Progressing Integrated Pest Management (Entomology) in macadamias

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Department of Primary Industries

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NSW Department of Primary Industries

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in Macadamias

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19 August 2011

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Purpose of the report:

The purpose of this study was to develop improved Integrated Pest Management (IPM) practices that are less reliant on broad spectrum insecticides in consultation with pest consultants and growers and provide these practices for the macadamia industry.

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1. Table of content

1.1. Abbreviations used in the text

AMS	Australian Macadamia Society
APVMA	Australian Pesticides and Veterinary Medicines Authority
BSB	Banana spotting-bug *Amblypelta lutescens lutescens* (Distant)(Hemiptera: Coreidae)
CTH	Centre of Tropical Horticulture site Alstonville NSW Department of Primary Industries, Formerly Industry & Investment NSW and NSW Agriculture
DPI	NSW Department of Primary Industries NSW
DEEDI	Department of Employment, Economic Development and Innovation (previously Queensland Department of Primary Industries (QDPI))
FSB	Fruitspotting bug (FSB) *Amblypelta nitida* Stål (Hemiptera: Coreidae)
gai	Grams active ingredient
GVB	Green Vegetable bug *Nezara viridula* (Linnaeus) (Hemiptera: Pentatomidae)
HAL	Horticulture Australia Limited
HRDC	Horticultural Research and Development Corporation
MNB	Macadamia nutborer *Cryptophlebia ombrodelta* (Lower) (Lepidoptera: Tortricidae)

2. Media summary

A range of pests cause significant losses to the macadamia industry. There is a need to improve pest management by using Integrated Pest Management (IPM), and approach which incorporates sustainable management options for the complex of key pests.

Key components:

During this study we developed a new IPM strategy that included insecticide screening for fruitspotting bugs (FSB), macadamia lace bugs and banana fruit caterpillar, a pest of macadamias in Bundaberg. We further investigated the potential of alternative FSB host plants as monitoring tools or trap crops, and investigated a parasitic fly as a potential biological control agent. Heat treatments currently used for nut drying were also evaluated for their effectiveness in killing nut in shell pests, which remains a quarantine issue for export.

Key results:

Our research identified a new chemical compound and a plant extract as the most effective alternatives to endosulfan in controlling FSB. Lepidex® gives mixed results; good control in the laboratory tests but not successful in the field. Actara® and the new chemical compound also effectively controlled macadamia lace bug and offer an effective alternative to endosulfan. Lannate® appears to be the most effective control option for banana fruit caterpillar. The shrub *Murraya paniculata* was particularly effective in attracting FSB and has potential as a monitoring tool and trap crop. A parasitic fly has some potential for biological control of FSB. Temperatures of 50ºC killed larvae and pupae of pests in nut in shell

Recommendations for practical application to industry:

Lepidex® is an alternative to endosulfan for FSB control, but low pH of spray solution is important. Lannate® at 2ml/L can be used for management of banana caterpillar. In relation to postharvest pests, the current macadamia nut-in-shell drying regimes are sufficient to kill macadamia pest species in nut in shell.

Recommendations for future R&D:

A better monitoring tool (i.e. trap hedge and pheromone trap) for FSB is needed for more targeted management. Alternatives to endosulfan still require further testing for future registration applications. Biological control for FSB needs to be further investigated. Other pests such as macadamia lace bug, banana fruit caterpillar, felted coccid and scirtothrips also need to be considered in a holistic approach.

3. Technical summary

A significant pest complex including the key pests: macadamia nutborer (MNB) *Cryptophlebia ombrodelta* and fruitspotting bugs (FSB) *Amblypelta* spp. poses a challenge for the macadamia industry and there is a strong desire to improve the current system by utilising Integrated Pest Management (IPM) and incorporate sustainable management options.

Key components:

As part of this research we developed a new IPM strategy that included insecticide screening for fruitspotting bugs (FSB) (*Amblypelta* spp.), macadamia lace bugs *Ulonemia concava* Drake and a *Physatochelia* sp. and banana fruit caterpillar *Tiracola plagiata* (Walker), a pest of macadamias in Bundaberg. We investigated the potential of alternative FSB hosts as monitoring tools and trap crops. The attractance of a plant host is used to modify the behaviour of fruitspotting bugs on the principle of a "Push and Pull strategy", from the main crop to a more attractive host. We investigated the Tachinid fly *Trichopoda giacomellii* (Blanchard) that had been introduced to Australia for biological control of green vegetable bug *Nezara viridula* (L.) as potential biological control agent for *Amblypelta* spp. Heat treatments currently used for nut drying were evaluated regarding their effectiveness in killing nut in shell pests, a quarantine issue for export, with a series of heat treatments investigated.

Key results:

Our research identified one new chemical compound and one plant extract which proved to be the most effective alternatives to endosulfan in controlling *Amblypelta* spp. We identified that there are two different lace bug species. Trichlorfon (Lepidex®) was effective in controlling macadamia lace bug in the loaboratory but not in the field. Thiamethoxam (Actara®) and the new chemical compound also effectively controlled macadamia lace bug and offer alternatives to endosulfan. Methomyl (Lannate®) appears to be the most effective control option for *Tiracola plagiata*. The shrub *Murraya paniculata* was particularly effective in attracting FSB. It has potential as a monitoring tool and trap crop. *Trichopoda giacomellii* has some potential as a biological control agent for *Amblypelta* spp. The fly laid eggs on FSB. Larvae hatched, killed the FSB adults but only developed to pupal stage. Temperatures of 50ºC killed larvae and pupae of pests in nut in shell.

Recommendations for practical application to industry:

Trichlorfon (Lepidex®) is an alternative to endosulfan for FSB control, but low pH of spray solution is important. Methomyl (Lannate®) at 2ml/L can be used for management of *Tiracola plagiata*.. The current macadamia nut-in-shell drying regimes are sufficient to kill macadamia pest species in nut in shell.

Recommendations for future R&D:

A better monitoring tool is needed for more targeted FSB management. The results from the trap crop research needs to be packaged for practical application and further research into pheromones will also be important in developing a monitoring tool for *Amblypelta* spp. Alternatives to endosulfan still require further testing for future registration applications. Biological control for FSB needs to be further investigated, including egg parasitoids and parasites. Other pests such as macadamia lace bugs, Scirtothrips, *Tiracola plagiata* and felted coccid (*Eriococcus ironsidei* Williams) also need to be considered in a holistic approach.

4. Introduction

A significant pest complex, including the key pests macadamia nutborer (MNB) *Cryptophlebia ombrodelta* (Lower) (Lepidoptera: Tortricidae) and fruitspotting bugs (FSB) *Amblypelta nitida* Stål and *Amblypelta lutescens lutescens* (Distant) (Hemiptera: Coreidae (Ironside, 1981; Gallagher *et al*., 2003), still challenges the macadamia industry. The focus of this study is to shift the management of FSB into Integrated Pest Management (IPM) that will also accommodate other pest issues which have arisen during 2006-2010.

The macadamia industry in Australia has been aiming towards:
- more sustainable pest management
- less reliance on broad-spectrum insecticides
- reduced negative environmental impacts
- reduced negative community reactions
- minimised risk of potential pesticide resistance issues
- minimised risk of pesticide residue issues in macadamia
- maintaining future market access

The macadamia industry are now faced with the reality of production without endosulfan for the first time, and our understanding of *Amblypelta* spp. needs to be complete if we are to maintain economic control..

Challenges with regards to FSB management are the following:

- The main problem identified is the constant migration of *Amblypelta* spp. adult into host crops that is diffuclut to monitor. (Waite *et al*., 1993).
- The control of *Amblypelta* spp. once detected was not difficult with access to endosulfan, beta-cyfluthrin or trichlorfon (Waite *et al*., 1993).
- The widespread adoption of cover-spraying to prevent *Amblypelta* spp. population development in the crop is touted as the solution and preferably with endosulfan as endosulfan was the least disruptive option to beneficials (Waite *et al*., 1993).
- The timing of fruitspotting bug control, which begins at flowering (Brimblecombe, 1948), also impacts on many other Hemipteran species of which FSB is the most common. There is an inherit risk in being too specific and not considering other pests.

Prior to 2000 the macadamia industry was characterised by programme and calendar spraying. The use of the egg parasitoid *Trichogrammatoidea fulva* sp. nov. (Nagaraja, 1978) was flagged as a possible leap towards a less spray dependent pest management system for the macadamia industry in Australia (Ironside, 1983). Major differences in the pest complexes in both Queensland and NSW growing districts and excessive pyrethroid use had caused major secondary pest problems (Ironside, 1983; 1987; Treverrow, 1983; 1987). Previous projects have investigated the feasibility of mass-rearing (part of MC96002) and evaluated field releases (MC99001) of *Trichogrammatoidea cryptophlebiae* Nagaraja (Hymenoptera: Trichogrammatidae), which is a biological control agent that was introduced to control MNB in macadamias in 2000 (Sinclair, 1974; Sinclair and Sinclair, 1980; Ironside, 1982; Waite, 1994; Campbell *et al*., 1999; Campbell & Maddox, 2000; Maddox *et al*., 2002). Insecticides were screened at the Centre for Tropical Horticulture (CTH) Alstonville on key pests and beneficials to determine wheter they were compatible with parasitoid use. The success of the parasitoid *T. cryptophlebiae* on the MNB populations also led an enthusiastic charge toward total organic nut production

(Treverrow, 2003), however, with that has come the realisation that other pests actually drive spray decisions and the crop-monitoring for those pests is inadequate.

Relying solely on MNB parasitoids when any of the potentially damaging FSB and GVB are around is not acceptable for total pest management. The need for improving the monitoring and managing FSB has become obvious.

This current project has changed focus from its original aim of finding replacement chemistry to reducing industry impact caused by the removal of endosulfan. The different components of the project are:

1. Laboratory bioassay mortality testing of insecticides on fruitspotting bugs, banana spotting bugs and green vegetable bugs.

2. The best bioassay options to be used in field trials over two or more seasons (to allow for the climatic variables).

    a. Intensive small scale field trials at CTH in Alstonville where FSB were released evenly into the orchard prior to each season including unsprayed and standard sprayed areas that have remained unchanged from 2001-2010.

    b. Commercial macadamia orchard field trials to compare standard chemical applications with softer IPM strategies.

3. Develop monitoring methodology for FSB that will allow year round detection, and test strategies to manipulate FSB behaviour to improve management using alternative hosts and trap hedges.

4. Evaluate the parasitic fly *Trichopoda giacomellii* (Blanchard) (Diptera: Tachinidae) as potential biological control agent for fruitspotting bugs.

Separate issues which are not directly related to fruitspotting bugs but are part of the pest management problem for macadamia include:

1. Insecticide screening (laboratory and field) for macadamia lace bugs (Tingidae).

2. Impact of heat treatment disinfestations on macadamia nutborer, kernel grub and bark beetles (Coleoptera: Scolytidae).

3. Chemical control of banana fruit caterpillar *Tiracola plagiata* (Walker) (Lepidoptera: Noctuidae) an outbreak pest in Bundaberg 2008-2011.

4. Colony maintenance for *Cryptophlebia ombrodelta* and *Trichogrammatoidea cryptophlebiae* at CTH Alstonville.

This project will provide the initial data and background for alternatives to endosulfan for FSB and lace bug control that will be valuable for future registration process. The project aims to obtain valuable ecological information improving the understanding of FSB and improve management, including biological control, trap cropping and monitoring options. The outcome of the heattreatment trials will support export market access by ensuring pest free nut in shell product. The project will further aspire to provide management options for other pest outbreaks such as the banana fruit caterpillar.

5. Laboratory insecticide screening for control of fruitspotting bug *Amblypelta* spp*.* and green vegetable bug *Nezara viridula*

The macadamia industry in NSW and Queensland has relied on endosulfan for the management of fruitspotting bugs (*Amblypelta* spp.) and green vegetable bug *Nezara viridula* (L.) (Hemiptera (Pentatomidae) for at least 20 years. Other compounds that are also currently in use, that are still effective, include the synthetic pyrethroid beta-cyfluthrin, the organophosphates trichlorfon, methidathion and acephate, and the carbarmates methomyl and carbaryl (Pubcris database, APVMA 2010). The feeding mode used by Hemipterans generates a difficult management problem that forced the continued use of contact insecticides until the development of systemic nicotinoid treatments (Corbett, 1974; Yamamoto & Casida, 1999). Neonicatinoids are very effective on aphids, thrips, mealy bugs, and a range of larger Hemiptera. Neonicatinoids are systemic and do pose residual risks in the fruit so that each crop will need to establish a withholding period so that fruit residues are below the limit determined for harvest. Secondary pest outbreaks have previously impacted on the macadamia industry (Treverrow, 1987). A study in avocado showed that methomyl gave similar results to endosulfan as a rotational product to minimise latania scale *Hemiberlesia lataniae* (Signoret) (Hemiptera: Diaspididae) when sprayed throughout the season on avocado. Methomyl was however far more toxic to bees during the flowering period. It is important that we find suitable chemical option within the flowering period as well.

Growers are constantly using mixtures to reduce the need to spray the entire orchard and during the early season, applications of fungicides for the control of Husk spot disease are often combined with insecticides. Trichlorfon has been registered for FSB in macadamias. The efficacy of trichlorfon however has also been linked to the pH of the carrier solution with alkaline hydrolysis claimed to reduce performance (NuFarm LI700 sales brochures regarding Lepidex® use 2008). The compatibility of trichlorfon and different copper based fungicides requires further testing.

The Narrabri based NSW DPI researcher with cotton industries Dr Robert Mensah provided us with two biopesticides (under patent) for testing, which have been effective in cotton work, for investigation in macadamias.

5.1. Materials and Methods

New experimental chemistries were supplied by the chemical companies (Table 5.1.1). Mortality was tested by dipping food sources, or by topical application (1µl) on an individual insect and measuring the mortality rate after a fixed period (2 hours, 1 day, 2 days, 3 days and 7 days). At CTH Alstonville we have been maintaining colonies of FSB, BSB and GVB to allow the rapid testing of potential new molecules before incorporation into larger more expensive field trials (see NSW DPI trials in Waite, 2000; Maddox *et al.,* 2000b and Huwer *et al.,* 2006). FSB are known to be very difficult to rear. Colonies of FSB are maintained and juveniles are kept separate from the breeding adults in all three species The colonies are always in decline so require field collections in South East Queensland during the spring and autumn breeding periods for both *Amblypelta* species to maintain numbers. Assays used F1 progeny from field collections to keep age relatively constant for the test animals.

Table 5.1.1: Pesticides supplied and used in the laboratory bioassay and field trials with green vegetable bug and *Amblypelta* spp. at CTH Alstonville

Chemical Company	Code number	Formulation	Species used in bioassay	Field Trial
Bayer	BYI 08831	240SC	GVB,FSB,BSB	No
Bayer		Thiacloprid 480SC**	BSB, GVB	2007/08
Bayer		Imidacloprid 200SC	BSB,GVB	
Bayer	DC083	200SC	BSB, GVB	
Bayer	DC084	100SC	BSB, GVB	
Bayer	NNI	480SC	BSB,GVB	
Bayer	DC091	200SC	FSB	
Bayer	DC092	200SC	FSB	2009/10/11+
Bayer	DC093	200SC	FSB	
Bayer / Nufarm		Endosulfan 350EC	FSB	2006-2011+ Standard
Bayer		Beta-cyfluthrin 25EC	FSB	2006-2011+ Standard
Nufarm		Lepidex 500EC	FSB	2008-10
Du Pont	HGW86	100SC	BSB, GVB	
Dow	GF2032	240SC	FSB	2010 *
Rhone Poulanc		Fipronil 200SC	FSB	2008/09
Syngenta		Thiamethoxam 250WG**	FSB	2006/07
CRC Cotton		Mensah BG639	FSB	
CRC Cotton		Mensah PE	FSB	2010/11+
		Indoxacarb 150EC	FSB	
		Indoxacarb 200WG	FSB	

* Formulation used in confidential field trial for Dow
**Formulations have been tested in previous studies
+ Formulations are in current field trial studies which will be harvested in August 2011.

Species used in bioassay
BSB = Banana spotting bug = *Amblypelta lutescens lutescens*
FSB= Fruitspotting bug = *Amblypelta nitida*
GVB=green vegetable bug = *Nezara viridula*

Laboratory bioassay techniques

Insecticides are normally provided with a recommended dose range and a suggested wetting agent. The information supplied was adapted and in most cases a serial dilutions in water were required. Solutions were labelled and mixed before the insects were collected for bioassay from the cages. The bioassays were conducted at a 25℃. Each bioassay included a negative control that of water and a standard positive treatment control of beta-cyfluthrin (0.5ml/L) which gives 100% kill at 3 DAT.

Adults (usually) or nymphs were collected in batches of five and immobilised at -18^{o}C for 2.5 - 3 minutes. The insects were placed legs down, using forceps, on a cryolyser and the dose applied to the dorsal thoracic plate with a Hamilton micro syringe delivering 1µl. Recovery from cold immobilisation was after about 1 minute. (Using more 5 individuals per group requires a longer time at -18^{o}C which increases the mortality rate). The treated insects were maintained in 750mL labelled plastic containers with lids and ventilation holes (1mm) for 7 days. Test insect were fed with a small piece of corn in the container Survival was assessed after 2 hours and then each day until day 7. The criterion for death is the lack of leg movement when probed.

Table 5.1.2: Pesticide solution volumes and rates used in the Cabrio compatibility and pH bioassay

Chemical	**Recommended Rates**	**gai used**	**Volume used in mixes**
SpinFlo® 500g/L Carbendazim	50ml/100L	0.0063	0.0125mls in 25mls
Cabrio® 250g/LPyraclostrobin	40ml/100L	0.0025	0.010mls in 25mls
Champ® 375g/Kg Cupric Hydroxide	140g/100L	0.013	0.035g in 25mls
Coppox® 500g/Kg Copper Oxychloride	250g/100L	0.031	0.0625g in 25mls
Thiodan® 350g/L Endosulfan	150ml/100L	0.11	0.3mls in 200mls
Lepidex® 500g/LTrichlorfon	100ml/100L	0.1	0.2mls in 200mls
Bulldock® 25g/L Beta-cyfluthrin	50ml/100L	0.0013	0.05mls in 100mls

Compatibility of insecticides and fungicides used were tested in a separate test with a mixture of both. For the pesticide mixture study, stock solutions of fungicides were prepared at the recommended rates (Table 5.1.2). The solutions were then decanted close to the required volume in 100ml flasks and the additional pesticides were added and then the final volume made up with the original stock solution and mixed under the fume hood. These solutions were allowed to stand (24 hours) and the pH measured with a calibrated TPS Aqua pH meter for a 1 minute period prior to topical application.

In order to establish pH of the pesticide solution the pH meter was calibrated. Calibration buffers were used to set the device each made on distilled water and store at 3^{0}C. These buffers were GB 4.0 (10% potassium hydrogen phthalate), GB 7.0 (10% potassium dihydrogen phosphate + 10% sodium phosphate), GB 9.23 (2% sodium tetraborate) and GB 10.00 (1% sodium bicarbonate + 1% sodium carbonate), and all were allowed to reach room temperature before use.

5.2. Results

Bioassays by topical application showed that Dow GF2032 was not effective at the rates suggested (4ml/L), and a rate greater than 4ml/L maybe required for FSB control (Tables 5.2.1).

Fipronil feeding bioassays from May 2008 were inconclusive, with fipronil only effective against GVB and ineffective against FSB (Table 5.2.2). In contrast, topical application of beta-cyfluthrin (Bulldock®) 0.5ml/L gave 100% mortality against FSB and BSB. Fipronil (0.4ml/L) was the most effective against GVB (Table 5.2.2). The Bayer experimental formulation BYI 08831 failed to give any control (Table 5.2.2) and Bulldock® at 0.05ml/L was only 50% effective (Table 5.2.2).

Topical application testing in June 2008 was restricted to BSB and GVB only. Only the standard treatment (Bulldock®) at 0.5ml/L gave 100% mortality (Table 5.2.3). Doses of fipronil at 0.4ml/L, DC084 at 4ml/L and DC083 at 0.3ml/L were able to kill all GVB but not BSB (Table 5.2.3). DuPont HGW86 failed to kill either test species (Table 5.2.3). Bayer NNI, BYI 08831, Confidor® and Calypso® were ineffective at the rates trialled (Table 5.2.3). Further dose range testing for the DC083 improved mortality rates to 90% at 0.6ml/L for BSB and GVB adults, and DC084 achieved similar efficacy at t 4-8ml/L on GVB adults (Table 5.2.4).

Bayer DC091, DC092 and DC093 were tested against FSB and macadamia lace bug. DC091 was effective at 0.6ml/L and DC092 in particular showed promise against both pest species at 1ml/L (Table 5.2.5 see chapters 6 and 9). DC093 failed to give effective control (Table 5.2.5). The plant extract was effective at 2% v/v against FSB adults and was used in a field trial in season 2010/2011 at CTH (Table 5.2.5).

Trichlorfon has been used against FSB for decades, (Ironside, 1983) its efficacy however was questioned by many growers who anecdotally preferred endosulfan. In June 2008 a Bayer Dipterex® formulation was trialled at 1ml/L on BSB and GVB adults with mixed results on BSB it gave 80% mortality and GVB 20% (Table 5.2.3).

Indoxacarb has been suggested as a possible replacement for endosulfan since 2001. Our early work on GVB with the Avatar® formulation of indoxacarb was not supportive and trials in 2010 with BSB and GVB adults were ineffective (Table 5.2.5). An alternative formulation labelled Steward® (150gm/L indoxacarb) was sourced from cotton growers in NSW and it appears to be more effective against FSB adults at 4ml/L (2011 lab assays).

The pH studies revealed that the addition of copper hydroxide formulations would give common insecticide/fungicide mixtures a pH over 10 (Table 5.2.5). When spray solutions were allowed to stand for 20 hours before use endosulfan and trichlorfon efficacy on BSB adults of was reduced when mixed with fungicides (only beta-cyfluthrin (Bulldock® at 0.5ml/L) and endosulfan alone at 1.5ml/L gave full control (Table 5.2.5). This test will be repeated with FSB adults when numbers permit a full trial and the dose rate can be shifted to include a 2.0ml/L trichlorfon (Lepidex ®) mixture series.

Table 5.2.1: Topical application results for 3 day exposures of *Amblypelta nitida* adults to 1µL doses of insecticide. Run 1 (5/11/2010) and Run 2 (09/11/2010). Mortality defined as no visible leg movement.

Run	Treatment	Adults tested	3 day mortality	Eggs laid
1	Untreated	6♀ 4♂	0♀ 0♂	37
1	Bulldock® (0.5ml/L)	3♀ 7♂	3♀ 7♂	0
1	GF 2032 (0.4ml/L)	5♀ 5♂	1♀ 3♂	7
1	GF 2032 (0.8ml/L)	4♀ 6♂	2♀ 5♂	9
2	Untreated	3♀ 2♂	0♀ 0♂	0
2	GF 2032 (1.0ml/L)	2♀ 3♂	0♀ 2♂	7
2	GF 2032 (2.0ml/L)	3♀ 2♂	1♀ 2♂	0
2	GF 2032 (4.0ml/L)	2♀ 3♂	1♀ 3♂	0

Table 5.2.2: Mortality after 3 day exposure to insecticides in either a contact (C) bioassay with 1µl dose applied topically or feeding (F) feeding with dietary corn dipped in the test compound and allowed to dry. Assay date 14/5/2008. Insects maintained 25 ^{0}C and a 14:10 dark: light photoperiod.

Chemical	**Rate**	**Number of test insects per replicate.**	**Assay Type**	**Mortality in numbers after 3 days**					
				Amblypelta nitida		***Amblypelta lutescens***		***Nezara viridula***	
	ml/L	**#**	**C/F**	**Adults**	**Nymphs**	**Adults**	**Nymphs**	**Adults**	**Nymphs**
Untreated control		5	F	2	2	1	1	2	2
Hasten® (H)	1.0	5	F	2	2	3	0	0	1
BYI 08831 240SC +(H)	0.4	5	F	1	1	3	0	1	1
Fipronil® 200SC +(H)	0.4	5	F	2	0	3	1	5	5
Bulldock® 25EC + (H)	0.5	5	F	2	0	4	2	4	4
Bulldock® 25EC +(H)	0.05	5	F	3	2	3	0	2	0
Untreated control		5	C	2	0	1	1	1	1
Hasten® (H)	1.0	5	C	1	1	3	1	1	0
BYI 08831 240SC +(H)	0.4	5	C	2	4	3	1	0	0
Fipronil® 200SC +(H)	0.4	5	C	5	3	5	5	4	5
Bulldock® 25EC + (H)	0.5	5	C	5	5	5	5	4	3
Bulldock® 25EC +(H)	0.05	5	C	5	1	4	4	1	3

Table 5.2.3: Mortality after 3 day exposure to insecticides in a contact (C) bioassay with 1µl dose applied topically. Assay date 3/6/2008. Insects maintained at 25 ^{0}C and a 14:10 dark: light photoperiodic regime.

Chemical	Rate	Company	Number of test insects per replicate	Assay Type	Mortality in numbers after 3 days	
					Amblypelta lutescens	***Nezara viridula***
	ml/L		**#**		**Nymphs**	**Adults**
Hasten® (H)	1.0		5	C	0	0
Agral®	2.0		5	C	1	0
DuWett®	1.0		5	C	1	1
Fipronil® 200SC +(H)	0.4		5	C	4	5
NNI 480SC +(H)	0.2	Bayer	5	C	1	0
BYI 08831 240SC +(H)	0.4	Bayer	5	C	1	2
DC084 100SC + (H)	4.0	Bayer	5	C	4	5
DC083 200SC +(H)	0.3	Bayer	5	C	3	5
Confidor® 200SC	0.9	Bayer	5	C	1	4
Calypso® 480SC	0.4	Bayer	5	C	2	2
HGW86 100SC +(H)	2.0	DuPont	5	C	2	0
HGW86 100SC +(H)	1.0	DuPont	5	C	1	0
Bulldock® 25EC + (H)	0.5		5	C	5	5

Table 5.2.4: Mortality after 3 day exposure to insecticides in a contact (C) bioassay with 1µl dose applied topically. Assays set 17/6/2008, and 8/7/2008. Insects maintained at 25 ^{0}C and a 14:10 dark: light photoperiodic regime.

Chemical	Rate	Company	Number of test insects per replicate	Assay Type	Mortality in numbers after 3 days	
					Amblypelta lutescens	***Nezara viridula***
	ml/L		**#**		**Nymphs**	**Adults**
Hasten® (H)	1.0		5	C	0	1
DC083 200SC +(H)	0.075	Bayer	5	C	2	2
DC083 200SC +(H)	0.15	Bayer	5	C	4	3
DC083 200SC +(H)	0.3	Bayer	5	C	2	5
DC083 200SC +(H)	0.6	Bayer	5	C	5	4
Dipterex® + (H)	1.0		5	C	4	1
Bulldock® 25EC + (H)	0.5		5	C	5	4
Hasten®	1.0		10	C		1
NNI 480 SC	0.2	Bayer	10	C		1
DC084 100SC + (H)	1.0	Bayer	10	C		2
DC084 100SC + (H)	2.0	Bayer	10	C		5
DC084 100SC + (H)	3.0	Bayer	10	C		9
DC084 100SC + (H)	4.0	Bayer	10	C		9
Bulldock 25EC + (H)	0.5		10	C		8

Table 5.2.5: Mortality after 3 day exposure to insecticides in a contact (C) bioassay with 1µl dose applied topically. Assays set 17/7/2008 and 18/5/2010 and 4/4/2011. Insects maintained at 25 ^{0}C and a 14:10 dark: light photoperiodic regime.

Chemical	**Rate**	**Company**	**Number of test insects per replicate**	**Assay Type**	**Mortality at 3 days**
	ml/L		**#**		***Amblypelta lutescens* adults**
Untreated control			10	C	0
Hasten®	1.0		10	C	0
DC091 200SC +(H)	0.6	Bayer	10	C	9
DC092 200SC +(H)	1.0	Bayer	10	C	10
DC093 200SC +(H)	0.3	Bayer	10	C	1
DC084 100SC + (H)	4.0	Bayer	10	C	8
Bulldock® 25EC + (H)	0.5		10	C	10
Untreated control					3
Avatar® 200WG indoxacarb	0.5gm		10	C	3
BioPest® pest oil	20		10	C	3
Canopy® pest oil	20		10	C	2
Mensah BG639	5	CRC cotton	10	C	2
Mensah BG639	10	CRC cotton	10	C	3
Mensah PE	10	CRC cotton	10	C	5
Mensah PE	20	CRC cotton	10	C	10
Bulldock® 25EC	0.5				9
Untreated control			10	C	2
Steward® 150gm/L indoxacarb	4.0		10	C	10
Steward® 150gm/L indoxacarb	2.0		10	C	9
Steward® 150gm/L indoxacarb	1.0		10	C	8
Bulldock® 25EC	0.5		10	C	9

Table 5.2.6: Fungicide compatibility with Lepidex® (trichlorfon) use and impact on efficacy against *Amblypelta lutescens*. (*C*ommon tank mixtures of chemicals used by macadamia growers in NSW and Queensland.) Solutions were left to stand for 24 hours before application and then shaken before two pH measurements. Adult mortality after 3 day exposure in a contact (C) bioassay with 1uL dose applied topically. Assays date 14/4/2009. Insect maintained at 25°C and a 14:10 dark: light photoperiodic regime.

Chemicals	**Rate**	**pH**		***Amblypelta lutescens***	
	ml/L	15 sec	30 Sec	No of tested adults	No. Dead adults
Water		7.4	7.2	10	2
Cabrio®	0.4	7.6	7.5	10	1
Spin Flo	0.5	7.4	7.3	10	3
Bulldock® 25 EC	0.5	7.5	7.3	10	10
Copper Oxychloride (CuOCl)	2.5g	7.8	7.9	10	0
Copper Hydroxride (CuOH)	1.4g	10.5	10.6	10	4
Lepidex®	1.0	4.4	4.3	10	3
Lepidex® + Spin Flo®		4.4	4.4	10	9
Lepidex® + Spin Flo® + CuOCl		6.2	6.1	10	5
Lepidex® + Spin Flo® + CuOH		7.6	7.7	10	3
Lepidex® + Cabrio®		4.2	4.1	10	1
Lepidex® + Cabrio® + CuOCl		6.1	6.0	10	4
Lepidex® + Cabrio® +CuOH		7.5	7.5	10	1
Endosulfan	1.5	5.1	5.1	10	10
Endosulfan+Spin Flo®		6.8	6.6	Not done	
Endosulfan+Spin Flo® + CuOCl		7.4	7.3	Not done	
Endosulfan + Spin Flo® + CuOH		10.1	10.2	10	3
Endosulfan + Cabrio®		5.6	5.5	10	5
Endosulfan + Cabrio® + CuOCl		7.4	7.3	10	5
Endosulfan+ Cabrio® +CuOH		10.0	10.1	10	5

6. Field trials at the Centre for Tropical Horticulture (CTH) in Alstonville and on commercial farms

This work has followed on directly from previous results (Huwer *et al.* 2006). Using both laboratory insecticide bioassay data and biological control agents for MNB and FSB (since 2000) we have been able to compare less conventional approaches to managing FSB within an orchard (both short and longer term). The work was done at an individual tree level with the aim of generating data on production yield, crop loss and spatial pest distribution. In essence, across four varieties of macadamia, we were able compare an integrated pest management programme incorporating both sprays and beneficial insects against a standard spray regime.

It is hard to justify any type of treatment unless the value of the damaged crop can be determined (Huwer *et al.,* 2006). Our methods for monitoring and treatment applications were based on the following findings:

- Researchers from all major macadamia growing districts of the world had investigated MNB monitoring (e.g. Jones, 1994; Newton & Odendaal, 1988; Sloan, 1998; Ironside, 1981, 1988; Sinclair, 1974; Chang, 1995, Campbell *et al.*, 1999).

- Due to significant periods of physiological drop prior to about 10 weeks post-anthesis (Trueman & Turnbull, 1994), it was important not to start monitoring until after that time when most of the nutlets would stay on the raceme through to maturity.

- With regards to the need for FSB monitoring (Brimblecombe, 1948) and subsequent spray threshold development (Ironside, 1981) The treatment thresholds used by pest consultants is around 3% damage to fallen green fruit (O'Hare *et al.,* 2004).

CTH trials

The aim was to compare the impact of four different treatments and four different macadamia varieties on key macadamia pests. The trial focussed on controlling MNB initially, however since 2004 the focus switched to FSB. Some areas within the trial were designated as long term biological control areas (rather than using a new randomised plot trial every year) to get a longer term view of what happens in a defined area. The treatments included biological control only, IPM and the industry standard. The experimental treatments varied with each season in order to study particular aspects of control. Blocks were seeded with FSB each season with live FSB to ensure pest pressure (not possible on commercial farms because of the damage risk to commercial crops).

Commercial farm trials

Field trials on commercial farms testing the industry standard broad-spectrum insecticide sprays versus IPM (including biological control and insect growth regulator for MNB management) were done. All monitoring for MNB and FSB in the on-field trials was done by professional pest consultants. Their input established an important link between the researchers and the growers.

6.1 Materials and Methods

Orchard design

The small scale field trial at CTH Alstonville which was planted in 1998, used macadamia trees of the varieties 246, 741, 849 and A4. Trees were spaced at 5 metres within rows and 7 metres between rows (equivalent to 285 trees per hectare). The experimental unit was defined as a block containing three rows of three trees. The varieties were allocated to blocks in a Latin square array and the blocks were separated by a buffer row of 246 trees (Figure 6.1.1). Variety 246 was chosen for cross pollination (McConchie *et al*., 1997).

Table 6.1.1: Insecticides tested and rates used from seasons 2006-2007 to 2009-2010

Pesticide			Rate of product applied	gai/L
Technical name	**Trade name**	**Formulation***		
Acephate	Orthene®	750 WP	1.0 g/L	0.75
Beta-cyfluthrin	Bulldock®	25 EC	0.5 ml/L	0.0125
Endosulfan	Thiodan®	350 EC	1.5 ml/L	0.525
Fipronil	Regent®	200 SC	0.4ml/L	0.08
Methoxyfenozide	Prodigy®	240 SC	0.4ml/L	0.096
Tefubenozide	Mimic®	700 WP	0.086 g/L	0.0602
Thiacloprid	Calypso®	480 SC	0.2ml/L	0.096
Thiamethoxam	Actara®	250 WG	0.3 g/L	0.075
Trichlorfon	Lepidex®	500 EC	1.5ml/L	0.75
New compound	Bayer 092	200 SC	1.0ml/L	0.2
Carbendazim	Spin Flo®	500 g/L	0.5ml/L	0.25
Pyraclostrobin	Cabrio®	250 g/L	0.4ml/L	0.1

* Level of active ingredient per kilogram or litre of product in a range of forms

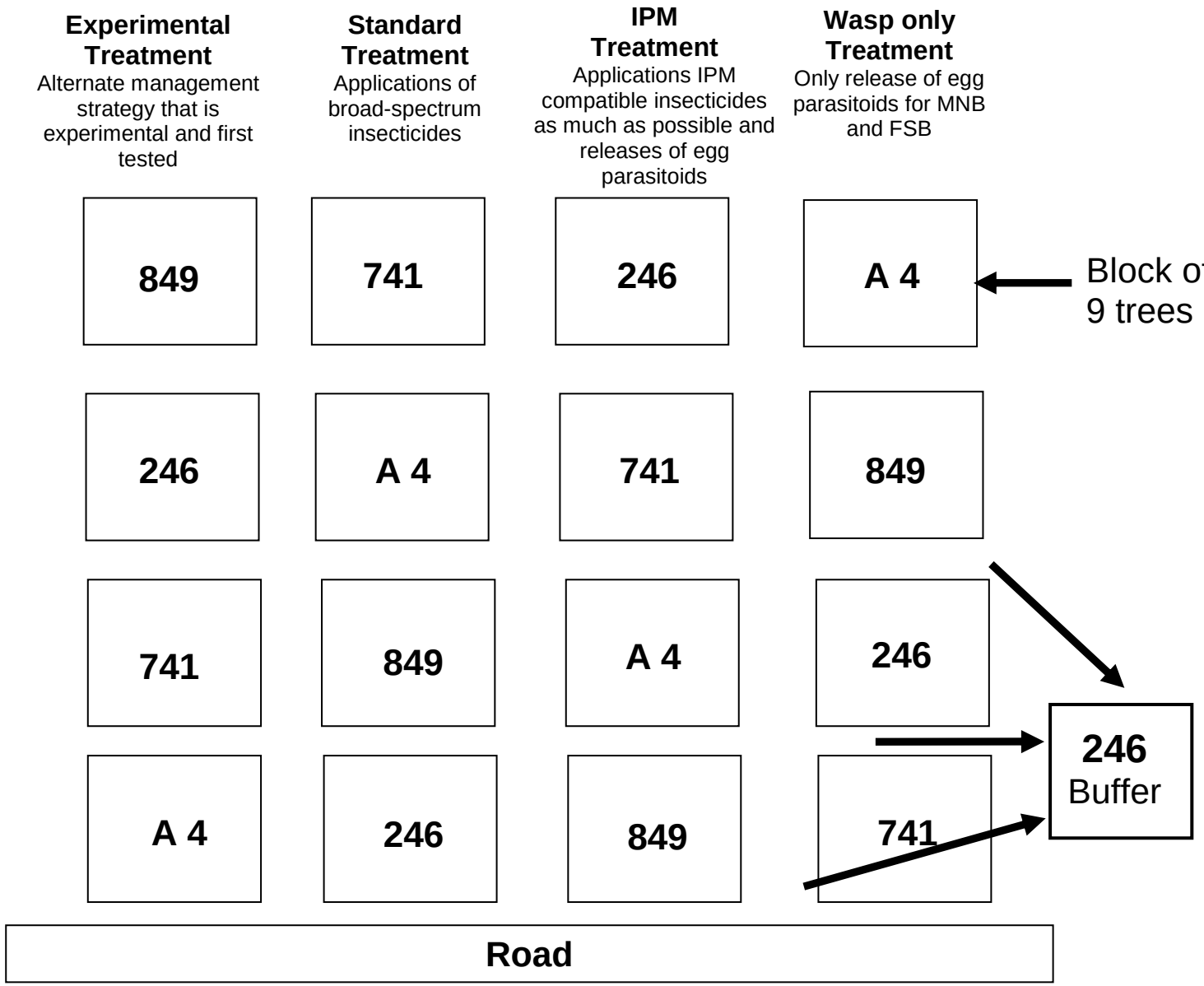

Figure 6.1.1: Entomology orchard layout: Four separate treatments (Experimental, Standard, IPM and Wasp only) were applied along 3 rows of trees that included blocks of 9 trees of 4 varieties of macadamias (246, 741 849 and A4). Each treatment block of 9 trees was buffered by trees of the variety 246.

Table 6.1.2: Schedule for pesticide applications and releases of biological control agents in different treatments in CTH orchard during the 2006/2007 season

Treatment	**September 2006**	**October 2006**	**December 2006**	**January 2007**	**February 2007**
Standard		Endosulfan	Acephate	Beta-cyfluthrin	Beta-cyfluthrin
IPM	Carbendazim Thiame-thoxam*	Thia-methoxam* Wasps *C.d.*x1	Beta-cyfluthrin Wasps *T.c.* x 1 *C.d.*x1	Tebu-fenozide Wasps *T.c.* x2	Beta-cyfluthrin Wasps *T.c.* x 1
Experimental (Standard + flower treatment)	Carbendazim Endosulfan	Endosulfan	Acephate	Beta-cyfluthrin	Beta-cyfluthrin
Wasp only		Wasps *C.d.*x1	Wasps *T.c.* x 1 *C.d.*x1	Wasps *T.c.* x2	Wasps *T.c.* x 1

*=not registered
Biological control agents: *T.c. = Trichogrammatoidea cryptophlebiae*
(egg parasitoids) *C.d. = Centrodora darwini*

Table 6.1.3: Schedule for pesticide applications and releases of biological control agents in different treatments in CTH orchard during the 2007/2008 season

Treatment	**November 2007**	**December 2007**	**January 2008**	**February 2008**
Standard (+ mulching)	Endosulfan	Acephate	Beta-cyfluthrin	Beta-cyfluthrin
IPM	Thiacloprid*	Thiacloprid*	Beta-cyfluthrin Wasps *T.c.* x1	Methoxy-fenozide
Experimental (- mulching)	Endosulfan	Acephate	Beta-cyfluthrin	Beta-cyfluthrin
Wasp only	Wasps *T.c.* x2 *C.d.*x1	Wasps *T.c.* x 1	Wasps *T.c.* x2	

*=not registered
Biological control agents: *T.c. = Trichogrammatoidea cryptophlebiae*
(egg parasitoids) *C.d. = Centrodora darwini*

Table 6.1.4: Schedule for pesticide applications and releases of biological control agents in different treatments in CTH orchard during the 2008/2009 season

Treatment	September 2008	October 2008	November 2008	December 2008	January 2009	February 2009
Standard		Endosulfan x2 Carben-dazim x2	Acephate	Beta-Cyfluthrin	Beta-Cyfluthrin	
IPM	Wasps *T.c.* x 1	Trichlorfon x2 Carben-dazim x2	Beta-Cyfluthrin Wasps *T.c.* x 1	Beta-Cyfluthrin Wasps *T.c.* x 1	Methoxy-fenozide Wasps *T.c.* x2	Wasps *T.c.* x 1
Experi-mental		Pyraclo-strobin x2	Fipronil*	Beta-Cyfluthrin	Beta-cyfluthrin	
Wasp only	Wasps *T.c.* x 1		Wasps *T.c.* x1 *C.d.*x1 (in block 4)	Wasps *T.c.* x 1	Wasps *T.c.* x2	Wasps *T.c.* x 1

*=not registered
Biological control agents: *T.c. = Trichogrammatoidea cryptophlebiae*
(egg parasitoids) *C.d. = Centrodora darwini*

Table 6.1.5: Schedule for pesticide applications and releases of biological control agents in different treatments in CTH orchard during the 2009/2010 season

Treatment	October 2009	November 2009	December 2009	January 2010	February 2010
Standard	Endosulfan Carbendazim	Endosulfan Carbendazim	Beta-Cyfluthrin	Beta-Cyfluthrin	
IPM	Trichlorfon Carbendazim	Trichlorfon Carbendazim	Beta-Cyfluthrin	Methoxy-fenozide Wasps *T.c.* x1	Wasps *T.c.* x1
Experi-mental	Bayer 092 Pyraclo-strobin	Bayer 092 Pyraclo-strobin	Beta-Cyfluthrin	Beta-Cyfluthrin	
Wasp only			Wasps *T.c.* x2	Wasps *T.c.* x2	Wasps *T.c.* x1

*=not registered
Biological control agents: *T.c. = Trichogrammatoidea cryptophlebiae*
(egg parasitoids) *C.d. = Centrodora darwini*

Treatments

The CTH trial compared four different treatments:

1. Wasp only: Only release of egg parasitoids for MNB and FSB
2. IPM: Applications IPM compatible insecticides as much as possible and releases of egg parasitoids
3. Standard: Applications of broad-spectrum insecticides
4. Experimental Experimental strategies including application of at the time unregistered insecticides and fungicides, new chemicals or new use for chemicals

Application rates for different insecticides used are listed in Table 6.1.1 while details and timing of treatments applied are listed in Tables 6.1.2 to 6.1.5. For orchard management reasons and separation of treatments, the treatments had to be overlayed in rows. (It would have been impossible to keep egg parasitoids out of standard and experimental treatments in a true randomised block design (Figure 6.1.1)). This design gave the opportunity of quantifying, in the longer term, the effects of a treatment with no pesticide application (since 2000).

During all four seasons (2006/2007 – 2009/2010) FSB adults were released in each trial block. A pair of FSB was released two weeks before the first insecticide applications. Males were released in tree 1 of each block and females in tree 9.

Orchard management

All trees received an equal fertilizer regime comprising a composite of Rustica® (superphosphate blend) and organic manures in alternate seasons at normal industry rates (O'Hare *et al.,* 2004). Soil tests were conducted annually to check soil pH and canopy pruning done to keep lower limbs high enough for inter row mowing and canopy maintenance to a 4x4m minimum size. (Storm damage destroyed some plots which were replanted but plants too young to crop have not been included in any analysis.)

6.1.2. Field monitoring methodology

Pest pressure and parasite levels were monitored and recorded each season. The crop loss caused by each pest within the treated area was also determined.

Pheromone trapping was used to establish the presence of MNB in the orchard. However, the count of MNB eggs on macadamia nuts provided a more robust decision making tool. Treatment thresholds based on the presence of live MNB eggs were established by Ironside (1988) and O'Hare *et al.* (2004) were as follows:

1 October – 15 December:	Number of live MNB eggs = 1 in 100 nuts
16 December – 31 January:	Number of live MNB eggs = 2 in 100 nuts
1 February – 28 February:	Number of live MNB eggs = 3 in 100 nuts

The orchard was monitored for macadamia lace bug during flowering by visual assessment. Monitoring for MNB and FSB was usually carried out between early November and March between 2007 and 2010.

Macadamia lace bug monitoring

Racemes were randomly checked with OptiVISOR® binocular headband magnifiers (5x magnification) for the presence of macadamia lace bug nymphs, cast skins or adults (see also Chapter 9).

MNB monitoring

MNB monitoring was a combination of pheromone trapping of moths and oviposition counts at fortnightly intervals (minimum) during the high risk periods of late spring and summer. During this study, pheromone traps were placed at the CTH orchard and at two locations in mangroves near Ballina (Ballina West – Burns Point and North Creek) to monitor the movement of MNB outside the orchard.

Pheromone lures were based on the Sinclair & Sinclair (1980) formulation and sold as impregnated rubber septa (Dr. Richard Vickers, Canberra). Delta traps used were made by IPM Tech Inc. (Portland, Origon and plates contain a TAC GEL® film which captures the moths as they land. Moths were generally removed from the plates at weekly intervals. Lures were stored in the freezer or eskies until required and changed fortnightly while TAC GEL® plates were changed every 6-8 weeks (Huwer *et al.,* 2006). Two week old lures catch 80% that of fresh lures, which needs to be considered when calculating catch numbers (Campbell *et al.,* 1999). Trap catches are presented as moth/trap/day (Campbell *et al.,* 1999).

From the first week of November each season 36 racemes (if possible four racemes per tree) were tagged and labelled in each block of nine trees at the CTH site. These tagged racemes were checked fortnightly until nut harvest in March each season. Magnifiers (5x) were used to count the number of hatched MNB eggs, live eggs, larval tunnels and fresh and emerged parasitised eggs. The treatment threshold used was one live egg per 100 nuts sampled. If parasitism of MNB eggs in the IPM area reached 30% insecticide applications were avoided.

Parasitism rate again was expressed as the ratio of live parasitised eggs per live host eggs per 100 nuts sampled (Knipling & McGuire, 1968; Catling & Aschenborn, 1974; Newton & Odendaal, 1990; Maddox *et al* 2002; Huwer *et al.,* 2006).

FSB monitoring

Early in the season nuts damaged by FSB drop and the nut drop can be easily monitored by sampling green nuts on the ground. Cutting with a knife through the middle of dropped green nuts reveals the FSB damage. Nut drop under each tree

was monitored fortnightly from the middle of October to mid December each season (Huwer *et al.,* 2006). A quadrat (0.4m x 0.4m) was thrown within 1 m of the trunk below each test tree and the green fallen nutlets collected. These nutlets were cut open with 60 mm hand knives and examined for FSB feeding marks using the OptiVISOR® magnifiers (Huwer *et al.,* 2006). Both the number of nuts per quadrat sample and those with visible feeding marks were recorded.

Thick shelled macadamia varieties are less susceptible to FSB and do not sustain as much late season damage. Thin shelled varieties can be susceptible to damage until maturity and need monitoring beyond the early nut drop phase in mid December. From mid December FSB damaged nuts usually stay in the tree, which greatly limits the fallen nut monitoring approach. From mid December to March, visual assessments of individual trees were made and sightings of FSB, including all life stages, were recorded.

6.1.3. Harvest methodology

Mature nuts were harvested during the first week of each month between March and August each year. Nuts under each treatment tree were collected by hand and weighed using a clock face scale (max. 25kg with a min. reading accuracy of 50g) mounted on a steel tripod. A standard 4kg weight was used for scale calibration. A random sub-sample of up to 30 nuts with green husk (if possible) was taken for each individual tree. The sample was put into a plastic net and the weight and number of nuts recorded. For March and April harvests the samples of nuts in husk were examined for MNB damage and eggs as previously described. Nut samples were then dehusked and processed as described in (Huwer *et al.*, 2006).

6.1.4. Kernel recovery methodology

Husk assessment

The husk of nut samples during the March and April harvests were checked for evidence of MNB and egg parasitoids.

The following parameters were recorded:

- Live MNB eggs (not hatched and not parasitised)
- Old MNB eggs (hatched MNB – not parasitised)
- Live parasitised MNB eggs (not hatched)
- Old parasitised eggs (hatched parasitoids)

After checking the husk of the nut samples for MNB eggs, the nuts were dehusked. Nut numbers and wet in-shell weights were recorded. Nut in shell samples were then put into plastic nets and placed in a dehydrator for drying: 48 hours at 38℃, followed by 48 hours at 45℃ and a further 48 hours at 60℃ to achieve 1.5% moisture content (AMS, 2001).

Once dried samples were counted, weighed and dry nut in shell weights recorded. Nuts were then cracked, poured on a sieve and any parts >2mm were sorted into the following categories:

- MNB damage
- FSB damage
- Kernel grub damage
- Fungus
- Discolouration
- Germination
- Immature kernel
- Sound kernel

After sorting, the sound kernel weight was determined. Sound kernel then floated in a bowl of water to separate mature kernel (with a higher oil content) that floated, from immature kernel (with a lower oil content) that oil sank. The immature kernels were discarded and the mature kernels counted, placed into plastic bags and returned to the dehydrator for 24 hours at 60℃. The nut samples were then re-weighed again and the A-grade kernel fraction of the nut sample calculated (Huwer *et al.*, 2006).

The average nut yield per tree was expressed as dry nut in shell (DNIS) at 10% moisture content.

6.1.5. Statistics

Strategy for the analysis

A Latin square experimental design was used and the appropriate analysis of variance table is shown in Table 6.1.5.1

Table 6.1.5.1: Analysis of variance table for the CTH entomology orchard trials for a complete single season

Term	Degrees of freedom
Row of blocks	3
Column of blocks (the treatments)	3
Varieties	3
Experimental error	6

It should be noted that the analysis does not allow a formal test of the interaction between variety and treatment as this term is confounded with the experimental error. If an interaction is truly present, the experimental error will be inflated and so the tests of variety and treatment impacts could be regarded as “conservative”.

Under the linear model that drives this analysis of variance table, prediction of the outcome for a particular combination of variety and treatment will be biased if an interaction is truly present. Therefore presentation of the results should be in terms of “main effects”. That is, a study of the variety outcomes averaged over treatments and the treatment outcomes averaged over varieties.

The repeated measures aspect of the data can be handled by constructing an analysis of variance that “nests” the effects of time and interactions with treatment and variety within the experimental blocks as listed in Table 6.1.5.2.

Reporting of the results will be in terms of the outcome of the hypothesis tests implied in the table above and presentations to display the response due to treatments and the response due to varieties over time.

Field monitoring sampling strategy

The blocks were monitored each season from November to February/March with measurements recorded at approximately fortnightly intervals. Each nut was examined for signs of MNB and parasitism of MNB eggs and the total counts per raceme were recorded on a total of 36 racemes monitored per block. Practical limitations meant that the number of racemes sampled per tree were not consistent. Tables 6.1.5.3 to 6.1.5.6 give the number of racemes recorded per tree at the first measurement of each season. While four racemes per tree was a common unit of measurement the raceme counts over all trees ranged from zero to ten.

Table 6.1.5.2: Analysis of variance table for the CTH entomology orchard trials for multiple observations within a season

	Term	Degrees of freedom
Block Stratum:	Row of blocks	3
	Column of blocks (the treatments)	3
	Varieties	3
	Main block error	6
Sub-block Stratum:	Time	7
	Variety:Time	21
	Treatment: Time	21
	Sub-block error	63

Aims of analysis for field monitoring

The aims of this analysis are to compare the performance of varieties and pest management strategies over time in terms of:

- “Yield potential” – the number of nuts per raceme retained over time
- Pest incidence – the number of MNB eggs, stung nuts and FSB evidence over time
- Parasitism of MNB – the number of MNB eggs showing evidence of parasitism over time.

Strategy for handling the field monitoring data

The experimental unit, the material to which a particular combination of variety and treatment was allocated, consists of a block of nine trees. Therefore it is sensible to aggregate the information to the block level. That is, the analyses will operate in terms of the total nuts per block, total green nuts per block, total damaged nuts per block etc.

Table 6.1.5.3: Number of racemes monitored at the first sampling of the 2006/2007 season on each of the 9 trees within each of the 16 blocks

		Tree number								
		1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
Block	**1**	4	4	4	4	4	4	4	4	4
	2	6	6	0	4	4	4	4	4	4
	3	6	0	6	4	4	4	4	4	4
	4	4	4	4	4	4	4	4	4	4
	5	4	4	4	4	4	4	4	4	4
	6	4	4	4	4	4	4	4	4	4
	7	4	6	0	6	4	4	4	4	4
	8	4	4	4	4	4	4	4	4	4
	9	4	4	4	4	4	4	4	4	4
	10	4	4	4	4	4	4	4	4	4
	11	4	4	4	4	4	4	4	4	4
	12	4	4	4	4	4	4	4	4	4
	13	4	4	4	4	4	4	4	4	4
	14	4	4	4	4	4	4	4	4	4
	15	4	4	4	6	6	0	0	6	6
	16	4	4	4	4	4	4	6	6	0

Table 6.1.5.4: Number of racemes monitored at the first sampling of the 2007/2008 season on each of the 9 trees within each of the 16 blocks

		Tree number								
		1	2	3	4	5	6	7	8	9
Block	1	4	4	4	4	4	4	4	4	4
	2	6	6	0	4	4	4	4	4	4
	3	6	0	6	4	4	4	4	4	4
	4	4	4	4	4	4	4	4	4	4
	5	4	4	4	4	4	4	4	4	4
	6	4	4	4	4	4	4	4	4	4
	7	4	6	0	6	4	4	4	4	4
	8	4	4	4	4	4	4	4	4	4
	9	4	4	4	4	4	4	4	4	4
	10	4	4	4	4	4	4	4	4	4
	11	4	4	4	4	4	4	4	4	4
	12	4	4	4	4	4	4	4	4	4
	13	4	4	4	4	4	4	4	4	4
	14	4	4	4	4	4	4	4	4	4
	15	4	4	4	6	6	0	0	6	6
	16	4	4	4	4	4	4	6	6	0

Table 6.1.5.5: Number of racemes monitored at the first sampling of the 2008/2009 season on each of the 9 trees within each of the 16 blocks

		Tree number								
		1	2	3	4	5	6	7	8	9
Block	1	4	4	4	4	4	4	4	4	4
	2	4	4	4	4	4	4	4	4	4
	3	6	0	6	4	4	4	4	4	4
	4	4	4	4	4	4	4	4	4	4
	5	4	4	4	4	4	4	4	4	4
	6	4	4	4	4	4	4	4	4	4
	7	4	4	4	4	4	4	4	4	4
	8	4	4	4	4	4	4	4	4	4
	9	4	4	4	4	4	4	4	4	4
	10	4	4	4	4	4	4	4	4	4
	11	4	4	4	4	4	4	4	4	4
	12	4	4	4	4	4	4	4	4	4
	13	4	4	4	4	4	4	4	4	4
	14	4	4	4	4	4	4	4	4	4
	15	4	4	4	6	6	0	4	4	4
	16	4	4	4	4	4	4	6	6	0

Table 6.1.5.6: Number of racemes monitored at the first sampling of the 2009/2010 season on each of the 9 trees within each of the 16 blocks

		Tree number								
		1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
Block	**1**	4	4	4	4	4	4	4	4	4
	2	4	4	6	0	2	4	6	7	3
	3	8	4	0	2	10	4	3	3	2
	4	4	4	4	4	4	4	4	4	4
	5	6	6	6	4	4	0	0	6	4
	6	4	4	4	4	4	4	4	4	4
	7	4	4	4	4	4	4	4	4	4
	8	4	4	4	4	4	4	4	4	4
	9	4	4	4	4	4	4	4	4	4
	10	4	4	4	4	4	4	4	4	4
	11	4	4	4	4	4	4	4	4	4
	12	4	4	4	4	4	4	4	4	4
	13	4	4	4	4	4	4	4	4	4
	14	4	4	4	4	4	4	4	4	4
	15	4	8	8	4	0	0	4	4	4
	16	6	4	6	2	6	4	4	4	0

Data Analysis

The average number of viable MNB eggs found on nuts, the percentage of parasitism by *T. cryptophlebiae*, the percentage of MNB damaged or tunnelled nuts, the number of MNB male moths caught in pheromone traps and the percentage of FSB damaged nuts early in the season (before shell hardening), were determined. The data were analysed for each season and over the whole period of the project (as in Huwer *et al.*, 2006).

The numbers of MNB eggs recorded and the numbers of male MNB moths caught in the pheromone trap were log transformed (Log (count +0.1)) to improve variance homogeneity before carrying out an ANOVA using Genstat © 13 (Lawes Agricultural Trust (Rothamsted Experimental Station)). MNB egg parasitism, MNB and FSB damage in the field were expressed as percentages and an ANOVA carried out (Huwer *et al.*, 2006).

Crop damage data for MNB and FSB was expressed as percentages and analysed by carrying out an ANOVA using Genstat© 13.

6.2. Results

6.2.1. Field monitoring results

The nut production per raceme was monitored over the nut development period for each season. The analysis of variance indicated that the impact of treatment (P<0.001), variety (P<0.001), time (P<0.001), and the interactions of treatment x variety (P<0.001) and variety x time (P<0.001) on the number of nuts per raceme was significant in each season (Table 6.2.1.1). The Standard treatment had significantly more nuts per raceme than all other treatments and the varieties A4 and 741 had significantly more nuts per raceme in season 2006/2007. The variety 741 had significantly more nuts per raceme while variety A4 had the least nuts per raceme in 2007/2008. The variety 741 had the most nuts per raceme while variety 246 had the least in 2008/2009. The Wasp only treatment had the significantly lowest number of nuts per raceme and the variety 741 had the most while the variety 246 the least nuts per raceme in season 2009/2010. Overall (2006-2010) the IPM and Experimental treatments had significantly more nuts per raceme than the Wasp only and Standard treatment and the variety 741 overall had significantly more nuts per raceme than the other varieties tested.

The pattern of nut drop over time for the different treatments and different varieties throughout each season is illustrated in Figures 6.2.1.1 to 6.2.1.4.

Table 6.2.1.1: Average nuts per raceme by season by variety and treatment. Within a season different letters indicate means are significantly different

Year	Treatment	246	741	849	A4	Average
	IPM	3.5	4.5	3.6	5.0	**4.2 b**
2006/2007	Standard	3.7	5.0	4.3	5.1	**4.5 a**
	Wasp only	3.4	4.2	4.3	4.6	**4.1 b**
	Experimental	3.6	4.8	4.0	3.9	**4.1 b**
	Average	**3.5 c**	**4.6 a**	**4.0 b**	**4.7 a**	
Year						
	IPM	3.1	5.0	4.8	2.7	**3.9 a**
2007/2008	Standard	2.7	4.4	3.6	2.8	**3.4 b**
	Wasp only	3.0	5.0	3.9	3.2	**3.8 a**
	Experimental	3.5	5.3	3.8	2.9	**3.9 a**
	Average	**3.1 c**	**4.9 a**	**4.0 b**	**2.9 d**	
Year						
	IPM	3.4	4.8	3.9	3.8	**4.0ab**
2008/2009	Standard	3.2	3.8	3.5	3.5	**3.5 c**
	Wasp only	3.4	4.8	4.1	3.7	**4.0 a**
	Experimental	3.5	4.4	3.6	3.8	**3.8 b**
	Average	**3.4 c**	**4.5 a**	**3.8 b**	**3.7 b**	
Year						
	IPM	2.4	3.7	3.2	4.1	**3.4 a**
2009/2010	Standard	2.7	3.6	2.6	3.5	**3.1 b**
	Wasp only	1.6	3.4	2.1	3.2	**2.6 c**
	Experimental	2.3	4.3	3.1	3.3	**3.3 ab**
	Average	**2.3 d**	**3.7 a**	**2.7 c**	**3.5 b**	
Overall						
	IPM	3.1	4.5	3.9	4.0	**3.9 a**
2006 to 2010	Standard	3.1	4.2	3.5	3.8	**3.6 b**
	Wasp only	2.9	4.4	3.6	3.7	**3.6 b**
	Experimental	3.3	4.6	3.6	3.5	**3.8 a**
	Average	**3.1 b**	**4.4 a**	**3.7 b**	**3.7 b**	

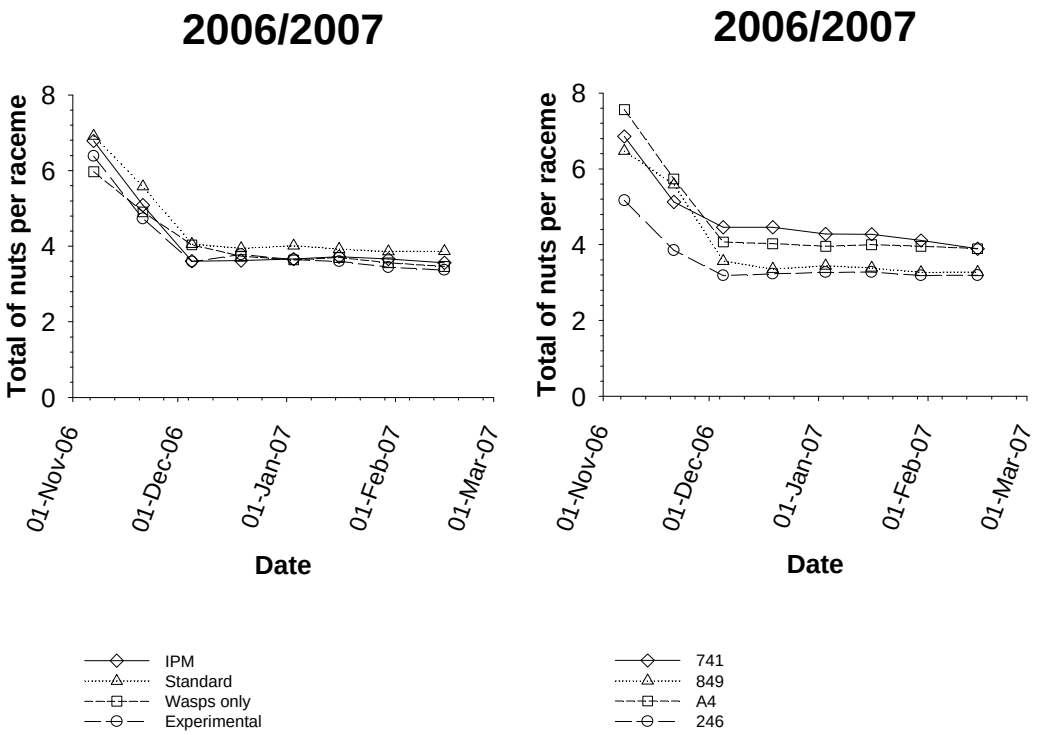

Figure 6.2.1.1: Nuts counted per raceme in different treatments (left) and different varieties (right) during 2006/2007 season

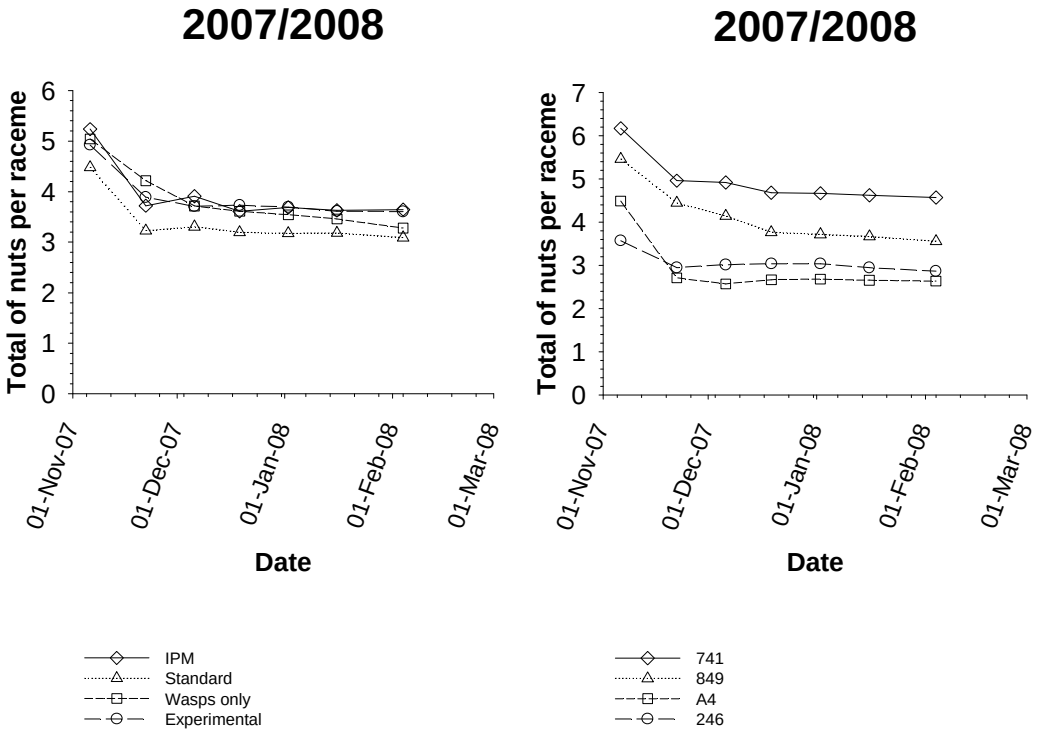

Figure 6.2.1.2: Nuts counted per raceme in different treatments (left) and different varieties (right) during 2007/2008 season

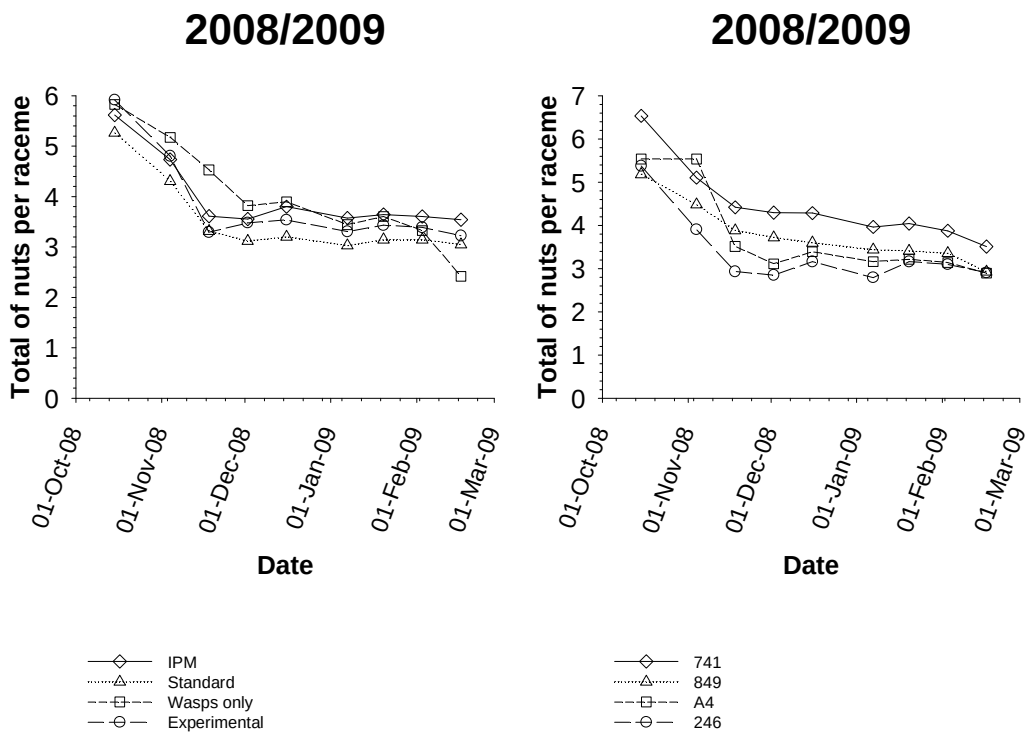

Figure 6.2.1.3: Nuts counted per raceme in different treatments (left) and different varieties (right) during 2008/2009 season

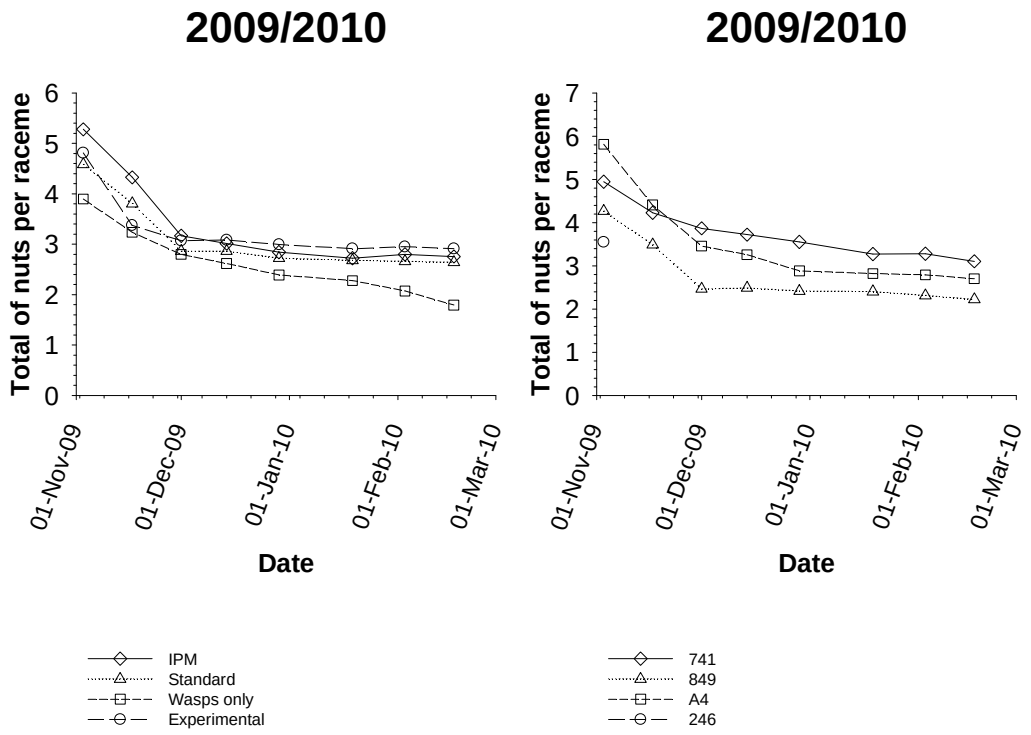

Figure 6.2.1.4: Nuts counted per raceme in different treatments (left) and different varieties (right) during 2009/2010 season

6.2.2. MNB monitoring results

MNB Pheromone trapping

Pheromone traps catches from the Entomology orchard at CTH and from the mangroves at Burns Point, West Ballina from 2006/2007 – 2009/2010 are shown in Figures 6.2.2.1 and 6.2.2.2. They illustrate high MNB activity in mangroves that precedes peaks in the orchard. The peaks between mid December and early January are the most important for timing of treatments for MNB management. Data records of this study confirm results from previous research (Huwer *et al.*, 2006).

MNB egg monitoring

Figures 6.2.2.1 and 6.2.2.2 show the number of live eggs recorded in different treatments in the CTH Entomology orchard over time. The wasp only treatment tended to have MNB oviposition above the treatment threshold. Oviposition varies over time during the season. Figures 6.2.2.3 and 6.2.2.6 show the total number of MNB eggs laid in the different treatments and varieties respectively over the duration of the study. Parasitism by the egg parasitoid *T. cryptophlebiae* usually increases with the increasing availability of viable MNB eggs over the season. Changes in the percentage of parasitism of MNB eggs recorded in different treatments and varieties respectively are shown in Figures 6.2.2.7 to 6.2.2.12. Only season 2008/09 showed oviposition approaching the threshold levels early enough to be a real problem to the crop with MNB generally a minimal issue until the commencement of nut maturity in February. Figures 6.2.2.1 to 6.2.2.12 illustrate the trend lines in different treatments and varieties over time, while Tables 6.2.2.1 to 6.2.2.5 give details on analysis of data.

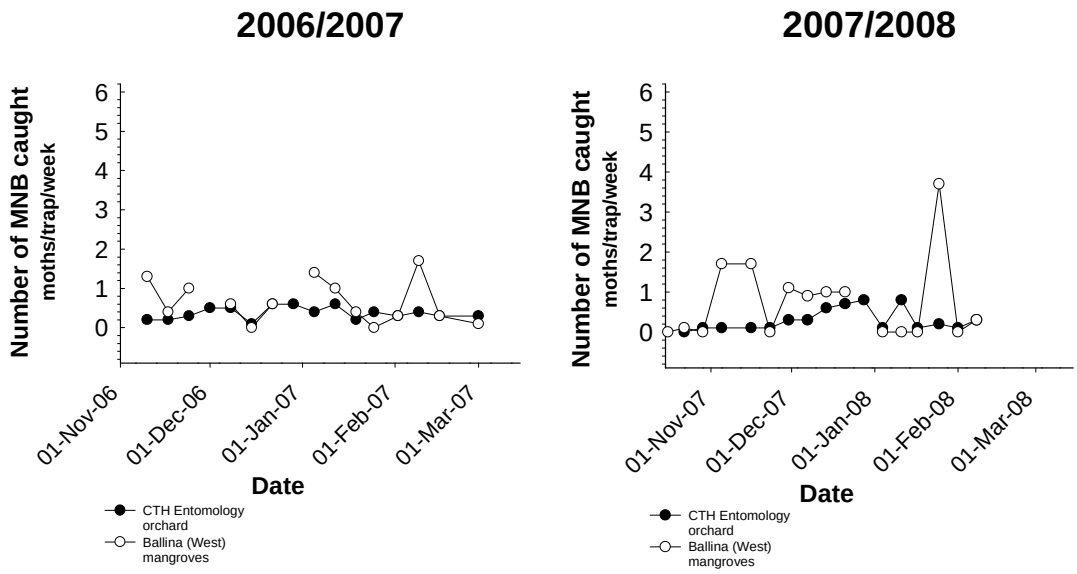

Figure 6.2.2.1: MNB pheromone trap catches 2006/2007 and 2007/2008

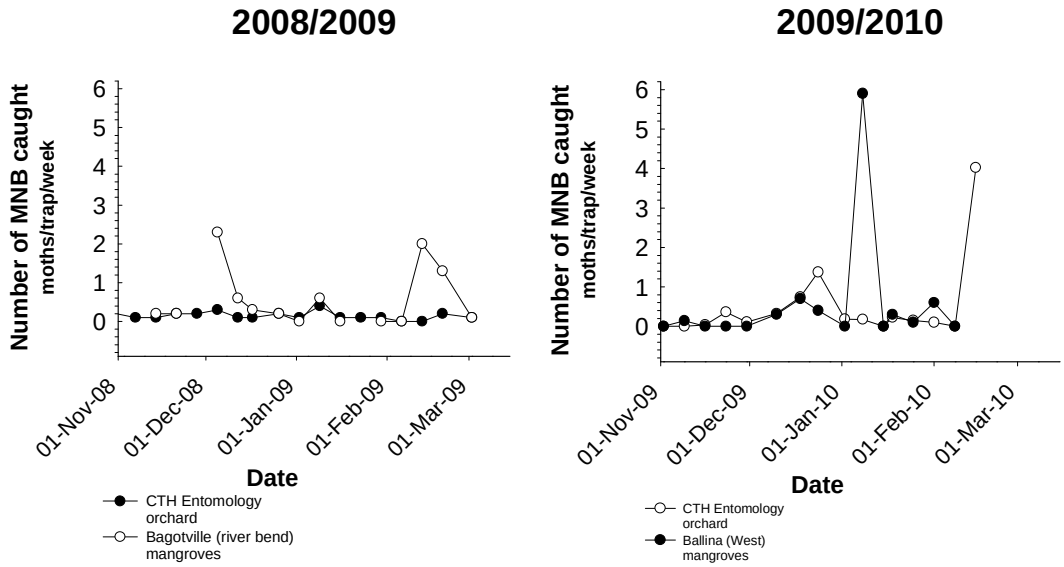

Figure 6.2.2.2: MNB pheromone trap catches 2008/2009 and 2009/2010

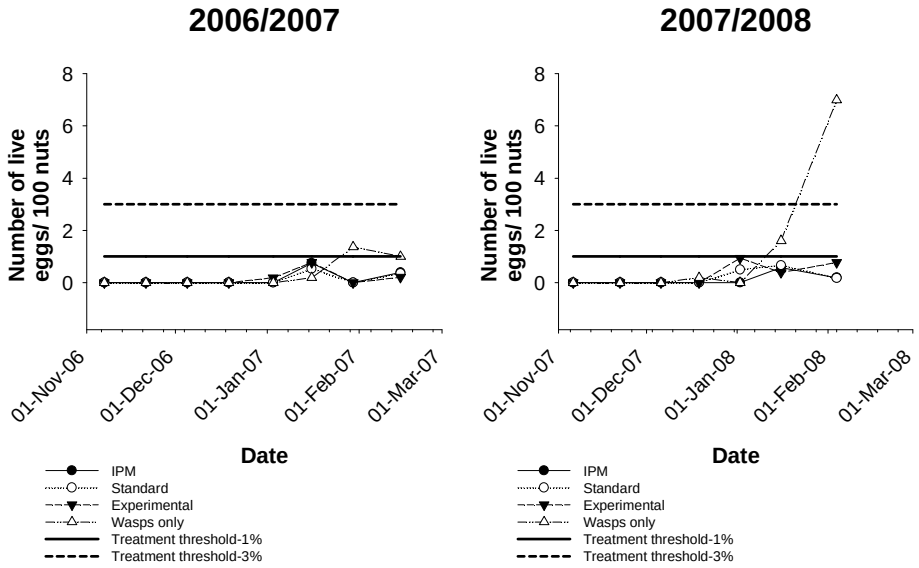

Figure 6.2.2.3: Number of live MNB eggs recorded in different treatments over time in seasons 2006/2007 and 2007/2008

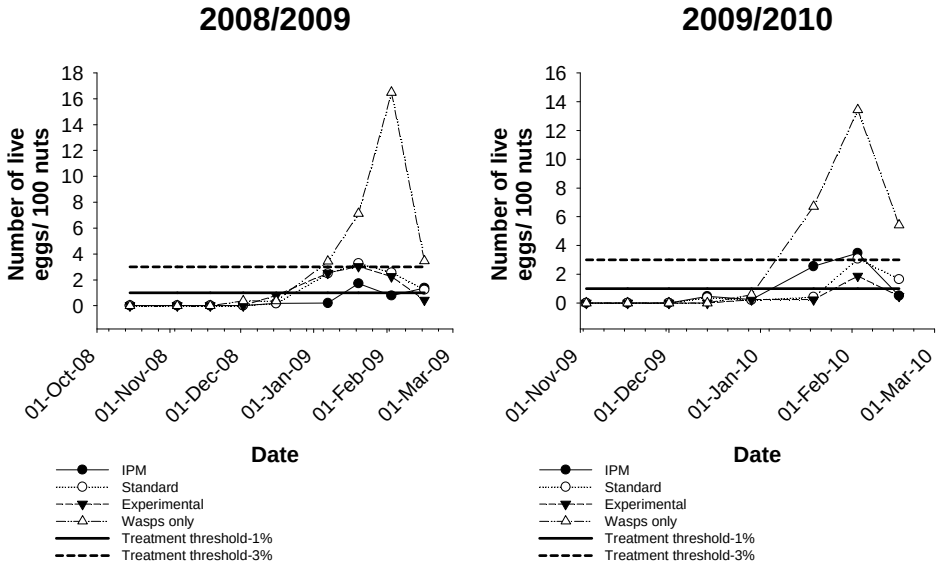

Figure 6.2.2.4: Number of live MNB eggs recorded in different treatments over time in seasons 2008/2009 and 2009/2010

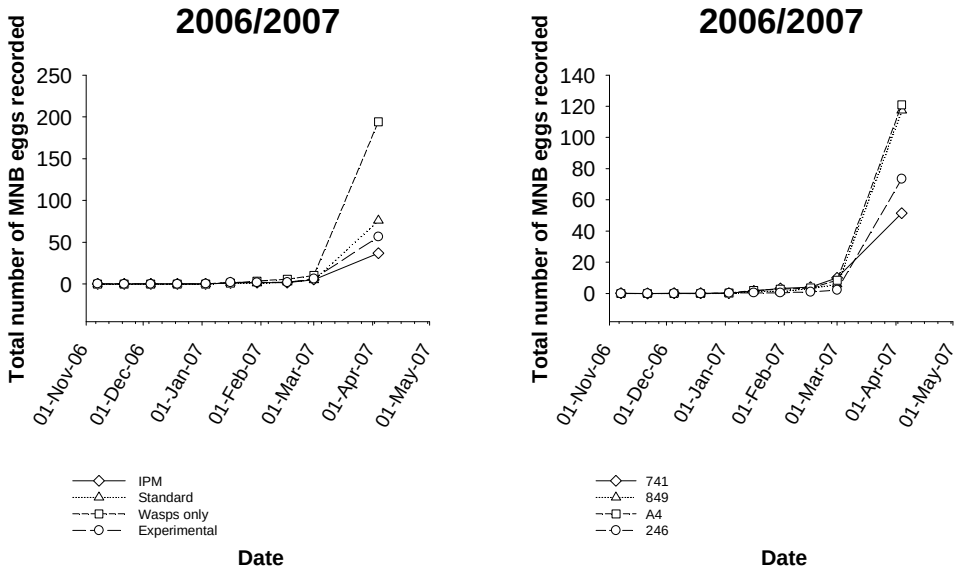

Figure 6.2.2.5: Total number of MNB eggs recorded over time in different treatments (left) and different varieties (right) in season 2006/2007

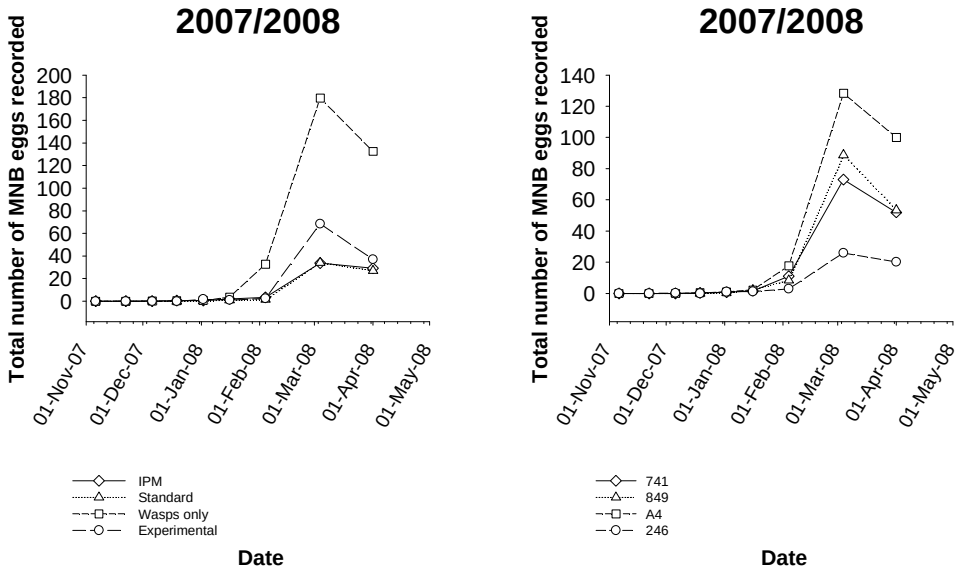

Figure 6.2.2.6: Total number of MNB eggs recorded over time in different treatments (left) and different varieties (right) in season 2007/2008

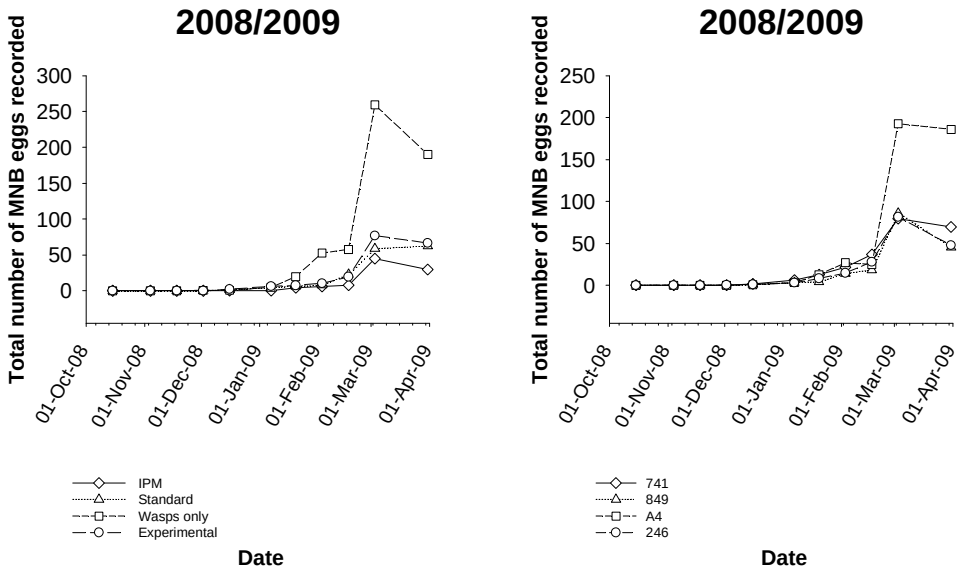

Figure 6.2.2.7: Total number of MNB eggs recorded over time in different treatments (left) and different varieties (right) in season 2008/2009

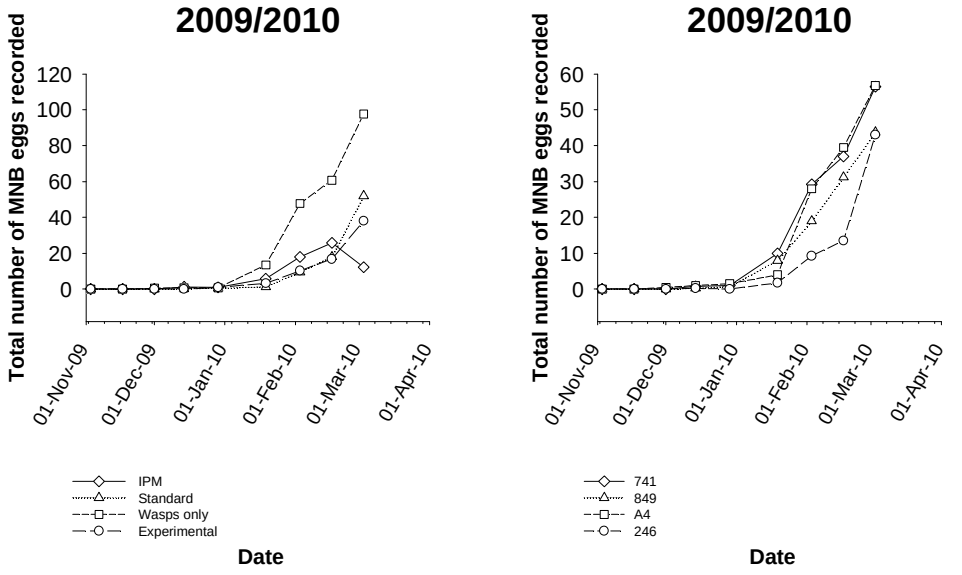

Figure 6.2.2.8: Total number of MNB eggs recorded over time in different treatments (left) and different varieties (right) in season 2009/2010

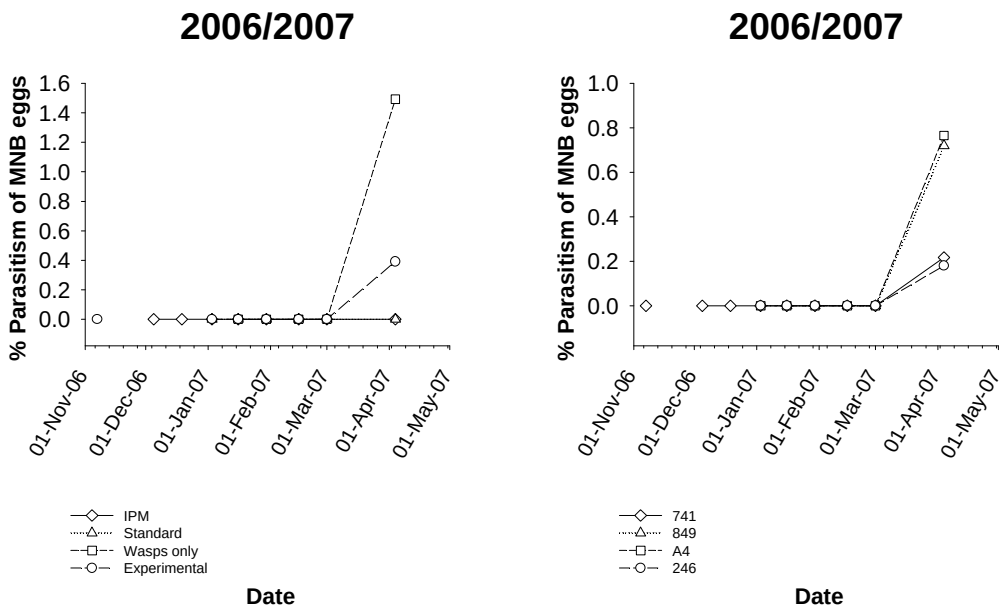

Figure 6.2.2.9: Percentage of MNB eggs parasitised over time in different treatments (left) and different varieties (right) in season 2006/2007

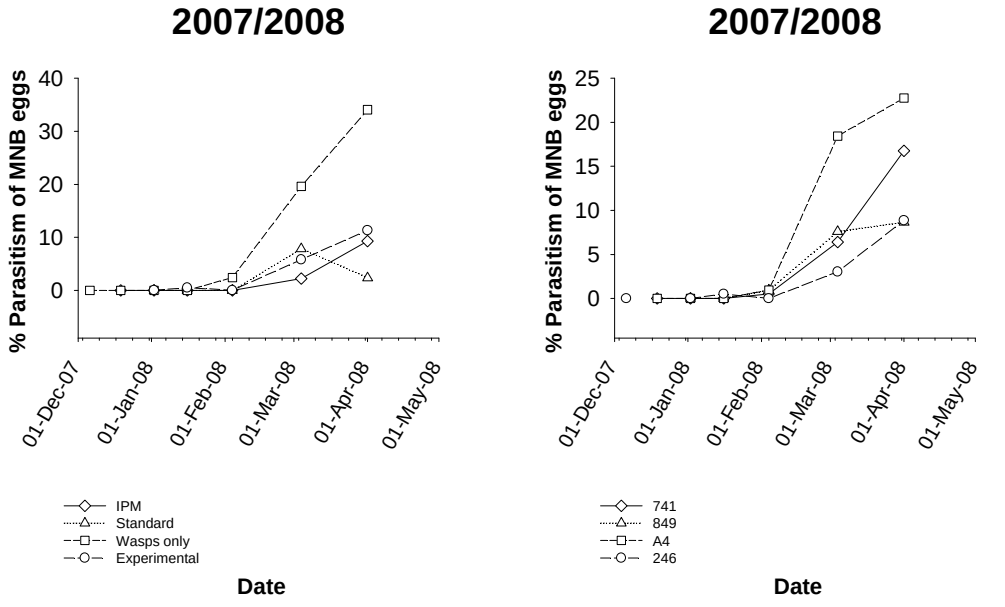

Figure 6.2.2.10: Percentage of MNB eggs parasitised over time in different treatments (left) and different varieties (right) in season 2007/2008

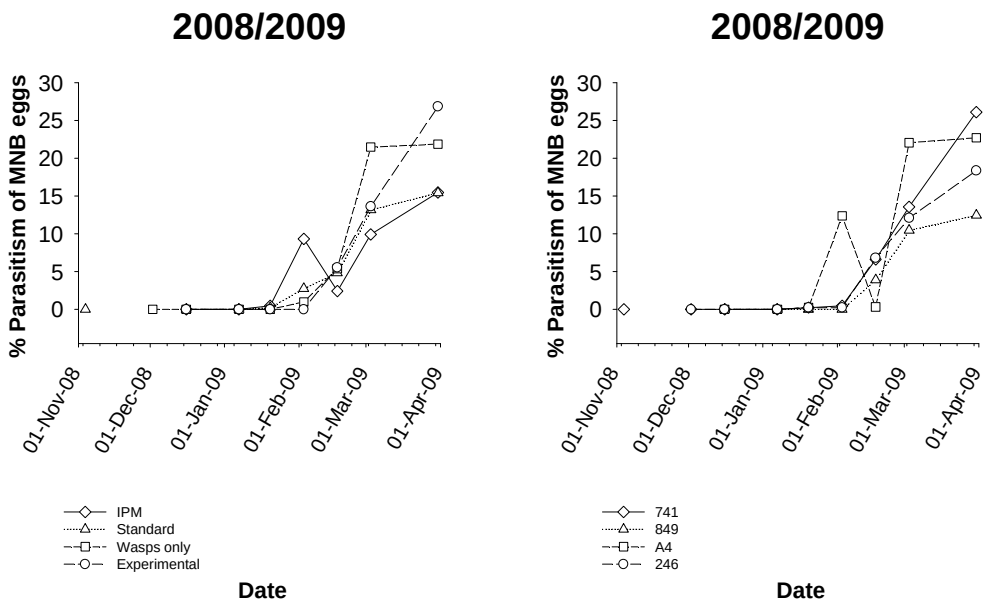

Figure 6.2.2.11: Percentage of MNB eggs parasitised over time in different treatments (left) and different varieties (right) in season 2008/2009

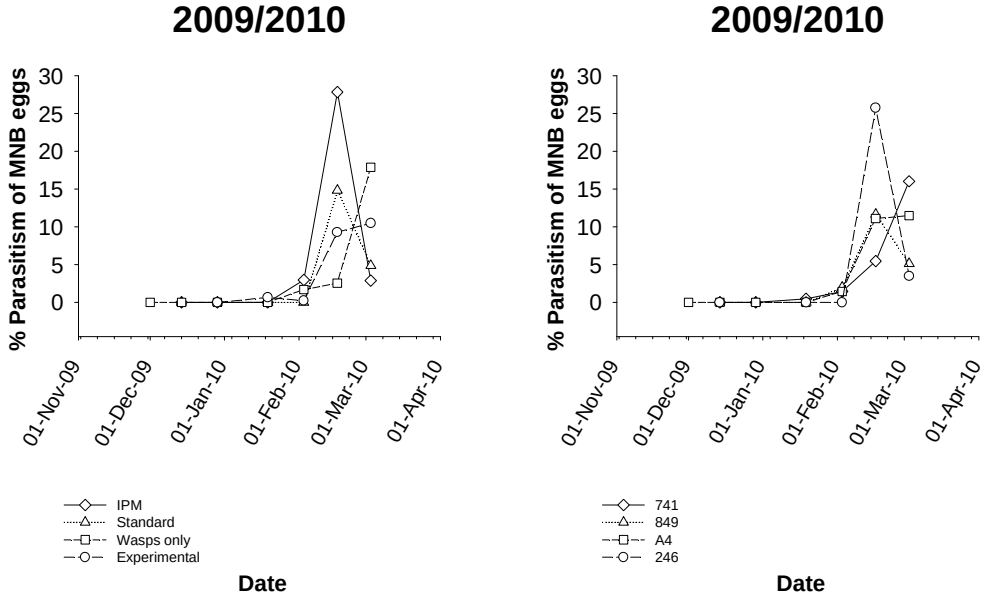

Figure 6.2.2.12: Percentage of MNB eggs parasitised over time in different treatments (left) and different varieties (right) in season 2009/2010

Table 6.2.2.1: MNB oviposition activity on macadamia nuts in 2006/2007 at CTH Alstonville. Averages followed by different letters are significantly different.

Year	Parameter	Treatment	246	741	849	A4	Average
2006/2007	MNB Eggs laid (average)	IPM	0.0	1.1	0.3	0.9	**0.6 a**
		Standard	0.9	1.1	0.1	0.1	**0.6 a**
		Wasp only	0.0	1.8	1.9	0.9	**1.1 a**
		Experimental	0.1	0.4	0.3	1.9	**0.7 a**
		Average	**0.3 a**	**1.1 b**	**0.7 ab**	**0.9 b**	
Year	**Parameter**						
2006/2007	% parasitism MNB Eggs (average)	IPM	0.0	0.0	0.0	0.0	**0.0 a**
		Standard	0.0	0.0	0.0	0.0	**0.0 a**
		Wasp only	0.0	0.0	0.0	0.0	**0.0 a**
		Experimental	0.0	0.0	0.0	0.0	**0.0 a**
		Average	**0.0 a**	**0.0 a**	**0.0 a**	**0.0 a**	**0.0 a**
Year	**Parameter**						
2006/2007	% nuts tunnelled (average)	IPM	0.10	0.11	0.08	0.18	**0.12 a**
		Standard	0.00	0.00	0.00	0.11	**0.03 a**
		Wasp only	0.00	0.07	0.41	0.56	**0.26 ab**
		Experimental	0.78	0.19	0.13	0.56	**0.41 b**
		Average	**0.22 a**	**0.09 a**	**0.15 a**	**0.35 a**	

MNB monitoring results 2006/2007

In 2006/2007 there was no significant treatment impact (*P*=0.267) on the total number of MNB eggs recorded on nuts on the tagged racemes. Variety however, did have a significant impact on number of eggs laid (*P*=0.057) (Table 6.2.2.1). Variety 246 had the lowest number of MNB eggs recorded while varieties A4 and 741 had the largest number of MNB eggs laid. During this season there was no parasitism of eggs recorded. Treatment had a significant impact on the percentage of nuts with tunnels (*P*=0.026) (Table 6.2.2.1). The Standard and IPM treatments had the lowest percentage of tunnelled nuts and the Experimental treatment the highest (Table 6.2.2.1).

MNB monitoring results 2007/2008

Treatment (*P*<0.001) and variety (*P*<0.001) both had a significant impact on MNB oviposition (Table 6.2.2.2). The Wasp only treatment had significantly more MNB eggs on nuts than the other treatments while a significantly highest number of MNB

eggs were recorded on variety A4 (Table 6.2.2.2). There was no significant difference in parasitism between different treatments (*P*=0.200) or variety (*P*=0.257) (Table 6.2.2.2). There was a significant effect of treatments (*P*<0.001) and varieties (*P*<0.001) on the percentage of tunnelled nuts recorded (Table 6.2.2.2). The number of tunnelled nuts was significantly higher in the Wasp only treatment and significantly higher in the A4 variety, which corresponds with the oviposition results (Table 6.2.2.2).

Table 6.2.2.2: MNB oviposition activity on macadamia nuts in 2007/2008 at CTH Alstonville. Averages followed by different letters are significantly different.

Year	Parameter	Treatment	246	741	849	A4	Average
2007/2008	MNB Eggs laid (average)	IPM	0.0	0.6	0.8	1.6	**0.7 a**
		Standard	0.0	0.4	0.4	0.3	**0.3 a**
		Wasp only	1.4	4.8	3.6	7.0	**4.2 b**
		Experimental	1.0	0.4	0.2	0.7	**0.6 a**
		Average	**0.6 a**	**1.6 b**	**1.3 ab**	**2.4 c**	
Year	**Parameter**						
2007/2008	% parasitism MNB Eggs (average)	IPM	0.0	0.0	0.0	0.0	**0.0 a**
		Standard	0.0	0.0	0.0	0.0	**0.0 a**
		Wasp only	0.0	3.7	4.2	3.5	**2.8 a**
		Experimental	6.3	0.0	0.0	0.0	**1.6 a**
		Average	**1.6 a**	**0.9 a**	**1.0 a**	**0.9 a**	
Year	**Parameter**						
2007/2008	% nuts tunnelled (average)	IPM	0.0	0.0	0.0	0.8	**0.2 a**
		Standard	0.0	0.1	0.0	0.2	**0.1 a**
		Wasp only	0.5	1.7	1.8	3.3	**1.8 b**
		Experimental	0.3	0.0	0.2	0.5	**0.2 a**
		Average	**0.2a**	**0.5 a**	**0.5 a**	**1.2 b**	

MNB monitoring results 2008/2009

Both treatments (*P*<0.001) and variety (*P*=0.011) had a significant impact on MNB oviposition (Table 6.2.2.3). The IPM treatment had a significantly lower number of MNB eggs than the Wasp only treatment while the variety 849 had significantly less MNB eggs than the varieties A4 and 741 (Table 6.2.2.3).Neither treatments (*P*=0.514) nor variety (*P*=0.275) had a significant effect on parasitism by the egg parasitoid *T. cryptophlebiae* (Table 6.2.2.3).Treatment (*P*<0.001) and variety (*P*<0.001) however did have a significant impact on the percentage of tunnelled nuts

(Table 6.2.2.3). The Wasp only treatment had a significantly higher percentage of tunnelled nuts than the other treatments and variety A4 also had a significantly higher percentage of tunnelled nuts (Table 6.2.2.3).

Table 6.2.2.3: MNB oviposition activity on macadamia nuts in 2008/2009 at CTH Alstonville. Averages followed by different letters are significantly different.

Year	Parameter	Treatment	246	741	849	A4	Average
		IPM	2.0	2.9	1.7	1.8	**2.1 a**
	MNB	Standard	3.2	8.8	2.7	4.8	**4.9 b**
2008/2009	Eggs laid	Wasp only	13.6	19.7	7.2	20.2	**15.2 c**
	(average)	Experimental	5.9	4.1	6.8	3.7	**5.1 b**
		Average	**6.2 ab**	**8.9 b**	**4.6 a**	**7.6 b**	
Year	**Parameter**						
		IPM	2.9	3.1	0.0	23.3	**7.3 a**
	%parasitism	Standard	6.3	6.7	1.5	4.2	**4.6 a**
2008/2009	MNB Eggs	Wasp only	5.1	2.3	4.2	0.9	**3.1 a**
	(average)	Experimental	3.3	20.8	0.0	0.0	**6.0 a**
		Average	**4.4 a**	**8.2 a**	**1.4 a**	**7.1 a**	
Year	**Parameter**						
		IPM	0.0	0.0	0.1	0.1	**0.0 a**
	% nuts	Standard	0.0	0.7	0.1	0.3	**0.3 a**
2008/2009	tunnelled	Wasp only	5.3	4.6	3.2	11.5	**6.1 b**
	(average)	Experimental	0.7	0.1	0.4	0.6	**0.5 a**
		Average	**1.5 a**	**1.4 a**	**0.9 a**	**3.1 b**	

MNB monitoring results 2009/2010

The preference for MNB oviposition was significantly affected by treatment ($P<$ 0.001) and variety ($P=0.071$). The Wasp only treatment had a significantly higher number of MNB eggs than the other treatments (Table 6.2.2.4). The variety 246 had the significantly lowest number of MNB eggs (Table 6.2.2.4).

During the season 2009/2010 treatments ($P=0.023$) had a significant impact on parasitism of MNB eggs but not variety ($P=0.961$) while the IPM treatment had significantly higher parasitism than the Wasp only treatment (Table 6.2.2.4).

The percentage of tunnelled nuts was significantly affected by treatment ($P<0.001$) but not by variety ($P=0.511$) (Table 6.2.2.4). The Wasp only treatment had a significantly higher percentage of tunnelled nuts than all other treatments (Table 6.2.2.4).

Table 6.2.2.4: MNB oviposition activity on macadamia nuts in 2009/2010 at CTH Alstonville. Averages followed by different letters are significantly different.

Year	Parameter	Treatment	246	741	849	A4	Average
		IPM	0.2	8.3	5.8	8.7	**5.8 a**
2009/10	MNB	Standard	0.9	4.8	2.6	4.9	**3.3 a**
	Eggs laid	Wasp only	9.2	17.8	15.3	12.8	**13.8 b**
	(average)	Experimental	0.7	3.8	2.7	6.8	**3.5 a**
		Average	**2.8 a**	**8.7 b**	**6.6 ab**	**8.3 b**	
Year	**Parameter**						
		IPM	0.0	16.0	18.5	20.9	**13.9 a**
2009/10	%parasitism	Standard	16.7	0.0	11.1	0.0	**6.9 ab**
	MNB Eggs	Wasp only	0.6	3.2	0.7	0.5	**1.3 b**
	(average)	Experimental	12.5	12.2	0.0	1.3	**6.5 ab**
		Average	**7.4 a**	**7.8 a**	**7.6 a**	**5.7 a**	
Year	**Parameter**						
		IPM	0.0	0.7	0.9	0.6	**0.6 a**
2009/10	% nuts	Standard	0.0	0.3	0.0	0.0	**0.1 a**
	tunnelled	Wasp only	4.4	6.3	7.1	5.0	**5.7 b**
	(average)	Experimental	0.0	0.0	0.3	0.8	**0.3 a**
		Average	**1.1 a**	**1.8 a**	**2.1 a**	**1.6 a**	

***MNB monitoring summary results* 2007-2010**

Overall, the impact of treatment (P<0.001) and variety (P=0.003) on MNB oviposition preference was significant (Table 6.2.2.5) and nuts from the Wasp only treatment had significantly more MNB eggs than nuts in all other treatments (Table 6.2.2.5). Nuts in the variety 246 had the lowest number of MNB eggs (Table 6.2.2.5). Overall treatment (P= 0.006) had a significant impact on MNB egg parasitism, but not variety (P=0.445) (Table 6.2.2.5). The IPM treatment had a significantly higher percentage of parasitism than the Standard and Wasp only treatments. (Table 6.2.2.5).Treatment (P<0.001) and variety (P=0.012) overall had a significant effect on the percentage of tunnelled nuts (Table 6.2.2.5). The Wasp only treatment had a significantly higher percentage of tunnelled nuts than the other treatments and the variety A4 had a significantly higher percentage of tunnelled nuts than the other varieties (Table 6.2.2.5).

Table 6.2.2.5: MNB oviposition activity on macadamia nuts from 2007to 2010 at CTH Alstonville. Averages followed by different letters are significantly different.

Year	Parameter	Treatment	246	741	849	A4	Average
		IPM	0.6	3.2	2.1	3.2	**2.3 a**
2007-	MNB	Standard	1.3	3.8	1.4	2.5	**2.3 a**
2010	Eggs laid	Wasp only	6.1	11.0	7.0	10.2	**8.6 b**
	(average)	Experimental	1.9	2.2	2.5	3.3	**2.5 a**
		Average	**2.4 a**	**5.0 c**	**3.3 ab**	**4.8 bc**	
Year	**Parameter**						
		IPM	2.0	7.0	7.4	13.9	**7.6 a**
2007-	% parasitism	Standard	7.1	2.5	4.7	1.9	**4.1 b**
2010	MNB Eggs	Wasp only	2.4	2.6	2.4	1.9	**2.3 b**
	(average)	Experimental	5.1	12.6	0.0	0.4	**4.5 ab**
		Average	**4.2 a**	**6.2 a**	**3.6 a**	**4.5 a**	
Year	**Parameter**						
		IPM	0.1	0.2	0.2	0.4	**0.2 a**
2007-	% nuts	Standard	0.0	0.3	0.0	0.1	**0.1 a**
2010	tunnelled	Wasp only	2.6	3.2	3.1	5.1	**3.5 b**
	(average)	Experimental	0.5	0.1	0.2	0.6	**0.3 a**
		Average	**0.8 a**	**0.9 a**	**0.9 a**	**1.6 b**	

Population forecasting for MNB has been done at CTH using the heat sum models described in Campbell et al 1999, Maddox et al 2002. These can predict the moth emergence within a block if flight arrival is known. The only seasons when the heat sum for the emergence of generation 2 is not within 2 weeks of the observed rise in oviposition rate are 2005/06 and 2008/09. Both of these seasons had significant flights detected in the southern Riverland district below the Alstonville plateau (Coraki & Bagotville), rather than the usual flights in from Ballina (Table 6.2.2.6, Figure 6.2.2.2). These were also the heavier pressure season which suggests monitoring here maybe more indicative of larger infestations

Table 6.2.2.6: Summary of climatic data (1 August - 31 March) from CTH Alstonville, 2003-2010. Incidence patterns of *Cryptophlebia ombrodelta* determined by moth catches in pheromone traps (Biofix date for each season) and the generation times calculated on degree days (408DD summed for hours above 15℃).

Climatic Data	**Aug 03 – Mar 04**	**Aug 04 – Mar 05**	**Aug 05 – Mar 06**	**Aug 06 – Mar 07**	**Aug 07 – Mar 08**	**Aug 08 – Mar 09**	**Aug 09 – Mar 10**	**30 year mean**
Average daily maximum (℃)	25.5	25.2	25.5	24.6	24.5	24.8	26.3	**24.8**
Average daily minimum (℃)	17.0	16.5	17.5	16.4	16.7	16.3	17.0	**16.3**
Ground minimum (℃)	-0.2	-0.5	0.2	3.1	-1.0	-1.5	1.2	
Biofix (Moth catch) (date)	18/9/03	1/9/04	1/9/05	28/8/06	17/8/07	29/9/08	22/8/09	
Heat sum G1 (date)	10/12/03	16/11/04	24/11/05	14/11/06	30/10/07	1/12/08	31/10/09	
Heat sum G2 (date)	**24/1/04**	**15/1/05**	**29/1/06***	**17/1/07**	**1/1/08**	**23/1/09***	**24/12/09**	
Heat sum G3 (date)	9/3/04	5/3/05	21/3/06	8/3/07	23/2/08	15/3/09	8/2/10	
G4 in season	no	no	no	no	yes	no	yes	
Threshold point for MNB oviposition	**3/2/04**	**4/2/05**	**6/1/06***	**18/1/07**	**5/1/08**	**20/12/08**	**6/1/10**	
Sun hours (Total hours)	1927.5	1971.1	1845.5	1918.3	1560.3	1760.3	1936.5	**1829.4**
Pan Evaporation (Total mm)	1233.3	1212.4	1177.2	1147.1	1081.4	1138.8	1228.8	**1200.7**
Rainfall (Total mm)	820.8	863.6	1319.8	884.8	1676.7	1212.9	987.0	**1169.5**
Days >0mm	94	88	117	108	144	121	104	**104**
Days>0.5mm	71	74	90	77	117	99	75	
Days > 1.0mm	63	71	79	64	111	82	69	
Wet days during Aug/Sep	9	12	20	29	24	17	12	**17**
Sun hours during Aug/Sep	526.3	533.2	449.1	473.6	437.7	478.1	552.5	**481.6**

* only seasons where heat sum for the emergence of generation 2 in the season was not within 2 weeks of the observed rise in oviposition rate

6.2.3. FSB monitoring results

Nut drop and damage to green nuts based on quadrat sampling under trees

The Standard treatments showed lower FSB sightings but no significant difference in the detected damage to green nut (Table 6.2.3.1, 6.2.3.2). During the monitoring of green nuts in quadrats in 2006/2007 there was neither a treatment (*P*=0.292) or a variety (*P*=0.710) effect on the percentage of FSB damaged green nuts (Table 6.2.3.1) on the ground. During the season 2007/2008 treatment (*P*=0.002) and variety (*P*<0.001) had a significant impact on the percentage of FSB damage on green nuts found on the ground. The FSB damage in the green nuts in the FSB IPM treatment was significantly higher than in all other treatments (Table 6.2.3.1). The green nuts on the ground in variety A4 had the lowest percentage of FSB damage (Table 6.2.3.1).

During season 2008/2009 treatment (*P*<0.001) and variety (*P*<0.001) effects were highly significant with regards to percentage of FSB damage on green nuts on the ground (Table 6.2.3.1).The green nuts on the ground in highest (Table 6.2.3.1). During the 2009/2010 season variety (*P*=0.089) but not treatment (*P*=0.785) had a significant effect on FSB damage in nuts on the ground. The variety A4 had the lowest percentage of FSB damage in green nuts on the ground (Table 6.2.3.1). Overall, variety (*P*=0.034) but not treatment (*P*=0.177) significantly affected the percentage of FSB damaged nuts on the ground (Table 6.2.3.1). Overall the lowest percentage of FSB damage on green nuts on the ground was in variety A4.

The sightings of FSB on tree were generally poorly correlated with the damage expressed in the final crop (Table 6.2.3.3). In season 2006/2007 the presence of FSB was significantly affected by treatment (*P*=0.001) and variety (Table 6.2.3.2). Numbers of FSB observed in the IPM treatment was significantly higher than in all other treatments (Table 6.2.3.2). The lowest number of FSB was recorded in variety 741 and the highest number in variety A4 (Table 6.2.3.2). Treatment (*P*<0.001) and variety (*P*=0.095) had a significant effect on presence of FSB in the 2007/2008 season (Table 6.2.3.2). Numbers of FSB recorded in the Experimental and Standard treatments were significantly fewer than in the Wasp only and IPM treatments (Table 6.2.3.2). The variety 246 had the lowest number of FSB recorded (Table 6.2.3.2). Treatment (*P*<0.001), but not variety (*P*=0.191) had a significant impact on the presence of FSB (Table 6.2.3.2). The number of FSB sighted was significantly higher in the Wasp only treatment than in all other treatments (Table 6.2.3.2). During the season 2009/2010 neither treatment (*P*=0.122) nor variety (*P*=0.951) had a significant impact on presence of FSB (Table 6.2.3.2). Overall, treatment (*P*<0.001) and variety (*P*=0.069) had a significant effect on FSB presence (Table 6.2.3.2). Overall, the Standard and Experimental treatments had significantly less FSB numbers than the IPM and Wasp only treatments (Table 6.2.3.2). Overall, the variety 246 had the lowest number of FSB recorded (Table 6.2.3.2).

Early season FSB damage results in nut drop that can be easily monitored, while mid and late season damaged nuts remain on the tree. Generally the early season monitoring is no indication of how extensive the final FSB damage will appear on the tree (Table 6.2.3.3). The lack of an adequate monitoring tool later in the season is the major short coming of the integrated pest management and wasp only treatments. Monitoring of pest infestation is an important tool of any IPM strategy, as treatments need to be targeted. To evaluate monitoring strategies tested it is important to relate them to the kernel damage attributed to FSB at harvest. Monitoring results were generally very poorly correlated with the FSB damage of kernel. The best result was

a 33% correlation of FSB sightings in trees with the FSB damage in Kernel in the 2007/2008 season (Table 6.2.3.3)

Table 6.2.3.1: Percentage FSB damaged nuts on ground, in green nut sampled at CTH Alstonville between 2006 and 2010. Averages with different letters indicate means are significantly different.

Year	Treatment	246	741	849	A4	Average
2006/ 2007	IPM	29.8	22.7	19.6	21.9	**23.5 a**
	Standard	24.8	29.6	17.3	20.7	**23.1 a**
	Wasp only	24.3	18.6	26.2	32.4	**25.4 a**
	Experimental	20.1	21.2	23.1	17.9	**20.6 a**
	Average	**24.7 a**	**23.0 a**	**21.5 a**	**23.2 a**	
Year						
2007/ 2008	IPM	76.6	64.0	77.6	62.2	**70.1 b**
	Standard	75.2	69.7	58.5	48.6	**63.0 a**
	Wasp only	63.1	57.2	60.8	57.6	**59.7 a**
	Experimental	62.7	56.9	68.7	48.3	**59.2 a**
	Average	**69.4 c**	**61.9 b**	**66.4 bc**	**54.2 a**	
Year						
2008/ 2009	IPM	62.5	51.8	51.0	57.3	**55.7 c**
	Standard	49.0	46.1	35.9	54.6	**46.4 a**
	Wasp only	56.6	51.3	49.5	52.0	**52.3 bc**
	Experimental	53.2	51.0	46.3	49.5	**50.0 ab**
	Average	**55.3 c**	**50.0 b**	**45.7 a**	**53.3 bc**	
Year						
2009/ 2010	IPM	64.3	59.4	65.5	47.3	**59.1 a**
	Standard	67.5	54.6	60.3	54.1	**59.1 a**
	Wasp only	65.6	51.7	64.1	70.9	**63.1 a**
	Experimental	60.7	66.8	62.8	46.2	**59.1 a**
	Average	**64.5 b**	**58.1 ab**	**63.2 ab**	**54.6 a**	
Overall						
2006- 2010	IPM	58.3	49.8	52.7	47.2	**52.0 a**
	Standard	54.1	50.0	43.0	44.5	**47.9 a**
	Wasp only	52.5	44.7	50.2	53.2	**50.1 a**
	Experimental	48.9	49.0	49.8	40.5	**47.0 a**
	Average	**53.5 b**	**48.4 a**	**48.9 ab**	**46.3 a**	

Table 6.2.3.2: Average number of FSB nymphs and adults seen from ground during sampling at CTH Alstonville between 2006 and 2010. Averages with different letters indicate means are significantly different.

Year	Treatment	246	741	849	A4	Average
2006/ 2007	IPM	0.4	0.4	0.9	1.4	**0.8 b**
	Standard	0.1	0.0	0.1	0.2	**0.1 a**
	Wasp only	0.0	0.1	0.0	0.7	**0.2 a**
	Experimental	0.0	0.0	0.0	0.0	**0.0 a**
	Average	**0.1 a**	**0.1 a**	**0.3ab**	**0.6 b**	
Year						
2007/ 2008	IPM	1.0	3.9	0.4	1.2	**1.6 b**
	Standard	0.0	0.3	0.1	1.0	**0.4 a**
	Wasp only	0.3	0.4	2.8	2.0	**1.4 b**
	Experimental	0.3	0.1	0.0	0.1	**0.1 a**
	Average	**0.4 a**	**1.2 b**	**0.8 ab**	**1.1 b**	
Year						
2008/ 2009	IPM	0.4	0.3	1.1	0.2	**0.5 a**
	Standard	0.3	0.4	0.1	0.2	**0.3 a**
	Wasp only	1.8	1.3	1.9	3.0	**2.0 b**
	Experimental	0.5	0.0	0.8	1.0	**0.6 a**
	Average	**0.8 a**	**0.5 a**	**1.0 a**	**1.1 a**	
Year						
2009/ 2010	IPM	0.2	0.3	0.1	0.8	**0.4 a**
	Standard	0.1	0.1	0.0	0.1	**0.1 a**
	Wasp only	0.5	0.5	0.4	0.1	**0.4 a**
	Experimental	0.0	0.2	5.0	0.1	**1.3 a**
	Average	**0.2 a**	**0.3 a**	**1.4 a**	**0.3 a**	
Overall						
2007- 2010	IPM	0.5	1.2	0.7	0.9	**0.8 b**
	Standard	0.1	0.2	0.1	0.4	**0.2 a**
	Wasp only	0.7	0.6	1.3	1.4	**1.0 b**
	Experimental	0.2	0.1	0.3	0.3	**0.2 a**
	Average	**0.4 a**	**0.5 ab**	**0.6 ab**	**0.8 b**	

Table 6.2.3.3: Correlation coefficient (R^2) between the final kernel damage due to FSB as a (%) and the early season monitoring of damaged green nutlets falling and the number of FSB seen from ground during sampling at CTH Alstonville between 2006 to 2010

Year	%of green nut drop with FSB Kernel damage at harvest	FSB Sightings in tree Kernel damage at harvest
2006/2007	0.11	0.15
2007/2008	0.001	0.34
2008/2009	0.01	0.07
2009/2010	0.10	0.05
2006-2010	0.28	0.04

6.2.3. Kernel damage results

Results from season 2006/2007

The main aim in 2006/2007 was to test value of flower spray and the comparison between early season thiamethoxam and endosulfan. Details of IPM, Standard, Wasps only and Experimental treatments are listed in Table 6.1.2 (page 28).

Results from the 2007 harvest for FSB damage in kernel indicate treatment (P<0.001) and variety (P<0.001) had a significant impact on FSB damage. Wasp only and Experimental treatments had significant less damage than Standard and IPM treatments. The varieties 246 and 741 had significantly less damage than the varieties 849 and A4 (Table 6.2.3.1). The latter is consistent with results from previous work (Huwer *et al.*, 2006) that mainly concentrated on MNB management in the first instance, but now has shifted the focus to FSB management.

Table 6.2.3.1: FSB damage in macadamia kernel (in %) in 2007

	246	741	849	A4	Average
IPM	4.5	7.4	22.3	18.4	**13.2 c**
Standard	2.6	6.1	6.8	7.0	**5.6 b**
Wasps only	1.6	0.9	2.9	10.2	**3.9 a**
Experimental	5.4	2.0	6.1	2.5	**4.0 a**
Average	**3.7 a**	**4.1 a**	**9.5 b**	**9.5 b**	

* Different letters indicate means are significantly different

Results from the 2007 harvest for MNB damage in kernel indicate treatment had no significant impact (P=0.623). Variety however did have a significant effect on MNB damage in kernel (P=0.003). The variety 741 had the significantly lower damage than the other varieties tested (Table 6.2.3.2).

The percentage of A-grade kernel in 2007 was significantly affected by treatment (*P*<0.001) and variety (*P*<0.001). The IPM treatment had significantly less A-grade kernel than all other treatments and the variety A4 had significantly highest and the variety 246 the significantly lowest percentage of A-grade kernel (Table 6.2.3.3).

Table 6.2.3.2: MNB damage in macadamia kernel (in %) in 2007

	246	741	849	A4	Average
IPM	1.0	1.0	0.9	0.9	**1.0 a**
Standard	2.8	0.2	1.0	1.3	**1.3 a**
Wasps only	1.1	0.2	2.5	2.0	**1.5 a**
Experimental	2.1	0.4	0.8	1.6	**1.2 a**
Average	**1.8 b**	**0.5 a**	**1.3 b**	**1.4 b**	

* Different letters indicate means are significantly different

Table 6.2.3.3: Percentage of A-grade kernel (in %) in 2007

	246	741	849	A4	Average
IPM	25.7	33.2	28.1	33.7	**30.2 b**
Standard	28.8	32.5	35.0	40.1	**34.1 a**
Wasps only	29.3	36.1	36.7	38.4	**35.1 a**
Experimental	28.9	35.1	33.9	42.6	**35.1 a**
Average	**28.2 c**	**34.2 b**	**33.4 b**	**38.7 a**	

* Different letters indicate means are significantly different

Table 6.2.3.4: Average nut yield (DNIS@ 10% moisture content) per tree in CTH trial across treatments and varieties in 2007

	246	741	849	A4	Average
IPM	17.1	23.0	17.3	10.9	**17.1 b**
Standard	23.0	22.8	19.2	14.4	**19.8 a**
Wasps only	19.0	22.7	14.8	15.7	**18.1 b**
Experimental	17.8	27.0	19.5	17.4	**20.4 a**
Average	**19.2 b**	**23.9 a**	**17.7 b**	**14.6 c**	

* Different letters indicate means are significantly different

Treatment (*P*<0.001) and variety (*P*< 0.001) significantly impacted on yield in 2007 (Table 6.2.3.4). Yield in the Experimental and Standard treatments had significantly higher yield than IPM and Wasp only treatments. The variety 741 had the significantly highest and the variety A4 the significantly lowest yield.

The results for 2007 suggested not a great benefit from the extra flower spray (endosulfan in the Experimental treatment) for 2 of the varieties (A4 and 849). There was an 18% increase in the crop on variety 741 and 20% on variety A4 and kernel quality was higher in both of those varieties (Table 6.2.3.4). MNB control was not

influenced by any of the treatments. The FSB damage however is strongly influenced by variety and in this case activity is also highest in the IPM treatment and in the thinner shelled varieties (Table 6.2.3.1). This expression of damage is most likely a result of the reliance on tebufenozide for management in January when all other blocks received beta-cyfluthrin. The thiamethoxam treatment also had significantly lower nut yields overall.

Results from season 2007/2008

The main aim in 2007/2008 was to compare early season thiacloprid and endosulfan application, and evaluate if MKG (macadamia kernel grub) was managed by mulching. Details of IPM, Standard, Wasps only and Experimental treatments are listed in Table 6.1.3 (see page 28).

The MNB damage in kernel was generally very low in 2008. Neither treatments nor (*P*=0.267) had a significant impact on MNB damage in kernel (Table 6.2.3.6.).

Table 6.2.3.5: FSB damage in macadamia kernel (in %) in 2008

	246	**741**	**849**	**A4**	**Average**
IPM	11.3	14.2	22.5	17.8	**16.4 b**
Standard	4.7	3.8	7.0	11.1	**6.7 a**
Wasps only	9.6	6.6	38.6	36.7	**22.9 c**
Experimental	2.6	1.3	12.4	8.5	**6.2 a**
Average	**7.0 a**	**6.5 a**	**20.2 c**	**18.5 b**	

Different letters indicate means are significantly different

In the 2008 harvest treatment (*P*<0.001) and variety (*P*<0.001) again had significant impact on FSB damage in kernel. The Experimental and Standard treatments had significant less damage than IPM and Wasp only treatments. The varieties 741 and 246 again had significantly less damage than the varieties A4 and 849 (Table 6.2.3.5).

Table 6.2.3.6: MNB damage in macadamia kernel (in %) in 2008

	246	**741**	**849**	**A4**	**Average**
IPM	0.2	0.3	0.1	0.1	**0.2 a**
Standard	0.7	0.3	0.1	0.3	**0.4 a**
Wasps only	0.8	0.1	0.2	0.5	**0.4 a**
Experimental	0.8	0.3	0.7	0.2	**0.5 a**
Average	**0.6 a**	**0.2 a**	**0.3 a**	**0.3 a**	

* Different letters indicate means are significantly different

The percentage of A-grade kernel in 2008 was again significantly affected by treatment (*P*<0.001) and variety (*P*<0.001). The Experimental and Standard treatments had significantly highest percentage of A-grade kernel and the Wasp only treatment had the significantly smallest percentage of A-grade kernel. Again, the

variety A4 had significantly highest and the variety 246 the significantly lowest percentage of A-grade kernel (Table 6.2.3.7).

Table 6.2.3.7: Percentage of A-grade kernel (in %) in 2008

	246	741	849	A4	Average
IPM	20.6	28.1	22.7	34.7	**26.5 b**
Standard	24.8	32.0	32.0	40.0	**32.2 a**
Wasps only	22.8	29.1	17.0	25.3	**23.6 c**
Experimental	25.6	33.7	29.3	41.6	**32.6 a**
Average	**23.5 d**	**30.8 b**	**25.2 c**	**35.4 a**	

*Different letters indicate means are significantly different

Treatment ($P< .001$) and variety ($P<0.001$) had a significant impact on yield in 2008. Yield in the Experimental treatment had the significantly highest yield and the IPM treatment the significantly lowest. This time the variety A4 had the significantly highest and the variety 246 the significantly lowest yield (Table 6.2.3.8).

Table 6.2.3.8: Average nut yield (DNIS@ 10% moisture content) per tree in CTH trial across treatments and varieties in 2008

	246	741	849	A4	Average
IPM	2.9	5.8	3.5	9.8	**5.5 d**
Standard	3.7	6.2	9.4	15.3	**8.6 b**
Wasps only	3.9	7.2	3.4	13.1	**6.9 c**
Experimental	6.6	11.1	7.7	17.3	**10.7 a**
Average	**4.3 d**	**7.6 b**	**6.0 c**	**13.9 a**	

* Different letters indicate means are significantly different

The results for 2008 suggest that thiacloprid in the IPM treatment is inferior to endosulfan in the Standard treatment (Tables 6.2.3.5 and 6.2.3.8). Nut quality is lower, nut yield is lower and the levels of FSB damage significantly higher in all varieties. Kernel grub did not eventuate in either treated area so spraying was effective. MNB was again at negligible levels.

Results from season 2008/2009

The main aim in 2008/2009 was to compare early season trichlorfon and endosulfan applications with a single fipronil application. Field test pyraclostrobin against carbendazim in the amount of husk spot infested nuts. Details of IPM, Standard, Wasps only and Experimental treatments are listed in Table 6.1.4 (see page 29).

In the 2009 harvest treatment ($P<0.001$) and variety ($P<0.001$) again had significant impact on FSB damage in kernel. The Standard treatment had the significant lowest damage and the Wasp only treatment the significantly highest damage. The varieties

246 and 741 again had significantly less damage than the varieties 849 and A4 (Table 6.2.3.9).

In 2009 treatment (*P*=0.011) and variety (*P*=0.021) had a significant effect on MNB damage in kernel. Standard and IPM treatments had significantly less MNB damage then Wasp only and Experimental treatments. The varieties 741 and 849 had significantly lower MNB damage in kernel than the varieties 246 and A4 (Table 6.2.3.10).

Table 6.2.3.9: FSB damage in macadamia kernel (in %) in 2009

	246	**741**	**849**	**A4**	**Average**
IPM	8.9	9.9	11.1	10.8	**10.2 b**
Standard	3.9	7.6	7.5	8.8	**6.9 a**
Wasps only	12.5	10.7	21.3	27.7	**18.0 c**
Experimental	9.1	7.1	12.7	10.9	**9.9 b**
Average	**8.6 a**	**8.8 a**	**13.2 b**	**14.5 b**	

* Different letters indicate means are significantly different

Table 6.2.3.10: MNB damage in macadamia kernel (in %) in 2009

	246	**741**	**849**	**A4**	**Average**
IPM	0.11	0.00	0.00	0.00	**0.03 a**
Standard	0.05	0.03	0.00	0.00	**0.02 a**
Wasps only	0.17	0.03	0.08	0.33	**0.15 b**
Experimental	0.16	0.00	0.13	0.36	**0.16 b**
Average	**0.13 b**	**0.01 a**	**0.05 a**	**0.17 b**	

* Different letters indicate means are significantly different

Table 6.2.3.11: Percentage of A-grade kernel (in %) in 2009

	246	**741**	**849**	**A4**	**Average**
IPM	20.3	28.2	32.4	33.8	**28.7 b**
Standard	28.9	29.2	34.7	34.6	**31.8 a**
Wasps only	16.4	25.5	20.0	20.6	**20.6 c**
Experimental	22.9	29.8	27.9	32.3	**28.2 b**
Average	**22.1 c**	**28.2 b**	**28.8 ab**	**30.4 a**	

* Different letters indicate means are significantly different

Table 6.2.3.12: Average nut yield (DNIS@ 10% moisture content) per tree in CTH trial across treatments and varieties in 2009

	246	**741**	**849**	**A4**	**Average**
IPM	15.0	18.2	10.5	9.5	**13.3 b**
Standard	20.3	19.6	14.6	10.3	**16.2 a**
Wasps only	9.1	14.2	5.3	4.6	**8.3 c**
Experimental	14.3	23.4	11.6	12.5	**15.4 a**
Average	**14.7 b**	**18.8 a**	**10.5 c**	**9.2 c**	

* Different letters indicate means are significantly different

The percentage of A-grade kernel in 2009 was again significantly affected by treatment (P<0.001) and variety (P<0.001). The Standard treatment had the significantly higher percentage of A-grade kernel and the Wasp only treatment had the significantly lowest percentage of A-grade kernel. Again, the variety A4 had the highest and the variety 246 the significantly lowest percentage of A-grade kernel (Table 6.2.3.11).

Treatment effect (P<0.001) and variety effect (P<0.001) had significant impact on yield during 2009. Yield in the Standard and Experimental treatments had the significantly highest yields and the Wasp only treatment had the significantly lowest yield. The variety 741 had the significantly highest and the varieties 849 and A4 had the significantly lowest yields (Table 6.2.3.12).

The results for 2009 suggest that trichlorfon in the IPM treatment is inferior to endosulfan in the Standard treatment (Tables 6.2.3.9 and 6.2.3.12). Nut quality is lower, nut yield is lower and the levels of FSB damage significantly higher in all varieties. MNB was again at negligible levels. The single fipronil application in the Experimental treatment did not give adequate control of FSB. Husk spot was not an issue in the pyraclostrobin treatment with nut yields similar to those treated with carbendazim (Table 6.2.3.12).

Results from season 2009/2010

The main aim in 2009/2010 was to compare early season trichlorfon, endosulfan and Bayer 092 applications. Field test pyraclostrobin against carbendazim in the amount of husk spot infested nuts. Details of IPM, Standard, Wasps only and Experimental treatments are listed in Table 6.1.5 (see page 29).

In 2010 treatment (P<0.001) and variety (P<0.001) again had significant impact on FSB damage in kernel. The Experimental treatment had the significant lowest damage and the Wasp only treatment the significantly highest damage. The variety A4 had significantly more FSB damage in kernel than the other varieties tested (Table 6.2.3.13).

Treatment (P=0.011) and variety (P=0.021) had a significant effect on MNB damage in kernel Standard and Experimental treatments had significantly less MNB damage and the Wasp only treatment significantly more damage. The variety 741 had the significantly lowest MNB damage in kernel and the variety 849 the significantly higher damage (Table 6.2.3.14)

Table 6.2.3.13: FSB damage in macadamia kernel (in %) in 2010

	246	**741**	**849**	**A4**	**Average**
IPM	15.2	21.4	31.5	30.3	**24.6 c**
Standard	11.9	14.1	13.8	17.1	**14.2 b**
Wasps only	33.2	27.0	18.2	61.7	**35.0 d**
Experimental	6.3	7.4	8.5	9.5	**7.9 a**
Average	**16.6 a**	**17.5 a**	**18.0 a**	**29.7 b**	

* Different letters indicate means are significantly different

Table 6.2.3.14: MNB damage in macadamia kernel (in %) in 2010

	246	741	849	A4	Average
IPM	0.3	0.1	2.0	1.3	**0.9 b**
Standard	0.5	0.2	0.2	0.0	**0.2 a**
Wasps only	1.9	0.2	3.4	0.0	**1.4 c**
Experimental	0.0	0.1	0.9	0.2	**0.3 a**
Average	**0.7 c**	**0.2 a**	**1.6 d**	**0.4 b**	

* Different letters indicate means are significantly different

The impact of treatment (P<0.001) and variety (P<0.001) on the percentage of A-grade kernel in 2010 was again significant. The Experimental treatment had the significantly highest percentage of A-grade kernel and the Wasp only treatment had the significantly lowest percentage of A-grade kernel. The variety 741 had the highest and the variety 246 the significantly lowest percentage of A-grade kernel (Table 6.2.3.15).

Table 6.2.3.15: Percentage of A-grade kernel (in %) in 2010

	246	741	849	A4	Average
IPM	23.2	30.0	26.2	31.0	**27.6 c**
Standard	27.9	31.9	31.2	37.2	**32.1 b**
Wasps only	14.5	26.7	19.4	13.5	**18.5 d**
Experimental	29.7	35.9	34.9	40.8	**35.3 a**
Average	**23.8 c**	**31.1 a**	**27.9 b**	**30.6 a**	

* Different letters indicate means are significantly different

Table 6.2.3.16: Average nut yield (DNIS@ 10% moisture content) per tree in CTH trial across treatments and varieties in 2010

	246	741	849	A4	Average
IPM	2.9	8.9	4.9	6.2	**5.7 b**
Standard	4.0	9.3	3.4	3.9	**5.2 b**
Wasps only	0.5	5.4	0.4	0.9	**1.8 c**
Experimental	4.1	15.1	6.0	7.2	**8.1 a**
Average	**2.9 c**	**9.7 a**	**3.7 bc**	**4.6 b**	

* Different letters indicate means are significantly different

In 2010, treatment (P<0.001) and variety (P<0.001) significantly affected yield. Yield in the Experimental treatment had the significantly higher yield and the Wasp only treatment had the significantly lowest yield. The variety 741 had the significantly highest and the varieties 849 and 246 had the lowest yields (Table 6.2.3.16).

The results for 2010 have finally given us a product that appears to have superior performance to endosulfan in the short term. Bayer 092 treated nuts in the Experimental treatment had higher quality, 50% higher nut yield and half the levels of FSB damage than the standard treatment in all varieties tested (Tables 6.2.3.13 and 6.2.3.16). This finding needs to be tested further and on different crops to ensure no secondary effects show and residue data should be gathered where possible to assist with the registration of the product. It will still take some more time (possibly 2-5 years) to get sufficient data for registrion for macadamia and other industries.

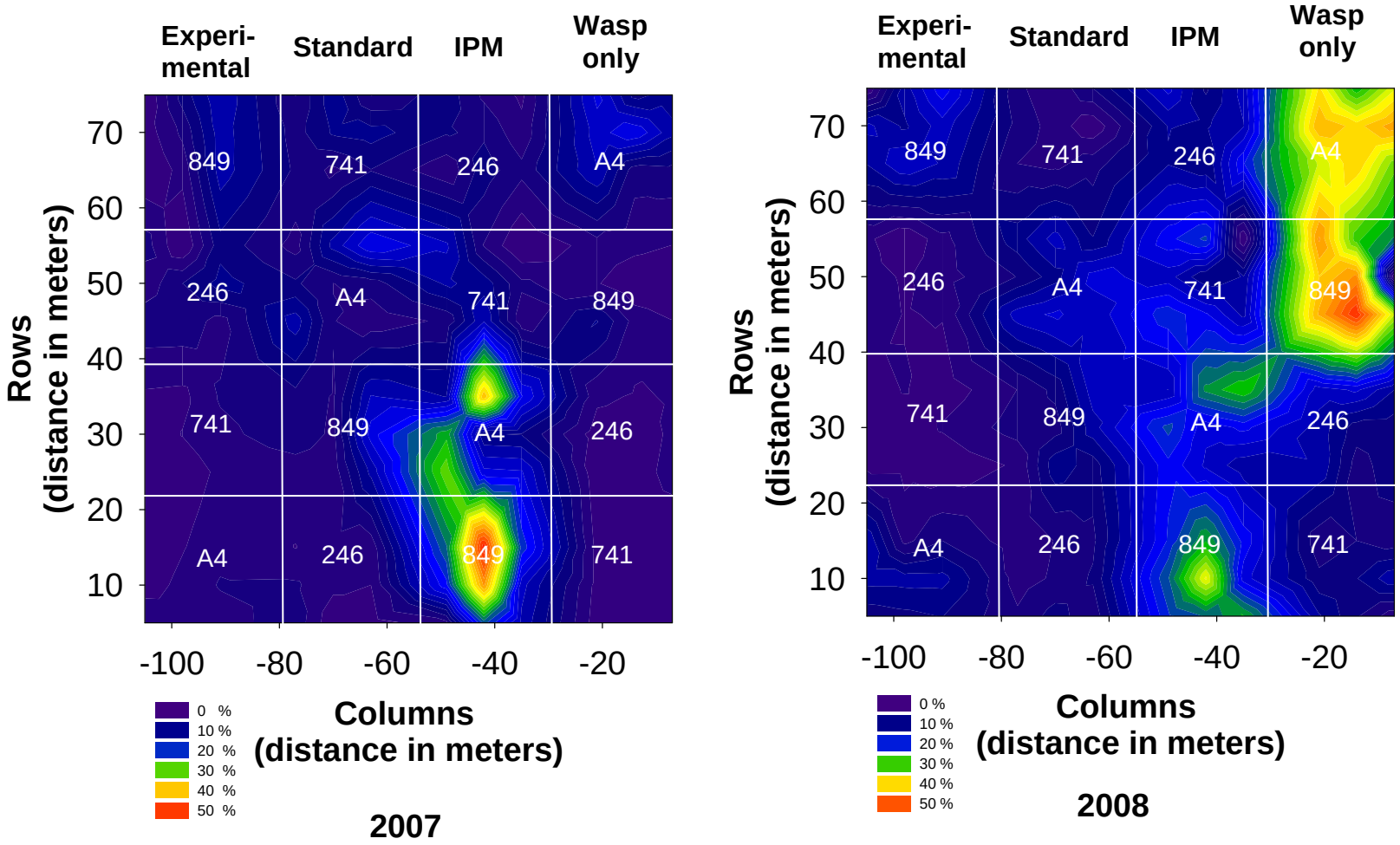

Figure 6.2.4.1: Spatial distribution of FSB damage in macadamia kernel in CTH trial orchard 2007 and 2008 (left to right)

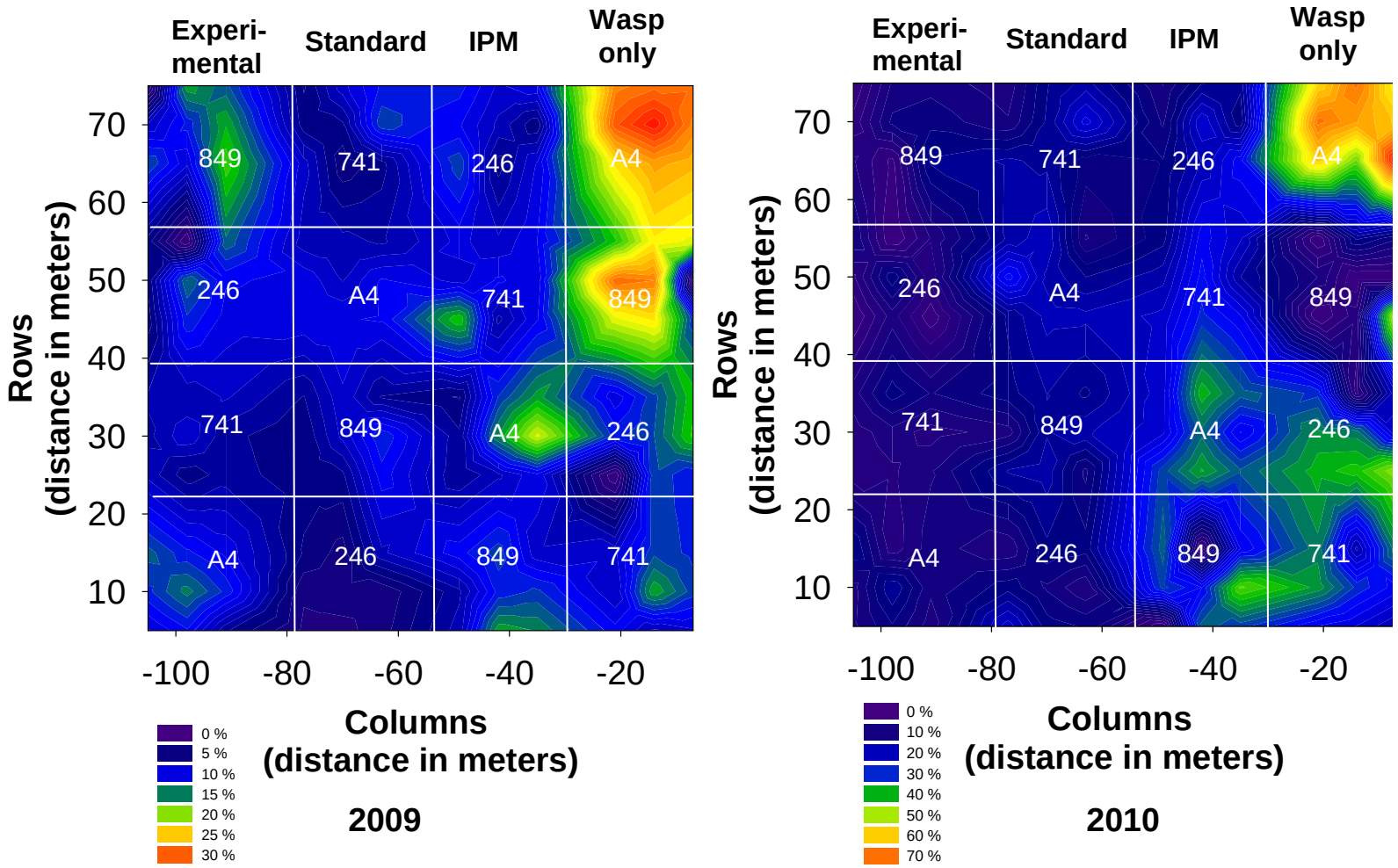

Figure 6.2.4.2: Spatial distribution of FSB damage in macadamia kernel in CTH trial orchard 2009 and 2010 (left to right)

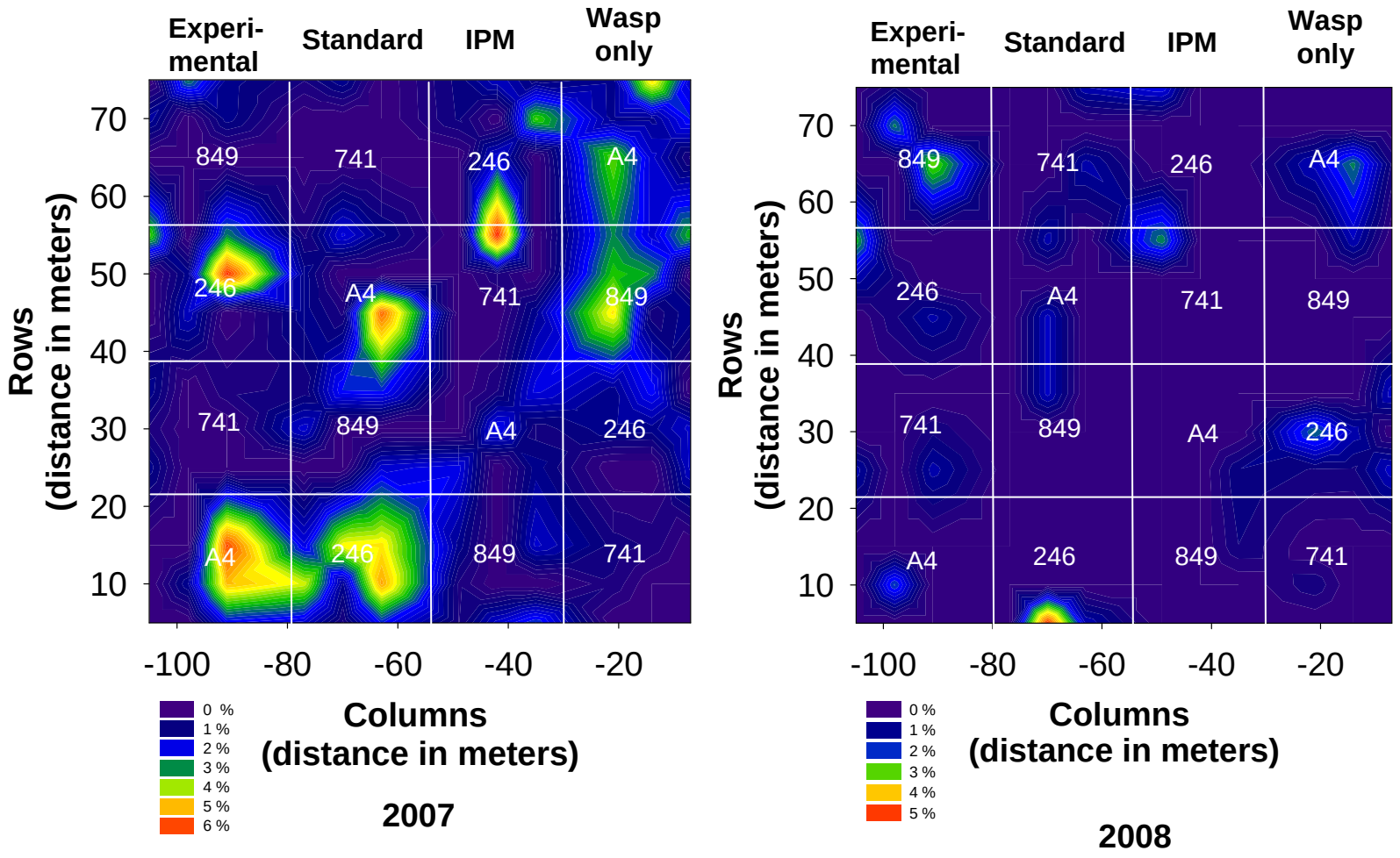

Figure 6.2.4.3: Spatial distribution of MNB damage in macadamia kernel in CTH trial orchard 2007 and 2008 (left to right)

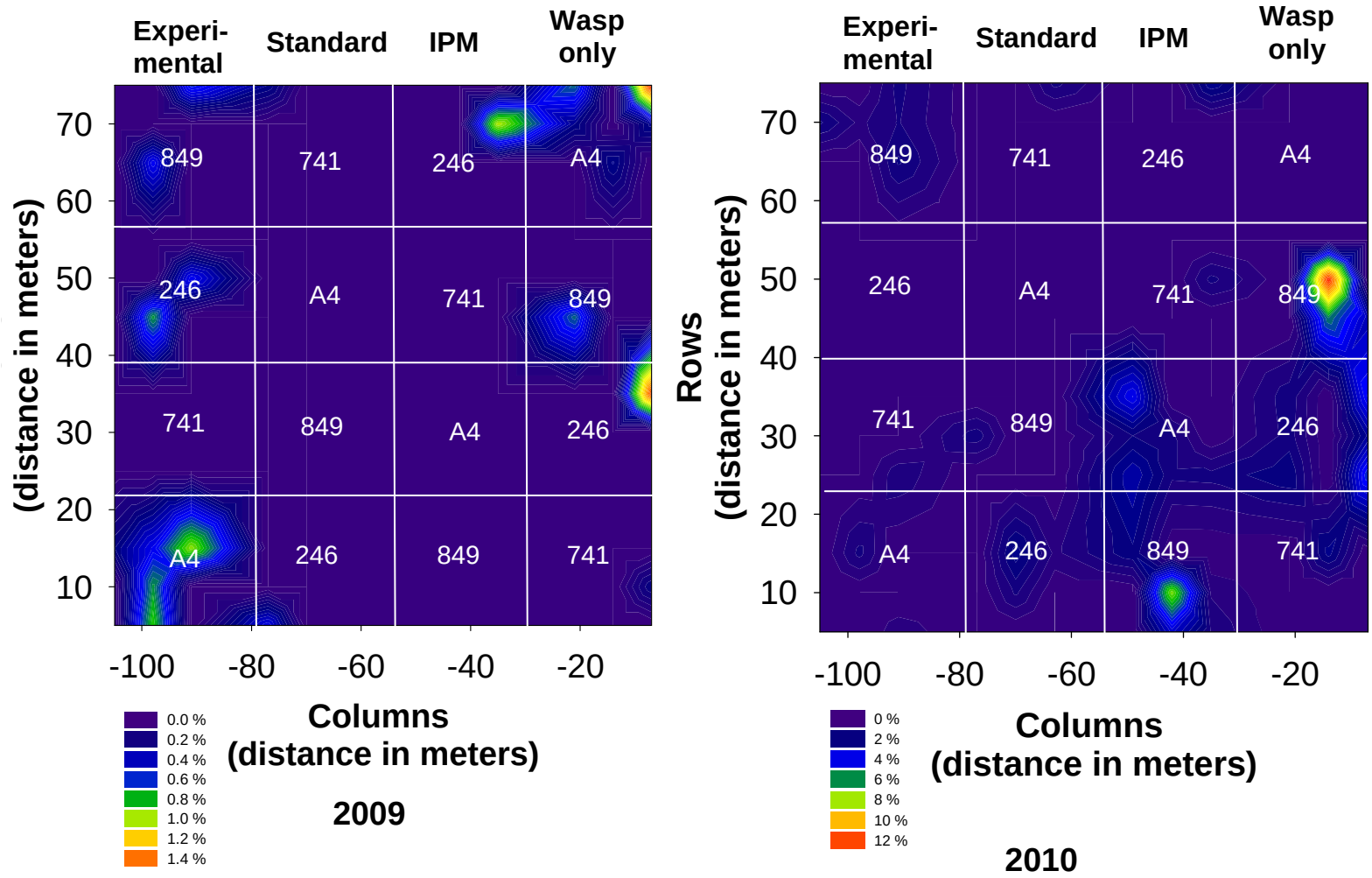

Figure 6.2.4.4: Spatial distribution of MNB damage in macadamia kernel in CTH trial orchard 2009 and 2010 (left to right)

6.2.4. Spatial damage distributions

By maintaining the individual tree damage layout and the longer term untreated and treated comparisons it is possible to establish whether the anecdotal claims are true that FSB attack the same plants in an orchard each year and often within a season. The even seeding of the plot with FSB adults prior to each season should produce a random damage array if all varieties were equally preferred and treatments equally effective. The spatial distribution of the FSB damage is shown for each season in Figures 6.2.4.1 and 6.2.4.2. The development of a clear “hotspot” is apparent on the neighbouring trees of the “Wasp only” A4 block as each season has progressed.

The implications of this finding were that indicator plants were feasible as a monitoring tool in macadamia (Maddox and Huwer, unpublished data). The use of these plants could be expanded to a trap crop if it can be shown that the bugs will return after a spray event to the same tree and the understanding of how that is done could make it possible to use alternate plants to establish FSB eggs parasitoids in close proximity to orchards.

This is commercially important information for the crop scouts because these particular sites when discovered in an orchard can dramatically reduce the monitoring time to make effective spray decisions for FSB.

For MNB the spatial damage pattern do not follow any expected pattern with only a mild preference for variety 849 visible in some seasons when late season activity has occurred (Figures 6.2.4.3, 6.2.4.4).

6.2.5. On farm trials

Field trials were further conducted on three commercial farms in the main macadamia production regions. This allowed us to make it more relevant for commercial farms and also provided tool to relay research outcomes to growers.

6.2.5.1. Materials and Methods

Field sites for on farm trials were located at Rouse, Northern Rivers (NSW) at Amamoor, Glasshouse Mountains and Bundaberg (QLD). An area of about 1 hectare each for IPM and Standard were used for a field trial on the commercial farms. In these on farm trials we tested our revised IPM strategy. In the IPM treatment tebufenozide (seasons 2006/2007 and 2007/2008) or methoxyfenozide (once it was registered, in seasons 2008/2009 and 2009/2010) and the egg parasitoid *T. cryptophlebiae* were used for MNB management instead of beta-cyfluthrin later in the season. Thiamethoxam (seasons 2006/2007 and 2007/2008) and trichlorfon (seasons 2008/2009 2009/2010) were used for FSB management instead of endosulfan. The kernel recovery methods and data analysis were the same as described for the trials conducted at CTH. Collaborators from commercial farms were asked to send 300 nut samples from IPM and Standard treatments for each harvest.

6.2.5.2. Results

Results from the on farm trials concentrated on FSB and MNB damage. Treatment had significant impact on FSB damage, in-kernel only, in 2007 and 2009, but not in 2008 and 2010 (Table 6.2.5.1). Overall, there was no significant different between IPM and Standard treatments with regards to FSB damage (Table 6.2.5.1).

Table 6.2.5.1: FSB damage in macadamia kernel (in %) from 2007 to 2010 on commercial farms.

Year	IPM	Standard	Fpr.
2007	**4.1 b**	**1.3 a**	*P*=0.007
2008	**2.0 a**	**1.2 a**	*P*=0.334
2009	**1.1 a**	**2.9 b**	*P*=0.035
2010	**0.8 a**	**0.7 a**	*P*=0.812
2007-2010	**2.0 a**	**1.5 a**	*P*=0.459

Different letters indicate means are significantly different

In the first year (2007) MNB damage in kernel was significantly higher in the IPM treatment than in the Standard treatment (Table 6.2.5.2). In the following years (2008, 2009, 2010 and overall) there was no significant difference in MNB damage in kernel between IPM and Standard treatment (*P*>0.05) (Table 6.2.5.2).

Table 6.2.5.2: MNB damage in macadamia kernel (in %) from 2007 to 2010

Year	IPM	Standard	Fpr.
2007	**2.1 b**	**1.3 a**	*P*=0.038
2008	**3.3 a**	**4.0 a**	*P*=0.702
2009	**1.2 a**	**1.4 a**	*P*=0.408
2010	**1.1 a**	**0.9 a**	*P*=0.492
2007-2010	**1.9 a**	**1.6 a**	*P*=0.998

* Different letters indicate means are significantly different

7. Semiochemistry, trap cropping and the possibility of monitoring the *Amblypelta nitida* population.

A branch of biological study has developed around chemical signals produced by herbivores and plants that can influence higher trophic levels of an ecosystem (Langenheim, 1994). Semiochemistry operates at several levels and can be separated by the source of the compound (e.g. floral odours or insect pheromones). Manipulation of chemical signals may allow the regulation of pest populations and minimise the need for blanket spraying. There are four main areas of semiochemical research.

1. Compounds that draw the pest to the crop.

2. Sex pheromones

3. Aggregation markers produced by the pest allow others to find the food source (and also allow parasitoids and predators to find the pest)

4. Repellents (prevent the pest from establishing).

The behaviour of FSB may be open to manipulation if the key to aggregation behaviour can be understood. Spatial analysis of the damage caused in macadamia has shown that in even a traditionally sprayed management system FSB can cause just as much damage after December on A4 macadamia as in an unsprayed system which implies crop re-entry after the traditional treatment period (Huwer *et al.,* 2006). The high focus of damage by FSB shown in this work (see previous chapter and Huwer *et al.,* 2006) supports the hypothesis that FSB populations are aggregated for at least some period during the fruiting season of macadamia. The studies also showed that when given a choice of macadamia varieties the levels of FSB damage on variety 741 were far lower later in the season than on variety A4 and 849.

The questions to answer include:

- Do FSB randomly select trees that they attack initially?

- Do FSB form reliable “hot spots” which could be used for monitoring more efficiently?

- What makes a “hot spot”?

- Can “hot spots” be artificially produced?

These are questions that have been studied but not yet resolved (Waite, 2000; Huwer *et al.,* 2006). The critical components are: why do FSB return to the same area, and if tree marking via semiochemistry is involved, exactly how does it work (Aldrich *et al*., 1993; Waite, 2000; Huwer *et al*., 2006)?

The reproductive state of overwintering *Amblypelta nitida (FSB)*

The long held belief that FSB over-winter as adults and reinfest orchards each spring was first suggested by Ironside (1981). There is evidence that FSB can live for over 12 months in a laboratory colony, but under field conditions the longevity of adults is considered to be 2-3 months (Waite, 2000). This adult longevity is a key to the problem posed by FSB, as individuals can cause damage on a large number of nuts during their life. Adults are very mobile and are difficult to monitor in older orchards with higher canopies. The normal reproductive rate for both *Amblypelta* species was previously listed as close to zero throughout May until August and linked to day length (Waite, 2000). To test the field relevance of this finding the build up of populations during winter needs to be observed on a variety of hosts. If temperature and day length restrict reproduction then few nymphs will be found in winter. If no breeding is taking place then finding how the insects survive the winter is crucial to restricting the following season's infestation. This approach was taken by us to manage MNB and infestation levels have been dramatically lowered by targeting the overwintering sites as well as the orchard generation of that pest. If they do breed through winter, is it possible to find the key winter host areas and restrict the breeding with egg parasitoid releases?

The local movement of *Amblypelta nitida*

The aim of the study was to obtain data on seasonality of FSB populations and determine the importance of proximity to other alternate FSB hosts (i.e. native scrub, orchards of other hosts) and migration between hosts. Previously population levels of FSB were inferred by using damage levels rather than incidence counts. Incidence counts require the establishment of a variety of sequentially flowering/ fruiting hosts and continuous monitoring to track FSB throughout seasons. This has been achieved at CTH Alstonville using several existing plantings (>30 years old) and establishing new trap hedges highly susceptible to FSB with the potential for easy monitoring (see Figures 7.1.2 and 7.2.1). Previous mark release recapture studies show high levels of recruitment into and out of regions on *M. paniculata* hedges in North and South Queensland through spring and summer (Waite, 2000). A similar result was obtained in papaya during autumn and winter 1999 with *A.l. lutescens* (Waite, 2000). High continual recruitment means that the plant is attracting the FSB population.

There may be, however, a much more dispersive behaviour pattern in summer for *A. nitida.* Previous studies showed importance of some *Eucalyptus* species as hosts for the shoot feeding *A.l. lutescens*, *A. cocophaga* (Waite, 2000) and it is suspected that gum nuts are a significant breeding host for *A. nitida* as well as many other unlisted rainforest species (Waite & Huwer, 1998) (see Figure 7.1.2). The role of the rainforest native ribbonwood tree *Euroschinus falcata* (Anacardiaceae) in the Nambour district is a case in point. The Nambour area, long considered a very high pressure district for FSB with many small orchards, forests and small rainforest pocket gullies, demonstrated unusual *Amblypelta* spp. behaviour. The swarming behaviour of adult *Amblypelta* spp. on flowering and fruiting of this native ribbonwood tree over a period of 2-3 weeks in mid – late December in 2008 and 2009 on a passionfruit farm in South East Queensland was confirmed (see Figure 7.1.2). This aggregation may prove useful for control because a concentration of *Amblypelta* spp. in a small area may allow for highly targeted spraying.

Semiochemistry and monitoring of *Amblypelta nitida*

Finding which compounds are biologically active as semiochemicals and how much is needed to draw a response from *A. nitida* is difficult. With the assistance of the Dr. Mike Russell and Professor Ian Southwell we investigated some of these questions for *A. nitida.* Using the solid phase microextraction (SPME) air trapping technique and comparative trace matching it is possible to determine the rate and ratio of compounds present in test samples (i.e. flowers, fruit etc.). SPME columns are exposed in controlled airspaces to known concentrations of standard components to cross check the chromatograph columns retention time and over known exposure periods to calibrate the amount of compound being emitted from a source (Anonymous, 1998; Pawliszyn, 2009).

Plant compounds that maybe involved in attracting FSB include benzaldehyde (which appears to be a key floral component that assists female *A. nitida* to find hosts). The problem with using benzaldehyde in the field is that it is unstable when in water and reacts to form benzoic acid. Under laboratory conditions, a mild attraction to benzaldehyde by FSB was shown. In the field using macadamia trees with no flowers, the addition of synthetic raw benzaldehyde could enhance damage to the crop of these trees significantly (Table 7.2.1) (Huwer *et al.,* 2006). The more interesting response to volatiles appears to be the movement of FSB to an autumn/winter host (*M. paniculata*) with the same compound in the flowers as macadamia. There is a strong correlation of FSB nymphs and adults collected on *M. paniculata* with damage levels seen on the crop in the neighbouring orchard (see Figures 7.1.2 and 7.1.3).

Sex pheromones of *A.l. lutescens* are known and some of *A. nitida* are also well defined (Aldrich *et al.,* 1993; Waite, 2004; Fay pers. comm). At this stage, no data showing the actual capture rates and population density and damage rates has been presented, nor the design of the traps that will be used to capture FSB adults. The traps used for some Hemipteran work overseas were tested at CTH by us on a *M. paniculata* hedge with a known density of *A. nitida* to see if the FSB adults would enter the trap baited with single volatile compounds (see Figure 7.1.2).

A. nitida aggregation markers are the main area where research by Fay (DEEDI) and Aldrich (USDA) is continuing and compounds that mark the presence of *A. nitida* on a host are known (Baker *et al.,* 1972). Hexyl hexanoate and hexyl acetate (Baker *et al.,* 1972; Aldrich *et al.,* 1993) are detectible with the SPME system whenever *A. nitida* feed directly on macadamia nuts (Huwer *et al.,* 2006).

Aggregation by FSB on *Euroschinus falcata* (ribbonwood) may indeed be explained this way, with FSB being attracted by the flower volatiles of the host (ribbonwood) and then emitting sex pheromones (Figure 7.1.3). The question is whether the tree is attractive before the adults arrive and feed or does feeding itself initiate the swarming. In the act of feeding FSB leaves residues of several compounds that will mark a tree as being fed upon. FSB leave a small sulphurdryl tube at the centre of each feeding mark, and an overflow of glucose rich plant fluid containing at least 3 enzymes injected by FSB will ooze to the surface and harden to a resin on some macadamia varieties leaving the characteristic spotting on the fruit surface (Miles & Taylor, 1994) (see Figure 7.1.2). A compound that shows where an insect is active will also assist parasitoids and predators searching and may even be the key behavioural switch.

Another avenue of investigation (not been pursued in this project) involves some Hemipterans releasing volatile defensive compounds. Some compounds can be

repellent to FSB (alarm pheromone), and some compounds may be species specific markers such as with litchi stink bug or bronze shield bug (that replace FSB on litchi and citrus in late spring at CTH Alstonville (Figure 7.1.2)).

Indicator trees and monitoring *Amblypelta nitida*

Following on from semiochemistry is the "hot spot" concept that certain areas are more prone to FSB attack within an orchard (Waite, 2000). Therefore placing the most appropriate bait in those areas should give the best indication of FSB activity. Trees in an avocado orchard neighbouring scrub or forest record significantly higher damage levels than avocado trees in the middle of the orchard, further away from the scrub (Waite, 2000). In longans double the population levels were recorded on longan trees neighbouring windbreak areas (Waite, 2000). Live FSB are rarely caught in conventional insect traps due to the high visual acuity of the FSB and their generally low population density. Such trapping problems increase the attractiveness of using indicator plants in some orchards (Waite, 1993). The aim of this project was to find a FSB monitoring hedge, suitable for macadamia and avocado orchards.

In various horticulture crops variety appears to be the most important factor for susceptibility to FSB damage within orchards. The carambola (star fruit) variety Thai Knight has the highest damage for carambola. However, some trees within uniform custard apple plots are always hit first and heaviest by FSB and these trees are used by growers to monitor FSB activity (Paxton, pers. comm.). These observations suggest that particular growth traits of an individual tree are also important.

The avocado variety Parida has double the damage level of variety Fuerte and the avocado variety Pinkerton preferentially sustains early season damage on fruit up to 4cm size (Waite, 2000). There is data showing clear edge effects and also showing damage halving each row from the margins in avocado orchards (Waite, 2000). Longans are a mature fruit host and provide an exception to the general observation of breeding FSB moving on to young developing fruit. In South African macadamia crops, a link between tree density and stink bugs (*Nezara viridula* and *Nezara pallodoconspersa*) damage were established (Schoeman, 2007). There is conflicting data linking the impact of tree canopy light and FSB damage. In mangoes FSB gave a clear preference for closed canopies, in carambolas they preferred more open canopy (Waite, 2000).

Beneficials and mass rearing release strategies

A composite of egg parasitoid species have been found on *Amblypelta* species in the field (Waite, 2000). Species included *Anastatus* sp*., Gryon* sp., *Ooencyrtus* sp. and *Centrodora* darwini (Girault) (Hymenoptera: Chalcidoidea) from almost all growing areas. Field egg parasitism rates (>90%) for *Amblypelta* species have been recorded in North Queensland (Fay & Huwer, 1993), and similar levels in South Queensland (Waite, 2000). Field data is available for Northern NSW where the FSB egg parasitoid *Centrodora darwini* has been trialled to investigate impact on FSB damage with limited success in macadamia (Huwer *et al.,* 2006, chapter 6). The egg parasitism of FSB by *C. darwini* has not been able to prevent crop damage.

To have any impact on FSB damage levels of parasitoids need to be boosted coming out of winter, which will require the introduction of the parasitoids on low density

hosts. Key release areas within winter breeding regions and neighbouring orchards also need to be identified through a carefully evaluation of searching behaviour and reproductive potential of each species and taking into account potential regional differences

7.1. Materials and Methods

Field monitoring of *Amblypelta nitida*

The aerial photograph (courtesy Col Cooksey) and legend (Figure 7.1.1) shows the proximity of the various orchard plantings to each other at CTH and the planting times. Within the CTH Entomology block trials (#6 Figure 7.1.1) between each season the trees (n=174 were checked fortnightly each season (October to March) and sightings of live FSB recorded for individual trees (see chapter 5).

On *M. paniculata* hedges (#8, #10 Figure 7.1.1 and 7.1.4) a period of 30 minutes were allocated weekly to collect live FSB from two hedges and data was recorded for individual plants. The observation and collecting is done between 10am and midday to ensure the FSB are exposed to enough heat in winter to be active. The data was presented as a monthly sum and pooled by FSB age (instars I, II & III = small, instars IV and V = large). Trees were numbered from the eastern edge of each hedge and surveyed every week each for 15 minutes. Visible FSB were collected and removed. Trees were ranked by the number of FSB collected during the capture period.

Survival of *Amblypelta* sp. on *M. paniculata* berries was determined by using 3 replicates of 5 first instar FSB nymphs in 6 x 750ml plastic containers over a period from 19/5/2009 until 5/6/2009. Berries were collected weekly from the sprayed hedge (highway hedge #9 figure 7.1.1) and the unsprayed hedge (germplasm area #2 figure 7.1.1). Developing FSB nymphs were transferred to sprayed or unsprayed fresh berries each week and mortality recorded.

The 23 trees of the *M. paniculata* hedge at CTH Alstonville contained 7 FSB capture traps (obtained from the USDA) between 21/5/2008 and 31/7/2008. Traps were fitted with 2ml lure vials containing nothing (blank), or purified hexyl hexanoate, hexyl acetate or benzaldehyde. Lures were changed monthly and the target population of FSB was collected from hedge each week to estimate the level of activity and to observe the presence or absence on traps.

The longan area of the farm (#9 Figure 7.1.1) and the floral sequence hedges were also monitored for FSB at fruit set (#7 Figure 7.1.1). Live FSB were collected from plants and fruit was examined for damage during autumn to determine if there was damage on various fruits. Visible lesions were counted on fruit from the Zuttano avocado, coffee berries, guava, custard apple and L64 macadamia (initiated in 2010).

Legend for figure 6.2.1:
1=germplasm block, 2000
2=germplasm hedge,2008
3=FSB sink block,2007
4=rainforest remnant area,
5=accession block, variety evaluation and parasite trapping;1978
6=CTH entomology macadamia block1998
7=flower sequence hedge2007
8=highway *Murraya* hedge 1990
9=longan block 1978
10= arboretum *Murraya* hedge 2004
11=CTH avocado block 2004

Figure 7.1.1: Aerial photograph of NSW Department of Industries & Investment Centre for Tropical Horticulture (CTH) Alstonville site (2010) Legend shows block and hedge locations and planting dates.

Figure 7.1.2:

Top left: *Musgraveia sulciventris* (bronze orange bug) aggregation on Tahitian lime trees at Wollongbar NSW

Top right: *Lyramorpha rosea* (litchi stink bug) aggregation in December 2001 at CTH Alstonville

Bottom left: Fresh exudate oozing from feeding by *Amblypelta nitida* on A4 macadamia variety nutlets, the sulphurdryl straw remains in the tissue.

Bottom right: Glucose rich exudate on the surface of A4 macadamia drying after feeding by *Amblypelta nitida.*

Figure 7.1.3:

Top left: Caged feeding by *Amblypelta nitida* adults on *Eucalyptus ficafolia* gumnuts (spring 2001 at CTH) Alstonville NSW

Top right: Damage to gumnuts caused by *Amblypelta nitida* adults feeding

Bottom left: Floral sequence hedge at CTH Alstonville 2010 used to attract and maintain *Amblypelta nitida* and to release and monitor egg parasitoids.

Bottom right: *Amblypelta lutescens* feeding on ribbonwood (*Euroschinus falcata*) fruit at *a* passionfruit farm Nambour Queensland December 2009 (Photo courtesy of Keith Paxton).

Figure 7.1.4:

Top left: Inverted bottle traps containing 2ml lure vials with either, hexyl hexanoate, hexyl acetate or benzaldehyde suspended on *M. paniculata* plants at CTH Alstonville to trap *Amblypelta nitida.*

Top right: *Amblypelta nitida* oviposition into macadamia florets (CTH Alstonville).

Bottom: FSB capture traps from USDA fitted with 2ml of lures and suspended in the 23 trees of *M. paniculata* hedge (CTH Alstonville 2008).

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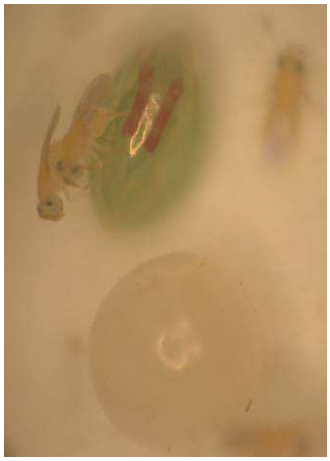

Figure 7.1.5:

Top left: *Amblypelta nitida* attractant bioassay setup (constructed from fish tanks, stainless steel plates with equal apertures and capture tanks below, one containing a test compound emitting from a 2ml lure vial.)

Top right: *Amblypelta nitida* females attracted to a 2ml lure vial containing benzaldehyde. Benzaldehyde is the main component of macadamia and *M. paniculata* floral essence and 100 times more potent in the hedge plant.)

Bottom: Left -Scelionid parasitoid emerging from *Amblypelta nitida* eggs (*Gryon* sp. has a distinctive equatorial stripe during development in the host 1 wasp emerges from 1 egg).

Bottom centre: Generalist Aphelinid egg parasitoid *Centrodora darwini* attacking *Amblypelta nitida* eggs (spined citrus bug egg below) (13-27 wasps emerged from a single egg. *C. darwini* has been released in the field trials 2001-2008 at CTH Alstonville.

Bottom right: - Egg parasitoid capture vessels (used with fresh *Amblypelta nitida* sentinel egg cards on Velcro strips) within 100mm diameter pipe sections at 1.5m through the canopy at CTH Alstonville.

The entomology orchard FSB observations (analysis in section 6.2.3) were of 36 trees fortnightly (9 trees of each 4 macadamia varieties, 741, 849, A4 and 246). The unsprayed areas of the orchard have not been sprayed since 2000. The standard treatment area (STD) is managed with monthly endosulfan and beta-cyfluthrin sprays. Floral sequence hedges were planted (50 m from the macadamia orchard), including avocado (Avo = Zuttano), macadamia (Maca = L64 a macadamia cultivar with a %KR>52%), custard apple pink mammoth (CustA), guava (Gua), longan (Long), and *M. paniculata* (M.P). The germplasm block contains all the known wild trees of the 4 known macadamia species (≥600 trees) and the trap hedge area was planted within 40 m of the western edge of the germplasm block (#1 Figure 7.1.1).

Parasitoids were trapped in various orchards using freshly laid FSB eggs (normally 5 target eggs) attached to Velcro® backed sticky paper labels ("trap egg" card). These were placed in 100mm pipe sections and suspended at about 1.8m in the macadamia orchards egg cards were collected after 4 days (#5 Figure 7.1.1). Other capture devices included dark green *Helicoverpa* spp. traps modified with a tray to hold the egg cards under the lids to prevent rain damage, these were hung in the upper macadamia canopy (8m), and transparent screw cap polycarbonate jars with 200mm walls and drilled out panels to allow parasitoid access in confined canopies (e.g. *M. paniculata*). Parasitism was determined under the microscope (12x magnification) after 6 days and parasitoid species was determined by internal egg structures (*Centrodora darwini, Gryon* sp*,* and *Anastatus* sp. eggs all look different during development (Figure 7.1.5)). The number of eggs that were successfully parasitised on egg cards was recorded during each month of trapping and labelled according to the plant they were collected on. The frequency of parasitoid trapping was dependent on the number of FSB eggs available and only *A. nitida* eggs were used for trapping in the field.

The macadamia germplasm area at CTH and the FSB sink block planting (#1, #3 Figure 7.1.1) were examined for damage at harvest and for presence of live FSB at monthly intervals (and before and after sprays). The mixed plant species hedge (#2 Figure 7.1.1) neighbouring both blocks was also examined weekly and live FSB collected. Releases of egg parasitoids (*Gryon* sp.) began in 2010/2011 to see if there was a difference in FSB numbers on the *M. paniculata* when parasitoids were present / absent.

7.2. Results

Climatic data comparisons with the FSB incidence patterns, crop damage levels confirmed anecdotal evidence of heavier *A. nitida* activity in macadamias during wetter seasons (Table 7.2.1). Data showed that simply managing the spring populations in the drier years 2003/2004, 2004/2005 and 2006/2007 (25% below average rainfall) gave effective control of FSB. However in wetter years (2005/2006 and 2007/2008) the possibility of re-infestation was much higher and the period requiring effective monitoring and management was extended. For macadamia growers in extended wet seasons a late spray (December/January) for FSB should be included to minimise the risk of late season damage. The only season where the standard treatment was inferior to unsprayed treatment was 2005/2006, (the only season when benzaldehyde lures were used to simulate late season flowering (Table 7.2.1, Figure 7.2.2)).

Monitoring data at CTH suggested that levels of FSB moving onto *M. paniculata* after each season followed a biennial pattern (Table 7.2.1).

The reproductive state of overwintering *Amblypelta nitida* (FSB)

Our work using field *M. paniculata* hedges suggested that FSB egg production is more tuned to host phenology than first realised. FSB will certainly progress through 1-2 generations during winter on fruiting *M. paniculata* in NSW (where cooler temperature and shorter days would be most noticeable) (Figure 7.2.1).

157 FSB were collected from the hedge during the volatile trapping experiment (12 weeks from 21/5/2008 until 31/7/2008). By instar the FSB were: I (0), II (10), III (23), IV (13), V (34), and adult FSB (77). No FSB were seen on the flight panels or collected in trap vessels during the 12 week trial, although many FSB were clearly visible on the surrounding fruit and foliage.

The local movement of *Amblypelta nitida*

FSB colonised all hedges that were planted on the CTH site for monitoring FSB from 2007. Within *M. paniculata* hedges on both hedges there was a strong FSB preference for particular plants (compared to the last 5 seasons when the FSB population was studied on a spatial level) (Table 7.2.2). Tree 8 in the highway hedge carried over 30% of the FSB population during that period, and Tree 15 in the arboretum hedge carried over 25% (Table 7.2.2). The FSB populations on the most favoured plants on these hedges were significantly different from the expected distributions (4.3% and 6.25% per plant respectively) (using chi squared test $pr<0.005$) (Table 7.2.2). Berries from these particular plants were used as seed stock for seedlings to become trap hedge plantings in the district.

The other interesting finding from this work was that after the older *M. paniculata* hedges were sprayed with systemic insecticides in November 2008, FSB did not colonise the plants as normal (Tables 7.2.2 and 7.2.3, Figures 7.2.1 and 7.2.2). Not only did FSB fail to establish on the hedges throughout 2009, but the survival rates were identical at 20% in both the treated an untreated *M. paniculata* berry feeding trials in May /June 2009. These data imply that recruitment onto the hedges, as seen in other mark recapture studies (Waite, 2000), is linked to an early colonisation in a fruiting season and FSB presence is due to some positive feedback either FSB, or FSB feeding related. The peak FSB activity period on the macadamia crop neighbouring the hedges appeared to be December to January each season which corresponded to damage in the final crop. However, oviposition in each crop definitely began at florescence each season (Figure 7.2.1, Table 7.2.3). In the unsprayed areas of the macadamia orchard, FSB populations were substantially higher on foliage (Table 7.2.3) and resulting crop damage levels were greater (Table 7.2.3, Figure 7.2.2). From 2007 onwards, FSB populations that built up in macadamia plots (especially unsprayed areas) (Table 7.2.3) were detected on neighbouring hosts. The floral sequence hedge had some encouraging early incidence data (Table 7.2.3) and the crop plants within the hedge (avocado and guava) showed significant damage (>50% in 2009/2010) (Table 7.2.4).

Statistical analysis of macadamia crop damage by FSB and impact on crop size is presented in chapter 6 (Tables 6.2.3.5 to 6.2.3.16). (For 2007 to 2010, confidence intervals were not included in the graph for clarity (Figure 7.2.2)). From analysis only varieties 246 and 849 had significantly more nut in the standard treatment in 2006/2007. All macadamia varieties bar A4 had lower FSB damage in the standard treatment. In 2007/2008 only variety 849 had significantly more nuts when standard sprays were applied, but all macadamia varieties were significantly cleaner in terms

of FSB damage. For the seasons 2008/2009, and 2009/2010 differences between the treatments for each macadamia variety were significant for both FSB damage and quantity of nut in shell produced per tree. The average levels of damage in the crop, and the crop size comparisons between unsprayed and standard spray regimes are shown in Figure 7.2.2. The average damage levels per tree increased with orchard age (up to 12 years). Varieties A4 and 849 were most consistently the high damage plots, and in 2005 and 2007 there was no real spraying effect (Figure 7.2.2, Table 7.2.2, and 7.2.3). Untreated plots within the trial also had an overall crop size reduction of which macadamia lace bug was partially responsible for the past two seasons (see Chapter 9).

Detection of FSB egg parasitoids was independent of the host trees. Parasitism was more efficient on *M. paniculata* hedges where the FSB densities were highest (Table 7.2.3). All parasitoid species (except *Ooencyrtus* sp.) were collected at CTH with *Gryon* sp. the most common parasitoid since 2008.

Table 7.2.1: Climatic variation measured at the weather station site for CTH Alstonville 2003-2010 for the period between August 1 of the year mentioned and March 31 the following year (243 days). Average *Amblypelta nitida* (FSB) damage levels (standard deviations) on the most susceptible macadamia variety (A4) and total FSB populations found on *Murraya paniculata* hedges.

Climatic Data	**Aug 03 – Mar 04**	**Aug 04 – Mar 05**	**Aug 05 – Mar 06**	**Aug 06 – Mar 07**	**Aug 07 – Mar 08**	**Aug 08 – Mar 09**	**Aug 09 – Mar 10**	**30 year mean**
Average daily maximum (℃)	25.5	25.2	25.5	24.6	24.5	24.8	26.3	**24.8**
Average daily minimum (℃)	17.0	16.5	17.5	16.4	16.7	16.3	17.0	**16.3**
Ground minimum (℃)	-0.2	-0.5	0.2	3.1	-1.0	-1.5	1.2	
Average FSB %damage on A4 Unsprayed trees (sd)	**27.0 (5.3)**	**2.1 (1.6)**	**17.6 (11.5)**	**9.8 (6.3)**	**37.2 (12.0)**	**32.3 (4.5)**	**62.1 (7.0)**	
Average FSB %damage on A4 Standard treatment block (sd)	**2.9 (5.1)**	**0.5 (1.1)**	**20.2* (15.0)**	**4.9 (6.9)**	**8.7 (7.1)**	**10.4 (2.7)**	**15.8 (6.1)**	
March FSB on *M. paniculata*	**25**	**3**	**38**	**1**	**44**	**0****	**14**	
April FSB on *M. paniculata*	**140**	**5**	**118**	**40**	**139**	**1****	**132**	
May FSB on *M. paniculata*	**343**	**1**	**182**	**65**	**198**	**2****	**187**	
Sun h Total (hours)	1927.5	1971.1	1845.5	1918.3	1560.3	1760.3	1936.5	**1829.4**
Pan Evaporation (Total mm)	1233.3	1212.4	1177.2	1147.1	1081.4	1138.8	1228.8	**1200.7**
Rainfall (Total mm)	**820.8**	**863.6**	**1319.8**	**884.8**	**1676.7**	**1212.9**	**987.0**	**1169.5**
Days >0mm	94	88	117	108	144	121	104	**104**
Days>0.5mm	71	74	90	77	117	99	75	
Days > 1.0mm	63	71	79	64	111	82	69	
Wet days during Aug/Sep	9	12	20	29	24	17	12	**17**
Sun hours during Aug/Sep	526.3	533.2	449.1	473.6	437.7	478.1	552.5	**481.6**

* Benzaldehyde lures used in all plots January to March 2006

** Hedges sprayed 11.11.08 to test if colonisation could be prevented in autumn, berries were suitable for complete development in laboratory feeding tests run in June 2009

Table 7.2.2: *Amblypelta nitida* numbers collected on individual plants within *Murraya paniculata* hedges at CTH Alstonville between 2005 and 2010.

Arboretum *Murraya paniculata* hedge								Highway *Murraya paniculata* hedge					
Year	2005	2006	2007	2008	2009**	2010		2005	2006	2007	2008	2009**	2010
Period	May-05 Feb-06	Mar-06 Jul-06	Nov-06 May-07	Aug-07 Apr-08	Jan-09 Dec-09	Jan-10 Jun-10		May-05 Feb-06	Mar-06 Jul-06	Nov-06 May-07	Aug-07 Apr-08	Jan-09 Dec-09	Jan-10 Jun-10
Tree #													
1	0	0	0	2	0	1		0	8	11	5	0	19
2	0	0	0	2	0	4		1	16	1	0	0	13
3	0	5	0	3	0	3		1	29	0	0	0	1
4	2	5	6	3	0	30		0	3	0	0	0	8
5	0	0	1	0	0	4		3	13	0	1	0	13
6	3	3	0	3	0	9		2	10	0	6	0	5
7	3	16	12	17	2	19		0	3	0	0	0	0
8	5	48	17	8	0	29		6	15	17	8	0	56
9	2	5	1	11	0	5		0	4	0	6	0	32
10	8	17	0	11	0	5		1	10	0	0	0	5
11	0	4	0	5	0	18		0	5	0	5	1	6
12	0	0	2	0	0	32		0	0	0	1	0	2
13	0	6	0	1	0	18		0	12	0	3	0	1
14	0	3	0	4	0	16		3	2	0	0	1	1
15	37	45	8	40	0	54		1	2	8	1	0	0
16	0	4	0	3	0	9		10	23	0	0	0	0
17	7	35	0	0	0	10							
18	10	13	0	1	0	14							
19	1	2	1	5	0	1							
20	0	2	0	3	0	8							
21	0	1	0	0	0	16							
22	0	0	0	3	1	11							
23	1	7	0	0	0	36							
Total FSB	79	221	48	125	3	352		28	155	37	36	2	162
FSB estimate per tree	3.4	9.6	2.1	5.4	0.1	15.3		1.8	9.7	2.3	2.3	0.1	10.1
X^2 (chi square)	398*	454*	211*	305*	30 n/s	251*		65*	102*	169*	52*	15 n/s	333*
d/f	22		X^2 Critical Value (pr 0.05) = 33.9					15		X^2 Critical Value (pr 0.05) = 25.0			

* Population significantly aggregated, most consistent trap trees shaded yellow and used for seed stock

** All trees sprayed November 2008 with systemic insecticides to test if infestation could be prevented.

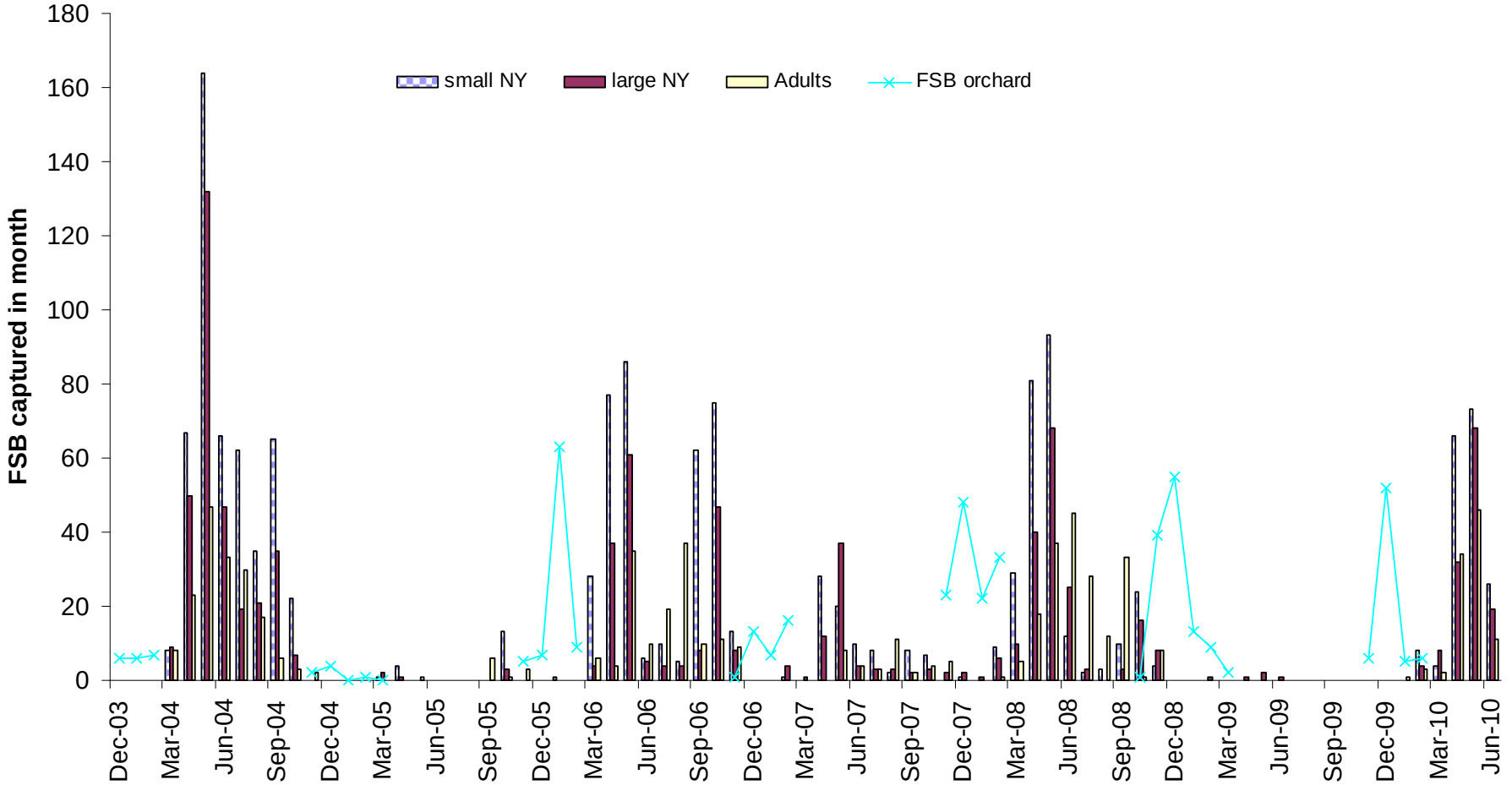

Figure: 7.2.1 Monthly total capture of *Amblypelta nitida* adults and nymphs (NY) showing the autumn/winter breeding cycle by on *Murraya paniculata* hedges at CTH Alstonville (in comparison with the levels in neighbouring macadamia orchards) during monitoring November to March each season.

Table 7.2.3: Monthly total captures of *Amblypelta nitida* (numbers of FSB collected) and egg parasitoids *Gryon* sp. (Scelionidae) and *Centrodora darwini* (Hymenoptera: Chalcidoidae) (numbers of parasitised eggs) at CTH Alstonville on a variety of hosts. ** Hedges sprayed with systemic insecticides

Monthly capture	Numbers of FSB seen in CTH Entomology macadamia orchard		Numbers of FSB collected on flower sequence hedge CTH Alstonville						Old hedges	Germplasm hedge area	Germplasm area	FSB egg parasitoid capture	
	unsprayed	STD	Avo	L64	CustA	Gua.	Long.	M.P.	M.P.	M.P.	Maca	Maca	M.P.
Nov 07	3	6											
Dec 07	16	5											
Jan 08	2	0							1				0
Feb 08	51	2							16				0
Mar 08									44				0
Apr 08									139				0
May 08	0	0							198				0
Jun 08									82			0	1
Jul 08									33			0	0
Aug 08									15			0	0
Sep 08									46			3	3
Oct 08	2	0							41			1	0
Nov 08	47	5							20**sprayed			0	0
Dec 08	51	7							0			0	0
Jan 09	8	2							0			0	
Feb 09	10	0							0	2		1	0
Mar 09									1				6
Apr 09									2				0
May 09									1				
Jun 09									0	0	3		
Jul 09								5	0				
Aug 09								7	0			0	0
Sep 09									0	0		0	0
Oct 09									0	0		0	0
Nov 09	8	0	2	2					0		46	0	0
Dec 09	32	7	1	1					0				
Jan10	3	0			2	1	0		1	1	0		
Feb 10	3	0				5	0	5	15	3			
Mar 10								7	14	18	0		5
Apr 10								25	132	20			15
May 10								15	261	42	0		4
Jun 10								5	56	32			0
Jul 10								2	42	17			
Aug 10				1					11	4	0		1

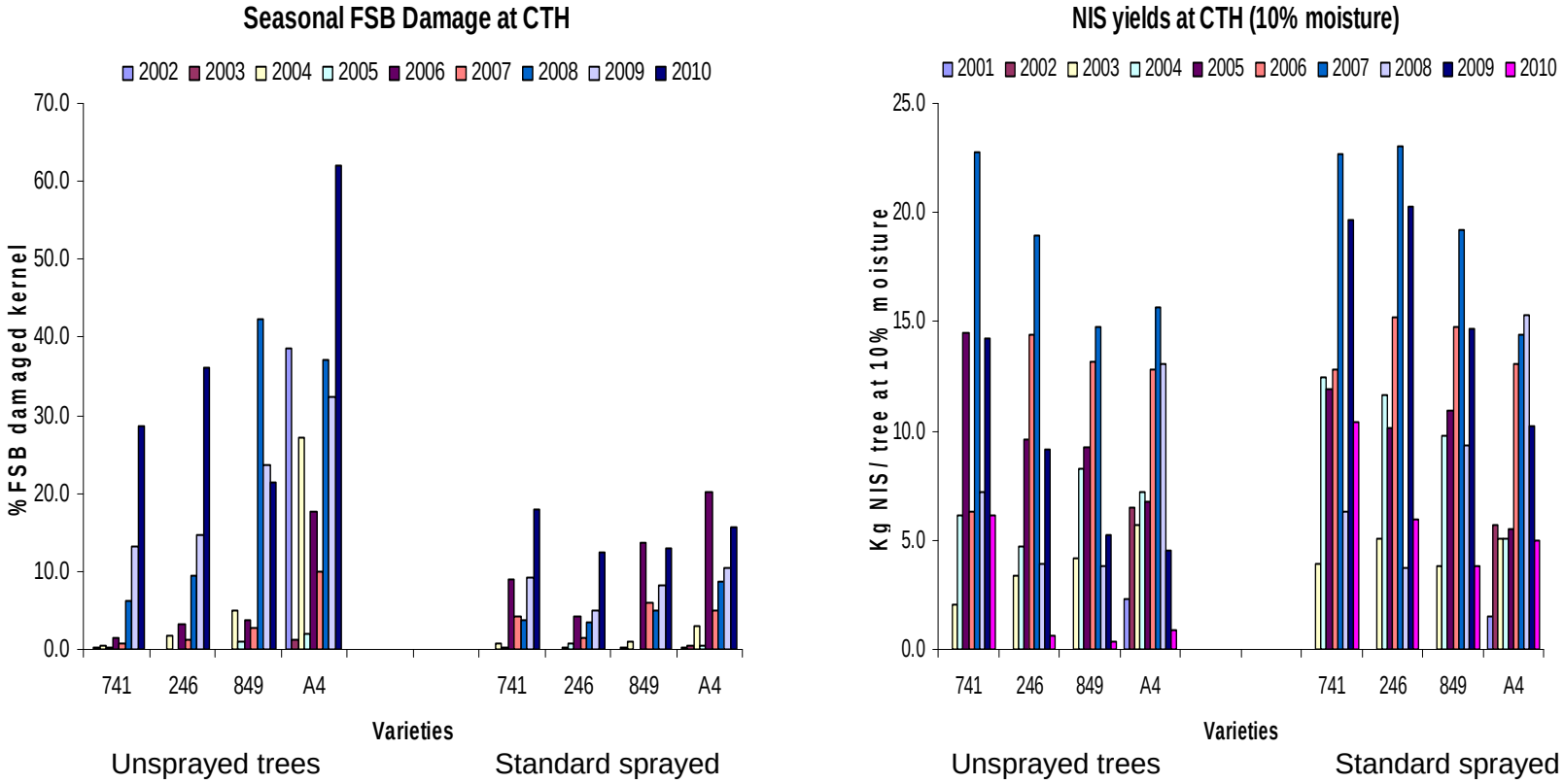

Figure 7.2.2: Two bar graphs showing seasonal impact of fruitspotting bug (FSB) on kernel quality (left) and total nut in shell yield (NIS) (right). In both charts the left hand columns represent the same unsprayed trees followed over the 10 year period and the right columns represent the standard sprayed treatments. All figures are average levels for 9 trees in each plot of each variety and all plots are buffered by a variety 246 ring of trees and pruning and fertiliser were identical for all trees in the study area.

Table 7.2.4: *Amblypelta nitida* damage to fruits from the sequential fruiting hedge crops at CTH Alstonville 2009/2010.

	Avocado Zuttano	**Avocado Zuttano**	**Avocado Pinkerton**	**Avocado Pinkerton**	**Coffee**	**Custard Apple**	**Guava**
	External	Internal	External	Internal			
Fruit count	34	34	44	44	34	34	50
FSB stings/fruit (average)	6.1	10.1	1.6	1.8	0	0.2	6.4
(Std Deviation)	5.5	8.7	1.8	3.0	0	0.7	7.6
#Fruit with>5 stings	17	24	4	5	0	0	29
%fruit with >5 damage	50	70.6	9.1	5	0	0	58
# Fruit with >10 stings	12	14	0	11.4	0	0	9
%fruit with >10 damage	35.3	41.2	0	11.4	0	0	18

7.3. Discussion

Amblypelta spp. have the capacity to feed on virtually any fruit. In macadamia and avocado there is strong evidence for varietal preferences with re-infestation occurring on specific host varieties well after fruit is fully developed (January to May). Waite, (2004) showed that FSB had a varietal preference in avocados, which the present study confirmed in macadamias.

This information may be useful in developing an improved pest monitoring system using hedge trap crops. Within the many macadamia variety plantings at CTH it was easy to find the more susceptible varieties. By FSB mapping various blocks in early spring (from 8-10m above the ground) FSB aggregations were noted and re-checked for FSB later in the year allowing the creation of a FSB sink block planting 2007 (#3 Figure 7.1.1). In doing this the macadamia varieties which FSB consistently favoured at the beginning of the season were identified as well the variety that carried the damaged nut in husk longest. FSB damage on the husk appears to give a signal back to re-invading FSB adults. A similar pattern of population incidence occurred in the *M. paniculata* study. When the FSB population was prevented (by a spring applications of broad spectrum insecticides) from marking the fruit early in the season (feeding/breeding) FSB did not recolonise the *M. paniculata* hedge later in the season.

Future work should quantify these observations with emphasis on measuring re-invasion of FSB onto specific plants after pesticide sprays. A practical outcome could be that a trap hedge could draw bugs out of an orchard and as well providing a monitoring strip in which incidence would precede a major FSB influx into an orchard. If the hedge was sited correctly then it could be used for FSB “trap and destroy” systems.

The benzaldehyde lure result confirmed a long suspected cause of treatment failure against FSB and late season attack. Benzaldehyde, which attracts FSB, is the main volatile component in both macadamia and *M. paniculata* flowers. Whenever macadamia crops fail to set fruit properly, the trees have capacity to compensate with out of season flowerings which may attract FSB. Thus, it is important to keep the flowering synchronised where possible or risk the need to calendar spray.

Work by the USDA with sorghum trap crops in cotton fields has shown that GVB parasitism rates of Tachinids like *Trichopoda pennipes* are enhanced by modifying the farmscape to generate a parasitoid source (Tillman, 2008). The FSB “hotspots” observed could be exploited in the same way. By knowing how quickly the pest can generate and where they are, FSB could be managed effectively with two insecticide applications sprays (on older Hawaiian macadamia varieties). The orchard manager should monitor the “hotspots” until there is an abundance of late instar nymphs, apply a cover spray throughout the whole orchard to remove all adults and large nymphs, then following up with another spray 3 weeks later to remove all the nymphs that hatched since the initial spray application. Such a spray regime would normally control a developing orchard FSB population, however, it may not effective when large populations develop on hosts outside the orchard and arrive as feeding adults on-mass (for this scenario, continual monitoring is required and pesticide spraying following each new invasion). On the sunshine coast in Queensland there is a large migration from hosts outside the orchards into avocado, passionfruit, custard apple, litchi and probably every other fruit crop orchard in the area.

From the results of this project, it is proposed that in high FSB pressure areas, to ensure sure invading adult FSB have a small site on each farm that is “actively calling in FSB ” throughout the year. If nectar was available FSB egg parasitoids could be directly maintained on such a planting. A trap hedge in macadamias, no more than 5-10m long, made up of 1-2 early flowering variety macadamias, 1-2 ribbonwood trees, 1-2 late flowering variety macadamias and 4-5 *M. paniculata* plants would achieve this purpose. In avocado, the hedge would vary slightly depending on which Hemipteran species was attracted. In NSW *A. nitida* to be the target and the macadamia hedge would include a Pinkerton avocado. In Queensland, there is also a need to control *A.l. lutescens* on shoots, plants like cassava and papaya would be added. From performance so far, the following conclusions can be made about trap crop hedges for FSB:

1) All selected trap hedge plants are terminal bearing. The trap hedge offers easy visual recognition of FSB activity and if kept below 3m the hedge allows easy FSB collection or spraying.

2) The trap hedge will provide a refuge for FSB egg parasitoids where parasitoids can build up and move into the crop. The trap hedge will allow us to quantify parasitism rates in FSB eggs and FSB population pressures more accurately.

3) The *Murraya* sp. hedges are ornamental and as such, have vastly different options for chemical control compared to horticultural food crops As an example, in November 2008 systemic insecticides were used to prevent FSB populations from establishing on *M. paniculata*. Very little FSB activity was observed on these *M. paniculata* from February through until the following May despite a large berry crop being present. Berries were collected in June 2009 to compare the FSB nymphal development and survival rates on the sprayed versus the unsprayed *Murraya* sp. and no difference was detected. Therefore the knockdown of initial expanding FSB populations was crucial to effective control. It also appeared that there was considerable variation in the *Murraya* spp. plants on site, with over 80% of FSB generated on 10% of the trees. These were the plants that fruited earliest and heaviest each year. These plants had the oldest nymphs present and females that laid into the budding florets (as happened with macadamia, litchi and avocado). The floral potency of the *M. paniculata* as a trap crop makes the trees useful in a hedge.

4) The FSB response to ribbonwood fruiting in December each year is quite different to that observed on other hedge plants. On ribbonwood there was a 2-3 week adult swarming phenomenon that could be used to target FSB for control.

8. Evaluating *Trichopoda giacomellii* as a potential biological control agent for *Amblypelta nitida* in Australia

The control of Hemipteran pest species has continued to rely heavily on broad spectrum insecticides since the end of World War 2. Various other strategies have been employed to improve the monitoring and augment the spraying programmes to manage these pests. These strategies include introductions of egg parasites, adult and nymphal parasites, predators, pheromone manipulation, vocalisation mimicry, genetic modification of host plants, and trap cropping. The green vegetable bug (GVB) has been the main pest investigated and is used as a representative species for Hemipteran management. GVB has shown remarkable resilience and adaptability to the different management techniques employed and the need for spraying still remains. Horticulture problems with FSB and BSB in tree crops are probably more severe than GVB because *Amblypelta* spp. are more difficult to monitor, are far more mobile and are true canopy insects (unlike GVB).

The problem with egg parasitoids is they always lag behind the invasive adult host pest population and significant damage can occur before pest numbers are brought in check (Knipling & McGuire, 1968). Beneficial insect control agents that can reduce the invading pest population are therefore desirable. Research projects have been carried out to identify such biological agents and so reduce the need for broad spectrum insecticide applications Spiders and insects such as the green tree ant *Oecophylla smaragdina* (Fabricius) (Hymenoptera: Formicidae) (Peng & Gibb, 2005) or coastal brown ant *Pheidole megacephala* (Fabricius) (Hymenoptera: Formicidae) (Ironside, 1981), assassin bug *Pristhesancus* sp. (Hemiptera: Reduviidae) (Waterhouse, 1998; Grundy & Maelzer, 2003) are known predators of Hemiptera. More recently, green lacewing *Mallada signata* (Schneider) Neuroptera: Chrysopidae) and *Chrysoperla rufilabris* (Burmeister) (Neuroptera: Chrysopidae) have been released for various lace bug, aphid and scale outbreaks (Papacheck, 2010 pers. comm.). Although relatively efficient at pest control green tree ant and coastal brown ant are particularly aggravating for farm workers pruning or harvesting crops and the ants predators need to be sprayed in order to pick the crop.

The Egyptian egg parasitoid *Trissolcus basalis* (Wollaston) (Hymenoptera: Scelionidae) was seen as the best candidate for biological control of GVB populations (Waterhouse, 1998). *T. basalis* was released into Western Australia in 1933 with great success in GVB control in general although performance on soybeans was notably below what is required. Soybean plants have many hairs on the leaves and stems that is thought reduces the wasps' capacity to search effectively (Waterhouse 1998). Summer survival of *T. basalis* is quoted as the limiting factor for good GVB control in Australia although *T. basalis* apparently parasitises GVB very well in the Ord River Area WA suggesting that temperature alone is not the key (Waterhouse & Norris, 1987; Coombs & Khan, 1998; Waterhouse, 1998; Sands & Coombs, 1999; Coombs & Sands, 2000).

Dipteran and Hymenopteran parasitoids have been collected from Hemipteran host pest species with Tachinid flies appearing to be the most effective group (Waterhouse, 1998). Tachinid parasitoids have been released in most regions of the globe for population management of common Hemipterous pests and mainly targeting GVB (Waterhouse, 1998). There were mixed results from numerous GVB trials including 4 attempts in Australia. There are 3 Tachinid species commonly used from North and South America;

these species are in the genus *Trichopoda* spp. (Diptera: Tachinidae)*, (T. pennipes* Fabricius*, T. pilipes* Fabricius and *T. giacomelli* (Blanchard)). An Ethiopian Tachinid *Bogosia antinorii* Rondani, which comes from the original area GVB range (Waterhouse, 1998), was also used. Australian native Tachinid species attack Hemipterans but these parasitoids are very rare in an orchard situation (parasitism by *Pentatomophaga bicincta* De Meijere in Bundaberg QLD on *A.l. lutescens*, has not been recorded since the 1950's (Ironside, 1981).

In Australia, entomologists tried unsuccessfully, to release *Trichopoda pennipes* and *T. pilipes* for GVB management in the 1940's, 1950's and again in the 1980's (Waterhouse, 1998). Releases of *T. pennipes* were also made during the 1950's in the south west Pacific island areas from Florida USA targeting *Amblypelta cocophaga* (coconut bug) in the Solomon Islands and Fiji but failed to establish (Waterhouse, 1998). In 1996 however, there were two well documented successful introductions of *T. pennipes,* one in California and another in Italy. The Californian introduction's key to success was to inundate a non target Coreid host population of *Anasa tristris* (squash bug) on nursery crops (Pickett *et al.,* 1996) with the parasite to build up parasite numbers in order to attack GVB on major vegetable crops. *T. pennipes* continues to register a 30% field parasitism rate of GVB (a major disease vector for cucurbits in California) (Decker & Yeargan, 2008). The accuracy of Tachinid taxonomy and therefore species distribution maps may be suspect given that earlier reports stated that *T. pennipes* attacked Pentatomid, Coreid, Pyrrhocorid and Largid bugs in California (Arnaud, 1978; Aldrich *et al.,* 2006). Field studies also suggested that 80% field parasitism of Coreid squash bugs from Massachusetts (Worthley, 1923), and 50% parasitism levels measured 7 years after release in Washington (Johansen, 1957) are possible. The Italian release of *T. pennipes* appears to be fortuitous and related to aircraft movements (Colazza *et al*., 1996). These examples suggest that *T. pennipes* is a far more generalist parasitoid than supposed and does take Coreid bugs as well as Pentatomids such as GVB. *T. pennipes* should therefore remain as an egg parasite insect of interest for FSB.

The Tachinid which Australian entomologists were able to successfully introduce was *Trichopoda giacomellii* from Argentina (Coombs & Khan; 1998; Coombs & Sands, 2000). *T. giacomellii* life cycle is shown in Table 8.1.1. In some parts of South America, field parasitism rates on GVB approach 100% (Buenos Aires) (Liljestrin, 1981) and up to 90% on some field crops in Argentina (La Porta, 1990). *T. giacomellii* also reduces fecundity of the adult female host by 70% and GVB male and immature female death normally occurs 2-4 days after the parasitoid emerges, fully mature GVB females normally live for another two weeks (Coombs & Khan, 1998). *T. pennipes* apparently kills its host more quickly than *T. giacomellii* (1 day after parasite emergence) (Shahjahn, 1968).

Table 8.1.1: Life cycle and fecundity of *Trichopoda giacomellii* reared on *Nezara viridula* at 26℃ 70% Relative Humidity and Light: Dark 16:8 from Coombs 1997.

Life stage	Time (days)
Egg	2.8
Larvae	13.0
Pupa	13.3
Adults + water and raisins	9.6
Adults + water	3.2
Fecundity	**Eggs laid**
Eggs per female+ water and raisins	163.1
Eggs per female	34.9

8.1. Materials and Methods

The aim of this study was to test the capacity of *T. giacomellii* to oviposit on FSB, complete development and kill the host. From March 2010 sufficient *T. giacomellii* parasitised GVB were located to investigate whether it could play a role in the management of FSB in the short and longer term. Adult *T. giacomellii* were introduced to GVB, FSB, BSB adults and late instar nymphs to see if the fly oviposition would occur. *T. giacomellii* eggs were also transplanted, and eggs were attached to surrogate FSB to test if *T. giacomellii* Tachinid development was possible in FSB.

Colony Establishment

With the assistance of Peter Entwistle (Horticulturalist) *T. giacomellii* infested GVB were collected from two sites on the Richmond River valley to establish a colony. *T. giacomellii* eggs were visible on GVB adults and nymphs. GVB were collected from Budda pea plant (*Aeschynomene indica)* at Coraki NSW, and black nightshade (*Solanum nigrum*) at Casino NSW.

Colony maintenance

All parasitised GVB adults were maintained in a 4 litre plastic container with a ventilated lid and were feed on fresh beans and corn. The containers were examined each week for fly pupal emergence. Dead GVB were examined to determine the level of parasitoid egg survival. Adult and about-to-eclose parasitoids (see Figure 8.1.1) were kept in a

Perspex fronted field cage 40cm x 40cm x 20cm with 1mm mesh gauze (see Figure 8.1.2. A crushed sugar cube and slivered raisins were placed in a suspended tray near a wet sponge cloth to nourish the adult flies. Pupae were kept on Petri dishes allowing them to emerge and fly directly into the cage. Adult GVB (up to 25 at a time) and sometimes adult and nymph FSB (up to 15) or BSB (up to 5) were also kept in the flight cage on green beans and corn and fruiting *Solanum nigrum* (Solanaceae) cuttings (changed weekly). Fly oviposition, feeding and mating was observed and photographed.

Weekly schedule for rearing Trichopoda giacomellii

1. Parasitised GVB adults were checked for mortality and emerging fly pupae. All dead GVB with parasitoid eggs were transferred into a date labelled Petri dish. All new Tachinid pupae were collected into a separate Petri dish and the total recorded. Old food was removed and replaced, all remaining live GVB adults were returned to the clean container.

2. In the main flight cage, live Tachinids were collected into vials, dead ones removed and the total numbers recorded (see Figure 8.1.1 and 8.1.2).

3. All GVB in the cage were collected live and dead. Individuals were examined for eggs and numbers recorded. All live GVB with egg parasitoids attached were transferred into the 4 litre container and all dead bugs were transferred to a Petri dish and checked for parasitoid emergence.

4. Old beans, corn and nightshade were emptied from cage. All fresh GVB eggs laid were transferred to the GVB colony.

5. The blotting paper floor of the cage, water sponge, sugar cube and raisins were replaced.

6. Unparasitised GVB (up to 20) and *Amblypelta* spp. and green stink bug *Plautia affinis* (Dallas) (Hemiptera: Pentatomidae) were re-introduced if possible to the cages.

7. Live Tachinid adults were then re-introduced into the flight cage.

Trichopoda giacomellii - Egg transplant experiments

With parasitoid cultures correct host population density is crucial to culture success. Host density needed to be altered in the confined colony cage as the Tachinids tended to lay multiple eggs on individual hosts (maximum 80+) with very little chance of any of the Tachinid eggs reaching maturity. The solution used in these experiments was to remove eggs from hosts to increase the chances of pupal survival and development.

Figure 8.1.1:

Top left: The Tachinid adult *Trichopoda giacomellii* bred from *Nezara viridula* adults at CTH Alstonville 2010 (male fly above, female below) (12x magnification).

Top right: The Tachinid adult *Trichopoda giacomellii* emerging from pupal cases that have come out of *Nezara viridula* adults (12x magnification).

Bottom left: The egg of the Tachinid *Trichopoda giacomellii* laid above the eye of a *Nezara viridula* adult (50x magnification).

Bottom right: The collapsed egg and entry point through the exoskeleton, near the eye of a *Nezara viridula* adult, made by the larva of the Tachinid *Trichopoda giacomellii* larva (50x magnification).

Figure 8.1.2:

Top left: Breeding cage used to house the Tachinid adults *Trichopoda giacomellii* with up to 25 *Nezara viridula* adults and 4-10 *Amblypelta nitida* at CTH Alstonville 2010.

Top right: Weekly removal of Tachinid fly *Trichopoda giacomellii* eggs from *Nezara viridula* adults using water and fine probes (12x magnification). Eggs were transplanted onto other hosts.

Bottom left: Eggs of the Tachinid *Trichopoda giacomellii* showing the different stages of development after removal from host *Nezara viridula.* (The left egg is beginning to develop the larval spiral drill to penetrate the host on hatching, the lower egg is hatched and the larva has left the egg, the top egg is freshly laid and the in central egg the larva is still in the egg but unsuccessfully attempted to enter the host. Fresh eggs and those still with the spiral forming can be transplanted to other hosts successfully) (50x magnification).

Bottom right: The entry point through the exoskeleton of a transplanted Tachinid fly *Trichopoda giacomellii* larva into a female *Amblypelta nitida* adult (30x magnification).

GVB were left with a single Tachinid egg after a 1 week exposure to avoid superparasitism. Remaining removed eggs were used for testing development on alternate hosts.

Egg transplantation was done under 20x magnification using cryolysers and fine probes to extract and replant the egg onto the dorsal thoracic or abdominal plates of a new host (where it would be difficult for the new host to remove the egg) (see Figure 8.1.2). FSB adults, with Tachinid eggs attached, were maintained in a separate 4 litre container and examined each week for survival of FSB. All FSB bodies were kept for three weeks to ensure the emergence of live parasitoids then remaining eggs were scraped off and examined for egg development and evidence of parasitoid invasion of the host.

8.2. Results

The field parasitism rates of the *Trichopoda giacomellii* two sites are shown in table 8.2.1. Levels around 12% were detected at Coraki (Budda pea plant *Aeschynomene indica*) and over 50% on black nightshade (*Solanum nigrum*) in at Casino in NSW.

Table 8.2.1: *Nezara viridula* field collections from 2010 from Budda pea plants at Coraki NSW and Black Nightshade weeds

Date Collected	No of large Nymphs	No. Large Nymphs Parasitised	No, Adults unparasitised	No. Adults Parasitised	Field % Parasitism
25/3/2010	111	15	47	11	14%
6/4/2010	142	13	28	5	10%
13/12/2010*	1	0	22	28	55%

* Within tea tree plantations at Casino NSW. All insects collected by hand with jars directly from host plants.

The Tachinid adults tended to attack the same GVB many times, which did not necessarily lead to a successful pupal emergence. However, after the removal of excess eggs from the hosts (leaving 1-2 eggs per GVB), an overall parasitism rate of 63% was achieved and produced a live Tachinid adults (Table 8.2.2). Very few adult GVB survived attack,(0-10%) From hosts that had eggs removed, from 77% of removed eggs larvae had already entered the host (Table 8.2.3). The average level of fresh eggs available for transplant each week was around 6% (Table 8.2.3) and a further 3% had spiral development but could still be used for transplanting.

Table 8.2.2: Weekly population levels of *Trichopoda giacomellii* at CTH Alstonville laboratory when caged with *Nezara viridula.*

Date Examined	Target number of GVB adults	Number of parasitoid eggs laid	Number of live flies in cage	Number of dead flies in cage	Number of live fly pupae
30/3/2010			0	0	5
8/4/2010			2	0	5
16/4/2010	22	184	6	0	2
23/4/2010	25	132	5	2	8
27/4/2010	22	295	6	1	10
3/5/2010	22	329	7	2	10
10/5/2010	18	258	8	1	8
17/5/2010	13	244	5	3	8
24/5/2010	26	214	2	5	2
31/5/2010	8	5	2	0	10
7/6/2010	11	4	0	2	12
16/6/2010	20	50	5	0	8
21/6/2010	14	39	1	5	8
28/6/2010	7	7	2	0	7
5/7/2010	11	0	1	1	13
11/7/2010	9	2	1	0	14
19/7/2010	9	0	1	1	13
26/7/2010	9	0	2	2	9
2/8/2010	17	94	3	0	7
9/8/2010	19	45	1	2	7
16/8/2010	5	0	0	0	4
23/8/2010	1	0	0	0	7
31/8/2010	1	0	1	0	10
7/9/2010	1	0			
13/9/2010	6	38	3	0	8
20/9/2010	11	47	2	2	7
28/9/2010	4	0	0	2	7
20/12/2010*			0	0	3
27/12/2010*			0	0	10

* New colony started from parasitised GVB at Casino field source on black nightshade plants

Table 8.2.3: Level of development of the *Trichopoda giacomellii* eggs examined from the bodies of adult *Nezara viridula* from CTH Alstonville laboratory colony

Date	Number of eggs examined	Number freshly laid eggs	Number of eggs with spiral inside	Number of eggs with emerged maggot	% Fresh	% Emerged
6/4/2010	5	0	0	5	0	100
27/4/2010	77	0	3	67	0	87
3/5/2010	35	0	2	24	0	69
10/5/2010	55	0	3	49	0	89
17/5/2010	35	0	1	31	0	89
24/5/2010	142	0	5	104	0	73
31/5/2010	26	0	1	23	0	88
3/6/2010	28	1	0	24	4	86
16/6/2010	4	0	0	4	0	100
28/6/2010	2	0	0	1	0	50
5/7/2010	8	1	1	5	12	63
11/7/2010	13	4	1	7	30	59
19/7/2010	17	1	0	10	6	59
2/8/2010	3	0	0	2	0	67
9/8/2010	1	1	0	0	100	0
16/8/2010	27	6	1	20	22	74
23/8/2010	4	4	0	0	100	0
31/8/2010	3	0	1	2	0	66
27/9/2010	1	0	0	1	0	100
5/10/2010	7	1	0	5	14	71
18/10/2010	13	0	0	9	0	69
Totals	507	19	19	393		
	For n>10			Average	6%	77%

Table 8.2.4: Level of development of transplanted *Trichopoda giacomellii* eggs examined from the bodies of adult and nymph *Amblypelta nitida* from CTH Alstonville laboratory.

Date	Number of eggs examined	Number freshly laid eggs	Number of eggs with spiral inside	Number of eggs with emerged maggot	% Fresh	% emerged
27/4/2010	1*	0	0	1*	0	100
15/5/2010	12	0	8	4	0	33
24/5/2010	21	0	4	10	0	48
31/5/2010	12	0	3	7	0	58
7/6/2010	1*	0	0	1*	0	100
28/6/2010	1	0	0	0	0	0
5/7/2010	1	1	0	0	100	0
9/8/2010	7	1	1	1	14	14
Totals	56	2	16	24		
				Average	14%	43%

(*)These were naturally laid *T. giacomellii* eggs on two adult *Amblypelta nitida* held in cages with *Nezara viridula* and adult *Trichopoda giacomellii* that pupated but failed to eclose.

When eggs were transplanted to FSB adults and late instar nymphs the effect was fatal if the larva was able to penetrate the new host. An average of 43% of eggs attached produced larvae that entered the new host (Table 8.2.4). In two instances adult *T. giacomellii* laid single eggs directly onto live FSB in the cage, larvae developed to pupation, however, adult eclosion failed (Table 8.2.4).

8.3. Discussion

Observations of the hunting and oviposition behaviour exhibited by adult *T. giacomellii* suggest that it selects oviposition area by shape recognition selection as *T. giacomellii* straddles a potential new host. *T. giacomellii* is able to locate a host on foliage or on cage walls (in the cage it probably done visually due to proximity). Other species of Tachinids are known to follow pest semiochemicals (Aldrich, 2006). The Tachinid adult rapidly swoops over the potential host grappling the host body with its tarsi and swinging its abdomen onto the host body to deposit one or more eggs. The eggs are normally laid on the thoracic margins dorsal or ventral and along ventral sternites of the abdomen margin.

FSB is a longer, thinner Hemipteran and is generally more mobile than GVB (GVB are easily caught in the cage by the Tachinid fly). FSB were caught and straddled readily by the caged Tachinids but oviposition did not normally occur. Other smaller Pentatomids with a similar body width to FSB (lesser green stink bugs), however, seemed to be acceptable as hosts, multiple ovipositions occurred on the thorax. These Hemipterans were accidentally introduced on the nightshade cuttings. It is debatable whether the *T. giacomellii* would complete larval development in those smaller hosts although they did kill the host.

Another surprising observation was the predatory behaviour of some adult GVB when they gained access to the Petri dish containing the fresh *T. giacomellii* pupae. GVB fed through the pupal case eliminating the developing parasitoid. The smaller FSB generated *T. giacomelli* pupae did not emerge may have been killed by feeding GVB.

From this work we concluded that *T. giacomellii* will not naturally parasitise FSB without some change in host preference. The positive aspect from this work was that the *T. giacomellii* larvae kill the FSB when eggs were transferred. *T. giacomellii* pupal eclosion is affected by many variables but eclosion from FSB hosts was witnessed twice and complete development maybe possible. The question is, whether this could lead to *T. giacomellii* with a modified host preference for FSB. It is highly desirable to augment biological control with parasitoids that attack migrating FSB adults as well as FSB eggs. The results from the CTH field trials have shown that the FSB egg parasitoid *Centrodora darwini* released into the plots at 4-6 wasps/tree, on the expanding October FSB population may reduce FSB numbers but will not prevent migrating adult feeding (Chapter 6). However, until an adult parasitoid is found that can be used from spring-autumn in Macadamia orchards, some pesticide spraying will remain necessary to control re-entering FSB. It is recommended that research continue to investigate native Tachinid? species on Coreids (e.g. *Myctis* sp., *Amblypelta* sp.) and re-investigate successful introductions of *T. pennipes*.

An alternative approach, which was used successfully to encourage the adoption of MNB egg parasitoid use, is to enhance pest population in the field to establish parasitoids in an orchard or in a small pest refuge area. Parasitoids can be released onto key overwintering hosts which will reduce the pest populations moving into orchards (e.g. *T. crytophlebiae* releases into mangroves during winter; see chapters 6, 7 and 11). Trap hedges, proposed for orchards in higher pressure areas, offer such an opportunity (if plants can be found that have greater attraction for the FSB than the crop). Such trap plants exist for macadamia and this knowledge will be tested, in the future, in avocado orchards to compare the behaviour of FSB. If FSB were allowed to establish in the hedge early in a season, trap plant use as a monitoring tool alone could help define spray timing, allow beneficial insect introductions and reduce the risk of re-infestation (see chapter 7).

New spray options, identified in this project, for FSB management will need residue studies and further optimal rate studies in the near future to progress product availability to growers. Older chemistries, which may be used in the short term, may not be as effective as endosulfan, which is now de-registered. Any decision by the Australian Pesticides and Veterinary Medcines Authority (APVMA) on what pesticides are available for use and what spray boundaries apply will impact greatly on future orchard spraying strategies. The macadamia industry needs to be aware of the consequence of large

areas of unsprayed macadamias that can harbour pests (especially macadamia lace bug and FSB) surrounding a commercial crop.

In 2009/2010 when FSB egg parasitoids were not released there were much higher damage levels and lower crop yields in unsprayed control plots. The cause of this steady decrease in crop yield was the emergence of macadamia lace bug as a pest within these unsprayed areas (chapter 9). A secondary benefit of spray applications in spray treatment areas was a reduction in lace bug impact. As endosulfan is no longer available, the potentially useful product Bayer 092 needs further evaluation as part of the registration process. This product also needs to be tested on lace bug infested plants to see what impact this new insecticide has on lace bug populations and the resulting nut set and final crop size (chapter 6). The increasing pressure of macadamia lace bugs is forcing some pre-emptive pesticides to be applied to set a crop.

9. Impact of macadamia lace bug species (Tingidae) on the macadamia crop, management implications, and insecticide assays

The original accounts of macadamia lace bug *Ulonemia concava* Drake (Hemiptera: Tingidae) (see Figure 9.1.1) suggested it could cause heavy damage to florescence but was rarely a major pest on macadamia (Ironside, 1981; QDPI, 2003). However, evidence is growing that macadamia lace bugs (Family Tingidae) are far more significant as a pest than originally suspected. Anecdotally the lace bug is more prevalent in northern NSW, especially in older closed canopy orchards, or near rainforest remnants. In South East Queensland lace bug is not an issue and this is probably due to flower caterpillar management with endosulfan or methidathion coincidently controlling lace bug. On the Atherton tablelands there may be lace bug activity and it has also been reported from the southern limits of NSW macadamia orchards around Nambucca Heads. There is virtually no data published on macadamia lace bug and in this project attempts were made to quantify the reproductive rate, behavioural traits, impact on crop load and the efficacy of various control options.

Discussions with crop consultants working on organic orchards point directly to lace bug more than any other pest as a production limiter. Correlation between light penetration into the macadamia canopy and reduced lace bug activity was suggested by several consultants (Shaun Stead, Dave Forrest, Phil McCarthy, and Alan Coates pers. com). Accordingly, this project compares unpruned with pruned treatments to test the hypothesis: that light could be a useful cultural management tool (Figure 9.1.5). We have also noted that the macadamia lace bug breeds when the tree flowers then moves onto the branches and potential new fruiting wood which are exposed to high adult populations between flowerings. A previous research project MC 05005 (Huwer, *et. al.* 2006) detected lace bug in unsprayed treatment plots of CTH Alstonville research trials and there was a reduction in crop yield of over 40% for the most productive variety (741). Since 2006 macadamia lace bug has flourished in all the unsprayed macadamia plots at CTH and this project has enabled assessment of impact more thoroughly.

A second lace bug species has been detected in NSW macadamia orchards, complicating the issue of lace bug management (from mid 2008 see Figure 9.1.2). The more recently discovered unnamed species of macadamia lace bug has been far more common than *Ulonemia concava* in all our trial work at CTH Alstonville. The new lace bug species was identified as *Physatochelia* sp. (NSW DPI taxonomy) and also examined by Dr Melinda Moir (Melbourne University). Taxonomy is being revised for the whole family of Tingidae and DNA marker work could assist in resolving the taxonomy.

9.1. Materials and Methods

Live adult lace bugs were collected July to Oct 2009 from infested sites in the Northern Rivers district and brought back to the insectaries at CTH Alstonville (Figures 9.1.3 and 9.1.4). The macadamia lace bugs were sexed under high magnification (20-40x) (see Figure 9.1.4) and allowed to breed in 300ml polystyrene containers held at room temperature of 25^{o}C on clean emerging macadamia florets provisioned every 2-3 days. Each floret was labelled by date of exposure to the macadamia lace bug adults and

nymphs were monitored until they had completed development. Examination of the florets every 2 days allowed the estimation of nymphal life stages and total nymphal development time, and oviposition sites (see Figure 9.1.1, 9.1.2).

Reproductive rates of both species of macadamia lace bugs were compared at two levels in the canopy of mature trees at CTH Alstonville. Breeding pairs were released into labelled fine gauze (0.2mm) bags on clean florets 3-4m above ground in low light areas (lower canopy) and 6-9m above the ground on well exposed racemes (upper canopy) 18/8/2009. Racemes were cut from the tree 24/8/2009 and number of nymphs counted the following day under 20-40x magnification.

It had been established that macadamia lace bug breeds on macadamia flowers, but nothing was known about how populations were maintained or continued between flowering events. We therefore conducted an incidence study of macadamia lace bug in an orchard at CTH with high pest pressure. Neighbouring trees within the orchard were sprayed with endosulfan at monthly intervals after the first flowering in spring until the appearance of the following florescence. Two hours after the insecticide application fallen insects were collected from tarpaulins under sprayed trees and counted (Figure 9.1.5).

With the removal of endosulfan as a control for lace bug candidate replacement pesticides were considered a priority. Products examined were Lepidex® (trichlorfon), Success® (spinosad) 5 new Bayer coded products (DC083, DC084, DC091, DC092, DC093), DuPont HGW086 and numerous unbranded oil and organic products suggested by growers. These insecticides were compared with a positive control, the pyrethroid beta-cyfluthrin (Bulldock ®).

The spray trials were conducted at three levels and in three arenas:

1. Preliminary trials were modelled on work done in 2005 where clean racemes were dipped in insecticide formulations, allowed to dry for 2 hours then placed in small vases and 3 live adult lace bugs were placed on each raceme. A total of 9 adults were tested per treatment originally and test populations of >20 insects per treatment were used when possible in following assays. Racemes were caged under inverted glass jars and mortality checked after 24 and 48 h.

   Subsequent bioassays in the laboratory involved field collection of infested stage 3-5 racemes, placing the racemes in small flasks with water and spraying test formulations directly onto the racemes (2mls). The sprays were applied with 500ml hand misters to run-off and raceme flasks were placed on metal lids with white filter paper inserts to collect dead lace bug adults and nymphs. Bioassays were caged in labelled Fowlers glass Vacola jars (800ml) to prevent adults flying off.

2. Three replicates for each treatment were done and mortality was assessed under a stereomicroscope, initially at 3 days but then 1 and 2 days because the florets become desiccated after 3 days. Adult death was determined by a lack of leg or antennal movement when prodded or a withered body for nymphs.

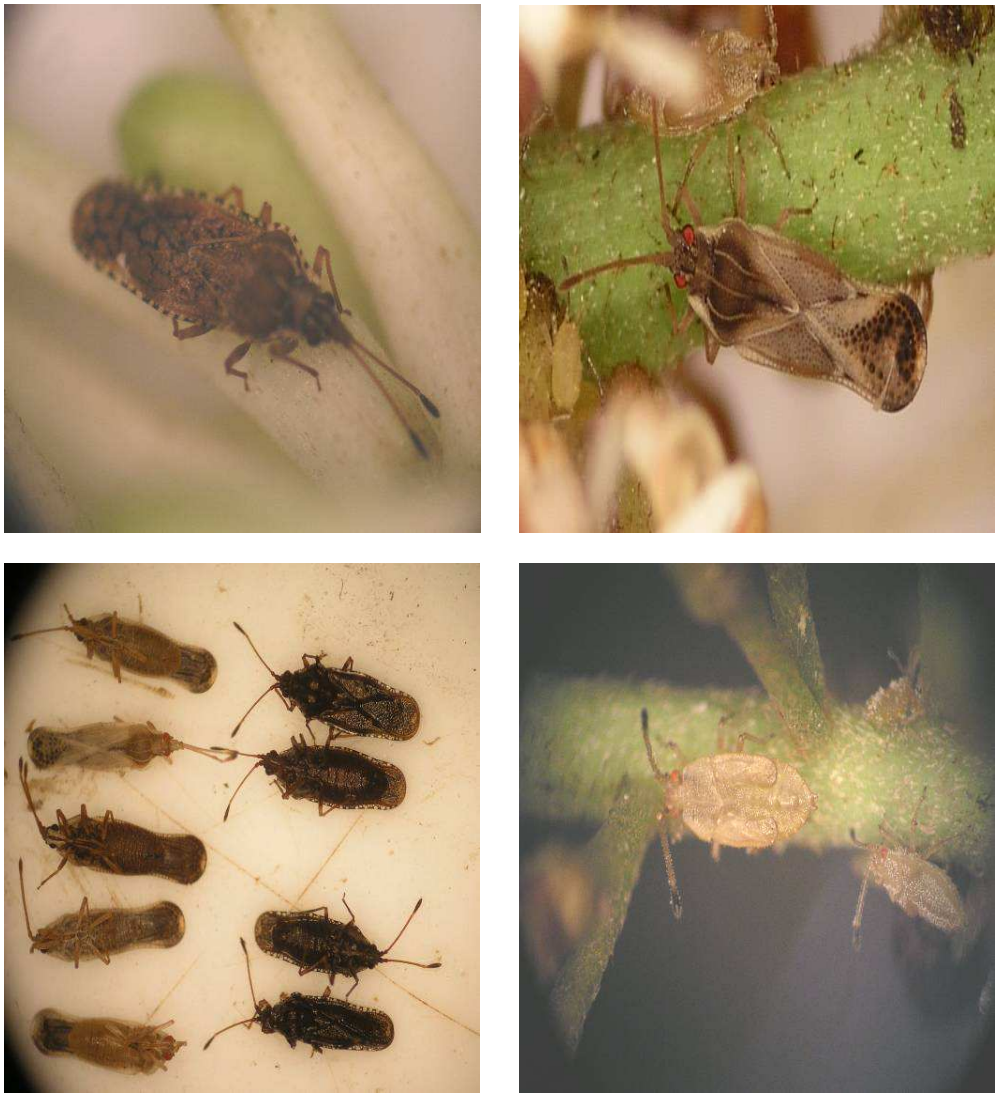

Figure 9.1.1:

Top left: Macadamia lace bug *Ulonemia concava* collected in Rous area 2009, (35x magnification) (see Ironside, 1981 p22).

Top right: The more common macadamia lace bug in Northern NSW orchards labelled as *Physatochelia* sp. (NSW DPI taxonomy branch ID (35x magnification), with second and 5th instar nymphs collected at CTH Alstonville 2007 on macadamia florescence.

Bottom left: Dorsal and ventral comparison of the two species of adult lace bug (25x magnification).

Bottom right: Wing bud variation between 4th and 5th instar macadamia lace bug (*Physatochelia* sp.) nymphs (35x magnification).

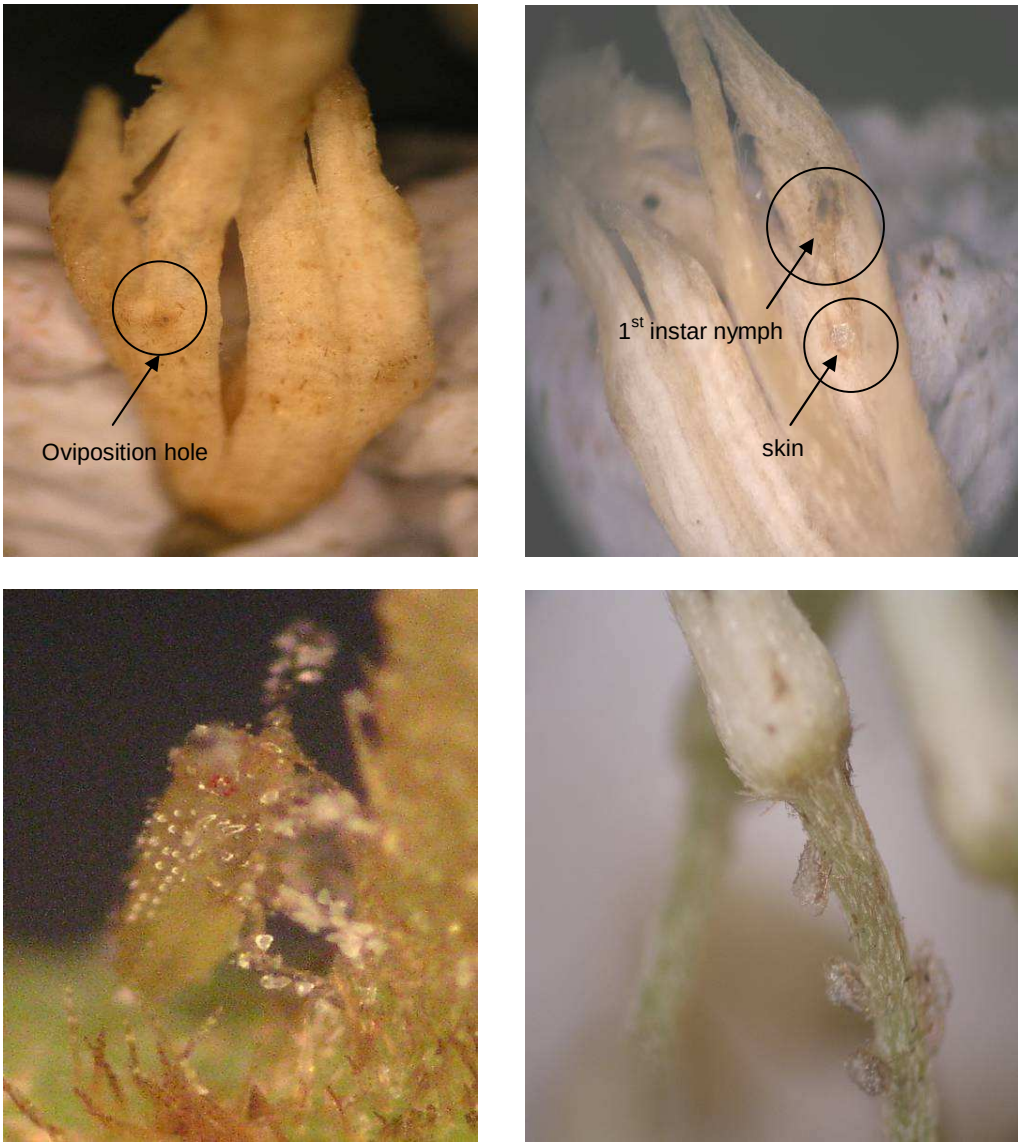

Figure 9.1.2:

Top left: An external oviposition entry hole on the left facing petal of a macadamia floret is the only evidence of macadamia lace bug *Physatochelia* sp. (50x magnification)

Top right: The empty egg case and freshly emerged first instar macadamia lace bug *Physatochelia* sp. on the interior wall of a macadamia floret (50x magnification).

Bottom left: 1st Instar *Physatochelia* sp. nymphs covered in macadamia pollen grains moving up the petiole of a macadamia floret (50x magnification).

Bottom right: *Physatochelia* sp. old nymph cast skins remaining on the macadamia floret petioles. Cast skins are a good activity indicator for macadamia lace bug (*Physatochelia* sp.) in the tree for crop scouts (12x magnification).

Figure 9.1.3:

Top left: Classic floret damage due to macadamia lace bug *Physatochelia* sp. spring 2007 CTH Alstonville.

Top right: The 5 tank spray unit built for spray trials at CTH Alstonville and nearby orchards. The unit slides on to a vehicle tray back and the 12 V DC pump drives the high volume application.

Bottom left: Trial at Doust farm Tuckombil NSW (The site of first spray trials on the *Physatochelia* sp. in September 2007) with the tagged racemes and raceme collection gear.

Bottom right: Second field spray trial site with tagged racemes at CTH Alstonville in the "Accession block" (September 2007) with 9m high canopy and constant lace bug activity.

Figure 9.1.4

Top left: Increasing floret damage due to macadamia lace bug *Physatochelia* sp. CTH Alstonville from left to right September 2009.

Top right: Flower dieback on racemes (macadamia variety A4, September 2009, Rous and CTH Alstonville) thought to be related to water stress but superficially similar in appearance to lace bug damage,

Bottom left: Sexual dimorphism in native Tingid species. The photo shows the ventral view with male specimens in top row and females in the bottom row. Species are olive lace bug (*Froggattia olivinia*) far left, *Ulonemia concava* centre and *Physatochelia* sp. on the right (10x magnification).

Bottom right: Sexual dimorphism in native Tingid species. The photo shows the dorsal view with male specimens in top row and females in the bottom row. Species are olive lace bug (*Froggattia olivinia*) far left, *Ulonemia concava* centre and *Physatochelia* sp. on the right (10x magnification).

Figure 9.1.5:

Top left: Increasing light penetration by opening 1m space between trees as well as along the rows to reduce the impact of macadamia lace bug *Physatochelia* sp. (CTH Alstonville Unsprayed Entomology block sections August 2008.)

Top right: Light penetration between trees shown along the rows at CTH Alstonville.

Bottom left: A beating tray and brush are used to extract and collect adult lace bugs from trunks of infested trees (October-November at CTH Alstonville) showing the carry over potential of *Physatochelia* sp. in unsprayed orchards.

Bottom right: Brown lace wing nymph attacking *Physatochelia* sp. populations in the field (25x magnification).

3. Small field trials on private farms or at CTH Alstonville were sprayed using a 5x 80L tank spray unit on the back of the 4WD (see Figure 9.1.3). Trial trees were selected on the presence of lace bugs on the flowers and tagged with flagging tape (Figure 9.1.3). Trees up to 6m high required a spray volume of 5L per tree. Spray coverage in the top of these trees was not possible so mortality was only compared on the racemes sprayed. Individual racemes were collected after 3-7 days. Racemes were cut from the tree then placed in labelled bags and examined later under the microscope. Survival rates of macadamia lace bugs on the racemes were counted as previously described. Other predatory or parasitic life forms associated with macadamia lace bugs were also noted (Figure 9.1.5).

4. The long-term yield loss trial which compared unsprayed areas with standard insecticides treatments estimated the damage on florescence in each plot. In this trial the number of lace bug damaged florets visible in the lower canopy (below 2m from the ground) were scored for each tree and compared that with the final seasonal yields. Sprays (5 litre per tree) were applied with a tractor mounted and PTO driven high volume air blast sprayer. Standard spray application dates for 2009 are listed (Chapter 5) and approximates commercial farm practices in the district.

9.2. Results

Macadamia lace bug biology

From development studies the Macadamia lace bug *Ulonemia* sp. appeared to complete its life cycle in 16+/-3 days (Table 9.2.1). There are 5 nymphal instars and the eggs are laid within the macadamia florets by insertion of the female adult ovipositor. The oviposition wound is almost undetectable and the first instar nymph emerges from the floret encrusted with pollen and begins feeding along the raceme (Figures 9.1.1 and 9.1.2).

Table 9.2.1: Summary of Life cycle stage durations for macadamia lace bug under laboratory conditions 25 +/- 3^{o} C and 50% Relative humidity

Life cycle stage	Duration (days)	Distinguishing features
Egg	1-2	Within opening floret
1st Instar	1-2	Pollen grains attached to newly emerged nymphs which have low mobility .
2nd Instar	2	
3rd Instar	2-3	
4th Instar	3-4	Wing bud started
5th Instar	5-6	Full wing buds
Full development	Average = 16	Emergence Range was 10 to 22 days

Table 9.2.2: Development rates of macadamia lace bug *Physatochelia* sp. under laboratory conditions at CTH Alstonville September 2009. The colony was maintained at 25 +/- 3^{o} C and 50% Relative humidity. Breeding pair placed on each raceme into 1 container on 7/9/2009. Data presented as numbers of individual bugs per life stage per raceme. Nomenclature used for nymphal stages roman I= 1st instar, II= 2nd, III= 3rd, IV=4th and V=5th and ad =adult emerged.

Date checked	Raceme 1	Raceme 2	Raceme 3	Raceme 4	Raceme 5	Raceme 6	Raceme 7	Raceme 8	Raceme 9	Raceme 10
10/9/09	2ad	2ad	2ad	1ad	2ad	2ad	2ad	1ad	2ad	2ad
11/9/09	2ad	1ad, 9 I	2ad	1ad,19II, 2I	2ad, 3I,4II	2ad,3II,2IV	2ad,3IV, 3V	1ad,2I	2ad	2ad, 1III
Adults removed										
14/9/09	1 I, 7 II	9 II	9I	5I,25II,4III	8II,2III	5II,1I,1V	2V	4I,9II	5I,5II	5I,1II,1III
16/9/09	9 II, 1I	3 I, 5 II, 4 III	20II,1 III	8II,13III,11IV	5II,3IV,1V	5II, 2V	2III, 1V, 1ad	9II,5III	5II,5III	2I,13II,5III, 1IV
18/9/09	8 III, 1 II	5II,6III,2IV	12II, 10III,1IV	6III,9IV,8V,1ad	6III,4IV,2V,1ad	5III,2ad	2II,1III, 1V, 1ad	10III,12IV ,3V	8I,6II,6III	12II,13III, 4IV,1V
20/9/09	8 IV	2V	4IV	12V	8V, 3ad	2V	2IV, 2V	3V	3IV	1V
22/9/09	3 IV, 5V	4V	10III,13IV ,2ad	9V,4ad	2III,10V, 3ad	4IV,4V, 1ad	4V, 1ad	3IV,9V	10III,4IV, 5V, 2ad	12IV,5V
24/9/09	7V	1IV, 4V	8III,9IV,11V	2V,5ad	2 IV,5V, 5ad	1IV,4V, 2ad	2IV, 2V	3IV,9V	1II,3IV,9V	5IV,7V
26/9/09	6V 1 ad	1V,3ad	4IV,22V	1V,1ad	7V	4V, 2ad	2IV,2V	1IV,8V, 1ad	11IV,9V	11V,2ad
29/9/09		1ad	12V,8ad		5V, 2ad	4V	2V,2ad	3V, 6ad	2ad	7V,3ad
Est. Nymph pop.	10	17	26	34	15	10	7	25	23	30
	1st Instar	2nd Instar	3rd Instar	4th Instar	5th Instar		adults			
Totals	196			146	102		68			

Table 9.2.3: Comparison reproductive rate (expressed in the number of resulting nymphs) of macadamia lace bug in upper and lower tree canopy on bagged racemes. (CTH Alstonville 18/8/2009).

Tree		Low1	Low2	Low3	Low4	Low5	Top1	Top2	Top3	Top4	Top5	Totals
White	2nd instar	0	0	0	0	0	0	0	0	0	0	0
	3rd instar	0	0	0	2	0	0	1	0	0	0	3
	4th instar	0	0	0	0	0	0	0	0	0	0	0
	5th instar	0	0	0	0	0	0	0	0	0	0	0
Yellow	2nd instar	7	7	0	3	0	0	0	0	0	0	17
	3rd instar	7	1	0	0	0	0	0	0	0	0	8
	4th instar	5	0	0	0	0	0	0	0	0	0	5
	5th instar	2	0	0	0	0	0	0	0	0	0	2
Pink*	2nd instar	0	0	0	0	0	0	0	0	0	0	0
	3rd instar	0	0	0	0	0	0	0	0	0	0	0
	4th instar	0	0	0	1	0	0	0	0	0	0	1
	5th instar	0	0	0	0	0	0	0	0	0	0	0
	Totals	35					1					36

*Pink tagged racemes contained breeding pairs of the dark bodied species (*Ulonemia concava*) (NSW DPI taxonomy branch)

While the levels of oviposition were difficult to detect on a single raceme without destroying the nymphs, data showed a single female produced up to 33 nymphs (20 average, n=10) and the survival rate to adulthood was close to 50% in the test conditions (Table 9.2.2). It is not known how many eggs were initially laid. Under field conditions the best reproductive rate observed was 17 nymphs from a single raceme and 97% of nymphs were found in the lower racemes where less sunlight penetrated. (Table 9.2.3).

Lace bugs remained on the bark of the macadamia trees between flowerings. (Figure 9.1.5).The population of lace bugs was estimated for an individual tree by using two $16m^2$ tarpaulins laid out below the tree, the trees were spayed with endosulfan and the fallen lace bugs collected 2 hours later. The dead lace bugs were counted. The November 2009 spray on a tree yielded over 500 adults, the December 2009 spray 40 adults and the January 2010 spray 15 adults. Macadamia trees that fail to set a crop from the normal spring flowering (August to October) will flower again in December/February and May/June the following year. We have established that macadamia lace bug adults can remain in significant numbers on macadamia trees between flowerings once they have established in an orchard and if no spray applications are made.

Macadamia lace bug insecticide bioassays

Laboratory screening

Perceived lack of effective insecticides in the aftermath of the withdrawal of endosulfan is a major issue for the macadamia industry because endosulfan provided a cheap and efficient method for managing macadamia lace bug. Preliminary work from 2005 (Table 9.2.4) indicated all other insecticides registered in macadamias were effective as was thiamethoxam which we were trialling at the time. A method for mortality screening needed to be developed for macadamia lace bug because laboratory survival on desiccating macadamia florets is very poor. Control mortality of macadamia lace bugs at 3 days was high (67%) due to senescing florets. Assays for macadamia lace bug were scored similarly to FSB (Table 9.2.5). Test chemicals in the bioassay were therefore scored at 24 hours (Table 9.2.6).

Anecdotally growers do not generally regard Lepidex® (registered @ 1ml/L) as an adequate alternative to endosulfan. Spray failures (mostly for FSB) with Lepidex® were often associated with alkaline hydrolysis when used with copper products. Our testing even after letting the mixtures stand for 20 hours before use suggested this was not the case (see Table 5.2.6) and we achieved 100% macadamia lace bug mortality with trichlorfon and copper hydroxide and copper oxychloride (Table 9.2.6). Bayer compound DC093 and both copper fungicides, and the Hasten® wetter had no impact on macadamia lace bugs (Table 9.2.6).

The spinosad formulation Success® was not effective against macadamia lace bug in either laboratory bioasays or field testing (Tables 9.2.6 & 9.2.9). Bayer compounds DC092, DC091, DC084 were all active at the rates suggested Table 9.2.6, & 9.2.7). DC093 was only 50% effective at 4ml/L rate and the BioPest® oil failed to control at 2ml/L. Pest oils have been traditionally used up to 2% concentration (20ml/L) but they can cause phytotoxicity at flowering. DuPont HGW086 showed some efficacy in the laboratory (Table 9.2.7).

Field spray trials

In small scale field trials re-invasion in high pressure areas is an issue. The presence of surviving nymphs is the key indicator in an assessment of a compounds efficacy (Table 9.2.8).

With macadamia lace bug it is important to detect infestation quickly as the population can expand rapidly. Samples were taken in the field prior to treatment from each site to determine macadamia lace bug pressure. At Doust farm Tuckombil average levels of 4.7 bugs/raceme (standard deviation 6) from 25 racemes were recorded and 1.4 bugs/raceme (standard deviation 2.4) from 27 racemes at CTH before the treatments were applied.

Only the endosulfan treatment at the Tuckombil 2007 trial appeared effective Table 9.2.8). During the 2009 trial at CTH Alstonville the new formulations of DC84, BYI and imidacloprid (i.e. Confidor®) all failed to kill macadamia lace bug on the racemes (Table 9.2.8).

Adult macadamia lace bugs constantly re-infested the CTH Trial site, while at the other sites patchy distribution and lower density caused problems for the field trials.

In the 2010 trial at CTH Alstonville, the Bayer compound DC092 (200SL) applied at 1ml/L gave similar control to endosulfan applied at 1.5ml/L (350EC). Adult mortality due to contact with DC092 was also good in laboratory bioassay and may be the best alternative to endosulfan found to date (Tables 9.2.5, 9.2.6 and 9.2.8).

The persistence of macadamia lace bugs within unsprayed areas made crop fruit setting virtually impossible until the population was reduced (Table 9.2.9). Adult macadamia lace bug feeding on the tree trunk may also affect fruit set and this aspect needs further investigation.

Table 9.2.4: Preliminary investigation of macadamia lace bug mortality after exposure to dipped racemes (September 2005). Aggregate mortality scored under 20x magnification 3 replicates of 3 adults for each treatment (n=9).

Treatment	Active	Registered rate ml/L	Dip Rate ml/L	Day1 No. live	Day 1 No. dead	Day 2 No. live	Day 2 No. dead
Bulldock ®	Beta-cyfluthrin	0.5	0.25	0	9	0	9
Endosulfan	Endosulfan	1.5	1.5	0	9	0	9
Actara ® **	Thia-methoxam	0.3**	0.3	0	9	0	9
Dipterex®	Trichlorfon	1.0	1.0	0	9	0	9
Water				6	3	6	3

** unregistered compound used in *Amblypelta nitida* field trials at CTH 2005-2007
Active Thiamethoxam 250 WG Actara®

Table 9.2.5: A study to determine an optimal time for scoring insecticide mortality of macadamia lace bug adult and nymphs on racemes sprayed with various insecticide formulations in two separate trials

Date	Treatment		Dose ml/L	gai applied	Test population number	18 hours %Mortality (sd*)	48 hours %Mortality (sd*)	72 hours. %Mortality (sd*)
25/8/2009	Control	Water			21			67(50)
	DC091		1.0	0.0004	21			97(5)
	DC092		1.0	0.0004	17			100
	DC093		1.0	0.0004	19			55(20)
	Endosulfan®	Endosulfan	1.5	0.002	23			100
	Lepidex®	Trichlorfon	1.0	0.001	18			93 (11)
	Lepidex®	Trichlorfon	2.0	0.002	24			100
	Success®	Spinosad	2.0	0.00192	21			89(8)
2/9/2009	Control	Water			38	0	11 (7)	
	Endosulfan®	Endosulfan	1.5	0.002	30	97 (4)	100	
	Lepidex®	Trichlorfon	0.5	0.0005	25	47 (22)	52 (27)	
	Lepidex®	Trichlorfon	1.0	0.001	27	91 (13)	100	
	Lepidex®	Trichlorfon	2.0	0.002	42	89 (8)	100	
	Lepidex®	Trichlorfon	2.5	0.0025	25	91 (7)	100	
	Lepidex®	Trichlorfon	4.0	0.004	20	100	100	
	Lepidex®	Trichlorfon	5.0	0.005	59	100	100	

Table 9.2.6: Comparison of 24 hour mortality rates of macadamia lace bug adult and nymphs on racemes sprayed with various insecticide formulations and mixtures with copper fungicides (trichlorfon mixtures were left to stand for 24 hours before use in the trial applied on (15/9/2009).The trial used recommended rates for trichlorfon, endosulfan and copper products and Hasten®

Date	Treatment	Active	Dose gm or ml/L	gai applied	Test population	%Mortality average	STD Dev. % Mortality
15/9/2009	Control	Water	0.0		32	0	0
	Lepidex®	Trichlorfon	1.0	0.001	20	100	0
	Champ®	Cu OH	1.4	0.001	12	0	0
	Coppox®	Cu OxyCl	2.5	0.0025	29	23	10
	Lepidex® + Champ®	Trichlorfon+ Cu OH	1.0+ 1.4	0.001+ 0.001	21	100	0
	Lepidex® + Cu OxyCl	Trichlorfon+ Cu OxyCl	1.0+ 2.5	0.001 0.0025	17	100	0
	DC092		0.25	0.0001	17	53	3
	DC092		0.5	0.0002	31	75	15
	DC092		1.0	0.0004	14	100	0
	DC093		0.3	0.00012	10	33	33
	Hasten®		1.0		24	21	6
	DC092 + DC093 + Hasten®		1.0+ 0.3+ 1.0	0.0004+ 0.00012	24	100	0
21/9/2009	Control	Water	0.0		27	0	0
	Endosulfan®	Endosulfan	1.5	0.002	35	100	0
	Success®	Spinosad	0.5	0.00048	41	39	13
	Success®	Spinosad	1.0	0.00096	32	21	12
	Success®	Spinosad	2.0	0.00192	23	5	7
	Success®	Spinosad	3.0	0.00288	23	55	39
	Success®	Spinosad	4.0	0.00384	29	80	4
	Success®	Spinosad	5.0	0.00480	59	75	24

Table 9.2.7: Mortality (24 h) of macadamia lace bug adult and nymphs on macadamia? racemes sprayed with various Insecticide formulations applied in separate trials.

Date	Treatment	Active	Dose gm or ml/L	gai applied	Test population number	%Mortality average	STD Dev. % Mortality
15/9/2009.	Control	Water	0.0		18	0	0
	DC084	N/A	1.0	0.0002	30	94	0
	DC084	N/A	2.0	0.0004	9	100	6
	DC084	N/A	4.0	0.0008	25	96	0
	DC091	N/A	1.0	0.0004	29	97	5
	DC091	N/A	2.0	0.0008	20	100	0
	DC091	N/A	4.0	0.0016	19	100	0
	DC093	N/A	1.0	0.0004	21	65	18
	DC093	N/A	2.0	0.0008	30	55	15
	DC093	N/A	4.0	0.0016	23	47	13
	Endosulfan®	Endosulfan	1.5	0.002	19	100	0
	BioPest®	Paraffinic oil	2.0	0.004	47	0	0
7/10/2009.	Control	Water	0.0		6	0	0
	Agral ®		0.25		3	25	35
	HGW086 + Agral ®	N/A	1.0 0.25	0.0002	5	84	30
	HGW086 + Agral ®	N/A	2.0 0.25	0.0004	6	100	0
	HGW086 + Agral ®	N/A	4.0 0.25	0.0008	10	100	0
	Endosulfan® + Agral ®	Endosulfan	1.5 0.25	0.002	5	100	0

Table 9.2.8: Results of insecticide efficacy trials conducted on flowering macadamia trees infested with macadamia lace bug. All selected and treated racemes were collected 3 days after application. Data are tabled cumulative counts from racemes from each treated trees

Site / Date	Treatment	Active	Rate ml/L	Racemes examined	gai applied in 2 ml	Live adults	Live nymphs	Dead adults	Dead nymphs	Cast skins
	Success ®	Spinosad	0.8	26	0.0076	16	96	2	5	32
Tuckombil Sept 2007	Success®	Spinosad	0.4	21	0.0038	0	37	0	1	35
	Lepidex ®	Trichlorfon	2.0	26	0.002	3	35	0	2	28
	Lepidex®	Trichlorfon	1.0	25	0.001	1	26	1	1	25
	Endosulfan®	Endosulfan	1.5	24	0.002	0	4	1	1	22
	BioPest®	Paraffinic oil	20.0	24	0.04	0	39	0	1	14
	unsprayed			29		9	77	0	1	35
	Water			28		7	68	0	2	51
CTH Alstonville Oct 2009	Endosulfan	Endosulfan	1.5	15	0.002	0	0			
	Bulldock ®	Beta-cyfluthrin	0.5	15	0.000025	0	0			
	Lepidex ®	Trichlorfon	1	15	0.001	0	0			
	Lepidex ®	Trichlorfon	2	15	0.002	0	0			
	BYI		0.4	15	0.00019	0	3			
	Calypso®	Thiacloprid	0.38	15	0.00038	0	0			
	Confidor®	Imidacloprid	0.9	15	0.00036	0	1			
	DC083		0.3	15	0.00012	0	0			
	DC084		4	15	0.0008	0	2			
	Hasten®		1	15		4	11			
	Rogor ®	Dimethoate	0.75	15	0.0006	0	0			
	unsprayed		0	15		12	4			
CTH Alstonville Sept 2010	BioPest®	Paraffinic oil	2.0	101	0.004	4	167	0	9	71
	DC092		1.0	86	0.0004	4	1	0	11	10
	Endosulfan®	Endosulfan	1.5	94	0.002	0	1	0	7	42
	Lepidex ®	Trichlorfon	1.5	96	0.0015	1	37	1	17	27
	Lacewings 125/Tree			95		3	41	0	3	30
	unsprayed			95		6	36	2	5	60

Table 9.2.9: Average dry nut in shell yield (kgs per tree @ 10% moisture) on CTH Alstonville macadamia trial since 2003. (Each plot was made up of 9 trees buffered by macadamia variety 246 trees. All plots were managed similarly. Broad spectrum insecticides were applied 4 times, at monthly intervals, beginning in October after flowering (2x endosulfan + carbendazim and 2x beta-cyfluthrin).

Treatment	Variety flower time	Block	2003	2004	2005	2006	2007	2008	2009 (+)	2010
Unsprayed	741 early	1	2.0	6.1	14.5	6.3*	22.7	7.3	14.2 (2)	6.1
	246	2	3.4	4.7	9.6	14.4	19.0	3.9	9.1(19)*	0.6*
	849 late	3	4.1	8.3	9.2	13.2	14.8	3.8	5.3 (24)*	0.4*
	A4 late	4	5.7	7.2	6.8	12.8	15.7	13.1	4.6 (67)*	0.9*
Standard	741 early	9	3.9	12.5	12.0	12.8	22.7	6.3	19.6 (4)	10.4
	246	10	5.0	11.6	10.1	15.2	23.0	3.7	20.3 (2)	6.0
	849 late	11	3.9	9.8	10.9	14.8	19.2	9.4	14.6 (6)	3.8
	A4 late	12	5.1	5.1	5.5	13.0	14.4	15.3	10.3 (13)*	5.0
Unsprayed	246	19				8.3	14.4	3.1*	2.3 (67)*	0.0*
Standard	246	20				11.4	13.9	8.4	18.9 (5)	4.5

*Macadamia lace bug presence noted at flowering (>10% florets infested). The 2009 bracketed figure is the average estimated % of lace bug damaged florets per tree in the lower canopy (7/9/2009) impacting on the 2010 crop.

10. Overall discussion

A significant pest complex, including the key pests macadamia nutborer (MNB) *Cryptophlebia ombrodelta* (Lower) (Lepidoptera: Tortricidae) and fruitspotting bugs (FSB) *Amblypelta nitida* Stål and *Amblypelta lutescens lutescens* (Distant) (Hemiptera: Coreidae) challenges the macadamia industry.

The macadamia industry has supported a system of crop scouting to ensure control treatments are targeted and only used when needed. However, a numbers of insecticide treatments are generally required. Due to the high costs associated with chemical application and the potential adverse impacts of insecticides on the wider rural urban interface, there is a strong desire to improve the current system of macadamia pest control by utilising IPM.

Many issues have arisen since the beginning of this project including the withdrawal of endosulfan registration from available insecticides. A new APVMA requirement for a spray buffer of 100 metres or more between orchards and adjacent specified sensitive areas (i.e. human dwelling, schools, cattle, water ways, national parks) presents a serious challenge. In order to reduce the impact of spray drift, new and improved technologies (Drift Reducing Technology (DRT)) and better spray drift management methods need to be adopted. The dilemma for macadamia growers is clearly demonstrated through the build up of pest populations (macadamia lace bug and FSB to a lesser degree) within the untreated buffer areas around a farm. These will impact heavily on the nutset within the main orchard making the farm economically unviable. Critical land use decisions are additionally required regarding rural or residential land to ensure arable land is maintained for agriculture. The best macadamias tend to be grown on very productive red Kraznozem soils in high rainfall areas of Northern NSW and South East Queensland (coincidentally the area highest human population growth in the Australia).

The current IPM strategy needed refining to incorporate sustainable management options to replace endosulfan for control of fruitspotting bugs (FSB) (*Amblypelta* spp.). FSB are considered the key pest associated with macadamia production and cause the majority of nut damage. The impact of FSB was confirmed by previous research and grower observation and FSB management is driving the macadamia pest management strategy.

To ensure unrestricted market access for all Australian macadamia products, particularly nut in shell, processors need to be able to guarantee that no kernel pest can survive post harvest heat treatment commonly used as part of nut processing.

This study therefore had two major aspects:

1. Improving the current IPM strategy by the following:

    a) Exploring the feasibility of using highly attractive varieties of macadamias and alternative hosts (i.e. *Murraya paniculata* (L.) Jack (Rutaceae) as a monitoring tool and/or trap crop to quantify pest movements and ensure treatment is applied in a timely manner.

    b) Evaluate IPM compatible insecticide options to replace endosulfan for FSB and macadamia lace bug control.

c) Investigate the potential of mangroves as an off-season host for MNB and determine whether MNB numbers in mangrove can be used to predict MNB migrations into surrounding orchards.

2. Management of macadamia pests for quarantine export

The aim was to:

a) establish if standard post harvest processing is lethal to all the relevant macadamia pests including: macadamia nutborer, kernel grub *Assara seminivale* (Turner) (Lepidoptera: Pyralidae) and the bark beetles *Hypothenemus eruditis* Westwood found in NSW and *H. seriatus* (Eichhoff) (Coleoptera: Scolytidae) found in QLD,

b) determine if the bark beetle species could be cultured in the laboratory for lethal temperature investigations and

c) establish the maximum temperature threshold values for the survival of different life stages.

Insecticide screening for fruitspotting bugs

a) Of the older chemistries screened fipronil 0.4ml/L was an effective alternative to endosulfan against FSB, BSB and GVB. Methomyl (i.e. Lannate®) is also known from other work to be effective and needs to be trialled. Indoxacarb when formulated as Avatar® was not effective but another formulation indoxacarb (Steward® at 4ml/L) was effective in controlling adult FSB.

b) Experimental Bayer insecticide DC084 at 4ml/L was effective against FSB, BSB and GVB. Bayer DC091 was also effective against FSB at 0.6ml/L and DC092 was equally if not more effective at 1.0ml/L against FSB. DC092 was the most effective of the new agents to manage macadamia lace bug (Chapter 8) and it is recommended that registration should be pursued and residue work commenced.

c) Dow GF2032 was not effective against FSB in laboratory tests at doses up to 4ml/L.

d) A novel insecticidal plant extract from Dr Robert Mensah (NSW DPI) was effective against FSB at 2% rate. This compound is currently being tested in a field trial this season.

e) Copper hydroxide formulations significantly enhance spray tank pH to levels over 10

f) Trichlorfon (i.e. Lepidex®) formulation at 1ml/L was inferior to endosulfan treatments at 1.5ml/L and all were less effective on BSB adults when mixed with fungicides. It is recommended that the experiment is repeated with using a 2ml/L series against FSB.

Alstonville trials

Macadamia nutset overall was significantly impacted by treatments and macadamia varieties tested. Out of the four varieties tested, variety 741 racemes consistently set

the most nuts and the final crop size was a 50% higher overall and with the least amount of FSB damage.

Cryptophlebia ombrodelta (MNB) pheromone trapping inside the orchard and at mangrove sites gave asynchronous peaks in activity (earlier in mangroves than in the orchards). From 2007 to 2010 the live egg/100nut benchmark treatment threshold showed that while season 2008/2009 had higher MNB pressure than other seasons overall, the combination of spraying of methoxyfenozide (i.e. Prodigy®) and releases of the egg parasitoid *T. cryptophlebiae* resulted in best control of MNB.

Heat sum development models for MNB when combined with pheromone traps gave an accurate prediction of timing of second generation of MNB to impact on the crop (within 10 days each year). The migration flight from mangroves in the southern estuary regions is not considered in this model. The model can be improved by including data from monitoring this new source of MNB in the mangroves.

Nut drop due to *Amblypelta nitida* (FSB) feeding during the season was highest in mid December each year. Nut drop most apparent under macadamia variety 246 and least apparent under macadamia variety A4. These results were contrary to the actual FSB damage in kernel of the total nut yield. This suggests that A4 is far less likely to drop damaged nut but increases the sorting cost to the grower. Numbers of FSB observed in the orchard were highest on the A4 and 849 varieties in the "Wasp only" and "IPM" treatment areas. These data are consistent with the final damage observations at an individual tree level but correlations were only significant in 2007/2008. By releasing FSB into the orchard each season a "hotspot was generated" in the unsprayed block 4 that consequently spread to the adjacent blocks 3 and 5 with thin shelled varieties. The spill over effect allowed the population generated in the untreated area to radiate out through the orchard as the season progressed and despite insecticide treatments in the other blocks. The movement of FSB out of the untreated areas of the orchard also complicated crop monitoring because damage may actually occur as late as March on some varieties when migration was not monitored. Ideally there should be a consistent flow of FSB through the orchard giving levels of damage where Wasp only treatment > IPM > Standard > Experimental treatment. Data from all the high pressure seasons has supported this hypothesis of FSB movement. Over the life of the orchard (12 years now), average FSB damage levels per tree have increased with orchard age. Varieties A4 and 849 gave most consistently high damaged plots (with the exception of 2005 and 2007).

On farm trials

The on-farm trials conducted in NSW and Queensland included no pest refuge area so pest build up was delayed. For this reason there was no difference in the major pest final damage levels either managed with IPM or standard spray regimes. The change from thiamethoxam (2007-2008) to trichlorfon (2009-2010) use in the early part of the season was more effective in controlling FSB. The overall loss to FSB during the trial period, on grower managed farms was only 1-4%. These data compare favourably to 8-18% loss (variety dependent) in treated plots maintained at CTH. Damage reached over 60% in 2010 within the untreated plots with thin shelled varieties (849 and A4) at CTH. As the FSB population builds up in macadamia throughout spring industry requires access to an effective spray programme with a series of options. Monitoring during October and November is reasonably efficient and damaged nut drop is a good indicator of FSB activity and control needs. In

summer the lag between feeding and nut drop is greater and seeing FSB in the upper canopy is often too difficult and time consuming for growers to cover the whole farm. Alternative means of finding FSB activity in summer need to be explored and the “Hotspot” phenomenon shows promise.

Semiochemistry, trap crop hedges of alternative hosts for the management and monitoring of fruitspotting bugs

Needs that have become apparent are: knowledge of the reproductive state of overwintering FSB, knowledge of any regional differences or local movement of FSB, and how this knowledge can be to used to manipulate FSB. The work using field *M. paniculata* hedges suggest that FSB egg production is more tuned to host phenology than first realised. FSB can progress through 1-2 generations during winter on fruiting *M. paniculata* even in NSW (cooler temperature and shorter days would be most noticeable). We observed lower late season FSB pressure during the drier years (in 2004, 2005 and 2007) CTH orchards and on *M. paniculata* hedges, the reasons for which are not clear. Crop size and kernel damage on some macadamia varieties (741 and A4 especially) in the untreated area also followed this pattern (low late season FSB presence and damage in drier years) apparently in-phase with FSB establishing on hedges. Peak activity of FSB on untreated macadamias neighbouring the hedges appears to be in December to January corresponding to damage. It is crucial to find a host plant that will be more attractive to FSB than macadamias at that time. The impact of simulating flowering (adding benzaldehyde) during the 2006 season to generate higher damage in the standard treated areas shows the importance of florescence as a signal to the pest.

Oviposition in each host crop, however, begins at florescence each season. The floral sequence hedge with alternative FSB hosts had some encouraging early FSB incidence data, and the host plants within the hedge showed significant damage (>50% in 2009/10) on avocado and guava. Levels of FSB activity found on longan were minor and only appeared in January to March during high FSB pressure seasons.

A small grid of trap hedges across the production area is recommended and should give a good indication of when and where FSB pressure can be expected and what parasitoids are present.

Semiochemicals (i.e. aggregation pheromomones, markers of feeding sites) can be produced by FSB on indicator trees and this offers a new FSB monitoring device for the grower. Pheromone components for *A.l. lutescens* have also been identified and should be trialled as attractants for trapping FSB. Findings of this project have delivered a series of plants that can be strong indicators of FSB abundance. Alternative hosts such as *M. paniculata* can function as a “live trap”. However trap plantings can take some years to attract FSB to fruit. From performance of trap crop hedges so far. The trap hedge offered easy visual recognition of FSB activity provided that varieties selected are terminal bearing and kept below 3m in height to allow easy FSB collection or application of pesticides.

The whole hedge also provided a refuge for the release of egg parasitoids for FSB control. The egg parasitoids built up populations on the hedge and follow their FSB host to the orchards. Finding significant numbers of FSB eggs in an orchard situation is next to impossible. With the imminent availability of FSB egg parasitoids for commercial releases their will be a need to quantify their impact for the grower. The trap hedge offers such a monitoring site with both host and parasitoid numbers at meaningful levels.

The *M. paniculata* hedges are ornamental and as such have vastly different restrictions to pesticide use in food crops. In November 2008 systemic insecticides were used to determine whether invading FSB populations could be prevented from establishing. Very little FSB activity was observed from February through until the following May despite a large berry crop on *M. paniculata*. Berries were collected in June 2009 to compare survival rates of developing FSB nymphs on the sprayed as opposed to unsprayed *M. paniculata* berries. There was no difference in the survival of FSB nymphs detected. These findings imply that the knockdown of the initial and expanding FSB population is crucial to the development of an effective control program. Data also suggested that there was considerable variation in attractiveness of *M. paniculata* plants at CTH with over 80% of FSB being generated on 10% of the trees. The most FSB attractive *M. paniculata* plants were those that fruited earliest and most heavily each year and have the oldest nymphs present early in the season. Female FSB are laying into budding florets as happens in macadamia, litchi and avocado. The floral potency of the *M. paniculata* appears to be the key as an FSB drawing agent.

The FSB response to ribbonwood fruiting in December each year is quite a different to that observed on the other hedge plants. For three to four weeks there is an adult swarming phenomenon that brings the bugs together and so provides an opportunity for targeted control.

New results from the germplasm blocks (2010 and 2011) identified a suitable smaller macadamia variety with greater potential as a monitoring tool than FSB attractive thin shelled macadamia varieties (L64). Seeds from the new plants were collected and will be incorporated into hedges as soon as possible (results will be published in the Macadamia breeding project report).

From the incidence data collected, it is predicted that damaged nut that remains hanging in the tree will serve as a marker for re–infestation. Varieties that carry the most damaged nut tend to be fed on by FSB later in the season. In this way, a trap hedge could indeed draw FSB out of an orchard and provide useful FSB monitoring data that could be used to predict a major FSB influx into an orchard.

Beneficials, mass rearing and releases strategies

Egg parasitoids of FSB can be collected on a number of host tree species but is most efficient on *M. paniculata* hedges where the FSB densities are highest. *Centrodora darwini* (Maddox *et al*., 2002) and all previously known species (Fay & Huwer, 1993; Huwer, 1996) except *Ooencyrtus caurus* were collected at CTH, with *Gryon* sp. the most common since 2008. *C. darwini* was reared and released at the rate of 5 wasps/tree each season in the untreated area at CTH (2002 and 2008) but no reduction in final FSB crop damage detected. A native *Anastatus* sp. that had been identified as and egg parasitoid of FSB (Fay & Huwer, 1993; Huwer, 1996) is under investigation for commercialisation. Measuring the impact of the egg parasitoid performance of FSB field population and FSB damage levels in orchards will be a challenge.

Climatic influences on FSB levels on the macadamia crop?

A climatic data comparison with FSB incidence and crop damage confirms anecdotal evidence of heavier FSB activity in macadamias during wetter seasons. Data showed that by simply managing the spring population in drier years (2003/2004, 2004/2005 and 2006/2007) gave effective FSB control. In contrast, in wetter years the possibility of FSB re-infestation is much higher and the period required for effective FSB management longer. During extended wet seasons, a later spray (December/ January) for FSB should be included in crop management recommendations to minimise the risk of late season damage.

Evaluating *Trichopoda giacomellii* as a potential biological control agent for *Amblypelta nitida* in Australia

A field population of *Trichopoda giacomellii* was sampled in March 2010. The field the parasitism rate of *Nezara viridula* (GVB) was around 12% at Coraki on the Budda pea plant *Aeschynomene indica* and over 50% on black nightshade (*Solanum nigrum*).

T. giacomellii were reared from parasitised GVB adults and nymphs. Female adult *T. giacomellii* in laboratory cages, tended to oviposit on the same GVB many times, causing superparasitism. By removal of excess eggs from hosts an overall parasitism rate of 63% of GVB was achieved. Very few GVB adults survived parasitism but 0-10% evaded detection by *T. giacomellii* in the cage. Maintaining the correct density of *T. giacomellii* is essential to maintaining the colony. Of the excess *T. giacomellii* eggs that were removed from the GVB adults, 23% were still viable and were used to transplant onto FSB adults and nymphs. When *T. giacomellii* eggs were attached to FSB adults and late instar nymphs the host was killed if *T. giacomellii* larva was able to penetrate into hosts. Fourty three percent (24 out of 56) of transplanted *T. giacomellii* eggs entered the surrogate host (FSB). In two cases, *T. giacomellii* females laid single eggs directly on live FSB in the cage and the larvae killed the FSB host. *T. giacomellii* pupae however the failed to eclose.

Our observations during the study suggest that it is a shape recognition selection that the *T. giacomellii* female makes as it straddles a potential new host. The FSB is a much thinner Hemipteran and generally more mobile than GVB (which is easily caught by *T. giacomellii* in the cage). FSB were straddled readily by the flies but oviposition did not normally occur. Many other much smaller Pentatomids (*Plaudia affinis*) however appeared to be equally suitable hosts as GVB in the cage.

Trichopoda pennipes is listed as attacking Coreids throughout a North American range and may be a potentially useful parasite for FSB. *T. pennipes* was recently established in Europe to manage GVB but *T. pennipes* failed to establish in Australia on three occasions. FSB adults are long living and nymphs and adults cause economic damage at a low density, so a biological control agent targeting the invading FSB adults is highly desirable. The Tachinid *Pentamophaga bicincta* (Cottre-Dormer, 1938) has been found in Queensland attacking late instar nymphs and adult *A. l. lutescens* during the 1930's and this parasite or one very much like it would be valuable in an integrated management plan for this pest.

Macadamia lace bugs - Biology and insecticide screening studies

There are two species of macadamia lace bug present in Northern NSW, *Ulonemia concava* and *Physatochelia* sp. with the later being the most common at CTH Alstonville. Development studies showed that *Physatochelia* sp. appeared to complete its life cycle in 16+/-3 days. There were 5 nymphal instars and eggs are laid within the macadamia florets by insertion of the female ovipositor (the oviposition wound is almost undetectable). First instar nymphs emerge from the floret encrusted with pollen and begin feeding on the raceme. Data showed that a single female could produce up to 33 nymphs (20 average, n=10) and that survival rate to adult was close to 50% in the laboratory. Under field conditions the highest reproductive rate observed was 17 nymphs from a single female (on a single raceme). Ninety seven percent of nymphs were found in the lower, darker racemes suggesting that increasing light intensity within the tree can reduce macadamia lace bug impact.

Lace bugs are found on the bark of the macadamia trees between flowerings. Numbers were high in November (500 adults/tree) and reduced as the season progressed (40 adults/tree in December and 15/tree in January respectively). Macadamia trees that fail to set a crop from the July /October flowering flower again in December/February and/or in May/June. These findings suggest that if not controlled, established adult lace bugs can remain in significant numbers between macadamia flowerings.

Endosulfan use against FSB also provided a cheap and efficient method for managing macadamia lace bug. Insecticides that were registered in macadamias were tested all were effective as was thiamethoxam. Alternative chemical options to endosulfan tested, included beta-cyfluthrin (which is particularly toxic to bees), and trichlorfon. Trichlorfon (Lepidex®) is perceived by a lot of macadamia growers to give poor control of lace bug. Field trial of rates of Lepidex® up to 1.5ml/L were not completely effective. When Lepidex® is used in fungicide mixtures with alkaline chemicals (such as copper hydroxide) in the field, hydrolysis of trichlorfon can occur, however, this could not be duplicated in the laboratory with mixtures left to stand for 24 hours.

Spinosad (Success®) was not efficacious against lace bug in either laboratory testing or field trials. Bayer compounds DC092, DC091, DC084 were all active at the rates suggested by the manufacturer. DC093 was 50% effective at 4ml/L rate and the Biopest® oil failed to kill at 2ml/L. Pest oils have been traditionally used up to 2% concentrations (20ml/L) but phytotoxicity is an issue at peak flowering. Results using DuPont HGW086 were inconclusive.

Trials (CTH Alstonville) with new insecticides (DC084, BYI and imidacloprid (Confidor®)) failed to remove macadamia lace bug from the racemes. Bayer DC092 (200SL) when applied at a lower rate of 1ml/L gave similar control to endosulfan (1.5ml/L (350EC)). Contact toxicity of DC092 on adults was good in laboratory trials making it a promising alternative to endosulfan. Problems encountered during the trials at CTH and elsewhere were: reinfestation of adult lace bugs at CTH and low pressure or uneven distributions at other sites.

Heat treatment on NIS pest in macadamia

Evidence suggests that current nut-in shell drying regimes does kill macadamia storage pest species. Exposure to temperatures above 50^0C for more than 24 hours

effectively controlled both common Lepidopteran pest species (*C. ombrodelta* and *A. seminivale*) although *C. ombrodelta* was more resilient to heat.

Results showed that the macadamia kernel grub *A. seminivale* oviposition could be monitored using navel orangeworm egg traps (with powdered macadamia shell as bait). Egg traps could be explored for disinfestations in macadamia processing plants and storage facilities.

Delate *et al.*, 1994 showed that high temperature exposure and high nitrogen atmospheres were effective in managing the tropical nutborer (*Hypothenemus obscurus*) in Hawaii. Native Scolytids cause similar nut-in shell storage problem in Australian macadamias and there is no reason to believe that the pests would not be controlled with currently used temperature treatment regimes.

Management options for banana caterpillar in Bundaberg

Outbreaks of the banana caterpillar *Tiracola plagiata* (Walker) (Lepidoptera: Noctuidae) have occurred in macadamia orchards around Bundaberg since 2008. Bioassays determined that methomyl (Lannate®) at 2ml/L was the most effective control option and an emergency use permit was sought from the APVMA. Chlorpyrifos at 1ml/L also provided some control of banana caterpillar. An effort needs to be made to find a better solution either by making the larvae more accessible earlier in the harvest to treat with softer options or mulching the fallen leaf earlier to prevent establishment of banana caterpillar.

MNB egg parasitoid culture maintenance

Over the period from 2002 to 2010 there was no detectible difference in parasitoid pupal numbers extracted from eggs laid by older MNB moths compared to pupal numbers from eggs laid by younger MNB females. There was however a higher output of MNB pupae during 2004 to 2007 compared to the other years. This may be due to a change of supplier for ingredients for MNB larval diet. Overall 1.5 million pupae have been reared at an average of 355 pupae per dietary tray. Seasonal production data showed that rearing of MNB larvae on diets from May to September produced the highest pupae numbers. Rearing failures occurred during summer due to higher ferment fly *Drosophila* sp. activity in MNB larval diet.

The wasp colony averaged 610 surplus cards with parasitised MNB eggs each year between 2006 and 2010. Production rates of parasitoids were maintained after halving the number of bottles used. Maintaining a strict 2 day feeding schedule for the egg parasitoid can increase production if the need arises. Egg cards were used to seed live wasps into estuarine regions with perennial MNB populations. In 2007 wasps were released into mangroves area at the Richmond River in mid spring with prior releases in the CTH and commercial macadamia orchards. In 2008 wasps were released in autumn and winter into the mangrove areas and in 2009 and 2010 focus was on autumn through to spring releases, aligning with the mangrove fruiting.

Wasp emergence rates from the wasp release cards averaged around 70%. Some egg cards placed did show signs of “superparasitism” (many unviable wasp eggs) but this occurred only when host egg densities were too low for wasp numbers. Poor parasitism of egg cards (<1%) was a far more common problem (than

superparasitism) and was caused when egg cards were introduced too early (before day 10) in cooler months or too late in warmer months. Cold storage of host eggs at (5℃ +/- 3℃) to feed egg parasitoids appears a use ful solution (although after 1 month storage MNB eggs become desiccated and unsuitable as host for the egg parasitoid).

Trapping data from estuarine areas found that where wasps had been released, recapture was common. Areas such as the North creek region around Ballina, Brunswick Heads, Wooyong and Yamba parasitoids were detected in all months except August. In other areas, wasps were trapped in lower numbers (<5 per card) from early spring through until early winter the following year. Highest wasp catches were in autumn. Based on this data, we adopted a plan of releasing wasps only in Winter/Spring (onto the most damaging MNB generation which re-infests orchards each year in late spring). In the future, a second area south of the Alstonville plateau (that is a source of MNB in some seasons) and will be treated with wasp parasitoids as the mangroves begin to fruit in mid winter.

11. Technology transfer

- 2-3 November 2006: A presentation including updates on the IPM research was given at the Pest & Disease workshop at the Australian Macadamia Society Conference, Broadbeach, Queensland.

- 31st of August 2007: A detailed update of the IPM program was given at the macadamia pest consultant meeting which was held at the Centre for Food Technology in Hamilton (Brisbane).

- October/November 2007: Presentations on the progress of the project were given at 6 MacGroup meetings to growers.

  16 October 2007: Macksville (NSW)
  23 October 2007: Beerwah (QLD)
  7 November 2007: Clunes (NSW)
  7 November 2007: Dunoon (NSW)
  8 November 2007: Wollongbar (NSW)
  9 November 2007: Newrybar

- 1-2 November 2007: Attended DPI stall and discussing pest issues with interested growers at Annual Macadamia Society Conference, Tweed Heads.

- 29 August 2008: Presentations "Progressing IPM in macadamias - Update 2008" and "Climate and crop sizes and FSB developments" were given at the macadamia crop consultant meeting in Brisbane. The meeting was well attended with 32 crop consultants, researchers and processors present. Topics discussed and presented were issues from the past season, pest and disease management, kernel quality, egg parasitoid rearing and internal discolouration of kernel. The IPM presentations were well received and resulted in useful discussions.

- 24 September 2008: Presentations "Macadamia pest management - Taking a closer look at alternatives" and "Cost of production, FSB developments and the new minimal spray trial" were presented at a field day organised by MPC (Macadamia Processing Company) on 24 September 2008 at Rosebank. Approximately 70 farmers attended the field day where five presentations were given on organic horticulture in general, principle and use of agrichar, organic macadamia production and pest management- alternatives, economics and future developments. Presentations were well received and lead to active discussions.

- 30 October to 1 November 2008: A presentation "Progressing IPM and tackling options for FSB – the next big challenge" was presented at the AMS Annual Conference in Ballina.

- 9 June 2009: Presentation on update of macadamia IPM project was given at AMS Research Forum in Brisbane.

- 18 June 2009: Meeting with pest consultants and update on IPM Project, Centre for Food Technology-Hamilton in Brisbane.
- 8 July 2009: Presentation on macadamia lace bug at MPC field day.
- 21-22 October 2009: Presentation at Annual Macadamia Industry Conference in Bundaberg on

  "Emerging Pests - What Are They?"

  "Do Organics and Sustainable Practices Work? New developments in biological controls"
- 28 July 2010: Presentation at the macadamia pest consultant meeting, Brisbane Technology Park.
- 28 September 2010: Bugs, wasps and Nuts (Maddox) Presentation at the Australian Entomology Conference, Perth, Vines Resort WA.
- 28 September 2010: Mangrove – important host for macadamia nutborer outside the orchard (Huwer) Presentation at the Australian Entomology Conference, Perth, Vines Resort WA.

Publications:

1. Huwer, R. and Maddox, C. (2007) Lace bug – increasing incidences in NSW orchards. *AMS News Bulletin November 2007*, Volume 34 Number 6, 41-43.
2. Huwer R.K., Maddox, C.D.A. and Purdue, I.M. (2008). Progressing IPM and tackling options for FSB – the next big problem Proceedings of the Australian Macadamia Society Conference 30 October - 1 November 2008
3. Huwer R.K. (2009) Workshop - Pest & Disease Management: Emerging Pests - What Are They? Annual Macadamia Industry Conference, 21-22 October 2009, Bundaberg
4. Maddox, C. D. Mitchell, A. and Dawes, M. (2009) Identification of Australian Scolytid beetles in macadamia tissue and the use of the "DNA barcodes" for the rapid identification of exotic pest incursions. *AMS News Bulletin March 2009*, Volume 36 Number 2, 42-44.
5. Mitchell, A and Maddox, C.D. (2010) Bark beetles (Coleoptera: Curculionidae: Scolytinae) of importance to the Australian macadamia industry: an integrative taxonomic approach to species diagnostics. *Journal of the Australian Entomological Society* **49**(2), 104 -113

Other:

- An improved monitoring technique for FSB is still being developed and still requires refinement for publication. Current updates for FSB monitoring and recommendations are published in this report.
- FSB management is a key to an improved IPM strategy in macadamias and an IPM suitable management option for FSB is still being developed. Project

results were presented to growers as they became available. Current best recommendations are published in this report.

- Craig Maddox did an extensive study on the genetic barcoding of the different bark beetle species in macadamia. Outcomes of this study were published in a DAAF report and also in an AMS newsletter (see references above).-

- There was no further insecticide survey conducted, as a lot of currently used insecticides are under review. There however was a recent survey commissioned separately to AgAware (Peter Dal Santo) as part of a pesticide use review for the macadamia industry.

12. Recommendations

1. *Continuation of insecticide studies*

   The new compounds tested in this project (MC06021) are looking promising, but need further field testing. There is also a need to screen new emerging insecticide compounds (conventional and biopesticides) for a better integration into IPM strategies and options for organic growers. Some of the new chemistries do appear effective with Bayer's 092 formulation the most likely candidate to replace endosulfan for FSB control. Of the older chemistries fipronil, methomyl and indoxacarb are effective but spray drift regulations are going to impact heavily on where these chemicals can be applied.

2. *Development of a better monitoring tool for FSB*

   There is still no suitable monitoring technique available to farmers. Preliminary research on trap hedges showed promising results in attracting FSB onto highly susceptible hosts for easy monitoring. Monitoring results were associated with crop damage in neighbouring orchards. Further trials of trap hedges should be undertaken on commercial farms in the coming seasons.

   There is also a need to continue with the pheromone work for *A.l. lutescens* and refine the mixture of pheromones and to test in the field. The pheromone work also needs to be adapted for *A. nitida*.

3. *Further investigations into biological control of FSB*

   A mass-rearing technique for FSB and egg parasitoids (i.e. suitable for a commercial insectary needs to be developed). Alternative hosts for FSB egg parasitoids need to be investigated.

   Investigations into the potential value of *Trichopoda* spp. as biological control agent for FSB needs to be further studied.

   Orchard management options that maintain biological control agents in or near the orchard need to be investigated.

   The potential value of the trap hedges on commercial farms as refuge for biological control agents of FSB needs to be investigated.

4. *Improvement on macadamia lace bug management*

   There is currently no effective monitoring system for macadamia lace bug and an effective monitoring technique needs to be developed.

   With the withdrawal of endosulfan registration for macadamia lace bug control alternative chemicals need to be investigated. Some new chemical compounds did appear effective with Bayer's 092 formulation the most efficacious. Other compounds are currently being investigated and this should continue. Older chemistries, such as methomyl and indoxacarb are effective, but not yet registered. Their role as management tools within an IPM system needs evaluation.

5. *Heat treatments for nut-in-shell pests*

   Exposure to temperatures above 50^0C for more than 24 hours will effectively control both common Lepidopteran pest species (*C. ombrodelta* and *Assara seminivale*).

   High temperature exposure was effective in managing the tropical nutborer (*Hypothenemus obscurus*). Use of nitrogen in silos should be applicable for use against Australian Scolytid species in silos, but studies are required for confirmation. .

6. *Management of banana caterpillar in Bundaberg*

   Methomyl (Lannate®) at 2ml/L is the most effective control option. As a non-chemical option, mulching of fallen leaves in spring to prevent pest establishment needs field evaluation. “Softer” chemical options like methoxyfenozide (Prodigy®) or the pathogen *Metarhizium* sp. could also be used for control of small larvae earlier in the season (winter) and needs to be tested.

7. *MNB egg parasitoid use*

   Parasitoid releases within a macadamia orchard are primarily to reduce the development of the second generation of MNB. Sole reliance on MNB parasitoids is only a suitable strategy and when FSB is removed from the orchard (particularly important for highly susceptible, thin shelled, macadamia varieties, where re-infestation by FSB is possible until harvest).

   Seasonal migration of MNB into orchards in early spring can be reduced by releases of wasps into the MNB overwintering areas (mangroves).

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15. Appendices

15.1. Appendix 1: Crop loss estimates from previous research

Adapted from tables published in Maddox *et al*. 2002 showing late bug effects page

Table 15.1.1: Macadamia total crop loss estimates based on the weight of nut collected at monthly nut assessment during the 2001/02 season in Northern NSW and proportions lost due to *C. ombrodelta* (MNB), and *Amblypelta nitida* (FSB) (8 trees at each site).

	Fernleigh (organic)	Macleans Ridges. (managed)	Jiggi (managed)	CTH Accession block (unsprayed)
Tree Height (average metres)	6m	8m	6m	6m
Harvest (SD) (Kgs NIH)/tree	20 (12)	8.5 (3.9)	39 (11)	28 (12)
% MNB loss average (Max)	3 (7.5)	4 (8.6)	2 (7.5)	5 (12.1)
Average % parasitism of live host eggs (SD)	93 (37)	53 (-)	0 (-)	97 (5.9)
Average number parasitised Eggs / 100 nuts on tree	3.5	3	1	41
% FSB loss average (Max.) Per tree**	**13(30)****	**2 (10)****	**2 (5)****	**8 (24)****

** well over 5% crop loss to FSB in unmanaged areas

Table 15.1.2: Crop loss estimates and *C. ombrodelta* (MNB) parasitism summary by tree at the CTH Entomology block over the 2001/2002 season.

Tree	Block	Total nut	% loss to MNB*	% Live eggs parasitised	Total parasitised eggs	**% loss FSB**	**% loss GVB**
1	1	605	5.4	80	74	**0**	**33**
2	1	729	7.7	97	107	**0**	**73**
3	**1**	**1122**	**2.0**	**100**	**23**	**1**	**9**
4	**2**	**622**	**3.8**	**100**	**39**	**0**	**0**
5	2	614	2.3	88	33	**1**	**0**
6	2	1027	1.9	100	16	**0**	**0**
7	3	645	5.6	68	68	**1**	**0**
8	3	158	14.3	86	51	**0**	**0**
9	**3**	**759**	**2.9**	**83**	**20**	**0**	**0**
10	**4**	**1386**	**1.3**	**-**	**6**	**10**	**0**
11	4	510	8.8	80	17	**0**	**1**
12	4	1278	4.0	0	7	**0**	**3**
Average (SD)							
Sprayed		**972 (350)**	**2.5 (1.1)**	**70**	**22 (14)**	**2.8 (4.9)**	**2.3 (4.5)**
Unsprayed		709 (360)	7.3 (4.7)	75	47 (35)	**0.3 (0.5)**	**14 (27)**

* *C. ombrodelta* loss includes early season drop plus immature and damaged kernel from nuts with larval holes in the husk.

Trees 3, 4, 9 and 10 were each sprayed in January, February and March with 3L of 0.5ml/L beta-cyfluthrin.
All trees were 3m high variety A4 planted 1998, the level of parasitism per 50 nuts sampled on tree and crop loss as determined from each tree at harvest and early nut drop collections.

15.2. Appendix 2: Heat treatments for nut in shell pests

Scolytid beetles are a major destructive faunal element in forest ecosystems around the world. Scolytid species are often saprophytic, “scavenging on” the dying and decaying trees in the forest, while species that invade healthy living tissue can become pests. These small beetles (1-3mm long) can bore into most woody tissue and breed in galleries under bark or inside the seed pods of their host. The feeding can disrupt sap flow causing tree or branch death and some Scolytid species are known vectors of major tree diseases (e.g. *Ophiostoma* spp., *Ceratocystis* spp.). The beetles can render timber and agricultural produce unmarketable, making quarantine inspection necessary for trade in those goods that might harbour these pests.

In the last few years Scolytid beetles were found frequently attacking the fruit, bark and wood of macadamia trees in all the major growing districts on the east coast of Australia, .At least 7 Scolytid species are now recognized from macadamia: *Hypothenemus seriatus* in Queensland, *H. eruditis* in NSW and an unidentified *Hypothenemus* species in the macadamia husk and nut tissue around Bundaberg (Figure 15.2.2 and 15.2.3). *Cryphalus subcompactus (*Lea) attacks the bark and husk (Figure 15.2.3) while *Cnestes* sp., *Xyleborus* sp. and *Euwallacea fornicatus* have been taken from the heartwood (Mitchell & Maddox, 2010). The Australian *Hypothenemus* species are smaller than tropical nutborer, and heat treatments are expected to be equally effective in control.

Nut-in-shell shipments for export could be rejected if significant quarantine pests are found within the kernel or shell. Thus we investigated whether the current heating regime to “dry down” nuts would prevent the survival of quarantine pests. In a preliminary study the most likely insect problems were identified and a series of heat exposure studies conducted to determine if the pests could survive the treatment. The pest most commonly found in both the shell and kernel is macadamia nutborer (*Cryptophlebia ombrodelta,* Tortricidae). As this insect is in laboratory culture it was used as convenient key test species. A smaller, more difficult to detect Lepidopteran pest is macadamia kernel grub (*Assara seminivale*, Pyralidae) which attacks exposed kernel in the field (Figure 15.2.1). Macadamia kernel grub is likely to be a problem after shells split in storage (Macadamia kernel grub is generally unable to penetrate a sound shell nut). Other pests in the field including the *Sigastus sp*. weevil and several other unidentified moth species could also potentially be found inside nut-in- shell.

The main exotic pest of macadamia, is tropical nutborer (*Hypothenemus obscurus* (Fabricius) (Coleoptera: Scolytidae) which commonly occurs throughout the macadamia growing areas of central South America and Hawaii (Figure 15.2.2). Tropical nutborer is managed with post harvest treatments such as heat (7 days at 45℃) or modified atmosphere (6 days in 95% CO_2, or N_2) (Delate *et al*., 1994) and within the orchard tropical nutborer is managed culturally by frequent harvesting (Jones, 2002).

Figure 15.2.1:

Top left: Open micropyle of the A16 nut allowing the entry of macadamia kernel grub (*Assara seminivale*)

Top right: Macadamia kernel grub (*Assara seminivale*) pupae within the kernel of a nut at processing.

Bottom left: Adult moths emerging from pupae within nut kernel macadamia nutborer (*Cryptophlebia ombrodelta*) moth on left and macadamia kernel grub (*Assara seminivale*) moths centre and right.

Bottom right: Damaged nut containing macadamia nutborer (*Cryptophlebia ombrodelta*) moth and macadamia kernel grub (*Assara seminivale*) (Bundaberg Queensland).

Figure 15.2.2:

Top left: The major exotic pest tropical nutborer (*Hypothenemus obscurus*) Scolytid beetle (and associated damage) in macadamia in Kona Hawaii April 2007.

Top right: Native Scolytid beetles (*Hypothenemus eruditis, H. seriatus*) attack macadamia husk and nut in shell Bundaberg, Gympie area (Qld) and Macksville (NSW).

Bottom left: The Scolytid bark beetles (*Cryphalus subcompactus)* attacking petioles on A4 nuts in the field. These beetles will kill trees by ring barking the cambium layer, attack husk and could affect the transfer of bud wood and planting material.

Bottom right: Native Scolytid beetle (*Hypothenemus seriatus*) in macadamia shell and kernel collected in Bundaberg Queensland winter 2010.




Figure 15.2.3:

Top left: Scolytid bark beetle *Cryphalus subcompactus* (Lea) adults (40x magnification) from Bundaberg Queensland

Top right: The emergence holes *Cryphalus subcompactus* on the tree trunk after *C. subcompactus* consumed the cambium layer of the macadamia May 2007.

Bottom left: The external appearance of macadamia shell damage due to *Hypothenemus obscurus* in Kona, Hawaii April 2007.

Bottom right: Macadamia shell damage due to *Hypothenemus eruditus* - Stuarts Point NSW.

15.2.1. Materials and Methods

The trial insects (*C. ombrodelta* and *A. seminivale* and *C. subcompactus*) were either cultured, using modified artificial diet (for Lepidopteran larvae), (Shorey & Hale, 1965) (MNB larvae) reared on raw kernel (kernel grub larvae) or collected from the field from areas where pest populations were causing problems (bark beetles). Screening for susceptibility to heat treatment required methods to identify each species and then testing for survival through drying regimes.) For long term storage nuts are subjected to 2 days at 38℃, 2 days at 45° C and 2 days at 60℃ (AMS Macadamia Growers Handbook 2001).

To allow entry of test insects into nuts a 1mm hole was drilled (Dremmel® high speed drill) in 50% of the tested nuts-in-shell (Figure 15.2.1.1). Insects of the required life stage and species were placed inside the drilled chamber and sealed with a small wooden plug (3mm long piece of wooden skewer). There were 5 replicates, containing 20 nuts, per treatment. Untreated controls were carried for each temperature regime tested. Following heat exposure the nut was opened and the insect mortality was assessed, (using 12 x magnifications). Data are shown in Table 15.2.1.1.

Nuts from variety 246 nut were collected and field moisture rates for batches of 25 nuts were measured. Moisture level changes with temperature over time. Moisture level was measured for intact nut-in-shell and nut that had been drilled to insert test insects. Moisture content was determined by placing the sample in a second oven running at 105+/-1℃) for 24 hours for final weight . Replicates of 25 nuts were used to estimate moisture levels at each temperature /exposure combination trialled. The percentage of moisture was calculated by weighing the nut samples before drying and then straight after drying. Dried nuts were compared to field collected nuts. Sub-groups from the field collected nuts were dried with drilled holes that were plugged to see if the desiccation rate was equivalent to the nuts that were dried without drill holes.

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Table 15.2.1.1: Heat exposure testing at CTH Alstonville showing numbers of pests used at each temperature and time combination.

Test Species	**Life Stage**	**30℃**			**40℃**			**45℃**			**50℃**		
		Exposure time in hours											
		24	48	72	24	48	72	24	48	72	24	48	72
Cryptophlebia ombrodelta	Larva	100	100	100	100	100	100	100	0	0	100	0	0
Cryptophlebia ombrodelta	Pupa	100	100	100	100	100	100	100	100	0	100	0	0
Assara seminivale	Larva	100	100	100	100	100	100	100	0	0	100	0	0
Assara seminivale	Pupa	*	*	*	*	*	*	*	*	*	*	*	*
Cryphalus subcompactus	Adult	100	100	0	100	100	0	*	*	*	*	*	*

* not attempted

Figure 15.2.1.1:

Top: Drilled macadamia nut with the holes exposed to allow the insertion of either live MNB or kernel grub larvae or pupae or adult bark beetles, depending on species and life stage tested. Batches of 20 replicate nuts were loaded up with each life stage of each species and tested at allocated temperature.

Bottom Left: Californian navel orangeworm egg trap tested in the field for trapping macadamia kernel grub (*Assara seminivale*).

Bottom Right: Size comparison of macadamia nutborer (*Cryptophlebia ombrodelta*) eggs (white cardboard) and macadamia kernel grub (*Assara seminivale*) (yellow cardboard).

Kernel grub (*Assara seminivale*) presents a challenge in the field at some sites and in "nut-in-shell" storage facilities. In-field monitoring is severely restricted due to *A. seminivale* egg size (Figure 15.2.1.1). Assess to navel orangeworm egg traps, (Figure 15.2.1.1), allowed potential attractants useful for monitoring to be investigated.

A small flight cage (0.4m x 0.3m x 0.4m) was set up containing at least 10 (usually 20) live adult female Kernel grubs. A dental wick, wetted with 10% sucrose, was hung from the cage roof and replaced every 3-4 days. A set of 20 labelled egg traps for kernel grub eggs were available for use to be filled with the test formulations. Six egg traps were used at any one trial, and these were placed inside the cage equal-distance apart. Numbered egg traps were packed with the various treatment options and examined for oviposition after 2-4 days. The number of eggs in each treatment was recorded as number of eggs laid per day. The process was repeated 14 times and the egg traps were cleaned after each use and the baits were reallocated to different egg traps on 4 occasions to ensure randomness of the experimental design and account for location as a factor (Table 15.2.1.2). Traps were assessed for oviposition at 20x magnification.

Table 15.2.1.2: The randomisation of Californian navel orangeworm traps in a series of 14 trapping choice tests.

Test Bait Formulation	**Egg Cage #**
Empty egg trap	5 , 17, 2, 7
Fresh nut in shell	10, 16, 17, 4
Old nut in shell	11, 19, 18, 5
Fresh kernel	12,15, 6, 19
Cracked shell	6, 18, 1, 8
Powdered shell	1, 20, 3, 9

15.2.2. Results

The drying rates for the kernels were not adversely affected by the drilling of holes for insect testing (Table 15.2.2.1). These give some validity to the bioassay method as insects in the drill holes were not drying faster than the rates for intact nuts (Table 15.2.2.1).

None of the test insects survived 24 h exposure at 50^0C, the resulting data suggests that the current nut drying regimes are effective (Table 15.2.2.2).

Table 15.2.2.1: Average percentage moisture content, by exposure and temperature regime, for intact and drilled and plugged macadamia nuts (n=25 per regime).

Temperature	Nut type	% Moisture			
		Initial (sd)	24 hrs	48 hrs	72 hrs
30^0C		19.3 (0.8)	9.7	6.5	5.5
30^0C	drilled	20.2 (1.5)	11.0	6.0	5.7
40^0C		12.0 (0.4)	5.5	4.5	4.1
40^0C	drilled	9.7 (0.3)	5.0	4.1	3.9
45^0C		12.5 (0.5)	4.7	3.5	3.3
45^0C	drilled	13.7 (0.8)	4.6	3.7	3.3
50^0C		12.7 (1.5)	3.7	2.4	2.1
50^0C	drilled	12.6 (0.8)	2.9	2.3	2.2

Table 15.2.2.2: The minimum duration for each temperature and exposure regime required for 100% kill (5 x 20 individuals) . (When the 72 hour temperature exposure failed to produce 100% mortality the difference between the heat treated mortality (T**) and the untreated control set (C**) kept at room temperature is shown).

Test insect	Life Stage	30^0C 24-48-72hr	40^0C 24-48-72hr	45^0C 24-48-72hr	50^0C 24-48-72hr
Cryptophlebia ombrodelta	Larva	Not fatal at 72 hrs 5%T** vs. 10%C**	Not fatal at 72 hrs 22%T vs. 8%C	24hr 100% fatal	24hr 100% fatal
Cryptophlebia ombrodelta	Pupa	Not fatal at 72 hrs 10%T vs. 10%C	Not fatal at 72 hrs 30%T vs. 10%C	48hr 100% fatal	24hr 100% fatal
Assara seminivale	Larva	Not fatal at 72 hrs 50%T vs. 10%C	72 hrs 100% fatal	24hr 100% fatal	24hr 100% fatal
Assara seminivale	Pupa	***	***	***	***
Cryphalus subcompactus	Adult	48hr * 100% fatal	48 hr* 100% fatal	***	***

*** not attempted

*control batches with 100 % mortality do not allow an effective test of Scolytid survival rates. Additional testing is required.

Cryptophlebia ombrodelta appeared able to withstand higher temperatures than *Assara seminivale* (Table 15.2.2.2). Control survival was above 90% for both Lepidopteran species at room temperature across all assays. In the trials with the Scolytid beetle, *Cryphalus subcompactus* we unfortunately could not get any of the beetles in the control treatment at room temperature to survive (Table 15.2.2.2).

The macadamia kernel grub oviposition baiting experiment showed that the females have a strong preference for the powdered shell (Table 15.2.2.3).

Table 15.2.2.3: *Assara seminivale* oviposition rates on various bait formulations inside navel orangeworm egg traps.

Test Bait Formulation	**Average Kernel grub eggs/day ***
Empty egg trap	1.2b
Fresh Nut in shell	1.2b
Old Nut in shell	1.7b
Fresh Kernel	1.6b
Cracked shell	1.8b
Powdered shell	5.0a*

* Means followed by different letters are significantly different using Genstat® analysis of variance (F value 9.55 pr<0.001)

Results as they were available were submitted to AMS.

15.2.3. Discussion

Further work since this project (Mitchell & Maddox, 2010) specifically on the taxonomy of the Scolytid species found in macadamias has shown that *Hypothenemus* species feed inside nut-in-shell and that *Cryphalus* is a cambium feeder. These data may explain why *Cryphalus* used for the experiments failed to survive kernel exposure treatments. None of the *Hypotheneumus* species have been cultured as yet. The Scolytids showed a preference for drier seasons and are becoming a problem in the Bundaberg district (Figure 15.2.1.2) and potential to damage crops should not be underestimated.

15.3. Appendix 3: Management of banana fruit caterpillar in Bundaberg on macadamia

Banana fruit caterpillar *Tiracola plagiata* (Walker) (Lepidoptera: Noctuidae) feeding is normally associated with leaf litter and banana leaves in banana plantations. Outbreaks of banana caterpillar have occurred in macadamia orchards around Bundaberg since 2008 (Figure 15.3.1.1). Emergency Pesticide Use Permits were sought by the macadamia industry for use of methomyl (Lannate ®) for short term pest management.

Banana caterpillars overwinter under macadamia leaf litter. In spring when the macadamia flowers and produces fruit the caterpillars migrate up the macadamia trunks in the evening to feed on young nutlets and return to the leaf litter before day light.

15.3.1. Materials and Methods

The larvae were counted after being sorted into species and divided by size into fourth and fifth instars. Three replicates of 5 individuals (15 in total) were exposed to ten different insecticide treatments plus an untreated control. Two bioassays were done, an ingestion feeding test and a contact spray.

Table 15.3.1.1: Insecticide treatments applied to Banana Caterpillar larva contact toxicity to screen for knockdown efficacy and a nut protectant.

Active	Trade Name	gai applied	Dose applied ml/L
Control (water)			
Methoxyfenozide	240 SC Prodigy®	0.00018	0.25
Methoxyfenozide	240 SC Prodigy®	0.00029	0.40
Fipronil	200 SC Regent®	0.00024	0.40
Chlorpyrifos	500 EC Lorsban®	0.0015	1.00
Methomyl	225g/L Lannate®	0.00135	2.00

Racemes of young nutlets (10-20 per raceme) were collected from unsprayed macadamia (CTH). A set of 3 racemes were placed through rubber seals of 20 ml water filled flasks and 200 ml of insecticide treatments mixed (A grade volumetric glassware (Brand®) and pipettes).

Pesticides were applied at rates suggested by Bundaberg pest consultants Spray mist volume of 3ml was used for contact bioassays and 6ml was used for feeding bioassays.

i. Feeding bioassay

Using a 500 ml hand sprayer on a fine mist setting, a series of 3 sprays were applied to each side of racemes in flasks (1 spray delivers 1-1.2ml). Flasks and racemes were allowed to stand on filter paper lined brass lids for 10 minutes to air dry then 5 larvae were placed on the paper and a labelled Glass jars (Fowlers Vacola, 800ml) caged the racemes, flask and larvae.

ii. Contact bioassay

Using a 500ml hand sprayer, filter papers (3 replicates) were sprayed (see Table 15.3.1.1) at a distance of 10cm 3 times sprays and then filter papers covered along with a flask of clean racemes with labelled glass jars (Fowlers Vacola, 800ml) after including a flask of clean racemes (Table 15.3.1.1). Water used in the control assay. The sprayer was rinsed between insecticides.

Insect were held in the dark at 23℃ ±5℃. Mortality was assessed after 1, 2, 3 and 7 days using lack of response to a fine probe was the criterion for death. Bodies were removed from day 2 onwards at each inspection, as cannibalism was evident and inflated mortality in the control by day 7. Assay was run in 23 hour darkness.

Figure 15.3.1.1: Banana caterpillar emerged adult moths (left) and 5th instar larva used for the bioassay (right) sourced from Bundaberg Queensland.

15.3.2. Results

The only effective contact pesticide (100% mortality) was Lannate® at 2ml/L and an emergency permit for its use in Bundaberg was sought. There was 93% mortality with Lorsban® at 1ml/L. From the feeding bioassay, the most effective pesticide was Lorsban® at 1ml/L. Banana caterpillar would have to be controlled earlier in the lifecycle if softer options like methoxyfenozide (Prodigy®) were to be effective (Table 15.3.2.1) as the latter softer option is most effective on small larvae.

Table 15.3.2.1: Mortality of banana caterpillar contact bioassay (C) or feeding assay or feeding bioassay (F) after 1, 2, 3 and 7 days.

Treatment	**Rate mls/L**	**gai applied**	**Bioassay type**	**Day 1 Cumulative deaths**	**Day 2 Cumulative deaths**	**Day 3 Cumulative deaths**	**Day 7 Cumulative deaths**	**Day 3 %mortality**	**Cannibalism**
Control			Contact	0	2	5	6	33%	Yes in 2/3 reps
Prodigy®	0.25	0.00018	Contact	1	3	5	8	33%	no
Prodigy®	0.4	0.00029	Contact	9	9	12	14	80%	no
Regent®	0.4	0.00024	Contact	0	0	3	10	20%	no
Lorsban®	1.0	0.0015	Contact	14	14	14	14	93%	no
Lannate®	2.0	0.00135	Contact	10	15	15	15	100%	no
Prodigy®	0.25	0.00018	Feeding	6	6	7	9	47%	Yes in 2/3 reps
Prodigy®	0.4	0.00029	Feeding	0	1	2	9	13%	Yes in 2/3 reps
Regent®	0.4	0.00024	Feeding	1	2	2	6	13%	no
Lorsban®	1.0	0.0015	Feeding	5	11	13	13	100%	Yes in 2/3 reps
Lannate®	2.0	0.00135	Feeding	0	1	9	9	60%	Yes in 1/3 reps

15.4. Appendix 4: Report on maintenance of colonies of MNB and their egg parasitoid *Trichogrammatoidea cryptophlebiae*

The use of the egg parasitoid *Trichogrammatoidea cryptophlebiae* Nagaraja (Hymenoptera: Trichogrammatidae) was flagged as a key component in a less spray dependent pest management system for the macadamia industry in Australia (Ironside, 1981). A permit to import and test specificity of *T. cryptophlebiae* from South Africa was obtained by DEEDI (Waite, 1994). The major steps towards this end were development of a larval diet and rearing system for the host *Cryptophlebia ombrodelta* (Campbell *et al*., 1999), and changing the oviposition substrate to e-fluted cardboard rather than the grease proof paper used in South Africa (Waite, 1994). These changes allowed many more eggs to be produced and exposed to the parasitoid for stinging (Maddox *et al*., 2002). Commercialisation of the system to enable rapid deployment of parasitised egg cards to growers at the key periods within a season has been undertaken by (BioResources "Mactrix" Richard Llewellyn).

Macadamia nutborer rearing (using a modified Shorey & Hale diet (Shorey & Hale, 1965)) has continued at a reduced level at Alstonville since commercialisation of *T. cryptophlebiae* by BioResources Pty. Ltd. The laboratory colony is maintained as a source of material for research trials such as insecticide compatibility, off-farm releases testing and parasitoid impact on invading MNB populations in the coastal mangrove area. Links of *Cryptophlebia* sp. with coastal mangroves and. was first noticed in Japan (Komai & Nasu, 2003) and pursued by Huwer *et al*., (2006). Coupling pheromone lures (manufactured by Dr. Richard Vickers, Canberra) for *C. ombrodelta*, with fresh MNB egg cards as parsitoid trapping tools now provides a means to investigate the MNB populations and parasitoids.

MNB parasitoid releases into orchards around NSW remain necessary because *T. cryptophlebiae* cannot survive temperatures below 13°C during winter in the orchard microclimate. However, moths and parasitoids remain active in the estuarine areas, where the microclimate does not go below 13°C (in mangroves) and parasitoid releases into these areas may have a significant effect on subsequent invading MNB populations in orchards later.

15.4.1. Materials and Methods

The colony of MNB larvae is usually maintained at 25°C +/- 3°C and 50% relative humidity. The original production target from each diet tray container was 200 live MNB pupae The MNB larvae population originated from either a "fresh egg card" where the moths laying on that card were no more than 2 days old, or a "reset egg card" on which the eggs laid came from moths at least 4 days old. (Figure 15.4.1.1). MNB moths (20-40 moths per container) were fed a 1% sucrose solution (on a dental wick) in a 280ml polycarbonate container lined with an e-flute cardboard strip as an oviposition substrate OH&S issues with fine wing scales required a separate area for the moth colony where staff used gas filtered masks when handling the moths. Moth scales on the egg cards were removed using compressed air before the egg cards were returned to the diet trays or delivered to the parasitoid sting chambers (Figure 15.4.1.3). Twice a week, 2 "reset" and 2 "fresh" egg cards, were taken from the adult moths cups, placed on fresh diet trays and labelled (container number, dates and type of egg card). After 10 days the cards were removed and at 28 days the MNB pupae were extracted (using water and sieves). Pupae were dried on absorbent

paper and weighed to estimate number (Campbell *et al.*, 1999). Pupae were placed in the flight cages to emerge as moths 8 days later. Egg cards were storable and still acceptable for parasitism at 13℃ for up to seven d ays and for up to 40 days at 5℃ (+/- 3℃).

The *T. cryptophlebiae* colony (Figures 15.4.1.2 to 15.4.1.4) was maintained in two rooms to ensure a backup source. A change-over from rolls of e-fluted cardboard to the pre cut and perforated cardboard (Richard Llewelyn (BioResources Pty. Ltd.)) was made in 2008. Excess parasitised egg cards were available to growers.

Data on all release sites and the emergence rates of the wasps from the host eggs were recorded in the following manner:

1. the date the MNB eggs were laid
2. the date MNB eggs were introduced to the parasite oviposition chamber
3. how long the parasite oviposition chamber was in use
4. whether the MNB egg cards were from fresh or reset moth cups

Samples of parasitised MNB egg cards were checked (under 12x magnification) to determine the proportion of parasitised eggs with wasp emergence holes or where wasps had failed to emerge. A minimum subsample of 10 cards with parasitised MNB eggs were examined per month. The key to a successful *T. cryptophlebiae* colony is to maintain a high fresh host egg density while the adult wasps are active. The main colony is caged in 2 litre glass jars with 18 micron quarantine mesh ventilation lids, two smaller colonies are caged in 280ml and 30ml vials respectably as backups to the main colony. Each MNB egg normally yields up to 3 wasps. MNB eggs containing up to 8 wasps have been dissected. This is a sign of super parasitism that can cause the colony to decline if the imbalance of host and parasitoid is not corrected.

Figure 15.4.1.1:

Top left: The *Cryptophlebia ombrodelta* colony showing 20-40 MNB moths on e-fluted corrugated cardboard lining polycarbonate cups and a dental wick saturated with 1% sucrose solution.

Top right: *Cryptophlebia ombrodelta* larvae tunnelling into the Shorey Hale diet trays after 10 days exposure to the freshly laid MNB egg cards.

Bottom left: *Cryptophlebia ombrodelta* pupal extraction.

Bottom right: *Cryptophlebia ombrodelta* moths emerging from pupae in the flight cage. .

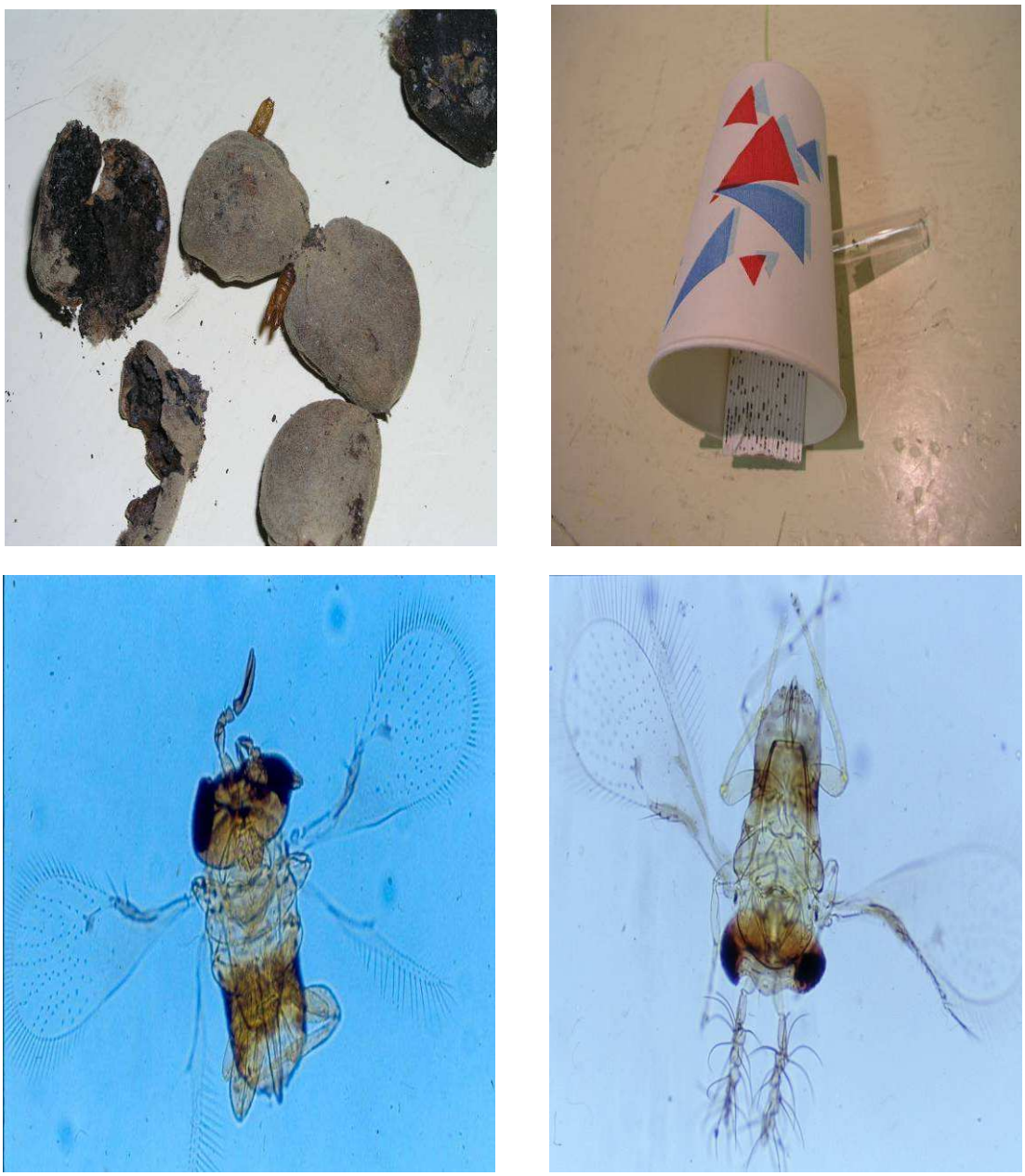

Figure 15.4.1.2:

Top left: *Cryptophlebia ombrodelta* damage to Mangrove seed pods

Top right: Release cups in trees provide shelter for parasitoids at emergence. TAC GEL® on the string prevents ant predation of eggs. A strip of parasitised *Cryptophlebia ombrodelta* eggs is attached with a paper clip to the inside of the cup. The polycarbonate vial holds *Amblypelta nitida* eggs containing *Centrodora darwini* parasitoids.

Bottom left: Female *Trichogrammatoidea cryptophlebia* (200x magnification)

Bottom right: Male *Trichogrammatoidea cryptophlebia* (200x magnification)

Figure 15.4.1.3:

Top left: Pheromone traps in macadamia orchard at height. Traps were suspended at 3metres on lengths of irrigation pipe.

Top right: Male *Cryptophlebia ombrodelta* moths caught on a TAC GEL® plate (7 days exposure).

Bottom left: Pheromone traps for *Cryptophlebia ombrodelta* in mangroves east of Brisbane airport

Bottom right: *Cryptophlebia ombrodelta* moths caught using a pheromone, over a 7 day period in an estuarine area well removed from any macadamias.

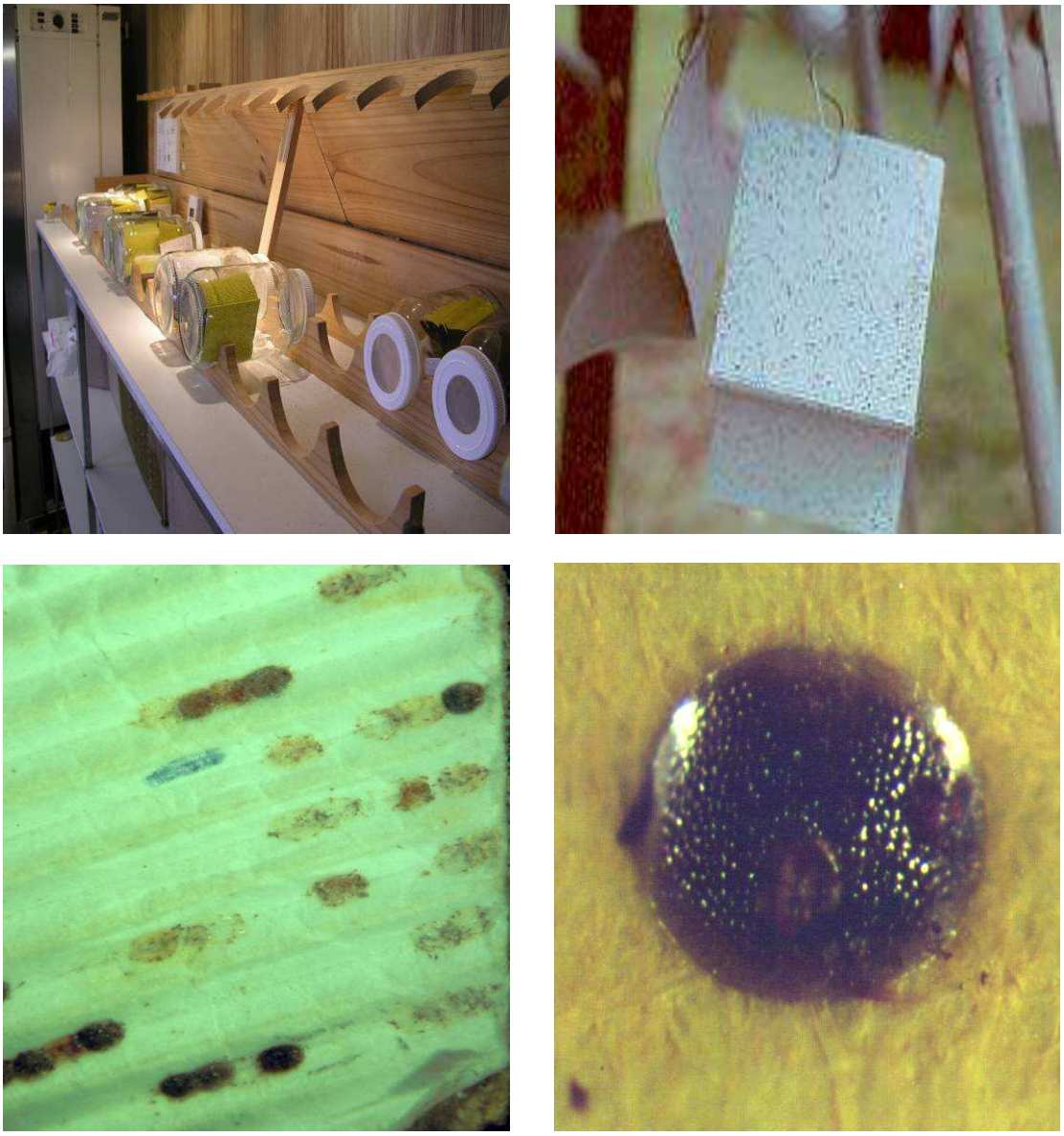

Figure 15.4.1.4:

Top left: The egg parasitoid colony rack showing wasp oviposition chambers.

Top right: Parasitised *Cryptophlebia ombrodelta* egg cards used in dispersal studies were hung or stapled to leaves or branches (if the ant populations were low).

Bottom left: Developing *Trichogrammatoidea cryptophlebia* inside MNB eggs (MNB eggs usually go black after 5 days parasitism). Empty MNB eggs indicate no parasitism.

Bottom right: *Trichogrammatoidea cryptophlebia* emergence holes chewed through the egg chorion (20x magnification) and an indicator of a successful wasp release.

15.4.2. Results

The production levels for the *C. ombrodelta* colony from 2002 to 2010 are given in Table 15.4.2.1. There was no detectible difference in oviposition and resulting pupal numbers extracted from older moths. There is higher output during the period 2004 to 2007 compared to the other years, which may be due to subtle differences in the larval diet ingredient formulations as a result of changing suppliers. Overall, >1.5 million pupae were produced over that period at an average of 355 per diet tray. The seasonal production data shows that diets from May-September were more productive (Table 15.4.2.2) and that most diet failures occur during summer. This is probably due to cooler weather restricting ferment flies that foul some diets and that the higher humidity in autumn-winter favours first instar MNB larvae survival.

The wasp colony has averaged 610 extra cards each year between 2006 and 2010 (Table 1.5.4.2.3) with production rates maintained using half the wasp oviposition chambers that were used in 2006. Excess parasitised MNB egg cards were used to "seed" live wasps into estuarine regions where field MNB populations are perennial. In 2007 wasps were released in the river areas in mid spring, just prior to orchard releases, while in 2008 wasps were released in autumn and winter at the rivers. In 2009 and 2010 releases were made autumn through spring to coincide with the fruiting of mangroves (Table 15.4.2.3). During summer excess cards were used to "seed" areas where MNB have become a problem in the orchards. The numbers of cards used in NSW are shown in Table 15.4.2.3.

The emergence rates from the wasp release cards averaged around 70% (Table 15.4.2.4). Fresh moths were no better an egg source than the older reset moths (Table 15.4.2.4). The day the cards were presented to the egg parasitoids (between day 7 and 16 in the wasp breeding cycle) also had no bearing on the emergence rate of wasps (Table 15.4.2.5). Some cards placed in the wasp oviposition chamber early did show signs of "superparasitism" (when the host egg densities were too low for the available wasp population). Poor parasitism of egg cards (less than 1%) is a far more common fault of the system. This usually occurs on egg cards introduced before the wasps were ready for oviposition. The cold storage of host eggs at (5℃ +/- 3℃) to feed to the egg parasitoids appeared useful although we recommend not to exceed 1 month storage t(o avoid egg desiccation).

Trapping of wasp parasitoids in estuarine areas showed that, in areas where wasps were released, recapture was common. In the North creek region around Ballina, Brunswick Heads, Wooyong and Yamba parasitoids were detected in all months except August (see Figure 15.4.2.1). In other area wasps were detected in low numbers from early spring through until early winter the following year. Highest catches were in autumn where releases were made (Figure 15.4.2.1). From this result it is suggested that wasps be released only in winter/spring (to parasitise the MNB generation which infests orchards each year in late spring.

Table 15.4.2.1: *Cryptophlebia ombrodelta* pupal production (at CTH Alstonville) from diet trays using eggs from freshly collected moths and older (Reset) moths (2002-2010).

Egg source	**Data**	**2002**	**2003**	**2004**	**2005**	**2006**	**2007**	**2008**	**2009**	**2010**	**Grand Totals**
Fresh moths (2-4day old)	Pupae	59796	64092	117044	115834	105511	105503	57572	63927	67192	756471
	Trays	260	311	276	247	212	206	195	205	206	2118
Reset moths (>5 day old)	Pupae	70668	80377	116596	113600	95190	105141	62003	70892	71804	786271
	Trays	317	347	276	221	205	213	184	210	214	2187
Totals	Pupae	130464	144469	233640	229434	200701	210644	119575	134819	138996	1542742
	Trays	577	658	552	468	417	419	379	415	420	4305*
Average per diet (sd)	Pupae	226 (177)	220 (153)	423 (240)	490 (289)	481 (279)	503 (292)	316 (185)	325 (210)	331 (177)	358 (249)

Table 15.4.2.2: Seasonal patterns to the pupae numbers recovered from the *Cryptophlebia ombrodelta* colony at CTH Alstonville 2002-2010. The colony is usually maintained at 25°C +/- 3°C and 50% RH. Data is the frequency of diet trays with pupal extraction values between the limits listed.

Pupae collected	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Grand Totals
0-20	28	17	30	19	4	4	3	13	3	20	25	40	206
21-50	13	34	33	24	7	6	9	12	12	17	18	24	209
51-100	35	35	49	20	24	9	14	41	25	30	28	43	353
101-150	40	22	42	29	38	18	11	17	27	24	29	46	343
151-200	32	20	30	19	39	23	15	22	38	37	36	31	342
201-300	59	40	67	60	47	35	50	32	54	67	58	71	640
301-400	40	42	48	46	45	42	41	35	41	39	50	59	528
401-500	49	50	25	42	24	36	38	33	44	56	41	50	488
501-600	27	37	22	32	40	41	52	34	40	49	38	25	437
601-700	13	22	13	17	34	46	43	32	28	25	21	17	311
701-800	18	13	9	24	25	29	33	43	25	22	22	12	275
801-900	5	7	3	7	18	25	15	20	17	9	9	3	138
901-1000	1	1	1	0	6	10	12	11	4	2	0	4	52
> 1000*	0	0	0	1	1	10	6	9	0	0	1	0	28
Totals	360	340	372	340	352	334	342	354	358	397	376	425	4350

* Maximum pupal extraction from a single diet tray was 1207, the eggs were laid between 4/8/2006-7/8/2006 tray extracted 4/9/2006

Table 15.4.2.3: Pattern of production of *Trichogrammatoidea cryptophlebiae* egg cards from the colony at CTH Alstonville (2006-2010). The colony is usually maintained at 25°C +/- 3°C and 50% RH. Data is the total number of extra cards produced each month available for release, number of cards released into the mangrove districts, the number of cards distributed to macadamia growers and the average number of sting chambers (ventilated 2 litre bottles) used each week to house the colony. Data does not include cards used to maintain the colony or those used in on site trials.

Year	Data	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Grand Totals
2006	Total cards	60	96	79	32	13	40	0	49	74	52	38	84	617
	Estuary release			8	8	8	8	8	8	8	8	8	8	80
	Farm release													
	Sting chambers	14	15	12	10	9	9	7	9	10	10	10	15	11*
2007	Total cards	37	83	108	96	24	39	25	0	0	38	103	66	619
	Estuary release	8	8	8	8	8	8	8	4		8	8	8	96
	Farm Release											6	2	8
	Sting chambers	11	10	10	10	5	5	6	10	5	9	10	10	8*
2008	Total cards	45	49	71	17	40	51	48	38	56	50	47	68	580
	Estuary release		4	4			4							12
	Farm release									3	6	25	26	60
	Sting chambers	10	10	10	6	6	6	6	6	6	6	6	6	7*
2009	Total cards	93	64	70	53	23	20	37	61	61	14	47	55	598
	Estuary release						4	1	2			1		8
	Farm release												7	35
	Sting chambers	6	6	6	6	6	6	6	6	6	6	6	6	6*
2010	Total cards	56	57	53	41	57	9	48	50	58	69	70	73	641
	Estuary release					10	4	2	2					18
	Farm release		13					2	3	6			15	39
	Sting chambers	6	6	6	6	6	6	6	6	6	6	6	6	6*

* Average number of sting chambers used each week to produce the wasp cards

Table 15.4.2.4: Seasonal patterns to the % emergence rate of *Trichogrammatoidea cryptophlebiae* from *Cryptophlebia ombrodelta* egg cards from the colony at CTH Alstonville 2006-2010. The colony is usually maintained at 25^{o}C +/- 3^{o}C and 50% Relative humidity. Data is the average of the emergence rate from at least 10 samples taken from the cards collected each month following the twice weekly clean out of the sting chambers.

Year	Data	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Overall average
2006	Freshly laid	82	77	74	66	48	35	57	82	73	73	74	75	67
	(s.d)	(6)	(8)	(11)	(13)	(22)	(18)	(18)	(7)	(12)	(7)	(10)	(11)	(19)
	Reset cards	75	74	71	64	46	22	50	75	74	73	69	65	63
	(s.d)	(8)	(8)	(8)	(17)	(19)	(8)	(10)	(2)	(5)	(15)	(5)	(11)	(18)
2007	Freshly laid	71	85	90	57	84	40	32	65	68	80	86	79	72
	(s.d)	(15)	(7)	(2)	(12)	(8)	(6)	(16)	(10)	(15)	(10)	(5)	(7)	(20)
	Reset cards	85	86	76	63	87	35	27	63	65	79	77	78	65
	(s.d)	(1)	(6)	(17)	(10)	(5)	(14)	(11)	(6)	(12)	(3)	(8)	(12)	(22)
2008	Freshly laid	86	77	83	68	77	71	57	62	80	85	86	96	77
	(s.d)	(6)	(14)	(11)	(17)	(12)	(10)	(15)	(15)	(7)	(6)	(5)	(2)	(15)
	Reset cards	82	75	72	73	78	55	44	56	80	82	83	91	73
	(s.d)	(10)	(10)	(0)	(9)	(7)	(18)	(18)	(22)	(7)	(9)	(8)	(6)	(18)
2009	Freshly laid	89	N/A	N/A	26	83	66	60	46	70	74	91	88	69
	(s.d)	(8)			(30)	(7)	(17)	(26)	(32)	(27)	(17)	(7)	(5)	(28)
	Reset cards	78	N/A	N/A	52	69	56	58	60	65	61	86	85	67
	(s.d)	(23)			(34)	(13)	(16)	(21)	(24)	(33)	(4)	(9)	(14)	(24)
2010	Freshly laid	88	91	83	83	79	58	65	75	79	88	88	90	81
	(s.d)	(6)	(5)	(15)	(14)	(8)	(21)	(18)	(17)	(24)	(3)	(15)	(7)	(17)
	Reset cards	72	92	71	82	57	61	85	67	65	76	91	91	75
	(s.d)	(11)	(6)	(24)	(7)	(16)	(20)	(6)	(9)	(30)	(12)	(5)	(8)	(19)

Table 15.4.2.5: Parasitoid performance when presented with fresh host eggs on different days in the wasp cycle (day number). Measured as % emergence of *Trichogrammatoidea cryptophlebiae* from *Cryptophlebia ombrodelta* egg (Alstonville 2006-2010). Data is the average of the emergence rate from at least 10 samples taken from the cards collected each month.

Day number in wasp cycle	**Number of samples examined**	**%emergence**	**(s.d)**
5	1	71	0
7	6	41	17
8	19	71	14
9 (usual emergence)	125	66	24
10 (usual emergence)	101	64	22
11	86	71	20
12	225	73	20
13	18	74	12
14	106	75	18
15	4	61	25
16	2	68	13

Table 15.4.2.6: Effect of storage *Cryptophlebia ombrodelta* egg cards at 4°C +/- 3°C on the % emergence rate of *Trichogrammatoidea cryptophlebiae* The colony is usually maintained at 25°C +/- 3°C and 50% Relative humidity. "Egg age" is the maximum age of the eggs when presented to the parasitoids.

Egg Age (days)	Number of cards	% emergence	(s.d)
2	359	71	19
3	165	70	21
4	31	71	20
5	39	71	18
6	4	62	20
7	32	55	31
8	2	69	4
9	10	52	29
10	6	63	32
11	5	80	12
12	5	62	31
13	1	81	0
14	9	74	17
15	2	70	1
16	2	55	45
17	1	96	0
18	1	81	0
19	2	92	1
20	1	58	0
21	1	76	0
23	2	73	14
25	1	78	0
28	1	96	0
30	2	82	15
32	1	58	0
33	2	93	2
35	1	63	0
42	1	69	0
90	1	78	0

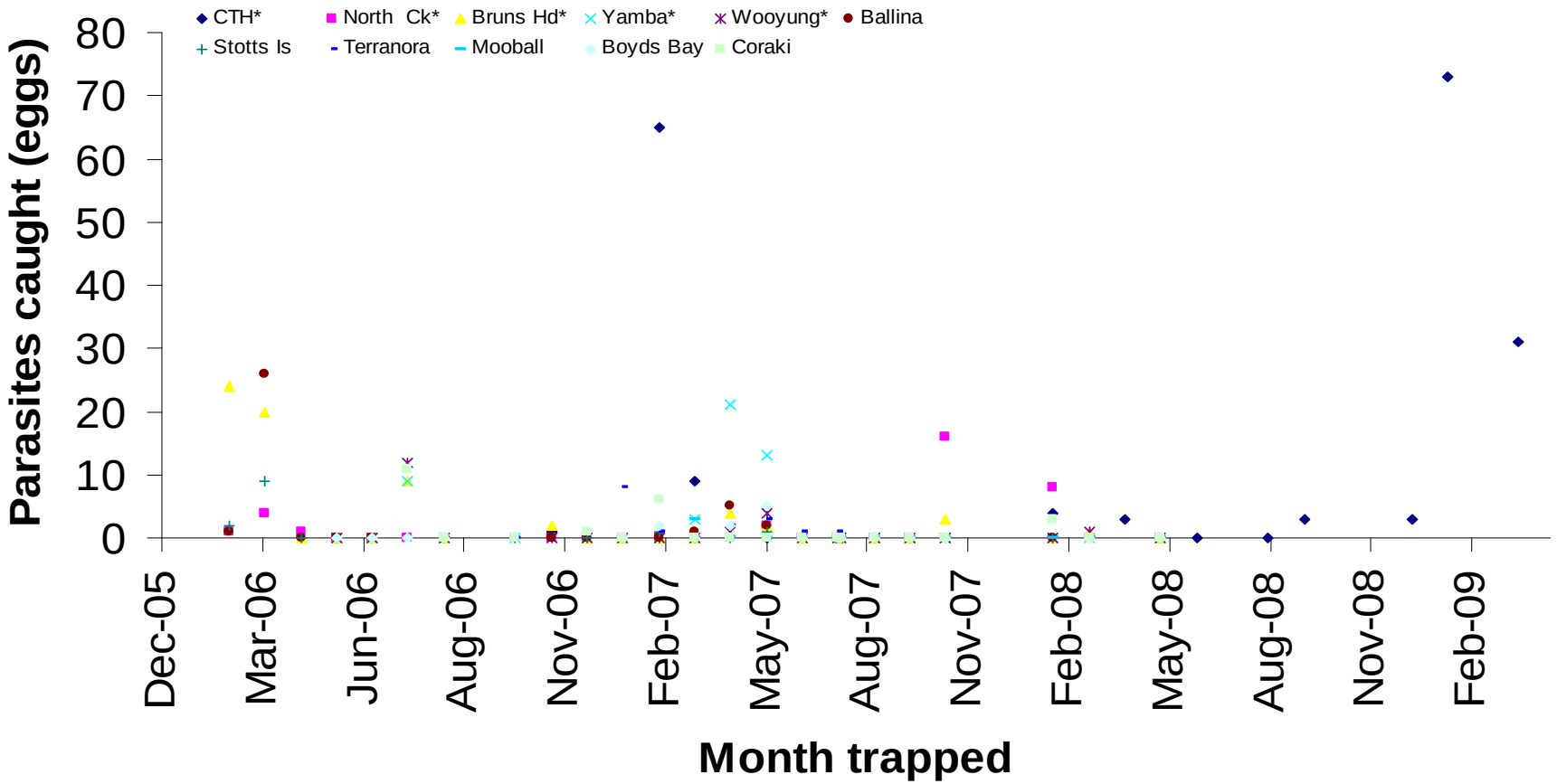

Figure 15.4.2.1: Numbers of parasitised *Cryptophlebia ombrodelta* eggs collected off trap MNB egg cards placed (for 1 week) at estuarine locations in mangroves in Northern NSW Sites with * have had wasps released at the trapping area

15.4. Appendix 5: Insecticide trial on felted coccid

Felted coccid, *Eriococcus ironsidei* Williams (Hemiptera: Eriococcidae) has been identified as a problem for a number of macadamia growe.rs in Queensland. A management option for this pest was therefore requested.

We endeavoured to set up a spray trial for management of felted coccid at CTH Alstonville in 2009. This needed a significant population to detect any treatment effect. This pest is only present at a low population density at CTH with natural parasitism. In order to set up the trial we needed to artificially enhance the felted coccid population by monthly applying beta-cyfluthrin starting in March 2009.. Despite this effort, we were not able to induce a population we could work with for a field trial. Two major climatic events did impact heavily in that season. Firstly a major storm with wind gusts of 140km/h hit the Alstonville Platoe in late May 2009, causing extensive damage. The following September dust storms occurred during flowering compromising most macadamia nutset experiments and our felted coccid population levels had failed to reach treatable level. We therefore had to abandon this, as the issues of macadamia lace bug and banana fruit caterpillar gained priority.

There are no doubt still issues with felted coccid which will be dealt with once the macadamia lace bug issue is solved.

Annecdotally, growers have found success using imidacloprid (Confidor®)off lable on non bearing trees in the Bundaberg district to control felted coccid. A trial may be necessary to legalise the option for the industry. Bayer 092 may actually an even better option in this case, as well. At present, finding minimum spray options for macadamia lace bug and scirtothrip attacks should be our priority to maximise production.