate	Total predatory beetles (suction samples)		Other predatory beetles (suction samples)		Total predatory Hemiptera (bugs) (suction samples)		Total wasps (Hymenoptera) (suction samples)		Total spiders Suction samples)		Jassids (suction samples)	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean[1]	%[2]	Mean[1]	%[2]	Mean[1]	%[2]	Mean[1]	%[2]	Mean[1]	%[2]
Cruiser Extreme		0.33	28.0	0.016	-62.9	0.033	314.4	0.49	-5.5	1.30*	23.5	2.13*	-34.8
CruiserX+Thiodicarb		0.35	34.6	0.004*	-90.6	0.008	3.1	0.48	-7.8	1.27*	20.3	2.01*	-38.5
CruiserX+Thiodicarb+ Fipronil		0.39	48.9	0.004*	-90.6	0.023	194.2	0.59	14.8	1.52*	44.1	2.47*	-24.3
Thimet		0.19	-27.2	0.020	-53.4	0.000	-100.0	0.74	43.0	1.05	-0.9	1.70*	-48.1
Control	---	0.26	0.0	0.043	0.0	0.008	0.0	0.52	0.0	1.06	0.0	3.27	0.0
P		0.16		0.03		0.08		0.34		0.044		<0.001	
LSD (p = 0.05)		ns		0.027		ns		ns		0.15		0.20	
df		(4, 79 suction samples, 4,72 leaf washes)											

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

ACRI Experiment 4 (2016/17)

Plant stand & Plant dry weight

Plants were counted on two dates after emergence and averaged between 21 and 22 plants per meter for all treatments except Cruiser X, where plant stand was significantly lower at 17.5 plants per m. By the second check emergence in Cruiser X plots had caught up and there were no significant differences between treatments (Table 24). There were no significant differences in plant damage between treatments as indicated by plant dry weights, suggesting that thrips pressure per plant may have been relatively low during seedling establishment.

Table 24: Effect of seed treatments on plant stand and dry weight, ACRI 2016/17

Treatment	Plant Stand 1 08/11/16	Plant Stand 2 15/11/16	Average plant dry weight (g)
Control	21.62[a]	21.12	0.36
Cruiser X	17.50[bc]	20.50	0.37
Cruiser X+ Thiodicarb + Fipronil	20.12 [a]	18.88	0.38
Imidacloprid+Thio+Fip	20.75 [ac]	20.12	0.38
Thimet	21.25 [ac]	18.15	0.37
P	0.038	0.689	0.608
LSD (p=0.05)	2.765	n.s.	n.s.
df	(4, 39)	(4, 39)	(4, 39)

Thrips

None of the seed treatments affected adult abundance but immatures and total thrips numbers were impacted significantly (Tables 25 & 26); the effect was interactive between treatment and date for both. All seed treatments decreased thrips numbers equally well at two weeks after emergence. Thrips number in the experiment increased overall during the third week but Thimet and the A1 treatment (Imidacloprid + Thiodicarb + Fipronil) remained effective. Overall thrips abundance fell slightly during the fourth week after emergence though Cruiser X maintained significantly higher numbers. By weeks 5 and 6 thrips abundance, which was relatively low throughout the establishment period, declined significantly in all treatments and any differences due to seed treatment disappeared. During the first three weeks after emergence, Thimet and A1 treatments had the longest lasting efficacy.

Table 25: Effect of seed treatments on abundance of thrips larvae/plant in each treatment for each sample date, ACRI 2016/17

Treatment * Date	08/11/16	15/11/16	22/11/16	29/11/16	07/12/16	14/12/16
Control	0.063	3.013[a]	3.225[a]	1.388[a]	0.625	0.250
Cruiser X	0.000	0.400[b]	3.825[a]	3.688[b]	0.825	0.250
CruiserX+Thiodicarb+ Fipronil	0.000	0.287 [b]	3.962[a]	2.275[a]	0.950	0.200
Imidacloprid+Thio+Fip	0.013	0.200 [b]	2.150[b]	2.150[a]	0.500	0.150
Thimet	0.038	0.538 [b]	1.413[b]	1.350[a]	0.675	0.250
P			0.002			
LSD (p=0.05)			1.339			
df			(4, 87)			

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Table 26: Effect of seed treatments on abundance of total thrips numbers/plant in each treatment for each sample date, ACRI 2016/17

Treatment * Date	08/11/16	15/11/16	22/11/16	29/11/16	07/12/16	14/12/16
Control	0.56	6.15[a]	4.50[a]	1.62[a]	1.30	1.00
Cruiser X	0.25	3.24[b]	5.41[a]	4.35[b]	1.77	0.80
CruiserX+Thiodicarb+ Fipronil	0.24	2.62 [b]	5.72[a]	3.09[b]	2.00	1.05
Imidacloprid+Thio+Fip	0.29	2.85 [b]	3.56[b]	2.72[a]	1.10	1.10
Thimet	0.20	2.52 [b]	2.79[b]	1.86[a]	1.57	1.55
P			0.003			
LSD (p=0.05)			1.702			
df			(4, 87)			

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Thrips ID

The main species of thrips identified were *Frankliniella occidentalis* and *Thrips tabaci*. There were no significant differences in species composition between the different seed treatments (Table 27). There were, however interactive effects (data not shown) that related to seasonal changes in thrips species composition where the proportion of *F occidentalis* increased during the hotter part of the season while *T. tabaci* abundance declined.

Table 27: Thrips species composition for different seed treatments, ACRI 2016/17.

Treatment	% F*rankliniella occidentalis*	% *Frankliniella schultzei*	% *Thrips tabaci*	% *Thrips imaginis*
Control	19.52	0	80.48	0
Cruiser X	9.95	0	90.05	0
Cruiser X+ Thiodicarb + Fipronil	13.18	0	86.82	0
Imidacloprid+Thio+Fip	17.31	0	82.69	0
Thimet	14.95	0	85.05	0
P	0.154		0.154	
LSD (p=0.05)	n.s.	n.s.	n.s.	n.s.
df		(4, 86)		

Impact of treatments on beneficial species

In D-vac samples, the Cruiser X, Cruiser X + Thiodicarb + Fipronil and A1 treatments significantly reduced ants (high – very high). There were interactive effects on wasps where the Cruiser X + Thiodicarb + Fipronil, Thimet and the A1 treatments significantly reduced *Telenomus* abundance during the fourth week of the experiment (Appendix 5c, Table 25). Cruiser X reduced tangleweb spider abundance by 62% (Appendix 5c, Table 26). All seed treatments appear to be effective against early sucking pests in general but the major contribution to this came from high effects on immature jassids and mirids (Appendix 5c, Table 27). Cruiser X + Thiodicarb + Fipronil had significantly higher mite numbers than the control and may contribute to mite flaring if Fipronil is sprayed against mirids as seedlings grow.

Yield

Yield differences between treatments were not significant and yields were relatively low, averaging 9 bales/ha (Table 28). There were no treatment effects on the number of bolls per metre or on boll weight. The maximum maturity delay of 4 days was seen in the Thimet treatment thought his effect was not significantly different from the control or other treatments.

Table 28: Boll counts, yield and boll weight for different seed treatments, ACRI 2016/17

Treatment	Bolls/m	Boll Wt (g/boll)	Yield (bales/ha)	60%OBollDAS
Control	104.75	4.83	9.12	163.51
Cruiser Extreme	98	4.65	8.47	164.73
Cruiser X + Thio + Fip	104.25	5.34	9.28	165.95
Imida& Thio & Fip	109	6.45	9.61	161.82
Thimet	106.375	4.86	9.40	167.67
P	0.621	0.18	0.31	0.13
LSD (p= 0.05)	n.s.	n.s.	n.s.	n.s.
df	(4, 19)			

Discussion

In 2016/17, plant stand in the Cruiser X treatment was lower than in other treatments but this effect disappeared by the second week. Plants did not show dry weight differences indicating that damage was low. Thrips pressure was low but there were significant effects of seed treatments on nymphs for the first 3 weeks of the experiment. Yield and maturity were not affected. Seed treatment with fipronil controlled jassids and mirids but also reduced ants, Telenomus and flared mites.

Conclusions

This series of treatments aimed to evaluate the effectiveness against seedling thrips of the neonicotinoid seed treatment thiamethoxam (Cruiser) at single and double rates. Later treatments included stacks of Cruiser with another neonicotinoid (imidacloprid), another systemic (fipronil) and a carbamate (thiodicarb). Control treatments were Untreated and Thimet. The first experiment also included alternative treatments such as Sero X (*Clitoria ternatea* extract), Fungus (*Metarhizium anisopliae*) and Blood and Bone meal.

Generally thrips numbers early in the season were low. In the first two experiments there was no significant damage from thrips and no yield effects whether yields were low (9-10 bales/ha) or high (14-15 bales/ha). In 2013/14 Cruiser and Cruiser X treatments matured 7 days earlier than the control though the reason was unlikely to be related to thrips damage. In 2013/14, there were no maturity effects from treatments. Blood and bone meal, Fungus and Sero X showed no effects on thrips or yields. *Frankliniella occidentalis* proportions tended to be higher in the Cruiser, Cruiser X and Thimet treatments indicating better control in *Thrips tabaci* compared to *F. occidentalis* which may be showing tolerance to insecticide treatments. Overall, *Thrips tabaci* is the dominating species in the region but weather conditions (relating to time in the season) may affect the relative species populations. Predatory Coleoptera were negatively affected by Cruiser, Cruiser X and Genero in 2014/15 though that effect was not as strong in 2015/16. Similarly, Cruiser X, Genero and Thimet strongly affected spiders in 2014/15 but in 2015/16 the treatments did not. All treatments in 2015/16 and 2016/17 significantly decreased jassid numbers. Other affected insects were the beneficial wasp *Telenomus* and ants. With regards to pests, all treatments reduced mirids and those containing fipronil also flared mites. This result is pertinent considering that fipronil, which is harmful to beneficials, is used to control mirids early to mid-season. Given the restrictions as to the number of permitted sprays, a seed treatment containing fipronil should also be counted and whitefly and mite numbers should be monitored carefully.

Seed treatments did not generally affect plant stand and emergence is more likely to be affected by planting depth and seed size. Wireworm damage was not assessed and judging by emergence in Control plots, it was not a problem. In 2015/16 overall thrips numbers were again too low to draw conclusions about treatment effects and yield and maturity were not significantly different from controls. In 2016/17 the combination treatments (Cruiser X, Thiodicarb, Fipronil, Imidacloprid) significantly reduced nymph numbers in the second week of the experiment and are likely to wear off between the 3rd and 4th week after planting. Total thrips numbers and plant damage during this time were also relatively low and effects on yield and maturity were not apparent. It has been difficult to draw meaningful conclusions about the effectiveness of the seed treatments under such conditions, given that cotton plants in this climate zone have a high chance of recovery. The results mostly reinforce the early season thrips work of Lewis Wilson which found that early season thrips damage in the Namoi has inconsequential effects on yield in 9 out of 10 years as long as plants typically have the climatic conditions and resources to compensate for the damage. While there is no reported resistance of *F. occidentalis* to neonicotinoid seed treatments in Australia to date, there have been reports from China of low levels of tolerance and the industry needs to carefully manage the treatments currently available. This could include tolerating some aesthetic damage in warm season areas and leaving seed treatments for the cool season areas where the risk of damage and yield loss is higher.

iv) GVB Damage Experiment

In response to industry questions about GVB damage to cotton bolls we investigated the relationship between boll age and susceptibility to GBV damage. GVB adults and nymphs damage bolls and leave visible marks on the outer boll husk and cause wart-like growths on the inner boll husk, as well as lint stains and tight locks, similar to mirid damage. Young bolls (7-10 d.o.) may be shed while older bolls (>15 d.o.) tend to be less prone to damage and hard bolls (>20 d.o.) generally do not sustain significant damage. Damage was assessed on bolls in the field and we documented the visible symptoms of feeding damage. We began the experiment in 2013/14 and followed up with another one in 2014/15, however, during the latter experiment, most of the young bolls which had been caged were aborted after a storm, therefore we only report data from 2013/14.

Methods

Experiment 1 (2013/14)

During flowering, we caged 120+ open flowers individually (10 replications) to be able to assess boll age correctly. Flowers were then exposed to either a GVB nymph (3/4th instar), male, female or no GVB (Control) at 5 days old, 10 days old or 30 days old. GVB were removed after 1 week and all bolls remained caged for the duration of the season. Open bolls were harvested and scored for damage (tightlock, stain, sooty mould, dead) and then assessed for dry weight. Photos were taken to illustrate the types of damage.

Results and Discussion

Experiment 1 (2013/14)

Dry Weights

Boll dry weight was significantly affected by both boll age at the time of damage ($P = 0.002$) and the GVB stage that inflicted the damage ($P < 0.001$). The mean dry weight for 30 day old bolls was 5.43 g/boll compared to 3.60 g/boll for 5 day old bolls and 3.95 g/boll for 10 day old bolls (Table 29). This reflects less damage on older bolls and significant damage on younger bolls.

Nymphs inflicted most damage reducing mean boll dry weight to 3.06 g/boll compared to 5.80 g/control boll. Females also caused significant damage, reducing boll weight to 3.86 g/boll. This was not significantly different from nymph or male damage. Males did not cause significant damage compared to controls (4.58 g/boll) but the damage was significantly less compared to damage from nymphs. In laboratory colonies of GVB, nymphs and adults feed on fresh green beans. Beans taken out of nymph colonies, are considerably drier and have more feeding damage than those taken out of adult colonies, indicating that nymphs either feed more frequently, more voraciously or both.

The interaction of boll age and GVB stage was not significant (P = 0.066, Table 29), however, the means indicate the trend that the younger the bolls, the more feeding damage they sustain and that nymphs cause more damage than females which cause more damage than males, especially in younger bolls.

Table 29: Mean boll dry weights for bolls of different ages damaged by GVB

Boll Age	**5 days**	**10 days**	**30 days**
Mean Boll Dry Weight (g)	3.60[a]	3.95[a]	5.43[b]
P	0.002		
LSD (p = 0.05)	1.050		
df	(2, 119)		

GVB	**Control**	**Male**	**Female**	**Nymph**
Mean Boll Dry Weight (g)	5.80[a]	4.58[ab]	3.86[bc]	3.06[c]
P	<0.001			
LSD (p = 0.05)	1.212			
df	(3, 119)			

Boll Age * GVB (mean boll dry weight) (g)	**Control**	**Female**	**Male**	**Nymph**
5 days	6.49	2.21	4.29	1.39
10 days	4.92	4.1	4.29	2.58
30 days	5.99	5.36	5.16	5.22
P	0.066			
LSD (p = 0.05)	n.s.			
df	(6, 119)			

Damage Scores

Damage scores were assessed after harvest (Table 30). The scores reflect the number of bolls possessing a particular type of damage (tight locks, staining of lint, presence of sooty mould and dead bolls) out of the total number of bolls per treatment (e.g. 0.2 = 2 bolls out of 10 were damaged) rather than giving a range of scores to each particular damage type (though this can still be done). Lowest boll recovery rates occurred in 5 day old bolls, 40% for those fed on by females and 50% for those fed on by nymphs. All other treatments had recovery rates of 80% or greater. All 30 day old bolls had 100% recovery. Bolls were assessed for the various types of damage (Fig. 49) and these scores were totalled to give an overall damage score.

The highest amount of tight locking (≥ 50%) occurred in 10 day old bolls fed upon by GVB. Control and 30 day old bolls had the least amounts of tight locks. No staining occurred on control bolls and 5 day old bolls fed on by nymphs, the latter presumably because most of those bolls aborted (60%). Sooty mould was least prevalent in Control bolls and 5 day old bolls, again for the same reason. Even though GVB were dead by the time that bolls were harvested, staining was likely incurred during feeding when bolls were closed and green and became visible upon boll opening. Feeding could also have introduced fungal organisms into the bolls

and fungi proliferated on physiological sugars upon boll opening. Of all recovered bolls, the lowest percentage of dead bolls (bolls that stopped growing and became necrotic) occurred in bolls not exposed to GVB until 30 days of age.

The total damage score shows that 10 day old bolls sustained the greatest amount of damage. This was followed by 30 day old bolls. This is in contrast to the dry weight data which identified 5 day old bolls as having the highest degree of damage, boll weight being a reflection of boll development. The total damage score comprises physical characteristics of the lint rather than lint quantity and several damages can occur in a single boll, for instance a boll may have 1 lock tight locked, with staining on the tips of locks and sooty mould overlaying most of the lint. In this regard it makes sense that the least damaged bolls (older bolls) would complete their development and be exposed to those forms of damage whereas the younger and mostly aborted or atrophied bolls would not sustain such damages.

Table 30: Ranked boll recovery, tight lock, stain, sooty mould, dead bolls and total score, GVB damage experiment 2013/14

GVB Stage	Boll Age	Bolls recovered	TightLock	Stained	Sooty Mould	Dead Bolls	Total Score
Control	30 DO	100%	0.4	0	0	0	0.4
Control	5 DO	100%	0.3	0	0.2	0	0.5
Nymph	30 DO	100%	0.4	0.2	0.4	0.1	1.1
Male	30 DO	100%	0.7	0.6	0.3	0	1.6
Female	30 DO	100%	0.4	0.8	0.4	0	1.6
Control	10 DO	90%	0.333	0	0.111	0.222	0.666
Male	5 DO	90%	0.555	0.444	0.111	0.222	1.332
Female	10 DO	90%	0.666	0.444	0.444	0.111	1.665
Nymph	10 DO	90%	0.666	0.666	0.444	0.111	1.887
Male	10 DO	80%	0.875	0.5	0.25	0.125	1.75
Nymph	5 DO	50%	0.2	0	0	0.6	0.8
Female	5 DO	40%	0.5	0.5	0	0	1
Nymph	10 DO	90%	0.666	0.666	0.444	0.111	1.887
Male	10 DO	80%	0.875	0.5	0.25	0.125	1.75
Female	10 DO	90%	0.666	0.444	0.444	0.111	1.665
Female	30 DO	100%	0.4	0.8	0.4	0	1.6
Male	30 DO	100%	0.7	0.6	0.3	0	1.6
Male	5 DO	90%	0.555	0.444	0.111	0.222	1.332
Nymph	30 DO	100%	0.4	0.2	0.4	0.1	1.1
Female	5 DO	40%	0.5	0.5	0	0	1
Nymph	5 DO	50%	0.2	0	0	0.6	0.8
Control	10 DO	90%	0.333	0	0.111	0.222	0.666
Control	5 DO	100%	0.3	0	0.2	0	0.5
Control	30 DO	100%	0.4	0	0	0	0.4
Male	10 DO	80%	0.875	0.5	0.25	0.125	1.75
Male	30 DO	100%	0.7	0.6	0.3	0	1.6
Female	10 DO	90%	0.666	0.444	0.444	0.111	1.665
Nymph	10 DO	90%	0.666	0.666	0.444	0.111	1.887
Male	5 DO	90%	0.555	0.444	0.111	0.222	1.332
Female	5 DO	40%	0.5	0.5	0	0	1
Female	30 DO	100%	0.4	0.8	0.4	0	1.6
Nymph	30 DO	100%	0.4	0.2	0.4	0.1	1.1
Control	30 DO	100%	0.4	0	0	0	0.4
Control	10 DO	90%	0.333	0	0.111	0.222	0.666
Control	5 DO	100%	0.3	0	0.2	0	0.5
Nymph	5 DO	50%	0.2	0	0	0.6	0.8

Table 30 cont.

GVB Stage	Boll Age	Bolls recovered	TightLock	Stained	Sooty Mould	Dead Bolls	Total Score

Female	30 DO	100%	0.4	0.8	0.4	0	1.6
Nymph	10 DO	90%	0.666	0.666	0.444	0.111	1.887
Male	30 DO	100%	0.7	0.6	0.3	0	1.6
Male	10 DO	80%	0.875	0.5	0.25	0.125	1.75
Female	5 DO	40%	0.5	0.5	0	0	1
Male	5 DO	90%	0.555	0.444	0.111	0.222	1.332
Female	10 DO	90%	0.666	0.444	0.444	0.111	1.665
Nymph	30 DO	100%	0.4	0.2	0.4	0.1	1.1
Nymph	5 DO	50%	0.2	0	0	0.6	0.8
Control	5 DO	100%	0.3	0	0.2	0	0.5
Control	10 DO	90%	0.333	0	0.111	0.222	0.666
Control	30 DO	100%	0.4	0	0	0	0.4
Female	10 DO	90%	0.666	0.444	0.444	0.111	1.665
Nymph	10 DO	90%	0.666	0.666	0.444	0.111	1.887
Female	30 DO	100%	0.4	0.8	0.4	0	1.6
Nymph	30 DO	100%	0.4	0.2	0.4	0.1	1.1
Male	30 DO	100%	0.7	0.6	0.3	0	1.6
Male	10 DO	80%	0.875	0.5	0.25	0.125	1.75
Control	5 DO	100%	0.3	0	0.2	0	0.5
Male	5 DO	90%	0.555	0.444	0.111	0.222	1.332
Control	10 DO	90%	0.333	0	0.111	0.222	0.666
Female	5 DO	40%	0.5	0.5	0	0	1
Nymph	5 DO	50%	0.2	0	0	0.6	0.8
Control	30 DO	100%	0.4	0	0	0	0.4
Nymph	5 DO	50%	0.2	0	0	0.6	0.8
Male	5 DO	90%	0.555	0.444	0.111	0.222	1.332
Control	10 DO	90%	0.333	0	0.111	0.222	0.666
Male	10 DO	80%	0.875	0.5	0.25	0.125	1.75
Female	10 DO	90%	0.666	0.444	0.444	0.111	1.665
Nymph	10 DO	90%	0.666	0.666	0.444	0.111	1.887
Nymph	30 DO	100%	0.4	0.2	0.4	0.1	1.1
Male	30 DO	100%	0.7	0.6	0.3	0	1.6
Female	5 DO	40%	0.5	0.5	0	0	1
Female	30 DO	100%	0.4	0.8	0.4	0	1.6
Control	5 DO	100%	0.3	0	0.2	0	0.5
Control	30 DO	100%	0.4	0	0	0	0.4


Nymph 5D
Nymph 5D
Nymph 10D
Nymph 10D
Nymph 30D
Nymph 30D
Female 5D
Female 5D
Female 10D
Female 10D
Female 30D
Female 30D
Male 5D
Male 5D
Male 10D
Male 10D
Male 30D
Male 30D

Fig. 49: Damage to bolls, GVB feeding damage experiment 2013/14. Photos depict the best and worst bolls of the treatment.

Conclusion

Dry weights showed that there was significant damage to younger bolls and less damage to older (10 and 30 days old) more developed bolls. These older bolls, however tended to display more staining, tightlocking and sooty mould since they were exposed to more damage opportunities during their development. Young damaged bolls (5 days old) were often aborted. Nymphs reduced boll dry weight significantly, followed by females, then males. These results have implications for GVB management since nymphs have a clustering habit, which means that they could cause significant damage in areas where they hatch. Adults, being more mobile, are likely to damage bolls in a wider area which make it less likely to find high numbers of damaged bolls unless GVB occur in high numbers.

(v) Improve understanding of insecticides used to manage whiteflies (Whitefly x Chemistry)

Whitefly management has become more difficult with the increasing resistance to pyriproxifen in some regions and the increasingly harsher management of early season pests such as thrips, cutworm, wireworm and mirids. Off-label use of pesticides at planting compounded by several applications of part-rate "soft" sprays are affecting the build-up of effective predator populations. While the cotton industry is very familiar with the modes of action and efficacies of Admiral (Pyriproxifen) and Pegasus (Diafenthiuron) for SLW control, other chemicals recommended for whitefly management in the CPMG are not used frequently. This is primarily due to lack of experience with the chemicals by both researchers and growers/consultants but price differences also play a part. This experiment aimed to better understand the activities of these other chemicals so that industry may have the full benefit of a wider range of IPM suitable products.

Methods

To be able to compare to commercial practices, we also had a standard early season mirid insecticide treatment (Regent (Fipronil)) to observe effects on beneficials and any related whitefly population increase. Ideally, insecticides were to be applied in optimum conditions, i.e. based on thresholds and pest developmental stages. We expected crop penetration of chemical to be an important issue later in the season. Plots for each treatment were managed

realistically, as they might be on a commercial property hence we monitored the crop closely from planting onwards. The experiment was laid out in a randomised block design with 9 treatments and 4 replications. Each of the 36 plots was 8 rows x 15 m and each row was sprayed as described in Section B (i).

The crop was monitored from emergence onwards for thrips and when tall enough for beat sheeting, 1 m was sampled weekly to estimate mirid numbers. The centre rows of each plot were infested with whiteflies in early January and each week, adult whiteflies were scored on fifteen Node 5 leaves from the centre rows of each plot while leaves from Node 8 were scored for 3rd & 4th instar nymphs. As whitefly populations built slowly, we sprayed all plots except Control plots with dimethoate in mid-January to reduce beneficial numbers. Mirids were also over threshold at that time and the Fipronil plots were sprayed. Paramite was applied on the 24th January 2018 as both two-spotted and strawberry spider mites were beginning to affect the experiment (Fig. 50). Insecticide treatments and rates are listed in Table 31 and were applied on the 14th of February despite whitefly numbers being below threshold as they were expected to crash.

Action thresholds were determined as:

- No thrips sprays as any plants in the region are expected to outgrow any damage.
- If mirids are over threshold, check for top damage and record and spray the Regent plots. Should mirids not exceed the action threshold we need to decide when to spray the Regent plots.
- Spray for whitefly in individual treatments when whitefly numbers/stages have reached the recommended action thresholds for each insecticide (see Cotton Pest Management Guide & Technical information).

At the end of the experiment, weekly maturity picks of 2 metres were collected from row 5 while row 4 was machine picked for yield assessment.

By the beginning of February we realised that we were struggling to reach threshold numbers of whiteflies and decided to apply the first spray based on the next count. To add value to the experiment we also decided to continue beat sheeting for mirids and to plant map 5 plants from each plot at the end of the season to assess whether there was a relationship between mirid numbers and boll numbers.

Figure 50: Strawberry (left) and two-spotted (right) spider mites and their effect on the experiment

Table 31: Insecticides applied to plots

Treatment	Application Rate	Adjuvant	Target Pest	Optimum application
1. Movento (Spirotetramat) **BAYER**	300 ml/ha	Hasten @ 200 ml/100L	SLW, Cotton Aphid	- use medium spray droplets - translaminar activity - best on nymphs at low populations - Max. 2 applications/season
2. Pegasus (Diafenthiuron) **SYNGENTA**	600 ml		Mites, Cotton Aphid, SLW	- 10-20% of leaves infested - not above 30°C - needs sun for vapour action - Max. 2 applications/season
3. Applaud (Buprofezin) **DOW**	1 L/ha		Mealybugs, Scale insects, GHWF, SLW	- Spray early nymph stages - Max. 2 applications 10 - 14 days apart
4. Exirel (Cyantraniliprole) **FMC**	600 ml/ha (Geoff recommends 800 ml/ha)	Hasten @ 500 ml/100L	SLW, Cotton Aphid, Helis	- Active on sucking and chewing pests (translaminar & local systemic) - May take 3-6 days for effect but feeding stops within a few hours - high activity on SLW eggs & early nymphs, med on adults, spray developing population (around 110 DAS or 1300 DD) -Tank mix (emulsion) needs to be agitated, don't leave standing, give a good stir before application - use medium spray droplets - Do not apply in heavy dew or imminent rainfall - Max. 2 applications 10 days apart
5. Starkle (Dinotefuran) **Agnova**	250 g/ha		SLW, Mirid	- Apply prior to canopy closure - Spray when threshold reached - Spray when crop is not water stressed - use medium spray droplets - Max. 2 applications at least 14 days apart
6. Mainman (Flonicamid) **ISK**	140 g/ha		Cotton Aphid, Mirid	- use medium spray droplets - Max. 2 applications/season
7. Biopest Oil **SACOA**	2 L/ha		Cotton Aphid	- Apply at low pest pressure - Acts as feeding /oviposition depressant -Contact poison - Do not apply to water stressed crop - Do not apply if temps are hot (above 35°) or in high humidity conditions - Max tractor speed 5 kph
8. Regent (Fipronil) **BASF**	62.5 ml/ha	Salt @ 10 g/L	Mirid, GVB, ADB, Thrips	- Apply at first sign of the pest - Takes 3-4 days for full effect - Use double the rate under heavy pest pressure - Max. XXX applications/season
9. Control				

Results & Discussion

Throughout the experiment whitefly numbers were very low and never reached threshold (Fig. 51) so we sprayed at below threshold levels in mid-February. Beneficial numbers were high (Fig. 52) despite a dimethoate spray to suppress them in mid-January. Effects of each single individual spray on beneficials can be found in Appendix 6. There was no significant effect of the one spray on yield or boll weight (Table 32), however, the Pegasus plots had significantly

higher boll numbers than the control. Since SLW numbers were very low and Pegasus is not registered for mirids, it is likely that Pegasus suppressed more mites. Analysis of mite numbers showed that 2-spotted mite numbers were lower in the Pegasus treatment but this was not statistically different (data not shown). Maturity was significantly slower in a number of treatments, but again, this was not related to SLW. Mite numbers in all these treatments were slightly lower than the control (n.s.), so they were also unlikely to be the reason. Figure 53 shows the relationship between cumulative mirid numbers and maturity and while there is a poor correlation, there appears to be a trend that maturity may be affected by mirid numbers.

As whitefly numbers were unlikely to increase after spraying we did not think that we would get good results from this experiment and decided to value-add by mapping 5 plants per plot at the end of the season to assess whether there was a relationship between cumulative mirid populations and yield parameters. Mean mirid numbers from beat sheets at each sampling date are shown in Table 33 but ranged from 2-30 per plot over 9 weeks. Numbers built up during squaring and flowering, dipping after the dimethoate spray in mid-January and again at cut-out in mid-February. There was no discernible relationship between mirid numbers and yield or boll weight (Fig. 53).

Plant mapping showed that the relationship between cumulative mirid numbers and bolls missing from fruiting positions on fruiting branches was poorly defined (Fig. 54). This suggests that loss of fruits on plants is also affected by other factors, such as, e.g. climatic conditions, plant nutrition, irrigation, alternative food sources, etc. This was true for first, second and third position bolls, however, the percentage of loss was higher in first and second positions (Fig. 54). This pattern appears to be common as it was also seen when plant mapping the fruit damage experiments.

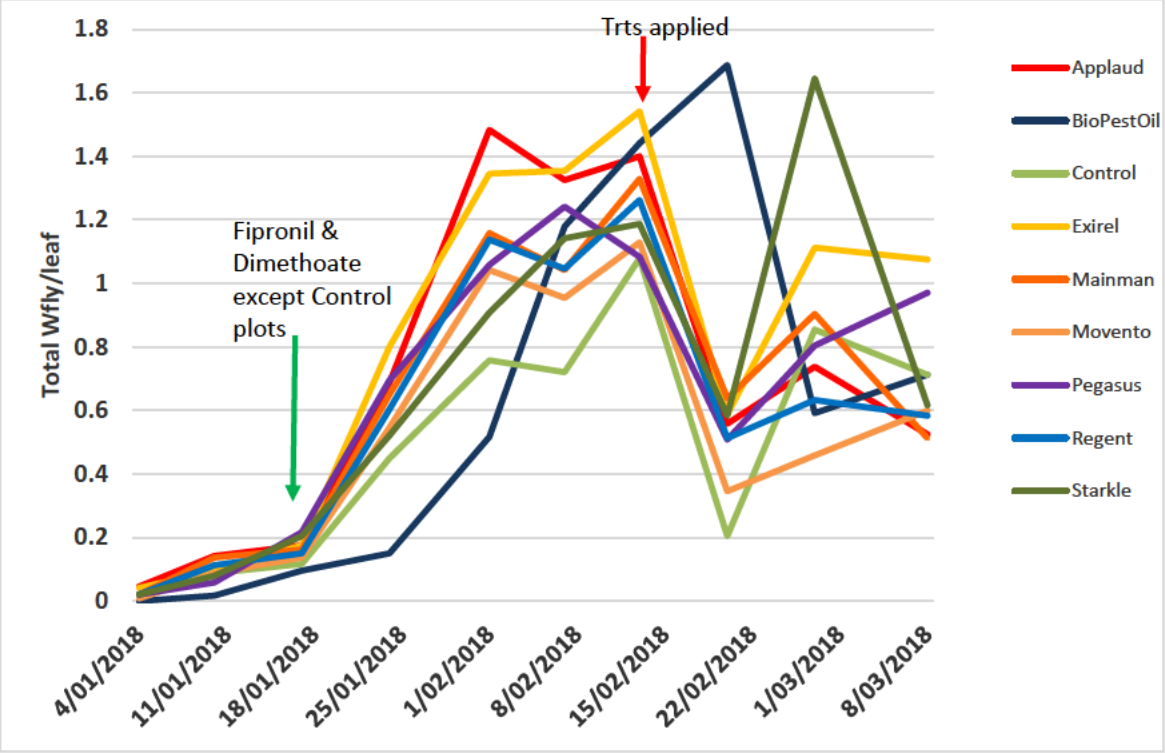


Figure 51: Total whitefly per leaf (adult & nymph counts), ACRI, 2017/18

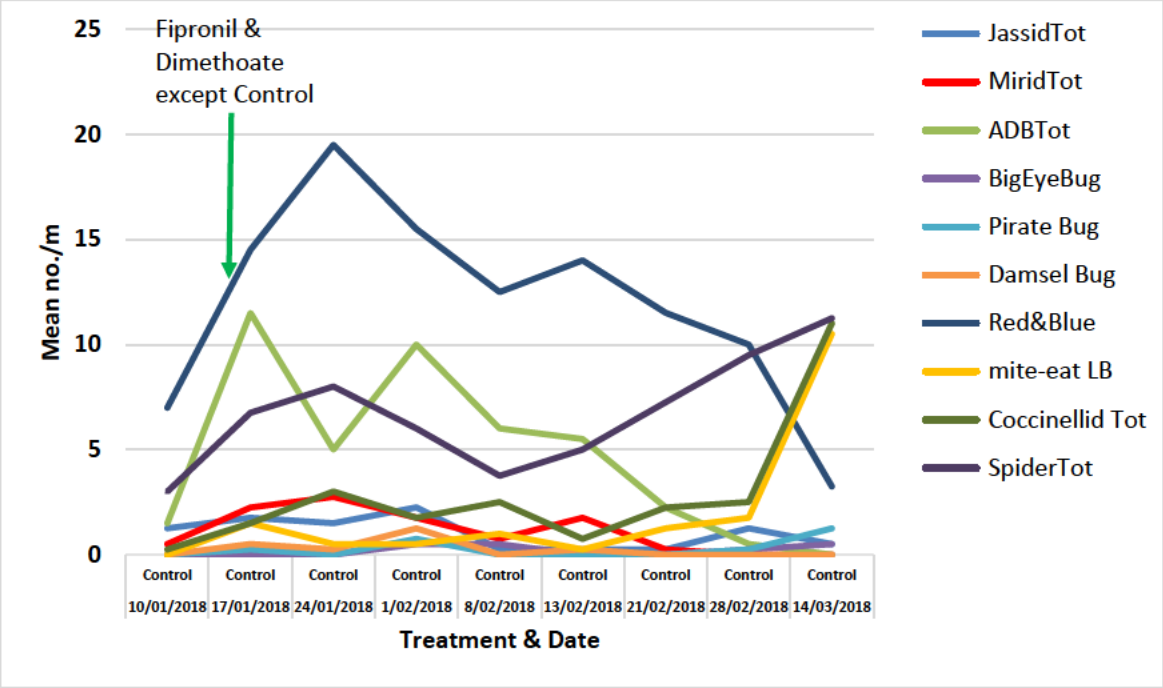


Figure 52: Control beneficials and mirids (mean number/m) from beat sheets, ACRI, 2017/18

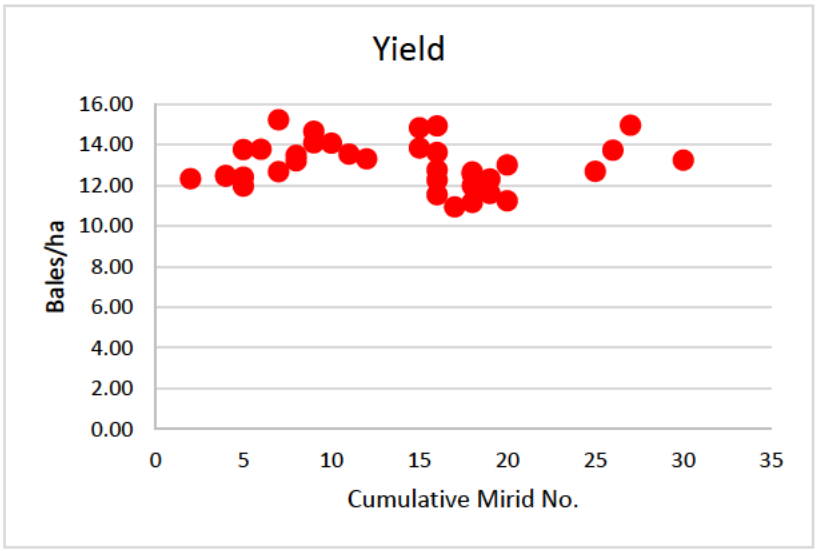
Table 32: Yield and maturity data Whitefly x Chemistry Experiment, ACRI, 2017/18

Treatment	Bolls/m	Boll_Wt (g/boll)	Yield (b/ha)	60% OBDAS
Control	138.38	4.31	13.55	147.49
Admiral/Lascar (Pyriproxifen)	130.75	4.38	12.59	148.66
Biopest Oil	132.88	4.48	12.76	150.69*
Exirel (Cyantraniliprole)	131.25	4.46	12.94	148.28
Mainman (Flonicamid)	133.75	4.35	12.34	150.23*
Movento (Spirotetramat)	131.75	4.42	12.83	147.84
Pegasus (Diafenthiuron)	154.50*	4.47	14.37	150.14*
Regent (Fipronil 1/2 rate + Salt)	138.50	4.64	13.39	151.44*
Starkle 20 SG (Dinotefuran)	128.38	4.67	12.51	150.45*
F (P=0.05)	0.042	0.592	0.109	0.033
LSD	14.62	n.s.	n.s.	2.558
df	(8, 35)			

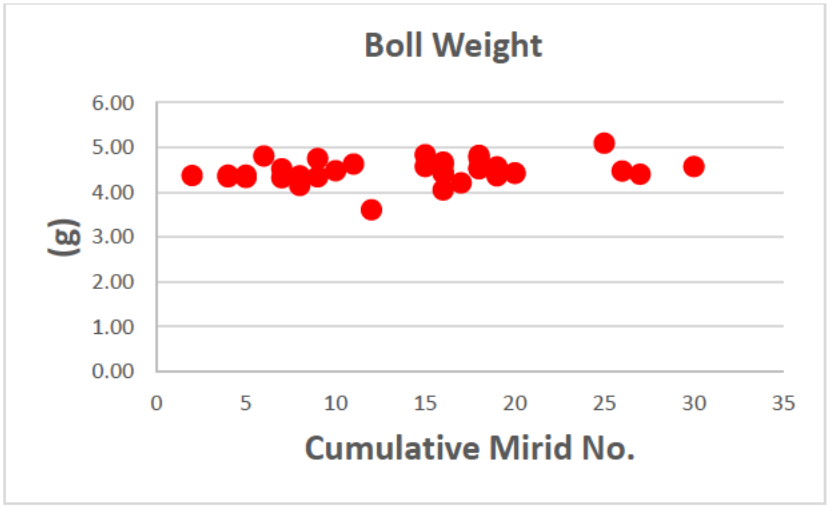
Table 33: Mean mirid numbers sampled, ACRI, 2017/18

Date	Mean Mirids/m
10/01/18	0.28
17/01/18	3.27
24/01/18	2.19
01/02/18	3.33
08/02/18	1.61
13/02/18	2.05
21/02/18	0.33
28/02/18	0.13
14/03/18	0

a)



b)



c)

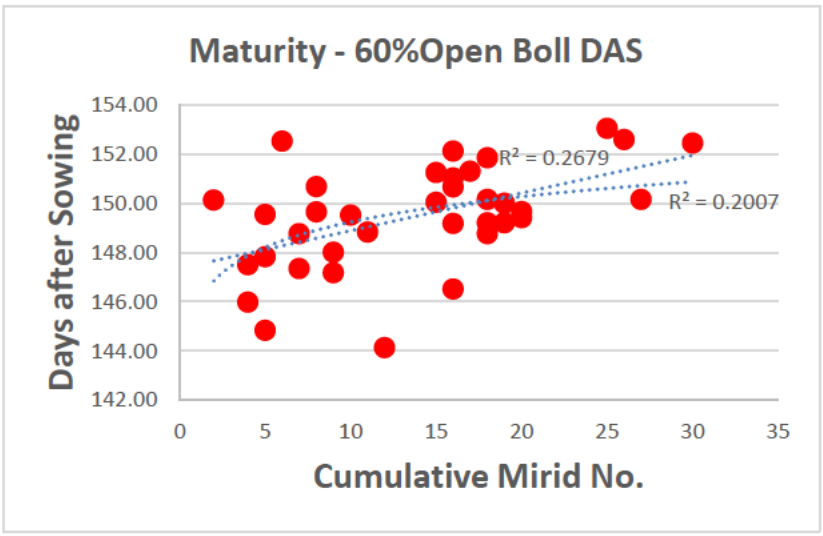
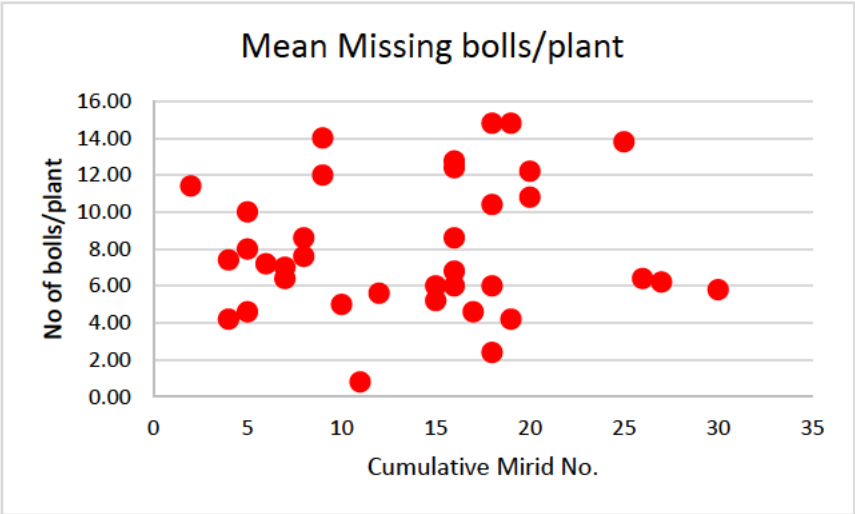


Figure 53: Effect of cumulative mirid number on a) yield, b) boll weight and c) maturity, ACRI, 2017/18

a)



b)

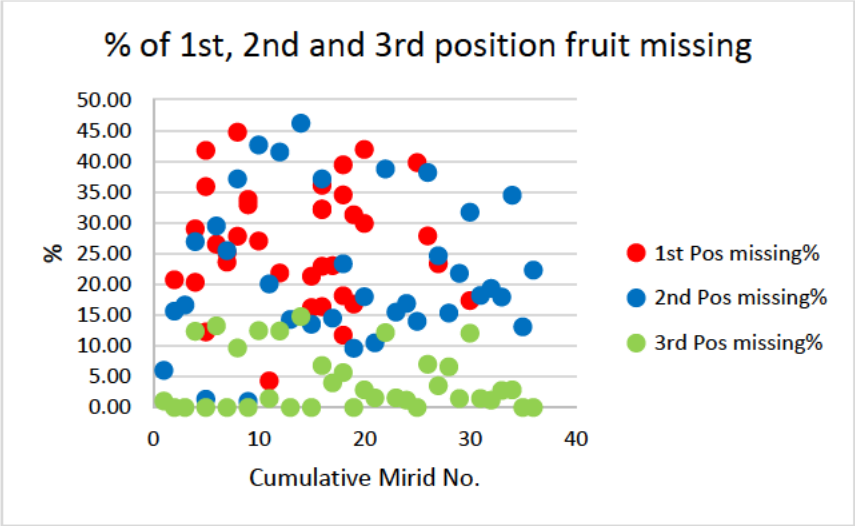


Figure 54: Effect of cumulative mirid number on a) missing bolls per plant and b) the percentage of missing 1st, 2nd and 3rd position bolls.

Conclusion

Low levels of whitefly populations hampered this experiment and we will attempt to repeat it as part of project CSP1905 during the 2018/19 season with better management of whitefly predators. This crop was hailed out before Christmas but area restrictions by Bayer prevented us from re-planting this experiment and it will be delayed until 2019/20. Early results indicating possible yield differences and relationships between mirids and yield parameters were possibly obscured by mite infestation which moved in from a neighbouring experiment. This demonstrated the difficulties in manipulating different insect populations in field experiments. The relationship between mirids and yield parameters needs to be investigated further and should we get an opportunity, we will attempt to collect incidental data from future experiments.

Section C. Management of early season damage – assessing seed treatments and measuring the effect of early season thrips and

mirid damage on plant growth, yield, maturity and fibre quality in southern regions

Cotton acreage in the Liverpool Plains area (Willow Tree, Quirindi, Spring Ridge, Pine Ridge) is gradually expanding. This area, south east of the Lower Namoi, is cooler and has a shorter growing season. This reduces the window for suitable planting conditions and increases the risk of early frosts. Given adequate soil moisture is available and there is opportunity for timely planting, cotton is an attractive crop for a number of reasons: managing cotton is now relatively straight-forward with the Bollgard II and Round-up Ready Flex system, and with reasonable cotton prices, gross margins are competitive with other crop options.

The shorter growing season in the Liverpool Plains means that growers and consultants believe that they cannot tolerate any seedling damage (leaf or terminal) or early fruit loss from thrips, mirids and other sucking pests, as this would lead to a significant delay in plant growth with a subsequent delay in fruit set. This could result in lower yield, cotton quality issues and a delay in harvest as bolls mature in cooler conditions. Far south NSW regions (Griffith, Hay) also have a shorter season but they have the advantage of longer day length which gives crops more time to compensate for earlier setbacks. Uncertainty around insect damage is encouraging foliar applications of insecticides to control thrips on seedling cotton or mirids during squaring and flowering, which is detrimental to survival of beneficial populations and can potentially increase risks from secondary pests. There is no information available for these areas and in order to provide better advice to southern cotton growers, we collaborated with CSD researchers Robert Eveleigh (Namoi/Liverpool Plains) and Jorian Millyard (Hay) evaluating the effects of thrips damage on early growth and consequences for yield, maturity and fibre quality (including novel seed treatment options and varieties as they become available).

In 2013-14 Jorian helped us to secure sites at Hay and Carathool, organised planting and implemented treatments. We also assisted Sandra McDougall (NSWDPI) in setting up early season thrips damage simulation experiments. Robert assisted us in locating and planting suitable sites in the Liverpool Plains: in 2015/16 at Willow Tree, in 2016/17 at Spring Ridge and Pine Ridge and in 2017/18 at Spring Ridge.

(i) Southern Seed Treatment Experiments

2013/14 Gravina and Daisy Lodge, Hay/Carathool

Two Experiments were set up at Hay ('Gravina') and Carathool ('Daisy Lodge'). These experiments aimed to evaluate the efficacy of the seed treatments to improve establishment and to measure if the protection against thrips in this short season area translated to a benefit in maturity or yield.

Methods

CSIRO assisted with the setup of experiments by demonstrating efficient thrips sampling and plant stand assessments on the first sample date, and the yield sampling from these sites at the end of the season. Jorian continued the thrips sampling and liaison with the growers at each site. The experiments used a replicated design with 4 replicates of each treatment. Treatments differed slightly at the two sites; at Gravina the treatments were Control, Cruiser, Cruiser Extreme (CruiserX), Genero and Lorsban, while at Daisy Lodge they were Control, Cruiser, Cruiser Extreme, Genero and Thimet.

Sampling in each experiment included weekly collection of plant or leaf samples from each plot, which were washed so that thrips and other insects could be counted and identified. Thrips adults and nymphs in each sample were counted and a sub sample of 20 adults (or less) was

taken for speciation into; Western flower thrips (*Frankliniella occidentalis*, F.o.), tobacco thrips (*Thrips tabaci*, T.t.), tomato thrips (*Frankliniella schultzei*, F.s.) or other species (mostly non-pest Tubulifera). Plants were retained after washing and dehydrated to assess dry weight.

Plant establishment and growth was also monitored to evaluate damage and assess how plants recovered. An initial plant establishment count was made once plants had emerged. Three sequential stand counts were then made at about 14 day intervals. Yield assessments were made at these site by hand harvesting three metres of cotton from each plot once crops were fully open.

Results & Discussion

Establishment Counts: first date only
These counts were made to assess potential differences in plants stand due to damage from wireworm (Table 34). At Daisy Lodge, analysis of the establishment counts showed a significant difference for the no. of plants/m. Cruiser treated strips had a significantly lower plant stand than the control, but at 9.6 plants per m this was still adequate and the difference between the highest (Genero 12.7) and lowest (Cruiser 9.6) was 3 plants per m. Plots with Thimet strips had a marginally higher plant stand than the Control. Plant stand at Gravina on the first date were overall low and not significantly different from each other. There was no significant difference between treatments at either location for the number of plants dead per metre or the % of plants showing wireworm damage.

Sequential establishment data for Gravina (28/10/13, 04/11/13 & 18/11/13)
At Gravina there were no significant differences in establishment between treatments for each date, significance for treatment (separate) or the treatment by date interaction (combined) analysis (Table 35), though there was a trend across all treatments for an increase in plant stand from 4.3 plants per m on the 28/10/11, to 8.3 plants per m on the 4/11/13 and finally to 7.3 plants per m on the 18/11/13 ($p < 0.001$, LSD = 0.69) which possibly reflects first further germination and then subsequent losses over this time.

Table 34: Plant emergence, established plants per m, dead plants per m and % of plants with wireworm damage (means), 2013/14

Treatment	Total Emergence	No. plant/m		No. dead/m		%Wireworm Damage	
	Daisy Lodge	Daisy Lodge	Gravina	Daisy Lodge	Gravina	Daisy Lodge	Gravina
Control	12.55 a	11.30 a	4.60	1.25	0	9.77	0
Cruiser	10.10 b	9.65 b	4.20	0.45	0	5.24	0
Cruiser X	12.10 a	11.40 a	5.10	0.70	0	5.77	0
Genero	12.70 a	11.7 5a	3.40	0.95	0	7.52	0
Lorsban	-	-	4.05	-	0	-	0
Thimet	13.20 a	12.70 a	-	0.5	-	4.29	-
df	5, 99	5, 99	5, 99	5, 99	5, 99	5, 99	5, 99
P	<0.001	0.001	0.252	0.074	-	0.247	-
LSD	1.297	1.394	n.s.	n.s.	n.s.	n.s.	n.s.

Table 35: Established plants per m for Gravina (means), 2013/14

	Date		
Treatment	**28/10/13**	**04/11/13**	**18/11/13**
Control	4.6	8.7	7.6
Cruiser	4.2	8.4	7.8
Cruiser Extreme	5.1	8.5	6.8
Genero	3.4	8.4	7.5
Lorsban	4.1	7.8	6.8
df	4, 299		
P	0.649		
LSD	n.s.		

Sequential establishment data for Daisy Lodge (28/10/13, 19/11/13)
For Daisy Lodge there were significant differences between treatments in the individual date analysis and for the two dates combined (Table 36). Overall, there was a slight but significant decline in plant establishment between the 28 Oct (11.4) and the 19 Nov (10.6) ($F_{2, 23} = 5.4$, $p = 0.02$). Thimet had a significantly higher plant establishment than the control, while Cruiser was significantly worse. In this experiment the differences were small, and probably reflect the interaction of seasonal conditions, disease and wireworm.

Table 36: Established plants per m for Daisy Lodge (means), 2013/14

	Date		
Treatment	**28/10/13**	**19/11/13**	**2 dates combined**
Control	11.3[b]	10.3 [a] (8.85%)	10.8 [b]
Cruiser	9.7 [a]	9.4 [a] (3.10%)	9.5 [a]
Cruiser Extreme	11.4 [b]	9.7 [a] (15.35%)	10.5 [ab]
Genero	11.8 [b]	10.8 [a] (9.10%)	11.3 [bc]
Thimet	12.7 [b]	12.7 [b] (0%)	12.7 [c]
df	4, 99	4, 99	4, 199
P	0.001	0.001	<0.001
LSD	1.40	1.66	1.1

Thrips Counts - Gravina
There were no significant differences in thrips abundance between the control and seed treatments at Gravina and no significant treatment by date interactions (Table 37). The majority of thrips were onion thrips.

Table 37: Effect of seed treatments on thrips numbers per plant at Gravina, 2013/14

TRT	**Thrips A/plant**	**Thrips I/plant**	**Thrips Total**	**Tubulifera /sample**	**%F.o.**	**%F.s**	**%T.t**	**% Unidentified**
Control	0.23	0.45	0.63	2.21	9.80	0	54.2	8.90
Cruiser	0.43	0.54	0.81	2.75	5.30	0	73.8	4.30
CruiserX	0.26	0.42	0.63	1.96	13.10	0	75.2	7.40
Genero	0.27	0.45	0.67	1.96	15.30	0	60.4	7.60
Lorsban	0.32	0.36	0.59	2.79	9.40	0	72.5	1.40
p	0.159	0.367	0.179	0.566	0.605	0	0.129	0.615
LSD (P=0.05)	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
df	4, 87							

There were significant date effects for the mean number of thrips per plant at Gravina. What is unusual is the build-up in thrips populations over dates, as usually thrips populations decline through mid-November (Table 38).

Table 38: Thrips abundance by date, Gravina, 2013/14

Date	Thrips A/plant	Thrips I/plant	Thrips Total/plant	Tubu-lifera/ sample	%F.o.	%F.s	%T.t	% Unidentified
28/10/13	0.06^{b}	0.05^{d}	0.03^{d}	0.15^{c}	20.00^{ab}	0	25.80^{c}	14.20
05/11/13	0.16^{b}	0.14^{d}	0.26^{c}	0.10^{c}	8.80^{abc}	0	73.30^{b}	7.80
12/11/13	0.14^{b}	1.3^{a}	1.43^{a}	0.20^{c}	5.80^{bc}	0	71.70^{b}	5.00
19/11/13	0.19^{b}	0.39^{c}	0.55^{b}	2.50^{b}	22.10^{a}	0	77.90^{ab}	0
26/11/13	0.13^{b}	0.23^{cd}	0.33^{bc}	1.40^{bc}	3.30^{c}	0	58.80^{bc}	7.90
03/12/13	1.14^{a}	0.60^{b}	1.39^{a}	9.65^{a}	3.50^{c}	0	95.80^{a}	0.60
p	<0.001	<0.001	<0.001	<0.001	0.025	0	<0.001	0.149
LSD (P=0.05)	0.185	0.194	0.214	1.474	14.23	n.s.	21.28	n.s.
df		5, 87						

Thrips Counts - Daisy Lodge

There were no significant treatment effects for the mean number of thrips per plant for Daisy Lodge nor any treatment by date interactions (Table 39). The majority of thrips were onion thrips.

Table 39: Effect of seed treatments on thrips numbers per plant at Daisy Lodge, 2013/14

Treatment	Thrips A/plant	Thrips I/plant	Thrips Total/ plant	Tubulifer a/sample	%F.o .	%F.s	%T.t	% Unidentified
Control	1.53	3.33	4.06	2.71	4.35	0	86.40	9.20
Cruiser	1.52	4.25	4.99	1.50	2.88	0	95.70	1.40
CruiserX	1.52	3.84	4.53	2.17	1.81	0	93.00	0.80
Genero	1.73	3.51	4.27	2.54	3.42	0	82.70	3.90
Thimet	1.84	3.93	4.65	3.00	6.35	0	89.80	3.60
p	0.571	0.577	0.598	0.119	0.652	0	0.329	0.076
LSD (P=0.05)	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
df	4, 87							

There were significant date effects for the mean number of thrips per plant at Daisy Lodge. Similar to Gravina, there was a build-up in thrips populations over dates, though usually thrips populations decline through mid-November (Table 40).

Table 40: Thrips abundance by date, Daisy Lodge, 2013/14

Date	Thrips A/plant	Thrips I/plant	Thrips Total/ plant	Tubu-lifera/ sample	%F.o.	%F.s	%T.t	% Unidentified
28/10/13	0.44[d]	0.49[d]	0.86[d]	0.20[c]	0[b]	0	85.80[b]	14.2[a]
05/11/13	0.46[d]	0.35[d]	0.72[d]	0.25[c]	13.68[a]	0	75.20[c]	6.10[b]
12/11/13	2.11[b]	2.01[c]	2.92[c]	0.45[c]	2.02[b]	0	97.70[a]	0.30[b]
19/11/13	1.12[c]	5.72[b]	6.52[b]	1.75[b]	3.87[b]	0	93.80[a b]	2.30[b]
26/11/13	1.19[c]	7.35[a]	8.29[a]	3.20[b]	1.25[b]	0	98.30[a]	0[b]
03/12/13	3.43[a]	6.71[ab]	7.70[ab]	7.45[a]	1.75[b]	0	98.30[a]	0[b]
F	<0.001	<0.001	<0.001	<0.001	0.001	0	<0.001	<0.001
LSD (P=0.05)	0.529	1.296	1.323	1.294	6.734	n.s.	10.07	6.92
df	5,87							

Dry Weights

There were no significant treatment or interaction (Treatment x Date) differences for Daisy Lodge or Gravina in the mean dry weight of samples (20 plants/sample) (Table 41).

Table 41: Effect of treatments on plant dry weight, 2013/14

Treatment	Daisy Lodge	Gravina
Control	8.70	5.24
Cruiser	7.92	5.41
CruiserX	7.68	4.83
Genero	9.13	4.92
Thimet (DL)/Lorsban (G)	8.74	4.66
p	0.161	0.416
LSD(P=0.05)	n.s.	n.s.
df	4, 87	

Yield

Subsamples of handpicks were ginned to calculate gin turnout. For Daisy Lodge mean ginout was 45.9% and for Gravina it was 47.0%. There were no differences in yield between treatments at Daisy Lodge. At Gravina, strips treated with Lorsban had significantly lower yield than the control (Table 42).

Table 42: Effect of treatments on yield (b/ha), 2013/14

Treatment	Daisy Lodge	Yield as % Control	Gravina	Yield as % Control
Control	11.56	100.00	10.86[a]	100.00
Cruiser	13.03	112.72	10.06[a]	92.63
Cruiser Extreme	11.83	102.33	9.92[a]	91.34
Genero	12.20	105.54	9.93[a]	91.44
Thimet	11.29	97.66	-	-
Lorsban	-	-	8.53[b]	78.55
p	0.211		0.021	
df	4, 44		4, 44	
LSD	n.s.		1.33	

Boll Weights

There were no treatment differences in mean boll weight (g/boll) for Daisy Lodge, but at Gravina strips treated with Cruiser Extreme, Genero or Lorsban had lower boll weights than the control (Table 43).

Table 43: Effect of treatments on boll weight (g), 2013/14

Treatment	Daisy Lodge	Gravina
Control	4.83	4.68[a]
Cruiser	4.56	4.58[a]
Cruiser Extreme	4.50	4.24[b]
Genero	4.47	4.21[b]
Thimet	4.30	-
Lorsban	-	4.00[c]
p	0.685	<0.001
df	4, 44	4, 44
LSD	n.s.	0.182

Discussion

Thrips numbers in both experiments were low and did not affect yield parameters. Hence the efficacy of seed treatments could not be determined with confidence. Establishment counts at Daisy Lodge were higher than at Gravina which was due to better seed bed preparation. Cotton at Daisy Lodge was planted into wheat stubble while the soil at Gravina was coarse, dried up quickly and set in a crust. However, Daisy Lodge also experienced a wireworm problem which was absent at Gravina. It is interesting to note that at both sites Thimet and Lorsban (Industry standards) had the lowest yields – marginal at Daisy Lodge (Thimet) and significant at Gravina (Lorsban).

2015/16 Berwicks, Willow Tree

In 2015/16 the first experiment in the Liverpool Plains was conducted at James Arnott's farm "Berwicks", about 50 km west of Willow Tree.

Methods

Soil moisture conditions at planting were marginal and the resulting plant stand was very poor and had strong competition from weeds. We began sampling shortly after emergence and continued for 4 sample dates. Plant/leaf sampling was carried out as described above. Plants/leaves were retained after washing, scored according to the scale below and dehydrated to assess dry weight. Treatments included Cruiser Extreme and Control (fungicide only). After the second sampling date the grower sprayed the rest of the field with dimethoate. We took the opportunity to sample four additional plots to assess if this spray affected thrips abundance or species complex. This was not a true replicated design as we simply allocated additional plots to the eastern side of the experiment but we still analysed the last 2 sample dates, including the dimethoate treatment to test for any differences. The crop performed poorly through the season and was not therefore not harvested as yield estimated would have been confounded.

Thrips Damage Scores

1	Undamaged leaves
2	2 true leaves slight crinkling but healthy
3	distinct crinkling, area reduced
4	at least 1 leaf severely crinkled
5	both leaves severely crinkled

Results

Plant stand - There was a slight difference in plant stand with the Cruiser Extreme treatment having a slightly higher plant stand than the control (Table 44) though leaf dry weights were lower than the control.

Table 44: Plant stand (plants/m), mean damage score per plant for cotyledons and true leaves, and plant dry weights (g), "Berwicks", Willow Tree, 2015/16.

TRT	Plant Stand	Coty	Leaf	Dry Weight
CruiserX	2.82*	0.16	1.88*	1.18
Control	2.18	0.13	2.54	1.15
P	0.005	0.306	<0.001	0.307
LSD (p=0.05)	0.432	n.s.	0.294	n.s.
df	(3, 47)			

Thrips counts - There was no significant effect of seed treatment on thrips abundance or on thrips species (Table 45 and 46). The damage score on leaves was also slightly lower for Cruiser Extreme treated plants, but there were no difference in plant dry weight.

Table 45: Abundance of adult, larval and total thrips per plant at "Berwicks", Willow Tree 2015/16.

TRT	Thrips Adults	Thrips Nymphs	Tubulifera	Total Thrips
CruiserX	5.38	3.25	0.00	8.63
Control	5.95	4.44	0.00	10.40
P	0.393	0.140	0.324	0.104
LSD (p=0.05)	n.s.	n.s.	n.s.	n.s.
df	(1, 47)			

Table 46: Thrips species complex (% of total) at "Berwicks", Willow Tree, 2015/16.

TRT	*F. occidentalis*	*F. schultzei*	*T. tabaci*
CruiserX	7.72	12.50	79.78
Control	4.67	9.45	85.88
P	0.170	0.677	0.403
LSD (p=0.05)	n.s.	n.s.	n.s.
df	(1, 47)		

In the last two sample dates that included dimethoate plots, both the Cruiser and Dimethoate treated plants had higher numbers of adult thrips than the controls, but nymph numbers were similar between treatments (Table 47). Interestingly there was a significant difference in the species complex where the proportion of *F. occidentalis* was higher and the proportion of *T. tabaci* lower in the dimethoate treated plots compared with the control (Table 48). This probably reflects the fact that the Cruiser Extreme treatment had ceased to offer any control and the dimethoate had selectively removed the dimethoate susceptible *T. tabaci*. Leaf scores for the dimethoate treatment were higher than for Cruiser X but equal to the Controls which probably means that the remainder of the field did not have Cruiser X as seed treatment, hence damage prior to the dimethoate spray would have been similar to controls. Total dry weights did not reflect this difference (Table 49).

Table 47: Abundance of adult, larval and total thrips per plant, including the dimethoate treatment for the last 2 sample dates at "Berwicks", Willow Tree 2015/16.

TRT	Thrips Adults	Thrips Nymphs	Tubulifera	Total Thrips
CruiserX	2.54*	5.12	0.00	7.66
Dimethoate	2.65*	6.74	0.00	9.39*
Control	1.68	4.31	0.01	6.00
P	0.002	0.139	0.382	0.038
LSD (p=0.05)	0.559	n.s	n.s.	2.551
df	(2, 35)			

Table 48: Thrips species complex (%), including the dimethoate treatment for the last 2 sample dates at Willow Tree 2015/16.

TRT	*F. occidentalis*	*F. schultzei*	*T. tabaci*	*T. imaginis*
CruiserX	6.85	25.00	68.15	0.00
Dimethoate	34.55*	0.00	23.78*	8.33
Control	5.59	18.90	75.50	0.00
P	0.009	0.098	<0.001	0.382
LSD (p=0.05)	19.810	n.s.	24.770	n.s.
df	(2, 35)			

Table 49: Mean damage score per plant and dry weight, including the dimethoate treatment for the last 2 sample dates at Willow Tree 2015/16.

TRT	Leaf	Dry Weight
CruiserX	1.85*	1.80
Dimethoate	2.41	1.83
Control	2.39	1.74
P	<0.001	0.252
LSD (p=0.05)	0.278	n.s.
df	2, 35	

Discussion

Experimental conditions at "Berwicks" were less than ideal and the experiment was most likely confounded by the poor plant stand at establishment and the high weed load. We decided to cut our losses and did not harvest this experiment but made sure to secure sites with better management for follow up experiments.

2016/17 Connamara, Pine Ridge & Dimby Plains, Spring Ridge

The next experiments in the Liverpool Plains were set up at more northerly locations: at 'Connamara' near Pine Ridge which is owned by Ian Carter and at 'Dimby Plains' near Spring Ridge which is owned by Dave and Gordon Brownhill.

At Connamara the treatments were Dynasty (Control), Cruiser Extreme and Thimet (@ 4 kg/ha). At Dimby Plains the grower did not have equipment to apply Thimet so the treatments were only Dynasty and Cruiser Extreme. Similar to previous experiments plants/leaves were sampled shortly after emergence as described above. Plant stand was also assessed after emergence and d-vac suction samples were taken at each site so that the non-target effects of the treatments could be determined. This was important to understand seed treatment compatibility with IPM and conservation of beneficial species.

Results

2016-17 Connamara

Plant stand & Plant dry weight

Plants were counted on two dates after emergence and on both dates, Cruiser X and Thimet had significantly higher plant stands (about 1 plant) than the Control (6 plants/m) (Table 50). Thrips damage was insignificant in all treatments as indicated by leaf areas and plant dry weights.

Table 50: Effect of seed treatments on plant stand and dry weight, Connamara 2016/17

Treatment	Plant Stand 1 21/11/16	Plant Stand 2 28/11/16	Leaf Area (cm^2)	Average plant dry weight (g)
Cruiser X	7.63^b	7.45^b	57.6	0.684
Thimet	7.70^b	7.68^b	59.9	0.672
Control	6.34^a	6.75^a	51.9	0.634
P	<0.001	0.011	0.341	0.707
LSD (p=0.05)	0.385	0.585	n.s.	n.s.
df	(2, 15)	(2, 15)	(2, 41)	(2, 41)

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Thrips

The Cruiser X treatment had significantly higher total thrips abundance compared to the Control and Thimet treatments (Table 51). Overall thrips numbers during establishment were relatively low (3-4/plant).

Table 51: Effect of seed treatments on abundance of total thrips numbers/plant in each treatment for each sample date, Connamara 2016/17

Treatment	Mean
Cruiser X	4.35[b]
Thimet	3.03[a]
Control	3.31[a]
P	0.036
LSD (p=0.005)	1.044
df	(2, 42)

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Thrips identification

The main species of thrips identified were *Frankliniella occidentalis* and *Thrips tabaci*. Treatment differences were highly significant, with *F. occidentalis* abundance significantly lower in the Cruiser X and Thimet treatments than in the Control, and conversely, abundance of *T. tabaci* significantly higher (Table 52). Thimet had the highest effect on *F. occidentalis* but the lowest effect on *T. tabaci*. There were highly significant interactive effects for both species which related to seasonal changes in species composition (Table 53).

Table 52: Thrips species composition for different seed treatments, Connamara 2016/17

Treatment	% F*rankliniella occidentalis*	% *Frankliniella schultzei*	% *Thrips tabaci*	% *Thrips imaginis*
Cruiser X	18.6[b]	0	81.4[b]	0
Thimet	8.7[c]	0	91.0[c]	0
Control	24.7[a]	0	74.3[a]	0
P	<0.001 (<0.001)		<0.001 (<0.001)	
LSD (p=0.05)	5.04	n.s.	5.30	n.s.
df	(2, 41); (2,8)			

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Effect on other species

Few effects on beneficial species were detected in D-vac samples. Cruiser X significantly decreased total ant abundance which was due to its effect on *Pheidole* sp. (Appendix 7, Table 1). Thimet also affected *Pheidole* spp. but to a slightly lesser extent. Both Thimet and Cruiser X significantly reduced abundance of red and blue beetles and total jassids, mainly by controlling immature jassid populations. Interestingly Rutherglen bug populations were significantly higher in the Cruiser X treatment (Appendix 7, Table 2).

Table 53: Thrips species composition for different seed treatments, Connamara 2016/17

Treatment	% F*rankliniella occidentalis*	% *Frankliniella schultzei*	% *Thrips tabaci*	% *Thrips imaginis*
21/11/16				
Cruiser X	7.0	0	93.0	0
Thimet	2.5	0	97.5	0
Control	7.6	0	92.4	0
28/11/16				
Cruiser X	17.4^{b}	0	82.6^{bc}	0
Thimet	1.1^{a}	0	98.6^{ac}	0
Control	3.6^{a}	0	96.4^{a}	0
05/1216				
Cruiser X	7.8	0	92.2^{a}	0
Thimet	0.0	0	100.00^{b}	0
Control	6.7	0	93.3^{a}	0
12/1216				
Cruiser X	10.1	0	89.9^{bc}	0
Thimet	12.1	0	87.9^{ac}	0
Control	10.5	0	84.5^{a}	0
19/12/16				
Cruiser X	50.6^{b}	0	49.4^{b}	0
Thimet	27.7^{c}	0	71.2^{c}	0
Control	95.2^{a}	0	4.8^{a}	0
P	<0.001		<0.001	
LSD (p=0.05)	5.04	n.s.	5.30	n.s.
df		(2, 41)		

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Yield

There were no significant effects of seed treatments on boll counts, boll weight, yield and maturity (Table 54). Yields average 8-9 bales/ha and maturity in the control was delayed by only 2 days.

Table 54: Boll counts, boll weight, yield and maturity for different seed treatments, Connamara 2016/17

TRT	Bolls/m	Boll Wt (g/boll)	Yield (b/ha)	60%OBollDAS
Cruiser X	127.00	4.88	8.42	164.81
Thimet	136.00	4.54	8.79	164.35
Control	132.75	4.64	9.60	166.66
F (P=0.05)	0.581	0.422	0.621	0.659
LSD	n.s.	n.s.	n.s.	n.s.
df	(2, 11)			

2016/17 Dimby Plains

Plant stand & Plant dry weight

There were no significant differences in plant stand between Control and Cruiser X treatment on either date with stands averaging 9.5 plants/m. Thrips damage, as assessed by leaf area and dry weight did not differ between treatments (Table 55).

Table 55: Effect of seed treatments on plant stand/m and dry weight/plant, Dimby Plains 2016/17

Treatment	Plant Stand 1 21/11/16	Plant Stand 2 28/11/16	Leaf Area (cm^2)	Average plant dry weight (g)
Cruiser X	9.42	9.50	111.2	1.263
Control	9.55	9.87	98.6	1.118
P	0.810	0.390	0.242	0.231
LSD (p=0.05)	n.s.	n.s.	n.s.	n.s.
df	(1, 15)	(1, 15)	(1, 27)	(1, 27)

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Thrips

The abundance of adults, immatures or total thrips was not affected by Cruiser X (Table 56).

Table 56: Effect of seed treatments on abundance of total thrips numbers/plant in each treatment for each sample date, Connamara 2016/17

Treatment	Mean
Cruiser X	6.37
Control	6.13
P	0.810
LSD (p=0.005)	n.s.
df	(2, 42)

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Thrips identification

The main species of thrips identified were *Frankliniella occidentalis* and *Thrips tabaci*, however, there was no significant treatment effect on species composition with proportions of *F.o.* to *T.t.* being approximately equal (Table 57). Date effects reflected the typical seasonal changes in species composition in mid-December (data not shown).

Table 57: Thrips species composition for different seed treatments, Dimby Plains 2016/17

Treatment	% *Frankliniella occidentalis*	% *Frankliniella schultzei*	% *Thrips tabaci*	% *Thrips imaginis*
Cruiser X	44.90	0	49.60	0
Control	44.70	0	49.10	0
P	0.969		0.564	
LSD (p=0.05)	n.s.	n.s.	n.s.	n.s.
df	(2, 42)			

NB: letters of significance are applied within dates, not across dates with reference to the Control treatment

Effect on other species

Cruiser X negatively affected the abundance of ants (Pheidole) and *Telenomus* sp. (Appendix 7, Table 3). Immature jassids were the main pest species controlled by Cruiser X (Appendix 7, Table 4)

Yield

There were no significant effects of seed treatments on boll counts, boll weight and maturity (Table 58). Cruiser X provided a significant 1.1 b/ha yield advantage compared to the control (9.16 b/ha).

Table 58: Boll counts, boll weight, yield and maturity for different seed treatments, Dimby Plains 2016/17

Trt	Bolls/m	Boll Wt (g/m)	Ginout %	Yield (b/ha)	60%OBollDAS
Cruiser X	146.25	4.60	42.99	10.27*	173.50
Control	127.25	5.41	43.01	9.16	174.50
F (P=0.05)	0.092	0.194	0.951	0.009	0.182
LSD	n.s.	n.s.	n.s.	0.584	n.s.
df	(1, 7)				

Discussion

Overall the results confirmed that the Cruiser and Cruiser Extreme treatments provided control of thrips, though this was difficult to show statistically at low thrips densities. Thimet generally provided slightly superior control to Cruiser X. At Dimby Plains Cruiser provided an extra bale per hectare in yield. For comparison, at Yarral – another CSD experimental site – the effect of the seed treatments and Thimet also carried through to slightly higher yield. At the southern experiment sites (Hay/Carathool) thrips numbers were increasing as seed treatments wore off though there was no effect on yield, most likely because the overall thrips abundance at the sites was low. At both sites Cruiser X mostly reduced the number of ants which predate pest species (such as jassids) that could have fed on treated seedlings and been poisoned. This is not surprising considering that thiamethoxam is also marketed as an ant bait. At Connamara Cruiser X and Thimet reduced Red and blue beetles while at Dimby Plains Cruiser X had a very high effect on the abundance of *Telenomus* wasps.

Conclusions

The issue of early season damage in southern areas remains somewhat unclear but most growers believe that they will suffer yield penalties if they do not control thrips or mirid damage. Higher thrips numbers and damage are often seen in the outer rows of fields located near harvested wheat fields. In our experiments, the highest number of thrips encountered were 10 thrips/plant at 'Berwicks' and we could not assess yield in that field; though at a plant stand of 2-3 plants/m, this crop had factors apart from insects limiting yield. The experiment at Yarral, mentioned above, experienced 20 thrips/plant indicating that such numbers have impact on yield. This was supported by other seed treatment experiments where <1 to 6 thrips/plant were too few to reduce yield, and which were reported to CSD in a separate report (not part of this project). To settle the questions about thrips damage in southern cotton growing areas, it may be worthwhile to find cotton seedlings close to a wheat field and sample at 20 m intervals into the field in order to capture gradients in thrips populations, implement control sprays in some sections and carry through to yield. From experiments here, we can project that under high thrips populations neonicotinoid seed treatments (and Thimet) would effectively control thrips on seedling cotton.

In the USA, thrips resistance to neonicotinoids has been reported for WFT and *Thrips tabaci* as neonicotinoids are used there in all crops. This is of concern for Australia since in dry springs thrips will at some stage pass through cotton, where they are likely to be exposed to neonicotinoids as seed treatments. Both thiamethoxam and imidacloprid are used in wheat against aphids; thiamethoxam as Cruiser Opti in combination with lambda-cyhalothrin, hence thrips can exposed to neonicotinoids all year round. There is anecdotal evidence that Cruiser has become less effective as a seed treatment in some areas. In experiments where we compared Cruiser (350 g/kg thiamethoxam) to Cruiser Extreme (600g/kg thiamethoxam) (CSD report),

Cruiser X always controlled thrips better than Cruiser though Cruiser was not always better than the Control. Increasing the rate of thiamethoxam is not a long term solution to potential resistance problems and it would be wise to reduce the use of neonicotinoid seed treatments in areas where seasonal thrips numbers tend to be low and cotton has the climatic opportunity to outgrow damage. In this way the life of neonicotinoids could be extended and regions where potential thrips damage risk is higher and control may be warranted, would benefit longer.

The impact of thiamethoxam and other seed treatments on beneficials overall is low, however, the impact on individual species such as Ants, *Telenomus* and Red and blue beetles can be very high and the importance of these species to the pest complex and ecosystem as a whole needs to be considered before using) seed treatments. Sublethal effects may also have impacts that cannot be captured with the methods we are using.

Section D. Cotton Bunchy Top Disease

i) Alternative host species (with Murray Sharman)

CBTv is still present in the environment, as affected plants can sporadically be found in cotton crops. However, IPM practices in combination with unfavourable host conditions have meant the abundance of host weed species has been very low and outbreaks of the vector species, Cotton Aphid (*Aphis gossypii*), have been rare. Hence CBT has in recent years had an insignificant effect on cotton and yield. We have consequently reduced effort against this milestone. In this project we have completed simple studies investigating the effect of seed treatments and aphid sprays on CBT transmission. We have also made a significant effort in identifying alternative hosts of CBT (in conjunction with Dr Murray Sharman, QDAFF) as this is important in understanding where CBT is in the environment and conditions likely to lead to higher disease risk. This work has been placed on hold temporarily as we concentrated on other projects (e.g. CSP1303) or priorities (e.g. evaluating SLW sampling strategies). To tidy up our CBTv work we had planned to complete processing of samples of potential CBTv hosts. These were generated by growing potential CBTv hosts in amongst CBTv affected cotton plants and deliberately infecting these plants with viriliferous aphids. Extraction of RNA from these hosts was attempted at ACRI but we encountered problems and despite several attempts, our extraction process failed to reliably isolate the virus from samples, as indicated by negative results with the positive controls. Dr Sharman picked this up in the first batch sent to him and suggested various modifications which failed to solve the problem.

We used our field CBT nursery to help generate conditions to test if potential hosts could be infected with CBTv. We grew cotton aphids in a colony in the glasshouse and used these to infest the field grown CBT affected cotton plants. We also collected seeds from potential host species and planted these seeds in amongst the CBT affected cotton plants in the CBT nursery. The nursery was sprayed and managed to keep predators out so that aphids multiplied and moved to the potential host species plantings. Once aphids reproduced on the plantings each plant species was collected, washed to remove aphids and virus residue they might carry, processed and frozen in preparation for extracting RNA. The potential host species we processed for extraction were from common weeds, crops or garden plants and are listed in Appendix 8, Table 1. Those in bold are poor or non-aphid hosts, which were opportunistically collected as species that germinated from the naturally occurring seedbank in the CBT nursery but are unlikely to host CBTv.

Our previous research with Dr Sharman showed that there were two strains of CBTv, Strain A and Strain B. Extraction of CBTv from a range of affected cotton crops showed that CBTvB is most important in disease manifestation. Plants can be infected by both strains at once. Where Strain A is present alone in cotton plants they do not appear symptomatic. Plants infected with Strain B alone or with Strains A and B simultaneously are symptomatic. We do need to keep

in mind that the relationship between Strains A & B hasn't been fully explored and it may be important for the process of disease development as well. Currently the family Malvaceae are known to contain the most important hosts of CBTv: *Gossypium hirsutum* ratoon, *Malva parviflora*, *Abutilon theophrasti* and *Anoda cristata* are particularly good harbourers of the disease and occur in and around cotton crops. Other Malvaceae such as *Malvastrum coromandelianum* and *Sida rhombifolia* are poor carriers, probably due to being poor aphid hosts. *Hibiscus sabdariffa* on the other hand is a moderately good host for cotton aphid but a poor disease host. Four other common weeds have been shown to be carriers sporadically and may not really be hosts. We have samples of *Medicago polymorpha* (Burr medic, a legume, tested positive to CBTvA only) and *Vigna radiata* (Mungbean, a legume) to re-test and *Rhyncosia minima* (Rhynco, a legume) and a *species of Macroptylium* (a legume) to test. They are all species of the order Fabaceae. *Lamium amplexicaule* (Deadnettle) and *Chamaesyce drummondii* (Caustic weed) are from families not well represented in cotton growing areas but both have tested positive to CBTvA & B so we have grown and collected them to re-test to confirm this result and rule out contamination. We also hope to test Watermelon, *Citrullus lanatus* (positive to B in the past) and several other Cucurbitaceae (*Cucumis* and *Cucurbita* spp.). The list of samples to test otherwise is prioritised with the commonness of the plant and its tendency to be infected with cotton aphid, and its presence in our trial in mind. We are maintaining a small field area of CBTv infected ratoon cotton for future use.

ii) Investigate the effectiveness of insecticide application to prevent spread of CBT.

These experiments aimed to test the effectiveness of seed treatments and aphid sprays to reduce transmission of CBT and used a similar methodology to experiments described in the Final Report for CRC1102.

Methods

Experiments 1 and 2 – Seed treatments

To test if two of the common seed treatments, Cruiser and Cruiser Extreme, could reduce transmission of CBT into cotton seedlings we transferred CBT infected aphids onto treated cotton seedlings at the one and 4 true leaf stage. We planted seed of a CBT susceptible variety (Sicot 71BRF) either untreated or treated with Cruiser or Cruiser Extreme in pots, 6 seeds per pot. We planted 10 pots with untreated seed, 15 with Cruiser seed and 15 with Cruiser Extreme seed. Of these pots five of each treatment served as controls for experimental method e.g. if there were 'stray' CBTv infected aphids that contaminated the experiment. After seeds germinated and seedlings emerged from the soil we thinned seedlings to four per pot. When plants had one true leaf 20 aphids were transferred, from CBT affected plants, onto each plant for five pots with untreated seed, and five each with Cruiser or Cruiser Extreme. When plants had four true leaves 20 CBT infected aphids were aphids were transferred onto each plant for five pots of Cruiser or Cruiser Extreme treated seed. The aphids used were from a neonicotinoid susceptible culture (a subsample was sent to Dr Grant Herron, NSW DPI for resistance testing in the previous season). Previous research showed that aphids can transmit CBT within one hour of feeding, so we allowed them to feed for 72 h then controlled them with pirimicarb. This was done to prevent aphid feeding causing damage to the plants that may mask symptoms of CBT disease. Plants were then monitored weekly for 10 weeks for the presence of CBT symptoms. We used the same protocol in the second experiment except that we had 21 control pots for the untreated seed which received 20 CBTv infected aphids per plant at one true leaf treatment.

Experiments 3 and 4 – Insecticide application

We also tested the effect on transmission of CBT of an insecticide applied 24 h before CBTv infected aphids colonised cotton plants, 10 minutes after colonisation or 24 hr after colonisation. We planted 45 pots with untreated seed of a CBT susceptible variety (Sicot 71BRF) at 6 seeds per pot. After seeds germinated and seedlings emerged from the soil we thinned seedlings to four per pot. Once plants had reached the four true leaf stage we imposed treatments. Five pots were left untreated as controls. Twenty-four hours before we infested pots with CBTv infected aphids we sprayed five pots with a 1% solution of Transform and another five pots with a 2% solution of transform. Twenty-four hours later we infested each plant in these ten pots plus those in an additional 25 pots with 20 CBT infected aphids. Five of these pots received CBTv infected aphid but were not sprayed as positive controls. Ten minutes later we sprayed five pots, plus five pots that received no aphids with Transform at 1% and a similar set of pots with Transform at 2%. This was repeated on a final set of pots at 24 h after infestation with aphids. After 72 h we controlled aphids on the treatment that received no insecticide and any aphids on other plants to prevent aphid feeding causing damage to the plants that may mask symptoms of CBT disease. Plants were then monitored weekly for 10 weeks for the presence of CBT symptoms. We used the same protocol in the second experiment, however we planted additional 'spare pots, that could be included for to replace replicates of treatments where we were unsure that the aphids had transferred well. This mean that some treatments had more replications than others (range 5 to 13 pots per treatment).

Analysis

We worked with NSW DPI statistician Bruce McCorkell to complete analysis of experiments across both years. The analysis of the experiments was quite complex due to differences in the number of pots per treatment in some experiments (2 and 4). Further, for the seed treatment experiments the +CBT aphids no seed treatment treatment is only present once, it has no 1 leaf and 4 leaf data. Similarly, for the insecticide prevention of transmission experiments the +CBT aphids no insecticide treatment is only present once, it has no -24h, +10 minutes, +24h data. Further, there are two ways to approach analysis, compare the number of CBT positive plants per pot or compare the number of pots with at least one CBT positive plant. Nevertheless, Bruce McCorkell was able to analyse using ASREML using a binomial distribution that allowed for differences in the numbers of replicates per treatment.

Results and Discussion

Experiments 1 and 2 – Seed treatments

The results are shown in Appendix 8a, Table 1. The controls for untreated seed and treated seed had no CBT infected plants, indicating the experimental method was effective. Considering the proportion of plants infested per pot there was no effect of stage of application of the aphids (nil, 1 leaf or 4 leaves, $p = 0.27$). Rate of insecticide (nil, cruiser, cruiser extreme) had a significant effect ($p = 0.003$), with Cruiser Extreme significantly lower than Cruiser or no insecticide, which were similar. There was no significant effect of rate by stage, so the effect of rate was similar regardless of stage (in other words the Cruiser Extreme was always lower regardless of when the aphids were placed on the plants). Considering the number of infested pots per treatment there was no effect of stage of application of the aphids (nil, 1 leaf or 4 leaves, $p = 0.66$). Rate of insecticide (nil, cruiser, cruiser extreme) had a significant effect ($p = 0.03$), with Cruiser Extreme significantly lower than Cruiser or control, but Cruiser and control not significantly different. There was no significant effect of rate by stage ($p = 0.8$), so the effect of rate was similar regardless of stage (in other words Cruiser Extreme was always lower regardless of when the aphids were placed on the plants).

In conclusion, Cruiser would not be effective in preventing transmission of CBT regardless of whether CBTv infected aphids colonised at 1 or 4 true leaves. Cruiser Extreme could reduce

the infection rate by 50% compared with the controls and results were similar at 1 and 4 true leaves.

Experiments 3 and 4 – Insecticide application
We evaluated the potential to use a new insecticide, highly effective against aphids, to prevent transmission of CBTv virus by cotton aphid. There was no effect of rate of insecticide for either the proportion of infested plants per pot ($p = 0.13$) or the proportion of infested pots ($p = 0.92$). There was a highly significant effect of timing of insecticides for both the proportion of infested plants per pot ($p < 0.001$) and the proportion of infested pots ($p < 0.001$). Application of the insecticide 24 h before aphids were placed on plants reduced transmission for the proportion of infested plants per plot (77-93%) but not for the proportion of infested pots (58-81%, Appendix 8a, Table 2) compared with the untreated control. Application of Transform at either rate 10 minutes after the plants were infested was not significantly different from the control for the proportion of infected plants or the proportion of infected pots, likely due to the short time span allowed for aphids to infect plants before being sprayed Application of Transform 24 h after infestation resulted in significantly higher transmission rates than the control, for both proportion of infected plants per pot and proportion of infested pots.

The results highlight that application of insecticide before a known migration of CBT affected aphids into cotton could effectively reduce the rate of transmission of CBTv resulting in a lower level of infection. For instance, if there are heavily infected ratoon plants near a cotton crop and these are sprayed with herbicide, then pre-treatment of the cotton crop may reduce transmission by aphids forced to migrate off the herbicide treated 'volunteers' or 'ratoons' (also known as rogue cotton). Application just after aphids entered the crop, or 24 hours after they entered the crop would not be effective.

If an influx of aphids carrying CBT occurs, then application of an insecticide would have to occur very quickly to prevent the initial transmission. If the number of aphids coming into the crop was high enough to result in aphids settling on every plant then, unless the insecticide was very well timed, application would be ineffective. However, if the infestation of aphids was at a lower level, with only a low proportion of the crop infested with aphids then the reduced aphid numbers following insecticide application may not prevent initial transmission but may reduce the risk of secondary transmission (e.g. from plants that became infected with CBT in the crop to new plants in the same crop).

Section E. Identify and manage emerging pests

The project has benefitted from experienced scientists and technical staff with connections to international and national expertise and good relationships with consultants, growers and industry extension to identify issues and deliver information quickly.

2012-13

Rutherglen Bugs
Phone calls from a number of growers and consultants indicated that Rutherglen bugs were an issue on seedling cotton planted next to fields that had hosted canola through winter. This was passed onto Susan Maas and extension material was made available to industry.

Late season thrips damage to flowers
There were reports of very high abundance of thrips in late season cotton. We had also observed this when we travelled to crops within the regions collecting mites or aphids for pesticide resistance testing. These thrips were mostly *Frankliniella schultzei*. Consultants were

concerned that thrips could cause damage to flowers late in the flowering cycle, causing them to shed, and potentially reducing yield. We approached this problem by asking "if thrips caused complete losses of flowers over a period, say a week, what would be the outcome for yield?" These experiments were carried out over three seasons: 2014/5, 2015/16 and 2016/17. Experiments are reported below under 'F. Effect of late season thrips damage to flowers on yield'.

2013/14

Symphyla

Symphylids emerged as an issue in a number of fields in the Gwydir and Namoi Valleys, the Darling Down and in Theodore. Simone Heimoana visited several fields with Mike Stone to see the situation first hand. Digging revealed symphylids in the soil. The insects had damaged the roots – resulting in virtually no tap root. Attempts to float symphylids from soil samples to quantify abundance were unsuccessful due to the creatures' fragility. It is unclear why these problems are occurring or whether the problem is becoming widespread. Ian Taylor co-ordinated a meeting with Dr Paul Grundy (QDAFF) and other interested researchers and consultants/agronomists to review the situation.

SLW

We met with consultants and growers in Moree in November 2013 to discuss whitefly management and the current guidelines which may need to be re-evaluated in high yield crop management situations. Further, at a grower meeting in St George, in our own experiments, in conversations with RDO's and consultants it became clear that the SLW sampling strategy could be unreliable under certain conditions. SLW in 2013/14 tended to stay lower in the canopy – and were not well represented in node 5 sampling. This needed further consultation with Richard Sequeira and possible research.

Rutherglen bug

Phone calls from consultants confirmed that Rutherglen bug was again a problem, especially where cotton was planted near fields that had canola in winter. Current control options are disruptive to natural enemies. It would be valuable to screen some compounds against this pest to identify effective and selective options as well as consider cultural controls.

Biosecurity

Wilson and Heimoana participated in the Cotton Biosecurity Committee meeting in Sydney in November 2013 and identified several potential threats. These included insecticide and/or Bt resistant *Spodoptera frugiperda* (fall armyworm). This species is a pest in the southern USA and in Brazil but has recently been reported from Africa. Bagrada bug, present through the SW USA, southern Europe and southern Asia was also identified as a species likely to eventually arrive in Australia, in particular since Australia imports fruits and vegetables from those areas.

2014/15

SLW

Through further discussions with the CCA and CRDC it was clear that there were concerns regarding management of whitefly. This included confidence in the main stem node 5 based sampling strategy developed by Dr Richard Sequeira. After discussions with Susan Maas at CRDC we met with Richard, reviewed these concerns and have started a collaborative research sampling effort to provide data to answer them (see sections on this topic above). These included (i) concerns that main stem node 5 (below the terminal) is too high on the plant and may not detect whitefly early enough for effective management, (ii) concerns that whitefly

distribution is affected by the time of day and this may affect the reliability of sampling in relation to the Threshold Matrix, and (iii) a desire to include some estimate of nymphal population development in decision making. Other concerns were around the presence of sooty mould growing on honeydew on leaves and bolls. Growers were concerned that honeydew/sooty mould on leaves reduced the effectiveness of defoliant applications. But more importantly there were concerns that even though sooty mould may help break down honeydew sugars, the moulds may contaminate the lint and result in colour downgrades (see above at Section A, part 'iv). Within plant and between plant distribution of SLW in central and southern regions' for a report on this research.

2016/17

Pest management in heavy pressure seasons

The 2016/17 cotton season was unusual in many ways. A wet winter resulted in early pest problems, a cool spring delayed crop development, a hot summer stressed plants even at night and resulted in fruit loss. Mirids were prevalent resulting in several applications of Fipronil and Rutherglen bugs (RGB) occurred in plague proportions in some cotton areas. Among the species already present in Australia there were some emerging trends. Firstly, the high abundance of early mirids in the 2016/17 season raised concerns about the need for high early fruit retention (> 90% at first flower) to achieve high yields (14 b/ha or more). Research to test the validity of this expectation was needed as trying to achieve this in a year with higher mirid abundance may lead to excessive spraying. Given that other pests such as spider mites were also in higher-than-usual abundance, there was increased risk of mirid sprays inducing mite outbreaks (or SLW outbreaks). As result of the experiences in this season we initiated new experiments to understand the responses of BG3 to early season damage from thrips and mirids, including, terminal damage and loss of early fruit resulting in reduced retention levels. For a report on this research and its rationale see section 'G. Effect of early season tip damage and fruit loss, damage due to thrips, mirids or other pests, on early season growth, yield, maturity and fibre quality in warm and cool regions'.

Rutherglen bug

Rutherglen bug has become a recurrent pest in cotton systems, especially for crops planted near canola stubble. This has given rise to two problems, firstly, high numbers of RGB larvae walking out of the stubble to feed in the cotton causing wilting and sometimes seedling death. Control options include physical barriers such as rough cloddy soil or channels filled with water. At present some growers are using 'border sprays' though insecticide spray options are limited and there are no registered 'selective' options for this pest. The second problem was that cotton fields may harbour significant numbers (dozens per plant) of RGB adults in the terminals and often also within squares. Past research at UQLD indicated that RGB is not a pest of vegetative cotton. However, the heavy numbers and anecdotal reports of reduced square retention mean that this issue needs clarification. To complement work done by others we have initiated some preliminary studies with RGB, see below.

Effect of RGB on cotton

During 2016/17 Rutherglen bugs (RGB) occurred in plague proportions in some cotton areas. Consultants were unsure if and how to manage RGB as it is generally classified as a non-pest, however, environmental conditions delayed boll setting which was attributed to insect damage by some. In order to clarify whether RGB adults caused square losses, 20 or 50 RGB were caged onto 5 day old bolls and checked after 1 week. While there were puncture marks on the outside of green bolls, the damage did not penetrate the boll wall (Fig. 55). This observation concurred with findings by Melina Miles (QDAF). We intended to investigate feeding damage by RGB on young cotton, however, numbers dropped off sharply by the time we had planted up seedlings and while we tried to breed RGB in the lab, numbers increased too slowly to replicate field infestations.

(a) (b)

Figure 55 a & b: Rutherglen bugs caged onto 5 day old cotton bolls, ACRI, 2016/17.

(c) (d)

e)

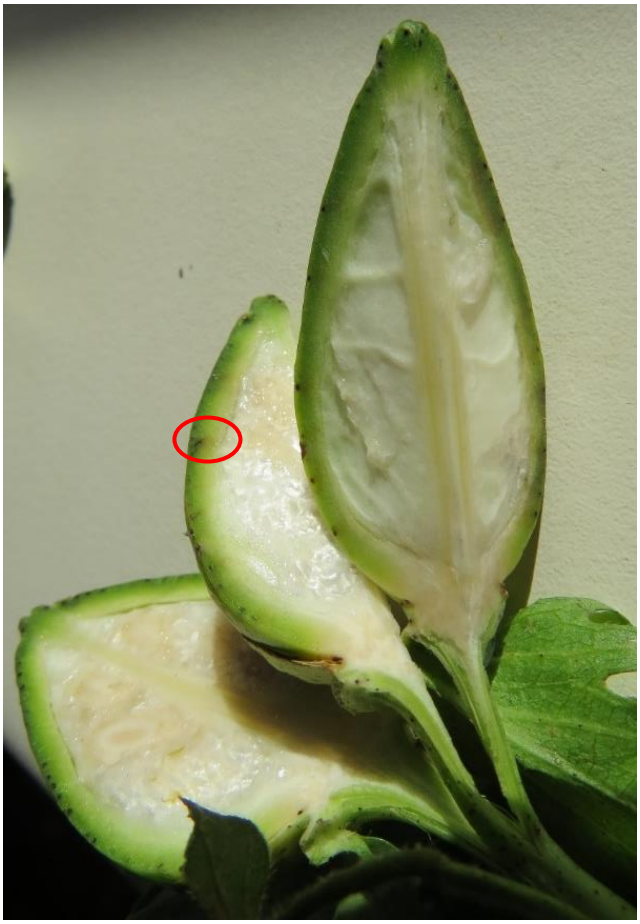


Figure 55 c, d & e: Twenty or fifty Rutherglen bugs per boll did not cause damage to developing cotton lint (d, e) though puncture marks were visible on the outside of the boll (c).

f)

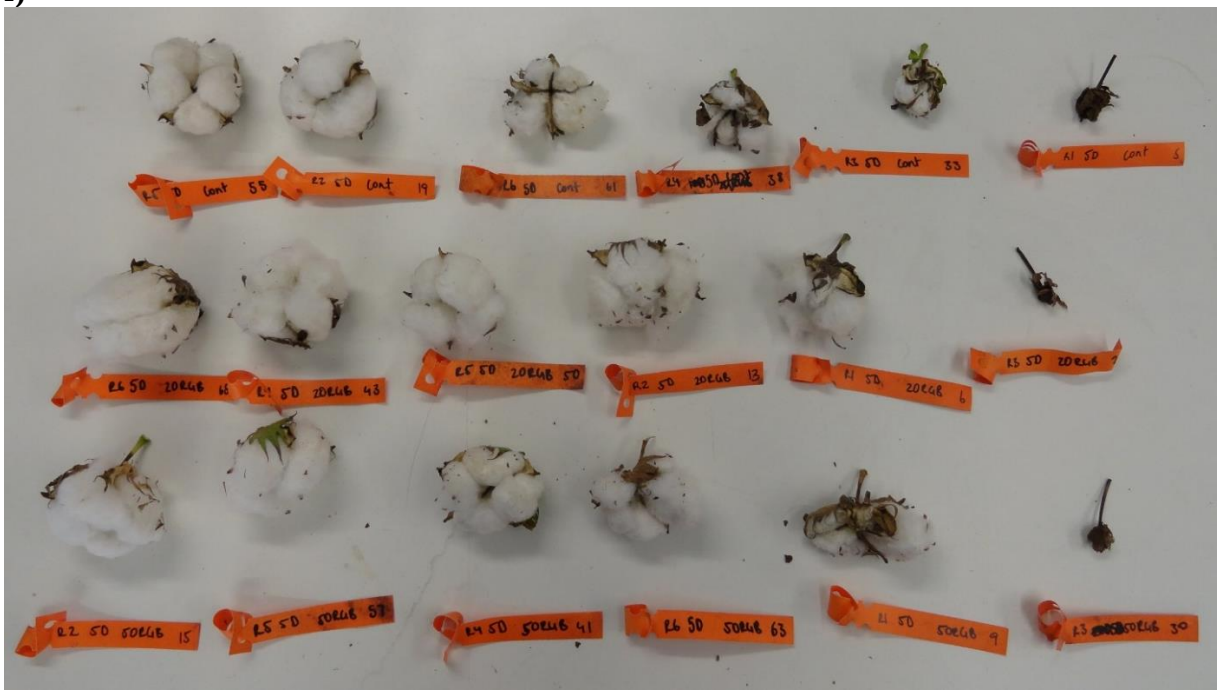


Figure 55 f: Post-harvest, RGB damage to bolls was either not apparent (f, bolls on left second/third row from top)) or caused by handling and other factors (f, bolls towards right second/third row from top) as it occurred in Control bolls as well (top row).

Biosecurity and SLW

Heimoana attended biosecurity training in Brisbane in July and has contributed to the Biosecurity Committee. A recent exotic pest alert for BMSB detection has been a timely reminder for staff training in staying alert to unknown pests seen in the field. During the winter we also had enquiries regarding the identification of aphids on wheat from Millie, Burren Junction (suspected *Diuraphis noxia* – negative), and SLW occurring on wheat as well. The SLW samples were sent for DNA testing and all the specimens found on wheat were grouped within *B. tabaci* MEAM1. Concerned that this may suggest wheat is a host for SLW Tanya Smith, a project Technical Officer, monitored the population on wheat regularly. However, the populations did not persist and gradually died out.

SLW Management

During the 2017/18 cotton season consultants expressed concern about penetration of chemicals to control SLW into dense canopies. As the cotton system changes (frequent irrigation, dense canopies, high N fertiliser, changing pest status and ecology), our management techniques need to change as well. SLW management has largely relied on Pegasus and Admiral, however, increasing resistance to pyriproxifen and application issues may require a different approach. We established a SLW x Chemistry experiment to become more familiar with the chemical control options available and their effects on SLW management. This experiment included Movento, Pegasus, Applaud, Exirel, Starkle, Mainman, Biopest Oil and Regent. We intended to infest the experiment with whitefly in early January and apply chemicals based on regular sampling and thresholds. Low whitefly numbers throughout the experiment meant that only one spray was applied and results were not as comprehensive as expected (see Section B v).

Solenopsis Mealybug spread

Solenopsis mealybug has been reported from the border region near Moree (Gurley, 1 specimen), St. George (2 specimen), Macintyre (1 specimen), Gunnedah (on okra), and the Namoi (Culgoora, in a patch) since 2016/17. In 2018/19, mealybugs were reported in a patch on a property 30 km south of Gunnedah (not Solenopsis, yet to be identified) while Solenopsis was reported from Boggabri. It is important to monitor the spread of this pest southwards and to ensure that the industry has ways and means to deal with potential outbreaks. While Solenopsis in Emerald does not appear to be a major pest problem if managed softly, different attitudes towards pest management in other regions could cause Solenopsis to become a difficult pest to manage in central regions.

2017/18

Pest management in low pressure seasons

The 2017/18 cotton season happened as the drought across eastern Australia worsened. It was a stark contrast to the previous season which saw the industry deal with very high pest populations, frequent pesticide applications and increasing pyriproxifen (Admiral, Lascar) resistance in Silver leaf whitefly (SLW). However, the industry proactively faced this threat by making changes to their mirid spray management, improving communications by forming Area Wide Management (AWM) groups and complying with regional 30 day spray windows for a single pyriproxifen application to minimise consecutive generations of SLW being exposed to resistance selection. Instead of relying largely on several applications of dimethoate or reduced rate fipronil (Regent) sprays, many growers applied other chemicals from the softer end of the spectrum such as sulfoxaflor (Transform), dinotefuran (Starkle) or flonicamid (Mainman). This should have contributed to lower overall whitefly numbers later in the season by being more selective with regards to the beneficials that control them. AWM was practised widely and was very successful with groups from different regions connecting and exchanging experiences. The benefits of AWM groups included mentoring of young agronomists new to the industry by more experienced consultants, and being encouraged to hold off on a spray when pests are close to or at threshold and considering all factors that feed into the decision making process. The voluntary Admiral window was set for each region at the beginning of the cotton season with input from consultants and, from feedback received, worked well for the irrigated crops. SLW numbers in dryland cotton were very low and no spraying was necessary.

Strawberry spider mites

We have been hearing about the changing mite complex in cotton for some time and 2017/18 was the first season where we experienced outbreaks of Strawberry spider mites (SSM) at the ACRI. They are similar in appearance to two spotted spider mites (TSM) but adults are much smaller and have six black spots instead of two. They also cause less damage to cotton - pale stippling on the underside of leaves is the only visible damage - and therefore have a higher threshold than TSM. How much higher is an unknown at the moment, but some research into the characterisation of SSM and Bean spider mite (BSM) damage being undertaken by Dr. Lisa Bird's Honour Student Chris Shafto at the ACRI should provide some answers in the future. From surveys carried out by Dr. Grant Herron in previous seasons and industry feedback, SSM have also occurred across other regions but consultants did not seem concerned as thrips were keeping them under control.

Unknown mite species

At the beginning of December Rob Holmes sent through pictures of mite damage on cotton seedlings and mites which he believed to be red-legged earth mites (Fig. 56). These mites usually attack winter cereal crops and occur much further south (Vic, SA) though there have

been reports that they are spreading north. Hence there was a suspicion that these mites were a different species. Tanya sent them for identification and we are awaiting results.

Figure 56: (left) Unknown mite species on cotton seedlings and (right) damage to seedlings, Gwydir, 2017/18 (Rob Holmes)

Section F. Effect of late season thrips damage to flowers on yield

In recent years there have been periods of very high abundance of thrips in late season cotton. These have mostly been *Frankliniella schultzei*. Consultants were concerned that thrips could cause damage to flowers late in the flowering cycle, causing them to shed, and potentially reducing yield. We approached this problem by asking 'if thrips caused complete losses of flowers over a period of, say a week, what would be the outcome for yield'. These experiments were carried out over three years from 2014-15 to 2016-17 (Experiments 1 to 3, respectively).

Methods

Each experiment was set up in a fully irrigated field with normal pest and weed control practices. We used a replicated design with 4 replicates (blocks). Each plot was 3 rows by 6m and we damaged all three rows. This plot length enabled us to machine harvest the central row for a more accurate estimate of yield. The treatments included an Undamaged Control, removal of one week's worth of flowers, removal of 50 % of leaf area of the top 6 leaves, and the combination of Flower removal and Leaf damage. Treatments were implemented at two different times, the first damage at 2-3 weeks after peak flowering (implemented at about 21 nodes average at the end of Jan or first week in February) – this coincided with the timing of major thrips infestations in commercial fields in the previous year, and the second damage 2 weeks later.

After each damage event we collected 1 m of plants from each treatment to assess dry matter to estimate the amount of leaf area removed. We also retained the leaf area removed from 1 m of plant row and the fruit from 6 m of plants so they could be measured and counted. Sequential harvests (maturity picks) were done in a 1 m section of row once bolls began to open and continued until all bolls had opened. At 60 % open boll, the crop was defoliated and the central low harvested with a single row picker.

Results and Discussion

Experiment 1 – 2014/15

We removed about 30 flowers per meter over a one week period in the Flower removal treatment and about 0.4 m^2 of leaf area in the leaf area treatment (Table 59). We were only able

to inflict one of the two damage events due to competition with other experiments for time. The results showed that the leaf removal and flower removal treatment individually reduced yield (Table 59). However, surprisingly the combination of the two did not affect yield. This is possibly because the removal of leaves allowed more light into the canopy and the removal of flowers decreased competition with bolls already set, resulting in compensation through increased boll weight. Though not statistically significant there was a trend for slightly higher boll weights in the combined flower and leaf damage treatment.

Experiment 2 – 2015/16

We had problems this season with poor plant stand in some plots due to residual herbicide damage, however, we managed to redesign the experimental layout to include all treatments. We were also able to include damage inflicted at peak flower and at cut-out. None of the damage treatments caused significant reductions in boll number, date of maturity (60% Open boll) or yield (Table 60). This was despite removing up to 40 flowers per m and 0.35 m^2 of upper canopy leaf tissue.

Experiment 3 – 2016/17

None of the damage treatments caused significant reductions in boll number, boll weight, date of maturity (60% Open boll) or yield (Table 61). This was despite removing up to 44 flowers per m and 0.41 m^2 of upper canopy leaf tissue. The unusual maturity profile this season, is possibly due to climatic factors around the time of harvest. First open bolls were observed on the 13th March 2017, on the 15th we had 75mm rain followed by 14 dry days at 28-34°C. By the time the fields had dried out and we did the first maturity pick on the 29th March, most plants were 35-60% open, skewing calculations for maturity delay.

Analysing the data across three years showed that there were significant differences between years for boll weight, Ginout% and hand-picked yield (Table 62). The interaction terms were not significant indicating broadly similar trend across the years, particularly for machine pick yields and maturity. There is a slight trend in treatments that include flower removal to have fewer bolls in the analysis across years (Table 63) which we expect to be due to the flowers we removed though the differences are not significant.

We noted marked variation in ginout even within experiments, particularly for machine picks which are ginned on the big gin at ACRI because of their trash content. The current cotton varieties that now produce such high yields, have smaller seeds that slip through in the ginning process causing higher lint weights due to higher seed content. This likely inflates machine pick yields. Hand-picked samples, which are cleaner, are ginned on smaller gins where the problem with seeds slipping into lint is not as acute.

Conclusions

We have previously shown that complete removal of the top 6-9 main stem node leaves of the plant during flowering and up to cut-out significantly reduced yield in both high (15 + bales/ha) and low yielding crops (< 15 bales/ha). We also showed that the yield loss potential is higher in high yielding crops. Here initially, either removal of half the main stem leaves of the top 6 nodes or removal of one week's worth of flowers significantly reduced yield by about 1.5 bales/ha. In two subsequent experiments where the control yielded 16.5 bales/ha, yield reductions of up to 2 bales/ha were not statistically significant. However, at prices of $600 per bale, a $1200 loss/ha would not be considered insignificant or acceptable. Overall the data show no consistent pattern of yield reduction due to leaf loss, flower removal or the combination of the two at the damage levels inflicted. However, it is probable that more severe flower loss would reduce yield and we began testing for this in subsequent experiments (Section G). Commercially it would be rare to see either severe damage to upper leaves, such as we have inflicted, or to late flowers, or damage so severe that it would cause yield loss. In

the latter case, the question whether thrips actually cause flower loss and if so, under what conditions, remains and if they do, controlling them would be difficult. Some critical questions remain – how much flower loss is too much? We are currently collaborating with Paul Grundy in addressing this question in the new IPM project 2019-2021.

Table 59: Effect of leaf and flower removal treatment imposed at about 2 weeks post peak flower on yield, yield components and maturity, ACRI, 2014/15

Treatment	Time of damage	Flowers removed/m	Leaf area removed (m^2)	S/C Boll weight (g)	Bolls/m	Maturity date (days after sowing)	Hand Pick Yield (b/ha)	Machine Pick Yield (b/ha)
Control		0.0	0.00	3.53	143.5	150.4	13.3	15.35[a]
Flowers removed	Peak Flower	32.6	0.00	3.83	142.5	150.8	13.6	13.75[b]
50% leaf area removed from top 6 nodes	Peak Flower	0.0	0.42	3.57	121.8	151.0	11.4	13.77[b]
Flowers and leaves removed	Peak Flower	30.2	0.43	3.95	133.0	150.2	13.3	14.86[ab]
LSD		**-**	**-**	**n.s.**	**n.s.**	**n.s.**	**n.s.**	**0.045**
P (3, 9 df)		**-**	**-**	**0.103**	**0.216**	**0.957**	**0.236**	**1.274**

Table 60: Effect of leaf and flower removal treatment imposed at about 2 weeks post peak flower on yield, yield components and maturity, ACRI, 2015/16

Treatment	Time of damage	Flowers removed/ m	Leaf area removed (m^2)	S/C Boll weight (g)	Bolls/m	Maturity date (days after sowing)	Hand pick yield (b/ha)	Machine Pick Yield (b/ha)
Control		0.0	0.00	5.04	153.8	157.1	13.1	16.6
Flowers removed	Peak flower	27.5	0.00	5.03	132.2	157.6	11.3	15.8
50% leaf area removed from top 6 nodes	Peak flower	0.0	0.38	5.17	159.0	157.0	13.3	14.9
Flowers and leaves removed	Peak flower	24.0	0.35	5.37	132.2	160.0	11.8	16.0
Flowers removed	Cut-out	38.0	0.00	5.17	158.2	155.9	12.9	15.7
50% leaf area removed from top 6 nodes	Cut-out	0.0	0.28	5.20	138.0	156.5	12.3	15.4
Flowers and leaves removed	Cut-out	42.5	0.36	5.21	126.3	157.4	11.3	16.1
LSD		-	-	n.s.	ns	ns	ns	n.s.
P (6, 27 df)		-	-	0.75	0.09	0.18	0.43	0.950

Table 61: Effect of leaf and flower removal treatment imposed at about 2 weeks post peak flower on yield, yield components and maturity, ACRI, 2016/17

Treatment	Time of damage	Flowers removed/ m	Leaf area removed (m^2)	S/C Boll weight (g)	Bolls/m	Maturity date (days after sowing)	Hand Pick yield (b/ha)	Machine Pick Yield (b/ha)
Control		0.0	0.00	4.69	136.3	153.4	12.0	16.5
Flowers removed	Peak flower	44.0	0.00	4.78	120.8	161.8	11.0	15.5
50% leaf area removed from top 6 nodes	Peak flower	0.0	0.38	4.62	139.3	161.6	12.1	14.7
Flowers and leaves removed	Peak flower	33.5	0.41	4.41	136.8	144.9	11.3	14.8
Flowers removed	Cut-out	15.0	0.00	4.72	132.0	153.3	12.2	14.5
50% leaf area removed from top 6 nodes	Cut-out	0.0	0.38	4.87	133.8	170.2	11.6	14.9
Flowers and leaves removed	Cut-out	15.0	0.40	4.31	122.8	170.9	10.6	14.8
LSD		-	-	n.s.	n.s.	n.s.	n.s.	n.s.
P (6, 27 df)		-	-	0.216	0.156	0.208	0.309	0.120

Table 62: ACRI Thrips Damage Experiment, Year ANOVA for Peak Flower damage, 2015-2017

Year	**Bolls/m**	**Boll Wt (g/boll)**	**Ginout%**	**Hand Pick Yield (b/ha)**	**60 %OBDAS**	**Machine Pick Yield (b/ha)**
2014/15	135.2	3.71[a]	43.91[b]	12.87[b]	150.52	14.43
2015/16	144.3	5.15[c]	46.57[c]	14.9[c]	157.91	15.22
2016/17	133.2	4.62[ab]	41.93[a]	11.61[a]	156.46	14.57
F	0.16	<0.001	<0.001	<0.001	0.071	0.060
LSD (P=0.05)	n.s.	0.211	1.945	1.195	n.s.	n.s.
df	(2, 47)					

Table 63: ACRI Thrips Damage Experiment, Year x Treatment ANOVA for Peak Flower damage, 2014-2017

Year	Treatment	Bolls/m	Boll Wt (g/boll)	Ginout%	Hand Pick Yield (b/ha)	%60%OBollDAS
2014/15	Control	143.5	3.53	40.54	13.29	150.04
2014/15	Peak Flower - 50% Leaf	121.75	3.52	43.33	11.36	151.00
2014/15	Peak Flower - 50% Leaf & Flower	133	3.95	46.72	13.29	150.25
2014/15	Peak Flower - Flower	142.5	3.83	45.06	13.55	150.80
2015/16	Control	153.75	5.04	46.66	15.79	157.66
2015/16	Peak Flower - 50% Leaf	159	5.17	44.95	16.04	157.47
2015/16	Peak Flower - 50% Leaf & Flower	132.25	5.37	47.24	14.16	158.55
2015/16	Peak Flower - Flower	132.25	5.03	47.42	13.61	157.98
2016/17	Control	136.25	4.69	42.45	12.04	154.43
2016/17	Peak Flower - 50% Leaf	139.25	4.62	41.42	12.12	162.64
2016/17	Peak Flower - 50% Leaf & Flower	136.75	4.41	42.17	11.34	145.94
2016/17	Peak Flower - Flower	120.75	4.78	41.66	10.96	162.82
F		0.111	0.121	0.264	0.174	0.435
LSD (P=0.05)		n.s.	n.s.	n.s.	n.s.	n.s.
df	(6,47)					

Section G. Effect of early season tip damage and fruit loss, damage due to thrips, mirids or other pests on early season growth, yield, maturity and fibre quality in warm and cool regions.

The 2016-17 season was characterised by heavy early season thrips damage, high and prolonged mirid abundance and high and prolonged populations of Rutherglen bug. The damage potential to cotton of the first two pests is generally well understood. Thrips could cause leaf stunting and perhaps cause tip damage if in very high numbers. Mirids, and potentially other sucking pests such as apple dimpling bugs, could cause tipping out and may also feed on young squares causing them to shed, resulting in reduced retention. However, the damage potential of RGB is still uncertain. This question been the subject of recent experimentation (see above).

In the warm season production areas the potential for cotton to recover from early pest damage is generally well understood. Previous research indicated that the risk of yield loss from early season thrips populations is about 1 year in ten in those regions. Research with simulated damage showed that from cutout onwards, plants could tolerate reductions in leaf area of up to 80% without loss of yield or delay in maturity. Responses to simulated thrips damage showed that plants could recover without loss of yield or delay in maturity from several tip damage events. However, as yield levels have increased considerably since that work was completed it is worthwhile considering if these results still apply.

The cotton production area has also expanded both south and east. These eastern areas, e.g. near Willow Tree, are characterised by similar day-length to nearby central regions, e.g. the Namoi Valley, but cooler, shorter seasons. However, with Bollgard III and RRFlex varieties cotton is an attractive proposition in terms of gross margins and consequently growers in these areas have been growing dryland cotton crops over the past 3-5 seasons. A key concern for growers in these cool regions is that early season damage from pests such as thrips and mirids could delay the onset of fruiting, and as a result delay crop maturity. In these cool regions delayed maturity could mean crops are finishing off in February-April under the cool conditions. This poses significant risks of reduced yield and fibre quality.

Previous experiments comparing yield of crops protected from thrips or not protected from thrips in ‘cool’ regions such as the Upper Namoi, Macquarie and Eastern Darling Downs suggest that the risk of suffering yield loss from thrips damage to seedlings is about 1 year in 2 (50%). However, these results are worth re-considering in these new cooler regions near Quirindi because (a) the research with thrips damage was done in non-Bt cotton or Ingard cotton where the plants would have been subject to ongoing fruit loss to *Helicoverpa*, especially in the mid and late season and (b) there is no data for these even cooler ‘cool’ regions where the time available for compensation may be limited. Further, there has been no systematic study using simulated damage to explore responses to leaf, tip and fruit damage as there has been in the warm regions. Hence, the thresholds for early season pests and damage may be different in these area and this needs to be assessed.

We carried out experiments with simulated damage to assess the impact of early season damage on leaf area, terminals and early fruit retention in both warmer, long season areas (ACRI) and the new cooler, short season regions centred around Quirindi/Willow Tree and Spring Ridge. We used tweezers to remove the terminal, simulating tipping out from mirids and removed young squares and flowers simulating early season fruit loss and poor retention caused by mirids or other pests. We also removed fruit later on close to cut-off. Such information will be important in helping to restore/gain the confidence of growers and consultants that current thresholds for managing these early season pests are correct.

2017/18 Geurie, Spring Ridge

Growers were concerned about fruit loss during the 2016-17 season, some of which related to high mirid numbers, heat and water stress, cold shocks and other physiological factors. We wanted to know how much fruit loss Bollgard 3 plants could sustain before experiencing a yield penalty and whether plants could compensate for fruit loss. Where does the compensation occur?

Method

Fruit removal experiments were carried out at ACRI (long season area) and Spring Ridge (short season area) to assess yield loss and damage compensation/tolerance. The experiment was set up with 7 treatments in 4 replications, outlined in Table 64. Damage involved +/- Tipping at Node 5/6, and/or the removal of all fruit (pin squares/squares/bolls) on a fruiting branch. Both the second and third damage events (fruit removal only) occurred between first flower and peak flowering (see Day Degree guidelines below) while the fourth damage (fruit removal only) occurred just before the first bolls opened. It was difficult to describe damage by the stage at which it occurred (e.g. peak flower) as the events fell somewhere between first flower and peak flower, hence damage descriptions are either as D1-D4 (chronological) or by severity (fruit removal below node 12).

Tip damage was simulated by pinching out the terminal with tweezers to set plant development back. Fruit damage was simulated by removing all fruit, including pin squares, squares, flowers and bolls, from designated fruiting branches. In order to quantify plant parameters at each damage event, we recorded plant height and nodes of 4 x 1m sections of row in control and damaged plots. The fruit removed from the centre row of each plot were kept and counted (Tables 65 and 66). Prior to defoliation, 5 plants from each plot were mapped to assess how plants had compensated for damage. All plots were maturity picked (1 m at ACRI, 2 m at Spring Ridge) and the centre row of the ACRI plots was also machine picked. Samples were ginned to assess yields. At both sites mirids were controlled based on regular bug checks.

Growth Phase	**Day Degrees (DD)**
Sowing to emergence	80
5th Leaf	330
1st Square	505
1st Flower	777
Peak Flowering	1302
Open Boll	1527
60% Open Boll	2050

Table 64: Damage treatments and timing of fruit removal at ACRI and Spring Ridge

TREATMENT	ACRI PD 06/11/17	SPRING RIDGE PD 20/10/17
Control	No damage	No damage
D1 Tipping at Node 5/6 **DAS** **DD (5th leaf)**	06/12/17 **30** **325**	05/12/17 **46** **391**
D1 + D2 Tipping at Node 5/6 + Fruit Removal below Node 16 **DAS** **DD**	06/12/17 & 11/01/18 **30 & 66** **325 & 956**	05/12/1746 & 10/01/18 **46 & 82** **391 & 924**
D1 + D3 Tipping at Node 5/6 + Fruit Removal below Node 12 **DAS** **DD**	06/12/17 & 23/01/18 **30 & 78** **325 & 1150**	01/12/17 & 30/01/18 **46 & 102** **391 & 1222**
D2 Fruit Removal below Node 16 **DAS** **DD (after first flower)**	11/01/18 **66 (F)** **956**	10/01/18 **82** **924**
D3 Fruit Removal below Node 12 **DAS** **DD (prior to peak flower)**	**23/01/18** **78** **1150**	30/01/18 **102 (F)** **1222**
D 4 Removal of 1st & 2nd position bolls below Node 12 **DAS** **DD (open boll)**	22/02/18 **108** **1628**	28/02/18 **131** **1613**

DAS = Days after sowing DD = Day degrees PD = Planting Date

Explanation of damage treatments:

D1 = Simulation of thrips damage. First damage where we tipped some plots at the 5th or 6th node. In some plots this was followed by fruit removal.
D2 = Simulation of early mirid damage. Second damage after first flower, where we intended to remove all fruit below node 12. However, when we mapped plants later, we realised that we had taken off more than intended and had actually removed all fruit below node 16, so this damage was quite severe.
D3 = Simulation of later mirid damage. Third damage prior to peak flower, where we removed all fruit below node 12.
D4 = Simulation of late fruit loss due to other possible factors (e.g. heat/water stress). Fourth damage just past first open boll, where we decided not to implement total fruit removal below node 12, but only removed 1st and 2nd position bolls from those nodes.

Table 65: Removal of mean fruit per meter from plots at ACRI and Spring Ridge, 2017/18

Treatment / Location	Squares		Candlewick		Yellow Flower		Pink & Purple flower		Green Bolls	
	ACRI	Spring Ridge	ACRI	Spring Ridge	ACRI	Spring Ridge	ACRI	Spring Ridge	ACRI	Spring Ridge
D1 & 2 Tipping & Fruit below N16	114.71	67.83	0	0	0.13	0	0.04	0	0	0
D1 & D3 Tipping & Fruit below N12	119.50	137.42	21.04	6.29	5.38	3.25	20.92	27.54	0	0
D2 Fruit below N16	149.67	72.17	0.17	0	0.29	0	0.08	0	0	0
D3 Fruit below N12	108.04	159.46	27.83	8.46	4.54	3.08	28.29	28.38	0	0
D4 Fruit Removal 1st & 2nd Pos below N12	0.08	0	0	0	0.04	0	0	0	50.04	45.25

Table 66: Ranges of removed fruit per meter from plots at ACRI and Spring Ridge, 2017/18

Treatment / Location	Squares		Candlewick		Yellow Flower		Pink & Purple flower		Green Bolls	
	ACRI	Spring Ridge	ACRI	Spring Ridge	ACRI	Spring Ridge	ACRI	Spring Ridge	ACRI	Spring Ridge
D1 & 2 Tipping & Fruit below N16	74-139	54-76	0	0	0-<1	0	0-<1	0	0	0
D1 & D3 Tipping & Fruit below N12	100-147	100-173	11-25	4-8	3-7	1-4	17-24	23-30	0	0
D2 Fruit below N16	1021-176	54-88	0-<1	0	0-<1	0	0-<1	0	0	0
D3 Fruit below N12	97-137	143-185	20-24	5-12	4-5	2-3	22-35	19-41	0	0
D4 Fruit Removal 1st & 2nd Pos below N12	0-<1	0	0	0	0-<1	0	0	0	39-65	40-49

Results

Yield

At ACRI, machine picks showed no significant differences in yield between the Control and treatments where fruit were removed below Node 16 (early flowering) and Node 12 (peak flowering), with or without tip damage, with yields around 16 bales/ha (Table 67). Plants that only received tip damage at Node 5/6 yielded significantly less (13.7b/ha). Plants where the 1st and 2nd position bolls on fruiting branches below Node 12 were removed very late (open boll) also yielded significantly less (11 b/ha) than the control. The result for the tipping damage is unusual as our previous research found early tipping rarely caused a yield reduction.

There were no statistical differences between treatments yields in ACRI hand picks though yield broadly followed a similar pattern to the machine harvests. The machine pick data are more reliable than the hand picks as the sample size was six-fold.

Table 67: Fruit removal experiment yield - hand and machine picks, ACRI, 2017/18

TRT	Hand Pick Y (bales/ha)	Machine Pick Y (bale/ha)	Machine Pick Ginout%
Control	13.5	16.9[a]	47.9
D1 Tipping N 5/6	12.4	13.7[b]	47.5
D1 + D2 Tipping N 5/6 Fruit Removal Below N16	14.4	15.6[a]	48.8
D1 + D3 Tipping N 6 Fruit Removal Below N12	12.9	16.2[a]	48.3
D2 Fruit Removal Below N16	15.0	16.6[a]	48.7
D3 Fruit Removal Below N12	14.2	16.0[a]	51.3
D4 Fruit Removal 1&2 Pos. Fruit Below N12	11.0	11.1[c]	48.2
F (P=0.05)	0.095	<0.001	0.188
LSD	n.s.	1.751	n.s.
df	(6, 27)	(6, 27)	(6, 27)

At Spring Ridge, however, the scenario was different (hand picks of 2 m^2 since there was no opportunity machine harvest). D1, tipping at Node 5/6 had no effect on yield. All fruit loss treatments (D2, D3, D4), with or without tipping (D1), significantly reduced yield (Table 63), probably because the shorter season did not allow plants time to set further fruit to make up for the early losses due to removal. The damage to fruiting branches close to peak flower (D3) was done at the end of January and Table 60 shows that at that stage about twice as many squares were removed than at D2 and D4 though yield for the D3 treatments was not significantly different from the yields for D2 and D4 (Table 68). Plants in this treatment did not have time to set new fruit further up on the plant to compensate for this loss. To compare this to CSD data from the same field (not shown) – their trial picked at 10.11 bales/ha with a Ginout of 43.9%.

Table 68: Fruit removal experiment yield - hand picks, Spring Ridge 2017/18

TRT	Hand Pick Y (bales/ha)
Control	11.43[a]
D1 Tipping N 5/6	11.49[a]
D1+D2 Tipping N 6 Fruit Removal Below N16	9.08[b]
D1+D3 Tipping N 6 Fruit Removal Below N12	7.85[bc]
D2 Fruit Removal Below N16	9.53[b]
D3 Fruit Removal Below N12	7.58[bc]
D4 Fruit Removal 1&2 Pos. Fruit Below N12	8.65[b]
F (P=0.05)	<0.001
LSD	1.340
df	(6, 27)

Boll Numbers and weight

There were no significant differences in boll numbers between treatments at ACRI (Fig. 57). The D1, D1 + D3 and D4 treatments had fewer bolls than the control (12-19 less), while the D 2 and D3 treaments had 12 and 6 bolls more, respectively. Boll weights of D1 + D2 and D1 + D3 (tipped treatments where bolls were also later removed) were significantly higher than the control (Fig. 59). Compensation in the fruit removal treatmend D1+D2, D2, D3 and D1 + D3 appears to be via increased boll number and or increased boll weight e.g. D1+D3 had fewer but heavier bolls, thus making similar yield to the control. Conversly some treatments did not recover yield, e.g. D1 and D4 had fewer bolls that were similar in weight to the control. Plants managed to recover from the combination of tipping and fruit removal quite well and it is likely that the removal of fruits in January during flowering allowed for better vegetative growth, a heavier boll load higher up in the plant and the capacity to mature that load.

At Spring Ridge, boll number was significantly reduced in the D2, D1+D3, D3 and D4 treatments and significantly increased in the D1 treatment (Fig. 58). There were no significant differences in boll weight but there was a trend for higher boll weight where fruit had been removed from nodes 16 and 12 (Fig. 60). Plants with fruit removal were unable to compensate for loss of fruit by either retaining more fruit later or by increasing boll weight.

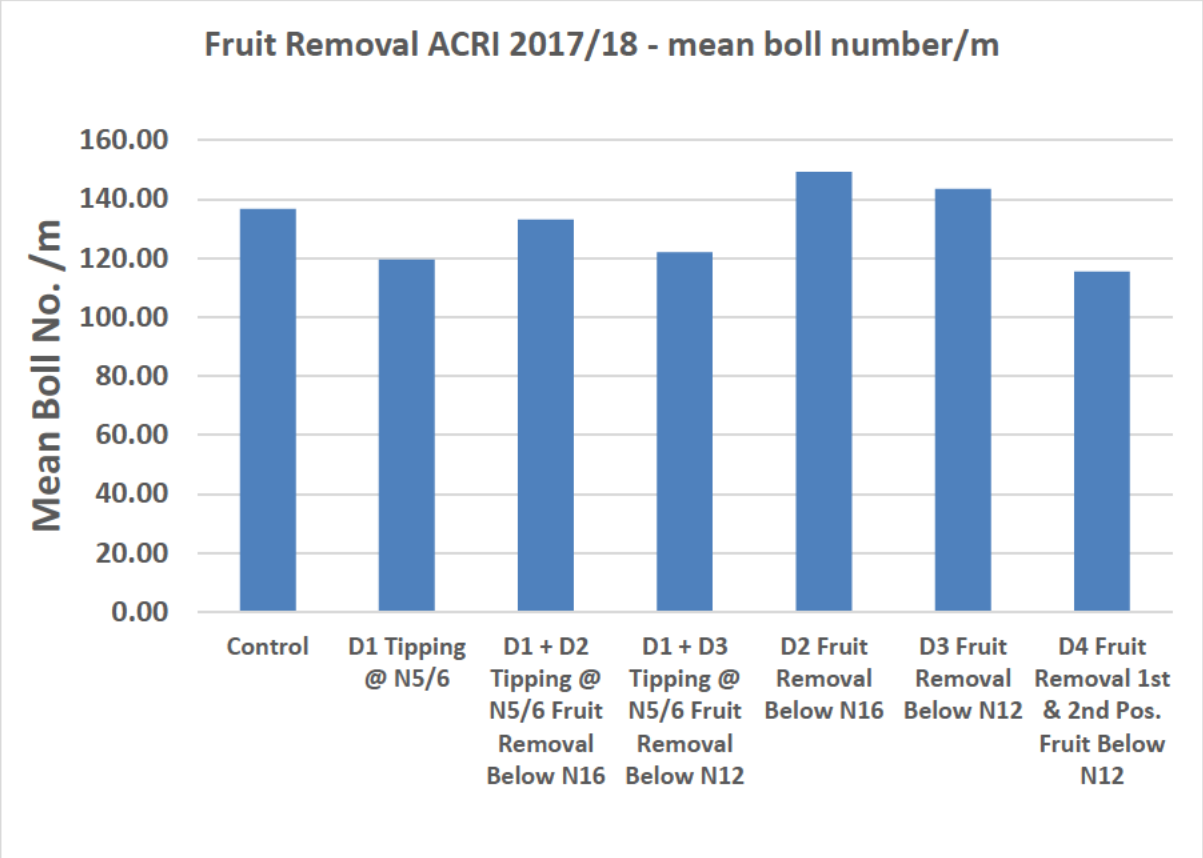


Fig 57: Fruit removal experiment – Mean boll numbers per meter from picks, ACRI 2017/18

P=0.05, F=0.156, n.s.

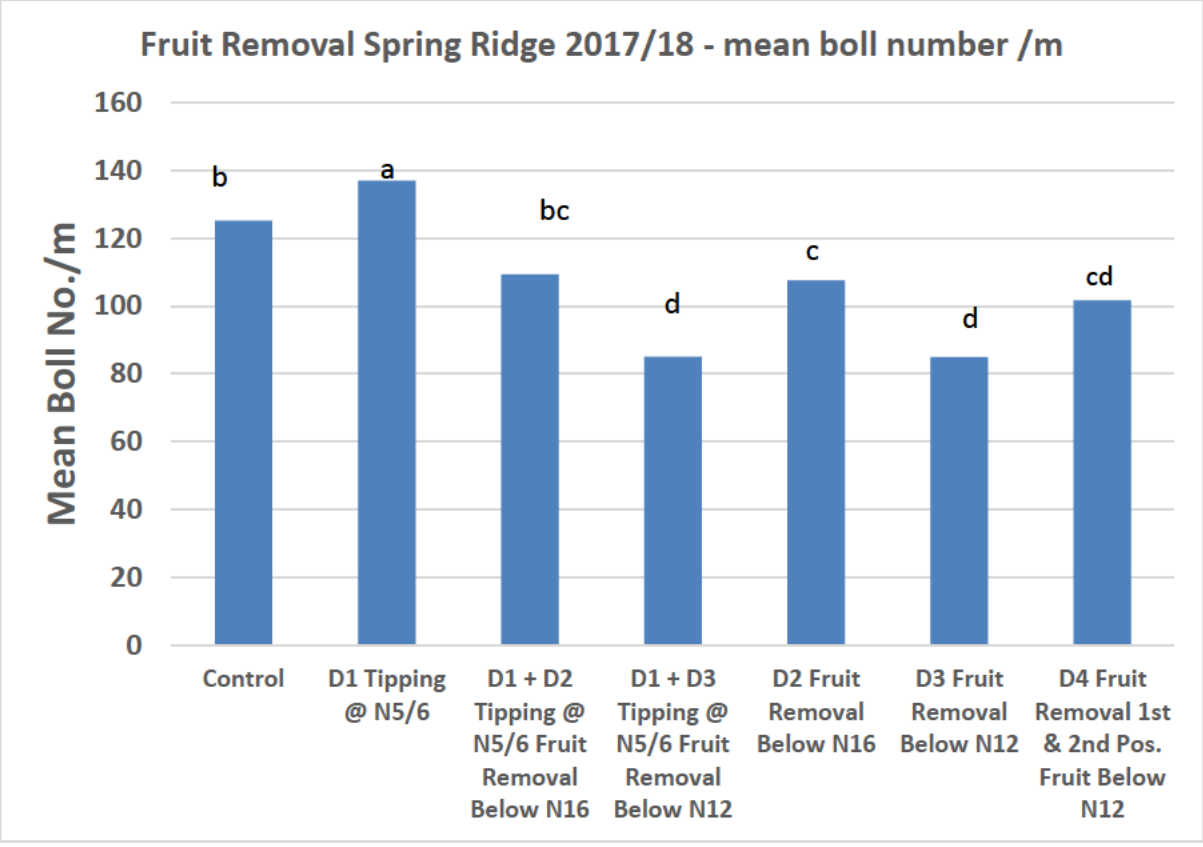


Fig 58: Fruit removal experiment – Mean boll numbers per meter from picks, Spring Ridge 2017/18, P = 0.05, F<0.001

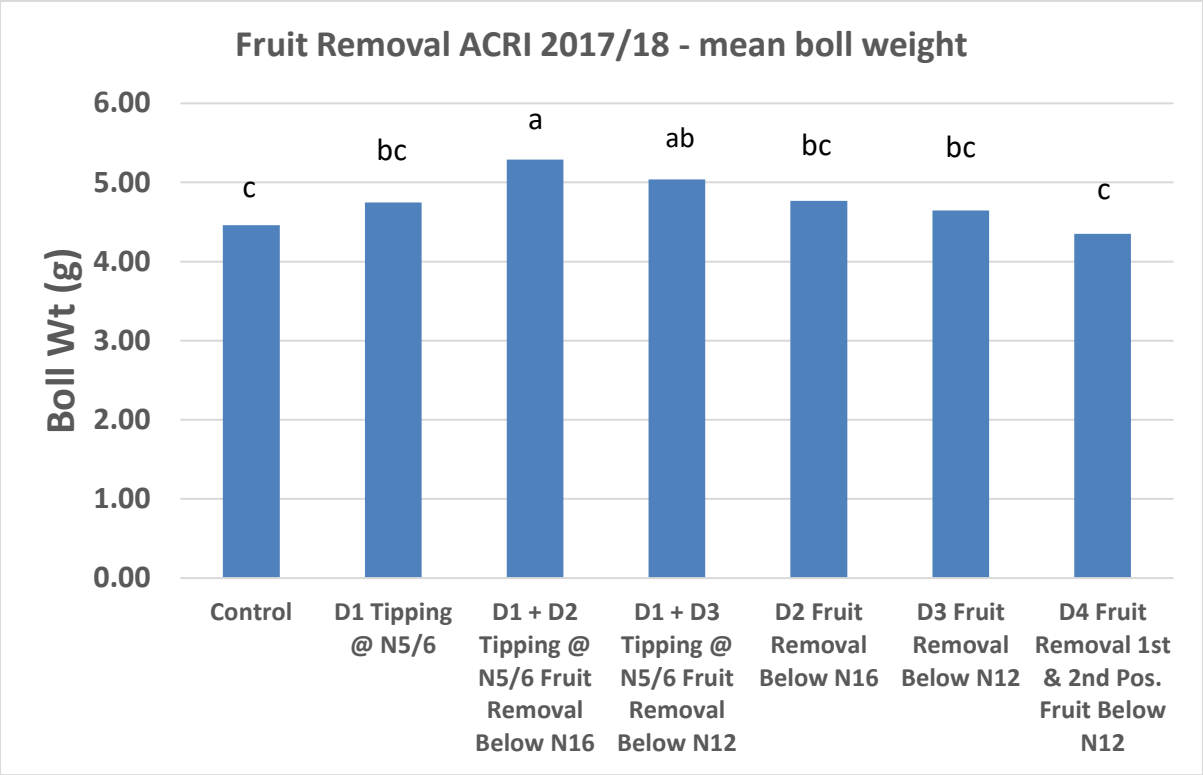


Fig 59: Fruit removal experiment – Boll Weight, ACRI 2017/18

P=0.05, F=0.016

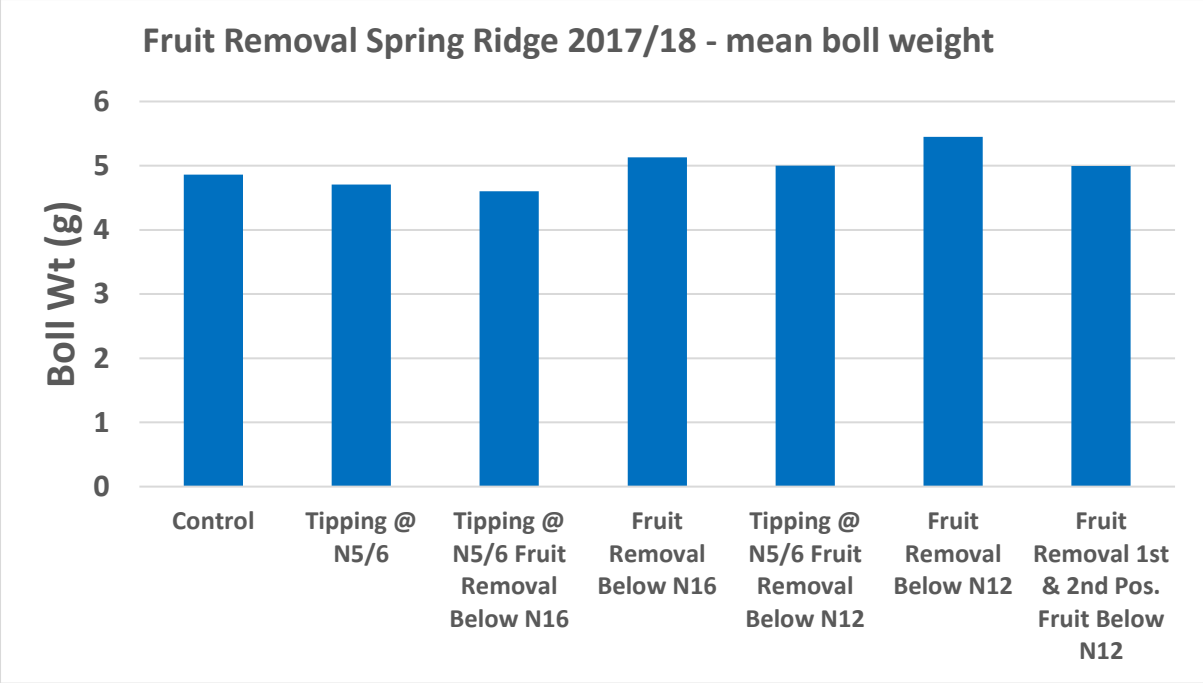


Fig 60: Fruit removal experiment – Boll Weight, Spring Ridge 2017/18

P = 0.005, F=0.185, n.s.

Maturity

At both sites there were significant lateness penalties for early fruit loss. D1 (Tipping) incurred no delay at ACRI (Fig. 61) and only 2 days delay at Spring Ridge (Fig. 62). All other treatments where extensive fruit numbers were removed were 11-12 days late at ACRI and 6-12 days late at Spring Ridge.

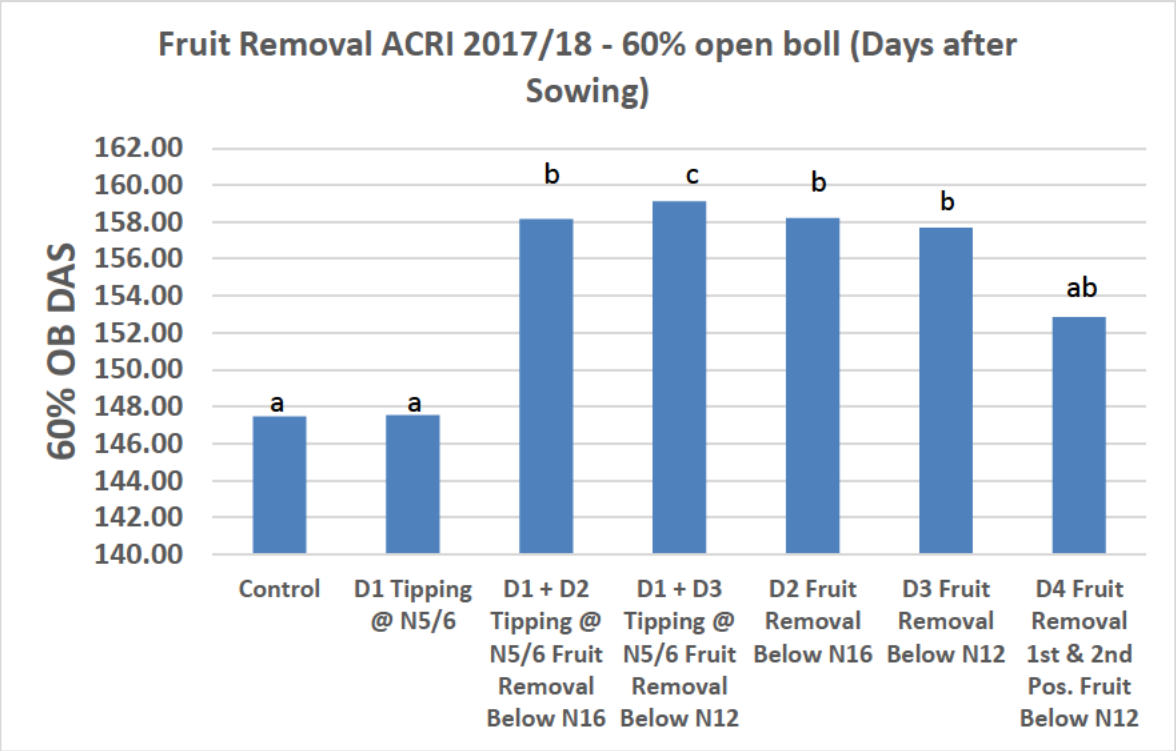


Fig 61: Fruit removal experiment – Maturity, ACRI 2017/18

P=0.05, F=<0.001

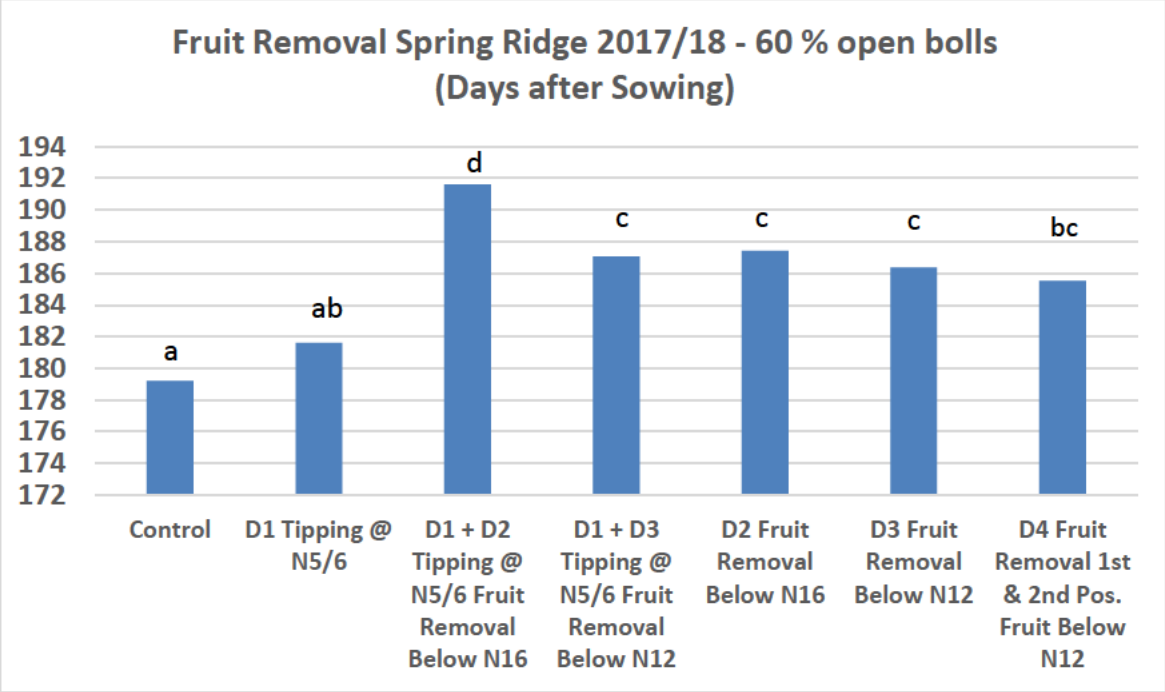


Fig 62: Fruit removal experiment – Maturity, Spring Ridge 2017/18

P = 0.005, F=<0.001

Plant Mapping

At ACRI, There was a significant effect of the D1 treatment (tipping) and also in the D1 + D3 (tipped + fruit removal below N12) treatment with respect to the number of fruiting branches and potential fruiting positions (No. of fruiting branches x 3 fruiting positions) (Table 69). The D1 + D2 (tipped + fruit removal below N16) treatment also trended lower but was not significant. This reduction in potential fruiting positions did not translate into a significant loss of actual fruit positions (no. of squares/flowers/green bolls & missing fruit on fruiting branches) though tipped treatments tended to have less positions than the Control. Retention ranged between 25 and 35% for the different treatments and was not significant. Retention was highest in the Control, with the differences in retention for treatments from the Control ranging from -3.73 to 10.39%. This indicates that plants were able to compensate to some extent for the fruit we removed. The only tipping treatment that showed a yield penalty was the D1 treatment. While the 10.39 % reduction of fruit from the last D4 treatment was not statistically significant, it caused a 5.8 bale reduction in yield! Appendix 9 shows the distributions of fruit and missing fruiting positions and when comparing the D4 treatment to the Control (Figs. 1 & 2), it has a very similar fruiting profile. The losses (2.6 fruit below Node 12 and 2.5 missing below Node 12) appear more marked on the fruiting branches of the lower canopy.

Differences in missing fruit were statistically insignificant, without any treatment, the control had lost about 60% of squares/bolls. During D2 we took off all fruits below node 16 which by the end of the season amounted to a 10 -11% difference from the control. Data from D3, where we removed all fruits below node 12, are less clear – where fruit was removed from untipped plants, about 8% more fruit were missing than from tipped or control plants. With respect to the vertical distribution of fruit and missing fruit across the three positions (P1-P3) on fruiting branches at the end of the season. About 14-23% of all fruit set at P1, 5-11% at P2 and 0.5-3% at P3. There were also 30-45, 16-25 and 3-8% of fruit missing from those positions, respectively. This pattern reflects across treatments highlighting that while a higher proportion of fruit are set and retained at position 1, there are also higher losses (missing fruit) from that position. This is also true for positions 2 and 3. Treatment differences for fruit in position 1 were not significant, even for D4, where we specifically removed first and second position bolls below node 12 (mean 50 removed bolls/m). Hence, while there were less fruit in this treatment, they were still distributed in a similar proportion to the control.

At Spring Ridge, there was no significant effect of tipping on the number of fruiting branches though there was a trend for tipped plants to have fewer fruiting branches (Table 70). Damage did not affect potential or actual fruiting positions, and differences in retention from the Control ranged from 5-19%. While this was not statistically significant, it translated into significant yield differences between treatments. Differences in missing fruit were also not significant and control plants had lost 43% of their fruit. The pattern of fruiting in relation to fruiting positions showed that more fruit set in position 1 than in position 2 and even fewer set in position 3. The same pattern was true for missing fruit from those positions with higher proportions of fruit missing from position 1 than from positions 2 and 3. Significantly fewer fruit were set at P3 in the D1 and D1 + D2 treatments while significantly more fruit were set at P3 in the D3 and D4 treatments.

Figures 63 and 64 depict the horizontal fruiting profiles of the different damage treatments (split into present and missing fruit in Appendix G), however, this division inadequately explained where compensation occurred (it was expected to occur in the upper canopy). Data will be re-analysed together with data from the 2018/19 season and will be split into more strata to answer more specific questions.

Table 69: Percentage fruit distribution for fruit removal treatments ACRI, 20017/18

Damage	Trt	Mean Fruiting Branches	Yield (Mach Pick) (b/ha)	Potential Fruit Positions	Act Fruit Positions	Retention %/plant	Act Total Missing %/plant	P1 Fruit%	P2 Fruit%	P3 Fruit%	P1 Missing%	P2 Missing%	P3 Missing%
None	Control	16.75	16.88[a]	50.25	29.9	35.41	59.59	21.96	11.40	2.05	30.73	21.30	7.56
D1	Tipping @N5/6	12.7*	13.66[b]	38.1*	22.95	31.33 (-4.08)	58.66 (-0.92)	23.66	7.15*	0.51	38.96	16.64	3.10*
D1 + D2	Tipping @N5/6 Fruit Below N16	14.85	15.62[a]	44.55	26.95	28.68 (-6.72)	71.31 (11.73)	19.39	7.76*	1.53	45.22*	21.62	4.47
D1 + D3	Tipping @N5/6 Fruit Below N12	13.3*	16.19[a]	39.9*	28.25	26.33 (-9.08)	58.66 (-0.92)	14.78	8.45	3.11	33.62	20.36	4.68
D2	Fruit Removal Below N16	15.95	16.63[a]	47.85	30.4	29.85 (-5.56)	70.14 (10.56)	18.13	9.76	1.96	38.89	22.73	8.53
D3	Fruit Removal Below N12	16.25	16.02[a]	48.75	29.3	31.68 (-3.73)	68.31 (8.73)	21.53	9.01	1.14	35.61	25.30	7.40
D4	Fruit Removal Below N12 1st & 2nd Pos.	15.15	11.08[c]	45.45	26.8	25.02 (-10.39)	64.97 (5.39)	18.98	5.05*	0.99	33.00	25.09	6.89
F (p=0.05)		0.004	<0.001	0.044	0.325	0.067	0.086	0.084	0.03	0.039	0.045	0 117	0.019
LSD		2.84	1.751	8.51	n.s.	n.s.	n.s.	n.s.	3.63	1.58	9.15	n.s.	3.45
df		(3, 139)	(6, 27)										

Percentage differences between treatments and the Control shown in red

Table 70: Percentage fruit distribution for fruit removal treatments Spring Ridge, 20017/18

Damage	Trt	Mean Fruiting Branches	Potential Fruit Positions	Act Fruit Positions	Retention %/plant	Act Total Missing %/plant	P1 Fruit%	P2 Fruit%	P3 Fruit%	P1 Missing%	P2 Missing%	P3 Missing%
None	Control	11.7	35.10	22.05	41.54	43.46	22.12	16.13	3.28	25.68	13.62	4.17
D1	Tipping @N5/6	8.25	24.75	18.70	33.56 (7.97)	36.44 (5.10)	20.67	12.51	0.38*	22.35	12.83	1.25
D1 + D2	Tipping @N5/6 Fruit Below N16	10.25	30.75	25.80	31.36 (10.18)	53.64 (-12.10)	18.19	11.38	1.79*	33.67	15.21	4.75
D1 + D3	Tipping @N5/6 Fruit Below N12	9.5	28.50	22.20	25.90 (15.64)	44.10 (-2.56)	13.29	9.66	2.95	27.26	12.15	4.70
D2	Fruit Removal Below N16	12	36.00	21.35	36.51 (5.03)	38.49 (3.05)	18.57	14.50	3.43	21.70	11.59	5.20
D3	Fruit Removal Below N12	10.95	32.85	24.95	32.82 (8.72)	37.18 (4.36)	15.19	12.90	4.73*	19.77	11.85	5.56
D4	Fruit Removal Below N12 1st & 2nd Pos.	10.4	31.20	18.90	22.13 (19.41)	47.87 (-6.33)	11.92	9.32	0.89*	26.39	16.44	5.03
F (p=0.05)		0.587	0.587	0.743	0.101	0.404	0.066	0.306	0.005	0.199	0.693	0.109
LSD		n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	1.201	n.s.	n.s.	n.s.
df		(3, 139)										

Figure 63: Distribution of fruiting positions in the canopy prior to harvest: Fruit damage experiment, ACRI 2017/18 (data includes formed squares/bolls and missing positions)

Figure 64: Distribution of fruiting positions in the canopy prior to harvest: Fruit damage experiment, Spring Ridge, 2017/18 (data includes formed squares/bolls and missing positions)

Discussion

The removal of an average of 120 squares/m between 1st and peak flower (D2 – fruit removal below N16) and just before peak flower (D3 – fruit removal below N 12) did not result in a significant yield loss at ACRI but did cause significant delay. Most of the damage treatments were able to compensate as the lower Namoi is a longer season region meaning there was sufficient heat and radiation during the delay period to mature fruit. This is likely the result of a shift in resource allocation by plants which can expend more energy into growth, having lost a significant fruit load earlier. Once plants develop a larger canopy, they continue to invest in fruiting structures on higher nodes as they have adequate resources to mature them, albeit somewhat later. Though differences in boll numbers were not significant, untipped damaged plants trended to have higher boll loads than the Controls while tipped plants had fewer. Yield increases were due to higher boll weights in damaged plants with tipped and damaged plants having the highest boll weights, hence any yield gains were due to a combination of boll numbers and weights. The removal of about fifty 1st and 2nd position bolls just prior to open boll reduced yield by 5 bales/ha which is not surprising given that plants had invested significant resources into these bolls. Yield was reduced by tipping at Node 5/6 (D1), yet a 3 bale/ha loss is not plausible given that previous data for early tip damage have repeatedly shown that it does not cause yield loss. However, the whole area experienced some herbicide damage early on and it may be that the tipped plants did not recover as well as undamaged plants. Maturity delays occurred in all damages implemented during flowering with 11-12 days being typical. There was no significant maturity delay in early (Tipping) or late (Boll removal) treatments as plants either compensated early or diverted resources into the upper canopy prior to boll opening.

At Spring Ridge, considered a short season area, the removal of an average of 108 squares/m between 1st and peak flower (D2 – fruit removal below N16) and just before peak flower (D3 – fruit removal below N 12) did result in a significant yield loss. All fruit removal treatments resulted in losses of 2-4 bales/ha. These losses were a result of lower boll numbers which were not made up for by higher boll weights. Hence compensation did not occur in this climatic zone. Tipping did not cause yield reductions and did not delay maturity which may indicate that a certain amount of early season thrips damage could be tolerated. Maturity delays from fruit loss were 6-12 days but may be longer depending on seasonal conditions.

In the long season area, controls finished at 147 days, leaving ample warm season length and day degrees for the other treatments to compensate, with the latest not even 165 days. In contrast, the controls in the short season area took almost 180 days to finish, pushing the crop into potentially cooler conditions and shorter day lengths with less radiation and hence, less opportunity for complete compensation in the damaged treatments.

Conclusion

There is a distinct difference between long and short season areas which needs to be considered in pest management guidelines when setting thresholds. Decision making for managers in short season areas is more precarious and less forgiving than for managers in long season areas. In long season areas, the compensation capacity of the crop should be considered when setting mirid thresholds. In cool season areas, protection against thrips, compatible with an IPM approach, should be considered. Over the top spraying with a broad-spectrum insecticide would not be a good option due to the damage to the beneficial population. However, seed treatments or at-planting treatments also have risks in terms of costs and selection of resistance in pests, but on balance are probably less damaging to beneficial populations.

The damage imposed on plants in this experiment was quite extreme as fruit removal in the lower strata temporarily reduced retention to zero. This occurred as plants had begun to invest in young bolls or had already invested musch into older bolls. If retention was reduced to only 30-40% at that time, the results may have shown less delay (ACRI) and less yield loss (Spring Ridge). And if fruit lost below nodes 12-16 can be compensated for, is there a need to spray for mirids between first and peak flower in long season areas? Damage experiments will be refined during the 2018/19 season and will be reported on later in the year.

Outcomes

4. Describe how the project's outputs will contribute to the planned outcomes identified in the project application. Describe the planned outcomes achieved to date.

A. To improve knowledge of and management of SLW by:

(i) Identifying factors contributing to reductions in honeydew on cotton and implications for cotton fibre quality and defoliation

- Rainfall and UV Radiation:

a) Rainfall and overhead irrigation can remove honeydew. Depending on how intensive the rainfall/irrigation, some honeydew may remain. The weather following these events, can hinder or promote sooty mould outbreaks on remnant honeydew. A new research project was developed to understand factors affecting cotton colour, including sooty mould.

b) Extended exposure of honeydew contaminated bolls to UV (solar) radiation (>3-4 weeks) did not breakdown the honeydew.

c) We found that night dew can promote sooty mould formation on contaminated bolls. This outcome reinforces whitefly and harvest management to reduce risks of sooty mould.

d) Bolls with greyish lint were infected with *Alternaria,* not contaminated with honeydew or sooty mould. This information contribututed to understanding of other factors affecting lint colour and potential management strategies.

e) The method of applying artificial honeydew was evaluated and confirmed to reduce variability in the data and ensure the results were reliable.

f) Honeydew mainly contaminates the outer surface of bolls, with little penetrating further. This explains why rainfall/overhead irrigation is effective at removing honeydew and the portion of a boll that can become contaminated with sooty moulds.

g) Mealybug honeydew had very high melezitose concentrations (56-70%) and very low trehalulose concentrations (<1%) and is more like aphid honeydew rather than whitefly honeydew.

h) Fructose, glucose and sucrose were rapidly metabolised by baker's yeast, followed by a slower reaction with trehalulose and a very small reaction with melezitose. Yeast may have

potential in remediation of honeydew, though risks to other fibre properties need consideration.

- Sooty Moulds

a) Treatment combinations of +/- Honeydew and +/- Rainfall showed that rainfall washed off most of the honeydew on contaminated bolls. Nevertheless, residual sugars made those bolls prone to sooty mould infection. This improved our understanding of the benefits and detriments of rainfall in relation to honeydew and sooty mould issues.

b) A scale was developed and validated to describe the severity of sooty mould contamination. This can help define contamination levels with respect to colour downgrades (CSP1703).

c) A significant outcome was the development of two projects investigating insect honeydew related colour issues of cotton: CSP1703 (2016-2018) and CSP1901 (2019-2021).

(ii) ***Identifying seasonal host use for SLW***

a) DNA analysis confirmed that 40 out of 55 host species tested harboured *B tabaci* MEAM1 with both nymphs and adults found

b) Important hosts include members of the Families Malvaceae, Brassicaceae, Cucurbitaceae, Fabaceae, Amaranthaceae, Solanaceae, Convolvulaceae, Asteraceae and Euphorbiaceae.

c) One unknown Bemisia sp. nymph was found on Chamaesyce drummondii (Caustic weed)

(iii) ***Assessing SLW mortality on cotton***

a) There was great varaibilityin SLW detection in the gut contents of predators. Apple dimpling bugs, Brown smudge bugs and Red & Blue betles usually had good detection rates if they ate 3-4 adult SLW in 0.5 to 3 hours.
b) Mite eating ladybird beetles, though observed to consume SLW, showed poor detection rates.
c) Detection is spiders was erratic with many false positives. This could have been associated with spider size, slow consumption rates or primers that did not work well or may even have been degraded prior to entering the gut.

(iv) & (v) ***Undertaking sampling to understand the within plant and within field distribution of SLW adults and nymphs***

a) A seasonal profile of SLW adult distribution on plants was established. Density of adults and nymphs increased with node number from 1800DD while the coefficient of variation decreased. Variability was highest in the top of the canopy at Node 5 and strongly affected by vertical movement of adults on the plant. This confirmed industry concerns about the reliability of sampling at Node 5.

b) Movement of adult SLW from upper to lower nodes appeared to be driven by relative humidity more than by temperature: as RH approached 70%, adult whitefly moved down into the canopy and moved up again as humidity decreased. Understanding this dynamic shift is crucial in developing improved sampling strategies.

c) Nymph populations grew exponentially leading up to the open cotton stage. Variability over time was higher at Node 5 than Node 8. Nymph counts lower down on the plant could add valuable consistency and reliability to sampling protocols.

d) Node 14 emerged as a consistent sampling site with low variability over time for third and fourth instar nymphs. Similar exponential growth profiles were found across different sites. These results strengthened the case for an improved sampling and monitoring system for SLW in cotton.

e) The superiority of nymph sampling in the lower canopy (below Node 10) to assess SLW populations more reliably was confirmed in artificially infested high and low density plots. Node 14 nymph population growth curves for various crops in the Namoi showed that the populations fell into 6 statistically different groups.

f) The Node 14 growth curves offered the best discrimination between seasons, sites and the effect of extraneous influences. A decision matrix can be constructed based on this knowledge. These outcomes led to the creation of Project DAQ1903 to achieve this goal.

B. To provide tools for IPM by:

(i) ***Assessing IPM fit and efficacy of new control options***
Research on the target and non-target effects of new insecticides continued. Fourteen new compounds were evaluated of which six have been added to the 'Impact of insecticides and miticides on beneficials' table in the Cotton Pest Management Guide for 2018/19.

(ii) ***Testing management options for mirids and GVB***
A number of insecticides were evaluated for their efficacy on GVB and mirids and incidental effects on SLW population development. Fipronil (+Salt) and Clothianidin emerged as the best options though both also affected the abundance of various non-target beneficials and flared mites. Yields were higher than the control for both insecticides. Fipronil increased risks from SLW populations that could become critical later in the season if not managed effectively and timely. These experiments highlighted the complexity of studies to understand interactions between insecticides, pests and beneficials in the cotton system, management strategies and crop yield.

(iii) ***Exploring options for alternatives to neonicotinoid seed treatments to control thrips***
A series of experiments assessed the effects of several chemical and biological seed treatments on seedling thrips in cotton. Low thrips abundance generally precluded conclusions about efficacy but thrips larvae were controlled for 3-4 weeks by neonicotinoids. Plant damage was low and did not translate into yield differences or maturity delays. Alternative treatments did not control thrips or improve yield. Neither thiamethoxam nor Thimet controlled *Frankliniella occidentalis* well, possibly indicating tolerance or resistance. Neonicotinoids variably affected predatory beetles, spiders, *Telenomus* and ants across different seasons and treatments containing fipronil tended to flare mites. Since fipronil is used to control mirids, any seed treatment containing it needs to be counted towards permitted number of sprays as per resistance management recommendations. Overall the experiments reinforced previous results that a degree of thrips damage can be tolerated in the Namoi Valley.

(iv) ***Assessing the relationship between boll age and susceptibility to GVB damage***

Male, female and nymphal GVB were caged onto 5, 10 and 30 day old cotton bolls for a week. All caused significant damage to 5 day old bolls, many of which were aborted. Older bolls continued to develop but sustained more staining and tightlocking. Nymphs caused more severe damage than females and males which has implications with respect to their clustering habit upon hatching and where there are high numbers of GVB, such as near watercourses and weedy edges.

(v) ***Improving understanding about insecticides used to manage SLW***
Commercial whitefly insecticides were evaluated in SLW infested plots to improve understanding about their efficacy. Despite artificial infestation, SLW numbers were low preventing a second spray and any meaningful conclusions. A continuation of the Whitefly x Chemistry experiment was hailed out in 2018/19, however we will continue to investigate at the earliest opportunity.

C. Manage early season damage by:

(i) ***Assessing seed treatments and measuring the effect of early season thrips and mirid damage on plant growth, yield and maturity in southern regions***
a) To provide answers to the pest problems experienced by growers in the southern cotton growing areas (Liverpool Plains/Hay Plain) a number of early season experiments were carried out in those regions. Neonicotinoid (Cruiser/Cruiser Extreme/Genero) and other (Thimet/Lorsban) seed treatments did not give a significant improvement at establishment and the delay in establishment at one site (Hay) was likely associated with field and weather conditions rather than with pests. At the other site (Carathool) Thimet provided good control of the wireworms that affected plant stand. At both sites thrips numbers were not affected by seed treatments and the dominating species was *Thrips tabaci*. Consequently, there were also no yield advantages from seed treatments. Thrips numbers at both sites did not exceed 5 thrips/plant, which was too low to draw definite conclusions. It is likely that higher thrips infestations will cause yield losses and under such conditions the performance of seed treatments would stand out.

b) We assisted Sandra Mc Dougall and Jinhua Mo (NSW DPI) in Griffith to carry out thrips damage simulations so they could investigate the impact of early season thrips in the southern growing areas.

c) The first seed treatment experiment in the Liverpool Plains to assess the performance of Cruiser Extreme occurred in less than ideal conditions with poor soil moisture and plant stand and heavy weed infestations. The dominating thrips species was *Thrips tabaci* but Cruiser Extreme did not affect numbers. It provided a small advantage in plant stand though yield data for this crop were not available. An additional dimethoate spray resulted in significantly higher thrips numbers and a shift in species to *Frankliniella occidentalis.* The confounding factors encountered at this site encouraged us to continue studies of early season pests at two other sites with improved management practices.

d) Two seed treatment experiments were set up at Connamara (Pine Ridge) and Dimby Plains (Spring Ridge) to continue early season research in the Liverpool Plains. Seed treatments included Cruiser Extreme and Thimet (at Connamara). Thimet increased plant stand by 1 plant/m at Connamara while Cruiser X had no effect at either site. While thrips numbers were low overall, at Connamara Cruiser X and Thimet both had significantly lower numbers of *Frankliniella occidentalis* and higher numbers of *Thrips tabaci*. Cruiser X reduced jassid

nymphs but also some beneficials including ants. Seed treatments did not affect maturity at either site but at Dimby Plains the Cruiser X treatment yielded an extra 1.1 bale/ha.

e) Low thrips numbers in the southern experiments were the most limiting factor in drawing definite conclusions as to the efficacy of seed treatments in those regions. Based on one successful demonstration of the effect of high thrips numbers on yield at Yarral (CSD Seed Treatment Experiments Final Report), we may surmise that under high thrips pressure, neonicotinoid seed treatments would emerge as effective control measures.

f) Experiments at Connamara and Dimby Plains were carried out in a year of intense insect pressure that prompted growers to spray for mirids and Rutherglen bugs beyond normal spray applications. The general perception was that mirids, and possibly Rutherglen bugs, caused loss of squares and young bolls to the extent that no bolls set below Node 12. The season also brought hot and cold shocks so it was difficult to pinpoint a single cause of square and boll losses. This uncertainty led us to investigate fruit loss and cotton compensation in the 2017/18 season (reported in section G).

D. Understanding Cotton Bunchy Top disease by:

(i) ***Identifying alternative host species***

a) CBT has become a sporadic disease of cotton due to current unfavourable conditions and low host and vector abundance.
b) Two strains of CBTv were previously identified, CBTvA, which is non-pathogenic on its own, and CBTvB which is required to manifest disease in a host, either alone or in combination with CBTvA.
c) The family Malvaceae contains the most important hosts of CBTv: *Gossypium hirsutum* ratoon, *Malva parviflora*, *Abutilon theophrasti* and *Anoda cristata* are particularly good harbourers of the disease and occur in and around cotton crops. Other Malvaceae such as *Malvastrum coromandelianum* and *Sida rhombifolia* are poor carriers, probably due to being poor aphid hosts. *Hibiscus sabdariffa* on the other hand is a moderately good host for cotton aphid but a poor disease host.

(ii) ***Investigating the effectiveness of insecticide application to prevent spread of CBT***

a) Cruiser was not effective in preventing transmission of CBT regardless of whether CBTv infected aphids colonised at 1 or 4 true leaves. Cruiser Extreme, however, could reduce the infection rate by 50% compared with the controls both at 1 and 4 true leaves.

b) Application of insecticide (Transform) before a known migration of CBT affected aphids into cotton could effectively reduce the rate of transmission of CBTv resulting in a lower level of infection.

E. Identifying and managing emerging pests by:

(i) ***Providing flexibility to undertake research to manage emergent/exotic pests, including those arising due to changes in the farming system***

a) Thrips: Some consultant believe that high abundance of thrips late season damages flowers causing them to shed and reduce yield. New research was initiated to address this (reported in section F).

b) Symphyla became a concern in particular fields in the Gwydir, Namoi, Darling Downs and Theodore in 2013/14. Investigations revealed that well mulched, friable and fertile soils contained a number of small arthropods amongst them symphilids. One of the symptoms in cotton was a "witches broom" root type. Heimoana researched the pest and presented during an information session in Moree with growers and consultants who gave feedback on the problem. Some had solved their issue with broad spectrum soil insecticides and replants. The problem has not been reported in any season since.

c) Rutherglen bug (RGB): Questions about the pest status of RGB in cotton have recurred over several seasons, especially where large numbers of nymphs and adults move from scenescent canola to adjacent cotton. Inappropriate management of RGB risks IPM in crops. To clarify if RGB adults caused square losses, 20 or 50 RGB were caged onto 5 day old bolls and checked after 1 week. While there were puncture marks on the outside of green bolls, the damage did not penetrate the boll wall (Fig. 55). We will continue to look for opportunities in the future to answer these questions.

d) SLW: Several meetings with growers and consultants were held in 2013 to discuss whitefly management alerted us to the inadequacy of Node 5 sampling in some areas. We initiated a collaborative research effort with Dr Richard Sequeira and Susan Maas to re-assess current sampling protocols (see Section A (iv) & (v)).

e) Growers also expressed concerns that honeydew and sooty mould covering leaves could potentially impact on the efficacy of defoliants. This will be opportunistically fitted into the new IPM and Cotton Colour projects (2019-2021).

f) Pesticide application: consultants have expressed concern about the penetration of insecticides into dense canopies. A meeting Heimoana had with chemical company reps regarding pesticide failure reiterated this problem. Intensive management of high yielding crops has created dense, closed canopies that hinder penetration of non-systemic insecticides and have implications on SLW management. Heimoana consulted with Peter Walters from United Phosphorus Ltd (UPL) to design an experiment that would test penetration of chemicals into the canopy when sprayed at different volumes by plane or ground rig. Experiments were carried out by Emma Ayliffe (Summit Agriculture, Griffith) and Peter presented results at a CCA meeting in Moree. Heimoana has discussed some ideas using the possibility of triggered release formulations of systemic insecticides with Susan Maas, however, more investigation into the feasibility of this idea needs to be done.

f) Biosecurity: Wilson and Heimoana remained involved in cotton biosecurity attending workshops and meetings. This included contributing to biosecurity protocols for potential invasive species such as MED biotype whitefly, Brown marmorated stink bug, Bagrada bug and *Spodoptera frugiperda*. Heimoana continues to participate in the Cotton Biosecurity Committee, having replaced Lewis Wilson after his retirement. During the winter we responded to queries about *Diuraphis noxia* (Russian wheat aphid) and had SLW from wheat DNA tested (*B. tabaci* MEAM1).

g) Solenopsis mealybug : mealybugs have crossed the northern border and re moving south having now been reported from St. George, the Macintyre, Gurley, south of Gunnedah (not

Solenopsis), Boggabri and the Namoi (Culgoora). There is concern about the spread of Solenopsis and uncertainty and inexperience about mealybug management in these areas, hence it may be beneficial to include the topic, tailored for this region, in future CCA meetings or workshops.

h) Mites: we experienced the first major strawberry spider mite outbreak at ACRI in 2017/18 and since we had artificially infested some experiments with two-spotted mites, their combination caused sufficient damage to warrant control. Questions about their damage potential and threshold levels remain relevant in particular if they are replacing two-spotted spider mites across the industry. Early in the 2018/19 season Rob Holmes (Consultant, Moree) sent through pictures of an unknown mite and its severe damage to cotton seedlings. The species resembled red-legged earth mite, however its host and range were incongruent with its activity in Moree cotton. Tanya Smith sent the mites for identification but results are yet to be received.

i) In 2016/17 the industry experienced heavy insect pressure after a wet winter and a cool spring that supported early insect populations and delayed crop development. In response several growing regions formed Area Wide Management (AWM) groups which worked on reducing or changing mirid sprays and improving communications. Heimoana attended a meeting with consultants in Walgett where the feasibility of AWM was discussed. The introduction of a 30 day single application pyriproxifen window by the Transgenic and Insecticide Resistance Management Committee aimed to slow the build-up of SLW resistance to pyriproxifen. In 2017/18 fipronil sprays, in many cases, were replaced by softer options (e.g. sulfoxaflor) lessening the impact on beneficials. This example shows the potential for peer to perr communication and commitment to help change practice.

F. Investigating the effect of late season thrips damage to flowers on yield

We found no consistent pattern of yield reduction due to leaf loss, flower removal or the combination of the two at the damage levels inflicted. More severe flower loss could reduce yield and we are collaborating with Paul Grundy to investigate this in the new IPM project 2019-202.

G. Investigating the effect of early tip and fruit damage on yield and maturity of Bollgard 3

To assess the damage tolerance of Bollgard 3 cotton to tip damage and fruit loss, plants were tipped at Node 5/6 and/or had fruits removed from the first 16 fruiting branches. Later treatments included fruit removal from the first 12 fruiting branches – either all fruits or only position 1&2 fruits. These experiments were done in a long season area (Narrabri) and a short season area (Spring Ridge). In the long season area, machine picks showed that most treatments recovered without yield loss, though there was delay in maturity. In the short season area hand picks showed that all fruit removal treatments had significantly reduced yield and delayed maturity. Across both regions compensation occurred by a combination of more bolls and/or heavier boll weights, though compensation was incomplete in the short season cooler region.

Other activites

Lewis Wilson and Simone Heimoana also contributed as a committee members to the TIMS Committee, the TIMS Insecticide and TIMS BT-Cotton Technical Panels, to REFCOM and the Australian Cotton Industry Bio-security Committee.

Lewis Wilson retired in April 2018 after 33 years in the cotton industry and was bid farewell in an official function well attended by industry members, to honour his dedication and achievements. And we had fun as well.

5. Please describe any:-

a) technical advances achieved (e.g. commercially significant developments, patents applied for or granted licenses, etc.); n/a

b) other information developed from research (e.g. discoveries in methodology, equipment design, etc.); n/a and

c) required changes to the Intellectual Property register; n/a

6. Provide an assessment of the likely impact of the results and conclusions of the research project for the cotton industry. What are the take home messages?

This project has fulfilled the majority of its objectives and provided knowledge on important IPM issues that have challenged the cotton industry. The project has

(a) provided a comprehensive understanding of the issues surrounding insect honeydew: its composition, stickiness characteristics, deposition patterns and distribution in the canopy, its ability to be washed off by rainfall and its suitability as a fungal substrate under certain conditions,
(b) begun to investigate the effect of honeydew on cotton lint colour, leading to two new Cotton Colour projects (CSP1703 & CSP1901).
(c) continued to provide information on the seasonal abundance and host use of SLW
(d) provided further information on the survival of SLW in cotton, mortality factors and potential predators
(e) investigated and validated the inconsistencies of SLW sampling, especially in central and southern areas, research which provided the basis for the new SLW Validation project DAQ 1903,
(f) evaluated target and non-target effects of a range of synthetic and biologically based insecticides,
(g) investigated management options for the control of GVB and mirids with consideration of their effects on beneficials and SLW,
(h) shown that tested alternative non-chemical options to replace neonicotinoid seed treatments do not adequately control thrips,
(i) provided information on the effect of neonicotinoid seed treatments on thrips and beneficials,
(j) assessed the damage done to bolls of various ages by GVB adults and nymphs,
(k) begun to improve our understanding of the effects of various chemicals used in SLW control,
(l) demonstrated no direct relationship between cumulative mirid numbers and yield parameters, though this needs more work,
(m) provided base information on the effect of thrips on seedling establishment in southern regions and how seed treatments may be of benefit,
(n) identified new hosts for cotton bunchy top disease
(o) evaluated the effect of seed treatments and a foliar applied insecticide on transmission of cotton bunchy top disease

(p) rapidly provided information where growers and consultants had critical questions, e.g. symphyla, Rutherglen bugs, strawberry spider mites, mealybugs, SLW, reports of insecticide failure, etc.,
(q) provided answers to the questions about late season thrips damage and has
(r) begun to investigate the effects of flower loss on yield.

The research in this project provides the basic knowledge that allows the industry to adjust pest management to new or improved crop management practices, changing pest scenarios and yield expectations. One of the most important outcomes is the re-evaluation of the SLW sampling methodology and the validation of more accurate sampling sites on the plant and SLW life stages to better predict populations, which will lead to better SLW management. This will eventually be underscored by an improved understanding of the insecticide options for SLW. The implications of better SLW management will greatly impact on lint quality and colour issues which have emerged in recent years. Despite research into remedies for honeydew related stickiness and discolouration of lint, timely and effective SLW management to prevent honeydew are the key to maintaining the industry's reputation for top quality, high value cotton. The research on the impact of predators and parasitoids on SLW has highlighted that its effective long term management can only occur through the incorporation of integrated pest management practices into the farming system. The continued testing of new insecticides supports IPM by providing the industry with IPM compatible choices. The 2016/17 high pest season has shown how quickly the industry can descend back into the "cycle of continual sprays" that ends with resistance, high costs and limited options. Underlying successful SLW management is the management of mirids. While there are chemical options to reduce the detrimental effects of consecutive fipronil spray on beneficials, demonstrating tolerance of Bollgard 3 to early fruit loss would reduce the need for early sprays and allow for the build-up of beneficials in the system. This is the ultimate aim of the fruit loss experiments. As we have seen with SLW sampling, thresholds do not apply across every cotton growing region and in order to provide regionally valid answers to common issues we have collaborated with Sandra Mc Dougal in the south (early season thrips work) and are currently collaborating with Paul Grundy and Richard Sequeira in central and northern regions, the outcomes of which will benefit the industry by being able to better relate to experimental results. Through our involvement with biosecurity and industry issues we have responded rapidly to issues of critical importance to the industry by providing extension material, fielding direct questions and visiting farms where problems occurred. This project has contributed valuable information on the management of sucking pests, an important component of the cotton pest complex, and the subsequent IPM project will build on the outcomes achieved here.

Extension Opportunities

1. Detail a plan for the activities or other steps that may be taken:
(a) to further develop or to exploit the project technology.
(b) for the future presentation and dissemination of the project outcomes.
(c) for future research.

Steps taken:
(i) The efficacy and IPM fit of new chemistry has been published for industry.
(ii) Wilson and Heimoana have presented to the CCA winter meeting at least once and often twice in each year of the project – discussing issues such as Symphyla, Rutherglen bugs, compatibility of cotton insecticides with bees, SLW management, early season pest

management, mite ecology and management, assessing the IPM fit of new insecticides, cotton damage results and movement of pests within the canopy.

(iii) Heimoana and Sandra Williams spoke to growers at Walgett regarding AWM implementation,

(iv) Heimoana and Smith have provided training in identification of mites, aphids, SLW and thrips to visiting groups of agronomist – including for Auscott each year.

(v) Heimoana and Smith have presented information on pest sampling, in collaboration with Sandra Williams, to consultants and agronomists in the Macquarie, Griffith, Moree, Narrabri and Warren during the project,

(vi) Wilson and Heimoana have answered frequent phone calls and email requests for identification of pests or discussions on management of particular situations. These requests come from widely across the industry and at peak times in the early season and in Jan –March can be 6-7 calls each week, and sometimes 2-4 per day.

(v) Wilson and Heimoana have spoken at field days at Griffith, Moree (x3), and have supported the CottonInfo team with requests to contribute to industry driven meetings or farm visits – for instance two meetings in late 2013 in Moree in response to problems with symphyla,

(vi) Wilson and Heimoana have presented research results at Australian Cotton Conferences and Australian Cotton Scientist Conferences.

Steps that need to be taken:

Please note that some of these correspond to steps previously identified but due to Lewis Wilson's retirement have not been written up.

(i) Information on the effect of late damage on yield (6 year's data now) needs to be packaged up into a Cotton Grower article for industry and published in a scientific journal.

(ii) Information on SLW host use and mortality factors needs to be published for industry.

(iii) New information on the interaction between seed treatments and foliar sprays and transmission of CBT can be made available to industry though a second year of data may would increase confidence in the results

(iv) Information on the interaction between transgenes, leaf shape and mirids sprays and implications for SLW and other secondary pests need to be packaged for industry and published in a journal.

(v) Outcomes from Dr Heimoana's thesis are being written up for scientific publication but should also be published for industry

(iv) Outcome of studies with the fate of honeydew have been widely disseminated verbally but need to be published for industry and scientifically.

(v) Information on GVB damage in relating to boll age need to be written up as a Cotton Grower article.

For future research.

Future research is detailed in a new project that has been funded by CRDC, CSP1905 "IPM to support the management of emerging pests". Aims for that project are to improve IPM adoption by scrutinizing invertebrate communities and insect/plant interactions that support sound IPM practice. Components of the project include:

1) Investigations into the impact of new insecticides on target pests and beneficials;

2) Identification of how cotton invertebrate communities (including "others" and ground-dwelling invertebrates) counter invasion and proliferation of pests including mealybugs;
3) Evaluation of the capability of Bollgard 3 to compensate for early tip damage and fruit loss in different climatic regions; measurement of the impact on fruit loss of physiological stress interacting with pest damage and testing these findings on industry leading farms;
4) Assessing the efficacy of various chemicals available for the control of SLW and consider their best fit in SLW management
5) Improving tactics to encourage the use of IPM principles.

However, a few other issues have also been identified that would warrant further investigations:

1. Continuing studies into our understanding of sooty moulds and possible remedial actions. This is being addressed in the CRDC project CSP1901 "Reducing the impact of weather, insects and microbes on cotton colour".
2. Assessing the impact of honeydew and sooty mould on the efficacy of defoliants. We will try and include this in the Cotton Colour project CSP1901.
3. Validation of the improved sampling methodology for SLW. This is being addressed in the CRDC project CSP1903 "Improved management of silverleaf whitefly on cotton farms".
4. Testing seed treatments under situations of high thrips pressure. This would assess whether neonicotinoid seed did provide sufficient protection from thrips and wireworms, especially in southern cotton growing regions. A strategically placed experiment could provide further answers.
5. Investigations into the relationship between cumulative mirid damage and yield parameters. We still do not have a quantitative understanding of how mirids contribute to yield loss.
6. Pesticide application into dense Bollgard 3 canopies. We need to find ways of improving the application of insecticides late in the season when SLW control may be critical to cotton quality.
7. Further investigations into the pest status of Rutherglen bugs in cotton, work which will need to be done opportunistically.

9. A. List the publications arising from the research project and/or a publication plan. (NB: Where possible, please provide a copy of any publication/s)

Scientific

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3. Lytton-Hitchins, J.A. Greenslade, P., Wilson, L.J. (2015) Effects of season and management of irrigated cotton fields on Collembola (Hexapoda) in New South Wales, Australia. Environmental Entomology 44**:** 529-545
4. Marshall, K.L. Collins, D; Wilson, L.J., Herron, G.A. (2015) Efficacy of two thiamethoxam pre-germination seed treatments and a phorate side-dressing against

neonicotinoid- and pirimicarb-resistant cotton aphid, Aphis gossypii (Hemiptera: Aphididae). Austral Entomology 54: 351-357
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1. Heimoana, S.C., Wilson, L.J and Smith, T.M. (2017). Management of emergent pests in cotton: refining experimental techniques for valid outcomes. Proceedings Australian Cotton Scientist Conference, Canberra, 05 - 07 September 2017.
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6. Heimoana, S. and Wilson, L. (2014). The fate of honeydew in cotton and impacts. In: 17th Australian Cotton Conference; 5-7 August 2014; Broadbeach. Cotton Research and Development Corporation.
7. Wilson, L. and Heimoana, S. (2014). Late season damage – worth worrying about? In: 17th Australian Cotton Conference; 5-7 August 2014; Broadbeach. Cotton Research and Development Corporation.
8. Wilson, L. and Smith, T. What's killing whiteflies? In: 17th Australian Cotton Conference; 5-7 August 2014; Broadbeach. Cotton Research and Development Corporation.
9. Herron, G., Suan, M., Woolley, L., Chen, Y. and Wilson, L. (2014) Resistance management of cotton aphid, two-spotted mite and mirids. In: 17th Australian Cotton Conference; 5-7 August 2014; Broadbeach. Cotton Research and Development Corporation.
10. Heimoana, S.; Wilson, L.; Constable, G. and George, D. (2013). The effect of honeydew on photosynthesis in cotton. Poster at the 1st Association of Australian Cotton Scientists Conference. (Narrabri, September 2013).
11. Heimoana, S.; Wilson, L.; Constable, G. and George, D. (2013). The effects of aphids on photosynthesis in cotton. In: Proceedings of the 1st Association of Australian Cotton Scientists Conference. (Narrabri, September 2013).
12. Wilson, L.; Heimoana, S.; O'Shea, M.; De Barro, P.; Priest, M. (2013). The fate of honeydew in cotton. In: Proceedings of the 1st Association of Australian Cotton Scientists Conference. (Narrabri, September 2013).
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2. Heimoana, S.; Wilson, L. (2017). How much rain is too much? The impact of rainfall on cotton colour grade. The Australian Cotton Grower 38(1):28-30.
3. Herron, G. and Wilson, L. (2016). Mite resistance danger from over-use of abamectin. The Australian Cottongrower 37(1): 14-16.
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5. Wilson, L. and Heimoana, S. (2015) Late-season pest damage – worth worrying about? The Australian Cotton Grower 36(1):46-48.
6. Sharman, M., Wilson, L., Smith, T., Grundy, P. and Webb, M. (2014) Cotton bunchy top disease and related biosecurity threats. The Australian Cotton Grower 35(3):30-31
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Spotlight Articles featuring project staff

1. **2017** Heimoana, S.; Wilson, L.J. et al. (2017). Impact of insecticides and miticides on predators, parasitoids and bees in cotton. In: The Australian Cotton Industry CottonInfo Team (eds) Cotton Pest Management Guide 2017-18. Cotton Research and Development Corporation, pp 7-9.
2. **2017** Know who's on your team. Spotlight on Cotton, Spring 2017. Cotton Research and Development Corporation. Australian Government, pp. 23-25. Featuring: Smith, T.M. and Hopkinson, J.
3. **2017** Cotton knows how to compensate. CRDC Spotlight Magazine, Autumn, 2017, pp 9-11. Featuring Wilson, L.
4. **2016** Keep IPM on your agenda. CRDC Spotlight Magazine, Summer, 2016, pp 9-11. Featuring Maas, S., Williams, S., Hopkinson, J., Herron, G., Wilson, L., Bird, L., Sequeira, R
5. **2015** Industry takes proactive approach to whitefly management. CRDC Spotlight Magazine, Autumn, 2015, pp 24-25 Featuring: Ceeney, S., Wilson, L., and Sequeira, R.
6. **2015** Integrated pest management: overcoming industry's greatest threat. CRDC Spotlight Magazine, Summer, 2015, pp 10-11. Featuring: Wilson, L.
7. **2014** Link to researchers vital for success. CRDC Spotlight Magazine, Winter, 2014, pp 14 Featuring: Wilson, L
8. **2014** Managing whitefly in the Macquarie. CRDC Spotlight Magazine, Winter, 2014, pp 21 Featuring: Thomas, A., Sequeira, R., Wilson, L. and Parlato, D.
9. **2013** Avoiding large discounts: Maintaining our reputation for quality by managing silverleaf whitefly. CRDC Spotlight Magazine, December-January, 2013, pp 6-7 Featuring: Wilson, L. and Heimoana, S.
10. **2013** Know when cotton aphids will affect your yield. CRDC Spotlight Magazine, December-January, 2013, pp 8-9 Featuring: Williams, S., Clancy, L., and Wilson, L.

Other extension activities (indicative not exhaustive list).

1. Heimoana, S. (2016). Honeydew and sooty mould update on cotton. In: Cotton Consultants Australia Whitefly Seminar; December 2016; Moree. Cotton IPM Project.
2. Heimoana, S. (2014). Symphyla. In: Cotton Consultants Australia Special Seminar; June 2014; Moree. Cotton IPM Project.

3. 2014 - Simone Heimoana provided training to Auscott staff in insect identification, sampling and thresholds
4. 2014 - Tanya Smith and Sandra Williams provided training in insect ID to agronomists in the Macquarie Valley
5. Heimoana and Smith presented and assisted at the IPM Course, Moree (21/08/17)
6. Heimoana and Smith presented and assisted at the IPM Course, Narrabri (26/08/17)
7. Heimoana and Smith presented and assisted at the IPM Course, Warren (13/09/17)
8. Smith presented and assisted at the IPM Course, Griffith (19/09/2017)
9. Heimoana presented at the UNE Cotton Production Course, Narrabri (14/09/17)
10. Heimoana trained Monsanto staff in thrips identification, Toowoomba (26/09/17)
11. Heimoana and Smith conducted an IPM training course for AUSCOTT bug checkers, Narrabri (23/11/17)
12. Heimoana and Smith attended the follow up field training for the Southern IPM course, Griffith (08/12/17)
13. Heimoana attended and presented at the Australian Cotton Scientist Conference, Canberra (05-07/09/17

B. Have you developed any online resources and what is the website address?

n/a

Part 4 – Final Report Executive Summary

Provide a one page Summary of your research that is not commercial in confidence, and that can be published on the World Wide Web. Explain the main outcomes of the research and provide contact details for more information. It is important that the Executive Summary highlights concisely the key outputs from the project and, when they are adopted, what this will mean to the cotton industry.

This report presents and summarizes the outcomes of five years of research into enhancing integrated pest management (IPM) in cotton production systems. Multi-year field and laboratory experiments aimed to answer pest management related questions developed with industry input and relevant across the different cotton growing regions. Analysis and synthesis of experiments have shown a number of important outcomes:

i. We have further unravelled the myriad factors influencing the effect of insect honeydew on fibre quality and factors that contribute to its reduction in the field. The presence of insect honeydew in cotton is complex and problematic since the remedial rainfall that reduces stickiness in cotton, has the potential, under some circumnstances, to increase the risk of sooty mould development, another factor that diminishes cotton quality and can lead to penalties.
ii. The host range of SLW was further confirmed and key hosts identified which is critical when considering management strategies to reduce overwinter survival and predict risks from outbreaks.
iii. Studies on the retention of SLW DNA in the gut of key predators wers completed and will be used to identify the most significant predators.
iv. Several new weed hosts of this disease were confirmed, potentially improving prediction of seasons with higher risks from CBT.
v. Seed treatments such as Cruiser would not be effective in preventing transmission of CBT but Cruiser Extreme could reduce the infection rate by 50%. Application of folair sprays against aphids just after aphids entered the crop, or 24 hours after they entered

the crop would not be effective at preventing initial transmission but may retard secondary transmission..

vi. We found in the central and southern cotton areas, the currently employed SLW sampling methodology inadequately predicts populations, potentially reducing the timeframe for optimal management decisions. The groundwork for development of new strategies was completed and will be developed in a subsequent project.

vii. The IPM fit of new insecticides was assessed and a number of new IPM compatible insecticides have been added to Table 3: "Impact of insecticides and miticides on predators, parasitoids and bees in cotton" in the Cotton Pest Management Guide.

viii. Fipronil and clothianidin were compared as management options for Green vegetable bug (GVB) and mirids. Both compounds were effective against target pests and improved yeild, but both also had negative impact on some predators (including Coccinellids) and one had high potential to flare mites. The complexity of these experiments highlighted the challenges involved in pest management decisions.

ix. Thrips control with alternative seed treatment options to neonicotinoids was poor, indicating that these biologicals could not replace currently used chemicals. In new cooler regions thrips larvae were controlled by neonicotinoid seed treatments but benefits to yield did not occur probably due to low thrips abundance.

x. Assessing the relationship between boll age and susceptibility to GVB damage. GVB nymphs and adults inflicted most damage on five day old cotton bolls which usually aborted within days of being damaged, while older bolls continued to develop but exhibited staining and tightlocking.

xi. Evaluating various insecticides used to manage SLW to understand their different modes of action We were unable to assess the effects of SLW insecticides as low whitefly numbers precluded conclusions resulting from a single application of whitefly insecticides, though our efforts will be continued in the future.

xii. Providing expert advice to consultants and growers throughout the season and rapid responses to critical pest problems Expert advice was provided to growers and consultants throughout the year via phonecalls, e-mails and personal interactions. Support was given particularly in 2016/17, a high pest year that threw up many questions about occasional pests and their management.

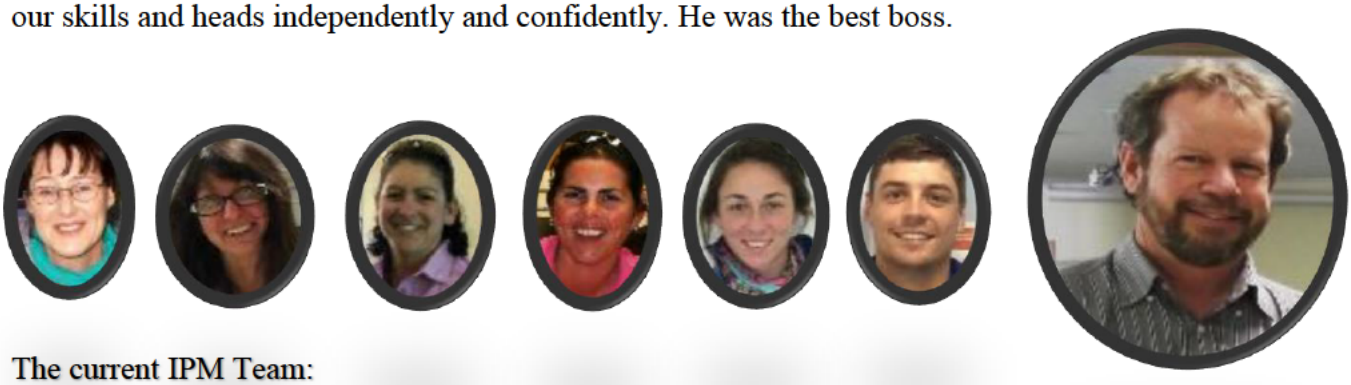
xiii. Investigating the simulated effects of early and late flower damage by thrips and mirids on yield and maturity With respect to simulated thrips damage, the removal of one weeks' worth of flowers at peak flower or cut-out was generally not severe enough to cause yield loss. More extreme, early season flower removal that simulated mirid damage had variable results on yield and maturity depending on the severity of the treatment and the regional climatic conditions, with higher risk of yield loss in southern areas. These experiments will be repeated across regions for verification.

The outcomes of this project have added to the current understanding of cotton pests, their assessment and impact on plants as well as their management and control options. They have also contributed to the better understanding of plant responses to pests which may potentially change management practices of some pests. Further, they have highlighted the interactions between plants, insects and other organisms, and the climatic factors that affect these interactions. What has become clear through the regional experiments is, that insect management decisions in the different cotton growing areas are governed by season length, and that the wrong decision in a short season area can have great consequences with respect to yield loss. These outcomes reinforce the importance of IPM in the cotton system and, once published and extended to the industry, will guide growers and consultants to make more

informed decisions and perhaps influence the degree to which some pests are managed and tolerated.

Part 5 – Achnowledgements

We would like to acknowledge the excellent leadership that Dr. Lewis Wilson (now retired) has provided to his team in the past 20 years. He has taught us good experimental methodology and guided us in our scientific thinking with enthusiasm, patience and humour. While his forward thinking often gave us extra work, it always paid off in the extra knowledge we gained and the preparedness for new things. He encouraged us to learn as much as we could and use our skills and heads independently and confidently. He was the best boss.



The current IPM Team:
Simone Heimoana, Tanya Smith, Ammie Foster, Dee Hamilton, Tianne Parker, Matt Nott

We also want to acknowledge the efforts of staff who temporarily supported us each season: Zane Stahn, Blake Hilderson, Mark Laird, Giulio Heimoana and others.

We thank our collaborators for providing analytical services which we could not access locally: Dr. Michael O'Shea (BSES, Brisbane), Dr. Anne Rae and Dr. Donna Glassop (CSIRO, QBP, St. Lucia)

For financial support we are grateful to the Cotton Researchand Development Corporation for sponsorship of this project.

APPENDIX 1: Section A(i) b)

2015/16 UV Experiment 4: this was also a repeat of Experiment 2 to clarify any packing or handling issues that could reduce honeydew from the freezer controls. It was set up the same as Experiment 2, except that this time the control bolls for the freezer were also pinned on the wash stand and then removed to ensure that all bolls were handled in the same way. They were then kept in trays instead of plastic bags. Experiments had shown that handling bolls with plastic gloves during pinning could remove between 0-25% of the honeydew (mean 12.1%). Frozen samples were taken to QBP for analysis, however, Donna contacted me later to say that QBP had a power outage over one weekend and that some of our samples had thawed out and that any analysis would be unreliable as the sugars in the warm samples would most likely have been degraded by micro-organisms. We therefore decided not to pursue the time-consuming and costly analysis.

2016-17 UV Experiments: Honeydew experiments were terminated since Dr. Anne Rae's project at CSIRO St. Lucia ended and the HPLC that had been used to analyse our samples was not serviced anymore. To cover for the continued use of the HPLC would have cost around $35,000 (including service, new columns and consumables).

APPENDIX 2: Section A(ii)

Table 1: Silver leaf whitefly (B tabaci MEAM 1) host testing: DNA analysis by Susan van Brunschot to confirm species collected from plant hosts. More important breeding hosts are in bold italics. A 'no' record for nymphs or adults indicates we were not able to find that whitefly stage on that host during the survey period. Where a result was inconclusive, Sharon tested the whitefly stage (adult or nymph) from the host but didn't return a result.

Plant Species	Common Name	Positive for nymphs of B. tabaci MEAM1	Positive for adults B. tabaci MEAM1
Sonchus oleraceus	**Milk Thistle**	**yes**	**yes**
Rapistrum rugosum	**Turnip weed**	**yes**	**yes**
Hibiscus trionum	**Bladder Ketmia**	**yes**	**yes**
Citrullus lanatus	**Camel melon/Water melon**	**yes**	**yes**
Chamaesyce drummondii	**Caustic weed**	**yes**	**yes**
Cucumis melo	**Ulcardo Melon**	**yes**	**yes**
Datura ferox	**Fierce Thornapple**	**yes**	**yes**
Ipomoea lonchophylla	**Common Cowvine**	**yes**	**yes**
Physalis ixocarpa	**Annual Ground Cherry**	**yes**	**yes**
Amaranthus macrocarpus	**Dwarf Amaranth**	**yes**	**yes**
Medicago polymorpha	**Burr Medic**	**yes**	**yes**
Amaranthus viridus	Green Amaranth	yes	yes
Lactuca serriola	Prickly Lettuce	yes	yes
Lamium amplexicaule	Deadnettle	yes	yes
Malva parviflora	Marshmallow	yes	yes
Solanum nigrum	Blackberry Nightshade	yes	yes
Tribulus micrococcus	Yellowvine	yes	yes
Tribulus terrestris	Caltrop	yes	yes
Urtica urens	Annual Nettle	yes	yes
Xanthium occidentale	Noogoora Burr	yes	yes
Bidens subalternans	Beggar's Tick	yes	yes
Euphorbia davidii	David's Spurge	yes	yes
Helianthus annuus	Sunflower	yes	yes
Rhyncosia minima	Rhynco	yes	yes
Ricinus communis	Castor Bean	yes	yes
Polygonum aviculare	Wireweed	yes	yes
Anoda cristata	Anoda weed	yes	yes
Bidens pilosa	Cobbler's Peg	yes	yes
Brassica rapa	Turnip	yes	yes
Glycine max	Soybean	yes	yes
Gossypium hirsutum	Cotton	yes	yes
Tetragona tetragonioides	New Zealand Spinach	yes	yes
Vigna radiata	Mungbean	yes	yes
Macroptylium lathyroides	Phasey bean	yes	yes
Verbena bonariensis	Purpletop	yes	yes
Sida rhombifolia	Paddy's Lucerne	yes	no

Einadia nutans	Climbing Saltbush	yes	no
Malvastrum coromeliandrum	False Mallow	yes	no
Chenopodium pumilio	Small Crumbweed	inconclusive	yes
Convolvulus erubescens	Australian Bindweed	inconclusive	yes
Boerhavia diffusa	Tarvine	inconclusive	yes
Fallopia convolvulus	Black Bindweed	no	yes
Haloragis aspera	Raspweed	yes	yes
Trianthema portulacastrum	Black Pigweed	inconclusive	yes
Portulaca oleracea	Pigweed	no	yes
Argemone ochroleuca	Mexican Poppy	no	yes
Commelina cyanea	Wandering Jew	no	yes
Cucumis myriocarpus	Prickly Paddy Melon	no	yes
Datura inoxia	Downy Thornapple	no	yes
Echium plantagineum	Paterson's curse	no	yes
Erodium crinitum	Stork's Bill	no	yes
Gomphrena celosioides	Gomphrena weed	no	yes
Solanum esuriale	Quena	inconclusive	yes
Vicia faba	Broadbean	no	yes
Abutilon theophrasti	Velvetleaf	no	yes
Ipomoea plebeia	Bellvine	yes	inconclusive
Silybum marianum	Variegated Thistle	inconclusive	inconclusive
Xanthium spinosum	Bathurst Burr	no	yes
Zea mays	Corn	no	inconclusive
Vicia sativa	Vetch	no	yes
Einadia hastata	Berry Saltbush	no	no
**Triticum aestivum*	Wheat	inconclusive	yes

*eggs and adults analysed by Wanaporn Wongnikong

APPENDIX 3: Section A(iii)

Table 1. Detection of SLW DNA in adult Apple dimpling bugs fed, 0, 1,2,3 or 4 SLW adults.

SLW eaten	**Total ADB testing positive**	**% ADB testing positive**	**Average time taken to feed/until preservation (mins)**	**Total insects tested**
0	0	0	0	6
1	4	40	57	10
2	6	86	114	7
3	2	100	155	2
4	no test	no test	no test	0

Table 2. Detection of SLW DNA in adult Mite-eating ladybeetles fed, 0, 1,2,3 or 4 SLW adults.

SLW eaten	**Total MELB testing positive**	**% MELB testing positive**	**Average time taken to feed/until preservation (mins)**	**Total insects tested**
0	0	0	0	10
1	5 (4)*	46 (36)*	22	11
2	4 (3)*	31 (23)*	118	13
3	7 (5)*	53 (38)*	157	13
4	2 (1)*	25 (13)*	207	8

* 5 samples showed very faint positives (if calculated as negative, result in bracket applies).

Table 3. Detection of SLW DNA in adult Red and Blue beetles fed, 0, 1,2,3 or 4 SLW adults.

SLW eaten	**Total R&B testing positive**	**% R&B testing positive**	**Average time taken to feed/until preservation (mins)**	**Total insects tested**
0	0	0	0	10
1	7	70	6	10
2	9	90	7	10
3	12	100	17	12
4	13	100	34	13

Table 4. Detection of SLW DNA in adult Lynx spiders fed, 0, 1,2,3 or 4 SLW adults.

SLW eaten	**Total Lynx testing positive**	**% Lynx testing positive**	**Average time taken to feed/until preservation (mins)**	**Total insects tested**
0	1	10	0	10
1	3	25	75	12
2	8	62	129	13
3	10	91	106	11
4	5	63	81	8

Table 5. Detection of SLW DNA in Nightstalker spiders fed, 0, 1,2,3 or 4 SLW adults in test 1.

SLW eaten	Total NS testing positive	% NS testing positive			Average time taken to feed/until preservation (mins)	Total NS tested
		All NS	Spiderling NS	Large NS		
0	3	30	50	28	0	10
1	3	25	27	0	12	12
2	6	50	55	33	16	12
3	9	69	60	100	29	13
4	10*	83	90	50	36	12

Table 6. Detection of SLW DNA in hatchling Nightstalker spiders fed, 0, 1,2,3 or 4 SLW adults in test 2.

SLW eaten	Total NS testing positive	% NS testing positive	Average time taken to feed/until preservation (mins)	Total NS tested
0	1	10	0	10
1	8	62	31	13
2	11	100	59	11
3	12	92	99	13
4	11	100	134	11

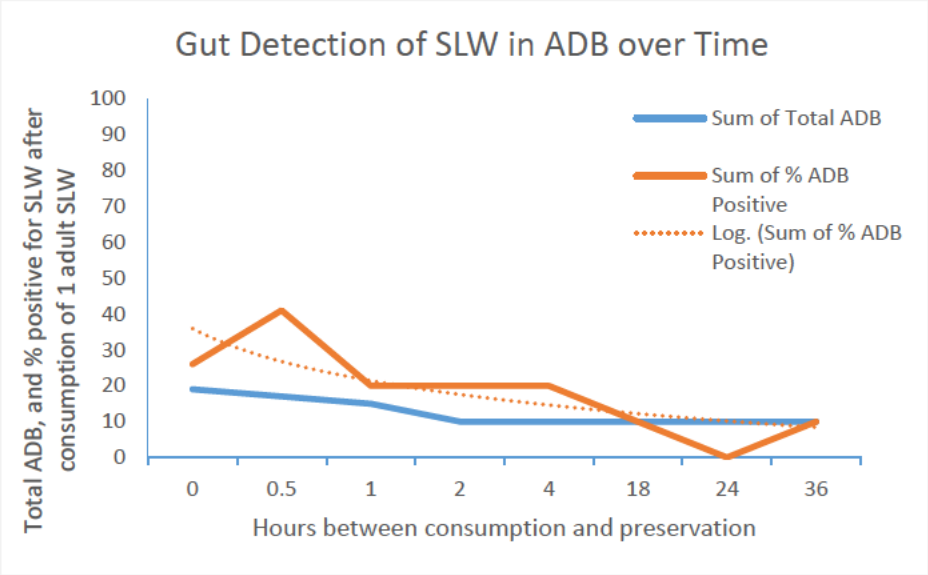


Figure 1: Detection of Silverleaf whitefly in the gut of Apple Dimpling bug after consumption of 1 adult Silverleaf whitefly

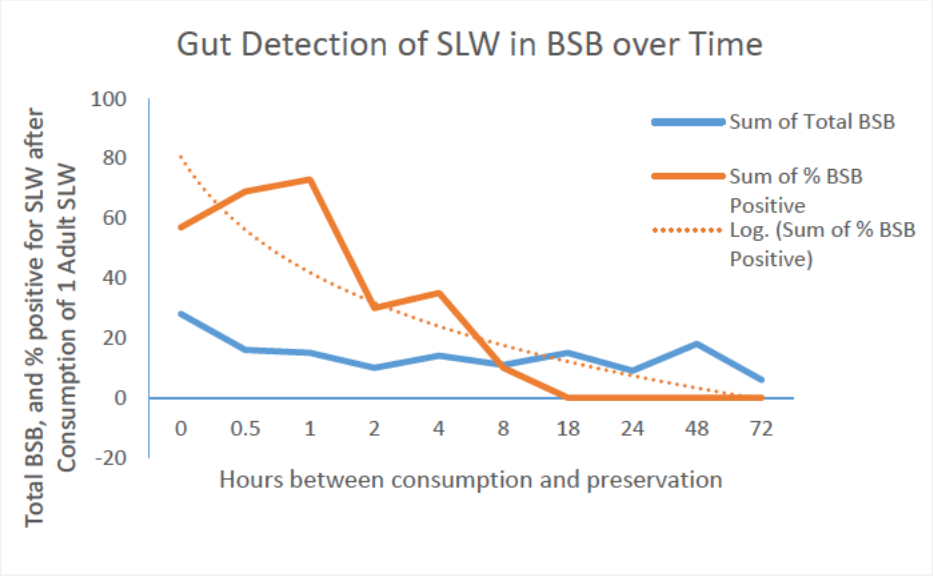


Figure 2: Detection of Silverleaf whitefly in the gut of Brown Smudge bug after consumption of 1 adult Silverleaf whitefly

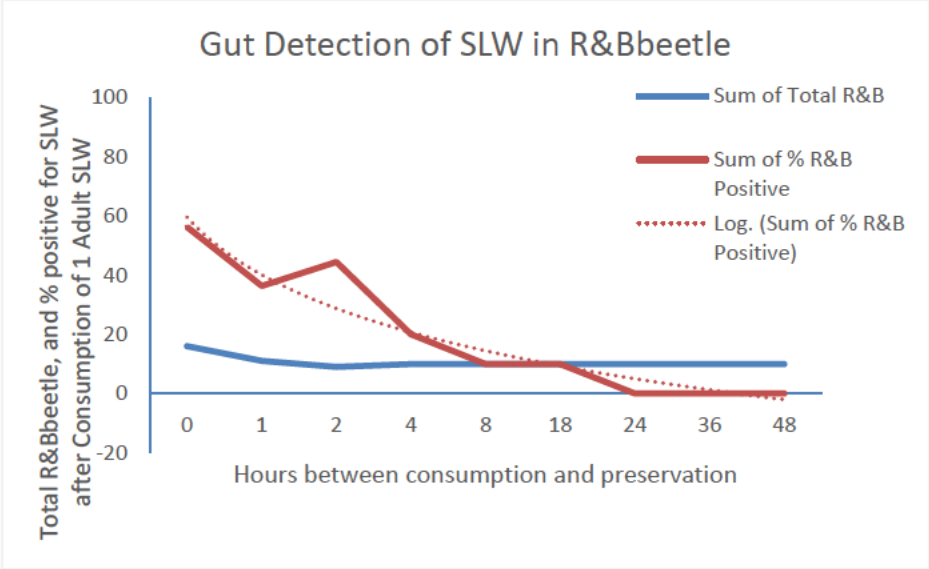


Figure 3: Detection of Silverleaf whitefly in the gut of Red and Blue beetle after consumption of 1 adult Silverleaf whitefly

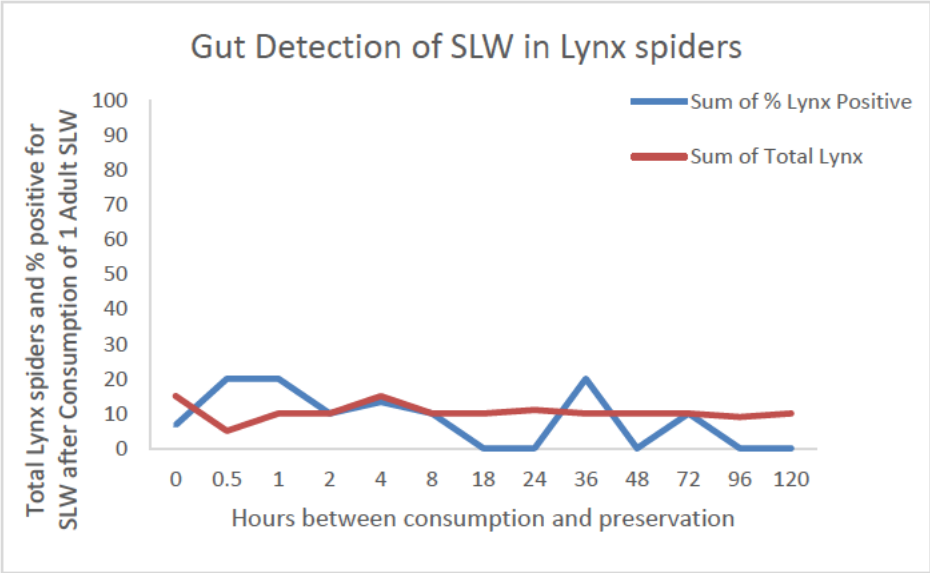


Figure 4: Detection of Silverleaf whitefly in the gut of Lynx spiders after consumption of 1 adult Silverleaf whitefly

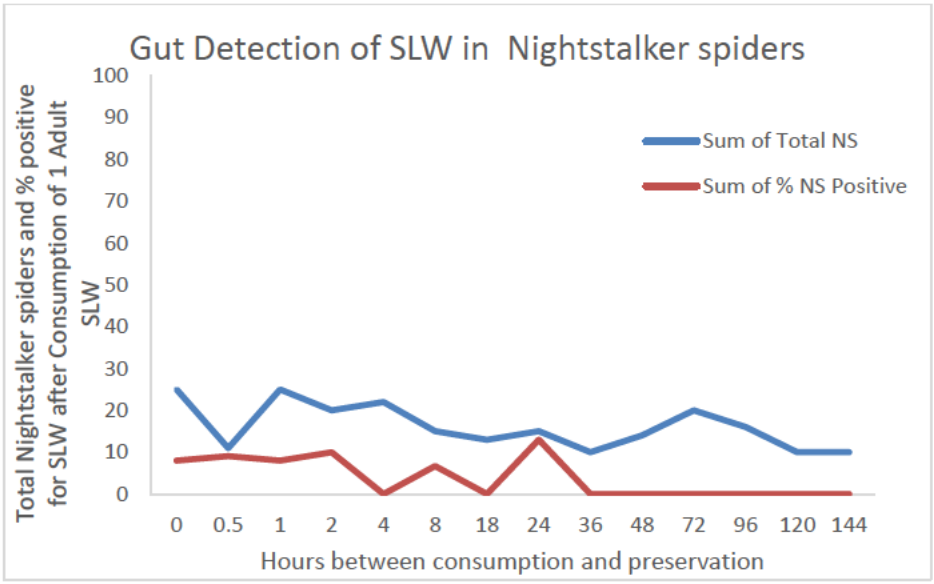


Figure 5: Detection of Silverleaf whitefly in the gut of Nightstalker spiders after consumption of 1 adult Silverleaf whitefly

APPENDIX 4: Section B (ii): Mirid and GVB Management

Methods

Experiment 1 (2012/13) and Experiment 2 (2013/14)

We established plots (8r x 13m) of cotton (Sicot71BRF) in a randomised block design with 6 treatments and 4 replications. There were 2 m buffers between plots. We relied on natural infestation for green mirids and beneficial species. However, we did not want to take a chance with SLW or GVB so we developed strategies to ensure they built up in plots. SLW were reared on kale in a shade house and when the kale plants were well infested, two plants were transplanted into the centre rows of each plot. SLW adults could then move onto cotton and SLW nymphs on the kale could continue to mature and emerge into cotton. We used a two pronged approach with GVB. We ramped up our laboratory GVB culture to produce larger numbers of adults. We also sowed a strip of mung bean within the experimental area as they are a good host for GV. Our intention was to boost numbers of GVB in the mung beans by regularly releasing adults from our culture into the strips. We assumed that some adult GVB would migrate into the cotton as numbers increased and the mungbeans aged. We helped this process along by running a steel bar suspended from a tractor across the mungbeans to disturb the adults, then slashing the mungbeans so that GVB would not be attracted back there but remain in the cotton. GVB did establish to some extent and build in the mungbeans (many nymphs were killed by the slasher) though parasitism by the Tachinid *Trichopoda* was an increasing issue.

The insecticides tested as options for mirid control (Table 1) were the same in both years except for the Plant X extract which used an older formulation in the first experiment and a newer formulation (SeroX) in the second experiment. In both experiments there were 3 spray events at about 2-3 week intervals. We used beat sheets to assess weekly GVB and mirid abundance, leaf sampling for whitefly development, and suction sampling for beneficial abundance.

Moving the GVB from the mungbeans was only partially effective in Experiment 1, so in Experiment 2 we directly infested two marked one metre sections in in each plot with third or fourth instar nymphs or adults. These areas could then be sampled using beat sheets before spraying, with bugs returned to the plots after spraying. In addition we added an extra strip of mungbeans and stocked it heavily from the culture.

Table 1: Insecticides tested for control of mirids and GVB and effects on build-up of SLW in Experiments 1 (2012/13) and 2 (2013/14).

Treatments	*Formulation ai/l or ai/kg*	*g ai/ha*	*Product Rate* **(ml or g/ha)**	**Comments**
1. Control (untreated)	-		-	
2. Dimethoate	400 g/l	140	350 ml/ha	Lower rate. Broad-spectrum, effective on sucking pests but short residual action
3. Fipronil + Salt (NaCl)	200 g/l	8.0	40 ml/ha + 1 kg NaCl/ha	One third full rate. One of the more selective options available, effective on

				sucking pests, short residual
4. Flonicamid	500 g/kg	70	140 g/ha	Higher rate. Reputed selective control of sucking pests (USA experience), not reported effective against pentatomids such as GVB
5. Clothianidin +MAXX	200 g/kg	50	250 ml/ha +0.02 l/l	Higher rate targeting GVB. Broad spectrum, effective on sucking pests but also suppresses SLW
6. 2012-13;CBS2 (Plant X extract formulation) 2013-14;SeroX (Plant X extract formulation)	xx g/L	xx	500 ml/ha	Plant extract from *Clitoria ternatea*, selective, short residual

Results & Discussion

Experiment 1 (2012/13)

CBS2 (Sero X), clothianidin, dimethoate and fipronil all provided effective control of GVB adults (Table 2). The abundance of GVB nymphs was substantially lower, making the chances of detecting a difference lower, and none of the treatments differed from the control, though numerically clothianidin and fipronil were lowest. Mirid abundance was too low to obtain useful results. All compounds significantly reduced abundance of ADB and red banded shield bug. Mite abundance was low and did not differ between treatments.

One of the goals of this study was to identify options to control green mirids and GVB without flaring SLW. In leaf counts SLW abundance was not significantly affected by insecticide treatment, though numerically the control and clothianidin had the lowest abundance for adults (Table 2, Fig. 1). Nymph abundance showed no consistent differences between treatments (Table 2, Fig. 2). In leaf wash samples CBS2, clothianidin, dimethoate and flonicamid had significantly few SLW (mostly adults). In suction samples fipronil had significantly more SLW (mostly adults) than the control and in suction samples clothianidin had significantly fewer (Appendix 4a, Table 17).

The insecticides did have effects on beneficial groups and species. Key results are summarised in Table 3. Clothianidin reduced the abundance of predatory beetles (moderate), though this group was overall more abundant in fipronil treated plots than the controls (Table 3). All of the insecticides reduced abundance (very high) of two spotted ladybeetle (*Diomus notescens*), a predator of mites and SLW. Clothianidin also significantly reduced abundance (high) of Coccinellids (ladybeetles). There were no significant negative effects of any insecticide on predatory bugs and surprisingly CBS2 and clothianidin had significantly more *Orius* spp (minute pirate bugs) than the controls. Lacewings (Neuroptera) were in very low abundance and there were no significant effects of insecticides. Wasps generally were not significantly

affected by insecticides but all insecticides had lower abundance of ants than the control, significantly so for CBS2, clothianidin and fipronil which all had very high negative effects. Flonicamid caused significant moderate reductions in total spiders, while both flonicamid and fipronil had significant moderate negative effects on ‘other spiders’, which was the largest grouping.

Yield was measured and showed no significant difference between treatments though this can be expected in an experiment such as this, where the crop is not managed for yield and where there is considerable repetitive sampling, hence yields were low.

Overall this first experiment highlighted effective control of GVB adults by some treatments and hinted at risk to SLW abundance.

Table 2: Effects of different compounds targeting mirids and GVB on these pests and other species, Experiment 1, ACRI, 2012/13.

Treatment	SLW Adults per leaf[1,4]	SLW Adults per leaf back-transformed	SLW Nymphs per leaf[1,4]	SLW Nymphs per eaf back-ransformed	GVB adults/m[6]	GVB nymphs/m[6]	Mirids /m[6]	Appl e dimpl ing bugs/ m[3,5]	Red-banded shield bug/m[6]	Thrips larvae /m[3,5]	Mites/l eaf [2, 5]	Yield (b/ha)
CBS2	1.18	2.2	2.4	10.0	0.04*	0.028	0.000	0.77*	0.009*	0.08	0.014	6.3
Clothianidin	0.96	1.6	2.3	8.6	0.04*	0.000	0.000	0.48*	0.019*	0.14	0.052	7.2
Dimethoate	1.07	1.9	2.3	9.1	0.04*	0.028	0.000	0.70*	0.019*	0.19	0.047	6.2
Fipronil + salt	1.15	2.1	2.6	11.7*	0.06*	0.014	0.000	0.72*	0.029*	0.07	0.046	6.9
Flonicamid	1.17	2.2	2.3	8.6	0.26	0.139	0.000	0.45*	0.019*	0.11	0.062	6.0
Control	0.99	1.7	2.2	7.7	0.29	0.069	0.014	1.00	0.088	0.23	0.077	6.9
P	0.035		0.038		0.001	0.017	0.42	0.18	0.019	0.14	0.23	0.07
df	5,177		5,177		5, 375	5,375	5,375	5,35	5, 375	5,35	5,191	3,39
LSD	0.21		0.32		0.16	0.085	-	-	0.056		-	0.93

1. Leaf counts or scores
2. Leaf washes
3. Suction samples
4. Values are ln(x+1) transformed.
5. Analysed using ln(x+1) transformed data, back-transformed means shown, statistics refer to transformed analysis, LSD not shown
6. Beat sheet samples

*treatments significantly different from the control at 0.05 using ANOVA/LSD.

Table 3: Whitefly x GVB Experiment: Mean abundance of predators in each insecticide treatment, ACRI, 2012/13. Only species with sufficient abundance for valid analysis are shown.

	Rate	Total Coleoptera Beneficial		*Diomus notescens*		Total other predatory beetles		Ants		Total spiders		Orius spp.	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
CBS 2		0.430	10.88	0.002*	-87.49	0.042	-4.77	0.007*	-87.09	0.291	12.29	0.204*	72.52
Clothianidin	100.0	0.308*	-25.50	0.000*	-100.00	0.040	-10.14	0.009*	-82.77	0.273	4.08	0.200*	68.88
Dimethoate	140.0	0.351	-13.21	0.000*	-100.00	0.036	-18.36	0.039	-27.22	0.236	-11.78	0.141	15.92
Fipronil	8.0	0.502*	34.53	0.007*	-62.39	0.087*	103.03	0.007*	-87.39	0.196	-28.08	0.150	23.88
Flonicamid	70.0	0.418	7.00	0.005*	-74.96	0.052	19.33	0.038	-27.97	0.190*	-30.49	0.135	10.75
Control		0.395	0	0.018	0	0.044	0	0.053	0	0.263	0	0.123	0
P		<0.001, (0.074)		<0.001, (0.032)		0.010, (0.089)		0.022		0.017, (0.227)		0.006	
LSD (p = 0.05)		0.083		0.009		0.029		0.034		0.069		0.052	
df		5,191, (5, 35)		191, (5, 35)		191, (5, 35)		5,191		191, (5, 35)		5,191	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as:
100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
* Asterisks in each column indicate treatments significantly different from the control.

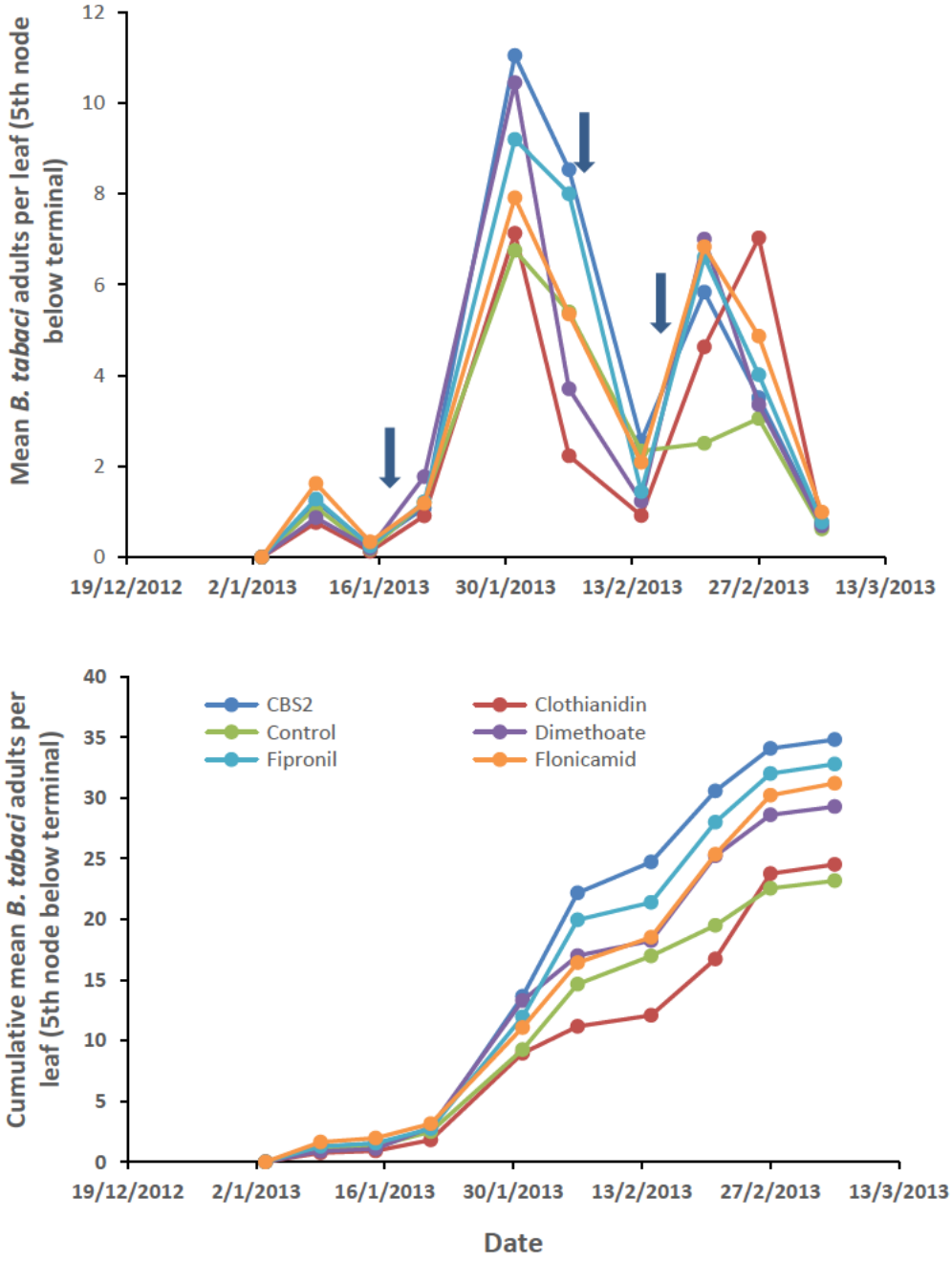


Figure 1: Development of SLW adult populations (Top) mean adults per leaf and (Bottom) cumulative adults per leaf, Experiment 1, ACRI, 2012/13. Arrows indicate spray applications

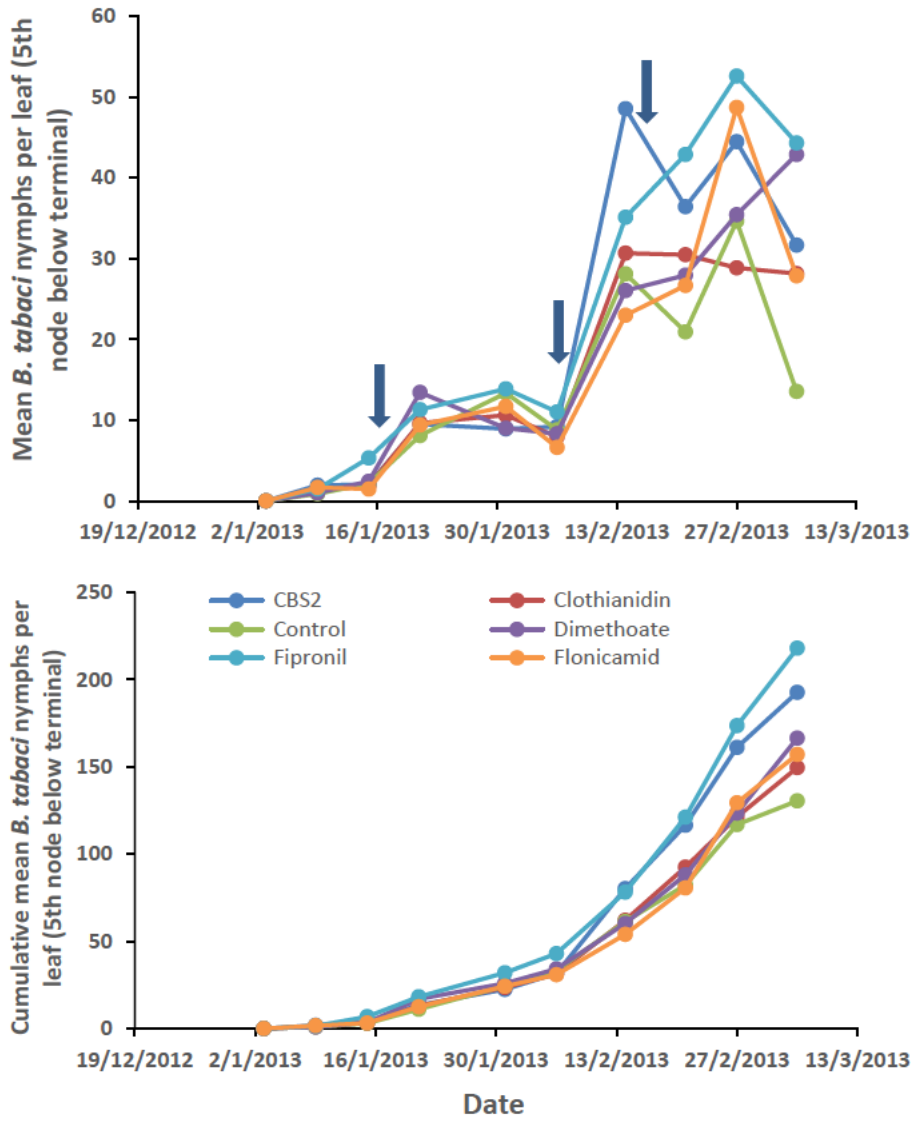


Figure 2: Development of SLW nymph populations (Top) mean nymphs per leaf and (Bottom) cumulative nymphs per leaf, Experiment 1, ACRI, 2012/13. Arrows indicate spray applications

Experiment 2 (2013-14)

This experiment used sections of cotton specifically infested with GVB adults and nymphs and resampled to try to gain a better insight as to the effectiveness of the treatments. This technique was successful for adults and showed effective control of GVB adults by clothianidin and fipronil in beet sheet samples (Table 4, Fig. 3). Nymphs released into the plots had high natural mortality, even in unsprayed control treatments, and this made differences due to insecticides difficult to detect (Table 4, Fig. 4). In suction samples there was no significant effect of insecticides on abundance of adults, but nymphs were significantly reduced by clothianidin, dimethoate and fipronil.

Mirids were more abundant in this experiment and both adults and nymphs were effectively controlled by clothianidin, dimethoate, fipronil and flonicamid (Table 4, Figs. 5 & 6). Similarly, all treatments including SeroX controlled ADB (Table 4). Mite abundance was significantly higher in all treated plots, especially those treated with clothianidin (Table 4). Jassids were significantly reduced in SeroX, clothianidin, dimethoate and flonicamid treated plots. Spider mites were significantly more abundant in all insecticide treatments than in the controls, especially in clothianidin treated plots.

SLW adults and nymphs were significantly more abundant in plots treated with SeroX, and significantly lower in plots treated with clothianidin (Table 4, Figs. 7 & 8). This result confirms the reported suppressive effect of clothianidin on SLW (P. Glover, Sumitomo, Pers. Comm.). There was more honeydew contamination of leaves in the SeroX treated plots and less in the clothianidin treated plots than in the control, in line with the trends in abundance of SLW.

Beneficial abundance was affected by the different treatments (Appendix 4b). In summary, predatory beetles (mainly red and blue beetles, *Dicranolaius bellulus*) were significantly reduced (high) by clothianidin. Total ladybeetles (Coccinellids, a small component of total predatory beetles) were higher in clothianidin, fipronil and flonicamid treatments. Predatory bugs were significantly reduced (high) in clothianidin treated plots. *Orius* spp (high) and *Nabis* spp. (damsel bugs, high) were also significantly reduced in clothianidin treated plots. There were no clear effects on Hymenoptera though clothianidin, dimethoate and fipronil treated plots tended to have lower abundance of some groups especially *Trichogramma*. Lacewings (Neuroptera) were significantly reduced in clothianidin treated plots (very high), including larvae (very high) and green lacewing adults (very high). Spiders were significantly reduced in fipronil treated plots (moderate), though tangleweb spiders, as a small portion of total spiders, were actually more abundant in SeroX, clothianidin and fipronil treated plots.

Table 4: Effect of different compounds targeting mirids and GVB on these pests and other species, Experiment 2, ACRI, 2013/14.

Treatment	SLW Adults per leaf[1,4]	SLW Nymphs per leaf[1,4]	GVB adults/m[5,4]	GVB nymphs/m[5,4]	Mirid adults/m[5,4]	Mirid nymphs/m[5,4]	Apple dimpling bugs/m[3,4]	Thrips larvae /m[3,4]	Mites/leaf (untransformed)[2,4]	Honeydew contamination score	Yield (b/ha)
SeroX	0.91*	1.45*	0.64	0.82	0.285	0.76	0.60*	1.17	0.83*	1.26*	9.7
Clothianidin	0.45*	0.79*	0.32*	0.67	0.072*	0.12*	0.15*	1.19	1.33*	0.67*	9.5
Dimethoate	0.70	1.17	0.63	0.84	0.095*	0.19*	0.33*	1.12	0.79*	0.92	10.3
Fipronil + salt	0.72	1.22	0.46*	0.64	0.058*	0.16*	0.22*	1.07	0.92*	0.84	10.8
Flonicamid	0.78	1.25	0.71	0.81	0.167*	0.19*	0.22*	1.01	1.03*	0.96	10.2
Control	0.71	1.09	0.85	0.76	0.348	0.75	0.80	1.06	0.61	0.96	10.0
P	<0.001	<0.001	<0.001	0.72	<0.001(0.009)	<0.001 (0.002)	<0.001 (<0.001)	0.023 (0.09)	0.001 (<0.001)	<0.001	0.051
df	5,177	5,177	5, 249	5,249	5,249 (5,25)	5,249 (5,25)	5, 159 (5,40)	5,159 (5,40)	5,159 (5,45)	5, 1185	5, 45
LSD	0.15	0.20	0.24	n.s.	0 120	0.16	0.071	n.s.	0 17	0.18	0.88

[1]Leaf counts or scores
[2]Leaf washes
[3]Suction samples
[4]Values are ln(x+1) transformed.
[6] Beat sheet samples
*treatments significantly different from the control at 0.05 using ANOVA/LSD.

Table 5: Whitefly x GVB Experiment: Mean abundance of predators in each insecticide treatment, Experiment 1, ACRI, 2013/14. Only species with sufficient abundance for valid analysis are shown.

	Rate	Total Coleoptera Beneficial		Total Predatory Hemiptera		otal Lacewings		nts		otal spiders		Orius spp.	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
CBS 2		0.89	-5.7	0.23	7.1	0.13	-5.6	0.030*	465.5	0.54	12.8	0.18	11.2
Clothianidin	100.0	0.62*	-43.2	0.11*	-51.6	0.04*	-70.4	0.012	126.2	0.53	8.8	0.09*	-48.8
Dimethoate	140.0	0.95	5.3	0.17	-23.2	0.15	13.8	0.008	55.7	0.49	-2.1	0.12	-29.3
Fipronil	8.0	0.94	3.6	0.29	38.8	0.14	2.1	0.009	64.9	0.36*	-32.1	0.26*	67.6
Flonicamid	70.0	0.91	-2.0	0.17	-23.3	0.11	-23.1	0.012	124.2	0.42	-18.1	0.14	-12.6
Control		0.92	0	0.21	0.0	0.14	0.0	0.005	0.0	0.49	0.0	0.16	0.0
P		<0.001, (<0.001)		<0.001 (<0.001)		<0.001 (0.002)		0.047		<0.001 (0.002)		<0.001 (<0.002)	
LSD (p = 0.05)		0.099		0.056		0.0.04		0.016		0.074		0.054	
df		5,159, (5, 40)		5, 159 (5, 40)		5,159 (5, 40)		5,159		5,159 (5,40)		5, 159 (5, 40)	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
* Asterisks in each column indicate treatments significantly different from the control.

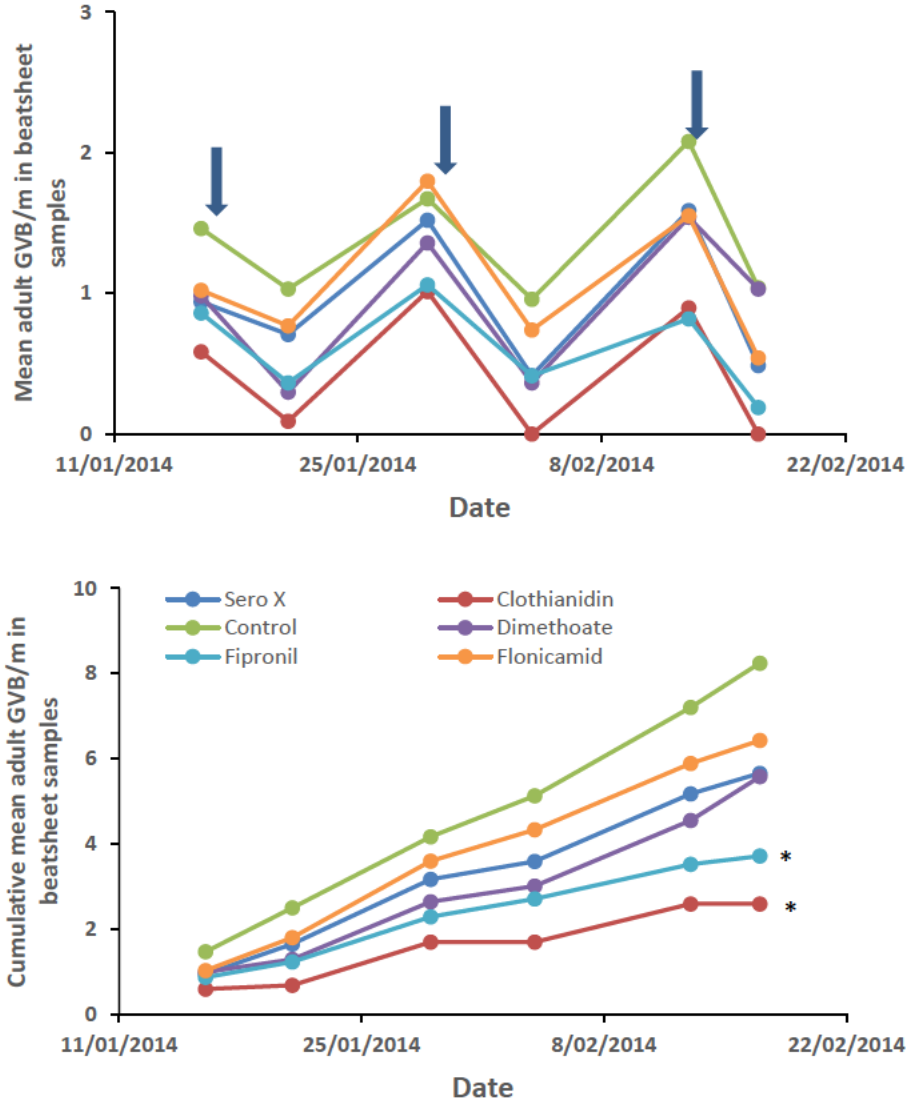


Figure 3: Adult populations of GVB (Top) mean adults from beat sheets and (Bottom) cumulative adults per beat sheet, Experiment 2, ACRI, 2013/14. Arrows indicate spray applications

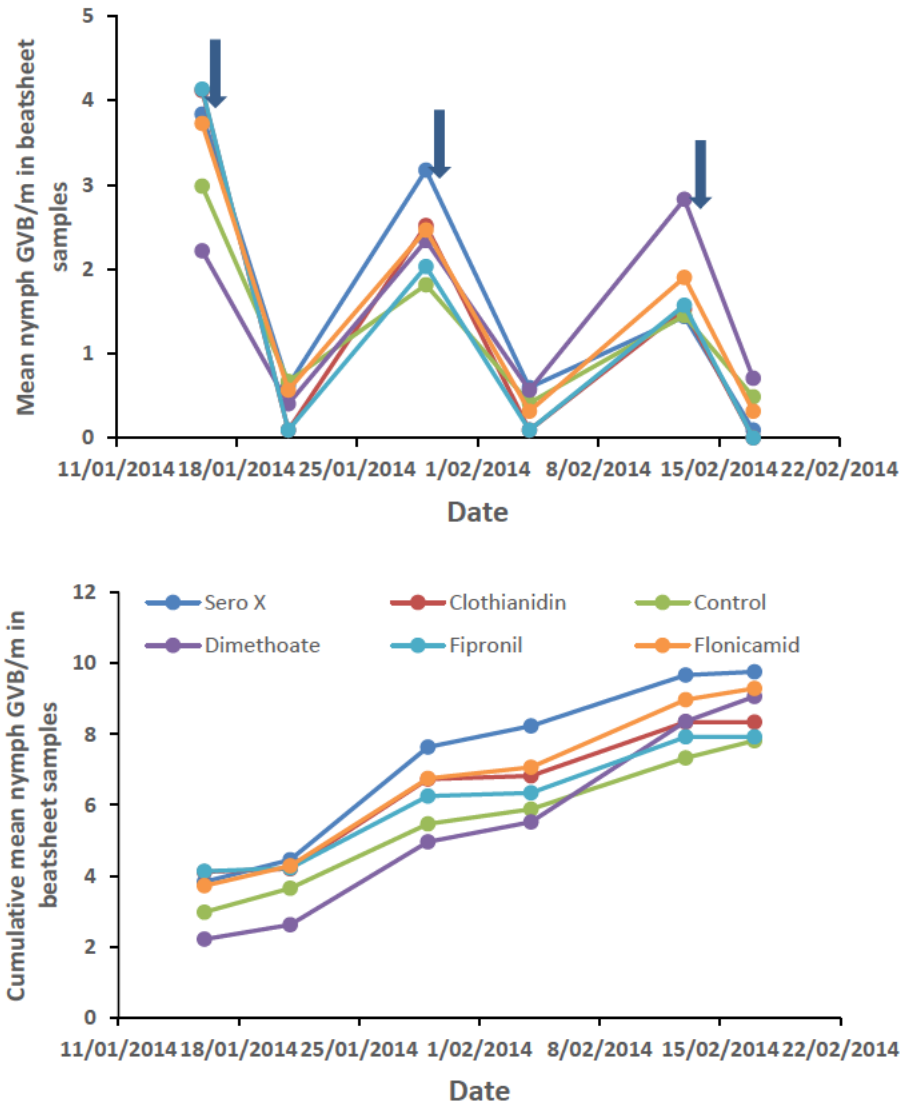


Figure 4: Nymph populations of GVB (Top) mean nymphs from beat sheets and (Bottom) cumulative nymphs per beat sheet, Experiment 2, ACRI, 2013/14. Arrows indicate spray applications

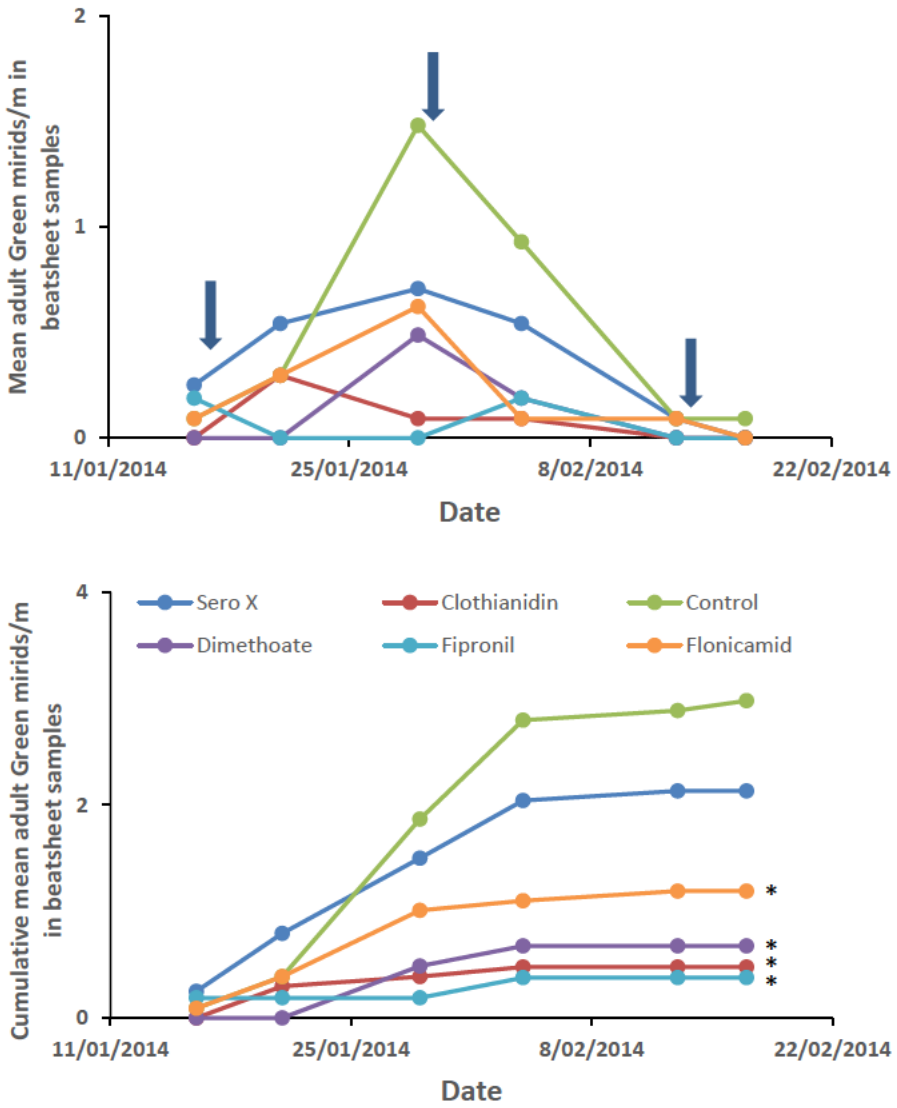


Figure 5: Adult populations of green mirids (Top) mean adults from beat sheets and (Bottom) cumulative adults per beat sheet, Experiment 2, ACRI, 2013/14. Arrows indicate spray applications

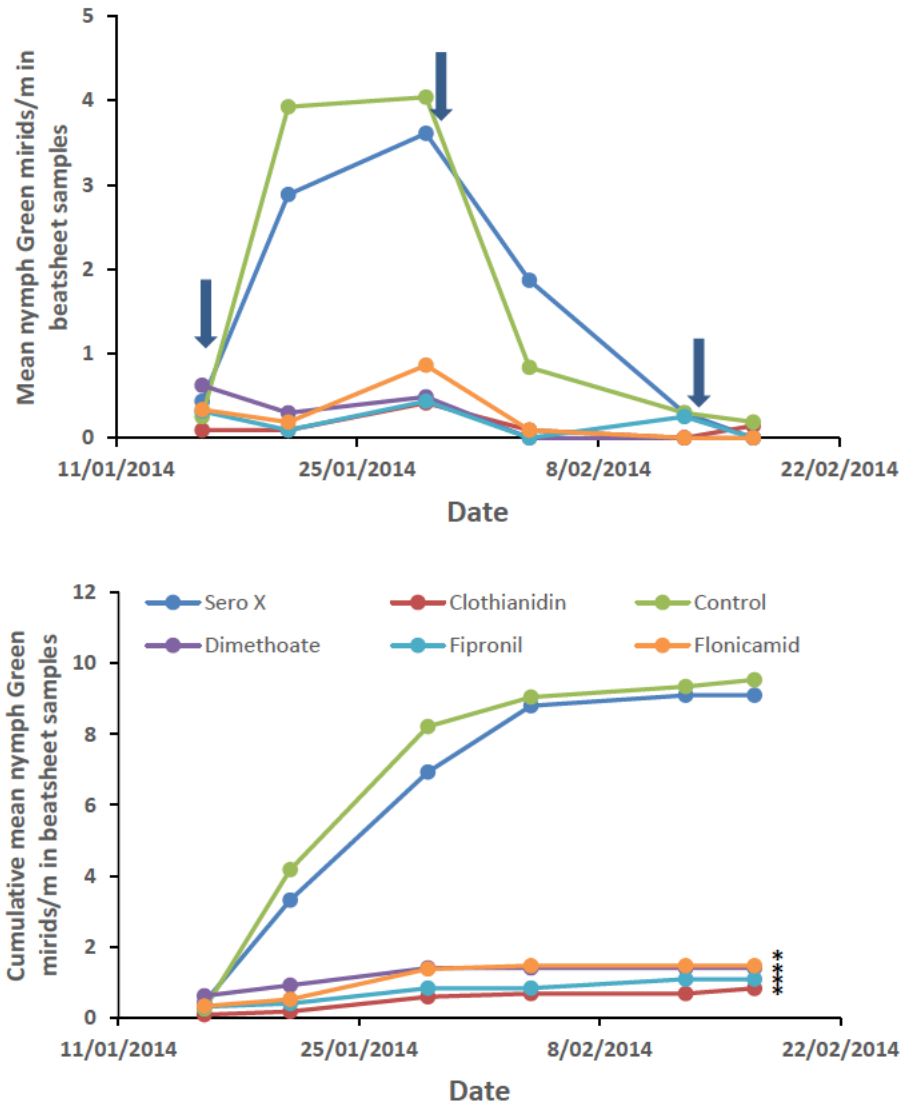


Figure 6: Nymph populations of green mirids (Top) mean nymphs from beat sheets and (Bottom) cumulative nymphs per beat sheet, Experiment 2, ACRI, 2013/14. Arrows indicate spray applications

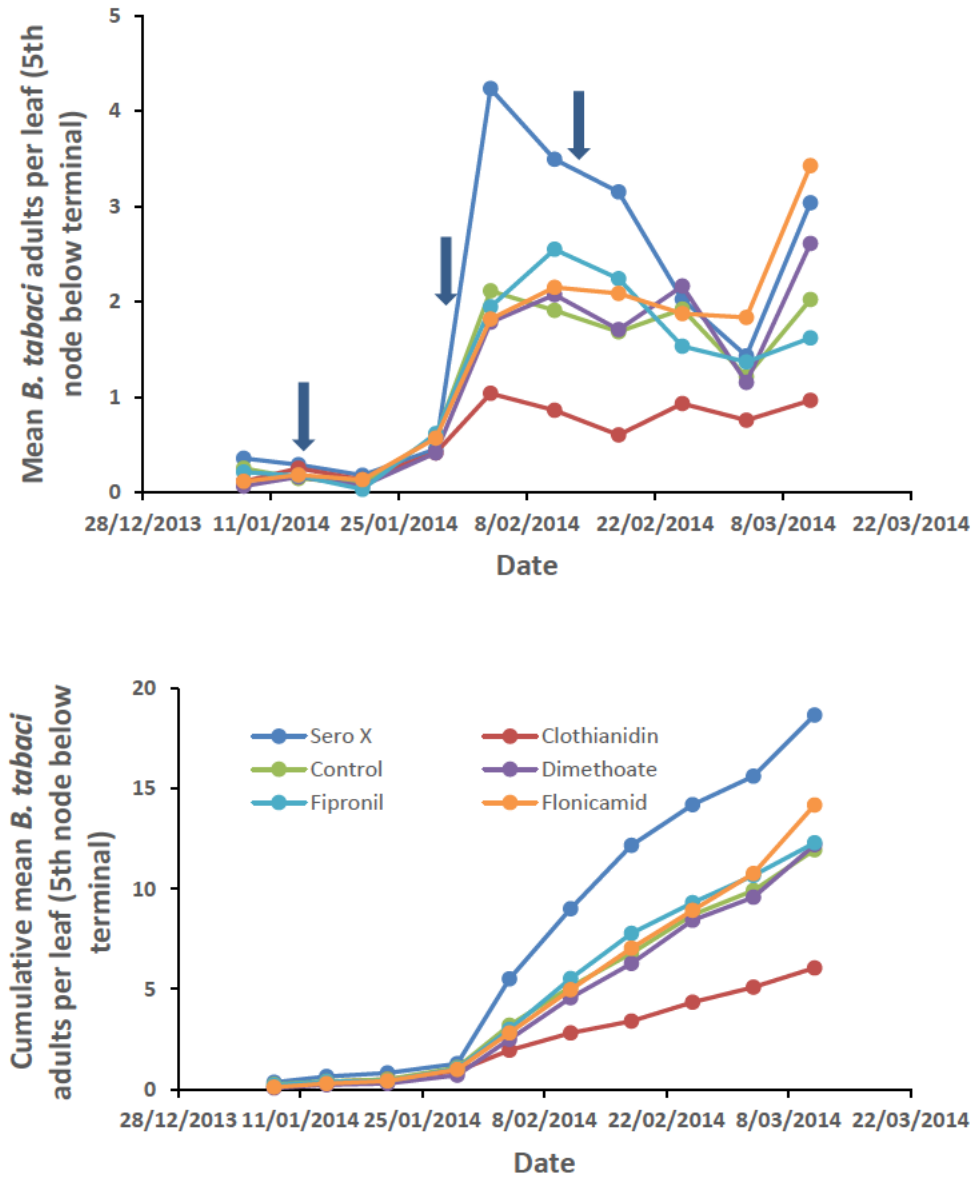


Figure 7: Development of SLW adult populations (Top) mean adults per leaf and (Bottom) cumulative adults per leaf, Experiment 2, ACRI, 2013/14. Arrows indicate spray applications

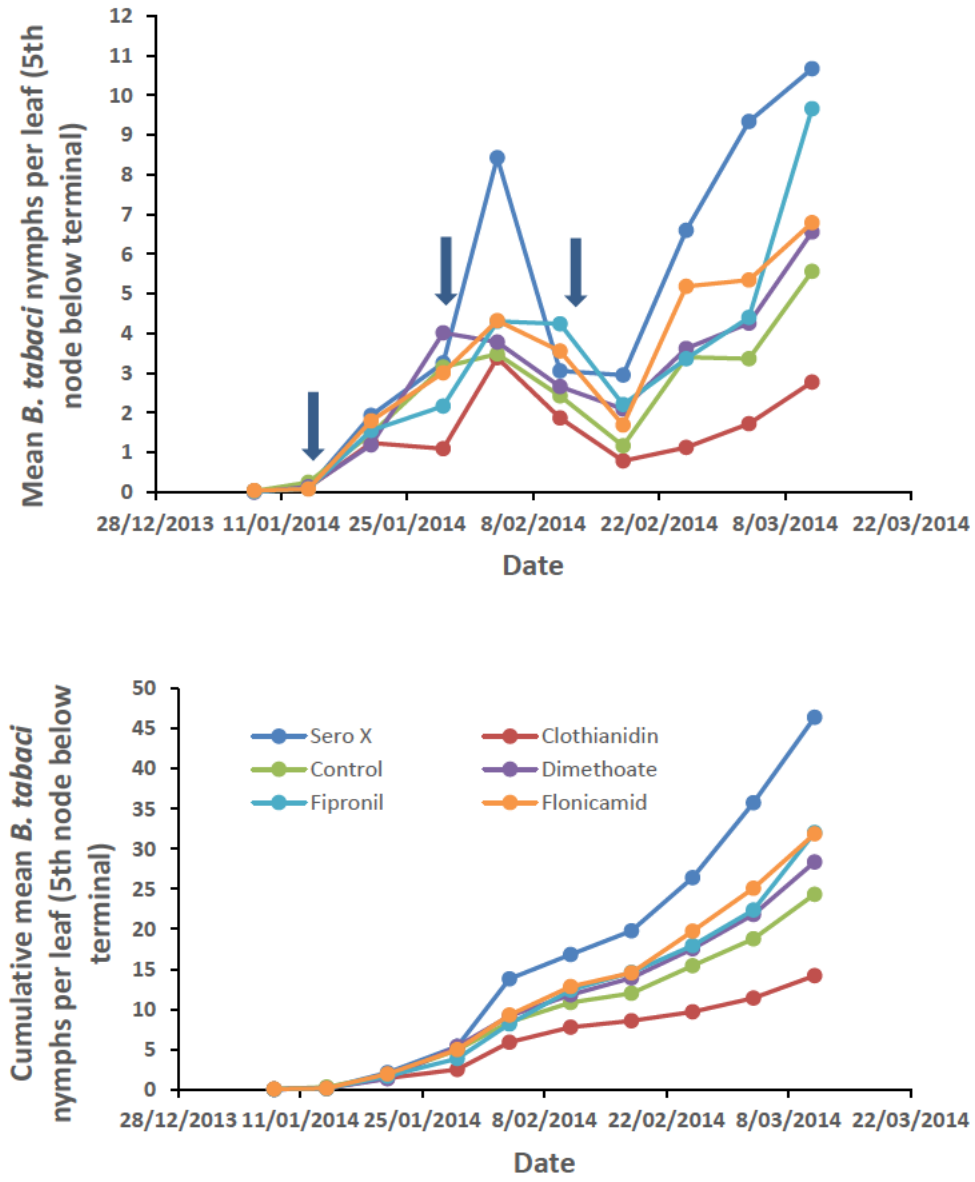


Figure 8: Development of SLW nymph populations (Top) mean nymphs per leaf and (Bottom) cumulative nymphs per leaf, Experiment 2, ACRI, 2013/14. Arrows indicate spray applications

Summary Experiments 1 and 2 (2012/13 and 2013/14)

Considering both the 2012/13 and 2013/14 experiments, some broad trends emerge. These have been summarised in Table 6. Firstly, green mirids were well controlled by all treatments except SeroX (no results for CBS2). Flonicamid was not quite as effective as clothianidin, dimethoate or fipronil. Only clothianidin, dimethoate and fipronil were effective against GVB adults and no compound appeared effective against nymphs, though poor survival of released nymphs and low numbers of naturally occurring nymphs precludes objective conclusions. Surprisingly flonicamid, which was highly effective against mirids, had little effect on GVB adults. All treatments except fipronil provided effective control of jassids. This may explain why jassids sometimes build in fields treated for mirids with fipronil. Red banded shield bug was well controlled by all compounds though as they were not abundant in 2013/14 no data is available for SeroX.

Amongst the compounds tested clothianidin had the most consistent negative effects on beneficial species (Table 6), causing significant reductions in a number of species across four Orders. Dimethoate, fipronil, CBS2 and flonicamid had lower impacts than clothianidin, affecting only a few species across two Orders.

The effects on beneficials were reflected in outbreaks of spider mites in the 2013/14 experiment, and the magnitude of this outbreak mirrored the severity of effects on beneficials, with clothianidin having the most severe mite outbreak.

In terms of managing mirids and GVB without creating SLW problems, the experiments highlight the challenges faced by industry. At the rates tested, clothianidin, dimethoate, fipronil and flonicamid all offered effective control of mirids and a low risk of 'flaring' SLW, though moderate to high risks with mites exist. Clothianidin is the most disruptive of the chemicals to beneficials, reflected in the worst mite outbreaks, but does not flare SLW as it provides suppression of this pest.

Control of GVB, however, is more challenging. Only CBS2, clothianidin, dimethoate and fipronil provided control. Surprisingly though while CBS2 was effective, SeroX was not. Of the options tested, clothianidin is probably the most robust against GVB. However, using clothianidin should be considered carefully due to broad disruptive effects on beneficials and risks of selecting neo-nicotinoid resistance in aphids.

Table 6: Summary of the effects of the insecticides tested on pest and beneficial species from experiments in 2012/13 and 2013/14. See code below the table to interpret symbols.

	Insecticide					
Grouping Beneficials[1]	CBS2	SeroX	Clothianidin	Dimethoate	Fipronil	Flonicamid
Predatory Coleoptera (beetles)			xx, xxx		x	
Total Coccinellids (ladybeetles			x			
Two-spotted ladybeetle (*Diomus notescens*)	xxxx		xxxx	xxxx	xxxx	xxxx
Red and blue beetle (*Dicranolaius bellulus*)			xxxx			
Predatory Hemiptera (true bugs)			xxx			
Minute pirate bugs (*Orius* spp.)			xxx			
Damsel bugs (*Nabis kinbergii*)			xxx			
Lacewings (Neuroptera)			xxxx			
Spiders					xx	xx
Other spiders					Xx,xx	xx
Ants	xxxx		xxxx	xxxx		
Pest and beneficial[1]						
Thrips	x		x		x	x
ADB	xx	xx	Xxx,xxxx	Xx,xxxx	Xx,xxxx	Xxx,xxxx
Pests[3]						
GVB adults	✓✓✓✓		✓✓✓✓, ✓✓✓✓	✓✓✓✓	✓✓✓✓, ✓✓✓	
GVB nymphs						
Mirid adults			✓✓✓✓,✓✓✓ ✓	✓✓✓✓, ✓✓✓✓	✓✓✓✓, ✓✓✓✓	✓✓✓,✓✓ ✓
Mirid nymphs			✓✓✓✓,✓✓✓ ✓	✓✓✓✓,✓✓ ✓✓	✓✓✓✓, ✓✓✓✓	✓✓✓✓,✓ ✓✓✓
Jassids	✓✓✓	✓✓	✓✓✓	✓✓,✓✓✓		✓✓,✓✓✓
RBSB	✓✓✓✓		✓✓✓✓	✓✓✓✓	✓✓✓✓	✓✓✓✓
Mites[2]		+	+++	+	+	++
SLW[2,3]		+	✓✓✓			

1. % suppression of beneficials compared with control
X = low (11-20%)
Xx = moderate (21-40%)
Xxx = high (41-60%)
Xxxx= very high (61%+)

2. % increase in pest abundance compared with control
+ = 40-100% increase
++ = 101-200% increase
+++ = 201% - 300% increase
++++ = 301%+ increase

3. % control of target pest
✓ = low (11-20%)
✓✓ = moderate (21-40%)
✓✓✓high (41-60%)
✓✓✓✓= very high (61%+)

Appendix 4a. Options to manage mirids and GVB without flaring SLW, mites or aphids. Beneficial data for 2012-13

Table 7: Whitefly x GVB Experiment: Mean abundance of predatory and pest Coleoptera in each insecticide treatment, ACRI, 2012/13.

	Rate	Total Coleoptera Beneficial		[redacted]		*Diomus notescens*		Total Coccinellids		otal other redatory beetles		Total Pest Coleoptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
CBS 2		0.430	10.88	0.161	-10.15	0.002*	-87.49	0.348*	38.48	0.042	-4.77	0.818	-22.64
Clothianidin	100.0 g/ha	0.308*	-25.50	0.161	-10.11	0.000*	-100.00	0.153*	-45.10	0.040	-10.14	0.866	-15.84
Dimethoate	140.0 g/ha	0.351	-13.21	0.149	-16.90	0.000*	-100.00	0.241	-9.15	0.036	-18.36	0.905	-10.15
Fipronil	8.0 g/ha	0.502*	34.53	0.182	3.04	0.007*	-62.39	0.372*	50.23	0.087*	103.03	0.799	-25.23
Flonicamid	70.0 g/ha	0.418	7.00	0.157	-12.38	0.005*	-74.96	0.311	21.70	0.052	19.33	0.945	-3.95
Control	---	0.395	0	0.177	0	0.018	0	0.262	0	0.044	0	0.970	0
P		<0.001, (0.074)		0.926		<0.001, (0.032)		<0.001, (0.008)		0.010, (0.089)		0.251	
LSD (p = 0.05)		0.083		n.s.		0.009		0.070		0.029		n.s.	
df		191, (5, 35)		191		191, (5, 35)		191, (5, 35)		191, (5, 35)		191	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 8: Whitefly x GVB Experiment: Mean abundance of predatory Hemiptera in each insecticide treatment, ACRI, 2012/13.

	Rate	*Geocoris lubra*		[redacted]		*Nabis kinbergiii*		*Deraeocoris signatus*		ther Beneficial emiptera		Total Predatory Hemiptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
CBS 2		0.002	0.00	0.204*	72.52	0.000	0.00	0.021	-50.59	0.007	0.00	0.230	39.11
Clothianidin	100.0 g/ha	0.007	193.75	0.200*	68.88	0.002	0.00	0.011	-73.06	0.002	0.00	0.222	33.92
Dimethoate	140.0 g/ha	0.005	100.23	0.141	15.92	0.000	0.00	0.021	-50.98	0.000	0.00	0.160	-6.64
Fipronil	8.0 g/ha	0.002	0.00	0.150	23.88	0.000	0.00	0.031	-25.60	0.000	0.00	0.181	6.96
Flonicamid	70.0 g/ha	0.002	0.00	0.135	10.75	0.000	0.00	0.020	-52.12	0.002	0.00	0.158	-7.69
Control	---	0.002	0	0.123	0	0.000	0	0.042	0	0.000	0	0.171	0
P		0.866		0.006		0.420		0.111		0.340			
LSD (p = 0.05)		n.s.		0.052		n.s.		n.s.		n.s.			
df		191		191		191		191		191			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 9: Whitefly x GVB Experiment: Mean abundance of Hymenoptera in each insecticide treatment, ACRI, 2012/13.

		Total Hymenoptera											
	Rate	Total Hymenoptera	(Wasp)	[redacted]		*Microplitis*		*Telenomus*		ther wasp spp.		Ants	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
CBS 2		0.280	-0.32	0.002	-50.06	0.069	41.60	0.029	161.92	0.209	-10.73	0.007*	-87.09
Clothianidin	100.0 g/ha	0.246	-13.79	0.007	50.17	0.064	30.09	0.025	129.27	0.168	-29.70	0.009*	-82.77
Dimethoate	140.0 g/ha	0.230	-20.22	0.002	-50.06	0.043	-12.25	0.030	171.85	0.166	-30.86	0.039	-27.22
Fipronil	8.0 g/ha	0.301	8.16	0.002	-50.06	0.070	43.57	0.023	105.13	0.229	-1.24	0.007*	-87.39
Flonicamid	70.0 g/ha	0.307	10.62	0.009	100.46	0.069	42.04	0.023	103.85	0.230	-0.61	0.038	-27.97
Control	---	0.281	0	0.005	0	0.049	0	0.011	0	0.231	0	0.053	0
P		0.194		0.440		0.470, (0.762)		0.304		0.135		0.022	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.		n.s.		0.034	
df		191		191		191, (5, 35)		191		191		191	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 10: Whitefly x GVB Experiment: Mean abundance of *Neuroptera* in each insecticide treatment, ACRI, 2012/13.

	Neuroptera								
	Rate	Total Neuroptera		LW Larvae		Green Adult		Brown Adult	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
CBS 2		0.009	-21.48	0.000	-100.00	0.009	31.17	0.000	0.00
Clothianidin	100.0 g/ha	0.009	-21.48	0.004	-3.46	0.005	-33.41	0.000	0.00
Dimethoate	140.0 g/ha	0.031	173.39	0.005	0.00	0.027	292.72	0.000	0.00
Fipronil	8.0 g/ha	0.029	154.31	0.009	96.99	0.021	199.76	0.000	0.00
Flonicamid	70.0 g/ha	0.025	117.32	0.009	96.99	0.016	129.76	0.000	0.00
Control	---	0.012	0	0.005	0	0.007	0	0.000	0
P		0.108		0.639		0.083		0	
LSD (p = 0.05)		n.s.		n.s.		n.s.		0	
df		191							

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 11: Whitefly x GVB Experiment: Mean abundance Arachnids in each insecticide treatment, ACRI, 2012/13.

		Arachnids					
		Total Spiders		Tangleweb		Other Spiders	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
CBS 2		0.291	12.29	0.057	270.02	0.254	1.36
Clothianidin	100.0 g/ha	0.273	4.08	0.030	93.45	0.250	-0.64
Dimethoate	140.0 g/ha	0.236	-11.78	0.009	-40.78	0.229	-9.84
Fipronil	8.0 g/ha	0.196	-28.08	0.029	90.44	0.170*	-34.90
Flonicamid	70.0 g/ha	0.190*	-30.49	0.014	-11.99	0.179*	-31.08
Control	---	0.263	0	0.016	0	0.251	0
P		0.017, (0.227)		0.256		0.017, (0.155)	
LSD (p = 0.05)		0.069		n.s.		0.062	
df		191, (5, 35)		191		191, (5, 35)	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 12: Whitefly x GVB Experiment: Mean abundance of *Creontiades dilutus*, *Nezara viridula* and *Campylomma liebknechti* in each insecticide treatment, ACRI, 2012/13.

	Rate	Total Hemiptera Pests		*Creontiades dilutus Adult*		*Creontiades dilutus Nymph*		*Total Creontiades dilutus*		*Nezara viridula*		*ampylomma liebknech*	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
CBS 2		4.562*	26.47	0.0023	0.00	0.0002	0.00	0.007	193.75	0.009	-51.44	0.571*	-22.90
Clothianidin	100.0 g/ha	4.305	-2.44	0.0023	0.00	0.0002	0.00	0.002	0.00	0.073	313.36	0.391*	-52.05
Dimethoate	140.0 g/ha	4.473*	15.60	0.0000	0.00	0.0000	0.00	0.005	100.23	0.017	-5.20	0.531*	-29.86
Fipronil	8.0 g/ha	4.697*	44.99	0.0000	0.00	0.0000	0.00	0.000	-100.00	0.008	-53.77	0.540*	-28.32
Flonicamid	70.0 g/ha	4.428	10.43	0.0000	0.00	0.0000	0.00	0.022	844.43	0.011	-36.93	0.374*	-54.51
Control	---	4.330	0	0.0000	0	0.0000	0	0.002	0	0.018	0	0.692	0
P		0.004		0.555		0.555		0.597		0.160		<0.001, (0.018)	
LSD (p = 0.05)		0.216		n.s.		n.s.		n.s.		n.s.		0.085	
df		191		191		191		191		191		191, (5, 35)	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 13: Whitefly x GVB Experiment: Mean abundance of leafhoppers and other Hemiptera pests in each insecticide treatment, ACRI, 2012/13.

	Rate	Jassids		Other Hemiptera Pests		Total Hemiptera Pests	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
CBS 2		0.059*	-48.18	0.025	-47.16	4.562*	26.47
Clothianidin	100.0 g/ha	0.099	-10.55	0.029	-38.36	4.305	-2.44
Dimethoate	140.0 g/ha	0.070*	-38.09	0.029	-39.21	4.473*	15.60
Fipronil	8.0 g/ha	0.127	16.34	0.055	15.72	4.697*	44.99
Flonicamid	70.0 g/ha	0.071*	-36.55	0.028	-42.25	4.428	10.43
Control	---	0.110	0	0.047	0	4.330	0
P		0.001, (0.039)		0.086		0.004	
LSD (p = 0.05)		0.036		n.s.		0.216	
df		191, (5,35)		191		191	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 14: Whitefly x GVB Experiment: Mean abundance of Lepidoptera and *Helicoverpa* spp. in each insecticide treatment, ACRI, 2012/13.

	Rate	Total Lepidoptera		*Helicoverpa* Eggs		*Helicoverpa* Larvae	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
CBS 2		0.014	-15.38	0.002	-66.74	0.011	23.41
Clothianidin	100.0 g/ha	0.007	-57.34	0.002	-66.74	0.002	-75.09
Dimethoate	140.0 g/ha	0.018	10.56	0.009	33.49	0.002	-75.09
Fipronil	8.0 g/ha	0.005	-71.59	0.000	-100.00	0.002	-75.09
Flonicamid	70.0 g/ha	0.019	14.42	0.005	-33.41	0.014	50.35
Control	---	0.016	0	0.007	0	0.009	0
P		0.339		0.256		0.150	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df		191		191		191	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 15: Whitefly x GVB Experiment: Mean abundance of thrips (*Thrips tabaci* and *Frankliniella schultzei*) in each insecticide treatment, ACRI, 2012/13.

		Total Thrips				Thrips Adults				Thrips Nymphs			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
CBS 2		0.607*	28.66	0.696*	-18.58	0.483*	29.61	0.662	-5.98	0.189	30.23	0.077*	-64.81
Clothianidin	100.0 g/ha	0.610*	29.53	0.908	19.62	0.471	25.57	0.855*	35.46	0.200*	38.85	0.130*	-39.57
Dimethoate	140.0 g/ha	0.434	-16.24	0.744	-10.72	0.345	-14.10	0.654	-7.41	0.128	-14.76	0.178	-14.91
Fipronil	8.0 g/ha	0.434	-16.11	0.563*	-38.91	0.355	-11.15	0.524*	-30.89	0.112	-25.85	0.066*	-70.11
Flonicamid	70.0 g/ha	0.464	-8.85	0.688*	-19.93	0.377	-4.53	0.636	-10.91	0.120	-20.48	0.109*	-49.66
Control	---	0.500	0	0.805	0	0.392	0	0.692	0	0.148	0	0.206	0
P		<0.001		<0.001, (0.001)		0.001		<0.001		<0.001		<0.001, (0.142)	
LSD (p = 0.05)		0.087		0.105		0.08		0.109		0.041		0.058	
df		191		191, (5, 35)		191		191		191		191, (5, 35)	

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1).
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 16: Whitefly x GVB Experiment: Mean abundance of mites (*Tetranychus urticae*) in each insecticide treatment, ACRI, 2012/13.

		Total Mites				Mites Adults				Mites Nymphs			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
CBS 2		0.014	-82.21	0.0023	0.00	0.011	-77.15	0.0000	0.00	0.002	-95.42	0.0023	0.00
Clothianidin	100.0 g/ha	0.051	-32.36	0.0000	0.00	0.041	-9.32	0.0000	0.00	0.013	-59.20	0.0000	0.00
Dimethoate	140.0 g/ha	0.046	-38.16	0.0046	0.00	0.028	-38.80	0.0023	0.00	0.013	-61.61	0.0023	0.00
Fipronil	8.0 g/ha	0.045	-40.70	0.0023	0.00	0.038	-17.08	0.0023	0.00	0.005	-86.45	0.0000	0.00
Flonicamid	70.0 g/ha	0.060	-18.87	0.0000	0.00	0.041	-10.07	0.0000	0.00	0.017	-48.30	0.0000	0.00
Control	---	0.074	0	0.0000	0	0.046	0	0.0000	0	0.033	0	0.0000	0
P		0.235		0.433		0.377		0.555		0.11		0.555	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.		n.s.		n.s.	
df		191		191		191		191		191		191	

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1).
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 17: Whitefly x GVB Experiment: Mean abundance of whitefly and aphids in each insecticide treatment, ACRI, 2012/13.

		Whitefly				Aphids			
	Rate	Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
CBS 2		1.631*	-66.21	4.502	24.92	0.591	35.81	0.011	-29.02
Clothianidin	100.0 g/ha	2.342*	-22.69	4.254	-2.83	0.553	24.50	0.009	-42.48
Dimethoate	140.0 g/ha	1.985*	-48.35	4.431	16.26	0.579	32.23	0.012	-28.02
Fipronil	8.0 g/ha	2.622	4.93	4.654*	45.69	0.416	-13.04	0.009	-42.48
Flonicamid	70.0 g/ha	1.909*	-52.73	4.395	12.04	0.566	28.34	0.013	-19.04
Control	---	2.577	0	4.282	0	0.465	0	0.016	0
P		<0.001		0.005		0.123		0.952	
LSD (p = 0.05)		0.213		0.221		n.s.		n.s.	
df		191		191		191		191	

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1).
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Appendix 4b: Options to manage mirids and GVB without flaring SLW, mites or aphids. Beneficial data for 2013-14.

Table 18: Whitefly x GVB Experiment: Mean abundance of predatory and pest Coleoptera in each insecticide treatment, ACRI, 2013/14.

	Rate	Total Coleoptera Beneficial		*Dicranolaius bellulus*		*Diomus notescens*		Total Coccinellids		Total other predatory beetles		Total Pest Coleoptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
SeroX		0.89	-5.7	0.78	-4.0	0.00	-74.0	0.09	-11.2	0.14	-11.1	0.56	-9.7
Clothianidin	100.0	0.62*	-43.2	0.39*	-61.4	0.00	-74.0	0.23*	150.3	0.08	-53.4	0.55	-11.7
Dimethoate	140.0	0.95	5.3	0.81	1.4	0.01	-61.0	0.16	66.4	0.14	-11.2	0.53	-16.2
Fipronil	8.0	0.94	3.6	0.74	-10.5	0.01	-48.8	0.24*	163.4	0.16	-2.8	0.55	-12.4
Flonicamid	70.0	0.91	-2.0	0.75	-8.9	0.00	-87.0	0.22*	134.9	0.11	-33.9	0.52	-16.9
Control	---	0.92	0	0.80	0.0	0.01	0.0	0.10	0.0	0.16	0.0	0.60	0.0
P		<0.001 (<0.001)		<0.001 (<0.001)		0.34		<0.001 (0.025)		0.001 (0.06)		0.07	
LSD (p = 0.05)		0.099		0.099		n.s.		0.063		n.s.		n.s.	
df		5, 159 (5, 40)		5, 159 (5, 40)		5, 159		5, 159 (5, 40)		5, 159		5, 159	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 19: Whitefly x GVB Experiment: Mean abundance of predatory Hemiptera in each insecticide treatment, ACRI, 2013/14.

	Rate	*Geocoris lubra*		*Orius spp.*		*Nabis kinbergiii*		*Deraeocoris signatus*		Other Beneficial Hemiptera		Total Predatory Hemiptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
SeroX		0.025*	130.0	0.18	11.2	0.027	-27.3	0.004	-50.1	0.002	-66.0	0.23	7.1
Clothianidin	100.0	0.002	-83.4	0.09*	-48.8	0.016*	-56.2	0.000	-100.0	0.007	34.1	0.11*	-51.6
Dimethoate	140.0	0.007	-33.5	0.12	-29.3	0.047	29.6	0.000	-100.0	0.002	-66.0	0.17	-23.2
Fipronil	8.0	0.011	-1.0	0.26*	67.6	0.021	-42.1	0.005	-25.1	0.000	-100.0	0.29	38.8
Flonicamid	70.0	0.000	-100.0	0.14	-12.6	0.019	-47.6	0.005	-25.1	0.000	-100.0	0.17	-23.3
Control	---	0.011	0.0	0.16	0.0	0.036	0.0	0.007	0.0	0.005	0.0	0.21	0.0
P		0.002		<0.001 (<0.002)		0.024		0.176		0.24		<0.001 (<0.001)	
LSD (p = 0.05)		0.012		0.054		0.02		n.s.		n.s.		0.056	
df		5, 159		5, 159 (5, 40)		5, 159		5, 159		5, 159		5, 159 (5, 40)	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 20: Whitefly x GVB Experiment: Mean abundance of Hymenoptera in each insecticide treatment, ACRI, 2013/14.

	Rate	Total (Wasp) Hymenoptera		*Trichogramma*		*Microplitis*		*Telenomus*		Other wasp spp.		Ants	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
SeroX		0.89	2.6	0.17	-33.3	0.040	-37.3	0.018	0.6	0.56	7.0	0.030*	465.5
Clothianidin	100.0	0.75	-21.1	0.12	-54.4	0.064	0.9	0.009	-49.6	0.54	2.1	0.012	126.2
Dimethoate	140.0	0.78	-15.7	0.12	-53.9	0.066	4.9	0.014	-19.2	0.50	-7.8	0.008	55.7
Fipronil	8.0	0.83	-8.5	0.17	-35.9	0.045	-29.8	0.018	0.6	0.59	14.6	0.009	64.9
Flonicamid	70.0	0.86	-3.0	0.13	-51.3	0.075	19.0	0.014	-21.0	0.55	4.3	0.012	124.2
Control	---	0.88	0.0	0.25	0.0	0.063	0.0	0.018	0.0	0.53	0.0	0.005	0.0
P		0.005 (0.6)		<0.001 (0.06)		0.10		0.80		0.5		0.047	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.		n.s.		0.016	
df		5, 159 (5, 40)		5, 159 (5,40)		5, 159		5, 159		5, 159		5, 159	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 21: Whitefly x GVB Experiment: Mean abundance of *Neuroptera* in each insecticide treatment, ACRI, 2013/14.

	Neuroptera								
	Rate	Total Neuroptera		LW Larvae		Green Adult		Brown Adult	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
SeroX		0.13	-5.6	0.077	18.9	0.051	-30.1	0.004	0.0
Clothianidin	100.0	0.04*	-70.4	0.018*	-73.3	0.023*	-69.0	0.002	-50.0
Dimethoate	140.0	0.15	13.8	0.082	27.9	0.076	4.7	0.002	-50.0
Fipronil	8.0	0.14	2.1	0.084	30.5	0.058	-20.8	0.002	-50.0
Flonicamid	70.0	0.11	-23.1	0.059	-10.0	0.052	-29.1	0.000	-100.0
Control	---	0.14	0.0	0.065	0.0	0.073	0.0	0.004	0.0
P		<0.001 (0.002)		<0.001 (0.04)		0.013		0.75	
LSD (p = 0.05)		0.04		0.032		0.032		n.s.	
df		5, 159 (5,40)							

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 22: Whitefly x GVB Experiment: Mean abundance Arachnids in each insecticide treatment, ACRI, 2013/14.

		Total Spiders		Tangleweb		Other Spiders	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
SeroX		0.54	12.8	0.056*	169.4	0.51	8.2
Clothianidin	100.0	0.53	8.8	0.048*	128.5	0.50	5.0
Dimethoate	140.0	0.49	-2.1	0.038	83.2	0.46	-4.9
Fipronil	8.0	0.36	-32.1	0.051*	145.0	0.32*	-38.1
Flonicamid	70.0	0.42	-18.1	0.025	18.2	0.40	-19.1
Control	---	0.49	0.0	0.021	0.0	0.48	0.0
P		<0.001 (0.002)		0.005		<0.001 (0.002)	
LSD (p = 0.05)		0.074		0.021		0.072	
df		5,159 (5,40)		5, 159		5, 159 (5, 40)	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 23: Whitefly x GVB Experiment: Mean abundance of *Creontiades dilutus*, *Nezara viridula* and *Campylomma liebknechti* in each insecticide treatment, ACRI, 2013/14.

	Rate	Total Hemiptera Pests		*Creontiades dilutus Adult*		*Creontiades dilutus Nymph*		*Total C. dilutus*		*Nezara viridula adults*		*Nezara viridula nymphs*	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
SeroX		3.49	10.4	0.075	-5.5	0.13	-24.4	0.19	-18.4	0.012	136.9	0.019	-15.6
Clothianidin	100.0	2.78*	-47.5	0.019*	-76.5	0.02*	-86.1	0.04*	-83.0	0.002	-66.0	0.000*	-100.0
Dimethoate	140.0	3.29	-10.4	0.030*	-63.6	0.03*	-80.8	0.06*	-75.1	0.007	34.1	0.002*	-91.9
Fipronil	8.0	3.30	-9.0	0.021*	-74.6	0.05*	-72.9	0.07*	-72.8	0.005	2.1	0.000*	-100.0
Flonicamid	70.0	3.31	-8.5	0.040*	-50.2	0.06*	-67.6	0.09*	-61.9	0.011	104.7	0.020	-9.8
Control	---	3.40	0.0	0.079	0.0	0.16	0.0	0.23	0.0	0.005	0.0	0.022	0.0
P		<0.001 (<0.001)		<0.001 (0.01)		<0.001 (0.002)		<0.001 (0.002)		0.42		0.046	
LSD (p = 0.05)		0.14		0.029		0.036		0.041		n.s.		0.02	
df		5, 159 (5, 40)		5, 159 (5, 40)		5, 159 (5, 40)		5, 159 (5, 40)		5, 159		5, 159	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 24: Whitefly x GVB Experiment: Mean abundance of leafhoppers and other Hemiptera pests in each insecticide treatment, ACRI, 2013/14.

	Rate	*Campylomma liebknechti*		Jassids		Other Hemiptera Pests	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
SeroX		0.60	-33.4	0.84*	-25.2	0.07	-28.3
Clothianidin	100.0	0.15	-87.1	0.70*	-42.9	0.06	-41.4
Dimethoate	140.0	0.33	-68.1	0.63*	-50.7	0.03	-66.2
Fipronil	8.0	0.22	-80.3	1.02	0.3	0.04	-61.1
Flonicamid	70.0	0.22	-80.0	0.72*	-40.7	0.05	-45.2
Control	---	0.80	0.0	1.02	0.0	0.10	0.0
P		<0.001, (<0.001)		<0.001, (<0.001)		<0.001 (0.023)	
LSD (p = 0.05)		0.071		0.095		0.051	
df		5, 159 (5, 40)		5, 159 (5, 40)		5, 159 (5, 40)	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 25: Whitefly x GVB Experiment: Mean abundance of Lepidoptera and *Helicoverpa* spp. in each insecticide treatment, ACRI, 2013/14.

	Rate	Total Lepidoptera		*Helicoverpa* Eggs		*Helicoverpa* Larvae	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
SeroX		0.046	-34.4	0.002	-88.6	0.044	-19.0
Clothianidin	100.0	0.049	-29.6	0.005	-65.7	0.041	-25.7
Dimethoate	140.0	0.036	-49.2	0.007	-54.2	0.027	-50.4
Fipronil	8.0	0.031	-55.8	0.005	-65.7	0.021	-62.5
Flonicamid	70.0	0.064	-7.8	0.005	-65.7	0.038	-30.6
Control	---	0.070	0.0	0.016	0.0	0.054	0.0
P		0.21		0.14		0.1	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df		5, 159		5, 159		5, 159	

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 26: Whitefly x GVB Experiment: Mean abundance of thrips (*Thrips tabaci* and *Frankliniella schultzei*) in each insecticide treatment, ACRI, 2013/14.

		Total Thrips				Thrips Adults				Thrips Nymphs			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
SeroX		1.17	18.6	2.34	-6.9	0.36	13.6	2.01	-1.4	1.04	19.9	1.27	-13.2
Clothianidin	100.0	1.19	22.0	2.50	12.0	0.40	27.9	2.05	3.1	1.03	18.8	1.54	24.8
Dimethoate	140.0	1.12	10.1	2.37	-3.4	0.33	3.6	1.91	-12.2	0.98	8.7	1.24	-16.7
Fipronil	8.0	1.07	2.2	2.24	-16.5	0.31	-4.5	1.84	-19.5	0.97	6.5	1.26	-14.2
Flonicamid	70.0	1.01	-7.0	2.38	-2.2	0.30	-6.9	1.94	-9.7	0.89	-5.4	1.26	-14.1
Control	---	1.06	0.0	2.40	0.0	0.32	0.0	2.03	0.0	0.93	0.0	1.37	0.0
P		0.023 (0.09)		0.005, (0.5)		0.065		0.012 (0.79)		0.196		<0.001, (0.52)	
LSD (p = 0.05)		n.s.		ns		n.s.		n.s.		n.s.		n.s.	
df		5, 177 (5, 45)		5, 159 (5, 40)		5, 177		5, 159 (5, 40)		5, 177		5, 159 (5, 40)	

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1).
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 27: Whitefly x GVB Experiment: Mean abundance of mites (*Tetranychus urticae*) in each insecticide treatment, ACRI, 2013/14.

		Total Mites		Mites Adults		Mites Nymphs	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
SeroX		0.83*	55.6	0.51*	50.0	0.54*	57.3
Clothianidin	100.0	1.33*	236.7	0.84*	196.8	1.06*	309.5
Dimethoate	140.0	0.79*	45.4	0.48	39.6	0.56*	61.5
Fipronil	8.0	0.92*	81.6	0.62*	94.9	0.61*	81.1
Flonicamid	70.0	1.03*	116.4	0.65*	104.7	0.76*	146.3
Control	---	0.61	0.0	0.37	0.0	0.38	0.0
P		<0.001 (<0.001)		<0.001(<0.001)		<0.001 (<0.001)	
LSD (p = 0.05)		0.17		0.13		0.15	
df		5, 177		5, 177		5, 177	

1. Values are means of transformed data from washes, i.e. ln (mean number of insects per m per sample +1).
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 28: Whitefly x GVB Experiment: Mean abundance of whitefly and aphids in each insecticide treatment, ACRI, 2013/14.

		Whitefly				Aphids			
	Rate	Washes (mainly adults)		Suction Samples (mainly adults)		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
SeroX		0.10*	112.5	3.29	21.7	0.018	1002.2	0.009	0.0
Clothianidin	100.0	0.07	46.8	2.58*	-42.2	0.005	182.9	0.009	0.0
Dimethoate	140.0	0.07	33.9	3.13	2.9	0.000	-100.0	0.012	40.7
Fipronil	8.0	0.09*	73.9	3.08	-1.6	0.012	632.0	0.005	-39.4
Flonicamid	70.0	0.09*	94.1	3.12	2.4	0.000	-100.0	0.004	-59.6
Control	---	0.05	0.0	3.10	0.0	0.002	0.0	0.009	0.0
P		0.019		<0.001 (<0.001)		0.3		0.7	
LSD (p = 0.05)		0.033		0.16		n.s.		n.s.	
df		5, 177		5, 159 (5, 40)		5, 177		5, 159	

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1).
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the controls

Appendix 4c: Options to manage mirids and GVB without flaring SLW, mites or aphids. Beneficial data for 2014-15.

Table 29: Mean abundance of predatory and pest Coleoptera in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

		Predatory Coleoptera											
	Rate	Total Coleoptera Beneficial		*Dicranolaius bellulus*		*Diomus notescens*		Total Coccinellids		Total other predatory beetles		Total Pest Coleoptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Clothianidin	100.00	0.641*	-53.01	0.496*	-60.72	0.104	74.77	0.108	48.22	0.105*	-40.61	0.743	-4.96
Fipronil + Salt	8.00	1.101	4.95	0.946	-3.72	0.157*	173.01	0.188*	168.83	0.152	-11.51	0.708	-11.09
Control	---	1.069	0.00	0.969	0.00	0.061	0.00	0.074	0.00	0.171	0.00	0.770	0.00
P		<0.001		<0.001		<0.001		<0.001		0.023		0.450	
LSD (p = 0.05)		0.094		0.088		0.048		0.050		0.048		n.s.	
df						(2, 83)							

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 30: Mean abundance of predatory Hemiptera in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

		Predatory Hemiptera											
	Rate	*Geocoris lubra*		*Orius spp.*		*Nabis kinbergiii*		*Deraeocoris signatus*		Other Beneficial Hemiptera		Total Predatory Hemiptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Clothianidin	100.00	0.023*	-53.99	0.151	54.36	0.012	-64.34	0.011*	-78.83	0.000	-100.00	0.195	-16.81
Fipronil + Salt	8.00	0.009*	-81.50	0.104	3.19	0.020	-36.40	0.039	-27.43	0.000	-100.00	0.166	-30.17
Control	---	0.049	0.00	0.101	0.00	0.032	0.00	0.053	0.00	0.002	0.00	0.231	0.00
P		0.008		0.071		0.107		0.007		0.374		0.055	
LSD (p = 0.05)		0.025		n.s.		n.s.		0.026		n.s.		n.s.	
df						(2, 83)							

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 31: Mean abundance of Hymenoptera in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

		Total Hymenoptera											
	Rate	Total Hymenoptera	(Wasp)	*Trichogramma*		*Microplitis*		*Telenomus*		*Eretmocerus* sp.		Other wasp spp.	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Clothianidin	100.00	1.038	8.18	0.043	2.38	0.053	-1.38	0.038	-0.15	0.648	-6.55	0.538*	31.77
Fipronil + Salt	8.00	1.146*	27.33	0.047	10.73	0.054	0.00	0.047	25.51	0.875*	43.46	0.434	0.46
Control	---	0.988	0.00	0.042	0.00	0.054	0.00	0.038	0.00	0.681	0.00	0.432	0.00
P		0.035		0.959		0.998		0.780		<0.001		0.044	
LSD (p = 0.05)		0.121		n.s.		n.s.		n.s.		0.111		0.094	
df						(2, 83)							

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 32: Mean abundance of *Neuroptera* in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

	Neuroptera								
	Rate	Total Neuroptera		LW Larvae		Green Adult		Brown Adult	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
Clothianidin	100.00	0.075*	-65.00	0.058*	-63.19	0.018	-64.81	0.000	-100.00
Fipronil + Salt	8.00	0.189	-6.66	0.132	-13.41	0.053	4.74	0.009	100.46
Control	---	0.201	0.00	0.151	0.00	0.051	0.00	0.005	0.00
P		<0.001		<0.001		0.065		0.111	
LSD (p = 0.05)		0.045		0.037		n.s.		n.s.	
df				(2, 83)					

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 33: Mean abundance Arachnids in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

		Arachnids					
	Rate	Total Spiders		Tangleweb		Other Spiders	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Clothianidin	100.00	0.788	-7.13	0.070	-0.96	0.754	-7.63
Fipronil + Salt	8.00	0.790	-6.68	0.056	-20.78	0.763	-6.11
Control	---	0.829	0.00	0.070	0.00	0.797	0.00
P		0.739		0.602		0.752	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df				(2, 83)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 34: Mean abundance of *Creontiades dilutus*, *Nezara viridula* and *Campylomma liebknechti* in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

	Rate	Total Pests	Hemiptera	*Creontiades dilutus Adult*		*Creontiades dilutus Nymph*		*Total Creontiades dilutus*		*Nezara viridula*		*Campylomma liebknechti*	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Clothianidin	100.00	4.247	-3.78	0.052*	-44.14	0.102*	-37.57	0.146*	-40.30	0.038	-54.65	0.871*	-52.06
Fipronil + Salt	8.00	4.216	-6.74	0.025*	-73.17	0.082*	-50.40	0.104*	-58.34	0.063	-24.66	0.855*	-53.30
Control	---	4.285	0.00	0.091	0.00	0.159	0.00	0.234	0.00	0.083	0.00	1.360	0.00
P		0.631		0.003		0.002		<0.001		0.086		<0.001	
LSD (p = 0.05)		n.s.		0.037		0.043		0.053		n.s.		0.090	
df						(2, 83)							

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 35: Mean abundance of cotton seed bug, Rutherglen bug, leafhoppers and other Hemiptera pests in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

	Rate	Rutherglen bug		Jassids		Other Hemiptera Pest	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Clothianidin	100.00	0.073	-22.64	0.871	-4.15	0.095	-13.65
Fipronil + Salt	8.00	0.081	-14.35	0.932	6.12	0.072	-36.07
Control	---	0.094	0.00	0.896	0.00	0.110	0.00
P		0.602		0.485		0.209	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df				(2, 83)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 36: Mean abundance of Lepidoptera and *Helicoverpa* spp. in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

	Rate	Total Lepidoptera		*Helicoverpa* Eggs		*Helicoverpa* Larvae	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Clothianidin	100.00	0.021	16.21	0.000	0.00	0.000	0.00
Fipronil + Salt	8.00	0.025	40.93	0.000	0.00	0.000	0.00
Control	---	0.018	0.00	0.000	0.00	0.000	0.00
P		0.082		0.000		0.000	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df				(2, 83)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 37: Mean abundance of thrips (*Thrips tabaci* and *Frankliniella schultzei*) in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

		Total Thrips				Thrips Adults				Thrips Larvae			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Clothianidin	100.00	0.765	7.49	1.489*	29.40	0.234	18.23	1.376	21.27	0.645		0.391*	113.67
Fipronil + Salt	8.00	0.518*	-36.50	1.184	-14.48	0.127*	-39.09	1.102	-17.60	0.434*		0.232	16.73
Control	---	0.727	0.00	1.296	0.00	0.201	0.00	1.236	0.00	0.624		0.202	0.00
P		<0.001		0.003		0.002		0.006		<0.001		0.003	
LSD (p = 0.05)		0.105		0.171		0.059		0.165		0.106		0.084	
df						(2, 83)							

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 38: Mean abundance of mites (*Tetranychus urticae*) in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

		Total Mites				Mites Adults				Mites Nymphs			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Clothianidin	100.00	0.871*	131.13	0.706*	192.48	0.485*	128.09	0.519*	307.33	0.658*	164.43	0.375*	140.92
Fipronil + Salt	8.00	0.758*	88.62	0.764*	227.00	0.447*	106.00	0.491*	279.74	0.516*	91.61	0.510*	252.57
Control	---	0.471	0.00	0.301	0.00	0.242	0.00	0.154	0.00	0.302	0.00	0.173	0.00
P		<0.001		<0.001		<0.001		<0.001		<0.001		0.001	
LSD (p = 0.05)		0.133		0.145		0.101		0.098		0.129		0.176	
df						(2, 83)							

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 39: Mean abundance of whitefly and aphids in each insecticide treatment, ACRI, 2014/15, Whitefly x GVB Experiment.

		Whitefly				Aphids			
	Rate	Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
Clothianidin	100.00	1.823	2.37	4.144	-2.17	0.000	-100.00	0.018	97.85
Fipronil + Salt	8.00	1.833	3.51	4.099	-6.48	0.011	27.70	0.030	225.29
Control	---	1.804	0.00	4.165	0.00	0.009	0.00	0.009	0.00
P		0.963		0.683		0.306		0.055	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.	
df						(2, 83)			

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

APPENDIX 5: Section B iii) Investigating alternative seed treatments for early season pests

Appendix 5a: Beneficial data, Seed Treatments, ACRI, 2014/15

Table 1: Mean abundance of predatory and pest Coleoptera in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

		Predatory Coleoptera											
	Rate	Total Coleoptera Beneficial		[redacted]		*Diomus notescens*		Total Coccinellids		Total other predatory beetles		Total Pest Coleoptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser		0.051*	-53.90	0.029	-10.97	0.004*	-92.57	0.023*	-70.65	0.000	-100.00	0.247	-11.53
Cruiser Extreme		0.055*	-50.03	0.029	-10.97	0.015*	-70.55	0.026*	-66.22	0.000	-100.00	0.300	10.82
Genero		0.061	-44.54	0.011	-66.62	0.022	-55.87	0.041*	-46.57	0.011	195.37	0.179	-37.85
Thimet		0.149	42.25	0.059	79.65	0.054	7.59	0.095	29.06	0.004	0.00	0.206	-27.51
Control	---	0.107	0.00	0.033	0.00	0.050	0.00	0.075	0.00	0.004	0.00	0.275	0.00
P		<0.001		0.216		0.001		<0.001		0.0235		0.353	
LSD (p = 0.05)		0.047		n.s.		0.032		0.032		n.s.		n.s.	
df							(4, 79)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 2: Mean abundance of predatory Hemiptera in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

		Predatory Hemiptera											
	Rate	*Geocoris lubra*		[redacted]		*Nabis kinbergiii*		*Deraeocoris signatus*		Other Beneficial Hemiptera		Total Predatory Hemiptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser		0.015	36.20	0.000	0.00	0.011	3.84	0.000	0.00	0.018	1.80	0.043	98.75
Cruiser Extreme		0.000	-100.00	0.000	0.00	0.011	3.84	0.000	0.00	0.000	0.00	0.011	-49.34
Genero		0.015	36.20	0.000	0.00	0.011	5.87	0.000	0.00	0.004	0.30	0.030	38.05
Thimet		0.022	101.12	0.004	0.30	0.011	5.87	0.000	0.00	0.000	0.00	0.037	71.43
Control		0.011	0.00	0.000	0.00	0.011	0.00	0.000	0.00	0.000	0.00	0.022	0.00
P		0.199		0.415		1.000		0.000		0.047		0.322	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.		0.014		n.s.	
df							(4, 79)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 3: Mean abundance of Hymenoptera in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

		Total Hymenoptera											
	Rate	Total (Wasp) Hymenoptera		[redacted]		*Microplitis*		*Telenomus*		Other wasp spp.		Ants	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser		0.518	-12.63	0.210	-8.55	0.050	-30.79	0.004	-74.78	0.348	-16.98	0.135	-23.00
Cruiser Extreme		0.497	-17.08	0.202	-12.04	0.070	-2.60	0.004	-74.78	0.327	-22.92	0.230	37.50
Genero		0.515	-13.34	0.254	13.36	0.062	-14.07	0.000*	-100.00	0.309	-27.87	0.147	-16.10
Thimet		0.511	-14.16	0.221	-2.85	0.070	-2.74	0.019	27.07	0.333	-21.29	0.130	-26.16
Control		0.575	0.00	0.227	0.00	0.072	0.00	0.015	0.00	0.407	0.00	0.173	0.00
P		0.646		0.836		0.810		0.021		0.170		0.638	
LSD (p = 0.05)		n.s.		n.s.		n.s.		0.013		n.s.		n.s.	
df							(4, 79)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 4: Mean abundance of *Neuroptera* in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

		Neuroptera							
	Rate	Total Neuroptera		LW Larvae		Green Adult		Brown Adult	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
Cruiser		0.004	0.30	0.004	0.30	0.000	0.00	0.000	0.00
Cruiser Extreme		0.000	0.00	0.000	0.00	0.000	0.00	0.000	0.00
Genero		0.004	0.30	0.000	0.00	0.004	0.30	0.000	0.00
Thimet		0.000	0.00	0.000	0.00	0.000	0.00	0.000	0.00
Control		0.000	0.00	0.000	0.00	0.000	0.00	0.000	0.00
P		0.571		0.415		0.415		0.000	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.	
df					(4, 79)				

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 5: Mean abundance Arachnids in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

		Arachnids					
	Rate	Total Spiders		Tangleweb		Other Spiders	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser		0.893	-11.58	0.052	-1.27	0.869	-12.31
Cruiser Extreme		0.835	-19.98	0.034	-36.62	0.821	-19.39
Genero		0.786	-26.78	0.055	4.66	0.761	-27.79
Thimet		0.855	-17.20	0.048	-9.02	0.836	-17.19
Control		0.968	0.00	0.053	0.00	0.947	0.00
P		0.162		0.740		0.164	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df				(4, 79)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 6: Mean abundance of *Creontiades dilutus*, *Nezara viridula* and *Campylomma liebknechti* in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15

	Rate	Total Hemiptera Pests		*Creontiades dilutus Adult*		*Creontiades dilutus Nymph*		*Total Creontiades dilutus*		*Nezara viridula*		*Campylomma liebknechti*	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser		1.609	-7.77	0.027	76.00	0.000	-100.00	0.027	4.84	0.000	0.00	0.044	-28.30
Cruiser Extreme		1.522	-17.44	0.011	-25.14	0.000	-100.00	0.011	-55.41	0.000	0.00	0.011	-81.95
Genero		1.419	-27.79	0.019	25.24	0.000	-100.00	0.019	-25.40	0.000	0.00	0.032	-48.22
Thimet		1.666	-1.05	0.025	68.88	0.000	-100.00	0.025	0.60	0.000	0.00	0.047	-23.67
Control		1.675	0.00	0.015	0.00	0.011	0.00	0.025	0.00	0.000	0.00	0.061	0.00
P		0.104		0.690		0.415		0.828		0.000		0.083	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.		n.s.		n.s.	
df						(4, 79)							

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 7: Mean abundance of cotton seed bug, Rutherglen bug, leafhoppers and other Hemiptera pests in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

	Rate	Rutherglen bug		Jassids		Other Hemiptera Pest	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser		0.402	-18.97	1.438	-2.71	0.064	-32.84
Cruiser Extreme		0.384	-23.42	1.360	-12.21	0.038	-61.12
Genero		0.384	-23.32	1.250	-24.53	0.044	-54.18
Thimet		0.477	0.01	1.433	-3.25	0.066	-31.27
Control		0.477	0.00	1.459	0.00	0.094	0.00
P		0.779		0.371		0.153	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df				(4, 79)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 8: Mean abundance of Lepidoptera and *Helicoverpa* spp. in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

	Rate	Total Lepidoptera		*Helicoverpa* Eggs		*Helicoverpa* Larvae	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser		0.037	24.92	0.019	151.43	0.015	-34.53
Cruiser Extreme		0.015	-49.67	0.004	-50.09	0.011	-48.86
Genero		0.015	-49.67	0.008	0.00	0.007	-66.95
Thimet		0.015	-49.67	0.007	-2.87	0.008	-65.97
Control		0.029	0.00	0.008	0.00	0.022	0.00
P		0.490		0.464		0.759	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df				(4, 79)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 9: Mean abundance of thrips in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

		Total Thrips				Thrips Adults				Thrips Larvae			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser		1.318	5.67	0.883	9.32	0.476	-2.52	0.733	-7.64	1.096	5.10	0.260*	108.47
Cruiser Extreme		1.066	-26.49	0.582*	-39.18	0.474	-3.11	0.514*	-42.57	0.817	-33.31	0.096	-29.03
Genero		1.202	-10.15	0.631*	-32.20	0.457	-7.43	0.538*	-39.08	0.977	-12.62	0.135	1.60
Thimet		1.242	-4.81	0.770	-10.51	0.526	10.58	0.697	-13.84	0.994	-10.18	0.130	-2.42
Control		1.278	0.00	0.832	0.00	0.486	0.00	0.775	0.00	1.063	0.00	0.133	0.00
P		0.182		0.010		0.789		0.016		0.149		0.016	
LSD (p = 0.05)		n.s.		0.191		n.s.		0.182		n.s.		0.098	
df		(4, 79) for suction samples, (4, 72) for washes											

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 10: Mean abundance of mites (*Tetranychus urticae*) in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

		Total Mites				Mites Adults				Mites Nymphs			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser		0.000	0.00	0.000	-100.00	n/a		0.000	-100.00	n/a		0.000	0.00
Cruiser Extreme		0.002	0.20	0.000	-100.00	n/a		0.000	-100.00	n/a		0.000	0.00
Genero		0.000	0.00	0.004	-50.09	n/a		0.004	-50.09	n/a		0.000	0.00
Thimet		0.000	0.00	0.099	1263.07	n/a		0.078	970.67	n/a		0.045	4.60
Control		0.000	0.00	0.008	0.00	n/a		0.008	0.00	n/a		0.000	0.00
P		0.413		0.405				0.456				0.324	
LSD (p = 0.05)		n.s.		n.s.				n.s.				n.s.	
df		(4, 79) for suction samples, (4, 72) for washes											

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 11: Mean abundance of whitefly and aphids in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

		Whitefly				Aphids			
	Rate	Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
Cruiser	n/a			0.127	25.20	0.476	-2.52	0.000	0.00
Cruiser Extreme	n/a			0.120	16.90	0.474	-3.11	0.004	0.30
Genero	n/a			0.058	-45.44	0.457	-7.43	0.000	0.00
Thimet	n/a			0.113	10.08	0.526	10.58	0.004	0.30
Control	n/a			0.103	0.00	0.486	0.00	0.000	0.00
P				0.343		0.798		0.571	
LSD (p = 0.05)				n.s.		n.s.		n.s.	
df				(4, 79) for suction samples, (4, 72) for washes					

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 12: Mean abundance of leafhoppers, spiders and wasps in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2014/15.

	Rate	Jassids		Spiders		Wasps	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser		0.049	-11.06	0.119	-27.52	0.085	55.80
Cruiser Extreme		0.033	-39.85	0.096*	-42.12	0.057	2.34
Genero		0.058	6.16	0.098*	-41.34	0.066	19.95
Thimet		0.041	-25.49	0.094*	-43.71	0.113*	108.23
Control		0.054	0.00	0.161	0.00	0.056	0.00
P		0.682		0.019		0.046	
LSD (p = 0.05)		n.s.		0.045		0.042	
df				(4, 72)			

1. Values are means of transformed data from washes, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Appendix 5b: Beneficial data, Seed Treatments, ACRI, 2015/16

Table 13: Mean abundance of predatory and pest Coleoptera in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

		Predatory Coleoptera											
	Rate	Total Coleoptera Beneficial		[redacted]		*Diomus notescens*		Total Coccinellids		Total other predatory beetles		Total Pest Coleoptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser Extreme		0.33	27.98	0.176	29.82	0.064	16.88	0.147	69.19	0.016	-62.93	0.073	-5.02
CruiserX+Thiodicarb		0.34	34.61	0.209	54.37	0.066	19.75	0.135	55.18	0.004*	-90.65	0.052	-32.11
CruiserX+Thiodicarb +Fipronil		0.39	48.94	0.218	61.24	0.097	77.00	0.175	101.86	0.004*	-90.65	0.050	-35.80
Thimet		0.19	-27.24	0.073	-45.97	0.081	47.89	0.098	13.25	0.020	-53.43	0.086	11.16
Control	---	0.26	0.00	0.135	0.00	0.055	0.00	0.087	0.00	0.043	0.00	0.077	0.00
P		0.034		0.148		0.708		0.200		0.034		0.582	
LSD (p = 0.05)		0.027		n.s.		n.s.		n.s.		0.027		n.s.	
df							(4, 79)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 14: Mean abundance of predatory Hemiptera in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

		Predatory Hemiptera											
	Rate	*Geocoris lubra*		[redacted]		*Nabis kinbergii*		*Deraeocoris signatus*		Other Beneficial Hemiptera		Total Predatory Hemiptera	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser Extreme		0.012	55.02	0.000	0.00	0.008	0.08	0.004	0.04	0.000	0.00	0.033	314.38
CruiserX+Thiodicarb		0.000	-100.00	0.000	0.00	0.000	0.00	0.008	0.08	0.000	0.00	0.008	3.14
CruiserX+Thiodicarb +Fipronil		0.008	0.00	0.000	0.00	0.008	0.08	0.000	0.00	0.008	0.08	0.023	194.20
Thimet		0.000	-100.00	0.000	0.00	0.000	0.00	0.000	0.00	0.000	0.00	0.000	-100.00
Control		0.008	0.00	0.000	0.00	0.000	0.00	0.000	0.00	0.000	0.00	0.008	0.00
P		0.469		0.000		0.421		0.171		0.112		0.084	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.		n.s.		n.s.	
df							(4, 79)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 15: Mean abundance of Hymenoptera in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

		Total Hymenoptera											
	Rate	Total (Wasp) Hymenoptera		[redacted]		*Microplitis*		*Telenomus*		Other wasp spp.		Ants	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser Extreme		0.489	-5.53	0.036	-46.72	0.033	103.18	0.114*	599.60	0.272	-32.37	0.286	15.52
CruiserX+Thiodicarb		0.477	-7.85	0.040	-41.26	0.016	0.00	0.004	-75.15	0.421	4.68	0.328	32.36
CruiserX+Thiodicarb +Fipronil		0.594	14.77	0.044	-34.70	0.041	153.81	0.000	-100.00	0.507	26.11	0.192	-22.33
Thimet		0.740	43.00	0.071	5.12	0.029	78.80	0.004	-75.15	0.629	56.40	0.511*	106.42
Control		0.517	0.00	0.068	0.00	0.016	0.00	0.016	0.00	0.402	0.00	0.247	0.00
P		0.343		0.600		0.221		<0.001		0.083		0.009	
LSD (p = 0.05)		n.s.		n.s.		n.s.		0.017		n.s.		0.131	
df							(4, 79)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 16: Mean abundance of *Neuroptera* in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

		Neuroptera							
	Rate	Total Neuroptera		LW Larvae		Green Adult		Brown Adult	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
Cruiser Extreme		0.008	0.00	0.008	0.00	0.000	0.00	0.000	0.00
CruiserX+Thiodicarb		0.016	97.74	0.012	50.30	0.000	0.00	0.004	0.04
CruiserX+Thiodicarb+Fipronil		0.020	151.52	0.012	50.30	0.000	0.00	0.008	0.08
Thimet		0.012	50.30	0.008	0.00	0.000	0.00	0.004	0.04
Control		0.008	0.00	0.008	0.00	0.000	0.00	0.000	0.00
P		0.676		0.960		0.000		0.395	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.	
df					(4, 79)				

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 17: Mean abundance Arachnids in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

		Arachnids					
	Rate	Total Spiders		Tangleweb		Other Spiders	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser Extreme		1.304*	23.49	0.033	-18.36	1.270	25.08
CruiserX+Thiodicarb		1.271*	20.32	0.159*	296.73	1.047	3.04
CruiserX+Thiodicarb+Fipronil		1.522*	44.06	0.098	143.89	1.429	40.68
Thimet		1.046	-0.93	0.076	89.78	0.971	-4.35
Control		1.056	0.00	0.040	0.00	1.016	0.00
P		0.044		0.016		0.020	
LSD (p = 0.05)		0.153		0.073		0.142	
df				(4, 79)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 18: Mean abundance of *Creontiades dilutus*, *Nezara viridula* and *Campylomma liebknechti* in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

	Rate	Total Hemiptera Pests		*Creontiades dilutus Adult*		*Creontiades dilutus Nymph*		*Total Creontiades dilutus*		*Nezara viridula*		*Campylomma liebknechti*	
Insecticide	g ai/ha	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser Extreme		2.775*	-27.85	0.052	-7.52	0.028	-0.79	0.080	-5.37	0.000	0.00	0.128	23.18
CruiserX+Thiodicarb		2.637*	-31.44	0.040	-29.04	0.028	-0.89	0.068	-18.97	0.000	0.00	0.161	55.71
CruiserX+Thiodicarb +Fipronil		3.240*	-15.74	0.076	33.94	0.024	-12.93	0.100	18.31	0.000	0.00	0.180	73.42
Thimet		2.372*	-38.32	0.068	19.86	0.004	-85.63	0.071	-15.31	0.000	0.00	0.159	53.13
Control		3.846	0.00	0.057	0.00	0.028	0.00	0.084	0.00	0.000	0.00	0.104	0.00
P		0.005		0.559		0.387		0.824		0.000		0.336	
LSD (p = 0.05)		0.196		n.s.		n.s.		n.s.		n.s.		n.s.	
df						(4, 79)							

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 19: Mean abundance of cotton seed bug, Rutherglen bug, leafhoppers and other Hemiptera pests in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

	Rate	Rutherglen bug		Jassids		Other Hemiptera Pest	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser Extreme		0.180	25.11	2.131	-34.79	0.052	-8.72
CruiserX+Thiodicarb		0.187	29.99	2.009	-38.52	0.057	-1.30
CruiserX+Thiodicarb+Fipronil		0.241	67.67	2.472	-24.35	0.080	39.13
Thimet		0.204	41.92	1.697	-48.07	0.056	-2.06
Control		0.144	0.00	3.268	0.00	0.057	0.00
P		0.404		<0.001		0.785	
LSD (p = 0.05)		n.s.		0.204		n.s.	
df				(4, 79)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 20: Mean abundance of Lepidoptera and *Helicoverpa* spp. in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

	Rate	Total Lepidoptera		*Helicoverpa* Eggs		*Helicoverpa* Larvae	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser Extreme		0.000	0.00	0.047	31.30	0.028	-0.80
CruiserX+Thiodicarb		0.000	0.00	0.023	-35.34	0.024	-13.04
CruiserX+Thiodicarb+Fipronil		0.000	0.00	0.040	12.43	0.020	-26.93
Thimet		0.000	0.00	0.032	-9.63	0.012	-57.23
Control		0.000	0.00	0.036	0.00	0.028	0.00
P		0.000		0.777		0.756	
LSD (p = 0.05)		n.s.		n.s.		n.s.	
df				(4, 79)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 21: Mean abundance of thrips in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

		Total Thrips				Thrips Adults				Thrips Larvae			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser Extreme		6.188	28.32	0.004	0.00	0.931	16.41	0.874	8.53	5.256	31.41	0.465	18.88
CruiserX+Thiodicarb		4.756	-1.36	0.008	100.40	0.850	6.25	0.883	9.64	3.906	-2.34	0.509	30.33
CruiserX+Thiodicarb +Fipronil		5.438	12.77	0.016	296.28	0.756	-5.47	1.257	56.06	4.675	16.88	0.723*	84.96
Thimet		3.138*	-34.93	0.012	195.09	0.666	-16.80	0.680	-15.57	2.472*	-38.20	0.201*	-48.62
Control		4.822	0.00	0.004	0.00	0.800	0.00	0.806	0.00	4.000	0.00	0.391	0.00
P		0.002		0.671		0.471		0.055		0.001		0.002	
LSD (p = 0.05)		1.460		n.s.		n.s.		0.182		1.280		0.171	
df							(4, 79)						

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m (or per plant) per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 22: Mean abundance of mites (*Tetranychus urticae*) in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

		Total Mites				Mites Adults				Mites Nymphs			
	Rate	Washes		Suction Samples		Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser Extreme		0.003	0.03	0.004	0.00	n.a.		0.004	0.00	n.a.		0.000	0.00
CruiserX+Thiodicarb		0.000	0.00	0.004	0.00			0.004	0.00			0.000	0.00
CruiserX+Thiodicarb+Fipronil		0.012	1.20	0.004	0.00			0.004	0.00			0.000	0.00
Thimet		0.000	0.00	0.004	0.00			0.004	0.00			0.000	0.00
Control		0.000	0.00	0.004	0.00			0.004	0.00			0.000	0.00
P		0.172		1.000				1.000				0.000	
LSD (p = 0.05)		n.s.		n.s.				n.s.				n.s.	
df							(4, 79)						

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m (or per plant) per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 23: Mean abundance of whitefly and aphids in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

		Whitefly				Aphids			
	Rate	Washes		Suction Samples		Washes		Suction Samples	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
Cruiser Extreme		0.000	0.00	0.200	155.35	0.000	0.00	0.000	-100.00
CruiserX+Thiodicarb		0.000	0.00	0.115	46.63	0.000	0.00	0.000	-100.00
CruiserX+Thiodicarb+Fipronil		0.000	0.00	0.118	50.83	0.000	0.00	0.008	0.00
Thimet		0.021	0.00	0.117	48.82	0.000	0.00	0.004	-50.10
Control		0.000	0.00	0.078	0.00	0.000	0.00	0.008	0.00
P		0.415		0.210		0.000		0.361	
LSD (p = 0.05)		n.s.		n.s.		n.s.		n.s.	
df						(4, 79)			

1. Values are means of transformed data from suction samples or washes, i.e. ln (mean number of insects per m (or per plant) per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 24: Mean abundance of leafhoppers, spiders and wasps in each insecticide seed treatment, Seed Treatment Experiment, ACRI, 2015/16.

	Rate	Jassids		Spiders		Wasps	
Insecticide	(g ai/ha)	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser		0.146	-31.66	0.155	20.26	0.084	-32.72
Cruiser Extreme		0.198	-7.13	0.269*	108.06	0.139	12.20
Genero		0.149	-30.19	0.173	33.98	0.087	-30.05
Thimet		0.134	-37.07	0.131	1.28	0.082	-33.90
Control		0.213	0.00	0.129	0.00	0.124	0.00
P		0.586		0.029		0.515	
LSD (p = 0.05)		n.s.		0.095		n.s.	
df				(4, 79)			

1. Values are means of transformed data from washes, i.e. ln (mean number of insects per plant per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Appendix 5c: Beneficial data, Seed Treatments, ACRI, 2014/15

Table 25: Mean abundance of beneficial Hymenoptera in each insecticide seed treatment, Seed Treatment Experiment, ACRI B17 2016/17

	Beneficial Hymenoptera											
	Ants Total		[redacted]		Wasp Total		*Trichogramma sp.*		*Microplitis sp.*		*Telenomus sp.*	
Insecticide	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser X	0.019	-81.61	0.000	-100.00	0.661	-3.25	0.255	-15.04	0.049	157.79	0.021	102.27
CruiserX + Thiodicarb + Fipronil	0.021	-79.05	0.000	-100.00	0.655	-4.26	0.237	-21.13	0.027	43.76	0.008	-23.53
Imidacloprid + Thiodicarb + Fipronil	0.035	-66.26	0.000	-100.00	0.572	-16.40	0.271	-9.77	0.021	11.09	0.000	-100.00
Thimet	0.082	-20.00	0.016	100.80	0.615	-10.03	0.265	-11.78	0.041	114.19	0.008	-23.53
Control	0.102	0.00	0.008	0.00	0.684	0.00	0.300	0.00	0.019	0.00	0.010	0.00
P	<0.001		0.025		0.046 (i)		0.018 (i)		0.003(i)		0.009 (i)	
LSD (p = 0.05)	0.040		0.012		0.220		0.177		0.056		0.031	
df						(4, 72)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control
4. (i) refers to the interaction values

Table 26: Mean abundance of beneficial coleopteran and spiders in each insecticide seed treatment, Seed Treatment Experiment, ACRI B17, 2016/17.

	Ben. Coleoptera Total		Red & Blue Beetle		Tangleweb Spiders	
Insecticide	Mean[1]	%[2]	Mean	%	Mean	%
Cruiser X	0.024	-25.19	0.008	1.75	0.030	-62.07
CruiserX + Thiodicarb + Fipronil	0.037	12.67	0.005	-32.26	0.062	-20.05
Imidacloprid + Thiodicarb + Fipronil	0.068	107.79	0.027	238.83	0.083	6.21
Thimet	0.021	-34.52	0.008	1.75	0.116	48.06
Control	0.033	0.00	0.008	0.00	0.078	0.00
P	0.024		0.013		0.002	
LSD (p = 0.05)	0.029		0.013		0.039	
df			(4, 72)			

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

Table 27: Mean abundance of Hemiptera pest species and mites in each insecticide seed treatment, Seed Treatment Experiment, ACRI B17 2016/17

	Pest Hemiptera Total		Jassid Tot[redacted]		Jassid Adults		Jassid Immatures		Mirid Immatures		Mite Immatures	
Insecticide	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Cruiser X	6.655	-9.98	5.867	-11.55	5.480	2.71	0.358	-65.17	0.022	-56.55	0.000	0.00
CruiserX + Thiodicarb + Fipronil	6.793	-8.12	6.089	-8.20	5.592	4.82	0.440	-57.20	0.013	-73.42	0.008	0.80
Imidacloprid + Thiodicarb + Fipronil	7.191	-2.73	6.405	-3.44	5.299	-0.67	0.883	-14.12	0.013	-72.88	0.003	0.03
Thimet	4.430	-40.08	3.880	-41.51	3.622	-32.12	0.252	-75.54	0.016	-67.07	0.000	0.00
Control	7.393	0.00	6.633	0.00	5.335	0.00	1.029	0.00	0.050	0.00	0.000	0.00
P	<0.001		<0.001		0.001		<0.001		0.012		<0.004	
LSD (p = 0.05)	0.156		0.160		0.150		0.132		0.022		0.005	
df						(4, 72)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

NB: New F values for significant interactions of TRT x Date at df = (4, 16):
Hemiptera Pest Total – P = 0.002*
Jassid Total – P = 0.007*
Jassid Adult – P = <0.001**
Jassid Immature – P = <0.001**
Mirid Immature – P = <0.001**
Mite Immature – P = 0.014*

APPENDIX 6: Section B (v)

Whitefly x Chemistry Experiment, ACRI 2017/18

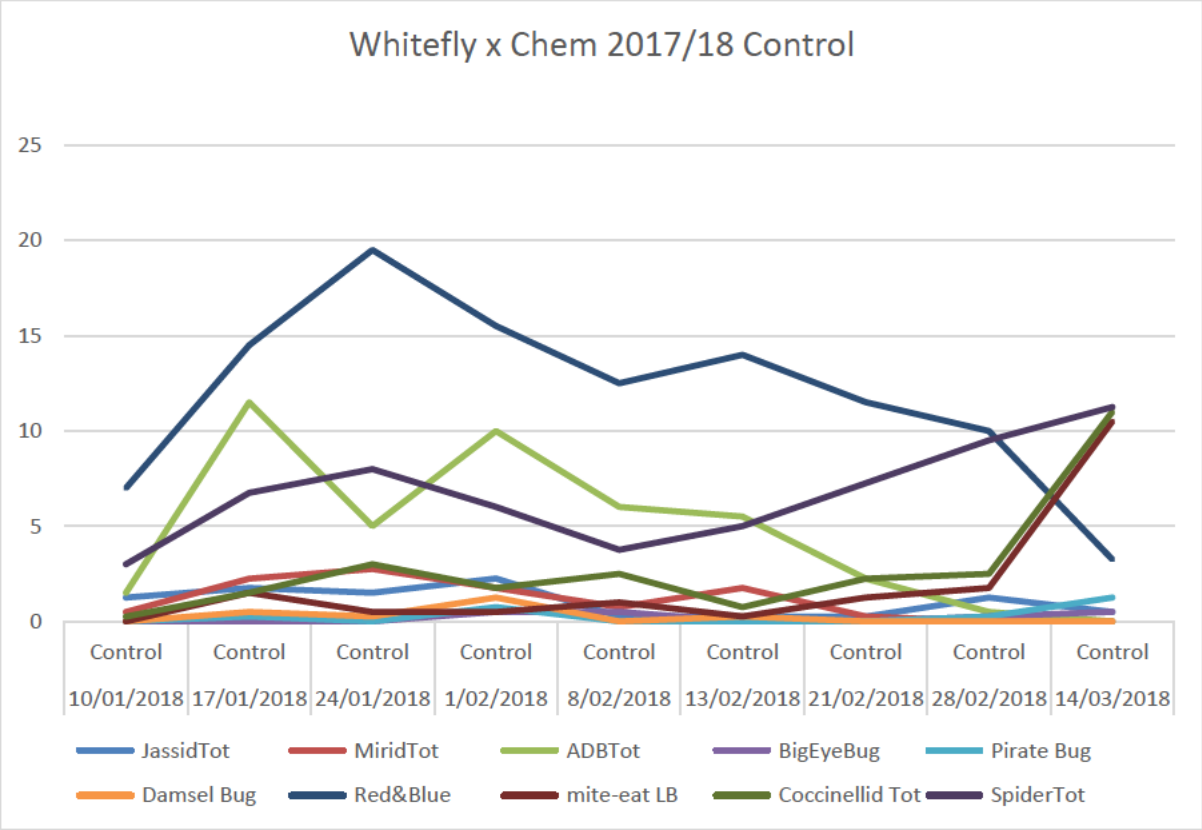


Figure 1: Effect of no sprays on mirids and beneficials

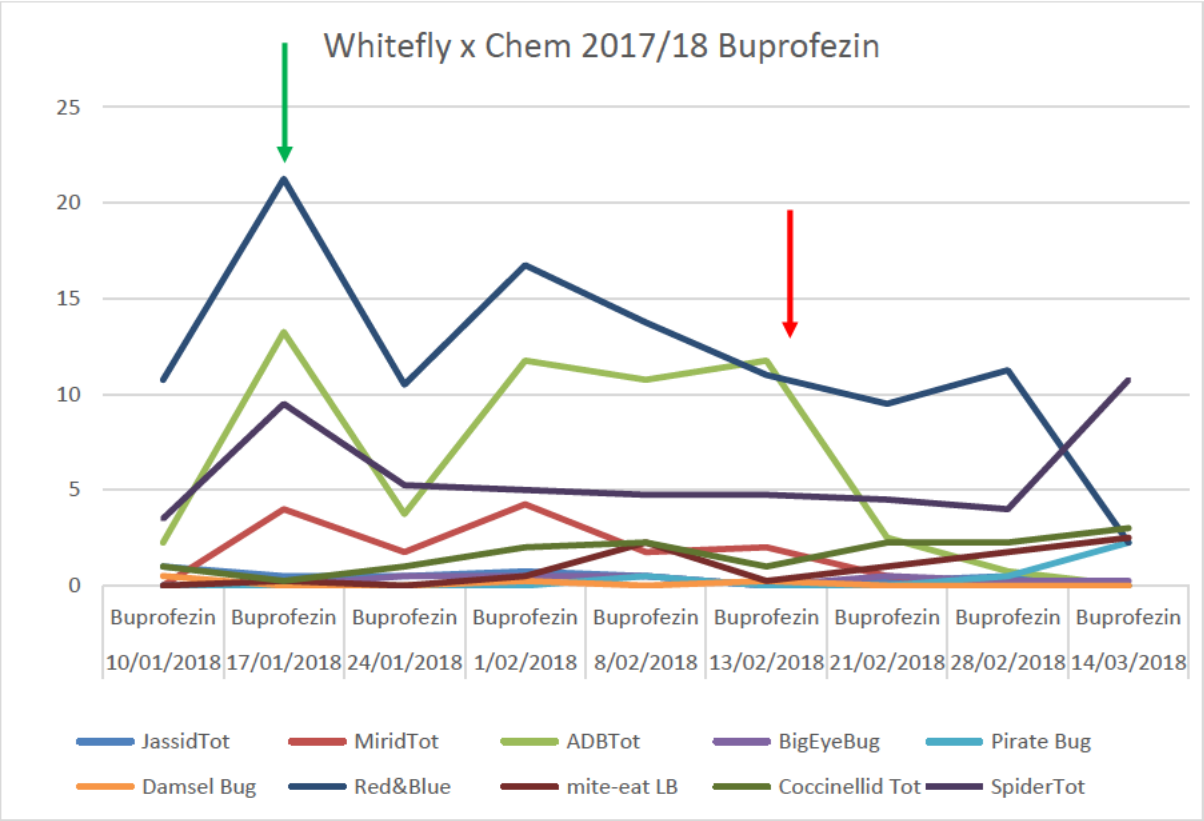


Figure 2: Effects of dimethoate spray (green arrow) and buprofezin spray (red arrow) on mirids and beneficials

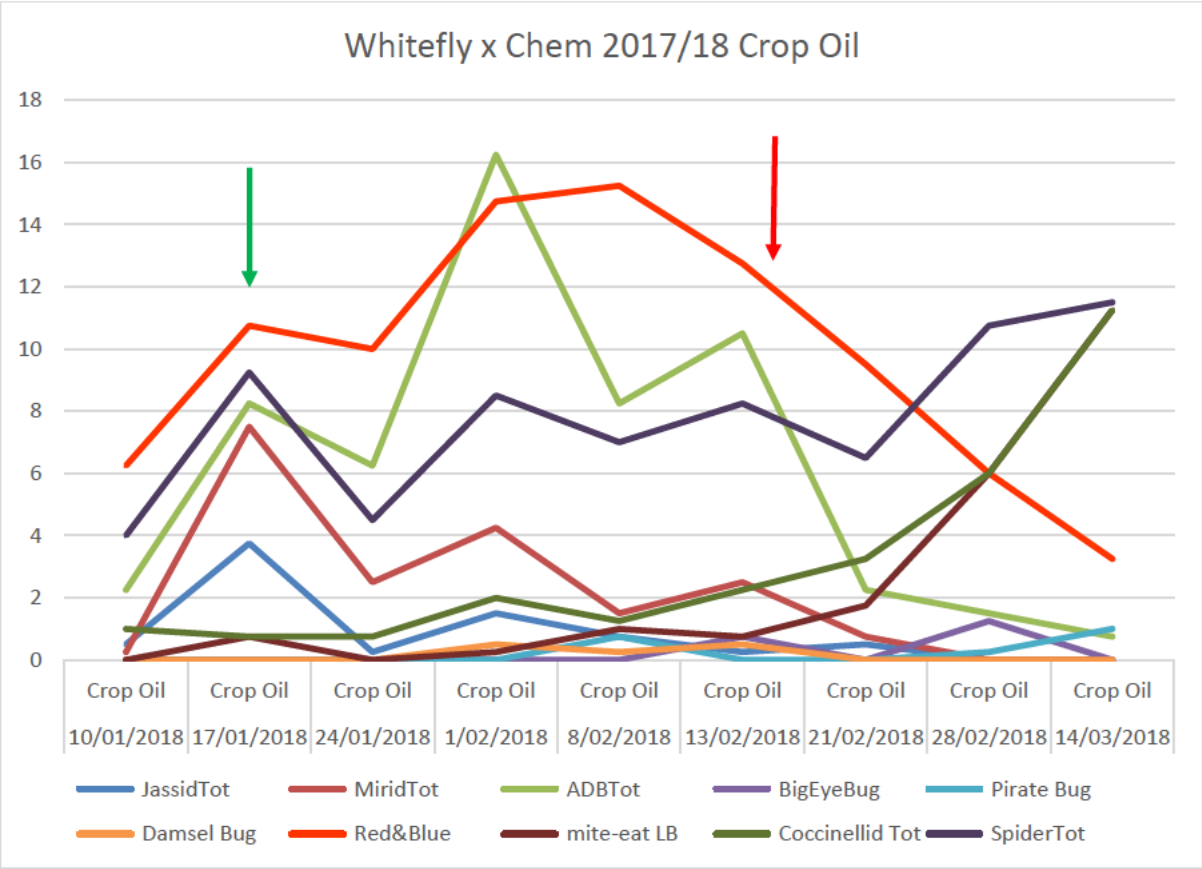


Figure 3: Effects of dimethoate spray (green arrow) and crop oil spray (red arrow) on mirids and beneficials

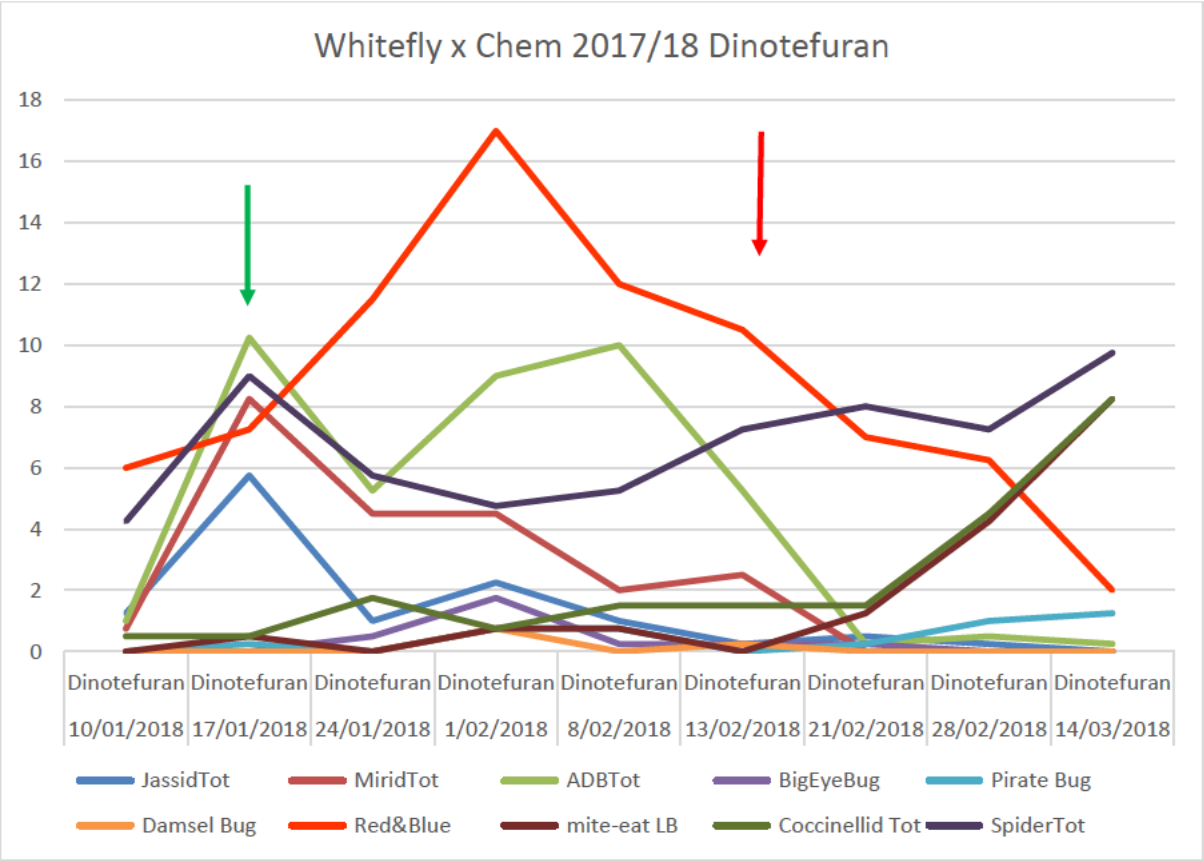


Figure xx: Effects of dimethoate spray (green arrow) and dinotefuran spray (red arrow) on mirids and beneficials

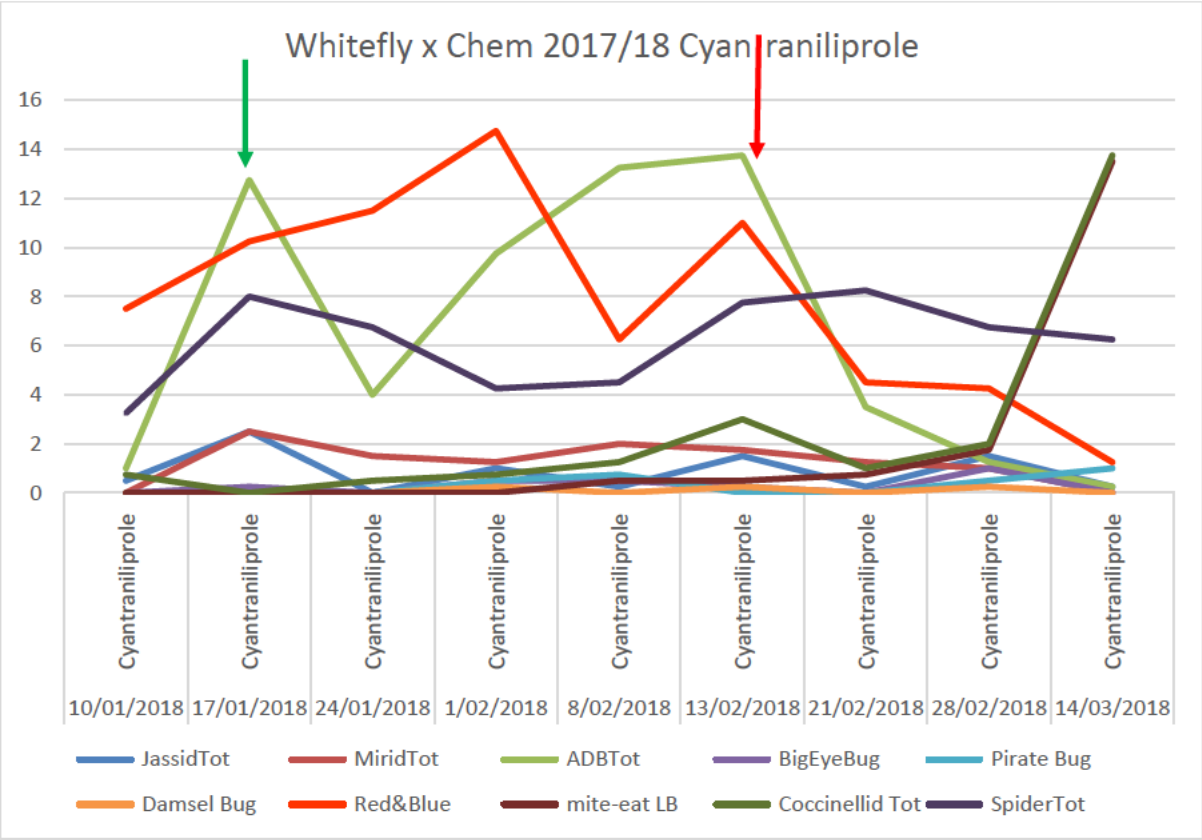


Figure 4: Effects of dimethoate spray (green arrow) and cyantraniliprole spray (red arrow) on mirids and beneficials

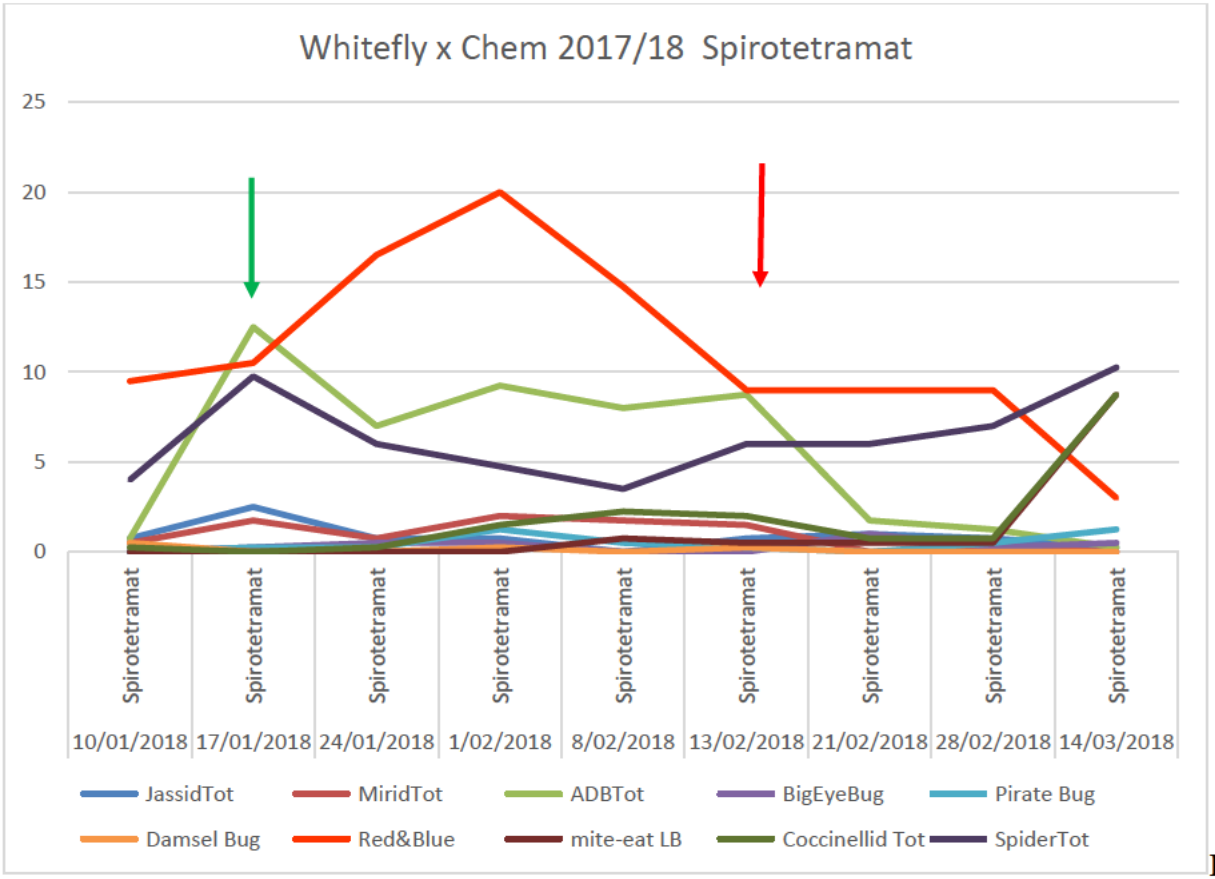


Figure 5: Effects of dimethoate spray (green arrow) and spirotetramat spray (red arrow) on mirids and beneficials

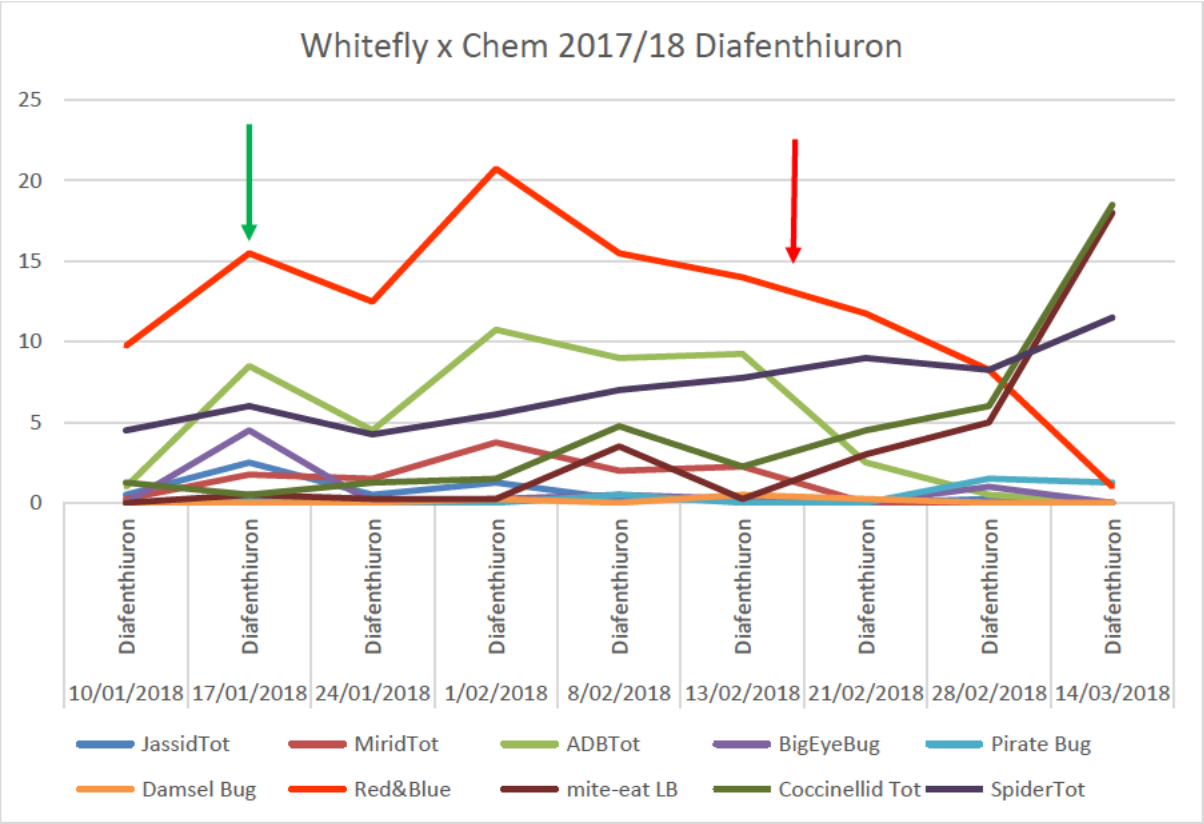


Figure 6: Effects of dimethoate spray (green arrow) and diafenthiuron spray (red arrow) on mirids and beneficials

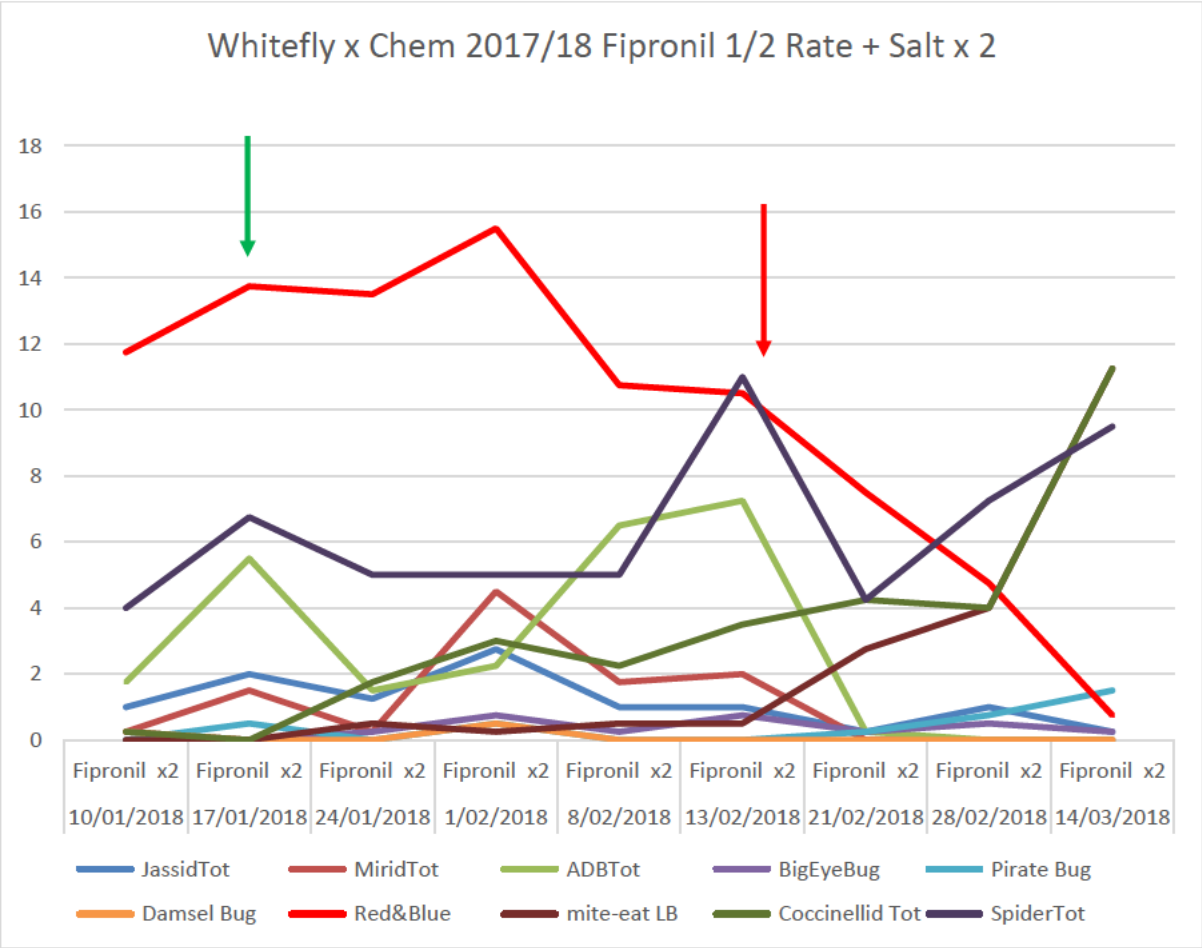


Figure 7: Effects of dimethoate spray (green arrow) and fipronil spray (red arrow) on mirids and beneficials

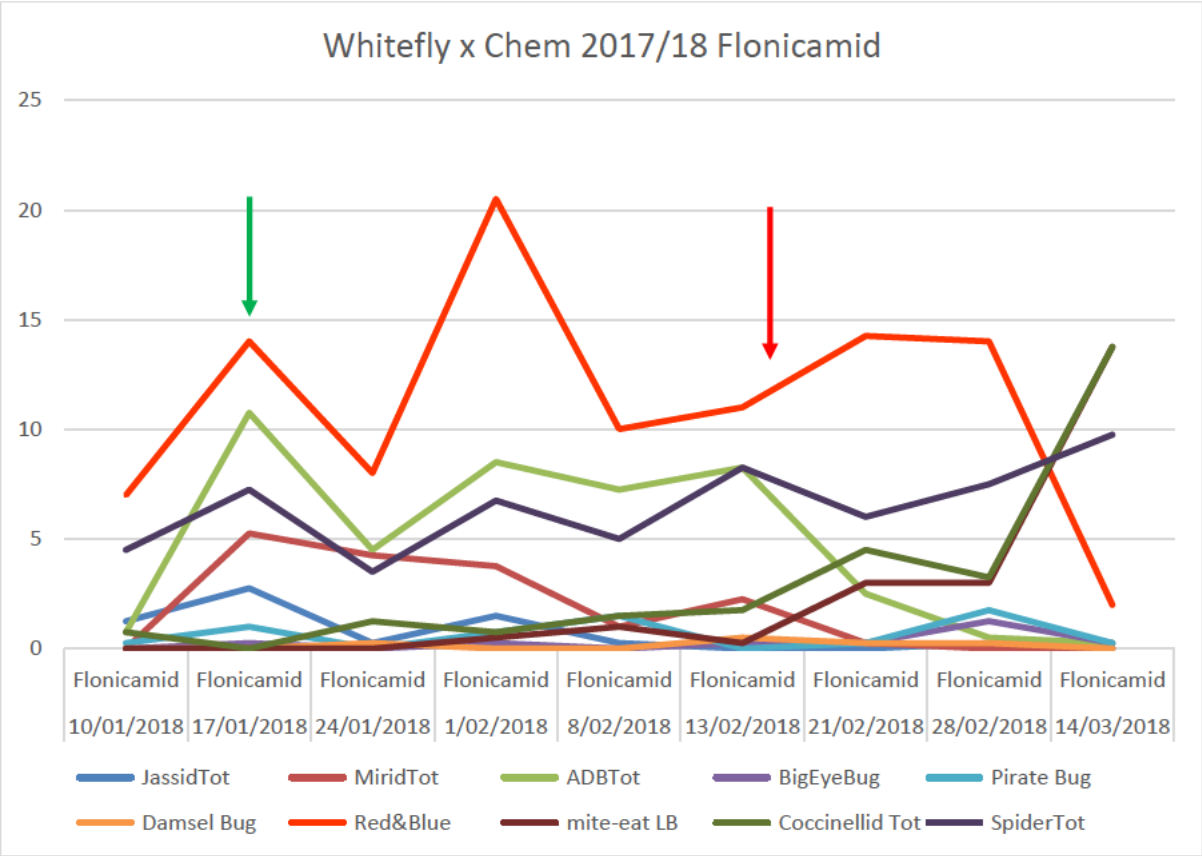


Figure 8: Effects of dimethoate spray (green arrow) and flonicamid spray (red arrow) on mirids and beneficials

APPENDIX 7: Section C (i) Southern Cotton Seed Treatment Experiments

Table 1: Mean abundance of Ants, beneficial Coleoptera and *Helicoverpa* spp. in each seed treatment, Connamara C3, 2016/17

	Ants Total		Pheidole sp.		Ben. Coleoptera Total		Red & Blue Beetle		Lepidoptera Total		*Helicoverpa* Total	
Insecticide	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Control	0.034	-68.93	0.019	-75.04	0.047	68.04	0.019	-17.84	0.073	8.35	0.073	8.35
Cruiser X	0.093	-14.84	0.027	-65.52	0.029	2.82	0.014	-37.14	0.096	41.78	0.096	41.78
Thimet	0.109	0.00	0.077	0.00	0.028	0.00	0.023	0.00	0.068	0.00	0.068	0.00
P	0.014		0.014		0.001 (i)		0.018 (i)		0.003(i)		0.003 (i)	
LSD (p = 0.05)	0.049		0.049		0.070		0.058		0.082		0.082	
df						(2, 42)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.
4. (i) refers to the interaction values

Table 2: Mean abundance of Hemiptera pest species and thrips in each seed treatment, Connamara C3, 2016/17

	Hemiptera Other		Jassid Total		Jassid Immature		Rutherglen Bug		Thrips Total		Thrips Adults	
Insecticide		%[2]	Mean	%	Mean	%	Mean	%	Mean	%	Mean	%
Control	0.132	100.98	2.275	-30.87	0.285	-71.36	0.558	176.73	1.706	67.38	1.630	67.63
Cruiser X	0.074	13.26	2.072	-37.04	0.246	-75.29	0.359	78.29	1.378	35.14	1.336	37.42
Thimet	0.066	0.00	3.291	0.00	0.994	0.00	0.202	0.00	1.019	0.00	0.972	0.00
P	0.019		0.018		<0.001		0.041		0.015(i)		0.015(i)	
LSD (p = 0.05)	0.045		0.240		0.177		0.199		0.543		0.240	
df						(2, 42)						

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide - back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.

NB: New F values for significant interactions of TRT x Date at df = (4, 16):
Ants Total – P =0.38 n.s.
Pheidole – P = 0.373 n.s. Jasssid Immature – P = <0.049*

Table 3: Mean abundance of various beneficial insects in each seed treatment, Dimby Plains, 2016/17

	Ants Total		Pheidole sp.		Green Lacewing Adult		Red & Blue Beetle		*Telenomus* sp.	
Insecticide	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%	Mean	%
Control	0.058	-78.99	0.041	-83.79	0.014	45.96	0.010	1.00	0.005	-83.29
Cruiser X	0.278	0.00	0.252	0.00	0.010	0.00	0.000	0.00	0.029	0.00
P	<0.001		<0.001		0.041 (i)		0.036 (i)		0.033	
LSD (p = 0.05)	0.096		0.094		0.046		0.025		0.021	
df					(1, 27)					

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.
4. (i) refers to the interaction values

Table 4: Mean abundance of various pest insects in each seed treatment, Dimby Plains, 2016/17

	Jassid Total		Jassid Immatu[redacted]		Thrips Immature		Looper	
Insecticide	Mean[1]	%[2]	Mean	%	Mean	%	Mean	%
Control	0.010	1.00	0.097	-77.40	0.186	59.74	0.010	1.00
Cruiser X	0.000	0.00	0.431	0.00	0.116	0.00	0.000	0.00
P	0.011		0.005				0.036(i)	
LSD (p = 0.05)	0.110		0.178				0.025	
df				(1, 27)				

1. Values are means of transformed data from suction samples, i.e. ln (mean number of insects per m per sample +1)
2. Values are percentage change compared to the control treatment using back-transformed means, calculated as; 100x ((back-transformed mean for insecticide – back-transformed mean for control)/back-transformed mean for control)
3. Asterisks in each column indicate treatments significantly different from the control.
4. (i) refers to the interaction values

NB: New F values for significant interactions of TRT x Date at df = (1, 4):
Ants Total – P =0.138 n.s.
Pheidole – P = 0.152 n.s.
Jasssid Immature – P = 0.156 n.s.
Jassid total – P = 0.218
Red & Blue Beetle –P = 0.374 n.s. Telenomus – P = 0.298 n.s.

APPENDIX 8: Section D (i) CBT

Table 1: Potential CBTv hosts grown at ACRI for extracting CBTvA and/or CBTvB after exposure to the viruses.

Plant species	**Number of Samples Processed, Ready for Testing**	**Number Previously Tested**	**Number Previously +ve to CBTvA**	**Number Previously +ve to CBTvB**	**Number Previously +ve to CBTvA & CBTvB**
Cicer arietenum	6	0	no previous test	no previous test	no previous test
Cucumis sativus	5	0	no previous test	no previous test	no previous test
Citrullus lanatus	4	0	no previous test	no previous test	no previous test
Ipomoea lonchophylla	4	0	no previous test	no previous test	no previous test
Albemoschus esculentus	3	0	no previous test	no previous test	no previous test
Ammi majus	**3**	**0**	no previous test	no previous test	no previous test
Conyza bonariensis	3	0	no previous test	no previous test	no previous test
Cucumis melo	3	0	no previous test	no previous test	no previous test
Cyperus rotundus	**3**	**0**	no previous test	no previous test	no previous test
Macroptilium sp.	3	0	no previous test	no previous test	no previous test
Mollugo cerviana	3	0	no previous test	no previous test	no previous test
Portulaca oleracea	3	0	no previous test	no previous test	no previous test
Chenopodium pumilio	2	0	no previous test	no previous test	no previous test
Cucurbita sp.	2	0	no previous test	no previous test	no previous test
Eragrostis sp.	**2**	**0**	no previous test	no previous test	no previous test
Oxalis sp.	**2**	**0**	no previous test	no previous test	no previous test
Stellaria media	**2**	**0**	no previous test	no previous test	no previous test
Alternanthera denticulata	1	0	no previous test	no previous test	no previous test
Echinochloa sp.	**1**	**0**	no previous test	no previous test	no previous test
Rhyncosia minima	1	0	no previous test	no previous test	no previous test
Medicago polymorpha	3	1	1	0	0

Rapistrum rugosum	3	1	0	0	0
Fallopia convolvulus	1	1	0	0	0
Lactuca serriola	**1**	**1**	**0**	**0**	**0**
Amaranthus macrocarpus	3	2	0	0	0
Datura ferox	**2**	**2**	**0**	**0**	**0**
Vicia sativa	**6**	**3**	**0**	**0**	**0**
Lamium amplexicaule	5	3	1	1	1
Xanthium occidentale	1	3	0	0	0
Chamaesyce drummondii	4	4	0	0	0
Physalis ixocarpa	4	4	0	0	0
Vigna radiata	3	4	0	1	0
Anoda cristata	5	7	4	7	4
Helianthus annuus	**2**	**7**	**0**	**0**	**0**
Sonchus oleraceus	**4**	**9**	**0**	**0**	**0**
Solanum nigrum	**3**	**11**	**0**	**0**	**0**
Trianthema portulacastrum	2	12	4	0	0
Abutilon theophrasti	4	13	2	5	2
Hibiscus sabdariffa	4	13	1	2	0
Sida rhombifolia	4	14	0	2	0
Hibiscus trionum	2	15	2	0	0
Malvastrum coromandelianum	2	20	1	1	1
Malva parviflora	**8**	**73**	**15**	**31**	**13**
Gossypium hirsutum	12	246	94	171	87

APPENDIX 8a: Section D (ii) CBT

Table 1. Transmission of CBT by aphids into unprotected plants (nil) or plants protected by Cruiser or Cruiser Extreme seed treatment.

Aphid Treatment	Insecticide treatment	Placement	2012/13 Proportion of plants per pot with CBT (of 4)	2013/14 Proportion of plants per pot with CBT (of 4.6)	Estimates from ASREML analysis both years (proportion of plants per pot)	2012/13 Proportion of pots with an infested plant (of 5 pots)	2013/14 Mean number of pots with an infested plant (of 5-13 pots)	Estimates from ASREML analysis both years (proportion of pots with an infested plant)
No aphids	Nil	-	0	0		0	0	
	Cruiser		0	0		0	0	
	CruiserX		0	0		0	0	
+ CBT aphids	Nil	1 true leaf	0.65	0.42	0.43	1	0.6	0.7
	Cruiser	1 true leaf	0.40	0.71	0.56	0.8	1	0.9
		4 true leaves	0.25	0.47	0.37	0.6	1	0.8
	CruiserX	1 true leaf	0.05	0.33	0.20	0.2	0.6	0.4
		4 true leaves	0.15	0.32	0.23	0.4	0.4	0.4

Table 2. Transmission of CBT by aphids into unprotected plants (nil) or plants protected by an insecticide sprayed onto plants at 1 or 2% concentration.

Aphid Treatment	Insecticide treatment	Timing in relation to aphid infestation	2012/13 Proportion of plants per pot with CBT (of 4)	2013/14 Proportion of plants per pot with CBT (of 4.6)	Estimates from ASREML analysis both years (proportion of plants per pot)	2012/13 Proportion of pots with an infested plant (of 5 pots)	2013/14 Mean number of pots with an infested plant (of 5-13 pots)	Estimates from ASREML analysis both years (proportion of pots with an infested plant)
No aphids	Nil	-	0.00 a	0.00	0.0	0.0	0.0	
+ CBT aphids	Nil	-	0.65	0.42	0.47	1.0	0.6	0.74
	Product 1 (1%)	nil	0.00	0.00	0.00	0.0	0.0	
		24 hr before	0.05	0.02	0.03	0.2	0.1	0.14
		10 min after	0.10	0.08	0.09	0.4	0.3	0.34
		24 hr after	0.55	0.47	0.50	1.0	0.8	0.91
	Product 1 (2%)	nil	0.00	0.00	0.00	0.0	0.0	
		24 hr before	0.15	0.04	0.09	0.4	0.2	0.31
		10 min after	0.26	0.22	0.24	0.6	0.4	0.49
		24 hr after	0.60	0.66	0.64	1.0	0.9	0.92

APPENDIX 9: Section G

Fruit Removal Experiments 2017/18

Figure 1: Distribution of fruit and missing fruiting positions in the canopy prior to harvest: Fruit damage experiment, ACRI 2016/17

Figure 1 cont.: Distribution of fruit and missing fruiting positions in the canopy prior to harvest: Fruit damage experiment, ACRI 2016/17

Figure 2: Distribution of fruit and missing fruiting positions in the canopy prior to harvest: Fruit damage experiment, Spring Ridge 2016/17

Figure 2 cont.: Distribution of fruit and missing fruiting positions in the canopy prior to harvest: Fruit damage experiment, Spring Ridge 2016/17