# Final report

Project title:

# Managing Flies for Crop Pollination

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### **Public summary**

Approximately 12 billion dollars/year of crop production in Australia is at least partially reliant on insect pollination. Honeybees provide most of this service, but over-reliance on a single species carries significant risk. Honeybee populations are under pressure from pests, colony collapse disorder, pesticide usage and climate change. These challenges are compounded by sustained growth of pollination dependent industries, declining apiarist numbers in Australia, and the understanding that no single species can optimise pollination in all crop types. In this context, access to a more diverse range of managed pollinators is important to protect and future-proof Australia's pollination-dependent industries.

Flies are the second most important pollinators after bees, but our knowledge of their role in crop pollination is limited and the potential to develop them as managed pollinators is mostly unexplored. Flies offer a good alternative or complementary option to bees because they occur in many regions and climates, are regular flower visitors and many can be mass reared.

This pioneering national project is the first major step towards developing fly pollinators for Australian horticulture, with teams from Department of Primary Industries and Regional Development (WA), University of Western Australia, University of New England (NSW), Western Sydney University, seedPurity (TAS) and industry stakeholders collaborating to:

- Improve understanding of the diversity, importance and habitat needs of fly pollinators in Australian horticulture;
- Evaluate efficiency of flies for pollination of model crops;
- Develop rearing and deployment protocols for promising fly pollinators; and
- Identify habitat augmentation measures to promote beneficial flies in crops.

Research conducted on avocados in WA and QLD, berries in NSW and TAS, mangos in QLD and NT and vegetable seed crops in TAS, SA and NSW highlighted the diversity of fly pollinators visiting horticultural crops and their contribution to crop production. We identified several Calliphorids (blowflies) and Syrphids (hoverflies) that visited a broad spectrum of crops in different production regions, performed well in pollinator efficiency evaluations and showed good potential for mass rearing.

Scalable rearing protocols were developed for *Calliphora vicina*, *C. dubia* and *Eristalis tenax*, and these were reared for evaluation across different crop types. In cage, polytunnel and glasshouse trials, *C. dubia*, *C. vicina* and *E. tenax* effectively pollinated avocados and blueberries, while *E. tenax* and *C. stygia* pollinated glasshouse strawberries, and *E. tenax* showed significant promise as a managed pollinator of blackberries, sweet cherries and vegetable seed crops. Open field experiments in vegetable seed and cherry crops (TAS) determined deployment protocols for *E. tenax*, ultimately demonstrating improved yields from complementary stocking of *E. tenax* and honeybees in both crop types.

Provision of habitat for fly pollinators, such as the use of 'stink stations' in mangos and habitat pools in seed carrot, promoted fly pollinator numbers, but this did not always translate to improved yields.

This comprehensive body of work highlights the importance of fly pollinators to Australian horticulture, the opportunity to develop flies as alternative managed pollinators to complement honeybees, and the importance of conservation and habitat augmentation measures to promote wild fly pollinators in agricultural landscapes.

# **Keywords**

Fly pollination, managed pollinators, alternative pollinators, complementary pollinators, crop pollination, vegetable seed pollination, cherry pollination, mango pollination, avocado pollination, blueberry pollination, raspberry blackberries, strawberry pollination, protected crop pollination, fly rearing, *Eristalis tenax, Calliphora stygia, Calliphora vicina, Calliphora dubia, Eristalinus punctulatus.* 

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Final report – Managing Flies for Crop Pollination



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## Introduction

Over 80% of the crops around the globe are either dependent on or have their yield enhanced by insect pollination (Aizen *et al.*, 2019). The annual gross economic value of crops requiring pollination services exceeds AUD\$9 billion in Australia and is estimated at USD\$780 billion worldwide (Hafi *et al.*, 2012). Managed and feral European honeybees (*Apis mellifera*) provide the vast majority of crop pollination services globally, including in Australia, with >90% of crops that depend on insect pollination serviced by honeybees (Free, 1993). Managed stingless bees and wild pollinator species provide the balance of insect-mediated crop pollination in Australia. This over-reliance on a single species carries significant risk, particularly since honeybee populations are under increasing pressure from pests such as varroa mite (*Varroa destructor*) and small hive beetle (*Aethina tumida*), as well as other threats including colony collapse disorder, climate change, pesticide usage and changing land use patterns (Cunningham *et al.*, 2002). For example, the recent arrival of varroa mite in Australia will inevitably devastate wild honeybee populations and, as a result, substantially impact pollination of some crops. This is likely to significantly increase the demand for managed honeybee pollination services in affected areas potentially restricting short- to medium-term availability of hives for pollination. Additionally, the continuing expansion of pollination-dependent industries (Clarke and Le Feuvre, 2022), combined with a decline in apiarist numbers and competing demands for honeybees for honey production and crop pollination will further exacerbate supply and demand challenges relating to managed pollinators.

An additional challenge faced by pollination-dependent industries is that honeybees, although generally efficient pollinators, are not well-suited to all crop types and cropping systems. Attracting and retaining foraging honeybees is difficult in some crops, while covered cropping systems, for example, can reduce honeybee efficiency (Evans *et al.*, 2019). Stocking honeybees in these environments can impact colony health and present a significant occupational health and safety risk for employees. In this context, ensuring access to a more diverse range of managed pollinators and abundant wild pollinator populations is crucial to protect and future-proof pollination-dependent industries.

Flies are one of the most diverse animal groups in the world and the second most important pollinator group after bees (Free, 1993; Larson *et al.*, 2001; Ollerton *et al.*, 2011; Rader *et al.*, 2020). As pollinators, flies are likely to represent a good alternative or supplemental option to bees, because different species are present all year round and they frequently visit flowers to feed on nectar and/or pollen to support key biological functions including flight and reproduction (Norris, 1965). Being hairy, they also pick up and move pollen from a wide variety of flowers (Stavert *et al.*, 2016). Fly taxa are highly variable in body size, allowing this to be matched to the floral morphology of a target crop for pollination — either via species selection, or within species by manipulating nutrition of the larval stage (Ireland and Turner, 2006). In addition, some fly taxa are easily mass reared with reasonably low inputs, have manageable health and safety requirements, and present negligible risk of disease transmission to existing managed and wild pollinators when reared under controlled conditions. Furthermore, they do not sting farm workers. However, although flies are recognised as being equally efficient to (or sometimes better than) bees for pollinating some crops (Ssymank *et al.*, 2008; Jauker and Wolters, 2008; Albano *et al.*, 2009; Orford *et al.*, 2015), and are often responsible for transporting high pollen loads in both natural and modified systems (Rader *et al.*, 2009; Orford *et al.*, 2015), large gaps exist in our knowledge of flies in crop pollination and their potential application as managed crop pollinators (Ssymank *et al.*, 2008).

This pioneering project was undertaken as the first major step towards developing managed fly pollinators as a strategy to improve pollination outcomes and reduce risks associated with over-reliance on honeybees in pollination-dependent horticultural industries in Australia. It involved a collaboration between Hort Innovation, pollination-dependent horticultural industries and five research organisations with expertise in managed pollination of horticultural crops, Dipteran entomology and pollination ecology:

- Department of Primary Industries and Regional Development (WA)
- University of New England
- seedPurity P/L
- University of Western Australia
- Western Sydney University

Several participating horticultural businesses were also integral to the project, including South Pacific Seeds, Bejo Seeds Australia and their subsidiary business Tasmanian Pollination Services, Biological Services, Costa, Reid Fruits, Jasper Farms Delroy Orchards. Avocado and mango growers in NSW, north Queensland and Northern Territory regions generously allowed access to their properties to conduct research on wild and managed pollinators.

**Hort Innovation** 

The broad aims of the project were to:

- 1. Improve understanding of the diversity, abundance, and lifecycle and habitat requirements of Dipteran pollinators in horticultural production systems in Australia;
- 2. Evaluate the efficiency of promising fly pollinator species for pollination of model horticultural crops in protected and open cropping systems;
- 3. Model developmental rates and develop rearing protocols for selected fly species displaying potential as managed pollinators;
- 4. Understand dispersal and retention of mass reared flies in model protected and open crops; and
- 5. Develop simple habitat augmentation strategies to promote beneficial fly numbers in cropping systems.

# **Fly Pollination of Avocados**

#### DPIRD (Western Australia research) and UNE (Queensland research)

David Cook and Shoaib Tufail; Romina Rader, Abby Davis, Lena Schmidt and Blake Dawson





# Introduction

Avocado production is rapidly increasing in Australia from 78,000 tonnes in 2021 (farmgate value of \$489M) to 150,000 tonnes in 2024 (farmgate value of \$590M) with total production area continuing to expand (Australian Horticulture Statistics Handbook 2023/24 Hort Innovation Australia). Avocado production occurs mainly in Queensland, northern New South Wales, the Tri-State region and south-western Western Australia (>850 growers nationally). Avocados require insect pollination and although managed honeybees are the most commonly used insect to achieve fruit set, avocado flowers are not particularly attractive to honeybees and no honey product is derived when placed in avocado orchards (Clarke and Le Feuvre, 2022). Insects facilitate avocado pollination, leading to increased fruit production, and yield improvements through improved pollination is evidenced (Dymond *et al.*, 2021). As with many insect-pollinated crops, avocado yields are at risk due to widespread pollinator declines (Biesmeijer *et al.*, 2006; Potts *et al.*, 2010), which has intensified the need to identify alternative insect pollinators.

In Australia, various Calliphorid species have been recorded visiting avocado flowers and may play a significant role in their pollination (Howlett, 2017; Cook, *et al.*, 2020). These include *Calliphora stygia*, *Calliphora augur*, *Calliphora vicina*, *Chrysomya rufifacies*, *Lucilia sericata*, *Lucilia cuprina* (Howlett, 2017)and *Chrysomya varipes* (Vithanage, 1986). Early findings in this project identified that the blow fly *Calliphora dubia* forages on avocado flowers three times more often than *Calliphora albifrontalis* (given equal access to inflorescences), which may explain their higher pollination rates of Hass avocados in paired-tree enclosures (Cook *et al.*, 2023). Both blow fly species improved avocado yield compared to no insect pollinators, with *C. dubia* enabling yields up to two-thirds of those produced on trees in the orchard (not in enclosures) and pollinated by managed honeybees.

Orchards typically have Type A (Hass) and Type B (e.g., Edranol, Ettinger) cultivars to promote cross-pollination and synchronise flowering of male and female flowers, with Type B trees around 8-11% of all trees. The literature suggests a honeybee hive density for avocado pollination of 2 to 3 hives/ha. Increasing hive density from 2 to 3 hives/ha has been shown to increase average fruit weight (Keogh *et al.*, 2010). As few as 2 hives/ha are used when there are unmanaged honeybee populations (feral bees), native bee and blowfly populations available to the grower (Garibaldi *et al.*, 2011; Osterman *et al.*, 2021). However, the arrival and inevitable spread of varroa mite on the east coast of Australia will eliminate many feral honeybee colonies and ultimately result in an increasing demand for managed pollination services within the avocado industry. Reliance on pollination from wild insects in the orchard is optimistic as the abundance of terrestrial

insects is estimated to be declining by around 10% per decade (Van Klink et al., 2020; Zattara and Aizen, 2021).

Avocado yields (like many insect-pollinated crops) are being increasingly affected by global pollinator decline (Biesmeijer *et al.*, 2006; Potts *et al.*, 2016). The need to optimise avocado yields is increasingly important as demand for this fruit is rising with 32.6M tonnes produced from 1999–2008 and 50.4M tonnes from 2009 to 2018 globally (FAO, 2020). In some avocado growing regions, expansion is having adverse environmental impacts (e.g., biodiversity decline, water depletion) (Magrach and Sanz, 2020), hence improving sustainable avocado production is crucial. More recent studies have examined the impact of either maintaining native vegetation around commercial orchards or establishing strips of flowering plants throughout the orchard. These strategies increase avocado production in trees in Chile most likely due to the increased flower visits by flies and other wild insects (Muñoz *et al.*, 2021). The use of wild and managed pollinators can supplement honeybees when other flowers are in bloom at the same time, to reach substantial pollination.

The industry average from a survey of growers is a honeybee hive density of 3.5 hives/ha. Most (66%) avocado growers are concerned with the price charged by beekeepers, supply shortages, hive quality and placement of hives within the orchard (Clarke and Le Feuvre, 2022). Urgent research priorities in relation to pollination are the use of native pollinators to fill the gap in honeybee availability, and cost and managing alternate pollinators such as flies.

## **Methods**

#### **Flower Visitation Studies**

#### Western Australia

Field surveys were conducted across two avocado orchards to determine what insect species were visiting avocado flowers. At 3 times during the avocado flowering period, i.e., the first, third and fifth week of flowering, five (5) trees within the same row along 11 different randomly chosen rows were visually observed both in the morning (0800–1200) and the afternoon (1230–1700). This was done at two avocado orchards (10 km apart near Busselton, Western Australia ( $-33^{\circ}44'17.919''$  S, 115°25′35.16″ E and  $-33^{\circ}38'23.75''$  S, 115°28′30.36″ E) over the 2018 and 2019 flowering seasons. A minimum of 2 min and up to a maximum of 3 min (depending upon the numbers of insects seen on the tree) was spent on a single avocado tree visually counting the number and species of insects in contact with and/or feeding on any avocado flowers. By moving through each quarter view of the tree in one direction, this minimised the chances of double counting any insects. Prior to the flower surveys, specimens of each insect species seen feeding on the flowers were caught and identified to at least family for photo identification reference guides during the transect surveys. Where unknown taxa were observed during transect surveys, a coded identification and a representative specimen was collected for later identification to fly family (according to Marshall *et al.*, (2017)). Other non-Dipteran insects found feeding on avocado flowers were also recorded. An entire day of observations between 8 am and 5 pm was done each week across two orchards and pooled for analysis into frequency of observations.

#### Queensland

Six (6) field sites were located at four large, commercial farms owned by third-party landowners located in the Atherton Tablelands, Queensland. All farms were located approximately 50 km from Mareeba, QLD, were located at least 500 m apart and grew commercial avocado trees (cultivar: 'Shepard'). Permission to conduct fieldwork on all sites was granted by either the farm owners or managers. All farms brought managed honeybees into fields to perform pollination services.

To identify the abundance and diversity of insects visiting avocado flowers, visitation surveys were conducted at six field sites selected across the four farms for a total of two to three days (at each site) from 2<sup>nd</sup> August 2022 to 25<sup>th</sup> August 2022 (peak bloom). Surveys were carried out on days with no rain and when temperatures were at least 15°C. Temperature, relative humidity (RH), and wind speed were recorded from nearby weather stations before each survey was conducted. For consistency, surveys were conducted along two, 10 m transects: one along the edge of the avocado orchards and an additional walk towards the middle (> 30 m within orchard rows). All transects were conducted walking slowly (one min per m) while looking at one row of avocado trees, with the observer looking for insects on flowers only from ground level to a height of 2 m. Edge and middle transect walks were at least 50 m apart. Insects were collected in the field for identification using keys or expert aid.

#### **Avocado Tree Enclosure Studies**

Six avocado tree enclosure studies testing different fly species' ability to pollinate Hass flowers were carried out from 2018-2024 (inclusive). The first two enclosure studies looking at the pollination ability of the endemic western golden-haired blowfly *C. albifrontalis* and the western blue-bodied blowfly *C. dubia* were tested within paired tree enclosures (Figure 1, a). The paired enclosures were spread throughout the orchard where a Type A Hass tree was enclosed alongside a Type B polliniser tree (Ettinger or Edranol) (Figure 1, b).



#### Figure 1: a) Paired-tree enclosures of Hass and Ettinger avocado trees, b) distribution throughout the orchard, c) multitree enclosures, Busselton, and d) Pemberton.

The third year (2020) of avocado tree enclosure studies looked at using larger mesh enclosures to give the trials a more meaningful result, with both a small 'nuc' hive of bees in an enclosure compared with flies placed into the large enclosures that covered 21 avocado trees (19 Hass and 2 Ettinger) (30m wide x 33m long enclosures - Figure 1, c). This methodology avoided the issue of trees in paired enclosures pushing up against the mesh sides of the enclosure and allowed the flies released into each enclosure the ability to forage amongst multiple trees. In addition, two trials sites were established, one at Ruabon (Jasper Farms, Busselton) and Delroy Orchards (Pemberton).

The fourth year (2021) of avocado tree enclosure studies used the same large mesh enclosures as in 2020, except that the avocado tree planting density was doubled at the Ruabon Farm site (Busselton). This included 39 avocado trees (spanning 3 rows), with 36 Hass (Type A) and 3 Ettinger (Type B) trees in the middle row within each of the 3 large enclosures (Figure 1, c). The fly species released into each of two separate enclosures were *C. dubia* and *C. vicina* (European blue bottle blowfly) that was introduced into Australia in the early 1900's and has since become established throughout Australia and is found worldwide (Figure 2). At Pemberton, 12 Hass trees were enclosed within a fly-proof mesh enclosure (Figure 1, d) and 3 separate enclosures contained either *C. vicina*, *C. dubia* or a small hive of bees.

#### **Hort Innovation**

Table 1 below provides a summary of the enclosure trials carried out during each flowering season in south-west Western Australia (Sept-Nov) on avocado orchards, and Figure 2 provides a snapshot of all the fly species tested on their ability to pollinate Hass avocado flowers to produce fruit.

Table 1: Site and year of enclosure trials assessing different fly pollination species (*Calliphora albifrontalis* (*C. albi*), *Calliphora dubia, Calliphora vicina* and *Eristalis tenax*) with predicted (italicised) and actual numbers of adult flies in each enclosure compared to honeybees (*Apis mellifera*) and open pollination treatments (control; honeybees and other insects) in avocado orchards in the south-west of Western Australia.

Sito	Year Ope	Onon	Insect Pollinator Species within Enclosures						
Sile		Open	A. mellifera	C. albi	C. d	ubia	C. vi	cina	E. tenax
	Predi Num	cted Iber	≈5k	150/tree	5k	10k	5k	10k	5k
Busselton	2018	Х	-	104					
Busselton	2019	Х	-	250					
Busselton	2020	Х	≈5k						
Busselton	2021	Х	≈5k		2,637	-	3,046		-
Pemberton	2021	Х	≈5k			-	3,244	-	-
Capel	2022	Х	-				3,899	7,899	-
Capel	2023	Х	-			11,050		9,783	3,532

*X* = All insects including managed bees in the open orchard.

5k = 5,000; 10k = 10,000 and 15k = 15,000 adult flies released in the enclosures.



Figure 2: The fly species tested for their ability to pollinate Hass avocados in either paired-tree enclosures or multitree enclosures in orchards in south-western Australia. Top L = *Calliphora albifrontalis* (Sheep blowfly); Second Row = *Calliphora dubia* (Australian sheep blowfly); Third Row = *Calliphora vicina* (Bluebottle fly); Bottom Row = *Eristalis tenax* (Drone fly). All photos were taken by David F Cook except for Top L (*C. albifrontalis* adult taken by Lochman Transparencies) and Bottom L (*E. tenax* adult taken by Matthew O'Donnell).

## Results

#### **Floral Visitation Surveys**

#### Western Australia

The first year of surveys (2018 flowering season) and monitoring insects visiting and feeding on avocado flowers showed that most avocado flowers were visited by lovebugs (*Bibio imitator*) (Figure 3). The hoverfly *Sphaerophoria macrogaster* (< 5mm) was the next most prevalent visitor followed by the blowfly *C. vicina*. The second year of surveys (2019) and monitoring of insects visiting avocado showed a shift from mostly bibionids in 2018 to mostly hoverflies and the two blowflies *C. albifrontalis* and *C. vicina* (Figure 3). Hoverflies were found mostly feeding on avocado flowers in the morning whilst the blowflies (*C. albifrontalis* and *C. vicina*) fed in the afternoon.



Figure 3: The proportion (%) of avocado flowers visited by insects (both dipteran and non-dipteran and excluding bees) in orchards in south-western WA during the 2018 and 2019 flowering seasons.



Figure 4: A pie chart representation of the insects found feeding on avocado flowers across an orchard in both the morning (LHS) and afternoon (RHS).

During the flowering season of 2020, both sites were dominated by hoverflies (>50% of all insect visits to flowers), which appeared in enormous numbers across the entire south-west of WA. The 3 species of hoverfly that were predominantly recorded were *Melangyna viridiceps, Simosyrphus grandicornis* and *Sphaerophoria macrogaster* (Figure 5).



Figure 5: The 3 dominant hover fly species throughout south-west WA in the 2020 flowering season, from L to R, *Melangyna viridiceps, Simosyrphus grandicornis* and *Sphaerophoria macrogaster* with approximate size indicated.

#### Queensland

In total, 99 floral visitation transect walks (16.5 hours) were conducted on 'Shepard' avocado trees. Out of the 770 insects observed, we identified 22 taxa (12 species and 10 morphospecies) from nine insect families (Table 2). Two species of honeybees were observed visiting avocado, including the European honeybee (*Apis mellifera*) and the Asian honeybee (*Apis cerana*). The three most common species seen visiting avocado flowers were *A. mellifera* (100 + honeybees in total; the rhinid fly, *Stomorhina discolor* (50 to 100 flies in total; and the syrphid fly, *Simosyrphus grandicornis* (50 to 100 flies in total) (Figure 6).

Table 2: Relative insect abundance of insect flower visitors recorded on avocado trees during peak bloom in the Atherton Tablelands, Queensland. Insects were identified to the lowest practical taxonomic level and sorted by pollinator group.

Orden	Family	0	Omeniae	Abundance		
Order	Family	Genus	Species	0 - 50	50 - 100	100 +
Diptera	Rhinidae	Stomorhina	Stomorhina discolor		$\checkmark$	
Diptera	Rhinidae	Stomorhina	xanthogaster	$\checkmark$		
Diptera	Calliphoridae	Chrysomya	saffrenea	$\checkmark$		
Diptera	Calliphoridae	Chrysomya	nrysomya flavifrons			
Diptera	Syrphidae	Syritta	luteinervis			
Diptera	Syrphidae	Simosyrphus	grandicornis		$\checkmark$	
Diptera	Syrphidae	Simosyrphus	bengalensis	$\checkmark$		
Diptera	Syrphidae	Eristalinus	punctulatus	$\checkmark$		

Diptera	Syrphidae	Melangyna	viridiceps	$\checkmark$	
Diptera	Tachinidae (Phasiini)		sp. 1	$\checkmark$	
Diptera	Tachinidae (Goniinae)		sp.	$\checkmark$	
Diptera	Tachinidae	Euvespivora	sp.	$\checkmark$	
Diptera	Tachinidae	Chaetoria	sp.	$\checkmark$	
Diptera	Sarcophagidae	Sarcophaga	sp.	$\checkmark$	
Diptera	Milichiidae		sp.	$\checkmark$	
Hymenoptera	Apidae	Apis	mellifera		$\checkmark$
Hymenoptera Hymenoptera	Apidae Apidae	Apis Apis	mellifera cerana	$\checkmark$	$\checkmark$
Hymenoptera Hymenoptera Hymenoptera	Apidae Apidae Apidae	Apis Apis Tetragonula	mellifera cerana carbonaria	√ √	✓
Hymenoptera Hymenoptera Hymenoptera Hymenoptera	Apidae Apidae Apidae Halictidae	Apis Apis Tetragonula Homalictus	mellifera cerana carbonaria sp.	✓ ✓ ✓	✓
Hymenoptera Hymenoptera Hymenoptera Hymenoptera	Apidae Apidae Apidae Halictidae Halictidae	Apis Apis Tetragonula Homalictus Lipotriches	mellifera cerana carbonaria sp. sp.	✓ ✓ ✓ ✓	✓ 
Hymenoptera Hymenoptera Hymenoptera Hymenoptera Hymenoptera	Apidae Apidae Apidae Halictidae Halictidae Pteromalidae	Apis Apis Tetragonula Homalictus Lipotriches	mellifera cerana carbonaria sp. sp. sp.	✓ ✓ ✓ ✓ ✓	✓



Figure 6: Common insect flower-visitors of avocado in the Atherton Tablelands, Queensland: (a) the European honeybee (*Apis mellifera* (Apidae) and (b) the snout-nosed fly (*Stomorhina discolor* (Rhinnidae). Photos taken by Abby E. Davis.

#### **Pollinator Efficiency Trials (WA)**

#### Trial # 1 (2018)

The first field trial showed that the blowfly *C. albifrontalis* was able to pollinate avocados compared with insects being excluded. Trees where insects were excluded bore very few fruit ( $\approx$  3 fruit/tree). When left open to bees and any other insects present, 254 fruit were produced by each Hass tree. The trees enclosed with *C. albifrontalis* flies produced a mean of 46 fruit, with as many as 107 fruit on one tree (Figure 7). After flowering had ended, the number of pupae that were "spent" (i.e., where the adult fly had successfully emerged) revealed that  $\approx$ 30% of the fly pupae placed into the enclosures did not emerge as they were parasitised by micro-hymenopteran insects (evident from small hole drilled into the side of the pupal case) (Figure 8). This can be eliminated in any future releases by placing the pupae in the enclosures when only 2-3 days away from adult emergence, where small, parasitic wasps are unable to kill the adult fly within the pupal case.



Figure 7: Mean number of fruitlets and final fruit harvest of Hass avocados when paired in fly-proof enclosures with either no insects present or *C. albifrontalis* flies compared with trees in the open pollinated by managed bees and other naturally occurring insect pollinators.



Figure 8: The micro-hymenopteran parasitic wasp *Tachinaephagus zealandicus* (left) and the exit hole left on a fly (right), source: https://doi.org/10.1590/1519-6984.21214)

#### Trial #2 (2019)

The second field trial fruitlet counts were significantly different between all 3 treatments (*C. albifrontalis, C. dubia* and Open) but there was no significant difference between treatments at final fruit harvest. When comparing each treatment mean, fruitlet counts were significantly higher in Open pollinated trees compared with both *C. albifrontalis* (q = 6.273) and *C. dubia*, but no different between fly species (Figure 9).



Figure 9: Mean number of fruitlets and mature fruit of Hass avocados when paired in fly-proof enclosures with either *C. albifrontalis* or *C. dubia* flies compared with trees in the open pollinated by bees in the orchard.

#### Trial #3 (2020)

This flowering season was simply a comparison between the performance of bees within an enclosure (small 'nuc' hive) and trees pollinated in the open by bees and any other insects present in the orchard during flowering. There was a massive influx of small hoverflies, which resulted in higher fruit production than usual in trees in the open orchard compared with trees enclosed with a small hive of bees (Figure 10). Trial sites were at both Busselton and Pemberton orchards.



Figure 10: Mean number of Hass fruitlets within enclosures with a 'nuc' hive of bees compared with trees in the open pollinated by managed beehives and all other insects in the orchard at both Pemberton and Busselton.

#### Trial #4 (2021)

Fruitlet counts showed that *C. vicina* pollinated equivalent numbers of flowers when compared with open pollinated trees (pollinated by bees and any other insects in the orchard) as seen in Table 3. Persistent cold and rainy periods significantly reduced the emergence of *C. dubia* adults when left in the enclosures in the pupal stage, resulting in 70% less adult *C. dubia* versus *adult C. vicina* in their enclosures. When correcting for the lower number *of C. dubia* in the enclosure so that they were equivalent to the number of *C. vicina*, then their pollination success was slightly more than open pollinated trees. At Busselton, a higher pollination rate within the bee enclosure compared to both fly and open pollination treatments was likely due to the thermal effect of the netting, increasing enclosure temperatures and promoting a wider time-period of bee foraging relative to open pollination. Within the enclosure, bees were limited to the trees available within and without the issue of competing bloom (as opposed to bees foraging in the open orchard treatment). Further, the shorter foraging distances from the hive within the enclosure compared to the position of the hives servicing the open pollination treatment may have contributed to the higher pollination observed.

At Pemberton, fruitlet counts in late January 2022 showed that *C. dubia* flies pollinated the most flowers, with fruit yield almost twice that of the open pollination treatment, which were pollinated by bees and other insects in the orchard. Pollination success by bees in the enclosure was slightly higher than pollination success within the open treatment. In contrast, *C. vicina* pollinated around three-quarters of the number of flowers than were pollinated in the open. Due to the lower number of trees assessed at this site, the treatment groups were not significantly different when statistical analyses were performed on the data (e.g. Open v *C. vicina* v honeybees).

At either field site, the fly species, *C. vicina* (Busselton) and *C. dubia* (Pemberton) were capable of pollinating avocados at a level equivalent to or higher than that accomplished by beehives placed in the orchard. The 2021 flowering season was particularly cold, and wet conditions persisted at both sites before and after flowering, pushing the start of flowering later than usual and resulting in lower than usual pollination events at both sites.

Both *C. vicina* and *C. dubia* spend an average of 30 seconds feeding on each flower they visit, which is a significant amount of time in contact with the flower and stigma, which would rub against the ventral surface of the flies during feeding. By comparison, honeybees visited flowers at the same site for an average of 5-6 seconds. Single visit pollen deposition data was collected during the 2021 avocado flowering season at both Busselton and Pemberton orchards. *Calliphora vicina* adults transferred the highest number of pollen grains in a single flower visit (1.72 grains/visit) followed by honeybees (*A. mellifera*) (1.43 grains/visit) and *C. dubia* (1.12 grains/visit). The two hoverfly species *Melangyna viridiceps* and *Simosyrphus grandicornis* transferred < 1 pollen grain per flower visit (Figure 11).



Figure 11: Single visit deposition of pollen grains by a range of fly species along with honeybees regularly found visiting avocado flowers in orchards in the south-west of WA (Data supplied by Sunil Shivananjappa, UWA).

#### Trial #5 (2022)

Fruitlet count data showed that doubling the fly density of *C. vicina* (Table 3) adults resulted in significantly higher avocado pollination (as measured by fruitlets formed), however it was still  $\approx$  50% of fruit formation expected at harvest. Problems with the emergence of all the adult *C. dubia* in each of the two enclosures they were placed in resulted in flies with crumpled wings, due to the extreme cold and high humidity. This highlighted the need for all future fly releases to be of only newly emerged adults. At final harvest, the number of fruit from trees enclosed with 10,000 flies of *C. vicina* was two-thirds the number produced by trees in the Open orchard pollinated by honeybees. The weight of the fruit harvested in each treatment indicated fruit was significantly bigger in trees placed with 5,000 *C. vicina* than in trees placed with 10,000 *C. vicina*, which were also bigger than fruit produced in the open orchard (i.e., bee-pollinated).

#### Trial #6 (2023)

All 3 fly species were able to effect significant fruit formation, particularly the newly assessed hoverfly (*Eristalis tenax*). Of the three fly species, trees in the *E. tenax* enclosures had nearly twice as many fruitlets formed compared with honeybee pollinated trees (Table 3) while trees enclosed with *C. dubia* produced 30% more fruit when compared with trees in the open pollinated by bees. The 2023 flowering period was an unusually warm and dry spring, which did not suit the blow fly *C. vicina*, which prefers cooler climatic conditions. In addition, there were very few insects seen in the avocado orchard during flowering, which indicates that any pollination by wild insects is declining, now a regularly reported event worldwide, with a decline in both biodiversity and abundance of insects in many agricultural production settings. This emphasises the need for the use of fly pollination agents to support and boost honeybee pollination to secure pollination into the future.

*Eristalis tenax* resulted in the highest mature fruit yield (73.0 ± 9.4), outperforming *C. dubia* (62.0 ± 10.8) (18% increase), open pollinated trees (48.5 ± 8.1) (50% increase) and *C. vicina* (27.4 ± 4.3) (Table 3). This trend was also reflected in mature avocado fruit weights, where *E. tenax* pollinated fruit was the heaviest compared to open pollination and calliphorid fly species tested (*C. dubia* and *C. vicina*). Table 3 also shows that *E. tenax* pollinated trees had the highest fruit yield/tree (18.4 ± 2.4 kg/tree) surpassing *C. dubia* (16.2 ± 2.5 kg/tree), open pollination (12.5 ± 1.9 kg/tree) and *C. vicina* (8.2 ± 1.3 kg/tree), respectively.

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*Eristalis tenax* produced the highest number of avocado fruit per tree (73.0), significantly (p < 0.05) outperforming open pollination (50.5), *C. vicina* (27.0), *C. dubia* (25.4) and bees (22.2). However, there was no significant difference (p > 0.05) between *C. vicina*, *C. dubia* and bees. Moreover, treatments (fly species and open pollination), fly density, site, and year as well as their interactions were all highly significant (p < 0.05) except the interaction between fly density and site, which was found non-significant (F = 1.711, df = 1, and p = 0.182) (Table 3). A fly density of 10,000 adults yielded significantly more avocado fruits (69.2) than 15,000 (43.2) and 5,000 flies (21.5) (p < 0.05), suggesting 10,000 adult flies as an optimal fly density for avocado pollination.

Table 3: Mean number of avocado fruitlets ( $\pm$  s.e.) 6-weeks after flowering had ended and mature fruit at harvest for each treatment at each trial site by year on avocado orchards in the south-west of Western Australia. WP = Wild pollinator insects in the orchard.

Site	Year	Treatment	Density (#)	Number of fruitlets	Number of mature fruits	Fruit yield (kg/tree)
				(mean ± s.e.)	(mean ± s.e.)	(mean ± s.e.)
		Open	Bees + WP	14 ± 3.7	17 ± 3.7	4.2 ± 0.9
Puscolton	2021	Bees	5,000	32 ± 4.8	22 ± 3.0	4.8 ± 0.6
DUSSEILUIT	2021	C. dubia	5,000	6 ± 1.5	5 ± 1.0	1.4 ± 0.3
		C. vicina	5,000	15 ± 3.5	12 ± 2.7	2.8 ± 0.6
		Open	Bees + WP	43.8 ± 8.21	69.2 ± 10.09	19.6 ± 2.8
		Bees	5,000	57.2 ± 15.28	62.9 ± 17.78	14.4 ± 3.9
Pemberton	2021	C. dubia	10,000	79.9 ± 20.09	124.3 ± 25.19	26.3 ± 4.5
		C. vicina	10,000	30.1 ± 6.09	20.5 ± 4.86	4.8 ± 1.2
		Open	Bees + WP	249 ± 22.9	113 ± 11.7	22.0 ± 4.6
Capel	2022	C. vicina	10,000	64 ± 12.6	69 ± 10.9	19.0 ± 5.8
		C. vicina	5,000	12 ± 3.8	9 ± 2.8	12.0 ± 3.9
		Open	Bees + WP	48 ± 6.0	48.5 ± 8.08	12.50 ± 1.88
Capel	2023	C. dubia	15,000	69 ± 11.7	62.0 ± 10.80	16.02 ± 2.49
	2023	C. vicina	15,000	31 ± 4.8	28.8 ± 4.27	8.22 ± 1.35
		E. tenax	5,000	80 ± 11.8	73.0 ± 9.44	18.41 ± 2.36

# Fly Pollination of Blueberry, Raspberry and Blackberry

# University of New England, seedPurity and DPIRD

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# Introduction

Berries are grown year-round in Australia, with peak production from April to September in New South Wales, and extended harvests from November – March during cooler months in Tasmania (Hort Innovations, 2024). Since 2017/18, *Rubus* berry production—mainly raspberries and blackberries—has more than doubled, now exceeding 13,386 tonnes annually and valued at over \$290.3 million (Hort Innovations, 2024). In 2024/25, blueberry production exceeded 27,000 tonnes annually and was valued at over \$500 million.

Insect-mediated pollination is beneficial to *Rubus* fruit production (Keep, 1968) and pollinator dependency varies with cultivar in blueberry (Kendall *et al.*, 2020). Each *Rubus* flower contains 50–150 pistils, and if enough pistils are properly pollinated, then the flower will develop into a full, well-formed aggregate fruit. While self-pollination can sometimes occur, insects are needed to pollinate the inner flower pistils (Nybom, 1985). Poorly pollinated fruits often develop fewer drupelets that fail to fuse into a single cohesive fruit, resulting in a disorder known as crumbly berry, or the fruits may become small and misshapen due to unfertilised (seedless) pistils, rendering it unmarketable (Graham *et al.*, 2015). Other factors like plant genetics, temperature, humidity, and viral infections can also contribute to poor fruit quality (Martin *et al.*, 2017; Linck and Reineke, 2019; Edgley *et al.*, 2020); however, low pollinator activity is often implied when berries are deformed, or yields are inadequate.

*Rubus* berry production frequently encounters pollination challenges due to weather conditions that limit honeybee activity—particularly when temperatures are too hot, cold, or variable (Woods *et al.*, 2005). These issues are especially pronounced during early-season production (October–December) in Tasmania, where frosts and near-freezing conditions can prevent honeybees from flying. Given the growing reliance on insect pollination for optimal fruit set and quality in *Rubus* crops, there is a need to explore alternative pollinators that remain active under conditions unfavourable for bees. Unlike bees, many fly species are more tolerant of cooler or variable weather (Inouye *et al.*, 2015) and may be better suited to

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provide pollination services during early-season or poor-weather periods.

The aim of this research was to 1) trial two species of managed flies (*Eristalis tenax* and *Calliphora stygia*) for pollinating blackberries and raspberries (NSW), 2) understand the impact of tunnel environment on fly dispersal in an enclosed system (TAS) and 3) compare fly and bee efficiency in blueberry tunnel production (WA).

# Methods

#### Preliminary Cage Efficiency Trials (NSW)

Cage trials were used to determine fly pollinator efficiency. All trials were conducted at one large commercial berry farm (Costa Exchange Group) on the Mid North Coast region of New South Wales ( $29^{\circ}59'13.2"S 153^{\circ}08'23.6"E$ ). At this site, *Rubus* berries are produced in polytunnels for most of the year. We used cage trials to measure the pollination efficiency of flies and compared them to open field conditions where flowers are visited by two common bee pollinators, the European honeybee (*A. mellifera*; Figure 12, a) and the Australian native stingless bee, (*T. carbonaria*; Figure 12, b). The two fly species tested in cages were the Australasian brown blowfly (*C. stygia*; Figure 12 c), and the European drone fly (*E. tenax*; Figure 12, d). We observed flower visits by flies and bees and measured single-visits (one insect visit to a flower before measuring fruit weight) and unlimited visits to a flower before measuring fruit weight. Cage trials were conducted in the austral autumn and winter months in 2020 and 2022 for blackberry, and 2021 and 2022 for raspberry. Raspberry and blackberry are grown in polytunnels at this location. Plants were caged in two different blocks for fly pollination trials (*n* = 19 cages in total). The cages ( $2m \times 0.5m \times 2m$ ) were built out of flexible conduit piping (Deta Electrical, Scoresby, Vic, AU) and 2 mm x 1.5 mm cross-woven aperture insect-proof netting (Bunnings, Burnley, VIC, AU). The number of cage replicates built over the two crops varied with the year of data collection due to COVID-19 travel restrictions and the availability to source the pollinators tested.

European honeybee hives, introduced for standard orchard management, were present at varying stocking rates (up to eight hives per hectare) and placed 20–50 m from the focal block where fly cage trials were being conducted. The performance of *T. carbonaria* was assessed as it was present at the site as both managed (bees brought to the farm within hives) and wild (bees locally found within the nearby bush) pollinator. Brown blowflies, *C. stygia*, were purchased as pupae and placed in portable mesh cages ( $40 \times 40 \times 60$  cm) to emerge per supplier instructions (Sheldon's Bait, South Australia). European drone fly, *E. tenax* pupae were sourced from seedPurity Pty. Ltd. (Margate, Tasmania) and similarly raised within cages. Both fly species were acquired 1–2 weeks before crop bloom to ensure timely emergence. If flies emerged early, they were held in cages (for up to four days) with access to water and store-bought bee pollen.



Figure 12: Managed pollinators used in experimental trials to gather Rubus pollination efficiency metrics: a) the European honeybee, *Apis mellifera* Linnaeus, 1758, visiting a blackberry (*Rubus fruticosus* L.) flower, b) Australian stingless bees, *Tetragonula carbonaria* Smith, 1854, visiting a blackberry flower, c) Australasian brown blowfly, *Calliphora stygia* (Fabricius, 1781), visiting a blackberry flower, and d) European drone fly, *Eristalis tenax* (Linnaeus, 1758), visiting a raspberry (*Rubus ideaus* L.) flower. All photos taken by Abby E. Davis.

The fly *E. tenax* was stocked at 10 flies per plant (20 for blackberry, 30 for raspberry), while *C. stygia* was stocked at higher rates—75 flies per plant (150 for blackberry, 225 for raspberry)—based on preliminary trials showing lower visitation rates. Once released into cages, flies were not supplemented with food or water. Fruits were harvested when ripe (4–6 weeks after flowering) and weighed within 24 hours using a 0.01g precision scale.

#### **Enclosed Polytunnel Trials (TAS)**

To test the efficiency of *E. tenax* for springtime blackberry pollination, we enclosed a  $376m^2$  tunnel at Costa farms, Dunorlan, Tasmania (41°29'29.61" S, 146°34'35.97" E) with insect proof netting and stocked the tunnel with flies during flowering (October – November 2022). We started at 8 flies/m<sup>2</sup> in early flower and increased the stocking rate to 30 flies/m<sup>2</sup> during peak bloom. To assess the impacts of tunnel environment on fly dispersal and activity of *E. tenax*, we conducted weekly surveys along 15 transects throughout the tunnel in which the number of foraging flies in each transect were counted. Surveys were also scored in an adjacent, open tunnel pollinated by honeybees. We compared the pollination efficiency of *E. tenax* with open pollinated by tagging flowers over 5 days throughout the flowering period in both the enclosed (fly) tunnel and an adjacent open (honeybee) pollinated tunnel. In each tunnel, tagged flowers were either insect pollinated, self-pollinated (flowers were bagged using fine mesh jeweler's bags prior to receptivity which were removed after flowering to allow fruit development), or hand pollinated to demonstrate maximum fruit set. There were 200 tagged flowers per treatment in each tunnel.

#### **Blueberry Glasshouse Trials (WA)**

#### Trial 1: Calliphora albifrontalis vs no flies

This experiment examined the ability of the western golden-haired blowfly, *Calliphora albifrontalis* Malloch (1932) to pollinate southern highbush blueberry (*V. corymbosum* hybrid, variety 8–17) (Figure 13, a and c) and improve berry yield (number and average weight). This fly is endemic to the south-west of WA, is large and hairy (Figure 13, b and d) and is often seen feeding on flowers in native bushland. For these reasons, it was considered a suitable fly to pollinate blueberry flowers.

Blueberry bushes (n=18) in 45L poly-weaved bags (southern highbush) were moved from a commercial blueberry production site into 2 adjacent glasshouses (9 bushes in each) on  $11^{th}$  June 2018 at DPIRD, South Perth, WA. The bushes were harvested the following day and two to three times every week thereafter until 9<sup>th</sup> November (150 days or 21 weeks). Temperature and humidity were recorded during the trial duration using data loggers within each house. Two identical quarantine insectary glasshouses ( $28m^2$ ) were used to house each of 9 blueberry bushes set up on a daily irrigation schedule of 2.5L - 4.0L/plant/day over 8-10 waterings with a leaching fraction of 30-50% of applied volume on the advice of the commercial producer. This was to keep the EC at 3,500 – 5,000 microsiemens per cm. Sulphate of ammonia was applied fortnightly to keep the soil pH at around 4.5. Each glasshouse had the air temperature controlled to maintain temperatures between 10° to 30°C (night/day), which dataloggers placed in each glasshouse confirmed along with a relative humidity of 35-80% over the 21-week trial duration.

A laboratory colony of *Calliphora albifrontalis* were reared through to the pupal stage (F3 generation) and 1,000 pupae placed under a 5cm bed of vermiculite into Glasshouse #1 (GH1) several days prior to adult emergence. Once all the adult blowflies had emerged in the glasshouse, spent pupal cases were counted to determine the exact number of flies in the house. Releases of 500 blowflies were then repeated every 4 weeks thereafter. The 9 blueberry bushes in Glasshouse #2 (GH2) had no insects in the house over the trial duration. The only source of sugar for the blowflies was from the blueberry flowers themselves; there was ample water leaching from the bottom of the plant bags after each irrigation to provide the flies with water. The number of blowflies released was based on the number of bees used in commercial blueberry production. The recommended rate of 8-10 hives/ha translates into 360,000-450,000 bees/ha (assuming 45,000 bees/hive). The density of blueberry plants under commercial production is 3,600 plants/ha, which equates to 400,000/3,600 or 111 bees/plant. Therefore, the equivalent number of blowflies to release with 9 plants was 111 x 9 or 999 flies.

#### Trial 2: Calliphora albifrontalis vs Calliphora dubia

A second blueberry pollination trial in the same quarantine glasshouse facilities was established in June 2019. This trial compared *Calliphora albifrontalis* with a blowfly endemic to mainland Australia (the western blue-bodied blowfly (*Calliphora dubia*) (Figure 14). As in the previous trial, blueberry bushes (n=8) were placed in the glasshouses and either had *C. albifrontalis* or *C. dubia* adult flies released over 5 months with blueberry yield per bush (weight and number) recorded twice weekly.

The yield from southern highbush blueberry plants (*Vaccinium corymbosum* hybrid) sourced from a commercial blueberry farm was recorded twice weekly from 8 blueberry bushes in each of 2 quarantine glasshouses – one house had adults of the blowfly *C. albifrontalis* and the other house adults of the blowfly *C. dubia*. Each house had adult blowflies in with the bushes over 5 months. The time from a flower being open till it is a mature berry ready to harvest is 2.5 months.

#### **Data Collection for both trials**

**1)** Adult Fly Mortality: A record of any dead flies within GH1 was kept to determine how long *C. albifrontalis* adults can survive under protected cropping conditions. The first release of flies was on 26/6/18. Subsequent releases were made  $\approx$  every 4 weeks to maintain a total house number of between 1,000 - 2,000 flies. Fly releases were made on: 28<sup>th</sup> June (230 flies); 5<sup>th</sup> July (500 flies); 16<sup>th</sup> July (493 flies); 13<sup>th</sup> Aug (471 flies); 30<sup>th</sup> Aug (500 flies) and 17<sup>th</sup> Oct (581 flies).

**2)** Effect of Water Leachate on Adult Fly Survival: A sample of the leachate from the plants was taken and it's EC measured at 1,200 microsiemens/decimetre and placed in with some newly emerged adult *C. albifrontalis* (100/cage) to see if the salt content in the leachate or any other component could affect adult fly survival compared with fresh, distilled water.

**3)** Number of Open Flowers: The number of open flowers on each blueberry bush were counted on the 29<sup>th</sup> June and the 2<sup>nd</sup>, 5<sup>th</sup>, 12<sup>th</sup>, 19<sup>th</sup> and 28<sup>th</sup> July to indicate how many flowers were available to the adult flies as a source of sugar, which is essential to their survival.

**4)** Blueberry Yield. Over a total of 46 harvest dates, the mature berry yield from each individual plant was recorded at every harvest (total fruit weight and number of berries) from the 9 plants in each glasshouse.



Figure 13: a) Blueberry bushes in flower in quarantine glasshouses (insect proof) and b) *Calliphora albifrontalis* flies visiting flowers



Figure 14: An adult Calliphora albifrontalis (a) and an adult Calliphora dubia (b) in blueberry pollination trials

# **Results and Discussion**

#### Preliminary Cage Efficiency Trials (NSW)

In blackberry cage trials, a total of 162 fruits were harvested across all single-visit treatment groups. Fruits that resulted from hand-pollinated flowers were heavier than those pollinated once by insects or left to self-pollinate (Figure 15). Handand bee- pollinated fruits had similar weights, suggesting that *A. mellifera* honeybees and *T. carbonaria* stingless bees were highly effective at transferring pollen to flowers in one visit—like the amount of pollen delivered to flowers in handpollination treatments. Further, self- and fly- pollinated fruits had similar weights, suggesting that flies may transfer less pollen per visit, more like natural self-pollination outcomes. (Figure 15). Bee-pollinated fruits, however, were 48.1% heavier than those visited once by flies, further indicating that bees may be more efficient at transferring blackberry pollen in one single visit compared to flies. Fruits that self-pollinated without insect help were the smallest, showing that insect pollination is essential for high-quality blackberry fruits.



# Figure 15: Mean blackberry fruit (+ SE) weight (g) after one visit to a blackberry flower by pollinator treatments. Pollinator treatments included a combination of managed bees (*Apis mellifera* and *Tetragonula carbonaria*), managed flies (*Eristalis tenax* and *Calliphora stygia*), hand-pollination, and self-pollination.

In raspberry cage trials, fruits pollinated once by insects weighed more than fruits that were hand-pollinated or left to selfpollinate (Figure 16). Fruits visited once by bees were 13.6% heavier on average than those visited once by flies, but ultimately single-visit fruit size was similar between both insect pollinator groups (Figure 16), suggesting that both bees and flies may deposit similar numbers of pollen grains onto raspberry flowers in one visit. Fruits that were left to self-pollinate were the smallest in size and tended to form crumbly berries, showing that insect pollination is essential to produce highquality raspberry fruits.



Figure 16: Mean raspberry fruit (+ SE) weight (g) after one visit to a flower by pollinator treatments. Pollinator treatments included a combination of managed bees (*Apis mellifera* and *Tetragonula carbonaria*), managed flies (*Eristalis tenax* and *Calliphora stygia*), hand-pollination, and self-pollination.

For both raspberry and blackberry, allowing managed flies to visit flowers multiple times resulted in heavier fruit than just a single visit (Figure 17). On average, raspberry flowers visited an unlimited number of times by flies produced nearly 20% heavier fruits than those visited once, and blackberries were over 60% heavier. This shows that repeated visits by flies can significantly improve fruit size, highlighting the value of maintaining high pollinator activity during flowering.



Figure 17: Mean (+ SE) raspberry and blackberry ('Crop') fruit weight (g) by a combination of managed flies (*Eristalis tenax* and *Calliphoria stygia*) after one visit ('single') or unlimited visits to *Rubus* flowers within cages in the field.

#### **Enclosed Polytunnel Trials (TAS)**

This experiment demonstrated that *E. tenax* is a highly effective blackberry pollinator both in terms of fruit set and foraging activity throughout the tunnel. In general, *E. tenax* dispersed evenly throughout the tunnel; however, foraging activity was greatest at all transect locations on the northern side of the plants. Activity ranged from 15.1% to 162.9% (mean = 64.4%) (Figure 18). This was driven both by flower abundance and environmental factors. There was little evidence of fly mortality over the 5-week flowering period. Fruit pollinated solely by *E. tenax* was 12% heavier on average than open pollinated fruit (honeybees) (Figure 19). The stocking rates used in this trial (from 8 flies/m<sup>2</sup> in early flower to 30 flies/m<sup>2</sup> at peak bloom) were adequate to pollinate blackberries. Fruit quality (shape) assessments based on a standardised criteria used by the industry partner was consistently better for flowers pollinated by *E. tenax* than open (honeybee) pollinated fruit.



Figure 18: Mean distribution of *Eristalis tenax* (foraging flies per plant) within an enclosed blackberry tunnel. Each location features two bars representing surveys conducted on the northern and southern sides of the row, respectively, as indicated by the compass



Figure 19: Distribution of blackberry fruit weight (g) between the different treatments in the Tasmanian trial; B – Apis mellifera, C – control, E – Eristalis tenax and HP – hand pollination.

#### **Blueberry Glasshouse Trials (WA)**

#### Trial 1: Calliphora albifrontalis vs no flies

Blueberry yields were monitored twice a week over six months in two quarantine glasshouses, each containing nine southern highbush blueberry plants (*Vaccinium corymbosum* hybrid) from a commercial farm. One glasshouse had no flies, while the other contained adult blowflies (*Calliphora albifrontalis*). Since it takes about 2.5 months for a flower to develop into a mature berry, differences in yield between the two glasshouses became apparent around the 2.5-month period, with fruit yield in the *C. albifrontalis* blowfly glasshouse increasing at this time (Figure 20). The green shaded area in Figure 20 highlights the additional yield attributed to the presence of *C. albifrontalis*, demonstrating that these flies contributed to improved fruit production.


Figure 20: Blueberry yield when pollinated by the blow fly *C. albifrontalis* ('+ flies') compared to bushes where no insects were present ('no flies') to promote blueberry pollination. Yield (extra fruit because of flies pollinating blueberry flowers) differences between the two treatments is shown by the green shading. Pollinator treatments (flies vs. no flies) are differentiated by colour.

Blueberry bushes exposed to adult *C. albifrontalis* blowflies produced both more and larger berries than those in the control group with no flies (Figure 21). Specifically, bushes with flies yielded 17.14 kg from 9,108 berries (average 1.88 g/berry), compared to 10.43 kg from 6,379 berries (average 1.63 g/berry) in the control.



Figure 21: Exponential plot of cumulative blueberry yield (kg) when pollinated by the blowfly *C. albifrontalis* (blue dots) compared with bushes where no insects were present (red dots).

Yield differences between the two treatments became noticeable 11 weeks after the flies were released. By the end of the study, bushes with *C. albifrontalis* had produced 11.29 kg from 6,177 berries (average 1.83 g/berry), while the control yielded only 4.98 kg from 3,427 berries (average 1.45 g/berry). Additionally, berry size was positively correlated with seed number, suggesting improved pollination where flies were present.

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#### Trial 2: Calliphora albifrontalis vs Calliphora dubia

In addition to the blowfly, *C. albifrontalis,* the blowfly *C. dubia* was also determined to be an effective blueberry pollinator. Over the 5-month trial, blueberry bushes pollinated by *C. dubia* flies produced both more and larger berries compared to those pollinated by *C. albifrontalis* (Figure 22). Specifically, bushes with *C. dubia* yielded approximately 14,500 berries at an average of 1.93 g per berry, while bushes with *C. albifrontalis* produced around 10,500 berries averaging 1.85 g per berry. (Figure 22). Further information about this study can be found in Cook *et al.* (2020).



Figure 22: Total blueberry fruit yield (kg), total berry number and mean berry size of blueberries when pollinated by either the blowfly *C. albifrontalis* (L) or *C. dubia* (R) over 130 days.

## **Key Outcomes and Recommendations**

This research shows that certain fly species, particularly *Eristalis tenax* and blow flies like *Calliphora albifrontalis* and *C. dubia*, can be highly effective pollinators of key berry crops such as blackberry, raspberry, and blueberry. In enclosed tunnel trials in Tasmania, *E. tenax* outperformed open-field honeybee pollination for blackberry, and stocking rates of 8–30 flies/m<sup>2</sup> were sufficient to ensure high quality fruit set. While bees deposited more pollen per visit in NSW blackberry trials, flies, including the blow fly *C. stygia*, still played an important role (especially when allowed multiple visits), resulting in significantly larger fruit. In NSW raspberry trials, both flies and bees produced similar fruit weights after a single visit, and repeated fly visits led to heavier fruit, reinforcing the importance of high pollinator activity during flowering. For blueberries, fly presence clearly boosted both berry number and size, with *C. dubia* outperforming *C. albifrontalis* across a five-month period. Together, these findings suggest that managed flies can serve as reliable, complementary pollinators to bees, particularly in protected or low-bee environments. With proper management, flies can help increase yields and improve fruit quality—providing growers with an alternative or backup pollination strategy to support consistent production outcomes.

## **Fly Pollination of Sweet Cherries**

## seedPurity

Raylea Rowbottom and Cameron Spurr



## Introduction

The Australian cherry industry comprises 700 growers and approximately 3000ha of crop, with more than 80% of production occurring in Victoria, New South Wales and Tasmania. Production volumes vary substantially from year to year due to climatic factors but in 2021 the industry grew more than 20,000 tonnes of cherries, representing a farm gate value of \$231 million (Clarke and Le Feuvre, 2022). The industry is currently in an expansion phase, targeting export opportunities in Asia and the United Kingdom (Cherry Strategic Investment Plan 2022-2026, 2022), with increased plantings occurring predominantly in Victoria and Tasmania.

One of the challenges faced by producers is that many sweet cherry varieties are self-incompatible and require insectmediated cross-pollination to ensure fruit set (Cachi and Wünsch, 2014) but flowering, which typically occurs in September in mainland orchards or October at cooler Tasmanian locations, is characterised by a relatively short main bloom period lasting just 3-4 weeks. To promote cross-pollination, orchard designs typically feature different cross-compatible cultivars planted either in alternating rows or within the same row. The selection and management of these different cultivars within an orchard to ensure synchronous flowering is critical for achieving successful pollination (Bright and Marte, 2004). Cherry orchards are normally stocked with honeybee hives for pollination. While 2-3 hives/ha is often considered adequate (Clarke and Le Feuvre, 2022), some researchers have concluded that higher stocking rates may be desirable (Somerville, 1999). Monck et al (2008) suggested hive requirements of between 2.5 and 5 hives/ha, while Goodwin (2012b) recommended that up to 10 hives/ha be used for pollinating cherries. Clarke and Le Feuvre (2022) estimated the annual demand for managed honeybees in the Australian cherry industry at 8,535 hives in 2021 and forecast 30% growth in hive requirements over the following decade. Many growers are also heavily reliant on feral honeybees and native bees to supplement the pollination services provided by stocked honeybees (Blaauw and Isaacs, 2014; Eeraerts et al., 2020; Holzschuh et al., 2012; Osterman et al., 2023). However, the arrival of varroa mite in Australia and its inevitable spread will eliminate many feral honeybee colonies and ultimately result in an increasing demand for managed pollination services within the cherry industry that outstrips industry growth.

Issues surrounding pollinator availability are further compounded by insufficient or ineffective pollination, which is known to be a key factor contributing to seasonal variation in fruit set in cherries (Somerville, 1999; Reilly *et al.*, 2020). Insufficient

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pollination is often the result of an orchard environment that is inhospitable to honeybee foraging during flowering. Cold, wet or windy weather conditions are relatively common during the spring bloom period in some production areas and can deter honeybees (Hansted *et al.*, 2015; Vicens and Bosch, 2000) while nets and plastic covers that are widely used to protect ripening fruit from birds, hail damage and rain cracking can also reduce honeybee foraging activity (Dag and Eisikowitch, 1995; Dag and Eisikowitch, 1999; Dag, 2008; Lang, 2014; Evangelista *et al.*, 2014; Ellis *et al.*, 2017; Nielsen *et al.*, 2017; Hall *et al.*, 2020; Kendall *et al.*, 2022). In addition, although cherry blossom is considered a relatively attractive forage source (Goodwin, 2012), competition from nearby alternatives such as weedy brassicas, capeweed, eucalyptus, Patterson's curse and white clover has been shown to draw honeybees away from flowering cherry crops (Keogh *et al.*, 2010; Warren *et al.*, 2024).

The contribution of sub-optimal pollination to variable yield outcomes is well-recognised within the industry, as evidenced by a recent review of pollination statistics for Australian cherry crops (Clarke and Le Feuvre, 2022) in which growers prioritised research into alternative pollinators and management of multiple pollinators in the crop. In this study, we investigated potential for the hoverfly *Eristalis tenax* to be used as a complementary managed pollinator in cherry orchards.

## **Methods**

Two years of field experiments were conducted in 2023 and 2024 in commercial blocks of sweet cherries at Reid Fruits orchard in Jericho, Tasmania (Figure 23, a). In 2023, a large cage trial was conducted under a retractable roof field-covering Cravo system (Cravo Equipment Ltd) (Figure 23, b) to test whether *E. tenax* could effectively pollinate cherries. In 2024, a large cage trial and an open field release were undertaken in an adjacent orchard block covered with bird netting (Figure 23, c). These trials were used to determine the relationship between *E. tenax* stocking rates and fruit set, and to obtain preliminary data on both fly deployment, dispersal and retention and yield outcomes from complementary stocking of honeybees and hoverflies in an open orchard setting.

The Cravo system was planted with alternating rows of varieties Kordia and Regina, interspersed with polleniser trees (var. Sylvia). The 2024 trial block was like the Cravo block except that it also contained 3 rows of variety Fertard interspersed between Kordia and Regina rows.



Figure 23: a) Google earth overview of Reids Cherry Orchard, Jericho, Tasmania. The Cravo system where we conducted trials in 2023 is highlighted in red and the 2ha trial block for the 2024 open release is highlighted in yellow, b) Cravo system used for large cage trials in 2023 and c) the netted 2ha block used for cage-based stocking rate and open release trials in 2024/25. Photo credits Raylea Rowbottom.

#### **Pollinator Efficacy Trials (2023)**

In late September, prior to flowering, four  $35m^2$  cages were erected over 2 rows of cherry trees, with each cage enclosing 6 cherry trees: 2 to 3 trees each of Kordia and Regina (depending on cage location) and a single Sylvia (polleniser) tree. The cages were netted with 3mm nylon mesh netting to prevent movement of pollinators between cages and external trees. At the time of construction, caged trees were inspected for potential pollinators and these were removed. At the onset of flowering, each cage was stocked with 1000 adult *E. tenax* (equivalent to 200,000 flies/ha). The surrounding open orchard was stocked with honeybees at 4.5 hives/ha (equivalent to 225,000 bees/ha). Ten flowering branch sections were tagged on Kordia and Regina trees in each cage, with corresponding sets of 10 branch sections/cultivar also tagged outside each cage, giving a total of 40 branch sections per treatment (open pollinated trees and caged trees pollinated exclusively by *E. tenax*) in each variety. The number of flowers on each marked branch section was counted and recorded for comparison with fruit set. After flowering, the cages were removed to allow for normal fruit development. On the 15<sup>th</sup> of December, approximately 5 weeks prior to commercial harvest, the number of fruit on each marked branch section was counted. Using corresponding flower and fruit count data, we then determined the percentage of flowers to set fruit in each treatment (Figure 24).



Figure 24: Images from cherry pollination trial at Reids, Jericho. Clockwise from top left: trial cage set up over Sylvia, Regina and Kordia trees within the Cravo system (4 cages in total, 9<sup>th</sup> October 2023); *E. tenax* feeding from cherry flowers; tagged branches for flower counts (27<sup>th</sup> October 2023); and Mark van Schilt from seedPurity assessing fruit set from the same tagged branches (15<sup>th</sup> December 2023). Photo credit: Raylea Rowbottom.

#### E. tenax Stocking Rate Trial (2024)

Cage design, cultivars and fruit set assessment methods used in this trial were identical to those used for the 2023 efficacy trial. Stocking rates ranged from 25,000 flies/ha to 200,000 flies/ha across six treatments.

#### Pilot Open Field Release (2024)

In October 2024 we stocked a small (2ha) commercial block of cherries under bird netting with *E. tenax* pupae at 60,000 flies/ha. The site was also stocked with honeybees at a reduced rate (3 hives/ha, equating to 90,000 – 120,000 bees/ha). The flies were deployed as pupae using flyscraper release boxes positioned in the centre of the block (Figure 25). Following deployment, we tracked hatching rates within the release boxes to determine the impact of climatic conditions in the orchard on fly hatching rates, and to ensure the target density of adult flies was met. Fly dispersal and distribution was tracked at approximately weekly intervals throughout flowering by visual counting of flies foraging on 90 trees (45 each of Kordia and Regina) located on a grid throughout the orchard. Visual counts were conducted between 10am and 2pm.



## Figure 25: Field site for open pollination trial (2024) at Reid Fruits, Jericho depicting the approximate locations of *E. tenax* flyscrapers and *A. mellifera* hives.

Fruit setting rates were assessed on Kordia and Regina within the trial block and in a commercial block with similar aspect, planting configuration, tree age and cover type (Refer to Figure 23) stocked only with honeybees at 3.5 hives/ha. At each site, we counted the number of fruit set on 5 tagged flowers on 100 trees (500 flowers in total/site). Fruit set was determined approximately 5 weeks before commercial harvest in January 2025. To minimise the potential for confounding site effects in the fruit set comparison between stocking with honeybees only or with both honeybees and hoverflies, we applied a supplemental hand pollination treatment (hand pollination plus insect pollination) under favourable pollinating conditions on the 23<sup>rd</sup> of October to a matching set of 500 flowers at each site. This treatment supplied sufficient pollen to individual flowers to allow measurement of fruit set at each site when pollination was non-limiting. We then standardised fruit setting rates for the insect pollination treatments across the two sites (honeybees or honeybees + hoverflies) against fruit set in the corresponding supplemental pollination treatment.

## **Results**

Trees caged with *E. tenax* set more fruit than open trees pollinated predominantly by stocked honeybees, with 7% increased fruit set in Kordia and 43% more fruit set in Regina (Figure 26). This result confirmed that *E. tenax* can effectively pollinate sweet cherries and identified that pollination in the adjacent orchard stocked with 3 bee hives/ha may have been sub-optimal.



Figure 26: Mean percent of flowers setting fruit for Kordia and Regina trees pollinated by either honeybees (A. *mellifera*) or hoverflies (*E. tenax*). Error bars indicate standard errors (n = 40). The letters above the columns denote statistically significant differences between treatment means.

We observed a statistically significant relationship between the stocking density of *E. tenax* and fruit set (Figure 27). When *E. tenax* was present as the sole pollinator in large cages, fruit set increased from 12.1% (Regina) and 19.6% (Kordia) in trees stocked with 350 flies/ tree (25,000 flies/ha) to 34.9 and 30.3%, respectively, at 1050 flies (75,000 flies/ha). However, at higher stocking rates (150,000 and 200,000 flies/ha), fruit set declined, possibly due to antagonistic behaviour between individuals when stocked at high density, over-utilisation of pollen as a food resource resulting in limited pollen availability for pollination, and/or starvation of flies due to insufficient nectar.



Figure 27: Effect of hoverfly stocking density (adult flies/ha) on the percentage of flowers setting fruit in Kordia (top - blue) and Regina (bottom - green). Error bars are standard errors; n=40. The letters above the columns denote statistically significant differences between treatment means.

Hatching of *E. tenax* in the open release trial was delayed by approximately 14 days and overall hatching rates were less than normal (57% in this trial vs >80% typically) due to both overstocking release towers and low temperatures experienced in the orchard (Figure 28). As cool conditions are common in spring in some cherry-growing areas of southern Australia, development of a pupal release box that maintain warm conditions around the hatching pupae may be important for improving timeliness and precision of fly deployment in early season crops.



Figure 28: Temperature comparisons between the open field (orange) and inside the pupal release towers (blue) during peak bloom (14<sup>th</sup> October – 30<sup>th</sup> Oct 2024).

Following an initial period of dispersal from the release points, *E. tenax* appeared to distribute relatively evenly throughout the orchard (Figure 29). On the 23<sup>rd</sup> of October, 21 days after pupae were placed into the trial block, we estimated numbers of hoverflies foraging on cherry blossom equivalent to 19% of hatched flies from pupae deployed in the crop, with many others observed foraging on clover on the orchard floor. While it is possible that wild *E. tenax* accounted for some of the surveyed flies, a baseline survey conducted at the beginning of peak bloom before flies hatched from the release boxes found almost no hoverflies at the site.

Although *E. tenax* was stocked into the trial release site at approximately one third the density of honeybees, hoverflies outnumbered honeybees in the counts of foraging insects on trees of both varieties by approximately 2:1, on average, during the survey period (Figure 29). The greatest difference in hoverfly and honeybee activity was observed on the 23<sup>rd</sup> of October (Figure 29) when air temperature during the survey period averaged 12.7°C.



Figure 29: Heat maps for both Kordia (top maps) and Regina (bottom maps) comparing the number of foraging *A. mellifera* and *E. tenax* within the orchard over time. Foraging activity is scaled from low (yellow) to high (red). No foraging activity was observed in grey areas.

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#### Fruit Setting Rates in Orchard Blocks With and Without Complementary Stocking of E. tenax.

Comparison of standardised fruit set data (open pollinated fruit set expressed as a percentage of corresponding supplemental hand pollinated fruit set) revealed that complementary stocking of *E. tenax* and *A. mellifera* (60,000 hoverflies and 3.7 hives/ha, equating to 185,000 bees/ha) improved fruit set during peak bloom by 44% relative to stocking honeybees only at 4.5 hives/ha (225,000 bees/ha) (Figure 30). This increase in fruit set is consistent with the overall improvement in pollinator activity observed from complementary stocking compared to stocking honeybees alone, especially under cooler conditions, and corroborates earlier results from 2023 cage trials suggesting that inadequate pollination was a yield limiting factor in trees grown in a Cravo system stocked with 3 hives/ha.



Figure 30: Comparisons between percent fruit set between supplementally pollinated fruit and open pollinated fruit in the commercial (bee only) pollinated block and the trial block that was stocked with both flies and bees for the cultivar Kordia in a commercial 2ha orchard. Error bars are standard errors.

### **Key Outcomes and Recommendations**

Data generated in this study supports earlier research findings pointing to inadequate pollination as a yield limiting factor in sweet cherry production. We demonstrated in large cage trials and open release studies on Kordia and Regina trees that *E. tenax* is an effective pollinator, dispersing evenly throughout the orchard. Delayed and reduced hatching rates caused by the low early season temperatures in this trial highlight the importance of a release box design that maintains warmth around the hatching pupae for early season releases. While this meant we were only able to evaluate yield effects of *E. tenax* stocking for the later part of the flowering period, the results provide a promising preliminary indication of the potential for *E. tenax* as a managed pollinator in cherry orchards. Replication of this experiment across multiple and larger sites, seasons and cultivars is essential so that the preliminary results can be confirmed, deployment protocols refined, and sufficient data generated to inform a cost-benefit analysis for the use of hoverflies as complementary managed pollinators in sweet cherry orchards.

## Pollination in Mango

## **University of New England and Western Sydney University**

Romina Rader, Abby Davis; James Cook and Jonathan Finch





## Introduction

Mango (*Mangifera indica*) is one of Australia's most important tropical and subtropical fruit crops, primarily grown across northern Queensland, the Northern Territory, and parts of Western Australia. Queensland leads national production, contributing around 70% of Australia's mango volume, followed by the Northern Territory with 20% (Hort Innovations, 2024). In 2024, over 63,000 tonnes of mangoes were produced nationally, with a total production value of \$220 million (Hort Innovations, 2024).

Despite these strong production figures, pollination remains a key limiting factor for fruit set in mango orchards (Ramírez and Davenport, 2016). Mango pollination is often inefficient and highly variable. Each panicle can produce thousands of flowers, yet only a small fraction (~0.1–1%) mature into fruit. This low fruit set is often attributed to insufficient pollination by insects, although studies suggest that environmental conditions (Dag, *et al.*, 2000; Sánchez *et al.*, 2024), genetic factors (Allen-Perkins *et al.*, 2022), management practices (Siqueira *et al.*, 2008), or a combination of all can also significantly influence mango pollination outcomes.

Mango flowers attract a diverse array of wild insects—including flies, native bees, and beetles—which are believed to play an important role in pollination (Anderson, *et al.*, 1982; Singh, 1985). In Australia, many growers rely on these naturally occurring insect visitors to support fruit production. While some growers introduce managed honey bee (*Apis mellifera*) hives during bloom, mango flowers produce little nectar and limited pollen (Sánchez *et al.*, 2024; Siqueira *et al.*, 2008), making them relatively unattractive to honey bees. As a result, reliance on managed bees alone may not ensure optimal pollination.

Research also indicates that pollination requirements vary across mango cultivars (Ramírez & Davenport, 2016), and the effectiveness of different insect groups can differ. For example, some European fly species have been shown to be effective mango pollinators (Huda, *et al.*, 2015; Sánchez, *et al.*, 2022). However, Singh *et al.* (2024) determined that Australian native bees were more efficient pollinators of mango than Australian hoverflies and European honeybees. Identifying which insects are the most effective pollinators—and finding ways to support these beneficial wild species on farms—is essential for improving mango yield and fruit quality.

Among the potential fly pollinators, blowflies (Calliphoridae) have been observed visiting mango flowers (Dag *et al.*, 2000; Hort Innovation Marcacci *et al.*, 2023; Saeed, *et al.*, 2016), and are known to pollinate other commercial crops (Cook *et al.*, 2020). The larvae of flower-visiting blowflies typically develop in decomposing organic matter (Davis *et al.*, 2023. Anecdotally, some growers in northern Australia have started using carrion, or "stink stations," to attract adult blowflies into orchards during flowering. However, the effectiveness of this practice in boosting fly abundance and improving pollination outcomes has not been scientifically tested.

Therefore, we conducted a series of experiments to 1) identify the diversity and abundance of mango floral visitors in Queensland and the Northern Territory, 2) determine how effective common, wild insects are at depositing pollen onto mango flowers, 3) determine if 'stink stations' promote the abundance of blowflies in turn promoting pollination and fruit set in Australian mango orchards.

## **Methods**

#### Mango Pollination – Queensland

#### University of New England

#### Field sites

Seven field sites were located at six large, commercial farms owned by third-party landowners within the Atherton Tablelands, Queensland. All farms were located approximately 50 km from Mareeba, Queensland, all sites were located at least 500 m apart and grew commercial mango trees. Mango cultivars grown at the farms included 'Kensington Pride', 'Keitt', 'R2E2', and 'Kent' (Table 4). All farms, except for Farm 4, brought managed honeybees into fields to perform pollination services. Permission to conduct fieldwork on all sites was granted by the farm owners.

Table 4: Mango cultivars grown at each farm site, including 'Kensington Pride' (KP), 'Keitt', 'R2E2', and/or 'Kent', depending on farm and location.

Farm	Cultivars				
1	'KP', 'Keitt', 'R2E2'				
2	'KP', 'R2E2'				
3	'KP'				
4	'KP'				
5	'Keitt', 'Kent'				
6	'KP', 'Keitt', 'Kent'				

#### Floral visitation surveys

To identify the abundance and diversity of insects visiting mango flowers, visitation surveys were conducted at seven field sites selected across the six farms for a total of two to three days (at each site) from 2 August 2022 to 25 August 2022 (peak bloom). Surveys were carried out on days with no rain and when temperatures were at least 15°C. Temperature, relative humidity, and wind speed were recorded from nearby weather stations before each survey was conducted. For consistency, surveys were conducted along two, 10 m transects: one along the edge of the mango orchards and an additional walk towards the middle (> 30 m within orchard rows). All transects were conducted walking slowly (one min per m) while looking at the flower panicles on one row of mango trees (Figure 31, a), with the observer looking for insects on flowers only from ground level to a height of 2 m (Figure 31, b). Edge and middle transect walks were at least 50 m apart. Insects were collected in the field for identification to the lowest possible taxonomic level using keys or expert aide.



Figure 31: Mango flowers and insect visitors in the Atherton Tablelands, Queensland: (a) a mango panicle used for insect visitor observations; (b) the snout fly (*Stomorhina discolor*; family: Rhinnidae) visiting a mango flower. Photos taken by Abby E. Davis.

#### Examining pollination effectiveness

To evaluate how effective fly species were at depositing pollen grains onto mango flower stigmas, we conducted singlevisit pollen deposition (SVD) trials—where one insect visits a flower, and the number of pollen grains the insect deposits is counted. These experiments were conducted during peak mango flowering (August 2022) at two commercial farms in the Mareeba region. Both farms grew the same mango cultivar ('Kensington Pride'), followed similar management practices, and relied solely on abundant wild pollinators, so they did not have managed bees or formal pollination protocols.

Because flies were commonly observed visiting mango flowers during the study period, we evaluated the pollination effectiveness of two fly families frequently found in the field: snout flies (Rhiniidae) and blowflies (Calliphoridae). SVD trials were conducted using four treatments: Calliphoridae, Rhiniidae, open-pollination, and self-pollination. To collect replicates for the Rhiniidae, Calliphoridae, and self-pollination treatments, prior to bloom, mango panicles were bagged individually with insect-proof organza bags. When the flowers on the panicle opened but were not yet receptive to pollen, all imperfect flowers (lacking stigmas) and the anthers from perfect flowers (containing both stigmas and anthers) were carefully removed before pollen release (when pink anthers darkened). Next, the female flowers were clipped from panicles and stored in a protected container for later use. This ensured that the remaining bagged flower stigmas were not exposed to self-, wind-, or accidental pollination prior to a controlled insect visit.

#### **Pollinator Visitation and Blowfly Habitat - Northern Territory**

#### Western Sydney University

The study was conducted across three major mango-growing regions in Australia: Darwin, NT (12.4°S, 130.0°E), Katherine, NT (14.4°S, 132.2°E), and Burdekin, QLD (19.5°S, 147.4°E). Farms in these regions primarily grew the *R2E2* and *'Kensington Pride'* mango varieties and were conventionally managed, using synthetic insecticides, fertilisers, and drip irrigation. Flowering occurred from May to July in Darwin and Katherine, and primarily in August in the Burdekin region.

To assess whether stink stations increased blowfly activity to mango flowers, pollinator surveys were conducted at control (no stink stations) and treatment (stink stations present) sites. Surveys were performed at varying distances (0m, 10-30m, and 30-50m) from stink stations at both treatment and control sites (Figure 32). Each tree was surveyed twice daily—between 9:00–11:00 am and 2:00–4:00 pm—on two non-consecutive days during flowering. Surveys were avoided during rain or heavy cloud cover, which occurred on two days in mid-June. During each survey, observers slowly walked around the tree for three minutes, recording all insects that contacted flowers within 3 m of ground level—which represented 50–75% of the flowers per tree. Both insect abundance and identity were recorded, with representative insects captured and identified to the lowest taxonomic level.



Figure 32: Design of experiment to test the effect of stink stations on blow fly visitation to mango trees. Insect surveys were performed 0m, 10-30m, and 30-50m from stink stations.

#### Results

#### **Mango Pollination in Queensland**

#### Floral visitation surveys

In total, 203 floral visitation transect walks (33.8 hours) were conducted on mango trees. From these transects, we identified 42 taxa (26 species and 16 morphospecies) from 16 insect families across four orders (Coleoptera, Diptera, Hemiptera, and Hymenoptera) (Table 5). The dipterans (flies) were the most diverse group, followed by the hymenopterans (bees and ants only), coleopterans (beetles), and hemipterans (true bugs).

Table 5: Relative insect abundance of insect flower visitors recorded on mango trees during peak bloom in the Atherton Tablelands, Queensland. Insects were identified to the lowest practical taxonomic level and sorted by pollinator group.

Order	Family	Genus	Species	Abundance			
				0 - 50	50 - 100	100 - 500	> 500
Diptera	Rhinidae	Stomorhina	discolor				$\checkmark$
Diptera	Rhinidae	Stomorhina	xanthogaster		$\checkmark$		
Diptera	Rhinidae	Metallea	incisuralis	$\checkmark$			
Diptera	Calliphoridae	Chrysomya	saffrenea			$\checkmark$	
Diptera	Calliphoridae	Chrysomya	rufifacies		$\checkmark$		
Diptera	Calliphoridae	Chrysomya	flavifrons	$\checkmark$			
Diptera	Calliphoridae	Chrysomya	varipes	$\checkmark$			
Diptera	Calliphoridae	Chrysomya	incisuralis	$\checkmark$			
Diptera	Calliphoridae	Onesia	tibialis	$\checkmark$			
Diptera	Calliphoridae	Calliphora	centralis	$\checkmark$			
Diptera	Calliphoridae	Calliphora	augur	$\checkmark$			
Diptera	Syrphidae	Syritta	luteinervis	$\checkmark$			
Diptera	Syrphidae	Simosyrphus	grandicornis			$\checkmark$	
Diptera	Syrphidae	Mesembrius	hilaris	$\checkmark$			
Diptera	Syrphidae	Mesembrius	bengalensis	$\checkmark$			
Diptera	Syrphidae	Austalis	resoluta	$\checkmark$			
Diptera	Syrphidae	Eristalinus	punctulatus	$\checkmark$			
Diptera	Syrphidae	Melangyna	viridiceps	$\checkmark$			
Diptera	Syrphidae	Citrogramma	sp.	$\checkmark$			
Diptera	Bombyliidae	Comptosia	sp.	$\checkmark$			
Diptera	Bombyliidae	Geron	sp.	$\checkmark$			
Diptera	Muscidae	Neomyia	timorensis	$\checkmark$			
Diptera	Muscidae	Musca	domestica	$\checkmark$			
Diptera	Muscidae	Hydrotea	chalcogaster	$\checkmark$			
Diptera	Tachinidae (Phasiini)		sp. 1	$\checkmark$			
Diptera	Tachinidae (Phasiini)		sp. 2	$\checkmark$			
Diptera	Tachinidae (Goniinae)		sp.	$\checkmark$			

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Diptera	Sarcophagidae	Sarcophaga	sp.	$\checkmark$	
Diptera	Culicidae		sp.	$\checkmark$	
Diptera	Milichiidae		sp.	$\checkmark$	
Hymenoptera	Apidae	Apis	mellifera		$\checkmark$
Hymenoptera	Apidae	Apis	cerana	$\checkmark$	
Hymenoptera	Apidae	Tetragonula	carbonaria		$\checkmark$
Hymenoptera	Apidae	Ceratina	sp.	$\checkmark$	
Hymenoptera	Apidae	Xylocopa	sp.	$\checkmark$	
Hymenoptera	Halictidae	Homalictus	sp.	$\checkmark$	
Hymenoptera	Halictidae	Lipotriches	sp.	$\checkmark$	
Hymenoptera	Evaniidae		sp.	$\checkmark$	
Hymenoptera	Pteromalidae		sp.	$\checkmark$	
Hymenoptera	Formicidae	Iridomyrmex	sp.	$\checkmark$	
Coleoptera	Tenebrionidae	Lagria	cyanea	$\checkmark$	
Hemiptera	Pentatomidae	Cuspicona	simplex	$\checkmark$	

Insect abundance varied based on pollinator groups, with wild bees and flies generally more abundant than managed honeybees (Figure 33). The three most common (> 500 observed in total) species seen visiting avocado flowers was the Australian native stingless bee (*Tetragonula carbonaria*), the European honeybee (*Apis mellifera*), and the rhinid (snout) fly *Stomorhina discolor* (Table 5). The large numbers of wild bees and flies observed on mango flowers suggest that the food sources and habitats to support these helpful insects are nearby, or within, the local mango farms.



Figure 33: Percentage of wild flies, wild bees, and managed bees seen on mango flowers in the Atherton Tablelands, Queensland.

#### Pollination effectiveness

In total, 161 mango flowers were assessed for pollen deposition. Open-pollinated flowers had the highest pollination rate, with 24% showing mango pollen on their stigmas (Figure 34) — suggesting the mango trees at the study sites were producing pollen and pollen-flow was occurring naturally in the system. The fly family that pollinated the greatest number of flowers after one visit was the Rhiniidae, with 12.5% of flowers pollinated (Figure 34). It was determined that two species of rhinid flies were depositing pollen onto mango flowers, including *Stomorhina discolor* and *S. xanthogaster*.



Figure 34: Percentage (%) of 'Kensington Pride' mango flowers with and without pollen on stigmas after a single visit, based on pollinator treatments near Mareeba, Queensland. Treatments included Rhiniidae flies, Calliphoridae flies, self-pollination (bagged flowers), and open pollination (natural conditions). Pollination success was measured by the presence ('pollinated') or absence ('unpollinated') of mango pollen on flower stigmas. Different colours represent pollination success.

In contrast, none of the flowers visited by Calliphoridae flies received pollen, despite using the same method that showed successful deposition by Rhiniidae flies. Similarly, no pollen was found on self-pollinated flowers, confirming that the organza bags effectively blocked wind and accidental pollination.

#### **Pollinator Visitation and Blowfly Habitat - Northern Territory**

Across all farms in the Darwin region, the highest percentage of visits (55 %) was made by a large hoverfly, *Mesembrius bengalensis* (Figure 35, a). Stingless bees (*Tetragonula* spp.) also made a high proportion of all visits (16%), but mostly on farms without stink stations (Figure 35, a). In contrast, European honeybees (*Apis mellifera*) were rare and accounted for less than 1% of all visits (Figure 35, a).



Figure 35: Insect floral visitation rates of on mango flowers with and without stink stations in Darwin, Northern Territory, Australia: A) Flower visitors by percentage of total visits to mango panicles on farms and B) Mean number of blow flies observed per tree (summed across pollinator surveys). Error bars show the standard deviation of the mean.

Mango farms with stink stations had more blowfly visits per mango tree than those without—34% of insect visits were from blowflies on treated farms, compared to just 3% on control farms (Figure 35, a). The number of blowflies seen on flower panicles was much higher on farms with stink stations (Figure 35, b). However, the increased abundance of blowflies did not result in increases in early or late fruit set. As such, although stink stations successfully increased the abundance of blowflies, we found no evidence that their use promotes yields in mango farms. Our finding that stink stations did not promote fruit set may have occurred because of the very high abundance of other non-blowfly pollinators during our experiment, namely a native hoverfly *Mesembrius bengalensis* (Syrphidae).

Blowfly numbers decreased the further trees were from the stink stations. The highest fly activity was recorded on trees closest to the stations (0–10 m), with fewer flies observed at 10–30 m and 30–50 m (Figure 36). However, even at 10–50 m away, blowfly numbers were still significantly higher than on farms without stink stations—showing that stink stations can attract and increase blowfly activity across a large area of the orchard. However, the beneficial effects of stink stations may only occur in years or regions where other pollinators are less abundant. More information about this study can be found in (Finch, Gilpin, & Cook, 2023).



Figure 36: Mean numbers of blow flies observed per tree during pollinator surveys at increasing distances from stink stations across seven mango farms around Darwin, Northern Territory, Australia. Boxes with a common letter are not significantly different.

## **Key Outcomes and Recommendations**

This study highlights the wide variety of insects visiting mango flowers in northern Australia, with wild insects—particularly native stingless bees and flies—making up most of the activity. Snout flies (family Rhiniidae) were frequently observed on flowers and were effective at depositing pollen, suggesting they are important mango pollinators in Queensland. In the Northern Territory, blowflies were successfully attracted to mango trees using stink stations, but this did not lead to higher fruit set. This was likely because other insects, such as the native hoverfly (*Mesembrius bengalensis*) and stingless bees, were likely providing strong pollination services to crop fields already. These results suggest that while stink stations can increase blowfly numbers in orchards, they may not be as effective for pollination as other fly species. More research is needed to understand the biology of Rhiniidae flies—particularly what they feed on and where their larvae develop. Knowing which insects are actively visiting flowers and are effective pollinators in mango orchards will help guide better pollinator management on mango farms.

# **Fly Pollination of Glasshouse Strawberries**

## Western Sydney University

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## Introduction

Strawberries are an important fruit crop both in Australia and worldwide. They are often grown in fields, but also increasingly in polytunnels or glasshouses, which facilitate crop management and mitigate the risks from extreme weather events. By their nature, glasshouses exclude wild pollinators, requiring pollination to be managed inside the structure for good cropping outcomes with pollination-dependent crops. Although honeybees are the main managed pollinators used in horticultural settings, they are less suitable for use in protected cropping and especially in glasshouses. This creates the need for other pollination options for glasshouse berry crops and some species of flies are potentially good candidates. Our major research aims were to:

- 1. Identify two species of flies available in Australia that readily visited strawberry flowers.
- 2. Test and compare the ability of these flies to pollinate strawberries in a glasshouse setting.
- 3. Establish appropriate stocking densities for good crop pollination in medium to large glasshouse settings.

## **Methods**

We performed a series of experiments to address our research aims, using the state-of-the-art research glasshouse facilities in the National Vegetable Protected Cropping Centre (NVPCC) facility at the Hawkesbury Campus of Western Sydney University in Richmond, NSW (Figure 37). First, we conducted a small-scale trial to test three candidate fly species for attraction to forage on strawberry flowers. This allowed us to identify two species for more detailed studies. We then proceeded to a glasshouse trial with a blowfly (*Calliphora stygia*) as the pollinating agent. We next performed essentially the same glasshouse experiment, but this time using a drone fly (*Eristalis tenax*). These trials showed that both species were very good glasshouse pollinators of strawberries, so we proceeded to the next stage of exploring stocking densities and comparing the relative performance of the two species. We reduced the stocking density of flies considerably from the previous experiments and still found very good pollination outcomes.



Figure 37: Top) Strawberries growing in hydroponic gutters in the NVPCC. Bottom) The NVPCC at the WSU Hawkesbury Campus near Richmond, NSW

#### **Experiment 1. Fly Species Screening**

Between December 2019 and February 2020, we trialed three species of fly as glasshouse pollinators of strawberry. The species used were *E. tenax* (Syrphidae), *C. stygia* (Calliphoridae) and *Hermetia illucens* (Stratiomyidae). The drone fly *E. tenax* was sourced from seedPurity, whilst the blowfly *C. stygia* was purchased from Sheldon's Baits, a commercial fly breeder and supplier in South Australia. *H. illucens* was obtained from a private colony maintained by Jon Finch. In a preliminary experiment, 20 *H. illucens* were placed in a cage containing several flowering strawberry plants, but flies showed no interest in visiting the flowers. As such, we chose not to continue with this species.

The remaining two species were placed in BugDorm<sup>™</sup> mesh cages (n=12) (Figure 38) within an experimental glasshouse (18-24°C, 70% RH) and allowed to forage on 1-3 flowering strawberry plants for three consecutive days. Two adult flies (3-8 days old) were placed in each cage and allowed a 12hr settling in period prior to any observations being made. None of the flies had previously foraged on flowers of any kind, being primarily maintained on an artificial nectar diet (1:1 sucrose-water solution) prior to the experiment. Every day, flower visiting behaviour was recorded for one hour at 8am, 11am, 1pm, 3pm and 5pm. We then assessed two key metrics of pollinator efficiency to compare the two species: flower visit duration and number of flower visits per hour.



Figure 38: Fly pollination trials setup for experiment 1. Photo credit James Cook

#### **Experiments 2-4. Glasshouse Pollination Trials**

The methods and experimental treatments were essentially the same across these experiments, apart from the strawberry variety and identity and stocking rate of the flies being tested. All trials were conducted in two fully enclosed glasshouse chambers at the NVPCC. There were six suspended gutters per glasshouse chamber that held 80 strawberry plants each. As in the first experiment, the drone flies (*E. tenax*) were supplied by project partners, seedPurity, from their breeding colony in Tasmania, whilst blowflies (*C. stygia*) were purchased from Sheldon's Baits. Flies were always additionally provided with an artificial nectar solution (1:1 water: sucrose) to promote longevity.

To assess the effects of fly pollination on fruit weight, development time and quality, we conducted bagging experiments with three experimental treatments:

- 1. **Control** This was a closed bag control with only unassisted self-pollination. Nylon mesh bags were added prior to the opening of flowers and not removed. Flowers from the control group were later unbagged when they had passed the pollination receptive stage.
- 2. Hand pollination Closed bag with hand pollination. In the hand pollination treatment, flowers were bagged prior to opening and then hand pollinated using a brush with pollen from an adjacent plant of the same variety. Hand pollinated flowers were then bagged again to prevent further pollination by flies. Flowers from the hand pollination group were later unbagged when they had passed the pollination receptive stage.

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3. **Fly pollination** – Open fly pollination with no bag. In the open fly pollination treatments, flowers were selected prior to opening and then left unbagged until the fruits were removed at harvest.

For all treatments, we recorded the date of treatment and date of harvest for each fruit. Ripe fruits were weighed and classified according to a commercial grading system with grades from A (best) to E (worst) (Figure 39).



Figure 39: Example of strawberry quality grading system from A grade with very little deformity to E grade, in which most part of the fruit is underdeveloped due to insufficient pollination intensity.

### **Results**

#### **Experiment 1. Fly Species Screening**

Both the drone flies and blowflies readily visited strawberry flowers and appeared to be good candidates for glasshouse pollinators. Both species visited flowers most frequently during the late morning (10am) (Figure 40). At this stage it is not clear whether this peak in activity towards the middle of the day was due to higher temperatures within the glasshouse or other factors such as diurnal variation in daylight and irradiance, or intrinsic behavioural rhythms of the insects.



Figure 40: Mean number of strawberry flower visits per fly in blow fly (*Calliphora stygia*) and drone fly (*Eristalis tenax*) between 7am and 5pm under glasshouse conditions.

The drone flies made significantly longer visits to strawberry flowers than the blowflies (t = 2.57, df = 15.4, p < 0.05) (Figure 41). However, blowflies made significantly more visits per hour than the drone flies (t = -2.43, df = 19.51, p < 0.05) (Figure 41). Both the number and duration of flower visits can be considered as proximate measures of the effectiveness of a pollinator, with longer durations and greater number of visits suggesting higher effectiveness. As such, both species show promising but contrasting attributes as pollinators of strawberries. Both species warranted further investigation, and more data were needed to determine the most effective pollinator of the two species.



Figure 41: A) mean duration of flower visits per fly and B) mean number of visits per fly per hour

#### **Experiment 2. Glasshouse Trials of Blowflies with High Stocking Density**

We placed 400 blowfly pupae into each of two chambers containing flowering strawberries (Red Rhapsody cultivar). We counted the number of flies that successfully eclosed after seven days to determine the number of adults in each chamber: 324 and 366 flies respectively. The number of flowers was also counted each day. Because the trial occurred late in the season, flowering density was low, providing a relatively high daily average of 0.3 and 0.06 flies per available flower. Analysis of the data from this trial revealed that fruit weight was considerably higher with fly pollination than with hand or no pollination (Figure 42).



Figure 42: Average fruit weight (SE) across three pollination treatments

#### **Second Blowfly Trial**

In October to December 2021, we conducted a second trial with the blowflies. We ordered two batches of blowflies. The first batch that arrived emerged with defective wings (Figure 43) and could not fly properly, possibly due to a delay in transportation or genetic defects caused by the breeding conditions of our supplier. Instead of discarding these flies, we released 50 deformed wing flies into each of the two chambers. We tagged >15 of the open flowers per chamber and designated them as a deformed-wing fly pollination treatment (DF). This additional treatment group helped us understand the impact of fly health on pollination efficiency.



Figure 43: An adult blow fly (Calliphora stygia) with deformed wings.

When deformed wing blowflies were no longer observed in the glasshouse chambers, we repeated the blowfly experiment with healthy flies. 450 and 504 healthy flies were introduced to the first and second chamber respectively. The numbers of flies were recorded by counting individual flies as they were released one-by-one into the chamber. Each chamber had at least 30 floral replicates of each of the three treatments: fly, hand and control flowers. Whole-day video footage of 6-8 flowers was used to confirm that flies visited strawberry flowers. After pollination, flowers were tagged and monitored until they developed into fruit. They were then harvested and assessed for size and quality.

We found that fly-pollinated fruits were the heaviest and developed the quickest, followed by hand pollinated flowers (Figure 44). Control (bagged) flowers and those pollinated by deformed wing flies had the lowest weights and longest development times to fully ripen. Furthermore, flowers pollinated by healthy flies produced the best quality fruits, again followed by the hand pollinated group, bagged group and deformed wing group respectively (Figure 44). In addition, 15% of bagged flowers, and 8.5% of deformed wing flowers did not develop into fruit at all.



Figure 44: Pollination outcomes from the blowfly glasshouse experiment. Graphs compare fruit weight (A) and development time (B) by pollination treatment. In both cases healthy fly pollination is significantly better than hand pollination, which is significantly better than the control group or deformed flies. The stacked column chart on the right shows proportions of fruits of different qualities from A (best) to E (worst). Again, the best outcome is with fly pollination.

The video footage confirmed that blowflies were foraging on the flowers (Figure 45) and the maximum time that a blowfly spent foraging on a flower was 148 seconds (n = 73).



Figure 45: A blowfly (Calliphora stygia) foraging on a strawberry flower.

#### **Experiment 3. Glasshouse Trial of Drone Flies with High Stocking Density**

In February to March 2022, we conducted a similar trial to assess the ability of drone flies to pollinate strawberry flowers. The same experimental design was used as for the second blowfly trial outlined above, with 30 replicates of each treatment in each of two NVPCC glasshouse chambers. We introduced 103 and 112 flies into chambers 1 and 2, due to the reduced density of flowers in this trial (because of the crop being older and later in the season).

We found that fly-pollinated fruits were the heaviest and developed the quickest, followed by hand pollinated flowers (Figure 46). Control (bagged) flowers had the lowest weights and longest development times. Furthermore, flowers pollinated by healthy flies produce the best quality fruits, again followed by the hand pollinated group, bagged group and deformed wing group respectively (Figure 46).



Figure 46: Pollination outcomes from the drone fly glasshouse experiment. Graphs compare fruit weight (A) and development time (B) by pollination treatment. In both cases fly pollination is significantly better than hand pollination, which is significantly better than the control group. The stacked column chart on the right shows the proportions of fruits of different qualities from A (best) to E (worst). Again, the best outcome is with fly pollination.

In addition, we trialed tagging individual flies using queen honeybee tags (Figure 47). This was successful and allowed continuous monitoring of the activity of an individual fly for up to 30 minutes. This novel approach allowed us to see that an individual fly would sometimes make repeated visits (up to 5 recorded) to the same flower in a single day and often spend up to 2 minutes (max 6.53) on the flower in a single visit. It may also allow better assessment of wider foraging patterns and fly lifespan in the glasshouse setting in future studies.



Figure 47: A tagged Eristalis tenax drone fly foraging on a strawberry flower in the glasshouse.

In summary, experiments 2 & 3 established that both fly species were effective pollinators of glasshouse strawberries. In our trials, we found that average fruit weight was higher in the blowfly trial and fruit quality was higher in the drone fly trial. However, as these were successive trials, the strawberry plants were older and the season later for the drone fly trial and this led to fewer and smaller flowers. To counteract this, we used about 100 flies per chamber with the drone flies, as opposed to about 400 with the blowflies. Smaller flowers, rather than the fly species, is probably the reason for slightly lower fruit weights with the drone fly trial and may also contribute to the difference in fruit qualities in the two trials. Nevertheless, the main result was that fruit weight and quality outcomes were very good with both species. To further compare the two pollinators and gain further insight to stocking densities we next planned to compare the two species side-by-side in different chambers, but at the same time and stage of crop maturity.

## Experiment 4. Simultaneous Glasshouse Trial of Drone Flies and Blowflies with Low Stocking Densities

In this final glasshouse trial, we continued work with the blowfly *C. stygia* and drone fly *E. tenax* that we showed previously to be effective pollinators of glasshouse strawberries. We did this experiment twice, stocking one chamber with blowflies and the other with drone flies. We used 80 flies per chamber in the first run and 120 in the second run. Based on previous trials we thought these fly numbers were about the lowest that could provide good pollination outcomes, and we also hoped to compare the relative performance of the two fly species. Each chamber contained 480 strawberry plants (Lowanna variety), and thus a ratio of one fly per 6 plants (run 1) or per 4 plants. However, because run 2 was with a new crop with more flowers, we believe that the fly to flower ratio was very similar. In addition, we individually tagged almost all of the flies in each chamber with honeybee queen tags (Figure 48).



Figure 48: Tagged flies (a) *Eristalis tenax* fly foraging on a strawberry flower, (b) *Calliphora stygia* fly trying to access bagged control flower, (c) *Calliphora stygia* fly on strawberry leaf.

After the flies were released, we bagged un-opened flower buds. When the flower buds opened, we then randomly assigned open flowers to the standard three pollination treatments: fly (open) pollination, hand pollination control and bagged control. We repeated the process daily until there were about 100 replicates per treatment. We also placed 5 video cameras on tripods per chamber to record fly visits to open flowers to monitor fly visitation rate and behaviour on the flowers. These videos were kept running for a full day on a single flower, apart from one that was used to monitor visitation at a sugar feeder. The video files were then transferred to external hard drives for later analysis.

Within this trial, we also conducted a single visit experiment to test the impact of one visit by a drone fly on fruit quality. After the flies were released into the experimental chambers, we bagged un-opened flower buds. When the flower buds opened, we then removed the bags and observed individual flowers until the first drone fly visited. We recorded the length of time the fly spent on the flower to test if this was correlated with fruit quality. The flower was then re-bagged until all petals had fallen off and pollen had been released from the anthers (7-10 days), after which bags were removed so that they did not affect fruit development. Ripe fruits were harvested for assessment of fruit quality as in previous work.

Tagging was fairly successful in both fly species, with only a few attempts leading to individual flies with reduced mobility, and plenty of observations of marked flies visiting flowers. As noted in previous trials, the drone flies were mostly observed visiting flowers, while the blowflies were observed landing in various places, including both strawberry leaves and flowers. In fact, blowfly visitation was not observed often enough to allow for a sufficient amount of single visit data to be collected.

As in the previous glasshouse trials, both fly species produced significantly higher quality fruits than either the hand pollination or control treatments (Figure 49 and Figure 50). In addition, we found that a single (controlled) drone fly visit, while better than no fly pollination, was significantly worse than hand or open fly pollination (Figure 50). The results for open drone fly pollination were particularly good (Figure 50), outperforming the blowflies (Figure 49) at a similar density and fly:plant ratio. Nevertheless, while these trials were performed at the same time, unlike previous ones, it is possible that a chamber effect could contribute to this difference. However, the repeated pattern across trials suggesting that drone flies are the better pollinators is also consistent with their more flower-centric behaviour and longer visit times to flowers, so is probably a genuine performance difference.



Figure 49: Fruit quality outcomes from different pollination treatments in the blowfly chamber of the final experiment. The chart shows proportions of fruits of different qualities from A (best) to E (worst) for the different treatments. Fly pollination is significantly better than hand pollination, which is significantly better than the control group.



Figure 50: Fruit quality outcomes from different pollination treatments in the drone fly chamber of the final experiment. The chart shows proportions of fruits of different qualities from A (best) to E (worst) for the different treatments. Fly (open) pollination is significantly better than hand pollination, which is significantly better than the other treatments. Finally, the single fly visit treatment is significantly better than the control (no flies) treatments.

In summary, experiment 4 provided further evidence that both fly species can be highly effective pollinators of glasshouse strawberries. This result was achieved with a stocking density of one fly per four plants, which produced excellent fruit quality results with drone flies and very good results with blowflies, which may still benefit from a slightly higher stocking density.

#### Key outcomes and recommendations

- 1. We have shown that both the blowfly, *C. stygia*, and the drone fly, *E. tenax*, are very good pollinators of glasshouse strawberries, capable of producing high quality strawberry crops with modest stocking rates in closed glasshouses.
- 2. Overall, our results suggest that *E. tenax* is a more effective pollinator (per fly) due to its stronger attraction to flowers and longer average visit time. However, the less flower-centric *C. stygia* also performed very well in a closed glasshouse environment. The difference might be greater in open protected cropping structures with the blowflies more likely to abandon the crop.
- 3. A stocking ratio of one drone fly per four plants yielded excellent fruit quality outcomes in our final experiment. A similar ratio for blowflies did not give quite such impressive results but was still very good. We would therefore recommend perhaps doubling the stocking rate for blowflies.
- 4. *C. stygia* is readily available from some suppliers of fishing bait (we used Sheldon's baits in SA) as packages of 500 pupae for about \$30 and it is fairly easy to hatch the flies and then introduce them to the crop. *E. tenax* is not yet easily available "off the shelf", but project partners seedPurity have developed good rearing methods during this project and *E. tenax* may soon be more readily available to buy.
- 5. These fly species offer a complementary glasshouse pollination option to managed bees. The flies can be bought in and applied to the crop as a one-off "treatment", whereas hive bees are valuable livestock that also need to be managed (or rented with a management contract).
- 6. While flies are effective pollinators for glasshouse strawberries, it is possible that fly-pollinated strawberries will be less appealing to customers, and this may warrant further consideration and study.

# Managed Fly Pollinators for Vegetable Seed Crops

## seedPurity and University of New England

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## Introduction

Australia produces approximately 2700ha of vegetable seed crops annually with an estimated farm gate value of \$40 million. Production is centred in south-eastern Australia (NSW, SA, VIC and TAS) and largely services export markets in Europe, Asia and North America, as well as the domestic market. The Australian industry has established a reputation for reliable, high quality carrot seed production, which has resulted in significant growth. Approximately 1800ha of carrot seed is currently produced in Tasmania (950ha), South Australia (600ha) and, to a lesser extent in New South Wales and Victoria. Other major insect-pollinated vegetable seeds grown in Australia include brassicas (cabbage, cauliflower, broccoli, kohlrabi, brussels sprouts and Asian greens); alliums (bulb and bunching onions and leeks); and other Apiaceae crops, including celery and parsnip. These crops typically feature small, unspecialised white, yellow or green flowers clustered in umbels (Alliaceae and Apiaceae) or racemes (Cruciferae), and are predominantly pollinated by honeybees, both managed and wild. A diverse assemblage of other insects including flies, native bees, wasps and beetles also contributes to pollination in open field crops (Howlett *et al.*, 2005; Rader *et al.*, 2009; Gaffney *et al.*, 2011), where most vegetable seed is grown. In other settings such as cages, glasshouses or polytunnels, which are commonly utilised for production of breeding lines and small, high-value seed lots, flies can be used as an alternative or supplement to honeybee pollination (Clement *et al.*, 2007).

Much of the vegetable seed grown in Australia is hybrid seed, which is produced by pollinating a seed bearing (female) parent line with pollen from a pollen donor (pollinator) parent line. Pollinator and female hybrid seed parent lines are typically grown in alternating 1m to 5m wide strips within the crop to facilitate pollen transfer and separation of the parent lines during harvest. To ensure hybridisation, the seed-bearing line features either male sterility (does not produce pollen) or strong self-incompatibility (produces pollen but is largely incapable of self-fertilisation) (Brown *et al.*, 2006).

The prevalence of hybrid seed production in Australia is important because inadequate pollination can be a yield-limiting constraint in many hybrid vegetable seed crops, including carrot (Erickson and Peterson, 1979; Spurr, 2003), brassicas

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(Brown *et al.*, 2006) and onion (Delaplane and Mayer, 2000). Underlying factors restricting pollination include the need for insects to move from a pollinator to a male sterile plant to effect pollination (Erickson and Peterson, 1979; Rodet *et al.*, 1991). Another important factor is the differences between hybrid parent lines, which elicit discriminatory foraging behaviour in honeybees and often result in one parent line being visited to the exclusion of the other due to variation in flower colour and morphology, nectar production rates, and/or nectar quality (Erickson and Peterson, 1979; Rodet *et al.*, 1991). In addition, honeybees sometimes find carrot and onion seed crops less attractive than other nearby forage sources and leave the target crop to forage elsewhere (Delaplane and Mayer, 2000). Enclosure coverings in protected crops can compromise the ability of honeybees to navigate and forage normally (Dyer and Gould, 1981; Hall *et al.*, 2019), whilst periods of inadequate pollination may also result from low temperature conditions limiting honeybee activity, particularly in early season brassica crops.

The challenge of optimising pollination is reflected in the relatively high stocking rates for managed honeybees in hybrid vegetable seed crops, which are typically around 10 hives/ha. This demand for hives can cause local supply issues and is reflected in the relatively high price growers pay for crop pollination services. For example, in Tasmania, where there are around 28,000 registered beehives (AgriGrowth Tasmania, 2022) approximately 10,000 will be required in mid-summer to pollinate the hybrid carrot seed crop but this need coincides with strong demand for hives from other pollination dependent industries and occurs during the peak period of leatherwood honey production, which is a mainstay of income for Tasmanian beekeepers. Given the substantial reliance on managed honeybees to pollinate vegetable seed crops, and the challenges that this can present for achieving reliable yields in some situations, efforts to develop alternative and complementary pollinators for use in this context are warranted.

After bees, flies are one of the most common pollinators to visit vegetable seed crops (Bohart and Nye, 1960; Bohart *et al.*, 1970; Howlett *et al.*, 2005; Rader *et al.*, 2009; Gaffney *et al.*, 2011) and are thus an obvious target for research in this area. To date, attention has focused largely on the use of flies for mass pollination in covered systems, including individually bagged plants, small cages, polythene tunnels and glasshouses (Clement *et al.*, 2007; Currah and Ockendon, 1984; Faulkner and Hinton, 1980; Howlett, 2012; Wilson *et al.*, 1991). Comparatively few studies have examined the potential for flies to be used as managed vegetable seed crop pollinators in open settings. Animal manure and carrion have sometimes been introduced into crops in the hope that flies and other insects will be attracted to the field from elsewhere and contribute to pollination (Roberton *et al.*, 2023). However, information regarding the timing of such introductions, their impact on pollinator species composition and abundance, and any demonstrated improvements in pollination and seed yield outcomes in open field settings is scarce.

Although there is some evidence that species such as the European Drone Fly (*Eristalis tenax*; Diptera:Syrphidae) could be well suited to mass rearing for crop pollination (Spurr, 2003; Rader *et al.*, 2009; Howlett *et al.*, 2017; Gaffney *et al.*, 2018), specific information relating to the use of mass-reared flies as managed field crop pollinators is lacking (Howlett and Gee, 2019). We undertook this study to identify and develop promising candidate fly species for use as vegetable seed crop pollinators in both open and protected cropping systems. The scope of our work included: crop surveys to identify promising candidate species in TAS, SA and NSW; evaluation of their effectiveness as pollinators relative to honeybees (the industry standard managed pollinator) in open and protected settings; and, in conjunction with developing the mass-rearing protocols and deployment strategies described elsewhere in this report, preliminary evaluation of fly dispersal, retention and crop yield outcomes following pilot-scale open field releases in Tasmania. Additionally, we investigated habitat augmentation strategies to promote wild populations of promising fly pollinators in and around carrot seed crops. This work will not only be of direct benefit to the vegetable seed industry but also to other horticultural industries interested in developing managed fly pollination through identification of non-specialised fly pollinators suited to mass rearing and experience gained in deployment of managed fly pollinators into commercial cropping systems.

## **Methods**

#### **Pollinator Surveys**

#### seedPurity, TAS and SA; University of New England, NSW

Pollinator surveys to identify candidate fly species with potential for development as managed pollinators were conducted in commercial hybrid carrot seed crops grown at 11 sites in 2018 and 2019 in the Derwent Valley, Coal River Valley and northern Midlands (TAS) and Naracoorte and Mt Gambier (south-east SA). In addition, 4 open pollinated and hybrid crops

#### **Hort Innovation**

grown near Griffith (NSW) were surveyed in 2021. Survey crops varied in size from 1 to 20ha.

To avoid sampling method bias, 3 survey techniques were employed in TAS: sweep netting (sweeping), flight intercept traps, and visual counts (Figure 51). In SA survey methods were visual counts only. Visual counts and sweeping surveys were conducted along five 30m transects of adjacent pollinator and male sterile beds in each field, with one transect located in each quarter and one in the centre. Each site was surveyed at weekly intervals on 4 to 5 occasions during the main bloom period (December - January) on days when weather conditions within the crop were favourable for pollinator activity (i.e. wind speed <18km/hour, no rainfall between 6am and commencement of surveys). Visual surveys were performed by walking (0.5m/second) along a 20m bed section within the transect and recording all insects foraging on flowering carrot umbels. Individual sweeping surveys comprised 10 back and forth net sweeps at canopy level over a 10m section of a single bed. Flight intercept traps were positioned 20m from the end of each survey transect between the pollinator and male sterile beds. Visual and sweeping surveys were conducted between 10am and 4pm to coincide with the main period of viable pollen and nectar availability within carrot seed crops (Spurr, 2003). The timing of individual surveys at each site was varied within this window, to prevent temporal bias in sampling. Flight intercept traps were deployed for a 24-hour period in each survey.



Figure 51: Tasmanian hybrid carrot field surveys. a) Jack Connell emptying a flight intercept trap; b) Emma Spurr conducting counts of floral visitors; b) Tracy Thornley sweeping a crop. Photo credits: Diane Spurr, seedPurity.
In NSW, pollinator surveys comprised visual counts along transects only, but the range of times over which surveys were conducted was extended to 05:00–9:00, 9:00–13:00, and 13:00–17:00, to account for hotter and drier climatic conditions. Surveys were conducted along two 10 m transects: one along the edge of the carrot field, and one positioned towards the middle of the field. Each transect consisted of 2 adjacent beds of plants; in hybrid crops this meant that transects incorporated adjacent pollinator and male sterile beds, with both being surveyed. Data loggers placed at each survey site recorded climate data to identify relationships between insect abundance patterns and environmental conditions.

Identifications of samples collected in sweeping and flight intercept surveys were derived from multiple sources, including the online Australian Pollinator Database subsection of PaDIL (Department of Agriculture, Water and the Environment); the citizen science site 'Insects of Tasmania: An Online Field Guide' (Daley and Ellingsen, 2012); and 'A Guide to Native Bees of Australia' (Houston, 2018). Pollinators observed in visual surveys were identified as far as practical under field conditions, based on defining visible features and (for the major pollinator groups) voucher specimens that were collected and formally identified.

### **Pollinator Efficiency**

#### seedPurity, TAS; University of New England, NSW

Pollinator efficiency studies were conducted in NSW in 2021, in a commercial hybrid carrot seed crop grown near Griffith. Studies were performed both in open conditions and in netted enclosures 3m (L) x 3m (W) x 2m (H) positioned over adjacent rows of pollinator and male sterile parent lines in the same crop.

In TAS, large cage trials investigating pollinator efficiency were conducted at South Pacific Seeds trial ground in Richmond (2020 and 2021). Three 30m (L) x 10m (W) 2.4m (H) netted seed production cages were erected over 6 beds of a model hybrid carrot seed parent line combination sown in a 4:2 (MS:pollinator line) row arrangement (Figure 52). Prior to flowering, each cage was divided into three 10m x 10m rooms with vertical, insect proof net walls. Open field work was also conducted in commercial hybrid seed crops and a hybrid carrot seed production trial located near Richmond (TAS). All field sites featured a brown-anther MS parent line grown in a 4:2 (MS:pollinator line) row arrangement.



Figure 52: a) Pollination cages used for pollinator efficiency trials in TAS; b) Hannah Allwright and Annik Witte (SPS TAS) scoring foraging behaviour within a pollinator efficiency trial.

### **Pollen Loading**

#### seedPurity, TAS

In 2020, representative specimens of 11 focal pollinator species were collected from MS line plants in a commercial crop at Richmond (TAS) to assess pollen loading. Target insects were collected directly from flowering umbels into sample phials, euthanised and assessed for pollen loading using a rinse method followed by microscopic examination of the rinsate (Spurr, 2003). The corbiculae were removed from *A. mellifera* prior to rinsing. To evaluate the viability of pollen carried by *E. tenax* and *A. mellifera*, we adapted the rinsing procedure to include FCR (fluorochromatic reaction) staining (Spurr, 2003) and evaluation under a UV fluorescence microscope.

#### Foraging Behaviour and Single Visit Pollen Deposition (SVD) Experiments

#### seedPurity, TAS; University of New England, NSW

Given the necessity of cross pollination for hybrid seed production and evidence that pollinators display discriminatory foraging of seed parent lines (Erickson *et al.*, 1979; Galuszka *et al.*, 1989), we investigated foraging behaviour by visually tracking the movement of three focal pollinator species (European honeybees (*Apis mellifera*), hoverflies (*Eristalis tenax*) and brown blowflies (*Calliphora stygia*) within and between parent lines in a 1ha open hybrid carrot trial in Richmond, TAS. On 4 days during peak bloom, we assessed pollinator activity in spot counts of 20m transects in 3 time intervals 10am to 12noon, 12noon to 2pm and 2pm to 4pm. During the peak period of activity for each species, 10 individuals of that species were located on a bed of the pollinator line and their next 10 moves to other umbels recorded in terms of parent line (pollinator or MS) and row position.

We assessed insect-mediated pollen transfer by offering umbellets excised from pre-bagged virgin MS umbels to target insects in the open or in cages (NSW) or allowing an individual to visit pre-bagged virgin umbels in large seed production tents stocked with the target pollinator species (TAS). A visit was recorded when the insect commenced feeding/probing flowers for nectar or pollen, and visits were timed until the insect left the umbellet. When visits were to *in-situ* umbels (TAS), we also counted the number of umbellets contacted by the insect during its visit. At visit completion, the umbellet was sub-sampled and 15 representative florets were scored for pollen deposition (in TAS we used the 1<sup>st</sup> umbellet contacted during the umbel visit). Pollen deposition per stigma and the number of pollinated flowers per umbellet were estimated by observing the number of carrot pollen grains adhered to the stigma surface of flowers mounted in basic fuchsin jelly and examined under a compound microscope (Spurr, 2003).

In NSW, focal insect species for the SVD experiments were *A. mellifera*, the native sweat bee (*Lasioglossum cognatum*) and two hoverflies (*E. tenax* and *Eristalinus punctulatus*). While the two bee species readily visited umbellets offered to them in open field conditions, the two hoverfly species did not, and so to collect SVD replicates, we caught wild hoverflies from the adjacent crop and placed them within cages. For both bee species and *E. tenax*, we collected SVD data in two time periods: before noon (05:00–12:00) and afternoon (12:00–17:00). Data for *E. punctulatus* visits were only collected in the morning period as these flies usually visited flowers before noon. In Tasmania, focal insect species for SVD experiments were the *A. mellifera*, the *C. stygia* and *E. tenax*, and all samples were collected from *in-situ* visits between 11:00 and 14:00. At both locations, we also sampled between 1-3 unvisited umbellets per virgin umbel to test if pollen flow occurred without pollinator visitation.

#### Seed Yields in Large Cage Trials

#### seedPurity, TAS

Seed yield data were collected from the Tasmanian carrot seed cage trials to compare the performance of two potential managed fly pollinators (*E. tenax* and *C. stygia*) with that of *A. mellifera* (the current industry standard managed pollinator). In 2020, the rooms within the cages were individually stocked with *A. mellifera* (1 nucleus hive with ca. 3000 bees), *E. tenax* (600 adults) or *C. stygia* (600 adults). In 2021, the rooms within the cages were individually stocked with *A. mellifera* and *E. tenax* (1 nucleus hive per room), *E. tenax* only (600 adults), or a combination of *A. mellifera* and *E. tenax* (1 nucleus hive plus 600 hoverflies). Each year, individual pollinator treatments were replicated 3 times (9 rooms total) in a randomised complete block design. Seed yields were determined for 40 whole plant samples taken from each room. Samples were dried, Hort Innovation

threshed, and cleaned to a commercial standard.

#### Mass Release of E. tenax in Tasmanian Hybrid Carrot and Brassica Vegetable Seed Crops

#### seedPurity, TAS

In collaboration with industry partners SPS, BSA and TPS, seedPurity monitored commercial-scale, open-field releases of mass-reared *E. tenax* as a complementary pollinator to honeybees in hybrid carrot and brassica seed crops between 2021 and 2024. The key objective was to understand fly survival, dispersal and retention within the target crop to inform deployment strategies. Field sites varied between 2ha and 17.5ha, with stocking rates ranging from 5,000 to 60,000 pupae per hectare. We arranged a grid of between 10 and 61, 20m long transects at each study site and, at weekly intervals during flowering, conducted visual counts of *E. tenax* and *A. mellifera* foraging on pollinator and MS line flowers within the transects. Surveys were conducted between 10:00 and 14:00 each day, under favourable conditions for pollinator activity. Survey data were visualised using the geocoordinate function with Heatmapper (Wishart Research Group, University of Alberta), a web-based application used under creative commons license.

Following on from a separate industry funded study that demonstrated pollination limitation of seed yields in hybrid Brassica oleracea seed crops in Tasmania (Spurr and Lucas, unpublished data), we conducted field experiments in 2023 and 2024 to determine whether complementary stocking with E. tenax and A. mellifera improved yield outcomes in hybrid cauliflower and hybrid broccoli seed crops. Three commercial sites, each approximately 2ha in size, were selected near Richmond (TAS) for the study. Each site was stocked with A. mellifera for the full flowering period, in accordance with commercial production standards, but drone fly stocking regimes varied between sites. Sites 1 and 3 were stocked with E. tenax only for the latter half of peak bloom, while Site 2 was stocked with them only for the first half of peak bloom. Stocked flies at Site 2 were eradicated after 2 weeks by application of a low residual contact insecticide, applied at night to avoid disruption to A. mellifera and native bees. Pollinator activity was surveyed 2 - 3 times per week for the duration of the flowering period, along a grid of twelve 20m transects spread throughout each crop. On each survey date, we applied a supplemental hand pollination treatment to 5 flowers on 2 plants in each transect and tagged a matching set of insect-only pollinated flowers (i.e. a total of 120 flowers per pollination treatment on each survey date). The supplemental pollination treatment was used to represent yield potential under conditions of unlimited pollination, whereas the insect-only treatment represented realised yield potential under the different pollinator stocking regimes. At maturity, pods from plants receiving each pollination treatment were collected for yield determination (% of flowers setting pods, and number and weight of seeds/pod). To compare complementary pollination with A. mellifera-only pollination, we standardised yield data from the insect-only pollinated flowers against the corresponding data for supplementally pollinated flowers from the same stocking regime.

#### Habitat Augmentation to Support Wild Fly Pollinators in Carrot Seed Crops

#### University of New England, NSW

In 2022 we investigated whether non-floral habitat could attract and support reproduction of *E. tenax* and *E. punctulatus*, using portable pools deployed at 4 commercial hybrid carrot fields in Griffith, NSW (Figure 53, a). Since *E. punctulatus* was observed ovipositing on decaying male carrot plants removed before harvest, we tested two habitat treatments: (1) decaying carrot plants with water, and (2) decaying carrot plants with water and farm soil. One of each treatment was placed at 1-2 locations at each site (14 pools in total) and left to decay for 12–21 days. Pools were then examined to count syrphid fly eggs and larvae, record where eggs were laid within the pools (e.g., stems, roots, umbels, leaves), and determine larval instar stages (the period in which the maggot is growing). Syrphid fly larvae were reared to adults to confirm species identifications (Figure 53, b).



Figure 53: Experimental design of the habitat pools deployed to attract eristaline flies: (a) habitat pool yet to be filled with water within a seed carrot field; (b) an adult, female *Eristalis tenax* (Linnaeus, 1758) fly within a deployed habitat pool. Arrowheads are pointing to the habitat pool and adult eristaline fly for clarity.

# Results

#### **Pollinator Surveys – TAS and SA**

#### seedPurity

Excluding insects <3mm in length, we observed 25,572 floral visitors in carrot seed crops in the TAS and SA surveys, comprising at least 78 different taxa (31 species and 47 morphospecies were identified). Although the abundance of different taxa varied between survey methods in Tasmania (Figure 54), overall trends were broadly similar. The main insect orders observed, beginning with the most abundant, were Hymenoptera (bees and wasps), dominated by *A. mellifera* from managed hives stocked into the crops; Diptera (flies); and, in TAS but not SA, Coleoptera (beetles), dominated by nectar scarabs, lady beetles and soldier beetles (Figure 55). We identified 24 families of flies that visited carrot flowers (Figure 56). The dominant species (by number) were the common housefly (*Musca domestica*); the blowflies, *C. stygia* and *Calliphora vicina* (Bluebottle Fly) and *Lucilia sericata* (Common green bottle fly); and the hoverflies: *E. tenax, Simosyrphus grandicornis* and *Melangyna viridiceps* (Figure 56 and 57).



Figure 54: Composition of surveyed floral-visiting insects within hybrid carrot seed crops in TAS. Survey data includes visual surveys, sweep nets, flight intercept traps and a total average of all survey methods. Composition is represented as the percentage of insects from three main orders (Hymenoptera, Diptera, and Coleoptera), and "other" orders.



Figure 55: Composition of surveyed floral-visiting insects within hybrid carrot seed crops in SA from visual surveying. Composition is represented as the percentage of insects from Hymenoptera and Diptera orders, with the Dipteran composition broken down into families. Other Syrphidae flies include the species *Simosyrphus grandicornis* and *Melangyna viridiceps* 



Figure 56: Abundance of different families of dipteran floral visitors in a representative subset of 4 hybrid carrot seed crops in the TAS.



Figure 57: Key floral-visiting fly (*Diptera*) species of hybrid carrot seed crops in Tasmania. Species include: a) *Eristalis tenax* (Syrphidae, Common Drone Fly); b) *Calliphora stygia* (Calliphoridae, Brown Blowfly); c) *Australophyra rostrata* (Muscidae, Black Carrion fly); d) *Melangyna viridiceps* (Syrphidae, Hoverfly); e) *Lucilia sericata* (Calliphoridae, Common green bottle fly); f) *Calliphora vicina* (Calliphoridae, Bluebottle fly). Photo credit: Amy Lucas, Cameron Spurr, and David Cook.

## **Pollinator Surveys - New South Wales**

# University of New England

Out of 26,083 insects observed in the Griffith (NSW) survey, we identified 52 different insect taxa (33 species and 19 morphospecies) from 26 families as floral visitors of carrot seed crops. The vast majority (87.3%) of all observed floral visitors belonged to four families, namely Coccinellidae (lady beetles), Syrphidae (hoverflies), Apidae (long-tongued bees), and Halictidae (sweat bees) (Figure 58). Within these families, six species were considered the dominant floral visitors based on their high abundance (> 800 visits in total): two bee species (*Apis mellifera* and *Lasioglossum cognatum*), two hoverfly species (*S. grandicornis* and *M. viridiceps*), and two lady beetle species (*Hippodamia variegata* and *Coccinella transversalis*). Other Syrphid species including *Eristalis tenax* and *Eristalinus punctulatus* appeared in the crops at low numbers.



Figure 58: Abundance of different families of insect floral visitors in carrot seed crops in the Riverina region of New South Wales, Australia. The insect order of each family name is differentiated by colour. The total numbers of individuals (n = 26,083) seen per family while conducting floral visitation surveys are listed on the figure for clarity.

Lady beetles are well-known biological control agents, with both adults and larvae feeding on aphids and other soft-bodied pests (Dixon, 2000; E. W. Evans, 2009). They are also known to consume floral resources (pollen and nectar) (Bertrand *et al.*, 2019), especially when prey is limited or scarce (Lundgren, 2009). While they were most likely present in survey crops due to the prevalence of aphids, they also likely acted as incidental pollinators. *A. mellifera* were present in large numbers due to hives stocked near the crops for pollination, while the sweat bee *L. cognatum*, a solitary ground-nesting species, was observed nesting in large numbers along flood irrigation levees around the field edges. Both bee species were most active at 30–40 °C and 0–66 % RH (Figure 59a and 59b). Unlike bees and lady beetles, the two syrphid fly species (*S. grandicornis* and *M. viridiceps*) peaked at mid-range RH levels (Figure 59b), being most abundant between 20–40 °C and 33–66 % RH (Figure 59a and 59b), which suggests that RH may exert a stronger influence on their flower visitation than temperature. These flies, like lady beetles, may contribute to both pollination and biological control in crops, as the immature stages of *M. viridiceps* and *S. grandicornis* prey on soft-bodied arthropods (Soleyman-Nezhadiyan, 1996), while adults rely on floral resources and often carry pollen on their bodies (e.g., thorax, legs) (Cook, Voss, *et al.*, 2020; Gross *et al.*, 2017).



Figure 59: Mean abundance of six dominant floral visitors ('Species') in commercial seed carrot crops grown in the Riverina region of New South Wales, Australia, in relation to environmental factors: (a) temperature (°C); and (b) relative humidity ('Humidity (%)'). Abundance data is a compilation of visitation surveys conducted on hybrid male fertile (n = 106), hybrid male sterile (female; n = 106), and open-pollinated (n = 60) seed carrot plant lines (n = 272 in total). Environmental condition categories are differentiated by colour for clarity. Significant differences between species abundance are shown as asterisks ('\*\*\*': p < .0001; '\*\*': p < .001; '\*': p < 0.05). No significant differences in abundance are denoted by 'n.s.'.

Observed numbers of both syrphid fly species did not vary according to physical location within survey fields, whereas *A. mellifera* were more abundant in the middle of the fields compared to the edges and sweat bees occurred in greatest numbers on the edge of the fields (Figure 60). It is unclear whether pollinator displacement was a factor in this study, however it appears unlikely as aggressive behaviours were not observed between individuals and it was commonplace for both bee species to share carrot umbels due to their large size (50 to 150 mm in diameter). We suspect that *L. cognatum* bees were less abundant in the middle of fields because hybrid carrot management practices in this region (e.g., inter-row cultivation and flood irrigation of fields) may damage the nests of *L. cognatum* in fields, confining them to the more permanent levee banks and uncropped edges.



Figure 60: Mean abundance of six dominant seed carrot floral visitors based on the physical location ('Edge' or 'Middle') of plants within crop fields. Surveys were conducted along the edge (n = 166) of crop fields and > 30 m towards the middle (n = 105) of crop fields. Physical locations of the plants that insect surveys were conducted on within crop fields are differentiated by colour for clarity. Significant differences between locations are shown in asterisks (p < .0001, \*\*\*), while 'ns' indicates no significant difference.

#### **Pollinator Efficiency - Pollen loading**

#### seedPurity, TAS

Pollen loads carried by representative specimens of different floral visitors sampled in a commercial hybrid carrot seed crop in TAS are shown in Table 6. Honeybees carried the most pollen (2444 grains per insect), followed by native sweat bees, soldier beetles and the drone fly, *E. tenax*. Pollen loads carried by *E. tenax* (1127 grains per insect) were 23 to 66 times greater than those carried by other fly species at this site (*Calliphora stygia* and *C. vicina*, *M. domestica* and the syrphids *Melangyna* and *Simosyrphis*). Assessment of pollen viability revealed that 20.6% of total pollen grains carried by *E. tenax* were viable, compared to 30.4% for *A. mellifera*.

Order	Genus	Species	Common Name	Mean Pollen Load	±S.E.	n
Hymenoptera	Apis	mellifera	European Honeybee	2444	560	78
	Homalictus		Native Sweat Bee	1877	1009	13
	Lasioglossum		Native Sweat Bee	1475	395	5
Diptera	Eristalis	tenax	Hoverfly / Drone Fly	1127	438	51
	Calliphora	stygia	Brown Blowfly	17	12	46
	Calliphora	vicina	Blue Bottle Fly	26	13	40
	Melangyna/ Simosyrphis		Hoverfly	48	18	42
	Musca	domestica	Common House Fly	49	24	54
Coleoptera	Phyllotocus	rufipennis	Ti Tree Beetle / Washing Beetle	788	214	45
	Cauliognathus	lugubris	Soldier Beetle	1428	481	55
	Coccinella	undecimpunctata	11 Spotted Ladybird	596	186	12

 Table 6: Pollen loading per individual insect for various common floral visitors observed in hybrid carrot seed crops.

 Standard error and sample size (n) are included. Corbiculae were removed from honeybees before washing.

We selected *E. tenax* and *C. stygia* as candidates for further evaluation as managed fly pollinators for vegetable seed crops in the TAS trials, and *E. tenax* and *E. punctulatus* for the NSW trials. *E. tenax* was chosen because it was a relatively common floral visitor in surveys at all TAS and SA sites and was observed to carry a comparatively large volume of carrot pollen on its body. It also appeared unlikely to cause nuisance when mass released into crops and has a life cycle compatible with mass rearing. Despite carrying much smaller loads of carrot pollen, *C. stygia* was selected for comparison because it was frequently observed in flowering carrot seed crops in the TAS and SA surveys and is already commercially reared in Australia. Although *E. punctulatus* was a less frequent floral visitor in the carrot seed crop surveys, it was included in the NSW trials because it occupies the same ecological niche as *E. tenax*, is morphologically similar (though smaller) and has the same life history, but favours warmer inland environments compared to *E. tenax*, which occurs more frequently in cooler and coastal locations. While both species overlap in the Riverina, it was hypothesised that the warm, dry conditions encountered in that area could favour *E. punctulatus* as a pollinator over *E. tenax*. Lastly, although the syrphids *Melangyna* and *Simosyrphis* may contribute to pollination and aphid control within crops, we did not consider either an ideal candidate for mass rearing because of the dietary requirement of their larval stage for aphids (Soleyman-Nezhadiyan, 1996).

#### **Foraging Behaviour and SVD Experiments**

#### seedPurity, TAS; University of New England, NSW

We compared the foraging behaviour of *A. mellifera*, *E. tenax* and *C stygia* under open field and cage conditions. In open field plots, more than 80% of moves between umbels or plants occurred within the one parent line (ie. pollinator umbel to pollinator umbel or male sterile umbel), as most movements took place within the same row of plants. This directional foraging was most likely driven by the greater proximity of umbels in a row compared with between rows. *E. tenax* crossed from the pollinator to the male sterile line most frequently (16.6% of moves), compared to 8.5% and 6.6%

of moves for *A. mellifera* and *C. stygia* respectively (Table 7). We observed active discrimination between parent lines based on floral preference when *A. mellifera* crossed between rows, but this was not apparent in both *E. tenax* and *C. stygia*. Most moves made by each species when crossing from the pollinator line to male sterile line occurred between adjacent rows of plants but *E. tenax* was most likely to cross to the male sterile line from plants in the inner rows of the pollinator block (33.5% of moves crossing from pollinator to MS line originated from inner rows) and both *E. tenax* and *C. stygia* were more likely than *A. mellifera* to cross directly from the pollinator line to inner rows in the male sterile block (Table 7). Discriminatory foraging as a yield limiting factor is a well-established phenomenon in hybrid vegetable seed production, as are declining seed yields across male sterile blocks with increasing distance from the nearest pollinator row. To combat these issues, breeders must carefully consider pollinator and MS floral traits when developing hybrid crosses, while growers have to balance pollinator and male sterile row ratios in crops, given that hybrid seed can only be harvested from the MS line. Behaviours displayed by the two fly species, including non-discriminatory foraging and a propensity to move greater distances between parent line plants, are desirable traits for managed pollinators in this production system (Table 7).

Table 7: Foraging behavior of *Eristalis tenax*, *Calliphora stygia*, and *Apis mellifera* in a hybrid carrot seed crop. "Selfing" refers to insect movement between/within pollinator rows; "Crossing" to movement from a pollinator row to a male sterile (CMS) row. CMS destination row indicates how many rows away from the origin (pollinator row) the insect traveled.

		E. tenax	C. stygia	A. mellifera
% pollinator	Selfing	83.4 ± 0.7	91.5 ± 2.1	93.4 ± 1.5
behaviour	Crossing	16.6 ± 1.5	8.5 ± 1.9	6.6 ± 0.6
	CMS destination row ♀(m)			
% of all crossing	1 (0.8)	80 ± 2.4	77.7 ± 22	100
moves (ơ to ♀)	2 (1.6)	20 ± 2.4	22.3 ± 22	0
	3 (2.4)	0	0	0

Under large cage conditions in TAS, the average amount of time each species spent foraging on an umbel before moving on was similar (around 55 seconds), but the number of umbellets covered per visit varied between species. *A. mellifera* visited 25 umbellets/minute, whereas *E. tenax* visited 15/minute and C. *stygia* visited 12/minute. Single visit pollen deposition rates varied between species and between trials (Table 8). In NSW, *Eristalinus punctulatus* pollinated more stigmas per umbellet (12%) and delivered more pollen to each pollinated stigma (8.2 grains) on average than either *A. mellifera* (8.1% and 3.5 grains) or *E. tenax* (4% and 1.3 grains) (Table 8). By contrast, in Tasmania, *E. tenax* pollinated more stigmas (35.8%) and delivered more pollen to each pollinated stigma (1.4 grains) on average than *A. mellifera* (32.2% and 1.1 grains) (Table 8). It is notable that in both trials improved pollination rates were associated with less time spent on individual umbellets, irrespective of species, possibly because more active foraging behaviour across the umbel surface improved coverage of individual flowers and pollen transfer rates. Table 8: Single visit pollen deposition (SVD) on hybrid carrot stigmas by common pollinators in NSW and TAS with comparison to self-delivered pollen (bagged). Includes total pollinated stigma percentages and visit durations; some umbellets were excised and presented to insects. Visit durations indicates the time spent on a single umbellet.

Species	Location	Umbelle presentati	t on	n	Stigmas/ n	Pollen grains/ stigma	Pollinated stigmas (%)	Visit duration (seconds)
Eristalinus punctulatus	Griffith NSW	Excised Offered	+	52	18	8.2	12.0	4.3
Apis mellifera	Griffith NSW	Excised Offered	+	51	19	3.5	8.1	12.1
	Richmond TAS	In-situ		15	30	1.1	32.2	2.4
Eristalis tenax Bagged (no insect- mediated pollination)	Griffith NSW	Excised Offered	+	59	17	1.3	4	13.7
	Richmond TAS	In-situ		15	30	1.4	35.8	4
	Griffith NSW	N/A		50	18	0.3	1.4	N/A
	Richmond Tas	N/A		15	30	0.1	2.2	N/A

Within the Tasmanian field and cage trials, we observed significant temporal and climatic effects on foraging behaviour (Figure 61 and Figure 62). *E. tenax* was most active in the morning (10am to midday), with a progressive reduction in activity into the afternoon. In contrast, *A. mellifera* most actively foraged on hybrid carrot umbels from midday into the afternoon. Foraging activity of *E. tenax* progressively reduced at temperatures exceeding 20 °C, a trend that typically coincided with later times of day (Figure 61). Peak foraging activity for *E. tenax* was consistently recorded within the 17–19 °C range (Figure 61). In contrast, *A. mellifera* exhibited optimal foraging activity at higher temperatures, between 21–24 °C (Figure 61).







Figure 62: Predicted insect activity at different temperatures and times of day modelled from observations of *Eristalis tenax, Apis mellifera and Calliphora stygia* foraging behaviour within TAS cage trials. The model was created using PROC MIXED in SAS/STAT version 9.4.

#### Seed Yields in Large Cage Trials - Tasmania

#### seedPurity, TAS

Average clean dry carrot seed yields from large cages (10m x 10m) pollinated exclusively by *E. tenax* were 92% (2020) and 135% (2021) of those from cages pollinated by *A. mellifera*. Yields from cages pollinated by *C. stygia* in 2020 were approximately 40% of those achieved by *E. tenax* or *A. mellifera* (Figure 63). Conditions during flowering in 2021 were cooler than average (0.15°C cooler than average for the whole of Tasmania) which likely favoured *E tenax* over *A. mellifera*, resulting in higher yields from cages stocked with *E. tenax*. This suggests that it is worthwhile attempting to mitigate climatic risks during flowering by stocking a mix of pollinators that perform optimally under different environmental conditions. The non-significant reduction in yield in the combined stocking treatment relative to *E. tenax* alone most likely reflects overstocking within the cage, leading to the over-utilisation of pollen for food. We observed no evidence of antagonistic behaviours between species that would contribute to this yield outcome.



Figure 63: Mean clean dry carrot seed yields from experiments comparing performance of single pollinator species (2020) and individual and combined stockings of *E. tenax* and *A. mellifera* (2021). Stocking rates per cage were *A. mellifera* - ca. 3000 bees; fly species – 600 flies and combination cages ca. 3000 bees and 600 flies. Error bars indicate standard errors (n=3), letters above columns denote statistically significant differences between treatments.

In summary, the European honeybee, *A. mellifera*, carried more pollen and foraged across carrot umbels at a faster rate than either the hoverfly, *E. tenax*, or the blowfly, *C. stygia*. While *A. mellifera* is undoubtedly the most important pollinator in hybrid carrot seed crops, its tendency to discriminate between parent lines and minimise the distance of crossing moves can impact its efficacy. In contrast, *E. tenax* and *C. stygia* were less likely to actively discriminate between parent lines and crossed more readily between pollinator and MS beds and over greater distances within the crop, all of which are desirable traits for hybrid seed production. In TAS, *E. tenax* and *A. mellifera* pollinated similar proportions of stigmas in single visits to umbellets but *E. tenax* deposited more pollen on each, while in NSW, the hoverfly *Eristalinus punctulatus* pollinated a greater proportion of stigmas in single visits and deposited more pollen per stigma than *A. mellifera*. In TAS, hybrid carrot seed yields from caged plots pollinated exclusively by *E. tenax* were comparable to or higher than those from plots pollinated exclusively by *A. mellifera*, but in the single year that it was tested in cages, *C. stygia* performed poorly. *E. tenax* most actively foraged carrot umbels of a morning and under cooler temperatures, whereas *A. mellifera* was most active in the afternoons and under warmer conditions. These differences highlight the potential complementarity of *E. tenax* and *A. mellifera* as managed vegetable seed crop pollinators.

## Development of E. tenax as a Pollinator for Commercial Tunnel-Based Seed Production in Tasmania

#### seedPurity

Based on successful efficiency and rearing studies, SPS (TAS) commenced commercial production of hybrid carrot and brassica vegetable stock seed in poly tunnels using mass reared *E. tenax* as the sole pollinator (Figure 64). Project partner SP facilitated the development of this enterprise by supporting mass fly rearing at SPS, designing deployment strategies, and co-supervising a master's research project at the University of Tasmania that investigated optimisation of pollination in this system. In 2025, yield outcomes for the program were 103% of target. This represented a significant improvement on previous attempts using honeybees in tunnel production, with the additional benefit of improved occupational health and safety conditions for employees working within the tunnels. There is significant interest from the local and international vegetable seed industry to expand tunnel-based vegetable seed production in Australia.



Figure 64: Images from commercial tunnel-based stock seed production of hybrid carrot and brassica (Komatsuna) at SPS TAS, utilising E. tenax for pollination. Photo credits: Raylea Rowbottom and Cameron Spurr.

### Dispersal and Retention of E. tenax in Open Field Releases

#### seedPurity, TAS

We mapped foraging *E. tenax* densities in open field releases in hybrid carrot seed crops under a range of different stocking scenarios (pupae deployments ranging from 5,000 to 32,500/ha) and observed foraging fly densities of up to 2.38 flies/m<sup>2</sup> (23,800 flies/ha) (Figure 65). Following the deployment of pupae, we observed progressive dispersal of hatched flies away from release points until a relatively uniform distribution throughout the crop was achieved after approximately 3 weeks (Figure 66). At sites with little or no pesticide usage during flowering, foraging *E. tenax* numbers ranging from 10% to 40.9% of flies hatched at the site were observed in crops up to 3 weeks after deployment. These surveys did not account for flies that were perched within the canopy (not actively foraging and often not easily located) or foraging weedy species at the site and so underestimate fly retention. Although we were unable to differentiate between mass reared and wild *E. tenax*,

baseline surveys conducted within each crop prior to the releases, levels of *E. tenax* activity in nearby unstocked crops and dispersal patterns observed from the release points all strongly indicated that most flies observed in post deployment surveys originated from the mass releases. These results compare favourably to reports that 4 to 15% of worker honeybees from hives stocked in carrot seed crops are typically observed foraging the target crop during visual surveys (Spurr, 2003). Site surveys following mass release also highlighted the importance of topography, amount and quality of floral resource, and agronomic practice on fly distribution and retention.

By far the greatest challenge to effective deployment of *E. tenax* in open vegetable seed crops, especially hybrid carrot seed crops, is the need for intensive management (at times) of migratory seed-feeding insect pests such as Rutherglen Bug (*Nysius vinitor*) with insecticides during the flowering period. While the industry's pesticide programs avoid adverse effects on honeybees through use of short residual synthetic pyrethroids applied only at night after the bees return to their hives, hoverflies are highly susceptible to this strategy because they remain within the crop. Staggering fly hatching within the crop by deploying different age classes of pupae or conducting multiple releases has proven effective in mitigating the adverse effects of low intensity spray regimes, but high intensity spraying remains problematic (Figure 67). The industry is focused on developing IPPM approaches to pest-management in vegetable seed crops that will help address this issue.



Figure 65: Distribution of *Eristalis tenax* in three hybrid carrot seed crops with varying fly stocking rates. From left to right: A) low (5,000 pupae/ha); B) medium (10,000 pupae/ha) and; C) high (32,500 pupae/ha) stocking rates. Each map is a single point in time, approximately 14 days after flies were released into the crop. Crop sizes were A) – 5.3ha, B) 5.2ha and C) 8.8ha.



Figure 66: Progressive dispersal of *Eristalis tenax* over time in a 5.2ha hybrid carrot seed crop in Tasmania in 2021-22, representing the time sequence of fly dispersal during the flowering period. *E. tenax* stocking rate was 10,000 flies/ha. Flies were released into the crop on 22nd December 2021.



Figure 67: Distribution heatmaps of *E. tenax* over time in three hybrid carrot seed crops in TAS with different intensities of pesticide spraying. From top to bottom; A) high spray intensity crop (stocked at 100,000 flies/ha over 3 releases, B) medium spray intensity crop (stocked once with 7,500 flies/ha), C) low spray intensity crop (stocked once with 10,000 flies/ha). (S) represents timing of pesticide spray events to control migratory flights of Rutherglen Bug (*Nysius vinitor*) into the crop. Fly release dates were: A) 29<sup>th</sup> December 2023 and 16<sup>th</sup> January 2024 (due to insecticide spray); B) 4<sup>th</sup> January, 2022; C) 22<sup>nd</sup> December, 2021.

#### Yield Benefits of Complementary Stocking of *E. tenax* with Honeybees in Hybrid Brassica Seed Crops.

#### seedPurity, TAS

We observed yield improvements (seed weight per flower) in 3 of the 4 commercial *Brassica oleracea* seed crops stocked with *E. tenax* in complement to honeybees in 2023/24 and 2024/25 in TAS, when compared to stocking with honeybees alone (standard commercial rate of 8 hives/ha) (Figure 68). Yield metrics for the different stocking treatments were quantified as the % of yield potential under conditions of unlimiting pollination (quantified by flowers receiving supplemental hand pollination in addition to insect pollination). Observed gains in yield in the 3 crops ranged from 12.4% to 65.7% (mean = 31.4%) of yield potential. While pod setting rates (/flower) were similar in both stocking treatments, complementary stocking with *E. tenax* resulted in more seeds set per pod and thus higher average weights of seed produced

per flower (Figure 68). The flowering period of *Brassica oleracea* seed crops in Tasmania occurs during September and October and typically feature periods of cool temperatures and rainfall, conditions which adversely impact honeybee activity. Furthermore, CMS hybrid parent lines can be less attractive to honeybees, and we observed both discriminatory foraging within crops and loss of honeybees to off-target forage sources within these experiments. The combined effect of these phenomena is that pollination limitation is frequently a yield-limiting constraint in Tasmanian hybrid *Brassica oleracea* seed crops (Spurr and Lucas, unpublished research conducted for the Tasmanian vegetable seed industry 2020/23). The results of this study highlight the potential of using *E. tenax* as a managed pollinator to improve seed yields from these crops.



Figure 68: Seed yield in *Brassica oleracea* (cauliflower and broccoli) seed crops during 2023/24 and 2024/25 comparing the percent difference in pod set per flower, seed number per flower and seed weight (g) per flower for *A. mellifera* only and *A. mellifera* plus *E. tenax* pollination treatments. . Total columns show average responses across all 3 sites. Statistical significance is indicated for difference in seed weight per flower (g) for all sites combined. *E. tenax* were deployed for the first half of flowering period at Site 1, and for the second half of flowering at Sites 2 (over two seasons 2023/24 and 2024/25). Error bars indicate standard errors (n=12).

## **Habitat Augmentation**

#### University of New England, NSW

The portable habitat pools introduced into carrot crops in Griffith (NSW) successfully supported the reproduction of the targeted syrphid fly species (*Eristalis tenax* and *Eristalinus punctulatus*). The European drone fly (*E. tenax*) was reared from all 14 pools, while the native golden drone fly (*E. punctulatus*) was reared from one pool containing decaying carrot plants with water and farm soil and two pools containing decaying carrot plants with water.

More eggs were laid on decaying carrot stems and roots than on other plant parts, suggesting these were the preferred sites for egg-laying. Larvae at all three stages of development (first, second, and third instar) were found in both types of habitat pools, demonstrating that the larvae were able to feed and grow successfully. However, the longer the pools were left in the field, the fewer larvae of all instars were found (Figure 69); p < .001 for all). This suggests that the larvae may have either left the pools to pupate, were eaten by predators, or died due to competition for, or lack of, food. However, given that all of the pools still contained plenty of decaying carrot plant material when the eggs and larvae were counted, it is unlikely that the larvae died from lack of food or competition.



Figure 69: The number of eggs oviposited by female eristaline syrphid flies within the deployed pools based on habitat (carrot plants + water only and the soil + carrot plants + water) and the location where the eggs were laid. Letters indicate significant differences between locations (p < 0.05). Individual data points representing each habitat pool (n = 14 in total) are jittered onto the figure for clarity. Lower to upper box boundaries indicate the inter-quartile range (IQR). Whiskers are extended to the furthest data point within 1.5x the IQR from each box end.

The results obtained in this pilot study suggest that providing non-floral habitats in agroecosystems could be an effective strategy to enhance the reproduction of beneficial fly pollinators. There were no larval instars more abundant than others within the habitat pools, suggesting that flies were consistently laying eggs daily within the pools. As the substrates placed within the habitat pools (soil, discarded carrot plants, and water) are locally available, cheap, and the pools are small and portable, enabling placement and removal at key flowering times, we hypothesise that this approach may increase the natural population of flies that provide critical pollination services to crops in intensely managed agricultural systems. Further research is required to better understand how to scale up these habitats to meet pollination service needs, the length of time the portable habitat pools should be placed on farms, the water conditions that eristaline syrphid fly larvae require to survive, the potential predators of the fly larvae, and whether these pools attract non-target or potential pest species to crop fields.

# **Key Outcomes and Recommendations**

Many fly species contribute significantly to pollination of vegetable seed crops as wild pollinators. In a series of crop surveys and crop and cage-based pollinator efficiency evaluations, we established that the hoverfly, *Eristalis tenax*, is a widespread and highly efficient pollinator in Australian vegetable seed crops (carrot and brassica). In experiments conducted in NSW, the related species *Eristalinus punctulatus* also demonstrated attributes that indicate it may also be an effective pollinator in warmer climates. Both species have life histories that are suited to mass rearing.

Hybrid carrot seed yields achieved with *E. tenax* as a sole pollinator in cage trials were comparable to, or greater, than those achieved with honeybees. Comparison of pollinator activity and climate data show that optimal activity of *E. tenax* and *A. mellifera* occurred at different temperatures, with *E. tenax* performing best under cooler conditions and *A. mellifera* preferring warmer conditions. This was reflected in *E. tenax* outperforming honeybees in carrot pollination cage trials conducted in TAS in 2021 when conditions during flowering were cooler than average. This difference between the two species highlights the potential advantage of using *E. tenax* as a complementary managed pollinator with honeybees to mitigate the risks of variable weather conditions during flowering in crops grown in cool-temperate or temperate regions.

In TAS, we effectively deployed *E. tenax* into open vegetable seed crops ranging in size from 2 to 17.5ha and established relationships between timing and rate of stocking, fly density, and dispersal patterns within the crop that will inform commercial release protocols. We were able to conclusively demonstrate a yield benefit from stocking hybrid cauliflower and broccoli seed crops with *E. tenax* in complement to honeybees. This is an important outcome for the Australian vegetable seed industry given that yields from these crops (which flower in early to mid-spring) are known to have been limited historically by inadequate pollination. To the best of our knowledge, this is the first example of deployment of mass reared hoverflies for open field crop pollination at a semi-commercial scale.

In addition to its potential as an open field pollinator, SPS have adopted mass reared *E. tenax* for pollination tunnel-based vegetable seed crops in TAS. Monitoring of crops produced in this system in 2024-25 identified that *E. tenax* performed well as a pollinator, with average crop yields exceeding commercial production targets. An additional benefit has been improved OH&S conditions for staff working alongside the pollinators within enclosed tunnel systems compared to the previous system in which honeybees were used for crop pollination.

Finally, our work in NSW demonstrated that it is possible to use relatively simple approaches for habitat augmentation to support the life cycle needs and promote populations of wild Syrphid pollinators (*E. tenax* and *E. punctulatus*) in crops. Further work is needed to improve our understanding of the impact that this approach could have in commercial cropping systems.

# Outputs

# Table 9: Output summary

Output	Description	Detail
News Article	David Cook	ABC News Online, South-West WA.
	Bushflies and their ecological role in pollination	November 27, 2015.
		https://www.abc.net.au/news/2015-11-27/swarms-of-tiny- bush-flies-leave-south-west-wa-struggling/6981034
Industry meeting	Cameron Spurr, Raylea Rowbottom and Amy Lucas	Annual presentation to SPS production teams and stakeholders.
	Annual Hoverfly R&D presentations	2018 (Naracoorte), 2019 (Ballarat), 2020 (online) 2021 (online), 2022 (Toowoomba), 2023 (Griffith) and 2024 (Hobart).
Conference	James Cook and Jonathan Finch	Mango Growers Conference, Darwin, NT,
	Field presentation about stingless bee pollination studies, referencing the Fly Pollination Project.	May 2019.
Industry Meeting	David Cook	Avocado Meeting, Gingin, WA
	Rob Deyl and Elliot Howse attended	June 6, 2019.
	Project	Handouts detailing the project and trial work on avocados were taken by attendees.
Industry	David Cook	Talking Avocados
article/magazine	"Avocado Pollination Trial – 2018"	Winter Edition, 2019.
Conference	Romina Rader	Hort Connections, Melbourne
	Apple pollination presentation	June 24–26, 2019.
Conference	Romina Rader chaired a session on pollination and gave an overview of pollination under protected	PCA Conference, Gold Coast

	cropping.	July 7–10, 2019.
	Amy Lucas	
	Presentation: "The dronefly (Eristalis tenax) as an alternative pollinator for vegetable seed production"	
Radio	David Cook	ABC Radio National Country Hour,
	Mangos and fly pollination – Jonathan Finch & Flies and	August 8, 2019,
	pollination –	https://www.abc.net.au/news/rural/2019-08- 09/researchers-investigate-the-role-of-flies-in- pollination/11395604
<b>N</b> I <b>1 1</b>		
News article	Cameron Spurr and Hannah Allwright	Tasmanian Country
	"Pollination blow-ins ready to take-	October 11, 2019.
	off"	
Presentation	David Cook	Leschenault Catchment Council, South-West WA
	National Pollination Week talk on fly pollination	November 13, 2019
Presentation	David Cook	WA Insect Study Group
	"Fly diversity in avocado pollination and project findings"	December 11, 2019, Kings Park Board Offices, Perth, WA.
Radio	Cameron Spurr and Adelina	ABC Country Hour Tasmania
		December 16, 2019.
	Management of alternative crop pollinators in Tasmania	
Industry	Jonathan Finch	Mango Matters
al ticle/ magazine	"Fishing for Flies"	January 2020, Volume 38; 20-21.
		https://www.industry.mangoes.net.au/cmsb/media/mm- spring-2023-final-(web).pdf
News Article	James Cook	ABC Rural on-line news article
	"Alternative pollinators to help	February 2020
	tarmers as bee populations suffer in drought and bushfires"	https://www.abc.net.au/news/rural/2020-02-06/bee-
		populations-die-alternative-pollinators-to-help-
		<u>iamers/11920000</u>

Webinar	David Cook	Southwest Catchment Council
	What's the Buzz with Avocado	August 12, 2020.
		Attendees from WA, QLD, and TAS.
Radio	David Cook and Jacinta Foley	GWN 7 (Regional South-West WA)
	Interview at Jasper Farms, Busselton. Focused on fly pollination in horticulture.	August 20, 2020.
Industry	David Cook	WA Grower Magazine,
article/ magazine	"Researching Native Flies as Pollinators of Horticultural Crops"	Spring 2020, pp22-23.
Industry article/magazine	David Cook	Farm Weekly Magazine
	"Good progress achieved on native flies as pollinators"	October 5, 2020
Industry	David Cook	Australian Tree Crop
article/magazine	"Study assesses native flies as nollinators"	November 2020
		https://www.treecrop.com.au/news/study-assesses-native-
		<u>Tiles-</u> pollinators/#:~:text=Native%20flies%20are%20showing%20
		potential%20as%20a%20supplementary,by%20Hort%20Inn ovation%20with%20partners%20from%20across%20Austral
		ia.
Industry	David Cook	Australian Berry Journal
article/magazine	"Native flies as pollinators"	December 2020
		https://issuu.com/berriesaustralia/docs/abj_edition_5_sum
		<u>mer_2020</u>
		Queensland NRM
		September 16, 2021
Industry article/magazine	Jonathan Finch and Romina Rader	Knowable
article, magazine	"The essential fly"	February 2021
		https://knowablemagazine.org/article/food- environment/2021/the-essential-fly
Industry article/magazine	Jonathan Finch	Smithsonian Magazine
מי נוכול/ ווומצמצווופ	"How much do flies help with	March 8, 2021

	pollination	https://www.smithsonianmag.com/science-nature/now-
Workshop	Cameron Spurr and Amy Lucas	Utas/TIA Industry and stakeholder workshop
		NA 2024
	Presented hoverfly research	May 2021
Industry Meeting	David Cook	Avocado Meeting, Pemberton, WA
	"Managing Flies for Crop Pollination	June 22, 2021
	Project" - tailored to avocado	
	growers	Approximately 100 people attended, including Avocados
		Australia and DPIRD. A one-page handout was distributed.
		Avocado Meeting, Wanneroo, WA
		June 24, 2021
		Approximately 100 attendees, including Avocados Australia
		and DPIRD. A one-page handout was distributed.
Webinar	David Cook	Landcare group and avocado growers in Northern
		Queensland
	Native flies and pollination	September 15, 2021
		September 15, 2021
Workshop	David Cook	Pollination and Predation Workshop, Gulf Savannah NRM,
		QLD
	"Managing Flies for Crop	
	Pollination"	September 16, 2021
Industry	Ionathan Finch & James Cook	Mango Matters
article/magazine		
	"Year of the fly - Native hover flies	October 2021, Volume 45, pages 20-21.
	dominate the Darwin mango	
	flowering season"	https://www.industry.mangoes.net.au/cmsb/media/mm-
		spring-2021-final-(web).pdf
Conference	Raylea Rowbottom and Abby Davis	Costa Exchange distribution centre, Devonport, Tasmania
	, , , , , , , , , , , , , , , , , , , ,	<b>.</b>
	"Hoverflies in berries"	July, 2022
Field day	Cameron Spurr, Raylea Rowbottom	Reid Fruits, TPS Field Day
		May 25, 2022
	Fly pollination research	
Workshop	Raylea Rowbottom	TIA/UTAS Industry and stakeholder meeting
	Fly pollination in vegetable seeds	May 2022
	research	1111 2022

Industry	David Cook	The Macadamia Magazine
	"Flies emerge as Key Pollinator for	Autumn 2022
	horticultural crops"	South African Macadamia Association
		https://themacadamia.co.za/2022/09/01/flies-emerge-as-
		key-pollinator-for-horticultural-crops/
Conference	Raylea Rowbottom	Tasmanian Fruit Growers Conference
	"Managing Flies for Crop Pollination"	May 26-27, 2022
Webinar	Cameron Spurr	"Plan Bee" webinar and Q&A session.
	Vegetable seed pollination	June 21, 2022
Industry Meeting	David Cook	WA Avocado Research Update, Manjimup Country Club,
	R&D presentation	June 22, 2022
		Approximately 35 growers attended.
Industry	James Makinson and Gaurav Singh	Australian Tree Crop Magazine
article/ magazine	"The role of stingless bee	June/July 2022
	pollination in N1"	https://www.treecrop.com.au/news/role-stingless-bee- pollination-nt-mango-orchards/
Radio	David Cook	ABC Melbourne Radio
	Flies as backup pollinators for avocados, berries, and vegetables	July 13, 2022
News	David Cook	ABC Landline (TV)
	Using flies in avocado and mango orchards. Filmed across Queensland and Western Australia.	July 24, 2022

Industry	David Cook	WA Grower Magazine,
article/magazine	"Flies as Alternate Pollinators"	Spring 2022, pp22-23
Media release	UNE	University of New England
	"Could flies fill the pollination gap?" Media article about the research for the University of New England	November 10, 2022 https://www.une.edu.au/about-une/news-and- events/news/2022/11/could-flies-fill-the-pollination-gap
News Article	Abby Davis	The Northern Daily Leader
	"Native flies potential pollinators, varroa mite reduces honeybee numbers, native flies act like bees"	November 30, 2022 <u>https://www.northerndailyleader.com.au/story/7987088/p</u> <u>retty-fly-for-a-pollinator-guy-native-flies-could-help-</u> <u>pollinate-crops/</u>
News article	SPS and BSA	ABC news
	"Hairy hover flies help honeybees pollinate Australia's food crops" Discuss the success of the Eristalis fly project/success in carrot crops	January 19, 2023 https://www.abc.net.au/news/rural/2023-01-19/hairy- hover-flies-help-honey-bees-pollinate-food- crops/101858910
Media release	Romina Rader and Hort Innovation	Hort Innovation
	"Scientists bring flies to Varroa Mite Red Zone"	March 29, 2023 Hort Innovation   Scientists bring flies to Varroa Mite Red Zone
News article	Cameron Spurr	ABC Lismore news
	"Pollinator fly to be trialed on berry farms in varroa mite zones on NSW Mid North Coast"	March 30, 2023 https://www.abc.net.au/news/2023-03-30/pollinator-fly- trialled-varroa-mite-zone-coffs-coast/102158020
Conference	Raylea Rowbottom	Tasmanian Fruit Growers Conference
	"Efficacy of Flies as Berry Pollinators and Current Pollination Research"	June 15-16, 2023
Conference	Romina Rader "Future-proofing Berry Pollination Services"	XIII International Rubus and Ribes Symposium, Portland Oregon, USA July 16-21, 2023
Journal Article	Romina Rader	Acta Horticulturae 1388:
	"Future Proofing Pollination to	10.17660/ActaHortic.2024.1388.3

	insect-pollinated crop production.	
Industry article/magazine	David Cook "Alternative Pollinators for Avocados"	Talking Avocados Volume 34(3) 2023, pp57-59. <u>https://avocado.org.au/wp-</u> <u>content/uploads/2024/05/AVO6451_TalkingAvocados_Spri</u> <u>ng_23_FA_Web.pdf</u>
Radio	Jonathan Finch "Research on many insects which can be used for pollination"	Country Hour June 22, 2023 <u>https://www.abc.net.au/listen/programs/tas-country-hour/alternate-pollinators/102512056?utm_campaign=abc_listen&amp;utm_</u>
Conference	Abby Davis "Dipteran pollination in Australian commercial cropping systems"	International Congress of Dipterology, Reno, Nevada, US July 2023
Workshop	Cameron Spurr and Raylea Rowbottom Presentation about pollination research in vegetable seed crops	TIA/UTAS May 2023 Strong industry and stakeholder engagement.
News video	TPS, SPS and seedPurity "Fly species emerges as alternative pollinator to bees"	ABC news interview September 22, 2023 <u>https://www.abc.net.au/news/2023-09-22/fly-species-emerges-as-alternative-pollinator-to-bees/102890024</u>
News article	Raylea Rowbottom and TPS "As Australia's bees face varroa mite infestation, it's hoped hover flies can fill the role of crop pollinator"	ABC news article September 22, 2023 <u>https://www.abc.net.au/news/2023-09-22/hover-flies-</u> <u>step-up-as-pollinator-as-varroa-mite-hits-bees/102880708</u>

Industry Meeting	Cameron Spurr, Romina Rader, Raylea Rowbottom "Hoverflies as Effective Berry Pollinators" David Cook and Shoaib Tufail interviewed to run a news story item on flies as pollinators	Berry Growers Meeting, Coffs Harbour November 19, 2023 Delivered to berry growers and industry stakeholders, highlighting results from recent pollination trials and efficacy comparisons in NSW. Channel 9 news, Perth, WA December 2023.
TV Program	Cameron Spurr, Raylea Rowbottom, Romina Rader Interview at Costa NSW to showcase the research conducted in the area, May 22-23 2024	ABC Landline Interview, NSW July 7, 2024 (broadcast) <u>https://www.abc.net.au/news/rural/programs/landline/20</u> 24-07-07/flower-flies:-using-flies-as-pollinators/104068838
Industry article/magazine	DPIRD "Could flies replace bees"	Australian Science Illustrated issue #107 May 16, 2024 <u>https://www.mymagazines.com.au/backissue/science-illustrated/issue-107?srsltid=AfmBOoqhzvUSG22zE74TIZFrzLoBc2_344EDojT-ulVeG9yTc57Z2V1B</u>
Conference	Raylea Rowbottom Fly pollination research in cherries.	Cherry Growers Conference, Tasmania July 1, 2024
News Article	Cameron Spurr, Romina Rader, Raylea Rowbottom "Flower fly pollination could alleviate Australia's reliance on honeybees for crops as varroa mite impact plays out"	Landline article and video Released July 6 and 7, 2024 <u>https://www.abc.net.au/news/2024-07-07/flower-fly-pollination-honeybees-trial-berries-crops/104054082</u>
Media release	David Cook "Flower Flies" pollinating commercial fruit crops	ABC Landline (National TV) July 6, 2024
Industry article/magazine	David Cook "New Pollination Agents: Fly Success in Avocado Orchards"	WA Grower Magazine Summer 2024, vol.59, no.4, pp. 40-41 <u>https://wagrower.vegetableswa.com.au/collections/wa-grower-summer2024/pdf?page=41&amp;fullscreen=false</u>

Webinar	David Cook	Queensland avocado growers meeting
	"Pollinator Diversity in Avocados" covering species observed and recommendations for fly-based pollination in orchards	September 25, 2024
Conference	Raylea Rowbottom and Abby Davis	BerryQuest International
	Independent presentations across research on flies as pollinators	February 27, 2025

# **Outcomes**

This project was funded by the Hort Frontiers Pollination Fund, which has the target outcome of a resilient and prepared horticulture sector equipped with the necessary research and capacity to meet ongoing and changing pollination needs. This project is specifically aligned to strategy 3.1 in the Hort-Frontiers Pollination Strategy, "increase the capability and capacity of alternate pollinators" to achieve Outcome 3, "alternate pollination options developed for increased productivity". The project also received co-investment from the Tasmanian Government Agricultural Development Fund (ADF) which invests in industry-driven agricultural research to address emerging opportunities and issues likely to have a direct impact on Tasmanian agriculture. The ADF supports projects that will deliver broad benefits to Tasmania and the state's agricultural sector; have strong support and partnership from industry; and that demonstrate a clear strategy to deliver on-farm impacts.

## Table 10: Outcome Summary

Outcome	Alignment to fund outcome, strategy and KPI	Description	Evidence
INTERMEDIATE OUTCOMES 1) Knowledge of fly diversity and abundance in target crops	All intermediate and end of project outcomes listed align directly to Outcome 3 and Strategy 3.1 of the Hort Frontiers pollination strategy.	Surveys of fly diversity and abundance in Mango crops in QLD and NT. Results communicated by way of Journal publication (Finch <i>et al.,</i> 2023) – refer to appendix.	Feedback from growers, agronomists and industry extension officers attending vegetable seed and berry seminars over increased awareness of the role of fly pollinators in vegetable seed crops.
target crops The outcomes for Tasmanian trials and also consistent with the Tasmanian Stat Govt ADF fundir objectives.	The outcomes for Tasmanian trials are also consistent with the Tasmanian State Govt ADF funding objectives.	Surveys of fly diversity, abundance and pollen loading in vegetable seed crops in TAS, SA and NSW across 2 seasons. Data were communicated to industry stakeholders by way of 1) Presentations at annual production conferences for South Pacific Seeds, attended by ca. 20 production staff from around Australia (2019 and 2020) and; 2) Annual Meetings with Bejo Tasmania's production team.	Attendees at Coffs Harbour berry seminar attendees
		SPS and BSA constitute > 80% of the vegetable seed industry in Australia. Dipteran pollinator taxonomy workshop conducted at seedPurity for industry partners. Presentation of research data on alternative pollinators in Party crops to	Both fly literature reviews included in Appendix.

	growers agronomists and	
	industry officers at Coffs Harbour (31/5/2023).	Data on journal article downloads/citations.
	Surveys of fly diversity and abundance in Avocados in WA and QLD. Results communicated in industry articles and media presentations – refer to outputs table.	
	2 literature reviews on the role of flies as crop pollinators and life history and habitat requirement of Dipteran Pollinators (Cook et al., 2020; Davis et al., 2023) completed by the research team and published in the Journal <i>Insects</i> to raise awareness of fly pollinators amongst researchers.	
	Numerous media presentations – refer to outputs table.	
2) Knowledge of pollinator efficiency of key fly species in target crops	FieldbasedtrialsconductedwithindustrypartnersinAvocado,cherriesandvegetableseedcropsbetween2019and2024.Glasshouse-basedpollinatorefficiencystudiesinstrawberries.	Ongoing provision of trial sites and in-kind support from Costa (TAS and NSW), Reid Fruits, SPS, BSA and WA Avocado producers for fly pollinator trials, indicating support for and understanding of the importance of fly pollinators within production systems. Participation of BSA and SPS staff in efficiency trial data collection, fly
	Participation in annual pollination workshops conducted by TIA and seedPurity for pollination dependent industries and agricultural advisors in Tasmania (2021, 22 and 2023)	rearing and deployment. Image of SPS staff Hannah Allwright and Annick Witte participating in fly pollinator efficiency evaluations in carrot seed crops.
	Presentations to industry partners at annual SPS	



		R. Rowbottom (SP) presentation to BerryQuest covering fly pollinator efficiency in Berries and Cherries included in Appendix.
3) Developmental rates and rearing protocols for key fly pollinator species determined.	Lab based experiments conducted at SP and DPIRD in research insectaries. Results for <i>E. tenax</i> communicated in fly rearing guide prepared for industry partners (see fly rearing chapter in report). Frequent informal meetings between researchers and commercialization partners.	Construction of research scale fly rearing facilities for <i>E. tenax</i> at BSA and SPS. Production of flies for pollination trials across multiple industries in this project by BSA and SPS. Images of BSA (Top) and SPS (Bottom) staff participating in fly rearing trials
4) Viability of commercial fly rearing; Risks associated with mass rearing	Potential risks associated with mass deployment of flies identified. A formal independent risk assessment and mitigation framework is being developed and will be implemented before large scale commercial releases are undertaken.	
END OF PROJECT OUTCOMES 5) Fly species available for use as a pollination service to the	Commericalisation of fly rearing by industry partners and widespread adoption of fly pollinators delayed by the need for thorough independent risk assessment around mass release of flies before large	Commercial adoption of <i>E. tenax</i> as sole pollinator for tunnel production of vegetable seed in Tasmania. Industry data show 2025 tunnel production using <i>E. tenax</i> achieved 103% of the commercial yield target. Pilot scale commercial open field

scale open field trials are	releases occurring in TAS carrot seed
conducted at mainland sites. Further work to develop the market and refine efficiency of fly rearing required before large scale commercial mass rearing commences Smaller scale commercial tunnel production and semi-commercial field releases occurring in the Tasmanian vegetable seed industry using flies reared by SPS and BSA	brassica seed and cherry crops.
<ul> <li>PHD and Honours/ Masters candidate completions in this project:</li> <li>Abby Davis – PHD (UNE) 2024</li> <li>Hui Jing Chong – Honours (SP-UTas) 2020</li> <li>Mark vanSchilt (HAS Internship seedPurity) 2021</li> <li>Annick Witte – Honours (UTas) 2021</li> <li>Bharat Dinakaran – Masters (SP-UTas) 2024</li> <li>Shilpa Kiorala – Masters (SP-UTas) 2025</li> </ul>	
	<ul> <li>conducted at mainland sites. Further work to develop the market and refine efficiency of fly rearing required before large scale commercial mass rearing commences</li> <li>Smaller scale commercial tunnel production and semi-commercial field releases occurring in the Tasmanian vegetable seed industry using flies reared by SPS and BSA</li> <li>PHD and Honours/ Masters candidate completions in this project:</li> <li>Abby Davis – PHD (UNE) 2024</li> <li>Hui Jing Chong – Honours (SP-UTas) 2020</li> <li>Mark vanSchilt (HAS Internship seedPurity) 2021</li> <li>Annick Witte – Honours (UTas) 2021</li> <li>Bharat Dinakaran – Masters (SP-UTas) 2024</li> <li>Shilpa Kiorala – Masters (SP-UTas) 2025</li> </ul>

# Monitoring and evaluation

# **Table 11: Key Evaluation Questions**

Key Evaluation Question	Project performance	Continuous improvement opportunities
1. To what extent has the project achieved its expected outcomes?	This project surveyed natural fly occurrence across a broad spectrum of crops and production regions across Australia. We identified and tested 5 fly species in different field settings across Australia. We conclude that 4 fly species have the best potential as managed pollinators in avocado, berry and vegetable seed crops. These are <i>Eristalis tenax,</i> <i>Calliphora dubia, Calliphora vicina</i> and <i>Eristalinus punctulatus.</i> A range of rearing substrates were tested and temperature dependent development times were determined for Eristalinae and Calliphoridae flies. Health risks associated with mass rearing of flies were identified for mitigation in future R & D activities. Preliminary data on cost-benefit analysis for fly stocking in vegetable seed and cherry crops were collected. Further data is required to complete cost-benefit analysis for other key crops pending completion of risk assessments relating to open release of flies and yield and retention studies. Further data is required on cost of rearing following refinement	While the results of this project have identified candidate fly species that show promise for development as managed pollinators, there is further research work that remains to be done to realise this opportunity to improve pollination security for Australian production systems.
	and scaling of rearing protocols and collection of additional fly performance and yield data from open releases.	
2. How relevant was the project to the needs of intended beneficiaries?	The project is directly relevant to the needs of beneficiaries as we have identified the most efficient flies that can also be mass reared. This means several flies could be progressed as alternative managed taxa to potentially be used alongside honeybees under specified conditions.	Opportunities to increase relevance to industry include the development of commercial rearing and mass release protocols, associated risk assessments and greater efficiencies in rearing technologies.
3. How well have intended beneficiaries been engaged in the project?	There has been continual engagement with the avocado, berry and seed production industries during this project along with the project reference group.	Increased engagement may result from on-farm site visits and open days for all growers.
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4. To what extent were engagement processes appropriate to the target audience/s of the project?	We used diverse communication channels to increase industry engagement. Regional forums, industry conferences, magazine articles and workshops have all been used to engage with industry (the target audience for this project)	
5. What efforts did the project make to improve efficiency?	Teams of researchers across different regions on target crops ensured field work efficiencies. Team meetings on- line as opposed to all travelling to one location for annual meetings;	

## **Recommendations**

This project is the first major step towards developing managed fly pollinators as a strategy to improve pollination outcomes and reduce risks associated with over-reliance on honeybees in pollination-dependent horticultural industries in Australia. The research findings reinforce the significant contribution that flies make to crop pollination across a range of model crops and highlight the importance of efforts to conserve and manage fly pollinators in agricultural landscapes. Our results identify candidate fly species that show promise for development as managed pollinators, whilst also highlighting the work that remains to be done to realise this opportunity to improve pollination security for Australian producers. Here, we provide conclusions and recommendations for industry participants wishing to adopt managed fly pollinators or enhance the contribution of wild fly pollinators in their production systems and offer recommendations on further research and development activities needed to support commercial development of fly pollination in Australia.

## Avocado

#### **DPIRD and UNE**

- Avocado growers should consider complementing the use of honeybees for pollination needs with the addition of fly species identified in this project.
- Cooler flowering seasons (<30 degrees) will favour the use of either the drone fly *Eristalis tenax* or blowfly *Calliphora vicina*, whereas warmer seasons will favour the blowfly *Calliphora dubia*.
- We recommend future research to better understand risks, challenges and opportunities for fly mass release. This includes trialling movement patterns and dispersal abilities of flies to better understand retention within the target crop. The use of smaller adult flies (either *Calliphora vicina* or *Calliphora dubia*) may be manipulated by modifying their diet.
- Snout flies (family Rhiniidae) were frequently observed in Queensland orchards and were effective at
  depositing pollen, suggesting they could have potential as managed pollinators. More research is needed to
  understand the biology of Rhiniidae flies—particularly what they feed on and where their larvae develop— to
  support wild populations in cropping systems, and to assess their suitability for mass rearing.

## **Blackberries, Blueberries and Raspberries**

### DPIRD, UNE and SP

Our research shows that certain fly species, particularly *Eristalis tenax* and blowflies (*Calliphora albifrontalis* and *C. dubia*) can be highly effective pollinators of berry crops such as blackberry, raspberry, and blueberry, providing growers with an alternative or backup pollination strategy to support consistent production outcomes.

- In springtime blackberry pollination trials in enclosed tunnels in Tasmania, *E. tenax* stocked at rates ranging from 8 flies/m<sup>2</sup> at early flower to 30 flies/m<sup>2</sup> at peak bloom provided excellent fruit set and quality, outperforming open-field honeybee pollination.
- Although bees deposited more pollen per visit in blackberry, flies—such as the blow fly *C. stygia*—still contributed significantly to pollination. When flies were allowed multiple visits to flowers, they produced much larger fruit, showing that repeated fly activity can improve fruit size. In raspberry, both flies and bees produced similar fruit weights after a single visit, and repeated fly visits led to heavier fruit, reinforcing the importance of high pollinator activity during flowering in these crops.
- In glasshouse trials with southern highbush blueberry (*V. corymbosum* hybrid, variety 8–17), berry yield (number and size) was increased by the introduction of fly pollinators during flowering. *Calliphora dubia* outperformed *Calliphora albifrontalis* across a five-month period.

## **Sweet Cherries**

### seedPurity

- The hoverfly *Eristalis tenax* appears to be a highly effective pollinator of sweet cherry crops in Tasmania and a good option for development as an alternative managed pollinator.
- As the sole pollinator, fly stocking rates of 75,000 flies/ha appear optimal. In a preliminary trial release under commercial orchard conditions, complementary stocking of 60,000 hoverflies and 3 honeybee hives/ha resulted in fruit set that was superior to stocking with honeybees alone (3.5 hives/ha).
- Further work is required to improve timelines of fly emergence (hatching) in cool, early season conditions encountered in Tasmanian cherry orchards, and to validate the results of open field stocking trials across sites and seasons.

### Mangoes

### **UNE and WSU**

A wide variety of insects visit mango flowers in northern Australia, with wild insects, particularly native stingless bees and flies, making up most of the activity.

- Snout flies (family Rhiniidae) were frequently observed in Queensland orchards and were effective at depositing pollen, suggesting they are important mango pollinators. More research is needed to understand the biology of Rhiniidae flies—particularly what they feed on and where their larvae develop— to support wild populations in cropping systems, and to assess their suitability for mass rearing.
- Stink stations can be used to attract blowflies to mango trees but, in trials conducted in the Northern Territory, this approach did not lead to higher fruit set. This was probably because other insects present in large numbers in the trial, such as the native hover fly (*Mesembrius bengalensis*) and stingless bees, already provided strong pollination services to crops.

### **Glasshouse Strawberries**

### WSU

- Flies offer a complementary or alternative glasshouse pollination option to managed bees. Flies can be brought in and applied to the crop as a one-off "treatment", whereas hive bees are valuable livestock that also need to be managed (or rented with a management contract).
- The brown blowfly, *Calliphora stygia*, and the hoverfly, *Eristalis tenax*, are very good pollinators of glasshouse strawberries, capable of producing high quality strawberry crops with modest stocking rates in closed glasshouses. Overall, our results suggest that *E. tenax* is a more effective pollinator (per fly) due to its stronger attraction to flowers and longer average visit time. However, the less flower-centric *C. stygia* also performed well in a closed glasshouse environment. The difference between the two species might be greater in open protected cropping structures with the blow flies more likely to abandon the crop.
- A stocking ratio of one hoverfly (*E. tenax*) per four plants yielded excellent fruit quality outcomes in our final experiment. A similar ratio for blowflies (*C. stygia*) did not give quite such impressive results but was still very good. We would therefore recommend perhaps doubling this stocking rate for blowflies.

## **Vegetable Seed**

## SP and UNE

Many different fly species contribute to pollination of vegetable seed crops in Australia. We identified that the hoverfly *Eristalis tenax* in particular is a widespread and highly effective pollinator of carrot and vegetable brassica seed crops in both open field and tunnel settings.

- E. tenax is an ideal complementary pollinator to honeybees in these crops because it forages optimally over a different (lower) temperature range, has foraging behaviours well suited to cross pollination of hybrid seed parent lines and, under favourable conditions and can be effectively deployed and retained in tunnel and open field crops throughout the 4 5-week flowering period typical of these crops.
- Stocking open field hybrid cauliflower and broccoli crops with *E. tenax* in complement to honeybees can improve seed yields. In 3 out of 4 field trials conducted in Tasmania over 2 seasons, complementary stocking of honeybees and *E. tenax* realised 31.4% more of the crop yield potential than stocking with honeybees alone.
- While *E. tenax* will effectively disperse and remain in carrot seed crops, it and other fly species are vulnerable to pesticide programs used to control invasive seed-feeding pests such as Rutherglen Bug. Further work to adapt approaches to fly deployment and develop IPPM strategies for carrot seed crops is needed before the full potential of *E. tenax* and other wild flies as complementary managed pollinators can be realised in these crops.
- The hoverfly *Eristalinus punctulatus* occupies a similar ecological niche to *E. tenax* in warmer and drier (northern and inland) climates. Preliminary work conducted in NSW during this project suggests that *E. punctulatus* has potential for development as a managed pollinator of carrot seed crops in these environments.
- Simple interventions, such as the provision of habitat for oviposition and larval development by placing decomposing plant material in pools of water, can be used to promote breeding of wild hoverflies (*E. tenax and E. punctulatus*) in vegetable seed crops.

## **Commercial Availability of Flies for Crop Pollination.**

- At the time of writing, the brown blowfly, *C. stygia*, can be purchased as pupae in limited volumes from fishing bait suppliers in Australia.
- *E. tenax* is not yet available "off the shelf", but scalable rearing protocols were developed in this project and are the focus of ongoing research.
- Several industry partners have expressed interest in developing commercial fly rearing capacity for promising Eristaline and Calliphorid pollinators and have committed funding and in-kind support to a new project to develop commercial scale rearing systems.

## **Recommendations for Future Research, Development and Extension Activities**

We identified 6 priority areas for RD&E activities to support progress towards the widespread commercial use of fly pollinators for Australian horticulture. These encompass quantification of yield and quality benefits from adoption of fly pollinators, deployment protocols for fly pollinators in commercial cropping systems, efficient mass rearing techniques, informing the business case for fly pollinators, and issues around risk management and market perception.

# 1. Assessing and Managing Potential Legal and Environmental Risks Associated with Mass Fly Releases for Pollination

While this project has intentionally focused on fly species that naturally distributed within the study areas and pose minimal Hort Innovation

health and safety risks to humans and livestock, it is important that any possible environmental or legal concerns surrounding mass fly releases for pollination are identified, evaluated and, if necessary, addressed before large scale commercial releases occur. This includes the need to irradiate released flies (rendering them infertile) and how that may affect their foraging behaviour, dispersal and longevity.

## 2. Deployment Strategies for Eristaline and Calliphorid Flies as Managed Pollinators

Progress towards developing protocols for the deployment of fly pollinators into commercial cropping systems has varied across the model crops studied in this project. Further work is required to address crop-specific knowledge gaps in:

- When, where and how to deploy flies, and the stocking rates required to achieve effective crop coverage during flowering;
- Effects of crop husbandry practices (for example pest management) on fly retention and survival;
- Potential management tools/habitat augmentation strategies to support and retain fly pollinators in crops; and
- Quantification of yield and quality gains that can be achieved by deploying fly pollinators into commercial production systems.

# **3.** Demonstrating the Potential of Managed Fly Pollinators in Other Pollination-Dependent Horticultural Crops

Other horticultural industries besides those involved in this project would also likely benefit from access to managed fly pollinators or enhanced wild fly pollinator populations, whether as a risk mitigation strategy or as a means of improving pollination in cropping systems less suited to honeybees (for example, protected cropping) or a combination of both. Demonstrating the potential of fly pollinators in crops with large volume demand for pollinators and/or demand during different times of the year is necessary to foster industry interest in fly pollinators and drive investment in large scale fly rearing for crop pollination.

## 4. Supporting Development of Commercial Rearing Capacity for Eristaline and Calliphoird Flies

During this project, we developed facilities and protocols for small-scale rearing capacity of target Eristaline and Calliphorid species (1-2 million flies/month). This was underpinned by research on rearing substrates, modelling of environmental effects on fly development and fitness, and identification of management options to synchronise fly production with demand for pollinators. Further work is required to optimise fly diets for rearing pollinators which are based on consistent, low-cost substrates, and to test scalable technologies for efficient fly rearing in commercial volumes.

## 5. Market Perception of Fly Pollinators

Understanding and addressing any negative or inaccurate market perceptions will be important for the adoption of fly pollinators into some cropping systems, for example soft fruits.

## 6. Identification and Development of Other Alternative Crop Pollinators

Research outcomes from this project highlight the importance of diversifying pollinator options to ensure reliable pollination in the face of variable climatic conditions and new crop production systems. While the immediate priority stemming from this project is to develop the candidate species already identified, it is also important to recognise that there is still much to be done in terms of understanding other wild pollinator species and exploring the potential roles they could fill for crop pollination in Australia.

At the time of writing, Hort Innovation is working with researchers and industry stakeholders to finalise a project addressing key research needs to progress fly pollination to a commercial option for Australian Horticulture.

## **Refereed Scientific Publications**

**Cook, DF, Deyl, RA**, Mickan, BS and **Howse, ET** (2020) Yield of southern highbush blueberry (*Vaccinium corymbosum*) using the fly *Calliphora albifrontalis*, (Diptera: Calliphoridae) as a pollinator. *Austral Entomology 59*, 345-352. https://doi.org/10.1111/aen.12455.

**Cook, DF, SC Voss, JTD Finch, R Rader, JM Cook,** and **CJ Spurr** (2020) Reviewing the role of flies as crop pollinators of Australian horticultural crops. *Insects*, *11*, 341. doi:10.3390 /insects11060341

**Cook, DF, SC Voss**, RA Deyl, ET Howse, J Foley, B Norrish, N Delroy, **SL Shivananjappa** and **MS Tufail** (2023) Blow flies (Diptera: Calliphoridae) ability to pollinate Hass avocado (*Persea americana*) trees within paired tree enclosures. *Journal of Applied Entomology* 147(8): 577-591, https://doi.org/10.1111/jen.13159

**Cook, DF, Tufail, MS**, Howse, ET and **Voss, SC** (2023) Manipulating larval rearing media to optimise mass production of the blow fly *Calliphora vicina* (Diptera: Calliphoridae). *Austral Entomology, 63*(1): 96-109, doi.org/10.1111/aen.12680.

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**Davis, AE**; LA Schmidt, **S Harrington**, **C Spurr**, and **R Rader** (2023) Provisioning Australian seed carrot agroecosystems with non-floral habitat provides oviposition sites for crop-pollinating Diptera. *Insects*, *14*, 439. <u>https://doi.org/10.3390/insects14050439</u>.

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# Intellectual property

No	Name of IP, if any	Type of Output	Usage	Nature of IP	Conditions of use	Confidentiality	Risks identified in relation to the IP
1	Protocols for Rearing and Deployment of Eristalinae and Calliphoridae Flies	Other	Commercialisation	Confidential Information	Non-Exclusive Licence	Confidential	Key industry stakeholders and potential commercial partners have invested 5- years of funds to the project and wish for this information to remain confidential for commercialisation.

Protocols have been provided to commercialisation partners for fly rearing within this project who invested funding to support fly rearing research. It is necessary for this information to remain confidential to these partners in order for them to justify the initial investment in large scale rearing required to supply flies for pollination to Australian horticulture. This is being managed on a first rights of refusal basis with HI having the option to bring in other commercialisation partners in the future based on uptake by the commercialisation partners and scale of demand.

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### Avocado

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#### **Hort Innovation**

# **Appendices**

## **Review Papers**

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Finch J, Gilpin A, & Cook J. 2023. Fishing for Flies: Testing the Efficacy of "Stink Stations" for Promoting Blow Flies as Pollinators in Mango Orchards. Journal of Pollination Ecology 33: 79-100. <u>https://doi.org/10.26786/1920-7603(2023)711</u>