Hort Innovation

Final Report

Integrated Pest Management of redberry mite, *Acalitus essigi*, on blackberries

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Delivery partner:

Tasmanian Institute of Agriculture

Project code:

RB17000

Project:

Integrated Pest Management of redberry mite, *Acalitus essigi*, on blackberries RB17000

Disclaimer:

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Content

Summary

Commercial blackberry production in Australia is expanding rapidly. An impediment to this expansion is the reliable production of high-quality fruit. Redberry mites, *Acalitus essigi* are a minute plant-feeding mite in the superfamily Eriophyoidea that infests blackberries in many growing regions. Redberry mite (RBM) feeding results in the incomplete and/or uneven ripening of blackberry fruit. The damaged fruit is unmarketable with very high RBM populations resulting in significant crop losses.

This project's aims were to identify and assess the impact of IPM strategies including cultural, biological, and chemical controls to enable the sustainable control of RBM in commercial blackberry production systems in Australia. To further enable these goals, we also developed optimised RBM monitoring protocols to enable blackberry producers and agronomists with the ability to develop a more predictive approach to their management of RBM.

These goals were achieved with a series of grower interviews examining their management of RBM's, a national fruit survey to assess their distribution and prevalence, and IPM field trials conducted in commercial production sites in Victoria's Yarra Valley and Tasmania. These trials consisted of both predatory mite releases and a '*spray reduction trial*' with RBM and predatory mite populations monitored on both blackberry fruit and within winter buds.

The results of these activities indicated that crop losses are cultivar specific and tended to be most prevalent in later season cultivars. Cultural control strategies including wild blackberry removal, cultivar selection and crop hygiene were highlighted as achievable cultural IPM strategies that will limit RBM populations.

Several predatory mite species known to predate Eriophyid mites were assessed in field trials. These assessments confirmed *Typhlodromalus lailae and Typhlodromus occidentalis* to be ineffective in controlling RBM with no evidence of control or establishment observed. However, *Typhlodromus doreenae* was isolated on both ripe fruit and within winter buds indicating they do persist in the crop and may predate RBM. Indeed, at all bar one site, where RBM numbers were already low, RBM populations were consistently lower in rows where *T. doreenae* were released. Another species not commercially available, *Typhlodromus dossei*, was also commonly observed in fruit sourced from Victoria. Little is currently known about *T. dossei* except that its presence is commonly associated with high numbers of Eriophyid mites.

The outcomes of the '*spray reduction trial*' confirmed that the spray program currently used by many Australian producers, although successfully reducing RBM populations, also has severe impacts on predatory mite populations. Furthermore, the adoption of a 'softer' program reduces RBM populations without impacting on predator populations or fruit quality.

These outcomes were extended to industry using multiple formats including but limited to, online communications, workshops and articles in industry magazines.

We recommend the following be further investigated:

- Research into the development of IPM strategies targeting sucking bugs including mirids and pentatomids (i.e. green vegetable bugs) to enable a more holistic approach to blackberry IPM.
- Further research into the efficacy of *Typhlodromus doreenae* and *Typhlodromus dossei* to determine the ability to control RBM and other pestiferous Eriophyid species.
- Further research in the potential impact of poor pollination and climatic effects on blackberry fruit quality.

Keywords

Blackberry; Integrated Pest Management; Redberry mite; *Acalitus essigi*

Introduction

Commercial blackberry production in Australia is expanding rapidly. An impediment to this expansion is the reliable production of high-quality fruit. Redberry mites, *Acalitus essigi* are a minute plant-feeding mite in the superfamily Eriophyoidea (Lindquist, 1996) (Figure 1A). Redberry mite (RBM) infests commercial and uncultivated blackberries in many growing regions of the world. The feeding activity of RBM is presumed to cause "redberry disease" (De Lillo and Duso, 1996), which consists of incomplete and/or uneven ripening of blackberry fruit (Figure 1B). The affected drupelets remain hard and have a green or bright red colour. The damaged fruit is unmarketable. Very high populations result in significant crop losses.

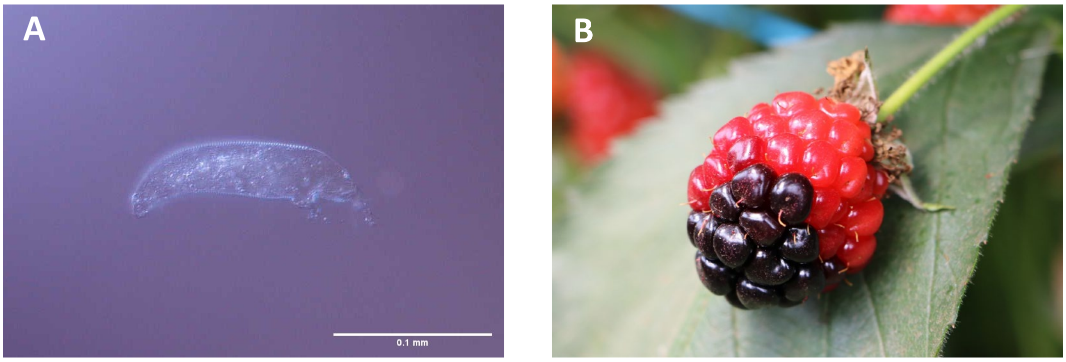

Figure 1. A) Redberry mite, *Acalitus essigi* isolated from commerial blackberry fruit; B) Blackberry displaying incomplete rippening caused by redberry mite infestation.

Aspects of the RBM lifecycle and seasonal population changes within wild blackberry plants were studied by Davies et al. (2001). Their limited motility and cryptic behaviours promote highly localised infestations and considerable seasonal population fluctuations. RBM overwinter in the leaf axils and under bud scales. In spring, mites move onto the developing shoots on the floricanes (fruit bearing two-year-old canes) and subsequently infest the developing fruit causing significant fruit damage. Later in autumn, populations decline, and mites move into overwintering sites.

Pesticides are currently the standard method of managing RBM, which should be deemed the last resort of controlling pests within Integrated Pest Management (IPM) programs. Rather than focusing solely on pesticide applications, IPM also utilises cultural and biological pest control methods and is relatively undeveloped in Australian blackberry production. Although chemical management has been demonstrated to successfully reduced RBM populations (Cross 2012), this ongoing reliance on pesticides is being increasingly deemed unsustainable by industry. Therefore, a greater emphasis on cultural and biological methods is required if sustainable pest management of Australian blackberries is to be achieved.

Cultural controls such as the selection of pest resistant or tolerant cultivars, crop hygiene and weed control are relatively simple strategies that undermine a pest's ability of establishing in a crop. Similarly, biological controls, primarily predatory mites, have been demonstrated to successfully control other eriophyid mites in several cropping systems (Sabelis 1996). However, no predatory species has been identified as predating RBM. Therefore, whether predatory mites are a viable alternative to chemical control of RBM in Australian blackberry production systems is currently unknown. Furthermore, a greater emphasis is needed regarding the selection of softer, predator friendly chemical applications if biological methods are to be employed.

In IPM, pest monitoring is seen as standard practice. If IPM of RBM populations can be achieved, a rapid method of monitoring RBM populations must be developed. Davies et al. (2002) developed a method to extract RBM from blackberries. However, this method requires prolonged processing times limiting its usefulness in IPM programs.

This project aims to develop a rapid method of Eriophyid mite extraction from blackberry fruit and winter buds in order to provide industry with a valuable tool for monitoring RBM populations as part of an IPM program. Furthermore, we identify and assess the impact of IPM strategies including cultural, biological and chemical controls with the aim of developing sustainable methods for RBM control in Australian commercial blackberry production systems.

Methodology

Initial Grower Survey

At the commencement of the project basic demographic information regarding blackberry in Australia was unknown. Therefore, a survey aimed to determine grower demographics, production system details, historical RBM damage and current IPM management understanding was conducted. A contact list of Australian berry producers growing commercial blackberry cultivars was developed. This was facilitated via an on-line registration process targeting the current RABA mailing list (450 contacts) and other mechanisms including presentations at the 2017 BerryQuest field day. The survey questions and participant information sheet are provided in Appendix 1 & 2, respectively.

National Fruit Survey

To facilitate the identification of cultivars susceptible to RBM and to identify sites for use in the IPM trails in years 2 & 3 of the project, the project team attempted to source fruit from all known commercial blackberry producers nationally. This survey yielded fruit from 19 growers across Tasmania, Victoria, New South Wales, and Queensland and comprised of 11 blackberry cultivars including seven patented Driscoll's cultivars. Mite extraction from fruit sourced from Tasmania was conducted at the University of Tasmania. Fruit sourced from mainland Australia was processed in quarantine facilities at the Department of Primary Industries, Parks, Water and Environment (DPIPWE) labs in New Town, Hobart. Fruit from wild blackberries was also collected from three commercial blackberry production sites and three roadsides in Tasmania to determine RBM and beneficial mite population sizes in wild blackberry. Fruit with disease symptoms was collected where possible to determine the relationship between damage incidence and severity and RBM population size.

Mite Extraction

The recommended method for mite extraction from blackberry involves placing the fruit on upturned adhesive tape (Figure 2), spraying the fruit lightly with a synthetic pyrethroid and waiting until the fruit is completely dried before quantifying the mite populations that had moved on to the tape (Davies et al., 2002). This process took ca. 4 weeks and did not extract all mites from the fruit and was therefore deemed unacceptable. Therefore, a rapid mite extraction protocol was developed. This new method required immersing and agitating the fruit in a vial of ethanol and quantifying the mites under a microscope. This method takes ca. 10 minutes per fruit rather than the 4 weeks required using the old method. This method was utilised for the remainder of the project.


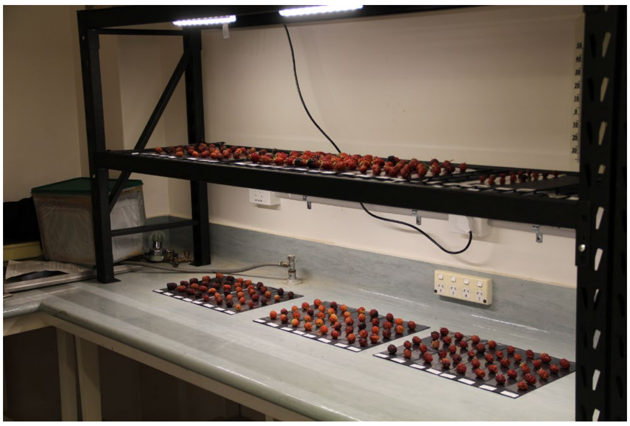

Figure 2. Blackberry fruit sourced from Tasmanian blackberry producers undergoing mite extraction at the Tasmania Institute of Agriculture's entomology laboratory

IPM field trials - 2018/2019

The 2018/19 trials utilised four sites: Blue Hills Berries and Fairview Hill (Yarra Valley, Victoria), Costa Group (Dunorlan, Tasmania) and Westaway Berry Farm (Westaway, Tasmania). The sites were chosen based on the results from the national fruit survey data previously described and consisted of RBM susceptible varieties 'Chester' (Blue Hills and Westerway) and 'BL454' (Costa Group and Fairview Hill) which possessed RBM populations. Both 'BL454' sites were under polytunnels and the 'Chester' locations were open field sites.

Predatory mite releases were conducted in consultation with consultant Dr Paul Horne (IPM Technologies), and industry representatives (Driscoll's Australia, YV Fresh and Costa Group) and biological control agent provider Biological Services (James Altman). Two species, *Typhlodromus occidentalis* and *Typhlodromalus lailae* were selected based on their known predation of Eriophyid mite species and were commercially available. Releases occurred at both Victorian locations on the 30th October and the 13th December 2018. Tasmanian releases were conducted at Westaway on the 29th November and 19th December and at Dunorlan on the 30th November and 21st December 2018. All biological control agents were supplied by Biological Services, Loxton, South Australia.

All participants were requested to utilise chemical controls that were less harmful on beneficial mite fauna. Two commonly used chemical controls with known impact on beneficial mites, the fungicide, Mancozeb and the acaricide, Abamectin were specifically requested not to be used after the release of biological control agents.

RBM and beneficial mite populations were monitored by randomly harvesting 40 fruit per row during the various fruiting stages (green, unripe red, and early season ripe and late season ripe fruit). During the 2019 winter, dormant buds (n = 20) were also collected from all locations to determine whether bud sampling could provide a predictive tool to forecast RBM population sizes on fruit. All fruit were assessed using the alcohol-based mite extraction protocol described previously with the buds first dissected to enhance mite extraction.

IPM field trials - 2019/2020

The production of the Driscoll's cultivar 'BL454' was removed from production nationally after the 2018/19 production season, which necessitated the sourcing of replacement field sites to enable continuation of the IPM field trials during the following season. This search included bud sampling of the new Driscoll's variety 'Victoria' at Costa Group's Tasmanian operations. A block of 'Victoria' blackberries containing moderate-high RBM populations (mean = 24.9 mites bud^{-1}) was identified and subsequently used in a '*pesticide reduction trial*'. Due to the constrained timeline, a block of the cultivar 'Victoria' containing RBM in Victoria's Yarra Valley could not be sourced as these plantings had already broken from dormancy preventing bud sampling prior to the commencement of the production season. However, an additional block of Chester in Silvan (Fruition by Nature, FBN) with a history of RBM infestation was secured and bud analyses confirmed the presence of a moderate RBM population size at this site.

Therefore, the 2019/20 field season consisted of the '*pesticide reduction trial*' conducted in one block of the cultivar 'Victoria' (Costa Group: Dunorlan, Tasmania) and the continuation of predatory mite trials this time involving the release of *Typhlodromus doreenae* and *Ty. lailae* in the Chester blocks located in Tasmania (Westerway Berry Farm) and Victoria (Blue Hills Berry Farm, Silvan) and at FBN. Mite releases and bud and fruit sampling were performed as previously described. All predators were again sourced from Biological Services, South Australia.

Pesticide reduction trial

A block of 'Victoria' blackberry (ca. 0.5 Ha) at Costa Group's property at Dunorlan was utilised for this trial. The aim of this trial was to determine whether fruit quality could be maintained whilst reducing the use of pesticide applications known to disrupt biological control agents. As part of this trial, the Costa Group agreed not to use the fungicide, mancozeb and the acaricide, abamectin after flowering commenced as these chemical controls are known to have deleterious effects on predatory mite populations. Costa Groups full spray program was utilised in the remaining half block to determine its impact on pestiferous and beneficial mite populations. Further assessments examined the ability of the predatory mites, *T. doreenae* and *Ty. lailae,* to reduce RBM populations and fruit damage. Predatory mite release timing and sampling protocols were as per the 2018/19 field season with fruit (n = 40) collected at intervals and buds (n = 20) collected the following winter. Samples were collected from rows that received the full and modified spray programs and rows that received both the modified spray program and the predatory mite releases. Predatory mite releases occurred on the 20th December 2019 and 16th January 2020.

Outputs

This project has actively sort to extend the outcomes of this project to industry stakeholders by utilising various formats. These have included:

Web Page

A Redberry mite page web page was produced and updated biannually. This page saw 393 unique visits throughout the life of the project and is being used as a repository for many of the extension materials developed.

E-News

The E-news to blackberry growers has included updates in a specific Redberry Mite News and also updates via the Tasmanian Institute of Agriculture's Berrylink newsletter to a wider audience.

RBM online project updates:

- RBM update Feb 2018: 60% open rate (33 recipients) - link
- RBM update Aug 2018: 55% open rate (41 recipients) - link
- RBM update Dec 2018: 51% open rate (45 recipients) - link
- RBM update July 2019: 64% open rate (55 recipients) - link
- RBM update Feb 2020: 60% open rate (85 recipients) - link
- RBM update Sept 2020: 50% open rate (86 recipients) - link

Berrylink articles:

- Berrylink May 2018: 49% of 188 - link
- Berrylink Aug 2018: 49% of 188 - link
- Berrylink Jan 2019: 36% of 233 - link
- Berrylink Mar 2020: 40% of 279 recipients link

Fact sheets

Three fact sheets produced and distributed annually

- Redberry Mite Update 2018 - link
- Redberry Mite Lifecycle and Monitoring - link
- Redberry Mite Update 2019 - link
- Redberry Mite Update 2020 - link
- Redberry symptom guide 'Is it redberry mite?' - link

Workshops

- **Launceston, Tasmania** - Nov 2018. 9 attendees – This workshop was also recorded and post on YouTube with a link also available on the project's webpage - link
- **Woori Yollock, Yarra Valley** - Nov 2018. 7 attendees

- **Western Junction, Tasmania** - Aug 2020. 18 attendees. The workshop presentation was posted on the Redberry Mite web page and distributed to growers - link

Industry Conferences

- Presenter at the BerryQuest field day (12^{th} – 15^{th} February 2018), which hosted > 380 growers, agronomists and industry representatives. Three presentations were given during the field day, which was attended by ca. 150 delegates. These interactions included an overview of the project and the recruitment of growers who had either not received or not responded to the earlier mail outs. Attendance at this event recruited another 3 growers and 2 agronomists.
- Display at the annual Fruit Growers Tasmania Conference 24^{th} – 25^{th} May 2019, which outlined the project and demonstrated mite extraction techniques to attendees

Industry Magazines

- **Australian Berry Grower** - Spring 2018: The secret life of redberry mite in blackberry (Appendix 3)
- **Australian Berry Grower** - Autumn 2019: Predators on the hunt for redberry mites (Appendix 4)
- **Australian Berry Magazine** - Summer 2019: The article gives a project update and discusses the interplay between predatory mite releases and chemical interventions for other pest species (Appendix 5).
- A final article discussing the project's outcomes is to be included in the Summer 2020 issue of the **Australian Berry Magazine**

News Media

- Burnie Advocate & The Examiner, Launceston, Tas. 7^{th} December 2017 "*TIA project studies blackberry colour*" (Appendix 6)
- The Examiner, Launceston, Tas. 28 February 2019 "*More Mites might be the key to help berries*" (Appendix 7)
- Tasmanian Country, 4^{th} September 2020 "*Doreen mighty on mite*" (Appendix 8)

Instructional Video

- Mite extraction demonstration video. The video is available on YouTube– link

Other Outputs

- Honours thesis and presentation. Honours student Hui Law completed Honours thesis on RBM has completed her thesis achieving First Class Honours. As part of her Honours year, Hui was required to present her research findings. Approximately, 60 people attended the event held on 12^{th} October 2018 at the University of Tasmania. The event was attended by a combination of academics and industry representatives including Costa Group employees and several members of the Ag Institute of Australia.

- Updates to the Berry Plant Protection Guide. The relevant updates (see Appendix 1) for the 2020 edition of the Berry Plant Protection Guide have been provided to Melinda Simpson (NSW DPI). The updates included mite monitoring, cultural, biological and chemical controls for pestiferous mites.

- Updates to growers and industry representatives regarding the outcomes of the fruit surveys have been delivered to each of the growers regarding the fruit samples they supplied to the project. Each grower received a table, which provides the percentage fruit that contained the various invertebrate fauna extracted from the fruit samples, the mean number per fruit and the maximum number quantified from a single blackberry. These communication channels have been maintained throughout the project.

- Literature review completed (Appendix 9)

Outcomes

The project's outcomes were aligned with the Raspberry and Blackberry Strategic Plan's Outcome 3 - increasing farm productivity - by aiming to reduce the input costs associated with the production of commercially grown blackberries in Australian production systems. To achieve this goal the project sort to:

1. Determine the extent (relative levels and distribution) of RBM infestation in Australia and its impact on blackberry production in Australia.

Australian blackberry producers and agronomists working with blackberry producers were interviewed to gain insights to current RBM management strategies employed by industry. Of those interviewed all bar one grower reported observing RBM damage. Of those that did report observing RBM damage, crop losses ranged from 0 to 60%. Participants indicated the following factors influenced RBM damage severity, in order of importance:

1. **Cultivar:** crop loss was variety specific and tended to be most prevalent in later season cultivars. Driscoll's variety 'BL454' and the open-source variety 'Chester' were believed to be most susceptible. Five cultivars (Elvira, Victoria, Karaka, Silvan and Murrindindi) were reported as being symptom free with two of these being relatively new cultivars.
2. **Management:** With appropriate management in place including improved crop hygiene and frequent pesticide applications, crop losses could be reduced to low (1-5%) levels.
3. **Age of the crop:** Although RBM symptoms could appear in a young crop, the level of crop loss increased with crop age.
4. **Season:** RBM symptoms were worse in some years than others although no specifics about the characteristics of the season were provided. 2016/17 was considered a bad season whilst the incidence was reduced in 2017/18 potentially due to higher chemical intervention.

To determine the true extent of RBM in Australian production systems, a national fruit survey was also conducted. This process yielded fruit from 14 commercial blackberry growers located in Tasmania, Victoria, NSW, and Southern Queensland. The results confirmed the findings of the grower interviews with the Driscoll's variety 'BL454', and the open-source variety 'Chester' containing elevated RBM populations (Figure 3). Similarly, RBM population sizes were found to vary greatly between locations, for example RBM populations extracted from ripe 'BL454' fruit harvested at site BB contained an average of 53 RBM fruit^{-1} (max = 534 RBM fruit^{-1}) and Chester fruit harvested from neighbouring property (site HC) contained average of 48 RBM fruit^{-1} (max = 245 RBM fruit^{-1}), whereas fruit from other locations yielded little-to-no RBM from these cultivars. Where high levels of RBM were not observed in these varieties the growers acknowledged that significant effort had been given to reduce RBM populations due to significant damage having occurred in previous years.

Generally, early season cultivars such as Karaka were shown to contain fewer RBM and exhibited less damage compared to most mid (i.e. Cowles) and late season (i.e. BL454, Chester, wild types) floricane cultivars. However, as observed in previous studies primocane fruiting cultivars such as Elvira contained few RBM due to the differing canopy management techniques used in these plantings (Murrietta and Gaskell 2016). Despite repeated sampling, no RBM were observed from any fruit sourced from producers in either Queensland or Northern NSW (Corindi Beach) indicating these sub-tropical regions may not sustain *Acalitus essigi* populations.

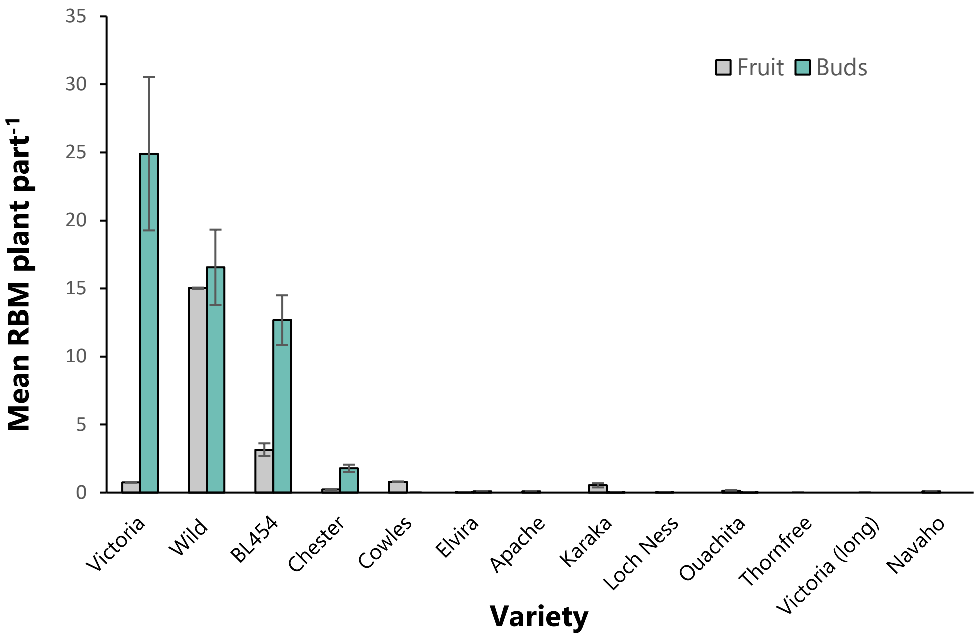

Figure 3. Mean (± SEM) redberry mite, *Acalitus essigi* extracted from ripe fruit and winter bud samples collected from 12 commercially grown blackberry cultivars and wild blackberry plantings sourced from various locations around Australia. The cultivar 'Victoria' was examined as both existing plantings and as newly established 'long cane' plantings (Victoria (long)).

During the winter of 2019 RBM was isolated within winter buds of 3-year-old plantings in the newly introduced Driscoll's cultivar 'Victoria" in Tasmania. However, despite their isolation in these plantings, no fruit damage had been observed in previous seasons. It is currently unclear whether 'Victoria' blackberry is susceptible to RBM as any fruit expressing RBM-like damage contained few, if any mites. Indeed, in January 2020, Quarrell was requested by industry stakeholders to examine a suspected severe outbreak of RBM in a block of tunnel grown 'Victoria' blackberry in the Yarra Valley. The block in question was estimated to be experiencing yield losses in excess of 60%. In some instances, the observed damage superficially appeared like that caused by RBM (Figure 4A). However, the unripe drupelets were soft, not hard, as is the case with RBM damage (redberry disease) with the red drupelets also showing signs that ripening was occurring all be it at a delayed rate. In more extreme instances, the fruit exhibited uneven fruit ripening and swollen individual drupelets (Figure 4B). As these symptoms are not characteristics of RBM damage it was believed to be caused by other factors. To confirm this hypothesis, green, unripe red and both damaged and undamaged ripe fruit samples were taken as per the project sampling protocols from the block in question and from another block of 'Victoria' blackberry from a different property under the same management that was not experiencing damage.

In the fruit samples taken from the site where damage was observed, low RBM populations (mean = 2.00, range = 0 – 12) were observed. Further, in the damaged ripe fruit assessed, only 8% of the ripe fruit contained RBM (n = 2), which would again lead to the conclusion that the observed damage is due to other factors.

It therefore currently believed that the underlying cause for this damage may be physiological in response to adverse climatic conditions, as this crop was experiencing extreme heat during this period, which was being managed with the use of overhead misters to cool the fruit which in turn generated a relative humidity in the crop exceeding 95%. This block was the only block in the area known to be using overhead misters to cool the fruit. Further resources may be required to help industry determine whether extreme climatic events can cause uneven fruit ripening in 'Victoria' blackberry as observed at this location.

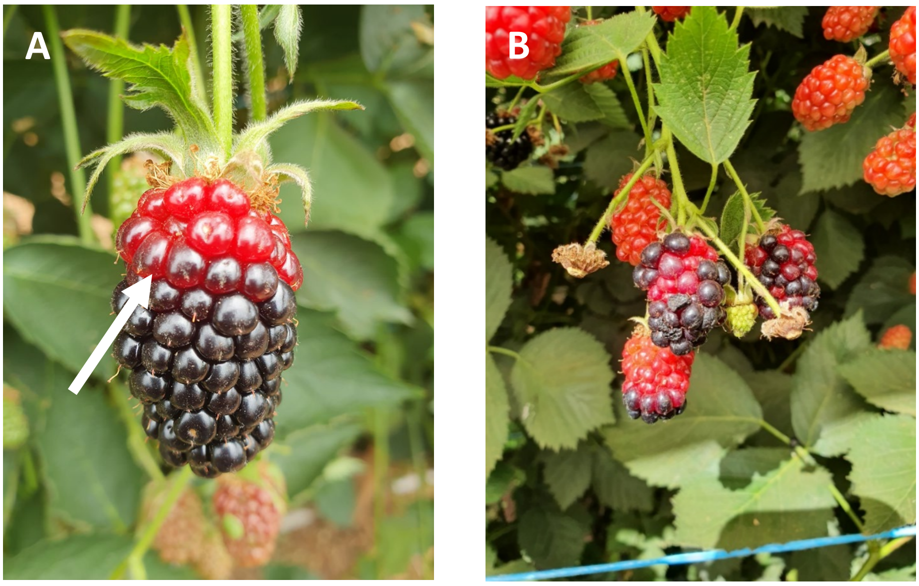

Figure 4. Examples of 'Victoria' blackberries exhibiting RBM-like damage during the 2019-20 production season. No RBM were extracted from these fruits. A) Blackberry exhibiting uneven fruit ripening at Costa Group's Bengeo Rd property at Dunorlan, Tasmania. The drupelets on these fruits were soft, not hard, which is one of the key characteristics of RBM damage. Furthermore, rather than a clear boundary between the red and black drupelets, the drupelets on the boundary are slowly ripening but by the time they do so the drupelets at the fruit's tip is overripe and the fruit is no longer marketable. Arrow indicates drupelets exhibiting delayed ripening B) blackberry drupelets displaying signs of what is indicative of poor pollination, leading to the appearance of severe RBM damage.

2. Identify Integrated Pest Management strategies that reduce the impact of RBM infestation on blackberry fruit production for the fresh market.

Numerous cultural, biological, and chemical strategies coupled with an increased monitoring effort were identified as strategies that will minimise the impact of RBM on Australian blackberries. These include:

Cultural controls

- improved farm hygiene including the removal of wild blackberries on-farm and the ongoing removal of canes and leaf litter after pruning
- the selection of cultivars less susceptible to RBM

Biological controls

- At the commencement of the project no predatory mites where known to predate RBM. Field trials were conducted into the release of predators currently commercially available in Australia (see below for predatory mite trial outcomes). Taxonomic studies were also used to identify any other predatory species to determine whether any yet to be identified species may provide RBM control.

Chemical controls

- Minimising the use of chemicals known to impact on predatory mites i.e. Mancozeb and Abamectin
- Improved chemical spray efficacy by applying sprays before RBM seek shelter in the fruit's complex structures

Monitoring

- Development of producer friendly monitoring protocols for RBM quantification in both blackberry fruit and buds
- Implementation of winter bud monitoring to understand background RBM population sizes prior to the commencement of the production season

3. Provide growers with better knowledge and understanding of management practices that impact on RBM infestation. Thereby ensuring growers are confident to implement an integrated pest management program that reduces the impact of RBM on blackberry fruit production for the fresh market

As previously described, wild blackberry removal, cultivar selection and crop hygiene were highlighted as achievable cultural IPM strategies that will limit RBM populations in Australian production systems. These measures have been extended to industry using various outputs (see Outputs). Indeed, 80% of growers surveyed at the end of the project have either commenced wild blackberry removal or intend to do so in the short term based on the findings of this project. Similarly, the removal of RBM sensitive varieties from production has already commenced with the Driscoll's variety 'BL454' no longer in production and the open-source variety 'Chester' also being phased out at several locations.

Similarly, 60% of growers interviewed at the project's end indicated they have commenced RBM monitoring of both fruit and winter buds. Winter bud monitoring provides producers with a better understanding of RBM populations in their crops enabling a more flexible IPM approach to RBM control.

Several predatory mite species were assessed for their ability to moderate RBM in field trials. Of these assessments *Ty. lailae and T. occidentalis* confirmed these species to be ineffective in controlling RBM with no evidence of control or establishment observed. Indeed, no *Ty. lailae* or *T. occidentalis* were extracted from any fruit collected at any of the trial sites during the 2018/19 season. These findings also correlate with the results from the 2019/20 season with larger RBM populations and smaller Phytoseiid populations observed in the *Ty. lailae* release rows. Whether this species failed to establish or did establish but resided on other plant parts is unclear.

However, trials during the 2019/20 field season indicated lower RBM numbers in rows where *Typhlodromus doreenae* were released at all sites except Westerway, Tasmania, where RBM populations were low in all treatments. Indeed, *T. doreenae* was isolated on both ripe fruit and in winter buds indicating they do persist in the crop and may predate RBM (Figures 5 & 6; Table 1). Further research is required to better understand the behaviour of *T. doreenae* in commercially grown blackberries. Another species not commercially available, *Typhlodromus dossei*, was also commonly observed in fruit sourced from the Yarra Valley. Little is currently known about *T. dossei* except that its presence is commonly associated with high numbers of Eriophyid mites (James et al. 1995), the family to which RBM belongs.

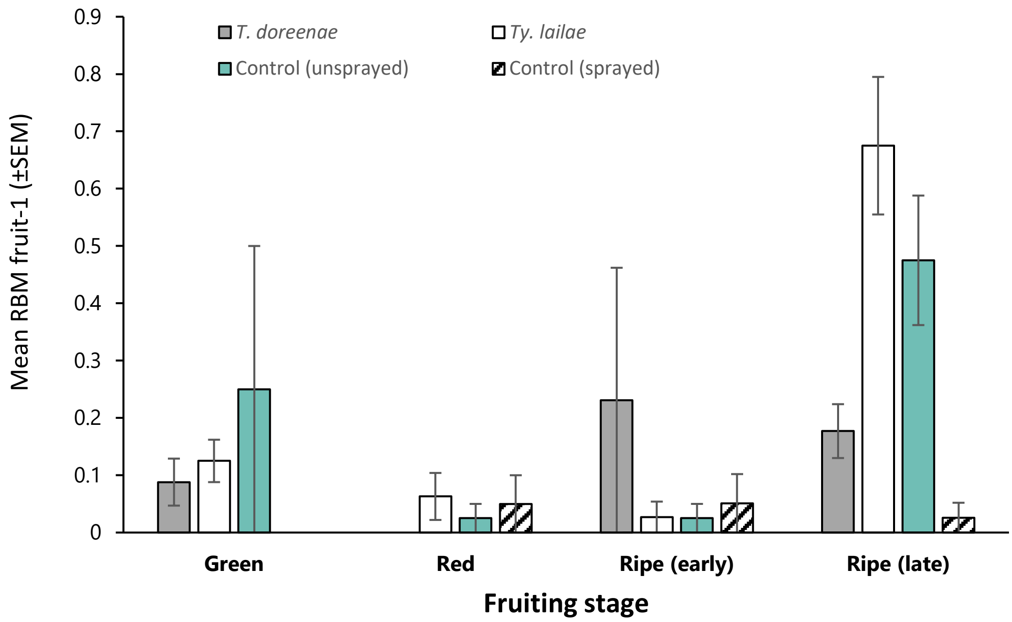

Figure 5. Mean (± SEM) redberry mite *Acalitus essigi* fruit^{-1} extracted from either green, unripe red, early season or late season ripe 'Victoria' blackberry fruit harvested from Costa Group's Dunorlan property in Tasmania during the 2019/20 IPM field trial.

The deleterious impacts of current chemical management practices have also been extended to industry using various formats. Indeed, the off-target impacts of current chemical management practices on beneficial species was further demonstrated during the spray reduction trial conducted at Costa Group's property at Dunorlan with lower numbers of beneficial mites extracted from fruit (Figure 6) and no predatory mites observed in the winter buds (Table 1) within the rows where the fungicide, Mancozeb and the acaricide, Abamectin were applied. It should also be noted that although the full spray program did reduce RBM numbers, it also reduced predatory mite populations.

Table 1. Mean invertebrates extracted from 'Victoria' blackberry winter buds (n = 20) collected in September 2020 from the 'Pesticide Reduction Trail' conducted at the Costa Group's Dunorlan property in Tasmania.

Treatment	RBM	Phytoseiidae	Acaridae	Broad mites	Spider mites	Thrips
Full spray program	4.65	0	0	0	0	0
Modified spray program	9.45	0	0	0	0	0
Typhlodromalus lailae	15.45	0	0	0	0	0
Typhlodromus doreenae	6.80	0.25	0	0	0.05	0

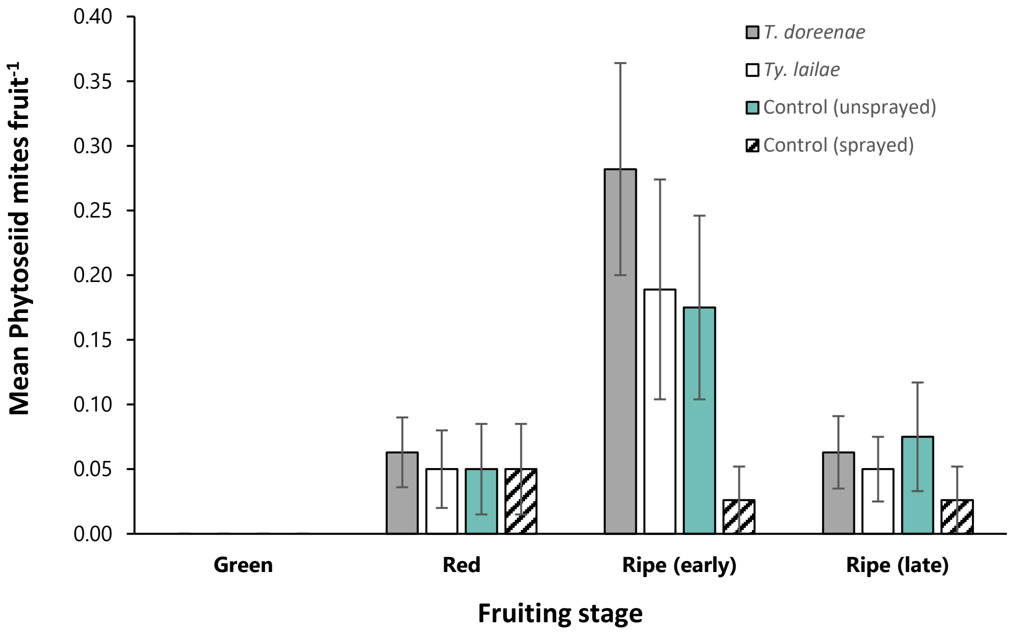

Figure 6. Mean (± SEM) predatory Phytoseiid mites fruit^{-1} extracted from either green, unripe red, early season or late season ripe 'Victoria' blackberry fruit harvested from Costa Group's Dunorlan property in Tasmania during the 2019/20 IPM field trial.

Although, producers have commenced limiting their use of products that are toxic to beneficial species, a more holistic approach to IPM in blackberry is currently being hampered by the impact of sucking bugs (Figure 7), which are only able to be controlled with the use of pyrethrum that is also toxic to predatory mite species (Castagnoli et al 2002). For this reason, the uptake of biological controls such as *T. doreenae* to control RBM are being considered; however, their uptake may be limited until IPM strategies to control sucking bugs are found.

Figure 7. Blackberry fruit damage observed at Costa Groups blackberry farm at Dunorlan, Tasmania, with collapsed unripened drupelets most likely from sucking insects i.e. mirids or pentatomids. The chemical management of these pests is limiting IPM uptake in commercially grown Australian *Rubus*.

Medium term outcomes (3 -7 years)

Increased implementation of IPM strategies that reduce the impact of RBM by Australian blackberry growers & increased packout of high-quality blackberry fruit by Australian blackberry producers

The findings of this project are already leading to the increased implementation of IPM in Australian blackberry production systems. Furthermore, with the removal of wild blackberries on-farm, the adoption of softer chemical management strategies, increased RBM monitoring and the movement away from the recognised RBM susceptible varieties including 'BL454' and 'Chester' the increased packouts are being already achieved. However, industry has indicated that it remains limited in its ability to take a more holistic approach to implementing IPM due to ongoing issues with sucking bugs i.e. mirids and pentatomids such a green vegetable bug (see above).

Monitoring and evaluation

Effectiveness:

Were useful IPM strategies developed for RBM including monitoring and management strategies?

Monitoring

A new technique for monitoring RBM in both winter buds and fruit. The research established how many buds or fruit were necessary to determine infestation of RBM in blackberries.

Management strategies

The following management strategies were developed for RBM in blackberries

- Removal of wild blackberries and general crop hygiene
- Monitoring protocols for presence of RBM in fruit and buds
- Identification of tolerant and sensitive varieties grown in Australia
- A predator friendly pesticide program
- Use of a new biological predator for RBM

Did the project establish the extent of RBM infestation in Australian blackberries and the impact on blackberry production?

A survey of blackberry producers and blackberry fruit in Australia established that RBM is currently only present in commercial blackberry crops grown in Tasmania and Victoria with none detected in fruit from NSW or Qld. Fruit from WA or SA was not available due to limited production in these states. The project established which commercial varieties were sensitive to RBM. The impact of RBM on blackberry production was evaluated by a survey of growers at the beginning of the research that indicated losses of between 30-60%. The indirect impact of RBM was the use of a spray program toxic to predatory mites and insects. This was practiced by a large proportion of growers of proprietary varieties in Australia.

Process Appropriateness / Relevance

The key stakeholders of this project were commercial blackberry growers and agronomists. The project was able to target the most relevant stakeholders through an initial industry survey, networking and on-ground knowledge of industry participants. Key to this was liaising with the industry association (RABA), major corporate producers (Costa, YV Fresh and Driscolls) and regional agronomists.

The engagement and communication methods included phone survey, workshops, farm visits, e-news, conference presentations, national berry journal, web page, YouTube and print media.

In a post-project survey, growers indicated which form of engagement they found most useful to their business (Table 2).

Table 2. Extension material format efficacy derived from post-project MER survey

Extension material format	Used this resource	Usefulness score
E-newsletter	70	4.2
Web page	50	2.5
Fact sheet	90	4
YouTube	10	3
Workshop	80	4.5

Comments received by growers highlighted the value they gained from workshops.

- "*I got the most out of the workshops*"
- "*Workshops are best, the practical aspect makes it the best way to learn and the ability to interact with others*"

Participation in the 3 workshops

Of the 19 growers first identified in the phone survey, the following numbers attended workshops

- Workshop 1: Nov 2018 – Tas: 9 attendees. This workshop is available on YouTube (see Outputs).
- Workshop 2: Nov 2018 – Vic (Woori Yallock): 7 attendees
- Workshop 3: Sept 2020 - Tas Driscoll's workshop: 18 attendees

The lower score for the website could be due to most communication around the project directing stakeholders to items hosted by the webpage rather than the actual pages (e.g. factsheets, resources, YouTube). The analytics from the page recorded 393 views as of June 2020.

Impact of the project

The change to grower's aspirations, knowledge and skills was monitored through direct feedback during the project and at the completion of the project via a phone survey of a cross section of the industry, primarily growers (60%) and in-house agronomists (40%).

Knowledge and skills

Eighty percent of growers/agronomists interviewed demonstrated knowledge of 3 or more useful strategies for managing RBM in blackberries. The most frequently identified strategy was removal of wild blackberries followed by a winter spray program. Other strategies also identified included crop hygiene, crop monitoring, use of predators, use of tolerant varieties and applying a suitable pesticide program.

Growers were then asked about their current practice change for the following key elements of a RBM management program:

- **Symptom recognition:** Of growers/agronomists surveyed, 70% had greatly increased their confidence and skill in recognising RBM symptoms. The growers whose level of confidence had not changed, was attributed to their skill at the start of the program (2 growers) or had only recently entered the industry (1 grower).
- **Removal of wild blackberries:** Eighty percent of growers interviewed were currently removing or had removed wild blackberries from their farm (Confident). There were issues still around removal of neighbouring wild blackberries. Those who were not currently doing this did not have a problem with RBM (NSW grower and one with no wild blackberries)
- **Use of tolerant cultivars:** Of those interviewed, 70% had changed cultivars (or recommended changing cultivars) to move away from RBM sensitive cultivars such as 'BL454' or 'Chester'.
- **Monitoring:** Of the respondents, 60% are now actively monitoring crops for RBM. Some growers monitor both winter buds and fruit whilst others monitor fruit and identify blocks with symptoms for targeted management.
- **Use of predators:** Growers were less confident about the introduction of specific predatory mites for RBM management. Thirty percent of growers are trialling specific predators for RBM, 50% would like to do so in the future but required more evidence. Thirty percent of growers indicated that mirids and green vegetable bugs limited their introduction of predators due to the need for using broad spectrum chemistry for these pests.
- **Applying an RBM predator friendly pesticide program:** Seventy percent of respondents were avoiding the use of specific pesticides known to be toxic to predators (acaricides/mancozeb) after flowering in blackberries, although this was currently limited to trial areas for 30% of respondents; 30% also indicated that they occasionally need to apply these pesticides due to other pest pressures (Ridomil Gold for downy mildew) or when other mite populations cannot be managed. Fifty percent of growers also indicated that other pests, specifically mirids and green vegetable bug required the use of broad-spectrum pesticides that were not predator friendly.

The greatest impediment to growers implementing a full IPM program for RBM was the threat from sucking bugs (i.e. mirids or pentatomids), which necessitated the use of a spray program unfriendly predator

Efficiency

The project was able to deliver on time and within budget. Resource efficiency was possible through engaging an honours student; engaging with local agronomists; event coordination with industry and the in-kind provision of biological control agents by James Altman, Biological Services.

Sustainability

The long-term impacts of this project are already apparent with a shift in the variety profile grown in Australia underway. The increased knowledge and skill within the industry also bodes well for the impact of this project to be sustained in the longer term. However, a serious impediment to the long-term sustainability of the benefits of this project is that it only addresses one pest. The impact of the more serious pests, the sucking bugs including mirids and green vegetable bugs, threatens the sustainability of this research. Seventy percent of growers indicated that the sucking bugs expose their crops to higher risk than RBM and their IPM is compromised by this. Some growers also indicated that the RBM program needed to be extended to provide them with more evidence and confidence with using predators for RBM.

Recommendations

We recommend the following be further investigated:

- Research into the development of IPM strategies target sucking bugs including mirids and pentatomids (i.e. green vegetable bugs) to enable a more holistic approach to blackberry IPM.
- Further research into the efficacy of *Typhlodromus doreenae* and *Typhlodromus dossei* to determine the ability to control RBM and other pestiferous Eriophyid species.
- Further research in the potential impact of poor pollination and climatic effects on blackberry fruit quality.

Refereed scientific publications

Journal article

Law, H.M., Allen, G.R., Davies, J.T. Corkrey, R. Buntain, M., Quarrell, S.R. 2020. Sampling, extraction and incidence of redberry mites (*Acalitus essigi*) on blackberries in Australia. *Experimental and Applied Acarology* **81**, 317-334 DOI: 10.1007/s10493-020-00509-y

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Intellectual property, commercialisation and confidentiality

No project IP, project outputs, commercialisation or confidentiality issues to report

Acknowledgements

We would like to acknowledge the following blackberry producers and agronomists for their time and the provision of site access to enable the field experiments to take place. Without your assistance we would not have developed the understanding of the impact RBM and other pestiferous species is having on Australian blackberry production:

- **Cost Group's** Nandita Sharma, Cameron Folder, Rohan Kile, and Alejandro Haro
- **Driscoll's Australia's** Jason Barnes, Sara Nour and Sally Kershaw
- Cindy Edwards, **YV Fresh**
- Tom Clarke, **Westerway Berry Farms**
- Kevin Chapman, **Blue Hills Berry Farm**
- Dean Chapman, **Fairview Hills Berry Farm**
- And the other industry stakeholders who provided fruit for the national fruit survey

We also wish to than James Altman from **Biological Services** for the provision of the predatory mites.

The project lead would also like to thank the project's team in particular **Michele Buntain** for her help with the extension, grower interviews and the MER components of the project. **Hui Law** for her hours spent at the microscope processing the seemingly endless number of fruit samples. **Jamie Davies**, DPIPWE for his mite identification and co-ordination of the import paperwork that enabled the processing of mainland fruit samples in Tasmania.

Appendices

Appendix 1

Redberry mite industry survey - 2018

The first part of this interview is about finding out a little about you and your production to provide some context. Then I will have some questions about symptoms of redberry mite you may have observed in your blackberries. To understand how the style of production affects redberry mite, I have some questions around how you grow and manage your blackberries.

If at any point you are uncomfortable or unsure about answering any of the questions or feel they may disclose confidential information, please say. You can just say that you would prefer not to answer the question or can't answer the question. That is perfectly OK.

A: Grower information/demographic

- Location – Where is your blackberry production?
- How long as a berry grower/blackberry grower
- What area of blackberries do you grow?
- Are you planning to increase blackberry production?
- Do you make decisions about crop protection or use an agronomist advisor?
- Are you familiar with redberry mite and the symptoms it causes in blackberries?

B: RBM observations:

- Have you observed symptoms that you think are due to RBM?
- Do you see RBM symptoms every year?
- Are symptoms across/over all varieties,
- Are symptoms spread evenly through crop, evenly throughout site/blocks/ tunnel?
- When (during the season) do you most often see symptoms? At what stage of the crop is this? At what time of year? – early, mid, late season?
- Do you see symptoms evenly along cane?
- Do you have any historical measures/estimates of crop loss due to RBM?
- What do you do with reject fruit?

C: Production system:

- Covered, outdoor, in-ground, substrate, long cane (artificial chill – if so where are these sourced)
- Training/trellis system
- Age of plantings
- Source of plants – tissue culture/tip cuttings in substrate or from soil grown nursery plants
- What time of year do you produce fruit
- What varieties do you grow? Primocane/floricane
- How do you manage your primocanes (when thinned / pruned/trained)?

D: Crop protection practices:

- What are your top 3 pest/disease (rank 1-3) issues in blackberries?
- How would you rate your crop protection across a scale of chemical through to biological?
- If you use IPM, how well established is your IPM?
- Do you believe any RBM IPM practices work better than others?

Spray program

Are you happy to share your spray diary? (confidential – not identifiable). If not can you indicate number / timing of

- Insecticides
- Fungicides
- Biologicals?

Weeds/interrow:

What type of cultural management / Inter-row management do you use? Crop/block hygiene?

Weed management? In crop/interrow/around crop?

Do you know of any wild blackberries close to your production? How close would these be to your blackberries?

Finishing up

Is there anything you think that may be relevant to redberry mite that we haven't talked about?

Appendix 2

Phone survey for the project 'Integrated pest management of redberry mite on blackberries'

Dear <Enter Name>,

Invitation

My name is Michele Buntain and I work at the Tasmanian Institute of Agriculture (TIA). I am currently working with Dr Stephen Quarrell (TIA) and Jonathan Eccles (Raspberries and Blackberries Australia) on the project 'Integrated pest management of redberry mite on blackberries'. As a provider of fruit from the national mite infestation survey, Jonathan Eccles and I would like to conduct a short phone interview with you or your farm manager to discuss redberry mite in blackberries on your farm.

What is the purpose of this study?

The phone interview is to help piece together historical information on redberry mite and link this with our fruit survey. This will help our research team understand which factors might be contributing to redberry mite infestation and to then develop the best possible field trials for the integrated pest management of redberry mite.

Why have I been invited to participate?

You are invited to participate in this interview survey because you have registered with RABA (Raspberries and Blackberries Australia) to take part in this project.

You are not obliged in any way to participate in the interviews which are completely voluntary. If you **do not** wish to participate, please indicate this by return email to the project leader, Dr Stephen Quarrell, Stephen.Quarrell@utas.edu.au or by phone on (03) 6226 6221

What will I be asked to do?

You will be asked to participate in a phone interview during March/April 2018, and again just prior to completion of the project in 2020. The interview will consist of approximately 25 questions relating to your production, your observations of redberry mite symptoms, your production system and crop protection practices. This should take around 40 minutes

You are not be obliged in any way to provide information or answer questions if you are not comfortable to do so, particularly if you believe this is sensitive or commercial in confidence information. You will be given the opportunity to review the completed transcript of the interview and correct or edit any parts of this.

Are there any possible benefits from participation in this study?

Your participation in this study will help the redberry mite research team find solutions that can be implemented on farms affected by redberry mite. It will guide the research trials undertaken by the redberry mite team over the next 2 years. This will help you as a blackberry grower manage pests including redberry mite.

Are there any possible risks from participation in this study?

The risks involved from participating in the interviews are low and protocols are in place to protect your privacy, including the use of summarised data for reporting that does not identify individuals or operations, security of information storage and a coding system to de-identify protected cultivars.

What if I change my mind during or after the study?

You can withdraw from this study without any explanation. This can be done by phoning or emailing a research team member. We will also ask if you still wish to participate when we contact you to conduct the phone interview. We will also send a reminder of your option to withdraw with the transcript in early May 2018. The option to withdraw will continue until 30th June 2018.

What will happen to the information when this study is over?

While the study is underway electronic data will be stored on the chief investigator's (Dr Stephen Quarrell) secure University of Tasmania electronic storage drive and any other material such as notes taken during the phone interview will be held in a lockable filing cabinet in the investigator's office. After the study is completed, all data and material will be transferred to the care of the chief investigator who will secure it until the eventual deletion date. Researchers who will have access to the data include the interviewers (Michele Buntain and Jonathan Eccles), research team members Dr Stephen Quarrell, Dr Geoff Allen and honours student Hui Law for the purposes of discussion and reporting to the funding body. Data will be de-identified 6 months after the completion of the project and will be destroyed after 5 years from the date of first publication. Data will not be archived, and will be treated in a confidential manner.

How will the results of the study be published?

The transcript of your interview will be read by the research team and only the summarised content used in project reporting to Hort Innovation. The summarised results may be presented at conferences, technical field days, in promotion to government and by other project partners and for scientific publication. A coding system has been arranged with Driscolls to protect the identity of individual cultivars so that these are not identifiable in external reporting.

What if I have questions about this study?

If you have any questions about this survey please contact Michele Buntain or a member of the research team listed below

Research team members:

Chief Investigator: Dr Stephen Quarrell | stephen.quarrell@utas.edu.au| 03 62266221
Michele Buntain | michele.buntain@utas.edu.au | 03 6226 6353 / 0429 957 975
Jonathan Eccles | jonathan.eccles@raba.com.au | 0407 242 757
Dr Geoff Allen | geoff.allen@utas.edu.au | 03 62262732
Hui Law | huileng.law@utas.edu.au

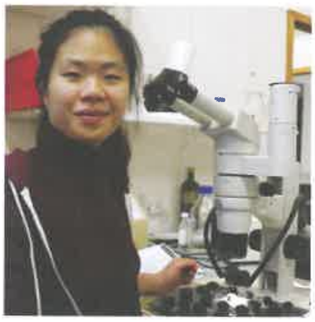
This study has been approved by the Tasmanian Social Sciences Human Research Ethics Committee. If you have concerns or complaints about the conduct of this study, please contact the Executive Officer of the HREC (Tasmania) Network on (03) 6226 7479 or email human.ethics@utas.edu.au. The Executive Officer is the person nominated to receive complaints from research participants. Please quote ethics reference number H0017182

This information sheet is for you to keep. Thank you for taking the time to read this information.

Appendix 3

The secret life of redberry mite in blackberries

Dr Stephen Quarrell and Michele Buntain, TIA
stephen.quarrell@utas.edu.au



Redberry mites can devastate blackberry crops, causing irreversible damage to fruit and up to 60 percent crop loss. The microscopic mites colonise the tiny spaces in the developing fruit and cause uneven ripening. Individual drupelets (often nearest the stem end of the fruit) remain hard, red and fail to ripen. The rest of the berry turns black, but the fruit is unsaleable and a potential disease risk if left unpicked.

This tricky pest is being tackled by a team from the Tasmanian Institute of Agriculture (TIA), Tasmanian Department of Primary Industries, IPM Technologies, and Raspberries and Blackberries Australia (RABA).

The team is working to develop integrated pest management (IPM) strategies to improve both monitoring and management of redberry mite in blackberries.

TIA entomologist Dr Steve Quarrell said one of the less endearing features of redberry mites is that they are small - *"bloody small"* - and they like to hide.

"Think half the size of a two-spotted mite that likes to play catch-me-if-you-can. They spend most of their life tucked away inside buds, under leaf scales and then under the fruit stem and between the actual fruit drupelets."

"It's their star survival strategy, which makes them so hard to manage," he said.

To make monitoring easier, TIA honours student Hui Law has developed a faster and more efficient method of keeping track of redberry mite populations.

The traditional method involves placing buds or fruit onto sticky strips, waiting around four weeks for the fruit to dry, and progressively catching the mites on the sticky strips as they crawl away.

"This is messy and slow so it really isn't a practical solution for IPM consultants or growers to use," Hui said.

Hui's new extraction method involves rinsing the buds or fruit in an ethanol solution for a few minutes.

She then pours this over black filter paper to collect mites and small insects that were hidden within the fruit.

The creamy white coloured redberry mites stand out on the black filter paper which makes counting them much easier and faster.

"We still need a reasonable level of magnification, around 15 to 20 times for counting, so slightly higher than a typical hand lens a grower would use," she said, *"but this method will make monitoring on farm a much more practical option"*.

KATIE MULDER (HILLWOOD BERRIES AGRONOMIST) AND STEPHEN QUARRELL (TIA)

HUI LAW (UTAS / TIA HONOURS STUDENT)

Dr Quarrell said an efficient counting method is the first step to understanding and then managing redberry mites. *"If we can monitor where and when the redberry mite population is on the plant, then we can better target our IPM strategies,"* he said.

Last season, Hui extracted and counted mites from over 1400 blackberry fruits from eleven different cultivars at eleven different commercial sites in the major blackberry growing regions of Australia. The count also included samples from wild blackberries, both on and off farm.

Mite numbers tell only part of the story. Growers have helped the research team fill in the bigger picture with historical information about their crops and how they manage them.

Dr Quarrell said this has really helped identify some hot spots for redberry mites, and management practices that could be improved.

"Wild blackberries were a real eye-opener for us. The mite counts from these were sky high compared to the numbers in the commercial blackberry crops."

"The danger is that nearly all the growers we interviewed said they have wild blackberries near their commercial crop, even as close as five metres. So managing these redberry mite populations could have quite an impact on preventing re-infestation of crops," he said.

Redberry mites show a definite preference for particular varieties over others, with the standouts being 'Chester' and one proprietary variety.

The research team is not sure if this preference is due to the lifecycle of the mites synchronising with the later fruiting varieties, or a particular characteristic of the plant that they find attractive. New late season floricane varieties becoming available will be ones to watch carefully in the coming season. Trials in 2018/19 will target the susceptible varieties, looking at the development of softer pesticide programs and the introduction of predatory mites as potential management tools. Whilst many crops had low or no redberry mites present this season, the intervention programs used were quite intensive due to a previous history of redberry mite infestation. It will be a positive step forward if the research program can help growers reliably produce high quality blackberry fruit with strategies that are more sustainable and friendlier to people and the environment.

The project is funded by Hort Innovation using the raspberry and blackberry research and development levy and contributions from the Australian Government, supported with in-kind contributions from TIA. TIA is a joint venture between the Tasmanian Government and the University of Tasmania. Hort Innovation is the grower owned, not-for-profit research and development corporation for Australian horticulture.

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Appendix 4

Predators on the hunt for redberry mites

Dr Stephen Quarrell,
Tasmanian Institute of Agriculture

MONITORING POST FOR MITE ACTIVITY

DR STEVE QUARRELL LOOKING FOR MITE ACTIVITY

The fruit damaging pest, redberry mite, is the target for some specially selected predatory mites in commercial blackberry crops this season.

The Tasmanian Institute of Agriculture (TIA) research team has been busy introducing two alternative predatory mites, *Typhlodromus occidentalis* and *Typhlodromalus lailae* (Biological Services) in commercial blackberry crops across Victoria and Tasmania.

Dr Steve Quarrell said *T. occidentalis* or 'Occi' is a robust choice for blackberry crops.

"This mite has been around for quite some time and is regarded as an efficient predator. It is also tolerant of a broader range of environments than some other predatory mites and fairly hardy when it comes to pesticide tolerance," he said.

In contrast, *T laliae* is the new kid on the block and currently being evaluated as a commercial option. Dr Paul Horne, team member from IPM Technologies, says that whilst this mite is new to integrated pest management it has features that make it a good bet for tackling redberry mite.

"This mite is naturally found in the cooler Southern half of Australia making it more likely to survive and persist in these climates. It is known to feed on thrips and broad mite putting it in good stead to tackle redberry mite, which comes from the same family of mites as broad mite," Dr Horne said.

Tasmanian Institute of Agriculture (TIA) project lead, Dr Steve Quarrell says the focus on commercial predatory mites is in response to the projects first season crop survey results.

'We found that predator numbers in commercial blackberry crops were extremely low relative to the number of redberry mites. This means we need to tip the balance in favour of predators by supplementing the natural population with some commercially reared predators', Dr Quarrell said.

The ratio of predator to pest is key to successful integrated pest management. Although there are currently no guidelines for redberry mite, Dr Quarrell says broad mites are a useful model pest.

"For every ten broad mites you need one predatory mite. This is a good guide as to the number we are likely to need to manage redberry mite," Dr Quarrell said.

The fate of both the predatory and redberry mites in the blackberry crops will be closely monitored through both winter and the cropping season to see if this method is a useful predictor of crop infestation levels.

Last season, winter bud sampling proved to be a simple and rapid method for monitoring the redberry mite population.

TIA honours student Hui Law found that in winter you need to sample fewer buds to get a good picture of the mite population compared to sampling during the growing season when mites become more mobile.

"Up to seven times as many redberry mites were detected in winter buds than in fruit. This means that a relatively small number of winter buds need to be sampled compared to fruit to get a good estimate of the population present," Ms Law said.

Pesticides are often necessary in intensive berry production and can be a useful tool in integrated pest management.

Dr Quarrell says selecting the most appropriate pesticide to use in integrated pest management is important to reduce any adverse impact on predators.

"In the case of redberry mite, knowing what pesticides to avoid is just as important as knowing what can be used to complement biological and cultural approaches.

Fungicides can often catch growers out as they don't necessarily see these as being harmful to mites and predators. Switching to registered fungicides that have low toxicity to predatory mites can help tip the balance in favour of predators," Dr Quarrell said.

Looking after predators makes good sense economically and for the future sustainability of intensive blackberry production.

Many thanks to our growers and IPM specialists participating in the predator release trials: Westerway Raspberry Farm, Costa BX (Nandita and the team at Durnorlan), Cindy Edward, Biological Services, Dean Chapman and Kevin Chapman.

More information on this project - RB17000 – Integrated Pest Management of Redberry Mite, *Acalitus essigi*, on blackberries - can be found on the TIA Redberry mite project page **www.utas.edu.au/tia/research/research-projects/projects/red-drupelets-on-blackberries** or by contacting project lead, Dr Stephen Quarrell - **stephen.quarrell@utas.edu.au**

Hort Innovation Strategic levy investment | RASPBERRY AND BLACKBERRY FUND

This project has been funded by Hort Innovation using the raspberry and blackberry research and development levy and funds from the Australian Government. For more information on the fund and strategic levy investment visit horticulture.com.au

'Doreen' to lend a helping hand in blackberry crops

Michele Buntain, Horticulturist, Tasmanian Institute of Agriculture

This project has been funded by Hort Innovation using the raspberry and blackberry research and development levy and funds form the Australian Government. Hort Innovation is the grower-owned, not-for-profit research and development corporation for Australian horticulture.

Redberry mite is a tricky pest of commercial blackberries, but it is not alone. Investigating redberry mite (RBM) has led Tasmanian Institute of Agriculture entomologist, Dr Steve Quarrell on a quest for better integrated pest management (IPM) in an industry which has doubled in size over the last two years.

Investigating redberry mite (RBM) has led Tasmanian Institute of Agriculture entomologist, Dr Steve Quarrell on a quest for better integrated pest management (IPM) in an industry which has doubled in size over the last two years.

"The more we look into RBM management, the more we realise how much there is to learn about the whole blackberry production system," he said. "It's impossible to manage one pest in isolation of other pests and diseases, particularly with the impact of sucking pests such as mirids and green vegetable bugs on fruit quality".

"When you intervene with pesticides for these, it is impacting the management of every other insect or mite in the crop and can undo all the good work you have achieved with beneficials if not managed carefully," Dr Quarrell said.

The good news is that this seasons Tasmanian field trials at Costa Groups Dunorlan and Bengeo farms will investigate how a reduced pesticide program can be married with management of other blackberry pests and diseases. A new native predatory mite from Biological Services, *Typhlodromus doreenae* or 'Doreen' for short, will be introduced alongside existing predators this season. 'Doreen' is a quality choice, feeding on similar pest mites to RBM. Dr Quarrell said the key to any integrated approach that includes predators is monitoring.

"If redberry mite numbers are low then it's time to back off with pesticide use and give predators a chance to do their job. However, I am realistic and know that at times pesticide intervention is needed, we just have to be a bit smarter with our timing of predator releases around that", he said.

Monitoring in the 2018/19 season revealed very low numbers of RBM in commercial blackberry crops. This was great news for growers but can be a little tricky when this is the pest you are trying to research. Dr Quarrell said despite this, his team were able to come up with some encouraging results with the use of predatory mite releases in four blackberry crops, two in Victoria and two in Tasmania.

"We released two predatory mites, *Typhlodromalus lailae* and *Typhlodromus occidentalis* on 3 dates through late spring to mid-summer, and again at the end of the season and monitored mite numbers including RBM, predators and other pest mites throughout the growing season. Over 2,000 fruit samples later, we found lower numbers of RBM and higher numbers of predators, particularly *T lailae*, in the release areas of all but one crop," Dr Quarrell said. The big question still remaining is does RBM cause redberry disease symptoms and how many do you need to cause damage? With low RBM numbers in commercial crops, Dr Quarrell used wild blackberries as a test case.

"There is definitely a positive relationship between redberry mite numbers and redberry disease, the more redberry mites there are, the more damage we observed on fruit. This season we will use a bigger sample size to get a more accurate figure of the numbers needed to initiate damage", Dr Quarrell said.

The complicating factor is that not all red drupelets are due to redberry mite and it can sometimes be a little tricky distinguishing which pest or physiological disorder is the culprit.

"This season we found very few typical redberry mite symptoms, but quite a lot of damage due to sucking pests, most likely caused by mirids and green vegetable bugs. The drupelets for redberry mite stay hard whilst these looked punctured and soft."

Steve sees the next challenge is finding alternatives to the non-selective pesticides used for sucking pests, or alternatively using pesticides more strategically or in trap crops which lure these pests away from the crop.

"Ideally I would like to see if we could introduce a whole systems approach to pest management in Rubus crops and be a bit smarter in how we tackle sucking pests", he said.

Many thanks to fantastic support from Cindy Edwards (YV Fresh), Biological Services, Costa Group, Fairvew Hills, Blue Hills and Lanoma Estate.

More information on this project – RB17000 – Integrated Pest Management of Redberry Mite, *Acalitus essigi*, on blackberries - can be found on the TIA Redberry Mite project page at: www.utas.edu.au/tia/research/research-projects/projects/red-drupelets-on-blackberries or by contacting project lead, Dr Stephen Quarrell on stephen.quarrell@utas.edu.au

Steve Quarrell monitoring RBM

Collapsed blackberry drupelets likely caused by sucking insects

Unripe red drupelets with the appearance of redberry disease

Appendix 6

07 Dec 2017
Launceston Examiner, Launceston TAS

Section: General News • Article type : News Item • Audience : 18,481 • Page: 33
Printed Size: 95.00cm² • Market: TAS • Country: Australia • ASR: AUD 520 • Words: 166
Item ID: 884001416

TIA project studies blackberry colour

BLACKBERRIES are meant to be black, but sometimes they are both red and black.

This discolouration is caused by a microscopic pest called redberry mite and researchers at the Tasmanian Institute of Agriculture are working on strategies to control its impact.

With commercial blackberry production expanding rapidly in Australia is expanding rapidly, the industry needs to stay productive and sustainable, project lead Dr Stephen Quarrell said.

"We are undertaking desktop, laboratory and field-based research to identify tools to prevent and manage redberry mite. As part of this, we will trial integrated pest management programs in select locations over two growing seasons," Dr Quarrell said.

"The project will include extension activities to share findings with growers and facilitate adoption of new strategies to support industry success, with the aim of enabling an increase in the yield per hectare of marketable blackberries," he said.

Blackberry varieties, climatic regions and growing conditions will be assessed. The $260,000 project is funded by Hort Innovation and supported by the institute.

07 Dec 2017
Burnie Advocate, Burnie TAS

Section: General News • Article type : News Item • Audience : 13,468 • Page: 21
Printed Size: 94.00cm² • Market: TAS • Country: Australia • ASR: AUD 389 • Words: 166
Item ID: 884040594

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PRINTED SIZE: 537.00cm² REGION: TAS MARKET: Australia ASR: AUD 3,859 WORDS: 416
ITEM ID: 1085237331

28 FEB, 2019

More mites might be the key to help berries

Launceston Examiner, Launceston

Appendix 7

More mites might be the key to help berries

BY **TIA**

REDBERRY mites cause blackberries to ripen unevenly, typically creating a berry that is half-black and half-red. This can reduce the number of first grade blackberries growers produce.

So how do you stop these micro monsters in their tracks? Researchers think that increasing the number of predatory mites might be the answer.

The Tasmanian Institute of Agriculture (TIA) is working with commercial blackberry growers across Victoria and Tasmania this season to measure the effectiveness of two different predatory mites, Typhlodromus occidentalis and Typhlodromalus lailae.

TIA Researcher Dr Stephen Quarrell said T. occidentalis—affectionately called ‘Occi’—is a seasoned veteran when it comes to protecting crops.

“Occi has been around for quite some time and is regarded as an efficient predator. It’s more tolerant of a broader range of environments than some other predatory mites and fairly hardy when it comes to pesticide tolerance.”

In contrast, T. laliae is the untested rookie with something to prove. Dr Paul Horne, a researcher from IPM Technologies, notes that T. laliae is a good bet for tackling redberry mite.

“This mite is naturally found in the cooler southern half of Australia, making it more likely to survive in these climates. It is known to feed on broad mite, which comes from the same family as redberry mite,” Dr Horne said.

Predatory mites were identified as a potential pest management solution after TIA surveyed crops last year in the early stages of the project. Though some predatory mites were found, the populations were unbalanced.

“We found that predator numbers in commercial blackberry crops were extremely low relative to the number of redberry mites. This means we may need to tip the balance in favour of predators by supplementing the natural population with some commercially reared predators," Dr Quarrell said.

So how many mites is the magical number?

“For every ten broad mites you need one predatory mite to effectively manage the pest. This is a good guide as to the ratio we are likely to need to manage redberry mite,” Dr Quarrell said.

Growers may be unknowingly giving redberry mites the edge over their hungry cousins. However, while pesticides are often necessary in intensive berry production and can be a useful tool in pest management, it’s important to minimise unwanted impact on potentially helpful predators.

TIA will monitor the fate of both the predatory and redberry mites in blackberry crops through both winter and the cropping season.

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PRINTED SIZE: 537.00cm² REGION: TAS MARKET: Australia ASR: AUD 3,859 WORDS: 416
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28 FEB, 2019

More mites might be the key to help berries

Launceston Examiner, Launceston

MITES: TIA researchers think more mites could help growers get more blackberries to market. **Picture: Supplied**


Doreen mighty on mite

04 Sep 2020
Author: CUPPA TIA
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Circulation: 17953

Appendix 8

Doreen mighty on mite

CUPPA TIA

TASMANIAN blackberry growers are embracing research findings from the Tasmanian Institute of Agriculture that offer a softer approach to managing the pest redberry mite.

Redberry mites feed on unripe blackberry fruit, which causes uneven ripening where part of the fruit stays hard and red while the rest is black and ready to eat, making the fruit unmarketable.

Redberry mites are so tiny they easily escape detection until they really start to cause problems.

Last season Tasmanian blackberry grower Costa Berries worked with TIA to trial a softer pesticide program to give predators a head start in managing redberry mite.

Alejandro Haro, horticulture manager for Costa Berries in Tasmania, explained the motivation for trialling the softer program.

"For us it is always important to investigate new control methods for our main pests and if they involve an alternative option to chemicals or the use of softer chemicals, we may find a greater benefit for our programs," Mr Haro said.

TIA entomologist and project lead Steve Quarrell emphasised that managing a pest such as redberry mite required a whole-of-system approach.

"There's quite a lot of detective work involved. It's critical to understand not just the pest but also how other farm management practices could affect the mite's predators, both the native species that are already there and the ones we deliberately introduce," he said.

Research trials in Tasmania and Victoria over the past two seasons involved introduction of three different predator mites and implementation of a reduced pesticide program.

The most promising candidate from these trials is the mite Typhlodromus doreenae commonly known as Doreen.

Dr Quarrell said the results looked promising for a combination of Doreen and a softer summer pesticide program.

"We were really stoked to see reduced redberry mite numbers in our Doreen test rows and even more so that this was matched with the reduced pesticide program," he said.

Sally Kershaw, Driscoll's southern field representative, urged a staged approach to introducing the new pest-management program.

"We are really excited by the results so far but would encourage growers to trial the softer program with a few tunnels or a block before extending it to the entire farm.

"It's about taking a level of risk that you are comfortable with so that you can hone your monitoring and management techniques on a smaller scale first," she said.

Dr Quarrell said the next step would be to confirm that Doreen is happy to persist in the blackberry crop over winter or whether reintroduction is necessary each season.

He said winter was the best time to see what levels of redberry mite were lurking in dormant blackberry buds to help growers plan for the next production season.

A recent workshop, hosted by blackberry supplier Driscolls, brought growers together to learn some simple techniques for monitoring redberry mite in their dormant blackberry crops.

At this workshop, Dr Quarrell demonstrated a grower-friendly technique for extracting mites from buds developed by former UTAS student Hui Law.

"We see real benefits in monitoring as it gives growers a greater appreciation of the pest levels in their crop and the beneficial insects and mites that need to be encouraged," he said.

CLOSE LOOK: Tae Ocean of Hillwood Berries at the microscope. Redberry mite is an issue for blackberry crops that is difficult to detect early.

Table of Contents

Chapter 1. Literature review

1.1 Introduction

Commercial blackberry production has expanded globally due to increased consumer demand (Strik *et al.* 2007). However, red berry disease, retarded or irregular fruit ripening, is causing serious damage and losses in commercial blackberry plantations globally (Gerding 1992; Amrine and De Lillo 2003; Cross, 2012). Several studies have suggested that red berry disease is caused by red berry mite (RBM), *Acalitus essigi* (Acari: Erophyoidea) (Brannon and Breakey 1946; Lindquist *et al.* 1996; Cross, 2012). To date, there is still no conclusive proof RBM causes red berry disease. Hence, the relationship between populations of RBM and the disease incidence of its blackberry host plant (*Rubus fruticosus* L.) will be examined to provide a background for the research questions in this study. Due to limited literature on redberry mite, the studies of superfamily Eriophoidea will be reviewed and then analogies will be drawn and related back to the specific interaction between redberry mite and blackberry plantings.

This literature review is comprised of five sections. The biology of blackberries (*Rubus fruticosus* L.) is examined in the first section as understanding of host plant biology is important in any study which relates to arthropod-plant interactions. In the second section, an overview of the economic importance of blackberries including both cultivated and wild blackberry production worldwide. Studies of eriophyoid mites are examined in the third section with topics covered including: (1) eriophyoid mite taxonomy with detailed characteristics used for identification; (2) the biology and life history of eriophyoid mites; (3) worldwide distribution of *A. essigi*, and association with *Rubus* species; (4) plant-eriophyoid mite interactions covering intraplant distribution of eriophyoid mites and the plant damage linked to eriophyoid mites; (5) dispersal mechanism of eriophyoid mite populations. Quantifying arthropod populations and their damage is critical for understanding their ecology and their impact on their plant host prior any decision making of management. For this reason, the methodologies currently employed to quantify eriophyoid mite populations will be reviewed in the fourth section. The final section will examine current integrated pest management (IPM) techniques utilised to control eriophyid mites including biological, chemical and cultural control methods.

1.2 Biology of blackberries

Blackberry (*Rubus* species) is a perennial berry growing plant. Blackberries are believed to have originated from both Europe and eastern North America (Gustafsson 1942). Blackberries generally have a lifespan of 5–20 years depending on the production region, management systems and cultivar (Strik *et al.* 2007). Cultivated commercial blackberries are hybrid plants and are only distantly related to the wild species which in Australia is classified as a weed and known as European blackberry, *Rubus fructicosus* aggregate. Blackberries undergo significant seasonal and temporal changes in their morphology as they have both vegetative, primocanes (first-year canes) and reproductive, floricanes (second-year canes), which are the primocanes from the previous growing season (Clark 2008) (Figure 1).

All the original cultivated blackberry production was basically produced from floricane-fruiting cultivars, requiring canes to be overwintered to produce a crop. However, numerous commercially grown primocane-fruiting cultivars, which have the potential of producing fruit on both primocanes and floricanes in the same year, have become commercially available since the first primocane releases of 'Prime-Jim' and 'Prime-Jan' by the University of Arkansas in 2004 (Clark and Moore 2005). Cultivated blackberries are often classified according to their cane architecture such as erect, semierect, and trailing canes (Strik *et al*. 2007). Blackberries with erect canes produce primocanes from buds at the base of floricanes, from the crown or from buds on roots, whereas trailing and semierect canes only produce new primocanes from buds on the crown (Figure 2). Erect-caned cultivars include the thorny 'Brazos', 'Cherokee' and 'Tupy'; and the thornless 'Arapaho' and 'Navaho'. Semierect cultivars include 'Loch Ness', 'Chester Thornless', 'Thornfree' and 'Cacanska Bestrna'. Trailing cultivars include 'Marion', 'Silvan', and 'Thornless Ever-green' (Strik *et al*. 2007). Except for the primocane-fruiting erect types, primocanes are vegetative in the first year and fruiting in the second year on the entire length of the floricane (Strik *et al*. 2007).

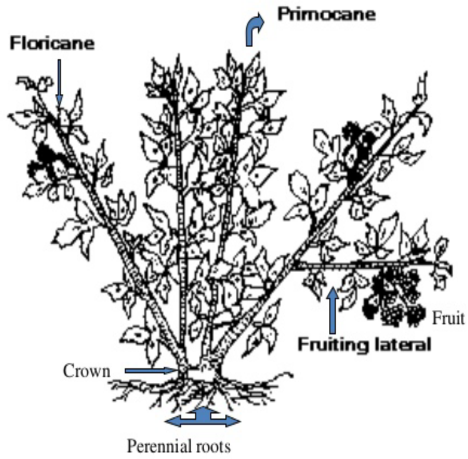

Figure 1. Blackberry plant morphology. Figure source from Poling and Fernandez (1996).

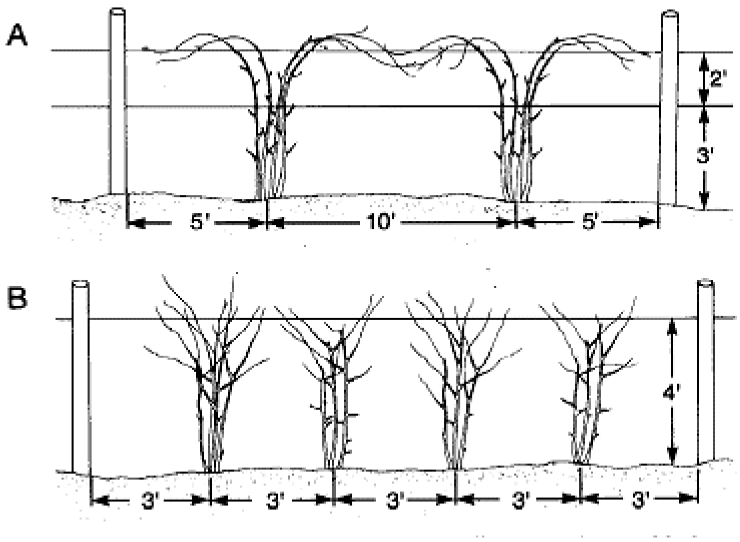

Figure 2. Blackberry cane architecture: A) trailing plants. (B) erect blackberry plants. Figure source from Poling and Fernandez (1996).

1.3 Economic importance of blackberries

1.3.1 Wild blackberries

Wild blackberries still make a significant contribution to worldwide production. For example, Chile processes and exports about one-third of worldwide wild production (5800 tonnes) for human consumption (Strik *et al*. 2007). However, the European blackberries, especially the

Rubus fruticosus L. aggregate, and other exotic *Rubus* spp. are important weeds of natural ecosystems and it is categorised as a Weed of National Significant in Australia (WoNS) (Thorp and Lynch 2000). Wild blackberries are considered as one of the worst weeds in Australia due to its invasiveness, environmental and economic impacts (Chalak and Pannell 2011). From an agricultural perspective, wild blackberries hinder site access, reduce grazing capacity, provide habitat for noxious animals (Adair and Bruzzese 2006) and provide shelter and food sources for insect pests and natural enemies, which can drastically alter insect population dynamics (Norris *et al.* 1982). Economic losses due to wild blackberry in Australia were estimated at $42 million per annum in 1984, but the actual losses had increased to approximately $70 million per annum by 1990 (Page and Lacey 2006). Members of the *R. fruiticosus* aggregate were naturalised in Tasmania by 1878 and the species composition of these introductions remains obscure (Evans and Weber 2003). Sixteen species of *R. fruticosus* agg. have been classified by a combination of DNA fingerprint data and morphological identification conducted by European and Australian *Rubus* specialists (Evans *et al.* 2007) who have listed the 16 species of common and widespread taxons of the *R. fruticosus* agg. in Australia (Table 1).

Table 1. Taxa of Rubus fruticosus aggregate and distribution of each taxon from Evans *et al*. (2007)

Taxon	Distribution
Rubus anglocandicans	The most common and widespread of the weedy *Rubus* in Australia. It is endemic to Britain.
Rubus cissburiensis	It is endemic to Britain where it is widespread.
Rubus echinatus	Rare in Victoria, but apparently more widespread in Tasmania. It is widespread in Britain and in west-central Europe.
Rubus erythrops	Found in Victoria, Tasmania and South Australia. It is endemic to Britain.
Rubus laciniatus	All southern states except Western Australia. It is widespread in Britain and in north-western Europe.
Rubus leightonii Lees ex Leight.	Found in New South Wales. It is widespread in Britain and occurs in northern France.
Rubus leucostachys Schleich. ex Sm.	Widespread in all southern states except Western Australia. It is endemic to Britain.
Rubus phaeocarpus	Recorded from South Australia, principally in the Lenswood area, and in New South Wales. It is widespread in south-eastern Britain and occurs in northern France.
Rubus polyanthemus Lindeb.	Recorded from Victoria and New South Wales. It occurs widely in Britain and north-western Europe.

Rubus riddelsdellii Rilstone	Recorded from Mt Lofty Ranges of South Australia but may have a wider distribution due to Rubus riddelsdellii is easily confused with R. leucostachys. It is endemic to southwest Britain.
Rubus rubritinctus	Recognised in Mt Lofty Ranges of South Australia. Also recorded from southern Tasmania. It is widespread and common in Britain, possibly extending to France.
Rubus sp. Tasmania (unknown species)	Only found in Tasmania.
Rubus ulmifolius Schott	Recorded from Western Australia, New South Wales, Victoria and South Australia.
Rubus ulmifolius Schott var. *ulmifolius*	Recorded and widespread in Western Australia, New South Wales, Victoria and South Australia. It is widespread in Britain extending as far north as Scotland in suitable sites. It occurs widely in continental Europe (except the north) and extends south to Italy, Spain, Portugal and to north-western Africa.
Rubus vestitus	Recorded from South Australia, Victoria, Tasmania and New South Wales. It is not common in Australia, but is widespread in Britain and throughout western and central Europe
Rubus laudatus	Native to Missouri, Kansas and Illinois in the USA. Well established in Western Australia, south-eastern Queensland and NSW.

1.3.2 Commercial blackberry cultivars

The majority of commercial blackberry varieties have been bred since 1850 from North American blackberry strains that are not members of the *R. fructicosus* aggregate (Amor and Richardson 1980). Cultivated blackberries (*Rubus fruticosus* L.) have become a common fruit in the global market, especially in North America and the European Union (Clark and Finn 2014). The worldwide growing area for cultivated blackberries increased 44% from 13,958 ha in 1995 to 20,036 ha in 2005 (Strik *et al*. 2007). The fresh blackberry industry in California expanded rapidly in the 1990s and has since expanded further into Mexico in the 2000s. In addition, the Pacific Northwest had a significant expansion of their fresh market in over a similar period of time (Clark and Finn 2014). According to recent figures, the blackberry production area in the USA has continued to expand from 2,468 ha in 2014 to 2,833 ha in 2016 (USDA 2016). In Australia, of the 120 *Rubus* growers nationally, only ca. 20% grow blackberries with the remainder focusing on raspberry production. However, the production volume of blackberries has doubled over the last five years with the value of production increasing from $40 million in 2013 to more than $50 million in 2014/15 (Mulderij 2016). The greatest expansion of fresh blackberry production has been driven by a combination of factors including improved cultivars, marketing efforts, stable fruit supply and increased interest of consumers in berries for health benefits as blackberries contain substantial antioxidant levels (Clark and Finn 2014; Strik *et al*. 2007).

1.4 Eriophyoid mites

1.4.1 Eriophyoid mite taxonomy

The taxonomy of mites, ticks and spiders (Class: Arachnida) is complex. Acari (mites and ticks), which is one of the sub-classes of Arachnida, has three major superorders: Acariformes, Parasitiformes and Opilioacariformes (Walter and Proctor 1999). The family Eriopyoidae, to which RBM belong, lie within the superorder Acariformes, the order Trombidiformes, the suborder Prostigmata and the superfamily Eriophyidea (Armine 1996). However, the taxonomy of the superfamily Eriophyidea has been poorly studied with less than 5 percent of the species believed to have been described. Although the identified Eriophyidea includes about 250 genera and over 2900 named species, it is estimated that ca. 18,000 to 19,000 new species are yet to be identified (Lindquist and Amrine 1996). The following list (Table 2) has been summarised by Lindquist and Amrine (1996) and is limited primarily to eriophyoid genera, which include at least some pest species of economic importance in agricultural or horticultural crops and ornamental plants.

Table 2. *Classification of the Superfamily Eriophyoidea From Lindquist and Amrine (1996)*

Family **Subfamily** Tribe	**No. of genera**	**Economically Important Genera**
Phytopidae	**18**	
Phytopinae	3	*Acarthrix, Phytoptus*
Sierraphytoptinae	8	
Sierraphytoptini	4	
Mackiellini	4	*Retrarcus, Mackiella*
Novophytoptinae	1	
Nalepellinae	6	
Nallepellini	3	*Nalepella, Setoptus*
Trisetacini	2	*Trisetacus*
Pentasetacini	1	
Eriophyidae	**168**	
Eriophyinae	27	

Eriophyini	10	*Eriophyes, Nacerimina*
Aceriini	16	*Acalitus, Acermina, Aceria, Kieferophyes, Paraphytoptus*
Diphytoptini	1	
Phyllocoptinae	108	
Phyllocoptini	36	*Acadricus, Acritonotus, Calepitremerus, Epitrimerus, Platyphytoptus, Phyllocoptruta, Rhombacus, Phyllocoptes, Vasates*
Tegonotini	11	*Dichrothrix, Scolocenus, Oxycenus, Shevtchenkella, Tegonotus*
Acaricalini	15	*Acaricalus, Acaphylla, Acaphyllisa*
Calacarini	6	*Calacarus*
Anthocoptini	40	*Aculus, Aculops, Anthocoptes, Ditrymacus, Heterotergum, Metaculus, Notostrix, Parulops, Tetra, Tetraspinus*
Nathopodinae	10	
Aberoptinae	3	*Cisaberoptus*
Cecidophyinae	19	
Cecidophyini	9	*Cecidophyopsis, Cecidophyes, Coptophylla*
Colomerini	10	*Cosetacus, Colomerus, Paracolomerus*
Ashieldophyinae	1	
Diptilomiopidae	**40**	
Diptilomiopinae	23	*Apodiptacus, Dialox, Diptacus, Diptilomiopus, Rhynacus, Trimerptes*
Rhynchaphytoptinae	17	*Catarhinus, Cheiracus, Rhyncaphytoptus*

1.4.2 Eriophyoid mite biology and life history

Eriophyoid mites are small, with body lengths ranging from 80 to 500 µm and an average length of approximately 200 µm. There are two major body forms of eriophyoid mites, either vermiform or fusiform (Figure 3). In general, eriophyoid mites have elongate, annulate and worm-like bodies, two pairs of anteriorly placed legs and, a transverse genital aperture close behind the legs (Lindquist 1996; Westphal and Manson 1996). The prodorsal shield may bear two pairs of setae or none at all. The tarsi lack claws but bear a feather-shaped empodium, which is commonly known as feather claw. The genital opening of adults of both sexes is positioned proximally, closely behind the bases of the legs. The setae on the body and appendages are mostly simple and tapered and rarely spinelike (Lindquist, 1996).

Eriophyoid mites have a relatively simple developmental history that may be completed in 6–14 days where they start life as an egg, pass through the larva stage and the nymphal stage and then finally emerging as an adult (Hull 2014). Female eriophyoid mites have two nymphal instars followed by a resting “pseudopupa” stage, but males are not often seen for which the reason is unknown (Hull 2014). The transfer of sperm without copulation also known as sexual dissociation is widespread in several groups of arthropods including Arachnida, Hexapoda, Pauropoda, Symphyla and Diplopoda (Alexander 1964; Proctor 1998). Sex dissociation can be complete or incomplete. For instance, sexual dissociation of some water mites is incomplete where spermatophore deposition is triggered only by previous contact with females or their odours (Proctor 1992). However, sexual dissociation in eriophyoid mites including *Aceria sheldoni* (Sternlicht and Griffiths 1974), *Epitrimerus pyri*, *Aculus schlechtendali* (Oldfield 1988), *Aculus fockeui* (Oldfield *et al.* 1970), *Aculus allotrichus* (Michalska and Boczek 1991) and *Cecidophyopsis hendersoni* (Michalska and Shi, 2004) is complete with the males depositing spermatophores in total isolation from both conspecifics and their odours (Oldfield et al. 1970; Michalska and Boczek 1991; Michalska and Shi 2004). Sexual dissociation has also been confirmed in RBM (Michalska and Boczek 1991).

Eriophyoid mites live gregariously on plants and often develop to high population densities (Sabelis and Bruin 1996). High density populations may favour sex dissociation, as it increases the availability of receptive females and thereby the probability that spermatophores will be visited by females (Proctor 1998). Eriophyoids are the only known group of obligatory herbivores exhibiting this mode of reproduction (Proctor 1998). Due to Eriophyoid mites having limited independent powers of dispersal, they tend to live and reproduce only on susceptible host-plant species (Westphal and Manson 1996). This host specificity is usually confined to one plant genus, or, at most, the members of a single family. They cannot survive for long periods away from a host plant, hence, most of the plant species on which they feed are perennial (Hull 2014). Many eriophyoid mite species including the RBM, *Acalitus essigi* (Hassan) and others such as *Aceria rubensis* Manson and *Calepitrimerus chemaemori* on *Rubus caesius*, and *Eriophyes rubifolii* on *Rubus racemosus* have been reported to feed on *Rubus* species worldwide including on *Rubus fruticosus* aggregate spp. (Amrine and Stasny 1994).

Eriophyoid mites have four typical life styles (vagrants, refuge inhabiting, bud inhabiting and gall/erinea inducing). However, some species may exist that fall in-between these categories (Sabelis and Bruin 1996). Vagrant eriophyoid mites often live openly on the leaf surface but may move to shelter such as under bud scales on host plant during winter. Refuge inhabiting eriophyoid mites hide in confined refuges on host such as the space between leaf axils and buds; within flowers, seed heads and fruits. Bud inhabiting eriophyoid mites live permanently under bud scales. Gall/erinea inducing eriophyoid mites can induce the host plant to create growth abnormalities that can then be used as shelters such as leaf edge curls, erinea and galls (Sabelis and Bruin 1996). RBM has been considered as a refuge inhabiting eriophyoid mite (Davies *et al.* 2001a) because it is commonly found in the area between buds and leaf axils and within berries (Hanson 1933; Hamilton 1950; Davies *et al.*, 2001a).

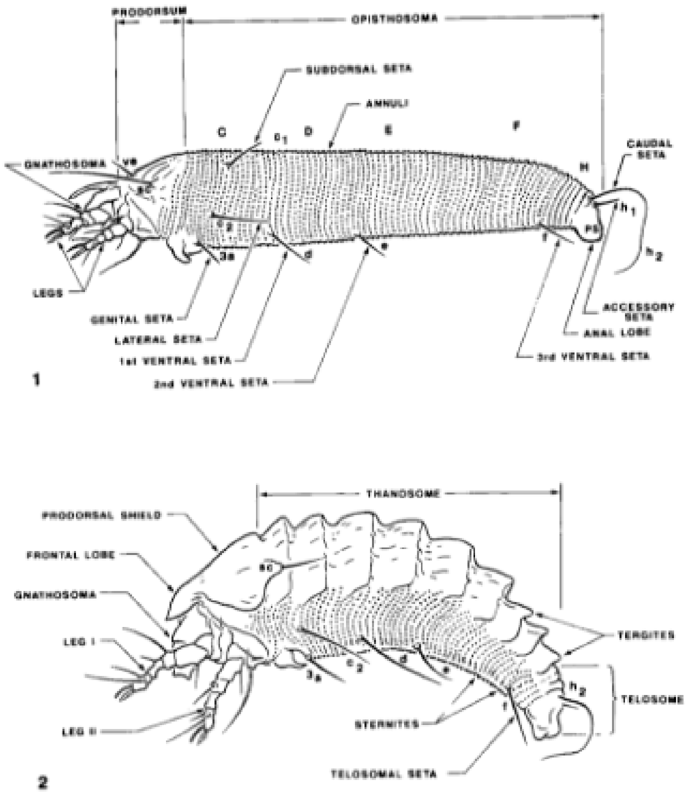

Figure 3. Two major body forms of eriophyoid mites in lateral view. (1) A vermiform mite; (2) A fusiform mite from Lindquist (1996)

1.4.3 Worldwide distribution of redberry mite (*Acalitus essigi*) populations

The red berry mite (RBM) is a refuge inhabiting species found in blackberries (Davies *et al.* 2001a) which has become an important pest species in agricultural sectors worldwide. Previous studies have reported that RBM can cause significant damage to both wild and modern blackberry cultivars (Essig 1925; Amrine and De Lillo 2003; Szendrey *et al.*, 2003) with the associated damage leading to bright red, hard and inedible fruit drupelets commonly known as red berry disease (Lindquist *et al.* 1996; Davies *et al.* 2001b). RBM can be found in leaf axil microhabitats and buds on both primocanes and floricanes, where in the latter it eventually occupies the fruit calyx and small crevices between the drupelets near the core of the developing fruit (Davies *et al*. 2001b). Fluctuations of RBM populations within the different microhabitats across the year are also evident (Davies *et al*. 2001a). RBM populations decline in bracts and leaf axils from summer to winter (December-August), but increase in number from February onwards within bud microhabitats where they overwinter. In spring, overwintering RBM populations within buds emerge from these overwintering sites (Davies *et al*. 2001a). Once emerged, RBM reside within in bracts and leaf axils until dispersing to flowers and developing fruits during spring and summer periods (Murrietta and Gaskell 2015).

RBM reside in most regions where both wild and commercial *Rubus* grow including America, Canada, Chile (Koch *et al.*, 2000, Strik *et al.*, 2007) and in many parts of Europe (Szendrey *et al.* 2003) including Italy, Poland (De Lillo and Duso, 1996, Knihinicki and Boczek, 2002), Germany and Hungary (Strik *et al.* 2007). In Hungary, RBM damage commercial blackberries in particular thornless varieties such as Thornfree, Dirksen Thornless and Loch Ness (Szendrey *et al.* 2003). RBM has also been noted as causing red berry disease in cultivated blackberry in New Zealand since 1946 (Hamilton 1950). In Australia, the first confirmed records of RBM are from raspberry, *Rubus idaeus* L. collected in Tasmania in 1976 (Knihinicki and Boczek 2002). The mite was subsequently identified on unknown species of wild blackberries within the *R. fruticosus* L. agg. in NSW in 1994 (Knihinicki and Boczek 2002) and from *R. leucostachys* Schleich. ex Sm. and *R. polyanthemus* Lindeb. in Neika, Tasmania in 2000 (Davies et al. 2001a). Although not formally identified, red berry disease which is believed to be caused by RBM on wild blackberry species has been noted as being wide-spread in Victoria (Scott *et al.*, 2008).

Despite several studies implicating RBM as the causal agent of redberry disease (Brannon and Breakey, 1946; Lindquist *et al.* 1996; Davies *et al*. 2001b), whether a relationship exists between the control of RBM population and incidence of red berry disease is still uncertain. According to Cross *et al.* (2012), attempts to reduce redberry disease symptoms with the application of acaricide treatments did not reduce redberry disease despite RBM populations being reduced by ca. 96%. Why the decline in mite numbers did not reduce disease symptoms is currently unclear and warrants further investigation.

1.4.4 Plant-eriophyid mite interactions

Eriophyoid mites are obligate plant parasites that feed on various plant organs, some eriophyoid mites can transmit plant pathogens and viruses that seriously impact on crop production. Feeding damaged by eriophyoid mites can sometimes be confused with those symptoms associated with viruses, nutrient deficiency and physiological disorders (Van Leeuwen *et al.*, 2010). Eriophyoid mite-plant interactions, mite species, life histories and host plant species are important factors that result in diversity of symptoms on plants (Petanović and Kielkiewicz 2009). The involvement of gnathosoma structures which include the mouth, chelicerae (specialized feeding appendages), and palps or pedipalps (Figure 3) are all believed to be involved in the feeding process of eriophyoid mites. Not all phases of the feeding process are currently understood (Nuzzaci and Alberti 1996). However, it is understood that the stylets of eriophyoid mouthparts are inserted into the cell wall mechanically and three glands which consist of paired and unpaired glands produce secretions. It is believed that the paired glands function as true salivary glands, but the function of the unpaired glands is still unknown (Nuzzaci and Alberti 1996).

Saliva of eriophyoid mites injected from the mouthpart which travels between cheliceral stylets and labrum enters the plant cell, whereas plant cell content moves through a canal formed by the infracapitular stylets and the labrum when eriophyoid mites are ingesting (Nuzzaci and Alberti 1996). However, some studies suggested that the cheliceral stylets are inserted into the plant cell during mite feeding (Jeppson *et al.* 1975) or that the mite withdraws its chelicera from the cell wall after it is perforated and then fixes the labrum to the hole in the cell wall to actively uptake the cell contents from plant tissue (Westphal and Manson 1996). RBM feed on the flowers, foliage and fruits with their stylets penetrating plant tissues. The injection of toxic saliva from RBM into the developing fruit drupelets is thought to result in red berry disease (Brannon and Breakey 1946; Westphal and Manson

1996; Cross *et al.* 2012; Lindquist *et al.* 1996; Davies *et al*. 2001b). RBM damage is greater in later fruiting blackberries cultivars such as Quachita than early season fruiting cultivars (Ferreira *et al.* 2015). This increase in damage is believed to be associated with higher RBM population sizes later in the season coupled with the mite's dispersal prior to overwintering (Ferreira *et al.* 2015).

a) b)

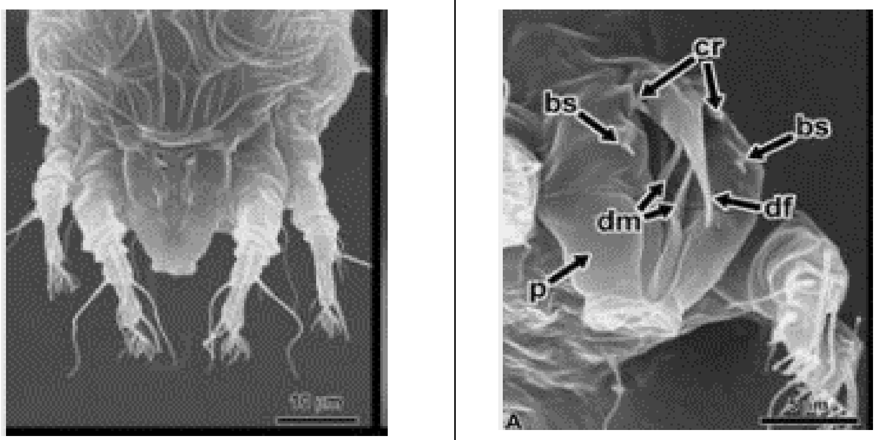

Figure 4. Floracarus perrepae mouth structure. a) dorsal view of anterior showing the gnathosoma and paired legs; b) gnathosoma with cheliceral stylets (digitus fixus (df) and digitus mobilis (dm)) extended, and cheliceral retainers (cr), basal (palpcoxal) setae (bs), and pedipalp (p) from Freeman *et al.* (2005)

1.4.5 Eriophyoid mite dispersal

Due to their restricted host plant range, finding a proper host plant is important for mite survival and reproduction. Therefore, the mode of dispersal utilised by a species is a key component of the foraging strategy for eriophyoid mites, which can influence their feeding behaviour and subsequent host acceptance (Michalska *et al.* 2010). Eriophyoid mites can stay on the host plant and continue to increase their population size or disperse to find and colonise a new habitat when the host plant becomes unavailable (Michalska *et al.* 2010).

The mode of dispersal utilised by eriophyid mites can be either active or passive. However, due to their tiny size, eriophyoid mites cannot actively seek new hosts, hence, the only active dispersal mode of eriophyoid mites is by walking or crawling within their current host plant, or from one plant to another if the plants are physically touching each other (Michalska *et al.* 2010). Generally, passive dispersal of eriophyoid mites depends on environmental conditions such as high temperature and humidity coupled with significant wind, initiated by approaching low pressure systems and is independent of the condition of the host plant (Michalska *et al.* 2010).

Passive dispersal of eriophyoid mites has been observed in many studies. One method used is phoresy or the use of animal carriers such as the mite, *Aceria cynodoniensis* which is transported on mole crickets (Cromroy 1983). Another method is by rain dispersal such as the grape rust mite*, Calepitrimerus vitis* (Duffner *et al.* 2001). Duffner *et al.* (2001) has suggested that rain dispersal is possible in *C. vitis* due to a large number of adults and juveniles *C. vitis* were caught in the rain run-off from the foliage. Higher number of adults *C. vitis* than juveniles in the water run-off has also suggested that adults are more exposed to rain when they moving from leaves to shoots, whereas juveniles are mainly accomplish their ontogenesis on the lower side of the leaves (Duffner *et al.* 2001). Transference by air currents or wind such as utilised by the gall forming mite, *Floracarus perreperae* (Ozman and Goolsby, 2005). Eriophyoid mites inhabiting hosts that are transient in space and time must disperse more frequently than if their hosts were more permanent, hence, eriophyoid mites are more likely to evolve on a transient host. Low host specificity has been proven for wind-dispersed species that live on short-lived or unpredictable hosts such as annual crops. For instance, *Aceria tosichella* is a monocot-infesting species, which is dispersed by wind, has a wide range of grass hosts (ca. 120 species) (Nault and Styer, 1969). Despite these other methods of dispersal, wind is believed to be the dominant mode of interplant dispersal for eriophyoid mites with eriophyoid dispersal by wind reported most frequently in many studies (Michalska *et al.* 2010) (see Figure 5). For instance, daily collections of possible eriophyoid mites that disperse aerially by using water tray trap (i.e. container with water) over one year has been documented that more eriophoid mites are captured on windy days compared to non-windy days (Zhao and Amrine Jr 1997). Lindquist and Oldfield (1996) has reported that eriophyoid mites do not show clear phoretic morphological adaptations, such as the pedicels or claws which can be found in other phoretic mites and no specific adaptations for selectivity toward a more favourable carrier. Hence, the use of carriers by eriophyoid mites is rather rare and accidental behaviour (Lindquist and Oldfield 1996).

Due to RBMs ambulatory ability being limited by its slow, deliberate movements (Murrietta and Gaskell, 2015), its dispersal also relies on methods other than aerial dispersal during windy periods (Hanson 1933; Davies *et al*. 2001b). Davies *et al*. (2001b) concluded that RBM utilise plant growth and the subsequent interactions this growth creates between neighbouring blackberry plants to facilitate their dispersal. This in turn localises the distribution of RBM creating aggregated populations on the plant. For this reason, adhesive barriers such as Tanglefoot® on the fruit's pedicle have also been successfully used to block RBM from infesting young fruits, indicating that crawling via pedicel is the main method of dispersal used by RBM within blackberry plants (Davies *et al*. 2001b).

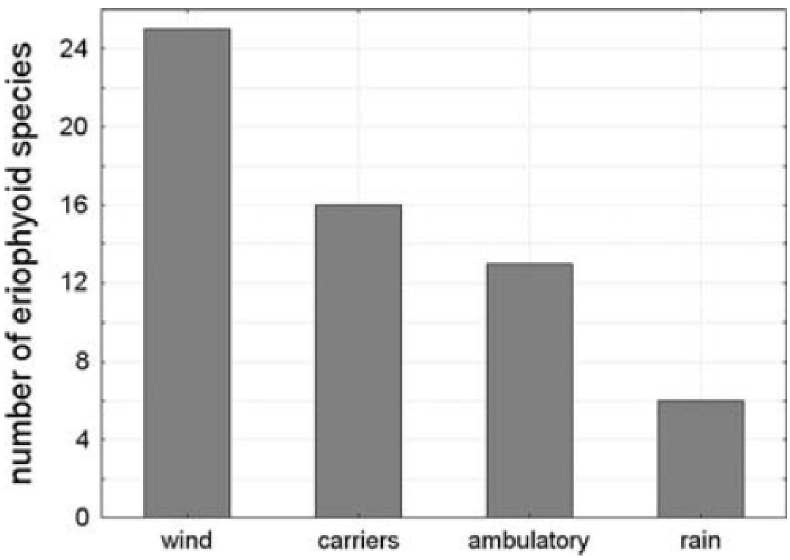

Figure 5. The number of eriophyoid species versus mode of dispersal derived from a search of the literature, from 1912 to 2008 summarised by Michalska et al. (2010).

1.5 Sampling and extraction methods of eriophyid mite

1.5.1 Extraction, detection and collection of eriophyoid mites

Sampling and monitoring of pest and natural enemy populations is critical in integrated pest management (IPM) for decision-making and allowing prediction of the optimal timing for the application of control treatments (Kelly and Mazomenos 1992). Due to their tiny body size and cryptic life-style, eriophyoid detection, collection and manipulation has become challenging, especially when they must be kept alive on plants for host specificity tests or further studies (Monfreda *et al.* 2007). Researchers mostly collect eriophyoid specimens directly from plant material under a dissection microscope or utilise cuttings to infest plants. Various trapping or extraction techniques have been used to collect eriophyoid mites including sticky tape or other adhesive traps, ultrasonic baths, centrifugal flotation, brushing machines or washing methods (Monfreda *et al.* 2007). Most of the applied methods utilise a sticky tape method or shake-and-wash technique to dislodge mites from leaves, shoots, buds and plant deformations (Monfreda *et al*. 2007). However, most sampling methods are often labour-intensive, time-consuming, and sometimes inaccurate with either fresh or dried samples (Monfreda *et al*. 2007).

1.5.2 Sticky tape method

Harvey and Martin (1988) first developed the use of sticky tape to quantify the populations of wheat curl mite, *Eriophyes tulipae* in wheat spikes by placing the excised spikes onto the sticky side of sticky tape. Allowing the spikes to dry forced the mites to crawl out of the complex structure with the mites subsequently stuck on the tape. This process required approximately two weeks. The mite populations were then counted under 15x magnification using a binocular microscope. Davies *et al.* (2002) adapted the sticky tape method with and without a supplementary synthetic pyrethroid spray to extract RBM population from blackberry fruits. This study demonstrated that the sticky tape method when supplemented with a synthetic pyrethroid spray extracted twice the number of RBM from blackberry fruits compared to the sticky tape method alone. It was subsequently hypothesised that the spray killed the more exposed predatory mites, leading to reduced mortality (and consumption) of RBM and a higher number of extracted RBM.

In addition, Bernard *et al.* (2005) used a sticky-method to assess *Calepitrimerus vitis* (Nalepa) emerging from buds during the spring migration period. Double sided sticky-tape was placed above the base of each grape spur and below each node, with the sticky tape replaced every three days. Mite numbers on the tape were then counted through transparent plastic sheets at 50–70x magnification, to assess the population density. Walton *et al.* (2007) has used a similar method to assess the incidence of eriophyoid mites on dormant plant tissues of grapevines. Sticky tape was wrapped around each cane, one at the base of a shoot close to the main trunk and another three buds away from the base of each cane. Sticky tape samples were replaced every 15 days.

Although sticky tape methods can be useful for spatial and temporal distribution studies of eriophyoid mites, these methods are time consuming and may create some difficulties in mite quantification and monitoring. Furthermore, the excessive period required to extract the mites leads to their desiccation on the sticky tape complicating identification and moisture on the tape from the fruit could affect stickiness over time also impacting on mite quantification. Similarly, detachment of the mites from the sticky tape without destroying them for formal taxonomic identification and storage is difficult and hence this method is unsuitable for some studies (Monfreda *et al.* 2010).

1.5.3 Shake and wash method

As it is often necessary to extract eriophyoids from plant material, especially from complex plant structures which can hide small aggregated mite colonies the ‘shake and wash’ method can be useful to estimate mite populations (Monfreda *et al*. 2007). This method has been widely used to extract and quantify eriophyoid mites in many studies (Zacharda *et al.*, 1988, Pérez-Moreno and Moraza-Zorilla, 1998, Monfreda *et al.*, 2009). A summary of the studies which have successfully used this method was summarised by Monfreda *et al.* (2010) (Table 3).

Table 3. Characteristics of washing techniques with suggestion for use from various studies from Monfreda *et al*. (2010)

Procedure	Chemicals or solutions	Devices	Suggested use
Zacharda et al. (1988)			
Shake and wash technique	80–90% ethanol	Separating funnel	Biological and taxonomical studies when live EMs are not required
Pérez-Moreno and Moraza Zorrilla (1998)			
Wash and sieve technique	70% ethanol Tap water 70% ethanol–5% glycerine	25-μm sieve	Biological and taxonomical studies when live EMs are not required
Duffner (1999)			
Shake, wash and sieve technique	Detergent solution in water Methylene-blue (1%)	90 and 32-μm sieves Vacuum pump	Biological and taxonomical studies when live EMs are not required
Gabi and Mészáros (2001)			
Shake, wash and sieve technique	Tap water Azur II eosin	Filtering apparatus (tissue mesh size: 300 and 40 μm)	Biological and taxonomical studies when live EMs are not required
Faraji et al. (2004)			
Shake, wash and sieve technique	Detergent solution in water 70% ethanol Methylene blue Paraffin	45-μm meshed sieve Centrifuge U-shaped Plexiglas® mite-counting channel	Mite quantitative determination, when live EMs are not required
Siriwardena et al. (2005)			
Shake and wash technique	Detergent solution in water	Equipment: tube; funnel; and Mohr's clip Rotor-mixture	Biological and taxonomical studies, EMs quantitative determination, especially when there is high population density
Monfreda et al. (2007)			
Shake and wash technique	Detergent and bleach solution in water Eggs: kaolin powder and $MgSO_4$	Sieves (mesh size: 850, 180, 53 and 25 μm) Filter holder (mesh size: 20 μm) Eggs: centrifuge	Biological, taxonomical studies, rearing, mass release, host specificity and biological assays, when live specimens are required

EMs eriophyoids

Zacharda *et al.* (1988) developed a shake and wash technique for monitoring both predatory and phytophagous mites in apple orchards, which was successfully employed to monitor *Aculus schlechtendali* populations*. Aculus schlechtendali* was extracted from infested plant material such as leaves, spurs and shoots in apple orchards by immersing the infected plant materials in 80-90% ethanol in a covered beaker, which was shaken for few seconds. The number of mites was counted under a dissecting microscope after alcohol evaporation. Similarly, Pe´rez-Moreno and Moraza Zorrilla (1998) developed a standardized method to sample *Calepitrimerus vitis* (Nalepa) using a washing and sieving method. Grapevine leaf samples were submerged in a 70% ethanol solution for five minutes to kill the mites. The leaves were individually washed under tap water and mites collected using a 25 µm sieve, and then transferred to a Petri dish using a solution of 70% ethanol and 5% glycerine. After the ethanol evaporated, the mites were embedded in the glycerine layer. The eriophyoids were then counted under a dissecting microscope at 70x magnification by placing a sheet of millimetre graph paper under the dish to facilitate accurate quantification. A similar procedure was applied to buds and basal cane portions, where the bud scales were carefully opened with pointed forceps, or the basal cane portions after lifting up of the bark, were placed in a jar with 70% ethanol and stirred for 10 min with a magnetic stirrer. The suspension was then poured through two sieves: the upper sieve (1 mm opening) retained coarse plant material and the lower sieves (25 µm opening) collected the eriophyoids. This method developed by Zacharda *et al.* (1988) and Pe´rez-Moreno and Moraza Zorrilla (1998) kills mites quickly without physical damage to extracted mites from infested plant material, which can also be preserved for further study. However, this method is not suitable for researchers who need to collect live specimens from infested plant material for use in behavioural studies (Monfreda *et al.* 2010).

Monfreda *et al.* (2007) has developed a more complex, but accurate shake and wash method for extracting and collecting different eriophyoid mites and their eggs. This method uses a bleach or detergent solution for washing and removing the mites from infested plant material. ASTM-stainless-steel sieves are then used for filtration of any sediment and eggs which are then separated from other mite stages by centrifugation. As bleach, detergent and tap water are readily available and cheaper than ethanol, it provides a cost-effective method of mite extraction. However, these solutions cannot be used for specimen preservation for further study and the associated equipment such as filter holders, nylon filters, and sieves are relatively expensive (Monfreda *et al.* 2010). However, the main advantage of this method is motile eriophyoid mite species can remain alive and active after collection, which can be valuable for taxonomic and biological studies (Monfreda *et al.* 2010).

1.6 Integrated pest management of eriophyid mites

IPM has been defined as a pest management strategy with the aim of long term prevention or suppression of pest populations without negative impacts on the environment, human health, and non-target organisms (Flint *et al.*, 1991). IPM uses a combination of practices such as cultural control for example habitat manipulation, biological control using natural enemies, including predators, and chemical control by using selective pesticides with lower toxicity to humans or non-target organisms (Flint *et al.* 1991). Monitoring pest and beneficial insect populations prior taking any action is an important aspect in IPM and involves the utilisation of economic thresholds. Economic thresholds are pest densities which indicate a control treatment should be taken to prevent an increasing pest population that will cause yield losses equal or greater than to the pest management costs (Higley *et al.* 1994). However, economic thresholds for RBM are yet to be determined (Murrietta and Gaskell 2015).

1.6.1 Cultural control

Cultural pest control is the management of pests by manipulation of the environment or implementation of preventive practices such as using alternate plant species or varieties that resist pests or the adoption of canopy management strategies i.e. pruning techniques that alter the microclimate in the plant canopy to make it less favourable to pest species (Flint *et al.* 1991). Successful cultural management of RBM may lie in cane management, field sanitation and cultivar selection (Murrietta and Gaskell 2015). The cultural control of RBM is mainly management of blackberry canes because the absence of annual pruning of floricanes is suggested to provide RBM with continual nutritional resources and shelter allowing RBM populations to increase rapidly after overwintering (Murrietta and Gaskell 2015). Annual pruning of primocane fruiting cultivars during early winter was suggested to interrupt the RBM's lifecycle by removing the overwintering sites (Murrietta and Gaskell 2015).

Similarly, old blackberry canes and plant residues should be removed and burned as soon as the blackberry harvest is complete to minimise the survival of RBM in drying and dead tissues, and subsequent dispersal to new spring flush (Brannon and Breakey 1946; Murrietta and Gaskell 2015). The differences in RBM populations between two cultivars of blackberries were studied by Murrietta and Gaskell (2015) to determine the effects of cane management on RBM population densities. The study demonstrated that RBM populations from vegetative, flower and fruit samples of 'Quachita' cultivar (floricane fruiting cultivar) were significantly greater than 'PrimeArk® 45' populations (primocane fruiting cultivar). The high density of RBM in floricane vegetative buds suggested that RBM migration to fruit could be more successful on floricanes than on primocanes, however, further studies are required to prove this assumption (Murrietta and Gaskell 2015).

1.6.2 Biological control

To date, the biological control of eriophyoid mites is little known, with suggestions from previous studies mainly based on the use of predators, and to a lesser extent, on pathogens (Van Leeuwen *et al.* 2010). Four different types of life histories have evolved in the predatory Phytoseiidae based on the differences in food utilization (McMurtry and Croft, 1997). The life histories of phytoseiid includes: specialized predators represented by the *Phytoseiulus* species (Type I) which are mainly associated with the spider mite *Tetranychus* species; selective predators of spider mites represented by *Galendromus*, some *Neoseiulus*, and a few *Typhlodromus* species (Type II); generalist predators represented by some *Neoseiulus* species and most *Typhlodromus* and *Amblyseius* species, as well as species in all other genera about which information is available (Type III) ; specialized pollen feeders or generalist predators represented by *Euseius* species (Type IV) (McMurtry and Croft 1997). Detailed examples of the genera and species of phytoseiids with four lifestyles have been listed by McMurty and Croft (1997) (see Table 4). Type I and II specialist phytoseiids is density-dependent as they tend to aggregate in increasing number of prey colonies whereas aggregation of Type III and IV generalist phytoseiids are unlikely to be density-dependent as they will forage more randomly and pick off wandering prey before extensive colonization occurs (McMurty and Croft 1997).

Table 4. Examples of genera and species of phytoseiids with four lifestyle that summarised by McMurty and Croft (1997).

Genus[a]	Species	Distribution[b]	Type of crop[c]	Strategy[d]
Type 1				
Phytoseiulus	*persimilis*	Nt, Pa, Na (I), Au (I), Et (I)	L, G	C, A, Im
	macropilis	Na, Nt, Et, Pa	L	A
	longipes	Et	L, G	A
Type II				
Galendromus	*annectens*	Na, Nt	T	C, A
	helveolus	Na, Nt	T	C, A
	occidentalis	Na, Pa (I), Or (I)	T, L, V	C, A, Im
Typhlodromus (Typhlodromus)	*rickeri*	Or, Na (I)	T	e
Neoseiulus	*bibens*	Et	L, Sh	C
	californicus	Pa, Na, Nt	T, L, V	C, A
	fallacis	Pa, Na, Nt, Au (I), Or (I)	T, Sh, L	C, A, Im
	idaeus	Nt, Et (I)	Sh, L	C, Im
	longispinosus	Or, Aus	T, Sh, L	C, A
	tiki	Et	Sh, L	C
Type III				
Typhlodromus	*pyri*	Pa, Na	T, V, Sh	C, A
(Typhlodromus)	*exhilaratus*	Pa	T	C
Typhlodromus	*caudiglans*	Pa, Na, Au	T	C
(Anthoseius)	*doreenae*	Au	V	C
Metaseiulus	*arboreus*	Na	T	C
	citri	Na	T	C
	pomi	Na	T	C
Paraseiulus	*soleiger*	Pa, Na	T	C
Phytoseius	*macropilis*	Na	T	C
	spoofi[h]	Pa	T	C
Amblyseius	*andersoni*	Pa, Na	T	C, A
	eharai	Or	T	C
	swirskii	Pa	T	C
Neoseiulus	*barkeri*	Pa, Na, Or	G	A
	cucumeris	Pa, Na	L, G	C, A
	umbraticus	Pa, Na	L, T	e
Kampimodromus	*aberrans*	Pa, Na	T, V	C, A
Typhlodromalus	*aripo*	Nt, Et (I)	Sh	C, Im
	limonicus	Na, Nt	T, Sh	C
	manihoti	Nt, Et (I)	Sh	C, Im
Typhlodromips	*sessor*	Na	T	e
	newsami	Or	T	C, A
Iphiseius	*degenerans*	Pa, Et	T, Sh, G	C, A
Type IV				
Euseius	*addoensis*	Et	T	C
	elinae	Au	T	C
	finlandicus	Pa, Na	T	C
	fructicolus	Nt	T	C
	sojaensis	Or	T	C
	tularensis	Na	T	C
	stipulatus	Pa, Na (I)	T	C, Im
	victoriensis	Au	T	C

[a]Use of genus and subgenus names follow Chant & McMurtry (23) for the subfamilies Typhlodrominae and Phytoseiinae and de Moraes et al (49) for the Amblyseiinae.
[b]Pa, Palearctic; Et, Ethiopian; Or, Oriental; Au, Australian; Na, Nearctic; Nt, Neotropical; (I), introduced.
[c]L, low-growing; Sh, shrub; T, tree; V, grapevine; G, greenhouse.
[d]C, conservation; A, augmentation; Im, importation and establishment.

Generalist Phytoseiidae (type III and IV lifestyle) which feed on spider mites (Tetranychidae) have been shown to be key predators of some eriophyoid mite species, although none of phytoseiid mites have been specifically reared on a commercial scale to control eriophyoid mites (VanLeeuwen 2010). For instance, the phytoseiid mite, *Cydnoseius negevi* has been demonstrated to control the fig mite, *Eriophyes ficus* and tetranychid mite, *Tetranychus urticae* under laboratory conditions. This study also demonstrated that *C. negevi* preferentially predates *E. ficus* over *T. urticae* (Abou-Awad *et al.* 1998). Two phytoseiid mites, *Neoseiulus paspalivorus* and *Neoseiulus baraki* have also been found to be the most abundant predatory mite species associated with the eriophyoid coconut mite, *Aceria guerreronis* (Lawson-Balagbo *et al.* 2007; Negloh *et al.* 2008). However, the behaviour of generalist phytoseiid mites and therefore their efficacy as biological controls agents may be highly influenced by leaf anatomy. Many phytoseiid mite species have been found to be localised on the underside of leaves, along the midrib or among leaf hairs and other protected areas where phytoseiids commonly lay eggs, thereby limiting their efficacy on other plant structures (McMurty and Croft 1997). In addition, the abundance of some phytoseiids such as *Amblyseius aberrans* (Oud.), *Typhlodromus pyri* Scheuten and *Amblyseius andersoni* (Chant) may be more influenced by the presence of sheltered habitats including leaf pubescence rather than food availability (Duso 1992).

A range of predatory mites belonging to other taxonomic groups are reported to be suitable as biological control agents of eriophyoid mites. For instance, *Zetzellia mali* Ewing, *Agistemus floridanus* Gonzalez and *Agistemus industani* Gonzalis (Prostigmata: Stigmaeidae) have been recognized as common predators of citrus rust mite *Phyllocoptruta oleivora* and apple rust mite *Aculus schlechtendali* respectively (Childers et al. 2001; Duso et al. 2008). Furthermore, *Homeopronematus anconai* (Baker) (Prostigmata: Tydeidae) has been found to reproduce very quickly and decrease the population of the tomato rust mite, *Aculops lycopersici* in Japan, although impact of the *H. anconai* (Baker) remains a matter of debate (Kawai and Haque 2004). In Northern Ireland, whirligig mite *Anystis baccarum* (Linnaeus) (Prostigmata: Anystidae) is the most abundant predator of *A. schlechtendali* (Cuthbertson and Murchie, 2006). Hence, the preservation of endemic mite fauna and the inclusion of selective pesticides in IPM programs is a fundamental requirement if significant populations of natural enemies are to be maintained and enable reduction in acaricide and other agrochemicals used with agricultural settings (VanLeeuwen 2010).

Other than predatory mites or other natural enemies, diseases can also cause a significant reduction of the mite populations (VanLeeuwen 2010). Entomopathogenic fungi, *Hirsutella thompsonii* were studied for the biological control of the coconut mite, *Aceria guerreronis* in India (Kumar and Singh, 2008) and the pathogenicity of *Beauveria bassiana* was tested on citrus mite, *Phyllocoptruta oleivora* in Brazil (Alves *et al.* 2005). Kumar and Singh (2008) have found that by combining the mycelial application of *H. thompsonii* with the adjuvants glycerol, malt extract broth or yeast extract powder can reduce the mite population significantly. Alves *et al.* (2005) reported that the lowest concentration ($1x10^6$ conidia/ml) of the *B. bassiana* based mycopesticide ''Naturalis L'' caused 24% mortality in 5 days after inoculation. Intermediate concentrations ($5x10^6$ and $1x10^7$ conidia/ml) and the highest concentration ($5x10^7$ and $1x10^8$ conidia/ml) resulted in 60% and 80–90% mortality respectively. Intermediate concentrations ($5x10^6$ and $1x10^7$ conidia/ml) were suggested the most economical because more applications could be carried out at the same cost of a single application with a higher concentration. *Hirsutella thompsonii* is the most common fungus found on eriophyoids (McCoy 1996), but, the availability of commercial strains may be restricted in some countries such as India (Faria and Wraight, 2007). However, commercial strains of *B. bassiana* are marketed worldwide (Faria and Wraight, 2007), therefore, the study of Alves *et al.* (2005) could be useful for eriophyoid pest management.

Several studies have proven blackberry plantings to be rich in beneficial mite fauna. Acarophagous mites from families Phytoseiidae, Tydeidae and Stigmaeidae have been found to be prevalent in blackberry crops (Ferreira *et al.*, 2015). Furthermore, insect species from families such as Cecidomyidae, Coccinellidae, Anthocoridae, Tripidae, Aeolothripidae and Phlaeothripidae have also been detected in blackberry crops and may play an important role in the reduction of RBM densities (Szendrey *et al.* 2003). Despite this, there are no known RBM specific predatory mites or beneficial insects thought to be capable of managing RBM (De Lillo E and Duso; Murrietta and Gaskell, 2015). Generalist predatory mites and beneficial insects that exist on blackberries could potentially control RBM, although long term evaluation is still required (Murrietta and Gaskell, 2015). Phytoseiid mites have been suggested as possible RBM predators since phytoseiid mites are the major predators of other eriophyoid mite species (Hanson, 1933; Lindquist, 1996). Damage assessment is fundamental to establish eriophyoid mite threshold levels, however, only limited research has been reported in this area (Lindquist *et al.* 1996). Once the economic threshold of RBM is determined, further studies are required to evaluate the ability of phytoseiid mites to control RBM below economic thresholds and crop management practices which optimise their population size in blackberry crops.

1.6.3 Chemical control

Eriophyoid mites are susceptible to most commonly used acaricides (Castagnoli and Oldfield, 1996). However, the main problem with chemical control of some eriophyoid species is their cryptic lifestyle, which limits their exposure to chemical controls and therefore their efficacy (Van Leeuwen *et al.* 2010). For instance, RBM hide in the small crevices between the drupelets making accurate timing of acaricide applications critical to access the life stages that leave the microhabitats to move onto the ripening fruit (Van Leeuwen *et al.* 2010). A previous study has shown that application of endosulfan to commercial a blackberry crop in late April-early May with two more sprays before flowering was effective to control RBM (Alford, 1979). However, this organochlorine insecticide and acaricide was withdrawn from the market over ten years ago, and no effective replacement treatment has been identified (Cross *et al.* 2012).

In a recent study by Cross *et al.* (2012), five acaricide treatments were evaluated to control RBM in four different blackberry varieties (Loch Tay, Carmel, Chester Thornless and Loch Ness). These were: 1) a full-season fortnightly spray programme of sulphur from bud burst; 2) a full season fortnightly programme of sprays of Codacide oil (rape seed oil); 3) two sprays of Dynamec (active ingredient: abamectin) at the start of flowering which is repeated two weeks later; 4) a combination of treatments 1+3; 5) a combination of treatments 2+3. All the acaricide treatments performed equally well in reducing blackberry mite populations. However, none of the treatments eliminated RBM populations. This was attributed to RBMs being present under the bud scales and therefore not exposed to the spray treatments. From this study, other issues were also observed with several of the treatments, for example, early application of Codacide when RBM are still under the bud scales, may be of little benefit, although it appears to be the best for growers due to its economic and environmental benefits upon biodiversity. Furthermore, sulphur spraying leaves unsightly deposits on the leaves and fruit which might be phytotoxic, hence, it is not recommended especially during fruiting. Dynamec is also believed to be lethal to predatory mite fauna including predatory phytoseiid mites (Cross *et al.* 2012).

Using acaricides to control eriophyoid mites could also have detrimental side effects on predator mites and other beneficial insects (Cross *et al.* 2012). Sublethal effects of abamectin have also been demonstrated on two different phytoseiid species, *Phytoseius plumifer* and *Neoseiulus longispinosus* with cumulative effects on adult female longevity, fecundity and fertility (Ibrahim and Yee, 2000, Hamedi *et al.*, 2011). Indeed, acaricides other than abamectin, such as etoxazole, spiromesifen, fenpyroximate, bifenazate, and acequinocyl have also been demonstrated to have sublethal effects on the Western predatory mite, *Galendromus occidentalis* used to control several pestiferous *Tetranychidae* species (Irigaray and Zalom 2006).

1.7 Conclusions and research outlines

RBM has been implicated in causing red berry disease in blackberries and quantifying their populations is an important aspect of their control in a successful integrated pest management (IPM) program. Two extraction methods of eriophyoid mites have been developed but these methods have limitations and might not be applicable for growers in quantifying mite populations. The sticky tape method (Harvey and Martin 1988; Davies *et al.* 2002) is time-consuming and unsuitable to be used in the field whereas equipment that is used in the shake and wash method (Monfreda *et al.* 2007) is expensive even though this method could be more efficient and accurate in extracting mite populations. Potential biological agents, including phytoseiid mites have been suggested to control RBM, however, whether they are effective and what impact the acaricides used to control RBM have on these predators is yet to be determined. Furthermore, there is still no conclusive proof RBM causes red berry disease (Cross *et al.* 2012). Therefore, the relationship between RBM populations, potential predator populations and the incidence of red berry disease needs to be determined as does the incidence across differing blackberry cultivars and management regimes.

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Appendix 10

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Sampling, extraction and incidence of redberry mites (*Acalitus essigi*) on blackberries in Australia

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Abstract
'Redberry disease' on blackberries is hypothesised to be caused by the redberry mite (RBM), *Acalitus essigi* (Hassan), and results in uneven ripening of blackberry drupelets, which become bright red, hard and inedible. This damage has been reported to cause significant crop losses in commercial blackberry production in most regions where commercial blackberries are grown. However, RBM are difficult to detect and manage due to their tiny body size. In this study, a new 'shake and wash' extraction method has been developed, enabling faster, more accurate mite detection and quantification. The 'shake and wash' method extracted significantly more RBM and predatory mites (Phytoseiidae) than the previously recommended 'sticky tape' method, where mite extraction using this technique was increased by 53 and 60%, respectively. RBM counts were then made from the fruit of wild and commercial blackberry cultivars. Significantly higher RBM populations were isolated in cultivars 'BL454' (mean = 12.1 per fruit) and 'Chester' (mean = 2.6 per fruit) from several sites indicating potential RBM susceptibility in these cultivars. The highest levels of disease incidence and RBM numbers were observed on wild blackberry fruit. The redberry disease incidence increased from 13.5 to 44.9% as the mean population of RBM increased on wild fruit. Further methods were developed to extract mites from winter buds on canes. RBM numbers were lower in the fruit compared to winter buds. RBM detection is best achieved in winter buds rather than fruit and may be an important tool for RBM detection and subsequent management in the cropping season.

Keywords Blackberry cultivars · *Acalitus essigi* · Sampling methods · Predatory mite · Mite detection

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Introduction

Commercial blackberry production has expanded globally due to increased consumer demand (Strik et al. 2007). In Australia, blackberry production volumes have doubled over recent years with the value of production increasing from \$40 million in 2013 to more than \$50 million in 2014/2015 (Mulderij 2016). This increase in demand for blackberries is due to several factors including improved cultivars, expanded marketing by production companies and an overall increase in berry consumption as fresh fruit (Clark and Finn 2014). However, redberry disease – that results in uneven ripening of a proportion of the blackberry drupelets, which become bright red, hard and inedible – has been highlighted as a cause of significant crop losses in commercial blackberry production (Amrine and Stasny 1994; Cross et al. 2012; de Lillo and Duso 1996; Gerding 1992). For instance, in 2010, 30% crop losses in commercial blackberries were observed in the UK due to redberry disease (Cross et al. 2012).

Redberry disease is hypothesised to be caused by the redberry mite (RBM), *Acalitus essigi* (Hassan) (Trombidiformes: Eriophyidae), either via an RBM-vectored plant virus or through the mite's feeding process (MacFarlane et al. 2015). Despite the plant virus hypothesis being explored, no mite-vectored plant pathogen has yet been identified although the possibility remains with some studies showing the redberry disease symptoms persisting after the successful treatment of RBM (Cross et al. 2012). The hypothesised damage induced by feeding is believed to be caused by the mite's stylets penetrating the developing blackberry drupelets and the injection of toxic saliva (Essig 1925; Szendrey et al. 2003).

Redberry mites reside among the bracts, fruits, bud scales and leaf axils of blackberry plants (Davies et al. 2001a, 2002). As the fruiting season progresses, RBM actively move from the fruit pedicel onto the berry as the fruit colour changes from green to red (Davies et al. 2001a). RBM was first identified in Australia in 1976 on raspberry, and in 1994 on both wild blackberry and commercial blackberries (Knihinicki and Boczek 2009). Previous studies have suggested that RBM damage is greater in later-fruiting blackberry cultivars than early-season-fruiting cultivars (Ferreira et al. 2015). Current management of RBM involves the regular application of acaricides which are not compatible with integrated pest management (IPM) programs due to potential impacts on predatory fauna. To date, no predators for RBM have been identified. A means of predicting the RBM populations prior to the onset of damage is important to any future IPM strategy as forecasting of RBM levels will enable better planning for pest control (Prasad and Prabhakar 2012).

Due to their tiny body size (82–350 µm), eriophyoid mites including RBM are difficult to detect and manage (Monfreda et al. 2010). Furthermore, RBM can be very patchy in the crop (Davies et al. 2001a), making the determination of sample sizes sufficient to detect RBM populations within commercial settings problematic. Few techniques exist for sampling eriophyoid mite populations, all of which are time-consuming and might not be suitable for grower use. Two main techniques are commonly employed to extract mites from complex fruit structures such as berries: the 'shake and wash' technique and the 'sticky tape' method. The 'shake and wash' technique has not been used on blackberries nor to extract RBM but involves the fruit being submerged and shaken in either ethanol (Zacharda et al. 1988), a detergent solution (de Lillo 2001), or the combination of bleach and Tween 80 (Monfreda et al. 2007) for a few seconds or minutes to extract the eriophyoid mites prior to separation either by separation funnel (Zacharda et al. 1988) or sieving (de Lillo 2001; Monfreda et al. 2007) and subsequent identification and quantification. Although the use of ethanol or detergent does enable the extraction of mites from complex structures such as berry drupes, their separation from these solutions can be problematic due to the need for specialised equipment or, in the case of sieving, may lead to a loss of extracted samples if they adhere to the sieve's mesh. The 'sticky tape' method was developed for extracting populations of RBM from blackberry fruit (Davies et al. 2002). However, this method is time consuming, as it takes approximately 3 weeks for the mites to be extracted (Davies et al. 2002). Faster, grower friendly methods need to be developed for extracting and quantifying eriophyoid mites from blackberries to enable timely and effective integrated pest control methods to be developed.

The aims in this study are: (i) to develop a better extraction and detection method for RBM on blackberry and associated plant structures; (ii) to determine susceptible blackberry cultivars to RBM; (iii) to determine whether there is a relationship between RBM populations and the levels of redberry disease on blackberry fruit; and (iv) to determine the sample size needed to detect RBM and test whether there is a relationship between winter bud RBM population levels and the blackberry fruit RBM population sizes in the previous season.

Materials and methods

Industry phone survey

A series of phone interviews were conducted to gather information on the prevalence of RBM in Australian commercial blackberry crops and management practices currently used by commercial blackberry growers that could impact on levels of RBM infestation. In total, 27 commercial blackberry growers, agronomists and industry representatives from around Australia were interviewed. The survey questions included grower demographics, perceived RBM and redberry disease incidence, production systems, crop sizes, and crop protection practices. The information from the phone surveys was undertaken to develop a greater understanding of improvements that could be made to current RBM management with an emphasis on the further development of IPM practices that could be used. These interviews were conducted with consent from the University of Tasmania's Social Sciences Human Research Ethics Committee ref no. H0017182.

Sampling of blackberry fruit across Australian production systems

To determine the prevalence of RBM and potential beneficial mite fauna in both commercial cultivars and wild blackberry fruit, fruit sampling was conducted from various locations around Australia's major berry growing regions. Blackberry material used in this study was collected from 19 sites around Australia with nine sites from Tasmania, eight sites from Victoria, one site from New South Wales and one site from Queensland. Of these blackberry samples, 16 were collected from commercial blackberry production sites and three from wild blackberries (*Rubus fruticosus* aggregate) found at roadsides (Table 1). Sampling was conducted from 1st February 2018 to 2nd May 2018 and 10th January 2019 to 10th April 2019. All commercially sourced fruits were supplied by growers who were interviewed in the phone surveys.

Fruit sampling was conducted by collecting at random 40 red, unripe blackberry fruits of each cultivar at each site. Whenever possible 10 fruits were collected from four

Table 1 Australian sample sites in Tasmania, Victoria, New South Wales and Queensland, where blackberry fruit were collected from

State	Locations	Production	Latitude	Longitude
Tasmania	Dunorlan Rd, Dunorlan	Commercial and wild	– 41.488630	146.5585
	Hillwood Rd, Hillwood	Commercial and wild	– 41.245337	147.0021
	College Road, Sandy Bay	Wild	– 42.907012	147.3242
	Redhills	Commercial	– 41.532398	146.5784
	Proctors Road, Dynnyrne	Wild	– 42.945638	147.3128
	Christmas Hills, Elizabeth Town	Commercial and wild	– 41.481317	146.5951
	Exton Road, Exton	Commercial	– 41.546103	146.7258
	Gordon River Road, Westerway	Commercial	– 42.674951	146.7877
	Wolfes Road, Neika	Wild	– 42.948863	147.2369
Victoria	Parker Rd, Silvan 1	Commercial	– 37.824976	145.4418
	Parker Rd, Silvan 2	Commercial	– 37.827556	145.4392
	Eacotts Rd, Hoddles Creek	Commercial	– 37.854923	145.5702
	Tooheys Rd, Pheasant Creek	Commercial	– 37.482566	145.3027
	Montrose, Yarra Valley	Commercial	– 37.802627	145.3592
	Hoddles Creek 1	Commercial	– 37.856044	145.5991
	Hoddles Creek 2	Commercial	– 37.857744	145.5983
	Monbulk-Seville Rd, Wandin East	Commercial	– 37.825636	145.4522
New South Wales	Range Rd, Dirty Creek	Commercial	– 30.006467	153.0946
Queensland	Border road, Applethorpe	Commercial	– 28.619032	151.9653

independent bushes located at least 10 m apart. If there were insufficient fruit numbers available at the time of sampling the 40 fruits were collected across more than four bushes. Red rather than ripe blackberry fruits were chosen as RBM is found mostly in the red, unripe fruits (Davies et al. 2001a, b). Fruits were harvested by using secateurs to remove both the pedicel along with the fruit as the pedicel is a known refuge of RBM (Davies et al. 2001b). All fruit from each bush were placed into a labelled zip-lock sandwich bag and stored in a cooler with icepacks for transport. In total, 11 blackberry cultivars from commercial blackberry production sites and untreated wild blackberry fruits were assessed, the details of sample collection are shown in Table 1 and within the Online Resource. The cultivars assessed included the non-patented cultivars 'Karaka' (n = 40, 1 site), 'Apache' (n = 40, 1 site), 'Chester' (n = 480, 7 sites), 'Thornfree' (n = 40, 1 site), 'Loch Ness' (n = 145, 3 sites), 'Ouachita' (n = 116, 3 sites) and 'Navaho' (n = 74, 2 sites), and the Driscoll's patented cultivars 'Cowles' (n = 240, 6 sites), 'BL454' (n = 440, 6 sites), 'Victoria' (n = 40, 1 site) and 'Elvira' (n = 160, 4 sites). Untreated wild blackberry fruits (n = 245, 6 sites) from both on- and off-farm were used to aid in determining the impact current management practices, including farm hygiene, may have on mite populations in commercial settings. The 'sticky tape' method and 'shake and wash' method as outlined below were used to extract mites from fruit samples. Fruit sourced from Tasmania for mite extraction was processed at the University of Tasmania's entomology insect rearing room. Mite extraction from berries sourced from the Australian mainland were conducted at the Tasmanian Department of Primary Industries, Parks, Water and Environment (DPIPWE) quarantine facilities in New Town. Importation of non-treated blackberry fruits from the

Australian mainland was conducted under authority from the Tasmanian Chief Veterinary Officer, Approval Number SA 17–84.

Assessment of blackberry fruits with redberry disease symptom

Ripe blackberry fruit showing signs of redberry disease were also sampled to determine the relationship between RBM population and the disease incidence. However, redberry disease was only found in sufficient quantities in wild blackberries from the Proctors Road site, which were collected on 14th March and 27th April 2018 and 10th April 2019. The presence and percentage of redberry disease symptoms on fruit was recorded prior to mite extraction. The percentage of redberry disease symptoms in a fruit was determined as: $100 \times$ total no. affected drupelets/total no. drupelets.

Improving mite extraction methods

Two sampling methods currently developed for eriophyoid mites are the ‘sticky tape’ (Davies et al. 2002; Harvey and Martin 1988) and ‘shake and wash’ methods with the latter not previously used for RBM extraction (Monfreda et al. 2007; Pérez-Moreno and Moraza-Zorilla 1998; Zacharda et al. 1988). As the sticky tape method is time-consuming and unsuitable to be used in the field, a mite extraction method modified from previous shake and wash methods was developed and its efficacy assessed in extracting RBM and other mite fauna from both fruit and winter buds. Extracted mites that were not RBM were first grouped into the families; Eriophyidae, Phytoseiidae, Tydeidae, Acaridae, Tarsonemidae and Tetranychidae and specimens prepared onto a slide for genus identification (Krantz and Walter 2009).

Sticky tape method

Collected blackberry fruit were placed on a A4 size black cardboard with four strips upturned of sticky tape (dimension: 48×66 m; brand name: 3M; product number: 3MSCOTCHCLR2), with 5–7 fruits placed on each length of sticky tape. Each strip of tape was labelled with the information of sample location, collection date, row number (if available) and cultivar. An insect surface spray (trade name: Mortein Naturgard) containing 8.0 g/kg permethrin and 6.0 g/kg transfluthrin was applied evenly on each fruit to initiate adult RBM extraction from the fruit (Davies et al. 2002). Fruit samples were then transferred into a 25 °C incubation room for approximately 3–4 weeks until the fruit had completely dried. The dried fruit were then removed from the tape and the number of RBM and other mite species and thrips on the tape recorded under a dissecting microscope at $40\times$ magnification. To ensure no double counting of the mite fauna on the tape across different fields of view, a black 5×5 cm Perspex card with approximately 1×1 cm grid etched into its surface was put underneath the tape.

Modified shake and wash method

Wild blackberry fruit (site: Proctors Road, $n = 80$), cultivar ‘Elvira’ (site: Hillwood, $n = 40$), cultivar ‘BL454’ (site: Dunorlan and Wandin East, $n = 200$) and cultivar ‘Chester’ (site: Silvan and Westerway, $n = 200$) were assessed by this method. The shake and wash method was modified from Zacharda et al. (1988) where apple rust mite, *Aculus* *schlechtendali*, was extracted from infested plant material by immersing the plant material in 80–90% ethanol in a covered beaker and then shaken for a few seconds. Individual blackberry fruit were placed into a 27-ml capped glass vial (brand name: ProSciTech; product number: L6227) containing 2 ml of 70% ethanol. The vial was then capped and shaken gently for 30 s to promote mite extraction from the fruit. After 30 s, the fruit was removed from the vial and the ethanol was poured out on a 4-cm-diameter Petri dish. To ensure the maximum extraction of mites and other fauna from the fruit, the extraction procedure was repeated another 2×. To prevent double counting the black Perspex card described as above was placed underneath the Petri dish and mite counts were again conducted using a dissecting microscope under × 40 magnification.

To enable sample storage and preservation upon completion of mite counts, the ethanol was poured into another Petri dish containing a 5-cm-diameter piece of black filter paper and allowed to dry. After the ethanol evaporated, the filter papers were kept individually in a labelled 65 × 75 mm of resealable plastic Ziplock bags.

To determine what proportion of mites were removed from the blackberry fruit using the sticky tape method, 10 of the 40 dried fruits collected at each site and analysed using the sticky tape method were randomly selected for re-analysis using the shake and wash extraction method. The same shake and wash method was conducted as previously described for fresh fruit, but the dried fruits were only shaken and washed once instead of 3×.

Blackberry winter bud sampling

Winter bud sampling for mites was also conducted to determine whether mite populations in buds related to mite populations in fruit. Bud sampling was done in Tasmania at two commercial sites (Hillwood and Dunorlan) and two roadsides (Proctors Road and College Road) between 25th July 2018 and 28th August 2019. To better understand the variability within a fruit block, 40 winter buds of each cultivar were collected randomly from multiple canes at each site to increase the chance of sampling RBM infested buds. Bud samples were collected using scalpel and forceps where forceps were used to hold the bud and the scalpel used to remove the bud from the cane. Each bud sample was placed individually into a labelled plastic vial and again stored in a cooler for transport. The cultivars sampled from Hillwood (21st August 2018) were ‘Elvira’, ‘BL454’ and ‘Cowles’, whereas samples from Dunorlan (21st August 2018) were ‘Karaka’, ‘BL454’, ‘Cowles’, ‘Ouachita’ and ‘Elvira’. Wild blackberry (*R. fruticosus* agg.) bud samples were also collected from the commercial sites and roadsides (n = 240). The extraction method of mites from buds is outlined below.

Bud extraction method

Each bud was dissected in a Petri dish (4 cm diameter) that contained 1–2 ml of 70% ethanol, again using scalpel and forceps. Each bud in the ethanol was cut in half lengthwise and gently ‘swirled’ for 30 s using the forceps to extract and eventually kill the mites within the bud. Bud material was then removed from the Petri dish by using the fine forceps. The counting of mites and storage of specimens in this method was the same as in the shake and wash method as described above.

Statistical analysis

SPSS v.25 was used to analyse all data except for sampling estimates which was analysed with R v.2.15.3 (R Core Team 2017). All data was first subjected to tests for normality, with most data not normally distributed. Spearman's correlation test was used to determine the relationship between the two mite extraction methods and between RBM and predatory mites extracted within fruit samples. Wilcoxon signed rank tests were used to determine any significant difference in mite removal from the same fruit between the 'sticky tape' and 'shake and wash' methods. Chi-square tests with Monte Carlo simulation ($\alpha = 0.01$) were used to determine any difference in mite populations between blackberry cultivars. To determine whether winter buds can be used as a predictive tool for fruit level RBM numbers, and the relationship between RBM populations isolated from the previous summer fruit samples and the winter bud samples Spearman's correlation tests were used. Mann-Whitney U tests were conducted to test for difference in RBM populations between fruit and winter buds.

The sample sizes needed to detect mites in fruit/buds were estimated by calculating the probability that a single fruit/bud in a sample of fruits/buds has at least one mite present. Beta-binomial distribution was used using the dbetabinom function in the VGM library in R v.3.6. This is an extension of the binomial distribution in which the probability of infestation (μ) is given by the beta distribution, $\mu \sim B(\alpha, \beta)$. The beta-binomial probability was calculated by equation where n is the infested fruits being present in a sample of size, m:

$$P(n|m, \alpha, \beta) = \binom{m}{n} \frac{B(n + \alpha, m - n + \beta)}{B(\alpha, \beta)}.$$

Results

Survey of redberry disease incidence

According to the industry phone survey, commercial blackberry producers reported redberry disease incidence ranging from 0 to 70% across Australian blackberry production regions (Table 2). Some cultivars were considered more susceptible to redberry disease than others with cultivars 'BL454' and 'Chester' mentioned the most in the grower survey for susceptibility (Table 2). Furthermore, the closer the commercial plantings were to wild blackberries the higher the disease severity reported. No disease incidence was reported by growers on farms where the distance from wild blackberries was greater than 400 m. Farms that reported having wild blackberries within 5 m of commercial plantings reported having the highest disease incidence (Table 2).

Modified extraction method for better detection of RBM and other mite species

A total of 435 dried blackberry fruit which had RBM analysed using the 'sticky tape' method were re-sampled by the 'shake and wash' method to test the efficiency of mite extraction methods. The 'shake and wash' method extracted significantly more RBM (53%) (Wilcoxon signed ranks test: $Z = -7.137$, $df = 434$, $p < 0.001$) and predatory mites

Table 2 Incidence of redberry disease (%) and the susceptible cultivars in commercial blackberry productions from industry survey

Grower	Susceptible cultivars	Redberry disease incidence (%)	Distance from wild blackberries (m)	Production system
C	None	0	None	Polytunnel
N	None	0	None	Polytunnel
Y	None	0	800	In ground (organic)
M	None	0	400	Polytunnel
H	None	0	50	Polytunnel
U	Chester, Loch Ness	1–2	50	In ground
K	BL454	1–5	25	Polytunnel
D	Chester	5–10	50–100	In ground
J	BL454, Ouachita, Navaho	5–15	10	Polytunnel
Z	Bl454, Chester	0–35	10–200	Polytunnel
P	Unknown trailing	50	20	In ground
X	Chester, Loch Ness	40–60	10	In ground
I	BL454, Chester	10–70	5	Polytunnel + soil
E	BL454, Chester	50	1	In ground

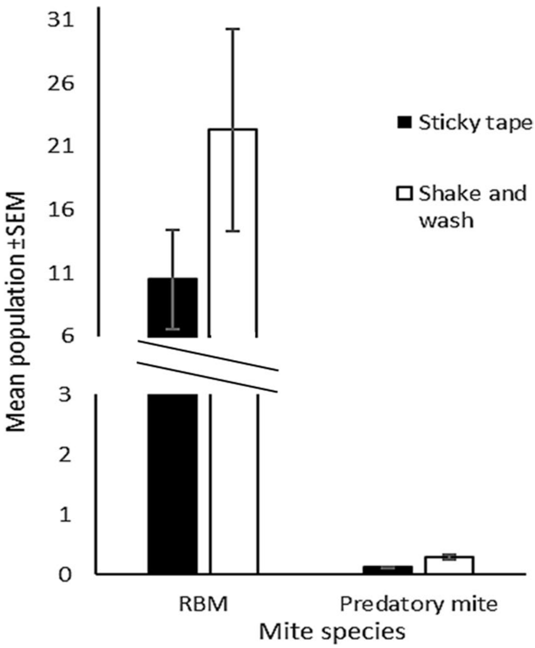


Fig. 1 Comparison of extracted mite population sizes (mean ± SEM; total n = 435) between 'sticky tape' and 'shake and wash' methods

(60%) ($Z = -4.066$, df = 434, $p < 0.001$) than the 'sticky tape' method (Fig. 1). Due to the increased efficiency of the 'shake and wash' method, RBM were also extracted from 8.5% of the fruit where no mites were extracted using the 'sticky tape' method alone with a mean of 2.8 extracted (range 1–7) from fruits where no mites where previously isolated.

Table 3 Taxa of other mite fauna found in blackberries

Family	Genus	Life habit
Phytoseiidae	*Typhlodromus* sp.	Predatory mite
Tydeidae		Neutral mite
Acaridae	*Tyrophagus* sp.	Neutral mite
Tarsonemidae	*Tarsonemus* sp.	Herbivorous
Tetranychidae	*Tetranychus* sp.	Herbivorous

Similarly, 3.3% of fruit where no predatory mites were extracted using the 'sticky tape' method were demonstrated to contain these mites when tested using the 'shake and wash' method (Fig. 1).

Mite fauna from families other than Eriophyidae were also found within the blackberry fruit (Table 3). Mites in the family Phytoseiidae are regarded as predatory mite species (McMurtry and Croft 1997). Mites in the families Tydeidae and Acaridae are considered as neutral mites in blackberry crops (Ferreira et al. 2015) whereas the mite families Tarsonemidae (broad mites) and Tetranychidae (spider mites) are mostly regarded as pestiferous in agricultural crops (Ferreira et al. 2015; Vincent et al. 2010). The identified genera for the Phytoseiidae, Acaridae and Tetranychidae were *Typhlodromus* sp., *Tyrophagus* sp. and *Tetranychus* sp., respectively (Table 3). The Tarsonemidae (broad mites) isolated were not the pest species known to infest blackberry crops (*Polyphagotarsonemus latus*) (Johnson et al. 2015) but were from a different genus (*Tarsonemus* sp.).

Differences in RBM and predatory mite populations on fruit between blackberry cultivars

The populations of RBM ($\chi^2 = 113.223$, df = 10, $p < 0.001$) and phytoseiid mites ($\chi^2 = 69.89$, df = 10, $p < 0.001$) were both different between blackberry cultivars. Wild blackberries from all 6 sample sites (n = 205) were infested with high numbers of RBM (Table 4). The proportion of infested fruit from each site ranged from 7.5 to 100%. Cultivar 'BL454' from 3 sample sites out of 6 (n = 245) also had high populations of RBM (mean = 12.1 ± 7.0, max = 534) with the proportion of infested fruit ranging from 0–38%. Cultivars 'Chester', 'Karaka', 'Ouachita' and 'Thornfree' had moderate to very low populations of RBM observed from single sites. No significant difference in RBM population size was observed between these four cultivars (Bonferroni adjustment Z-test: $Z = -1.48$, df = 10, $p = 0.13$). Hence, cultivar 'BL454' appears to be the most susceptible cultivar to RBM currently grown in Australia. A positive correlation was observed between the presence of RBM and predatory mites ($r = 0.161$, $p < 0.001$; n = 1616 fruits). Cultivar 'Karaka', the only early season fruiting cultivar that was assessed in this study had the highest mean populations of predatory mites (mean = 0.53 ± 0.15, max = 4) compared to other cultivars (Table 4), which is probably due to the management practices on this particular farm that included the use of commercially sourced predatory mites early in the season for the control of two-spotted mite, broad mite and western flower thrips.

There was a positive correlation between RBM populations and the level of redberry disease symptoms in wild fruit at Proctors Road (Spearman's correlation: $r = 0.561$, $p < 0.001$, n = 100). The redberry disease incidence increased from 13.5 ± 2 to 44.9 ± 4%

Table 4 Mean population size (number of mites per fruit) of redberry mite (RBM) and predatory mites between blackberry cultivars

Blackberry cultivar	RBM	Predatory mites
Wild	26.81a	0.3hk
BL454	12.08b	0.25hi
Chester	2.57c	0fg
Karaka	0.025cd	0.53k
Ouachita	0.017cde	0.14ij
Thornfree	0.098cd	0abcdefg
Apache	0cdef	0.075abcdefghij
Cowles	0f	0.075dej
Elvira	0f	0.038abcde
Loch Ness	0def	0.007bg
Navaho	0cdef	0.095ceij
Victoria	0cdef	0abcdefg
P	<0.001	<0.001

Means within a column followed by the same letter are not significantly different (χ^2 tests with Monte Carlo simulation: P > 0.05)

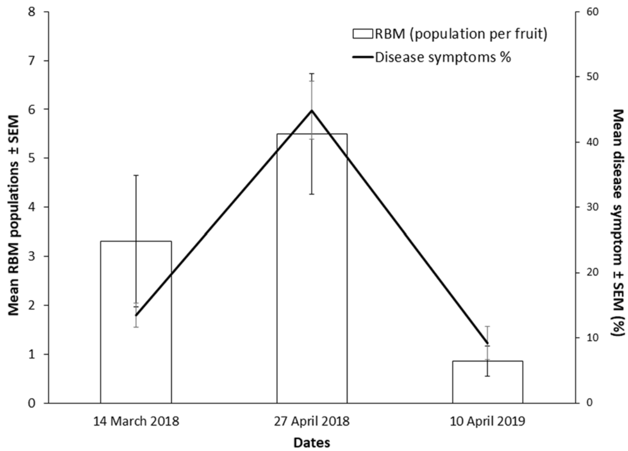


Fig. 2 Mean (± SEM) population size of redberry mite (RBM) in wild (*Rubus fruticosus* aggregate) blackberry fruit and the correlation between RBM and redberry disease of wild blackberry fruit on three dates (n = 40, 20 and 40, respectively)

and then dramatically decreased to 9.3 ± 2.5% as the mean population of RBM increased from 3.3 (± 1.4) to 5.5 (± 1.2) and then decreased to 0.9 (± 0.3) (Fig. 2).

Winter bud sampling

Higher absolute numbers of RBM were observed in winter buds compared to the previous season's fruit across cultivars 'BL454' (Mann–Whitney U: Z = − 6.85, p < 0.001), 'Chester' (Z = − 6.27, p < 0.001), 'Karaka' (Z = − 2.24, p = 0.025) and 'Ouachita' (Z = − 2.04, p = 0.042) and wild blackberries (Z = − 7.63, p < 0.001) (Fig. 3). However, there was no

RBM observed in both the fruit and winter buds in cultivars ‘Cowles’ and ‘Elvira’. A positive correlation exists between the populations of RBM within previous-season fruit and the following season’s winter buds (Spearman’s correlation: $r=0.396$, $n=418$, $p<0.001$).

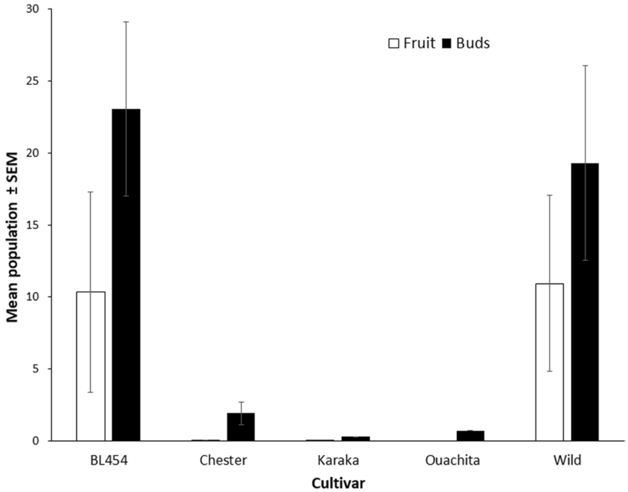


Fig. 3 Comparison of mean (±SEM) population size of redberry mite (RBM) between fruit and bud samples within blackberry cultivars ‘BL454’, ‘Chester’, ‘Karaka’, ‘Ouachita’ and wild blackberries. Sample sizes: n=120, 74, 40, 40 and 108, respectively, for each cv. and wild (*Rubus fruticosus* aggregate) blackberry fruit samples; n=90, 40, 40, 40 and 160, respectively, for each cv. and wild blackberry bud samples

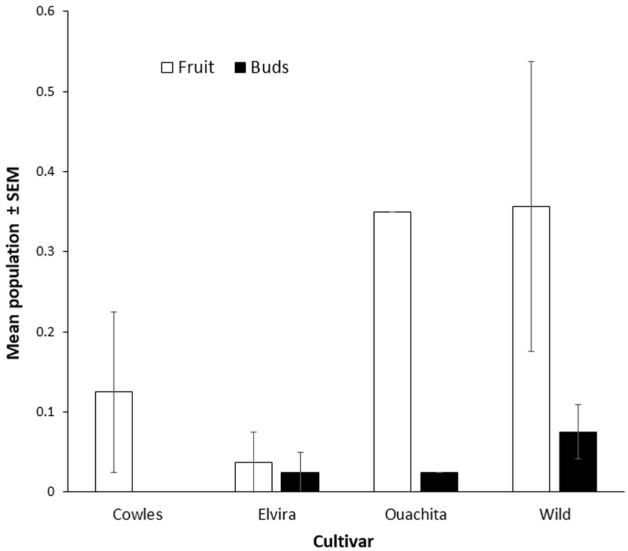


Fig. 4 Comparison of mean (±SEM) population size of predatory mites between fruit and bud samples within blackberry cultivars (‘Cowles’, ‘Elvira’, ‘Ouachita’) and wild (*Rubus fruticosus* aggregate) blackberries. Sample sizes: n=80, 80, 40 and 108, respectively, for each cv. and wild blackberry fruit samples; n=80, 80, 40 and 160, respectively, for each cv. and wild blackberry bud samples

Fig. 5 Sample estimates (probability of detection vs. sample size needed) to detect both redberry mite (RBM) and predatory mites (Phytoseiidae) in commercial (BL454) and wild (*Rubus fruticosus* aggregate) blackberry fruit and winter bud samples. Arrows indicate relative sample sizes required for detection probabilities of either 50, 70 or 80%

Predatory mites showed a decline in number from fruit to winter buds across cultivars ‘Cowles’ (Mann–Whitney U: $Z = -2.89$, $p = 0.004$), ‘Elvira’ ($Z = -2.70$, $p = 0.007$), ‘Ouachita’ ($Z = -2.72$, $p = 0.007$) and wild blackberries ($Z = -5.07$, $p < 0.001$) (Fig. 4). However, no significant difference was observed for predatory mite populations between the fruit and winter buds in cultivars ‘BL454’ ($Z = -1.31$, $p = 0.19$), ‘Chester’ ($Z = -1.06$, $p = 0.29$) and no predatory mites were observed in both fruit and buds of cultivar ‘Karaka’.

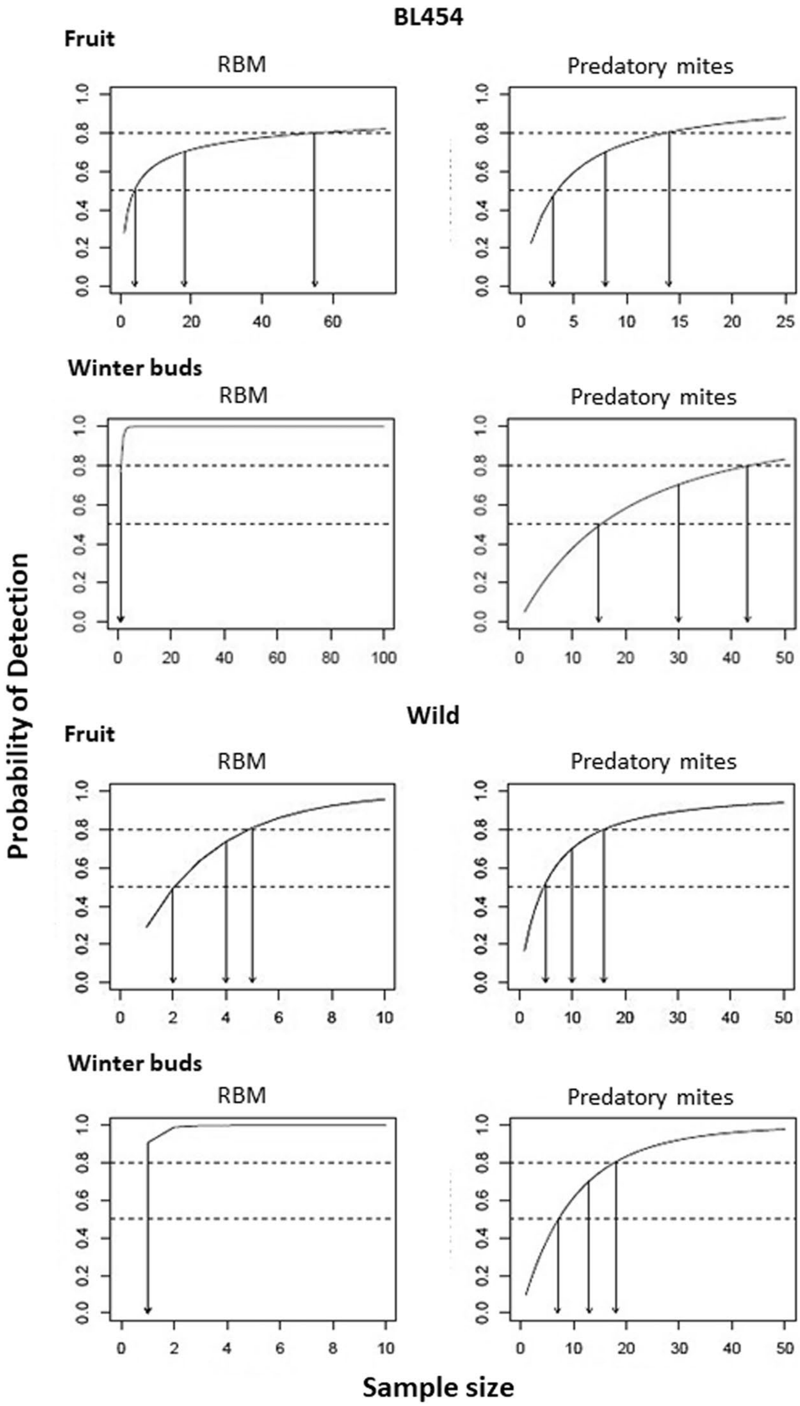
Sample sizes needed to detect RBM and predatory mites

Winter bud sampling requires a far smaller sample size than fruit sampling to detect RBM in both cultivar ‘BL454’ and wild blackberries (Fig. 5). The chance of 50, 70 and 80% detection for RBM in the fruit of cultivar ‘BL454’ require 4, 18 and 55 fruits, respectively, whereas wild blackberries require just 2, 4 and 5 fruits, respectively. Only one winter bud was required to achieve 80% chance of RBM detection in both cultivar ‘BL454’ and wild blackberries (Fig. 5). For the detection of predatory mites in cultivar ‘BL454’, the chance of 50, 70 and 80% detection in fruit require 3, 8 and 14 fruit, respectively, whereas winter bud sampling requires 15, 30 and 43 buds, respectively. Similarly, the chance of 50, 70 and 80% detection for predatory mites in wild blackberries require 5, 10 and 16 fruits, respectively, whereas the number of winter buds requires either 7, 13 and 18 buds, respectively (Fig. 5).

Discussion

The ‘sticky tape’ method is the only published mite extraction method that has been reported for RBM detection (Davies et al. 2002). However, this method is slow, requiring approximately 3–4 weeks before mite counting can occur. In addition, the ‘sticky tape’ method did not successfully extract as many mites as the ‘shake and wash’ method, with 8.5% of fruit that had no RBM mites extracted using the sticky tape method having RBM extracted with the shake and wash method. One possible reason for this occurring is the insecticide spray killing the mites before they moved out from the fruit. The concept of the spray in the ‘sticky tape’ method was to initiate the mites leaving the fruit, which crawl onto the tape when they run out of food (i.e., fruit becomes dried) (Davies et al. 2002; Harvey and Martin 1988). However, the limited mobility and cryptic behaviour of RBM (Davies et al. 2001a) may lead to them staying within the fruit and dying there instead of crawling onto the tape.

Furthermore, fruit placed on the sticky tape were on occasion observed to become mouldy during drying at 25 °C, which prevented the quantification of RBM and other mite fauna from these samples. This was due to the drying temperature also being the optimum temperature for fungal growth (Robbins and Morrell 2002). Although either lower or higher temperatures could be utilised during the drying process these temperatures may further promote fungal growth or dry the fruit too quickly thereby reducing mite extraction


BL454
Fruit
RBM
Predatory mites
Winter buds
RBM
Predatory mites
Wild
Fruit
RBM
Predatory mites
Winter buds
RBM
Predatory mites
Probability of Detection
Sample size

efficiency. This slow drying process also may have amplified RBM numbers with some fruit having excessively high mite numbers (max = 1440), which were not apparent during sample collection. This may be due to the 3–4 weeks required to dry the fruit possibly enabling the mites to produce offspring on the fruit during the extraction period. However, the generation time of RBM is currently unknown and therefore warrants further research to determine whether this may indeed have been the cause for these elevated RBM numbers on some fruit.

For these reasons, the modified 'shake and wash' method was deemed more efficient than the 'sticky tape' method as the use of 70% ethanol in the 'shake and wash' method kills the mites quickly (Monfreda et al. 2010), and the shaking leads the mites to moving out from the fruit within a few minutes instead of weeks. Despite this, the 'sticky tape' method does appear to effectively enable the larger, more predatory phytoseiid mites to potentially leave the fruit due to their being more active or mobile than RBM, and therefore most likely to move onto the tape faster. Despite the efficiency of the sticky tape method for the predatory mites, the 'shake and wash' method still provided better extraction than the 'sticky tape' method, which could also be due to direct contact with the insecticide in the 'sticky tape' method killing some mites within the fruit before they had the opportunity to move onto the tape. Another benefit of the 'shake and wash' method is that the chance of RBM mite detection can be increased as higher mite populations can be extracted, hence, samples sizes required to determine population sizes within crops can be reduced. Despite these advantages, the use of ethanol does pose issues due to its lethality if the mites extracted are required for culturing or host specificity testing. It is therefore, recommended that if the extracted fauna is required for culturing or behavioural assays that detergent or a combination of Tween 80 and bleach as outlined in Monfreda et al. (2007) be utilised. However, if the specimens are required for taxonomic identification including DNA analysis or long-term storage as is standard practice in research settings, that ethanol be used to hasten sample processing by negating the need to sieve the samples from the extraction solution, which may lead to sample loss if specimens adhere to the sieve's mesh (SR Quarrell, pers. obs.).

To date, there is still no conclusive proof that RBM causes redberry disease. In this study, higher redberry disease incidence significantly correlated with higher RBM populations in wild blackberries. A previous study has demonstrated that RBM may disperse from wild blackberries to the farms via aerial dispersal during windy periods, which is the dominant dispersal mode of most eriophyoid mites (Michalska et al. 2010; Valenzano et al. 2019). Proximity to wild blackberries was also evident in our grower survey in relation to self-reported incidence of redberry disease. Hence, if growers have wild blackberries nearby their commercial blackberry plantings, they should also manage these blackberry plants, which where possible, should include the removal of these bushes. The cultivars 'BL454' and 'Chester' which were both highlighted by most growers as susceptible to RBM has been supported in this study. A previous study observed that RBM damage is greater in the later fruiting blackberries such as the cultivar 'Ouachita' compared to early season fruiting cultivars such as 'Karaka'. This increase in damage is believed to be associated with higher RBM population sizes later in the season coupled with the mite's dispersal prior to overwintering (Ferreira et al. 2015). All the cultivars assessed in this study were late-season fruiting cultivars, except for 'Karaka'. In this study, cultivar 'Karaka' (sourced from only one site) had very low RBM populations in the fruit and no historical damage of redberry disease. In addition, no RBM was observed in the late season fruiting cultivars 'Apache', 'Cowles', 'Elvira', 'Loch Ness', 'Navaho' and 'Victoria'. This might be due to either a preference by RBM for feeding on specific cultivars and/or differences in cane management. Primocane cultivars such as 'Elvira' are often managed by pruning the floricanes to ground level and have been observed as having lower levels of RBM (Murrietta and Gaskell 2016). However, further studies on the differences of chemical composition of potential attractants and deterrents between cultivars are required to determine if there are cultivar preferences by RBM.

Ongoing research has aimed at controlling eriophyoid mite species including RBM by biological control methods primarily with the conservation of predatory mites (Van Leeuwen et al. 2010). In this study, mite species from families of Phytoseiidae (predatory mites), Tydeidae (neutral mites), Acaridae (neutral mites), Tarsonemidae (broad mite) and Tetranychidae (spider mites) were found in blackberry crops across Australia. Although a positive correlation between RBM and predatory mites was observed, what role they might play in reducing RBM populations is unclear. Generalist Phytoseiidae not only feed on spider mites, but also prove to be key predators of eriophyoid mites (Van Leeuwen et al. 2010), though none have been specifically implicated in the predation of RBM. In this study, most of the predatory mite species were identified as *Typhlodromus* sp. which can be either a specialist or generalist predator and are known to predate on eriophyoid species including rust mites (McMurtry and Croft 1997). For instance, *Typhlodromus pyri* and *T. athiasae* predate the apple rust mite, *Aculus schlechtendali* and tomato rust mite, *Aculops lycopersici*, respectively (Van Leeuwen et al. 2010). In addition, some farms had introduced *Phytoseiulus persimilis* (specialist predator) and *Neoseiulus cucumeris* (generalist predator) to their blackberry crops, although they were mainly intended for control of spider mites and thrips (McMurtry and Croft 1997). It is therefore recommended that the phytoseiid fauna observed in Australian blackberry crops be identified to species level, to better understand their dietary ranges and that already commercially available phytoseiid mite species known to predate eriophyid mites such as *Typhlodromus occidentalis* (McMurtry and Croft 1997) or *Typhlodromips montdorensis* (Schicha 1979) be investigated for the biological control of RBM in Australian blackberry crops.

Fluctuations of RBM populations within the different microhabitats across the year are evident (Davies et al. 2001a). RBM populations decline in bracts and leaf axils from summer to winter. In spring, overwintering RBM populations within buds emerge from these overwintering sites and populations increase (Davies et al. 2001a). Therefore, detection of overwintering RBM colonies may prove important when developing pest management programs for the following fruiting season. In this study, higher populations of RBM were found in winter buds compared to the previous season's fruit samples, whereas very low populations of predatory mites were observed in winter buds. This is possibly due to the tiny body size of RBM (< 200 μm), which enables them to overwinter in locations where the larger predatory species may not have access such as in the buds. In addition, intensive chemical controls are mostly utilised on-farm during the cropping season, hence, winter buds might become the preferable shelter for RBM due to low or no chemical applications being able to penetrate the internal tissue within winter buds where these species are able to overwinter. For the susceptible cultivar 'BL454', RBM populations were approximately 70% higher in the winter buds compared the other cultivars assessed. Due to higher number of RBM populations residing in the winter buds of 'BL454' and wild blackberries, an 80% chance of RBM detection in the winter buds can be achieved with a sample size needed of only 1 bud, which is much lower (approximately 55 × and 5 × for 'BL454' and wild samples, respectively) than detection sample sizes needed from fruit. Therefore, winter bud assessment may be an important tool for RBM detection and management in both cultivated and wild blackberries during the cropping season. It should be noted that although some predatory mites are known to overwinter in buds (Murrietta and Gaskell

2016), others may overwinter in other plant organs and therefore their presence in buds should only be deemed as a course indicator of predatory mite abundance, not an absolute estimate of their population size. It is therefore also recommended that further research be conducted to better understand what proportion of predatory fauna do reside in buds over-winter and if those species residing in buds are dominated by a limited number of the total predatory fauna present within the crop to enable more accurate forecasting of predatory mite population sizes during the production season.

In conclusion, the 'sticky tape' method is not recommended for growers for mite detection because it is time-consuming, unsuitable for field use, and does not extract the majority of mites within plant organs being sampled, which can lead to false negative recordings. A further modified extraction method from that proposed by Zacharda et al. (1988) has been developed with more efficient mite detection. The 'shake and wash' method extracted significantly more RBM and predatory phytoseiid mites than the 'sticky tape' method. Smaller but representative sample sizes are required when using this method, which benefits growers or agronomists by saving time and crop scouting costs when conducting IPM monitoring programs. Therefore, it is recommended that the 'shake and wash' method be adopted for mite extraction and detection in complex fruits such as blackberry with the exclusion of the sieving step proposed in other studies to both allow more rapid mite sampling and the elimination of the expensive fine sieves needed to separate the mites from the extraction solution. Due to relatively high populations of RBM residing in wild blackberries and incidence of redberry disease shown in wild blackberries, growers should consider managing or removing wild blackberry bushes nearby the commercial blackberry plantings to protect their crops. Significant numbers of RBM were observed in cultivars 'BL454' and 'Chester', however, further studies investigating the chemical composition differences between susceptible and non-susceptible cultivars of blackberry are required. The presence of predatory phytoseiid mites may impact on the RBM populations but the roles of these mites on RBM populations remains unclear. Winter bud assessment may be an important tool for RBM detection and management during the cropping season as a better chance of RBM detection can be achieved in winter buds rather than fruit.

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