Horticulture Innovation Australia

Final Report

New in-field treatment solutions to control Fruit Fly (1)

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Project Number: VG13041

VG13041

This project has been funded by Horticulture Innovation Australia Limited with co-investment from Vegetable (R&D Levy), The Department of Agriculture and Fisheries (DAF) and funds from the Australian Government.

ISBN 978 0 7341 3954 2

Published and distributed by:
Horticulture Innovation Australia Limited
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Content

Summary

In-field management of fruit flies in fruiting vegetable crops has relied heavily on regular cover sprays with dimethoate and fenthion. However, recent restrictions in their use, and the possibility of further future restrictions, mean that alternative control options are required. The project aimed to assess a combination of perimeter protein baiting and male annihilation, and alternative chemical options, as well as obtaining data on seasonal fruit fly activity in vegetables.

Semi-field trials were performed to assess eight insecticides, applied as cover sprays to fruiting capsicum and zucchini plants, for efficacy against Queensland fruit fly, *Bactrocera tryoni*, and cucumber fly, *Zeugodacus cucumis*. Clothianidin was very effective against Queensland fruit fly and cucumber fly. Thiacloprid, imidacloprid, cyantraniliprole and alpha-cypermethrin were also very effective against Queensland fruit fly, but less so against cucumber fly. Bifenthrin, spinetoram and abamectin demonstrated a suppressive effect. Alpha-cypermethrin, bifenthrin and dimethoate were linked to higher incidence of aphid and silverleaf whitefly infestation. A laboratory trial, in which Queensland fruit fly were exposed to dried insecticide residues on capsicum fruit, found that efficacy of thiacloprid was comparable with dimethoate, and spinetoram had a suppressive effect. Chlorantraniliprole and flubendiamide were ineffective.

A trial was performed in a commercial chilli crop in Bundaberg to assess a combination of perimeter protein baiting and male annihilation for management of fruit fly. Sampling of fruit from the trial block throughout harvest found that the treatments successfully prevented infestation: no fruit fly larvae were found in any sampled fruit (a total 6966 fruit, 91 kg), with an upper infestation level of 0.04% (95% confidence). For comparison, sampling was also conducted in a second block, where regular cover sprays with dimethoate, trichlorfon and methomyl were applied. Seven flies were found in fruit sampled from the comparison block (3048 fruit, 49 kg), with an upper infestation level of 0.21% (95% confidence).

The trial was repeated on a smaller scale in a research planting of capsicums at Bundaberg Research Facility, using a combination of perimeter protein baiting, male annihilation, and fortnightly cover sprays with spinetoram. A total of 12,995 fruit (2488 kg) were sampled. No larvae were recovered during winter, when fruit fly activity was low; however the control measures were not sufficient to prevent infestation outside of this period. The high local fruit fly pressure at the trial site coupled with the smaller size of the area over which control measures were applied were most likely critical factors.

Monitoring was performed to obtain more information on the seasonal activity of fruit flies in vegetable crops in the Bundaberg region. Peak trap catch occurred in the spring, with a second peak in the summer. Monitoring also indicated an edge effect, with more flies caught in traps located along a tree-line, or within the crop close to the tree-line, compared with those further within the crop. A trial targeting cucumber fly, using traps baited with a cucumber volatile lure, found that the BioTrap (BioTrap Australia Pty Ltd) was a better performing trap type for this species, catching an average of 9.1 cucumber flies per trap per day, compared with 0.7 cucumber flies per trap per day caught by Bugs for Bugs traps (Bugs for Bugs Pty Ltd). The average sex ratio of trapped flies was 74:26 female:male. However, further trials performed in the Fassifern Valley failed to catch cucumber flies.

Keywords

Fruit fly, Queensland fruit fly, *Bactrocera tryoni*, cucumber fly, *Zeugodacus cucumis*, protein baiting, male annihilation, insecticide cover sprays, monitoring, seasonal activity

Introduction

In-field management of fruit flies in fruiting vegetable crops has relied heavily on regular cover sprays with dimethoate and fenthion. However, recent restrictions in their use (APVMA, 2011, 2015, 2017) and the possibility of further future restrictions, will make control more difficult. Regular use of broad spectrum cover sprays also precludes the successful implementation of integrated pest management (IPM) in these crops, and pests such as silverleaf whitefly are becoming difficult to manage with insecticides in many areas due to resistance. If alternative effective controls are not found, growing vegetables in endemic fruit fly areas will cease to remain viable.

A combination of protein bait spraying and male annihilation technique (MAT), coupled with field hygiene and fruit inspection, was developed for management of fruit fly in tree crops (Lloyd et al, 2007, 2010; Fay et al, 2011). This has become a model approach and was further developed in later work in table grapes (Oag et al, 2010), and more recently in strawberries, based on a winter window but also incorporating bait sprays applied to fruit fly resting sites on the perimeter of strawberry blocks (Missenden, 2014). Queensland fruit fly is thought to behave differently in low growing crops compared with orchards, invading the crop from the field margins (Balagawi et al, 2014). Perimeter baiting, where protein bait is applied to vegetation surrounding a crop, was developed for control of melon fly, *Zeugodacus cucurbitae*, in curcurbits in Hawaii (Prokopy et al, 2003). This species has been shown to roost in vegetation on the field margin, only entering the crop to oviposit (Nishida and Bess, 1957). Aspects of perimeter bait application in vegetable crops were also explored by DeFaveri (2013), however the efficacy of this technique to control fruit flies in vegetable crops in Australia is currently unproven.

Two trials were performed to evaluate the use of perimeter protein baiting and MAT, the first in a commercial chilli crop, and the second on a smaller scale in a research capsicum crop. As perimeter baiting and MAT alone may not be sufficient to achieve full control, the project also evaluated the efficacy of a range of insecticides as alternatives to dimethoate. A number of studies have examined the toxicity of novel pesticides to tephritid fruit flies in the laboratory, and small plot trials have assessed efficacy of insecticides for management of cucurbit specific fruit flies such as melon fly, but the literature on insecticide efficacy for Queensland fruit fly and cucumber fly is scarce. Moreover, as vegetable crops are often subject to intensive insecticide regimes for management of other pest species, it is important to understand the potential impact of these insecticides on fruit flies. For instance, Subramaniam et al (2011) demonstrated that pre-harvest cover sprays for pests other than fruit flies was sufficient to minimise fruit fly pressure during the Bowen harvest season and prevent infestation in tomato and capsicum fruit. Likewise, laboratory trials confirmed that several insecticides used as cover sprays for pests other than fruit flies had efficacy against fruit flies (Senior and Wright, 2012). Fruit fly monitoring was also undertaken throughout the project in order to obtain data on seasonal fruit fly activity and to evaluate a cucumber volatile blend lure shown to have potential for use with cucumber fly in Australia (Royer et al, 2014). Additional trials investigated activity within a capsicum crop in order to better understand the edge effect observed by Balagawi et al (2014).

Methodology

Comparative efficacy of insecticides, applied as cover sprays, for control of Queensland fruit fly and cucumber fly (Appendix 1)

Semi-field trials were conducted over two years to evaluate the efficacy of eight insecticides against Queensland fruit fly and cucumber fly: clothianidin (assessed at two application rates), thiacloprid, imidacloprid, bifenthrin, alpha-cypermethrin, cyantraniliprole, spinetoram and abamectin. Treatments were compared with the industry standard (dimethoate) and an untreated control. Trials were conducted from January to April in 2014 and 2015 at Gatton Research Facility (Gatton, QLD). Insecticides were applied to plantings of capsicum, for evaluation against Queensland fruit fly, and to zucchini, for evaluation against cucumber fly. There were seven treatments in each trial, replicated four times. Treated fruit were exposed to flies using two methods. In the first, plants were removed from the field, placed in large pots, and transferred into four large field cages (3 m x 3 m base). Each cage contained plants from all seven treatments and represented one replicate. This method was intended to simulate actual use conditions whilst ensuring an even infestation pressure. In the second method, fruit were removed from the plants and exposed to fruit flies in small laboratory cages, in order to assess adult mortality and efficacy of aged residues. All fruit were held for approximately two weeks under controlled conditions and insecticide efficacy determined from the number of pupae recovered.

Comparative efficacy of insecticides against Queensland fruit fly in a fruit dip bioassay (Appendix 2)

Laboratory trials were conducted to evaluate four insecticides against Queensland fruit fly: chlorantraniliprole, flubendiamide, spinetoram and thiacloprid. Treatments were compared with dimethoate and a control (water). The insecticides were applied to capsicum using a fruit dip method, and fruit flies exposed to the dried residues. Four replicates were performed per treatment, each consisting of a cage of 20 male and 20 female fruit flies, exposed to three treated capsicum fruit. Efficacy was assessed in terms of mortality of the adult flies and development of pupae from the treated fruit.

Evaluation of perimeter protein baiting and MAT for control of fruit flies in a commercial chilli crop (Appendix 3)

A trial was performed in a commercial chilli crop at Austchilli Pty Ltd (Bundaberg, QLD) to assess a systems approach, using a combination of perimeter protein baiting and MAT, for management of fruit fly. Protein bait was applied weekly to a tree-line on three sides of the trial block and to a planting of sugarcane on the fourth side. MAT wicks were placed on the perimeter of the block at 20 m intervals and replaced every three months. Austchilli applied cover sprays for management of other pests, but these were soft option chemistries, not expected to affect fruit flies. Assessments were made in a second block, to enable comparison with Austchilli's standard procedures for fruit fly management, which included regular cover sprays with dimethoate, trichlorfon and methomyl. Chillies were sampled at weekly intervals from October to December 2015 and inspected for the presence of fruit fly larvae after a period of incubation. A total of 6966 red fruit (90.9 kg) were sampled from the trial block, and 3048 red fruit (48.8 kg) from the commercial comparison block over this period. Reject dropped fruit was also sampled from between the rows in both blocks, a total of 18.0 kg from the trial block and 17.9 kg from the comparison block. Freshly deposited reject waste fruit was sampled from a waste pile (5.2 kg). Cue-lure monitoring traps were installed at the trial and commercial comparison blocks, in the tree-line and in the crop, and adjacent to the waste pile.

Evaluation of perimeter protein baiting and MAT for control of fruit flies in a small-scale capsicum trial (Appendix 4)

A trial was performed in a research planting of capsicum, at Bundaberg Research Facility (Bundaberg, QLD). The trial

site was a block used for vegetable research trials, with a tree-line on two sides and sorghum planted on a third side. Sequential plantings of capsicums were made in one area of the block for the purpose of sampling fruit, and other fruit fly susceptible crops were present in the block at the time of the trial (capsicums, tomatoes and strawberries). Protein baiting and MAT were applied to the perimeter of the trial block. Fortnightly applications of Success Neo (spinetoram) were made as an additional measure when baiting and MAT alone proved inadequate. Insecticides were applied as necessary for control of other pests, but the majority were not expected to affect fruit flies. An exception was Vertimec (abamectin), applied at intervals to control infestations of mites, which may have had a suppressive effect on fruit flies. Capsicum fruit were sampled at weekly intervals from February to December 2016 and assessed for the presence of fruit fly larvae. Fruit were sampled from three areas at varying distances from the tree-line and assessed separately. A total of 12,995 capsicum fruit (2488 kg) were sampled. Cue-lure monitoring traps were installed in the tree-line and in the capsicum crop, as well as at various locations elsewhere on the research station. From May onwards, reject fruit were also examined for the presence of fruit fly larvae, and from June onwards, additional reject fruit were placed in emergence traps and monitored for the presence of adult flies.

Seasonal fruit fly activity in the Bundaberg region (Appendix 5)

Cue-lure monitoring traps were installed in or adjacent to capsicum and tomato crops at four vegetable farms in the Bundaberg region. Traps were checked every two weeks from July 2014 until March 2017 in order to monitor the seasonal activity of Queensland fruit flies. A series of short-term trials were performed using traps baited with a new cucumber volatile blend lure produced for melon fly (Scentry Biologicals Inc, Montana, USA), to assess the efficacy of the lure for cucumber fly. Trials were performed in commercial pumpkin crops between December 2014 and October 2015.

Activity of fruit flies within the vegetable crop (Appendix 6)

Trials were performed to further examine activity of Queensland fruit fly within vegetable plantings (capsicums and tomatoes). Monitoring traps were installed at two locations at a vegetable farm in the Bundaberg region. At location 1, traps were placed in a tree-line and in adjacent plantings of tomatoes (ca. 65 m from the tree-line) and capsicums (ca. 120 m from the tree-line). At location 2, two trials were performed, with traps placed in the tree-line at either end of a planting of capsicums, and within the capsicum crop at varying distances from 3 m to 254 m from the edge of the crop. In all trials, pairs of traps were deployed, one baited with cue-lure and one with BioTrap Fruit Fly Attractant Gel (BioTrap Pty Ltd).

Outputs

Comparative efficacy of insecticides, applied as cover sprays, for control of Queensland fruit fly and cucumber fly (Appendix 1)

- Clothianidin (Sumitomo Samurai Systemic Insecticide) was the most effective of the eight insecticides assessed, consistently demonstrating efficacy comparable to dimethoate. No Queensland fruit fly pupae developed from fruit treated with clothianidin applied at the higher rate of 40 g/L in a field cage trial, and efficacy against cucumber flies was comparable to dimethoate. Three day aged residues were comparable to dimethoate for both species. Clothianidin was the only insecticide other than dimethoate to significantly affect mortality of adult flies.
- Thiacloprid (Calypso 480 SC Insecticide), imidacloprid (Confidor 200 SC), cyantraniliprole (DuPont Benevia Insecticide) and alpha-cypermethrin (Nufarm Fastac Duo Insecticide) demonstrated efficacy comparable to dimethoate against Queensland fruit fly, but were generally less effective against cucumber fly.
- Bifenthrin (Talstar 250 EC), spinetoram (Success Neo Insecticide) and abamectin (Vertimec Miticide/Insecticide) were all relatively less effective than the other treatments, although all demonstrated a suppressive effect.
- Higher numbers of aphids and silverleaf whitefly were observed in plots treated with alpha-cypermethrin, bifenthrin and dimethoate compared with other treatments.
- Efficacy was generally lower for cucumber fly than for Queensland fruit fly, possibly due to the greater vigour of the cucumber fly and hence higher infestation pressure.

Comparative efficacy of insecticides against Queensland fruit fly in a fruit dip bioassay (Appendix 2)

- Thiacloprid (Calypso 480 SC Insecticide) was the most effective of the four assessed insecticides in terms of number of pupae developing from treated fruit.
- Spinetoram (Success Neo Insecticide) also had a significant effect compared with the control.
- Chlorantraniliprole (DuPont Coragen Insecticide) and flubendiamide (Belt 480 SC Insecticide) had no effect compared to the control.
- At 48 hours post initial exposure of flies to treated fruit, an average of 73% of flies in the spinetoram treatment were knocked down or dead, compared with 96% in dimethoate. Mortality in all other treatments was below 9%.

Evaluation of perimeter protein baiting and MAT for control of fruit flies in a commercial chilli crop (Appendix 3)

- A large-scale trial performed in a commercial chilli crop demonstrated the efficacy of perimeter protein baiting and MAT to control fruit fly in vegetables in the Bundaberg region.
- A total of 6966 red chilli fruit (90.9 kg) were sampled between October and December 2015. No fruit fly larvae were detected, resulting in an upper infestation level (95% confidence) of 0.04%. This compared to seven larvae (from two fruit) detected in a total of 3048 red fruit (48.8 kg) sampled from the commercial comparison block, with an upper infestation level of 0.21% (95% confidence).
- Sampling of reject fruit collected from between the rows (a total of 18.0 kg from the trial block and 17.9 kg from the comparison block) found no larvae in any samples.

- Two empty pupal cases were found in 5.2 kg of freshly deposited reject waste fruit sampled from a waste pile.
- Trap catches in cue-lure traps installed in the trial block and commercial comparison block remained at or below one fly per trap per day for the majority of the trapping period. Trap catches at three other vegetable farms monitored over the same period were generally substantially higher, indicating low pest pressure at Austchilli.
- The highest trap catches were observed in traps placed on the southern tree-line in each block, followed by traps placed a short distance into the crop (20 - 25 m from the southern tree-line). Traps placed further into the crop (160 m or more from the southern tree-line) caught the fewest flies.

Evaluation of perimeter protein baiting and MAT for control of fruit flies in a small-scale capsicum trial (Appendix 4)

- A small-scale trial to assess the efficacy of perimeter protein baiting, MAT and regular cover sprays with Success Neo (spinetoram) in a research planting of capsicums sampled a total of 12,995 red fruit (2488 kg) between February and December 2016. No fruit fly larvae were detected in cooler months, between 18th May and 5th September, when fruit fly pressure was low. However, treatments were not sufficient to prevent fruit fly damage under conditions of high fruit fly pressure during the summer and autumn.
- Background fruit fly pressure was much higher compared to the commercial chilli crop, peaking at 3.6 flies per trap per day in February in citrus blocks adjacent to the trial block. Higher pressure coupled with the small-scale nature of the trial may have contributed to the lack of suppression during summer and autumn.
- Large numbers of *Atherigona* sp. were observed in rotten fruit. The larvae of these flies can easily be mistaken for fruit fly larvae, but *Atherigona* sp. does not attack healthy fruit. It therefore is important that growers are able to distinguish between the larvae of these two species so that cover sprays are not applied unnecessarily.

Seasonal fruit fly activity in the Bundaberg region (Appendix 5)

- The majority of flies caught in cue-lure traps were Queensland fruit fly or lesser Queensland fruit fly.
- The highest trap catches generally occurred between September and November, with a second smaller peak in January/February, and low trap catch over the winter period. A broadly similar pattern was observed in the first two years of monitoring, however in the third year trap catch was also high in December.
- Trap catches differed between farms and location within the farm. More flies were caught in traps located along a tree-line adjacent to native vegetation compared with traps located within the crop or on a fence-line.
- Trials performed in cucurbit crops in the Bundaberg region, using a new cucumber volatile blend lure, found that Bugs for Bugs traps baited with the lure caught an average of 0.7 cucumber flies per trap per day, compared with an average of 9.1 cucumber flies per trap per day caught by BioTraps. The average sex ratio was 74:26 female:male.
- Two further trials with the cucumber volatile lure were performed in commercial plantings of pumpkins in Kalbar (SE QLD). The aim was to further compare the two trap types and to compare trap catches in different locations in the blocks as the crop matured. However, despite the presence of cucumber flies in the field, only a single fly was trapped in each trial. Based on these results, and variable results obtained using the lure in project MT12050 (Farm-wide fruit fly management systems for the east coast of Australia), trapping with the cucumber volatile blend lure was discontinued.

Activity of fruit flies within the vegetable crop (Appendix 6)

- Traps located along a tree-line, or in a crop close to the tree-line, generally caught more flies than traps placed in the crop further from the tree-line. However, results from one of the three trials performed indicated a change in this behaviour as the crop matured, with more flies trapped within the crop once fruit were present.
- Traps baited with BioTrap Fruit Fly Attractant Gel generally caught few fruit flies. They were observed to trap a mix of male and female flies, of a variety of species.

Communications and publications

- Regular communication has been maintained with Bundaberg Fruit and Vegetable Growers (BFVG) throughout the project.
- Project results were presented to industry at intervals:
    - Workshop and growers' forum held in collaboration with BFVG on 21st November 2014. There were approximately 40 attendees: growers (32%), agronomists/advisors (25%), industry representatives (18%) and others such as industry stakeholders and researchers.
    - Horticulture Innovation Australia Ltd fruit fly roadshow, held in Bundaberg on 9th December 2015. There were approximately 50 attendees, a mix of growers, agronomists, resellers and researchers.
    - Gatton stakeholder event, held at Gatton Research Facility on 22nd March 2016. Attended by local growers.
    - National Horticultural and Innovation Expo, organised by the Lockyer Valley Growers Inc, held at Gatton Research Facility on 27th July 2016. Estimated attendance over the two day event was 1500.

Horticulture Innovation Australia Ltd fruit fly roadshow, Bundaberg

National Horticultural and Innovation Expo, Gatton

National Horticultural and Innovation Expo, Gatton

Outcomes

Several insecticides with efficacy against fruit fly (*B. tryoni* and *Z. cucumis*) were identified from the ten evaluated in a series of field and laboratory trials. One of these, clothianidin (Samurai), is now permitted for use in fruiting vegetable crops (APVMA permits PER80100 and PER80101), although its usefulness is limited by a seven day withholding period. Cyantraniliprole (Benevia) demonstrated good efficacy against Queensland fruit fly. This insecticide is currently registered for use in fruiting vegetable crops for control of pests other than fruit fly, with a one day withholding period. Its use for control of these pests is likely to have a significant impact on fruit fly within the crop. Other insecticides commonly used for control of pests other than fruit fly in vegetable crops were demonstrated to have a suppressive effect on fruit fly, namely spinetoram (Success Neo), abamectin (Vertimec), imidacloprid (Confidor) and bifenthrin (Talstar). These insecticides could be used in conjunction with other control methods, such as protein baiting and MAT, to reduce fruit fly damage to below economic thresholds.

A successful demonstration of the efficacy of perimeter protein baiting and MAT was achieved in a commercial chilli crop in the Bundaberg region. Infestation of fruit and monitoring trap catches were comparable to a commercial comparison block, where fruit fly management included weekly broad spectrum cover sprays (dimethoate, trichlorfon and methomyl). The research failed to demonstrate efficacy when protein baiting, MAT and Success Neo cover sprays were applied on a smaller scale, under high local fruit fly pressure. However, successful control was achieved during winter, when fruit fly pressure was low. Results therefore suggest that a combination of perimeter baiting, MAT and Success Neo cover sprays may be a viable management option during a winter window. In contrast, the success of the large scale field trial shows that the size of the area over which protein baiting and MAT are applied is most likely a critical factor for successful control.

No fruit fly larvae were found in reject fruit sampled from the commercial trial site or the research facility trial site. However, larvae of *Atherigona* sp. were commonly observed in reject fruit, and these larvae can easily be mistaken for those of fruit fly. This highlights the importance of distinguishing between the larvae of these two species so that cover sprays are not applied unnecessarily.

Monitoring of fruit fly activity in the Bundaberg region over three successive seasons confirmed a population peak in September/October, and suggested a second, smaller peak in January/February. Growers can utilise these data to help plan fruit fly management strategies.

Results from trapping and fruit sampling demonstrated an edge effect in fruiting vegetable crops. This needs further research to confirm results in terms of infestation of fruit within the field, and if validated could be incorporated into growers' fruit fly management practices, such as targeting cover sprays to the crop edges rather than the centre of the crop. The findings also confirmed that application of protein bait to vegetation on the border of the crop, rather than to the crop itself, is likely to be an effective strategy for control of fruit flies in vegetable crops.

Evaluation and discussion

This project has successfully identified a number of management options for fruit fly in fruiting vegetable crops, as alternatives to dimethoate cover sprays. Currently the vegetable industry is highly reliant on dimethoate for management of fruit flies, and this has hindered the adoption of IPM for other serious vegetable pests, such as silverleaf whitefly. Restrictions to the use of dimethoate have made fruit fly control in certain crops much more difficult, threatening production. Very few effective alternative insecticides are registered for fruit fly in fruiting vegetable crops, and published data on insecticide efficacy against Queensland fruit fly and cucumber fly is scarce. Although the combination of protein baiting and MAT has been used successfully to manage Queensland fruit fly in tree crops, there is insufficient knowledge regarding its efficacy in low growing crops such as fruiting vegetables. There is also limited understanding regarding the behaviour of fruit flies in these types of crops.

A number of insecticides currently commonly used in vegetable crops demonstrated moderate to good efficacy against Queensland fruit fly and cucumber fly, namely spinetoram (Success Neo), abamectin (Vertimec), imidacloprid (Confidor), bifenthrin (Talstar) and cyantraniliprole (Benevia). Cyantraniliprole was particularly effective against Queensland fruit fly, with efficacy similar to dimethoate in semi-field trials. Although not registered for control of fruit fly, the use of these insecticides for management of other vegetable pests is likely to have at least a suppressive effect on fruit fly. Two neonicotinoid insecticides, not currently registered for use in vegetable crops, had excellent efficacy against Queensland fruit fly and cucumber fly: clothianidin (Samurai) and thiacloprid (Calypso). Although not registered, clothianidin is now permitted for control of fruit fly in vegetable crops (PER80100 and PER80101, expire 30 September 2018), although its effectiveness is limited by a seven day withholding period. Spinetoram (Dow AgroSciences, n.d.) and cyantraniliprole (DuPont, 2015) are claimed by the manufacturers to have limited impact on beneficial insects and to be compatible with IPM. Likewise, clothianidin is claimed to have no effects on predatory mites, and a moderate effect on beneficials generally (Sumitomo Chemical, n.d.). Recent literature has revealed that many newer pesticides, such as cyantraniliprole and spinetoram, may not be as selective as claimed, due to sublethal effects (Mills et al, 2016; Amarasekare et al, 2016). However, they are still likely to have less impact on natural enemies than current chemical options: examining the impact of pesticides on a range of beneficials, Maas and Redfern (2016) ranked cyantraniliprole, abamectin and clothianidin as less harmful than either dimethoate or methomyl. The research also represents the first successful trial of the efficacy of insecticides, applied to a vegetable crop as cover sprays, against Queensland fruit fly and cucumber fly. The method developed during the research allowed for the comparison of several insecticides under semi-realistic conditions: insecticides were applied to plants and the residues aged under field conditions; fruit flies were exposed to entire plants bearing fruit; and fruit flies were able to choose where to land and oviposit. This allowed for evaluation of effects other than mortality, such as repellent effects resulting in reduced oviposition, which were not apparent in laboratory bioassays.

A combination of perimeter protein baiting and MAT achieved successful control of fruit fly in a commercial chilli crop in the Bundaberg region, comparable to that achieved through weekly cover sprays with dimethoate, trichlorfon and methomyl. However, the same combination of baiting and MAT was not successful when applied on a smaller scale in an experimental capsicum crop, even when regular cover sprays with Success Neo were included as an additional control measure. This was likely due in part to the differing fruit fly pressures at the two trial sites. Monitoring showed that the local fruit fly population at the commercial site was low throughout the trial, even during the spring and early summer, when fruit fly populations in the Bundaberg area are typically at their highest. The low local population at the commercial site was likely due to the existing farm-wide fruit fly management practices, which included regular cover sprays with broad spectrum insecticides as well as protein bait spraying and MAT. The trial results suggest that if a low local fruit fly population can be achieved, the combination of protein baiting and MAT is sufficient to prevent infestation. In contrast, the local fruit fly population at the research facility trial site was very high, making control

much more difficult to achieve; the trial site was adjacent to large blocks of citrus where fruit flies were not controlled effectively. Nevertheless, good control was achieved during a period between mid-May and the beginning of September, when temperatures were low and fruit fly activity was reduced. Results from this project demonstrate that if protein baiting and MAT is to be employed in vegetable crops, efforts must be made to maintain low populations in the locality. During periods of high pressure further control methods may also be required, such as cover sprays (preferably with the more IPM-compatible insecticides), as well as pack-house inspections and culling.

Monitoring in the Bundaberg region over three successive seasons showed a consistent peak in fruit fly populations in the spring/early summer, and generally lower trap catches over the winter. This activity pattern was reasonably consistent between locations and between years, although comparatively high trap catches were observed in December in the third year of monitoring. Previous trapping work carried out as part of Hort Innovation project BS09022 (Missenden, 2014) also indicated low populations during the winter, followed by an increase in trap catch in August/September, although trapping did not continue beyond this date.

Monitoring at several locations in the Bundaberg region also demonstrated an edge effect, with more male flies caught in cue-lure traps placed in a tree-line, or a short distance into the crop, compared with trap catch from traps placed further into the crop. This is further evidence that perimeter protein baiting, rather than application of the protein bait to the crop itself, is likely to be an effective strategy for fruit fly management in fruiting vegetable crops. Sampling of fruit from the research trial also indicated that infestation was higher in fruit close to the tree-line. A similar finding was reported in Hort Innovation project BS06002, Alternative fruit fly treatment for interstate market access for strawberries (Gu, 2010), with higher trap catches from traps placed near the border of the block, and higher infestation recorded from fruit sampled from the border compared to 80-100 m from the border. Unfortunately trapping using BioTrap Fruit Fly Attractant Gel (BioTrap Pty Ltd), which also attracts female flies, did not catch sufficient fruit flies to allow comparison between traps on the tree-line and in the crop.

Trapping of cucumber flies using a new cucumber volatile lure produced variable results. A trial conducted in the Bundaberg region to compare two commercial trap types was successful, demonstrating a clear difference between the traps. However, subsequent trials performed in Kalbar resulted in very poor trap catches, despite large numbers of cucumber flies observed at the trap site. Similar mixed results were also obtained using this lure in Hort Innovation project MT12050 (DeFaveri, 2013), in which significant trap catches occurred at only one of the monitoring sites, with no or very few cucumber flies trapped at all other sites. Further research is required with this lure before it can be recommended as a monitoring tool.

Recommendations

The following are recommendations for growers to improve management of fruit fly in fruiting vegetable crops:

- Perimeter protein baiting and MAT can be effective during periods of low fruit fly pressure, such as during the winter or if effective farm-wide management of fruit flies can be achieved, but are not sufficient when fruit fly populations are high.
- A number of insecticides commonly used for control of pests other than fruit fly can help to suppress fruit fly populations. These include some products which are less damaging to natural enemies than dimethoate, the current chemical option. Despite the manufacturers' claims of IPM compatibility, products such as Benevia (cyantraniliprole) and Success Neo (spinetoram) do have side effects on some non-target organisms, including bees. They should therefore be used with caution and not applied while bees are actively foraging in the crop, as per label instructions.
- Based on trapping and fruit sampling it is recommended that particular attention should be paid to crops near the border of a block when applying cover sprays, particularly those near a tree-line, as results indicate that these fruit are more likely to be infested than those in the centre of the block.
- Growers should pay particular attention to fruit fly management when populations increase in September.
- Larvae of *Atherigona* sp. can easily be confused with those of fruit fly, but do not attack healthy fruit. Correct identification of larvae can avoid unnecessary pesticide applications.

The following are recommendations for further research to develop in-field treatments for fruit fly in vegetable crops:

- Large scale field trials with cyantraniliprole should be conducted to generate data to allow its use, through permit or registration, for management of fruit fly in vegetable crops. This would give growers an alternative to the current limited options, with less impact on natural enemies.
- Further large-scale trials in commercial vegetable crops are required to validate the efficacy and cost-benefit of perimeter protein baiting and MAT during the winter window, and to improve the use of these techniques under conditions of high fruit fly pressure.
- Further trials are required to better understand the behaviour of Queensland fruit flies within a vegetable crop, to include: monitoring using a protein lure; monitoring using a female bias lure; sampling of fruit from different locations within the field to assess infestation. This would, firstly, confirm the optimum location for application of protein bait. Secondly, if trials confirm that fruit located some distance from a tree-line is less susceptible to infestation, this would allow growers to target cover sprays more effectively.
- Research is required to improve the use of the cucumber volatile lure as a monitoring tool for cucumber fly.

Scientific refereed publications

Journal article

Senior, L.J., Missenden, B.P., Wright, C., 2017. Comparative efficacy of insecticides on *Bactrocera tryoni* and *Zeugodacus cucumis* (Diptera: Tephritidae) in laboratory and semifield trials in fruiting vegetables. *Journal of Economic Entomology*, doi: 10.1093/jee/tox109.

Intellectual property/commercialisation

No commercial IP generated.

References

Amarasekare, K.G., Shearer, P.W. & Mills, N.J. 2016. Testing the selectivity of pesticide effects on natural enemies in laboratory bioassays. *Biological Control* **102**, 7-16.

APVMA 2011. Use of the insecticide dimethoate suspended in many food crops. (http://apvma.gov.au/node/11771)

APVMA 2015. Fenthion chemical review. (http://apvma.gov.au/node/12541)

APVMA 2017. Dimethoate chemical review. (http://apvma.gov.au/dimethoate)

Balagawi, S., Jackson, K. & Clarke, A.R. 2014. Resting sites, edge effects and dispersion of a polyphagous *Bactrocera* fruit fly within crops of different architecture. *Journal of Applied Entomology* **138**, 510-518.

DeFaveri, S. 2013. Farm-wide fruit fly management systems for the east coast of Australia. *HAL project MT12050*. Horticulture Innovation Australia.

Dow AgroSciences n.d. Success™Neo Insecticide Vegetable technical manual. (http://www.dowagro.com/webapps/lit/litorder.asp?filepath=au/pdfs/noreg/012-10782.pdf&pdf=true)

DuPont 2015. Benevia® brochure. http://www.dupont.com.au/content/dam/dupont/products-and-services/crop-protection/documents/en_au/15_1350_AGRIMC_DuPont%20Au%20Benevia%208PP_EDIT_PROOF2%20(1).pdf

Fay, H., Lloyd, A., Gu, H., Hamacek, E., Missenden, B. & Peek, P. 2011. Area wide management of fruit fly – Central Burnett Phase 2. *HAL project CT06046*. Horticulture Australia Limited.

Gu, H. 2010. Alternative fruit fly treatment for interstate market access for strawberries. *HAL project BS06002*. Horticulture Australia Limited.

Lloyd, A., Hamacek, E., Peek, T., Kopittke, R., Wyatt, P., Neale, C., Eelkema, M. & Smith, D. 2007. Area wide management of fruit fly – Central Burnett. *HAL project AH03002*. Horticulture Australia Limited.

Lloyd, A. C., Hamacek, E. L., Kopittke, R. A., Peek, T., Wyatt, P. M., Neale, C. J., Eelkema, M. & Gu, H. N. 2010. Area-wide management of fruit flies (Diptera: Tephritidae) in the Central Burnett district of Queensland, Australia. *Crop Protection* **29**, 462-469.

Maas, S. & Redfern, R. 2016. Cotton pest management guide 2016-17. Cotton Research and Development Corporation. CottonInfo. (http://www.cottoninfo.com.au/sites/default/files/documents/CPMG%202016%20PrintVersion.pdf)

Mills, N.J., Beers, E.H., Shearer, P.W., Unruh, T.R. & Amarasekare, K.G. 2016. Comparative analysis of pesticide effects on natural enemies in western orchards: a synthesis of laboratory bioassay data. *Biological Control* **102**, 17-25

Missenden, B. 2014. Alternative fruit fly treatment for interstate market access for strawberries: winter window option. *HAL project BS09022*. Horticulture Australia Limited.

Nishida, T. & Bess, H.A. 1957. Studies on the ecology and control of the melon fly *Dacus (Strumeta) cucurbitae* Coquillett (Diptera: Tephritidae). *Technical Bulletin Hawaii Agriculture Experiment Station* **34**, 44 pp.

Oag, D., Gu, H. & Missenden, B. 2010. Domestic market access for table grapes. *HAL project TG08001*. Horticulture

Australia Limited.

Prokopy, R.J., Miller, N.W., Piñero, J.C., Barry, J.D., Tran, L.C., Oride, L. & Vargas, R.I. 2003. Effectiveness of GF-120 fruit fly bait spray applied to border area plants for control of melon flies (Diptera: Tephritidae). *Journal of Economic Entomology* **96**, 1485-1493.

Royer, J.E., De Faveri, S.G., Lowe, G.E. & Wright, C.L. 2014. Cucumber volatile blend, a promising female-biased lure for *Bactrocera cucumis* (French 1907) (Diptera: Tephritidae: Dacinae), a pest fruit fly that does not respond to male attractants. *Austral Entomology* **53**, 347-352.

Senior, L. & Wright, C. 2012. Efficacy data to support fruit fly control systems for export trade in tomatoes and peppers. Department of Agriculture, Fisheries and Forestry, Queensland, Australia.

Subramaniam, S., Jackson, K. Lloyd, A., Kopittke, R., Hamacek, E. & Kreymborg, D. 2011. Alternative fruit fly control and market access for capsicums and tomatoes. *HAL Project VG06028*. Horticulture Australia Limited.

Sumitomo Chemical n.d. Sumitomo Samurai Systemic Insecticide technical manual. (http://www.sumitomo-chem.com.au/sites/default/files/literature/samurai_manual_blank.pdf)

Acknowledgements

The following QDAF staff members were involved in this project:

- Contributing authors: Lara Senior, Carole Wright, Brendan Missenden, Pauline Wyatt
- Bundaberg project team: Tim Behrens, Leslie Lowe and Graeme Pomroy
- Brisbane project team: Thelma Peek, Chloe Jacobs, Linda Clarke, Ania Ganowicz, Alexandra Gardiner, Kellie Bilney, Marianne Eelkema, Jaye Newman
- Gatton project team: John Duff, Mary Firrell
- Jane Royer assisted with identification of fruit fly samples
- Bundaberg Research Facility farm staff maintained the 2016 perimeter baiting plus MAT evaluation trial site
- Gatton Research Facility farm staff maintained the insecticide trial sites

We also thank:

- Austchilli Pty Ltd for allowing us to perform the 2015 perimeter baiting plus MAT evaluation trial in a commercial chilli block, maintaining the trial site and applying protein bait.
- Stephen Hawkey, Josh Crow, Barbera Family Farms and Dennis Uccisella for allowing monitoring on their properties.
- DuPont Crop Protection Australia and Sumitomo Chemical Australia Pty Ltd for providing insecticide samples.
- Bundaberg Fruit and Vegetable Growers for providing advice and liaison with growers.

Appendices

Appendix 1 Comparative efficacy of insecticides, applied as cover sprays, for control of Queensland fruit fly and cucumber fly

Appendix 2 Comparative efficacy of insecticides against Queensland fruit fly in a fruit dip bioassay

Appendix 3 Evaluation of perimeter protein baiting and MAT for control of fruit flies in a commercial chilli crop

Appendix 4 Evaluation of perimeter protein baiting and MAT for control of fruit flies in a small-scale capsicum trial

Appendix 5 Seasonal fruit fly activity in the Bundaberg region

Appendix 6 Activity of fruit flies within the vegetable crop

Appendix 7 Paper accepted for publication in Journal of Economic Entomology: Comparative efficacy of insecticides on *Bactrocera tryoni* and *Zeugodacus cucumis* (Diptera: Tephritidae) in laboratory and semifield trials in fruiting vegetables, by Lara Senior, Brendan Missenden and Carole Wright.

Appendix 1 Comparative efficacy of insecticides, applied as cover sprays, for control of Queensland fruit fly and cucumber fly

Semi-field trials were conducted to evaluate the efficacy of eight insecticides against Queensland fruit fly and cucumber fly (Table 1). Treatments were compared with the industry standard (dimethoate) and an untreated control, and were chosen based on a literature search and through consultation with a range of industry representatives (growers, agricultural chemical suppliers, Bundaberg Fruit and Vegetable Growers and Hortus Technical Services). The trials have been written up as a scientific paper and accepted for publication in the Journal of Economic Entomology. A copy of the paper is attached (Appendix 7).

Table 1 Treatments and application rates

Product	**Active ingredient**	**Product application rate**
Sumitomo Samurai Systemic Insecticide	clothianidin	40 g/100 L; 30 g/100 L
Calypso 480 SC Insecticide	thiacloprid	37.5 ml/100 L
Confidor 200 SC	imidacloprid	25 ml/100 L
Talstar 200 EC Insecticide/Miticide	bifenthrin	24 ml/100 L
Fastac Duo Insecticide	alpha-cypermethrin	55 ml/100 L
DuPont Benevia Insecticide	cyantraniliprole	100 ml/100 L
Success Neo Insecticide	spinetoram	40 ml/100 L
Vertimec Miticide/Insecticide	abamectin	60 ml/100 L
Dimethoate (industry standard)	dimethoate	75 ml/100 L

Trials were conducted over two years (January to April 2014 and January to April 2015) at Gatton Research Station (Gatton, QLD). Insecticides were applied to field grown capsicums and zucchinis in a small plot trial (Plate 1). Plants bearing fruit were then dug up and placed in large field cages, with plants from all treatments placed in a cage (Plates 2 and 3). Fruit flies were then released into the cages. This method ensured treatments were exposed to equal pest pressure. Additionally, fruit were removed from the plants in the trial plots at one and three days after treatment application and exposed to fruit flies in laboratory cages (Plates 4 and 5). This allowed assessments of adult mortality and efficacy of aged residues.

Clothianidin (Samurai) was very effective against Queensland fruit fly and cucumber fly, both in terms of preventing pupae developing from treated fruit and mortality of adult flies. Thiacloprid (Calypso), imidacloprid (Confidor), cyantraniliprole (Benevia) and alpha-cypermethrin (Fastac Duo) were also very effective against Queensland fruit fly but less effective against cucumber fly. Bifenthrin (Talstar), spinetoram (Success Neo) and abamectin (Vertimec) were all relatively less effective than other treatments, although all demonstrated a suppressive effect. Detailed results are provided in the attached paper (Appendix 7).

Plate 1 Field trial layout

Plate 2 Treated fruit exposed to fruit flies within field cages

Plate 3 Treated plants within field cage

Plate 4 Laboratory cages

Plate 5 Treated fruit in laboratory cage

Appendix 2 Comparative efficacy of insecticides against Queensland fruit fly in a fruit dip bioassay

A laboratory trial was performed to assess insecticides, applied as a dip, for efficacy against Queensland fruit fly. Five insecticides were assessed: a commercial in confidence product at two application rates; chlorantraniliprole (Coragen); flubendiamide (Belt 480 SC); spinetoram (Success Neo) and thiacloprid (Calypso 480 SC). However, at the request of the manufacturer the results for the commercial in confidence product are not reported here. These treatments were compared with dimethoate (the industry standard) and a control (water). All were applied at the label specified rate (Table 1). No surfactants were used. Chlorantraniliprole, flubendiamide and spinetoram are currently commonly used in vegetable crops for control of pests other than fruit fly. Thiacloprid is not registered for use in vegetable crops, however it has demonstrated efficacy against Queensland fruit fly in stone fruit (Reynolds et al, 2014) and Mediterranean fruit fly in stone fruit and pome fruit (Rahman & Broughton, 2016). The trial was performed in November 2014.

Table 1 Treatments and application rates

Product	**Active ingredient**	**Product application rate**
DuPont Coragen Insecticide	chlorantraniliprole	10 ml/100 L
Belt 480 SC Insecticide	flubendiamide	10 ml/100 L
Success Neo Insecticide	spinetoram	40 ml/100 L
Calypso 480 SC Insecticide	thiacloprid	37.5 ml/100 L
Dimethoate (industry standard)	dimethoate	75 ml/100 L

Capsicum fruit were obtained from an organic supplier. Three fruit were placed in a net bag and submerged in the insecticide solution for one minute (Plate 1). Dipped fruit were allowed to dry outside in the shade for 30-60 minutes, and then returned to the laboratory. The three fruit per treatment replicate were placed in wire frame, net cages (21 cm x 33 cm base, 21 cm height) containing 40 Queensland fruit flies (20 male, 20 female). Flies were obtained from the colony maintained in the DAF Market Access laboratories in Brisbane (detailed in Appendix 1), were 14 days post emergence, and had been protein fed. Sugar and water were provided for the duration of the trial. Cages were held in a controlled environment room (26°C, 70% relative humidity) with natural and artificial lighting (Plate 2). Four replicates were performed for each of the treatments, with treatments assigned to cages using a randomised complete block design.

Assessments of the number of knocked down and dead flies were made at ten minute intervals for one hour following placement of treated fruit into the cages. The fruit were then removed from the cages. Further assessments of knocked down and dead flies occurred up to 48 hours post introduction of the treated fruit. Fruit removed from the cages were placed on drip trays (shallow plastic containers covered with net to allow drainage) in ventilated containers, with each replicate held separately (Plate 3). A layer of vermiculite on the base of the container was provided as a substrate for pupation. The fruit were held at 26°C, 70% relative humidity, for approximately two weeks, after which the vermiculite was sieved and pupae counted.

The number of knocked down and dead flies were expressed as a proportion of the total number of flies per cage. A repeated measures residual maximum likelihood (REML) analysis was used to investigate trends over time. The most

appropriate correlation model was the identity model, which assumes no correlation between the time points. The proportion of knocked down and dead flies at 48 hours was also analysed separately using a generalised linear mixed model (GLMM) assuming a binomial distribution and a logit link function. The assumptions underlying the model were checked using various diagnostics, such as investigating the residuals and the dispersion parameter, and no violations were identified. Pupal counts were analysed using a GLMM assuming a Poisson distribution and a log link function. The effect of replicate was fitted as a random term in all analyses. Where a significant difference was found pairwise comparisons were made using the 95% least significant difference (LSD). Means with an over-inflated standard error were not included in the pairwise comparisons. The commercial in confidence product results were included in all analyses but results are not reported here.

Plate 1 Dipping capsicums in insecticide

Plate 3 Treated fruit on drip trays

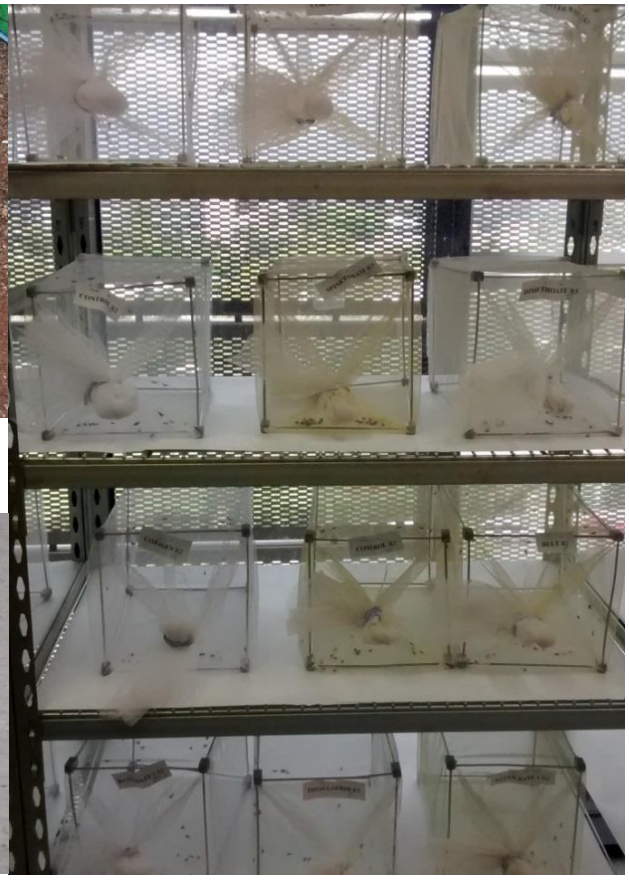

Plate 2 Flies held in cages following removal of treated fruit

Results

Treatment had a significant effect on the number of pupae developing in the fruit (P=0.016) (Figure 1). The standard treatment, dimethoate, was most effective, with no pupae developing in any replicates. Thiacloprid was also very effective, with an average 2.25 pupae per cage. The spinetoram treatment also resulted in significantly fewer pupae than the control (P<0.05). Chlorantraniliprole and flubendiamide had no effect on number of pupae.

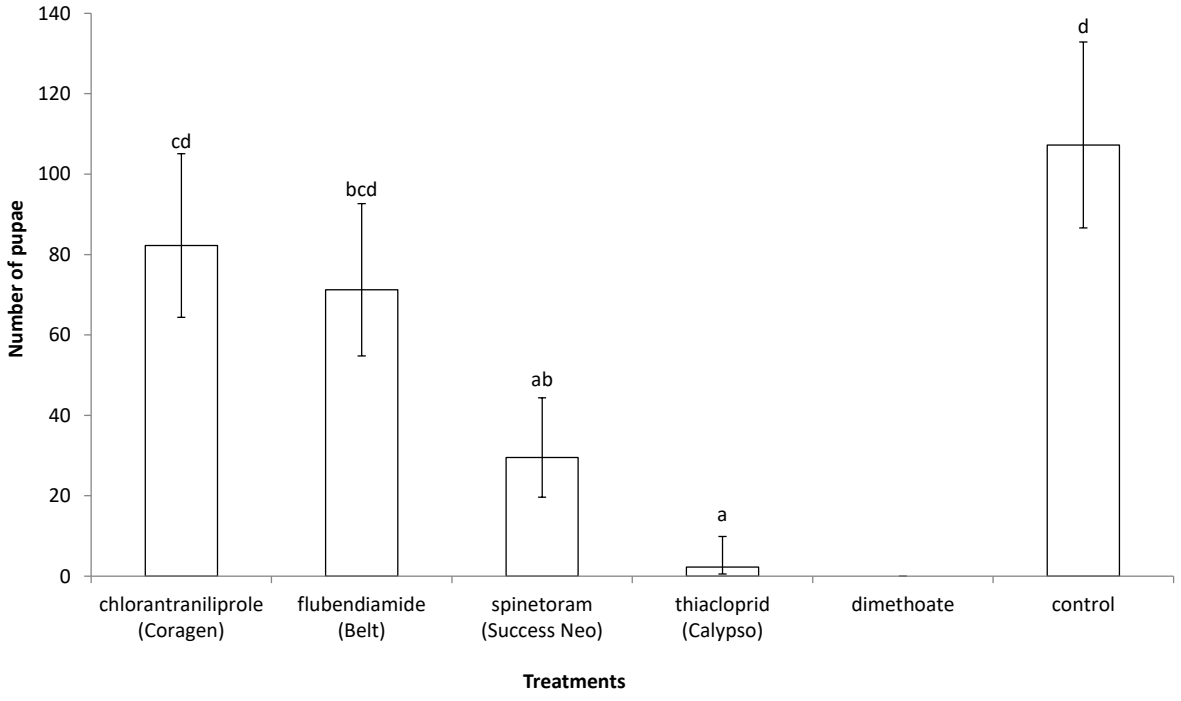

Figure 1 Number of pupae developing in treated capsicum fruit (back transformed means ± 1 standard error). Means with a letter in common are not significantly different (P>0.05). Dimethoate was excluded from the pairwise comparisons due to an over-inflated standard error.

REML analysis combining assessments over time found a significant effect of treatment on knockdown and mortality of adult flies (P<0.001). The overall mean proportion of affected flies was significantly higher in dimethoate and spinetoram compared to all other treatments (P<0.05). No other treatments had a significant effect. Analysis of the 48 hour mortality data confirmed this result (Table 2). However, knockdown and mortality in the spinetoram treatment occurred slowly compared with dimethoate (Figure 2).

Table 2 Results from a GLMM of the proportion of flies observed knocked down or dead at the 48 hour assessment. Means with a letter in common are not significantly different (P>0.05).

Treatment	**Proportion of knocked down or dead flies at 48 hours**	
	Mean (± SE)	**Back transformed mean**
chlorantraniliprole (Coragen)	-3.421 (±0.527) a	0.03
flubendiamide (Belt)	-2.820 (±0.398) ab	0.06
spinetoram (Success Neo)	1.016 (±0.209) c	0.73
thiacloprid (Calypso)	-2.338 (±0.324) ab	0.09
dimethoate	3.252 (±0.482) d	0.96
control	-2.813 (±0.398) ab	0.06
P<0.001		

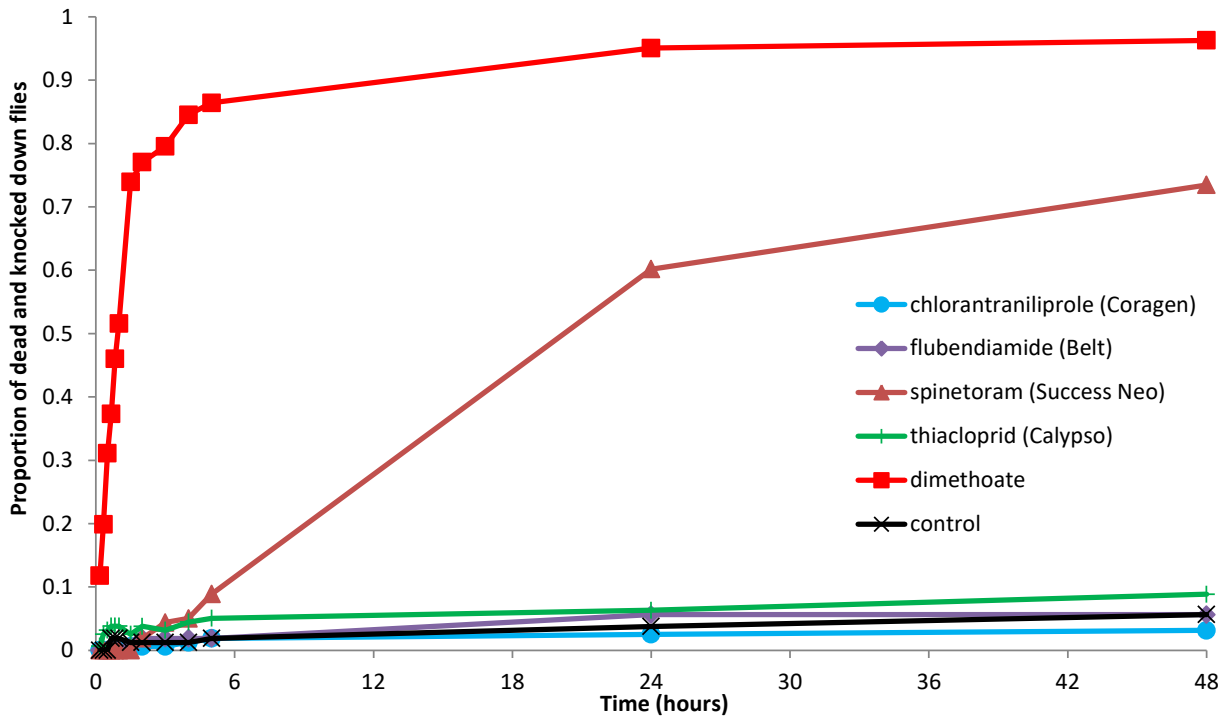

Figure 2 Mean affected (knocked down or dead) adult *B. tryoni* exposed to dried residues of insecticide on capsicum, expressed as a proportion of the total flies per cage. Average standard error (REML) = 0.014.

It can be concluded that thiacloprid demonstrated efficacy comparable with dimethoate in terms of preventing pupae from developing from treated fruit. However, this insecticide did not affect adult flies. Spinetoram had a suppressive effect on fruit infestation, and also affected adult flies, although the rate of knockdown was much slower than for dimethoate. Following this laboratory trial, thiacloprid and spinetoram were assessed further in the second season of semi-field trials (Appendix 1).

References

Rahman, T. & Broughton, S. 2016. Evaluation of thiacloprid and clothianidin (neonicotinoids) as alternative to fenthion (organophosphate) for control of Mediterranean fruit fly (Diptera: Tephritidae) in deciduous fruit orchards. *Crop Protection* **90**, 170-176.

Reynolds, O.L., Jessup, A., Osborne, T., Bodnaruk, K., Archer, J., Eamens, K. & Barchia, I. 2014. Efficacy of potential controls for Queensland fruit fly management in summerfruit. *HAL Project SF12012*. Horticulture Australia Limited.

Appendix 3 Evaluation of perimeter protein baiting and MAT for control of fruit flies in a commercial chilli crop

A trial was performed in a commercial chilli crop at Austchilli Pty Ltd (Goodwood Rd, Bundaberg, QLD) to assess a systems approach for management of fruit fly in a fruiting vegetable crop, consisting of a combination of perimeter protein baiting and male annihilation technique (MAT). This combination of protein bait spraying and MAT, coupled with field hygiene and fruit inspection, was developed for management of fruit fly in tree crops (Lloyd et al, 2007; Lloyd et al, 2010; Fay et al, 2011). This has become a model approach and was further developed in later work in table grapes (Oag et al, 2010). More recently Missenden (2014) assessed management of fruit fly in strawberries based on a winter window but also incorporating bait sprays applied to fruit fly resting sites on the perimeter of strawberry blocks. Perimeter baiting, where protein bait is applied to vegetation surrounding a crop, was developed for control of melon fly, *Zeugodacus cucurbitae*, in curcurbits in Hawaii (Prokopy et al, 2003). This species has been shown to roost in vegetation on the field margin, only entering the crop to oviposit (Nishida and Bess, 1957). There is evidence that Queensland fruit fly, *Bactrocera tryoni*, may exhibit a similar behaviour in low growing crops (Balagawi et al, 2014; Gu, 2010). Aspects of perimeter bait application in vegetable crops were also explored by DeFaveri (2013), but the efficacy of this technique to control fruit flies in vegetable crops in Australia is currently unproven.

The test system, based on perimeter protein baiting and MAT, was applied in a trial block (ca. 5 ha), where the first sequential planting of chillies was made in mid-June 2015 (Plate 1). Protein bait was prepared at recommended rates as follows: 2 L Fruit Fly Lure (Bugs for Bugs Pty Ltd, Mundubbera, QLD) mixed with 435 ml Hy-Mal (maldison) and 100 L water, plus 1 L prepared Fruit Fly Lure Thickener (xanthan powder)/water mix. MAT wicks were obtained from Bugs for Bugs Pty Ltd. Fruit fly management at the trial site followed Austchilli's standard procedures, with some modifications to maximise efficacy: protein bait was applied to vegetation at a height of approximately 1.5 m; where natural vegetation was not present bait was applied to a border crop planted prior to the start of the trial; MAT wicks were installed on the crop perimeter at an increased density (20 m spacing). MAT wicks were installed on 8th July and replaced every three months. The protein bait was applied weekly to a tree-line on three sides of the trial block, and to a planting of sugarcane on the fourth side (Plate 2). However, the sugarcane was harvested approximately two months into the trial (17th September) and Austchilli elected not to plant a replacement border, hence baiting was not made around the full perimeter of the crop following this date. Harvest of the sugarcane also resulted in removal of the MAT wicks from one side of the trial block for a one month period. The wicks were removed at the request of Austchilli in order to avoid interference with the harvesting machinery, and delays in harvesting extended the period during which the wicks were not in place. Insecticide cover sprays were applied to the trial crop for control of other pests, but no broad spectrum insecticides were used, with the exception of Lorsban (chlorpyrifos) applied at the seedling stage for control of soil pests, and two applications of Lannate (methomyl) to young plants, the last occurring approximately one month prior to the appearance of the first fruit and approximately three months prior to the start of fruit sampling (Table 1). Assessments were also made in a second block of similar size to enable comparison with Austchilli's standard procedures for fruit fly management, which included regular cover sprays with dimethoate, trichlorfon and methomyl (Table 1).

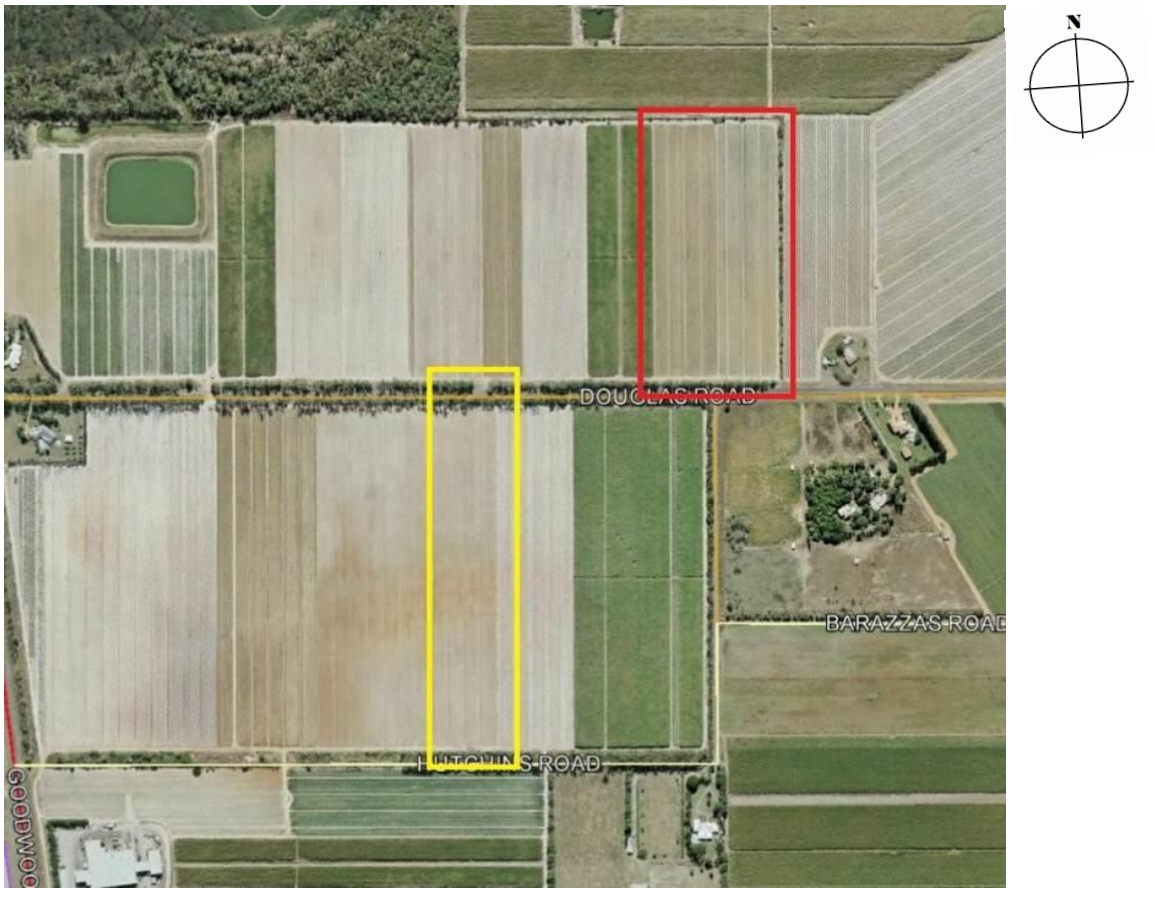

Plate 1 Trial block (outlined in red) and commercial comparison block (outlined in yellow) at Austchilli Pty Ltd.

Plate 2 Tree-line on perimeter of trial block (left) and sugarcane adjacent to crop (right)

Table 1 Insecticides applied to the trial block and commercial comparison block. Products with efficacy against fruit fly in bold.

Date	Trial block	Commercial comparison block
1/7/15	**Lorsban (chlorpyrifos)**	**Lorsban (chlorpyrifos)**
6/7/15	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)
13/7/15	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)
20/7/15	Microthiol (sulphur) **Lannate (methomyl)**	Microthiol (sulphur) **Lannate (methomyl)**
27/7/15	Microthiol (sulphur) **Lannate (methomyl)**	Microthiol (sulphur) **Lannate (methomyl)**
3/8/15	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)
10/8/15	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)
17/8/15	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*
24/8/15	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*)	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*)
31/8/15	Microthiol (sulphur) Coragen (chlorantraniliprole) Transform (sulfoxaflor) ParaMite (etoxazole) (miticide)	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus) **Rover (dimethoate)**
6&7/9/15	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*)	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus) **Lepidex (trichlorfon)**
14/9/15	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus) **Rover (dimethoate)**
21/9/15	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus) **Lepidex (trichlorfon)**
28/9/15	Vivus Max (nucleopolyhedrovirus)	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*) **Rover (dimethoate)** **Lannate (methomyl)**
6/10/15	Microthiol (sulphur) Vivus Max (nucleopolyhedrovirus)	Microthiol (sulphur) **Lannate (methomyl)** **Lepidex (trichlorfon)**
12/10/15	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*)	Microthiol (sulphur) **Lannate (methomyl)** **Rover (dimethoate)**
19/10/15	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*) Movento (spirotetramat)	Microthiol (sulphur) **Lannate (methomyl)** **Lepidex (trichlorfon)**
26/10/15	Microthiol (sulphur) Dipel (*Bacillus thuringiensis* subsp. *kurstaki*)	Microthiol (sulphur) **Lannate (methomyl)** **Lepidex (trichlorfon)**
2/11/15	Microthiol (sulphur) Movento (spirotetramat)	Microthiol (sulphur) Movento (spirotetramat)

	Belt (flubendiamide)	Belt (flubendiamide) **Lannate (methomyl)** **Lepidex (trichlorfon)**
9/11/15	Avral (*Bacillus thuringiensis*)	
16/11/15	Microthiol (sulphur) Avral (*Bacillus thuringiensis*)	

The first green fruit was observed in the trial block on 3rd September. Sampling commenced when red fruit were observed and occurred weekly from 20th October to 2nd December 2015 (Table 2). A total of 6966 red fruit (90.9 kg) were sampled from the trial block, and 3048 red fruit (48.8 kg) from the commercial comparison block over this period. Fruit were picked randomly from five marked areas within each block. Three chilli varieties were sampled (Table 2), all of which were of the Cayenne type. Sampled fruit were sent to the DAF laboratories in Brisbane where they were incubated for a minimum of one week (26°C, 70% relative humidity), then inspected for the presence of fruit fly larvae after a period of incubation (Plate 3). Reject dropped fruit was also sampled from between the rows in both blocks at weekly intervals in an attempt to evaluate its potential as a source of infestation. This dropped fruit may have fallen from the plants or been rejected during picking as unsuitable for harvest (e.g. due to sunburn). A total of 18.0 kg of reject fruit was sampled from the trial block and 17.9 kg from the comparison block (Table 3). Reject waste fruit was sampled from a waste pile, situated between the packing sheds and the comparison block, at approximately fortnightly intervals. A total of 5.2 kg freshly deposited waste was sampled. The upper limit of infestation percent at the 95% confidence level was calculated for the assessed chilli samples, using the program CQT STATS (Liquido and Griffin, 2010).

Sampling of pack-house fruit, to assess the impact of rejection of damaged or poor quality fruit during picking and grading, could not be evaluated as planned as in-field treatments yielded no infestation with which to make a comparison.

Plate 3 Incubation and assessment of sampled fruit

Cue-lure monitoring traps (BioTrap Australia Pty Ltd, Ocean Grove, VIC), that targeted male fruit flies, were installed at the trial block, commercial comparison block, and adjacent to the waste pile on 7th July 2015 and remained in place until 2nd December 2015 (Plate 4). Five traps were placed in the trial block and five in the commercial comparison block. At each site, the traps were placed in a line across the block, such that there was a trap placed in the tree-line on either side of the block (north and south end) and the remaining three traps were within the crop at varying distances from the border. The distances from the border varied between the two blocks due to their differing size and were positioned approximately: close to the southern tree-line (20 – 25 m), middle of the block (160 – 210 m from each tree-line) and mid-distant to the northern tree-line (215 - 300 m from the southern tree-line). The traps were serviced weekly (with the exception of one week's missed sampling at the beginning of October) and contents sent to the DAF laboratories in Brisbane for identification.

Plate 4 Cue-lure monitoring traps installed in the trial block

Results

No larvae were found in any fruit sampled from the trial block (Table 2). The upper infestation level with 95% confidence was estimated to be 0.04%. Two fruit sampled from the commercial block were found to contain a total of seven larvae, with the upper infestation level (95% confidence) estimated at 0.21%. No larvae were found in any of the reject fruit samples (Table 3). Two empty pupal cases were recovered from the final waste sample (collected 2/12/15), however no adults were found. It is possible that the pupal cases were present in the waste sample when it was collected, however the possibility of contamination while setting up the fruit cannot be ruled out. Therefore it cannot be stated conclusively that there were fruit fly present in this waste sample.

Table 2 Number and weight of sampled fruit picked from the trial block and commercial comparison block

Sampling date	Block	Variety	Number of fruit	Weight of fruit (g)	Number infested fruit	Number larvae	Upper % infestation level (95% confidence)
20/10/15	Trial	Blade	379	5336	0	0	0.79
20/10/15	Comparison	Caysan	203	4163	0	0	1.48
20/10/15	Comparison	Hong Kong	197	5434	0	0	1.52
27/10/15	Trial	Blade	520	9503	0	0	0.58
27/10/15	Comparison	Caysan	151	1982	0	0	1.98
27/10/15	Comparison	Hong Kong	150	4134	0	0	2.00
2/11/15	Trial	Blade	504	7547	0	0	0.59
2/11/15	Trial	Hong Kong	411	8392	0	0	0.73
2/11/15	Comparison	Caysan	54	1053	0	0	5.55
2/11/15	Comparison	Hong Kong	82	1929	0	0	3.65
11/11/15	Trial	Caysan	404	5303	0	0	0.74
11/11/15	Trial	Hong Kong	612	7985	0	0	0.49
11/11/15	Comparison	Caysan	275	4490	0	0	1.09
11/11/15	Comparison	Hong Kong	428	6216	0	0	0.70
24/11/15	Trial	Blade	1976	21892	0	0	0.15
24/11/15	Comparison	Caysan	401	5312	0	0	0.75
24/11/15	Comparison	Hong Kong	603	8841	2	7	1.04
30/11/15	Trial	Blade	2160	24899	0	0	0.14
2/12/15	Comparison	Blade	504	5249	0	0	0.59
TOTAL	**Trial**		**6966**	**90857**	**0**	**0**	**0.04**
TOTAL	**Comparison**		**3048**	**48803**	**2**	**7**	**0.21**

Table 3 Weight of reject fruit collected from between the rows and the waste pile

Sampling date	Block	Variety	Weight of fruit (g)	Number larvae
20/10/15	Trial	Blade	3465	0
20/10/15	Comparison	Caysan & Hong Kong	5936	0
21/10/15	Waste		1176	0
27/10/15	Trial	Blade	1812	0
27/10/15	Comparison	Caysan & Hong Kong	2663	0
2/11/15	Trial	Blade & Hong Kong	5109	0
2/11/15	Comparison	Caysan & Hong Kong	2434	0
2/11/15	Waste		670	0
11/11/15	Trial	Caysan & Hong Kong	5744	0
11/11/15	Comparison	Caysan & Hong Kong	3856	0
24/11/15	Trial	Blade	1916	0
24/11/15	Comparison	Caysan & Hong Kong	1612	0
24/11/15	Waste		1965	0
2/12/15	Comparison	Blade	1400	0
2/12/15	Waste		1389	0 *
TOTAL	**Trial**		**18046**	**0**
TOTAL	**Comparison**		**17901**	**0**
TOTAL	**Waste**		**5200**	**0 ***

* Two empty pupal cases were recovered from the waste fruit collected on 2/12/15. However, no adults were present.

The majority of flies caught in monitoring traps were Queensland fruit fly, *B. tryoni*, and lesser Queensland fruit fly, *B. neohumeralis* (Figure 1). Small numbers of non-pest species (*B. bryoniae*, *B. chorista* and *Dacus aequalis*) were also trapped. Trap catch remained at or below one fly per trap per day for the majority of the trapping period (Figure 2). This was substantially lower than trap catches at three other commercial vegetable farms in the Bundaberg area, monitored over the same period, which peaked at between 3.4 and 33 flies per trap per day, dependent on location (Appendix 5). Trap catch was higher at the final collection (6.75 flies per trap per day in the commercial block), however this collection was made after the blocks had been slashed and hence the high catches may have been a result of this disturbance.

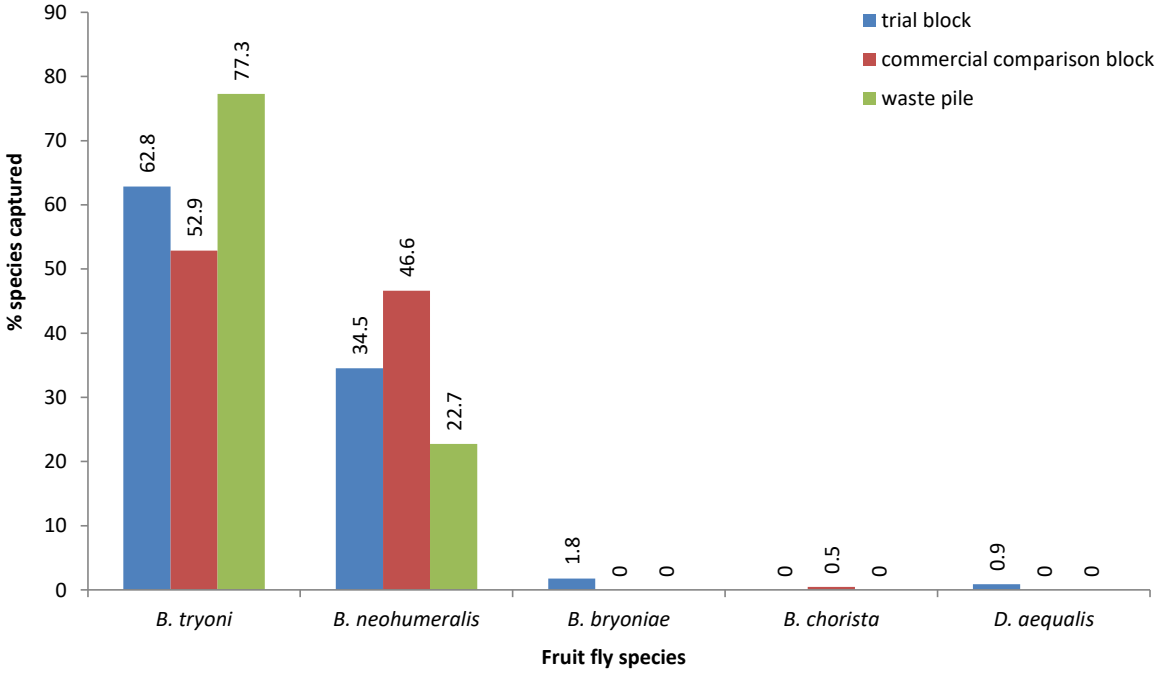

Figure 1 Percentage of male fruit fly of each species caught from cue-lure monitoring traps at the Austchilli trial sites between 7th July and 2nd December 2015

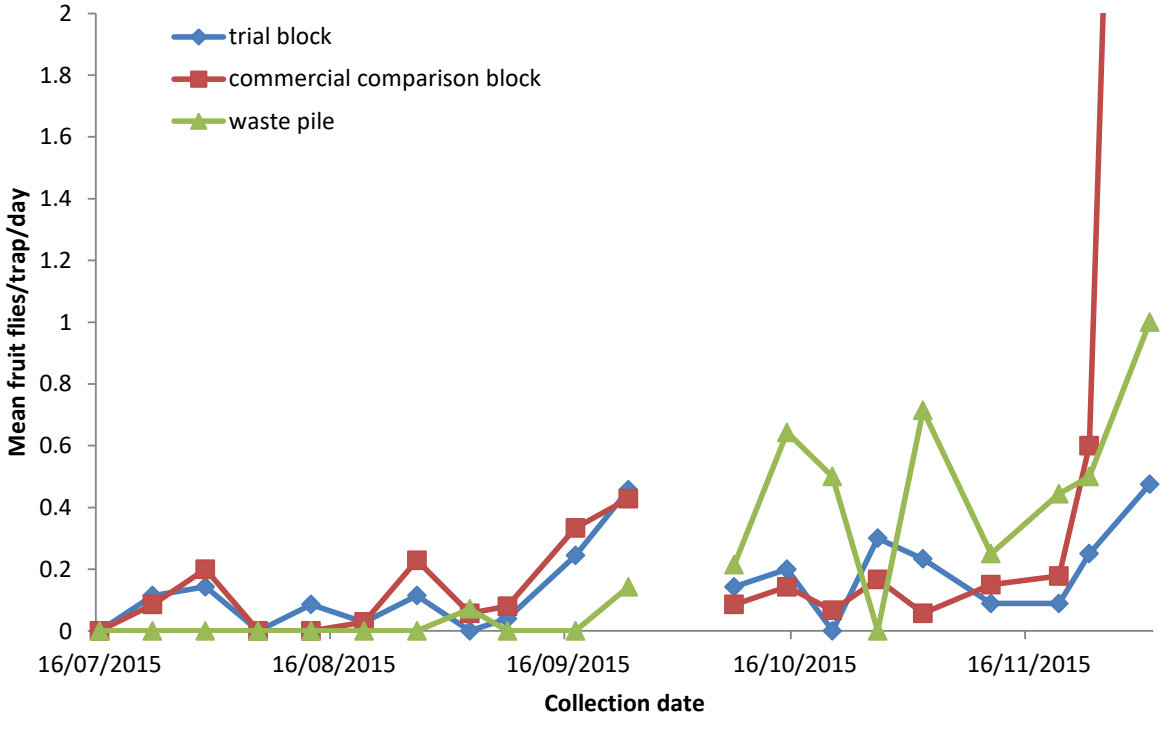

Figure 2 Male Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) cue-lure trap catches from 7th July to 2nd December 2015. Final trap catch at commercial comparison block was 6.75 flies (Y axis truncated to show detail)

Trap catch was affected by the location of traps within the blocks. In both blocks at Austchilli, the highest trap catches were observed in traps placed on the southern tree-line, followed by traps placed a short distance (20 – 25 m) into the crop, close to the southern tree-line (Table 3). Traps placed further into the crop caught the fewest flies.

Table 3 Total trap catch of Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) from traps placed at various locations within the vegetable crop and in bordering tree-lines

Trap location (distance from southern tree-line)	**Total trap catch over the trial period**	
	Trial block	**Commercial comparison block**
Southern tree-line	40	154
20 - 25 m in crop	30	22
160 - 210 m in crop	4	6
215 - 300 m in crop	13	7
Northern tree-line	23	18

Weather data for the trial period are displayed in Figure 3.

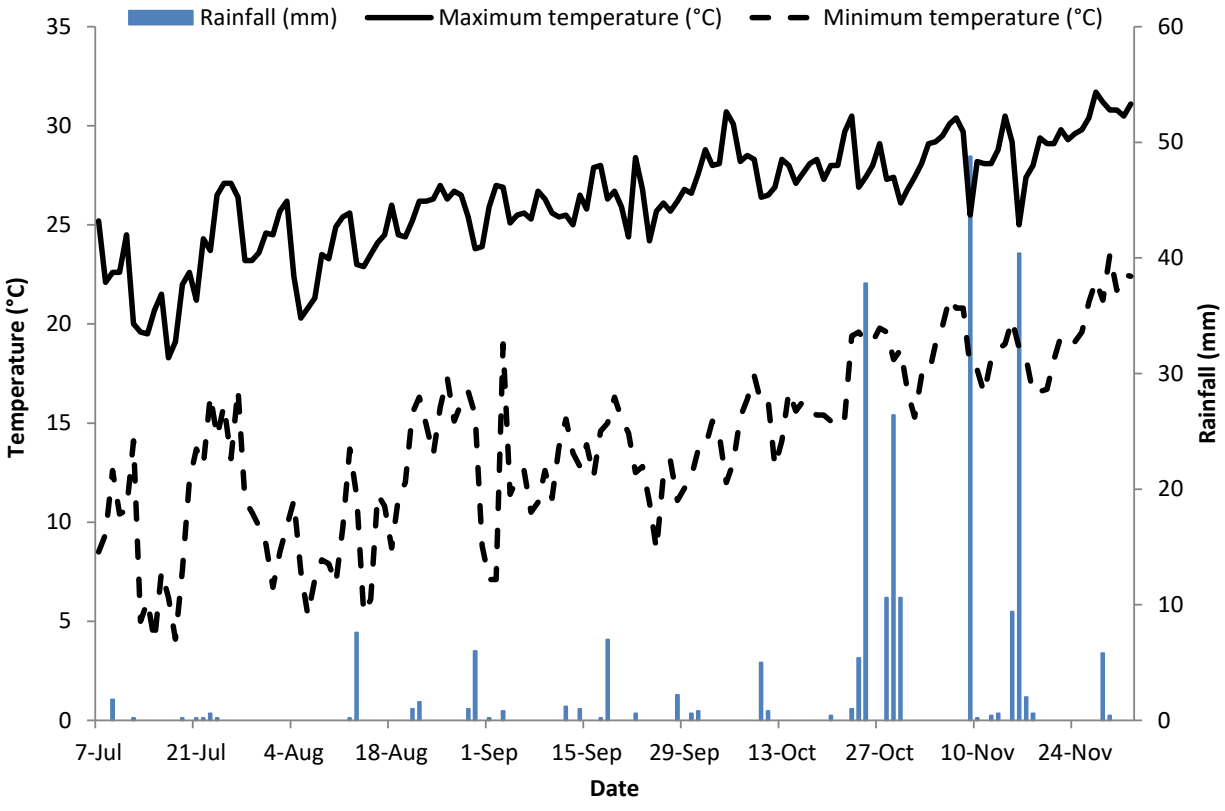

Figure 3 Weather data for the trial period: monitoring traps installed 7th July; observation of first green fruit in trial block 3rd September; start of sampling 20th October; end of sampling 2nd December 2015. Data obtained from the Bureau of Meteorology website for the closest weather station (39128, Bundaberg Aero).

In conclusion, trial results indicated that in-field treatments (perimeter protein baiting and MAT) were effective in controlling fruit fly in the trial block, without the use of broad spectrum cover sprays. As in-field treatments were so effective during this season, the impact of other parts of the system, such as rejection of damaged or poor quality fruit during picking and pack-house grading, could not be evaluated. Infestation in fruit sampled from the trial block was comparable to that in a commercial comparison block, where fruit flies were controlled using cover sprays of dimethoate, trichlorfon and methomyl. It should be noted that although trial sampling occurred during spring and early summer, a time of year when high fruit fly pressure is expected (Appendix 5), monitoring indicated that fruit fly numbers were low at both the trial block and commercial comparison block. It is likely that the fruit fly management system employed on a farm-wide scale at Austchilli resulted in low local fruit fly pressure.

It had originally been planned that the trial would continue in a second trial block, allowing sampling to continue for another season. However, following discussions with Austchilli management it was apparent that there was no suitable site with an existing border of vegetation on at least two sides of the crop, and management were unwilling to plant a sorghum or sugarcane border. The lack of a suitable perimeter planting would have compromised the efficacy of protein baiting. Therefore it was decided to continue the trial at Bundaberg Research Facility (Appendix 4).

References

Balagawi, S., Jackson, K. & Clarke, A.R. 2014. Resting sites, edge effects and dispersion of a polyphagous *Bactrocera* fruit fly within crops of different architecture. *Journal of Applied Entomology* **138**, 510-518.

DeFaveri, S. 2013. Farm-wide fruit fly management systems for the east coast of Australia. *HAL project MT12050*. Horticulture Innovation Australia.

Fay, H. 2011. Area wide management of fruit fly – Central Burnett phase 2. *HAL project CT06046*. Horticulture Australia Limited.

Gu, H. 2010. Alternative fruit fly treatment for interstate market access for strawberries. *HAL project BS06002*. Horticulture Australia Limited.

Liquido, N. J. & Griffin, R. L. 2010. Quarantine Treatment Statistics. United States Department of Agriculture, Center for Plant Health Science and Technology. Raleigh, NC. http://cqtstats.cphst.org/index.cfm

Lloyd, A. 2007. Area wide management of fruit fly - Central Burnett. *HAL project AH03002*. Horticulture Australia Limited.

Lloyd, A.C., Hamacek, E.L., Kopittke, R.A., Peek, T., Wyatt, P.M., Neale, C.J., Eelkema, M. & Gu, H.N. 2010. Area-wide management of fruit flies (Diptera: Tephritidae) in the Central Burnett district of Queensland, Australia. *Crop Protection* 29: 462-469

Missenden, B. 2014. Alternative fruit fly treatment for interstate access for strawberries: winter window option. *HAL project BS09022*. Horticulture Australia Limited.

Nishida, T. & Bess, H.A. 1957. Studies on the ecology and control of the melon fly *Dacus (Strumeta) cucurbitae* Coquillett (Diptera: Tephritidae). *Technical Bulletin Hawaii Agricultural Experiment Station* 34, 44 pp

Oag, D. 2010. Domestic market access for table grapes. *HAL project TG08001*. Horticulture Australia Limited.

Prokopy, R.J., Miller, N.W., Piñero, J.C., Barry, J.D., Tran, L.C., Oride, L. & Vargas, R. 2003. Effectiveness of GF-120 fruit fly bait spray applied to border area plants for control of melon flies (Diptera: Tephritidae). *Journal of Economic Entomology* 96: 1485-1493

Appendix 4 Evaluation of perimeter protein baiting and MAT for control of fruit flies in a small-scale capsicum trial

A trial was performed in a research planting of capsicum at Bundaberg Research Facility (Ashfield Road, Bundaberg, QLD) to assess a combination of perimeter protein baiting and male annihilation technique (MAT) for management of fruit fly in a fruiting vegetable crop. This trial followed the successful completion of a trial in a commercial chilli crop in which the use of perimeter baiting and MAT, without the use of broad spectrum cover sprays, resulted in zero fruit fly larvae in sampled fruit (Appendix 3). It was not possible to conduct a second trial at Austchilli, therefore this second trial was performed on a smaller scale at Bundaberg Research Facility. The trial was established in November 2015 and continued until December 2016.

The trial was conducted within an area approximately 300 m x 100 m, used for a variety of vegetable research trials (Plate 1, trial site outlined in red). There was an established tree-line to either side of the site. Bamboo was planted at one end, approximately six months prior to the start of the trial, however at the time the trial started the plants were very small. A sorghum border was therefore planted (outlined in green), which provided a substrate for application of protein bait over the first seven months of the trial. The sorghum border was well established when the first capsicum were planted, but deteriorated over the course of the trial and was removed in August, after which time protein bait was applied to the bamboo. It was not possible to plant a border crop along the northern end of the trial site and therefore protein bait application was not made to the full perimeter. Capsicum seedlings (variety Caldo) were planted in an area of the trial site (ca. 100 m x 50 m area) (Plate 2). The first planting was made on 25th November 2015, with further sequential plantings at 8 to 13 week intervals, to enable fruit to be sampled over the course of several months. The planting area was divided into three blocks, each ca. 0.1 ha, with each planting consisting of three rows of capsicum within each block. The three blocks of capsicum were at varying distances from the tree lines (Plate 2): block 3 was approximately 15 m from the closest (western) tree line, and block 1 approximately 48 m from the western tree line. During the period of the trial, other crops within the trial site included capsicums, tomatoes and strawberries. To either side of the trial site were blocks of citrus. Fruit flies were managed in these blocks using a combination of protein baiting and cover sprays.

MAT wicks (Bugs for Bugs Pty Ltd, Mundubbera, QLD) were installed around the perimeter of the trial site at 20 m intervals on 24th November, immediately prior to the first capsicum planting. The MAT wicks were placed in the tree-line, on a wire fence-line on the southern border of the trial area, or on stakes on the northern border. New wicks were added every three months, leaving the older wicks in place as per the manufacturer's instructions. Weekly applications of protein bait to the tree-line and sorghum on the perimeter of the trial site commenced in December, several weeks prior to the start of fruiting of the capsicum crop (Plate 3). The protein bait consisted of 2 L Fruit Fly Lure (Bugs for Bugs Pty Ltd) and 25 ml Vertimec (abamectin) in 100 L water. Any reject fruit (damaged due to sunburn, bacterial rots or grub damage) were collected and disposed of. Due to high numbers of fruit fly in sampled fruit, fortnightly applications of Success Neo (spinetoram) at the label rate of 40 ml/100 L were made from 30th May 2016 onwards as an additional measure, increasing to weekly from 8th July. Insecticides were applied as necessary for control of other pests, however broad spectrum insecticides were avoided where possible, and most of the applied insecticides would have had little or no effect on fruit flies. The exceptions were Lorsban (chlorpyrifos) at 100 ml/100 L, applied to seedlings for control of soil pests, and Vertimec at 60 ml/100 L, applied at intervals to fruiting plants to control infestations of mites (Table 1).

There was a fruit fly management program in place at Bundaberg Research Facility prior to the start of the trial, primarily for control of fruit fly in citrus blocks situated to either side of the trial site (Plate 1). However, it became apparent that it was not implemented effectively. Application of insecticide cover sprays and protein bait was sporadic

and not performed throughout all blocks. MAT devices were not used and fallen fruit was not removed from the orchards. An attempt was made to improve control in the citrus blocks through installation of MAT traps (18th February) and increasing the frequency and scope of protein baiting.

Red capsicum fruit were sampled at weekly intervals from 3rd February to 5th December 2016 (Table 2). Fruit were sampled randomly from each of the three blocks, and fruit from each block was held and assessed separately. A total of 4360 fruit (823 kg) were sampled from block 1, 4264 fruit (829 kg) from block 2 and 4371 fruit (836 kg) from block 3 over the trial period. Sampled fruit were sent to the DAF laboratories in Brisbane where they were placed on drip trays, held over vermiculite, in lidded, ventilated containers (Plate 4). The fruit was incubated for a minimum of one week (26°C, 70% relative humidity), then inspected for the presence of fruit fly larvae and pupae. Larvae and pupae found in sampled fruit were retained until emergence for identification of adults. The upper limit of infestation percent at the 95% confidence level was calculated for the assessed chilli samples, using the program CQT STATS (Liquido and Griffin, 2010).

From May onwards, ten reject fruit picked for disposal every week were first dissected and examined for the presence of fruit fly larvae in the laboratory at Bundaberg Research Facility. From June onwards, a further 30 to 40 reject fruit from each block were placed into emergence traps and monitored for the presence of adult fruit flies. Fruit from each block was placed into a separate emergence trap.

Cue-lure monitoring traps (BioTrap Australia Pty Ltd, Ocean Grove, VIC), targeting male fruit flies, were installed at the trial site on 25th November 2015. The lures were replaced every three months. Two traps were placed in the capsicum plantings (one each in blocks 1 and 3), and three in the tree-line on the perimeter of the trial site (Plate 1). The traps were serviced weekly and contents sent to the DAF laboratories in Brisbane for identification. Additional cue-lure monitoring traps were installed at the Bundaberg Research Facility on 22nd December 2015 in the citrus blocks and elsewhere, in order to monitor the fruit fly population in the immediate vicinity of the trial site. These flies were counted *in situ* and not identified to species. Average daily trap catch at the trap site and elsewhere at the Research Facility was correlated with the average daily minimum and maximum temperatures, using Pearson's correlation.

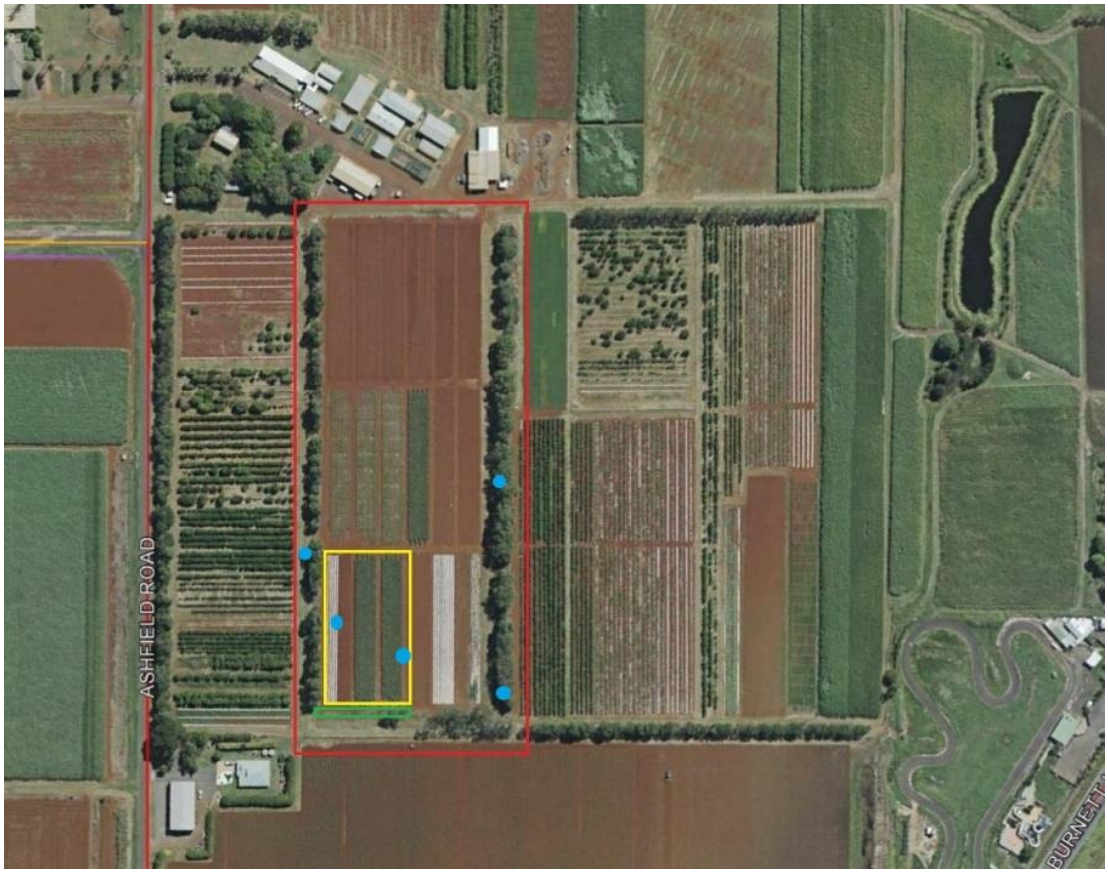

Plate 1 Bundaberg Research Facility trial showing the trial site (outlined in red), location of capsicum plantings (outlined in yellow), sorghum (outlined in green), and traps (blue dots)

Plate 2 Three blocks of capsicum adjacent to a tree-line at the trial site

Plate 3 Application of protein bait to vegetation on the perimeter of the trial site

Plate 4 Incubation of sampled fruit

Table 1 Insecticides applied to the capsicums at the Bundaberg Research Facility trial. Products with efficacy against fruit fly in bold.

Date	Insecticide	Date	Insecticide
26/11/15	Bacchus WG (*Bacillus thuringiensis* subsp aizawai)	9/6/16	**Success Neo (spinetoram)**
30/11/15	**Lorsban (chlorpyrifos)**	17/6/16	Belt (flubendiamide)
8/12/16	**Lorsban (chlorpyrifos)**	23/6/16	Transform (sulfoxaflor)
14/12/15	**Success 2 (spinosad)**	24/6/16	**Success Neo (spinetoram)**
21/12/15	Proclaim (emamectin) Bacchus WG (*Bacillus thuringiensis* subsp aizawai)	1/7/16	Transform (sulfoxaflor)
30/12/15	Belt (flubendiamide)	8/7/16	**Success Neo (spinetoram)**
8/1/16	Proclaim (emamectin)	13/7/16	**Lorsban (chlorpyrifos)** * **Success Neo (spinetoram)**
15/1/16	Transform (sulfoxaflor) Bacchus WG (*Bacillus thuringiensis* subsp aizawai)	21/7/16	**Success Neo (spinetoram)**
22/1/16	Movento (spirotetramat)	28/7/16	**Success Neo (spinetoram)**
28/1/16	Chess (pymetrozine) Belt (flubendiamide)	5/8/16	**Vertimec (abamectin)** **Success Neo (spinetoram)**
11/2/16	Belt (flubendiamide)	11/8/16	**Success Neo (spinetoram)**
18/2/16	Belt (flubendiamide) Movento (spirotetramat)	19/8/16	**Success Neo (spinetoram)**
26/2/16	Belt (flubendiamide)	26/8/16	**Success Neo (spinetoram)**
3/3/16	Proclaim (emamectin)	2/9/16	**Success Neo (spinetoram)**
10/3/16	Belt (flubendiamide)	9/9/16	**Success Neo (spinetoram)** Transform (sulfoxaflor)
17/3/16	Belt (flubendiamide)	14/9/16	**Success Neo (spinetoram)**
23/3/16	Belt (flubendiamide)	20/9/16	**Lorsban (chlorpyrifos)** *
31/3/16	Belt (flubendiamide)	23/9/16	**Success Neo (spinetoram)**
8/4/16	Belt (flubendiamide)	28/9/16	**Success Neo (spinetoram)**
13/4/16	**Lorsban (chlorpyrifos) ***	7/10/16	**Success Neo (spinetoram)**
15/4/16	Proclaim (emamectin)	14/10/16	**Success Neo (spinetoram)**
22/4/16	Belt (flubendiamide)	21/10/16	**Success Neo (spinetoram)**
29/4/16	Belt (flubendiamide)	27/10/16	**Success Neo (spinetoram)** Movento (spirotetramat)
6/5/16	**Success 2 (spinosad)**	3/11/16	**Success Neo (spinetoram)**
16/5/16	Belt (flubendiamide)	11/11/16	**Success Neo (spinetoram)**
20/5/16	Transform (sulfoxaflor)	17/11/16	**Success Neo (spinetoram)**
27/5/16	**Vertimec (abamectin)**	24/11/16	**Success Neo (spinetoram)**
30/5/16	**Success Neo (spinetoram)**	2/12/16	Belt (flubendiamide) Transform (sulfoxaflor)
2/6/16	**Vertimec (abamectin)**		

* Applications of Lorsban were made to newly planted seedlings only

Results

A total of 12,995 capsicum fruit (2488 kg) were sampled over the trial period (3rd February to 5th December 2016), from which a total of 482 fruit fly pupae were recovered (Table 2). All adults emerging from pupae were identified as *B. tryoni* or *B. neohumeralis*. No pupae were recovered from fruit sampled in the first two weeks, and only one larva was found in fruit sampled on 17th February (Table 2, Figure 1). However, infestation in sampled fruit then increased and detections of pupae occurred in fruit sampled from one or more blocks at each sampling date between 24th February and 9th May, the highest number occurring in the sampling of 2nd March. No pupae were recovered from 18th May to 5th September, a period coinciding with lower temperatures (Figure 2) and lower trap catches (Figures 4 and 5). This indicates a decrease in activity of the flies: it is known that *B. tryoni* activity decreases at lower temperatures (Meats, 1989). However, the efficacy of cue-lure is also influenced by weather conditions (reviewed by Clarke et al, 2011) so it cannot be assumed that low trap catches indicate low fruit fly populations. Fortnightly applications of Success Neo (spinetoram) also commenced during this period, from 30th May onwards, increasing in frequency to weekly applications from 8th July. However, infestations in sampled fruit were detected once again in the spring (12th September) when temperatures began to rise, indicating that the additional Success Neo cover sprays coupled with perimeter baiting and MAT were not sufficient to manage fruit fly under warmer conditions. Numbers of pupae then increased in subsequent weekly samplings, with a total of 67 pupae recovered from the final fruit sample in December.

The majority of pupae were recovered from fruit taken from block 3, which was closest to the tree-line, a total of 297 pupae over the trial period (Table 2). This compared with 127 pupae recovered from the middle block (block 2) and only 58 pupae from block 1, the furthest from the tree-line. This could be a result of higher fruit fly activity close to the tree-line, which would fit with observations from monitoring in commercial crops (Appendices 5 and 6), and trap catches in the large-scale trial at Austchilli (Appendix 3).

Table 2 Number and weight of capsicum fruit sampled from the Bundaberg Research Facility trial

Sampling date	Block *	Number of fruit	Weight of fruit (kg)	Number of pupae	Upper % infestation level (95% confidence)
3/2/16	1	244	23.8	0	1.2
	2	315	29.0	0	1.0
	3	205	14.9	0	1.5
10/2/16	1	72	14.3	0	4.2
	2	134	24.6	0	2.2
	3	165	28.5	0	1.8
17/2/16	1	103	19.9	0	2.9
	2	102	18.6	0	2.9
	3	143	20.7	1 (larva)	3.3
24/2/16	1	157	28.1	0	1.9
	2	167	30.5	8	8.6
	3	199	31.1	28	19.3
2/3/16	1	172	28.5	13	12.0
	2	157	28.4	22	20.0
	3	171	28.8	50	37.0
9/3/16	1	179	28.6	0	1.7
	2	167	27.8	6	7.1
	3	163	24.5	24	20.7
16/3/16	1	136	20.6	0	2.2
	2	139	20.8	0	2.2

	3	132	19.9	13	15.7
Seven week break in sampling between sequential plantings					
9/5/16	1	121	27.8	0	2.5
	2	120	27.2	0	2.5
	3	135	29.4	30	30.1
18/5/16	1	122	22.4	0	2.5
	2	144	25.8	0	2.1
	3	137	26.0	0	2.2
One week break in sampling					
1/6/16	1	156	29.2	0	1.9
	2	163	29.0	0	1.8
	3	158	27.8	0	1.9
8/6/16	1	157	25.9	0	1.9
	2	165	28.7	0	1.8
	3	172	30.6	0	1.7
15/6/16	1	160	28.5	0	1.9
	2	139	26.9	0	2.2
	3	154	27.5	0	1.9
22/6/16	1	163	29.2	0	1.8
	2	155	28.8	0	1.9
	3	154	25.6	0	1.9
Two week break in sampling due to insufficient fruit					
12/7/16	1	115	21.1	0	2.6
	2	124	22.3	0	2.4
	3	142	25.3	0	2.1
20/7/16	1	95	33.0	0	3.2
	2	89	28.7	0	3.4
	3	93	30.6	0	3.2
27/7/16	1	94	32.2	0	3.2
	2	94	30.6	0	3.2
	3	93	31.9	0	3.2
Two week break in planting due to heavy rain					
15/8/16	1	130	36.2	0	2.3
	2	119	34.6	0	2.5
	3	122	37.1	0	2.5
22/8/16	1	142	36.0	0	2.1
	2	142	38.4	0	2.1
	3	142	39.2	0	2.1
29/8/16	1	159	36.4	0	1.9
	2	156	36.0	0	1.9
	3	156	35.7	0	1.9
5/9/16	1	129	26.4	0	2.3
	2	117	28.0	0	2.6
	3	129	29.1	0	2.3
12/9/16	1	131	26.2	0	2.3
	2	116	26.9	0	2.6
	3	126	26.3	6	2.4
19/9/16	1	155	25.0	0	1.9
	2	141	25.2	0	2.1

	3	139	25.3	11	13.1
26/9/16	1	185	25.4	17	13.8
	2	176	25.4	17	14.5
	3	166	25.6	4	5.5
4/10/16	1	173	21.2	0	1.7
	2	67	8.1	0	4.5
	3	159	22.8	0	1.9
Two week break in planting between sequential samplings					
24/10/16	1	104	26.0	0	2.9
	2	100	25.3	10	17.0
	3	102	26.3	0	2.9
31/10/16	1	111	27.4	0	2.7
	2	105	27.3	0	2.9
	3	99	26.2	14	22.1
7/11/16	1	108	27.2	0	2.8
	2	103	28.1	0	2.9
	3	101	26.8	0	3.0
15/11/16	1	111	26.5	8	13.0
	2	109	26.5	15	21.2
	3	110	27.3	20	26.4
21/11/16	1	123	25.3	10	13.8
	2	121	24.9	1	3.9
	3	120	24.9	35	38.7
28/11/16	1	170	22.9	2	3.7
	2	153	23.4	41	34.8
	3	152	22.8	9	10.3
5/12/16	1	183	21.7	13	11.3
	2	165	23.6	7	8.0
	3	132	16.9	54	51.3
Totals per block	**1**	**4360**	**822.8**	**63**	**1.8**
	2	**4264**	**829.2**	**127**	**3.5**
	3	**4371**	**835.6**	**299**	**
Grand total		**12995**	**2487.7**	**489**	**

* Block 1 was approximately 48 m from the nearest tree line and block 3 approximately 15 m

** Upper % infestation level could not be calculated due to high infestation

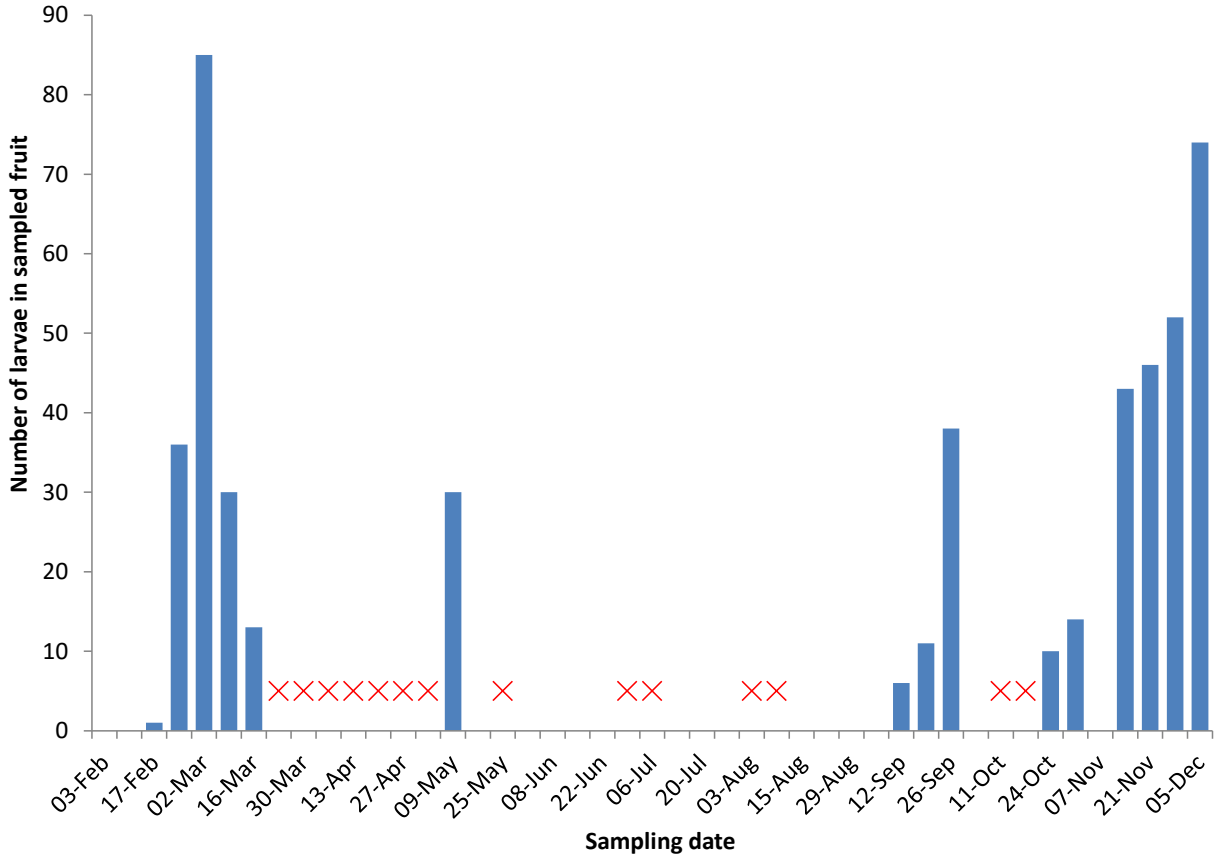

Figure 1 Summary of number of pupae recovered from sampled fruit. Red crosses indicate no fruit sampling occurred that week.

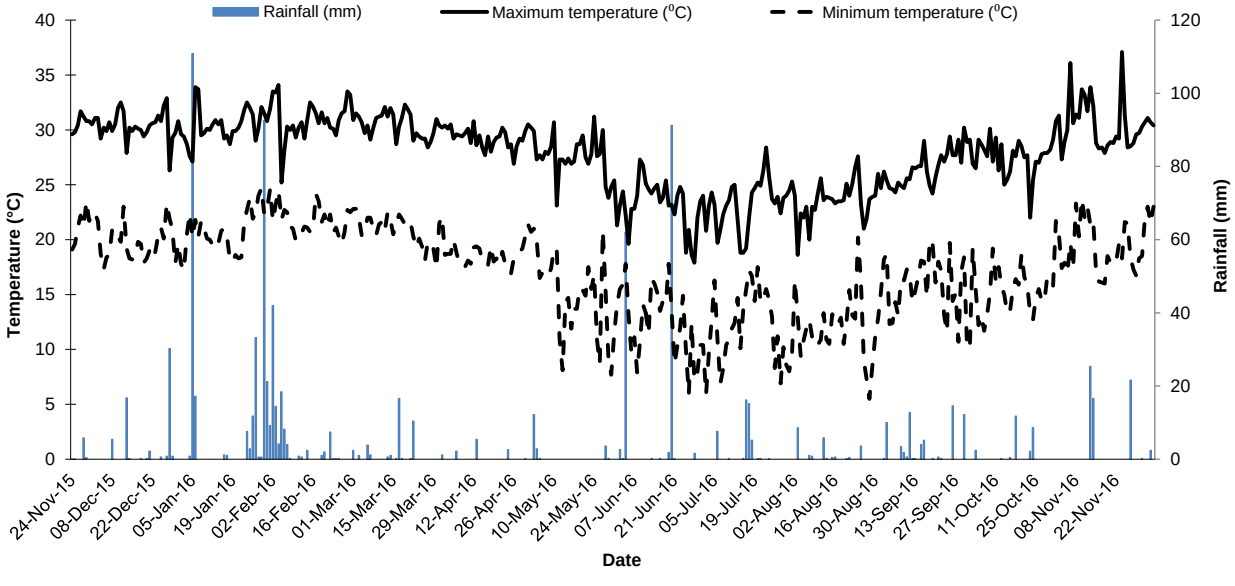

Figure 2 Weather data for the trial period: monitoring traps installed 25th November 2015; start of sampling 3rd February 2016; end of sampling 5th December 2016. Data obtained from the Bureau of Meteorology website for the closest weather station (39128, Bundaberg Aero).

No fruit fly larvae were observed in any of the sampled reject fruit examined in the laboratory, and no adult flies were recovered from emergence traps holding reject fruit. However, larvae of *Atherigona* sp. were commonly observed in rotting fruit. These flies do not attack intact fruit, breeding in rotting fruit and organic matter, but the larvae can easily be mistaken for those of Queensland fruit fly (Plate 5). It is therefore important that growers are able to distinguish between the larvae of the two species so that cover sprays are not applied unnecessarily.

Plate 5 Larvae of *Atherigona* sp. (left) showing distinguishing black spiracles, and *Bactrocera tryoni* (right), with no spiracles visible

The majority of trapped flies were Queensland fruit fly (*Bactrocera tryoni*) and lesser Queensland fruit fly (*Bactrocera neohumeralis*) (Figure 3). Small numbers of non-pest species (*B. bryoniae* and *B. quadrata*) were also caught. A single fly classified as 'other' was a damaged specimen unable to be identified. Average trap catch (Queensland fruit fly and lesser Queensland fruit fly) from traps installed at the trial site at the start of the trial in December was 2.7 flies per trap per day, decreasing to less than 1 fly per trap per day from 11th February onwards, and remaining below 1 fly per trap per day for the remainder of the trial (Figure 4). Trap catch was low over the winter period, remaining at or below 0.1 fly per trap per day between mid-June and mid-October. This was broadly consistent with the results of monitoring performed at commercial vegetable farms in the Bundaberg region (Appendix 5), and with the findings of similar trials carried out in strawberry crops in the Bundaberg region: Missenden (2014) found average trap catch from commercial strawberry farms in the Bundaberg region was less than 0.25 male flies/trap/day between May and September 2012, and less than 0.5 male flies/trap/day from April to mid-August 2013, with trap catches increasing subsequently. Trap catch from monitoring traps installed at Bundaberg Research Facility, in the citrus blocks and elsewhere, was generally similar to that at the trial site, however trap catch in the citrus blocks was occasionally higher. As these flies were not identified to species, data represent total trap catch. Trap catches from monitoring traps installed at the trial site and elsewhere at the research facility were positively correlated with both the daily minimum and daily maximum temperature (P<0.001) (Table 3).

Table 3 Correlation of trap catches (traps placed at the trial site and elsewhere at the Research Facility) with minimum and maximum temperatures

	Minimum temperature	Maximum temperature
Trial site trap catches	r=0.560 *P*<0.001	r=0.543 *P*<0.001
Research facility trap catches	r=0.579 *P*<0.001	r=0.546 *P*<0.001

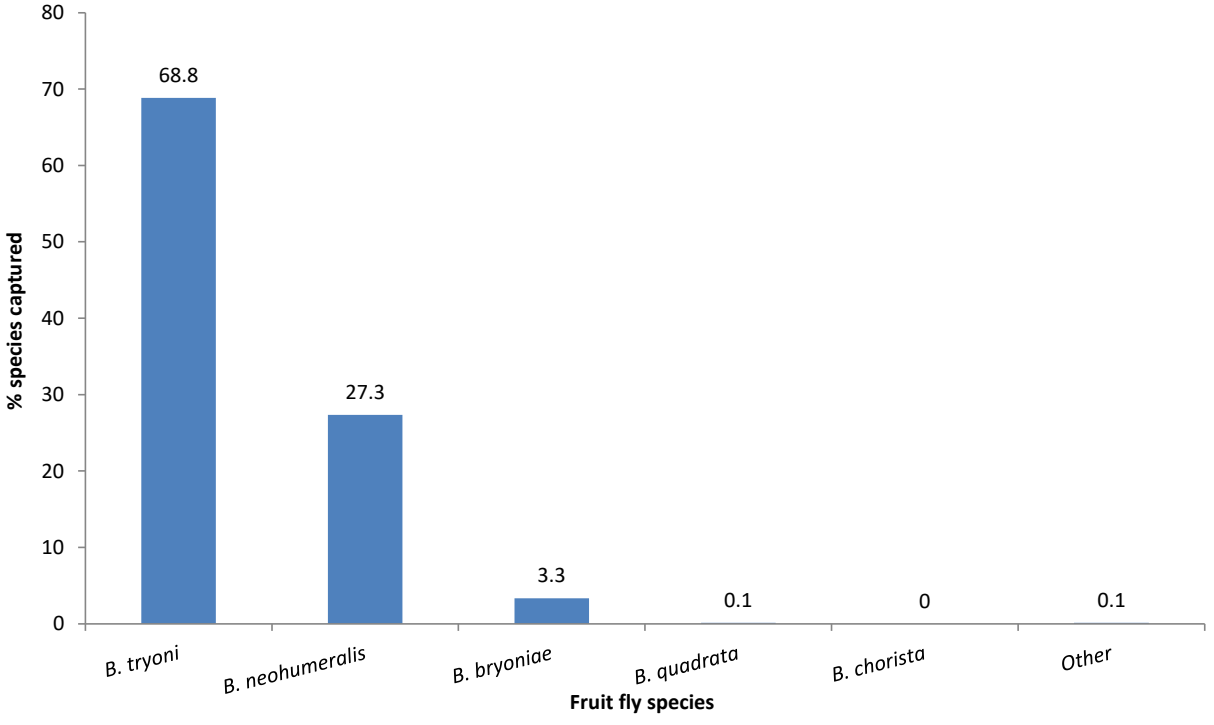

Figure 3 Percentage of male fruit fly of each species caught from cue-lure monitoring traps at the Bundaberg Research Facility trial site between 24th November 2015 and 5th December 2016

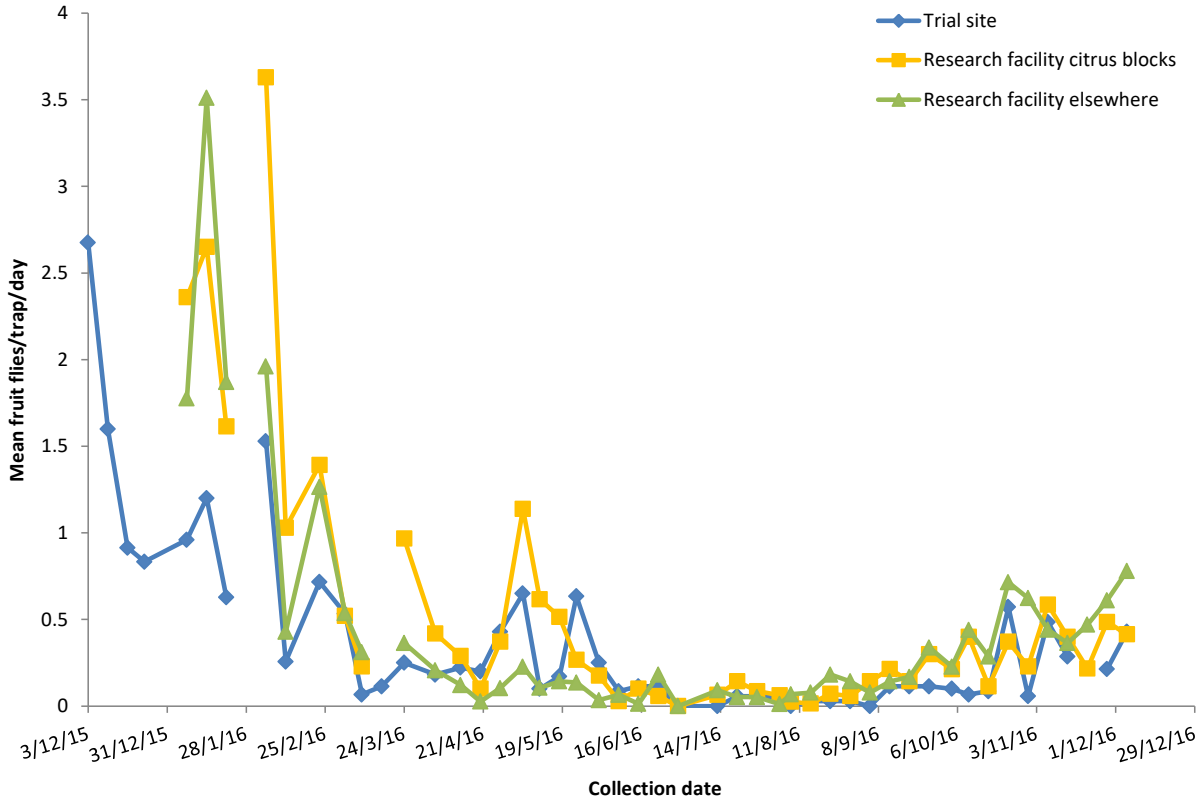

Figure 4 Male fruit fly trap catches from cue-lure monitoring traps at the Bundaberg Research Facility trial site and surrounds between 25th November 2015 and 5th December 2016. Trial site catches are Queensland fruit fly (*B. tryoni* and *B. neohumeralis*); Research Facility trap catches were not identified to species.

Although not the purpose of the trial, there may be interest in a comparison of trap catch and fruit infestation (Figure 5). These data reflect the results from one trial site, over one year, for one fly species and one commodity and therefore it is not possible to infer any major conclusions. Even though there are components that appear to show a relationship (e.g. fruit infestation and trap catch are both very low over winter) it could definitely not be said that trap catches are a good indicator of infestation levels from these data alone.

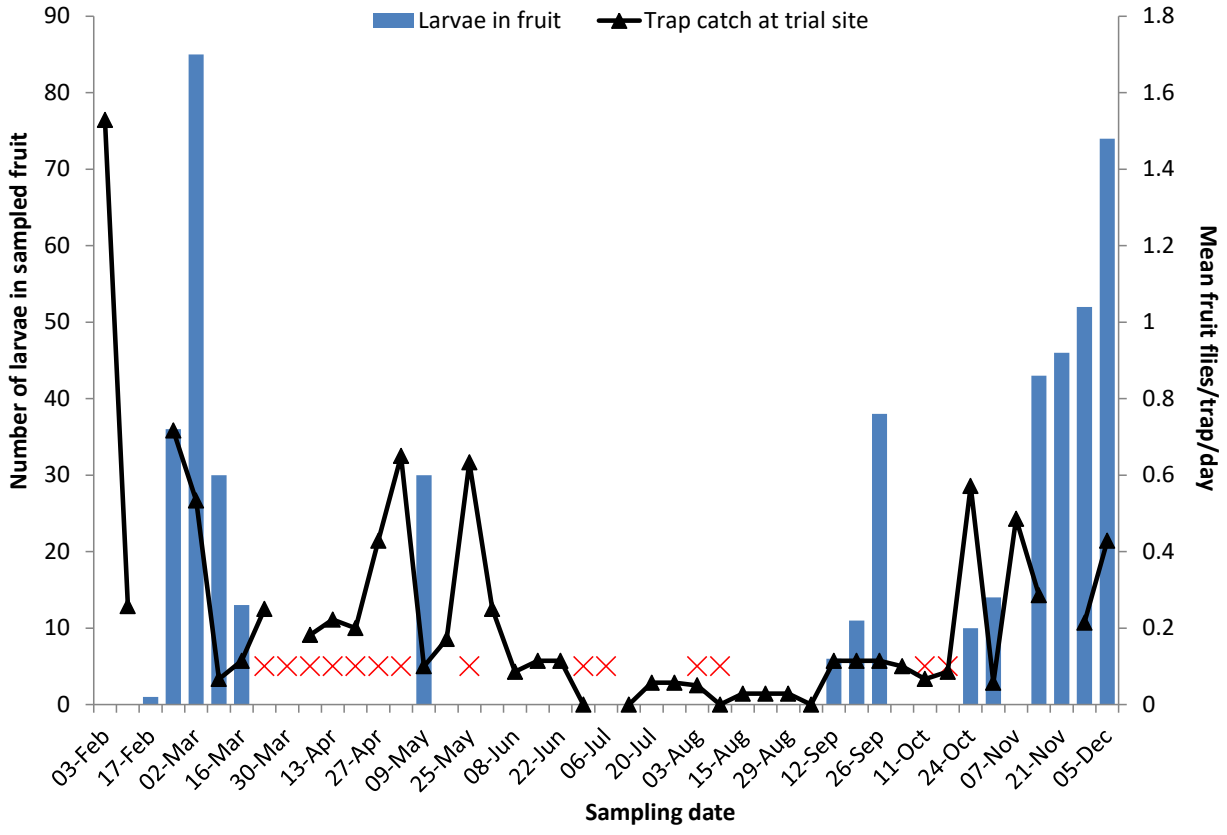

Figure 5 Summary of number of pupae recovered from sampled fruit and trap catch at the trial site. Red crosses indicate no fruit sampling occurred that week.

Trap location had little effect on trap catch (Table 4), unlike the large-scale trial performed in a commercial chilli block (Appendix 3), where the highest total trap catch occurred in a trap placed in a tree-line. This may have been due to the fact that the research facility trial was performed on a much smaller scale compared to the commercial crop trial.

Table 4 Total trap catch of Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) from traps placed at various locations within the trial site

Trap location	**Total trap catch over the trial period**
Western tree-line	124
Crop 15 m from closest tree-line	172
Crop 48 m from closest tree-line	202
Eastern tree-line	141
Eastern tree-line to north of crop *	142

* The trap placed on the eastern tree-line to the north of the crop was initially adjacent to a crop of mature capsicum (not part of the trial), which were harvested a few weeks into the trial

In conclusion, trial results indicate that the combination of perimeter protein baiting, MAT and regular cover spray applications of Success Neo were effective in controlling fruit fly in a small block of capsicum during winter when fruit fly pressure was low: no pupae were recovered from sampled fruit from 18th May until 5th September. The threshold for female fruit fly ovarian development has been found to be 13.5°C, with 1.6 degree days above this threshold required for maturation of ovaries to occur (Pritchard, 1970; Fletcher, 1975). Daily minimum temperatures regularly dropped below 13.5°C between mid-May and mid-October. Treatments were not sufficient to prevent fruit fly damage under conditions of high fruit fly pressure during the summer and autumn. However, it should be noted that local fruit fly pressure at the research facility where this trial was performed was much higher than in the trial performed in a commercial chilli crop (Appendix 3). This high pressure was due in large part to the presence of fruit fly in citrus blocks adjacent to the trial site. Although an attempt was made to improve control in the citrus blocks, it was likely that this was not sufficient to greatly reduce the fruit fly population in the area during the course of the trial. It is likely that the resulting high fruit fly pressure impacted on the trial, particularly as the trial site was relatively small.

References

Clarke, A.R., Powell, K.S., Weldon, C.W. & Taylor, P.W. 2011. The ecology of *Bactrocera tryoni* (Diptera: Tephritidae): what do we know to assist pest management? *Annals of Applied Biology* **158**, 26-54

Fletcher, B.S. 1975. Temperature-regulated changes in the ovaries of overwintering females of the Queensland fruit fly, *Dacus tryoni*. *Australian Journal of Zoology* **23**, 91-102.

Liquido, N. J. & Griffin, R.L. 2010. Quarantine Treatment Statistics. United States Department of Agriculture, Center for Plant Health Science and Technology. Raleigh, NC. http://cqtstats.cphst.org/index.cfm

Meats, A. 1989. Abiotic mortality factors: temperature. In *Fruit flies: Biology, natural enemies and control*. pp. 231-234. Eds A.S. Robinson and G.H.S. Hooper. Amsterdam, the Netherlands: Elsevier.

Missenden, B. 2014. Alternative fruit fly treatment for interstate market access for strawberries: winter window option. *HAL project BS09022*. Horticulture Australia Limited.

Pritchard, G. 1970. The ecology of a natural population of Queensland fruit fly, *Dacus tryoni*. III. The maturation of female flies in relation to temperature. *Australian Journal of Zoology* **18**, 77-89

Appendix 5 Seasonal fruit fly activity in the Bundaberg region

Cue-lure traps

Cue-lure monitoring traps (Bugs for Bugs Pty Ltd, Munduberra, QLD), that targeted male fruit flies, were installed in or adjacent to capsicum and tomato crops at four farms in the Bundaberg region in July 2014 in order to monitor seasonal activity of fruit flies in relation to fruiting vegetable crops (Table 1). At two properties, traps were placed within the crop as well as in the tree-line at the perimeter of the vegetable crop (Plate 1). These traps placed within the crop were relocated at harvest during the first year of monitoring such that all traps were along the tree-line, and they remained in this position for the remainder of the monitoring period. Traps were checked every two weeks, the contents emptied into labelled containers, and sent to the DAF laboratories in Brisbane for identification of trapped flies. The wicks were replaced every three months. Monitoring continued until 21st March 2017. Monitoring at Property B was discontinued on 16th February 2015 at the request of the property owner.

Table 1 Monitoring trap installation details

Property	**Date installed**	**Number of traps and location at installation**
Property A	3/7/14	Six on tree-line at the perimeter of capsicum crop
Property B *	15/7/14	Nine total: 3 - fence-line at perimeter of capsicum crop 3 - tree-line at perimeter of capsicum crop 3 - in capsicum crop
Property C	16/7/14	Three on tree-line at perimeter of capsicum crop
Property D †	16/7/14	Nine total: 3 – in tomato crop (on wire trellis at end of rows) 3 – in capsicum crop 3 – tree-line at perimeter of tomato crop

* Monitoring at Property B was discontinued on 16th February 2015 at the request of the property owner

† Traps in the tomato and capsicum crops were relocated to the tree-line in November 2014 when crops were harvested, and subsequently tomato was planted over a large proportion of the block

Plate 1 Cue-lure monitoring traps placed (clock-wise from top left) on the tree-line, on a fence-line, within a capsicum crop and on trellis in a tomato block

The majority of trapped flies were Queensland fruit fly (*Bactrocera tryoni*) and lesser Queensland fruit fly (*Bactrocera neohumeralis*) (Figures 1-3). Small numbers of non-pest species were also trapped. Flies categorised as 'other' were predominately damaged specimens that could not be identified with confidence. This was a particular problem following periods of heavy rainfall, causing fly samples to become mouldy and unidentifiable. Further trapped flies categorised as 'other' were a small number of *Zeugodacus cucumis*, *Dacus signatifrons*, *D. newmani, B. breviaculeus* and either *B. mayi* or *B.* sp. nr. *quadrata* (identification performed by Senior Entomologist Jane Royer, DAF BQ).

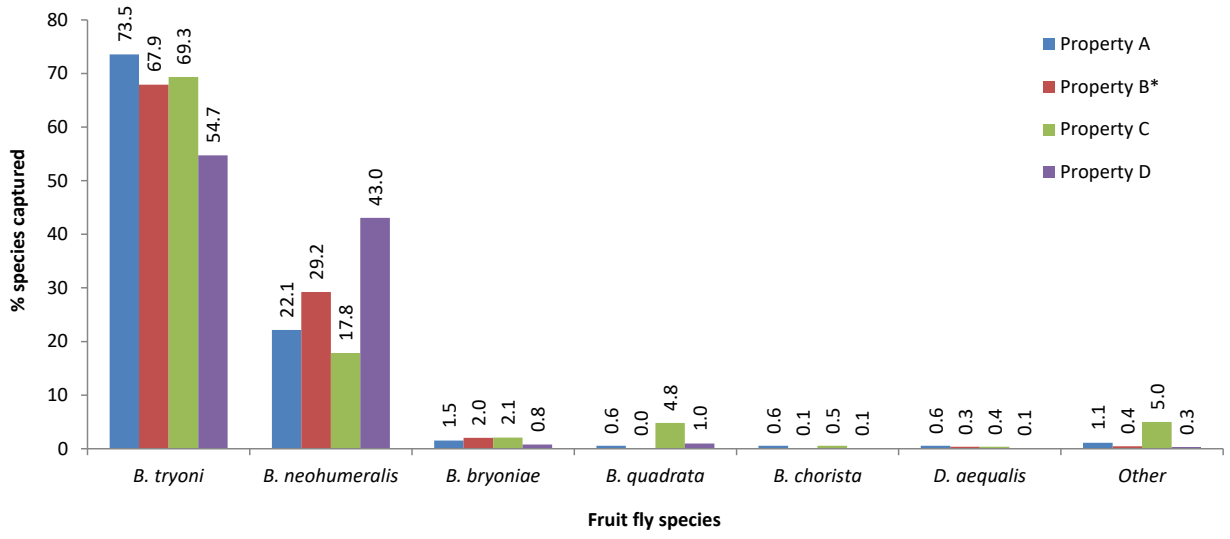

Figure 1 Percentage of male fruit fly of each species caught from cue-lure monitoring traps at each of four vegetable farms between July 2014 and June 2015. Monitoring at Property B was discontinued in February 2015 at the request of the property owner.

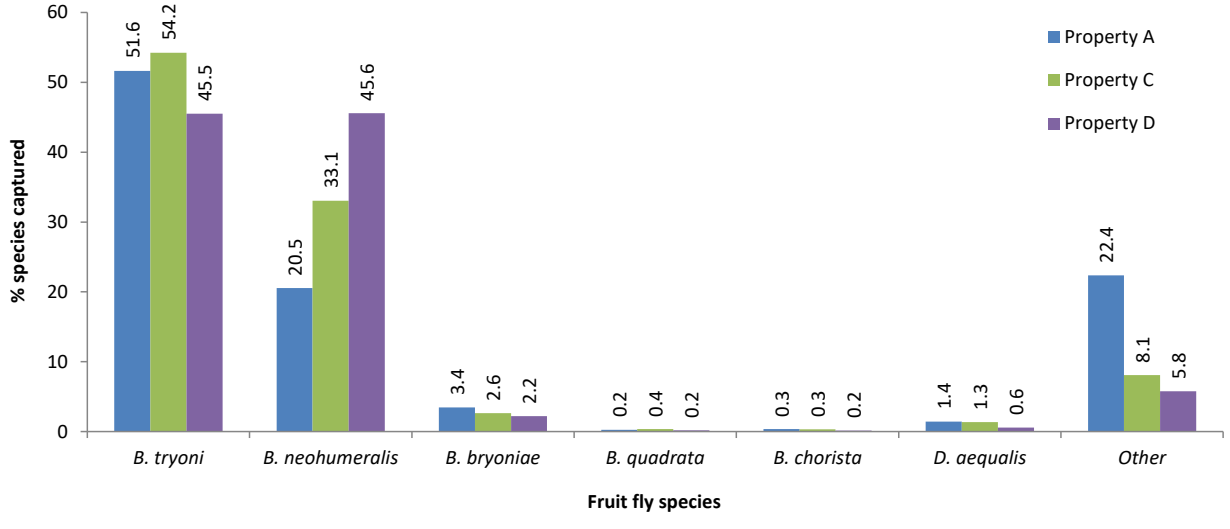

Figure 2 Percentage of male fruit fly of each species caught from cue-lure monitoring traps at each of three vegetable farms between July 2015 and June 2016. The large proportion of flies categorised as 'other' was due to a particularly large number of damaged specimens.

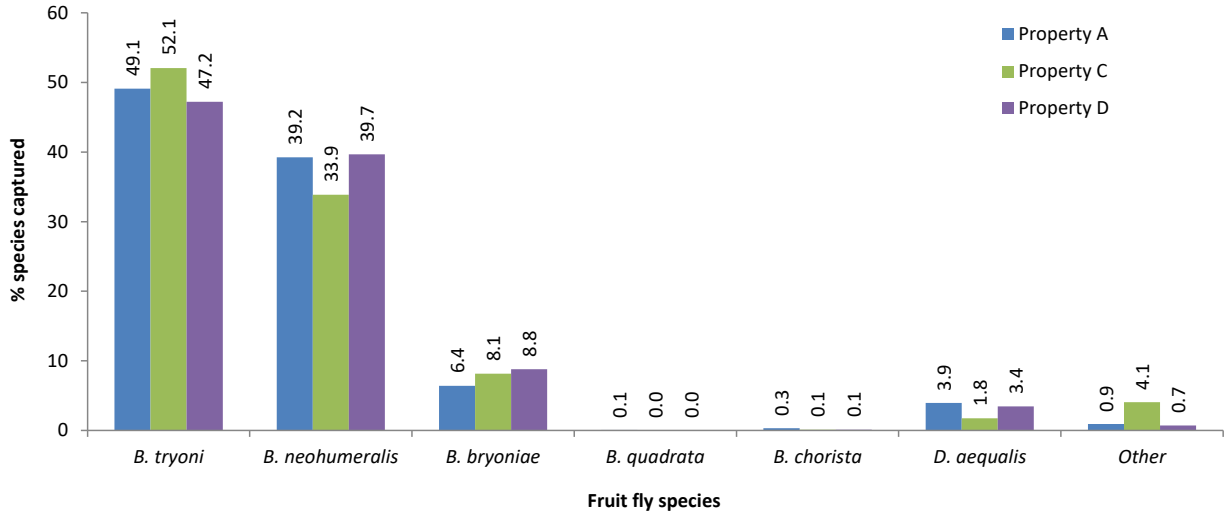

Figure 3 Percentage of male fruit fly of each species caught from cue-lure monitoring traps at each of three vegetable farms between July 2016 and March 2017

Average trap catch (Queensland fruit fly and lesser Queensland fruit fly) per day varied substantially with season, between years and between properties (Figures 4-6; Y axis truncated on Figures 4 and 5 to enable comparison between the three years). However, the trend indicated a peak in trap catch in spring, and a second peak later in the season in the summer. The spring peak was particularly pronounced at Property D in 2014 and 2015. The reason for much lower spring trap catches at Property D in 2016 is not clear, but may be linked to a greater proportion of the land planted to tomato than capsicum in this year. Likewise, differences in trap catches between properties could be due to a number of reasons, such as differences in landscape (e.g. presence of alternative hosts or bodies of water), differences crop management and in area planted to susceptible crops. Much larger data sets from different locations would be required in order to understand this relationship, which is beyond the scope of this project.

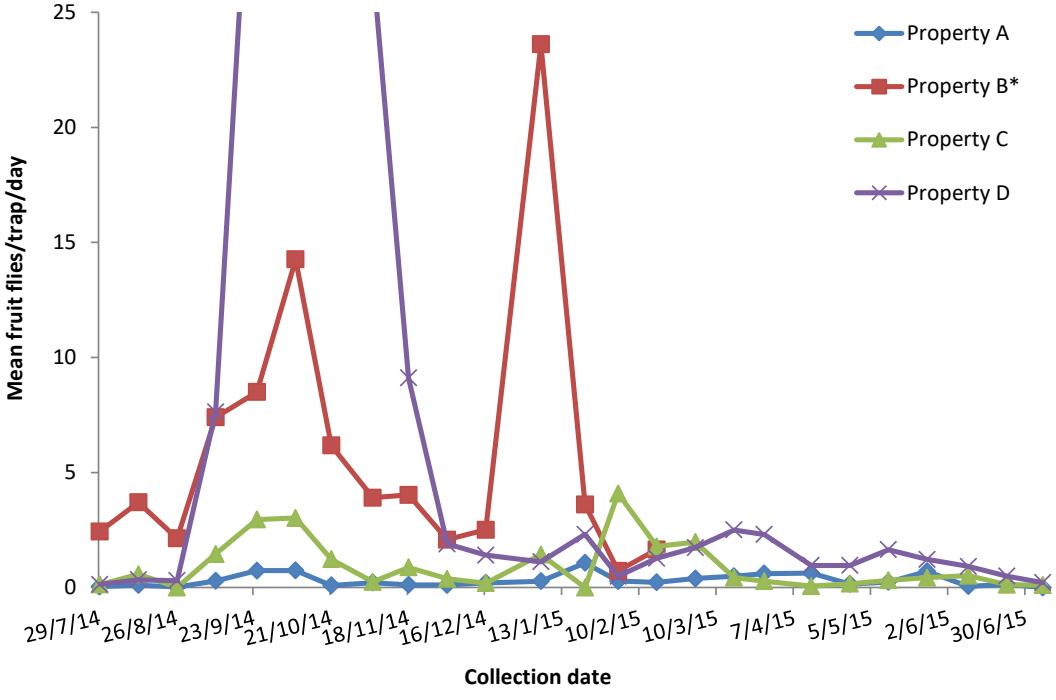

Figure 4 Male Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) trap catches from cue-lure monitoring traps at each of four vegetable farms between July 2014 and June 2015. Monitoring at Property B was discontinued in February 2015 at the request of the property owner. Trap catches at Property D between 24th September and 5th November were 38, 100, 50 and 27 flies per trap per day (Y axis truncated to show detail at other locations).

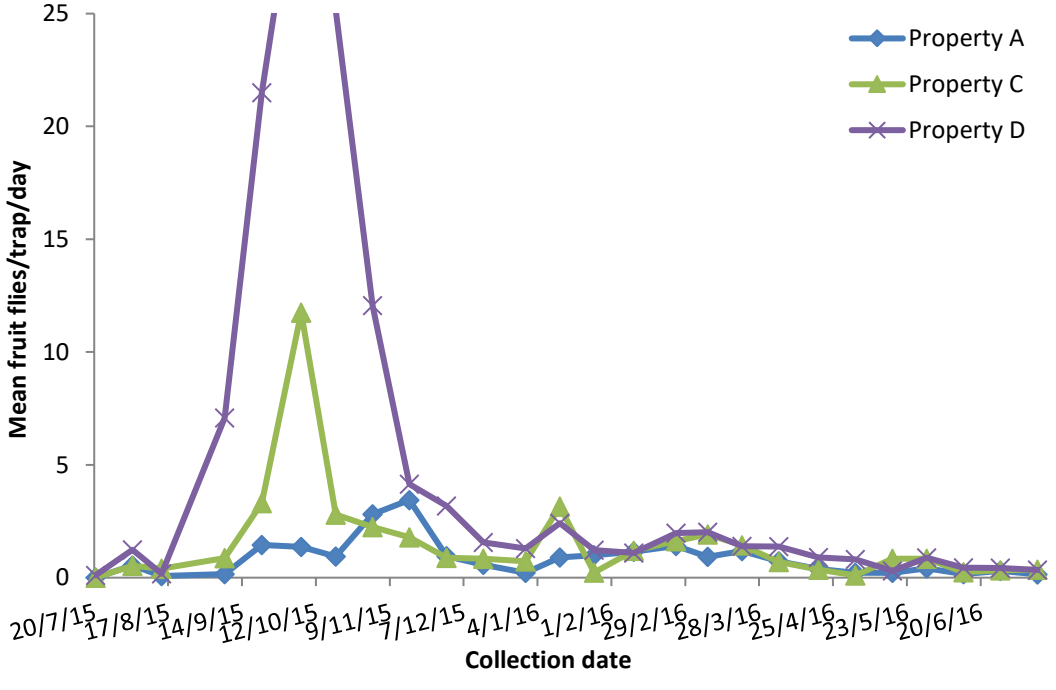

Figure 5 Male Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) trap catches from cue-lure monitoring traps at each of three vegetable farms between July 2015 and June 2016. Trap catch at Property D on 6th October was 33 flies per trap per day (Y axis truncated to show detail at other locations).

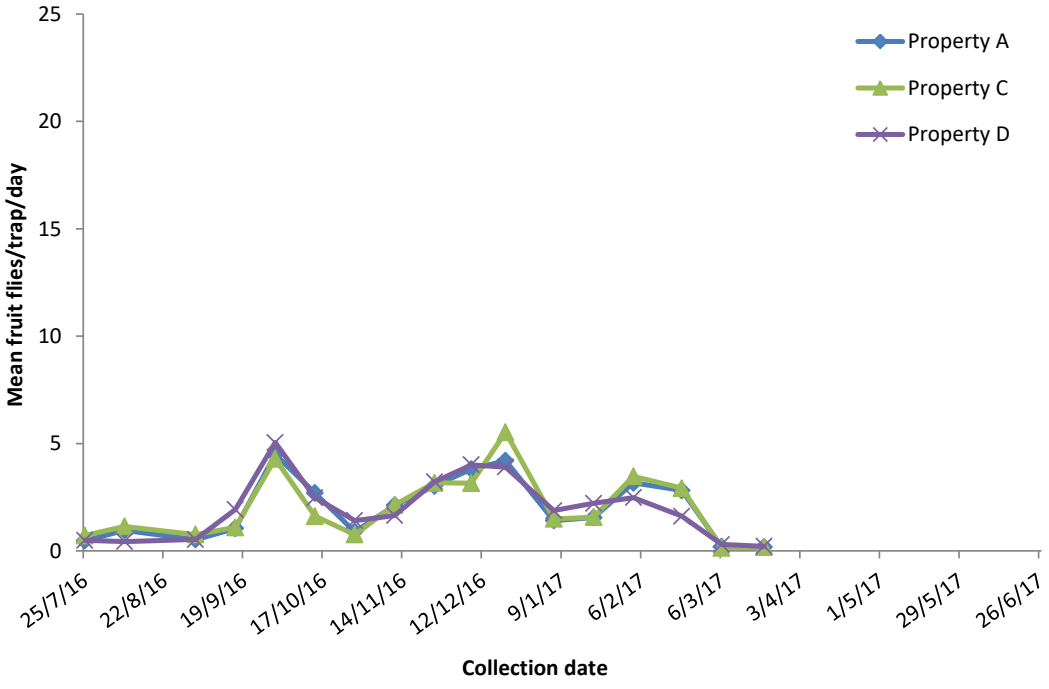

Figure 6 Male Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) trap catches from cue-lure monitoring traps at each of three vegetable farms between July 2016 and March 2017.

In the first year, traps at Property B and Property D were placed in different locations: on a tree-line, fence-line and in a capsicum crop (Property B), or on a tree-line and in a capsicum crop (Property D) (Table 1). Traps were also placed in a tomato crop at Property D however these failed to catch any flies before the crop was finished and removed in September 2014. At both properties, higher numbers of flies were caught in traps located along a tree-line compared to those within a capsicum crop or on a fence-line (Figures 15 and 16). Trapping at Property B was discontinued in February 2015 and traps at Property D were all relocated to the tree-line in November 2014. It should be noted that the placement of traps in different locations is unlikely to be responsible for the overall differences in trap catch between the different properties, as these differences were also observed after all traps had been relocated to the tree-line at all properties.

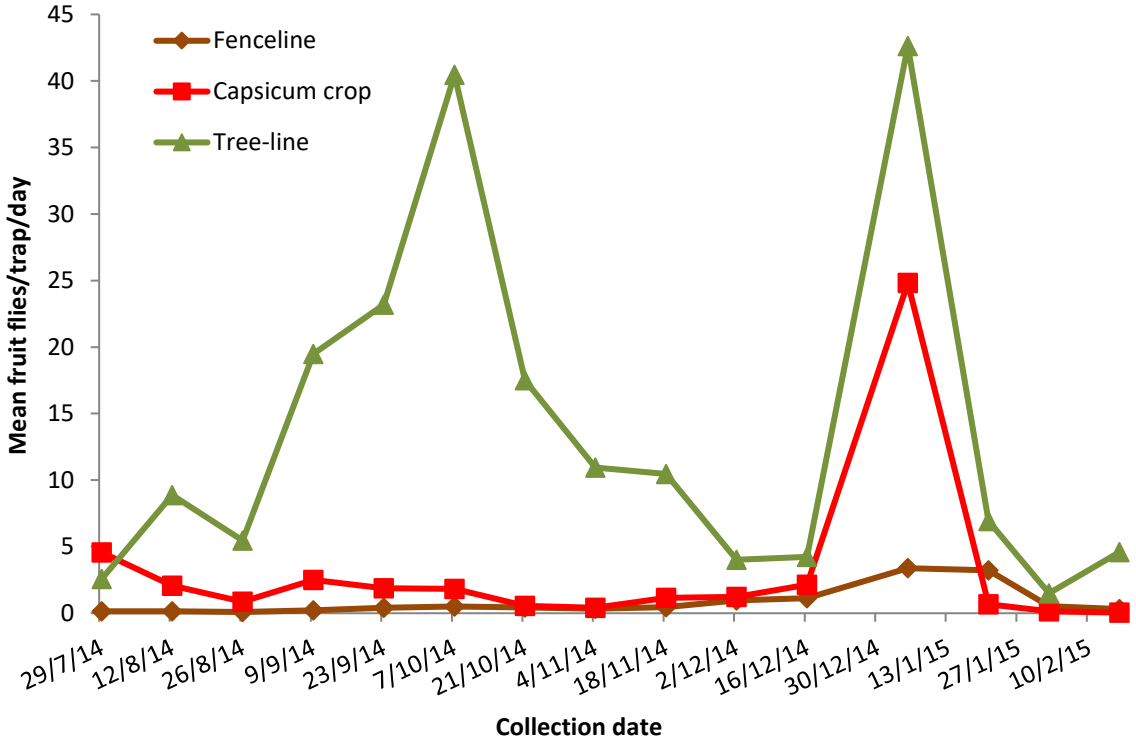

Figure 7 Male fruit fly trap catches from cue-lure monitoring traps placed in three locations at Property B

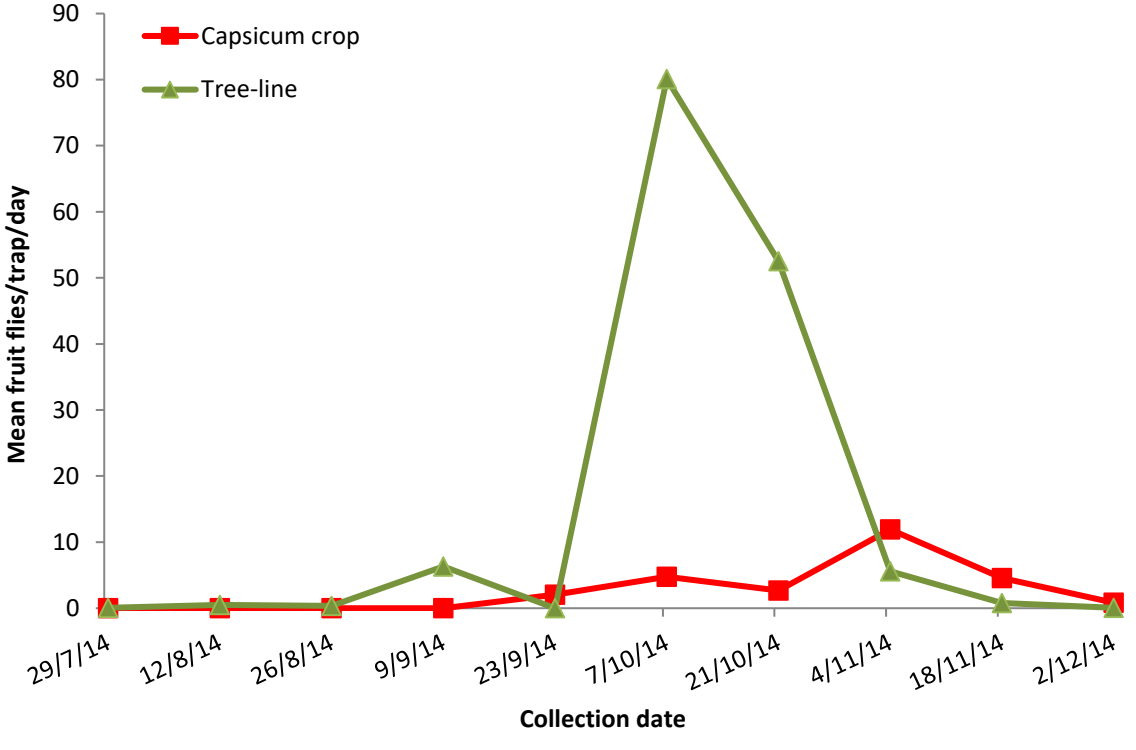

Figure 8 Male fruit fly trap catches from cue-lure monitoring traps placed in two locations at Property D

The percentage of *B. tryoni* and *B. neohumeralis* (excluding other species) was found to vary between farms and between years (Figure 9).

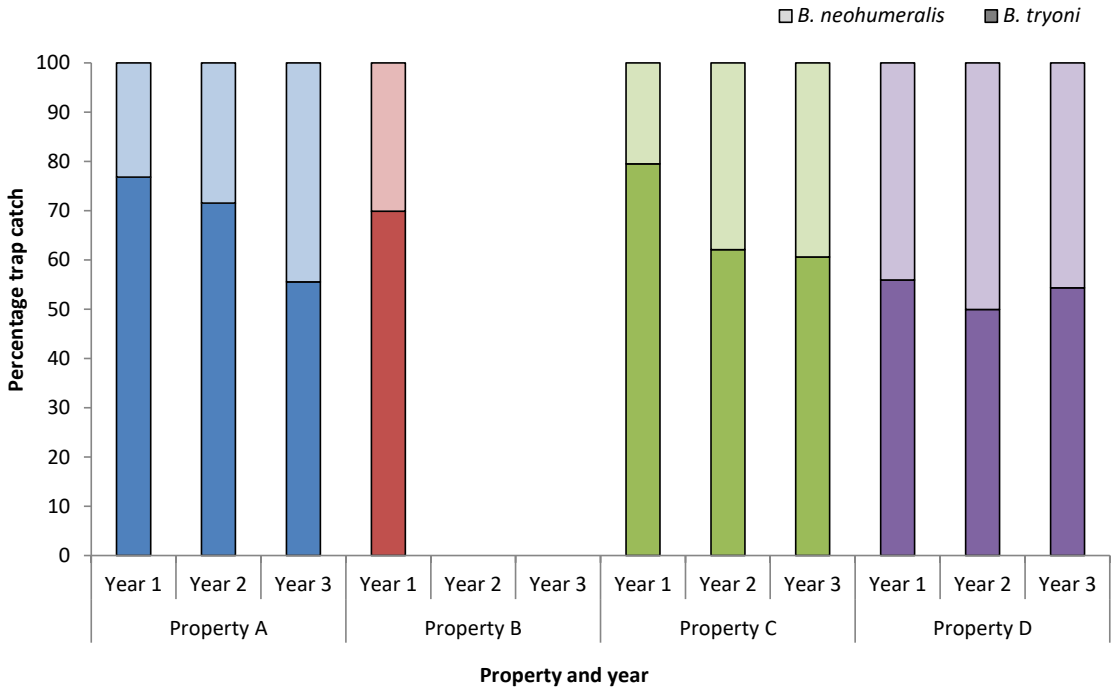

Figure 9 Percentage of *B. tryoni* and *B. neohumeralis* males trapped over each year of monitoring at each of four vegetable farms

Examination of numbers of trapped *B. tryoni* and *B. neohumeralis* over time shows that the two species generally followed a similar trend (Figures 10-13; Y axis truncated on Figures 11 and 13 to show detail). However, there were times when peaks in *B. tryoni* were observed, with no similar corresponding peak in *B. neohumeralis* trap catch.

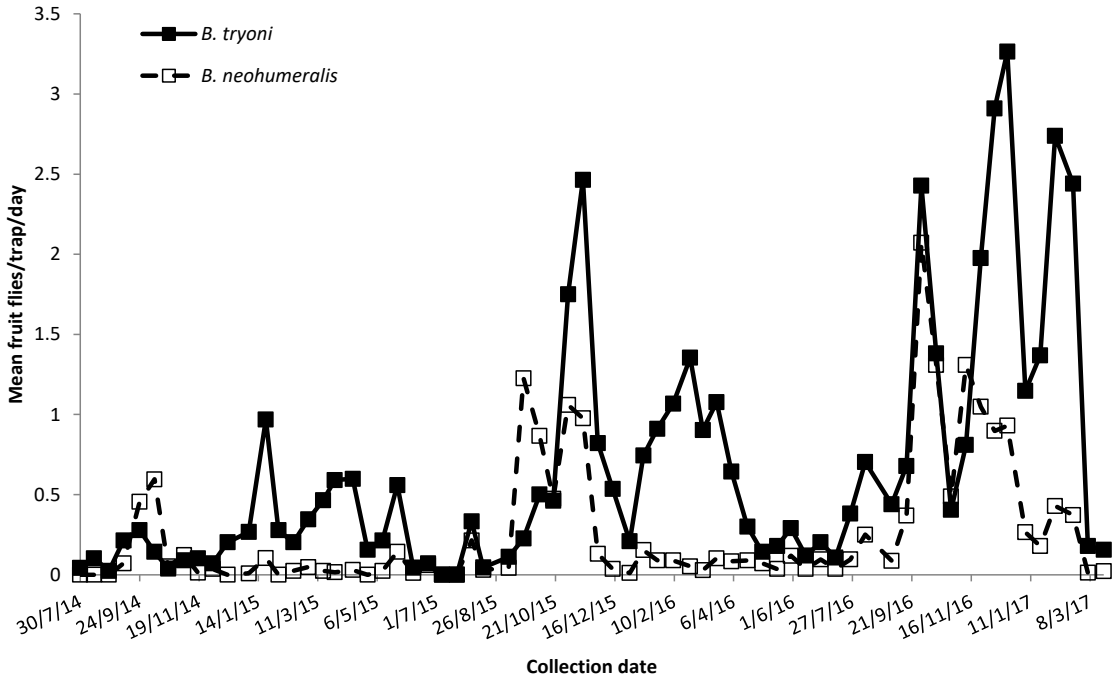

Figure 10 Trap catches of *B. tryoni* and *B. neohumeralis* males from cue-lure monitoring traps at Property A over three consecutive years.

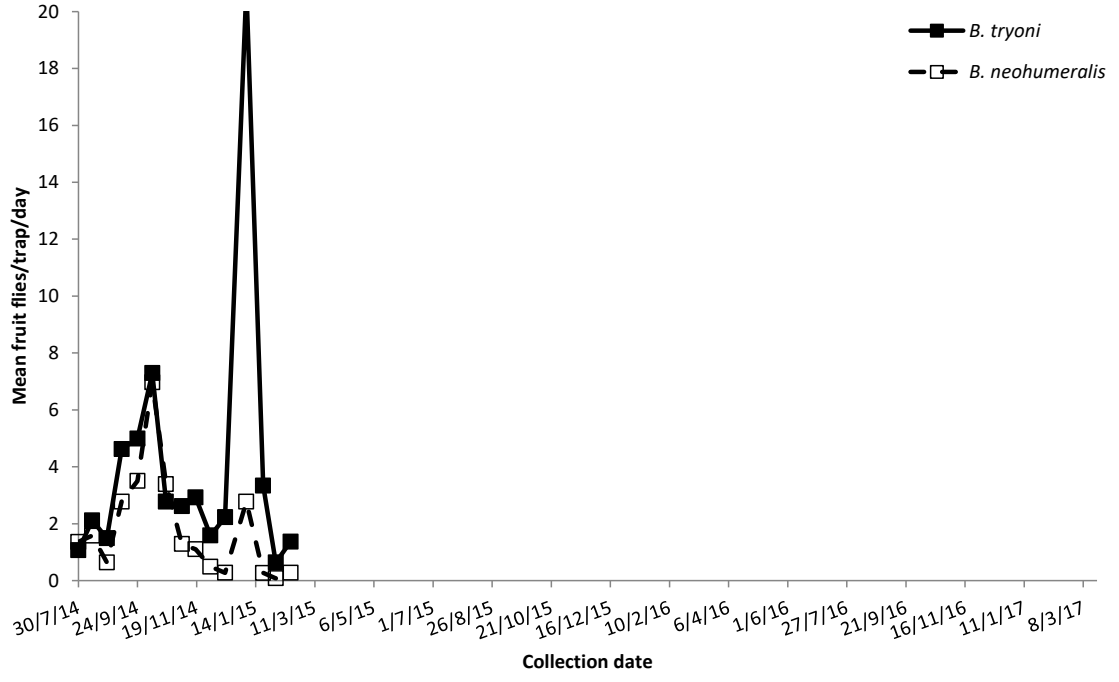

Figure 11 Trap catches of *B. tryoni* and *B. neohumeralis* males from cue-lure monitoring traps at Property B. Y axis truncated to show detail.

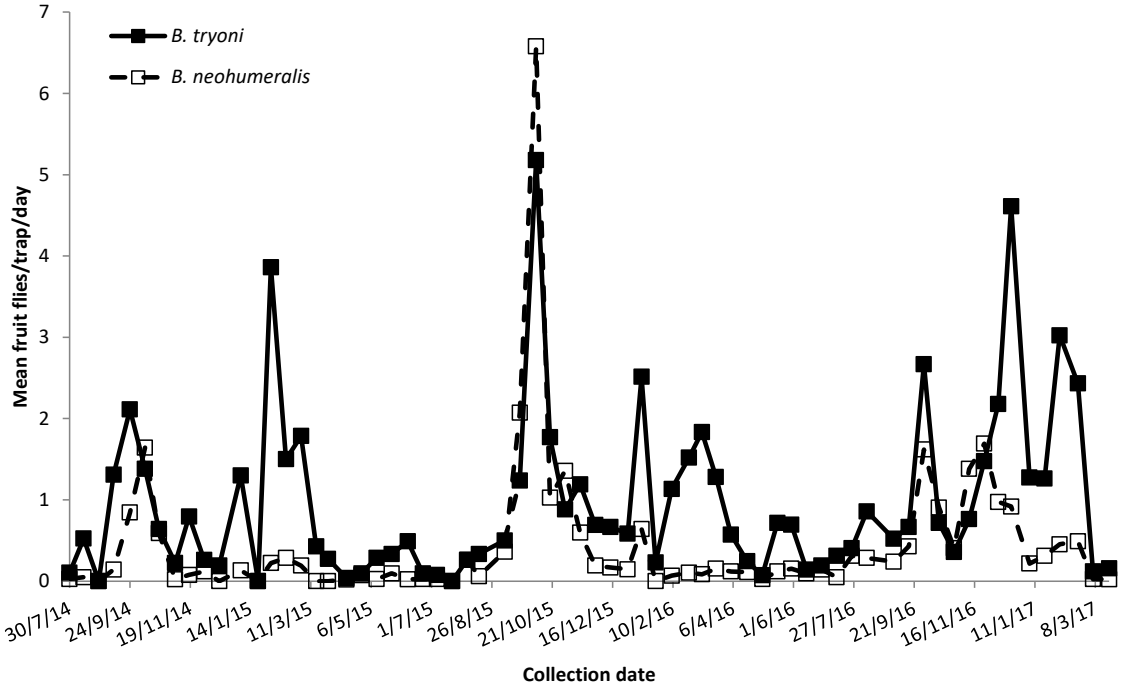

Figure 12 Trap catches of *B. tryoni* and *B. neohumeralis* males from cue-lure monitoring traps at Property C over three consecutive years.

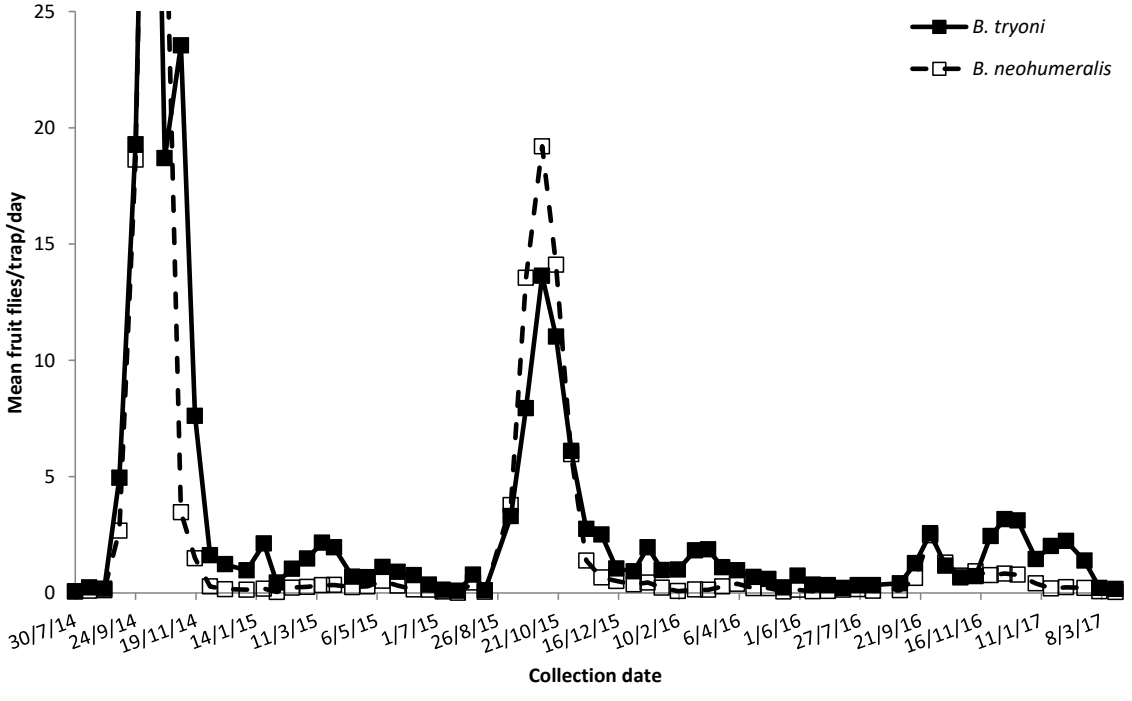

Figure 13 Trap catches of *B. tryoni* and *B. neohumeralis* males from cue-lure monitoring traps at Property D over three consecutive years. Y axis truncated to show detail.

Weather data for each year of monitoring are displayed in Figures 14-16.

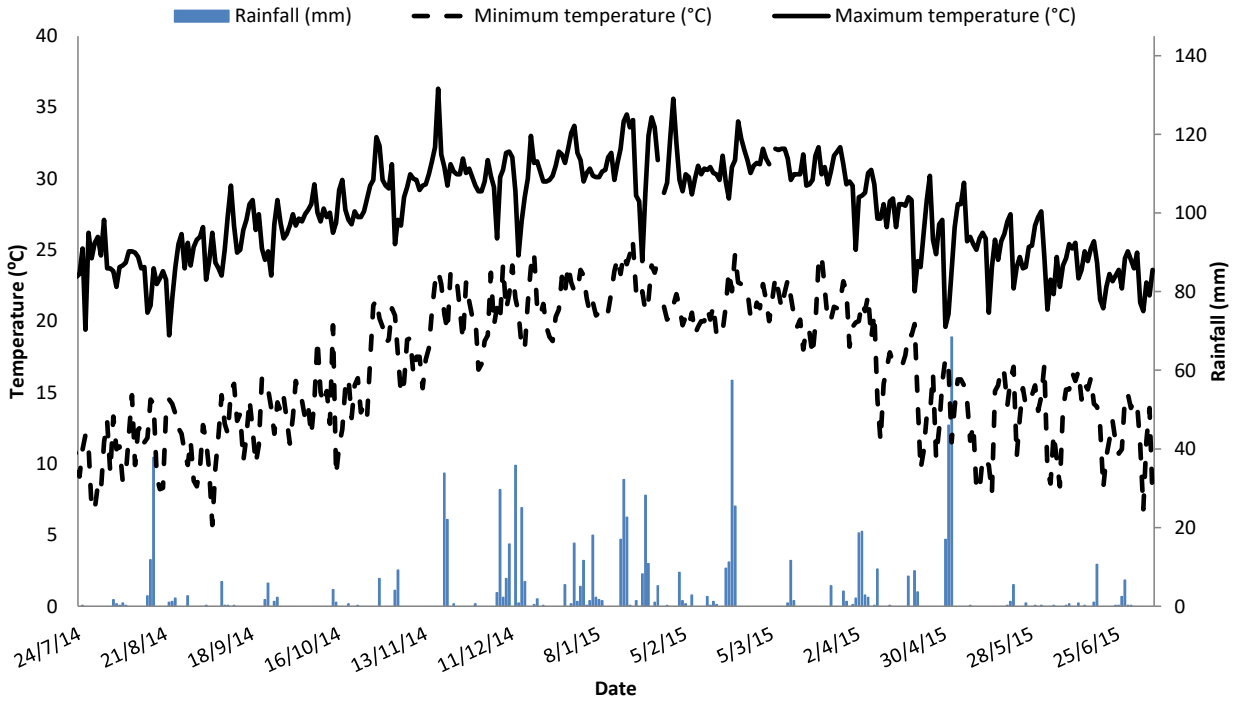

Figure 14 Weather data for July 2014 to June 2015. Data obtained from the Bureau of Meteorology website for the closest weather station (39128, Bundaberg Aero).

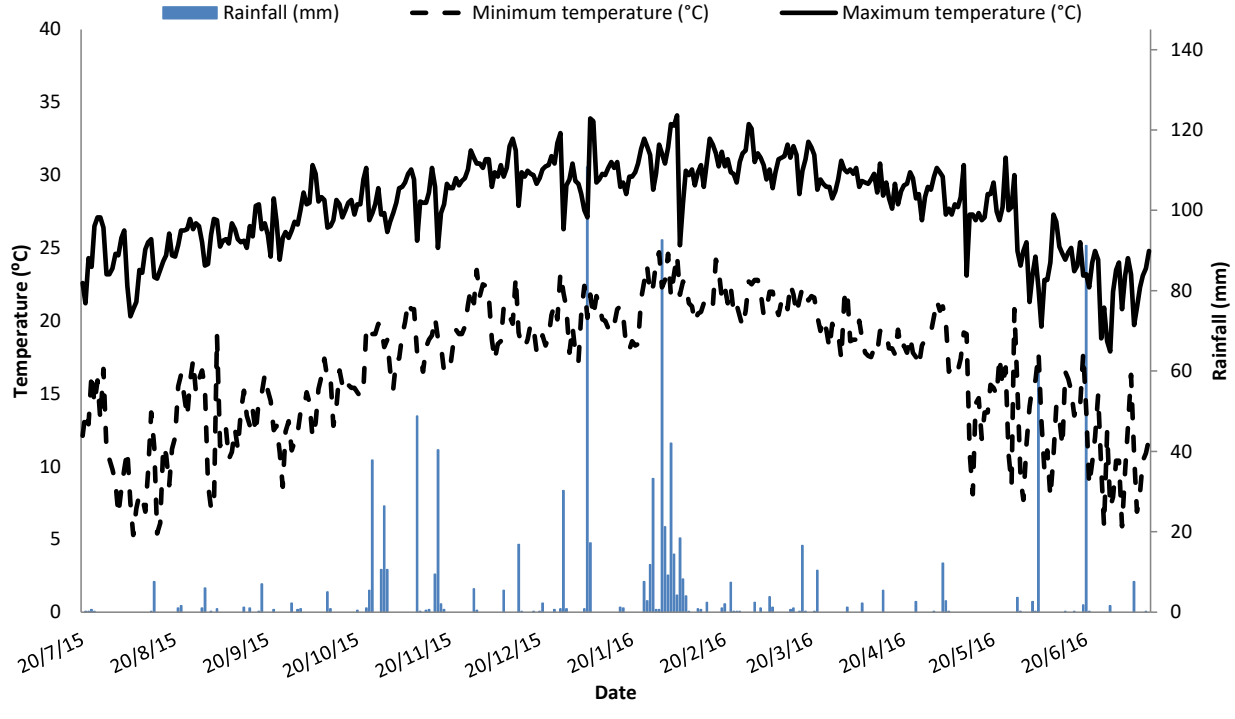

Figure 15 Weather data for July 2015 to June 2016. Data obtained from the Bureau of Meteorology website for the

closest weather station (39128, Bundaberg Aero).

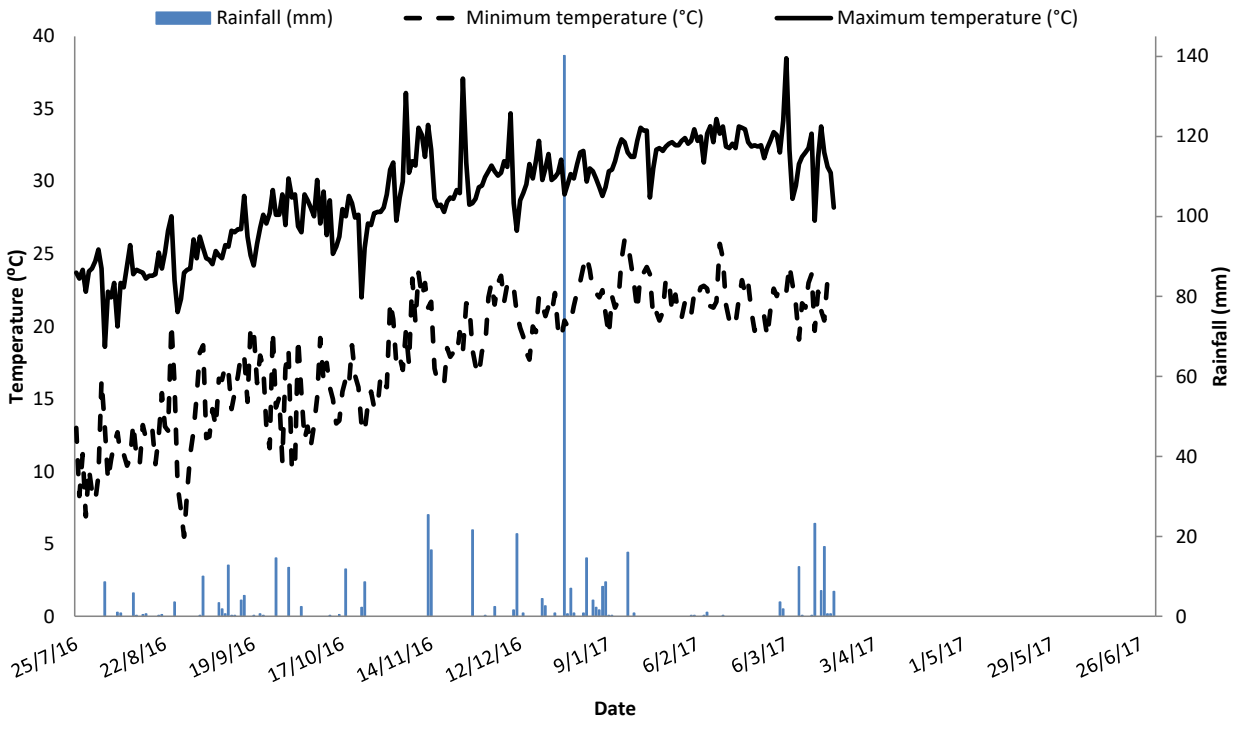

Figure 16 Weather data for July 2016 to March 2017. Data obtained from the Bureau of Meteorology website for the closest weather station (39128, Bundaberg Aero).

Cucumber volatile lure traps

A series of trials was performed using traps baited with a new cucumber volatile blend lure produced for melon fly, *Zeugodacus cucurbitae* (Scentry Biologicals Inc, Montana, USA), in order to monitor the activity of cucumber fly, *Z. cucumis*. In all trials, the cucumber volatile lure was placed into a small mesh bag suspended within the trap (Plate 3). A dichlorvos block was used as the toxicant, placed within a Petri dish on the base of the trap. Traps were hung from wire hoops at a height of approximately 30 cm above the ground (Plate 3).

Trial 1. Trapping conducted with the cucumber volatile lure in 2013-2014 for project MT12050 (Farm-wide fruit fly management systems for the east coast of Australia), suggested that although the lure was attractive to cucumber flies, the type of trap used (Bugs for Bugs Pty Ltd, Mundubbera, QLD) did not appear to be suitable. Therefore in December 2014 a trial was performed to compare the Bugs for Bugs trap with an alternative design (BioTrap Australia Pty Ltd, Ocean Grove, VIC) (Plate 3). Five traps of each type were placed in and around a commercial pumpkin crop and an adjacent home garden cucurbit crop on 4th December 2014 (Wallaville, QLD), and trap contents examined seven days later. The Bugs for Bugs traps caught an average of 0.7 cucumber flies per trap per day, compared with an average of 9.1 cucumber flies per trap per day caught by BioTraps. The average sex ratio was 74:26 female:male. This confirms the female-biased attraction observed by Royer et al (2014), who found that traps baited with the cucumber volatile lure caught between 60% and 95% females. In comparison, unpublished data collected by the DAF Market Access Team shows the sex ratio of flies emerging from cucurbits collected from the field, to establish cucumber fly colonies, to be closer to 50:50.

Trial 2. In May 2015 a further trial was carried out in a commercial planting of pumpkins (Kalbar, QLD) to compare the Bugs for Bugs trap with the BioTrap. Three traps of each type were placed in the pumpkin crop on 22nd May 2015, and the trap contents examined six days later. Unfortunately, despite the observed presence of cucumber fly in the crop, only one individual was trapped and therefore the comparative efficacy of the trap types could not be compared.

Trial 3. In October 2015, BioTraps were placed in a newly planted commercial crop of pumpkins (Kalbar, QLD). The aim was to monitor traps regularly to observe the increase in cucumber fly population as the crop matured, and then compare trap catches in different locations within the field. However, only one fly was trapped.

Due to the poor trap catches, trapping with the cucumber volatile blend lure was discontinued.

Plate 3 Traps used to monitor cucumber fly. From left to right: cucumber volatile lure suspended within the trap within a mesh bag; Bugs for Bugs trap; BioTrap

References

Royer, J.E., De Faveri, S.G., Lowe G.E. & Wright, C.L. 2014. Cucumber volatile blend, a promising female-biased lure for *Bactrocera cucumis* (French 1907) (Diptera: Tephritidae: Dacinae), a pest fruit fly that does not respond to male attractants. *Austral Entomology* **53**, 347-352.

Appendix 6 Activity of fruit flies within the vegetable crop

Three trials were performed to further examine the activity of Queensland fruit fly within plantings of capsicum and tomatoes. Monitoring traps (BioTrap Australia Pty Ltd, Ocean Grove, VIC) were installed at various locations at Property D (property details in Appendix 5). For all trials, pairs of traps were deployed, baited with either cue-lure (which targets male fruit flies) or BioTrap Fruit Fly Attractant Gel (BioTrap Australia Pty Ltd) (which is claimed to attract both male and female fruit flies). Traps were examined weekly, the contents emptied into labelled containers, and sent to the DAF laboratories in Brisbane for identification of trapped flies.

Trial 1: traps were placed in the tree-line on the perimeter of the planting, and in adjacent plantings of tomatoes (ca. 65 m from the tree-line) and capsicums (ca. 120 m from the tree-line) (Plate 1). At each of these positions there were four traps, two baited with cue-lure and two baited with BioTrap Fruit Fly Attractant Gel, lure type alternating, spaced approximately 15 m apart. Traps were installed on 17th August 2016, and the first sampling occurred on 9th September 2016, at which point the tomato crop was fruiting and the capsicum crop was not yet flowering. The tomato crop was harvested and traps removed in early October, hence from 13th October onwards collections were only made from traps in the tree-line and capsicum crop. Fruit were present in the capsicum crop from late October onwards. The final sampling occurred on 2nd December 2016.

Trial 2: traps were placed in the tree-line at either end of a planting of capsicums (western tree-line and eastern tree-line), and within the capsicum crop at varying distance from the eastern edge of the crop: 3 m, 80 m, 130 m, 180 m (Plates 2 and 3). The eastern tree-line was approximately 18 m to the edge of the crop, and the western tree-line approximately 26 m to the edge of the crop. As in trial 1 there were two of each lure type at each position, a minimum of 6 m between traps. All except the 3 m traps were installed on 11th October and the first sampling occurred on 13th October. The 3 m traps were installed on 25th October and monitoring commenced on 28th October. The final sampling occurred on 30th December 2016. Plants were immature at the start of sampling, fruit were present from mid-November onwards and ripe fruit from late November onwards.

Trial 3: traps were placed as for trial 2, but with additional traps at 254 m from the eastern tree-line and at 3 m from the western tree-line. Traps were installed on 30th January 2017 and sampling occurred weekly from 6th January to 27th March 2017. Plants were immature at the start of sampling, fruit were present from late February/early March onwards and ripe fruit from mid-March onwards.

Plate 1 Trial 1 showing traps placed in capsicum crop, tomatoes to the left and tree-line to the far left

Plate 2 Site used for trials 2 and 3 showing the tree-line to the west and east of the crop

Plate 3 Traps placed in a capsicum crop at varying distances from the tree-line, trial 2.

Results and discussion

The majority of flies trapped in cue-lure baited traps were Queensland fruit fly and lesser Queensland fruit fly.

Table 1 Total catches over trapping period in each trial from traps baited with cue-lure. Note trial period varied for each trial.

Trial	All fruit fly species	*B. tryoni* & *B. neohumeralis*
1	2576	2411
2	2862	2695
3	4986	3565

In trial 1, traps placed in the tree-line caught the most flies, followed by those in the capsicum crop, with very few trapped in the tomato crop (Figure 1). In trial 2, the highest trap catches were from traps in the tree-lines and the traps in the crop closest to the tree-line (3 m into the crop) (Figure 2). The start of fruiting did not appear to affect trap catch in this trial. Results from trial 3 were similar over the first few weeks of sampling, with traps on the tree-line or in the crop close to the tree-line catching the most flies (Figure 3). However, from the start of fruiting trap catch increased substantially and traps within the crop caught more flies.

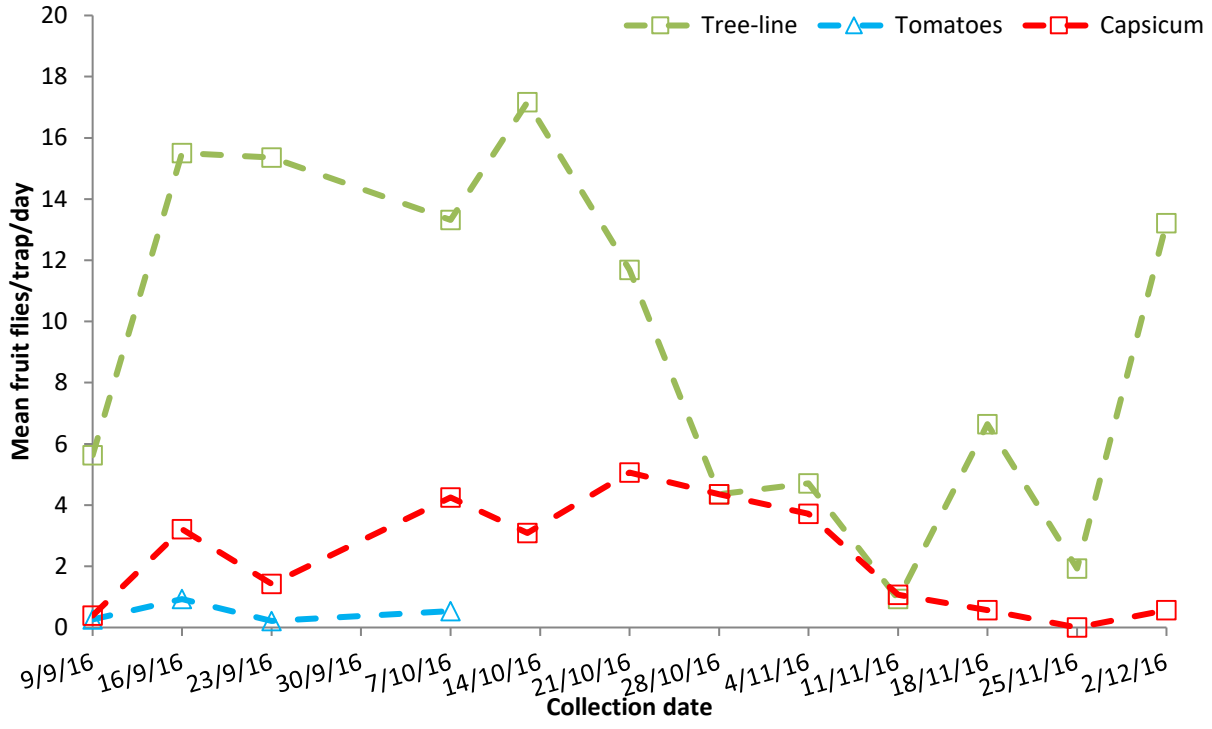

Figure 1 Male Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) trap catches from cue-lure baited monitoring traps placed in a tree-line and an adjacent tomato or capsicum crop, trial 1.

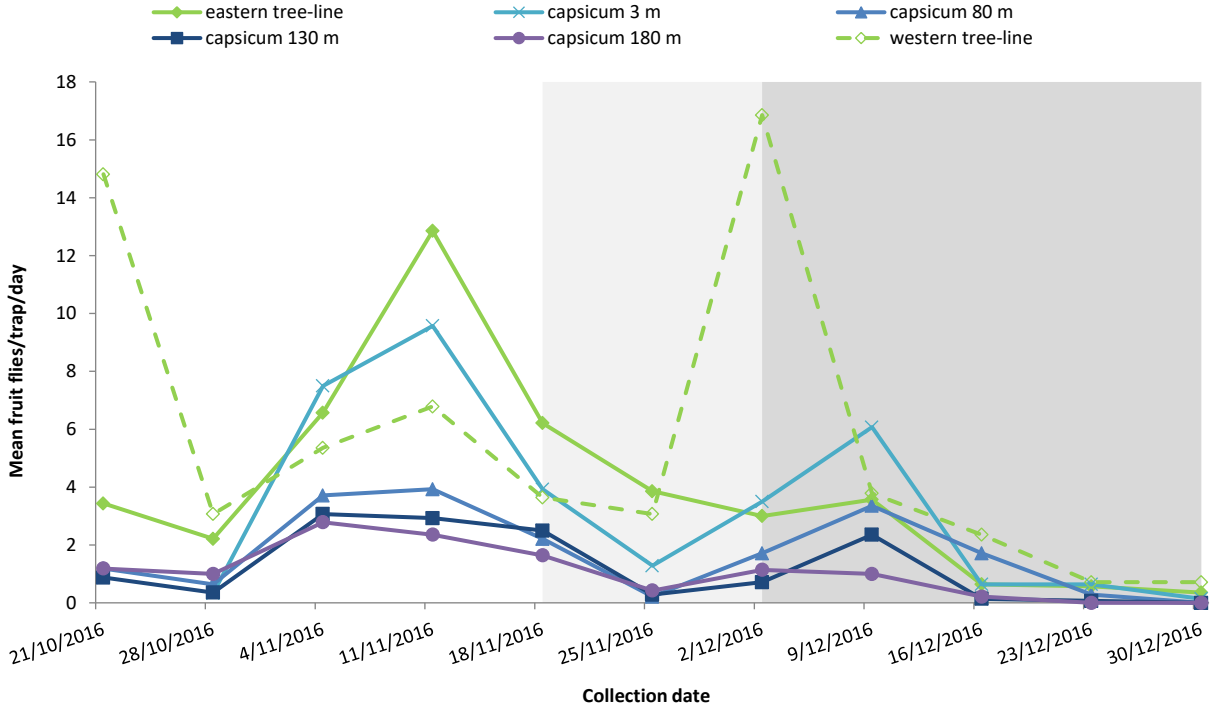

Figure 2 Male Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) trap catches from cue-lure baited monitoring traps placed in a tree-line or an adjacent capsicum crop, trial 2. Fruit were present from mid-November (indicated by light grey shading) and ripe fruit from late November (dark grey shading).

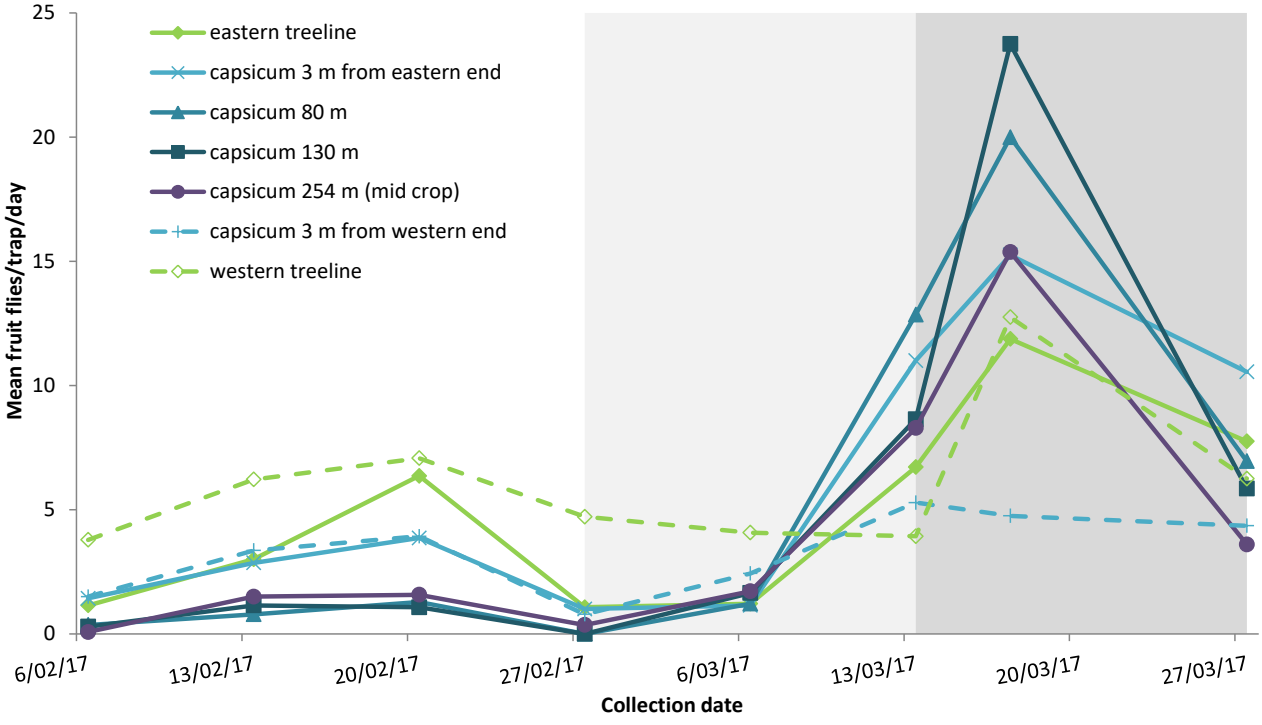

Figure 3 Male Queensland fruit fly (*B. tryoni* and *B. neohumeralis*) trap catches from cue-lure baited monitoring traps placed in a tree-line or an adjacent capsicum crop, trial 3. Fruit were present from late February/early March (indicated by light grey shading) and ripe fruit from mid-March (dark grey shading).

Trap catch from all BioTrap gel baited traps was very low in all three trials (Table 2). Unlike cue-lure baited traps, they caught a mix of males and females, and a high proportion of fruit fly species other than *B. tryoni* and *B. neohumeralis*. They also caught flies other than fruit flies, including *Drosophila* sp., *Lamprolonchaea* sp. and *Atherigona* sp. Due to the low trap catches in traps baited with BioTrap Fruit fly Attractant Gel, these results were not analysed further.

Table 2 Total catches over the trapping period in each trial, from traps baited with BioTrap Fruit Fly Attractant Gel

Trial	All fruit fly species	*B. tryoni* & *B. neohumeralis*
1	41 (25♀, 6♂, 10 unsexed)	27
2	107 (59♀, 40♂, 8 unsexed)	32
3	98 (67♀, 29♂, 2 unsexed)	85

Appendix 7 Paper accepted for publication in Journal of Economic Entomology

Journal of Economic Entomology, 00(0), 2017, 1–11
doi: 10.1093/jee/tox109
Research article

Comparative Efficacy of Insecticides on *Bactrocera tryoni* and *Zeugodacus cucumis* (Diptera: Tephritidae) in Laboratory and Semifield Trials in Fruiting Vegetables

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Subject Editor: Anthony Clarke

Received 24 January 2017; Editorial decision 19 March 2017

Abstract

In-field management of *Bactrocera tryoni* (Froggatt) and *Zeugodacus cucumis* (French) (Diptera: Tephritidae) in fruiting vegetable crops has relied almost exclusively on organophosphate cover sprays. Laboratory and semifield trials were performed to compare a number of alternative insecticides for efficacy against these species. A novel semifield method was used whereby the insecticides were applied to crops as cover sprays under field conditions, and treated plants bearing fruit were transferred to large cages and exposed to fruit flies. Efficacy was assessed in terms of numbers of pupae developing from treated fruit. A laboratory cage method was also used to assess effects on adult mortality and comparative effects of 1- and 3-d-aged residues. The neonicotinoids clothianidin and thiacloprid were very effective against *B. tryoni* and *Z. cucumis*. Clothianidin was the only insecticide other than dimethoate to affect adult mortality. The synthetic pyrethroid alpha-cypermethrin was also very effective, particularly in semifield trials, although higher incidence of aphid and whitefly infestation was observed in this treatment compared to others. Cyantraniliprole was effective against *B. tryoni*, but less effective against *Z. cucumis*. Imidacloprid, bifenthrin, spinetoram, and abamectin were all relatively less effective, although all demonstrated a suppressive effect.

Key words: *Bactrocera tryoni*, *Zeugodacus cucumis*, Tephritidae, insecticide

Tephritid fruit flies are serious pests of horticultural crops in Australia. *Bactrocera tryoni* (Froggatt) attacks a wide range of fruit and vegetable crops in eastern Australia (Drew et al. 1982, Hancock et al. 2000), causing crop loss and threatening market access. There have been increasingly frequent incursions of this pest into the Fruit Fly Exclusion Zone in the southern states of Australia (Dominiak et al. 2015). *Zeugodacus cucumis* (French) causes damage to cucurbit crops and tomatoes in Queensland, northern New South Wales, the Northern Territory, and Torres Strait Islands (Drew et al. 1982, Hancock et al. 2000).

In Australia, in-field management of fruit flies in fruiting vegetable crops has relied almost exclusively on organophosphate cover sprays. However, recent restrictions in the use of dimethoate (Australian Pesticides and Veterinary Medicines Authority [APVMA] 2011, APVMA 2017) and fenthion (APVMA 2015) have greatly limited the available control options. While the use of dimethoate for control of fruit fly in certain fruiting vegetable crops has been retained (e.g. capsicum, melons, zucchini), for others it has been suspended from use, or the withholding period extended rendering it largely ineffective. Alternative approaches for managing fruit fly in host crops have been explored, such as the use of toxic baits and male annihilation technique (Clarke et al. 2011, Vargas et al. 2015). However, development of these management methods has primarily been focused on tree crops, or exotic tephritid species in vegetable crops. For instance, perimeter baiting, in which a protein bait plus insecticide is applied to a nonhost plant adjacent to the crop, is a commonly used technique for management of melon fly, *Zeugodacus cucurbitae* (Coquillett) in Hawaii (McQuate 2011). It exploits the observation that *Z. cucurbitae* females roost and forage for protein in certain favoured nonhost plants (Nishida and Bess 1957, Prokopy et al. 2003). The efficacy of toxic baits for control of *B. tryoni* and *Z. cucumis* in fruiting vegetable crops is currently unproven. Moreover, vegetable crops are often subject to intensive insecticide regimes for management of other pest species, and it is important to understand the potential impact of these insecticides on fruit flies.

A number of studies have examined the toxicity of pesticides to tephritid fruit flies in the laboratory using a range of methods such as direct application to adult flies (Wang et al. 2013), exposure to residues on artificial substrates (Mosleh et al. 2011), exposure to residues on fruit applied and aged in the laboratory (Maklakov et al.

2001, Yee 2008, Yee and Alston 2012) or the field (Yee et al. 2007, Rahman and Broughton 2016), or application of insecticide sprays to infested fruit (Wise et al. 2009). Small plot field trials have been employed to assess efficacy of insecticides for management of cucurbit-specific fruit flies such as *Z. cucurbitae* in cucurbits (Oke 2008, Khursheed and Raj 2012, Oke and Sinon 2013), and field trials conducted in tree crops for management of *Rhagoletis* spp. (Reissig 2003, Yee and Alston 2006). However, the literature on insecticide efficacy for *B. tryoni* and *Z. cucumis* is scarce. Reynolds et al. (2014) compared insecticides for efficacy against *B. tryoni* in the laboratory and in semifield trials in stonefruit, in which fruit flies were introduced into mesh sleeves enclosing treated fruit on peach trees. Subramaniam (2013) evaluated insecticide cover sprays for management of *B. tryoni* in eggplant; however, efficacy formed part of a systems approach and was not compared with a control. Kay (2004) assessed efficacy of insecticides against *B. tryoni* in a small plot field trial in capsicum, but failed to find significant differences amongst treatments due to high variability of infestation across replicate plots. Atuahene and Hooper (1971) investigated the susceptibility of *B. tryoni* and *Z. cucumis* to DDT, with no recent literature relating to the efficacy of insecticides against the latter species.

Laboratory trials can provide useful information on relative efficacy of insecticides but are limited by their artificial nature (Macfadyen et al. 2014). When insecticides are applied directly or insects are confined in close contact with residues, repellent effects cannot be measured. Laboratory trials often provide no information on the insecticide's performance in the field, such as resistance to weathering, movement within the plant, or the effect of nonuniform application.

Small plot field trials are the conventional method used for many horticultural pests to assess comparative efficacy of insecticides under field conditions. However, there is evidence that because *B. tryoni* invades low growing crops from the field margins, this results in a typically uneven infestation in such crops. Balagawi et al. (2014) recorded higher numbers of male *B. tryoni* in traps placed in vegetation bordering a strawberry crop than traps within the crop, and from the same study Gu (2010) found rates of infestation were correspondingly higher in fruit near the border than within the crop. This makes it difficult to compare treatments within a trial area due to large variability between treatments and replicates, as observed in the small plot field trial performed by Kay (2004) to compare cover sprays for control of *B. tryoni* in capsicum. Kay (2004) speculated that the higher infestation in certain plots was due to proximity to bordering trees and a citrus block. Steiner and Hinman (1952) encountered similar difficulties in small plot tests in tree crops, noting that populations of oriental fruit fly, *Bactrocera dorsalis* (Hendel), in control plots were depressed by nearby treated plots, and that greater movement of fruit flies into the windward side of the trial area resulted in larger infestations in these plots.

Trials were carried out to compare insecticides as cover sprays for efficacy against *B. tryoni* and *Z. cucumis* under semifield conditions. The aims were twofold: first, to obtain information on the comparative efficacy of a series of insecticides, and second, to assess a novel semifield methodology. The insecticides were applied to crops in the ground, and therefore subject to actual use conditions. An artificial infestation method was then used to ensure all fruits from all treatments were subjected to a similar fruit fly pressure.

Materials and Methods

Small Plot Trial Layout

Semifield trials were conducted over two years to evaluate the efficacy of a range of insecticides against *B. tryoni* in capsicum, *Capsicum annuum* (commercial variety Warlock), and *Z. cucumis* in zucchini, *Cucurbita pepo* (commercial variety Congo F1). Trials were conducted from January to April 2014 (season one) and January to April 2015 (season two) at Gatton Research Facility (Lockyer Valley, QLD, Australia; 27° 32′ S, 152° 19′ E, elevation 98 m). Crops were planted in January into plastic mulch and irrigated using trickle tape. The trial layout was a randomized complete block design with seven treatments, replicated four times. Treatments were arranged lengthwise, with 2 m no-crop buffers between plots in the lengthwise direction, and 2 m no-crop buffers between replicate blocks. Between-plant spacings were 0.5 m for capsicum and 0.75 m for zucchinis. Each plot was on a 1.5-m-wide bed and plot length varied according to season and crop: season one 9 m (14 capsicum plants) or 9.5 m (11 zucchini plants); season two 10 m (17 capsicum plants) or 10.5 m (12 zucchini plants). Rows of forage sorghum on either side of the trial block provided protection from wind and potential spray drift. Fungicides were applied for disease control: Polyram DF (BASF Australia Ltd, Southbank, VIC), Kocide Blue Xtra (Du Pont (Australia) Pty Ltd, Macquarie Park, NSW), and Dithane Rainshield Neo Tec Fungicide (Dow AgroSciences (Australia) Ltd, Frenchs Forest, NSW). In season one, the insecticides Transform (Dow AgroSciences (Australia) Ltd; active ingredient sulfoxaflor) and Talstar 250 EC (FMC Australasia Pty Ltd, Murarrie, QLD; active ingredient bifenthrin) were applied for control of whitefly and aphids. Application of these insecticides to young plants was made prior to the application of the trial treatments, and was not expected to affect the fruit flies.

Treatments

In each trial, five insecticide treatments were assessed and compared with dimethoate (the industry standard) applied at the rate of 75 ml/100 liter and with an untreated control. In season one (2014) the five insecticide treatments were Sumitomo Samurai Systemic Insecticide (Sumitomo Chemical Australia Pty Ltd, Epping, NSW; active ingredient clothianidin) applied at 40 g/100 liter; Confidor 200 SC (Bayer CropScience Pty Ltd, Hawthorn East, VIC; active ingredient imidacloprid) at 25 ml/100 liter; Talstar 250 EC Insecticide/Miticide (FMC Australasia Pty Ltd, Murarrie, QLD; active ingredient bifenthrin) at 24 ml/100 liter; Fastac Duo Insecticide (BASF Australia Ltd, Baulkham Hills, NSW; active ingredient alpha-cypermethrin) at 55 ml/100 liter; and DuPont Benevia Insecticide (Du Pont (Australia) Pty Ltd, Macquarie Park, NSW; active ingredient cyantraniliprole) at 100 ml/100 liter. In season two (2015) the five insecticide treatments were Sumitomo Samurai Systemic Insecticide (Sumitomo Chemical Australia Pty Ltd; active ingredient clothianidin) applied at 40 g/100 liter and 30 g/100 liter; Calypso 480 SC Insecticide (Bayer CropScience Pty Ltd; active ingredient thiacloprid) at 37.5 ml/100 liter; Vertimec Miticide/Insecticide (Syngenta Australia Pty Ltd, Macquarie Park, NSW; active ingredient abamectin) at 60 ml/100 liter; and Success Neo Insecticide (Dow AgroSciences (Australia) Ltd; active ingredient spinetoram) at 40 ml/100 liter. Agral spray adjuvant (Syngenta Australia Pty Ltd) was added to all treatments at the rate of 10 ml/100 liter, with the exception of the Samurai treatments, where Maxx Organosilicone Surfactant (Sumitomo Chemical Australia Pty Ltd) was used at the rate of 50 ml/100 liter.

Insecticides were applied using a gas pressurised sprayer, with a 1.2-m four-nozzle boom. The spray was applied at an operating boom pressure of ~230 kPa and a volume of 700–760 liter/ha (dependent on crop and plot size), achieved through two passes of each plot in order to ensure good coverage. Treatments were applied weekly from first fruit set onwards, until all trials had been

Table 1. Dates of spray application and trials, season one (2014)

Date	Capsicum	Zucchini
18 Feb. 2014		Spray application
25 Feb. 2014	Spray application	Spray application
26 Feb. 2014		Laboratory cage trial one 1 DAT
4 Mar. 2014	Spray application	Spray application
5 April 2014		Field cage trial
		Laboratory cage trial two 1 DAT
11 Mar. 2014	Spray application	
12 Mar. 2014	Laboratory cage trial one 1 DAT	
18 Mar. 2014	Spray application	Spray application
21 Mar. 2014		Laboratory cage trial 3 DAT
25 Mar. 2014	Spray application	
1 April 2014	Spray application	
2 April 2014	Field cage trial	
	Laboratory cage trial two 1 DAT	
4 April 2014	Laboratory cage trial 3 DAT	

DAT—day after treatment.

Table 2. Dates of spray application and trials, season two (2015)

Date	Capsicum	Zucchini
24 Feb. 2015		Spray application
3 Mar. 2015	Spray application	Spray application
4 Mar. 2015		Field cage trial
10 Mar. 2015	Spray application	Spray application
11 Mar. 2015		Laboratory cage trial 1 DAT
13 Mar. 2015		Laboratory cage trial 3 DAT
17 Mar. 2015	Spray application	
24 Mar. 2015	Spray application	
25 Mar. 2015	Laboratory cage trial 1 DAT	
27 Mar. 2015	Laboratory cage trial 3 DAT	
31 Mar. 2015	Spray application	
1 April 2015	Field cage trial	

DAT—day after treatment.

completed, hence the number of treatment applications varied dependent on season and crop (Tables 1 and 2). In season one (2014) four applications were made to the zucchinis and six to the capsicums. In season two (2015) three applications were made to the zucchinis and five to the capsicums. However, due to the rapid rate of development of the zucchini fruit, it was necessary to remove large fruit at regular intervals to ensure continuous production; hence zucchini fruit used in trials had received a maximum of three applications.

Treated plants were exposed to fruit flies using two methods. First, fruit flies were exposed to intact fruit on plants removed from the field and placed into large field cages. Second, fruit flies were exposed to fruit in small laboratory cages, in order to assess adult mortality and efficacy of aged residues.

Fruit Flies

All fruit flies were obtained from colonies maintained by the Market Access research group at the Department of Agriculture and Fisheries (DAF) (Brisbane, QLD, Australia). *Bactrocera tryoni* colonies were established from collections of host fruit (*Endiandra* sp, *Barringtonia calyptrata*, and *Terminalia catappa*) in the Cairns region in January 2012 and reared according to the method of Heather and Corcoran (1985). Adults used in season one (2014) were 10–16 d post emergence and 16–18 d post emergence in season two (2015). *Zeugodacus cucumis* colonies were established from collections of zucchini in the Ayr and Cairns regions in September and October 2010. They were reared according to the method of Heather and Corcoran (1985) using the pumpkin diet described by Swaine et al. (1978). *Zeugodacus cucumis* were 13–15 d post emergence. Prior to use in the tests all fruit flies were provided with sugar, water, and protein (autolysed yeast) and allowed to mate, hence were ready to oviposit.

Field Cage Infestation

Trials were performed in four metal frame, netted cages (3 m by 3 m base, 2.5 m high), each cage representing one replicate. One day following treatment application, treated plants bearing fruit were selected at random from each replicate of each treatment, dug up, placed in large pots, watered and transferred to the field cages. Fruit flies were therefore exposed to residues on the foliage as well as on the fruit. Three zucchini plants or four capsicum plants were used per treatment replicate, with plants from each treatment grouped together, meaning that all seven treatments were present in each replicate cage. Approximately 600 mixed sex adult fruit flies, determined by pupal weight, were released into each cage. In the first season trial, all fruit flies were left for ~4 h to oviposit. In the second season trial, based on results from the first season, the exposure time was decreased to 3 h for *Z. cucumis*, and due to adverse weather conditions (cool with light rain), *B. tryoni* were left to oviposit overnight (~ 20–21 h). Fruit were then harvested and transported to the laboratory for subsequent assessment of infestation. The size and number of fruit per treatment replicate varied between replicates and treatments, due to variable fruit production of the plants. Therefore, fruit in each treatment replicate were weighed (not counted), and the weight range for each of the field cage trials presented in the results.

In the laboratory, the harvested fruit from each treatment replicate were placed on shallow plastic containers covered with net, within ventilated containers. A layer of vermiculite on the base of the container was provided as a substrate for pupation. The fruit were held in a controlled environment room (26 °C, 70% relative humidity) for ~2 wk to allow any eggs laid to develop to the pupal stage. The vermiculite was then sieved and the number of pupae counted. Additional fruit were harvested from untreated control plants in the trial block to assess the level of infestation in the field, prior to artificial infestation in the field cages. However, no pupae developed from any of these fruit and therefore these results were not included in the analyses.

Laboratory Cage Infestation

Fruit were removed from plants in the trial plots either 1 or 3 d after treatment application (1 and 3 DAT). Two 1 DAT and one 3 DAT laboratory cage trials were conducted for the first season; one 1 DAT and one 3 DAT trial were conducted for the second season. Bifenthrin (Talstar 250 EC Insecticide/Miticide) was omitted from the 3 DAT first season cucumber fly trial due to poor efficacy at 1 DAT. Three fruit from each treatment replicate were exposed to fruit flies in small laboratory cages (wire frame, netted cages, 21 cm wide, 21 cm high, 33 cm deep). Each cage contained 10 male and

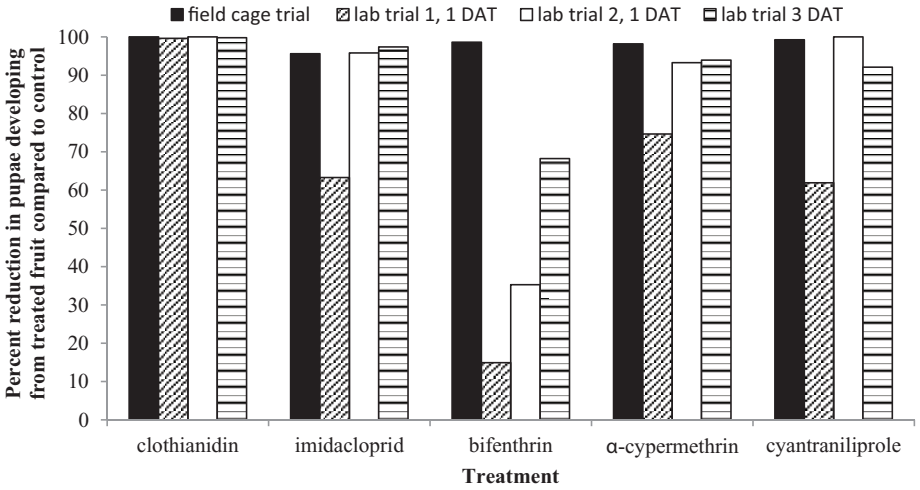

Fig. 1. Effect of treatments on reduction in numbers of *B. tryoni* pupae developing from treated capsicum compared with control fruit in season one trials.

10 female fruit flies, provided with sugar and water. Exposure times varied dependent on the fruit type, fruit fly species, and time of day that fruit were harvested. In season one, zucchinis in the 1 DAT trials were picked and placed into cages in the afternoon, and hence infested overnight (13 and 18 h infestation periods). This was reduced to 3 h for the 3 DAT trial. Capsicums were infested for 3 h (1 DAT trial one), overnight (~ 15 h, 1 DAT trial two) or 2.5 h (3 DAT trial). In season two, exposure was reduced due to overinfestation in season one, and all laboratory cage trial fruit was infested for between 2 and 3 h. Following exposure to the fruit flies, fruit were held in ventilated containers under controlled conditions as described previously, and the number of pupae counted. Mortality of adult fruit flies was assessed 1 d (~ 24 h) after placement of fruit into the cages.

Statistical Analyses

Numbers of pupae developing from the fruit were analyzed using a generalized linear mixed model (GLMM) assuming a Poisson distribution and a log link function. Due to variability in number and size of fruit sampled from each replicate of each treatment, fruit weight was initially used as a covariate in the field cage analyses. However, with the exception of season one (2014) trial with *B. tryoni* in capsicum, the effect of fruit weight was found to be not significant and therefore removed. Where a significant effect of treatment was found, pairwise comparisons between the transformed means were made using the 95% least significant difference (LSD). In some instances, no pupae developed in a treatment, or pupae developed in only one replicate of a treatment, resulting in an overinflated standard error. Means with an overinflated standard error were not included in the pairwise comparisons; however, when no pupae developed we can intuitively say that there was a significant effect of treatment. Control corrected means (Abbott 1925) were also calculated and presented to facilitate comparison of treatment efficacy.

The proportion of dead fruit flies at one day after exposure to the treated fruit was analyzed using a GLMM assuming a binomial distribution and logit link function. Where a significant treatment effect was found, pairwise comparisons between the transformed means were made using the 95% LSD. Mortality was expressed as the mean number of dead fruit flies per cage for presentation in results.

Statistical analyses were performed in GenStat for Windows 16th Edition (VSN International 2013).

Results

Season One (2014) *B. tryoni* in Capsicum

Results of a field cage trial found a significant effect of treatment on development of pupae from treated fruit ($P = 0.002$; Table 3). All insecticide treatments resulted in significantly fewer pupae than the control. Weight of fruit harvested from each replicate of each treatment varied between 908 g and 1984 g. The first 1 DAT laboratory cage trial and the 3 DAT laboratory cage trial both found a significant effect of treatment ($P < 0.001$), with significantly fewer pupae in all treatments compared to the control at 3 DAT and significantly fewer pupae in all treatments except bifenthrin compared to the control at 1 DAT. There was no significant effect of treatment in the second laboratory cage trial ($P > 0.05$), due in part to large variability in the control treatment (pupal counts ranged from 0 to 108). The effect of treatment on development of pupae from treated fruit was also expressed as control corrected means (Fig. 1). Clothianidin consistently resulted in circa 100% reduction in development of pupae in all four trials; imidacloprid, alpha-cypermethrin, and cyantraniliprole resulted in greater than 90% reduction in development of pupae in three of the four trials; and bifenthrin was effective only in the field cage trial.

Mortality of the adult fruit flies in the laboratory cage trials was assessed at ~ 24 h (Table 4). There was a significant effect of treatment on fruit flies exposed to 1-d residues (1 DAT), in both trials ($P \leq 0.011$), with significantly higher mortality in the dimethoate treatment compared to the control, and in the clothianidin treatment compared to the control in the first trial only. Three-day-old residues had no significant effect on mortality of the adult fruit flies ($P > 0.05$).

Season One (2014) *Z. cucumis* in Zucchini

Results of a field cage trial found a significant effect of treatment on the number of pupae developing from the fruit ($P = 0.011$; Table 5). Clothianidin, alpha-cypermethrin, cyantraniliprole, and dimethoate resulted in significantly fewer pupae compared to the untreated control. Weight of fruit harvested from each replicate of each treatment varied between 892 g and 1200 g. Two laboratory cage trials conducted at 1 DAT and a laboratory cage trial at 3 DAT all found a significant effect of treatment ($P \leq 0.004$). Clothianidin and dimethoate consistently resulted in the fewest pupae, and bifenthrin

Table 3. Mean number of pupae developing from capsicum exposed to *B. tryoni* in season one trials; back-transformed means (BTM) and predicted means on the log scale ± 1 standard error (PM)

Treatment		Field cage trial, 1 DAT		Lab cage trial 1, 1 DAT		Lab cage trial 2, 1 DAT		Lab cage trial, 3 DAT	
		BTM	PM	BTM	PM	BTM	PM	BTM	PM
Clothianidin		0.0	−16.82 ± δ	0.5a	−0.69 ± 1.90	0.0	−16.48 ± δ	0.3	−1.39 ± δ
Imidacloprid		3.1a	1.12 ± 1.13	46.7b	3.84 ± 0.20	0.7	−0.32 ± 1.66	3.3a	1.18 ± 1.28
Bifenthrin		1.0a	−0.02 ± 1.62	108.2c	4.68 ± 0.14	11.1	2.41 ± 0.80	39.3a	3.67 ± 0.37
α-Cypermethrin		1.3a	0.24 ± 1.65	32.2b	3.47 ± 0.24	1.2	0.15 ± 1.38	7.5a	2.02 ± 0.85
Cyantraniliprole		0.5a	−0.63 ± 2.22	48.5b	3.88 ± 0.20	0.0	−16.48 ± δ	9.8a	2.28 ± 0.74
Dimethoate		1.0a	−0.04 ± 1.63	24.0ab	3.18 ± 0.28	1.3	0.26 ± 1.32	7.3a	1.98 ± 0.86
Untreated control		70.2b	4.25 ± 0.48	127.2c	4.85 ± 0.13	17.2	2.85 ± 0.77	123.5b	4.82 ± 0.21
GLMM	*F*	5.41		12.59		2.10		6.31	
	df	6, 17.7		6, 20		6, 18.2		6, 21	
	P	0.002		< 0.001		0.104		< 0.001	

Means with a letter in common are not significantly different (LSD test; $P > 0.05$).
δ indicates an overinflated standard error.

Table 4. Mean mortality per cage of 20 adult *B. tryoni* exposed to insecticide residues on capsicum in season one trials; back transformed means (BTM) and predicted means on the logit scale ± 1 standard error (PM)

Treatment		Lab cage trial 1, 1 DAT		Lab cage trial 2, 1 DAT		Lab cage trial, 3 DAT	
		BTM	PM	BTM	PM	BTM	PM
Clothianidin		5.5d	−0.97 ± 0.24	3.5bc	−1.55 ± 0.38	1.5	−2.51 ± 0.48
Imidacloprid		0.5a	−3.66 ± 0.68	1.5ab	−2.51 ± 0.54	1.5	−2.51 ± 0.48
Bifenthrin		2.5bc	−1.95 ± 0.32	0.5a	−3.66 ± 0.91	0.5	−3.66 ± 0.80
α-Cypermethrin		1.3ab	−2.71 ± 0.44	0.5a	−3.66 ± 0.91	1.5	−2.51 ± 0.48
Cyantraniliprole		1.8ab	−2.35 ± 0.38	1.5ab	−2.51 ± 0.54	1.5	−2.51 ± 0.48
Dimethoate		4.8cd	−1.17 ± 0.25	5.5c	−0.97 ± 0.32	3.3	−1.64 ± 0.34
Untreated control		1.8ab	−2.35 ± 0.38	0.8ab	−3.25 ± 0.75	2.0	−2.20 ± 0.42
GLMM	*F*	5.62		3.76		1.25	
	df	6, 21		6, 21		6, 21	
	P	0.001		0.011		0.320	

Means with a letter in common are not significantly different (LSD test; $P > 0.05$).

was ineffective. Only clothianidin resulted in over 90% reduction in development of pupae in all four trials (Fig. 2).

There was a significant effect of treatment on adult fruit fly mortality in all three laboratory cage trials ($P < 0.001$; Table 6). Mortality was highest in the dimethoate and clothianidin treatments at 1 DAT, and in the dimethoate treatment at 3 DAT.

Although not quantified, it was observed that occurrence of aphids (*Myzus persicae* and *Aphis gossypii*) and sweetpotato whitefly (*Bemisia tabaci*) was much higher in plots treated with alpha-cypermethrin, bifenthrin, and dimethoate compared with other treatments.

Season Two (2015) *B. tryoni* in Capsicum

Results of a field cage trial found no significant effect of treatment on the development of pupae from treated fruit ($P > 0.05$; Table 7). Weight of fruit harvested from each replicate of each treatment varied between 421 g and 2836 g. Laboratory cage trials at 1 DAT and 3 DAT both found a significant effect of treatment ($P \leq 0.018$), with no pupae developing in the clothianidin treatments, and significantly fewer pupae in the thiacloprid and dimethoate treatments compared to the control. In addition, pupal counts were significantly lower than the control for abamectin in the 1 DAT trial and spinetoram in the 3 DAT trial. Only the higher rate of clothianidin (40 g/100 liter) and thiacloprid resulted in greater than 90% reduction in development of pupae in all three trials (Fig. 3).

There was a significant effect of treatment on adult fruit fly mortality in both the 1 DAT and 3 DAT laboratory cage trials ($P \leq 0.007$; Table 8). Significantly higher mortality compared to other treatments was observed for dimethoate and both rates of clothianidin, in both trials.

Season Two (2015) *Z. cucumis* in Zucchini

Results of a field cage trial found no significant effect of treatment on the number of pupae developing from the zucchinis ($P > 0.05$; Table 9). Weight of fruit harvested from each replicate of each treatment varied between 1172 g and 2696 g. Laboratory cage trials at 1 DAT found a significant effect of treatment on number of pupae developing from the fruit ($P < 0.001$), with significantly fewer pupae in all treatments compared to the untreated control. Clothianidin and dimethoate resulted in the fewest pupae, and abamectin was the least effective treatment. There was no significant effect of treatment for 3 DAT residues ($P > 0.05$). Only clothianidin resulted in close to 100% reduction in development of pupae in all three trials (Fig. 4).

There was a significant effect of treatment on adult fruit fly mortality in both the 1 DAT and 3 DAT laboratory cage trials ($P \leq 0.008$; Table 10). The highest mortality was observed in the clothianidin and dimethoate treatments.

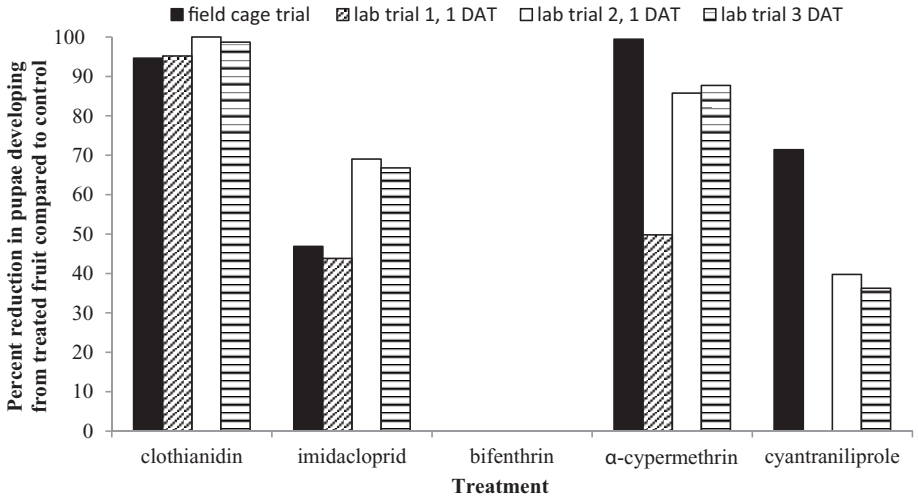

Fig. 2. Effect of treatments on reduction in numbers of *Z. cucumis* pupae developing from treated zucchini compared with control fruit in season one trials.

Table 5. Mean number of pupae developing from zucchini exposed to *Z. cucumis* in season one trials; back-transformed means (BTM) and predicted means on the log scale ± 1 standard error (PM)

Treatment	Field cage trial, 1 DAT		Lab cage trial 1, 1 DAT		Lab cage trial 2, 1 DAT		Lab cage trial, 3 DAT	
	BTM	PM	BTM	PM	BTM	PM	BTM	PM
Clothianidin	23.4a	3.16 ± 1.21	20.5a	3.02 ± 0.92	0.0	−8.69 ± δ	11.0a	2.40 ± 0.97
Imidacloprid	231.5ab	5.45 ± 0.49	238.6b	5.48 ± 0.29	177.0c	5.18 ± 0.19	281.2c	5.64 ± 0.19
Bifenthrin	601.7c	6.40 ± 0.39	432.0b	6.07 ± 0.23	590.8e	6.38 ± 0.10		[a]
α-Cypermethrin	2.5	0.91 ± δ	213.1b	5.36 ± 0.31	81.5b	4.40 ± 0.28	104.0b	4.64 ± 0.32
Cyantraniliprole	124.7a	4.83 ± 0.60	432.0b	6.07 ± 0.23	344.5d	5.84 ± 0.14	539.6d	6.29 ± 0.14
Dimethoate	18.7a	2.93 ± 1.35	50.4a	3.92 ± 0.59	17.8a	2.88 ± 0.60	17.2a	2.85 ± 0.77
Untreated control	435.8bc	6.08 ± 0.41	424.6b	6.05 ± 0.23	571.8e	6.35 ± 0.11	846.6e	6.74 ± 0.11
GLMM *F*	3.92		4.83		17.22		18.28	
df	6, 17.9		6, 17.8		6, 21		5, 15	
P	0.011		0.004		< 0.001		< 0.001	

Means with a letter in common are not significantly different (LSD test; $P > 0.05$).

δ indicates an overinflated standard error.

[a] Bifenthrin was omitted from the 3 DAT trial due to poor results in the 1 DAT trials.

Table 6. Mean mortality per cage of 20 adult *Z. cucumis* exposed to insecticide residues on zucchini in season one trials; back transformed means (BTM) and predicted means on the logit scale ± 1 standard error (PM)

Treatment	Lab cage trial 1, 1 DAT		Lab cage trial 2, 1 DAT		Lab cage trial, 3 DAT	
	BTM	PM	BTM	PM	BTM	PM
Clothianidin	9.0b	−0.20 ± 0.48	5.7b	−0.91 ± 0.27	1.7a	−2.35 ± 0.44
Imidacloprid	1.0a	−2.93 ± 0.65	0.3a	−4.38 ± 0.96	0.3a	−4.38 ± 1.08
Bifenthrin	0.2a	−4.62 ± 1.15	0.5a	−3.67 ± 0.69		[a]
α-Cypermethrin	0.6a	−3.48 ± 0.76	1.7a	−2.35 ± 0.40	0.3a	−4.38 ± 1.08
Cyantraniliprole	0.6a	−3.48 ± 0.76	0.0	−16.75 ± δ	0.0	−16.75 ± δ
Dimethoate	3.4b	−1.60 ± 0.52	7.5b	−0.51 ± 0.25	11.0b	0.20 ± 0.27
Untreated control	0.0	−17.71 ± δ	0.3a	−4.38 ± 0.96	0.0	−16.75 ± δ
GLMM *F*	9.95		9.36		10.64	
df	6, 18.2		6, 17.9		6, 15	
P	< 0.001		< 0.001		< 0.001	

Means with a letter in common are not significantly different (LSD test; $P > 0.05$).

δ indicates an overinflated standard error.

[a] Bifenthrin was omitted from the 3 DAT trial due to poor results in the 1 DAT trials.

Table 7. Mean number of pupae developing from capsicum exposed to *B. tryoni* in season two trials; back-transformed means (BTM) and predicted means on the log scale ± 1 standard error (PM)

Treatment		Field cage trial, 1 DAT		Lab cage trial, 1 DAT		Lab cage trial, 3 DAT	
		BTM	PM	BTM	PM	BTM	PM
Clothianidin 40 g/100 liter		0.0	−13.68 ± δ	0.0	−11.69 ± δ	0.0	−18.71 ± δ
Clothianidin 30 g/100 liter		15.3	2.73 ± 0.74	0.0	−11.69 ± δ	0.0	−18.71 ± δ
Thiacloprid		1.4	0.36 ± 2.29	13.2a	2.58 ± 0.64	3.4a	1.22 ± 1.10
Abamectin		35.0	3.56 ± 0.52	47.7ab	3.87 ± 0.34	41.8bc	3.73 ± 0.37
Spinetoram		42.2	3.74 ± 0.49	103.7bc	4.64 ± 0.24	11.1ab	2.41 ± 0.63
Dimethoate		6.5	1.87 ± 1.10	20.4a	3.02 ± 0.51	2.2a	0.78 ± 1.37
Untreated control		35.5	3.57 ± 0.52	182.5c	5.21 ± 0.19	75.2c	4.32 ± 0.31
GLMM	*F*	0.94		6.32		3.53	
	df	6, 18.2		6, 18.1		6, 17.7	
	P	0.492		0.001		0.018	

Means with a letter in common are not significantly different (LSD test; $P > 0.05$).
δ indicates an overinflated standard error.

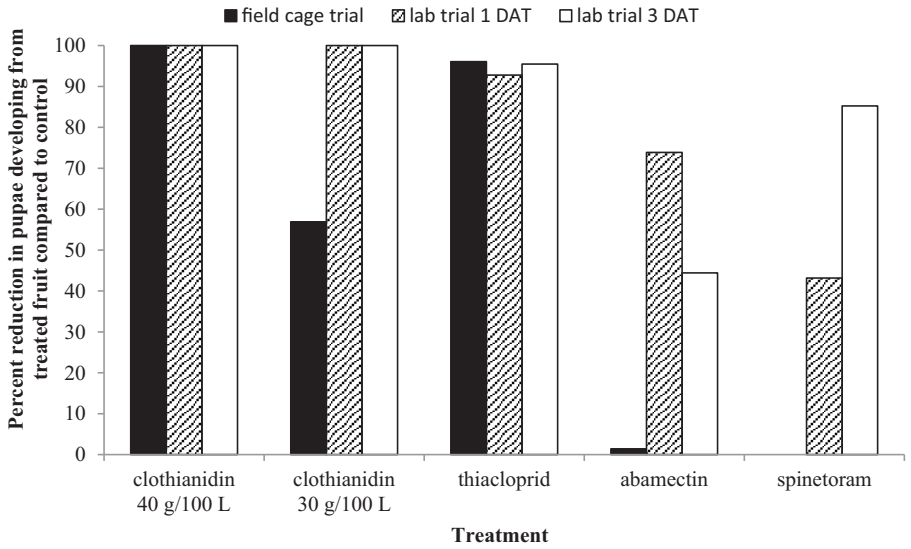

Fig. 3. Effect of treatments on reduction in numbers of *B. tryoni* pupae developing from treated capsicum compared with control fruit in season two trials.

Weather Data

Rainfall for the 2014 trial period was minimal except for a period following the penultimate spray application made to capsicum (25th March). Rainfall during the 2015 trial period was minimal in the 48-h periods following spray applications with the exception of those applied to capsicum on 17th and 24th March.

Discussion

All the insecticides demonstrated some level of efficacy compared to the untreated control. Efficacy was generally lower for *Z. cucumis* than *B. tryoni*. This may in part have been because the zucchinis developed more quickly than capsicums, were picked more frequently and hence received fewer sprays. However, it is likely that the comparative vigour of the two fruit fly species also had an effect; the number of *Z. cucumis* pupae developing in all treatments, including the control, was generally much higher than for *B. tryoni*. Unpublished data collected for the two fruit fly colonies used in the trials found that egg hatch and total survival to adult in *Z. cucumis* (94–98% and 78–87%, respectively) was higher than *B. tryoni* (76–87% and 65–79%).

Clothianidin (Sumitomo Samurai Systemic Insecticide) was the most effective of the eight insecticides assessed, with both 1- and 3-d-aged residues consistently demonstrating efficacy comparable to dimethoate in terms of numbers of pupae developing from treated fruit. Clothianidin was the only insecticide other than dimethoate to significantly affect mortality of adult fruit flies. Two other neonicotinoid insecticides, thiacloprid (Calypso 480 SC Insecticide) and imidacloprid (Confidor 200 SC), also demonstrated efficacy comparable to dimethoate against *B. tryoni* in terms of reduction in pupal development. However, they were generally much less effective against *Z. cucumis*, and had no effect on adult mortality in either species. Reynolds et al. (2014) found clothianidin to be moderately effective in semifield trials with *B. tryoni* in stonefruit. Neonicotinoids have also proven to be effective against a number of other tephritid species. Rahman and Broughton (2016) found clothianidin and thiacloprid significantly reduced infestation of stonefruit by Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), in laboratory trials of 24-h field aged residues. Efficacy was reduced when residues were aged for 7 d. Yee and Alston (2006) found thiacloprid and imidacloprid significantly suppressed infestation by western cherry fruit fly *Rhagoletis indifferens* Curran in field trials

in cherry orchards. Reissig (2003) found imidacloprid to be more effective than thiacloprid in laboratory trials with apple maggot, *Rhagoletis pomonella* (Walsh), but thiacloprid was more effective in orchard trials. The author suggested efficacy was primarily a result of reduced oviposition; neither insecticide resulted in high adult mortality. Hu and Prokopy (1998) demonstrated that whilst imidacloprid was effective against *R. pomonella* via contact (residues applied to glass) and ingestion, it was ineffective when applied to foliage. Similarly, Hu et al. (1998) found imidacloprid to be ineffective for control of *R. pomonella* in apple orchards, suggesting that this was due to rapid absorption by the foliage and degradation by sunlight.

Alpha-cypermethrin (Nufarm Fastac Duo Insecticide) was effective against *B. tryoni* in capsicums in laboratory and field cage trials, with efficacy of 1-d and 3-d residues comparable to dimethoate. Alpha-cypermethrin was also effective against *Z. cucumis* in zucchini when tested in the field cage trial, but was less effective in laboratory trials. Reynolds et al. (2014) found that efficacy of alpha-cypermethrin against *B. tryoni* was comparable to fenthion in semifield tests. Various isomers of cypermethrin have demonstrated efficacy against tephritids, for example, cypermethrin reduced infestation and increased yield in a trial to manage *Bactrocera* spp in cucumber (Sharma et al. 2016); residues of zeta-cypermethrin on cherries effectively reduced oviposition by *R. indifferens* (Yee and Alston 2012); fresh cypermethrin residues on cucumber prevented oviposition by *Dacus ciliatus* Loew, the lesser pumpkin fly (Maklakov et al. 2001). Efficacy of the second synthetic pyrethroid assessed, bifenthrin (Talstar 250 EC Insecticide/Miticide) was also comparable with dimethoate in a field cage trial against *B. tryoni*. However, efficacy in laboratory trials was generally low and this insecticide was ineffective against *Z. cucumis*. Maklakov et al. (2001) found that fresh bifenthrin residues prevented oviposition by *D. ciliatus* in cucumbers, suggesting that cypermethrin and bifenthrin had a repellent effect on fruit flies. Repellency of pyrethroids has been documented in a variety of insects, including mosquitoes, houseflies, honey bees, Lepidoptera, and mites (Virgona et al. 1983, Rieth and Levin 1988, Hirano 1989, Siegert et al. 2009). A repellent effect could explain the better efficacy achieved for alpha-cypermethrin and bifenthrin in field cage trials, where fruit flies were presented with a choice of treated and untreated fruit, as opposed to laboratory cage trials, where fruit flies were confined in close proximity with treated fruit.

Cyantraniliprole (DuPont Benevia Insecticide) demonstrated efficacy in field cage trials against both fruit fly species, and laboratory cage trials against *B. tryoni*. It was less effective against *Z. cucumis*. There is relatively little published data on the effect of this new insecticide on tephritid flies. Reynolds et al. (2014) found cyantraniliprole to have little impact on mortality of adult *B. tryoni* in laboratory tests. Cyantraniliprole significantly reduced adult emergence of *B. dorsalis*, *Z. cucurbitae*, and *C. capitata*, when applied as a soil drench (Stark et al. 2013), and resulted in high adult mortality of *B. dorsalis* when ingested (Zhang et al. 2015).

Spinetoram (Success Neo Insecticide) was not effective in the field cage trials and had mixed efficacy in the laboratory cage trials. Reynolds et al. (2014) found spinetoram to be moderately effective on mortality of *B. tryoni* in laboratory tests. Likewise, Yee and Alston (2012) found that this chemical resulted in 81% mortality of *R. indifferens* females exposed to residues on cherries at

Table 8. Mean mortality per cage of 20 adult *B. tryoni* exposed to insecticide residues on capsicum in season two trials; back transformed means (BTM) and predicted means on the logit scale ± 1 standard error (PM)

Treatment		Lab cage trial, 1 DAT		Lab cage trial, 3 DAT	
		BTM	PM	BTM	PM
Clothianidin 40 g/100 liter		6.3b	−0.79 ± 0.25	3.2b	−1.65 ± 0.29
Clothianidin 30 g/100 liter		8.0b	−0.41 ± 0.23	2.5b	−1.95 ± 0.31
Thiacloprid		0.3a	−4.37 ± 1.03	0.3a	−4.38 ± 0.86
Abamectin		0.0	−14.75 ± δ	0.0	−16.75 ± δ
Spinetoram		0.3a	−4.37 ± 1.03	0.3a	−4.38 ± 0.86
Dimethoate		9.0b	−0.20 ± 0.23	2.7b	−1.84 ± 0.30
Untreated control		0.0	−14.75 ± δ	0.3a	−4.38 ± 0.86
GLMM	*F*		5.62		4.32
	df		6, 21		6, 18.2
	P		0.002		0.007

Means with a letter in common are not significantly different (LSD test; $P > 0.05$).

δ indicates an overinflated standard error.

Table 9. Mean number of pupae developing from zucchini exposed to *Z. cucumis* in season two trials; back-transformed means (BTM) and predicted means on the log scale ± 1 standard error (PM)

Treatment		Field cage trial, 1 DAT		Lab cage trial, 1 DAT		Lab cage trial, 3 DAT	
		BTM	PM	BTM	PM	BTM	PM
Clothianidin 40 g/100 liter		2.8	1.02 ± 4.25	1.7a	0.56 ± 1.75	0.0	−13.69 ± δ
Clothianidin 30 g/100 liter		0.0	−15.52 ± δ	12.7a	2.54 ± 0.65	1.5	0.39 ± 3.97
Thiacloprid		376.4	5.93 ± 0.47	90.3b	4.50 ± 0.25	189.1	5.24 ± 0.38
Abamectin		375.3	5.93 ± 0.47	478.3c	6.17 ± 0.12	385.9	5.96 ± 0.28
Spinetoram		361.0	5.89 ± 0.47	151.2b	5.02 ± 0.20	253.8	5.54 ± 0.34
Dimethoate		44.8	3.80 ± 1.09	9.0a	2.20 ± 0.77	236.1	5.46 ± 0.35
Untreated control		423.7	6.05 ± 0.45	797.4d	6.68 ± 0.10	243.7	5.50 ± 0.34
GLMM	*F*		0.91		32.06		0.87
	df		6, 17.9		6, 18		6, 18.1
	P		0.507		< 0.001		0.538

Means with a letter in common are not significantly different (LSD test; $P > 0.05$).

δ indicates an overinflated standard error.

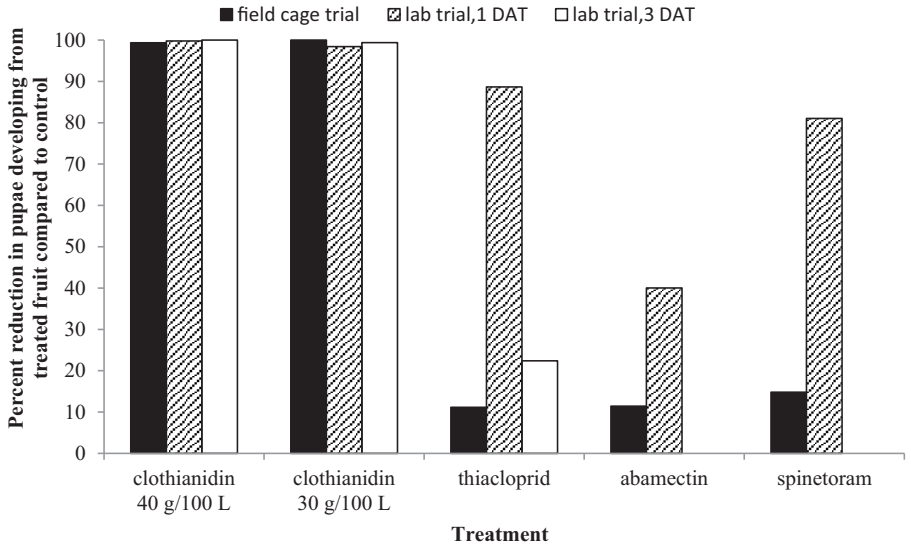

Fig. 4. Effect of treatments on reduction in numbers of *Z. cucumis* pupae developing from treated zucchini compared with control fruit in season two trials.

Table 10. Mean mortality per cage of 20 adult *Z. cucumis* exposed to insecticide residues on zucchini in season two trials; back transformed means (BTM) and predicted means on the logit scale ± 1 standard error (PM)

Treatment	Lab cage trial, 1 DAT		Lab cage trial, 3 DAT	
	BTM	PM	BTM	PM
Clothianidin 40 g/100 liter	7.5b	−0.52 ± 0.25	5.5bc	−0.97 ± 0.38
Clothianidin 30 g/100 liter	4.5a	−1.25 ± 0.28	2.5ab	−1.95 ± 0.50
Thiacloprid	0.0	−18.75 ± δ	0.3a	−4.37 ± 1.45
Abamectin	0.0	−18.75 ± δ	0.3a	−4.37 ± 1.45
Spinetoram	0.0	−18.75 ± δ	0.3a	−4.37 ± 1.45
Dimethoate	11.3c	−0.25 ± 0.25	10.0c	0.00 ± 0.34
Untreated control	0.0	−18.75 ± δ	0.0	−15.75 ± δ
GLMM *F*		4.24		9.36
df		6, 18		6, 18.1
P		0.008		0.004

Means with a letter in common are not significantly different (LSD test; $P > 0.05$).

δ indicates an overinflated standard error.

24 h, and reduced oviposition compared to controls. Both Reynolds et al. (2014) and Yee and Alston (2012) applied treatments and aged residues under laboratory conditions. Yee et al. (2007) found that aging of spinetoram residues under field conditions resulted in reduced mortality of adult *R. pomonella*; however, residues remained effective up to 14 d in terms of preventing oviposition.

Abamectin (Vertimec Miticide/Insecticide) had no effect in field cage trials. One-day residues had a small but significant effect on *B. tryoni* and *Z. cucumis* in laboratory trials. Kay (2004) found abamectin residues on capsicum resulted in up to 70% mortality of *B. tryoni* up to 4 h after dipping, but 24-h residues were less effective. Reynolds et al. (2014) found abamectin residues on stonefruit resulted in high mortality of adult *B. tryoni* and reduced oviposition. Abamectin is rapidly degraded by exposure to light and air (MacConnell et al. 1989).

In summary, the neonicotinoid clothianidin demonstrated efficacy comparable to dimethoate against both *B. tryoni* and *Z. cucumis*. Thiacloprid and imidacloprid were also generally effective against *B. tryoni*. However, there are concerns about effects of neonicotinoids on bees and other pollinator species (Blacquière et al. 2012, Godfray et al. 2014). The synthetic pyrethroid alpha-cypermethrin was also very effective. However, observations of increased aphid and whitefly activity in alpha-cypermethrin and bifenthrin plots compared with other treatments suggest a disruptive effect on natural enemies. Cyantraniliprole was effective against *B. tryoni* and is claimed by the manufacturer to be soft on beneficials. Spinetoram, abamectin, and bifenthrin were generally less effective than the other treatments. However, these insecticides are registered in fruiting vegetable crops for control of other pests, and it is likely that their use would have a suppressive effect on fruit flies. It should also be noted that treated fruit were exposed to much higher fruit fly pressure than could be expected in the field, and it is possible that greater efficacy of treatments would be observed in actual use conditions. Large variability between replicates was a problem and may have obscured some treatment effects; incomplete coverage by insecticides may have accounted for some of this variability.

These data represent the first successful trial of the efficacy of insecticides, applied to a vegetable crop as cover sprays, against *B. tryoni* and *Z. cucumis*. Although Kay (2004) applied insecticides using a similar small plot layout, background fruit fly pressure alone was not sufficient to result in a significant difference amongst treatments. Reynolds et al. (2014) also used a semifield method to compare insecticides for efficacy against *B. tryoni*; however, although insecticides were applied to fruit in the field, fruit flies were then caged in close proximity to the treated fruit. The field cage method described here allowed for comparison of a number of insecticides under semirealistic conditions. Insecticides were applied to plants and the residues aged under field conditions; fruit flies were exposed to entire plants bearing fruit; and fruit flies were able to choose where to land and oviposit. This allowed for evaluation of effects other than mortality, for example, repellent effects resulting in reduced oviposition. This is significant, as few of the insecticides affected adult mortality in the laboratory cage trials, but the majority

demonstrated at least a suppressive effect on infestation of fruit in field cage trials.

Acknowledgments

This study formed part of project VG13041 "New in-field treatment solutions to control fruit fly (1)," funded by Horticulture Innovation Australia using the vegetable industry levy and matched funds from the Australian Government. We thank Kellie Bilney, Ania Ganowicz, Chloe Jacobs, Mary Firrell, and John Duff for technical assistance, Thelma Peek for providing fruit flies, Gatton Research Facility staff for maintaining the trial site, and Geoff Cornwell (DuPont Crop Protection Australia) and Doug Paton (Sumitomo Chemical Australia Pty Ltd) for supplying insecticide samples. Earlier versions for this manuscript were improved by comments from Pauline Wyatt, Peter Leach and two anonymous reviewers.

References Cited

Abbott, W. S. 1925. A method of computing the effectiveness of an insecticide. J. Econ. Entomol. 18: 265–267.

(APVMA) Australian Pesticides and Veterinary Medicines Authority. 2011. Use of the insecticide dimethoate suspended on many food crops. (http://apvma.gov.au/node/11771) (accessed 30 March 2017).

(APVMA) Australian Pesticides and Veterinary Medicines Authority. 2015. Fenthion chemical review. (http://apvma.gov.au/node/12541) (accessed 30 March 2017).

(APVMA) Australian Pesticides and Veterinary Medicines Authority. 2016. Dimethoate chemical review. (http://apvma.gov.au/node/12496) (accessed 30 March 2017).

(APVMA) Australian Pesticides and Veterinary Medicines Authority. 2017. Dimethoate chemical review. (http://apvma.gov.au/dimethoate), accessed 30 March 2017.

Atuahene, S.K.N., and G.H.S. Hooper. 1971. Insecticide susceptibility of, and metabolism of DDT by *Dacus tryoni* (Froggatt) and *Dacus cucumis* French (Diptera: Tephritidae). J. Aust. Entomol. Soc. 11: 135–142.

Balagawi, S., K. Jackson, and A. R. Clarke. 2014. Resting sites, edge effects and dispersion of a polyphagous *Bactrocera* fruit fly within crops of different architecture. J. Appl. Entomol. 138: 510–518.

Blacquière, T., G. Smagghe, C.A.M. van Gestel, and V. Mommaerts. 2012. Neonicotinoids in bees: A review on concentrations, side-effects and risk assessment. Ecotoxicology 21: 973–992.

Clarke, A. R., K. S. Powell, C. W. Weldon, and P. W. Taylor. 2011. The ecology of *Bactrocera tryoni* (Froggatt) (Diptera: Tephritidae): What do we know to assist pest management? An. Appl. Biol. 158: 26–55.

Dominiak, B. C., B. Wiseman, C. Anderson, B. Walsh, M. McMahon, and R. Duthie. 2015. Definition of and management strategies for areas of low pest prevalence for Queensland fruit fly *Bactrocera tryoni* Froggatt. Crop Prot. 72: 41–46.

Drew, R.A.I., G.H.S. Hooper, and M. A. Bateman. 1982. Economic fruit flies of the South Pacific region, 2nd ed. Queensland Department of Primary Industries, Brisbane, Australia.

Godfray, H.C.J., T. Blacquière, L. M. Field, R. S. Hails, G. Petrokofsky, S. G. Potts, N. E. Raine, A. J. Vanbergen, and A. R. McLean. 2014. A restatement of the natural science evidence base concerning neonicotinoid insecticides and insect pollinators. Proc. R. Soc. B. 281: 20140558.

Gu, H. 2010. Alternative fruit fly treatment for interstate market access for strawberries. Horticulture Australia Project Number BS06002 Final Report, Horticulture Australia Ltd, Sydney, Australia.

Hancock, D. L., E. L. Hamacek, A. C. Lloyd, and M. M. Elson-Harris. 2000. The distribution and host plants of fruit flies (Diptera: Tephritidae) in Australia. Queensland Department of Primary Industries Information Series QI99067, Brisbane, Australia.

Heather, N. W., and R. J. Corcoran. 1985. *Dacus tryoni*, pp. 41–48. *In* P. Singh and R. F. Moore (eds.), Handbook of insect rearing, vol 2. Elsevier, Amsterdam, The Netherlands.

Hirano, M. 1989. Characteristics of pyrethroids for insect pest control in agriculture. Pestic. Sci. 27: 353–360.

Hu, X. P., A. Kaknes, and R. Prokopy. 1998. Can apple maggot fly control benefit from sprays of Provado aimed at killing leafminers and leafhoppers? Fruit Notes 63: 4–6.

Hu, X. P., and R. J. Prokopy. 1998. Lethal and sublethal effects of imidacloprid on apple maggot fly, *Rhagoletis pomonella* Walsh (Dipt., Tephtritidae). J. Appl. Entomol. 122: 37–42.

Kay, I. 2004. Heliothis and fruit fly integrated pest management strategies for tomato, vegetable and melon crops. Horticulture Australia Project Number VX99035 Final Report, Horticulture Australia Ltd, Sydney, Australia.

Khursheed, S., and D. Raj. 2012. Bio-efficacy of certain insecticides and biopesticides against melon fruit flies, *Bactrocera* spp. Pest Manag. Hort. Ecosyst. 18: 143–148.

MacConnell, J. G., R. J. Demchak, F. A. Preiser, and R. A. Dybas. 1989. Relative stability, toxicity, and penetrability of abamectin and its 8,9-oxide. J. Agric. Food Chem. 37: 1498–1501.

Macfadyen, S., J. E. Banks, J. D. Stark, and A. P. Davies. 2014. Using semifield studies to examine the effects of pesticides on mobile terrestrial invertebrates. Ann. Rev. Entomol. 59: 383–404.

Maklakov, A., I. Ishaaya, A. Freidberg, A. Yawetz, A. R. Horowitz, and I. Yarom. 2001. Toxicological studies of organophosphate and pyrethroid insecticides for controlling the fruit fly *Dacus ciliatus* (Diptera: Tephritidae). J. Econ. Entomol. 94: 1059–1066.

McQuate, G. T. 2011. Assessment of attractiveness of cassava as a roosting plant for the melon fly, *Bactrocera cucurbitae*, and the oriental fruit fly, *B. dorsalis*. J. Insect Sci. 11: 30.

Mosleh, Y. Y., S.F.M. Moussa, and L.H.Y. Mohamed. 2011. Comparative toxicity of certain pesticides to peach fruit fly, *Bactrocera zonata* Saunders (Diptera: Tephritidae) under laboratory conditions. Plant Prot. Sci. 47: 115–120.

Nishida, T., and H. A. Bess. 1957. Studies on the ecology and control of the melon fly *Dacus (Strumeta) cucurbitae* Coquillett (Diptera: Tephritidae). Tech. Bull. Hawaii. Agric. Exp. Stn. 34: 44. pp.

Oke, O. A. 2008. Effectiveness of two insecticides to control melon fruit fly (*Bactrocera cucurbitae* Coq.) in cucumber (*Cucumis sativus* L.) crop at Anse Boileau Seychelles. Eur. J. Sci. Res. 22: 84–86.

Oke, O. A., and S. G. Sinon. 2013. Effectiveness of three insecticides to control *Bactrocera cucurbitae* (Diptera: Tephritidae) on the cucumber crop at Praslin, Seychelles. Fla. Entomol. 96: 120–123.

Prokopy, R. J., N. W. Miller, J. C. Piñero, J. D. Barry, L. C. Tran, L. Oride, and R. I. Vargas. 2003. Effectiveness of GF-120 fruit fly bait spray applied to border area plants for control of melon flies (Diptera: Tephritidae). J. Econ. Entomol. 96: 1485–1493.

Rahman, T., and S. Broughton. 2016. Evaluation of thiacloprid and clothianidin (neonicotinoids) as alternative to fenthion (organophosphate) for control of Mediterranean fruit fly (Diptera: Tephritidae) in deciduous fruit orchards. Crop Prot. 90: 170–176.

Reissig, W. H. 2003. Field and laboratory tests of new insecticides against the apple maggot, *Rhagoletis pomonella* (Walsh) (Diptera: Tephritidae). J. Econ. Entomol. 96: 1463–1472.

Reynolds, O. L., A. Jessup, T. Osborne, K. Bodnaruk, J. Archer, K. Eamens, and I. Barchia. 2014. Efficacy of potential controls for Queensland fruit fly management in summerfruit. Horticulture Australia Project Number SF12012 Final Report, Horticulture Australia Ltd, Sydney, Australia.

Rieth, J. P., and M. D. Levin. 1988. The repellent effect of two pyrethroid insecticides on the honey bee. Physiol. Entomol. 13: 213–218.

Sharma, S. K., Unam, and R. Kumar. 2016. Management of fruit fly (*Bactrocera* spp.) in cucumber (*Cucumis sativus* Linn.) grown organically. J. Biopest. 9: 73–79.

Siegert, P. Y., E. Walker, and J. R. Miller. 2009. Differential responses of *Anopheles gambiae* (Diptera: Culicidae) modulate mortality caused by pyrethroid-treated bednets. J. Econ. Entomol. 102: 2061–2071.

Stark, J. D., R. I. Vargas, S. L. Souder, A. J. Fox, T. R. Smith, and B. Mackey. 2013. A comparison of the bioinsecticide, spinosad, the semi-synthetic insecticide, spinetoram and synthetic insecticides as soil drenches for control of tephritid fruit flies. Biopestic. Int. 9: 120–126.

Steiner, L. F., and F. G. Hinman. 1952. Field tests of insecticides for control of oriental fruit fly. J. Econ. Entomol. 45: 388–395.

Subramaniam, S. 2013. Alternative fruit fly control for market access and to enhance integrated pest management in eggplant. Horticulture Australia Project Number VG09023 Final Report, Horticulture Australia Ltd, Sydney, Australia.

Swaine, G., R. J. Corcoran, and M. Davey. 1978. Commodity treatment against infestations of the cucumber fly, *Dacus* (Austrodacus) *cucumis* French, in cucumbers. Qld. J. Agric. Anim. Sci. 35: 5–9.

Vargas, R. I., J. C. Piñero, and L. Leblanc. 2015. An overview of pest species of *Bactrocera* fruit flies (Diptera: Tephritidae) and the integration of biopesticides with other biological approaches for their management with a focus on the Pacific region. Insects 6: 297–318.

Virgona, C. T., G. Holan, and E. Shipp. 1983. Repellency of insecticides to resistant strains of housefly. Entomol. Exp. Appl. 34: 287–290.

VSN International 2013. GenStat for Windows, version 16th ed. VSN International, Hemel Hempstead, United Kingdom.

Wang, J.-J., D. Wei, W. Dou, F. Hu, W.-F. Liu, and J.-J. Wang. 2013. Toxicities and synergistic effects of several insecticides against the oriental fruit fly (Diptera: Tephritidae). J. Econ. Entomol. 106: 970–978.

Wise, J. C., R. Vanderpoppen, and C. Vandervoort. 2009. Curative activity of insecticides on *Rhagoletis pomonella* (Diptera: Tephritidae) in apples. J. Econ. Entomol. 102: 1884–1890.

Yee, W. L. 2008. Effects of several newer insecticides and kaolin on oviposition and adult mortality in western cherry fruit fly (Diptera: Tephritidae). J. Entomol. Sci. 43: 177–190.

Yee, W. L., and D. G. Alston. 2006. Effects of spinosad, spinosad bait, and chloronicotinyl insecticides on mortality and control of adult and larval western cherry fruit fly (Diptera: Tephritidae). J. Econ. Entomol. 99: 1722–1732.

Yee, W. L., and D. G. Alston. 2012. Behavioral responses, rate of mortality, and oviposition of western cherry fruit fly exposed to malathion, zeta-cypermethrin, and spinetoram. J. Pestic. Sci. 85: 141–151.

Yee, W. L., O. Jack, and M. J. Nash. 2007. Mortality of *Rhagoletis pomonella* (Diptera: Tephritidae) exposed to field-aged spinetoram, GF-120, and azinphos-methyl in Washington State. Fla. Entomol. 90: 335–342.

Zhang, R., E. B. Jang, S. He, and J. Chen. 2015. Lethal and sublethal effects of cyantraniliprole on *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae). Pest Manag. Sci. 71: 250–256.