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Rural R&D for Profit Program

Final Report

Novel technologies and practices for the optimisation of pollination within protected cropping environments

By Hort Innovation
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Disclaimers

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The project was led by Hort Innovation with research partners including the University of Adelaide, Plant and Food Research Australia, NSW Department of Primary Industries, University of New England and the University of Tasmania.

The success of this project was only possible thanks the many external collaborators, including willing growers who allowed the research to be conducted on their farms and the trust of beekeepers who permitted access to their hives.

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Plain English summary

As Australian horticultural industries increasingly turn to protected cropping to mitigate the impacts of extreme weather, pests, and diseases, understanding their impact on honey bees and pollination is vital. This collaborative research project set out to enhance the quality and yield of fruit and vegetables in protected cropping environments.

The overarching goals of the program included enhancing insect pollinator efficacy by improving honey bee performance under covers, manipulating plant floral and reproductive traits for improved fruit production and quality, optimising the placement of pollen donor plants (pollinisers) to sustain high fruit yield and quality, and advancing innovative methods of mechanical pollination to achieve artificial pollination in the absence of insects.

Funded by Hort Innovation, the project commenced in 2019 and brought together teams from the University of Adelaide, Plant & Food Research, the NSW Department of Primary Industries, the University of New England, and the University of Tasmania. The comprehensive findings presented here provide valuable insights into optimising pollination practices and improving overall crop productivity in protected cropping environments.

> Bee performance and vigour was improved by allowing bees to forage outside and by raising covers high above the crop.

Research from apple orchards in the Adelaide Hills has revealed that crop covers have significant effects on bee orientation, activity, and foraging returns. Bees' performance and vigour was improved when covers were held high above the crop. Allowing bees to forage outside, either through open sides or penetrable covers, enhanced hive foraging returns and facilitated hive growth. However, allowing bees to access other floral resources is likely to reduce pollination services in the protected crop, potentially requiring more hives to achieve adequate fruit set.

The combined evidence from molecular, behavioural, and fruit set data highlighted the importance of orchard design under netting. Ideal orchard configurations include limiting row lengths and including simultaneously flowering pollinisers in every seventh row. Unmanaged pollinators (mostly native bees and hoverflies) in orchards were also examined, and shown to benefit when their preferred nesting locations, namely in open soil in headlands and under trees, were managed. Native bees visiting apple flowers proved to be generalists, benefiting from the presence of flowering weeds in the orchard.

The impact of hail netting on pollination was explored in depth in apple orchards across New Zealand and southeast Queensland. When comparing partially covered and uncovered apple blocks, data showed that flowers in the uncovered block produced fruit with significantly more seeds than the covered block. Supplementing bee pollination with hand-applied pollen increased seed numbers and reduced empty carpels in both environments.
While the literature suggests an optimal increase in fruit size with seed numbers up to 5-6 seeds per apple, no overall increase in fruit weight with seed number was found in the trial.

> Placing hives in the centre of blocks, and providing gaps in netting, increased the distance bees travelled while mitigating bee losses.

The New Zealand study also showed that when hives were positioned at the end of a covered block, the number of bees on apple flowers decreased with increasing distance from the hive. Placing hives in the middle of the block under cover resulted in bees traveling farther from their colony, a similar result was observed for hives under gaps in the roof of the netting. The study also showed that creating temporary gaps in the roof can mitigate bee losses in fully covered blocks, with colonies in partially covered blocks performing well, regardless of their proximity to gaps. No evidence was found to suggest that gaps in the roof had a detrimental effect on apple flower pollination or fruit quality. However, it is possible that a different outcome might have been observed if there were more floral resources available in the area surrounding the apple orchards, which was the case in the Adelaide Hills.

In northern NSW, research focused on advancing berry pollination techniques under protective crop covers on farms. This research resulted in the development of innovative strategies to augment pollination in berries within protected cropping environments.

The position of blueberry flowers was more important than floral morphology in attracting bees

Structured around two principal themes, the research examined the floral characteristics of blueberries and their attraction to pollinators, particularly honey bees. Evidence that variations in floral morphology among blueberry cultivars affect the attractiveness of flowers to honey bees was limited; instead, the positioning of cultivar flowers, and their access to honey bees, played a pivotal role. The study also delved into the effects of protected cropping systems on access to, and abundance of, floral nectar resources, incorporating diverse cover types such as plastic tunnels and bird netting. The impact of varying strengths of fertigation on nectar resources was also explored and highlighted that practices affecting the density of flowers within the crop are tools to manage the available nectar resources for pollinators.

Despite finding no significant differences in honey bee visits between contrasting covered crops, increases in berry weight and fruit sugars were noted and attributed to better crop nutrition and light conditions.

Although bees visit fewer flowers under polythene, pollination may still be optimised, especially if tunnel length is optimal

Additional studies in blueberries focused on the impact of cover types on bee visitation and colony health. Results of these studies suggest that using polythene covers instead of bird-netting can reduce bee visitation, but under certain conditions, this lower visitation rate may still lead to optimal yields, providing a potential advantage for polythene covers over bird-netting. Results also indicated that choosing an optimal tunnel length for blueberry cultivation can increase pollinator abundance and potentially boost yields.

Regarding colony health, colonies placed under protective netting on blueberry farms experienced decreased brood production, pollen, honey storage, and hive weight within the first four weeks. Although these metrics gradually increased, hives under netting showed smaller gains in brood production and pollen storage compared to those outside the covering. The findings emphasise the need for protected crop management strategies to consider both pollinator health and crop pollination requirements for maintaining bee health and pollination services in such systems.

Mechanical pollination shows some promise but application methods need further work to maximise yield

To explore alternative pollination techniques, studies in Tasmania developed, refined, and evaluated prototypes and protocols for mechanical pollination of model perennial and annual crops, namely sweet cherry, and seed onions. In particular, the technical feasibility of collecting commercial quantities of sweet cherry and onion pollen was demonstrated. For onions, a portable vacuum unit fitted with cyclone separators has proven to be the most efficient pollen collection system, enabling harvest rates of 20-25 g of purified pollen per hour per operator. For sweet cherry, the development of a mill prototype allowed for high throughput processing of fresh closed flowers while maintaining pollen quality. The harvested pollen was then applied to commercial crops as part of extensive field trials. Throughout these trials, yield outcomes varied, and the method of application requires further refinement to increase reliability.

This project lays the groundwork for new and integrated crop pollination methods to reduce the risks associated with the high dependence on honey bees and enhance pollination outcomes in protected cropping systems across a range of horticultural crops.

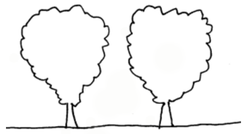
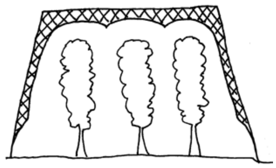
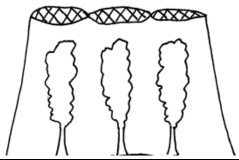
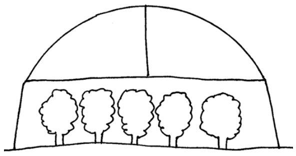
Working towards a common goal, the outcomes of this project are relevant to both the protected cropping grower community and apiarists. The insights gained from the extensive data collected over several seasons from a range of environments highlight the impact of protected covers and will help both growers and beekeepers the resources to make informed decisions about their practices and orchard designs. The ongoing extension of this work through best practice case studies, already commissioned, and other communication avenues, will be critical to ensure uptake.

Abbreviations and glossary

Abbreviations

AGMARDT	The Agricultural and Marketing Research and Development Trust (AGMARDT).
AHBIC	The Australian Honey Bee Industry Council
ALGA	Australian Lychee Growers' Association
APAL	Apple and Pears Limited
DPI	Department of Primary Industries
FOB	Frames of Bees
MPU	Mobile Polliniser Unit
NSW	New South Wales
NSWAA	The New South Wales Apiarists' Association Inc.
NZ	New Zealand
PCA	Protected Cropping Association
PFR	Plant & Food Research
Qld	Queensland
SA	South Australia
Tas	Tasmania
UNE	University of New England
UoA	University of Adelaide
UTas	University of Tasmania

Glossary of protected cropping styles and materials

Open orchard	No covers installed during the pollination period.	
Fully covered	Orchard is fully enclosed with netting covers both top and sides of crop.	
Partially covered	A roof or partial roof and some or all sides left open during the pollination period.	
Tunnel house	Open at ends, sides may be open or closed, solid structure covered with plastic or shade netting. Typical use – blueberries. Plastic film covers the roof with netted sides for blueberry.	
Hail net	A type of netting that is typically used to protect crops from hail damage. Hail netting is usually made from a sturdy, hail-resistant material, such as high-density polyethylene. The weave design and colour is variable. Shown here is a white quad hail netting.	
Bird net	Low (1-2m) and relatively floppy, lightweight material, mesh size 16mm x 16mm, commonly black or white.	
Rice net	A fine weave, flexible, apex/gable style net that allows hail to collect above and drop through into the interrow. Not permeable for bees, but can be opened above the interrow, as it is connected with bungees.	

1 Project rationale and objectives

Protected cropping systems are high value and production intensive. They offer many benefits, as crops are sheltered from the extremes of wind, sun and, in some cases, rain and hail. By protecting plants from environmental extremes, such systems help growers adapt to a hotter, drier and more changeable climate, supporting farm viability as well as maintaining food security. As a result, the area under protected cropping is expanding by at least four to six percent annually.

However, these benefits do not come without cost. The altered environmental conditions within protective environments do not necessarily suit the needs of the insects' growers rely on to pollinate their crops (e.g. Kendall et al. 2021; Evans et al. 2019; Hall et al. 2020). Inadequate pollination reduces both yield and quality. Growers and apiarists are therefore grappling with the challenges of balancing the needs of crop and pollinator.

This collaborative project is focused on practical outcomes focused research, optimising the efficiency of pollination services in order to maintain and improve productivity within protected cropping systems. This will allow beekeepers to better prepare hives, companies to improve their technology, and growers to improve pollination practices within protected cropping environments

This research project, managed by Hort Innovation, involves a collaboration of five key organisations:

- NSW Department of Primary Industries
- Plant & Food Research
- The University of Adelaide
- University of Tasmania
- The University of New England

Several participating horticultural businesses were also integral to the project, including ALGA, Seed Purity, APAL, Hansen Orchards, Costa, Reid Fruits, OzGroup, Mountain Blue Farms, and South Pacific Seeds.

The overarching aims of the program were as follows:

Increase insect pollinator efficacy and pollination by improving the performance of honey bees under covers.

- Pollination in protected cropping may be compromised by a range of factors specific to conditions under covers that affect honey bee (*Apis mellifera*) pollinator performance.
- Understanding the effect of these conditions can lead to better advice regarding the maintenance of hive health and performance.
- Advice could include hive size and placement, as well as the choice and manipulation of the covers themselves to maximise yield and fruit quality.

Manipulate plant floral and reproductive traits to improve fruit production and quality.

- Quantify the impact of protected cropping and flower physiology on bee behaviour
- Provide insight into how environmental conditions can be optimised to maximise attractivity of flowers and promote pollination.
- Improve understanding of floral biology, encompassing nectar secretion patterns, nectar sugar concentration, and pollen production in flowers of various *Vaccinium* (blueberry) and *Rubus* (raspberry and blackberry) varieties.
- Optimise the placement of pollen donor plants (pollinisers) to enable growers to manipulate crop/orchard configuration to sustain high fruit yield and quality.

Study bee foraging behaviour in netted apples in order to optimise the configurations of pollinisers.

- Examine the potential of low-cost robotic mobile polliniser units, to facilitate the improved delivery of pollen.

Improve and advance innovative methods of mechanical pollination.

- Mechanical pollination (MP) is an emerging technique that offers several key advantages to horticultural producers, including a reduced reliance on insect pollinators and the ability to overcome the effects of asynchronous flowering.
    - Pollen quality can easily be assessed to ensure maximum viability
    - Storage and transport of pollen reduces the need for on-site polliniser cultivars, which are typically of less commercial value.
- To facilitate use of MP, improve understanding of the underlying pollination biology of the target species, including flower morphology, stigma receptivity, pollen type, growth habits, agronomic requirements, susceptibility to environment conditions and risk of pollen vectored viruses.

Investigate non-crop floral resources

- Pollination in open and covered crops is provided by a suite of flying insects, which may be supported in different ways.
    - For example, ground-nesting generalist bees require a place to nest and floral support when the crop is not in flower.
- Investigate how the non-crop floral resources and other features of the orchard floor affect the presence of pollinators under protected covers, and identify practices that can increase their presence.

2 Locations and Methodology

2.1 Activities by organisation

This project was carried out across multiple sites in Australian and New Zealand, with each organisation contributing to two or more of the research subprograms, as summarised in Table 1.

Table 1 Project sub programs and institute involvement

Research activity/organisation	**UoA**	**PFR**	**DPI**	**UNE**	**U Tas**
1. Understand floral biology to inform development of mechanical and bee-vectored pollination to optimise productivity.					
a) Conduct field experiments to characterise pollen quality and nectar quality of berry (NSW DPI), onion and sweet cherry (UTas) flowers					
b) Develop pollen collection, storage and application protocols and prototypes for annual - onion, and perennial - sweet cherry, systems					
c) Assess pollinator visiting rates and their flower visiting behaviour along polytunnels/ under enclosures					
d) Relate pollinator behaviour undercover and open-field to floral resource quality and fruit set					
e) Assess the relationship between initial colony size and colony size post flowering in apple orchards					
2. Improved the performance of honey bee colonies deployed under covers through orchard manipulation					
a) Determine whether opening up the sides of netting enclosures improves hive performance and/or pollination in covered apples					
b) Determine whether modifying the length, tunnel type or configuration of tunnels improves hive performance and/or pollination in covered blueberries					
c) Determine whether the height of netting above the canopy affects colony performance/strength in covered apples					
d) Assess how different types of covers and nettings affect light transmittance, honey bee orientation (PFR) and activity (UoA; UNE) and fruit production (UNE)					
3. Assess the importance of varietal pollen presence and proximity for apple production under cover					
a) Assess the diversity of varietal pollen carried by honey bees on bodies under and outside the net					
b) Experimentally assess the importance of paternal variety for fruit set and quality					
c) Experimentally assess the utility of mobile pollinisers units for fruit set					
d) Use the information generated under 5.4 (a-c) to develop mobile polliniser units					
4. Investigate how non-crop floral resources and other features of the orchard floor affect pollinators under protected covers					
a) Investigate wild pollinator diversity and density in relation to under row and inter-row management					
b) Investigate how co-flowering non-crop floral resources influence pollinator visitation to crop flowers)					
c) Investigate how varying configurations of co-flowering plants influence crop fruit production and quality					

Each group collaborated with local growers and grower groups to conduct field experiments. A summary of research locations is provided in Figure 1.

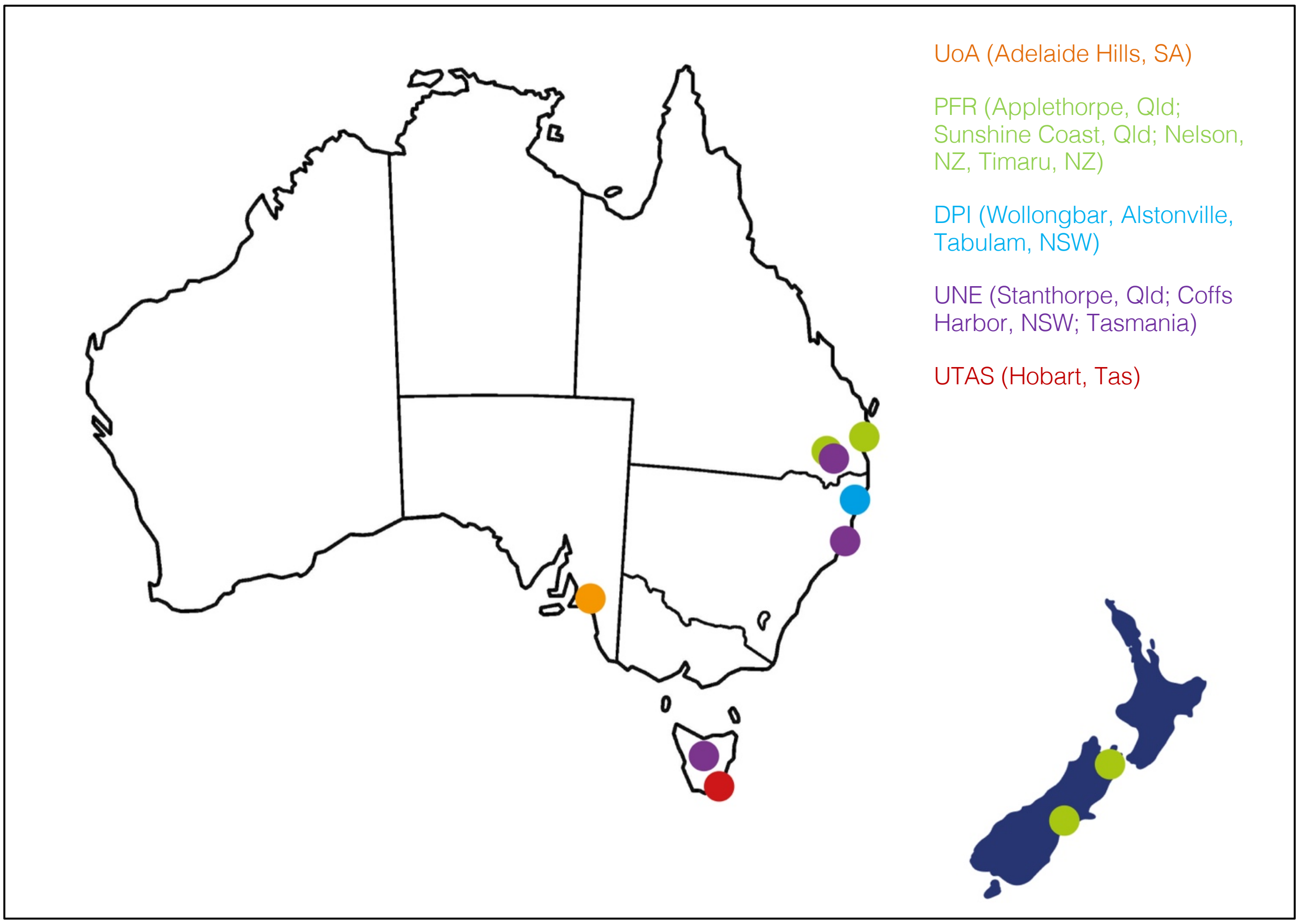


Figure 1. Research locations

2.2 University of Adelaide

Key researchers: Dr Katja Hogendoorn, Jay Iwasaki, Scott Groom, Michael McLeish, Elisabeth Williamson

Research from the University of Adelaide contributed to multiple subprograms including 1e, 2a,c,d; 3,a,b,c,d; and 4a,b (refer to Table 1Table *1* for details) and included seven investigations in apple orchards across the Adelaide Hills/Peramangk country (Figure 2) (eleven netted and one open).

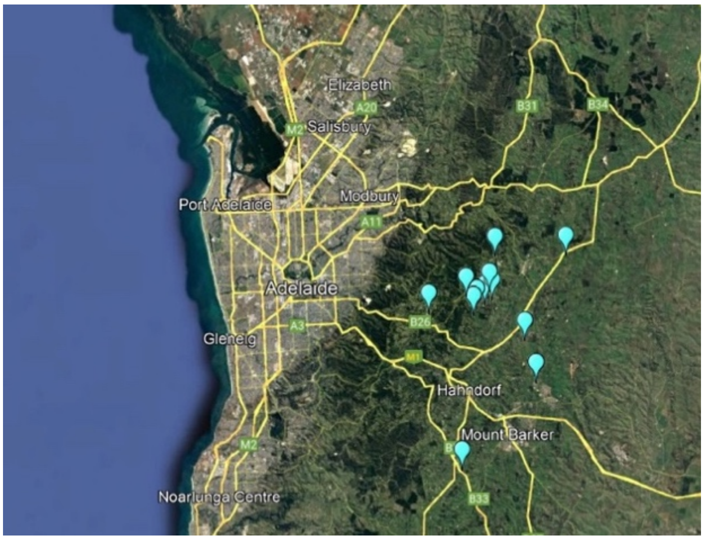


Figure 2. Location of apple orchards involved in this study in the Adelaide Hills

Orchards were either covered by rice net, which is floppy and low (1-2m) above the top of the trees, or by hail net, which is high (5-7m) above the crop and taught, (Figure 3).

Figure 3. Rice net, partially open (left), and hail net (right).

The size and shape of the netted areas varied between orchards, as did the layout, apple varieties and their relative placements, pruning strategies, orchard floor, stocking rates of bees and hive sizes, maintenance, and placement. In most orchards, the roof was closed by nets, but the sides were open during flowering. In two orchards, the area above the hive was covered by bird net which is permeable for bees.

Observations of bees

Activity and orientation

Bee activity on apple flowers was quantified by counting the number of bees seen per ‘panel’, which was the distance between two poles that upheld the net. This was carried out regularly, in a single large orchard, to assess the effect of distance to the edge and visitation in relation to fruit set. In addition, this bee activity was assessed over three days in 2023, comparing foraging activity under hail net and in the open where part of hail netting was damaged and had been removed over a distance of 90 m length and four rows wide.

Orientation of bees out in the open, under hail net and rice net was investigated using a method modified from Free (1966). Modifications included: (a) marking bees over a stretch of 2m of flowering apple (single trees are not meaningful in current, ‘wall-type’ orchards); (b) later quantifying the proportion of marked bees in the area where they were initially marked, rather than checking the whole orchard for marked bees; (c) marking bees by placing a dot of the thorax with a Posca pen (www.posca.com , PC-3M and PC-5M, 4 colours per orchard, 50 bees per colour) as they were collecting pollen or nectar on flowers, rather than catching and painting them.

At three orchard blocks, one in the open, one with a closed hail net, and one with a closed rice net, four marking locations per orchard were selected. These locations were dispersed throughout the orchard; 50 bees that visited flowers at each 2-meter-wide location were color-marked, with different colors used at each site. At each site where bees had been marked, a repeated ten-minute observation was conducted to count the number of marked and unmarked bees visiting later in the day and during one or two consecutive days. A minimum of three observations per marking site were collected. The average number of marked bees observed was then calculated for each orchard, marking site, and day.

Hive health and pollen collection

The number of bees stuck in nets per m^2 was quantified for hail net, and for open and closed rice net. This was quantified by walking 500m and counting the number of bees stuck in nets at the end of the flowering period. For hail net, the number of bees stuck was analysed aginst net height.

Hive health was assessed over two years using the Liebefeld method (Imdorf 1987, Dainat et al. 2020). This involved assessing the number of adult workers covering the side of each comb, as well as the surface area (dm^2) of the comb that is occupied by the open brood (developing larvae), capped brood (pupating larvae), honey and pollen stores.

The change in hive health was then quantified by subtracting the first from the second set of observations. Data from both years was combined and changes in hives with easy access to the outside: i.e., that had been placed under gaps in the roof, in the open or under bird net, which is permeable to bees (n = 32 hives) were compared to those in the middle under closed net (n = 22 hives), for each of the variables measured. Changes in hives placed under closed hail (n = 10 hives) and closed rice net (n = 12 hives) were also compared.

To investigate the reasons for hive declines under net, pollen traps were used to investigate the pollen collected by bees in hives situated in the open (n = 12 hives) or under permeable (bird net or opened rows; n = 6 hives; Figure 3) and under closed roofs in the middle of the orchard block, far removed from the open sides (n = 10 hives). The hives had been in position for more than a week, and days were selected with good foraging conditions (i.e., >17°C and sunny).

The pollen was collected after 24 h, weighed, and ground. A representative sample from each hive was mounting on a slide with glycerin (Erdtman 1960) and the morphology of 400 kernels assessed under the microscope. Pollen from apple, capeweed, other Asteraceae, Photinia and Myrtaceae were recognised and classified as such, and all other pollen species were classified into morphospecies. T-tests were used to investigate how the position of the hive affected the weight of the pollen collected, the number of morphospecies, and the proportion and total weight of apple pollen kernels collected.

Effects of opening rice net on bees and yield

To investigate whether it is worthwhile to open the rice net above the interrow during flowering, we quantified bee activity, fruit set (applets per flower cluster), seed set (seeds per apple), fruit symmetry in rows under opened and closed rice net (Figure 2).

Orchard layout

The effect of proximity of pollinisers was investigated in two ways.

1. Set was quantified throughout a large orchard block and analysed in relation to the proximity of polliniser varietals and distance to the edge of the row.

2. Molecular markers were used to investigate the varietal origin of pollen that was deposited and had led to the production of a seed. To do this, 'Cripps Pink'/Pink Lady® apples were harvested (15 April 2020; 18 April 2021) in a grid through the orchard. Seeds were then removed from the individual apples, stored at 4°C before germinating and growing to seedlings. The microsatellite loci of the seedlings were analysed to identify the fathers, and therefore which apple cultivar had pollinated the Pink Lady apples. The closest possible tree of the inferred pollen donor was mapped to assess the minimum distance over which pollen had been deposited.

Mobile polliniser unit

Investigation into the selection of varietals for the mobile polliniser unit involved experimental hand pollination with different varietals. While research intensive, this did not yield any new insights and will not be further reported. However, this work confirmed that requirements for suitable pollinisers are:

1. Flowering phenology that is synchronous with the target variety.
    - If there is a difference, flowering can be protracted or delayed by altering environmental circumstances before flowering

2. Different S-alleles. This implies that they are descendants from crosses with different parental lines.

A robotic mobile polliniser unit was simulated by arranging 12 potted flowering apple trees, roughly 1.5m tall, (polliniser cultivar: 'Granny Smith') on a vehicle tray (Figure 4), and systematically driving back and forth down an orchard row (cultivar: Pink Lady), stopping for 30 minutes at measured intervals. At each interval, the number of honey bees crossing between flowers on the polliniser unit and the apple rows were counted to quantify the number of potential pollination events, defined as bees crossing from the MPU to the apple rows.

Bees were observed successfully using the simulated MPU and crossing between the cultivars. 'Granny Smith' trees were selected because they had the best temporal overlap and known effectiveness in cross-pollinating Pink Lady cultivars (see Activity 5.4b on p 50).

Figure 4. (left) Jay Iwasaki scoring bees transitioning between the pollinisers on the MPU and the fruiting crop.

Figure 5. (right) Aerial photo of apple orchard under rice netting. Trial row is in white, and control rows are in green. All experimental rows are separated by one row not used in field experiments.

Apple trees were arranged in lines across the tray to encourage honey bee movement between rows. The number of pollinisers used was sufficient to create an unbroken corridor of two rows along the bed between rows (two rows of 6-10 trees).

Trials were undertaken in late September and early October 2021 and 2022, between 10am and 6pm, when temperatures were at least 12°C, for roughly seven hours per day during six suitable days out of a total of two weeks during full flowering. Trials commenced when bees began foraging and ended when there were relatively few bees active on the crop. There was one treatment row with the MPU, and four control rows. Control rows were on either side of the treatment row with single rows as buffers (Figure 5).

To verify pollen transfer, fruit and seed set were recorded after bees transitioned from the flowers on the MPU to the flowers in the adjacent rows and compared this with transitions from flower to flower in control rows. To assess efficacy of the MPU, fruit set (applets per cluster) and seed set in the MPU row and control rows were compared.

Because the MPU was not economically feasible, implementation was not further developed.

Native bees and the orchard floor

A total of 14 orchard blocks from eight properties were evaluated for factors that may influence wild pollinator diversity and density during the apple flowering period.

Within a single row of each orchard block 15 trees were selected and all floral visitors counted over a 1 minute period. The floor under the trees of each orchard block was classified for two factors: i) the vegetation cover, either vegetated or bare, and ii) the shape of the orchard floor, either mounded or flat.

Using two generalised linear models with Poisson distributions, the effect of tree floor management on the presence of wild pollinators and non-Apis bees was assessed. In both models the importance of under-tree cover, floor shape, temperature, and the mean number of flowers per tree as predictors of wild pollinator or non-Apis bee presence were evaluated.

Non-crop floral resources

In 2021, presence of native bees on weeds before flowering in the orchard was assessed whenever the weather permitted (on 11 September, 18 September, 1 October).

In 2022 and 2023, flowering weeds were assessed in more detail for the presence of pollinators. Up to 100 flowers of different species were counted and quantified the number of visitors in four categories: honey bees, hoverflies, other flies, and native bees.

In 2022 and 2023, native bee visitation was assessed in a trial led by UTas and coordinated in South Australia by Fruit Growers SA. This trial investigated the effect of orchard floor treatments on the adjacent apple trees. The inter-row treatments were:

a. grower's standard inter-row sward (Lenswood orchard mix)
b. native grass mix
c. meadow mix (a mixture of mostly North American annuals and perennials (Table 3), supplied by Meadow Flowers Australia).

Of these combinations, only the meadow mix contained flowers that were useful for generalist bees; their visitation was quantified.

The three types of interrow plantings were alternated in neighbouring rows, each with one untreated row in-between. The aim of the UTas study was to monitor effects on IPM, yield and fruit quality.

The trial did not investigate the effects on pollination as the treatments were too close together, i.e., all within 100m, which is within the flight range of native bees and most other insects. However, the trial did facilitate assessments of how the native bees present in the orchard made use of the meadow mix. This was only feasible during 2022, because the meadow mix was either not resown in 2023 or did not produce flowers.

2.3 Plant & Food Research

Key researchers: Dr Lisa Evans, Brian Cutting, Dr Melissa Broussard, and Dr Mateusz Jochym.

Trials were undertaken in 30 blocks across 10 apple orchards and four lychee orchards. Orchards include:

- One large orchard near Applethorpe, Queensland (Sep – Oct 2019),
- Five orchards in Timaru ,New Zealand (Oct – Nov 2019 and 2022),
- Four orchards in Nelson, New Zealand (Oct 2020, 2021).
- Four lychee orchards (11 blocks) on the Sunshine Coast, Queensland (Sep – Oct 2019).

Relate pollinator behaviour undercover and in the open-field to floral resource quality and fruit set

Pollinator behaviour

Observers used audio recorders to follow individual honey bees, annotating their visits to flowers. Observed honey bees were chosen haphazardly while they foraged on flowers underneath full netting, in uncovered blocks, and underneath gaps in the roof netting (netting rolled up in one row out of every four). Each honey bee was followed until it flew out of sight, or 5 minutes had elapsed.

Fruit set and quality

Fruit set and quality was assessed in covered versus open grown apples, apple flowers were prepared and marked in a partially covered and uncovered block of both 'Smitten®' and 'Ambrosia' (the same blocks used as above) as follows:

1. Supplementary cross (allogamous) pollination with a fully compatible variety (no shared S-alleles); 30 flowers were hand-pollinated with a different cultivar and then left exposed to floral visitors;
2. Open pollination; 30 flowers were left exposed to floral visitors;
3. Closed pollination; 30 flowers were enclosed in greaseproof paper bags to prevent insect visitation/pollen transfer.

Figure 6. Pollinating apple flowers and bagged and marked apple flowers

Determine whether gaps in netting / opening sides improves colony performance and/or pollination.

Hive performance

To determine whether cover type affects the health of hives deployed for apple pollination, adult bee numbers (as frames of bees; FOB a measure of colony strength) were compared in blocks fully covered with white hail netting, partially covered (roof only or a roof and partial sides), and uncovered (control) blocks.

Fully netted blocks with a gap in the roof of the netting (e.g., Figure 7) and partially netted blocks with a gap in the roof of the netting were also compared. Similar assessments were undertaken to determine whether cover type affects the health of hives deployed for lychee pollination; adult bee numbers in four blocks fully covered with black hail netting and five blocks partially covered (a roof and some sides) with black hail netting, were compared

Figure 7. Example of a gap in the roof of a hail netting covered apple orchards. The size of 'gaps' used in the current study varied between blocks and orchards as they were installed by the growers.

All hives were moved into the blocks at night at approximately 10% flowering. Colony strength was assessed the morning after hives were deployed in orchards using a modified cluster-count method; the number of frames covered by bees was estimated to nearest quarter of a frame in both the top and bottom boxes (e.g., Figure 8; Nasr et al. 1990). Colony strength was re-assessed prior to hive removal from the blocks. All assessments were conducted starting at first light, before the bees began to forage.

Figure 8. Honey bee colony strength assessment method; photo of bee cluster over the top of hive frames. The number of frames covered by bees ('cluster size') was estimated to nearest quarter of a frame.

In addition to looking at the effect of netting structure on colony performance, the effect of hives' position within the orchard was also examined. Colonies were categorised as being

1. Open/uncovered (no netting)
2. At edge of partially (roof only) covered block
3. In the middle of a partially covered block
4. Under gaps in roof netting of partially covered blocks
5. On the edge of fully covered blocks
6. In the middle of fully covered blocks, and
7. Under gaps in roof netting of a fully covered block.

Bee movement and pollination

To assess bee activity on apple flowers relative to hive position, a fluorescent powder was used to mass mark bees as they left their hives (Figure 9). Powder dispensers were attached to all the colonies within two covered and one uncovered block. Where there were multiple groups of hives within a block, a different coloured powder was used for each group. On average, the powder dispensers marked 80% of bees exiting the hives (the data have been corrected to reflect this), so any unmarked bees observed in surveys were more likely to have come from another (unmarked) hive outside of the block.

Surveys of bees on apple flowers (marked and unmarked) were completed in each of the three blocks between 1000–1030 h, 1200–1230 h, and 1400–1430 h on one fine weather day. These data were collected with co-funding from The Agricultural and Marketing Research and Development Trust (AGMARDT).

To determine whether cover type affects bee activity on lychee flowers, surveys of honey bees and other pollinators were conducted on 36 trees across each of the nine trial blocks. Surveys were completed between 8:00–9:00, 10:00–11:00 and 12:00–13:00 h, and during each survey period a different 12 trees were observed; six on the edge and six located in the middle of the block (i.e., 50–100m from edge). On the sunny side of each survey tree, 1m^2 of flowers was be observed for 2.5 minutes and all insects seen visiting female and male flowers were recorded. On the same day as insect surveys, open flowers were counted on six of the 36 survey trees to enable insect visitation rates to be standardised.

Figure 9 (A). Powder marking dispensers attached to the front of colonies. (B) The dispensers deposited coloured powder on the dorsal-thorax of incoming and outgoing bees, which was visible whilst foraging on flowers.

Assessment of how different types of covers and nettings affect light transmittance and honey bee orientation

Light transmittance

To assess light transmittance photos were taken under various netting conditions during the period of apple bloom in two different years: 2-13 October 2021 (Nelson, New Zealand) and 16-21 November 2023 (Stanthorpe, Australia). A polarisation sensor (Polarsens camera, The Imaging Source) was fitted with a wide-angle fisheye lens (Fujinon FE185C086HA-1, Fujifilm) and mounted on a tripod 1.4m above the ground facing directly up. The angle between north and the direction the camera was facing was recorded for each measurement. Images were taken under a minimum of two exposures, including one where there was minimal overexposure from the sun, and one where the periphery of the image was well-exposed. The angle and degree of light polarisation was recorded in three channels (RGB), but only the blue channel was used for further analysis as bees are most able to detect polarisation in blue and ultraviolet light bands.

Polarisation measurements: The effect of nets on the degree of linear polarisation was measured at selected points, generally where the degree of linear polarisation was highest. Points were selected along the arc passing approximately through peak-polarisation, centred at the sun position and as near to a line passing through the image centre as practical. Image artefacts, such as lens flare, and orchard structures were avoided. In cases where distinctly different polarisation was observed along net or structural boundaries, multiple points were selected (Figure 10).

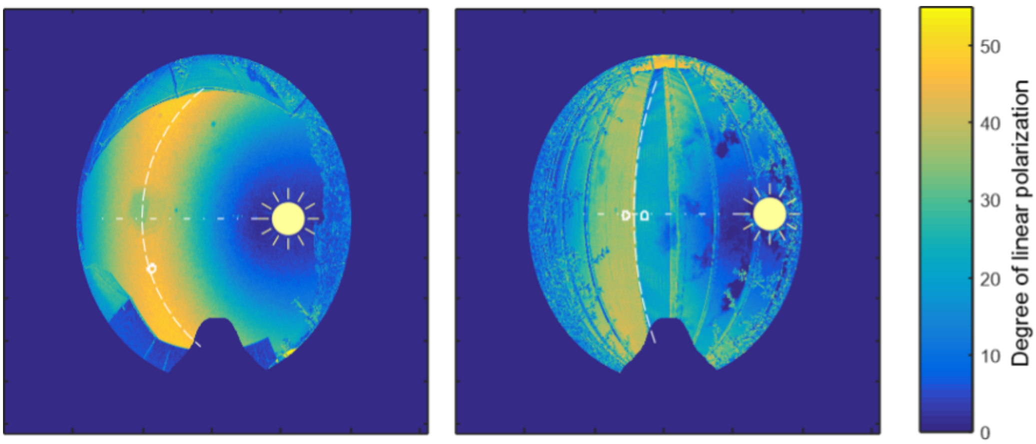


Figure 10. Peak polarisation was estimated at selected points (left); multiple points were estimated along boundaries created by the net and/or orchard structures (right). Figure by Paul Martinsen.

Forager navigation

To determine how different nettings affect honey bee orientation, a four frame observation hive was deployed under two different types of hail netting (black and white), as well as an uncovered (control) apple orchard at Stanthorpe, QLD. The trial was conducted outside of apple flowering and marked bees were trained to a sucrose feeder ~150m from the colony and the dance behaviour of returning foragers was video recorded in the colony using infrared cameras (Figure 11). The direction of the bees' waggle dances were analysed from video footage collected between 12:20 and 14:20, for three to four days per treatment. We assessed the variation in dance direction and mean deviation from expected direction for both netting types compared to the control.

To investigate if netting type influenced the angle of bee dances relative to the true angle (delta angle = observed angle - true angle) we used a generalized linear mixed effects (GLMM) model in R (R Core Team, 2022) using the brms package (Bürkner, 2017). Fixed effects were an interaction between netting type and true angle, with each dance observation specified as a random effect (multiple dances from the same bee were treated as nested). A von Mises distribution was selected to account for circular data, which produces two parameters: the mean angle (μ) and the concentration parameter (κ). Model selection was based on leave-one-out cross-validation (LOO) and model validation was performed using prior and posterior predictive checks, as well as effective samples sizes and r-hats (McElreath, 2020).

Figure 11. Setup for analysing bee navigation under netting in covered (A) and uncovered (B) growing environments. Foragers were trained to a feeder ~150m and marked with paint and numbered tags (C) to enable their dance behaviour to be tracked in their observation hive (D) using infrared lighting and night vision cameras (E).

Bee movements away from pollinisers

How far bees were foraging from target polliniser trees on which they were caught and marked, both under netting and in the open, was also examined. Bees were captured on two pollinisers in each of four blocks (two blocks completely covered in white hail netting and two blocks uncovered).

Captured bees were placed in a honey bee queen marking tube (Ecrotek) and given a unique colour dot on the back of their thorax (e.g., polliniser 1 was white, polliniser 2 was blue). The number of bees marked varied between pollinisers and to increase numbers, additional bees were typically marked prior to Day 2 surveys (described below). The maximum number of bees marked per polliniser ranged from 100 to 128.

Surveys of marked bees on flowers were conducted over two days (typically in the afternoon of Day 1 and in the morning and afternoon of Day 2). The area survey expanded over 10-row of 10-bays (approx. 500 trees; Figure 12), for the first pair and was extended to 20 rows of 5 bays for the second pair of open and covered blocks (Figure 13. Location *of* trees within the 10-row, 5-bay sample grid. Pollinisers with marked bees are red. Trees highlighted in green were included in extended surveys of the orchard.

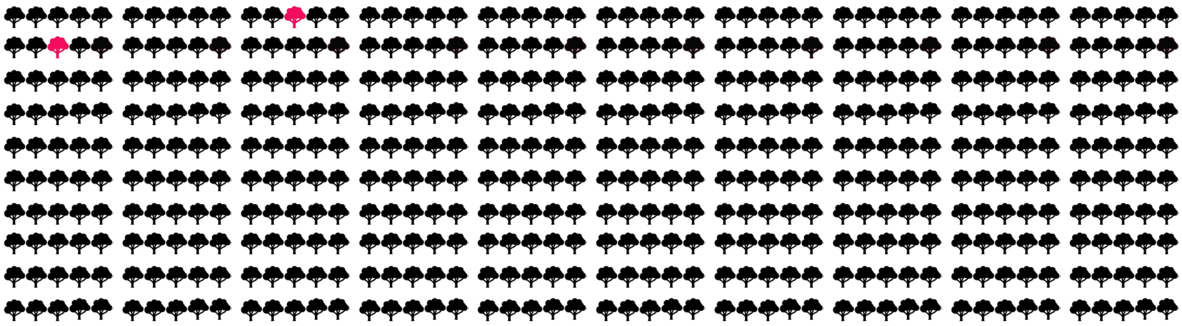


Figure 12. Idealised location of trees within the 10-row, 10-bay sample grid. Pollinisers with marked bees are red.

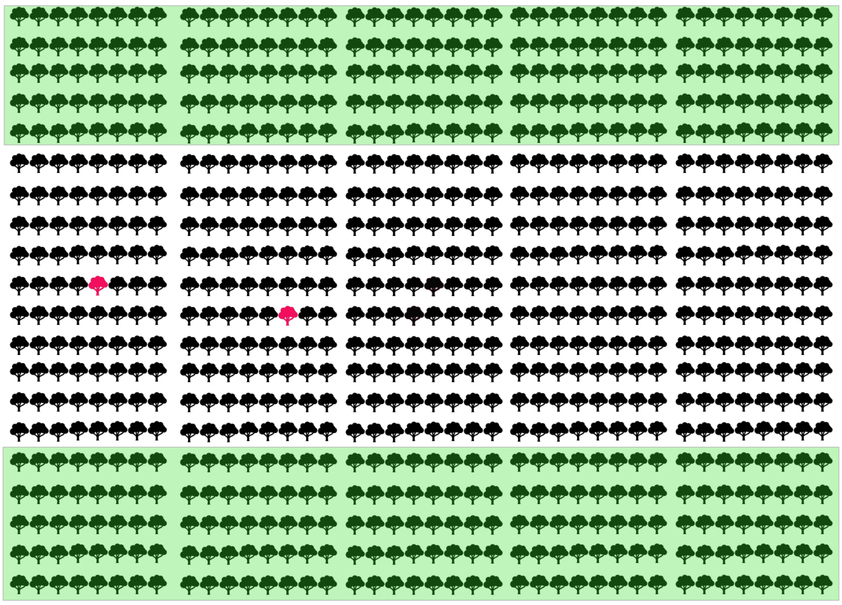


Figure 13. Location of trees within the 10-row, 5-bay sample grid. Pollinisers with marked bees are red. Trees highlighted in green were included in extended surveys of the orchard.

2.4 NSW Department of Primary Industries

Key researchers: Dr Sophie Parks, Melinda Simpson and Dr Madlen Kratz

Department of Primary Industries (NSW) undertook five investigations to understand the floral biology, including nectar secretion patterns, nectar sugar concentration, and pollen production, in flowers of several Vaccinium (blueberry) and Rubus (raspberry and blackberry) varieties used in Australian production systems. This research contributed to subprogram 1, part a,d (refer to Table 1 for details).

Initial plans included pollen studies in collaboration with Dr Evans from PFR. However, due to travel restrictions imposed during the COVID 19 pandemic in 2021, experts from PFR were unable to travel to NSW to participate in the studies. Research was also limited to blueberry, with UNE undertaking the Rubus work.

Field and controlled experiments conducted/completed in northern NSW (Figure 1) included:

Description of variation in floral morphology among seven blueberry cultivars.

Flowers were sampled from seven cultivars of low chill southern highbush blueberry (*Vaccinium corymbosum* interspecific hybrids) including 'Dazzle, '11-11', 'Eureka', 'Eureka Sunrise', 'First Blush', 'Masena' and 'Splash', growing in the same location of northern NSW. The flowers were preserved in alcohol, and their morphological characteristics described using methods in microscopy. The floral characteristics described are shown in Figure 14.

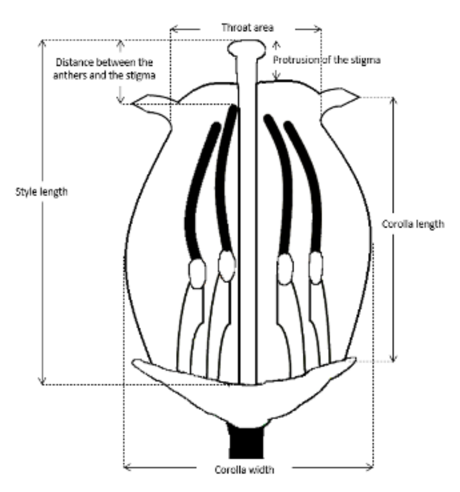


Figure 14. A diagram of a blueberry flower indicating the morphological characteristics described for seven blueberry cultivars.

Effect of floral age and time of day on nectar characteristics in blueberry *Vaccinium corymbosum* 'First Blush'.

Individual closed flowers growing in the field were tagged and covered with mesh bags to exclude pollinators and observe the dynamics of nectar production. Nectar was collected from some flowers over several hours on the first day of opening and then from other flowers over the next 8 days. Nectar was removed by 'washing' the flowers in water, and the sugars (including sucrose, fructose, and glucose) were analysed in the extracts using an enzymatic kit method.

Effect of covers (tunnel or bird net) on blueberry pollination.

In July 2021, over a 2-day period, bee visits to flowers were counted and floral nectar of individual flowers collected and tagged for later assessment of nectar sugars, berry weight and berry sugars, (indicated by total soluble solids (degrees Brix)), under two blueberry blocks with contrasting cover.

Bird net covered one block, and high tunnels covered with plastic film were installed above the adjacent block.

For the control treatment, mesh bags were placed over flowers, preventing honey bees from taking the nectar and introducing pollen, which would otherwise facilitate pollination (Figure 15).

Sensors were also placed within the crop to record temperature, relative humidity, and wind characteristics.

Figure 15. One of the mesh bags placed over a blueberry flower used to exclude pollinators from entering the flower. This was the 'no pollination' control. Three to five of these were placed on each experimental plant.

Effect of fertigation strength on plant nutrition and nectar production in potted blueberry *Vaccinium corymbosum* 11-11.

Glasshouse experiments were conducted at the Wollongbar Primary Industries Institute with 30 mature blueberry plants (variety 11-11), growing in potted substrate. Three fertigation treatments of low, moderate, and high concentrations of nutrient salts (electrical conductivities of 0.7, 0.9 and 1.2 dS/m), at 10 plants per treatment, were applied. Pollinators were excluded.

During peak flowering, nectar from six flowers of the same age, per plant, was extracted using a pipette. The nectar samples were weighed and analysed for their sugar concentrations. Other measurements included plant canopy area, flower count per plant and temperature and relative humidity during the experiment.

Effect of cultivar on honey bee visitation to blueberry flowers (cultivars Masena, 11-11, Eureka Sunrise).

To examine the impact of cultivar on floral attractiveness to honey bee visitation, three contrasting cultivars that flower concurrently, Masena, 11-11 and Eureka Sunrise were selected to assess their nectar reward (sugars, plus proportions of sucrose, fructose, and glucose), and their ability to attract honey bees in the field.

Floral stations containing bunches of all three cultivars were placed in interrow spaces within each varietal block, replicated three times. Honey bee visits to floral stations and to the adjacent crop were counted over two days. Floral nectar was also collected from each cultivar over two days with four replications.

2.5 University of New England

Key researcher/s: Dr Romina Rader, Dr. Karen Santos, Carolyn Sonter and Jeremy Jones

Researchers from the University of New England (UNE) contributed to multiple projects in each subprogram, specifically, 1c,d; 2a-d, 3a,b; 4a,b,c (refer to Table 1 for details).

Research was conducted from 2019 to 2024 at 10 apple sites in Stanthorpe, QLD, seven blueberry sites in Coffs Harbour, NSW and three cauliflower sites in Tasmania (Figure 1). Industry partners were consulted for blueberry (Costa and OzGroup) and apple (Orchard Services Stanthorpe). Research was conducted on commercial farms for all trials.

The team focused on the following research topics to meet subprogram objectives:

1) To determine how netting and poly tunnels impact pollen flow, flower visitation and honey bee health
2) To determine how the addition of floral resources impacts flower visitation in protected cropping

Figure 16. Monitoring honey bee hive health under protected cropping netting (left), honey bee foraging on a blueberry flower (right) (Photo credit: J. Jones, UNE)

Effect of blueberry cover type on insect visitation and yield

Eighteen 100-metre-long blueberry tunnels were grouped into consecutive sets of three and covered alternately with bird netting or polyethylene. This was repeated for two cultivars with different flowering periods (winter or spring at the Coffs Harbour study site). Within the middle tunnel of each set of three, two groups of plants were identified to represent the central and edge sections of the tunnel. In each group, two plants were left uncovered, two were completely enclosed with bee-proof netting to act as controls for insect visitation, and the remaining two were partially enclosed with netting to allow bees access to the flowers while serving as controls for shading effects.

Twenty hours of observations on insect visitation were recorded for the open and partially enclosed plants during the peak bloom period of each cultivar, along with counts of the open flowers on each plant. As the berries ripened, they were harvested and weighed from each plant. Additionally, transect walks were conducted across polyethylene tunnels of different lengths to evaluate the effect of tunnel length on pollinator density within the tunnels.

Effects of different protected cropping structures on pollen composition on stigmas

This study was conducted on 10 farms located between Macksville (30◦41′56"S, 152◦54’ 11"E) and Halfway Creek (29◦56’ 20"S, 153◦07’ 27"E), NSW. We focused on 6 cultivars of southern highbush blueberry (*Vaccinium corymbosum* L. interspecific hybrid) as a model system, to determine the extent to which pollen flow (conspecific and heterospecific pollen) on stigmas would vary in fully netted, partially netted and unnetted fields (example in Figure 17 below).

Figure 17. a) unnetted, b) partially netted, and c) fully netted blueberry blocks.

Stigma sampling

The study commenced data collection 14 days after initial flower bud opening, coinciding with hive deployment and 15% bloom across all sampled blocks. Blueberry stigmas were collected every two weeks for 13 weeks, ending at the conclusion of flowering. In the first week, eight branches were tagged per block - one at the edge and one in the middle of four randomly chosen rows to allow to analyse if the stigmatic pollen load composition would differ between the edge and middle of blueberry blocks.

Stigmas from one flower per tagged branch were mounted on slides immediately after collection. Flower buds were tagged early in development, taking 4–5 days to open. A 48-hour period post-flower opening ensured pollen deposition before stigma collection. Pollen grains on stigmas were quantified and classified as conspecific or heterospecific, with heterospecific pollen identified to the family level using reference collections and existing literature.

Comparison of netted and unnetted apple blocks upon insect visitation to apple flowers

Insect visitors to apple flowers were observed during the peak bloom period across nine netted and three unnetted orchard blocks on eight farms in the Stanthorpe region, Queensland. Observations were made along randomly positioned transects in each block, covering 10 consecutive apple trees at least 10 meters away from any edge, focusing on a three-dimensional search area around each tree. Each tree was observed for one minute to record all insect visits to flowers, and a tally of open flowers was made. Observations were conducted up to twice per day during specific time windows. Tree under rows that had overhead netting opened in an effort by growers to increase visitation to apple flowers were recorded, together with the mean height of the trees within the block and the mean net height.

Effect of protective covers on bee colony development and access to resources

The investigation was conducted on four commercial blueberry farms located on the Mid North Coast of New South Wales, Australia, involving a total of 14 bee hives, with half situated under netted blocks and the other half outside of the nets. Sampling took place fortnightly from late May to late August, involving detailed inspections of each hive's brood box, weight measurements, and ambient temperature recordings. The research utilised Langstroth bee hives with a configuration of 8 to 10 frames in a single brood box and an initially empty honey super, ensuring that at the start, all bee colonies were of comparable size, health, and resources. Additionally, the study assessed honey bee access to external resources by tracking their attempts and successes in exiting netted enclosures, measuring both the duration of attempts and the frequency with which bees collided with the nets, and included floral surveys of the area under the nets and surrounding the farms.

Effects of co-flowering plants on blueberry floral visitation, berry weight and pollen flow

The study was conducted in southern highbush blueberry orchards in Coffs Harbour, NSW, focusing on the pollination dynamics involving honey bees, stingless bees, and other insects, against the backdrop of blueberry's partial pollinator dependency. To enhance pollination, Mexican heather and basil, previously found to be compatibility with blueberry cultivation and attractive to key blueberry pollinators, were introduced into the orchards. Observations and data collection spanned various scales, including individual plant pairs and whole rows, to assess the impact of these added plants on pollinator visitation and pollen flow. In 2022, a before-after control-impact experiment further explored the effects of adding or removing basil plants on insect visitation and berry weight in both netted and polytunnel-protected orchard blocks for two cultivars.

Effects of protective nets on pollen flow and composition along and between rows in apple orchards

The study took place from September to October 2020-2022 across 24 apple orchard blocks on nine farms near Stanthorpe, Queensland, Australia. Orchards ranged from 0.97 ha to 6.31 ha, with trees spaced 4 m apart in rows and planted 1.2 m apart within rows. Most growers used protective covers, either flat or pitched nets, though some orchards were open (Figure 18). Common cultivars included 'Cripps Pink'/Pink Lady®. and 'Gala', often grown together.

Figure 18. Apple orchard blocks a) unnetted and covered with b) pitched net and c) flat net.

We selected nine blocks covered with pitched nets, 8 with flat nets and 6 blocks were unnetted. In each block we randomly chose two pairs of rows at least 30 m apart. Pollinator efficiency work was conducted to investigate number of visits by bees to result in fruit set (see Stavert et al. and Lobaton et al.) In each row, we tagged 50 trees and applied a fluorescent powder on 20 flowers in every fourth tree. We always chose flowers that were still attractive to pollinators (i.e., with all the petals and anthers), and each row received a different colour so we could track if bees were moving along or across the rows.

Using a small paintbrush, we applied the fluorescent powder around the pistil base and on the anthers of the flowers. We tagged the inflorescences that received the powder with a reflective flagging tape so we would not count those flowers when scanning the blocks with the UV light at night.

The powder was applied on the flowers in the morning of day 1 and we scanned the block in the evening of day 2 using a 365nm UV LED. In all the blocks, we always scanned the 50 trees tagged within the four rows we used in the experiment in case the received a different colour other than the one applied. We searched for the powder in all the flowers at eye height (approximately 1.65 m) and below. We counted the number of inflorescences with the powder and not the number of flowers, and we only considered as a visit when the powder was on the reproductive structures (i.e., stigmas and/or anthers) (Figure 19).

Figure 19. a) Apple flowers receiving fluorescent powder, b) honey bee visiting painted flower, and c) apple flowers with fluorescent powder under UV light at night.

2.6 University of Tasmania

Key researchers: Dr Alistair Gracie, Dr Alieta Eyles, Dr Ryan Warren, Dr Cameron Spurr

Researchers from the University of Tasmania (UTas) contributed to subprograms 1a and b (refer to Table 1 for details) on onion farm and cherry orchards in Tasmania (Figure 1).

This project was undertaken from 2019 to 2024 in southern Tasmania. Industry partners were consulted for sweet cherry (Hansen Orchards and Reid Fruits) and onion seed (South Pacific Seeds) and included in the planning. Commercial production sites owned by the industry partners were utilised for all trials.

The two model systems selected were a perennial (sweet cherry) and annual (onion seed) crop. These are reported separately in each section. For the sweet cherry sub-project, the main activities were two-fold:

1. Improve our understanding of the floral biology of the main commercial cultivars grown in Australia
2. Develop protocols and equipment for evaluation of pollen quality, pollen collection, handling and storage, and pollen application.

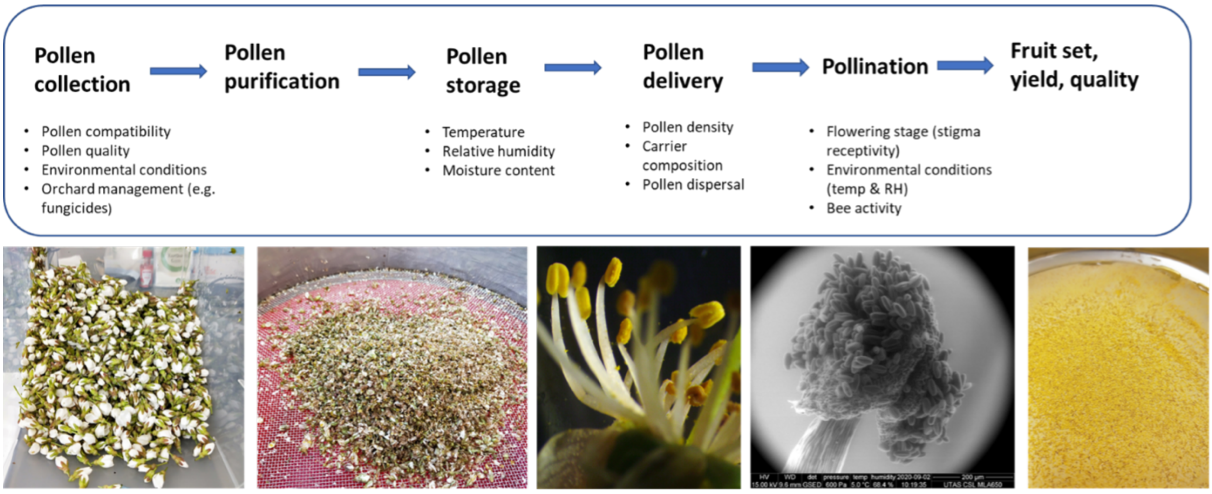


Figure 20. Key steps and factors in mechanical pollination systems.

Pollen collection

Sweet cherry

Unfortunately, the ‘sticky’ nature of cherry pollen meant that mechanical harvesting of pollen was not possible using a vacuum system. Furthermore, it was not possible to extract pollen from dried flowers.

Due to this, the optimum harvest strategy was handpicking unopened fresh flowers at the closed-bud stage (Figure 21). These flowers could be harvested at an approximate rate of 6-8 kg per day by four trained pickers, but mechanical collection systems could be developed in orchards designed for the specific purpose of pollen collection. Flowers had to be closed to ensure the pollen remained genetically pure and of high quality.

Onion

Several broad collection methods were tested for harvesting pollen directly from onion inflorescence, including a vacuum, a tumbling barrel and hand processing with a sieve (control). The efficiency of each method for pollen yield per umbel and quality were compared. The vacuum was the most efficient method.

Several modifications were tested including in-line filters, a cyclone, baffles, and variable air velocity. The optimum time of day for harvesting pollen was also assessed. This included single and multiple pollen harvests of the same onion inflorescence. All harvested umbels were held in the laboratory at 25°C and 55%RH.

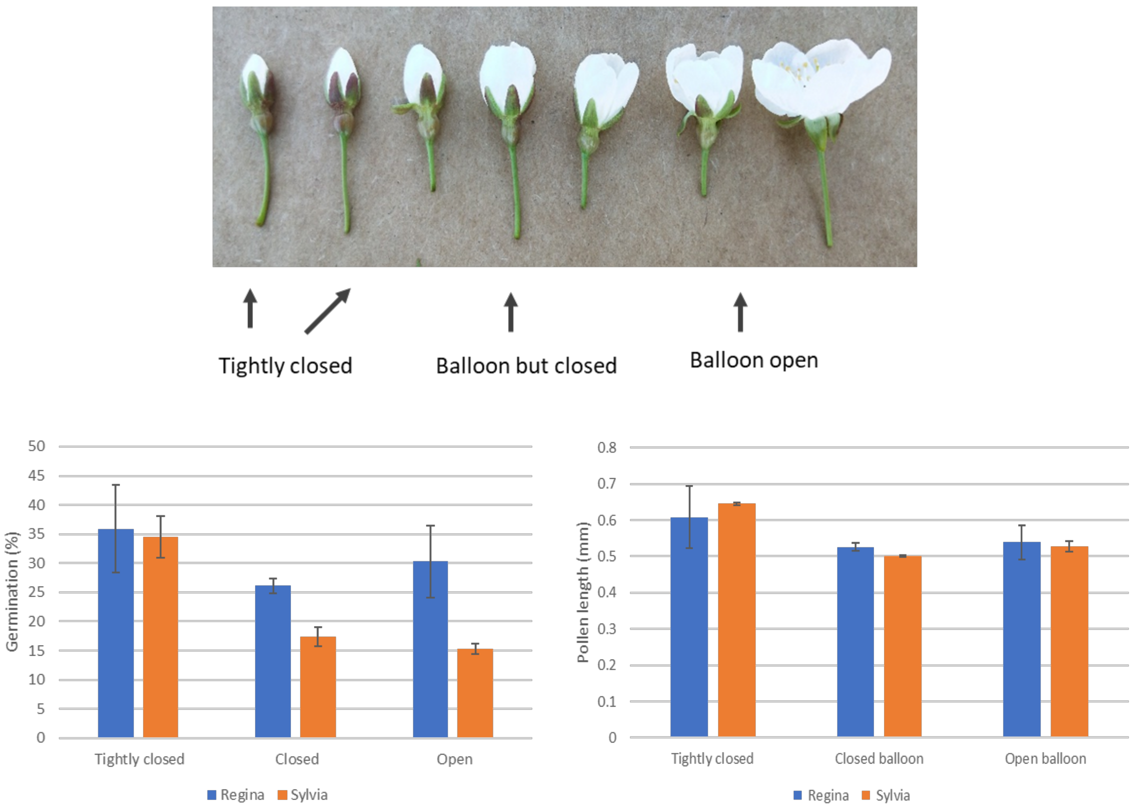


Figure 21. Effect of flowering stage (tightly closed, balloon but closed, balloon open) on pollen viability (germination and pollen tube) of Regina and Sylvia. Bard = standard errors, n= 3.

Evaluating Pollen Quality (in vitro and in vivo methods)

In developing mechanical pollination systems, an in-depth understanding of pollen biology and rapid in-vitro techniques for predicting pollen performance in the field were required.

Sweet cherry – in vitro

A variety of methods that assessed pollen viability were evaluated. The conventional fluorochromatic reaction (FCR) biochemical test used for in vitro assessment was found to overestimate pollen germination. More consistent results were obtained with agarose-sucrose medium using 15% sucrose stored at 20°C for 24hr.

This was confirmed through complementary laboratory experiments that examined a range of factors, which may influence pollen germination rate and tube length e.g. incubation temperature (10, 15, 20, 25 and 30°C), concentration of sucrose (0, 5, 10, 15, 20, and 30%) and boric acid (0, 5, and 100 ppm) in media, media type (i.e., agar versus liquid media) and the effects of Tween 80 (0, 10, 100 and 1000ppm) in reducing pollen clumping.

Sweet cherry – in vivo

An in vivo method was also optimised for sweet cherry based on a protocol by Kho and Baer (1968) testing both fresh and stored pollen. Closed flower buds on detached ~ 20 cm branches were emasculated by removing petals and stamens. Pollen treatments were applied to pistils and incubated for 48hr at 20°C. Subsequently, pistils were excised from the receptacle using a scalpel and placed in an Eppendorf tube for digestion in 250 - 500uL 4M NaOH (enough to cover the pistils) at 60°C for 40m. Pistils were carefully rinsed eight times in double distilled water, then stained by submerging them in 250-500uL 1% Aniline Blue in 0.1M K_3PO_4 for a minimum of 2hr. Pistils were gently squashed onto a glass slide with a coverslip and a drop of 80% glycerine. A Leica DRMB fluorescence microscope (Leica Microsystems (Switzerland) Ltd) fitted with a mercury vapour lamp (wavelength 350-400 um) was used to view pollen tube growth.

Unlike the original protocol of Kho and Baer (1968), best results were obtained by digesting pistils 4 M NaOH at 60 °C for 40 min and staining the rinsed pistils with 1% Aniline Blue in 0.1 M K_3PO_4 for a minimum of 2 hr. Both fresh and dried pollen of cv. Tamara®, Kordia®, Sweetheart® and 'Lapins' were directly applied onto the pistil, either as dry pollen or in a 15% sucrose carrier.

Onion

In-vitro pollen quality assessment compared fluorochromatic reaction (FCR) and agar-based tests (Brewbaker and Kwack 1963). Agar was selected because of its suitability for in-field testing of pollen tube growth. The sugar concentration of the agar media was refined to optimise onion pollen germination. The use of Tween® 80 in solution to reduce pollen clumping was investigated, including the use of aniline blue (flooded agar plates) for visualising pollen tube growth on media. Cultured plates were left to incubate for 24 hours at room temperature (~20°C) before examination under a UV fluorescent microscope.

In-vivo pollen quality assessment examined pollen tube growth down the style of onion flowers. This involved excising the style and upper section ovaries of sampled flowers. These were tissue cleared using 4M NaOH, heated in oven for 45 minutes at 65°C, and rinsed with deionised water. Then they were stained and examined using Kho and Baer (1968) protocol

Pollen Handling/Purification

Sweet cherry

A range of options were tested for mechanical processing of fresh flowers. In the first trial, bulk quantities of pollen were processed using a standard blender. Although effective at dispersing the pollen, this method significantly reduced pollen viability for some cultivars, regardless of milling duration (5, 10, 20 seconds) and speed (2000 – 3000rpm). Subsequent scanning electron microscope studies revealed distortion of pollen following milling (Figure 22).

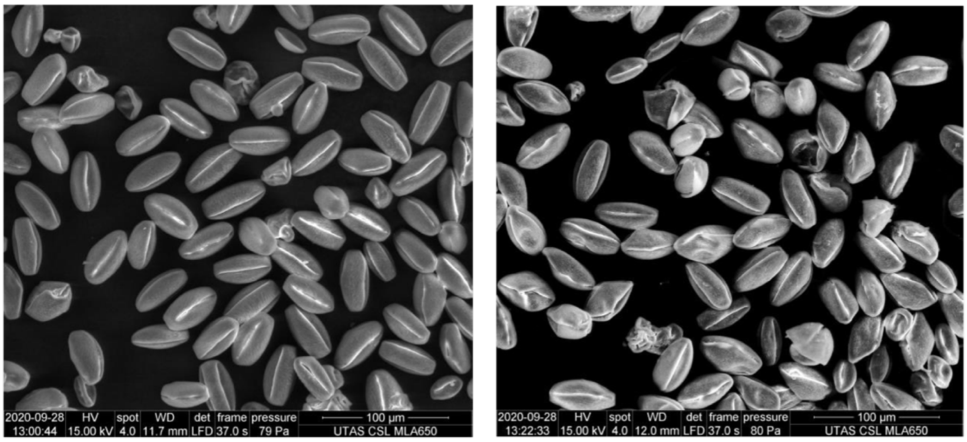


Figure 22. Scanning electron microscope of un-milled pollen and milled pollen of fresh flowers of sweet cherry in 2019.

A second trial used a new mill prototype (based on similar principles to a commercial tri-mill (https://frasergear.co.nz/products/pollen-extraction/tri-mill/) used to collect pollen from kiwifruit flowers. The mill was designed to split the closed flowers open to allow the separation of anthers from the rest of the flower. Recovery of anthers/pollen was improved by milling the same batch of flowers for a total of two times.

The anthers were separated from rest of the flower debris by gentle sieving (2mm then 1mm). This procedure provided clean production of anthers/pollen, which were then dried at 20°C for at least 24hr or until the moisture content of the anther/pollen mix had reduced to around 6%. Once the pollen had been dehisced, pure pollen was obtained by gently brushing the anthers through a 125µm sieve. The pollen was thereafter stored at -20°C for up to 2 weeks prior to application.

Figure 23. Mill prototype developed in 2020 to split closed flower open to separate the anthers from the rest of the flowers (240 V geared electric motor, 173 rvp, 300 mm x 6 blades, 24 'teeth' on the blade and blade is 3 mm wide.

Onion

Pollen harvested using the modified, cyclone-fitted vacuum was relatively pure. However, collected pollen samples were still refined using a 300-micron sieve and a paintbrush to remove inert particles e.g. anthers and dirt. The handling and storage of pollen was assessed. Desiccation of pollen prior to storage was investigated to determine the effect on pollen viability. Desiccation involved storing pollen in a chamber with silica gel for 24hr at 20°C.

The combined effect of storage temperature (20°C, 4°C, -18°C, -80°C) and duration (multiple assessments between 24hr to 12 months) on desiccated and non-desiccated pollen were compared. Furthermore, various lengths of pollen rehydration (0, 15, 30, 60mins) were compared in their effect on pollen germination. Rehydration was performed with pollen on a petri dish in a sealed water bath.

Pollen Delivery

Sweet cherry

Results from the 2020 field trial confirmed it was possible to hand pollinate sweet cherry flowers that had been processed and stored at -20°C for up to 2 weeks. However, the inherently low fruit set (< 15%) of the selected cultivars meant that experimentation at the branch level was insufficient to determine if hand pollination had improved fruit set. In the subsequent seasons, evaluation of mechanical pollination in improving fruit set was undertaken at the tree-level, consistent with previous studies in apple (Webber et al. 2020).

Multiple field trials were conducted on commercial orchards from 2019 to 2024 (

Table 5). Initial trials improved understanding of sweet cherry pollination biology, whilst developing new protocols and processes for collecting, handling and storage of pollen. The latter field trials continued the refinement of protocols, as well investigating the efficacy of pollinating key sweet cherry cultivars (Kordia and Regina) with processed pollen.

The optimised supplementary pollination treatments involved applying 100 mg of purified pollen in 100 mL of 15% sucrose carrier per tree. This pollen was applied twice during flowering (40-50% and 60-70% flower bloom). The pollen was applied with a hand-held positive electrospray instrument, which produced a fine and even mist in low volumes (i.e. 100 mL, Tpshke 2.5 AH Max 12 V).

Onion

A range of pollen carriers, comprising commercially available products, organic and salt-based solutions and dry powders, were assessed for suitability. Suitability was determined by:

- In-vitro germination assessment of pollen suspended in the carrier
- Pollen delivery to the stigma surface in suspension

Comparison of in-vivo germination and seed setting in cytoplasmic male sterile (hybrid seed parent line) or emasculated flowers (open pollinated line) of plants grown in dedicated field trials. Hand pollination and insect pollination treatments were used as controls

For liquid carriers, the effects of adding thickening agents (gum arabic and carboxymethyl cellulose) to maintain pollen in suspension was tested. Effects of pollen-to-pollen carrier ratio and electrostatic charging on pollen delivery to the stigma surface were also assessed.

For all trials liquid pollen suspensions were applied using either a modeler's airbrush or handheld electrostatic sprayer. Powder based pollen suspensions were applied either by hand (brush), in a modified venturi gun or with an electrostatic powder applicator.

Additional complementary experiments

Effect of fungicides on pollen quality

In vitro assays using detached fresh flowers of sweet cherries indicated that pollen viability may be significantly reduced by commercial fungicides (e.g. Delan®) by up to 50%. This impact may be less for closed as compared to open flowers. This work is ongoing by a PhD student.

DNA analysis of S-allele profile of sweet cherries

A range of DNA methods were developed and optimised to allow S-allele analysis of sweet cherry leaves, pollen and embryo including quantitative PCR (qPCR), microsatellite analysis, DNA sequencing and digital PCR. These methods were developed to understand pollen sources and flow in orchards.

3 Project Outcomes

3.1 Project level achievements

KPIs for earlier stages of the project have been met, and reports sent as part of Performance Reports 1, 2 and 3.

Table 2 End of project KPIs

KPI number	KPI description	Achievements against each KPI
Activity 2	Project planning and management	
KPI 3.1	Provide a summary of project planning and management activities implemented	
Activity 3	Communication and extension	
KPI 3.2	Provide a summary of communication and extension activities conducted	See Appendix 104
Activity 5	Research activities	
Activity 5.1	Understand floral biology to inform development of mechanical and bee-vectored pollination to optimise productivity	
KPI 2	Provide update for trials and data analysis for Activity 5.1 (UTas, PFR, NSW DPI)	Refer to p33, p52, p64, p79
Activity 5.2	Improve the performance of honey bee colonies deployed under covers through orchard manipulation	
KPI 1	Provide a progress report on the assessment of the effects of open sides on colony performance/strength. Activity 5.2a (PFR, UoA, UNE)	Refer to p36, p52, p71
KPI 2	Provide a progress report on the assessment of the effect of height above canopy on colony performance/strength for activity 5.2d (PFR, UoA, UNE)	Refer to p41, p58, p71
KPI 3	Provide a progress report for Activity 5.2d (PFR, UNE)	As above
Activity 5.3	Assess the importance of varietal pollen presence and proximity for apple production under cover	
KPI 1	Evaluate and provide a progress report on the importance of multiple pollinisers under net. Activity 5.3b (UoA)	Refer to p44, p**Error! Bookmark not defined.**
KPI 2	Provide a progress report on varietal selections made for mobile polliniser unit. Activity 5.3b (UoA)	As above

KPI 3	Provide a progress report on the assessment of utility of mobile polliniser units for apple under net. Activity 5.3c (UoA)	Refer to p45
KPI 4	Provide a progress report on the Development of MPU. Activity 5.3d (UoA)	Refer to p47
Activity 5.4	Investigate how maintenance of floral resources and other features of the orchard floor affects pollination by bees (native and honey) under nets	
KPI 4	Provide a progress report on the analysis of the importance of non-crop resources and under row management for native bees Activity 5.4a,b,c (UoA, UNE)	Refer to p47

3.2 University of Adelaide

Understand floral biology to inform development of mechanical and bee-vectored pollination to optimise productivity

Assess the relationship between initial colony size and colony size post-flowering in apple orchards (5.1.e)

Changes in worker numbers

Colony checks using the Liebefeld method were analysed for overall differences between placements of hives, comparing those with easy access to outside and hives under closed net. Additionally, the difference between roofs closed with hail and rice net was further explored.

The average number of workers in hives under closed net decreased by 44 workers over three weeks (n = 22 hives). In contrast, the average number of bees in hives with easy access to the outside (n = 32 hives) increased by 1,955 workers (Figure *24*a; F = 3.56, p = 0.05). However, there was a large variation between hives.

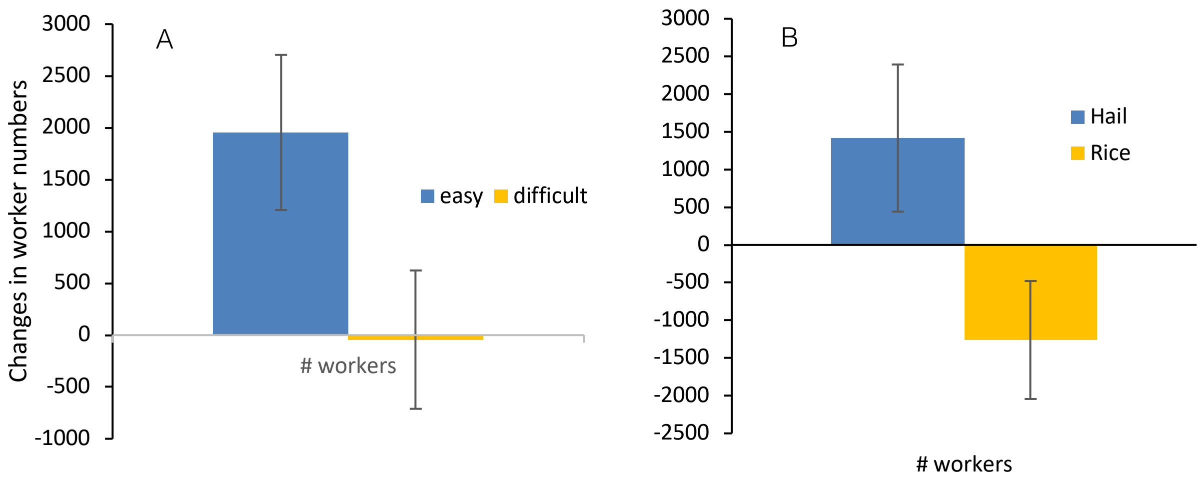


Figure 24. *The changes in number of workers in hives that a) had easy (opened or bird net; n = 32) or difficult (under closed roofs; n = 22) access to the outside of the crop; (b) were placed centrally under closed hail (n = 10) or rice net (n = 12) (n = 12).*

The number of workers declined significantly when hives were placed centrally under areas of closed rice net (an average decrease of 1,261 workers), whereas populations in hives placed centrally under closed hail net increased by an average of 1,417 workers (Figure 24; F = 4.70; p = 0.04).

The difference in the increase in the number of workers between hives placed under closed hail net and outside was not significant (p = 0.23).

The decrease in number of workers under rice netting is likely due to a lack of ability to orient in these conditions, as described in the next pages.

Worker numbers decreased by about 8 % when hives are placed in the middle of closed areas under rice net, whereas under closed hail net they increased slightly.

Changes in brood

Placement of the hives significantly affected the area of capped brood (prepupae and pupae; $F = 9.52$, $p < 0.001$). In hives that had easy access to the outside, the area of capped brood increased by 5.75 dm^2 which, at 400 brood cells/dm^2, amounts to an increase of 2,300 brood cells. The area of open brood (developing larvae) in these hives also increased (Figure 25). In contrast, when hives were placed under a closed roof, the average area of capped brood declined by an average of 4,620 brood cells (Figure 25a; $F = 15.42$, $p < 0.001$), while the area of open brood remained stable. In the case of open brood, the difference between hives with easy and difficult access to outside was not significant ($F = 0.77$, $p = 0.39$).

The area of capped brood generally decreased when hives were placed under closed roofs. While closed brood decreased more under rice than under hail net (Figure 25), this was not a significant difference ($F = 1.72$; $p = 0.20$). Overall, net type (rice or hail) did not seem to affect the amount of open brood.

These decrease in capped brood is consistent with a decline in the number of workers under rice net.

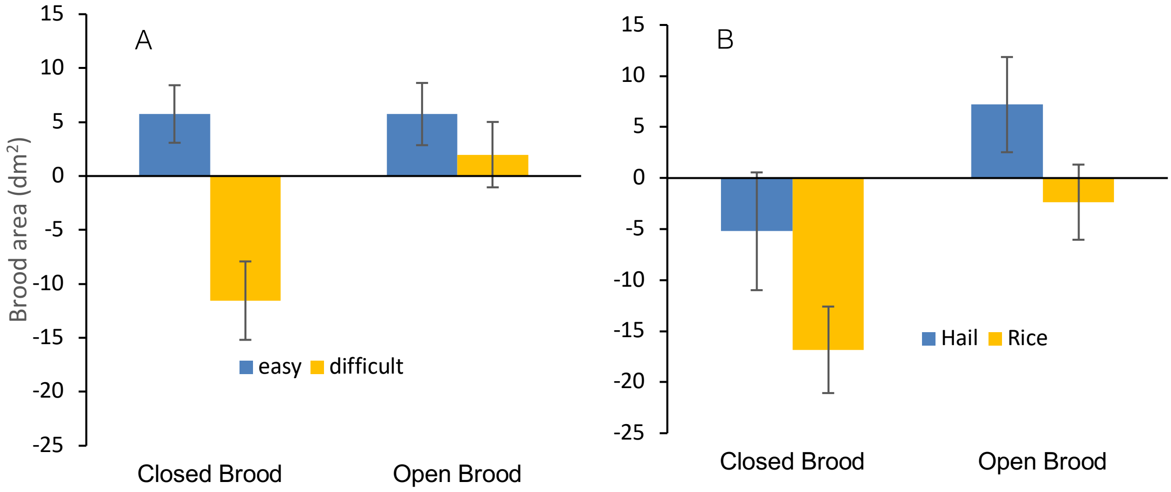


Figure 25. The changes in area of capped brood and open brood (dm^2) in hives that (a) had easy (opened or bird net; n = 32) or difficult (closed net; n = 22) access to the outside of the crop; (b) were placed centrally under closed hail (n = 10) or rice net (n = 12)

Hive health, based on the number of capped brood, declined when access to the outside of the crop was difficult.
The most impact was observed under fully closed rice net.
There was less impact on open brood (eggs and developing larvae)

Stored food

The area of stored pollen and honey decreased more in the hives that had easy access to the outside compared to those placed in the middle under closed roofs (F = 8.31, p = 0.006, and F = 7.95, p = 0.007 for pollen and honey respectively, Figure *26*), while there was no significant difference in nectar storage.

Pollen and honey stores remained similar under rice net but were reduced in hives that were placed in the open or under hail net. It is possible that the reduction in stored food in the latter colonies was related to the higher number of brood cells produced, as well as differences in incoming food.

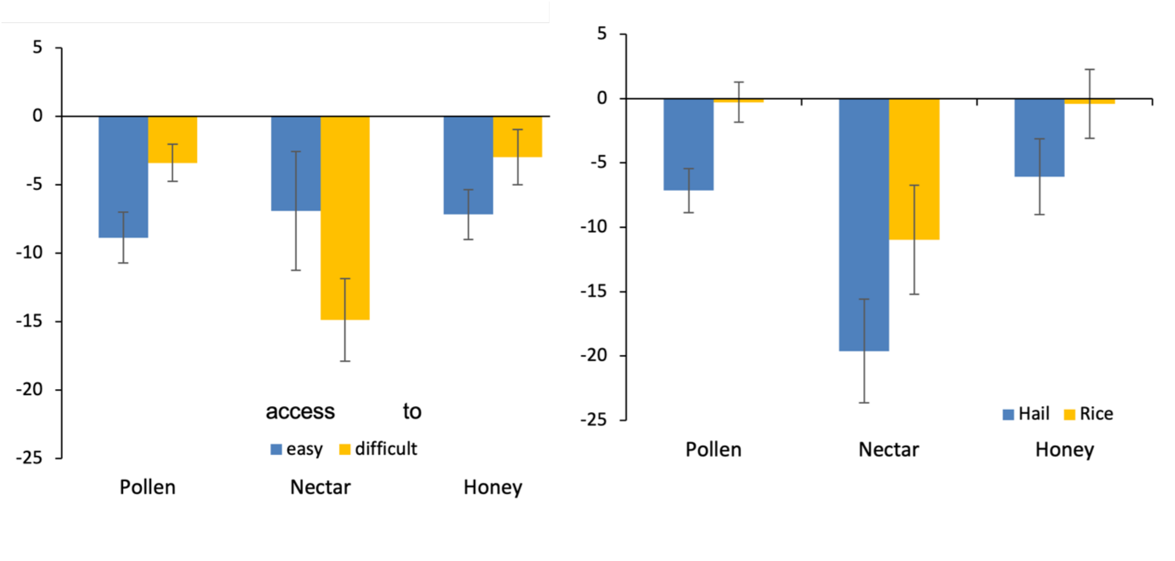


Figure 26. The change in the area (dm^2) of stored resources in hives *(a) had easy (opened or bird net; n = 32) or difficult (closed net; n = 22) access to the outside of the crop; (b) were placed centrally under closed hail (n = 10) or rice net (n = 12).*

The amount of stored food declined in all hives, even those with easy access to the outside, with the greatest changes in hives that had the most brood cells.

Improve the performance of honey bee colonies deployed under cover through orchard manipulation

Determine whether opening of sides improves pollination in covered apple orchards (5.2.a)

To examine whether opening rice net above the inter-row, to allow light in, would benefit bee activity and apple production, we opened net above individual rows in two orchards. The growers left every fourth (orange) and third (blue) row opened (Figure 27). We assessed how this affected bee activity, fruit set (applets per flower cluster on120 trees), seed number (seeds per apple) and fruit symmetry (120 apples per orchard), in rows under opened and closed rice net in 15 rows per orchard.

There was significantly more bee activity ($F = 9.13$, $p = 0.003$) under opened rows than under closed rows. Fruit set was higher under opened rows compared to closed, but not significantly so ($F = 4.80$, $p = 0.27$). After thinning, the seed number in apples under opened rows was significantly higher in one orchard ($F = 9.13$, $p = 0.003$), but not in the other, and there was no significant difference in apple symmetry between opened and closed rows for either orchard.

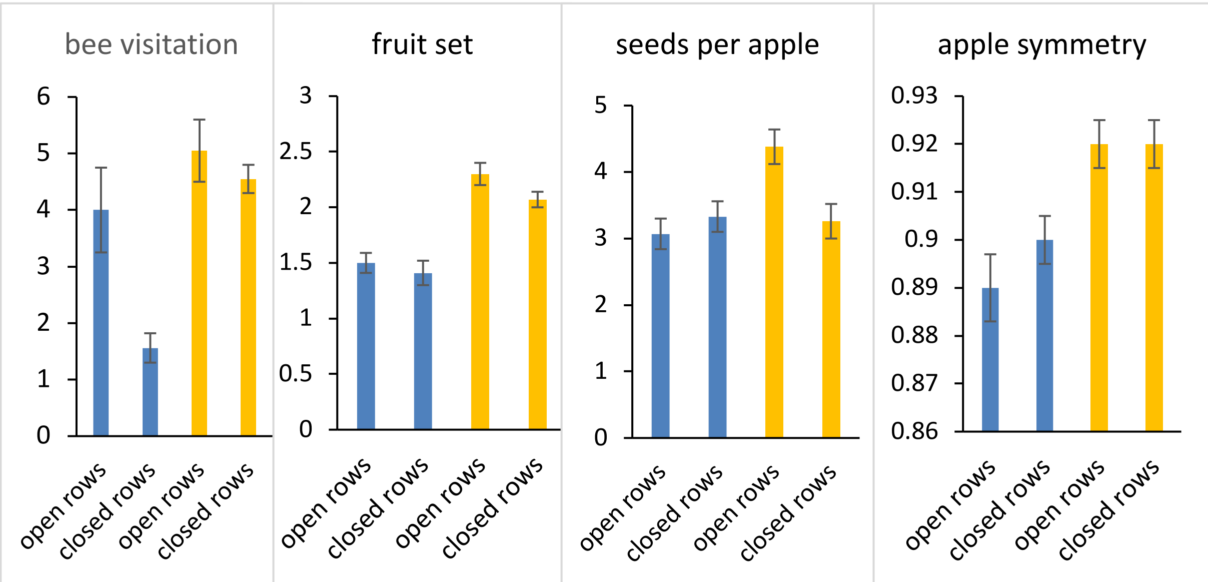


Figure 27. Bee visitation, fruit set, seeds per apple and apple symmetry under rice net, where rows that where either open or closed during pollination at two orchards (blue and orange). The roof was open over every third (blue) or fourth (orange) row.

These results are comparable to results of Murphy-White (2019) for apple, and Evans et al. (2019) for kiwifruit, where decreased bee activity in netted orchard blocks did not lead to decreased seed set, quality or pack out.

Given that opening and closing the rice nets requires special machinery and handling time at a cost of approximately $1,200 per hectare, and may lead to increased wear and tear on the nets, as well as increased risk of hail damage, our advice to growers is to leave rice nets closed, and we advise the beekeeper to provide the hive with ample resources for the relatively short period under net for apple pollination (see the following pages).

Although opening the net over every 3 – 4 rows increases bee activity on the crop, it did not lead to higher fruit set or better apple symmetry.

Determine whether the height of netting above the canopy affects colony performance or strength in covered apples (5.2.c)

As reported previously (p36) hive health declined in hives placed under roofs closed with rice net compared to those placed under roofs closed with hail net or outside. In the following section, the reasons for these differences are explored by investigating:

- The attrition due to bees getting stuck in the nets
- Orientation by bees under net
- Pollen collection by bees under net

Attrition due to bees getting stuck in nets

The count of dead bees involved walking through the rows, looking upwards, and recording bees stuck in the net. The count was conducted across entire orchard blocks and calculated per square meter. The counting process was carried out in five orchard blocks with roofs closed with hail nets and six with roofs closed with rice nets. The rice nets had cross bungees, except for one with a lighter weave and straight bungies, under which no hive was placed.

Few bees died due to being trapped in the rice net with straight bungees. However, this could also be attributed to the absence of a hive directly beneath this type of net, and the fact that the covered area was narrow with open sides.

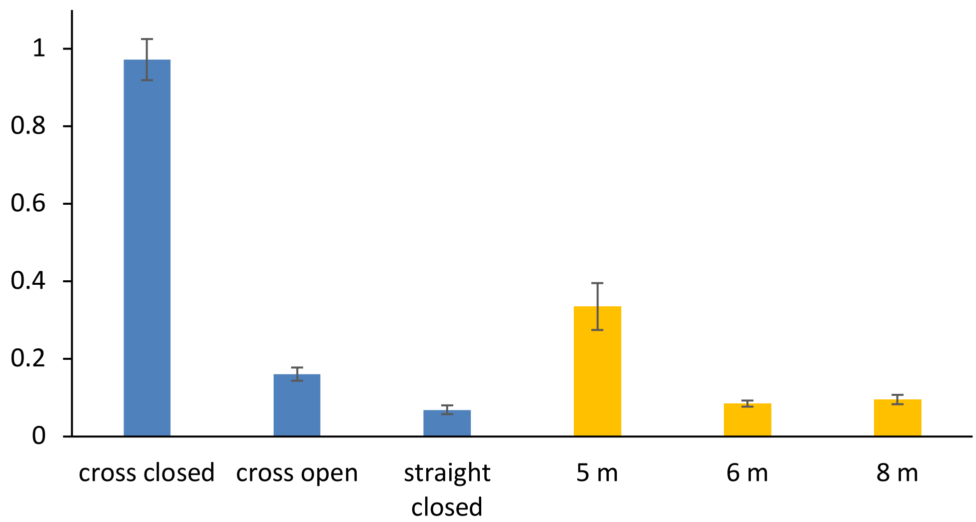


Figure 28. The number of bees stuck per m² in rice net (blue) and hail net (orange) after three weeks of pollination.

Under hail net, there was less attrition (bees stuck in the nets) when nets were >5 m high than under nets that were lower. Under rice net with cross bungees, the attrition was significantly lower when the roof was opened every fourth row.

Observations of bee flight behaviour indicated that many bees flew out of the netted area through the opened sides. Placing the hive in the centre under the net, where bees had to travel a relatively long distance (50 – 100m) to exit, likely resulted in many foragers not finding their way back, especially shortly after placement of the hive under the net. This is because, on their return flight, the bees fly in a straight line to the hive, inadvertently ending up above the hive, on top of the net. They may then attempt to find an opening or to crawl through the mesh that is too small.

The higher hail net provides space between the trees and the roof, and, compared to lower nets, this may offer better opportunities for orientation, allowing bees to return under the net. This may explain why attrition was lower under the high hail net and opened rice net (Figure 28). However, birds were observed landing on the top of hail nets and removing dead bees, a scenario not observed in the folds of the floppy rice net. Consequently, attrition under the hail net may be underestimated.

In the Adelaide Hills, while all nets had open sides, some nets had an overhang along the sides lower than the roof of the net. This presented a significant challenge for bees attempting to leave from under the net. Particularly in the late afternoon, many bees were observed in these corners, struggling to exit. Although not quantified, this aspect should be considered in net design, as there appears to be no evident need for this overhang.

Bees can easily become trapped in the corners if nets have an overhang on the sides, compared to presenting a clean exit with no side panels

Orientation by bees under net

Apart from getting stuck in the nets, it is also likely that the light and lack of height above the trees prevents the bees from orienting and finding their way around the crop and back to the hive. To investigate this, foraging patterns of groups of bees in an outside orchard, under hail net and under rice net, were investigated.

On each of four spread-out stretches per orchard, 2m in length, 50 bees were marked. The orchards included one that was open, another covered with closed hail, and a third with closed rice net. Subsequently, bees were observed visiting the same stretches for 10 minutes later in the day (at least more than 1 hour later) and on two consecutive days, to quantify the proportion of marked bees returning to the same spot.

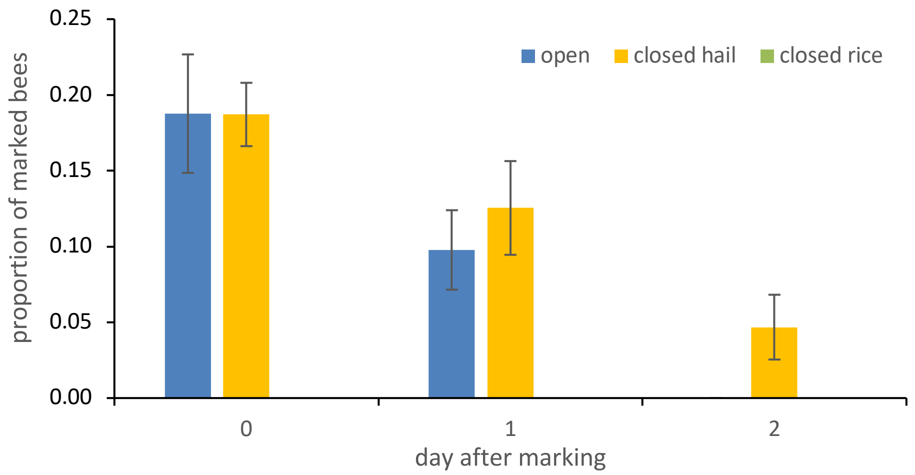


Figure 29. Proportion of marked bees observed at the spot where they were marked, in the open (blue), under closed hail net (red) and under closed rice net (not shown, because no marked bees were observed).

On the day of marking, the proportion of marked bees observed foraging in the proximity of the spot where they had been marked was ~0.18 for bees in the open orchard, and under roofs closed with hail net (Figure 29).

Under roofs closed with rice net, no marked bees were observed to revisit the spot where they had been marked. A day after marking, the proportion of marked bees foraging in the same areas was ~0.09 in the open and under hail net, while again, no marked bees were observed at any of the four areas under rice net. This indicates that bees lack the ability to orient under rice net.

Bees find it difficult to orient under closed rice net, likely due to interruption of the light and lack of height above the trees

Pollen collection by bees under nets

Similar to the effects on hive health, no significant difference was found in pollen collecting behaviour between hives placed under open edges or bird net, and those placed outside. Therefore, the hive placement data was categorised into two classes: hives with either easy or difficult access to the outside. In addition, after checking that there was no effect of year, and no interaction between year and placement on any of the variables, data from the two years was combined.

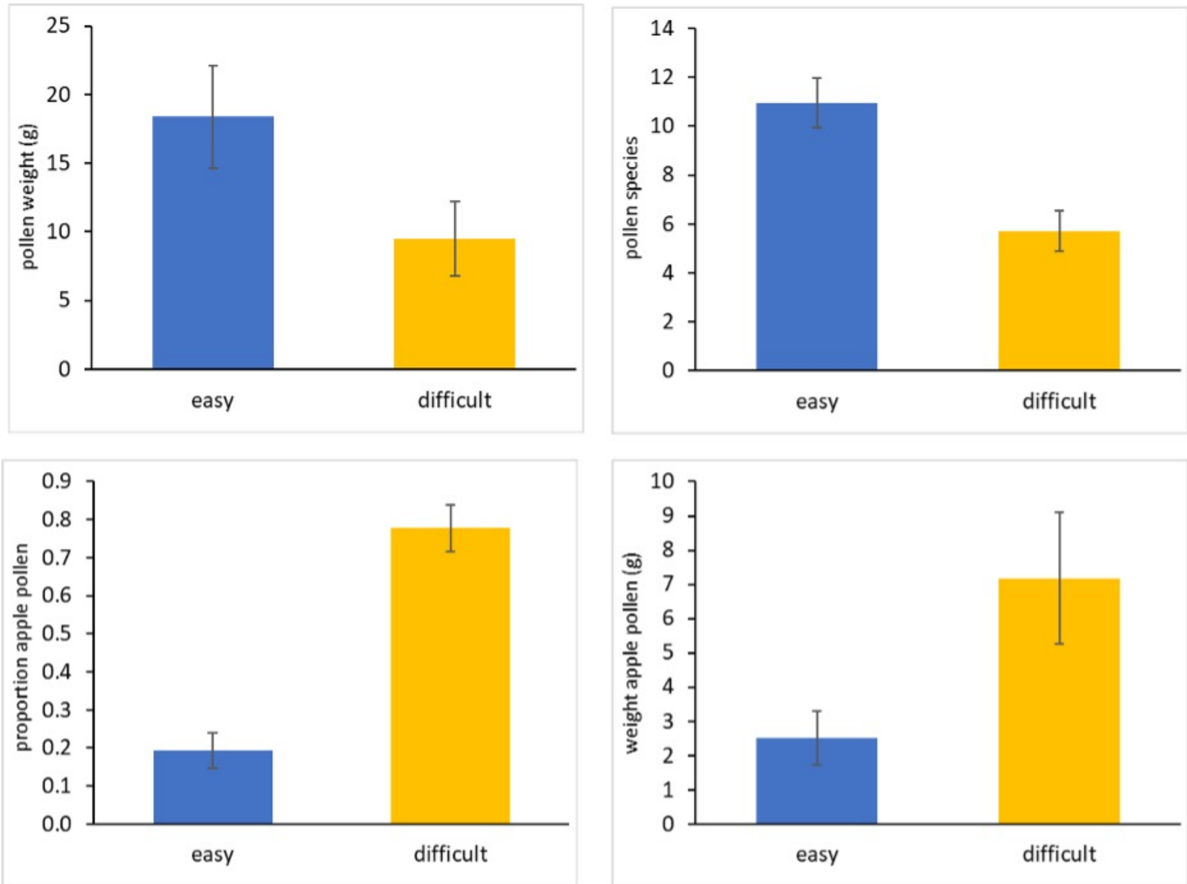


Figure 30. The total weight of pollen collected (a) the number of pollen species collected (b), the weight of apple pollen collected (c) and the proportion of pollen that was apple pollen (d), by hives with easy (blue) and difficult access to foraging outside.

The weight of pollen collected was lower when the hives were placed in the centre under a closed roof, than when access to the outside was relatively easy, but not significantly so ($t = 1.91$; $p = 0.07$; Figure 30a).

Compared to hives with easy access to the outside, hives under a closed roof collected pollen from about half the number of plant species ($F = 11.88$, $p = 0.002$; Figure 30b).

To assess which placement would result in the most apple pollen collected, and hence, the most apple pollination, a representative sample was taken, pollen collected per hive mixed, and the number of apple pollen kernels in 400 kernels per sample counted. The pollen from hives with access to the outside contained on average 19% apple pollen, while the pollen collected by hives with limited access to outside contained 78% apple pollen (Figure 30c).

Assuming that the weight of pollen species was proportional to the relative number of pollen kernels in the sample, it can be concluded that, while hives with limited access to the outside collect less pollen, they collect nearly three times as much apple pollen than hives with easy access to the outside (Figure 30d).

As opposed to foragers with easy access to outside, bees from hives under closed roofs mostly focus on collecting pollen, and probably also nectar, from apple flowers. The increase in visitation to apple flowers may partly compensate for the negative effects on the number of foragers, foraging activity, and hive size.

Assess how different types of covers and nettings affect light transmittance, honey bee activity (5.2.d)

It was near impossible to collect meaningful data about how the net influenced bee activity, as the orchards differed in block sizes, tree varieties, size and pruning, flower densities, covers used, height of covers, hive stocking rates, hive sizes, and placement of hives. However, in one orchard, hail netting was damaged in 2023 and had been removed over a length of 90 m and four rows wide, in a large block.

This offered an opportunity to investigate the activity of bees in the open and under high hail net in the same orchard (same stocking rate), on the same apple variety (Pink Lady), at the same distances to the hive and under the same weather conditions at different times of day (Figure 31).

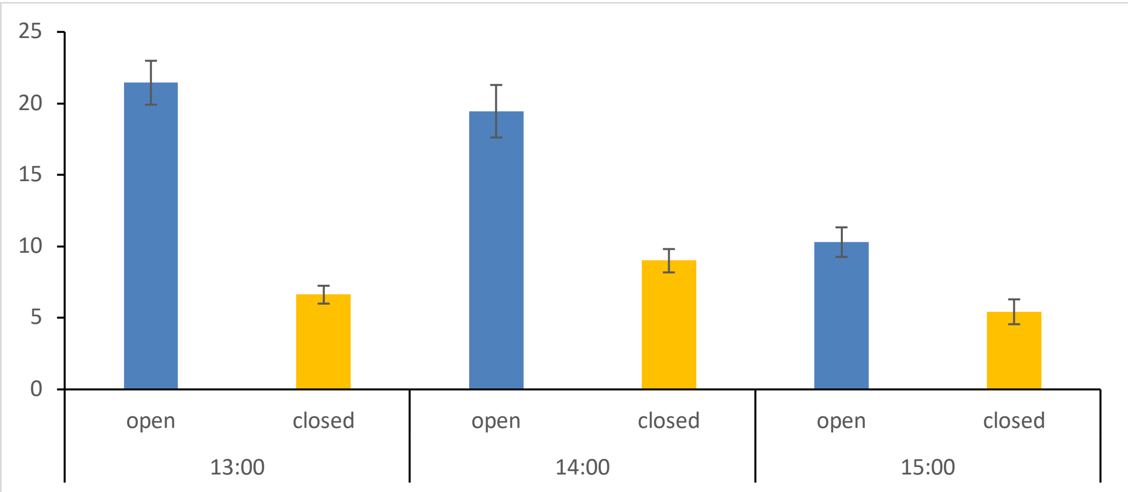


Figure 31. Bee activity under open rows compared to under closed hail net in the same orchard at 1pm, 2pm and 3pm.

On average, there were 2.5 times more bees active on the trees that were not covered by hail net than on the trees that were covered by hail net (Figure 31). This result is consistent with the findings of Murphy-White (2017) for apple, and Evans et al. (2019) for kiwi fruit.

Other factors being equal, the presence of high hail net more than halved bee activity on apples compared to bees foraging in the open. It is likely that this is related to changes in the quality and quantity of light under the net.

Summary of the findings regarding bee health and pollination services under net

Hive health decreased substantially when placed under roofs closed with rice net. This is likely due to many foragers not being able to find their way back to the hive. This is deduced from:

- The large number of workers stuck in nets
- Reduction in the numbers of active bees
- Reductions in closed brood
- Lack of the ability of foragers to return to the spot where they were foraging earlier

It seems likely that low netting does not allow bees to orient well in relation to the hive. There may also be effects of differences in the quality and quantity of light, but we did not research this. However, light also seemed to profoundly affect foraging activity of bees under hail net, which was much reduced compared to out in the open.

In hives placed under roofs closed with hail or rice net, total pollen collection was reduced (both amount and species of pollen). However, it seems unlikely that this caused reduced hive health, because (a) it was similar for hives under closed hail and rice net, whereas declines were higher under rice net, and (b) when a hive is well stocked, which most hives were, the effects of reduced foraging returns on hive strength and brood production would be delayed and not observed during the ~3 weeks of apple pollination.

Under closed rice and hail net, the bees largely focused on collecting pollen from apple flowers, and this may benefit apple pollination, and counteract the effects of lowered foraging activity.

Assess the importance of varietal pollen presence and proximity for apple production under cover

Assess the diversity of varietal pollen carried by honey bees on bodies under net and outside using molecular markers (5.3.a)

Direct assessment of the varietal pollen carried by honey bees under net and in the open was not feasible, due to both methodological difficulties of assessing the varietal composition of pollen carried by bees, as well as the costs involved to analyse a meaningful number of samples.

Instead, it was decided to identify

a. The importance of proximity of pollinisers for fruit set
b. The minimal distance over which pollen varieties were successfully deposited on stigmas.

Both were carried out in the same single large orchard block of Pink Lady (25 consecutive of 200 m long rows) covered entirely by rice net. The net in each row was supported by 15 – 17 posts; the distance between two posts (15m) is referred to as a ‘panel’.

Set was assessed over two years by counting the applets per flower clusters on two branches per tree, a high and a low branch, of two trees per panel, throughout the large Pink Lady block (Figure 32), just after flowering. This was possible because the grower did not chemically thin after flowering.

The numbers of applets per cluster varied depending on the position in the orchard (Figure 32).

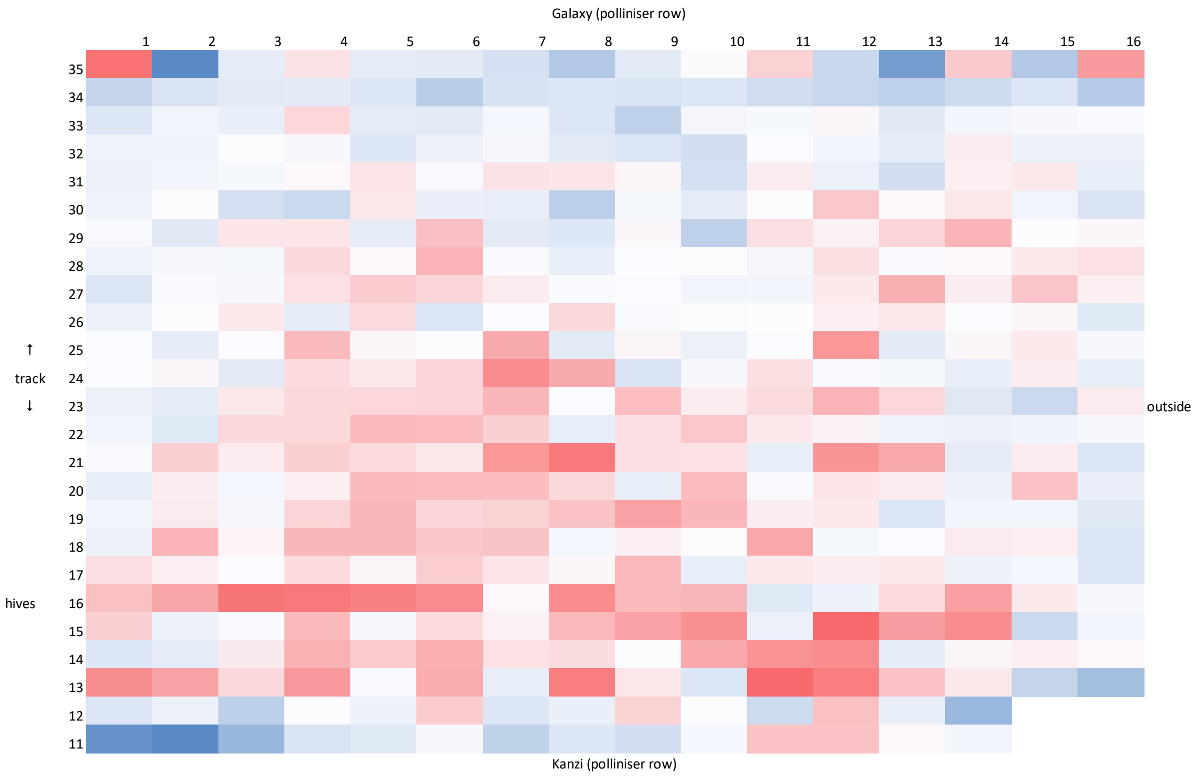


Figure 32. Heat map of fruit set (apples per cluster, apc) relative to position in the orchard. Panel number is given horizontally, row number vertically. Red and pink: 0.8 – 1.7 apc; white: 1.7 – 2 apc; blue: 2 – 4.5 apc.

The number of applets per cluster was significantly higher for the first two rows next to a polliniser row ('Galaxy' on the one and Kanzi® on the other side; Figure 32; Figure 33). Interestingly, the fruit set next to the 'Galaxy' was higher than the set next to Kanzi. It is possible that this was caused by higher visitation due to closer proximity to the outside. There were only three rows of 'Galaxy' between the Pink Lady and the end of the netting, while on the other side of the orchard, there were 10 rows of Kanzi before the net ended.

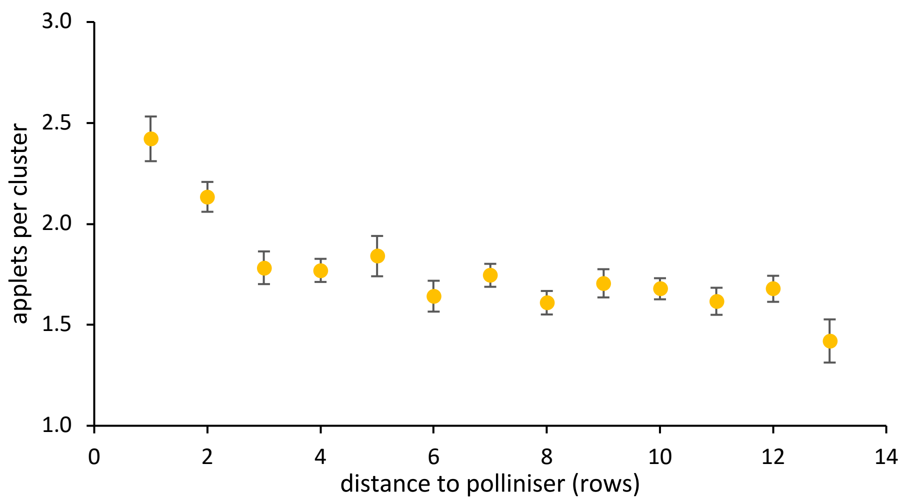


Figure 33. Applets per cluster relative to the distance to polliniser rows. Rows were 4.5 m apart

For the rows that were further removed from the Kanzi and ‘Galaxy’ polliniser rows and the outside (i.e., rows 31 – 13; Figure 32), the flowers in the panels closer to the track and to the outside were better pollinated than those further towards the middle of the rows (Figure 34).

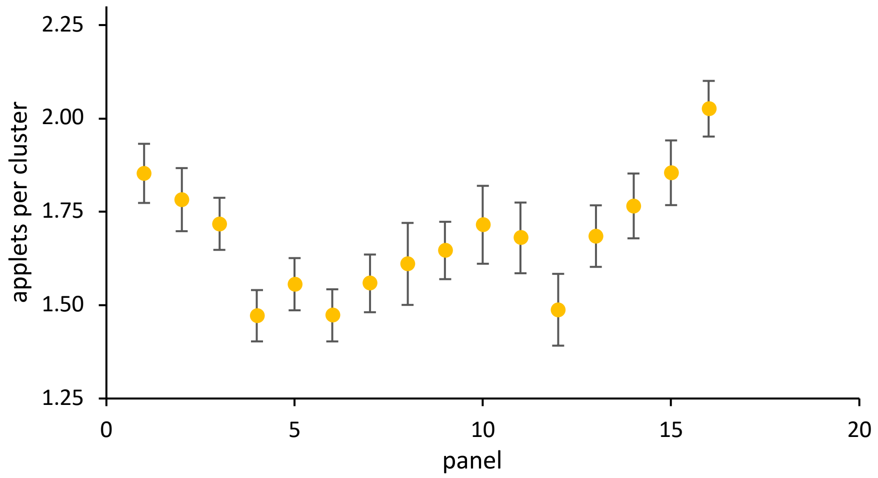


Figure 34. Average number of apples per cluster in rows that were not close to polliniser rows (i.e. rows 13 - 31).

This is likely to be the result of higher bee activity closer to the edges. In 2021 and 2022, bee activity was documented along the treatment and control of the mobile polliniser trial, and in 2022, this was quantified for an additional six rows in the centre of the Pink Lady block.

This again shows that bee activity is consistently higher at the edge of the row (Figure 35). Overall, bee activity was lower in 2022 than in 2021, which is probably because in 2022, the hive numbers on the track were reduced, and some hives were less active. The activity on the last four panels closer to the edge was higher in 2022. This was because in 2022, hives had been placed outside the net, next to panel 16 at row 19, while in 2021, hives were only present on the track close to panel 1.

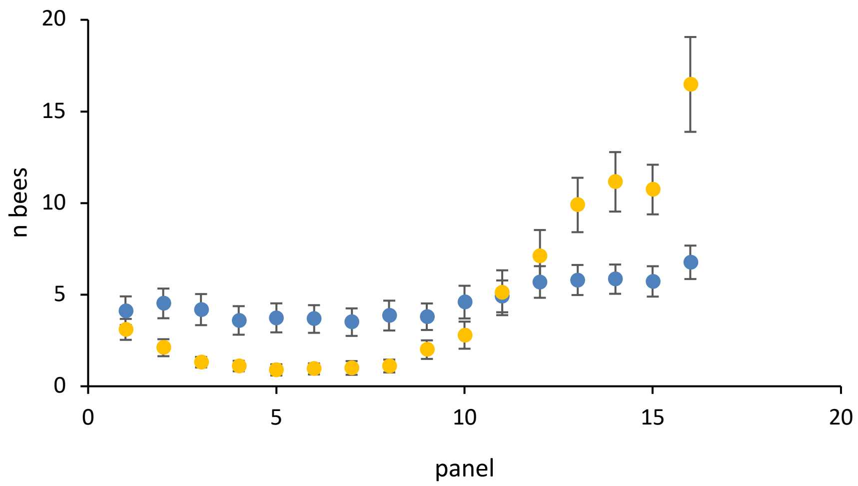


Figure 35. Average number of bees observed per panel stretching 15 m of flowering apple trees, +/- s.e during 2021 and 2022.

The effect of hive placement outside the net could be seen up to five panels (75m) into the crop, whereas the effect of hive placement on a covered track, while less prominent, led to increased activity along most of the row. This indicates that it is beneficial for bee activity on the crop to place the bees under the net.

Bee activity is higher close to the edge of the nets. When sides are open, the effect of proximity to the outside on bee activity can be seen approximately 75 m along the row, from the outside.

Placement of hives in the middle of rows under rice net provides a better overall distribution of bees in the orchard.

The importance of proximity of pollinisers

A symmetrical apple fetches a higher price, and there is a strong correlation between seed number and apple symmetry. The data above show that proximity of pollinisers is important for fruit set, but whether this reflected the paternity of the seeds warranted further investigation.

For 170 seeds, the closest possible pollen donor has been identified. In total, the closest possible pollen donor for 73.5% of seeds was within 10 m of the flower (

Figure 36). This does not necessarily imply that the pollen was donated by the closest donor, but it demonstrates the significance of the proximity of the pollinisers.

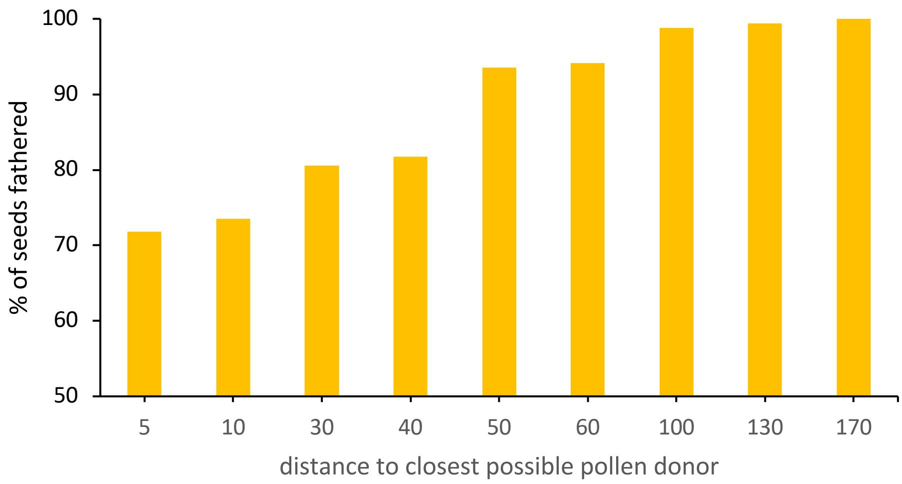


Figure 36.The distance to the closest possible pollen donor in relation to the cumulative % of seed donors.

Nearly all pollen donors could be found within 50m of the pollinated flowers, with less than 6% of seeds fathered by pollen donors more than 60m away. This result underscores the importance of the proximity of pollinisers, is consistent with the findings regarding fruit set in relation to distance from the polliniser row (Figure 33), and with observations of bee behaviour (Free, 1962) which all indicate that individual bees rarely move further than 50 m in the crop.

The observations presented in the above section can be translated into design advice for orchards, as follows:

1. If placing hives on the outside areas of net, avoid rows longer than 100m; most bees will not venture further than ~50 m under net
2. Ensure that one or more rows of pollinisers are present every six rows under net
3. High hail net is better for hive health and encourages bees to forage more on the crop compared to low rice net
4. Placing hives centrally under the net, rather than outside or on the edge, forces the bees to forage on apples; increased presence of apple pollen may counterbalance the negative effects of central hive placement on bee vigour and foraging activity

Experimentally assess the importance of paternal variety for fruit set and quality (5.3.b)

Hand pollination trials were carried out using different pollen sources to pollinate Pink Lady flowers. Initially, this led to differences in fruit set, but not in apple quality. However, later experiments indicated that these differences were likely related to the quality of the pollen, which is greatly affected by presence of moisture during harvest as well as the method and period over which pollen is allowed to dry.

Further literature studies indicated that genetic compatibility (i.e., different alleles on the S-locus) is the only factor that influences seed set. Because this has been well documented, and S-alleles are known for all varieties, no further experiments were performed to identify the optimal paternal variety for use in the mobile polliniser units (MPU).

Experimentally assess the utility of MPUs for fruit set (5.3.c)

Initial observations suggested that visitation to the pollinisers was influenced by the shape of the display. This was investigated by testing two displays:

a. A presumably denser and visually more attractive clustered presentation
b. A wide presentation, with placement of the pollinisers perpendicular to the direction of the rows (Figure 37).

With the same number of polliniser trees, the wide presentation attracted significantly more bees ($p < 0.05$; Figure 38).

Moreover, after spending time on the clustered display, bees often flew up and left the area, possibly returning to the hive. This would not achieve cross-pollination. In contrast, the bees that visited the wide presentation largely used the MPU to cross to the flowers in the neighbouring row (Figure 37). Therefore, the wide presentation showed more potential for cross pollination.

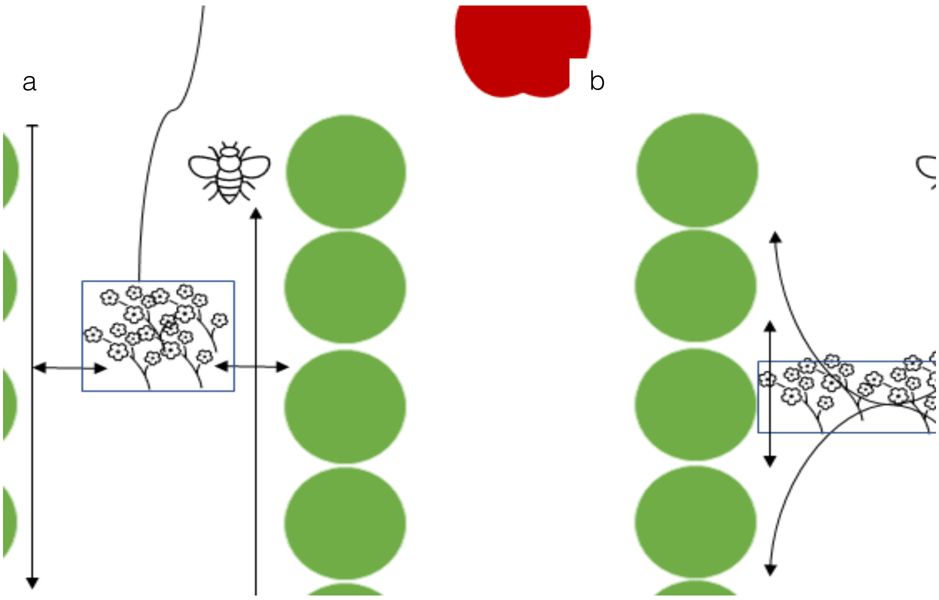


Figure 37. Two polliniser arrangements: a) cluster and b) wide presentation. The movement patterns of bees along rows and to pollinisers on the MPU are represented by arrows.

The movement of the MPU down the row would also likely extend these benefits to all segments of the orchard, as opposed to the local area where bees forage before returning to the hive. The MPU thus acts more like a bridge between apple rows than a pollination unit, where each use results in an encounter with a polliniser, then an apple flower. Bees are also incentivised to use the polliniser bridge because there are denser resources visible on the tray unit than one a single tree directly adjacent.

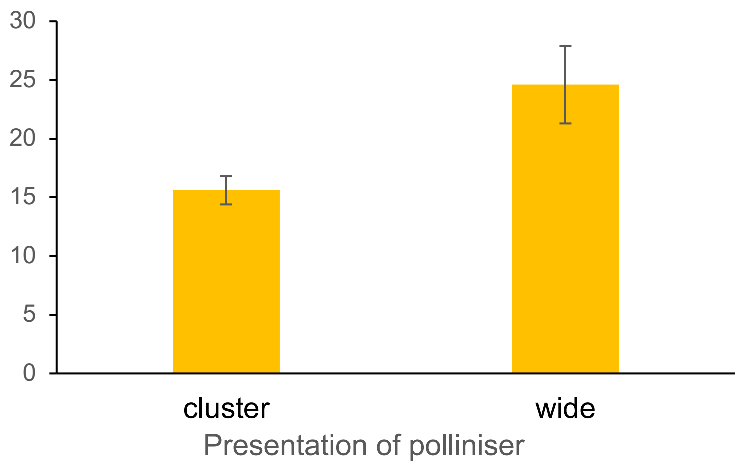


Figure 38. The average number of honey bees attracted per 10 minutes to presentations of pollinisers that were either clustered or wide (see Figure 37)

Bee activity within rows

To check whether any potential difference in set between the MPU and the control rows was caused by a higher background activity of bees in the MPU treatment rows, or whether the presence of the MPU affected bee activity in the row, instantaneous bee activity was assessed as the number of bees observed foraging on flowers per panel (15m), while walking at a steady pace.

While there were relatively large numbers of bees, only bees observed interacting with a flower during a survey encounter were counted to avoid pseudo-replication. Activity surveys were conducted three times per day.

Bee activity (number of bees per row) varied per panel but was not significantly different between the treatment and control rows (control: 14.1 ± 0.47 [mean ± s.], n = 786, treatment: 15.4 ± 0.86 n = 256, $p = 0.18$).

Bee visits to the MPU

Unsurprisingly, bee activity on rows was positively correlated with crossings between the MPU and rows ($p < 0.001$). All activity was also positively correlated with increase in flowering, higher temperatures, and fewer clouds, although p-values varied with different models.

Fruit set and parentage

Fruit and seed set after bees transitioned from the flowers on the MPU to the flowers in the adjacent rows was compared to fruit set after transitions from flower to flower in control rows.

Fruit set (number of apples per visited flower) was significantly higher after transition from the MPU than after transition from flowers in the row (treatment: 0.56 ± 0.04, n = 154, [mean ± standard error] vs. control: 0.2 ± 0.024, n = 260, $p < 0.0001$). Control/treatment rows were the only significant variable in the model, which included row panel and flowers tagged as visited by either pollen or nectar collecting bees. Thus, a transition from the MPU to flowers in the row more than doubled the fruit set. However, the apples in the MPU row did not contain more seeds than those in control rows.

While transition from the MPU to flowers in the row caused high fruit set, there was no difference between control and treatment rows in fruit set (measured in applets per cluster; $X^2= 0.30$, $p = 0.59$). This is because there were not enough transitions between the MPU and the rows to make an overall difference. While there was an effect of year on fruit set ($X^2=18.49$, $p<0.001$), there was no interaction between year and treatment. Hence, overall, there was no benefit of the MPU in terms of fruit set (Figure 39).

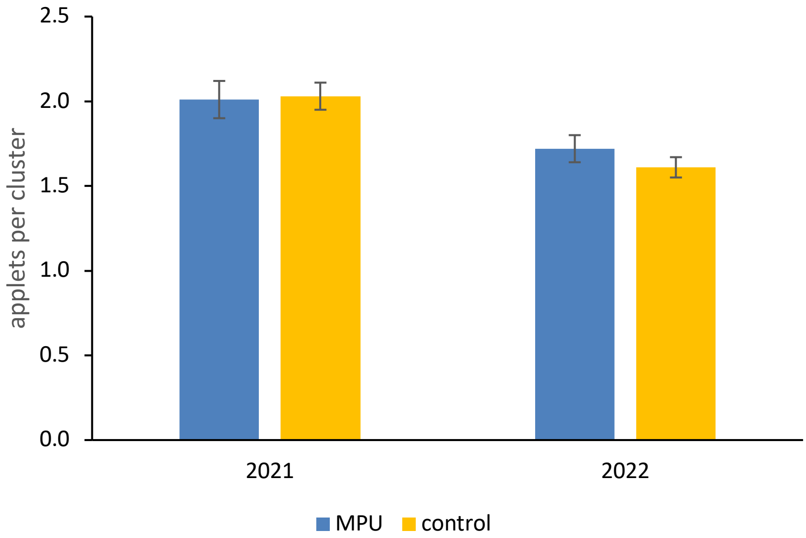


Figure 39. Average fruit set in mobile polliniser and control rows in 2021 and 2022.

Although there was no effect on average fruit set, strong differences in fruit set between panels were observed in relation to their distance to the edge (Figure 34). It is possible that benefits of the MPU are present in locations in the orchard where fruit set is naturally lower.

To explore this, fruit set (below or above average), found in the MPU panels and the control panels was classified for each year and each panel. A matched-pairs t-test was then employed to investigate whether MPU panels had a higher average fruit set than control panels. This was not the case ($t = 0.90$, $p = 0.38$).

Overall, permanent presence of MPUs may somewhat increase the set, but maintaining MPUs is costly and labour intensive. The trees need to be maintained in pots, watered regularly, and strategies need to be developed to ensure synchronous flowering. In addition, the costs involved in operating several robots in the orchard are substantial. Therefore, the costs of MPUs are highly unlikely to outweigh the benefits. One might consider adding or grafting some pollinisers in areas with low bee visitation and fruit set.

Use the information generated to develop mobile polliniser units (5.3.d)

Because there was no indication that the use of mobile polliniser units would be cost effective, we did not pursue this.

At high density, mobile pollinator units have the potential to increase fruit set in areas where set is low. However, the labour and cost involved are prohibitive, and growers are better off either adding a hive, or a polliniser row, to such areas.

Investigate how non-crop floral resources and other features of the orchard floor affect pollinators under protected covers

Investigate wild pollinator diversity and density in relation to under row and inter-row management (5.4.a)

A total of 14 orchard blocks from eight properties were evaluated for the factors that may influence diversity and density of apple flower visitors during the flowering period.

Within a single row of each orchard block, 15 trees were selected and all floral visitors over a 1 min period were observed. The floor beneath the trees of each orchard block was classified for two factors: i) the vegetation cover, either vegetated or bare, and ii) the shape of the orchard floor, either mounded or flat. Both factors are believed to influence the available nesting substrate for groups such as ground-nesting bees, a common visitor to flowering apple trees, which show preference for clear and well-drained soils.

Across the 14 blocks, there was great variability in average number of wild-flower visiting insects and non-Apis bees present (Figure 40).

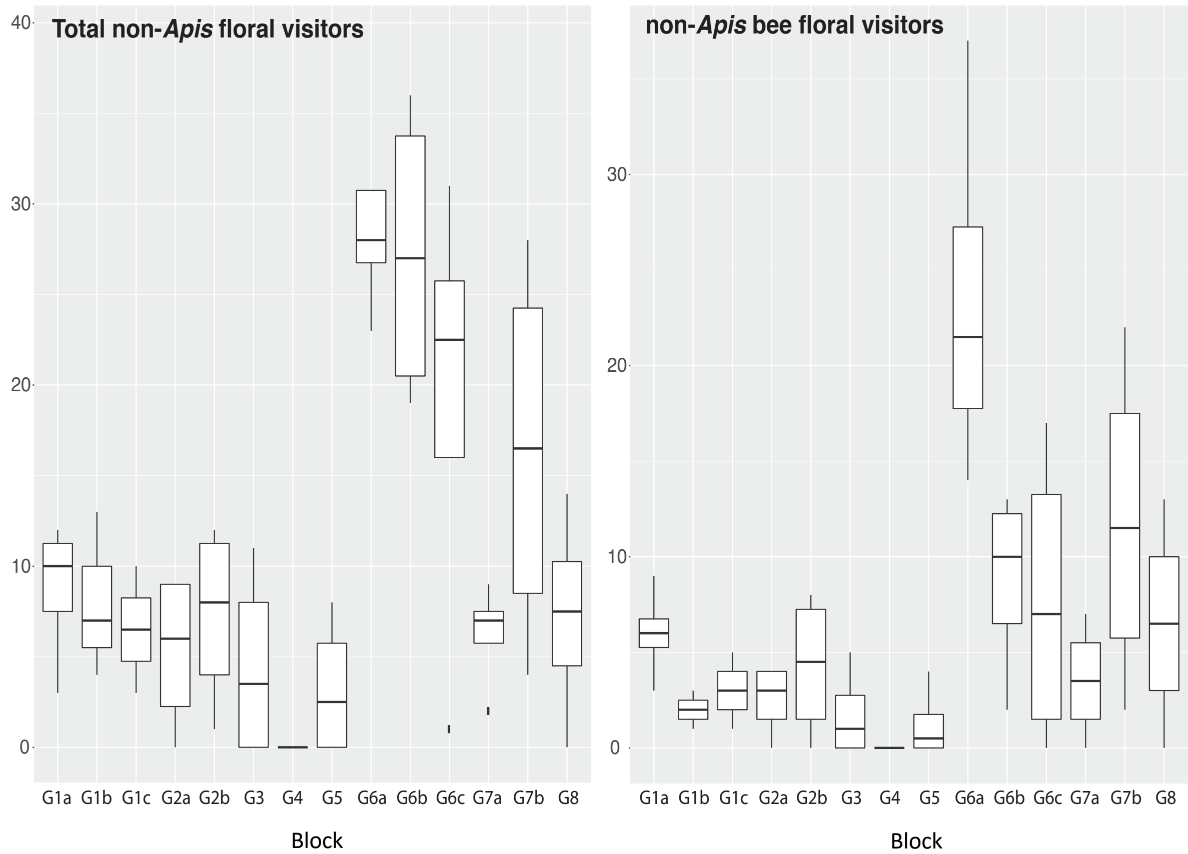


Figure 40. Box plot comparison of a) non-Apis insect visitors per orchard row and b) non-Apis bees per orchard row for our 14 blocks from 8 growers. Boxplots labelled as Grower 1-8 (G1-8) with multiple blocks per grower indicated by lower case letters (i.e. G1a-c)

Two generalised linear models with Poisson distributions were used to assess the effect of tree floor management on the presence of wild insects and non-Apis bees visiting apple flowers. In both models the importance of under tree cover, floor shape, temperature, and the mean number of flowers per tree were evaluated as predictors of wild insect or non-Apis bee presence.

Under tree floor, temperature and the number of flowers were found to all have a significant effect ($z = 4.8$, 7.9, 5.7, $p < 0.05$, respectively) on the number of wild insects present per orchard row. Post-hoc comparison of under tree cover (Figure 41) showed that bare floors under the trees had significantly higher (z ratio: 4.8, $p < 0.05$) numbers of wild insects (model estimated means – bare: $\mu = 10.67$ (CI: 9.53 - 11.94), vegetated: $\mu = 5.78$ (CI: 4.49 - 7.45)).

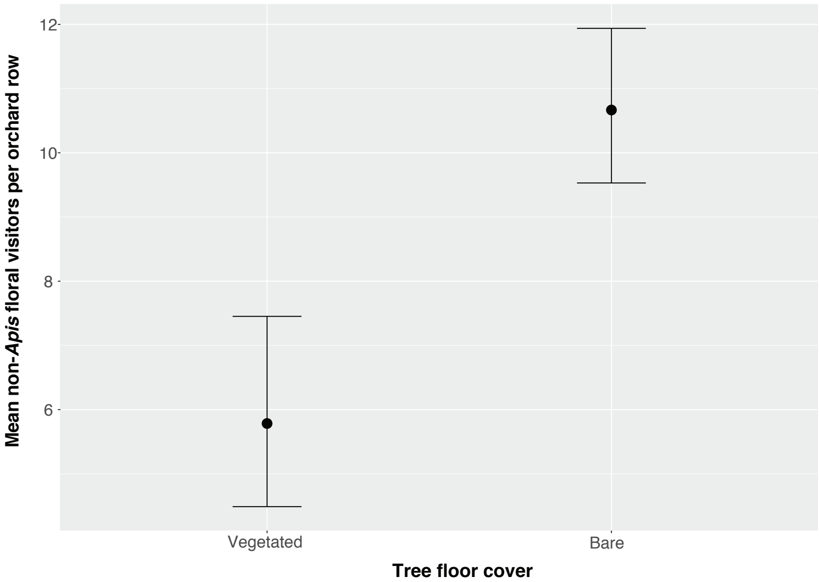


Figure 41. Model predicted mean non-Apis floral visitors per orchard row under different tree floor vegetation management conditions with 95% confidence intervals.

In comparison, the second model also found tree floor shape (z: 3.9, $p < 0.05$), in addition to under-tree cover (z: 2.7, $p < 0.05$) and temperature (z: 6.451, $p < 0.05$), to have a significant effect on non-Apis bee abundance.

Post-hoc pairwise comparisons of the two main factors of under-tree cover and shape (Figure 42) revealed that orchards that were bare and mounded had significantly higher (model estimated mean(μ): 9.8, CI: 7.7-12.4) numbers of non-Apis bees per row than those that were vegetated and mounded (μ: 5.9, CI: 3.7-9.4, z ratio: 2.7, $p < 0.05$), bare and flat (μ: 5.0, CI: 4.1-6.1, z ratio: 3.9, $p < 0.05$), and vegetated and flat (μ: 3.06, CI: 2.25 - 4.17, z ratio: 6.073, $p < 0.05$).

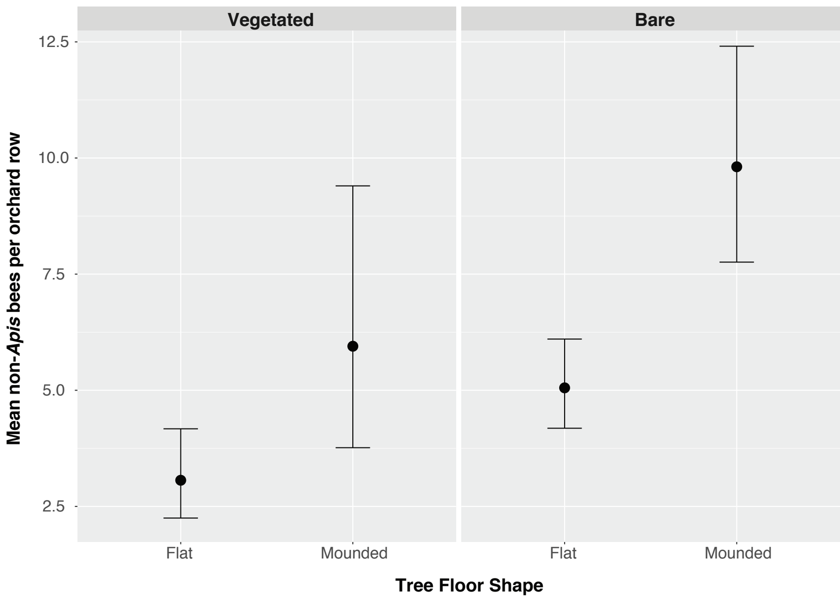


*Figure 42. Model predicted mean non-*Apis *bees per orchard row under different vegetation management conditions and tree floor shape with 95% confidence intervals*

Investigate how co-flowering non-crop floral resources influence pollinator visitation to crop flowers (5.4.b)

On the weeds in the orchard, *Lasioglossum (Chilalictus)* species, *Homalictus*, and syrphid flies were found to be actively foraging on *Arctotheca calendula* (cape weed), *Taraxacum officinale* (Dandelions), and *Sinapis arvensis* (wild mustard).

After the apple flowering season, the unmown rows mostly had introduced grasses, sometimes accompanied by flowering subterranean clover (*Trifolium subterraneum*), plantain (*Plantago lanceolata*) and chickweed (*Stellaria media*), and occasionally red chickweed (*Anagallis arvensis*), all of which were not attractive to native bees. When some capeweed or other yellow Asteraceae such as flatweed (*Hypochaeris radicata*) and hairy hawkbit (*Leontodon taraxacoides*) or dandelion were present, species of *Lasioglossum* and *Homalictus*, were often observed, which are the main wild bee visitors of apple in this region.

The plants in the meadow mix trial led by UTas and coordinated in SA by Fruit Growers SA were not growing or flowering well in either year, and were mown soon after the apples finished flowering. The number of native bee visitors per flower or plant was established.

Table 3. Plants in the meadow mix trial, and adventive in apple orchards and bees observed per flower. Yellow: attractive hoverfly plant. Blue: attractive bee plant

	Flowering?	N flowers observed	Honey bees	Hover flies	Butterflies	Native bees
Meadow mix						
Alyssum benthamii (Sweet alyssum)	yes	500	0	200	2	1
Ammi majus (Queen Anne's Lace)	no					
Anethum graveolens (Common Dill)	no					
Aubrieta hybrida (Rock Cress)	few	0	0	0	0	0
Cheirianthus chieri (Wallflower)	no					
Chrysanthemum maximum (Shasta Daisy)	no					
Coreopsis lanceolata (Mayfield Giants)	no					
Coreopsis sativum (Coriander)	no					
Cosmos bipinnatus (Garden/Dwarf Cosmos)	no					
Eschscholzia (California Poppy)	no					
Gaillardia aristata (Blanket Flower)	no					
Iberis umbellata (Candytuft)	no					
Liatris spicata (Dense Blazing Star)	no					
Monarda fistulosa (Wild Bergamot)	no					
Nemophila menziesii (Menzies' Baby Blue Eyes)	yes	140	2	0	0	2
Adventive weeds in orchard						
Arctotheca (Cape Weed)	yes	2500	127	121	5	67
Veronica persica (Creeping Speedwell)	yes	500	0	7	4	15
Trifolium repens	yes	400	8	9	1	2
Trifolium subterraneum	yes	400	0	0	0	0
Oxalis pes-caprae (Sour sob)	yes	100	2	0	0	1
Taraxacum officinale (Dandelion)	yes	200	6	1	0	1
Hypochoeris radicata (Flat Weed)	yes	400	20	56	4	16
Anagallis arvensis (Red Chickweed)	yes	150	0	0	0	0

The weeds already present were more attractive than most of the plants in the meadow mix, with cape weed and flat weed standing out. However, cape weed flowers only in spring, and will not provide sustenance for the bees during summer. It can be maintained in the interrow, or just outside the orchard block by disturbing the soil (e.g., every two years). Of the meadow mix flowers, *Nemophila menzies* seemed the most useful for native bees. However, the plants in the meadow mix did not flower well, and most are perennials that require a year to become established.

Of native plants in the Adelaide Hills, *Scaevola* and *Pultenaea pendiculata* are among the most useful plants for generalist bees (Spronk et al, 2023). While these can be planted on the edge of orchard blocks, it is unlikely that they would survive in the interrow, and it is possible that they would cause slipping hazards in steep orchards.

Alternate mowing of rows seems to lead to a reduction in flowering weeds, and an increase in grass, and may therefore not be a good way to improve floral resources for bees. However, it is possible that seed production and therefore proliferation of flowering weeds is influenced by the frequency of mowing.

Ground nesting bees benefit from access to open soil under the trees or in headlands, which is kept clear of dead leaves and pruned branches. Under-tree herbiciding in winter creates this accessible soil. Some weeds, in particular yellow Asteraceae can provide additional pollen and nectar sources for the native bees.

3.3 Plant & Food Research

Understand floral biology to inform development of mechanical and bee-vectored pollination to optimise productivity

Relate pollinator behaviour undercover and open-field to floral resource quality and fruit set of apples (5.1.d)

To assess pollinator behaviour in an apple orchard, foraging bees were followed in covered and uncovered blocks, and blocks with gaps in the roof of the netting.

Flowers visited/min was similar in all three growing environments: 5.87 ± 0.23 (n = 111); 6.31 ± 0.22 (n = 109); and 5.95 ± 0.39 (n = 37) respectively (p = 0.379; Kruskal-Wallis).

To assess fruit set in covered versus open apples, flowers were prepared and marked in a covered and uncovered block of both ‘Smitten®’ and ‘Ambrosia’ (the same blocks as above) as follows:

1. 30 hand pollinated + bee visits
2. 30 bee visits only
3. 30 bagged (controls).

Overall, flowers in the uncovered block produced fruit with significantly more seeds compared to the covered block (5.5 versus 3.5 seeds on average; GLM: χ^2 = 9.190, 1 df, p=0.002; Figure 43).

Supplementing bee pollination with hand-applied pollen increased the number of seeds and reduced the number of empty carpels, irrespective of cover type (glm: χ^2 = 44.200, 1 df, p<0.001; Figure 43). With the addition of pollen, both environments produced fruit with ≥ 7 seeds on average.

While this suggests there may have been a pollen deficit in both growing environments, only the ‘bee only’ flowers under netting produced fruit with fewer than five seeds on average. The literature suggests that fruit size in apples typically increases with increasing number of seeds up to five to six seeds per apple (after which there is diminishing returns), e.g. Sheffield 2014), so the fruit in this treatment may have been smaller than optimal. However, we did not find an overall increase in fruit weight with seed number in either variety assessed in our trial (Figure 44).

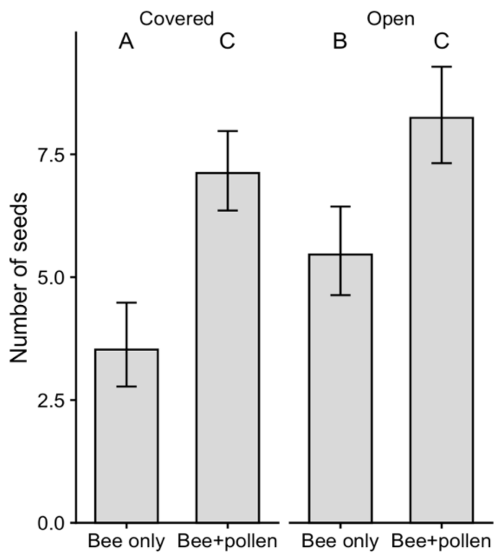


Figure 43. Number of seeds in fruit that were hand pollinated (bee + pollen) versus those that were open pollinated (bee only) in partially covered (bee only n = 19; bee + pollen n = 42) and uncovered (bee only n = 26; bee + pollen n = 33) blocks. Model estimates and 95% confidence intervals shown. Different letters indicate significant differences

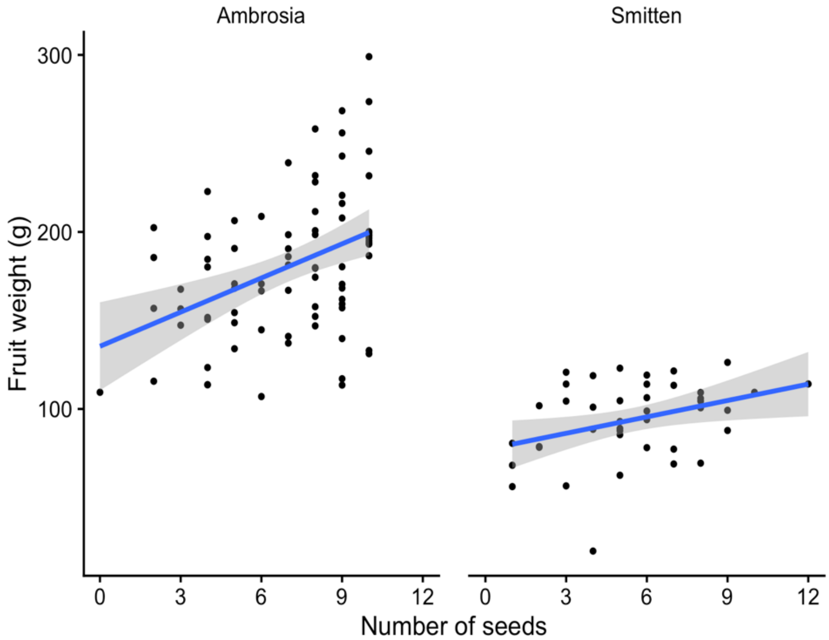


Figure 44. Relationship between seed number and fruit weight in Ambrosia and Smitten apples

Assess the relationship between initial colony size and colony size post-flowering of apples (5.1.e)

Pre-flowering and post-flowering colony strength assessment data (frames of bees – FOB) collected from New Zealand apple orchards in 2019, 2020, 2021, and 2022 have been pooled to provide data from 372 different colonies. The colonies were situated at the following combinations of location within block and netting type:

1. Open/uncovered (no netting)
2. At edge of partially (roof only) covered block
3. In the middle of a partially covered block,
4. Under gaps in roof netting of partially covered blocks
5. On the edge of fully covered blocks
6. In the middle of fully covered blocks
7. Under gaps in roof netting of a fully covered block.

The size of honey bee colonies going into flowering is a significant predictor of the change in colony strength coming out of flowering; bigger colonies lost more frames of bees than smaller colonies; some small colonies increased in worker strength (Figure 45).

The strength of this relationship differed across treatments; with colonies located in the middle of fully covered blocks showing greatest decrease in FOBs with pre-flowering colony strength (-0.908 FOB with every additional frame 'pre') and colonies in the open or under a gap in a fully covered block changing independently of pre-flowering colony strength.

Recommendations for the maximum colony strength (in terms of FOBs) for colonies deployed for apple pollination under different cover structures and positions within the orchard are provided in Table 4. To ensure that the desired stocking rate is achieved when following the below recommendations, a larger number of colonies may need to be deployed, particularly if small colonies are used (i.e. ≤ 6 frames of bees).

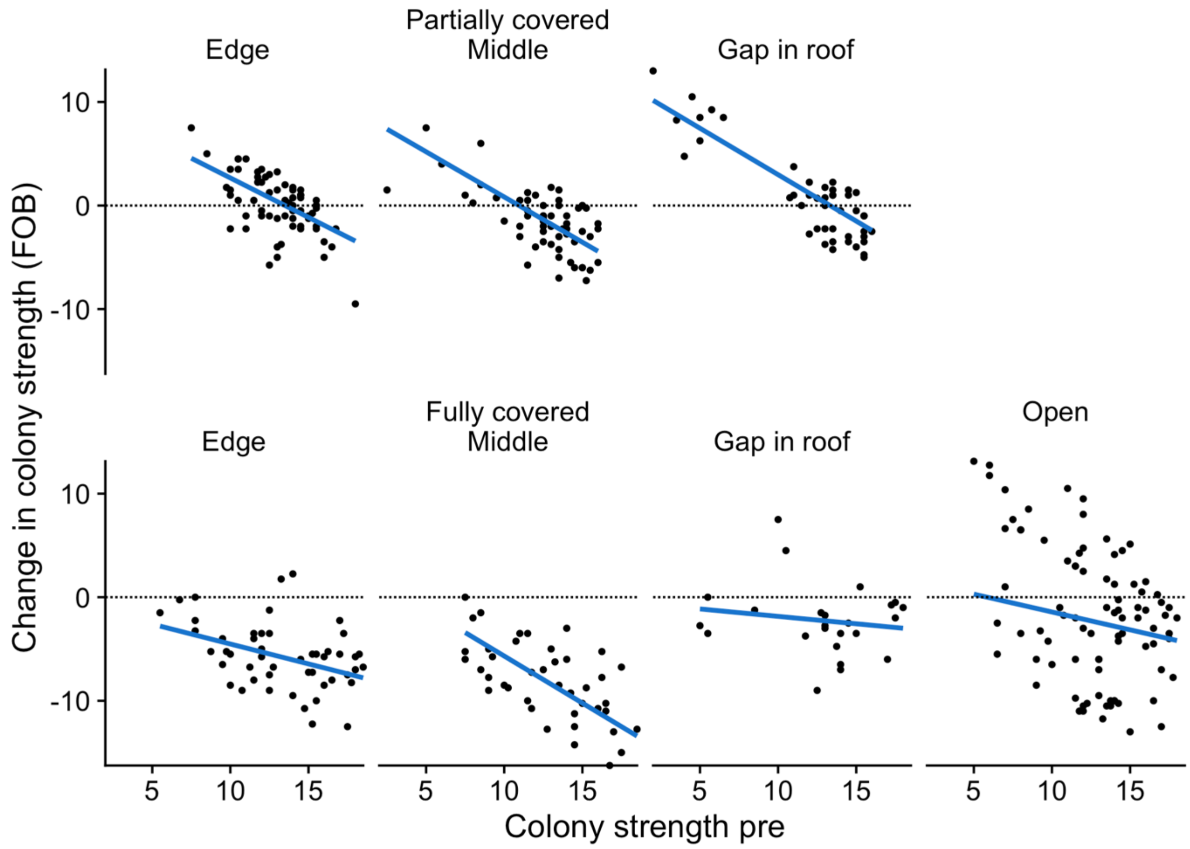


Figure 45. Change in frames of brood (colony strength) during apple flowering in relation to the number of frames of brood in the colony at the beginning of apple flowering ('Colony strength pre'). 0 = no change colony strength. The top three graphs correspond to partially covered apple blocks and the bottom four graphs correspond to fully covered apple blocks. Data points represent individual colonies. Blue lines represent the relationship between change in colony strength and colony strength at the beginning of flowering based on the statistical model fit to the data.

Table 4. Modelled change in frames of bees (FOB) during apple flowering in relation to the number of frames of brood in the colony at the beginning of flowering ('pre'). The maximum number of 'pre' frames before FOBs (frames of bees) decreased is our recommended colony size.

Treatment	Change in FOB with every additional frame 'pre'	Maximum number of 'pre' frames before FOBs decreased	SE	DF	Lower CL	upper.CL
Fully covered middle	-0.908	3.75	0.157	358	-1.218	-0.599
Part covered gap in roof	-0.897	13.3	0.208	292	-1.306	-0.487
Part covered middle	-0.873	10.9	0.154	354	-1.176	-0.571
Part covered edge	-0.762	13.5	0.163	323	-1.082	-0.442
Full covered edge	-0.384	NA*	0.117	319	-0.614	-0.155
Open	-0.345	NA*	0.102	331	-0.546	-0.143
Full covered gap in roof	-0.143	NA*	0.142	303	-0.422	0.136

* Weak relationship between frames of brood pre flowering and FOB delta.

In locations with poor pollination, standard strength colonies could be replaced with a larger number of small, growing colonies to reduce bee losses. This strategy may be particularly effective for colonies in partially covered blocks and those located in the middle of fully covered blocks (not under gaps). We note that this strategy not been tested directly to assess its efficacy for pollination.

Improve the performance of honey bee colonies deployed under cover through orchard manipulation

Determine whether opening of sides improves pollination in covered apple orchards (5.2.a)

Colony performance: Colonies were visually assessed at the beginning and end of flowering; colonies were situated at the same locations described above.

Our combined assessments from 2019-2022 showed that on average, the colonies in the partially covered bocks performed similarly well compared with the open/uncovered blocks and maintained their strength over the course of flowering regardless of their position within the block.

Conversely, colonies in the middle of fully covered blocks declined significantly compared to colonies in the open/uncovered blocks (Figure 46; Figure 47). In fully covered blocks, creating a gap in the netting above the colonies significantly reduced bee losses over the flowering period (Figure 46; Figure 47; linear mixed-effects model: χ^2 = 42.6 at 6 df; p<0.001; significance of treatment established with a likelihood ratio test).

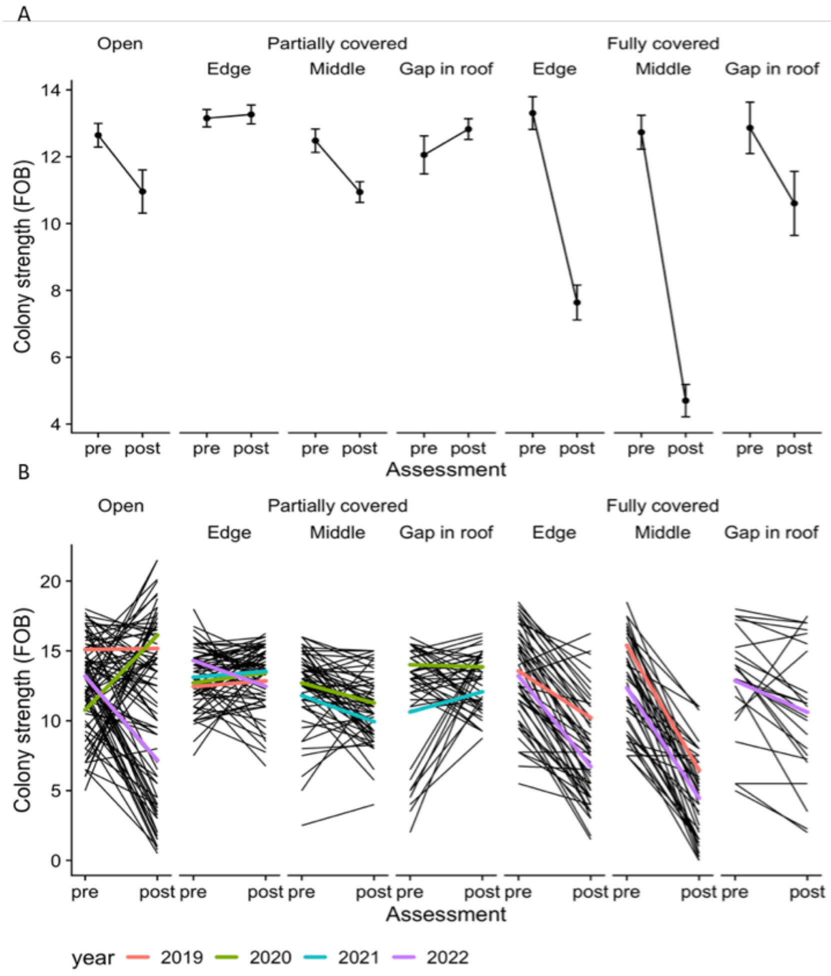


Figure 46. Mean differences in colony strength (frames of bees) overall (A) and each season (B) at the beginning and end of flowering, for colonies in different locations within apple blocks. The different locations (treatments) compared included open blocks (n = 85 colonies), on the edge of partially covered blocks (n = 67), in the middle of partially covered blocks (n = 61), under a gap in the roof of partially covered blocks (n = 45), near the wall in a fully covered block (n = 49), in the middle of a fully covered block (n = 41) and under a gap in the roof of fully covered blocks (n = 24). Coloured lines in (B) are the group means in different years. Black lines in (B) are raw data from individual colonies. Note – A cold snap in 2022 (purple lines) may have affected the colony strength in the open orchard (netted orchards did not frost, but open one did). (FOB=frame of bees)

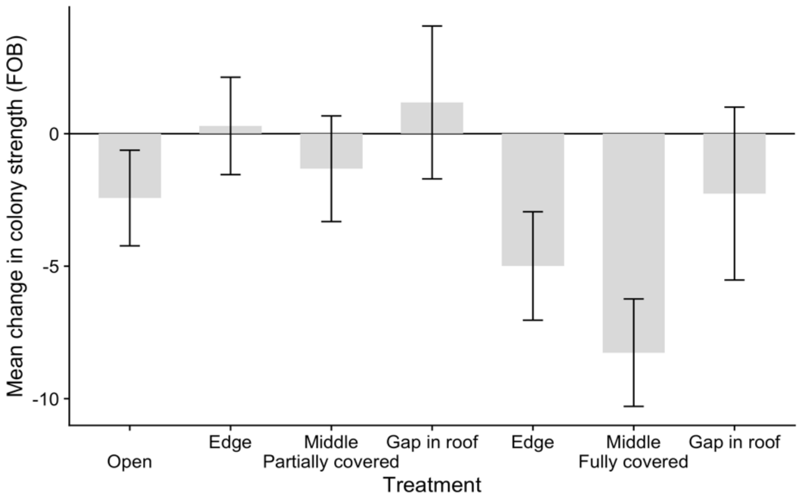


Figure 47. Change in colony strength (frames of bees) over the course of flowering, for colonies in different locations within apple blocks. Colony strength data has been modelled to account for hive drop, orchard, and year combination. Colonies in "Fully covered middle" positions lost significantly more bees than colonies in "Open" (control blocks) locations. There were no statistically significant differences between "Open", and the partially covered treatments or the "Fully covered gap in roof" treatment. Nor between the "Fully covered edge", and "Fully covered middle" treatments. Error bars indicate 95% confidence intervals around group means. (FOB=frame of bees)

Bee movement/Pollination: The number of bees on apple flowers dropped off with distance from hives more rapidly when the hives were located at one end of a covered block (i.e., an open side).

Overall, bees foraged further from their colony when their hive was located in the middle of the block, but bees from hives under gaps still foraged significantly further down rows compared to bees leaving hives on the edge of blocks (Figure 48).

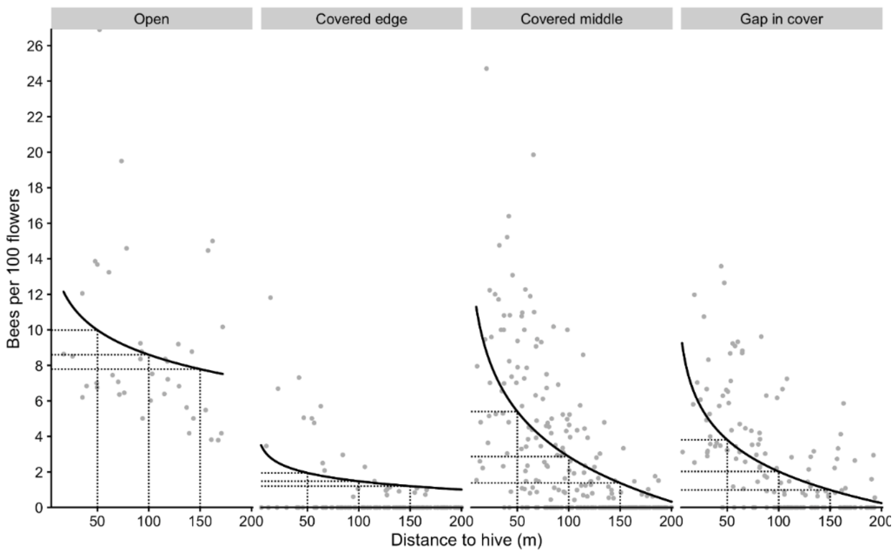


Figure 48. Honey bee density (bees/100 flowers/minute) compared at different distances for hives that were located either in open/uncovered blocks; on the edge of covered blocks; in the middle of covered blocks; and in the middle of covered blocks under a gap in the netting. Data points represent daily sums for each individual transect point. The slope lines represent the relationship of honey bee density and distance to hive drop based on the statistical model fit to the data.

Installing temporary gaps in the roof of the netting may help reduce honey bee losses in fully covered blocks. Opening the sides is another option for improving honey bee colony performance under covers.

Optimal pollination results were achieved by placing hives in the centre of partially covered apple blocks, either under cover or beneath a gap in the netting.

Determine the effects of covers on colony performance and pollination in a lychee orchard (5.2.a)

Colony performance: Forty hives were assessed across nine blocks; five fully covered blocks and four blocks with one to two open sides. The number of adult bees in colonies remained similar in the fully covered blocks (t(19) = 0.6, p = 0.5), and increased in blocks with open sides (t(19) = -6.6, p < 0.001; Figure 49).

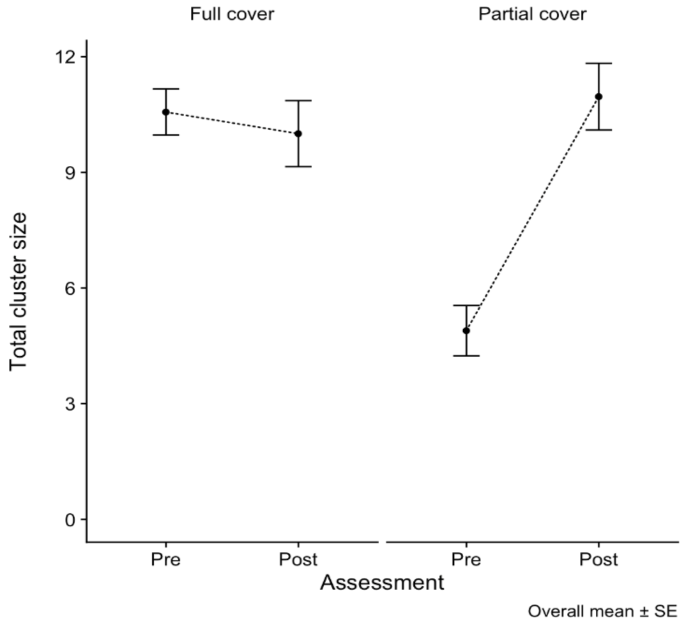


Figure 49. Mean colony strength (frames of adult bees) in hives at the beginning (pre) and end (post) of lychee flowering under full cover and partial cover. Vertical lines are standard errors of the mean.

Pollination: Across all hives/blocks, bee activity (honey bees per 1000 flowers) was not significantly affected by cover treatment (p = 0.6) but was affected by location within the block (p = 0.02), with fewer honey bees being found in the centre of blocks (model effect size = 0.2 and 0.1 honey bees per 1000 flowers on the edge vs middle of the block; Figure 50). This pattern of activity was not influenced by hive location (Figure 51).

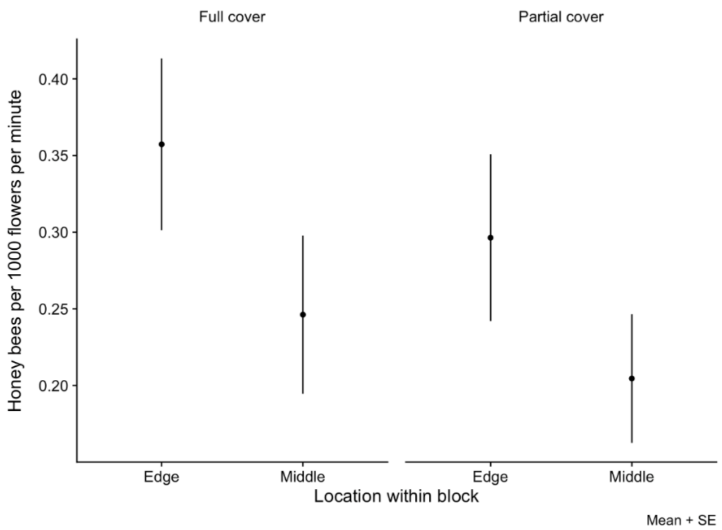


Figure 50. Mean counts of honey bees per 1000 lychee flowers on the edge of the block versus in the middle of the block, in seven fully covered and four partially covered (roof and two sides). Vertical lines are standard errors of the mean.

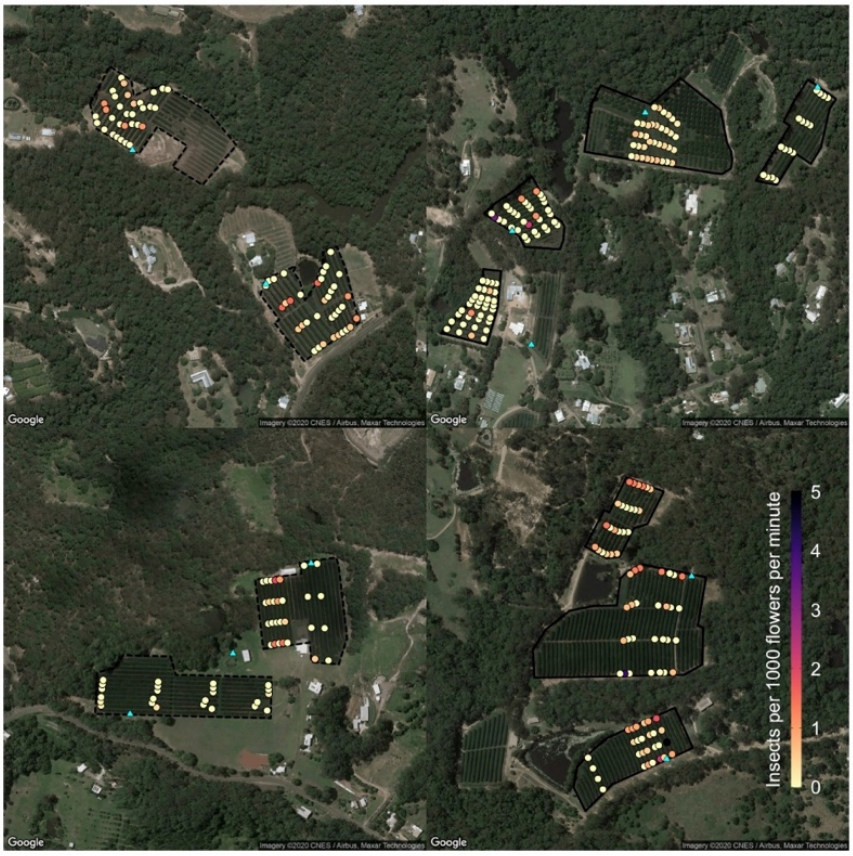


Figure 51. Average distribution of flower visits in lychee blocks (honey bees per 1000 flowers per minute; coloured dots). Black polygons denote covered blocks (solid line: full cover; dashed line: partial cover). Cyan triangles indicate hive locations

Honey bee colony strength pre and post flowering of lychees was unchanged in fully covered blocks but increased in those with some sides open (noting that these colonies were initially smaller).

Honey bees were more active at the edges than the middle of the blocks, regardless of netting type.

Improved the performance of honey bee colonies deployed under covers through orchard manipulation

Assess of how different types of covers and nettings affect light transmittance and honey bee orientation (5.2.a,d)

Light Transmittance

The degree of linear polarisation visible in the blue channel through darker materials was similar to an uncovered sky at approximately 45% (Figure 48, for example). Polarisation was generally lower under lighter materials, dropping to approximately 20% for white quad netting (Figure 49). Darker materials absorb more light than they scatter, which could explain this difference. Some variation in polarisation is expected from time-of-day and the section of sky selected, however we believe the choice of covering to be the predominate affect in these data. Bees can be quite sensitive to polarisation in the blue-to-ultraviolet band, needing only 5-10% polarisation in a patch of sky to successfully navigate (Wehner 1984). All netting types measured exceeded this threshold (except one measurement of the white netting) in sunny conditions.

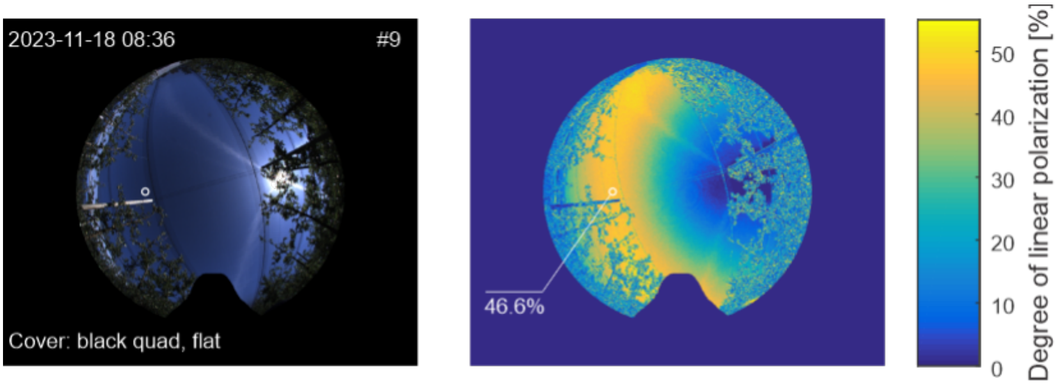


Figure 52. Example of natural light (left) and degree of linear polarisation at approximately 450 nm (right) collected through 'black quad' nets. White markers indicate the locations of polarisation measurements

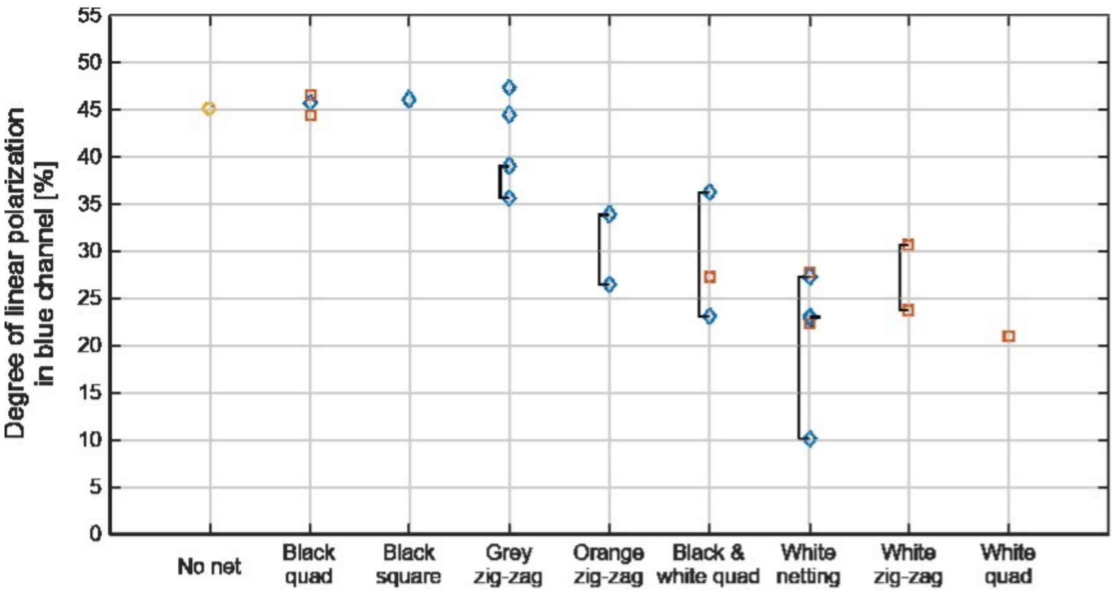


Figure 53. Highest degree of linear polarisation observed at selected locations grouped by covering material. Observations at slightly different positions in a single image are connected by black brackets; flat coverings are marked with squares while pitched cover

Orientation of the covering, whether parallel to the ground (Figure 54A) or pitched (Figure 54B,C), affected polarisation observed, particularly in the lighter coloured coverings. With pitched cover sections perpendicular to the central solar axis, polarisation changed suddenly from 27% to 10% between sections (Figure 54B). This was not observed with pitched cover parallel to the central solar axis, with polarisation of 23% observed in nearby locations (Figure 54C). This could occur if light is not scattered uniformly off the covering material, which is unlikely.

Scattering direction may be affected by the microstructure of the covering material, and the way it is stretched, so the effect is still apparent in flat coverings. Stacking several cover layers can also create discontinuities. Discontinuities in polarisation were not observed under the white quad cover, which also showed the largest difference to open sky polarisation at 20%. This could be caused by a high degree of scattering randomising the polarisation and scattering more light away from the canopy. Further work will be needed to quantify these effects.

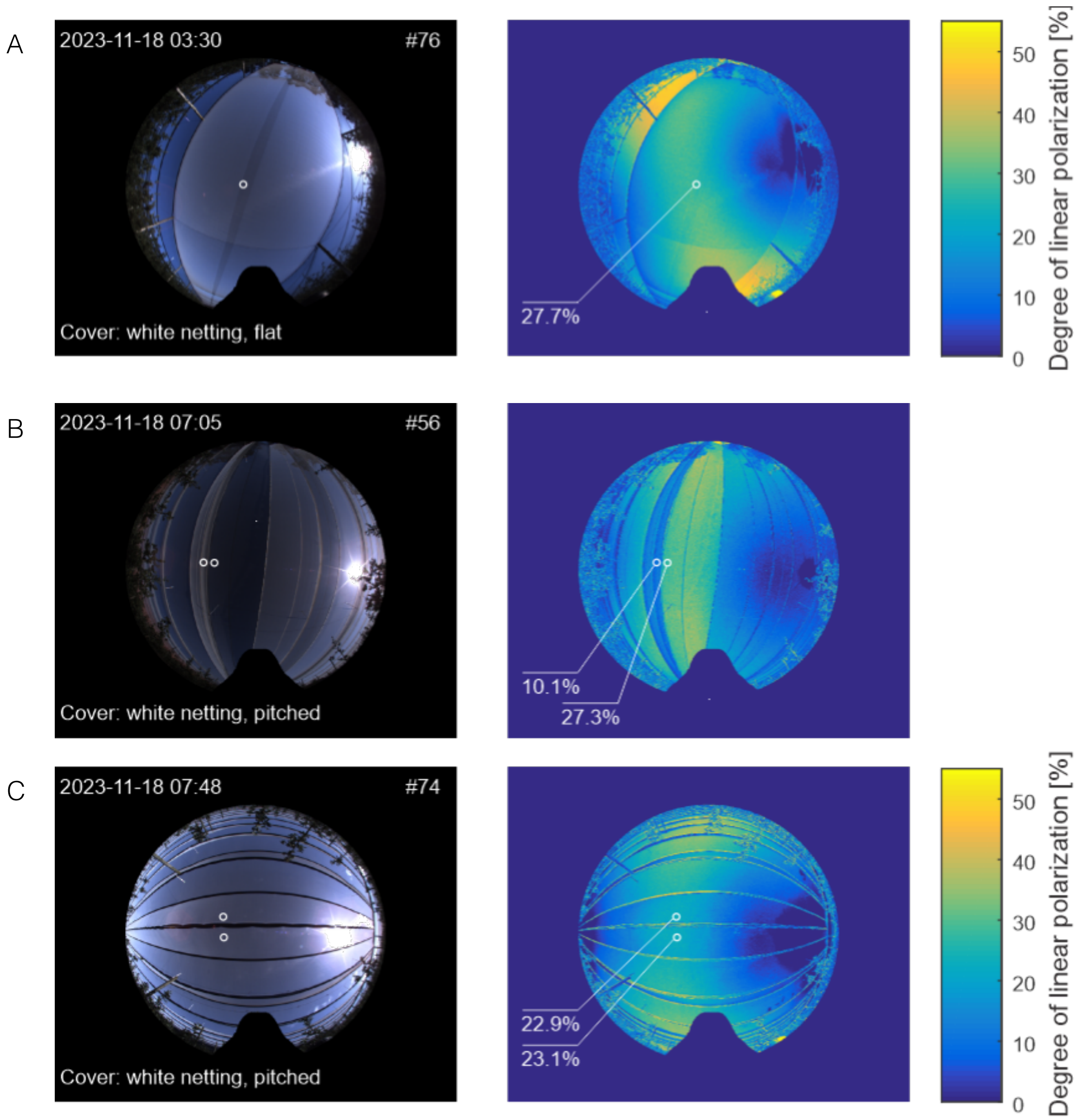


Figure 54. A) Example of natural light (left) and degree of linear polarisation at approximately 450 nm (right) collected through white netting suspended parallel to the ground covering a clear sky. B – C) Example of natural light (left) and degree of linear polarisation at approximately 450 nm (right) collected through white pitched netting completely covering clear skies. Longitudinal pitch is perpendicular to (B) and parallel to (C) solar rays passing through the centre of the sky.

Clouds also scatter light, reducing the degree of linear polarisation observed (Figure 55). Under overcast sky, polarisation was largely uniform and less than 5% across both netted and uncovered regions (Figure 55A). Discontinuities in polarisation, congruent with netting boundaries, are observed in partly cloudy skies (e.g. Figure 55B). However, quantifying the affect under clouds is difficult because the characteristic polarisation seen in Figure 52 is not available to aid in locating the peak. In a small region of the sky, with a mix of cloud and net cover, polarisation varied from 8.4 to 23%. Generally, polarisation was lower in the presence of nets, thick clouds or both, and higher under clear patches, whether covered by net or not (Figure 55C). Artificial coverings may alter the natural pattern of polarisation insects may 'expect' to observe in the sky, but perhaps no more than variation in cloud cover. However, cloud cover may have a short-term temporal component that is not present in fixed coverings.

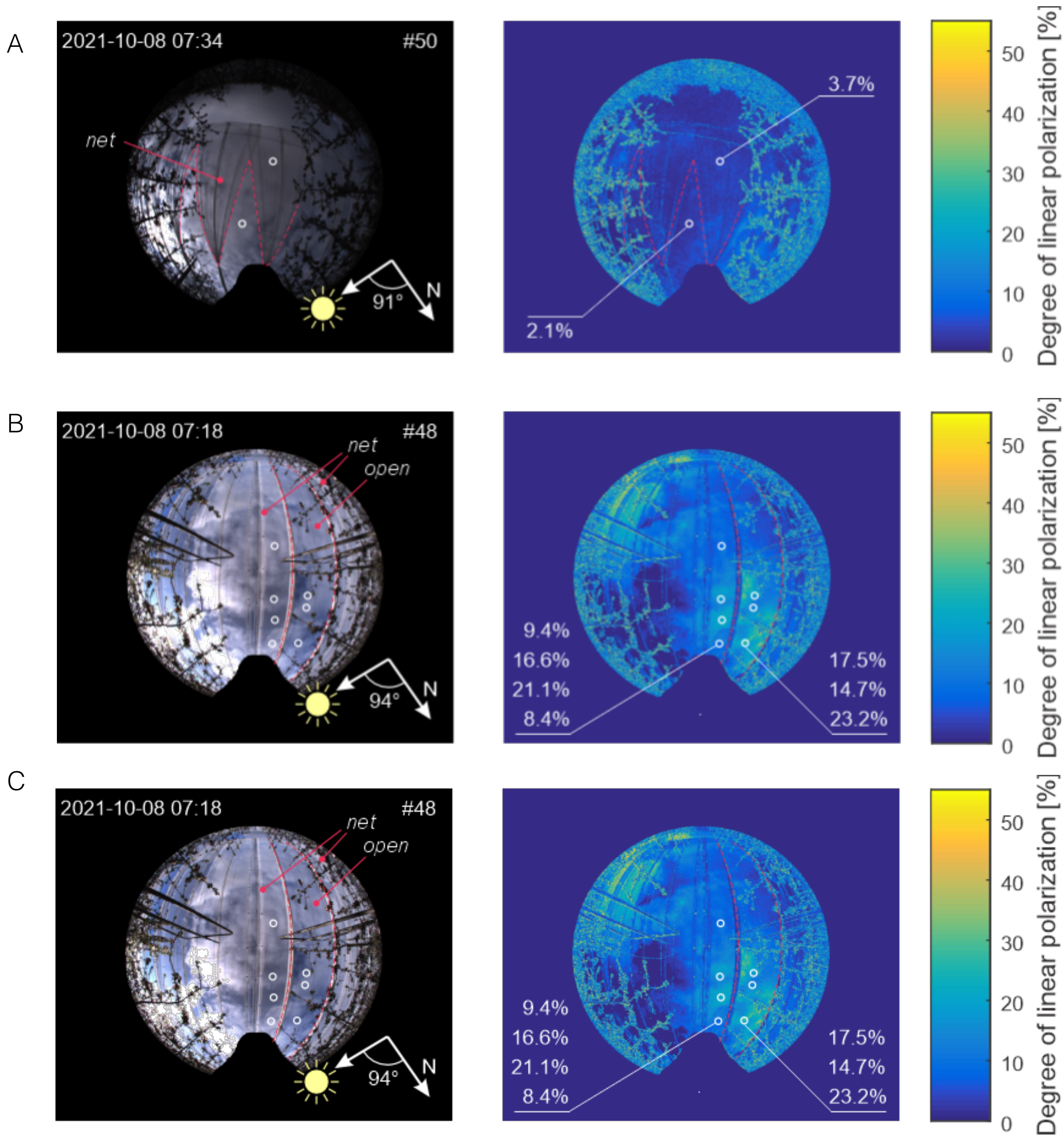


Figure 55. Example of natural light (left) and degree of linear polarisation at approximately 450 nm (right) collected through partially folded back nets under overcast sky (A), through partially open netting under partly cloudy sky (B), and through partially open netting under mostly cloudy sky (C). Approximate location of sun and north, along with net boundary and selected polarisation measurements are shown.

Forager navigation

In terms of delta angle (the difference between actual honey bee dance direction and the true angle of the feeder), there was no statistically significant difference between the uncovered control and the white netting treatments. Bees in these treatments exhibited dances that closely aligned with the actual direction of the feeder. However, dances performed under black netting showed a significantly lower delta angle compared to both the control and white netting treatments across all observations, and the direction of bee dances under black netting differed significantly from the actual feeder direction (Figure 56A).

No statistical differences were observed between netting types regarding the variation in dance direction (kappa, the concentration parameter; Figure 56B). However, the white netting treatment exhibited the highest variability, followed by the control and black netting treatments. A larger sample size of bees would be necessary to detect subtle differences in dance variability, given the considerable variation observed in dance direction both among bees.

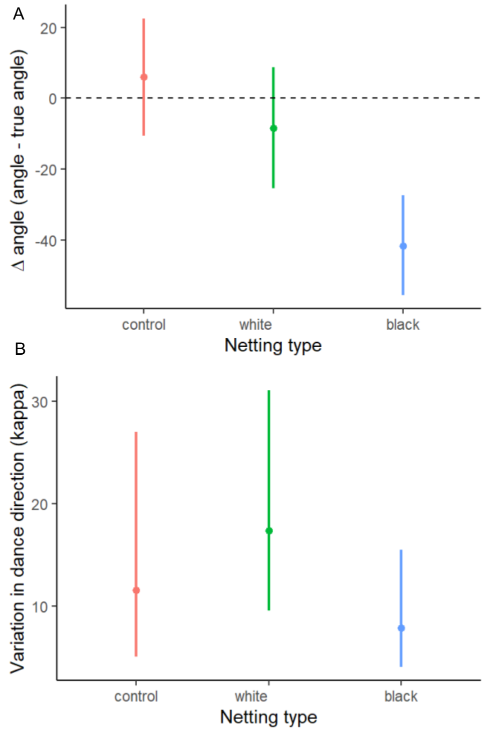


Figure 56. Conditional effect plot of the difference between netting types (white, black, and no netting/control) on: A) honey bee waggle dance direction/angle in radians (-pi,pi); and B) variation in dance direction (kappa). The dashed line at 0 represents the expected direction of the waggle dance (directly towards the feeder). The number of bee dances observed differed among the treatments: black netting n = 14; white netting n = 48, and control n = 47.

The accuracy of honey bee waggle dances was compromised by the presence of black netting, leading to communication of incorrect feeding locations to nest mates. This disruption does not appear to stem from a reduction in the polarisation of light caused by the black netting. Instead, it may be attributed to a decrease in the amount of light entering the growing system. These findings suggest that bees are more susceptible to becoming disoriented or lost while foraging under this type of netting.

Bee movements in relation to pollinisers

The average number and location of marked bees found in our surveys are shown Figure 57. Overall we found no effect of orchard netting type on the number of marked honey bees visiting the flowers (negative binomial generalised linear model: p = 0.1322, or on the rate of decay of honey bee visits with increasing distance from the polliniser (p = 0.9334; Figure 58). The model accounted for the variation in flowering intensity between orchard and survey days and for differences between survey bouts in the number of marked bees.

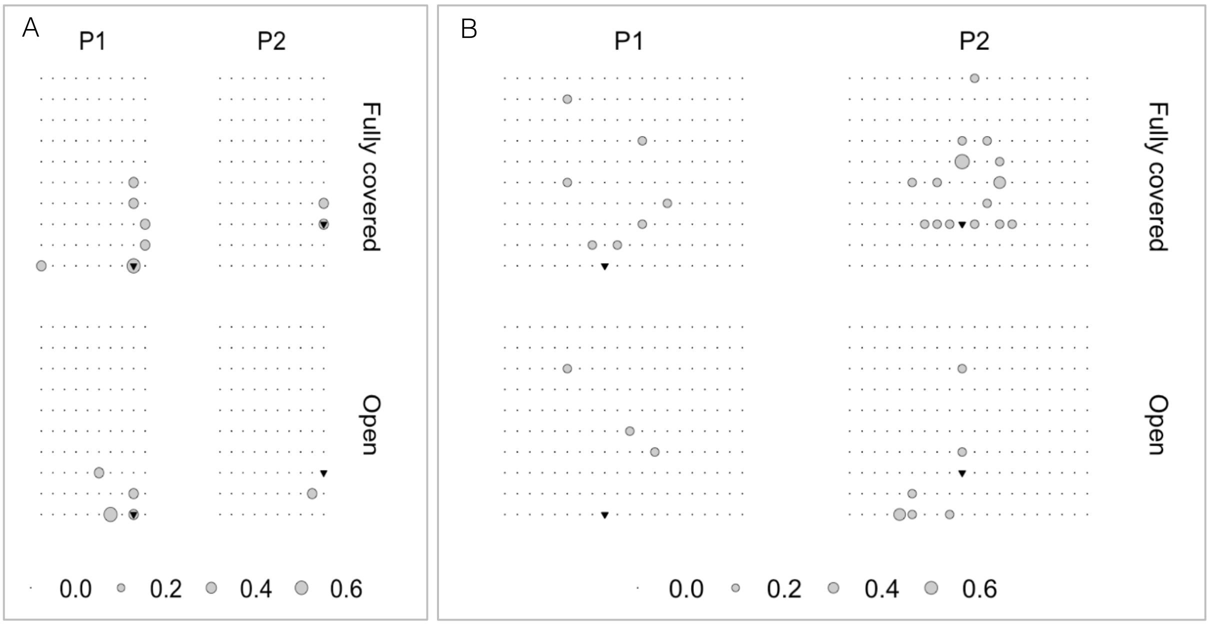


Figure 57. (A) Average numbers of observed marked bees in 10 row by 10 bay areas and (B) average numbers of observed marked bees in 20 row by 5 bay areas (extended surveys). Location of pollinisers (P1, P2) is indicated with upside down triangles.

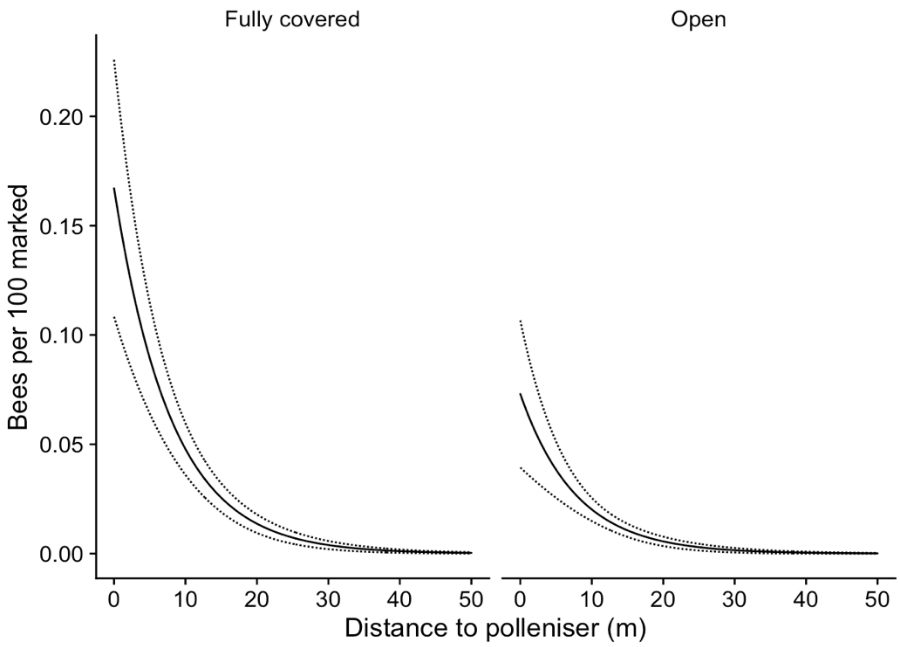


Figure 58 Numbers of marked honey bee visits with distance from polliniser. Dotted lines indicate +/- standard error of the model fit. There were no statistically detectable differences between orchard netting types.

3.4 NSW Department of Primary Industries

Understand floral biology to inform development of mechanical and bee-vectored pollination to optimise productivity

Conduct field experiments to characterise pollen quality and nectar quality of berry; relate pollinator behaviour undercover and open-field to floral resource quality and fruit set; Assess pollinator visiting rates and their flower visiting behaviour along polytunnels (5.1.a,d, c)

Variation in floral morphology among seven blueberry varieties

Could the shape or morphology of a flower be important to the attraction of a bee to that flower? To answer this question, the NSW DPI researchers investigated some floral traits that may affect the attraction of European honey bees (*Apis mellifera*) to blueberry flowers.

Previous research suggests that blueberry flowers with a large opening, or throat area, can provide greater access to the nectar found at the base of the flower. However, this trait and its 'attractiveness' has not been examined among the cultivars used in Australia. And, if influential in pollination, cultivars with more 'attractive' flowers could be developed for utilising under high tunnels where honey bees are less inclined to visit and where poor pollination is a problem.

The first step in this investigation examined the floral morphology of seven blueberry cultivars with a view to selecting cultivars with contrasting flowers to test in the field for their attractiveness to honey bees.

The flowers of the seven blueberry varieties Dazzle, 11-11, Eureka, Eureka Sunrise, First Blush, Masena and Splash, were obtained from plants growing in the same location on the North Coast of NSW. They were picked fresh and preserved in alcohol until they were examined.

The variability of flower shape was recorded using methods in microscopy including photographs of the magnified flowers and imaging software to measure the corolla (the group of petals) width and length and the corolla throat area (Figure 59).

Closely examining corolla length and corolla throat area shows that Eureka has the longest flower with the largest opening, and therefore the greatest contrast in floral morphology compared with all the other varieties (Figure 60). However, the corolla width was relatively similar among these varieties.

As the cultivars Eureka Sunrise, 11-11 and Masena contrast in their flower shape, and since they flower at a similar period and in our study were planted in the same location, these were compared for their ability to attract bees in a commercial crop.

Floral nectar weights did not differ among the cultivars, but cultivar Masena contained a higher concentration of sugars in nectar compared with Eureka Sunrise, with 11-11 having variable concentrations.

Eureka Sunrise appeared to have the highest visitation number from honey bees (to be statistically verified) perhaps related to its potential access (larger corolla width and throat area compared with 11-11 and Masena). Floral bunches on the edge of the stations were also more frequently visited compared to those in the centre of the stations, highlighting the effect of position of floral resources on visitation by honey bees.

All cultivars tested would have required honey bees to enter the corolla to access the floral nectar, having longer corolla tubes than the length of honey bee tongues. We corroborate others in highlighting that ease of access to floral resources, in terms of spatial context, is a determinant in floral attraction for honey bees.

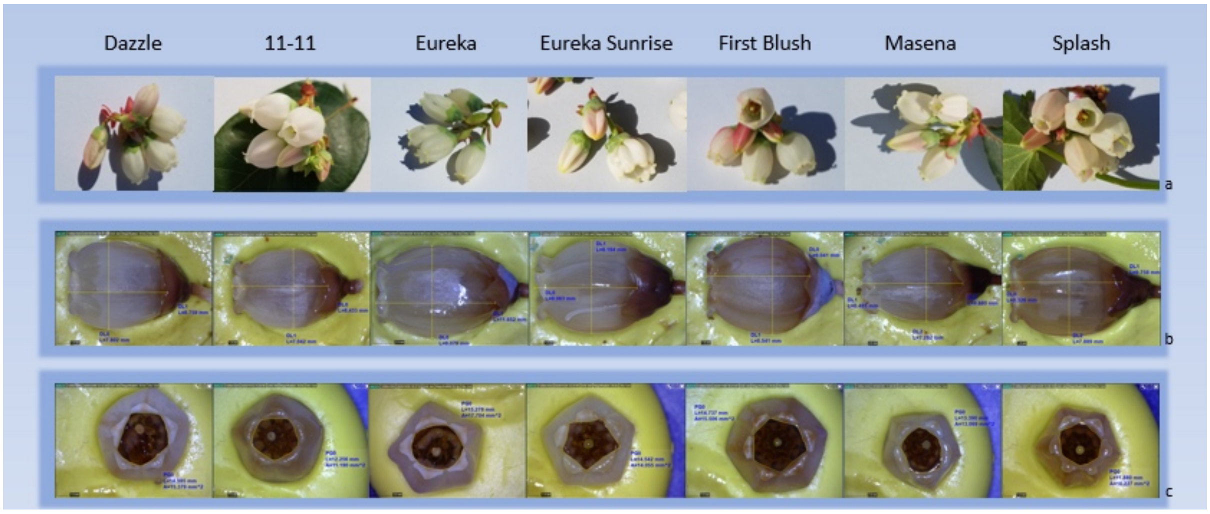


Figure 59. Images of flower racemes (top row), corolla width and length (middle row), corolla throat area (bottom row). Magnification is 30-35x.

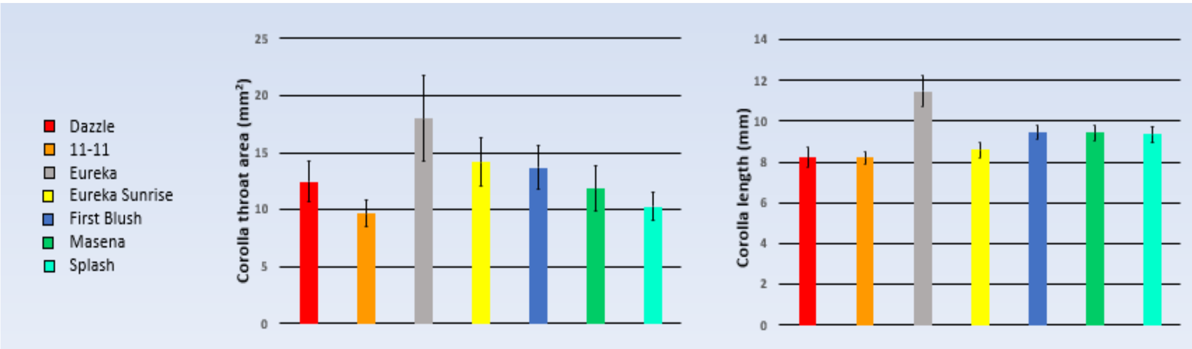


Figure 60. Corolla throat area (left) and corolla length (right) for seven blueberry varieties. The columns represent the average of the measurements of between 21-71 flowers and the bars are standard deviation.

Blueberry cultivars differ in the length, width, and area of the floral tube entrance, and in the length of the style and stigma.

Effect of floral age and time of day on nectar characteristics in blueberry *Vaccinium corymbosum* 'First Blush'

Using the southern high bush cultivar ‘First Blush’ we showed that blueberry flowers, when protected from foragers, can produce nectar sugars during the day and can continue sugar production for up to several days, which helped us to set the parameters for our following studies. In this study, where floral washes were conducted, we did not assess nectar volume over time. However, in our other work, we highlighted how nectar mass and sugar concentrations are likely to be affected by the date of sampling potentially related to contrasting light, temperature, and humidity conditions. The sugar types present in blueberry nectar were also determined and are predominately hexose types (fructose and glucose) with smaller proportions of sucrose (<10%).

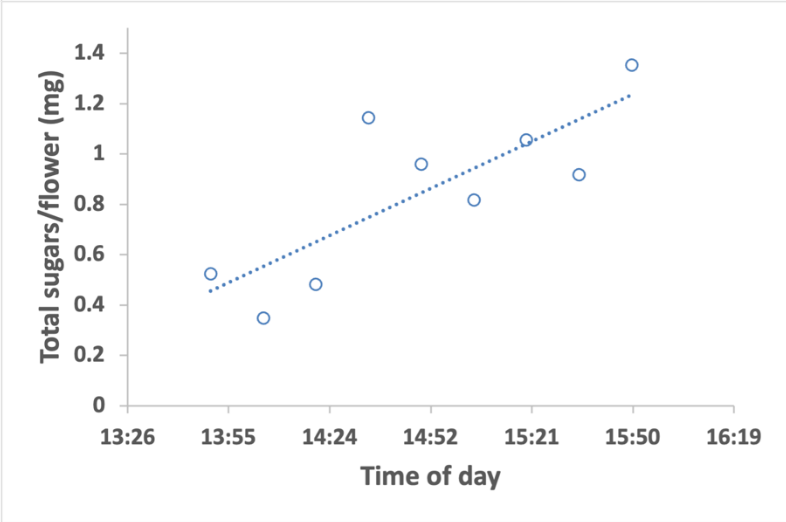


Figure 61. Sugar mass accumulated in individual flowers during the day in the blueberry cultivar 'First Blush'. Values are means of three replicates with trendline fitted (r2=0.64)

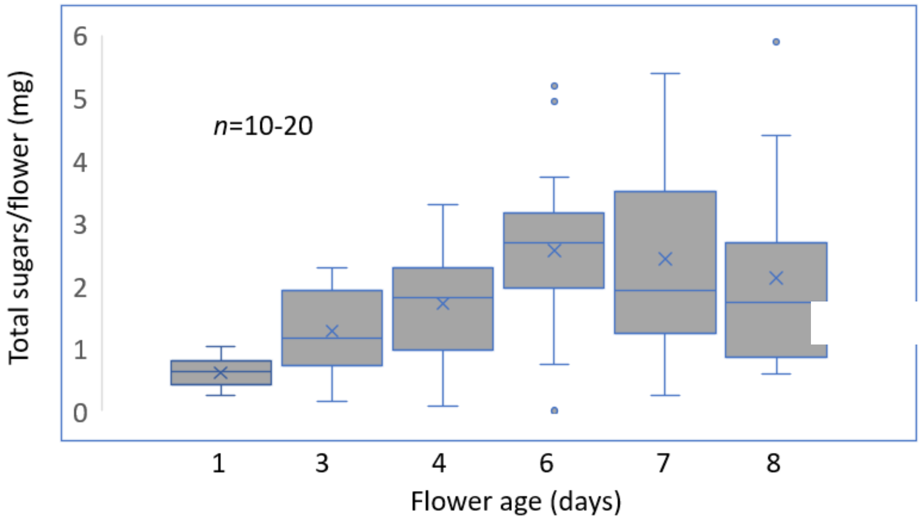


Figure 62. Box plots showing sugar mass accumulated in individual flowers during several days in the blueberry cultivar 'First Blush'

Flowers can accumulate nectar sugars over the day or over several days when pollinators are not present.

Effect of covers (tunnel or bird net) on blueberry pollination

Pollination in highbush blueberry (cultivar 11-11), grown as an evergreen crop underneath bird net and under high tunnels covered with plastic film, was investigated. Data collected on honey bee visits to flowers, floral nectar, and berry quality were used to evaluate pollination success in this crop under the two cover types with some unexpected results.

Figure 63. The study site showing the intersection between the crops under the tunnels and bird net.

Previous research has shown that crop covers can be a barrier to honey bees, limiting their access to the crop and its floral resources. While bird net generally has holes large enough for honey bees to pass through, in contrast, a plastic film-covered tunnel is less accessible with entry possible only via the tunnel ends.

In seeking to answer the question ‘Do tunnels present a barrier to pollination success in blueberry?’ a study was conducted on a commercial farm in northern NSW in three 100m long tunnels and under bird net (Figure 63). The honey bees were placed in two groups of 16 hives outside and they had to enter through bird net covering the sides of the adjacent blocks to access the plants in the tunnels and under bird net (Figure 64).

Figure 64. The honey bee hives placed outside the blueberry crop next to the tunnels. Other hives were placed outside near the intersection between the tunnels and bird netted crops.

Honey bee counts

The numbers of honey bees landing on flowers were counted for 5 minutes at the 36 experimental plants on four occasions across the day. Counts appeared to be little influenced by the type of crop cover, except perhaps in the tunnels where the presence of bees was sometimes greater towards both ends of the tunnels. In contrast, the day of recording appeared more influential with the second day having 25% fewer bees than the first, probably related to cloudier and cooler conditions. The maximum temperature was 25^{o}C on the first day compared with 21^{o}C on the second day and the people counting on this day noticed a complete absence of bees when there was cloud cover.

Sugars in floral nectar

The floral nectar was extracted by shaking flowers in a small vial of water. The total sugars analysed in these suggest that the honey bees were actively harvesting sugars from the flowers under both cover types to a similar degree. The total weight of nectar sugars in the open flowers in the early morning, before the honey bees were flying, was about 1 milligram (mg) per flower. This declined to below 1 mg per flower in the afternoon compared with those flowers where bees were excluded. With bees absent, the sugars in the bagged flowers accumulated during the day to about 2-3 mg per flower.

Fruit quality

This study shines a light on the importance of pollinator presence for fruit development in the 11-11 blueberry variety since fruits from the pollinated flowers were almost 2.5 times the weight of fruits from the unpollinated flowers. In contrast, the effect of the cover types on fruit weight appeared negligible as berry weights from pollinated flowers were similar under both (2.8 grams per berry).

However, the sugar levels in the berries from the tunnels at 16.6 degrees Brix appeared to be higher than those under bird net at 13.1 degrees Brix (although the statistical significance of these results is yet to be confirmed).

Given the similar presence of honey bees and characteristics of floral nectar between the two covers, factors other than pollination appear to be contributing to the higher fruit sugars from the tunnels. The tunnel plants were not rain fed and were receiving more fertiliser via fertigation compared with the bird-netted crop and may have had superior nutrition and therefore a greater capacity to support the developing fruits such as having a larger canopy. We are currently collecting canopy size, and flower and fruit load data from photos taken of each experimental plant to explore this hypothesis further.

Although a plastic-covered tunnel can present some challenges for honey bees accessing crops, in this case study we do not believe the tunnels inhibited pollination success in this blueberry cultivar. Honey bee abundance was similar, and the floral nectar appeared to be as well utilised, under both crop covers. Here, we have not only highlighted the essential role of honey bees and the role of floral nectar to pollination and berry development in cultivar 11-11, but also to the importance of non-pollination factors to fruit quality.

Honey bee visits and pollination were similar under tunnels and bird net. Pollination was essential for good fruit quality, approximately doubling the weight of individual fruits. North facing rows and row edges closest to hives had heavier and sweeter fruits.

Fruits from tunnels were 30% heavier and sweeter than bird-netted fruits, which may be due to better nutrition under the rain shelters.

Effect of fertigation strength on plant nutrition and nectar production in potted blueberry *Vaccinium corymbosum* 11-11.

Floral nectar is the main attractant to blueberry crops for honey bees, with the pollen ignored due to its low protein value compared with other pollens. Access to nectar resources, however, can be limited for honey bees in blueberry crops under covers used to exclude birds and provide protection, as these covers form barriers that affect the bee's flight path. Our research aims to better understand the attributes of nectar from blueberry flowers with a view to increasing the 'attractiveness' of this crop.

One potential intervention to modify nectar production in blueberry flowers is through fertigation. Blueberry crops are often fertigated (irrigation containing fertiliser salts), and we hypothesised that adjusting fertigation strength has the potential to modify nectar production to suit the needs of pollinators.

Can fertigation modify nectar in blueberry flowers?

The nectar weights, total amount of sugars in nectar, including sucrose, glucose and fructose and their concentrations, contrasted between the two days of sampling. As highlighted by other studies, contrasting light, temperature and humidity conditions may have affected these. However, the fertigation treatments did not affect any of these nectar characteristics but instead affected the floral resources by affecting flower number.

As the fertigation strength increased from the low to high treatments, the number of flowers per plant increased, suggesting that higher fertigation strength is a way of increasing the nectar available for pollinators. Although this appears to be promising, the plant needs to be able to support the development of the extra flowers into fruits, without limiting fruit size, which can occur when resources are limited. This problem can be avoided by using practices that ensure crops are growing optimally, such as conducting leaf mineral analysis in summer after harvest, to evaluate the nutrient status of the crop.

Following peak flowering, nectar was extracted from blueberry flowers for mineral analysis. Flowers and nectar were limited and only enough nectar was extracted for one sample each of the medium- and high-fertigation treatments. Nectar volumes were not sufficient to measure nitrogen, but potassium, calcium, manganese, copper, and zinc were detected in these nectars, with the other elements being below levels of detection. The potassium concentrations in nectar were low compared with reports for avocado and onion nectar, which can deter honey bees, but higher than those reported in citrus, which is highly attractive.

This work has highlighted the important role that microclimate can play in nectar production but also the potential effect that fertiliser practices, or other practices such as pruning, can have on crop floral resources.

A blueberry flower with nectar droplets visible on the stigma and inside the floral tube

Leanne Davis, Technical Officer (NSW Department of Primary Industries) in the experiment at Wollongbar, NSW, where nectar from blueberry flowers was extracted for analysis of sugars, and chemical elements in some samples (right).

Fertigation modified the nutrient status of plants which affected flower production but not nectar characteristics.

3.5 University of New England

Improve the performance of honey bee colonies deployed under covers through orchard manipulation

The results presented below represent a summary of activities for activities 2 a-d (including KPI 5.2d)

Researchers from UNE have evaluated the effects of the different cover types on insect visitation and yield using polythene and bird-netting covers over blueberries and apples.

Results suggest that while polythene reduces bee visitation compared to bird-netting, in some circumstances this lower visitation rate may still be sufficient for optimal yields. In these situations, polythene covers may provide a yield advantage over bird-netting, independently of insect visitation, but this effect may depend on cultivar, climate, or other factors.

Effect of blueberry cover type on insect visitation, pollen flow and blueberry yield

Over 22 hours of observations, 563 floral visits were recorded to Snowchaser and 356 to Arana, with honey bees being the dominant visitors to Snowchaser (94.8%) and stingless bees to Arana (56.7%). Visitor proportions varied between the two cover types; stingless bees visited more under bird-netting than under polythene for both cultivars. Insect visitation rates were higher on average under bird-netting than polythene across both cultivars, but yield effects varied by cultivar. For Snowchaser, total berry yield was 17% greater under polythene compared to bird-net, in contrast, Arana yields under bird-net were greater (39%) than under polythene (Figure 65). For Arana, there were no significant differences in yields between pollinator excluded plants and plants exposed to pollinators under polythene, and for both cultivars, there was a trend of higher yield under polythene compared to bird-net for pollinator excluded plants. This suggests that polythene provided a yield benefit independently from pollination, but that this effect is context specific, and depends on pollinator density and cultivar-specific pollinator dependency traits. Benefits of polythene are likely to be greatest when insect visitation rates are already optimal for pollination, whereas if insect visitation rates are suboptimal, higher visitation rates under bird-net may result in greater yields than polythene.

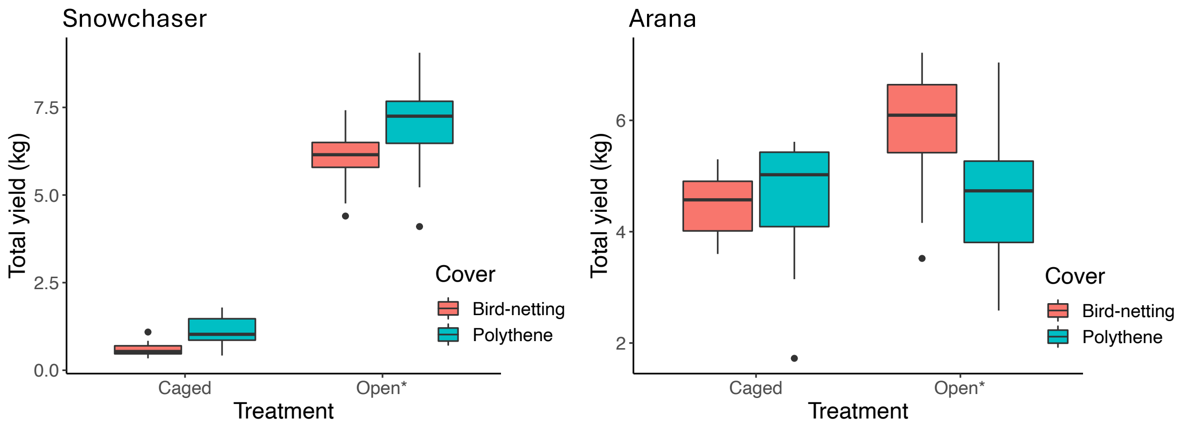


*Figure 65. Total berry yield for Snowchaser (left) and Arana (right) blueberry varieties under two different cover types, for caged (pollinator excluded) plants and plants open to pollinators (*pooled for open and partially enclosed plants).*

Effects of different protected cropping structures on pollen flow and composition on blueberry stigmas

We counted a total of 41,779 pollen grains on 840 blueberry stigmas, finding conspecific pollen amounts affected by netting type. Unnetted blocks had the highest conspecific pollen counts (96.67±49), followed by partially netted (58.88±48) and fully netted blocks (18.30±45). Additionally, 31 pollen morphospecies from 20 families were identified on blueberry stigmas, with netting significantly affecting heterospecific pollen deposition. Unnetted blocks had the highest heterospecific pollen counts (17.85±16), followed by partially netted (9.55±14) and fully netted blocks (2±13). While netting impacted pollen deposition, mesh size did not correlate significantly. Blocks transitioning to fully netted showed reduced pollen counts (Figure 66).

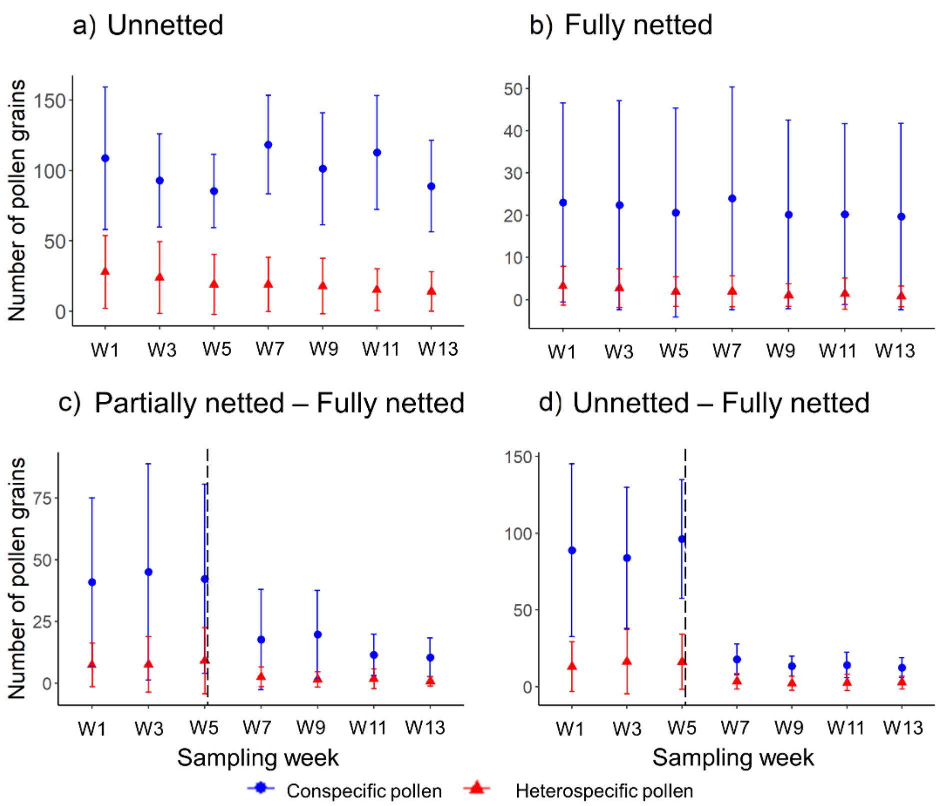


Figure 66. Number of conspecific (blue) and heterospecific (red) pollen grains observed on the stigmas throughout the blueberry flowering season in (a) unnetted (2 blocks), (b) fully netted (5 blocks), (c) partially netted changed to fully netted (4 blocks) and (d) unnetted changed to fully netted blocks (3 blocks) in each of the 7 sampling rounds. Dashed black lines indicate when the netting was changed.

Stigmas at the edge of the block received more conspecific and heterospecific pollen compared to those in the middle. In unnetted blocks, edge stigmas had significantly more conspecific pollen (103±42 grains) than middle stigmas (89±40). Similarly, edge stigmas had more heterospecific pollen (24±21 grains) than middle stigmas (11±15). Partially netted blocks showed no difference in conspecific pollen between edge and middle stigmas but had significantly more heterospecific pollen at the edge (14±17 grains) than in the middle (5±7). Fully netted blocks had the lowest pollen amounts, with only heterospecific pollen significantly differing between edge (3±4 grains) and middle (1±3 grains). Conspecific pollen showed no significant difference between edge and middle stigmas in fully netted blocks (Figure 67).

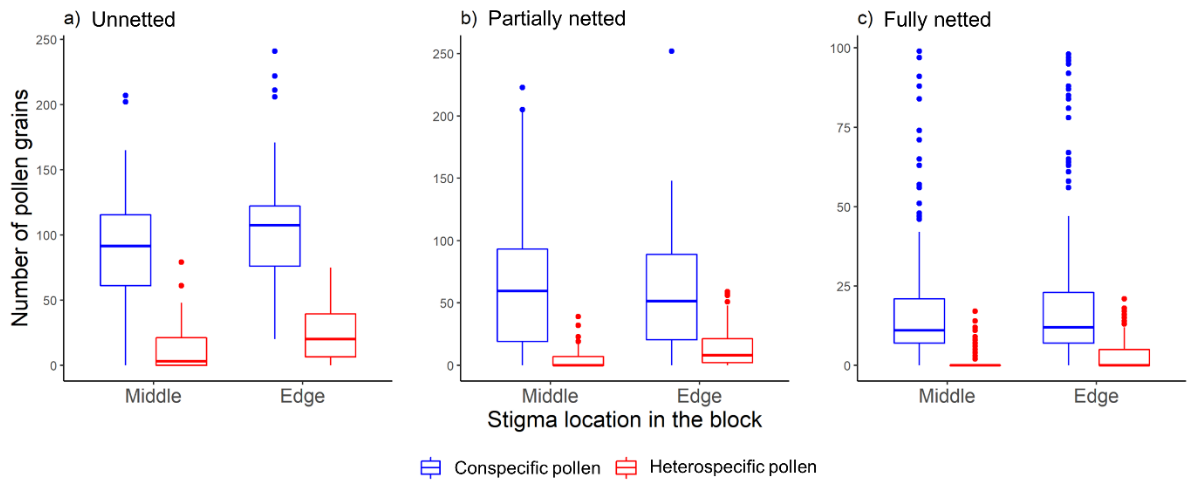


Figure 67. Amount of conspecific (green) and heterospecific (red) pollen grains on stigmas in the middle and at the edge of a) unnetted, b) partially netted, and c) fully netted blocks.

Determine whether modifying the length, tunnel type or configuration of tunnels improves hive performance and/or pollination in covered blueberries (5.2b)

Tunnel length results show that plants in shorter tunnels have higher pollinator abundance (Figure 68). These findings suggest that blueberry yields may be increased by using an optimal tunnel length and cover type for a given blueberry cultivar.

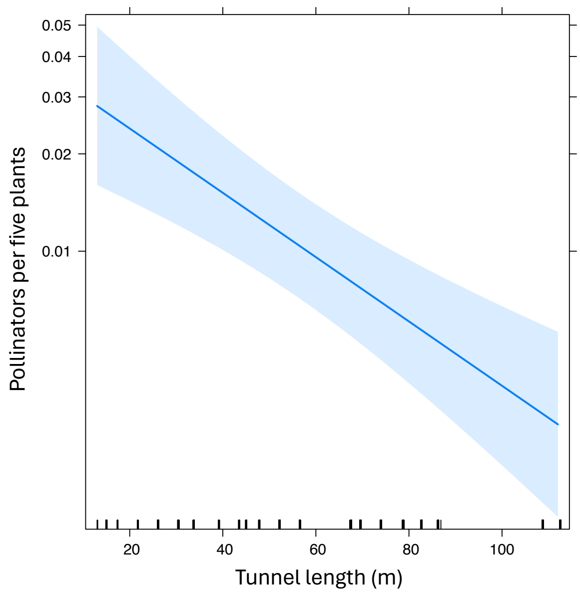


Figure 68. Relationship between pollinator density and blueberry polythene tunnel length.

Blueberry yields may be increased by using an optimal tunnel length and cover type for a given blueberry cultivar.

Determine whether opening up the sides of netting enclosures improves hive performance and/or pollination in covered apples (5.2.a)

During 52.5 hours of observations, 4,704 insect visits to apple flowers were recorded, with honey bees accounting for 94% of these visits. Insect flower visitation was higher in open blocks compared to netted blocks (Figure 69a). For the netted blocks, there was a positive relationship between the amount of clear space above trees and floral visitor abundance (Figure 69b). Trees under rows that had temporarily opened overhead netting had higher floral visitor abundance compared to trees under inter-row netting that had remained closed during bloom (Figure 69c).

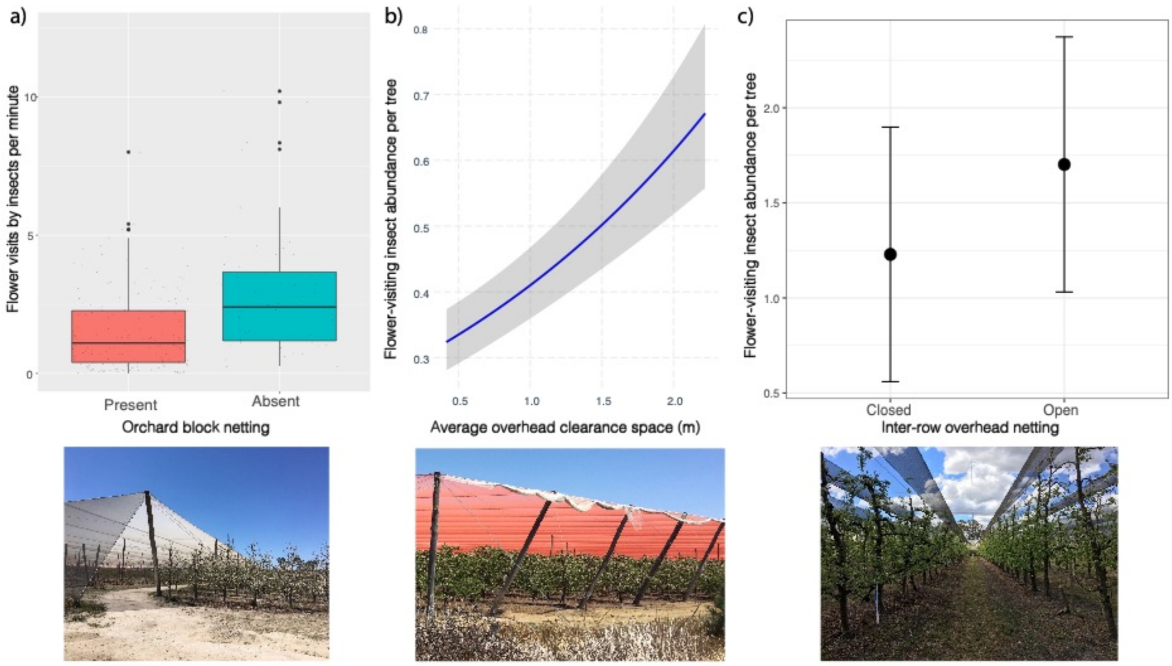


Figure 69. Comparison of insect visitation between open and netted orchard blocks (a); abundance of floral visitors in relation to the space between the tops of trees and the nets overhead (b); and the abundance of floral visitors in rows with inter-row net openings versus rows without openings.

Data on the relationship between height above apple canopy on honey bee visitation showed that the higher the netting above the canopy and the more open areas above the canopy (where netting has been drawn back), the greater the bee visitation to flowers.

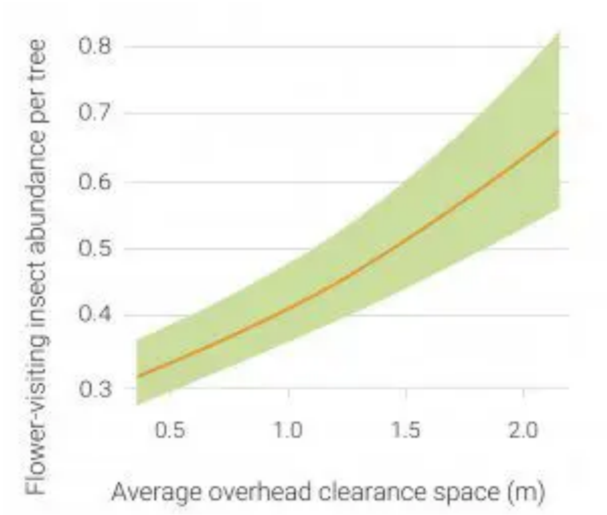


Figure 70. Insect visitation to apple flowers in relation to the overhead clearance space in blocks.

Higher overhead net clearance (i.e. more space between crop canopy and overhead net height is better for insect visitation

Effect of protective covers on bee colony development and access to resources

To determine the impact of protective cropping structures on honey bee brood production and resource storage, 14 honey bee colonies were placed inside and outside of protective covers on four blueberry (*Vaccinium corymbosum* and *V. virgatum*) farms of northern NSW, Australia. We measured changes in brood production, pollen and honey storage, hive weight and ambient hive temperature fortnightly over a 12-week period.

Brood production, pollen and honey storage, and hive weight all decreased within four weeks of placement under net cover Figure 71. Although brood and honey metrics gradually increased in all hives during the remainder of the 12-week monitoring period, hives located under protective netting had relatively smaller gains in brood production and pollen storage when compared to hives located outside the protective covering. Hives under protective netting that were in the shade and/or whose entrances were facing south were most impacted by the end of the monitoring period. Additionally, bees took significantly longer to exit from blocks covered by hail nets, averaging 12.65 seconds, compared to just over 9 seconds for bird nets, and had greater success in exiting when the cover was bird netting (Figure 72). Furthermore, the incidence of bees contacting and rebounding off the netting was significantly higher in the context of hail netting (mean = 9.0 contacts, SD = 1.3) than with bird netting (mean = 4.6 contacts, SD = 0.4) during individual flight observations.

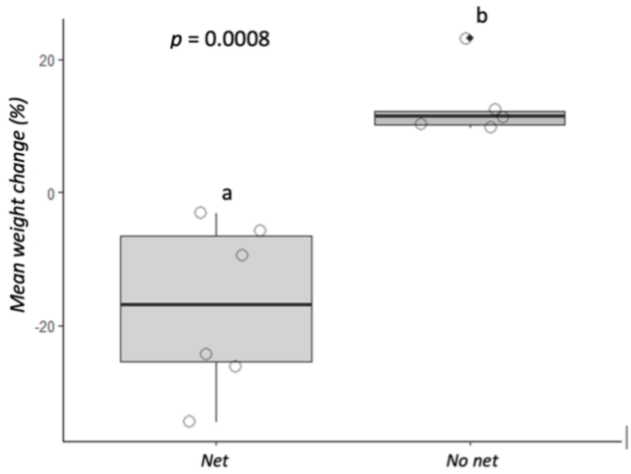


Figure 71. Percentage weight change in hives from initial to final monitoring events for hives inside or outside of netted blueberry blocks.

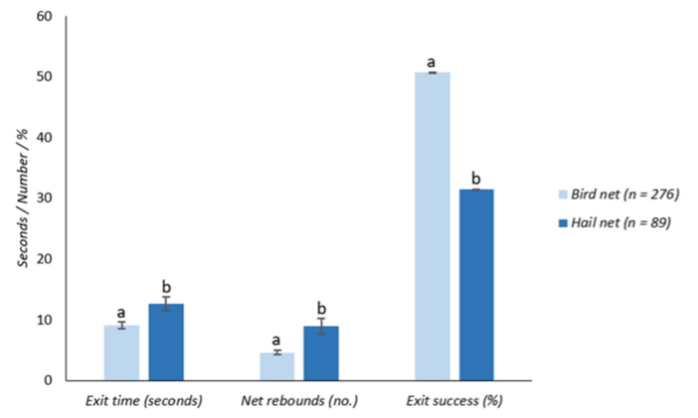


Figure 72. Bee exit time (seconds), rebound incidents and percentage of bees that successfully exited the netted environment.

Hive health can be negatively affected by placement under netting, especially if the hives are south facing and shaded.

Investigate how non-crop floral resources and other features of the orchard floor affect pollinators under protected covers

Effects of co-flowering plants on blueberry floral visitation and berry weight (5.4.b,c)

Basil plants in blueberry orchards were frequently visited by honey bees and stingless bees, indicating basil could serve as a complementary pollen resource, enhancing pollinator diets and potentially reducing foraging distances. However, Mexican heather was less attractive to pollinators, suggesting it is a poor complementary forage plant. Added co-flowering plants did not affect blueberry visitation at the row scale, but at the plant neighbour scale evidence of a negative impact on blueberry visitation was found. The presence of added plants led to higher conspecific pollen deposition on blueberry flowers late in the bloom season, indicating a potential benefit to pollination when blueberry flower availability was low. Nonetheless, there was no significant impact on blueberry fruit weight from the presence of adjacent basil plants, suggesting limited direct benefits to crop yield from these pollinator support strategies (Figure 73).

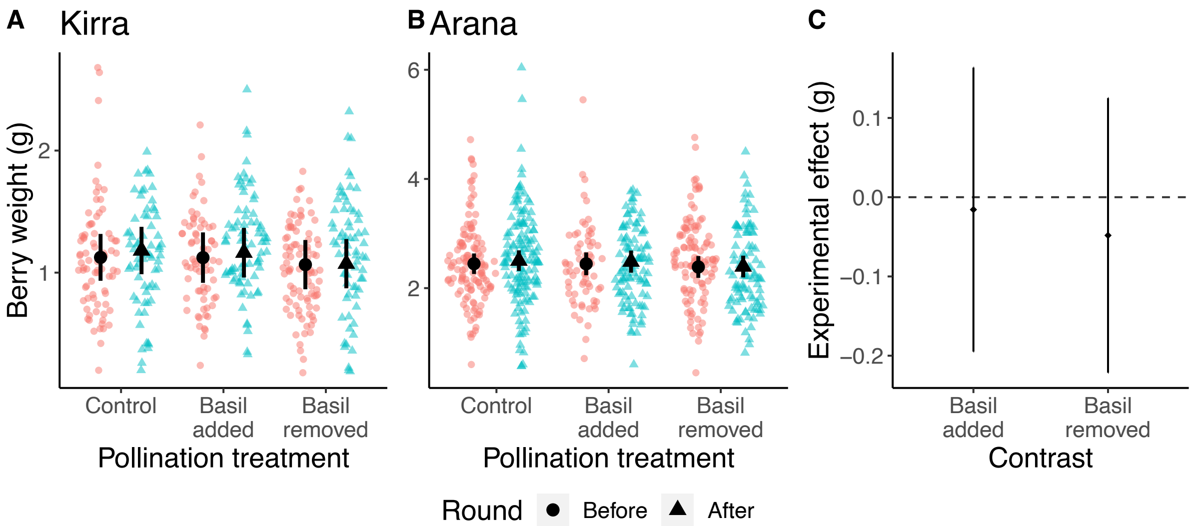


Figure 73. Estimated open-pollinated berry weight marginal means across the two periods for Kirra(a), Arana (b), and experimental effect for both cultivars (c). Coloured points represent individual berry weights (jittered). Solid lines represent 95 % confidence intervals. The dashed line (c) represents no experimental effect on berry weight.

Assess the importance of varietal pollen presence and proximity for blueberry and apple production under covers

Blueberry: Fluorescent dye was used on flowers to determine how the spacing of blueberry plants of two different cultivars impact pollen flow. Comparing single rows of each cultivar to rows with mixed cultivars, the researchers found that single cultivar rows result in little pollen transfer from other cultivars. Rows in which blueberry plants were spaced in a 1:3 ratio within the same row provided the highest pollen flow across different cultivars.

Apple: Honey bees more often moved pollen from a single cultivar after a single visit and generally only deposited pollen from other cultivars after multiple visits to a given flower (Stavert et al. 2020, Lobaton et al. 2021).

Pollen flow was impacted by the configuration of the netting over apple orchards. From the three different netting treatments (open, pitched and flat), blocks with pitched netting accounted for 59% of the total pollen flow, blocks with flat netting accounted for 27%, and blocks with no net present (open) accounted for only 14%. When pollen movement was compared along versus across the rows, pollen flow along the rows was significantly higher than across rows for all netting treatments (87% of the total flow was along the rows as opposed to across rows) (Figure 74). When tracking honey bee foraging behaviour, individual bees tended to stay on the same row for more than 1 minute until it would fly away, but rarely moving to one of the rows next to where they were foraging.

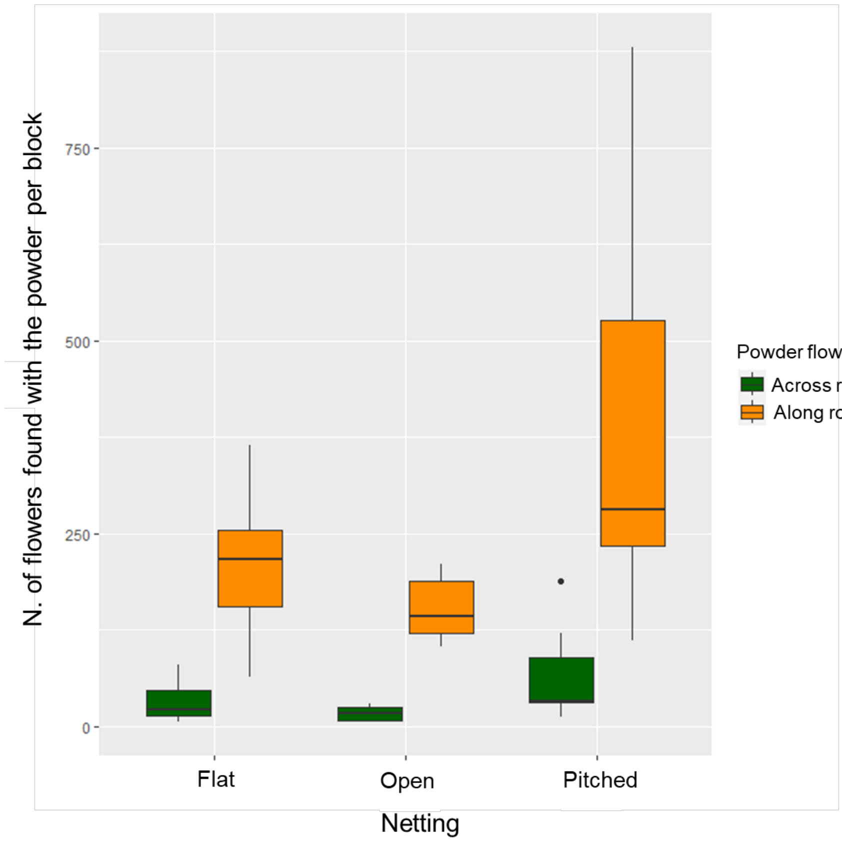


Figure 74. Number of flowers found with the inflorescent powder across (green) and along (orange) painted rows per block under different netting treatments (flat, open, and pitched).

Summary of outcomes

1. Bird-netting is better for bee foraging activity than polythene tunnels.
2. This means industry could focus on bird netting rather than polytunnels if cultivars used are highly pollinator dependent. For self-pollinating cultivars, this is not as important, although fruit size and quality can be increased with insect pollination.
3. Honey bee hives placed under protected covers (i.e. bird netting and polytunnels) impacted honey bee colony performance relative to those placed in uncovered positions outside netting. Hive weight and pollen storage was lower in hives under nets than outside nets.
4. Netting height above apple plants is important to bee movement in apple orchards, higher netting is better.
5. Blueberry pollen abundance on stigmas was reduced by up to 81% under full netting and 36% by partial netting, relative to open, uncovered blueberry plants. This means varieties that are highly pollinator dependent would receive highest pollen transfer in uncovered systems. Varieties are less pollinator dependent will be less impacted by netting.

3.6 University of Tasmania

Understand floral biology to inform development of mechanical and bee-vectored pollination to optimise productivity

Conduct field experiments to characterise pollen quality and nectar quality of onion and sweet cherry flowers (5.1.a)

Pollen collection

Sweet cherry

In each of the three seasons, it was possible to harvest approximately 40-50 kg of fresh flowers over 8 days. These samples were processed with a recovery rate of ~1.6% dried raw pollen/anthers.

Onion

For onions, a portable vacuum unit fitted with cyclone separators proved to be most efficient for pollen collection. Yields from the vacuum unit were comparable with those achieved with hand harvesting (rubbing dried inflorescences on a screen), confirming the efficiency of the vacuum method. A tumbling barrel unit was highly ineffective.

Optimum vacuum head air velocity was 12-16km/hr. Modifications to the cyclone separator were needed to ensure that pollen was not damaged at greater vacuum head air velocities. The best time of day to harvest pollen for yield and quality was midday (compared to morning and afternoon).

Repeated harvests over a 10-day period (in-situ or cut umbels held at 20°C and 55% RH) optimised pollen yield per umbel (30mg of pure pollen per umbel ~0.1g/plant). Using the harvesting equipment and protocols developed in this project, we can harvest 25 grams of pollen per hour, per operator, with average viability of ca. 30%.

Evaluating pollen quality

Sweet cherry – in vitro

When assessing in vitro methodologies, incubation temperatures of 20 and 25°C yielded the highest germination rates (Figure 75). There was no evidence that boric acid affected pollen germination rate or tube length, however sucrose concentrations at 10%, 15%, and 20% gave the highest germination rate. The addition of Tween 80 significantly reduced pollen clumping by up to 98%, with no reduction in pollen viability.

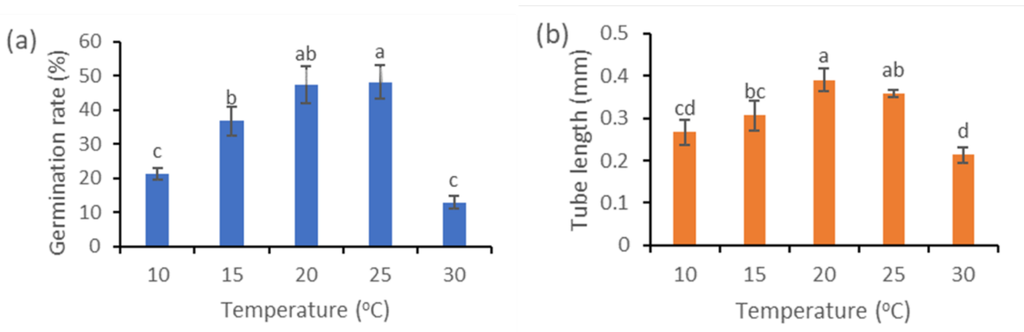


Figure 75. Effect of incubation temperature on in vitro pollen germination rate (A) and tube length (B) of Regina. Bars = standard errors, n = 3, letters indicate significant differences between treatments.

Overall, pollen viability was higher in agar medium when compared to liquid medium. Pollen germination ranged from 20 to 80%, depending on variety and season. Viability of the main cultivar, ‘Lapins’, varied with season, and orchard, ranging from 30 to 55% (Figure 76). Kordia and Regina cultivars typically had the lowest germination (15-20%), regardless of year, which was consistent with previous studies (Radičević et al. 2013).

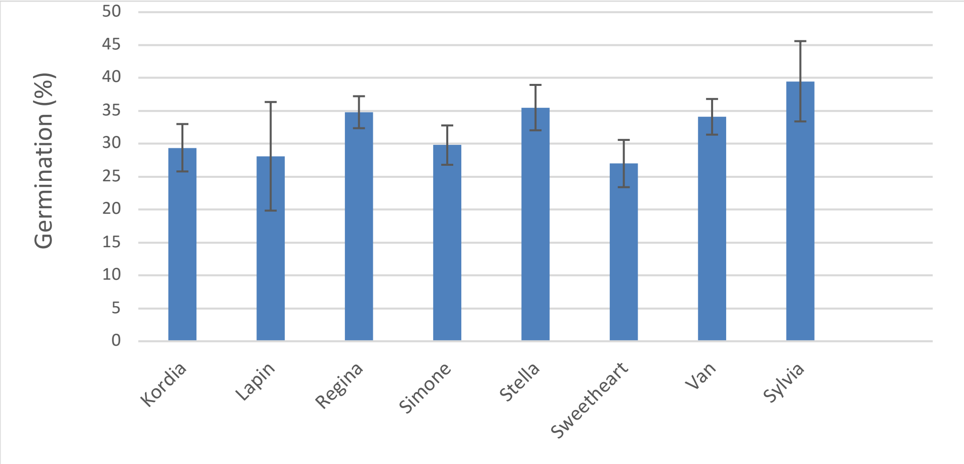


Figure 76. Effect of variety on average in vitro pollen germination rate. Bars = standard errors, n = 3

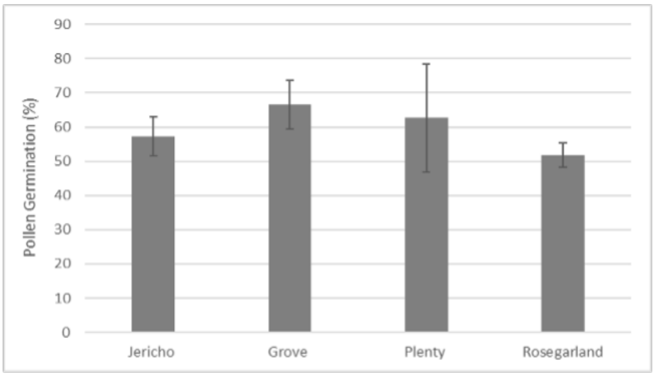


Figure 77. Effect of location on average in vitro pollen germination rate. Bars = standard errors, n = 3

Sweet cherry – in vivo

The optimised in vivo method confirmed that it was possible to fertilise fresh flowers of commercial varieties with processed pollen that had been stored at -20°C for up to one year. This was achieved with and without a sucrose carrier on detached branches. Using fluorescence microscopy, it was possible to track the pollen tube down the style to enter the ovule (Figure 78).

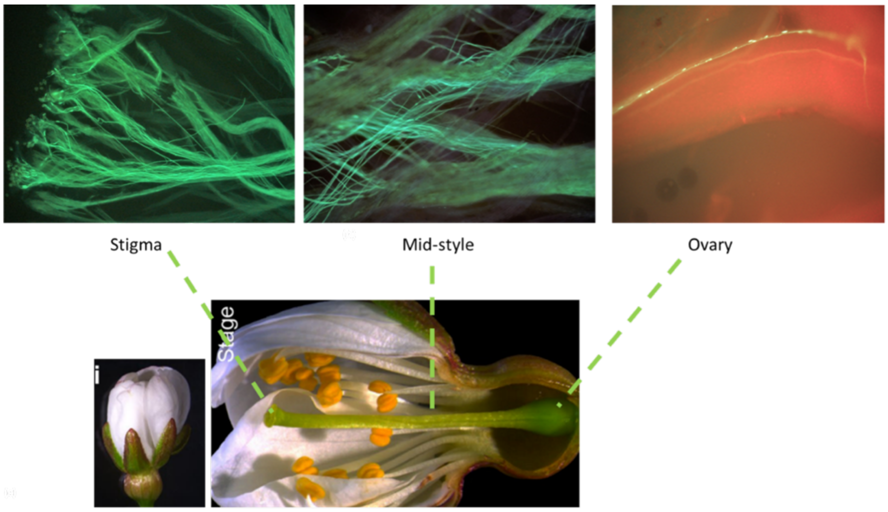


Figure 78. In vivo germination of fresh flowers confirmed that pollen that had been prepared for mechanical pollination were viable, as confirmed by the growth of a pollen tube down the style into the ovule.

Onion – in vitro

Optimal pollen germination media was composed of 20% sucrose in BBK media agar. Inclusion of Tween 80 at assessment stage was beneficial for separating pollen tubes growing on the media, while inclusion of aniline blue greatly improved visualisation of pollen tubes. Pollen commenced germination within 2hrs or application to the media and completed germination within 12hrs.

In vitro and in vivo trials demonstrated that harvested pollen was viable, and could be used to mechanically pollinate flowers

Pollen handling and storage

Sweet cherry

Large-scale purification of raw pollen from fresh flowers was achieved, first by milling and then by multiple sieving. Assessment over two seasons showed that the milling prototype had variable effect on pollen quality, depending on cultivar. The cultivars Van and 'Lapins' were found to have highly robust pollen, whilst Tamara was highly susceptible to milling damage (no germination after third milling) (Figure 79, Figure 80).

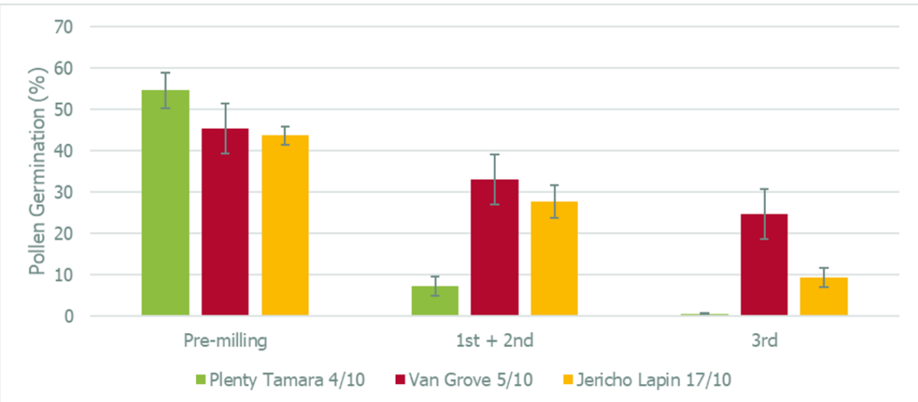


Figure 79. Impact of milling of pollen viability of three sweet cherry cultivars (Tamara, Van, and' Lapins') harvested from three locations (Plenty, Grove and Jericho) tested in 2022.

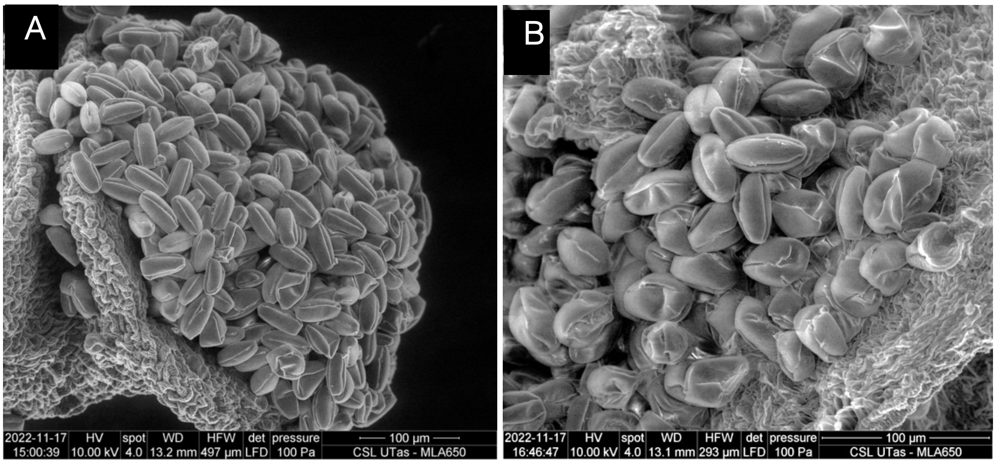


Figure 80. Electron microscope images of A)- pre-mill and B)- 3rd mill pollen of Tamara pollen

Viability of pollen from the main pollinator variety 'Lapins' varied significantly between different locations, as well as at different stages of the milling and purification process.

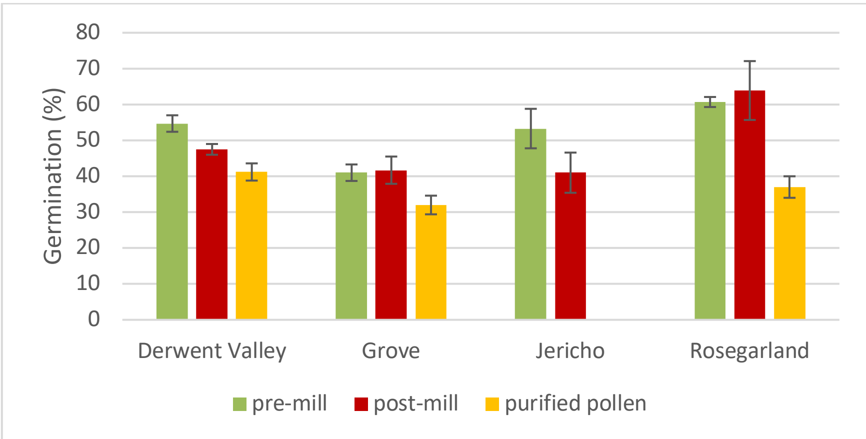


Figure 81. Viability of 'Lapins' pollen after milling and sieving; pollen was harvested from four orchards in Southern Tasmania in 2021.

Purified pollen was shown to be viable for over 12 months post-harvest at both 20 and 80°C (Figure 82). The literature suggests that long-term storage of pollen is influenced by moisture content (Towill 1985), with an optimal level of 11%. However, the results from this project found the optimal moisture content of sweet cherry pollen to be 6-7% for storage (Figure 83).

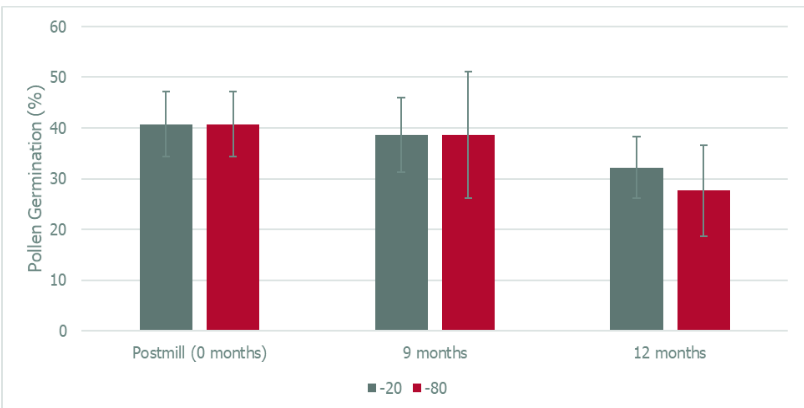


Figure 82. Purified pollen remained viable for over 12 months when stored at both -20 and -80°C.

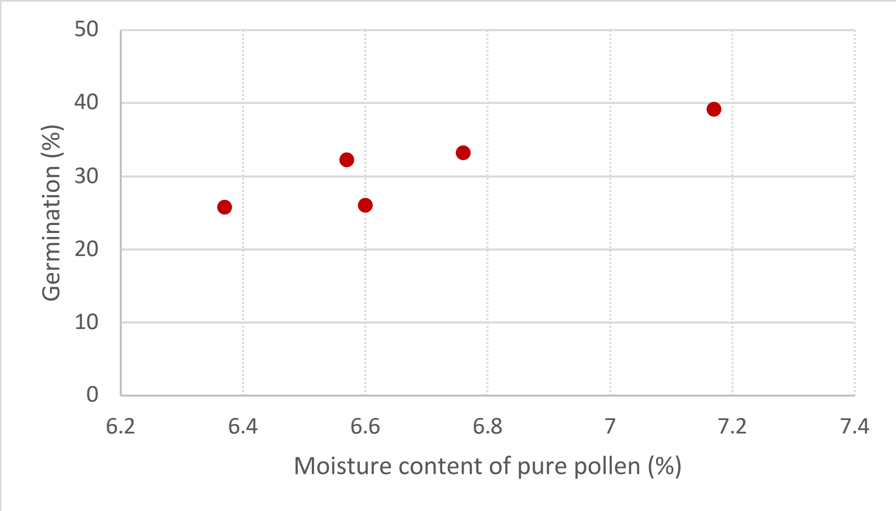


Figure 83. Optimal moisture content of dried anthers/pollen after desiccation at 20°C for around 2 days drying.

Onion

Onion pollen dried over silica gel for 48hours at 20°C to 7% moisture content immediately following harvest, has successfully been held at -18°C or -80°C for more than 12 months without a detectable loss in viability. At higher temperatures (4 to 20°C), loss of pollen viability was observed after relatively short periods of storage (2 to 4 weeks). Rehydration of stored pollen (30 minutes over a water bath at 20°C) was shown to significantly improve pollen germination rates after storage.

The milling process used to extract pollen from sweet cherry flowers had greater negative impacts on viability for some varieties than others, with pollen from Van and 'Lapins' more robust than that fromTamara.

Pollen viability for both sweet cherry and onion was unaffected by 9 months storage at -20°C, and only slightly lower when storage extended to 12 months. There was no benefit in reducing the storage temperature to -80°C

Pollen delivery

Sweet cherry

The field trials conducted at up to four commercial sweet cherry orchards in Southern Tasmania are summarised in
Table 5.

Field trials in 2021/2022 (eight field trials) and again in 2022/2023 (four field trials) indicated that supplementary pollination did not improve fruit set or individual fruit weight at the tree level. While the lack of improvement in 2021/2022 could be attributed to the extremely wet spring, supplementary pollination also failed to improve yield in 2022/2023, despite a typical spring.

These results suggest that further refinements are needed to optimise pollen delivery (Figure 84). In 2023/2024 refinements to the pollen:carrier ratios and spray volumes led to yield increases of approximately 30-50% in experiments conducted at the branch level. Further work investigating the role of electrostatic charge in attaching the pollen to the stigma is recommended.

Table 5. Summary of the major field trials conducted from 2019 to 2024 for sweet cherry commercial orchards

Season	Objectives of the field trials	Main results
2019/2020	Understanding pollination biology of sweet cherry Developing protocols and processes of mechanical pollination (focus on assessing pollen collection and in vitro quality assessments)	Identified genetic variation and environmental interaction in sweet cherry pollen quality (germination percentage and pollen tube growth)
2020/2021	Hand pollination of Kordia and Regina (branch-level design) (6 field trials) Developing protocols and processes of mechanical pollination (focus on pollen handling)	Controlled hand pollination field trials confirmed it was feasible to use processed and stored sweet cherry pollen to pollinate selected main cultivars.
2021/2022	Supplementary pollination of Kordia and Regina (tree-level design) (8 field trials) Developing protocols and processes of mechanical pollination (focus on pollen handling, delivery and storage)	The spring of 2021 was unusually wet with the rainfall nearly double to that of the long-term rainfall for Sep/Oct at Huonville and Jericho. Supplementary pollination improved overall tree yield of Regina at one site but had no effect on Kordia. Increased yield did not appear to affect individual fruit mass
2022/2023	Supplementary pollination of Kordia and Regina (tree-level design) (8 field trials) Developing protocols and processes of mechanical pollination (focus on in vivo pollination) Special field trial to use DNA analysis to confirm that supplementary pollination worked.	Despite setting up 8 field trials, only 4 were harvested due to factors beyond the control of the project (e.g. accidental harvests, wet weather during flowering for one orchard). Supplementary pollination did not significantly impact on fruit set, tree yield or individual fruit weight. The DNA analysis confirmed Tamara was the pollen donor of 3 out of the 290 seeds tested for Regina.
2023/2024	Supplementary pollination of Kordia and Regina (branch-level design) (1 field site with 150 branches) Refine protocols and processes of mechanical pollination (focus on pollen handling and delivery) Continuing DNA analysis to confirm that supplementary pollination success.	Fruit was harvested in January 2024. Yield increases of approximately 30-50% were recorded at the branch level.

Figure 84. Photo of cordless hand-held electrostatic sprayer (TPSHKE) used to apply pollen

Supplementary mechanical pollination failed to improve yield of sweet cherry in the main field trials. However, smaller trials with a refined protocol increased yield by up to 50%, suggesting that mechanical pollination may have significant yield benefits if an effective application method can be developed.

Onion

Field trials conducted on onions are summarised in

Table 6. Initial field application trials were conducted in a commercial onion seed crop in Southern Tasmania in January 2022. The electrostatic sprayers described for sweet cherry pollination were shown to be effective in delivering pollen in liquid carriers to onion flowers.

Two supplemental applications of pollen to receptive umbels within a four-day period increased pollination rates (% of pollinated flowers and pollen grains per stigma), increasing seed yields by up to 18% compared to control umbels (natural pollination only). No effects of supplemental pollination on seed size were observed.

Data collected in the field trial demonstrated the importance of timing of pollen application, with early morning (6am) applications proving more effective than mid-day applications. This result may reflect laboratory data on impaired pollen performance if the carrier media desiccates before pollen germination.

Table 6. Summary of the major trials conducted from 2019 to 2024 for onion

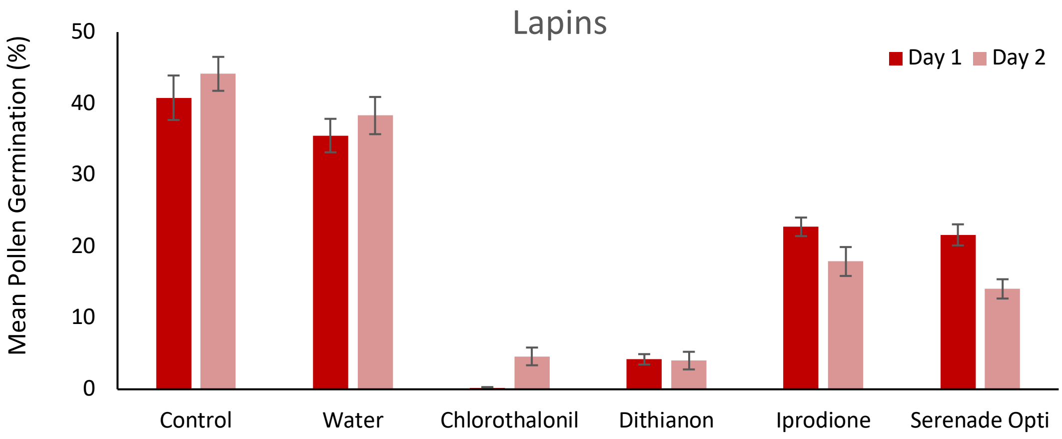
Season	Objectives of the trials	Main results
2019-2021	Preliminary laboratory work to develop carriers and application protocols.	Commercially available liquid and dry pollen carriers for MP were shown to be unsuitable for onion pollen. In in-vitro and in-vivo tests, pollen delivered in suspension germinated over a 2-hour period, but germination was inhibited if the carrier solution dried out. Liquid carrier candidates were narrowed down to polyethylene glycol, calcium nitrate, mannitol and sucrose solutions, each at two concentrations to cover an osmotic range. Similarly, dry carrier candidates were condensed to xanthan gum, brewer's yeast and lycopodium spores Optimal pollen:carrier concentration for liquid suspensions was 1:100 W:V.
2021/2022	Preliminary trial of candidate liquid carriers Evaluation of time of day of application	The electrostatic sprayers described for sweet cherry pollination were shown to be effective in delivering pollen in liquid carriers to onion flowers. Two supplemental applications of pollen to receptive umbels within a four-day period increased pollination rates (% of pollinated flowers and pollen grains per stigma) and also increased seed yields by up to 18%, compared to control umbels (natural pollination only). Data collected in the field trial demonstrate the important of timing of pollen application, with early morning (6am) applications proving more effective than mid-day applications. This result may reflect laboratory data on impaired pollen performance if the carrier medium desiccates before pollen germination.
2022/2023	Optimise liquid carrier options for pollen delivery in hybrid onion crop Preliminary trial of dry carrier options for pollen delivery in open-pollinated onion crop	PEG presented as the most effective liquid carrier to maintain pollen viability at ~30% (comparable to control). Xanthan gum present as most effective dry carrier option. Pollen delivered in dry carriers (applied by hand) was more effective than liquid carriers for setting seed. Yields were limited by relatively low deposition for both dry and liquid carriers.
2023/2024	Evaluate electrostatic application to improve pollen deposition Develop delivery technology and protocols for dry carriers	Pollen was successfully applied and yield responses will be recorded in March 2024.

Additional complementary experiments

Effect of fungicide on pollen viability

Treatment with Dithianon, Captan and Fenhexamid significantly reduced pollen viability for an unknown *Prunus* spp., apricot (*Prunus armeniaca*) and sweet cherry flowers by approximately 40%, 20% and 29% respectively, as compared to the control.

Interestingly pollen viability of flowers that had been closed during fungicide application was ~50 % higher than for open flowers. Work is ongoing in optimising the best analytical method to quantify fungicide residue concentrations in flowers and bees.



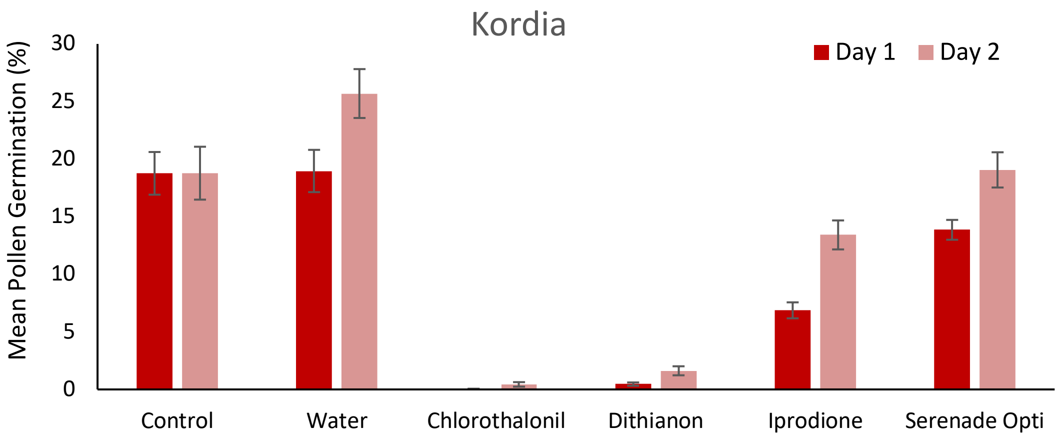


Figure 85. Effect of fungicide treatment on the viability of pollen from fresh sweet cherry flowers on detached, watered branches of 'Lapins' and Kordia

Common fungicides, as well as the biological product Serenade® Opti, significantly reduced pollen viability of 'Lapins' and Kordia one day after application, with mixed impacts after two days.

DNA methods for S-allele analysis of sweet cherry

In order to understand the relative contribution of pollen cultivars (specifically those applied using artificial means) to crop yield, a quantitative PCR (qPCR) method was optimised for S-allele analysis of sweet cherry leaves and pollen. The qPCR method developed was highly sensitivity, as evidenced by low detection limits for pollen, with the equivalent of 8 to 23 pollen grains in 2 μL of DNA.

This project also unequivocally determined the S-allele profile of two commercial cultivars (Simone and Sweet Georgia) that had not been genetically catalogued for the S-locus. DNA sequencing revealed that both cultivars have S1 and S4' alleles. Ongoing parental DNA analysis of ~400 individual sweet cherry seeds from commercial field sites collected over two seasons confirmed the identity of the pollen donors for the main cultivars of Kordia and Regina. This identification of effective pollinisers will assist farmers to optimise orchard design, particularly in the selection and introduction of new cultivars.

3.7 Contribution to program objectives

Refer to Appendix 7.3 for full details on monitoring and evaluation

4 Collaboration

<table>
<tr><th>UoA</th><th>COLLABORATOR</th><th>DETAILS</th></tr>
<tr><td colspan="3">Dr Hogendoorn and her team at UoA collaborated with 14 apple growers and provided annual presentations to the Protected Cropping Association and the Fruit growers SA (Suzie Green and later Jose Gil).

Apple growers were consulted about the information they wanted to get out of the project, both during our annual presentations to the Fruit Producers SA, and on an unofficial basis during visits to the orchards. They allowed us ample access to the orchard. In particular, Andy Flavell, Robert Green, John Vickers Joe Ceravolo and Joel Brockhoff were very helpful. They opened or closed the nets when asked, provided flowering branches when the mobile polliniser units were not flowering in time, and allowed the use of their tractor/gator when the orchard was too wet for the university's truck. They also allowed the team to pick all the apples needed for assessments.

They also worked with the hives of five beekeepers and we thank them for their trust. The beekeepers allowed them to open the hives for assessments and/or place pollen traps on the hives. They also notified the team when the bees were going in and when they were going to come out, which was important for the timing of the assessments.

The polliniser trees were grown and maintained by Balhannah Nurseries.

UoA enjoyed the academic collaborators in this project, in particular Dr Lisa Evans and Dr Romina Rader.

We are grateful to Ashley Zamek for project management and APAL and PCA, for their invitations to present at conferences and grower days.

Work in apple orchards will continue as a PhD student will be starting in 2024 to investigate biological control agents under net.</td></tr>
<tr><td rowspan="13">Dr Katja Hogendoorn</td><td>Dr Scott Groom</td><td>University of Adelaide</td></tr>
<tr><td>Dr Jay Iwasaki</td><td>University of Adelaide</td></tr>
<tr><td>Dr Michael McLeish</td><td>University of Adelaide</td></tr>
<tr><td>Nicholas Timbs</td><td>University of Adelaide</td></tr>
<tr><td>Nicholas Congedi</td><td>University of Adelaide</td></tr>
<tr><td>Elisabeth Williamson</td><td>University of Adelaide</td></tr>
<tr><td>Keely Bryars</td><td>University of Adelaide</td></tr>
<tr><td>Robbert Green</td><td>Apple grower, Cold store road, Lenswood SA</td></tr>
<tr><td>Matt Flavell</td><td>Apple grower, Stentiford Road Forest Range SA</td></tr>
<tr><td>John Vickers</td><td>Apple grower, Tiers road, Peacock road, Lenswood SA; Swamp Road, Forest Range SA</td></tr>
<tr><td>Joe Ceravolo -</td><td>Apple grower, Mappinga Road Woodside SA; Chambers Road, Nairne SA; Strathhalbyn Road, Echunga SA</td></tr>
<tr><td>Dean Nichols</td><td>Apple grower, Basket Range Road, Uraidla SA</td></tr>
<tr><td>Joel Brockhoff</td><td>Apple grower, Swamp Road, Forest Range SA</td></tr>
</table>

	Andrew Flavell	Apple grower, Schocroft Road, Mount Torrens SA
	Aaron Woolston	Beekeeper
	Troy Stidiford	Beekeeper
	Brock Jeffreys	Beekeeper
	Danny Le Feuvre	Beekeeper
	Kym Schultz	Beekeeper
	Balhannah Nurseries, Hartmann Road, Charleston SA	
PFR	COLLABORATOR	DETAILS
Dr Lisa Evans	Dr Melissa Broussard	Plant and Food Research
	Brian Cutting	Plant and Food Research
	Dr Mateusz Jochym	Plant and Food Research
	Sam Read	Plant and Food Research
	Franziska Schmidlin	Plant and Food Research
	Grant Fale	Plant and Food Research
	Max Buxton	Plant and Food Research
	Tama Nathan	Plant and Food Research
	Sam Neitherclift	Plant and Food Research
	Dr Paul Martinsen	Plant and Food Research
	Dr Tom Moore	Plant and Food Research
	General academic collaboration	Insightful and collaborative discussions were held with all project partners. Data was collected in collaboration with Dr Hogendoorn and her UoA team to help address regional differences in apple production in protected cropping environments.
	T&G Global	• Partnered with PFR to obtain further funding (from AGMARDT) to help expand the project work in NZ during COVID-19 when travel to and around Australia was not possible for our team • Supported research objectives by assisting PFR to connect with apple growers using protective covers • Attended annual project debriefs • Helped to promote project findings

	Australian Lychee Association	• Provided in-kind support during the program (farm access, grower time) • Members attend annual steering group meetings • Research presented at annual growers association meetings
	AGMARDT	• Provided additional funding to expand our protected cropping research
	Key growers	• Vailima Orchard Limited (Richmond NZ) • MA Orchards (Timaru NZ) • Daniel Nicoletti (Stanthorpe QLD) • Tristram Hoddy - Vailima Orchard Limited • Morten Tønder - MA Orchards • Daniel Nicoletti - Nicoletti Orchards • John Francis – Lychee grower • Bryan Pocock – Lychee grower • Chris Salta – Lychee grower
	Beekeepers	• Murray Elwood – Mountain Valley Honey • Shane Hutton – Huttons Honey
DPI	COLLABORATOR	DETAILS
Collaboration between NSWDPI and UNE groups provided a data set with broader inferences regarding the effects of protective covers on pollination in berries than would otherwise have been gained by the separate groups. Collaboration with the Mountain Blue farms group has led to a valuable area of research beyond the project – namely, developing a water use model for blueberry that will help growers improve water use efficiency in blueberry.		
Dr Sophie Parks	Melinda Simpson	NSW Berry Industry Development Officer, extension activities
	Madlen Kratz	NSW DPI Honey bee Industry Development Officer, extension activities
	Key growers	Mountain Blue Farms Group (Tabulam, Alstonville) including contacts Damian Clothier, Bep Pera and Ian Hendry Steve Fuller, Apiarist, Bee Services.
UNE	COLLABORATOR	DETAILS
In addition to collaborations among research partners, PhD students and postdocs employed under this grant, UNE researchers collaborated with industry groups and growers for research and extension activities.		
Dr Romina Rader	Costa Staff	Berry research
	OzGroup	Berry research
	Perfection	Berry research
	Apple and Pear Australian Limited (APAL)	Research extension and grower discussions.

	Key growers	
	Agronomists	
UTas	COLLABORATOR	DETAILS
Industry partners were consulted included in the planning. Commercial production sites owned by the industry partners were utilised for all trials. The collaboration was extended to the broader sweet cherry and vegetable seed production specialists including growers, managers, agronomists and beekeepers. The team thanks Hansen Orchards (Nigel Bartel and Wayne Trengove) and Reid Fruits (Andrew Hall, Nick Owens and Brad Ashlin), and South Pacific Seeds (Craig Garland) for supporting the field trials. Their willingness to share their knowledge and experiences on all aspects of pollination was much appreciated. The team also sincerely thank Dr Morag Glen for her expertise and support of the sweet cherry DNA work.		
Dr Alistair Gracie	Hansen Orchards	Sweet cherry research
	Reid Fruits	Sweet cherry research
	South Pacific Seeds	Onion research

5 Extension and adoption activities

ORGANISATION	DATE	ACTIVITY	DETAILS
UoA – Katja Hogendoorn	5/8/2019 23/7/2021	Apple growers' workshops	In Lenswood to present the results of MPU and opening of netting experiments in August 2019 and hive health July 2021. Events attracted 30 and 23 apple growers and two consultants. Future activity: Fruit Producers SA to for final winter workshop in 2024.
UoA – Katja Hogendoorn	August 2021	Newsletter article	Fruit Producers SA's newsletter "Around the Orchard" titled "Orchard floor management and how to make the most of the wild pollinators of Apple https://heyzine.com/flip-book/d93edbe353.html#page/6)
UoA – Katja Hogendoorn	September 2019	Presentation	APAL Grower R&D day https://apal.org.au/protecting-our-pollinators/ Discussed project and contributed to its legacy in an interview at the APAL conference https://t.co/3aRar8iBAm
UoA – Katja Hogendoorn	September 2022	Presentation	APAL Grower R&D day 2022 https://t.co/3aRar8iBAm
UoA – Katja Hogendoorn	December 2021	Presentation	The conference of the Australian Entomological Society (Adelaide)
UoA – Katja Hogendoorn	March2021	Presentation	PCA Conference (Coffs Harbor)
UoA – Katja Hogendoorn	July 2023	Presentation	PCA Conference (Brisbane)
UoA – Katja Hogendoorn	June 2022	Presentation	The conference of the Australian Native Bee Association (Sydney)
UoA – Katja Hogendoorn	October 2023	Presentation	The conference of the International Commission for Plant Pollinator Relationships (South Africa)
UoA – Katja Hogendoorn	November 2023	Presentation	The conference of the Australian Entomological Society (Albany)
UoA – Katja Hogendoorn	August 2022	Presentation	AGM of the Fruit Producers SA, attended by ~50 fruit growers from the Adelaide Hills
UoA – Katja Hogendoorn	August – October 2022	Press release	The press release regarding the mobile polliniser unit has resulted in • A radio interview with the SA country hour (9 September 2021),

			• A radio interview with ABC North & West (4 August 2021) • An article in The Land (Home delivery for pollinators, 21 October 2021) https://www.theland.com.au/story/7459725/home-delivery-for-pollinators/).
UoA – Katja Hogendoorn	Duration of project	Project management	• Project management committee meeting (20 August 2021), which included industry representatives. Their assessment of the project's progress was very favourable and is available SLIDO Results - ST19000 - Project Management Meeting • Met with the evaluation team (15 March 2022) • Participated in the project meeting of (7 April 2022) and presented our progress. • Attended the project management meeting (19 July 2022) in Brisbane
UoA – Katja Hogendoorn	Throughout the duration of the project	Conversations with several apple growers about the project	

ORGANISATION	DATE	ACTIVITY	DETAILS
PFR - Lisa Evans	July 2019	Presentation	Costa PCA Conference, Gold Coast (Qld)
PFR - Lisa Evans	September 2019	Invited presentation: Honey bee health and foraging under netting	Berry Growers workshop (Tas) approx. 60 people present)
PFR - Brian Cutting	September 2019	Invited presentation	Lychee Pollination. Annual meeting of the Australian Lychee Growers Association, Sarina Beach Qld.
PFR - Lisa Evans	November 2019	Presentation followed by a panel discussion	on optimising pollination under covers at the APAL Grower R&D meeting in Melbourne
PFR - Lisa Evans	November 2020	Webinar	The future of horticulture and pollinators: how to ensure pollinator health in protected cropping systems, (Hosted by HI – Ashley Zamek , PFR – Lisa Evans, UoA – Katja Hogendoorn), 33 people participated
PFR - Lisa Evans and Melissa Broussard.	September 2021	Zoom workshop	1.5 hour zoom apple growers workshop conducted -Attended by all invited participants involved in research: 4 x industry representatives 5 x apple growers.
PFR - Brian Cutting	September 2021	Lychee growers workshop	Attended by approximately 50 growers and industry representatives. This presentation resulted in a related write-up in the Living Lychee industry newsletter.

PFR - Melissa Broussard.	August 2022	Zoom workshop	Apple growers workshop Attended by all invited participants involved in research including two industry representatives and three apple growers
PFR - Melissa Broussard	September 2022	Presentation	Attended by 12 industry representatives (T&G and Apples and Pears) Presentation was recorded by T&G and shared with T&G operational staff and growers, Apples & Pears, & M A Orchards operational staff
PFR Lisa Evans			• Project management committee meeting (20 August 2021), which included industry representatives. Their assessment of the project's progress was very favourable and is available SLIDO Results - ST19000 - Project Management Meeting • Met with the evaluation team (15 March 2022) • Participated in the project meeting of (7 April 2023) and presented our progress.
PFR – Lisa Evans, Melissa Broussard, and Brian Cutting	Throughout the duration of the project	Conversations with several growers about the project	
ORGANISATION	DATE	ACTIVITY	DETAILS
DPI – Melinda Simpson, Sophie Parks	March 2020	Trade show and workshop	Bee pollination expo with NSW DPI and growers
DPI – Sophie Parks, Madlen Kratz, L. Davis	Spring 2020	Magazine article	What's the attraction of honey bees to blueberry flowers? Australian Berry Journal, Edition 4 pp 83-84
DPI – Sophie Parks, M. Simpson, L. Davis	Spring 2021	Magazine article	The potential effect of flower shape on pollination success in blueberry. Australian Berry Journal, Edition 8 pp 37-40
DPI – Madlen Kratz, Sophie Parks, M. Simpson, L. Davis.	May 2021	Poster	Variation in floral morphology among blueberry varieties presented at the NSW Apiarists' Association conference, Tamworth.
DPI – Madlen Kratz and Sophie Parks	August 2021	Webinar	Supporting honey bees in your crop and a bit about our research on improving blueberry pollination under crop covers, hosted by the Protected Cropping Australia Webinar Series
DPI – Sophie Parks, M. Simpson, Madlen Kratz, L. Davis	August 2023	Magazine article	A tale of two cover types and the pollination of blueberry flowers by honey bees. Australian Berry Journal, Edition 12, 2022 pp 105-107
DPI – Sophie Parks	December 2021,	Presentation	The effect of covers (bird net and tunnel) on honey bee activity and pollination in blueberry: To Mountain Blue team (December 2021); at the Protected

	March and June 2022		Cropping Australia Conference in Coffs Harbour (March 2022), and at the 4th Australian Bee Congress in Sydney (June 2022)
DPI – Sophie Parks	May 2023	Report	The report, Honey bee pollination of blueberry flowers, which provides a research update on fertigation effects on floral nectar, was distributed at the Annual General Meeting of the NSW Apiarists Association in May 2023.
DPI – Sophie Parks	August 2023	News article	Rewarding pollinators with sweet-talking blueberry flowers. Australia's Honey bee News, volume 16 (4)
DPI – Sophie Parks	Throughout the duration of the project	Conversations with several growers about the project	
ORGANISATION	DATE	ACTIVITY	DETAILS
UNE - Jeremy Jones, Stephen Tancred, Karen Santos, Romina Rader	Winter 2021	Magazine article	Does orchard netting affect insect visitation to apple flowers in Stanthorpe, Queensland? Australian Fruit grower winter edition Magazine Article pg. 52-54. https://apal.org.au/wp-content/uploads/2021/08/AFG-2021-Winter-final-proof-v3.pdf
UNE – Romina Rader	June 2019	Presentation	Pollinators in Protected Cropping Systems: How to Move Forward. PCA conference (R. Rader hosted the pollination session)
UNE – Romina Rader	November 2020	Webinar	Pollination in Protected cropping. PCA webinar series
UNE – Romina Rader	August 2021	Web article	Does orchard netting affect insect visitation to apple flowers in Stanthorpe, Queensland? https://apal.org.au/does-orchard-netting-affect-insect-visitation-to-apple-flowers-in-stanthorpe-queensland/
UNE – Jeremy Jones	2021	Video presentation (YouTube)	Video presentation on insect visitation under apple netting for APAL YouTube channel https://www.youtube.com/watch?v=N1DglSuMGe8
UNE – Jeremy Jones	June 2022	Presentation	Protected cropping pollination of blueberries: Bird-netting is better for bees, but is polythene better for plants? PCA conference (Coffs Harbour)
UNE – Karen Bezerra da Silva	December 2022	Presentation	Impacts of protective nets on pollen flow in blueberry orchards. British Ecological Conference (Scotland, online)
UNE – Karen Bezerra da Silva	July 2023	Presentation	Protective nets reduce pollen flow in blueberry orchards. PCA conference (Brisbane)
UNE – Romina Rader	Throughout the	Conversations with several growers about the project	

	duration of the project		
ORGANISATION	DATE	ACTIVITY	DETAILS
UTas – Alistair Gracie	May 2022	Presentation	Keynote presentation delivered at the Fruit Growers Tasmania Conference, Hobart
UTas – Alistair Gracie	May 2023	Invited article	Published feature article in the July issue of the Nutfruit magazine published by the International Nut and Dried Fruit Council, Spain. Electronic copy can be made available if requested.
UTas – Alistair Gracie	May 2023	Pollination expo	Showcased a broad range of applied pollination research undertaken by TIA and Seed Purity
UTas – Alistair Gracie	October 2020	Newspaper article	Article detailing the project. Published in the Tasmanian Country newspaper and available to entire readership. https://www.utas.edu.au/about/news-and-stories/articles/2020/building-back-up-for-bees
SeedPurity - Cameron Spurr	August 2021	Oral presentation	Oral presentation at the South Pacific Seeds annual production conference, Electronic copy can be made available if requested.
UTas – Joshua Lomax	November 2021	Oral presentation	Oral presentation at the Australian Society of Plant Science Conference, Hobart. Electronic copy can be made available if requested.
UTas – Dugald Close	May 2022	Invited presentation	Oral presentation at the Cherry Growers Australia Conference, Melbourne Electronic copy can be made available if requested.
UTas – Alistair Gracie Alieta Eyles Cameron Spurr Stephen Quarrell Ryan Warren	2021	One day industry workshop on pollination	25 participants. Audience included key industry partners (Reid Fruits, Hansen Orchards, Seed Purity, SPS) and agronomists (Serve-Ag, Nutrien Ag Solutions). Evidence of engagement of the past industry workshops was assessed by a survey. We also received follow-up communications from industry participants thanking us for organising such an informative workshop. 
UTas – Alistair Gracie Alieta Eyles Cameron Spurr	2022	One day industry workshop on pollination	40 participants. Audience included Ashely Zamek (Hort Innovation), key industry partners (Reid Fruits, Hansen Orchards, Seed Purity, Bejo Seeds, SPS), agronomists (Serve-Ag, Nutrien Ag Solutions, Elders) and additional industry stakeholders (Costa, Tasman Apiaries, Driscoll's Australia).

<table>
<tr><td>Stephen Quarrell

Ryan Warren</td><td></td><td></td><td>Evidence of engagement of the past industry workshops was assessed by a survey. We also received follow-up communications from industry participants thanking us for organising such an informative workshop.

</td></tr>
<tr><td>UTas – Alistair Gracie

Alieta Eyles

Cameron Spurr

Stephen Quarrell

Ryan Warren</td><td>2023</td><td>One day industry workshop on pollination</td><td>33 participants. Audience included key industry partners (Reid Fruits, Hansen Orchards, Seed Purity, Bejo Seeds, SPS), agronomists (Serve-Ag, Nutrien Ag Solutions) and additional industry stakeholders (Costa, Tasman Apiaries, Driscoll's Australia, NRM South).

</td></tr>
<tr><td>UTas – Alistair Gracie

Alieta Eyles</td><td>Throughout the duration of the project</td><td colspan="2">Conversations with growers about the project</td></tr>
<tr><td>UTas – Alistair Gracie

Alieta Eyles

Cameron Spurr</td><td colspan="3">Supported several undergraduate and post graduate projects.

Results were collated in a formal thesis. Written for an academic audience. Submitted to the University of Tasmania for assessment. Electronic copy can be made available if requested.

Postgraduate was also presented at workshops, where survey feedback revealed that the study had changed their agronomic practices of pollination</td></tr>
</table>

Recommendations for further adoption

Further adoption of the recommendations can be achieved by adding this report, as well as a summary to the APAL website, and for the project, to producing a list with links to outputs on the grantee's (Hort Innovation) website, classified by information content and type of information, allowing growers easy choice between a podcast, video, factsheet or journal article.

For program leaders in primary industry, the coordinated production of podcasts and short videos should be standard practice, as this allows lasting availability of information for industry and hence creation of legacy.

6 Lessons learnt

Comment from Dr Katja Hogendoorn

No apple orchard can be compared with another, and with a short flowering time, it is virtually impossible to include a meaningful sample size of orchard blocks without inducing enormous variance that renders data uninformative. After realising this, we chose to do most of the work in different spots in the same orchards (e.g. MPU data, proximity of polliniser data).

At the start of the project, researchers were asked what advice to growers would be generated, how this would advance or change the industry, and how long adoption would take. These questions are likely motivated by a desire to ensure that levy paid funds are used to benefit industry. However, most are near-impossible to answer at the start of a research project, due to the exploratory nature of research, the context dependence of outcomes, the inability to incorporate all operational or economic benefits or constraints involved in management decisions, and the fact that we live in changing times. While researchers strive to create advice that growers can implement, not all research outcomes will result in changes in crop management or adoption.

Better ways to ensure informative outcomes for industry includes active engagement between researchers and industry during all phases of the project. Surprisingly often, it is left to the researchers to pursue and organise extension at the end of a project, ignoring the fact that regular updates and feedback can help shape the progress, and is in the interest of both the researchers and industry. Ongoing engagement should therefore be pursued by both parties, and this should be a milestone requirement not only for the researcher, but also for the industry body. APAL has been very good in this respect, and so was the expert project management by Ashley Zamek.

In addition, soon after a project is awarded, researchers working on similar questions in different cropping systems should convene separately to co-design and streamline their approaches and methodology. At later program management meetings, time should be set aside for discussion and evaluation of methods among researchers that work on similar questions, and to forge a combined way forward.

Comment from Dr Lisa Evans

One of the challenges encountered by the project team in this research program was the large variation in both the protected cropping structures and the honey bee colonies used for pollination within these structures. Addressing this variation required more time than initially anticipated, particularly for certain objectives where extensive replication was necessary. Aligning methods across collaborative research teams was important for overcoming this challenge and verifying findings. In this context, it is necessary to strike a balance between exploring various research questions and delving deeply into a select few questions to achieve more comprehensive understanding.

Consistent communication has been an important part of the success of this collaborative project. Allowing time for discussions among researchers and to prepare updates for our steering group has enabled the research teams to refine their approaches and align methodology. Ultimately this has allowed us to begin to understand disparities in landscapes/regions and produce more robust recommendations for growers and industries. Additionally, early discussions of our findings with representative growers and beekeepers in our steering group have ensured that the mitigation strategies we are assessing or working towards are feasible and practical for the industry.

Our outreach and extension efforts faced challenges due to the travel and social restrictions imposed by COVID-19. Despite completing updates for growers through virtual means, the research team would have preferred to conduct more presentations in person. Consequently, we plan to extend the dissemination of our findings beyond the original timeline of this program to make sure industries benefit from our research.

Comment from Dr Sophie Parks

Often, research results in more questions than answers! Several new lines of inquiry have emerged during our research and are being pursued. In this project, we were unable to obtain enough nectar in volume from flowers to conduct a survey of the elemental profile of blueberry floral nectar. The elemental value of nectar would assist in the understanding of how nectar may contribute to honey bee health.

The light conditions under the covers used in protected cropping in Australia appear highly variable given the range and age of cover types used. A better understanding of these covers and associated light conditions would contribute to the development of guidelines that promote plant growth and pollinator health alike, such as choice of cover type and the timing of cover replacement. Preliminary work on this is being undertaken as part of a PhD in partnership with the University of Western Sydney.

In our discussions with blueberry growers, the need for better methods of estimating plant water use was highlighted. In response, we have developed a new research trial at Wollongbar Agricultural Institute, NSW, aimed at developing a simple water use model for blueberry. This is being undertaken as part of the Clean Coastal Catchments project, an initiative of the NSW Marine Estate Management Strategy Delivery.

Comment from Dr Romina Rader

Protected cropping structures are used widely and expanding throughout the horticultural sector. This research aimed to better inform industry as to the types of conditions and contexts in which the structures may be impacting pollination service delivery to horticultural crops. We discovered that impacts are not uniform and predictable across cover types or taxa so a one-size-fits-all approach is not feasible. This research has informed several important insights:

- Flower visitation by insects and pollen flow on floral stigmas are both generally reduced under protected cropping structures when compared to open conditions (i.e. without netting or tunnels). Bird netting resulted in higher insect visitation than polytunnel covers.

- Honey bees moved down rows more frequently than across rows but the effect was unclear in response to variations in netting type. This has implications for cross-pollination if blocks are planted with single cultivars.

- The above impacts upon pollinator visitation under covers will ultimately impact yield and/or quality of fruit if particular crop cultivars need multiple insect visits to achieve high-quality fruit. In particular, the risk of insufficient pollination is highest for those varieties that are highly pollinator dependent (i.e. the cultivars that require insect visitation to transfer pollen to achieve optimal fruit quality).

Comment from Dr Alistair Gracie

Pollination in sweet cherry and onions is a major constraint to yield and must be a priority. Without adequate management, pollination is likely to be highly variable among seasons. The following have emerged as key considerations and/or outcomes from the research:

- The continued refinement of a mechanical pollination methodology (application) is required before broader commercial use.

- Pollen viability varied among genotypes and growing environments. In some cases, it was significantly reduced by the application of some fungicides. In some situations, spray practices could be modified to reduce the potential negative effects of fungicide during flowering.

- The S-allele DNA work captured the interested of growers as it provided insights into pollen donors and crop planting arrangements.

- Mechanical pollination improved yield of sweet cherry in some trials. The project provided practical protocols and processes for pollen collection, handling, storage, and quality assessment. Researchers should build on these findings to further optimise application/delivery of pollen.

- Optimal wet or dry carriers, the ratio of pollen to carrier and the role of electrostatic charge in deposition of the pollen onto the stigma should be the focus of future investigations.

- The S-allele DNA method developed in this project will be a useful tool for identifying pollen donors in future sweet cherry pollination trials.

- Further work is required to determine which fungicides impact negatively on pollen quality and bee health. For pollen quality, the current study utilised detached branches under indoor conditions, however, the magnitude of the impact may be reduced in field conditions from exposure to more extreme environmental conditions.

- Dormancy breakers appear to be extensively used by growers to manage flowering, particularly of later-flowering sweet cherry cultivars. However, there is little understanding of the long-term impact of using strong dormancy breakers on orchard health.

- The feedback provided by the 2023 survey of industry stakeholders indicates that pollination continues to be a major concern for all stakeholders in the sweet cherry and onion industries. They also expressed strong interest in research in alternative pollinators, and additional plant host species for wild and bee pollinators.

7 Appendix - additional project information

7.1 Project, media and communications material and intellectual property

7.1.1 Media, communications, and conferences

All media, communication, and conference activities are listed in Section 5

7.1.2 List of publications

Broussard MA, Coates M, Martinsen P. 2023. Artificial Pollination Technologies: A Review. Agronomy. 13. 1351.

Carisio L, Diaz SS, Ponso S, Manino A, Porporato M. 2020. Scientia Hortculturae. 273: 109629.

Eyles A, Quarrell S, Allen G, Close D, Spurr C, Whiting M, Gracie A. 2022. Feasibility of mechanical pollination in tree fruit and nut crops; a review. Agronomy. 12(5), 1113.

Gagic V, Kirkland L, Kendall LK, Jones J, Kirkland J, Spurr C, Rader R. 2021. Understanding pollinator foraging behaviour and transition rates between flowers is important to maximize seed set in hybrid crops. Apidologie, 52(1), p. 89-100, issn:1297-9678

Hall MA, Jones J, Rocchetti M, Wright D, Rader R. 2020. Bee visitation and fruit quality in berries under protected cropping vary along the length of polytunnels. Journal of Economic Entomology, 113(3), p. 1337-1346, issn:1938-291X

Kendall LK, Evans LJ, Gee M, Smith T, Gagic V, Lobaton JD, Hall MA, Jones J, Kirkland L, Saunders ME, Sonter C, Cutting B. Parks S, Hogendoorn K, Spurr C, Gracie A, Simpson M, Rader R. 2021. The effect of protective covers on pollinator health and pollination service delivery. Agriculture, Ecosystems and Environment. 319: 107556.

Santos, KCBS, Saunders ME, Samnegård U, Rocchetti M, Scalzo J, Rader R. 2023 Protective nets reduce pollen flow in blueberry orchards. Agriculture, Ecosystems and Environment 353 (2023) 108544.

Spronk AL, Guerin GR, Martín-Forés I, Lowe AJ, Hogendoorn K. 2023 Evaluating remnant vegetation management practices adjacent to apple orchards to support native bee pollinators. Ecological Management and Restoration. https://doi.org/10.1111/emr.12588

Webber SM, Garratt MPD, Lukac M, Bailey AP, Huxley T, Potts SG. 2020. Quantifying crop pollinator-dependence and pollination deficits: The effects of experimental scale on yield and quality assessments. Agriculture, Ecosystems and Environment. 304, 107106.

In press/under review

Bailey C, Sonter C, Rader R. Protected covers impact the composition of pollen collected by honey bees (in prep.)

Hogendoorn K, Evans LJ. The use of solitary bees in protected cropping.

Glen et al. 2023. Using DNA analysis for the identification of pollen donors in sweet cherry. In preparation.

Jones J, Rocchetti M, Rader R. Bird netting is better for bees but polythene is better for plants. (in prep.)

Jones J, Santos R, Rader, R. Netting impacts honey bee foraging activity in apple orchards. (in prep.)

Sonter CA, Jones J, Dawson BM, Reid JN, Santos KCBS, Fuller S, Tighe M, Wilson SC, Rader R. (in review). Protective covers impact honey bee colony performance and access to resources

7.2 Equipment and assets

Institute	Equipment/asset acquisitions
UoA	N/A
PFR	N/A
USW DPI	N/A
UNE	N/A
UTas	N/A

7.3 Monitoring and evaluation

See over for inserted pages from M&E report

7.3. Monitoring and Evaluation

Rural Research and Development (R&D) for Profit program

Pollination protected cropping program: Novel technologies and practices for the optimisation of pollination within protected cropping environments

Project code:

ST19002

Milestone number:

End of Project Evaluation MSR 112

February 2024

ACKNOWLEDGEMENTS

This project is being delivered by Hort Innovation with support from the Australian Government Department of Agriculture and Water Resources *(*restructured as the Department of Agriculture, Water and the Environment [DAWE] in January 2020)* as part of its Rural R&D for Profit program.

Delivery partners (Alpha listed):

- Plant and Food Research Australia
- Primary Industries NSW
- Tasmanian Institute of Agriculture
- University of Adelaide
- University of New England

Industry partners (Alpha listed)

- ALGA
- APAL
- Costa
- Daintree fresh
- G2 Netting System
- Hansen orchards
- OzGroup
- Reid Fruits
- Seed purity
- South Pacific Seeds

Note on the report

This M&E Report is based on data collected over the life of the project. It has been a collaborative exercise dependant on the cooperation on the different people involved in the project in using agreed data collection processes and sharing their data to inform the M&E needs. They, and other stakeholders, have also willingly participated in on-going interviews and surveys to ensure that we had the understanding and insights as the project developed and could report with confidence on its progress and outcomes.

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SUMMARY

Context

The aim of the Pollination protected cropping program: "*Novel technologies and practices for the optimisation of pollination within protected cropping environments*" has been to enhance the yields and quality of fruit and vegetable seeds cultivated in protected cropping environments through the development of cutting-edge pollination technologies.

Protected cropping, while advantageous, shows growing signs of adverse effects on crop plant physiology, reproduction, and insect pollinators, impacting the pollination services they offer. Consequently, numerous growers face insufficient pollination, leading to reduced yields and fruit quality.

This report while referencing the research activities and outcomes achieved throughout the project, does not include detailed descriptions of the research methodologies, datasets or analysis undertaken by project partners. This information can be found in the consolidated 'RnD4Profit – ST19000 Final Report' and individual project partner milestone and final reports.

Monitoring + Evaluation team synopsis

The Monitoring and Evaluation (M&E) reporting indicates that the project has met its Key Project Indicators across the complex research elements undertaken throughout the project. The finalisation of Best Practice Guidelines remains in abeyance at the time of writing this report.

An assessment of the project's objectives [explored shortly], highlights that solid outcomes have been achieved across all four of the project's research objectives, while the application of the research outcomes broadly across industry remains in its early stages.

Project objectives / Key Project Indicators

1. ***Enhancing and advancing innovative mechanical pollination methods to achieve artificial pollination in the absence of insects.***
2. ***Enhancing the effectiveness of insect pollinators, specifically honey bees under covers, to enhance pollination.***
3. ***Manipulating plant floral and reproductive traits to elevate fruit production and quality.***
4. ***Optimising the arrangement of pollen donor plants (pollinisers) to empower growers to adjust crop/orchard configuration for sustained high fruit yield and quality***

Five project partners

The project involved five individual projects / project partners, each with their own research who were located geographically across Australia and in New Zealand. The M&E expertise extended to both the five individual projects and those five projects at a collective impact level.

The M&E team observe that amongst the five project partners communication and relationships were strong, professional, collegiate, and broadly supportive. However, comments from all the partners indicate that collaboration and shared findings could have been strengthened formally, prospectively enhancing the program outcomes. COVID 19 travel restrictions, along with bushfire and flood impacts, hindered early collaboration efforts and opportunities for the sharing of information and investigating integrated trials and data aggregation were not able to be optimised.

External Factors

In the first 18 months, the project had to operate within the unprecedented global health pandemic environment of Coronavirus (COVID-19). This meant that major adjustments were made to meet the legal requirements of physical distancing and the inability to cross state boundaries. As a result, the opportunity for research activities to be replicated across multiple interstate locations in this period were lost as were opportunities for collaboration between researchers. In some cases, COVID 19 resulted in the delay of some project components and increased costs in the delivery of some components.

A drought in New South Wales and Queensland was a major issue when the project commenced in 2019. The drought has been marked by three consecutive extremely dry winters and prolonged periods of low rainfall. These conditions adversely affected the amount of data that could be collected due to the impacts on crop production cycles and the reduced windows of opportunity to undertake research activities.

The project has also been impacted by the unprecedented bushfire season of 2019 - 2020 that started early (in early Spring rather than early Summer) due to drought conditions. In mid-January 2020, 17 million hectares (46.03 million acres) was burnt or was burning across all Australian States and territories. One Adelaide Hills grower participating in the research was burnt out and left with only 80% of their trees. They have had to re-net sections of their orchards which has impacted on annual work cycles. Continued participation post fire was restricted to crop areas that were unburnt.

Varroa destructor (Varroa mite) attacks European honeybees (Apis mellifera) and is thought to be one of the greatest threats to Australia's honey and honey bee pollination plant industries. Varroa mite was detected during the project period in sentinel hives in NSW on 22 June 2022. Two main areas of infestation were identified covering the greater Newcastle area, the Hunter Valley, and Central Coast region, as well as around Kempsey on the mid-north coast.

In the early days of detection of Varroa mite, limitations were imposed on the movement of bees, beehives, and beekeeping equipment both within and from areas affected by infestation. These restrictions also had an adverse impact on some project activities.

The convergence of the above-mentioned factors has created unprecedented conditions and project circumstances for this project to which project partners have had to respond.

Importance of Industry relationships

What has become apparent throughout the life of the project has been the ***importance of relationships*** with government, industry, growers, and other researchers to the achievement of the project's outcomes.

Many project partners entered the project with pre-existing relationships in their sector. These pre-existing relationships demonstrate to stakeholders that the project partners are known within their area of expertise, and that they both understand local / sectoral issues and offer solutions for continuous improvement.

The reliance on relationships, new and existing, became apparent when overcoming the challenges brought about by COVID.

Project partners commented that connection and trust with industry and growers have been the most significant relationships developed through the project. This has been demonstrated through the level of access to properties and ongoing on-property research that has been achieved throughout the project life.

Observation: grower extension adoption practices

The M&E team has observed a commendable level of collaboration between project partners and a close-knit network of growers across Australia. These relationships are predominantly characterised by strength, positivity, and professionalism.

As the project timelines continued the M&E team observed an increased effort in communicating with, and educating industry stakeholders, learning jointly, and in some cases seeking active feedback.

Based on the information provided to the M&E team, it is evident that the project has reached numerous growers through a variety of channels, including forums, newsletters, formal events, and on-site presentations. The data supplied to the M&E team suggests that a small number of growers have actively participated and directly benefited from the adaptive research and initial discoveries of the project. This is not unusual for field based research which is reliant on grower involvement for trial site access.

Given that the development of Best Practice Guidelines hinges on research outcomes, the project timeline did not allow adequate time for translating these findings into such guidelines, nor for their subsequent testing and refinement. While the M&E team is confident that valuable information has been successfully disseminated to growers, the degree of awareness and implementation of these techniques across a wider spectrum of growers remains uncertain, indicating a potential gap in the project's findings.

Figure 1. Photo of attendees at the workshop held at UTAS on 2 July 2021 with industry project partners. (Source: University of Tasmania)

Project Scope

In this project, the ongoing focus on research activities, data collection and analysis was driven by the need to overcome early research setbacks arising from COVID-19, bushfires and floods. The successful achievement of the project's research objectives is worthy of mention given the challenges faced by project team members.

Whilst not always feasible due to funding constraints/requirements, future projects would benefit from including contingencies in their timeframes / planning to allow for unexpected external factors that can adversely impact on undertaking research on seasonal crops. Findings from this project also highlight the time challenges in not only gathering but also analysing data.

With the benefit of hindsight, it is possible that the original project scope was overly optimistic in relation to understanding and measuring the success of extension activities and uptake of practices by growers and or apiarists across industries.

Cost Benefit

When assessing the cost benefit of the project, the extension and application of key findings by industry across any of the crops explored in this project has been minimal and occurred at individual research property level only. As such, claims regarding the cost/benefit of the project have not been tested at either a micro or macro level and are based on assumptions underpinned by the project's program logic.

Assessment of the impact of the Evaluation process

In addition to gathering information and evidence to support the evaluation of the Pollination project itself, the M&E Team also reviewed its own performance against the objectives driving their activities.

Core findings demonstrate that the inclusion of an Evaluation Team from the commencement of the Pollination project has contributed to increased confidence amongst project team members in the realm of evaluation. The M&E work was perceived to be valuable and is held in high regard.

What is less evident is the capacity building of the M&E activities on the project team. It appears that the full capacity of the M&E process was never fully adopted throughout the project. This trend appears to be consistent with the lack of embedding of M&E, feedback loops and capturing of the impact of industry extension, spoken about earlier in this summary.

Despite this, another key finding illustrates that the evaluation process has delivered learnings for future project management and delivery including building capacity across key principles of good evaluation practice, and confidence of how evaluation can be built throughout the monitoring and evaluation.

Detailed findings on the impact of the Evaluation process can be found in the Evaluation Team Impact section of this report.

Contents

1 ABOUT THIS REPORT

1.1 Rationale for Evaluation

This is the 'end of project' Monitoring & Evaluation Report for the 4-year Rural Research & Development 'for profit' program - Novel technologies and practices for the optimisation of pollination within protected cropping environments (Pollination project). It follows the development of an overarching Monitoring and Evaluation(M&E) plan and has included the provision of support to 5 individual projects to ensure ongoing M&E.

The M&E strategy was undertaken independently by the monitoring and evaluation experts from Innovative Influence and Strategic Matters

The primary aims of the evaluation process have been to:

- Assess whether the project has achieved its intended goals.
- Understand how the project achieved its intended purpose, or why it may not have done so.
- Identify how efficient the project was in directing resources into activities, objectives, and goals.
- Assess how sustainable and meaningful the project outcomes are for participants.
- Inform decision makers about how to build on or improve the project.

The report builds on the progress and findings presented in the mid-term M&E report to capture activity, feedback, impact, and issues. While this report references the research activities and outcomes achieved throughout the project, it does not include detailed descriptions of the research methodologies, datasets or analysis undertaken by project partners. This information can be found in the consolidated 'RnD4Profit – ST19000 Final Report'.

1.2 Project Context

"Protected cropping" describes the growing of crops under glass, plastic or netted covers and includes production systems within greenhouses, glasshouses, polytunnels and under netting. Australian horticultural industries are increasingly reliant on protected cropping to reduce yield variability in association with the impacts of extreme weather events, pests and diseases and the efficiency of inputs such as water, fertilisers, labour, land and energy. In Australia, a number of high value industries are grown under protected cropping systems including raspberries, blueberries, apples and vegetables. These high value industries are significantly expanding in production across Australia at an average rate of 4-6% growth each year. Blueberry production as an example has increased from 5,500 tonnes to 17,000 tonnes in the last 5 years representing more than 80% of Australia's total berry production.

Despite the benefits of protected cropping, there is increasing evidence to suggest that the altered environmental conditions caused by protective environments can negatively impact crop plant physiology and reproduction, insect pollinators and the pollination services they provide. This means that many growers are experiencing inadequate pollination resulting in lower yields and fruit quality than could otherwise be achieved under these growing conditions. The causes of these yield deficits have been attributed to lower plant and pollinator performance under enclosed conditions – both of which could prospectively be rectified with the development and adoption of new technologies and adaptive, grower-driven research.

Project objectives / Key Project Indicators

The program's aims have focused on enhancing the yields and quality of fruit and vegetable seeds cultivated in protected cropping environments through the development of cutting-edge pollination technologies. More specifically the focus has been on:

1) Enhancing and advancing innovative mechanical pollination methods to achieve artificial pollination in the absence of insects.

Mechanical pollination offers an exciting technology to improve crop performance, yet existing approaches are rudimentary in design with few analyses of the net benefits. The project aimed to develop protocols and technology for pollen collection, handling, storage, and quality assessment, and to lay the groundwork for new pollination methods to reduce the risks associated with the high dependence on insects and enhance pollination outcomes.

2) Enhancing the effectiveness of insect pollinators, specifically honey bees under covers, to enhance pollination.

Pollination in protected cropping may be compromised by a range of factors specific to conditions under covers (for example, greater humidity and temperatures, low UV light) that affect honey bee (*Apis mellifera*) pollinator performance. Understanding the effect of these conditions can lead to better advice regarding the maintenance of hive health and performance. This can include advice for e.g. hive size and placement, as well as the choice and manipulation of the covers themselves to maximise yield and fruit quality.

3) Manipulating plant floral and reproductive traits to elevate fruit production and quality.

Quantifying the impact of protected cropping and flower physiology on bee behaviour can provide insight into how environmental conditions can be optimised to maximise attractivity of flowers and promote pollination. Specifically, the focus is on understanding floral biology, encompassing nectar secretion patterns, nectar sugar concentration, and pollen production in flowers of various Vaccinium (blueberry) and Rubus (raspberry and blackberry) varieties.

4) Optimising the arrangement of pollen donor plants (pollinizers) to empower growers to adjust crop/orchard configuration for sustained high fruit yield and quality.

Pollination in open crops is provided by a suite of flying insects, which may be supported in different ways. Changes in pollinator movement under cover may cause the need to change the advice regarding varietal proximity. Therefore, a further objective was to understand and design optimal configurations of pollinisers that maximize yield and fruit quality. In addition, the potential of a low-cost robotic mobile polliniser units, to facilitate the improved delivery of pollen to the maternal fruit bearing plants was explored. The project also investigated how the non-crop floral resources and other features of the orchard floor affected the presence of pollinators under protected covers, and identified some practises that can improve their presence.

1.3 About the M&E Program

The role of the M&E team was to deliver design, implementation and impact benefits while also providing an exemplar for the take up of monitoring and evaluation as integral components of program and project planning. Working across this program and with associated project team members, the M&E team provided many ‘touch points’ to demonstrate evaluation approaches and techniques whilst also building the capacity of project teams involved.

The M&E team’s intentional approach is anticipated to have flow on benefits for future project management and delivery by demonstrating:

- Different types of evaluation and how they might be used.
- A strategic approach to evaluation that prioritises evaluation, and scales evaluations based on the characteristics of different types of programs.
- Key principles of good evaluation practice.

This project’s M&E is based on evaluation Project Logic which clearly links inputs, activities and outputs to desired outcomes and impacts.

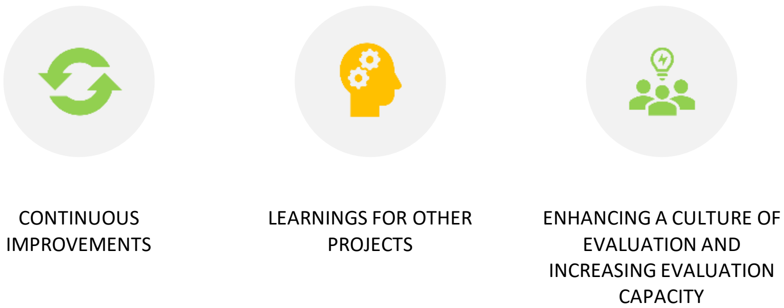
The content of this logic is based on the project proposal and plan. The direct responsibility for the project stops at the Project Outcome level – which describes the specific contribution to the Long-Term Goal expected of the project within its resources and time- frame. The framework looks at the 'logical' performance, measured at each stage and level of project delivery, and then the 'logical' and practical M&E data collection methods to capture this data.

1.4 Evaluation Outcomes

Underpinning the scope of works were three evaluation outcomes.

- Continuous improvement.
- Learnings for other projects.
- Enhancing a culture of evaluation and increasing evaluation capacity.

These are diagrammatically illustrated below:



1.5 Methodology

A range of qualitative and quantitative data collection and analysis methods approaches, and tools are in use to gather the necessary information to address the project's evaluation and reporting requirements. Focused discussion sessions have been held with project team members to capture ongoing insights and information about project progress, implementation, and stakeholder engagement.

This has included:

1. Receipt and analysis of Milestone Reports from project partners.
2. Provision of email and telephone support to project team members.
3. Development of a survey for partners re: project progress, outcomes and preliminary findings.
4. Exploration of ways in which evaluation data can be garnered from industry and / or grower representatives as part of the project partners' ongoing commitment to M&E throughout the life of their projects.
5. Staying in touch with broad project trends and continuously looking for opportunities to support the project partners with their M&E requirements.

Project partner milestone reports and associated publications and research papers were reviewed along with any evaluation data collected directly by project partners. In addition, interviews were undertaken with external key industry personnel to better understand how meaningful the project outcomes have been and the extent of their impact on the proposed beneficiaries.

This report includes information from:

- Previous M&E reports and their data sources.

- Narratives.
- Interviews with project team members and stakeholders.
- Online survey of project team members.

The Monitoring and Evaluation Framework used to guide the process is included as Appendix 1 which provides more information on the performance measures, process and methods used.

Appendix 2 identifies the key outputs / products that the evaluation team produced throughout the life of the project.

1.6 Evaluation Team Impact

In addition to gathering information and evidence to support the evaluation of the Pollination Protected Cropping Program itself, the Evaluation Team also reviewed its own performance against the objectives driving their activities. Results are detailed in Appendix 3.

Enhancing a culture of evaluation and increasing evaluation capacity

The inclusion of an evaluation team from the commencement of the Pollination project has contributed to increased capability and confidence amongst project team members in the realm of evaluation.

The top benefits associated with having a dedicated elevation team were:

- Early planning and incorporation of evaluation into projects.
- Holding the team to account on evaluation.
- Time management (that the evaluation team members held project partners to account timewise).

The most significant improvements occurred across the project team members' attitudes towards evaluation and recognition of the importance of evaluation.

Learnings for other projects

This evaluation project has delivered learnings for future project management and delivery by demonstrating:

- Different types of evaluation and how they might be used.
- A strategic approach to evaluation that prioritises evaluation, and scales evaluations based on the characteristics of different types of programs.
- Key principles of good evaluation practice.
- How capacity and confidence of evaluation can be built throughout the monitoring and evaluation process.

2 PROGRAM LOGIC M&E FRAMEWORK: POLLINATION

2.1 Program Logic Framework

The following program logic framework was developed by Innovative Influences and Strategic Matters in January 2020. The framework had substantial input from all project partners.

More information is available in Figure 2 and in the following end of project and intermediate outcomes table as identified by the project partners and articulated in the program logic framework.

Table 1: End of Project Outcomes/Intermediate Outcomes

End of Project Outcomes	Intermediate Outcomes
Uptake and application of guidelines to improve honey bee colony performance and pollination of apples, lychees, and berries within protected cropping.	Development of techniques and methods to improve pollination of apples, lychee and berries as well as honey bee colony performance, under protective crop covers on farm.
Improved awareness of techniques to improve pollination within protected cropping environments in berries, and growers are interested in applying recommended techniques.	Development of techniques to improve pollination of berries under protective crop covers on-farm such as identification of flowering stage or time when floral attractiveness peaks. Industry endorsement of guidelines and recommendations.
Increased awareness and commitment to adopt best practise of pollinator management undercovers.	Development of management regimes and practices to improve/maintain pollination services under protected cropping covers.
Actions taken by growers and beekeepers to modify previous management in line with research findings.	Increased awareness of and commitment to adopt best practice.
Increased awareness amongst apple orchardists and netting industry of best practise farm management or better hive health and pollinator activity and apply quality.	Increased body of knowledge about: • The effects of net type, colour, height above canopy and management of hive health and bee activity. • The diversity of apple pollen varieties carried by honey bees under net and outside. • The effect of paternity on apple set quality for three major varieties. • The utility of mobile pollinator units. • Wild pollinator diversity in relation to under row and inter row management. • How co-flowering crop and non-crop resources influence pollinator visitation to crop flowers. Development of strategies & recommendations.
Development and validation of alternate pollination options for sweet cherry and onion.	Hand-held prototype equipment for mechanical pollination of sweet cherry and onion is developed. In-vitro methods for rapid assessment of pollen quality are validated.

End of Project Outcomes	Intermediate Outcomes
	The effect of environment and handling on sweet cherry and onion pollen quality is understood and protocols for collecting, handling, storing and delivering pollen are developed. The technical feasibility and benefits of mechanical pollination systems for commercial onion seed and sweet cherry production are understood.
Increased knowledge/ use of alternate pollination options for sweet cherry and onions by growers.	Increased knowledge/use of information by growers.

Figure 2. Program Logic for Pollination Protected Cropping Program (ST19000)

Relative SIP Outcome(s)	Improved management of European honey bee for pollination Crop pollination requirements understood and integrated into best practice				Alternate pollination options developed and available
	PFRA	**NSW DPI**	**UNE**	**UOA**	**UTAS**
End-of-project outcomes	Uptake and application of guidelines to improve honey bee colony performance and pollination of **apples**, **lychee** and **berries** within protected cropping	Improved awareness of techniques to improve pollination within protected cropping environments in **berries**, and growers are interested in applying recommended techniques	Increased awareness of and commitment to adopt best practice of pollinator management under covers. Actions taken by growers and beekeepers to modify previous management in line with research findings	Increased awareness amongst **apple** orchardists and netting industry of best practice farm management for better hive health and pollinator activity and apply quality.	Development and validation of alternate pollination options for sweet **cherry** and **onion** Increased knowledge/use of information by growers
Intermediate outcomes	Development of techniques and methods to improve pollination of apples, lychee and berries, as well as honey bee colony performance, under protective crop covers on-farm	Development of techniques to improve pollination of berries under protective crop covers on-farm, such as identification of flowering stage or time when floral attractiveness peaks. Industry endorsement of guidelines and recommendations	Development of management regimes and practices to improve / maintain pollination services under protected cropping covers. This may include modifying honeybee stocking practices and/or removing tunnel / netting during flowering Increased awareness of and commitment to adopt best practice.	Increased body of knowledge about: 1. the effects of net type, colour, height above canopy and managements on hive health and bee activity (5.1 e, 5.2 a, c, d) 2. the diversity of apple pollen varieties carried by honey bees under net and outside (5.3 a) 3. the effect of paternity on apple set quality for three major varieties(5.3 b) 4. the utility of mobile pollinator units (5.3 c) 5. wild pollinator diversity in relation to under row and inter-row management (5.4 a) 6. how co-flowering crop and non-crop resources influence pollinator visitation to crop flowers (5.4 b) Development of strategies and recommendations	Hand-held prototype equipment for mechanical pollination of sweet cherry and onion is developed In-vitro methods for rapid assessment of pollen quality are validated. The effect of environment and handling on sweet cherry and onion pollen quality is understood and protocols for collecting, handling, storing and delivering pollen are developed The technical feasibility and benefits of mechanical pollination systems for commercial onion seed and sweet cherry production are understood Increased knowledge/use of information by growers
Outputs	Research results. Consultation outputs. Publications & Articles. Guidelines and recommendations to industry (lychee, apple, and berry growers and apiarists) to improve honey bee colony performance and pollination within protected cropping environments	Guidelines and recommendations to improve pollination under protected crop covers are disseminated to industry (berry growers and apiarists), and industry commitment to these is evaluated. Literature review. Field trial results. Industry events delivered. Articles published. Survey results.	Research results relating to optimizing pollination service delivery; Research publications and outreach material from trial results; activities to disseminate findings	Literary review produced, Academic publications, guidelines and recommendations, preliminary design mobile polliniser unit, grower engagement activities, steering committee meetings	Prototype mechanical pollination systems for two high-value perennial and biennial horticultural crops and protocols for pollen collection, handling and delivery; A blueprint document for development of mechanical pollination prototypes for other horticultural industries Commercialisation plan for technologies developed in the program Presentation of project results at relevant national (e.g. Protected Cropping Australia annual conferences) and international conferences where appropriate; Presentation of project results at relevant industry conferences and/or field days; Publication of manuscripts using study results in peer reviewed scientific journals within 2 years of project completion. Guidelines and recommendations to industry (Onion and Cherry growers) to improve pollination under protective crop covers developed in this program. Inclusion of these guidelines in a cross-commodity pollination manual produced by the overall program.
Activities	Conduct research trials, present results to growers and industry members, development of guidelines and recommendations, consult with industry on guideline development. Publish both popular and research articles summarising our findings.	Conduct a literature review and field trials for greater scientific understanding of berry pollination. Conduct industry events such as workshops, presentations, webinar and publish articles to inform the industry of project activities and findings. Conduct surveys/interviews/casestudy to assess grower commitment to applying developed guidelines.	Review literature on protected cropping; conduct research trials to collect data on activity of honeybees and other pollinators under covers; collect data on plant responses to variations in environmental and management conditions under covers; conduct workshops, compile data for journal articles. Conduct surveys/interviews to assess grower commitment to modifying practices.	In collaboration with other researchers, investigate: 1. the effects of net type, colour, height above canopy and managements on hive health and bee activity (5.1 e, 5.2 a, c, d) 2. the diversity of apple pollen varieties carried by honey bees under net and outside (5.3 a) 3. the effect of paternity on apple set quality for three major varieties(5.3 b) 4. the utility of mobile pollinator units (5.3 c) 5. wild pollinator diversity in relation to under row and inter-row management (5.4 a) 6. how co-flowering crop and non-crop resources influence pollinator visitation to crop flowers (5.4 b) Produce a literature review on pollination in protected cropping. Present plans, progress and outcomes to local steering committee and wider apple, orchard and netting industries using a variety of extension methods, publish papers, assist with	Conduct field trials Conduct laboratory experiments Develop instruments Communicate information at science and industry communication events, conferences and publications
Foundational activities	· Research to improve the performance of honey bee colonies under covers through colony manipulation (in collaboration with UoA and UNE). · Research to improve the performance of honey bee colonies under covers through orchard manipulation in apple and lychee orchards (in collaboration with UoA and UNE). · Research to understand relationship between plant physiology and bee visits in covered blueberries (in collaboration with NSWDPI & UNE). · Collate information from wider project team and present summaries to industry	Research to understand blueberry pollen & nectar production, & relationship with bee visits under protected crop covers Development to extrapolate information from NSWDPI, UNE and PFR research for industry	Collaborate with NSWDPI, PFR, Seed Purity, UTas and UoA to coordinate field work and collate data from existing studies; Baseline data collection	Get a signed contract. Attract staff. Consult with growers and beekeepers about their issues re pollination in protected cropping. Plan research. Check research plan with industry partners and consult them about desired outputs and end-of-project outcomes.	Project administration Project planning

3 POLLINATION KEY PROJECT OUTCOMES

As mentioned in the summary, all project partners in the Pollination project were required to respond to the following Key objectives / Key Project Indicators:

These objectives shape the overall evaluation of the project:

1. ***Enhancing and advancing innovative mechanical pollination methods to achieve artificial pollination in the absence of insects.***
2. ***Enhancing the effectiveness of insect pollinators, specifically honey bees under covers, to enhance pollination.***
3. ***Manipulating plant floral and reproductive traits to elevate fruit production and quality.***
4. ***Optimising the arrangement of pollen donor plants (pollinisers) to empower growers to adjust crop/orchard configuration for sustained high fruit yield and quality***

The M&E team has observed a commendable level of collaboration among project partners and a close-knit network of growers across Australia in the delivery of the project objectives. These relationships are predominantly characterised by strength, positivity, and professionalism.

An assessment of the project's objectives highlights that solid outcomes have been achieved across all four of the project objectives, with the application of the research outcomes across industry more broadly, still in its early stages.

The evaluation process indicates that the project has met its Key Project Indicators across the complex research elements undertaken throughout the project.

3.1 Enhancing and advancing innovative mechanical pollination methods

Mechanical pollination offers an exciting technology to improve crop performance. This KPI focused on devising protocols and technology for the process of pollen collection, handling, storage, and quality assessment. Additionally, it aimed to pave the way for novel pollination methods, with the overarching goal of mitigating the risks associated with overdependence on insects and enhancing the effectiveness of pollination.

Through the term of this project, one project partner (UTAS) focused specially on innovative mechanical pollination methods for yield forecasting across sweet cherries, onions, and blueberries.

UTAS research illustrates that:

- Hand-held prototype equipment for mechanical pollination of sweet cherry and onion has been developed and tested extensively in the field.
- It is feasible to scale generation vacuum collector for onion pollen. This technology can collect quantities of pollen required for commercial mechanical pollination, with an individual operator collecting up to 20g of purified pollen per hour.
- Trials demonstrated that there was no decline in pollen viability during harvesting operations.
- In-vitro and in-vivo methods for rapid assessment of pollen quality and pollination capacity have been developed and validated.
- Large scale purification of raw (Cherry) pollen was achieved using a prototype mill, with germination significantly reduced by the third pass through the mill.
- The effect of milling was also found to vary significantly according to cherry variety, with Van and Lapins pollen being highly robust, whilst Tamara highly susceptible to milling damage. The pollination milling process can cause damage to pollen. Milling flowers twice, rather than three times can significantly reduce mechanical damage and maintain pollen yields.
- Purification, handling, and storage protocols for onion pollen have been established with

research illustrating that Desiccated pollen stored for one year at -18 and -80oC with no loss of viability. In contrast, desiccated pollen samples stored at temperatures ranging from 20oC to 4oC progressively lost viability over a period of days to weeks.

- Storage of pollen, particularly regarding the temperature can have a significant impact on the pollen's viability.
- UTAS findings believe that the understanding of sweet cherry pollination has progressed significantly.

3.2 Enhancing the effectiveness of insect pollinators under covers

Through the term of this project, four of the five project partners (Department of Primary Industries-NSW, Plant & Food Research Australia, University of Adelaide, University of New England) focused specially on the effectiveness of insect pollinators, specifically honey bees undercovers to enhance pollination. The emphasis of their research focused on the following crops: apples, blueberries, lychees.

Research findings demonstrate the following:

- The exact science / process of how pollen moves through an orchard remains unclear
- Honey bee hives placed under protected covers (i.e., bird netting and polytunnels) negatively impact honey bee colony performance relative to those placed in uncovered positions outside netting. Hive weight and pollen storage was lower in hives under nets than outside nets.

Position of flowers

- Work undertaken by NSW DPI demonstrated the following findings: In blueberry crops, the location or position of flowers within the crop may affect their likelihood of being pollinated, and achieving a maximum fruit weight or fruit sweetness
- The ends of blue berry rows, closest to hives, or a north facing side of a row were more likely to achieve maximum weight and sugar contents (total soluble solids). The ends of blue berry rows, closest to hives, or a north facing side of a row were more likely to achieve maximum weight and sugar contents (total soluble solids).

Netting and covers

- Generally, Bird-netting is better for bee foraging activity than polythene tunnels
    - This differs to NSW DPI results that illustrates that under the conditions of their experiment, honey bee visits to flowers were not different between the tunnels and bird netted crops. However, this occurred *when honey bees were abundant.*
- If enclosing the hive under the net and placing them inside the crop, growers may require fewer hives, but should expect to pay a small additional hive lease to compensate for the reduction in hive strength.
- Honey bee hives placed under protected covers (i.e. bird netting and polytunnels) negatively impact honey bee colony performance relative to those placed in uncovered positions outside netting. Hive weight and pollen storage was lower in hives under nets than outside nets.
- The University of Adelaide's research highlights that crop covers affect orientation, activity, and foraging returns and reduces hive health by about 8% over a period of about 3 weeks.
- Allowing the bees to forage outside, either by opening sides or by making part of the cover penetrable for bees improves the foraging returns of the hive and allows the hive to grow but is *also likely to reduce pollination services* in the protected crop.
- UNE research indicated that blueberry pollen on stigmas was reduced by up to 81% under full netting and 36% by partial netting, relative to open, uncovered blueberry plants.
- In the DPI blueberry work, pollination success did not differ between the cover types. However,

the mean berry weight and fruit sugars (total soluble solids) were both about 30% higher from the tunnels than the bird net. This outcome is thought to be attributable to favourable fertigation and light conditions. Pollination in protected cropping may be compromised by a range of factors specific to the condition under covers conditions under covers (i.e. greater humidity and temperatures, low UV light)

Netting height

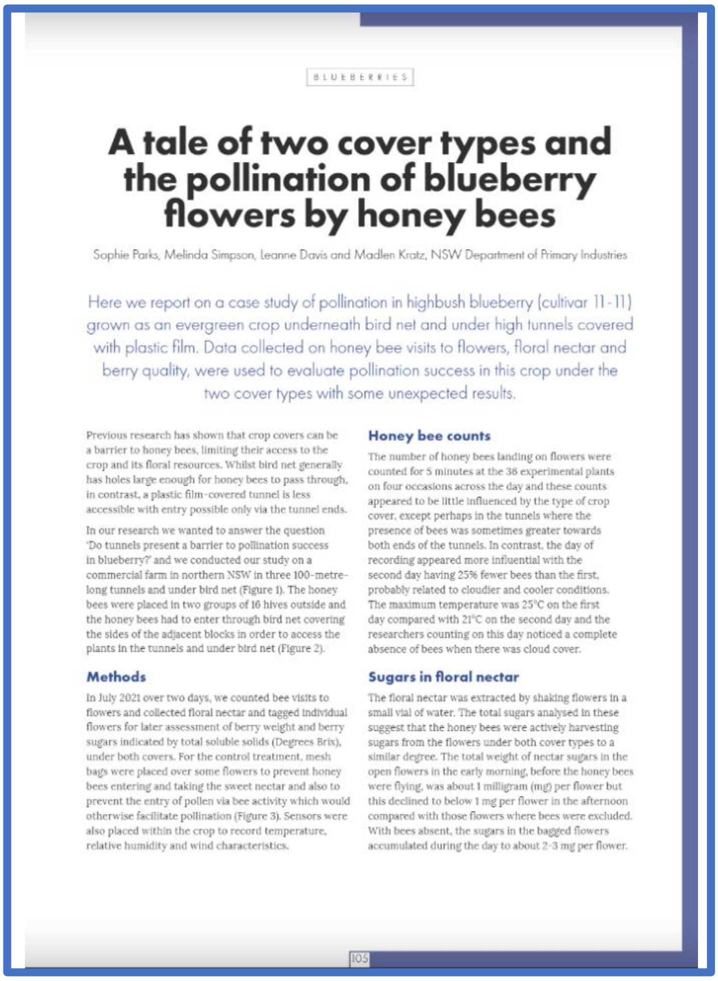
- Netting height above apple plants is important to bee movement in apple orchards, higher netting is better.
- Both UoA and UNE observes that Bees perform better under covers that are high above the crop, as it allows them to orient under the cover. Netting height above apple plants is important to bee movement in apple orchards, higher netting is better.

Hive strength, foraging and pollination:

- UoA' research demonstrates that allowing the bees to forage outside, either by opening sides or by making part of the cover penetrable for bees improves the foraging returns of the hive and allows the hive to grow but is also likely to reduce pollination services in the protected crop.
- Plant and Food Research Australia (PFRA) findings note that native bush surrounding blocks may impact negatively on the pollination of crops – due to alternative foraging sources If enclosing the hive under the net and placing them inside the crop, growers may require fewer hives, but should expect to pay a small additional hive lease to compensate for the reduction in hive strength.
- Unmanaged pollinators in orchards included mainly native bees and hoverflies (Uni of Ad)
- General research indicates that bees are not always reliable pollinators

Other findings

- Unmanaged pollinators in orchards included mainly native bees and hoverflies. These may play an important role in pollination in both open crops as well as under cover. For example, ground nesting generalist bees
- Research from Uni of Adelaide + UNE suggests that: The exact science / process of how pollen moves through an orchard remains unclear



BLUEBERRIES

A tale of two cover types and the pollination of blueberry flowers by honey bees

Sophie Parks, Melinda Simpson, Leanne Davis and Madlen Kratz, NSW Department of Primary Industries

Here we report on a case study of pollination in highbush blueberry (cultivar 11-11) grown as an evergreen crop underneath bird net and under high tunnels covered with plastic film. Data collected on honey bee visits to flowers, floral nectar and berry quality, were used to evaluate pollination success in this crop under the two cover types with some unexpected results.

Previous research has shown that crop covers can be a barrier to honey bees, limiting their access to the crop and its floral resources. Whilst bird net generally has holes large enough for honey bees to pass through, in contrast, a plastic film-covered tunnel is less accessible with entry possible only via the tunnel ends.

In our research we wanted to answer the question 'Do tunnels present a barrier to pollination success in blueberry?' and we conducted our study on a commercial farm in northern NSW in three 100-metre-long tunnels and under bird net (Figure 1). The honey bees were placed in two groups of 16 hives outside and the honey bees had to enter through bird net covering the sides of the adjacent blocks in order to access the plants in the tunnels and under bird net (Figure 2).

Methods

In July 2021 over two days, we counted bee visits to flowers and collected floral nectar and tagged individual flowers for later assessment of berry weight and berry sugars indicated by total soluble solids (Degrees Brix), under both covers. For the control treatment, mesh bags were placed over some flowers to prevent honey bees entering and taking the sweet nectar and also to prevent the entry of pollen via bee activity which would otherwise facilitate pollination (Figure 3). Sensors were also placed within the crop to record temperature, relative humidity and wind characteristics.

Honey bee counts

The number of honey bees landing on flowers were counted for 5 minutes at the 38 experimental plants on four occasions across the day and these counts appeared to be little influenced by the type of crop cover, except perhaps in the tunnels where the presence of bees was sometimes greater towards both ends of the tunnels. In contrast, the day of recording appeared more influential with the second day having 25% fewer bees than the first, probably related to cloudier and cooler conditions. The maximum temperature was 25°C on the first day compared with 21°C on the second day and the researchers counting on this day noticed a complete absence of bees when there was cloud cover.

Sugars in floral nectar

The floral nectar was extracted by shaking flowers in a small vial of water. The total sugars analysed in these suggest that the honey bees were actively harvesting sugars from the flowers under both cover types to a similar degree. The total weight of nectar sugars in the open flowers in the early morning, before the honey bees were flying, was about 1 milligram (mg) per flower but this declined to below 1 mg per flower in the afternoon compared with those flowers where bees were excluded. With bees absent, the sugars in the bagged flowers accumulated during the day to about 2-3 mg per flower.

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Figure 3. Spring edition 2022 [an article in the Australian Berry journal]
(Source: NSW Department of Primary Industries)

3.3 Manipulating plant floral and reproductive traits.

The intention of this element of the project was to research options to modify plant features related to flowers and reproduction to increase both the quantity and quality of fruits produced.

Manipulating plant floral and reproductive traits can involve changing traits such as flower size, colour, and reproductive processes to enhance fruit yield and improve characteristics like taste, size, and nutritional content.

Floral characteristics and attraction

The flowers of seven blueberry cultivars were examined that varied in the length, width, and the area of the floral tube entrance. The flowers of three cultivars contrasting in their morphological characteristics were selected and honey bee visits to 'flower stations' positioned within blocks of these cultivars on-farm were observed. Although the floral morphology, and the concentration of nectar sugars, contrasted among the three cultivars, it appeared that only the position of the cultivar flowers at the flower station influenced the number of visits by honey bees (data analysis is ongoing).

The production of floral nectar (volume or mass) and the concentration of sugars of nectar during this project was variable. Nectar sugars accumulated in flowers, when pollinators were excluded, during the day or even over several days. However, both nectar mass and the concentration of sugars contrasted greatly in some experiments between the days of sampling which may have related to environmental factors, known to affect nectar production, such as light intensity, temperature and relative humidity.

Covers, attraction and pollination

Under the conditions of the research, honey bee visits to flowers were not different between the tunnels and bird netted crops when honey bees were abundant. Further, pollination success did not differ between the cover types. However, the mean berry weight and fruit sugars (total soluble solids) were both about 30% higher from the tunnels than the bird net. This was attributed to greater nutrition provided to the tunnel crop via fertigation, and more favourable light conditions for growth under the tunnel plastic film. Position within the row also influenced fruit weight for both cover types with generally heavier berries at the row edges where the honey bees were accessing the crop. This work highlights that although pollination was fundamental to fruit quality in this cultivar, other practices significantly improved fruit quality, beyond pollination.

Using fertigation to modify floral resources

Fertigation, the inclusion of nutrients with irrigation, was used as a treatment to observe the effect of fertigation strength on nectar production and quality. Although plant nutritional status was affected by fertigation, there was no relationship observed between the concentration of fertilisers in the irrigation and nectar production or concentration of nectar sugars, or between plant nutritional status and nectar characteristics. However, the most concentrated fertigation treatment was associated with the largest flower number, yielding the largest nectar source per plant at sampling. The role of plant nutrition in the availability of nectar resources through the timing and production of flowers is highlighted by this work.

In the project three partners (DPI [NSW], University of Adelaide and University of Tasmania) undertook research in this area. Findings demonstrated:

Fertigation

In trials undertaken by NSW DPI, the following results were shared:

- Fertigation cannot be used to adjust nectar volume or content of sugars but it can be used to manipulate flower timing and density.
- The mean blueberry weight and fruit sugars (total soluble solids) – which were both about 30% higher from the tunnels than the bird net and are likely to be attributable to fertigation rather

than pollination success.

- Furthermore, the most concentrated fertigation treatment was associated with the largest flower number, yielding the largest nectar source per plant at sampling.

Molecular testing for paternity

The University of Adelaide undertook some testing of the paternity of pollen in apples. This involved molecular testing. This process is commonly used in plant genetics and breeding to identify the male parent (pollen donor) in a cross-pollination event.

The work that the University of Tasmania undertook demonstrated the following:

- UTAS's research observed an 18% seed yield increase in an open pollinated onion parent line sprayed with a liquid pollen suspension, but pollen applied to an inbred hybrid onion seed parent line germinated poorly in-vivo and did not improve seed set

Amount of pollen

- In Onions, with both liquid and dry carriers, the major constraints to seed setting were the amount of pollen that could be delivered onto the stigmas of the seed-bearing line and pollen viability
- Pollen applied in dry carriers was also capable of effecting Onion seed set in an open pollinated parent line, in which other forms of pollination (self or insect pollination) were prevented by emasculation and bagging.
- UTAS's research observed an 18% seed yield increase in an open pollinated onion parent line sprayed with a liquid pollen suspension, but pollen applied to an inbred hybrid onion seed parent line germinated poorly in-vivo and did not improve seed set Pollen applied in dry carriers was also capable of effecting Onion seed set in an open pollinated parent line, in which other forms of pollination (self or insect pollination) were prevented by emasculation and bagging.
- In Onions, with both liquid and dry carriers, the major constraints to seed setting were the amount of pollen that could be delivered onto the stigmas of the seed-bearing line and pollen viability

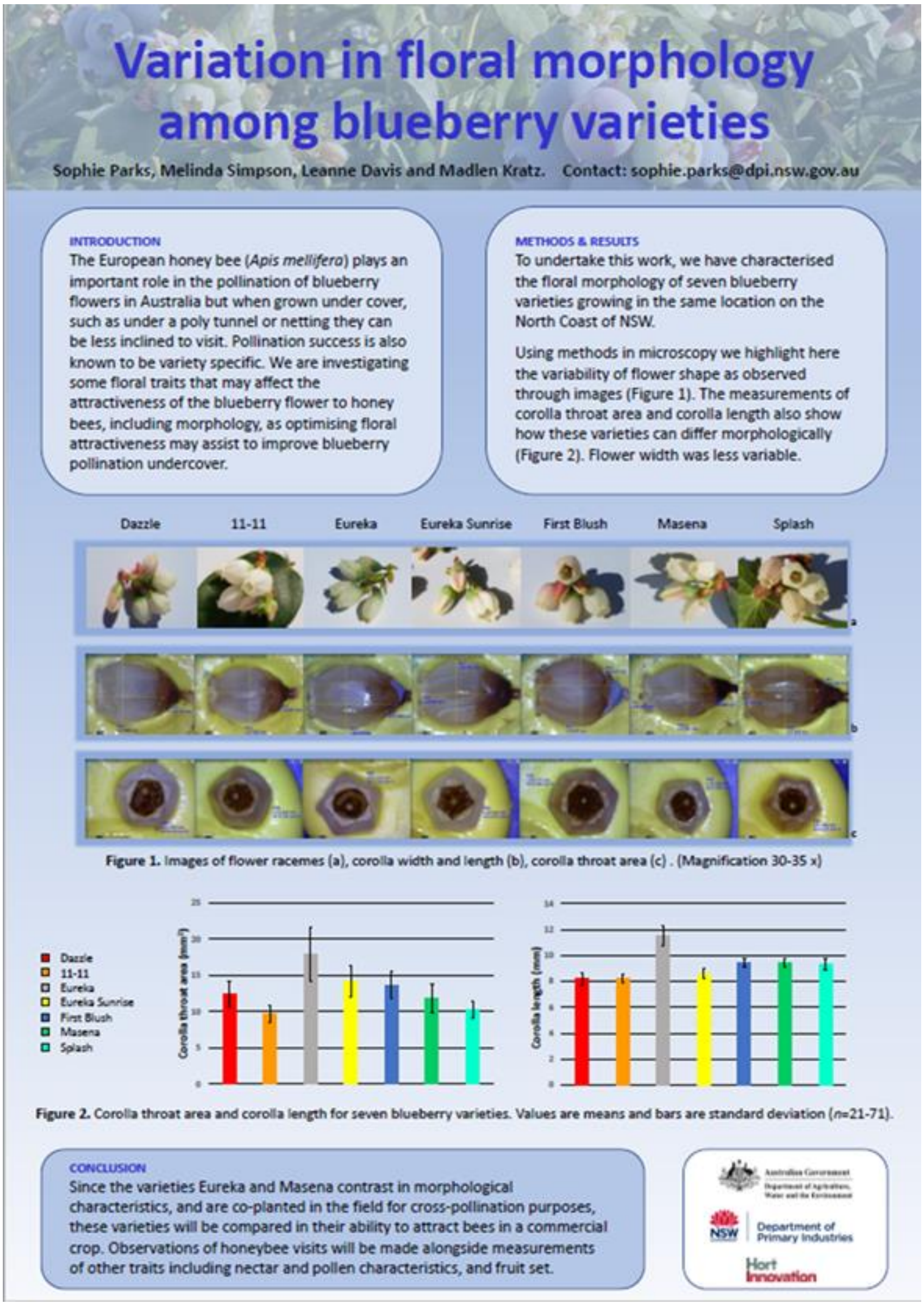


Figure 4. Poster presented at the NSW Apiarists' Association conference 19-20 May, Tamworth.
(Source: NSW Department of Primary Industries)

3.4 Optimising the arrangement to empower growers to adjust crop/orchard configuration

Understanding the local conditions surrounding an orchard is important when attracting pollinators (e.g.: amount of alternative foraging plants / material)

"Optimising the arrangement to empower growers to adjust crop/orchard configuration" project findings are drawn from work undertaken by the University of Adelaide (UoA) and DPI (NSW).

Most findings are of a broad nature, and are specified below:

- A systematic approach to pollination of crops is required particularly when requiring increasing yield
- Orchard design and placement of pollinisers is crucially important for the set of apple under net.

- Research from the UoA illustrates that combined evidence from molecular, behavioural and fruit set measures showed that orchard design and placement of pollinisers is crucially important for the set of apple under net. Ideally, apple rows should be limited to about 90 m in length, and one or more rows of compatible, simultaneously flowering pollinisers should be present every seventh row.
- Access and proximity to bee hives also plays a key role in increasing yield and production of fruit.
- Understanding the local conditions surrounding an orchard is important when attracting pollinators (e.g.: amount of alternative foraging plants / material)

Position of plants

- NSW DPI work demonstrated that the position of a plant within the row influenced fruit weight for both cover types with generally heavier berries at the row edges where the honey bees were accessing the crop.

3.5 Extension & Industry Outcomes

The M&E team acknowledges the diverse channels employed for the dissemination of information and insights arising from the research, including national conferences, newsletters, online content, and talks.

Engagement with key growers and stakeholders by the M&E team were limited due to timing and availability of stakeholders to participate in interviews and surveys.

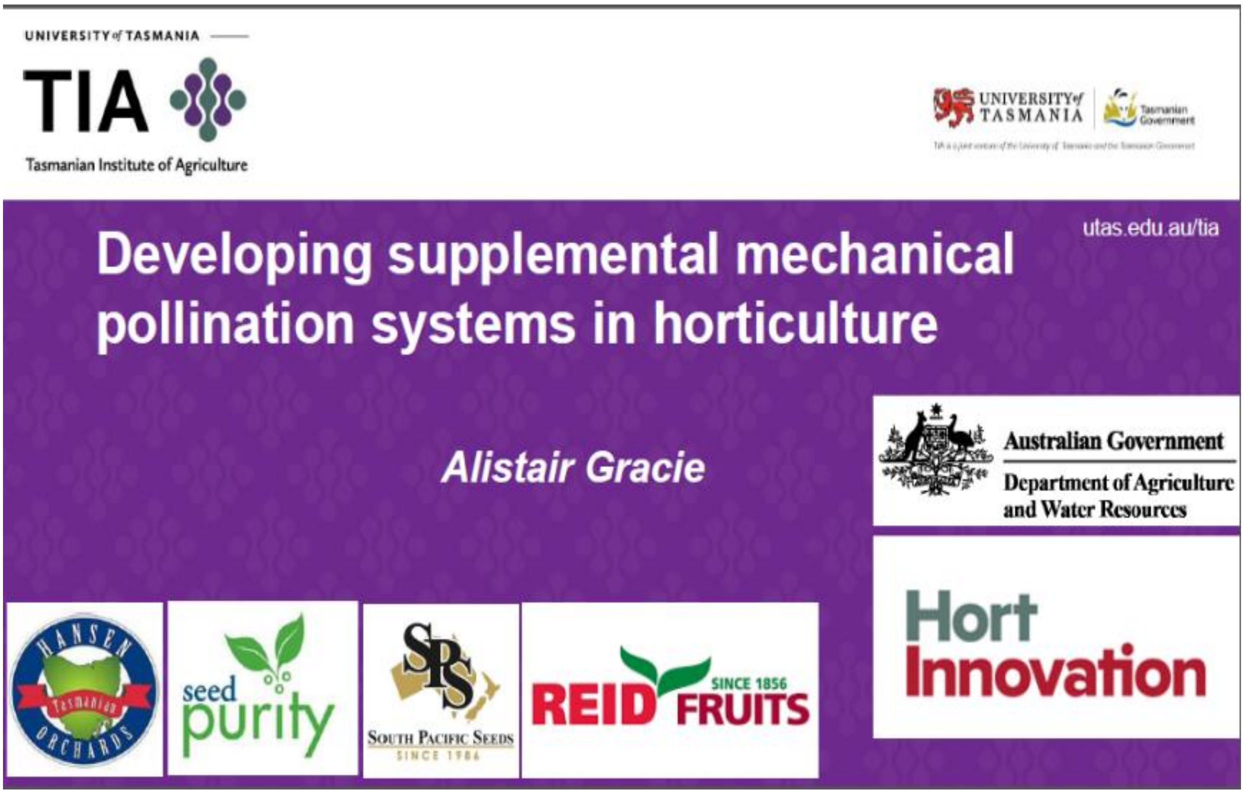
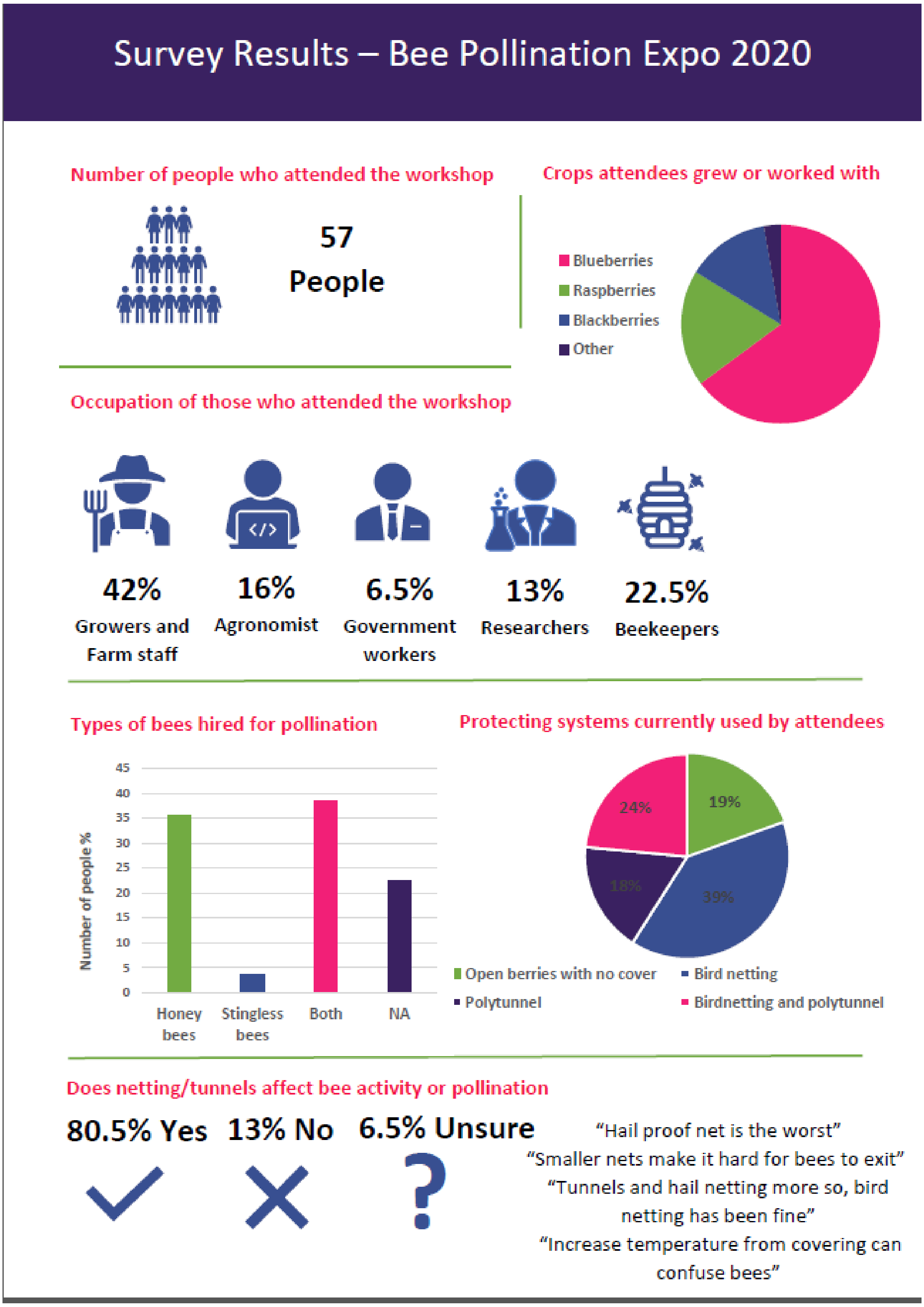


Figure 5. Presentation at PCA conference (2022)
(Source: University of Tasmania)

We commend the effort made by members of the project team to evaluate their activities aimed at raising awareness among growers and landholders, exemplified by the initiatives undertaken by the NSW Department of Primary Industries, as demonstrated at the Bee Pollination Expo (Figure 6).

Surveys undertaken by UTas in the final six months of the project (Figure 7) also provide an example of how data can be collected to demonstrate shifts in awareness, attitude and practice. The survey responses were not available at the time of preparing this report however it is anticipated that they will provide useful insights into any observed shifts by growers.



Is anything being done besides hiring bees to improve pollination?

35.5% Yes

29% No

35.5% NA

"Encourage flies"
"Own stingless bees"
"Trialling stingless bees"
"Improve bee health and honey"
"Cultivars"

How attendees rated their knowledge on pollination prior to and after the workshop

Information received that was of most use

"Hive standards and how to assess these"
"Information from a beekeeper's point of view"
"Location and positioning of hives"
"Communicate with bee-keepers"
"Q&A discussion"
"Increased pollen diversity leads to healthier bees"

What action will you take as a result of the workshop

"Monitor beehive health better"
"Not spray on full moon night"
"Monitor hives under netting"
"Re-think use of chemicals"
"Research sprays less harmful to bees"
"Work on contracts"

Further questions that need to be answered about pollination, bees or covers

"Exact numbers of hives/ha developed by research for highest costs vs reward benefit"
"More information on sprays that effect bees"
"Pros and cons of single vs double box hives"
"Is rotation of hives necessary"
"Should beekeeper's remove hives from growers during spraying for a few days"
"How does hive orientation affect/improve pollination"
"Should growers lift side netting for pollination at all?
"What biosecurity means in bee-keeping"
"Efficiency of non-bees"
"Environmental factors"
"How protecting crops can affect pollen and pollen diversity"

Figure 6. Survey results [Pollination expert Expo, February 2020]
(Source: NSW Department of Primary Industries)

UNIVERSITY of TASMANIA
TIA
Tasmanian Institute of Agriculture

Industry Pollination Workshop Survey 2023

What is your occupation?	☐ Farmer/Landholder ☐ Student ☐ Private Consultant ☐ Farm Manager ☐ Researcher ☐ Government ☐ Rural supplier ☐ Agribusiness ☐ Other (please specify) ____________
How many workshops have you attended?	☐ 2021 ☐ 2022
Which industry do you represent?	☐ Vegetable seeds ☐ Cherries ☐ Apples ☐ Berries ☐ Apiary
Have you found the workshops relevant?	On a scale of 1-5 how do you rate the relevance of the information presented? (Please circle) Irrelevant Highly Relevant 1 2 3 4 5
Do you see pollination as an ongoing issue for your industry? Are you able to provide details?	
Have the workshops improved your understanding of crop pollination?	☐ Fungicide use.......... ☐ Impact of dormancy breakers on flowering.......... ☐ Selection of pollinisers and orchard design (s-allele pollen donors).......... ☐ Alternative pollinators.......... ☐ Bee health and foraging behaviour.......... ☐ Other?..........
Have the workshops changed any of your production practices. If so, how?	
Do you support continued work to develop mechanical pollination systems?	
What other projects would you like to see in pollination research?	
Please provide any other comments or suggestions:	

Figure 7 . Utas Industry Pollination Survey 2023
(Source: University of Tasmania)

While the M&E team is confident that valuable information and insights on enhancing pollination within protected cropping environments have been disseminated and actively shared, the degree of awareness and implementation of these techniques across a wider spectrum of growers is still to be optimised with the finalisation and promotion of Best Practice Guidelines.

The development of Best Practice Guidelines aimed at enhancing honey bee colony performance, specifically targeting the improved pollination of apples, lychees, and berries, is currently underway in the project's final stages. Although proposing this outcome in the final stages of the project is valuable, and the capture of early recommendations in fact sheets (Figure 9) has been achieved, the absence of the Guidelines at the time of reporting is noted.


NSW Department of Primary Industries primefact

Best practice bee management - berries

April 2020, Primefact [number], First edition

Melinda Simpson, Blueberry development officer, Horticulture, Wollongbar

Bees (Honey and Native bees) and other insects play a vital role in pollinating berry crops. Inadequate pollination results in smaller or imperfect fruit since not all seeds and drupelets are formed. Thus, bees and other insect pollinators require protection during flowering to ensure successful pollination of berry crops.

Strategies to consider for protecting bees

Communication

- Communication between beekeeper, grower, spray operator and neighbours are vital especially as honeybees can easily fly two kilometres from their hive to forage on flowers.
- The BeeConnected app is a great digital tool for growers who would like to be informed of, and connected with, beekeepers near their farm, and beekeepers who want to be informed of crop protection activities near their beehives.
- As a BeeConnected user, growers input the location of their property which if within 10 km of a BeeConnected registered hive the app will notify both grower and beekeeper to enable proactive discussion around orchard activities and maintenance of hive health.
- When engaging in commercial pollination services growers and beekeepers should agree upon each other's responsibilities in the following areas:
  - Pesticide applications
  - Number of frames/health of hives
  - Date and location for placement
  - Payment terms
  - Accessibility of colonies to beekeepers
  - Date of hive removal from the orchard

Hive placement and bee care

- Hives should be placed:
  - Where they are accessible and convenient at all hours for servicing and removal
  - With northern and eastern exposures for hive openings to encourage bee flight
  - Away from areas prone to shade or flooding
  - To maximise pollination and guard the bees from drinking pesticide-contaminated water, beekeepers and growers should ensure clean water is always available. Covering or removing water sources for bees before a pest control treatment or emptying and refilling water after a treatment is made is good practice. Water supplies will need to be cleaned and refreshed regularly.
  - Providing alternative pollen sources for good colony strength and bee health should be considered. This could include flowering boundary plants.
  - Hive quality and strength should be assessed upon arrival, either by the beekeeper or a third-party auditor. This inspection should be observed by a grower. Hives should be inspected for colony strength and the other parameters agreed upon between the grower and the beekeeper.
  - Place hives evenly throughout the orchard for better pollination.

Spray management

- Always warn nearby beekeepers of your intention to spray in time for steps to be taken to protect the bees. Give at least two days' notice and advise nearby farmers/ neighbours.
- Spraying should occur late in the afternoon or evening (be aware of inversion layer conditions), when bees are not foraging, and pollen is not present.
- Turn off nozzles when near beehives, even if at night.
- Avoid directly spraying bees in flight or beehives, bees that come in contact with agricultural sprays will not be able to fly because of the weight of the spray droplets on their wings.
- **Always avoid spray drift.** Do not spray in conditions where spray might drift onto adjacent fields which could be supporting foraging bees.
- Mow all flowering weed growth before spraying orchards.

2 | NSW Department of Primary Industries, July 2021

Chemical Choice

- Always choose short acting chemical. Application should finish at least 6 hours prior to bee activity.
- Follow the warnings on pesticide container labels.
- Dispose of waste chemicals or used containers correctly.
- Surfactants are used to improve the effectiveness of chemicals, to decrease the droplet size or to increase the penetration of chemicals on a plant or insect. Surfactants allow the water to penetrate the body hairs, which will kill bees. Reinforcing the message, **do not spray when bees are active**

Acknowledgements

[The (name) program is] a collaboration between the NSW Department of Primary Industries and [other state/industry/NSW agency].

If you are required to use a program identifier or other agency logo, click the icon below and browse for your file, otherwise delete the square. Acknowledgements and copyright/disclaimer should be on the last page of your document.

3 | NSW Department of Primary Industries, July 2021

Figure 8. NSW DPI Primefact on best management practices for bee management in berries (in review). (Source: NSW Department of Primary Industries)

4 COST BENEFIT ANALYSIS

The Rural Research and Development (R&D) for Profit program boosts funding to the rural Research and Development Corporations (RDCs) for nationally coordinated, strategic research that aims to improve farm-gate productivity and profitability and deliver real outcomes for Australian farmers.

All research and development and marketing levy investments undertaken by Horticulture Innovation Australia Limited (Hort Innovation) are guided and aligned to specific investment outcomes, defined through a Strategic Investment Plan (SIP). The SIP guides investment of the levy to achieve each industry's vision.

This cost-benefit analysis follows a conceptual framework for the economic evaluation of projects and programs in the public sector that considers all gains (benefits) and losses (costs), regardless of to whom they accrue.

This analysis involved identifying and briefly summarising the principal economic, environmental, and social impacts in a triple bottom line framework.

4.1 Pollination Economic Value Profiles

Table 2: Economic profile of project crops

Crop	Economic Profile
apples	Apple production is spread across various regions in Australia, with different varieties thriving in different climates. Some of the major apple-producing states include Victoria, New South Wales, South Australia, and Tasmania. According to apple and pear Australian limited[1], for the year 2020- 21, Australia's apple industry was valued at $527 million. There are over 500 commercial apple and or pear growers in the country. All states produce apples. in 2018- 19, Australians ate on average 10 kilograms of apples per year which amounts to more than one apple a week. Australia currently exports one- 2% of our total marketable production of apples. The Australian apple industry has seen investments in technology and research to enhance production efficiency and improve apple varieties in terms of taste, appearance, and storage capabilities.
blueberries	The blueberry industry in Australia has been experiencing significant growth in recent years. Favorable climate conditions in certain regions, along with increased consumer awareness of the health benefits of blueberries, have contributed to the expansion of blueberry farming. Blueberry production has increased from 5,500 tonnes in the year ending 2014 to 17,000 tonnes in the financial year ending June 2018. [2] Most blueberries grown in Australia are for the domestic market. Australia has been exporting blueberries to various international markets, particularly to countries in Asia. The rising awareness of the health benefits of blueberries has also led to

[1] https://apal.org.au/programs/industry-data/industry-stats/

[2] Berries Australia (2023)

Crop	Economic Profile
	increased domestic consumption. Blueberries are often promoted as a superfood, Australian farmers have been investing in research and development to improve blueberry varieties that are well-suited to local growing conditions. This focus on varietal development aims to enhance yield, quality, and overall economic viability.
cherries	Cherries are a seasonal fruit in Australia, typically harvested during the summer months (November to February). The seasonality of cherry production can influence market dynamics and pricing. Some of the major cherry-producing regions in Australia include Tasmania, Victoria, New South Wales, South Australia, and Western Australia. Different varieties of cherries are grown in these regions, contributing to the overall diversity of the Australian cherry market. Australia exports a significant portion of its cherry production to international markets, particularly Asia. Cherries are popular among Australian consumers, and domestic consumption also contributes to the economic value of the cherry industry. Supply chain inefficiencies and barriers to trade may limit export trade, particularly in developing Southeast Asian markets as the quality of product may deteriorate before it reaches the final consumer. Investments in technology, orchard management practices, and research contribute to improved cherry production, helping maintain high-quality standards and potentially increasing economic returns for growers.
onions	Onions are a significant vegetable crop in Australia, and they are grown across various regions of the country. Different varieties of onions are cultivated to meet both domestic and export demands. The Australian onion industry provides year-round production, with most of the production based in South Australia (48%) and Tasmania (22%). Lower seasonal volumes are available from Queensland (12%), along with smaller volumes in Western Australia (10%), Victoria (6%) and New South Wales (2%). 271,930 tonnes of onions were produced in 2020/21, with a farm gate value of $203.2 million. Fresh domestic demand continues to be the focus of the industry, which accounted for 75% of the Australian onion supply in 2020/21. [3] Onions were Australia's second largest vegetable export by volume after carrots in 2020/21, reporting an export volume of 44,885 tonnes with a value of $30.4 million. The Export value of onions fluctuates year on year based on short-term global trading conditions. In addition to fresh onions, there is a market for processed onion products and value-added items such as onion powder and onion-flavoured products. This diversification can contribute to the overall economic value of the onion industry.

[3] Source: https://www.horticulture.com.au/globalassets/hort-innovation/levy-fund-financial-and-management-documents/annual-investment-plans-pdfs-202223/hort-innovation-aip-2022-23-onion.pdf

4.2 Project Investment

An investment in the Pollination project by the Department for Agriculture, Fisheries and Forestry through the Rural R&D for Profit program has been made through a contract established with Hort Innovation who subsequently established contracts with project partners for the delivery of key activities and outcomes while retaining responsibility for managing and administrating the project.

In addition to this investment, further contributions to the project were provided though investments from:

- Growers to support research trials.
- Industry sectors in the promotion of research findings through publications, conferences and online.
- Project partner research institutions in the provision of postgraduate students, facilities and internal collaborations.

4.3 Impacts

Table 3 provides a summary of the principal types of impacts delivered by the project. Impacts have been categorised into economic, environmental, and social impacts.

Table 3: Triple Bottom Line Categories of Principal Impacts from the Pollination Project

	Grower	Industry	Government
COSTS (Direct & Opportunity)			
Economic			
Pollination	Reduced production associated with suboptimal pollination management.	Reduced production associated with suboptimal pollination management.	Opportunity cost arising from impact on Gross Domestic Product
Production costs	Increased cost of hives to achieve pollination as beekeepers reluctant to jeopardise hive health.	Reduced ability to attract growers to the industry if perceived low profitability.	
Social			
	Impact on household income due to decreased profitability		Negative regional community wellbeing as a spill-over impact from less productive crop growing regions.
Environmental			
	Production risk associated with loss of honey bee as pollinator.		Biosecurity risk associated with honey bee health and subsequent impacts on production and food security.
BENEFITS			

	Grower	Industry	Government
Economic			
Pollination	Increased fruit size and quality increasing returns.	Market access and reputation Ability to grow the sector	Gross Domestic Product impact
Social			
Whole of project	Increased knowledge and research capacity.		
	Some potential future contribution to improved regional community wellbeing as a spill-over benefit from more productive crop growing regions		
Environmental			
	Optimising pollinators other than honey bees (including mechanical methods)	Effective management of biosecurity risks associated with honey bees	

4.4 Public versus Private Impacts

There is a large reliance on honey bees for pollination in all the target industries which exposes the industry to risks related to availability of managed honey bees, seasonal variations in wild honey bee populations and issues associated with use of honey bees in covered cropping. Mechanical pollination, manipulation of floral traits and beehive management, thus offers exciting opportunities to significantly improve pollination of horticultural and vegetable seed crops.

Impacts identified in this evaluation are both public and private in nature. At the national and regional scale increased productivity arising from optimal pollination practices will contribute to stronger industries, reliable production for regional economies; whilst at the farm level, growers can better plan orchard design and improve pollinator management leading to increased productivity. All these aspects can improve profitability for growers and industry stakeholders.

Some public benefits may occur and include improved food security outcomes through increased productivity as well as potentially increased incomes in the horticultural crop growing communities/regions associated with a more productive industry.

In the public realm, food security can be supported by improved pollination practices and management of risks associated with honey bee health.

Impacts on Other Australian Industries

Impacts on industries other than the crop industries targeted in this project may include potential gains to other horticultural industries (e.g. other fruits, nuts and vegetables reliant on honey bee pollination) via potential future spill-overs from the increase in knowledge and scientific capacity.

4.5 Match with National Priorities

The Australian Government's Science and Research Priorities and Rural R&D 4Profit priorities are reproduced in Table 4. The project findings and related impacts will contribute to Rural R&D 4Profit Priorities 1, 4 and potentially some contribution to Priorities 2 & 3, and to Science and Research Priority 1.

Table 4: Australian Government Research Priorities

Rural Research and Development for Profit Priorities	Australian Government Science and Research Priorities
1. Advanced technology 2. Biosecurity 3. Soil, water and managing natural resources. 4. Adoption of R&D	1. Food 2. Soil and Water 3. Transport 4. Cybersecurity 5. Energy 6. Resources 7. Advanced Manufacturing 8. Environmental Change 9. Health

Source: Australian Government, 2023

5 EVALUATION INSIGHTS

5.1 External factors

In the first 18 months – 2 years, the project had to operate within the unprecedented global health pandemic environment of Coronavirus (COVID-19). This meant that major adjustments were made to meet the legal requirements of physical distancing and the inability to cross state boundaries. As a result, the opportunity for research activities to be replicated across multiple interstate locations in this period were lost as were opportunities for collaboration between researchers. In some cases, COVID 19 resulted in the delay of some project components and increased costs in the delivery of some components.

Drought was a major issue when the project commenced in 2019.The drought has been marked by three consecutive extremely dry winters, adversely affecting the amount of data that could be collected due to the impacts on crop production cycles and the reduced windows of opportunity to undertake research activities.

The project has also been impacted by the unprecedented bushfire season of 2019 - 2020 that started early (in early Spring rather than early Summer) due to drought conditions. In mid-January 2020, 17 million hectares (46.03 million acres) was burnt or was burning across all Australian State and territories. One Adelaide Hills grower participating in the research was burnt out and left with only 80% of their trees. They have had to re-net sections of their orchards which has impacted on annual work cycles. Continued participation post fire was restricted to crop areas that were unburnt.

The combination of the above mentioned three factors has created unparalleled conditions and project circumstances for this project to which project partners have had to respond. Added to this has been the project extension with no additional resources allocated.

5.2 Value of pre-existing relationships

Many partners have pre-existing relationships in the sector that they are working within.

These pre-existing relationships demonstrate to stakeholders that the project partners both understand local / sectoral issues and offer solutions for continuous improvement. This high quotient of trust has proved invaluable in terms of the ability to:

- Engage growers in the research activities.
- Build industry understanding and value of research overtime.
- Encourage growers to try new things.
- Increase understanding of grower needs and expectations.

5.3 Collaboration

Project partners viewed the opportunity to collaborate with each other very positively and spoke highly of the professional and collegiate relationships established.

Notwithstanding these sentiments, the evaluation team has found that the level of collaboration did not achieve that which was originally anticipated or expected. The project was initially designed to build on the collaborative efforts of researchers to ensure that research findings could be applied across regions. The intention of aligning methodologies, replicating trials, and site-visits, was not able to be achieved.

The COVID pandemic halted early collaboration efforts and dampened the momentum for collective work. Consequently, disparities in methodologies and trial designs emerged, hindering the replication of research to the extent initially anticipated. There is a sense amongst project partners that project team activity has been more insular resulting in reduced geographical datasets along with modified involvement of project team contributions to research elements.

This resulted in some reticence from researchers regarding the applicability and appropriateness of individual findings beyond their local regions.

5.4 Project Extension

The Pollination project received a twelve-month extension due to the impacts of external factors outlined previously. The government weren't able to provide additional resources to support the extended timeframe.

One of the project challenges has been the ability to maintain research momentum including the retention of graduate students and high-level research personnel. A range of factors can impact on this including the following: limited funding to attract and retain top tier PhD students and research personnel; Competition amongst university and research institutions to recruit talent; Wage disparities for PHD students and research personnel; funding availability; and that many research positions are offered on short term contracts which can create uncertainty and instability for project teams.

Notwithstanding any resource challenges, project partners used the extension to prioritise the continuation of research activities conducted in somewhat isolated units by individual researchers across various project elements.

5.5 Research Risks and Limitations

The complexity and multi-faceted nature of the research undertaken by project partners is worthy of acknowledgement.

The Literature Review undertaken early in the project proved to be a piece of work that had long been overdue for researchers in this field. In developing this project, the knowledge gaps regarding enhancing and optimising pollination were 'guessed' as there had not been a comprehensive review of the literature in the past to identify what was known and where there were knowledge gaps. The Literature Review clearly established that this project would contribute to addressing these gaps and building the body of knowledge in this field.

Across the project team and with individual project partners several limitations and risks emerged in relation to research activities.

Inherent in any research there are design and methodology challenges. Some of those identified by project partners included:

- Getting access to suitable crop sites for trials to enable replication and trial robustness proved difficult both within Australia and New Zealand.
- Data sets in NZ and Australia are different across pollination research activities.
- Managing the number of variables inherent at research sites – varieties, cover types (colour, material, open, closed).
- Reliance on published works to inform methodology.
- Blueberries bud, flower, and fruit at the same time, making the window for research smaller and shorter.
- For best / most robust research outcomes ideally like crops both under cover and in the open would be compared – this has not always been possible, meaning that results may need to be extrapolated.

Beneficial elements of the research approach have included:

- Undertaking the Literature review early in the project established gaps in knowledge and confirmed the value of research questions that were funded.

- Engaging statisticians early in the project has been beneficial in supporting research design and ensuring trial data robustness.

5.6 Research funding

In general, research funding opportunities are short term, low investment and extremely competitive and as such generally don't have the time, breadth of expertise or technologies nor funds to achieve the required outcomes. The 'RR&D for Profit scheme' has provided a unique opportunity for collaboration between multiple crop industries, research institutions, and government to work together to address common themes. The successes achieved by this project are the direct result of this collaborative, well resourced, multi-industry funding model.

5.7 Cost Benefit Analysis

The investment in the Pollination project has delivered insights into pollination, along with improved knowledge and scientific capacity. Though the project produced no direct, quantifiable impacts, the Pollination project is likely to contribute to future potential impacts for the horticulture industries involved in the project, including improved pollination for crops under covers, improved management of risks associated with honey bee biosecurity threats, and enhanced regional community wellbeing as a spill-over benefit from more productive horticultural industries. Any such future benefits may be partially attributable to the investment in this project.

5.8 Project Management

The Project's governance structures, delegations, and reporting requirements supported the effective delivery of the project and provided appropriate levels of accountability. Feedback early in the project identified some areas where effectiveness and efficiency could be improved.

The late contract signing from the Federal Government created an approximate 3-month delay in formal project commencement for Delivery partners. There was no factoring in of a contract buffering period within project plan timeframes despite contracts taking up to 6 months in some instances.

The provision of stable project management by Hort Innovation throughout most of the project was viewed as a positive by project team members. The responsiveness of Hort Innovation in supporting and seeking an extension of time for project delivery was welcomed and enabled project partners to overcome some of the challenges brought about by COVID-19. Project partners indicated their appreciation of the level of communication and support provided by Hort Innovation.

However, the project would have benefited from some additional collaborative and or governance structures to both encourage / drive better integration of the dual benefits of research and findings and sharing of communication and outcomes particularly given the challenges arising from the external factors detailed previously.

An integrated project management model, delegating responsibility for a level of project coordination to a project team member, could have assisted with streamlining the broader integration and application of findings across industry personnel.

5.9 Project Scope

It is possible that the original scope was overly optimistic and that this project would have benefited from:

- Recognising the possibility of a multi-layered research approach may be required as opposed to assuming a simple cause and effect scenario.
- Establishing realistic timeframes for research activities and data analysis taking into account

seasonality of data collection.

- Recognising the need for separate research and extension phases, with a focus on developing extension activities based on research outcomes.
- Including contingencies in timeframes / planning to allow for unexpected external factors that can adversely impact undertaking research on seasonal crops.

Findings from this project also highlight the time challenges in not only gathering but also analysing data. In future, researchers may also wish to factor in more time to analyse data including the possibility of needing to revisit sites to verify data, and time to ensure that adequate resources are in place to undertake the work (e.g.: PhD students and other specialists).

5.10 Challenges of extension with growers

Based on the information provided to the M&E team, it is evident that the project has reached numerous growers through a variety of channels, including forums, newsletters, formal events, and on-site presentations. The data supplied to the M&E team also suggests that a small number of growers have actively participated and directly benefited from the adaptive research and initial discoveries towards the culmination of the project.

Given the development of Best Practice Guidelines hinges on research outcomes, the project timeframe left little time for translating these findings into such guidelines. The development of Best Practice Guidelines is currently underway in the project's final stages.

While the M&E team is confident that valuable information has been successfully disseminated to growers, the degree of awareness and implementation of these techniques across a wider spectrum of growers remains uncertain and will be enhanced by the finalisation and promotion of the Best Practice Guidelines.

6 RECOMMENDATIONS

6.1 Project Management

- In future projects, incorporate a Contract buffering process that includes a timely/well communicated contractual negotiation period.
- Streamline governance structures and reporting requirements within the project team structure to enhance project efficiency.
- Ensure stable project management, governance, and effective communication support throughout the project lifecycle.
- Incorporate regular meetings with project collaborators, and partners, keeping the focus on milestone due dates and at the same time providing opportunities for knowledge sharing, collaboration, and networking.
- Ensure that monitoring and evaluation is:
    - undertaken by an impartial third party
    - Embedded from the project inception
    - Focuses on (amongst other things) building capacity and knowledge across the team
    - Part of the researchers' KPIs.

6.2 Adapting to External Factors

- Develop flexible programs and funding models that can extend research to address external factors like pandemics, natural disasters, resource, travel, and climate-related challenges.
- Establish remote collaboration platforms and methodologies to ensure project continuity during crises.
- Allocate resources for unforeseen delays and increased costs due to external factors.

6.3 Research Activities

- Embed collaboration efforts into the project expectations / KPIs from the outset to encourage and / or facilitate interdisciplinary collaboration to maximise the project's impact.
- Prioritise practical collaboration over 'siloed' research activities.
- Minimise disparities in methodologies and trial designs to ensure research findings can be applied across regions effectively.
- Where trial disparities arising field conditions and external factors are likely to occur, the inclusion of statistical methods analysis expertise will become critical to addressing these challenges.
- Identify strategies to attract PhD students to project e.g. incentives, high value scholarships.
- Incorporate contingencies in project timeframes to accommodate unexpected external factors.
- Allocate sufficient time for data gathering, analysis, and resource management.
- Maximise the benefits of project extensions by maintaining momentum, retaining key personnel like graduate students, and reviewing collaboration opportunities.

6.4 Engagement and Extension

Internal focus

- Provide adequate support and resources for extension activities alongside research efforts through an additional 'extension phase' with a focus on developing extension activities based on completed research outcomes.

- Establish clear internal feedback mechanisms to facilitate information sharing and collaboration among project team members.
- Build confidence among researchers to translate early findings into actionable industry recommendations through active testing and refinement.
- Focus on developing key products/collateral etc that consolidate project findings and recommendations.

External focus

- Keep industry engagement: easy, uncomplicated, non-time consuming and inexpensive.
- Connect to existing industry engagement platforms i.e. websites, papers, magazines, seminars etc.
- Utilise industry and regional 'champions' to support increasing grower awareness and adoption of project outcomes.
- Continue to identify opportunities for involvement of growers and industry personnel in the delivery/achievement of project activities and milestones to enhance engagement and awareness outcomes.
- Continue to keep industry informed of updates (big and or small) to ensure project momentum, interest, and buy in.

6.5 Evaluation

- Incorporate evaluation support into future projects.
- Build evaluation into projects early in the process and engage all partners / team throughout the development.
- Undertake pre- and post-project grower/industry surveys to better understand shifts in knowledge, attitude, skills and practice change arising from project activities.
- Identify key project beneficiaries for involvement in evaluation activities throughout the project.
- Make evaluation easy to participate in.
- Build knowledge, capability, and confidence in your partners / team.
- Assess the long-term impacts and potential benefits of the project beyond direct, quantifiable measures such as: industry productivity, community well-being, and scientific knowledge.
- Continue to build capacity of project partners in evaluation through training and skill development.

7 Concluding Statement

The Pollination program's aim has been to enhance the yields and quality of fruit and vegetable seeds cultivated in protected cropping environments through the development of cutting-edge pollination technologies.

The evaluation process indicates that the project has met its Key Project Indicators across the complex research elements undertaken throughout the project. An assessment of the project's objectives highlights that solid outcomes have been achieved across all four of the project objectives, with the application of the research outcomes across industry more broadly, still in its early stages.

The M&E team has observed a commendable level of collaboration among project partners and a close-knit network of growers across Australia in the delivery of the project objectives. These relationships are predominantly characterised by strength, positivity, and professionalism.

APPENDIX 1: MONITORING AND EVALUATION FRAMEWORK

APPENDIX 2: M&E TEAM ACTIVITY SUMMARY

Table 1. Activity summary

Output	Description	Detail
Project Plan (Oct 2019)	Project Plan developed	Project management plan finalised
Workshop (Oct 2019)	Project Team Workshop	Delivery of workshop session which resulted in increased capacity of project team members to develop and apply program logic and evaluation frameworks.
Program Logics (Dec 2019)	Five program logic frameworks	Five program logic frameworks were developed. These were based project partner outcomes: 1. Department of Primary Industries NSW 2. University of New England 3. University of Adelaide 4. University of Tasmania/Seed Purity Pty Ltd 5. Plant & Food Research
Consolidated Program Logic (Jan 2020)	Program Logic	Five program logics consolidated into one program logic.
Evaluation Plan (Feb 2020)	Evaluation Plan	Evaluation plan developed encompassing monitoring and evaluation activities across all five project areas.
Project Team support (Nov 2019 – January 2024)	Email, telephone & online meeting support	Ongoing support provided to project team members as they implemented their Monitoring and Evaluation activities across their respective project activities.
Project Team Meetings • **29 April 2020** • **12 August 2020** • **22 April 2021** • **20 August 2021** • **7 April 2022** • **15 May 2023**	Project Team Progress meetings	Attendance at project team meetings (online and face-to-face). Provision of M&E update presentations.
Evaluation Interviews	20 x meetings	• (21/5/20) Sophie Parks • (22/5/20) Katja Hogendoorn & Scott Groom • (27/5/20) Romina Rader • (28/5/20) Melinda Simpson & Sophie Parks • (3/6/20) Lisa Evans • (5/6/20) Alistair Gracie • (19/6/20) Alistair Gracie • (11/12/21) Romina Rader • (11/12/21) Katja Hogendoorn + Scott Groom • (14/1/212) Alistair Gracie • (3/2/22) Dr. Sophie Parks • (25/2/22) Romina Rader • (9/3/22) Alistair Gracie • (15/3/22) Katja Hogendoorn • (7/12/23) Sophie Parks • (7/12/23) Romina Rader • (18/12/23) Robert Green • (18/12/23) Katja Hogendoorn • (9/1/24) Alistair Gracie • (15/1/2024) Lisa Evans
Survey Results (Dec 2020)	Distribution of online survey to project partners	Questions explored the following issues: • Milestone achievement • Project management

Output	Description	Detail
		• Stakeholder engagement • Project Beneficiaries • Challenges Survey results are here: https://www.surveymonkey.com/stories/SM-DXZNWR72/
Survey Results (Feb 2022)	Distribution of online survey to project partners	Questions explored the following issues: • Milestone achievement • Project management • Stakeholder engagement • Project Beneficiaries • Challenges Survey results are here: https://www.surveymonkey.com/stories/SM-tPJ4TRVfETBnyYaG_2F7h3kQ_3D_3D/
Survey Results (Jan 2024)	Distribution of online survey to project partners	Questions explored value and impact of evaluation team Survey results are here: https://www.surveymonkey.com/stories/SM-mkkou4YEjTyVgnDJ0Ny21A_3D_3D/
Analysis (April 2020 – Jan 2024)	Review of Project Partner milestone reports	Receipt and analysis of Milestone Reports from the following project partners: •
Milestone Reports & Annual Reports	MS101 MS102 MS103 MS104 (Annual report) MS105 MS106 MS107 MS108 (Annual report) MS109 MS110 MS111 MS112	Milestone and Annual reporting as per project schedule requirements
Midterm Evaluation Report (Jan 2021)	Midterm Evaluation Report	Midterm evaluation report
Evaluation Team Performance Survey Results (Jan 2024)	Online survey	Survey distributed to project team members seeking feedback on the effectiveness and impact of the Evaluation Team. Survey results are here: https://www.surveymonkey.com/stories/SM-GK5C3gBgWDkfYNQfSuz2Ig_3D_3D/
Final Evaluation Report	Final Evaluation Report	End of project evaluation report (February 2024)

APPENDIX 3: EVALUATION TEAM PERFORMANCE

Figure 1: Evaluation Team performance

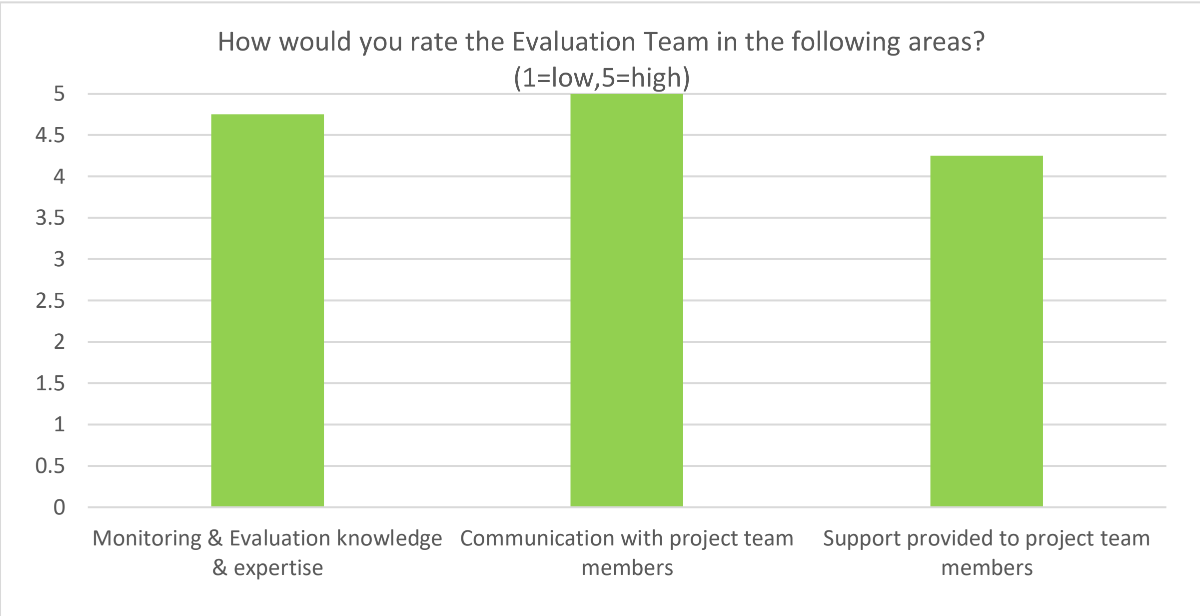


Figure 2: Impact on project team behaviour

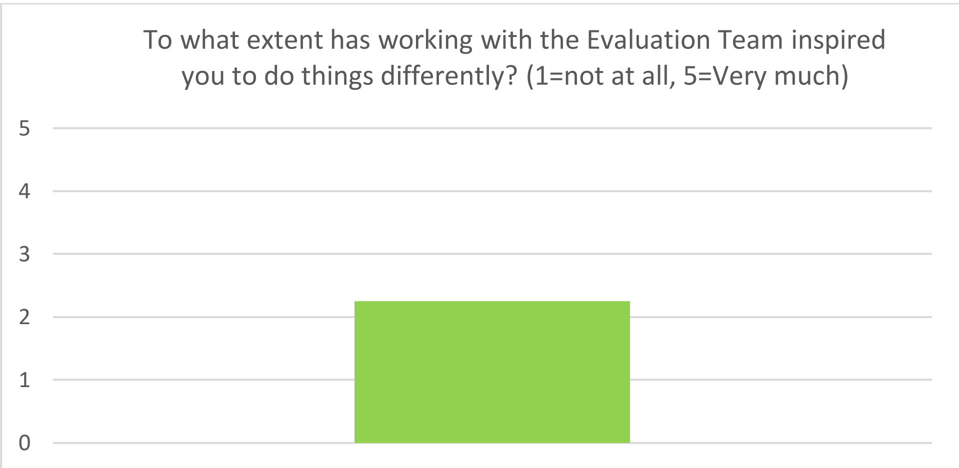


Figure 3: Recommendation for use of Evaluation Team

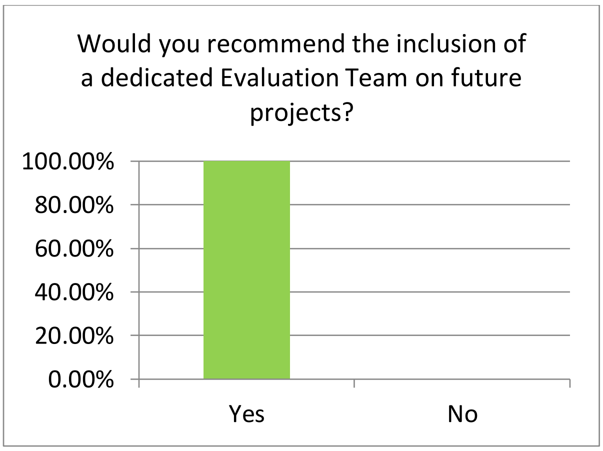


Figure 4: Benefits arising from Evaluation team involvement

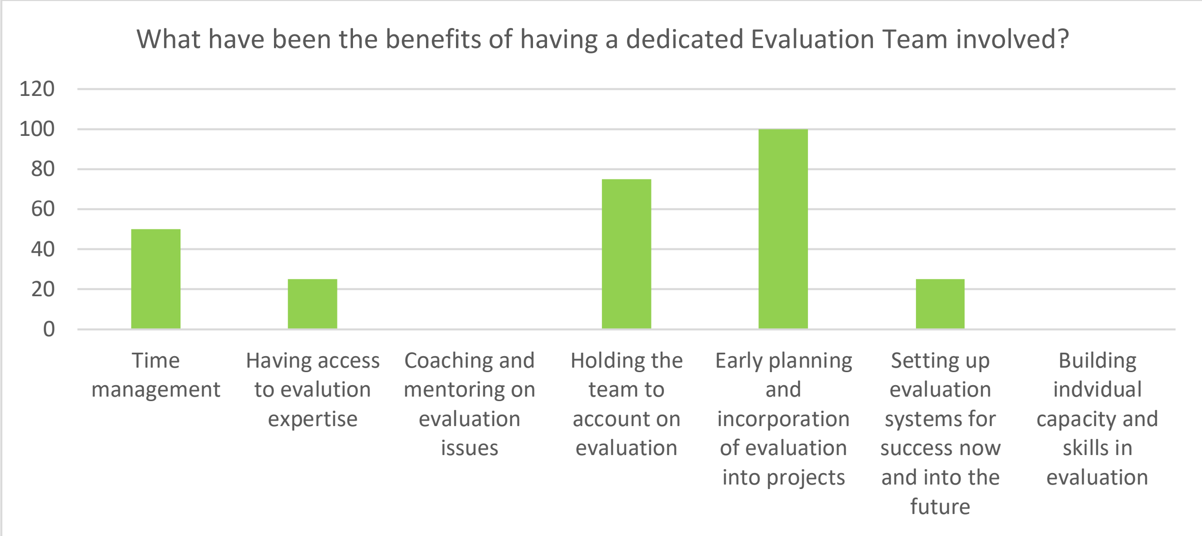
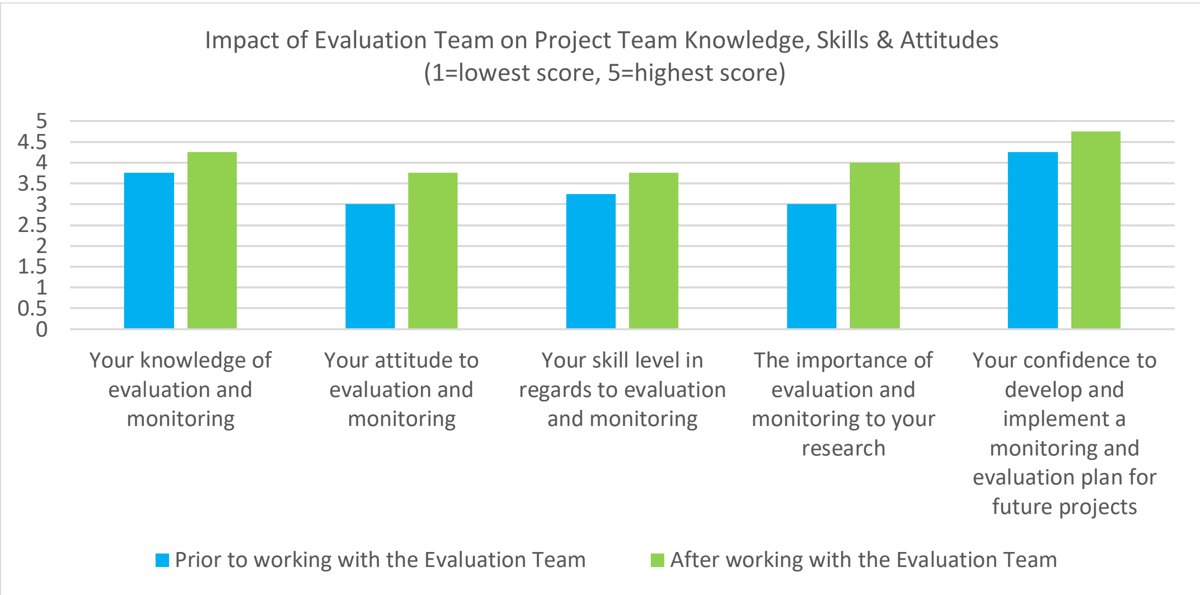


Figure 5: End of project impact of Evaluation Team



Project Team Feedback

- Funnily enough, your involvement provided some cohesion in the project.
- You have been great.
- I would not want to do a project without external M&E.
- Nothing. They were good. It was great not to have to do the M&E myself, as it is awkward to produce your own FIGJAM, and isn't credible.
- You have been great but could have picked up on the need of the researchers to produce a sound complementary project and facilitated the development of that.
- (Support for inclusion of external evaluation team) In principle, yes, but in practise, it depends on the costs. Whether it is cost effective depends on the size and the aims of the project.
- My only issue - systemic to the funding providers' expectations- is that the adoption outcomes are always unrealistic when the aim of a project is to better understand a problem rather than create a solution for adoption. This makes evaluation less valuable than it should be.

The full results of the survey can be found here.
https://www.surveymonkey.com/stories/SM-mkkou4YEjTyVgnDJ0Ny21A_3D_3D/

7.4 Budget

Budget and Costs

Description of expenditure	Expenditure Item	Grant Contributions	Other Contributions		Total Costs
			Grantee	Third Parties	
Expenditure in 2018-19					
Hort Innovation					
Contract set up	finalisation of project methodology, project plan put in place, milestones date and payments agreed upon, signed contracts executed		2,000.00		2,000.00
Steering committee set-up	preliminary list put together, members recruited, ToR developed and agreed, first meeting organised		1,000.00	1,000.00	2,000.00
University of Tasmania					
Staff	Inkind: Fraction of staff time from TIA, SeedPurity, SPS, Hansen Orchards & Reid Fruits in planning and designing experiments			57,549.00	57,549.00
University of New England					
Staff resources	All fixed term and casual staff including postdoc, research fellows and technical officers	98,952		15,970	114,922.00

Expenditure in 2019-20					
University of Adelaide					
Staff resources	UoA senior postdoc salary (0.5 FTE plus on-costs), liaison with industry and extension, project management, administration and industry support, advice, steering committee /hive management PhD scholarship	170,000.00		215,250.00	385,250.00
Equipment	experimental netting, robotic unit			20,000.00	20,000.00
Field work expenses	pots and plants, travel to field sites, hive hire, consumables, molecular analysis			13,000.00	13,000.00
lab work	molecular analysis			10,000.00	10,000.00
Plant and Food Research Australia					
Staff resources	Plant and Food Research scientist salaries (0.3 FTE and 0.06 FTE), Senior RA salary (0.3 FTE), Technicians salaries (0.1 x 3), and funds to cover casual workers	197,200.00		244,000.00	441,200.00
Equipment and services contracts	Honey bee hives + transport, consumables, compensation for fruit removal	24,650.00		30,500.00	55,150.00
Travel	Flights to field sites and conference/ workshop presentations,	24,650.00		30,500.00	55,150.00

	Accommodation for staff, vehicle hire + fule				
University of New England					
Staff resources	All fixed term and casual staff including postdoc, research fellows and technical officers	180193.00		378349.00	558,542.00
Facilities	laboratory facilities, library, glasshouse use			20500.00	20,500.00
Equipment	mechanical and cross pollination technologies / equipment to facilitate field insect and plant collection/ field trial equipment	18754.00			18,754.00
field work expenses	travel to field sites, hive hire, consumables	38900.00			38,900.00
lab work	flower reproduction and physiology analyses	25300.00			25,300.00
University of Tasmania					
Staff	2 FTE Level B Step 6 @ $130,248 including oncosts; laboratory and Field Research Technician 0.75FTE@ $99,041; 600hrs casual labour @ $40.71/hr including oncosts. Inkind: Fraction of staff time from TIA, SeedPurity, SPS, Hansen Orchards & Reid Fruits in planning and designing experiments	226,165.48		288,832.52	514,998.00

Operating costs	Laboratory and office consumables ($22,000), initial engineering of pollen harvesting and applicator equipment ($46,000 for perennial and annual systems); Ampha Z32 impedance flow cytometer with software, chips, buffers and sampling equipment for pollen production and viability testing ($86,792); field trial costs ($25,000) . In kind contributions from industry partners	110,486.07		123,505.93	233,992.00
Travel	Interstate travel: 8 return airfares Hobart to trial site and meeting locations in NSW and SA per year (@$1500 ea), 32 days accommodation including self catering (@$200/day); Intra state travel @ $130 car hire/day for 25 days per year each for annual and perennial crops	18,394.00		6,506.00	24,900.00
NSW DPI					
Staff resources	NSW DPI salaries (1 person 20% of time, 1 person 5% of time and another at 10% of time), and funds to cover casual workers and a part time technical officer	88,750		68,080	156,830.00

Equipment	Site preparation and monitoring equipment and installation soil moisture probes, sap flow sensors, temperature humidity sensors. Spanish high tunnels and netting.	60,000		225,000	285,000.00
Field work expenses	Accommodation for staff, vehicle hire + fuel, compensation for fruit removal	20,000		10,000	30,000.00
Travel	Flights to field sites, conference/ workshop presentations	10,000		5,000	15,000.00
Operating	Coordination and delivery of industry meetings, industry publications, consumables for data collection at field sites and laboratory analyses of samples.	20,000		5,000	25,000.00
Hort Innovation					
Project consortium governance	Provide an account of completed activities undertaken through the project.	23,991.74	1,000.00	1,000.00	25,991.74
	Four Project Reference Committees held	119,958.72	5,000.00		124,958.72
	Monitoring and Evaluation Plan reported on, Benefit cost analysis undertaken	23,991.74	1,000.00		24,991.74
	Project management, Financial audit conducted, Report on project legacy arrangements	47,983.49	2,000.00		49,983.49
Expenditure in 2020-21					

University of Adelaide					
Staff resources	UoA senior postdoc salary (0.6 plus on-costs), PhD scholarship, advice, steering committee /hive management, liaison with industry and extension, project management, administration and industry support	171,000.00		191,750.00	362,750.00
Equipment	experimental netting			3,000.00	3,000.00
field work expenses	travel to field sites, hive hire, consumables			9,000.00	9,000.00
lab work	molecular analysis			10,000.00	10,000.00
travel	conference presentations			1,500.00	1,500.00
Plant and Food Research Australia					
Staff resources	Plant and Food Research scientist salaries (0.3 FTE and 0.06 FTE), Senior RA salary (0.3 FTE), Technicians salaries (0.1 x 3), and funds to cover casual workers	197,200.00		244,000.00	441,200.00
Equipment and services contracts	Honey bee hives + transport, consumables, compensation for fruit removal	24,650.00		30,500.00	55,150.00
Travel	Flights to field sites and conference/ workshop presentations, Accommodation for staff, vehicle hire + fule	24,650.00		30,500.00	55,150.00
University of New England					
Staff resources	All fixed term and casual staff including postdoc,	179473		381749	561,222.00

	research fellows and technical officers				
Equipment	mechanical and cross pollination technologies / equipment to facilitate field insect and plant collection/ field trial equipment	27,500			27,500.00
field work expenses	travel to field sites, vehicle hire/accommodation; nets, vials other consumables, chemical use	38,900			38,900.00
lab work	sample analyses	98,574			98,574.00
travel	conference presentations	3,500			3,500.00
University of Tasmania					
Staff	2 FTE Level B Step 6 @ $133,504 including oncosts; laboratory and Field Research Technician 0.75FTE@ $101,517; 870hrs casual labour @ $41.73/hr including oncosts. Inkind: Fraction of staff time from TIA, SeedPurity, SPS, Hansen Orchards & Reid Fruits in planning and designing experiments	239,644.00		297,338.00	536,982.00
Operating costs	Engineering costs associated with modification of prototype pollen harvesting and delivery systems for perennial and annual systems ($35,334); Laboratory and office consumables ($25,151);	53,526.00		91,159.00	144,685.00

	field trial costs ($30,000). In kind contributions from industry partners.				
Travel	Interstate travel: 8 return airfares Hobart to trial site and meeting locations in NSW and SA per year (@$1500 ea), 32 days accommodation including self catering (@$200/day); Intra state travel @ $130 car hire/day for 25 days per year each for annual and perennial crops	18,394.00		6,506.00	24,900.00
NSW DPI					
Staff resources	NSW DPI salaries (1 person 20% of time, 1 person 5% of time and another at 10% of time), and funds to cover casual workers and technical officer costs	88,750		105,226	193,976.00
Field work expenses	Accommodation for staff, vehicle hire + fuel, compensation for fruit removal	10,000		10,000	20,000.00
Travel	Flights to field sites, conference/ workshop presentations	10,000		10,000	20,000.00

Operating	Coordination and delivery of industry meetings, industry publications, consumables for data collection at field sites and laboratory analyses of samples.	20,000		10,000	30,000.00
Hort Innovation					
Project consortium governance	Provide an account of completed activities undertaken through the project.	23,991.74	1,000.00	1,000.00	25,991.74
	Four Project Reference Committees held	119,958.72	5,000.00		124,958.72
	Monitoring and Evaluation Plan reported on, Benefit cost analysis undertaken	23,991.74	1,000.00		24,991.74
	Project management, Financial audit conducted, Report on project legacy arrangements	47,983.49	2,000.00		49,983.49
Expenditure in 2021-22					
University of Adelaide					
Staff resources	UoA senior postdoc salary (0.5 plus on-costs), PhD scholarship, advice, steering committee /hive management, liaison with industry and extension, project management, administration and industry support	175,800.00		178,250.00	354,050.00
Equipment	experimental netting			3,000.00	3,000.00
lab work	molecular analysis			7,000.00	7,000.00

field work expenses	travel to field sites, nest, consumables			8,000.00	8,000.00
travel	conference presentations			1,500.00	1,500.00
Plant and Food Research Australia					
Staff resources	Plant and Food Research scientist salaries (0.3 FTE and 0.06 FTE), Senior RA salary (0.3 FTE), Technicians salaries (0.1 x 3), and funds to cover casual workers	197,200.00		244,000.00	441,200.00
Equipment and services contracts	Honey bee hives + transport, consumables, compensation for fruit removal	24,650.00		30,500.00	55,150.00
Travel	Flights to field sites and conference/ workshop presentations, Accommodation for staff, vehicle hire + fuel	24,650.00		30,500.00	55,150.00
University of New England					
Staff resources	All fixed term and casual staff including postdoc, research fellows and technical officers	174493		368332	542,825.00
Equipment	mechanical and cross pollination technologies / equipment to facilitate field insect and plant collection/ field trial equipment	35,361			35,361.00
lab work	analyses and processing of samples	23,680			23,680.00
field work expenses	travel to field sites, vehicle hire/accommodation; nets, vials other consumables, chemical use	38,900			38,900.00

travel	conference presentations	3,520			3,520.00
University of Tasmania					
Staff	2 FTE Level B Step 6 @ $136,841ncluding oncosts; laboratory and Field Research Technician 0.75FTE@ $104,055; 900hrs casual labour @ $42.77/hr including oncosts. In kind: Fraction of staff time from TIA, SeedPurity, SPS, Hansen Orchards & Reid Fruits in planning and designing experiments	244,703.41		268,855.59	513,559.00
Operating costs	Engineering costs associated with modification of prototype pollen harvesting and delivery systems for perennial and annual systems ($30,000); Laboratory and office consumables ($24,594); field trial costs including crop management (31,500). In kind contribution from industry partners	55,030.11		80,263.89	135,294.00
Travel	Interstate travel: 8 return airfares Hobart to trial site and meeting locations in NSW and SA per year (@$1500 ea), 32 days accommodation including self catering (@$200/day); Intra state travel @ $130	15,843.83		9,059.07	24,902.90

	car hire/day for 25 days per year each for annual and perennial crops				
NSW DPI					
Staff resources	NSW DPI salaries (1 person 10% of time, 1 person 5% of time and another at 5% of time), and funds to cover casual workers and a part time technical officer	88,750		68,449	157,199.00
Field work expenses	Accommodation for staff, vehicle hire + fuel, compensation for fruit removal	5,000		10,000	15,000.00
Travel	Flights to field sites, conference/ workshop presentations	5,000		10,000	15,000.00
Operating	Coordination and delivery of industry meetings, industry publications, consumables for data collection at field sites and laboratory analyses of samples.	20,000		10,000	30,000.00
Hort Innovation					
Project consortium governance	Provide an account of completed activities undertaken through the project.	23,991.74	1,000.00	1,000.00	25,991.74
	Four Project Reference Committees held	119,958.75	5,000.00		124,958.75

	Monitoring and Evaluation Plan reported on, Benefit cost analysis undertaken	23,991.74	1,000.00		24,991.74
	Project management, Financial audit conducted, Report on project legacy arrangements	47,983.49	2,000.00		49,983.49
Total Costs		**4,318,514.00**	**30,000.00**	**4,531,480.00**	**8,879,994.00**

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