Horticulture Innovation Australia

Final Report

Investigating management of green stink bugs in raspberry crops

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Summary

The green stink bug *Plautia affinis* (Dallas) (Hemiptera: Pentatomidae) and other closely related pentatomid bugs have become a pest in raspberries at the Costa Berry Exchange in Corindi, Northern NSW. Large numbers of young nymphs were appearing amongst the fruit trays during picking and adult bugs were obviously breeding in significant numbers in the crop.

- **Project objectives**

The general aim of the study was to investigate a low pesticide management strategy for *Plautia affinis* which involved different components.

1. Establishment of a laboratory colony to enable screening of insecticides and rearing of biological control agents
2. Evaluation of insecticides for their efficacy in controlling *P. affinis*
3. Investigations for potential biological control
4. Development of monitoring and management strategies

- **Target audience**

The target audience includes growers, owners, managers and consultants in the *Rubus* industry.

- **Project activities**

Establishment of a laboratory colony

A laboratory colony was an essential part of the project in order to provide insects for laboratory screening and rearing of egg parasitoids.

Evaluation of insecticides

This was done in the laboratory in the first instance (10 chemicals) and 9 selected insecticides were evaluated in a field trial. Pyrethrins (i.e. PyGanic®) and tolfenpyrad gave the best results in the insecticide trials. However, pyrethrins (i.e. PyGanic®) are the more IPM compatible and therefore the most suitable chemicals for management of *Plautia affinis* in raspberries.

Care has to be taken when interpreting the laboratory data as number of insects and replicates were small due to limited availability of insects.

Investigations for potential biological control

Raspberry plantings at Corindi were monitored for *P. affinis* and parasites. Adult bugs, nymphs and eggs were collected and taken back to the insectary at the Wollongbar Primary Industries Institute (WPII). Three fly species, parasites of adults and nymphs and 3 different egg parasitoids were recorded. Two egg parasitoids were identified as *Trissolcus* sp. and *Telenomus* sp. (Hymenoptera: Scelionidae) and one species has not been identified. *Trissolcus* sp. and *Telenomus* sp. were reared in the laboratory and small field releases were made and evaluated. The general background of parasitism in areas without releases however, was 18.6% (2013) 43.87 % (2014) and could not be significantly improved in the second season with small releases.

Monitoring and Management strategy

Treatment thresholds and a management strategy have been developed through monitoring *P. affinis* and egg parasitoids in the field. Part of the management strategy would need to be weekly or fortnightly monitoring of *P. affinis* and egg parasitoids once the plants have started to set berries.

P. affinis adults were observed to shelter in the dry leaves of the raspberry plants during winter and it would be important to culturally manage this population.

Pyrethrins (i.e PyGanic®) are the preferred chemical option. A maximum of 2 applications per crop cycle should be used when indicated by monitoring to reduce the peak of the bug population.

- **Key outcomes (results, consequences or impacts)**

1. A monitoring management strategy for raspberries that includes biological control and minimal insecticide input has been developed.
2. Preservation of natural enemies through minimal reliance on pesticides
3. Better understanding of the biology and ecology of green stink bugs and its natural enemies.
4. Maintaining market access into the future by minimising the risk of potential pesticide residues in raspberries

- **Recommendations: Future R&D and practical application to industry**

1. Use of regular monitoring and recommended thresholds to commence chemical treatments
2. Preservation of natural enemies on site by minimising pesticide input
3. Implementing cultural control of bug population in dried foliage
4. Registration for pyrethrins (i.e. PyGanic®)
5. Investigations into fully integrated pest and disease management

Keywords

Raspberries; *Plautia affinis*; Integrated Pest Management; biological control, egg parasitoids, *Trissolcus* sp.; *Telenomus* sp.; cultural control; chemical control; insecticides

Introduction

The tunnel house techniques for growing temperate crops of raspberries in a sub-tropical environment have been pioneered at Costa Berry Exchange Corindi in Northern NSW. To this point the pest pressure on the crop has been minimal. The green stink bug, *Plautia affinis* (Dallas) (Hemiptera: Pentatomidae) and other closely related pentatomid bugs appear to have developed into serious contaminant pests that can also cause significant the crop damage.

Large numbers of young nymphs appear amongst the fruit trays during picking and adult bugs are obviously breeding in significant numbers on the crop. Given there is a very short shelf life for the crop unless it is frozen there is a need to reduce the *P. affinis* population at harvest.

Coombs and Khan (1998) reported on *P. affinis* causing damage in raspberries at Caboolture, south-eastern Queensland. They conducted a study monitoring populations of the *P. affinis* and their natural enemies (Coombs and Khan, 1998).

The problem has also been recorded in the US (Anonymous, 2013), and if a biological solution could be found it would be the most desirable outcome.

Our aim was to develop a more sustainable and long term management system that was practical and easily applied by growers.

Aspects considered for investigation were:

1. Establishment of laboratory colonies of *P. affinis* and biological control agents
2. Chemical control (investigation of IPM compatible insecticides)
3. Biological control (release and evaluation)
4. Cultural control (investigation of management of bug population in dried foliage)
5. Development of monitoring and management strategies – Best Practice

Chemical control

Previously there has been no permit or registration of chemicals for management of stink bugs for raspberries and options that are compatible with biological control and suitable for an Integrated Pest Management (IPM) approach needed to be investigated. As part of this, it was important to also consider the need for presence of bees in the planting during a prolonged period of flowering.

Biological control

There has been information on biological control from previous studies. Coombs and Khan (1998) reported on parasitism by *Trissolcus basalis* (Wollaston), *Trissolcus oenone* Dodd and *Telenomus* sp. (Hymenoptera: Scelionidae).

Egg parasitoids like *Trissolcus basalis* and the Argentinian tachinid fly *Trichopoda giacomellii* Blanchard (Diptera: Tachinidae), which have been introduced already to control the closely related green vegetable bug (GVB) *Nezara vridula* (L.) (Hemiptera: Pentatomidae) in many crops in Australia (Waterhouse and Norris, 1987; Clarke, 1990; Sands and Coombs, 1999; Coombs and Sands, 2000; Coombs, 2003) were evaluated.

An investigation of natural enemies from the field and their assessment in the laboratory and field were also considered to be an important aspect of the research.

Cultural control

It was important to explore if there was potential for any cultural management options that would fit into an IPM strategy. Coombs and Khan (1998) had anecdotal evidence that *P. affinis* overwinters in the leaf litter.

Monitoring and management strategy - Best Practice

A monitoring strategy and development of treatment thresholds would be an important part of an IPM approach. A practical, quick and easy way to assess population density frequently was required in order to make it practical for commercial plantings. It was also important to correlate the population with potential to damage and from that develop the treatment threshold.

Methodology

1. Laboratory rearing of bugs

Plautia affinis

A colony of *P. affiinis* was established in one of the insectaries at Wollongbar Primary Industries Institute (WPII). The temperature in the laboratory room was set at 25°C (±2).

Nymphs and adults were reared successfully and the colony expanded well. Nymphs and adults were kept separate in plastic containers (4 litre volume) with a lid. To allow air into the container a circle was cut out of the centre of the lid and covered with gauze.

P. affinis nymphs and adults were fed fresh French beans, pieces of corn cobs and tobacco weed berries (the latter depending on availability). Bugs were also provided with a moist piece of sponge in a Petri dish. Containers were cleaned out and food changed once a week. Newly emerged adults were removed from the nymph container and added to the adult container. Dead adults and eggs were collected from the adult container. A proportion of the collected eggs were given to the egg parasitoids and the rest of the eggs were used to maintain the bug colony.

McDonald (1971) described the life cycle of *P. affinis* at 26°C (±2) (room temperature). The duration from egg to adults took an average of 44.4 (±10.3) days.

2. Biological control

2.1. Laboratory rearing of egg parasitoids

In October 2012 stink bug eggs that were found on fruit were collected and brought back to WPII. Eggs were monitored and parasitism observed. Two egg parasitoids were found and identified as *Trissolcus* sp. and *Telenomus* sp. Specimens of both egg parasitoids were sent to the Agricultural Scientific Collections Unit in Orange and for DNA barcoding to the Molecular Biology Unit at Wagga Wagga Agricultural Institute to confirm identification.

Small colonies of the two egg parasitoids *Trissolcus* sp. and *Telenomus* sp. were maintained in one of the insectaries at WPII at Wollongbar. The temperature in the laboratory room was set at 25°C.

Both wasp species were kept in separate glass jars (2 litre volume) with a ventilated screw top plastic lid. *Trissolcus* sp. and *Telenomus* sp. were given dental cotton wicks soaked in 5% sugar solution once a week. They were also presented with fresh eggs from the *P. affinis.* The eggs were placed on the sticky surface strip of Post-it® notes and placed in the jars with the egg parasitoids.

While parasitoids are alive and active they were fed with fresh *P. affinis* eggs (about 20 eggs per jar, once a week).

Dead bodies of *P. affinis* were kept and checked for parasitism. We found 3 different fly species parasitising *P. affinis*, which still need to be identified.

2.2. Field releases

Trissolcus release 2013

Trissolcus sp. was released in March 2013 in raspberry tunnels at Berry Exchange at Corindi. One tunnel was selected for each egg parasitoid species. The number of *P. affinis* eggs available from the laboratory colony was limited and we therefore concentrated on rearing larger numbers of *Trissolcus* sp. first, as *Trissolcus basalis* (Woll.) (Hymenoptera: Scelionidae) had been successfully released for biological control of *Nezara viridula*. We wanted to evaluate *Trissolcus* sp. first before rearing larger numbers of *Telenomus* sp. for releases.

Population pressure of *P. affinis* (nymphs and adults) was sampled before release. A total of 150 *Trissolcus* wasps were released in the raspberries at Berry Exchange at Corindi on three release dates, 29/10/2013, 22/11/2013 and 19/12/2013.

Containers with sentinel cards with fresh *P. affinis* eggs were put out after releases to see if *Trissolcus* sp. could be recaptured. Ten raspberry bushes next to the release points (5 bushes to the left and 5 bushes to the right) were monitored for presence of *P. affinis* adults, nymphs and eggs and their parasitism. To get a baseline of presence of bugs and egg parasitoids, the same amount of raspberry bushes were monitored in 3 tunnel rows where no releases of *Trissolcus* sp. had been made.

Telenomus release 2014

Telenomus sp. was released at Corindi on 20 May 2014 (153 parasitised eggs) and collected on 29 May 2014. The release was made alternately in the northern and southern end of two rows in two adjacent tunnels.

Telenomus release 2015

161 *Plautia affinis* eggs parasitised by *Telenomus* sp. were released again in block S1 at Costa Berry Exchange on 14 January 2015.

Releases of parasitised eggs were made in the middle row of each of 4 tunnels, alternating at the northern or southern end.

The trial was monitored for development of parasite population by putting out sentinel eggs of *P. affinis* in the trial area. After a week, the containers were collected and brought back to the laboratory and were checked regularly for parasitism.

On the 25 February 2015 Row 13 next to the releases was also monitored for background parasitism.

Refer to Appendix 1 for further details.

3. Chemical control

3.1. Laboratory screening of insecticides

Selected insecticides were initially screened in the laboratory using the drop test techniques first and then the treated surface technique as described below. We included chemicals that we knew would fit into an IPM system, would be effective on bugs (i.e. Admiral®, Lepidex, PyGanic® etc.) and also newer chemistry that we wanted to test for their effectiveness on the bugs (i.e. DC142, MCW9540, flonicamid, tolfenpyrad etc.) to look at their potential role in management of *P. affinis*.

Drop test

Insecticides were tested at recommended rates.

One microlitre of the test solution was applied on individual bugs from the colony at WPII. Mortality was checked after 1, 2, 3 and 7 days. Six different chemical were tested and water was used as a control. Each treatment was replicated twice and 4 adults were used in replicate 1 and 3 adults in replicate 2.

Numbers of insects available for laboratory trials were a limiting factor and care has to be taken when interpreting the results.

Treated surface test

A number of chemicals were screened against adult *P. affinis* using the treated surface test. Five chemicals were compared and water was used as a control treatment. Five adult bugs were placed in a 750 ml square plastic food container with a piece of corn cob which was dipped into the insecticide solution. The mortality of *P. affinis* adults was recorded after 1, 2 and 3 days. Each treatment was repeated twice.

3.2. Insecticide field trial

A field trial was set up in March 2015. The middle row under a tunnel was used as a treatment row. Within each row, 9 treatment blocks were assigned. The trial was monitored for presence of dead and alive *P. affinis* adults and nymphs after 1 day and 7 days. Eight chemicals were tested and water was used as a control treatment. Each treatment covered 3 or 4 plants in a row and was repeated 9 times. One treatment row had 1 replicate of each treatment and there were 3 buffer rows between each treatment row.

4. Monitoring and Management strategy - Best Practice

A practical way of monitoring commercial plantings was required. Discussions were held with Costa Berry Exchange and as a consequence, thresholds and a monitoring protocol were developed.

5. Technology Transfer

During the course of the project we worked closely with Costa Berry Exchange with regards to the project, its progress and discussions of management options. Costa Berry Exchange contributed to the research funding with a voluntary contribution. The general target audience includes growers, owners, managers and consultants in the *Rubus* industry.

We published an update in the industry journal and produced a fact sheet to be put on the industry website for grower access.

Unless otherwise stated, all measures of variation reported are standard errors.

Outputs

1. **ID of pest species confirmed**

We found a complex of pentatomid bugs in the raspberry planting, however, the majority was *Plautia affinis* Dallas and smaller populations of the green vegetable bug *Nezara viridula* (Linnaeus) and the green potato bug *Cuspiconia simplex* Walker (Hemiptera: Pentatomidae).

2. **Laboratory colony of *Plautia affinis* established**

At the beginning of the project a small laboratory colony of *P. affinis* was established that enabled us to supply eggs for rearing of egg parasitoids and insects for insecticide screening. The colony was maintained throughout the project.

3. **Collection of biological control agents**

We collected 3 different egg parasitoids in the field over a period of 10 months. One was identified as *Trissolcus* sp., one as *Telenomus* sp. and one unidentified species. We also found 3 fly species; one unidentified tachinid fly species, a *Trichopoda* sp. and a phorid fly species, which still need to be identified. No identification has been provided yet for *Trissolcus* sp. an unidentified species. The genetic investigation of the *Telenomus* sp. showed 95% match with *Telenomus turesnsis* and 94% with *Telenomus cloropus* but no absolute matches. It is therefore possibly a new species (D. Goporenko pers. com., Jan. 2016).

4. **Laboratory colonies of biological control agents**

As availability of *P. affinis* was limited, we had to prioritise the use of the insects available from the colony. The two dominant egg parasitoids species were therefore selected for rearing and small laboratory colonies were established and maintained over time. The *Trissolcus* sp. allowed us to

conduct 3 field trial releases, and a small colony of *Telenomus* sp. that allowed us to make 3 small field releases.

5. Efficacy of selected insecticides tested in laboratory

Eleven different insecticides have been screened in the laboratory and a field trial was conducted with selection of 9 chemicals. From the field trial pyrethrins (i.e. PyGanic®) and tolfenpyrad gave the best results. With regards to IPM compatibility and compatibility and presence of bees in the planting, pyrethrins (i.e. PyGanic®) are considered the more suitable chemicals. The industry now has a permit available for the use of PyGanic® (PER80070).

6. Efficacy of selected biological control agents tested in the laboratory

Trissolcus sp. and *Telenomus* sp. were collected in the field and tested in the laboratory. Both species gave parasitism in the laboratory of over 60%.

7. Selected biological control agents evaluated in field trials

Trissolcus sp. and *Telenomus* sp. were also both tested in small field releases with variable results. When general background parasitism was high, small releases of *Telenomus* sp. did not increase parasitism rates. The total parasitism was about 45%. The two species accounted for approximately 15% of parasitism present each. A third species (potentially hyperparasite) also accounted for further 15%.

8. Recommendation for pest management strategy (Best Practice) for industry at the end of the project.

In consultation with industry representatives a management strategy has been developed (details see under Outcomes point 3.1. - pp. 12-13 and Appendix 2, pp. 37-38), including a monitoring strategy, treatment thresholds, IPM compatible insecticides (pyrethrins), boosting natural enemy populations with small releases and cultural control (removing *P. affinis* in dried leaves after harvest).

9. Industry Publications

An article titled *'Investigating management of green stink bugs in raspberry crops',* giving a project update, was submitted to the industry newsletter on 9 May 2014.

A fact sheet on *P. affinis* management can be found on the industry (RABA) website.

Outcomes

1. Better understanding of the biology and ecology of *Plautia affinis* and its natural enemies.

1.1. Laboratory rearing of bugs

The detailed life-cycle of *Plautia affinis* has been described in earlier studies by McDonald (1971) (Table 1). We successfully maintained a small colony of *P. affinis* at WPII on commercially available French beans and corn. We also provided the bugs with wild tobacco, *Solanum mauritianum* Scopoli (Solanaceae) when available. Wild tobacco tended to increase their oviposition.

The lifespan of *P. affinis* adults was generally short (2-3 weeks). The colony however was sufficient to provide insects for insecticide screening and rearing of egg parasitoids at different times.

1.2. Biological control

1.2.1. Laboratory rearing

Trissolcus sp.

We maintained a small colony of *Trissolculs* sp. between 2012 and 2013, which enabled us to undertake 3 small field releases. At a room temperature of 25°C the development time for *Trissolcus* sp. took an average of 23.5 days. The average parasitism was 69.4% and the percentage of *Trissolcus* sp. emerging from eggs was 35.0%.

Telenomus sp.

Between 2013 and 2015 we also maintained a small laboratory colony of *Telenomus* sp. which supplied insects for 3 small field release trials.

At a room temperature of 25°C the development time for *Telenomus* sp. had a mean duration of 23.1 days. The average parasitism was 61.7%. Live egg parasitoids emerged from 30.6% of the parasitised eggs.

The rearing process itself for both egg parasitoids was easy but the number of *P. affinis* eggs available as a food source was the limiting factor for the laboratory colonies.

Dead bodies of *P. affinis* were checked for parasitism in the laboratory. Phorid flies appeared to be the dominant parasite. Of dead adults collected, parasitism by phorids varied between 20-80%. Parasitism by the other fly species was very sporadic.

1.2.2. Field releases

Trissolcus release 2013

In the release area, only 9 eggs, (5.8%) were parasitised by *Trissolcus* sp. and 8.4% were parasitised by *Telenomus* sp. *Telenomus* sp. naturally occurs in the raspberry plantings and to this point had not been released.

Monitoring the natural presence of the two egg parasitoids found *Trissolcus* sp. had only 0.4% parasitism and *Telenomus* sp. 18.2% parasitism. This lead to the conclusion that *Telenomus* sp. appears to be the stronger candidate for biocontrol of *P. affinis* in the raspberry tunnels at Corindi.

Telenomus releases 2014 and 2015

In May 2014, the average rate of *Telenomus* hatching from parasitised eggs was 86.3% of the eggs released (hatching rate 77.1 – 100.0%). Parasitism and presence of *P. affinis* was monitored in control and release areas.

There was no difference between control and release treatments in any of the different categories we looked at; no difference in parasitism and no difference in the presence of *P. affinis*. With the small release of *Telenomus* sp. however we managed to increase parasitism by 43.8%.

Parasitism observed was exclusively done by *Telenomus* sp. and it seems to be quite persistent in the field. This suggested that it would be the more effective egg parasitoid but doesn't respond to rearing and releasing.

In January 2015, the row next to the release site we collected 18 egg masses, of which 6 were parasitised. Two egg masses were parasitised by *Trissolcus* sp., 1 egg mass by *Telenomus* sp. and 3 egg masses by an unidentified egg parasitoid.

P. affinis populations were generally low during the past season and the general background parasitism was high (43.9%). Small scale releases of *Telenomus* sp. therefore did not increase parasitism or have a significant impact on the pest population.

2. **Minimal pesticide use encourages biological systems**

2.1. Laboratory screening of insecticides

Drop test

Results were inconclusive due to high mortality (50%) in the control treatment after 3 days. However, results suggest that the only chemicals resulting in 100% mortality of *P. affinis* adults at day 7 were methomyl and Sivanto. Trichlorfon and Exirel gave 92% mortality at day 7.

The treated surface test was considered a better screening technique.

Treated surface test

We tested 11 new and old insecticides. The new insecticides Sivanto™, Exirel® but also Lepidex gave promising results in initial tests. Lannate® and tolfenpyrad gave the best results overall. Due to their detrimental effect on bees while they are active, these chemicals are not a suitable option that would fit into an IPM system.

As numbers of insects available for trials were limited, results have to be interpreted with caution.

2.2. Field insecticide trial

In the field trial the PyGanic® treatment had significantly more dead *P. affinis* than the three new products Admiral, DC142 and DC099. PyGanic® and tolfenpyrad gave the best overall results in the insecticide trials and also the field trial. As mentioned above, due to issues with bee compatibility pyrethrins (i.e. PyGanic®) are the more appropriate candidates for management of *P. affinis*.

3. Integrated pest management strategy for raspberries

3.1. Monitoring and Management strategy – Best Practice

During this study, we noticed that *P. affinis* adults shelter during winter (Coombs & Khan, 1998) in the dry leaves of the raspberry plants. It would be important to culturally manage this population.

Part of the management strategy would need to be weekly or fortnightly monitoring of *P. affinis* and egg parasitoids once the plants have started to set berries.

It is important to monitor when *P. affinis* starts to appear in different blocks during spring and then use a knock down spray after *P. affinis* adults are detected. Afterwards (possibly 7 days) releases of parasitoids should commence to clean up residual bug eggs.

Monitoring can be done by walking the planting visually checking and counting *P. affinis* nymphs, adults and egg masses. Egg batches need to be collected and checked for parasitism.

Pest pressure was estimated and actions thresholds developed as described in Table 1.

It will be important to get egg parasitoid populations to build up early and release them in tunnels as soon as the pest is observed in the planting.

PyGanic® is the preferred chemical option, as its residual time is very short and it can easily be used when bees are around if applications are done late in the afternoon or at night when bees are in the hive. A maximum of 2 applications per crop cycle should be used when indicated by monitoring to reduce the peak of the bug population.

However if pest pressure at the end of the season is high and considered necessary, and taking appropriate precautions regarding bees Lannate® could be used as a clean-up option only after harvest is completed (residues must be avoided) to prevent carry over to the next crop.

4. **Maintain future market access by reducing potential pesticide residues in raspberries**

By taking the approach of minimum insecticide input (a maximum of 2 applications per crop cycle) and choosing natural pyrethrum (pyrethrins) as a preferred control option pesticide residues can be significantly reduced. Pyrethrins have a low toxicity and do not persist for long in the environment. The active chemical pyrethrin, is degraded by high temperature and UV light (from 100% to less than 1% within 5 hours) (Gunasekara, 2005).

Table 1: Evaluation of monitoring population of *P. affinis* in raspberries

Pest pressure	**Observation**	**Action**
Low	<0.5 adult/plant (1adult/10 plants) 0.2-0.3 egg masses per plant	Keep monitoring or get ready for wasp release
Medium	0.6-2.0 adult/plant <0.1 egg masses/plant 1.5-3.0 nymphs/plant	Get ready for chemical treatment or release wasps asap
High	>2.0 adults/plant >2.0 egg masses/plant >10.0 nymphs/plant	Apply chemical treatment and release wasps after withholding period

Evaluation and Discussion

1. Laboratory rearing of bugs

Adults of *Plautia affinis* only lived for 2 to 3 weeks and the colony had to be restocked from field collections regularly, which was a major drawback. The colony would have been much more effective if it had a regular supply of field stock closer to the laboratory.

It was not possible to maintain a reasonable sized colony of *P. affinis*, *Trissolcus* sp. and *Telenomus* sp. and sacrifice *P. affinis* for insecticide screening at the same time. It was therefore decided to concentrate on biological control first and postpone the insecticide screening in the first instance. This delayed the project and 2 variations for time extension were required.

2. Biological control

2.1 Laboratory rearing

The actual process of rearing *Trissolcus* sp. and *Telenomus* sp. in the laboratory was fairly simple. The problem was keeping up the supply of *P. affinis* eggs to maintain and build up the colonies of either of the two selected egg parasitoids. We had to set priorities and decided to start with rearing

Trissolcus sp. first and then evaluate the parasitism in the field before rearing *Telenomus* sp. For a commercial insectary this would be a major impediment. An alternative host (i.e. *Nezara viridula*) or an artificial medium to rear the egg parasitoids could be an option which could be explored.

The economics of mass-rearing the egg parasitoids for a commercial insectary and also the economics of releases of a commercial product would need to be investigated. However, the potential for commercial rearing of an egg parasitoid for *P. affinis* as well as *Nezara viridula* was discussed with Dan Papacek (Bugs for Bugs). There was definitely interest in looking at the feasibility for commercial rearing.

Realistically it would be easy for individual growers to take on rearing of *P. affinis* and egg parasitoids on a small scale on site and release them as needed to boost the natural population of egg parasitoids in the field. Costa Berry Exchange is planning to take on small scale rearing of *Telenomus* sp. for small releases in the future to increase field populations in the first instance.

A rearing methodology for *Plautia affinis* and egg parasitoids used in this study is described in Appendix 1 (points 1. and 2.). A rearing methodology for *P. affinis* is also described in Coombs and Khan, 1998.

2.2 Field releases

Given that we managed to increase parasitism with releases of only small numbers of egg parasitoids in one instance, it should be possible to further increase parasitism with inoculative commercial size releases.

We also noticed a third species of egg parasitoids present, which will be identified if possible and which will tell us whether it is a primary parasite or a hyperparasite. If the latter is the case, this would be impacting on the effectiveness of *Telenomus* sp.

3. Chemical control

3.1 Laboratory screening of insecticides

The problem was finding a product that was effective in controlling the pest that had a very short withholding period due to a daily picking schedule. It also needed to have a limited impact on bees and other beneficials.

We screened 11 different chemicals and 4 of them at different rates for their efficacy. Five of these products gave about 80-100% mortality at day 3 and were selected for the field trial.

3.2 Field insecticide trial

In consultation with Costa Berry Exchange we selected 8 different chemicals (5 from the screening, 2 new products and Success™ Neo due to its IPM compatibility) and tested them in a field trial. The assessment was difficult due to the general low pest pressure of bugs.

PyGanic® and tolfenpyrad had the best knock-down effect and also gave the best results after 1

week. Pyrethrins (i.e. PyGanic®) are the preferred chemical option. A maximum of 2 applications per crop cycle should be used when indicated by monitoring to reduce the peak of the bug population. Only if necessary and if appropriate precautions regarding bees are taken Lannate® could be used as a clean-up option only after harvest is completed to prevent carry over to the next crop.

Costa Berry Exchange is already following the chemical recommendations and a permit for PyGanic® has been issued to the industry.

4. Monitoring and Management strategy – Best Practice

It was important to develop a monitoring and management strategy that was practical and easily adoptable by growers. An emphasis on biological control and a minimal of pesticide input were key requirements for the management strategy (under Outcomes point 3.1. – pp. 12-13 and Appendix 2, pp. 37-38).

Monitoring has to be a crucial part of the management strategy and resources need to be allocated for this task. The monitoring could be done by the grower, farm staff or professional pest consultants. A weekly or fortnightly check of blocks with fruit will be sufficient. We are able to provide a general management strategy that can be adapted to meet individual grower's requirements.

The monitoring protocol is a fairly simple one – walking along the rows of the planting and recording numbers of different life-stages of *P. affinis* and collecting egg batches found. The eggs need to be kept and monitored in order to assess parasitism. The latter is important in order to decide whether biological control agents need to be released.

Finding suitable chemical solutions was challenging. Pyrethrins were preferred due to their short activity time and therefore short withholding period. The impact on bees however still needs to be considered. Spraying needs to be carried out when bees are not foraging.

With regards to cultural practices, the removal of dried dead foliage when harbouring the pest is important. Particularly after harvest in late autumn/ winter in which overwintering bugs are hiding is crucial. Plants need to be removed within 2-3 weeks after harvest, when the cropping cycle for the plant is completed (Haro pers. comm., 2015).

The monitoring and management strategy was developed in collaboration with Costa Berry Exchange to ensure that this is a practical way of monitoring and managing the pest on a commercial farm scale. This has formalised a management strategy that Costa Berry Exchange is planning to follow and has also been published for the broader industry to adopt.

In Summary

We can demonstrate that the project generally achieved our project goals and new pest management practices have been introduced by Costa Berry Exchange representing a major part of the industry.

The project aims and evaluation of the project against project outcomes are as follows:

1. *Establishment of laboratory colony and Better understanding of the biology and ecology of Plautia affinis and its natural enemies*

We successfully established and maintained a laboratory colony, which provided enough insects for insecticide screening and maintaining of colonies of 2 egg parasitoids. The short longevity of adults in the laboratory limited the size of the colony; reasons for the short longevity need to be further investigated. We have achieved a better understanding of *P. affinis* and have been able to collect some important biological data.

Three parasitic flies and 3 egg parasitoids were found. We were able to maintain laboratory colonies of 2 of the egg parasitoids and make 3 small field releases with each of them and evaluate the egg parasitoids on the basis the small scale releases. We collected basic biological data and ecological data of the natural enemies. With regards to the egg parasitoids, again the *P. affinis* colony was the limiting factor.

2. *Minimal pesticide use encourages biological systems*

This project target has been successfully met providing a suitable chemical control option that would be compatible with biological control and bees and a short withholding period. We established that a natural pyrethrin (i.e. PyGanic®), is suitable for an IPM system. Numbers of insects available for screening were low and the field population at the time of the field experiment were a limiting factor. However, results were statistically valid.

The plantings had been monitored for natural enemies and 3 egg parasitoid species and 3 parasitic fly species were recorded.

3. *Integrated pest management strategy for raspberries*

In collaboration with Costa Exchange we have developed a monitoring protocol and action thresholds (Table 1) as well as a management strategy (under Outcomes point 3.1. – pp. 12-13 and Appendix 2, pp. 37-38) that can be adopted on commercial farms. The monitoring protocol and management strategy developed are now being adopted by Costa Berry Exchange. A permit for a suitable insecticide (PyGanic®) has been issued and onsite rearing of *Telenomus* sp. is planned. Plants will be destroyed and removed within 3 weeks after harvest and production of the block is completed. Costa Berry Exchange covers the major grower in Australia, but it now has to be promoted to the wider industry.

4. *Maintain future market access by reducing potential pesticide residues in raspberries*

The project developed a management strategy with minimal chemical input with minimal risk for residues, integrates biological and cultural control. It therefore achieved the initial aim of the project of minimising chemical residues in order to maintain market access. Again, this strategy while being adopted by the largest grower, needs to further promoted to the wider industry.

Recommendations

1. Implementation of regular monitoring and using recommended thresholds (Table 1) to commence chemical treatments
2. It is very important to remove dried dead leaves in which bugs are hiding, particularly in late autumn/ winter in when they are overwintering. All plants need to be removed within 2-3 weeks after harvest and production of block is completed.
3. Preserve natural enemies on site by minimising pesticide input
3. Identify the third species of egg parasitoids present and establish whether it is a primary parasite or hyperparasite.
4. Registration for pyrethrins (i.e. PyGanic®)

   PyGanic® proved to be the more appropriate candidate. It gave good results in the insecticide trials and is the most suitable option with regards to bee compatibility and given that bees are present most of the time. We would encourage industry to discuss registration with chemical companies in the long-term.
5. Investigations into fully integrated pest and disease management

Scientific Refereed Publications

None to report

Intellectual Property/Commercialisation

No commercial IP generated

References

Anonymous (2013) Berries Northwest, Management detail stinkbug, in raspberries.

URL: http://www.berriesnw.com/DisordersDetail.asp?id=64

Clarke, A.R. (1990) The control of *Nezara viridula* L. with introduced egg parasitoids in Australia - a review of a 'Landmark' example of classical biological control. *Australian Journal of Agricultural Research*, **41**:6, 1127-1146.

Coombs, M. (2003) Post-release evaluation of *Trichopoda giacomellii* (Diptera: Tachinidae) for efficacy and non-target effects. In: *Proceeding of the 1st International Symposium on Bioloigical Control of Arthropods* – 14-18 January 2002,Honolulu, Hawaii, USA, USDA Forest Service FHTET-03-05. June 2003.

Coombs, M. and Khan, S.A. (1998) Population levels and natural enemies of *Plautia affinis* Dallas (Hemiptera: Pentatomidae) on raspberry, *Rubus idaeus* L., in south-eastern Queensland. *Australian Journal of Entomology*, **37**: 2, 125-129.

Coombs, M. and Sands, D.P.A. (2000) Establishment in Australia of *Trichopoda giacomellii* (Blanchard) (Diptera: Tachinidae), a biological control agent for *Nezara viridula* (L.) (Hemiptera: Pentatomidae)-Part 1. *Australian Journal of Entomology*, **39**, 219-222.

Gunasekara, A.S. (2005) Environmental Fate of Pyrethrins. Environmental Monitoring Branch Department of Pesticide Regulation, 1001 I Street Sacramento, CA 95812. 22pp.

URL: http://cdpr.ca.gov/docs/emon/pubs/fatememo/pyrethrin_efate2.pdf

McDonald, F.J.D. (1971) Life cycle of the green stink bug *Plautia affinis* Dallas (Hemiptera: Pentatomidae). *Journal of the Australian Entomological Society*, **10**, 271-275.

Sands, D.P.A. and Coombs, M.T. (1999) Evaluation of the Argentinian parasitoid, *Trichopoda giacomellii* (Diptera: Tachinidae), for biological control of *Nezara viridula* (Hemiptera: Pentatomidae) in Australia**.** *Biological Control*, **15**: 1, 19-24.

Waterhouse, D.F. and Norris, K.R. (1987) *Nezara viridula* (Linneaeus) *In*: Biological Control, Pacific Prospects. Inkata Press, Melbourne, 81-89.

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Appendices

1. Appendix 1-Detailed Methodology
2. Appendix 2- Detailed Results

Appendix 1: Detailed Methodology

1. Laboratory rearing of bugs

Plautia affinis

A colony of *P. affiinis* was established in one of the insectaries at Wollongbar Primary Industries Institute (WPII). The temperature in the laboratory room was set at 25°C.

Nymphs and adults have been reared successfully and the colony is expanding. Nymphs and adults were kept separate in plastic containers (4 litre volume) with a lid. To allow air into the container a circle was cut out of the centre of the lid and covered with gauze. A commercially available insect rearing cage with gauze cover could have also been used.

P. affinis nymphs and adults were fed fresh beans, pieces of fresh corn cobs and tobacco weed berries (the latter depending on availability). A piece of cleaning sponge (approximately 35x45mm) soaked in water and put in a Petri dish was used to provide a source of moisture. Containers were cleaned out and food changed once a week.

Newly emerged adults were removed from the nymph container and added to the adult container. Dead adults and eggs were collected from the adult container. A proportion of the collected eggs were given to the egg parasitoids and the rest of the eggs were used to maintain the bug colony.

McDonald (1971) described the life cycle of *P. affinis* at 26°C room temperature as listed in Table 2. The duration from egg to adults took an average of 44.4 days (± 10.3 (no measure stated)).

Coombs and Khan (1998) found in their laboratory studies that females had a mean longevity of 74.2 days (±SE = 4.3), a mean fecundity of 423 (± 30.3) eggs per female, and a mean pre-oviposition period of 7.9 days (± 0.5). Adult males had a mean longevity of 44.8 days (± 6.4) (Coombs and Khan, 1998).

Table 2: Life cycle table for *P. affinis* after McDonald (1971)

Life stage	Duration	Variation (Measure not stated in reference)
Egg	6.2 days	± 1.5
1st instar nymph	5.0 days	± 0.6
2nd instar nymph	5.5 days	± 1.6
3rd instar nymph	8.2 days	± 2.1
4th instar nymph	9.1 days	± 1.0

2. Biological control

2.1. Laboratory rearing of egg parasitoids

In October 2012 stink bug eggs that were found on fruit were collected and brought back to WPII. Eggs were monitored and parasitism observed. Two egg parasitoids were found and identified as *Trissolcus* sp. and *Telenomus* sp. Specimens of both egg parasitoids have been sent to the Agricultural Scientific Collections Unit in Orange and for DNA barcoding to the Molecular Biology Unit at Wagga Wagga Agricultural Institute to confirm identification.

Small colonies of the two egg parasitoids *Trissolcus* sp. (Figure 1) and *Telenomus* sp. (Figure 3) were maintained in one of the insectaries at WPII at Wollongbar. The temperature in the laboratory room was set at 25°C.

Both wasp species were kept in separate in glass jars (2 litre volume) with a ventilated screw top plastic lid. *Trissolcus* sp. and *Telenomus* sp. were given dental cotton wicks soaked in 5% sugar solution once a week. They were also presented with fresh eggs from the *P. affinis.* The eggs were placed on the sticky surface strip of Post-it® notes and placed in the jars with the egg parasitoids. Eggs parasitised by *Trissolcus* sp. are shown in Figure 2 and eggs parasitised by *Telenomus* are shown in Figure 4.

We also found 3 fly species parasitising *P. affinis* (Figure 5). The fly species still needs to be identified.

2.2. Field releases

Trissolcus release 2013

Trissolcus sp. was released in March 2013 in raspberry tunnels at Berry Exchange at Corindi. One tunnel was selected for each egg parasitoid species. The number of *P. affinis* eggs available from the laboratory colony is limited and therefore we concentrated on rearing larger numbers of *Trissolcus* sp. first, before rearing larger numbers of *Telenomus* sp. for releases.

Population pressure of *P. affinis* (nymphs and adults) was sampled before release. A total of 150

Trissolcus wasps (50 wasps at end of 3 selected tunnel rows) were released in the raspberries at Berry Exchange at Corindi on 29/10/2013, 22/11/2013 and 19/12/2013 (Figure 6). Parasitised eggs (35-40 were placed on the sticky surface of Post-it® notes, which were attached with Velcro® tape to the bottom of polycarbonate containers. The containers had two large slots for ventilation on two sides which also allowed egg parasitoid to disperse once emerged. The polycarbonate containers were attached to the horizontal wire (about head height) on the trellises.

Containers with sentinel cards with fresh *P. affinis* eggs (Figure 7) were put out after releases to see if *Trissolcus* sp. could be recaptured. Ten raspberry bushes next to the release points (5 bushes to the left and 5 bushes to the right) were monitored for presence of *P. affinis* adults, nymphs and eggs and their parasitism (Figure 8). To get a baseline of presence of bugs and egg parasitoids, the same amount of raspberry bushes were monitored in 3 tunnel rows where no releases of *Trissolcus* sp. had been made.

Telenomus release 2014

Telenomus sp. was released at Corindi on 20 May 2014 (153 parasitised eggs) (Figure 9) and collected on 29 May 2014. The release was made alternately in the northern and southern end of 2 rows in 2 adjacent tunnels. A diagram of the experiment is shown in Figure 10. Each release point (1 per row) received between 35 and 42 parasitised eggs (average 38.3), as described above.

Telenomus release 2015

In January 2015, 161 *P. affinis* eggs parasitised by *Telenomus* sp. were released again in block S1 at Costa Berry Exchange Corindi on 14 January 2015.

Releases were made in the each middle row of 4 tunnels, alternating at the northern or southern end. A diagram of the release trial is shown in Figure 11. Each release site received 45-50 parasitised eggs. Releases were made as described above. The release row was checked for presence of *P. affinis* to ensure an even pest pressure at release sites.

The trial was monitored for development of parasite population by putting out sentinel eggs of *P. affinis* in the trial area. Each end of the treatment rows (control and release ends in Figure 11) received 10 fresh eggs of *P. affinis* target eggs on Post-it® notes in polycarbonate containers as described above at each end of the treatment rows after each release in 2013 and 2014 and 26-72 (see Table 3) eggs in 2015.

Figure 1: *Trissolcus* sp.

Figure 2: *Plautia affinis* eggs parasitised by *Trissolcus* sp.

Figure 3: *Telenomus* sp.

Figure 4: *Plautia affinis* eggs parasitised by *Telenomus* sp.

Figure 5: Fly species parasitising *Plautia affinis* - left: unidentified tachinid species adult bugs, fly pupae and adult flies - right: *Trichopoda* sp. adult bugs with fly eggs, fly pupa and adult fly

Figure 6: Release of *Trissolcus* sp. in raspberry tunnel at Berry Exchange Corindi

Figure 7: Sentinel cards with *Plautia affinis* eggs in polycarbonate containers

Figure 8: *Plautia affinis* eggs on raspberry fruit

Figure 9: Eggs being parasitised

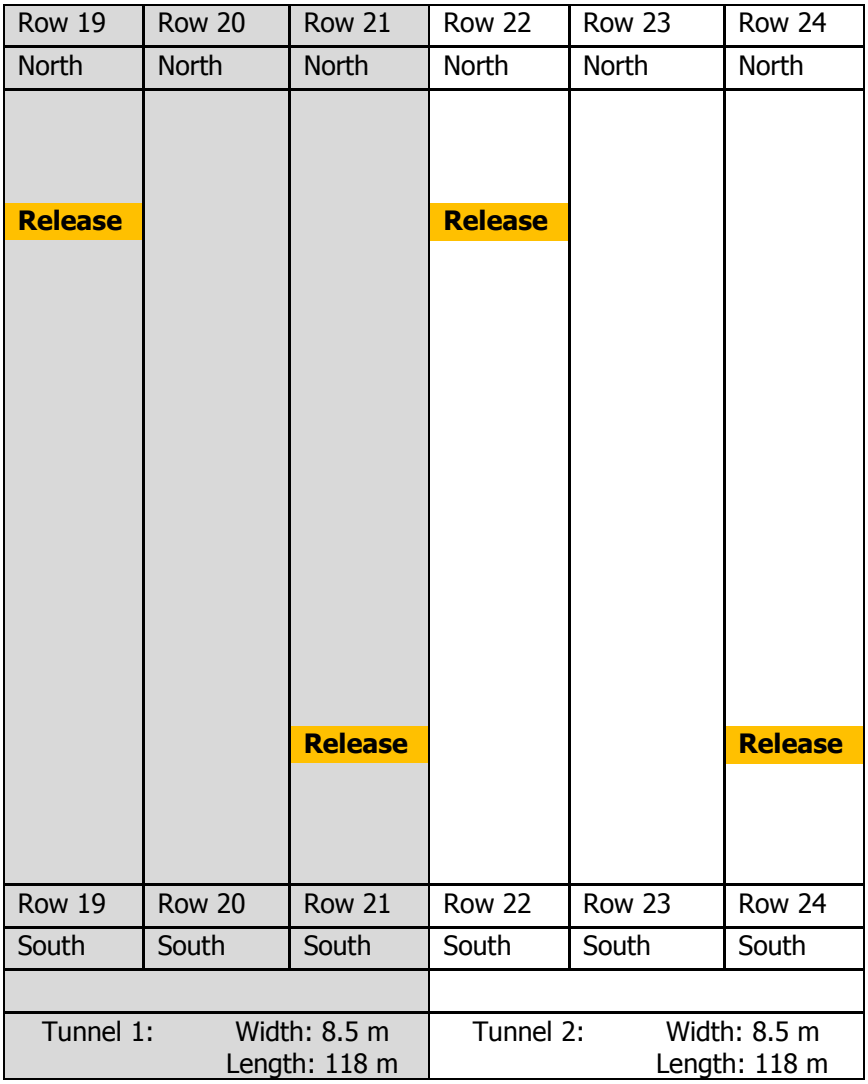


Figure 10: Diagram of *Telenomus* release trial May 2014

After a week, the containers were collected and brought back to the laboratory. The eggs on the paper were put into test tubes with ventilated lids and they were checked regularly for emergence of nymphs or parasitoids. After about 4 weeks, eggs were counted, resulting wasps and nymphs recorded and eggs without emergence of nymphs or wasps were dissected.

Table 3: Sentinel card egg numbers released in January 1015

Row	Treatment	Replicate	Egg numbers
2	*Telenomus*	1	32
2	Control	1	56
5	*Telenomus*	2	57
5	Control	2	72
8	*Telenomus*	3	53
8	Control	3	
11	*Telenomus*	4	26
11	Control	4	26

On the 25 February 2015 Row 13 next to the releases was also monitored for background parasitism, by collecting 18 egg batches in the row and checking them for parasitism.

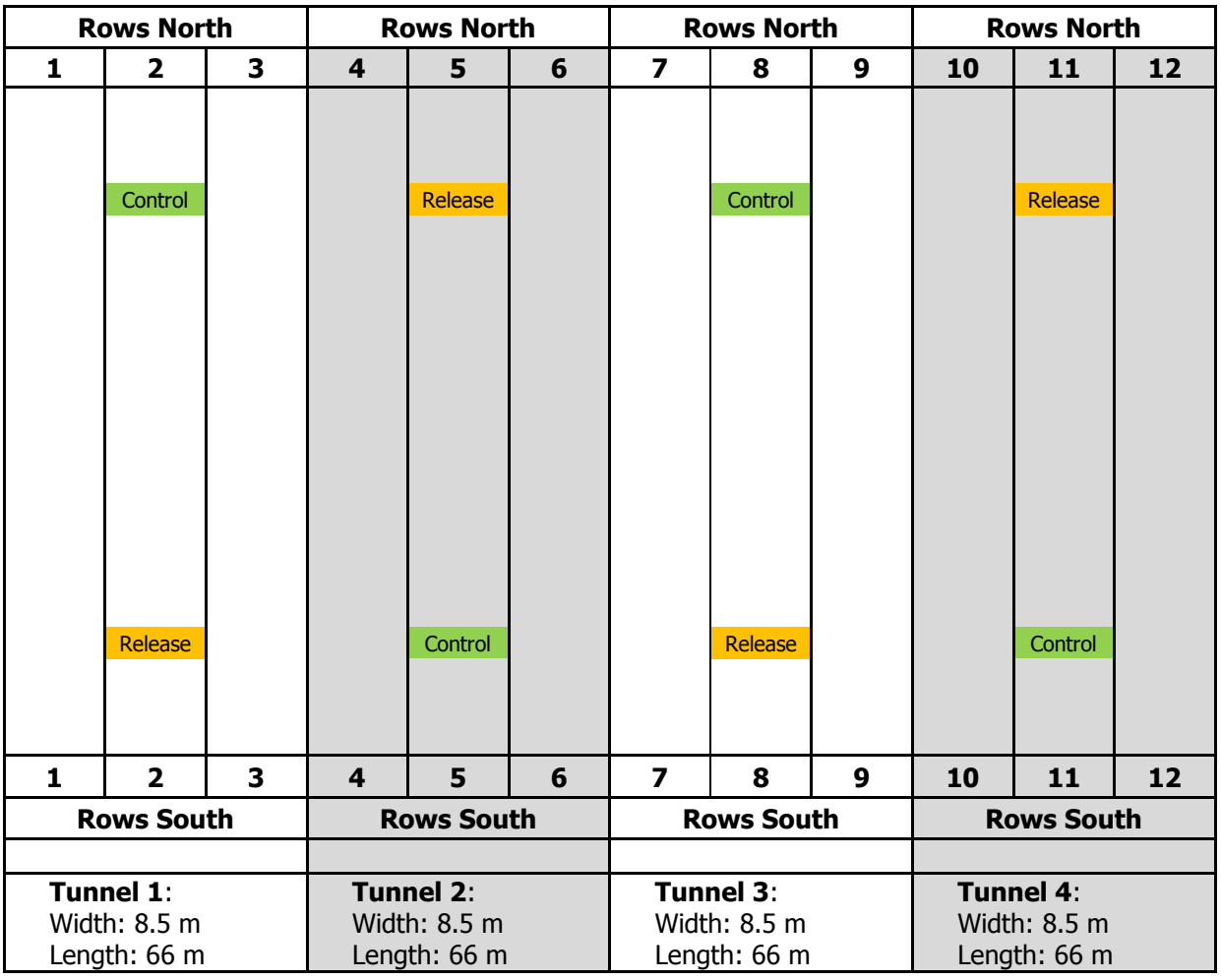


Figure 11: Diagram of *Telenomus* sp. release trial January 2015

3. Chemical control

3.1. Laboratory screening of insecticides

Selected insecticides were initially screened in the laboratory using the drop test techniques first and then the treated surface technique as described below. We included chemicals that we knew would fit into an IPM system, would be effective on bugs (i.e. Admiral®, (Lepidex, PyGanic® etc.) and also new chemistry that we wanted to test for their effectiveness on bugs (i.e. DC142, MCW9540, flonicamid, tolfenpyrad etc.) and look at their potential role in management of *P. affinis*.

Drop test

Insecticides were tested at recommended rates (see Table 5). Trichlorfon (i.e. Lepidex) and an untreated control were used as benchmark treatments.

One microlitre of the test solution was applied on the thoracic plate of individual bugs from the colony at WPII, using a calibrated Hamilton micro-syringe. Mortality was checked after 1, 2, 3 and after 7 days. Mortality rates of different treatments were compared. Insecticides tested and results are listed in Table 5.

Treated surface test

A number of chemicals were screened against adult *P. affinis* using the treated surface test. Five adult bugs were placed in a 750 ml square plastic food container with a treated food source, pieces of corn cobs which were dipped into the insecticide solution (Figure 12). Mortality of *P. affinis* adults was recorded after 1, 2 and 3 days. Each treatment was replicated 3 times. Different treatments were compared to a water treated control. Insecticides tested and results are listed in Table 6.

Numbers of insects available for laboratory trials were a limiting factor and care has to be taken when interpreting the results.

3.2. Insecticide field trial

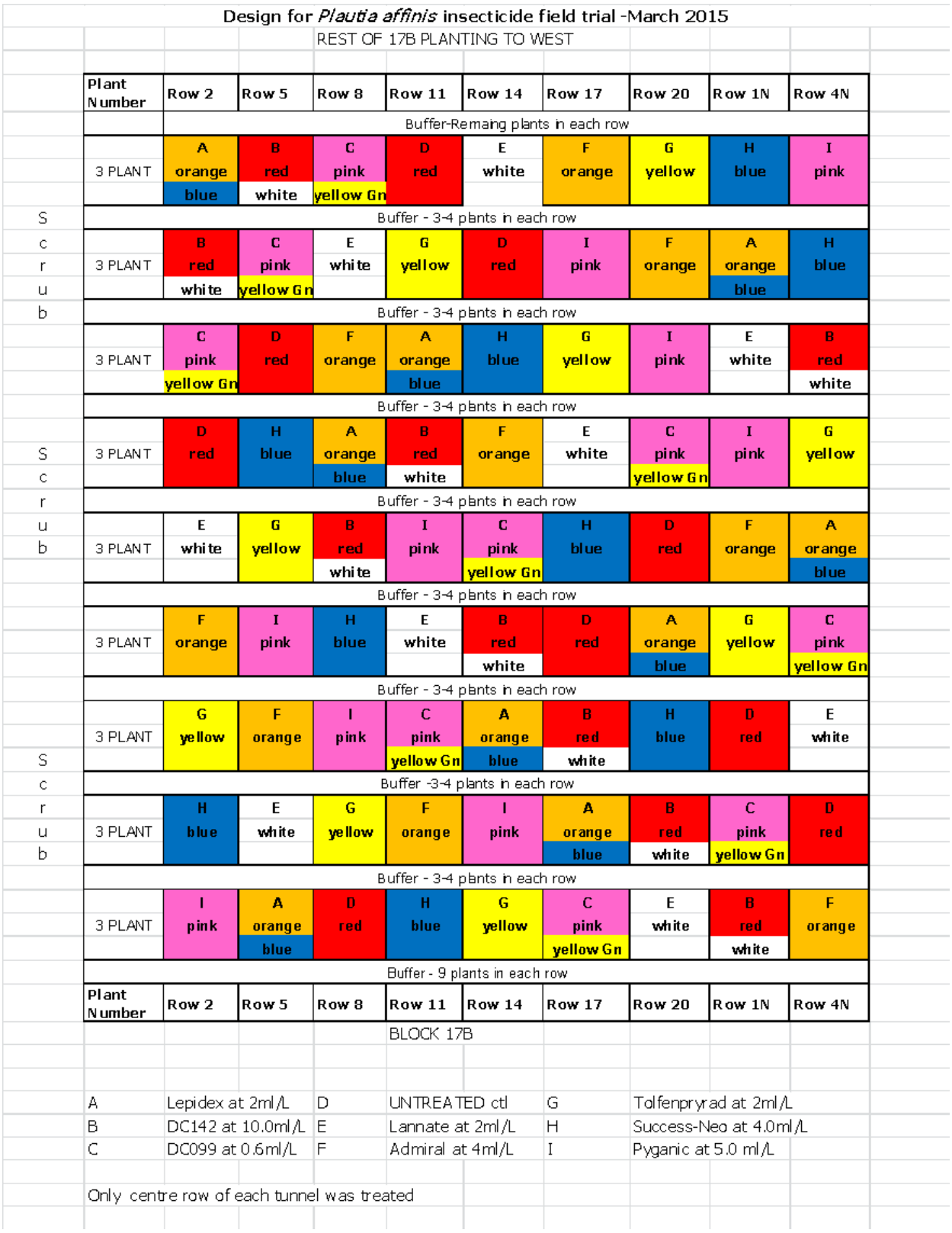
A field trial was set up in March 2015. A block of tunnels with raspberries that had young fruit was selected and a Latin square design was used for the trial. Details of the design are shown in Figure 13. The middle row under a tunnel was used as a treatment row. The rows to the right and left were kept untreated to avoid any drift issues. Within each row, 9 treatment blocks were assigned. Each treatment block consisted of 3 raspberry plants. Between each block/treatment replicate we had 3-4 buffer plants also to account for potential drift between treatments.

Plants were sprayed with a 15L Solo knapsack sprayer on 29 March 2015. Spay applications were undertaken late afternoon into the evening when bees were back in their hive, to minimise chemical impact on bees. The trial was monitored for presence of dead and live *P. affinis* adults and nymphs the next day (30 March 2015) and again after one week (7 April 2015). This was done by visually checking treated plants and ground for evidence of bugs. We wanted to get information on the knock down effect of chemicals and also wanted to check the effect over one week, including potential re-colonisation of bugs. Results are shown in Table 7.

4. Monitoring and Management strategy-

A practical way of monitoring commercial plantings was required. Discussions were held with Alejandro Haro and Brittney Landsberry from Costa Berry Exchange at Corindi and as a consequence, thresholds and a monitoring protocol were developed (Table 8).

Figure 12: Insecticide screening set up for *Plautia affinis*



Design for *Plautia affinis* insecticide field trial -March 2015

REST OF 17B PLANTING TO WEST

	Plant Number	Row 2	Row 5	Row 8	Row 11	Row 14	Row 17	Row 20	Row 1N	Row 4N
		Buffer-Remaing plants in each row								
	3 PLANT	A orange blue	B red white	C pink yellow Gn	D red	E white	F orange	G yellow	H blue	I pink
S		Buffer - 3-4 plants in each row								
c	3 PLANT	B red white	C pink yellow Gn	E white	G yellow	D red	I pink	F orange	A orange blue	H blue
r		Buffer - 3-4 plants in each row								
u	3 PLANT	C pink yellow Gn	D red	F orange	A orange blue	H blue	G yellow	I pink	E white	B red white
b		Buffer - 3-4 plants in each row								
	3 PLANT	D red	H blue	A orange blue	B red white	F orange	E white	C pink yellow Gn	I pink	G yellow
S		Buffer - 3-4 plants in each row								
c	3 PLANT	E white	G yellow	B red white	I pink	C pink yellow Gn	H blue	D red	F orange	A orange blue
r		Buffer - 3-4 plants in each row								
u	3 PLANT	F orange	I pink	H blue	E white	B red white	D red	A orange blue	G yellow	C pink yellow Gn
b		Buffer - 3-4 plants in each row								
	3 PLANT	G yellow	F orange	I pink	C pink yellow Gn	A orange blue	B red white	H blue	D red	E white
S		Buffer -3-4 plants in each row								
c	3 PLANT	H blue	E white	G yellow	F orange	I pink	A orange blue	B red white	C pink yellow Gn	D red
r		Buffer - 3-4 plants in each row								
u	3 PLANT	I pink	A orange blue	D red	H blue	G yellow	C pink yellow Gn	E white	B red white	F orange
b		Buffer - 9 plants in each row								
	Plant Number	Row 2	Row 5	Row 8	Row 11	Row 14	Row 17	Row 20	Row 1N	Row 4N

BLOCK 17B

A	Lepidex at 2ml/L	D	UNTREATED ctl	G	Tolfenpyrad at 2ml/L	
B	DC142 at 10.0ml/L	E	Lannate at 2ml/L	H	Success-Neo at 4.0ml/L	
C	DC099 at 0.6ml/L	F	Admiral at 4ml/L	I	Pyganic at 5.0 ml/L	

Only centre row of each tunnel was treated

Figure 13: Design of raspberry insecticide trial at Corindi

Appendix 2: Detailed Results

1. Laboratory rearing of bugs

We successfully maintained a small colony of *Plautia affinis* at WPII, as described above. We successfully maintained a small colony of *P. affinis* at WPII on commercially available green beans and corn. We also tried to give them wild tobacco, *Solanum mauritianum* Scopoli (Solanaceae) when available, which were collected from orchard and roadsides in the area close to Wollongbar. Wild tobacco tended to increase their oviposition.

The lifespan of *P. affinis* adults was generally fairly short (2-3 weeks). The colony however was sufficient to provide insects for insecticide screening and rearing of egg parasitoids at different times.

2. Biological control

2.1. Laboratory rearing

Trissolcus sp.

We were able to maintain a small colony *Trissolcus* sp. between 2012 and 2013, which enabled us to undertake one field release. At a room temperature of 25°C (±2) the development time for *Trissolcus* sp. took an average of 23.5 days (±1.18). The average parasitism was 69.37% (±4.09) (Figure 14) and the percentage of *Trissolcus* sp. emerging from eggs was 35.03% (±4.46) (Figure 15).

Telenomus sp.

Between 2013 and 2015 we maintained a small laboratory colony of *Telenomus* sp. We were able to carry out two small field release trials.

At a room temperature of 25°C (±2) the development time for *Telenomus* sp. took an average of 23.1 days (±0.99). The average parasitism was 61.74% (±4.75) (Figure 14) and the percentage of *Telenomus* sp. emerging from eggs was 30.57% (±4.20) (Figure 15).

The rearing process itself for both egg parasitoids was easy was but the number of *P. affinis* eggs available as a food source was the limiting factor for the laboratory colonies.

Due to the limited number of fresh eggs from *P. affinis*, we were only able to rear one parasitoid species for releases at the time.

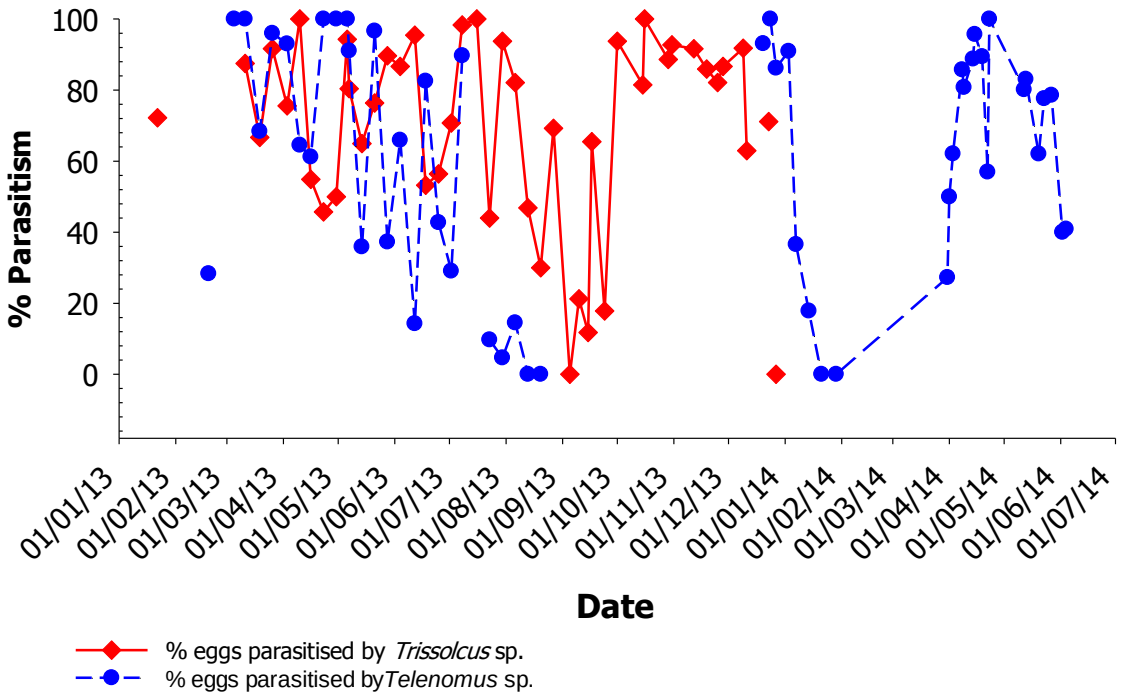


Figure 14: Parasitism rates by *Trissolcus* sp. and *Telenomus* sp.

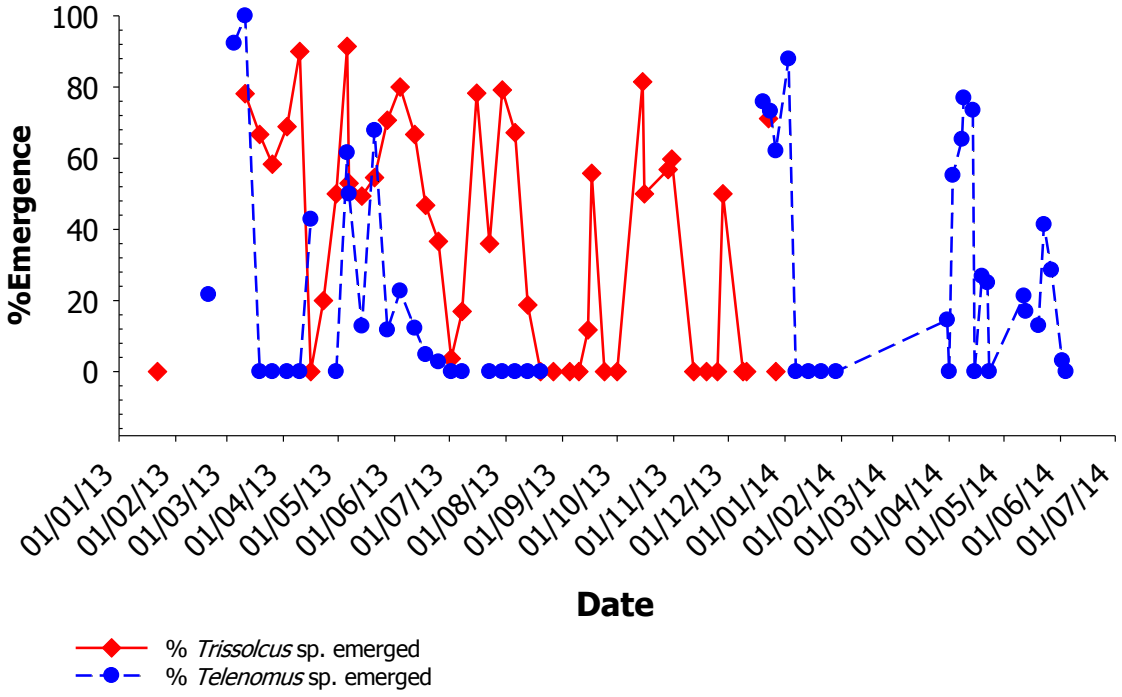


Figure 15: Emergence rates by *Trissolcus* sp. and *Telenomus* sp.

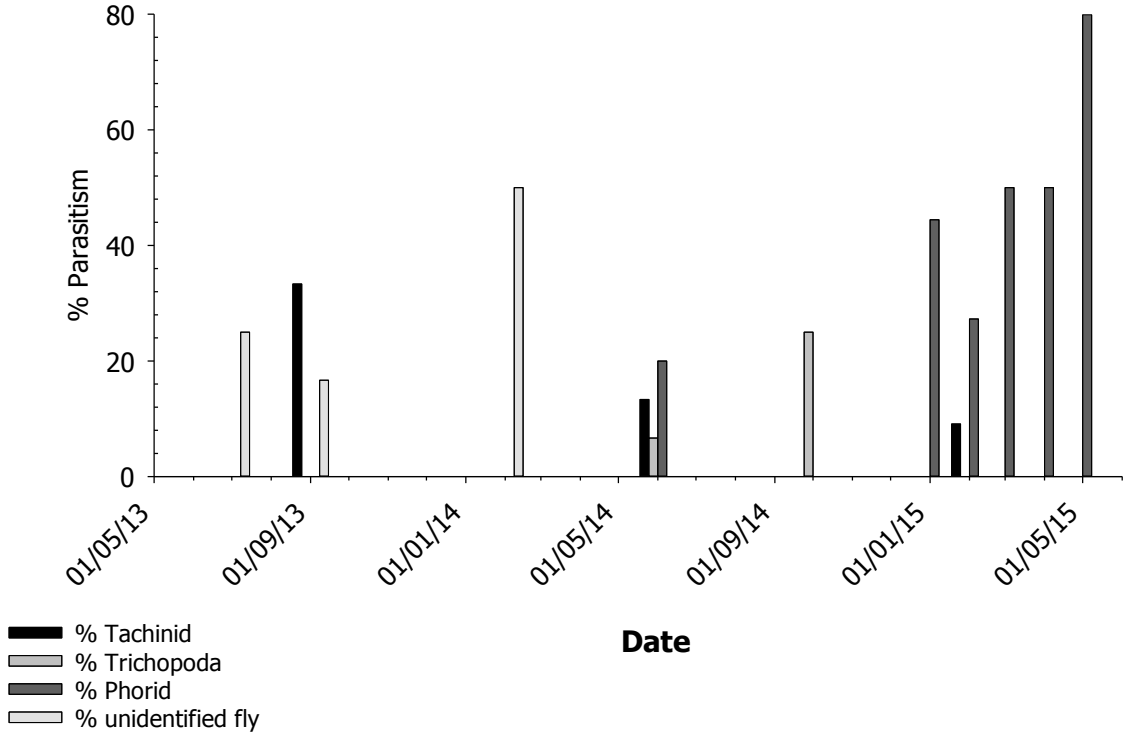


Figure 16: Parasitism by different fly species, in dead *Plautia affinis* bodies collected from the field

Dead bodies of *P. affinis* were checked for parasitism in the laboratory. Results are shown in Figure 16. Phorid flies appeared to be the dominant parasite. Of dead adults collected, parasitism by phorids varied between 20-80% at times (Figure 16). Parasitism by the other fly species was very sporadic.

2.2. Field releases

Trissolcus release March 2013

In the release area, a total of 154 eggs were collected. In the release area 9 eggs (5.8%) were parasitised by *Trissolcus* sp. and 13 eggs (8.4%) were parasitised by *Telenomus* sp. (Figure 9). *Telenomus* sp. naturally occurs in the raspberry plantings and to this point had not been released.

In the control areas with no release 242 *P. affinis* eggs were collected. One of the collected eggs (0.4%) was parasitised by *Trissolcus* sp. and 44 eggs (18.2%) were parasitised by *Telenomus* sp.

In summary, a total of 396 *P. affinis* eggs were collected, 57 eggs (14.4%) were parasitised by *Telenomus* sp. and 10 eggs (2.5%) by *Trissolcus* sp.

Monitoring the natural presence of the 2 egg parasitoids (*Trissolcus* sp. only 0.4% parasitism and *Telenomus* sp. 18.2% parasitism) lead to the conclusion that *Telenomus* sp. appears to be the stronger candidate for biocontrol of *P. affinis* in the raspberry tunnels at Corindi.

Telenomus release May 2014

The average rate of *Telenomus* hatching from parasitised eggs was 86.3% of the eggs released (hatching rate 77.1 – 100.0%). Parasitism and presence of *P. affinis* was monitored in control and release areas (Table 4, Figure 10).

There was no significant difference ($P > 0.05$) (ANOVA followed by LSD) between control and release treatments in any of the different categories we looked at, no difference in parasitism and no difference in the presence of *P. affinis*. With the small release of *Telenomus* sp. however, we managed to increase parasitism by 43.79% (from 18.45% to 26.53%) (Table 5).

Table 4: Parasitism after release in May 2014

Row	**Row end**	**Number of eggs in**	**Eggs parasited**	**Species**	**% emergence**
19	North	41	41	*Telenomus*	100.00
21	South	35	28	*Telenomus*	80.00
22	South	42	37	*Telenomus*	88.10
24	South	35	27	*Telenomus*	77.14
Total		153	133		
Average		38.25	33.25		86.31

Table 5: Monitoring of *Plautia* after *Telenomus* release results 2014. Numbers indicate the mean number of individuals.

Treatment	**Control site (SD)**	**Release site (SD)**
Plautia adults	23.75 (9.22)	21.25 (2.99)
Plautia small nymphs	2.75 (0.96)	1.25 (1.89)
Plautia large nymphs	17.25 (7.93)	8.00 (3.74)
Total *Plautia* nymphs	20.00 (8.72)	9.25 (5.32)
Total *Plautia*	43.75 (16.78)	30.50 (5.97)
Telenomus parasitism (%)	18.45 (22.12)	26.52 (23.68)

Comparing the results with earlier releases of *Trissolcus* sp., taking in consideration that releases were made at different seasons, it appears that *Telenomus* sp. would be the better biological control agent. Parasitism observed was exclusively done by *Telenomus* sp. and it seems to be quite persistent in the field. This suggests that it would be the more effective egg parasitoid.

Telenomus release January 2015

In the row next to the release site we collected 18 egg masses out of which 6 egg masses were parasitised. Two egg masses were parasitised by *Trissolcus* sp., 1 egg mass by *Telenomus* sp. and 3 egg masses by an unidentified egg parasitoid.

The trial was monitored on 21 January 2015 for field parasitism and emergence of parasite release. 82.2% of the released parasites emerged.

Number of wasps, nymphs and eggs without emergence of nymphs or wasps, resulting from dissections of the re-capture at the release site are shown in Table 6. Between the control and the release treatment, there was no significant difference in *Plautia affinis* nymphs hatching (P=0.937), total percentage of parasitism of eggs (P=0.215) (ANOVA followed by LSD).

Averages of parasitism per treatment are shown in Table 7. Between the control and the release treatment, there was no significant difference in parasitism by *Telenomus* sp. (P=0.443), parasitism by *Trissolcus* sp. (P=0.368), parasitism by unidentified parasite (P=0.209) or percentage of infertile eggs (P=0.230) and there was no difference between parasitism of the different parasitoid species in the control and release treatment (P=0.207) (ANOVA followed by LSD). *P. affinis* populations were generally fairly low during the past season and the general background parasitism in row 13, adjacent to release site was relatively high (Table 8). Small scale releases of *Telenomus* sp. therefore did not increase parasitism or have a significant impact on the pest population.

Table 6: Details of re-capture trial at *Telenomus* release site after release in January 2015

Row	Treatment	Rep	Egg numbers	*Plautia* nymphs	% Total Parasitism	% Parasisim by *Telenomus*	% Parasisism by *Trissolcus*	% Parasitism by unidentified parasitoid	% dead parasitoids	% infertile eggs
2	*Telenomus*	1	32	5	25.00	15.63	0.00	0.00	9.38	59.38
2	Control	1	56	0	41.07	30.36	0.00	10.71	0.00	58.93
5	*Telenomus*	2	57	19	7.02	0.00	0.00	7.02	0.00	59.65
5	Control	2	72	1	51.39	9.72	1.39	11.11	29.17	47.22
8	*Telenomus*	3	53	2	86.79	3.77	0.00	22.64	60.38	9.43
8	Control	3	missing							
11	*Telenomus*	4	26	0	7.69	7.69	0.00	0.00	0.00	92.31
11	Control	4	26	11	7.69	0.00	0.00	0.00	7.69	50.00

Table 7: Average parasitism per treatment re-capture trial at *Telenomus* release site after release in January 2015

Treatment	Egg numbers	*Plautia* nymphs	% Total Parasitism	% Parasitism by *Telenomus*	% Parasitism by *Trissolcus*	% Parasitism by unidentified parasitoid	% dead parasitoids	% infertile eggs
Control	51.33	4.00	33.38	13.36	0.46	7.28	12.29	52.05
Telenomus	42.00	6.50	31.63	6.77	0.00	7.41	17.44	55.19

Table 8: Background parasitism (average over 18 egg batches) adjacent to *Telenomus* release site (Row 13)

Total egg numbers	Average egg numbers	*Plautia* nymphs	% Total Parasitism	% Parsitism by *Telenomus*	% Parasitism by *Trissolcus*	% Parasitism by unidentified parasitoid	% dead parasitoids	% infertile eggs
197	10.94	7.36	43.87	14.80	16.75	16.75	0.35	13.63

3. Chemical control

3.1. Laboratory screening of insecticides

Drop test

Results or the drop test were inconclusive due to high mortality (50%) in the control treatment (water) after 3 days and a different screening method needed to be chosen. However, results suggest that the only chemicals resulting in 100% mortality of *P.* affinis adults at day 7 were Lannate® and Sivanto™. Lepidex and Exirel® gave 92% mortality at day 7 (Table 8).

Table 8: List of insecticides screened and results from drop test as % mortality

Insecticide	Active	Rate (ml/L)	Number of insects (=n)	Mortality%		
				Day 1	Day 3	Day 7
Success™ Neo (Dow AgroSciences)	Spinetoram 120g/L	0.4	11	36.4	54.5	54.5
Stealth® (CROPPRO)	Abamectin 18 g/L	1.0	12	41.7	58.3	83.3
Sivanto™ (Bayer CropScience)	Flupyradifurone	1.0	12	58.3	83.3	100.0
Exirel® (DuPont)	Cyantraniliprole	1.0	12	8.3	50.0	91.7
Lannate® (Crop Care)	Methomyl 225 g/L	2.0	12	41.7	66.7	100.0
Lepidex (Nufarm)	Trichlorfon 500 g/L	2.0	12	16.7	100.0	91.7
Control	Water	1000.0	12	16.7	50.0	83.3

Treated surface test

Over time a number of screening trials were conducted and the summary results of all screening trials are shown in Table 9.

As a conclusion, we have tested a number of new and old insecticides. There is a couple of new insecticides Sivanto™ and Exirel®, but also Lepidex that gave reasonable results in previous tests and tolfenpyrad that might be useful. These results need to be confirmed in further tests. Overall Lannate® and tolfenpyrad gave the best results in the laboratory screening.

Lannate® and tolfenpyrad, due to their detrimental effect on bees while the chemicals are active, are not a suitable option.

Table 9: Summary list of insecticides screened and the overall results from a treated surface test

Treatment	**Active**	**Gramm of active ingredient/L (gai/L)**	**Average % mortality at Day 3 (se)***	
Admiral® (Sumitomo)	Pyriproxyfen 100gm/L	0.1000	79.73 (16.60)	a,b
Admiral® (Sumitomo)	Pyriproxyfen 100gm/L	0.2000	40.00 (23.47)	a,b
Control	Water	0.0000	16.39 (9.59)	a,b
DC142(10 ml/L) (Bayer CropScience)	**	0.0000	59.73 (16.60)	a,b
Flonicamid (UPL)	Flonicamid 500WP	1.0000	49.73 (16.60)	a,b
Lannate® (Crop Care)	Methomyl 225gm/L	0.4500	99.73 (16.60)	a
Lepidex (Nufarm)	Trichlorfon 500gm/L	0.1000	100.00 (13.58)	a
Trivor (0.125 ml/L) (ADAMA)	Acetamiprid 186g/L +	0.0233	9.73 (16.60)	a,b
	Pyriproxyfen 124g/L	0.0155		
Trivor (0.25 ml/L) (ADAMA)	Acetamiprid 186g/L +	0.0465	0.00 (16.60)	b
	Pyriproxyfen 124g/L	0.0310		
Trivor (0.50 ml/L) (ADAMA)	Acetamiprid 186g/L +	0.0930	9.73 (16.60)	a,b
	Pyriproxyfen 124g/L	0.0620		
Trivor (1.0 ml/L) (ADAMA)	Acetamiprid 186g/L +	0.1860	80.73 (13.58)	a,b
	Pyriproxyfen 124g/L	0.1240		
Trivor (2.0 ml/L) (ADAMA)	Acetamiprid 186g/L +	0.3720	49.73 (16.60)	a,b
	Pyriproxyfen 124g/L	0.2480		
Trivor (4.0 ml/L) ADAMA	Acetamiprid 186g/L +	0.7440	79.73 (16.60)	a,b
	Pyriproxyfen 124g/L	0.4960		
Primal® (ADAMA)	Acetamiprid 200g/L	0.1000	39.73 (16.60)	a,b
Primal® (ADAMA)	Acetamiprid 200g/L	0.2000	49.73 (16.60)	a,b
PyGanic® (OCP)	Pyrethrins 13g/L	0.0130	40.00 (23.47)	a,b
PyGanic® (OCP)	Pyrethrins 13g/L	0.0260	59.73 (16.60)	a,b
Steward® (DuPont)	Indoxacarb 150gm/L	0.6000	9.73 (16.60)	a,b
Success™ Neo (Dow AgroSciences)	Spinetoram 120gm/L	0.2400	59.73 (16.60)	a,b
Tolfenpyrad (UPL)	Tolfenpyrad 150gm/L	0.3000	87.23 (16.60)	a,b

*Averages with different letters indicate means are significantly different (P >0.05). (ANOVA followed by LSD)

**Withheld by manufacturer

Numbers of insects available for laboratory trials were a limiting factor and care has to be taken when interpreting the results.

2.1. Field insecticide trial

Results from field trials are shown in Table 10. The results are showing the percentage of dead *P. affinis* individuals found 1 day and 1 week after treatment application.

There was a significant difference between treatments (P<0.001) (ANOVA followed by LSD). The PyGanic® treatment had significantly more dead *P. affinis* than the three new products Admiral®, DC142 and DC099. Overall, PyGanic® and tolfenpyrad gave the best results in the insecticide trials and also the field trial. Due to to their compatibility with bees when appropriate precausions are taken, pyrethrins (i.e PyGanic®) are the more appropriate candidates for management of *P. affinis*.

Table 10: List of treatments and the results of insecticide field trial at Corindi from assessments at 1 day and 1 week after treatment.

Treatment	**Active**	**Applied rate (ml/L)**	**Gramm of active ingredient/L (gai/L)**	***% Plautia* (nymphs and adults) dead***	
Admiral	Pyriproxyfen 100gm/L	4.0ml/L	0.2000	0.00	b
Control	Water	0.00	0.00	11.00	a,b
DC-099 (Bayer CropScience)	**	0.6ml/L	**	4.40	b
DC-142 (Bayer CropScience)	**	10 ml/L	**	0.90	b
Lannate® (Crop Care)	Methomyl 225gm/L	2.0ml/L	0.4500	6.10	a,b
Lepidex (Nufarm)	Trichlorfon 500gm/L	2.0ml/L	0.1000	15.00	a,b
PyGanic® (OCP)	Pyrethrins 13g/L	5.0ml/L	0.0260	44.60	a
Success™ Neo (Dow AgroSciences)	Spinetoram 120gm/L	4.0ml/L	0.2400	11.00	a,b
Tolfenpyrad (UPL)	Tolfenpyrad 150gm/L	2.0ml/L	0.3000	35.10	a,b

*Averages with different letters indicate means are significantly different (P >0.05). (ANOVA followed by LSD)

**Withheld by manufacturer

Best candidates

3. Monitoring and Management strategy – Best Practice

During the course of the study, we noticed that *P. affinis* adults shelter during winter (Coombs & Khan, 1998) in the dry leaves in the raspberry plants. It would be important to culturally manage this population.

An essential part of the management strategy would need to be weekly or fortnightly monitoring of *P. affinis* and egg parasitoids once the plants have started to set berries.

It is important to monitor when *P. affinis* starts to appear in different blocks during spring and then use a knock down spray after *P. affinis* adults are detected. Afterwards (possibly 7 days) releases of parasitoids should commence to clean up residual bug eggs.

Monitoring can be done by walking the planting visually and counting *P. affinis* nymphs, adults and egg masses. Egg batches need to be collected and checked for parasitism.

Pest pressure is estimated and actions thresholds developed as described in Table 11.

It will be important to get egg parasitoid populations to build up early and release them in tunnels as soon as pests are observed in the planting.

PyGanic® is the preferred chemical option, as its residual time is very short and it can easily be used when appropriate precautions are taken and bees are not active (i.e. at night when bees are in the hive). A maximum of 2 applications per crop cycle should be used when indicated by monitoring to reduce the peak of the bug population.

Table 11: Evaluation of monitoring populations of *Plautia affinis* in raspberries

Pest pressure	**Observation**	**Action**
Low	<0.5 adult/plant (1adult/10 plants) 0.2-0.3 egg masses per plant	Keep monitoring or get ready for wasp release
Medium	0.6-2.0 adult/plant <0.1 egg masses/plant 1.5-3.0 nymphs/plant	Get ready for chemical treatment or release wasps asap
High	>2.0 adults/plant >2.0 egg masses/plant >10.0 nymphs/plant	Apply chemical treatment and release wasps after withholding period