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Sustainable control of small hive beetle and varroa mite in Australia.

Abstract

Honeybees are under threat globally from a range of sources including pesticide use and a range of potentially devastating pests and diseases. This review focuses on two of these pests: the small hive beetle, which has been in Australia for almost a decade, and the varroa mite which has yet to arrive. In Western countries, the default method of controlling these pests has been the use of synthetic chemicals, however issues with contamination of hive products and the development of resistance to these substances necessitates the exploration of other strategies. The use of organic acids and essential oils, physically and cultural control techniques and breeding strategies are examined from an Australian perspective. In the long-term, the development of adaptations for the coexistence of honeybees with small hive beetle and varroa mite should be the ultimate goal, but the widespread use of chemical control methods will hinder this process.

Introduction

Worldwide, honeybees (*Apis mellifera* L.) are experiencing many different threats to ongoing colony health and stability. These include widespread use of synthetic pesticides on crops, diseases such as American Foul Brood, invasive populations of other bee species, various mite parasites, and other invertebrate pests of hives (see for example Stankus 2008). This review focuses on two pest arthropods that live in the hives of honeybees: one currently in Australia, the small hive beetle (*Aethina tumida* Murray) (hereafter referred to as SHB) and one as yet absent, the varroa mite (*Varroa destructor* Anderson and Trueman) (hereafter referred to as varroa).

Both pests, particularly the varroa mite, have caused damage to local honeybee populations, and widespread economic losses, when they have moved from their natural range. Besides the provision of honey, honeybees are critical to agriculture because of their pollination services, so it is critical that sustainable methods of control are found that will work safely and effectively into the long-term to prevent colony losses and reduce the economic impacts of these pests. Current and potential control methods for varroa and the SHB are here evaluated from an Australian perspective.

The Australian situation

The Australian honeybee industry is worth around $80 million per year in honey and wax production (RIRDC 2011), however the pollination services that honeybees provide are worth even more(Keogh *et al* 2010). Pollination by honeybees is critical to an estimated 65% of agricultural and horticultural crops grown in Australia, and boosts many others through increased yields. It is estimated that $1.8 billion worth of Australian agricultural commodities are to some degree reliant on honeybee pollination services (Keogh *et al* 2010). Thus, whilst any threats to the honeybee industry will impact those directly involved with bee keeping, there could be much wider flow-on effects to the horticulture industry through reduced pollination (Keogh *et al* 2010).

Pollination services are by no means limited to managed hives; Australia's extensive feral bee populations play a critical role. Whilst economic importance of feral bees to the horticulture industries has not been specifically assessed, it is likely to be vitally important (Cunningham 2010), and may well exceed that of managed hives (Keogh *et al* 2010). Thus in assessing any threats to honeybees in Australia, both managed hives and feral bees must be taken into account.

Being an island continent, Australia is able to maintain stricter quarantine measures than is possible in most areas of the world. These cannot be relied upon however, as the discovery of the SHB near Sydney in 2002 clearly demonstrates. Likewise, varroa has reached the local island state of New Zealand despite strict quarantine laws (Keogh *et al* 2010; Goodwin 2010). It is generally acknowledged that it is not a matter of if varroa reaches Australia, but when (for example Keogh *et al* 2010; Anderson 2010).

Since its arrival in Australia, SHB has spread rapidly and now occurs in NSW, Victoria, ACT, Queensland and the Kimberley region of WA (Annand 2011). Whilst the SHB prefers warm, humid conditions, its distribution on other continents suggests that it could live anywhere in Australia except the very arid interior (Annand 2011). Countrywide economic losses to apiarists through SHB infestations have been estimated at $4.5 million dollars per annum (Annand 2011). This figure does not include losses in other industries reliant on pollination, so the overall figure is likely to be even higher. There is some good news however, as it appear that SHB might not cause as much damage as was originally feared, for instance in the warm coastal areas of northern NSW, where the SHB is likely to be the most prolific, around 10% of hives are lost to SHB damage(Annand 2011). This compares favourably with the widespread damage caused by the introduction of the SHB to the US for example (Neumann & Elezen 2004). It is hard to quantify whether the introduction of SHB has had similar effects in Australia as the US however, as the in the US varroa became established before SHB - it is likely that the combination of varroa and SHB is worse than either alone.

Biology and lifecycle

Small hive beetle

The SHB is endemic to Sub-Saharan Africa where it co-exists with the local honeybee subspecies. It rarely causes damage except in already weakened and diseased colonies, and generally does not reproduce in healthy colonies (Ellis & Hepburn 2006). Ellis & Hepburn (2006) also suggest that it confers a positive benefit to local subspecies of bee as it acts as a scavenger, disposing of weak and abandoned hives that could otherwise be a reservoir for honeybee diseases. When introduced to a new area however, the SHB can have a deleterious effect on European subspecies of honeybee, as has been shown by its introduction to North America and Australia (Neumann & Elzen 2004; Annand 2011).

The morphology of the SHB makes it difficult for honey bees to remove them from the hive: they possess a thick, smooth exoskeleton; short legs and antennae; and retractable legs and head making it difficult for bees to grip, bite or sting them (Ellis & Hepburn 2006). They also exhibit behavioural adaptations to avoid capture by hiding in cracks and crevices (Ellis & Hepburn 2006).

In African subspecies of honeybees, SHB are generally confined by guard bees to corners and crevices at the periphery of the hive and are thus rarely able to feed directly from the honey or brood (Ellis & Hepburn 2006). This isolation is further facilitated by the creation of propolis (made by honeybees from tree resins and wax) structures in which guard bees confine SHB (Neumann *et al* 2001; Ellis *et al* 2003c). Despite this, SHB is able to survive in their captive state for many months by feeding trophallactically - guard bees regurgitate drops of honey directly into SHB mouths (Ellis *et al* 2002b). It is unclear by what mechanisms this occurs, although making antennal contact in mimicry of normal bee communication plays a role, it is possible that olfactory signals also play a part (Ellis & Hepburn 2006).

Confinement of the SHB to cracks in the hive has also been observed to occur in European honeybee hives, suggesting that this is not a specific adaptation to SHB by their original host but rather a generalised response to small nest invaders (Ellis & Hepburn 2006). It has also been found that the confinement/guard behaviour of Cape and European honeybees does not differ substantially and thus is unlikely to be the sole reason for the different tolerance levels (Ellis *et al* 2003c). There is, however, much more variation between hives in confinement and guarding behaviour in the European honeybees than African honeybees (Ellis *et al* 2003a).

Where possible, females SHB bite into the waxy caps of brood to oviposit directly on the bee pupae in the cell allowing the larvae to develop on a concentrated food source, but they can lay anywhere in the hive, particularly if disturbed (Ellis & Hepburn 2006).

Both adult and larval stages of the SHB feed on honey, pollen and growing bee brood. Once fully grown, the larvae move out of the hive and burrow into the surrounding soil to pupate, with the emerging adults re-entering the hive or dispersing (Ellis & Hepburn 2006). The SHB larvae are able to survive for more than six weeks without food whilst looking for a suitable site for pupation, however with suitable soil conditions most pupation occurs within 30 cm of the hive entrance (Spooner-Heart 2008).

Significantly, it has also been shown that the SHB is able to survive and reproduce outside of a bee colony, feeding on fungi, fallen fruits and other decaying matter (Arbogast *et al* 2010), although this has not yet been documented to occur under field conditions (Neumann & Elzen 2004). Whilst the SHB has the potential for a far greater rate of reproductive success within the environment of the colony than on alternative food sources (Ellis *et al* 2002a), the ability to survive on other food sources provides an adaptive advantage facilitating dispersal to distant hives, and allowing it to persist in the absence of a hive (Arbogast *et al* 2009b).

SHB locates hives through olfactory cues, and is particularly attracted to honeybee alarm pheromones (Ellis & Hepburn 2006).

SHB is a carrier of the yeast *Kodamaea ohmeri*, which inoculates hive pollen and honey stores when SHB larvae feed and defecation on combs (Ellis & Hepburn 2006). This leads to the fermentation, or 'sliming' of the honey making it unusable to both humans and bees (Aannand 2011). This fermentation also produces volatile compounds that mimic honeybee alarm signals, thus indicating to other local SHB that there is a good supply of pollen and honey (Torto *et al* 2007). Once the SHB population becomes too high in the hive, or the amount of sliming reaches a critical level, the hive is abandoned (Ellis & Hepburn 2005; Annand 2011)

SHB thus causes problems to bee colonies, and consequently apiarists, in a number of ways. As well as the direct ingestion of honey, pollen and growing brood by both adult and larval SHB, and the fermentation and subsequent abandonment of hives, losses also occur through the removal of brood cells with SHB eggs by worker bees (Ellis *et al* 2003b; Ellis & Hepburn 2006). It is also possible that the need to guard the beetles prevents some workers from foraging – a negative correlation has been found between the rate of incoming bees to the colony and the number of SHB in the colony (Ellis *et al* 2003a).

Furthermore, SHB can also act as a carrier of pathogenic viruses. Eyer *et al* (2009) demonstrated that SHB can transmit the deformed wing virus through feeding on infected brood, feeding on dead bees, trophallaxis, and making contact with contaminated wax. This possibly has more serious implications than virus transmission through varroa, as SHB can fly up to 16 km, enabling it to easily transmit viruses directly to other hives (Eyre *et al* 2009).

Varroa

The varroa mite is a parasite of the Asian honeybee, *A. ceranus*. It has jumped hosts to infect *A. mellifera* with devastating consequences and has spread to every continent of the world except Australia.

Varroa lives its whole life on bees or within the hive, being an obligate parasite feeding on haemolymph from both adult and developing bees. Female mites enter brood cells before they are capped and conceal themselves in the jelly at the bottom. Once the cell is sealed, the mite will lay an unfertilised egg, which develops into a male, and then two to four fertilised, female eggs. These hatch within 12 hours of laying, and nymphs feed on the pupating bee larvae (Donze & Guerin 1994). Before is brood is open and the adult bee emerges, the male will mate with his sisters. This inbreeding allows resistance to chemicals to spread rapidly (Floris *et al* 2001). When the adult bee emerges, the female mites, including the original, leave with it, and the male mite dies (Donze & Guerin 1994).

Varroa show a strong preference for drone brood cells, and will more successfully reproduce on these, but can also reproduce in worker cells of honeybees (Calderone 2005). On their original hosts, varroa are unable to reproduce on the worker brood as it is more closely tended and they are detected removed by worker bees (Donze & Guerin 1994). *A. ceranus* also keeps varroa under control through mutual cleaning and grooming behaviour that is rarely expressed in *A. mellifera* (Donze & Guerin 1994).

Varroa mites also spread viruses throughout honey bee colonies. These can be more damaging than the mites themselves. For instance, Martin (2001) found that quite low infestations of virus-infected mites can cause a colony to collapse over winter due to the death and disablement of healthy bees.

Parasitation by the varroa mite has been shown to cause learning deficits and behavioural changes in individual honeybees – in particular bees show a reduced ability to find their way back to the hive from foraging (Kralj *et al* 2007). This has been suggested as a way in which the varroa mite's spread is facilitated: 'drifting' bees have increased chances of arriving at the wrong colony and thus transmitting the mite. This may be an evolutionary advantage to bees too however; if heavily infected bees are less able to return to the hive, the total parasite load on the hive is reduced (Kralj *et al* 2007). Whether such behavioural changes are directly the result of the mite is hard to ascertain however – Iqbal & Mueller (2007) found that infection with deformed wing virus, commonly associated with varroa (Martin 2001), is linked to similar learning defects. This raises the intriguing idea that harbouring these viruses provides an evolutionary advantage to varroa to despite leading to increased colony mortality. This emphasises that varroa cannot be successfully analysed or controlled without recognising it as part of a mite-virus complex.

Implications for control

The SHB can be targeted at a number of stages in its life-cycle: free living adults and larvae within the hive; pupating larvae in the soil; dispersing adults outside of hives; and larvae and adults on frames that have been harvested. There are fewer options for targeting varroa as the mites are always within the hive. As varroa spends so much of its lifecycle within brood cells, controlling the mite only when it on the comb or adult bees would miss most individuals during most parts of the year. In cooler parts of the world, varroa is often successfully treated over winter when there are no brood cells present and all mites in a hive are attached to adult bees. Australian conditions mean that this window of opportunity is reduced, because there is less time in the year when brood is not present. Chemical control measures must be able to penetrate the brood cell to have an effect on the varroa populations if there is extensive brood present.

It is also important to acknowledge that both pests have achieved a stable coexistence with honeybees in their original habitat – the varroa with the Asian honeybee and the SHB with a subspecies of *A. mellifera.* Thus the adaptations these bees show towards the presence of these pests could be encouraged in the European honeybee.

Quarantine

The single most important strategy for controlling varroa in Australia is prevention. Varroa has yet to reach Australia and there are strict quarantine measures in place (Anderson 2010). Despite Australia's isolation and quarantine measures however, SHB entered and established the country, showing that these quarantine measures are far from failsafe.

Strategies to deal with an initial infestation of varroa would be different from strategies to deal with an established pest in the long-term. For instance, if a presence in Australia is detected early enough, it may be possible to eradicate the pest altogether if there are few colonies involved. In this case the complete destruction of hives, and even apiaries, and the use of strong synthetic chemicals might be justified, however these would not be effective or affordable strategies if a population becomes entrenched.

Because an Australian population of varroa is seen as inevitable, and that a population of SHB is already present, this piece focuses on strategies that might be appropriate for control of entrenched populations rather than initial quarantine breaches.

Possible control strategies

Chemical controls

A number of synthetic chemical controls have been used to combat varroa and SHB. For varroa, these include flumethrin, fluvalinate, and coumaphos (Gregorc & Planinc 2001; Bogdanov 2006). Chemicals may be applied in a number of ways including mists, trickling into the hive, impregnated strips and, in the case of SHB, drenches on the ground targeting the pupae stage. Whilst synthetic chemicals may be effective initially, there are a lot of problems associated with their use.

One of these problems is that beeswax and honey tend to absorb and store chemicals used in the hive. If chemical residues are present on hive products they become unsaleable, and they may also have implications for bee health. Because foundation for combs is repeatedly emptied and replaced into hives, chemicals used in the hives can build up over years in beeswax (Wallner 1999). These chemicals can then diffuse from the wax into the honey (Wallner 1999; Bogdanov 2006). One way to reduce the impacts of chemicals on hives would to be to halt the practice of repeated reuse of wax (Wallner 1999). This would lead to a reduction in honey production however, as more energy would need to be expended by bees in wax production.

Another problem with synthetic chemicals is safety - for bees, apiarists and the environment. The method of application can influence the safety of chemical use. For instance, while fipronil has been shown to be very toxic to bees and to negatively affect bee behaviour at sub-lethal doses (Levot 2008), confined to bee-proof traps for the control of SHB, it can be an effective control measure within hives. A test of this system found no detectable residues in the honey or effects on bees but the numbers of live SHB detected after six weeks had reduced by 96% compared with the untreated control traps (Levot 2008). This method of application could not be used with varroa however as the mite is constantly in contact with bees or brood.

Although the chemical must disperse throughout the hive for varroa control, there are ways in which this can be done more safely than by direct liquid application, particularly for the human operator. Strips impregnated with flumethrin, fluvalinate or coumaphos placed in the top of the hive are now one of the most common controls for varroa (Floris *et al* 2001), and are also used in SHB control Neumann & Hoffmann (2008).

The development of resistance is another problem with synthetic chemicals. Resistance has been noted for all the major synthetic chemicals used against varroa (Nasr 2010). The appropriate use of chemicals is an important consideration in preventing resistance. Using a range of chemicals, and rotating their use, may slow the development of resistance (Goodwin 2010). Also, Floris *et al* (2001) found far less resistance in varroa mites from apiaries using commercially prepared chemical strips than from apiaries where homemade chemical remedies are used, such as wood soaked in these chemicals and placed in the hives.

As well as resistance and residues, there are more fundamental issues associated with synthetic chemicals use. The use of chemicals to destroy varroa not only encourages resistance in mites, it also allows less well adapted colonies to survive. Fries and Carmazine (2001) suggest that current apicultural practices are responsible for maintaining virulent forms of the varroa mite as there is no chance for a benign host-parasite relationship to form. There are many instances where honeybee populations have been found to achieve coexist with varroa, and the adaptations allowing this coexistence to develop appear to be different in different areas, suggesting that they developed independently (Rosencrantz 1999; Fries & Camazine 2001). The potential for longer term coexistence thus exists for honeybee, but the process of developing this coexistence is being inhibited by the widespread use of chemicals.

Organic chemicals

The use of biological-based chemicals is a possible alternative to synthetic chemicals. The two main groups of these are essential oils and organic acids. Such chemicals may be more acceptable than synthetic chemicals to producers and consumers but have generally been found to be less effective than the standard chemical arsenal (Goodwin 2010).

Organic acids, such as formic acid, acetic acid and oxalic acid, have been investigated in the control of varroa. They exhibit many of the same problems as synthetic chemicals: they can be toxic to bees, dangerous to work with and can lead to resistance. They have also been found to be quite variable in their effectiveness against varroa (Eguaras *et al* 2001; Goodwin 2010). They are unlikely however to cause problems with residues in wax or honey – neither formic nor oxalic acid can be detected in hive products using these treatments, however there is a possibility that formic acid may change the taste of the honey if used long term (Bogdanov 2005). Also, these organic acids do not accumulate in wax even if present in honey (Gregorc & Planinc 2001), and formic acid is a natural component of honey anyway (Eguaras *et al* 2001).

The application of organic acids is also a possible control measure for SHB. Schafer *et al* (2009) found that treating combs with either formic or acetic acid reduced the number of both adult and larval SHB on and in the combs. Critically, they also found that these acids inhibited the growth of the *K. ohmeri* yeast, providing not only control of the pest but also the factor that causes the major economic and colony losses.

Like with synthetic chemicals, the method of application can make a difference to organic acid effectiveness and safety. The trickle method involves liquid being trickled directly over the frames in an active hive. This has been shown to be an effective method for using oxalic acid to control varroa, although the timing of applications is critical, as the acid cannot reach those mites in the capped brood cells (Gregorc & Planinc 2001). This is risky for the human operator however; using the acid in this way requires respirator, eye protection and gloves (Gregorc & Planinc 2001).

Other methods of application are both safer for the apiarist and more effective. Formic acid bound within gel packs also ensures that the acid is released over an extended period of time. Tests of such a product by Eguaras *et al* (2001) suggest this is an effective and relatively safe way to treat a varroa infestation, with a much lower variability in its effectiveness than other formic acid application methods. Another method of application, the fumigator, uses the heat of the hive and the fanning actions of the bees to disperse formic acid throughout the hive (Amrine & Noel 2006). Use of formic acid in this way was particularly effective at controlling varroa, with the formic acid killing mites within brood cells as well as on adult bees. Whilst formic acid was a useful tool to control varroa, there are increasing cases of resistance emerging, and its usefulness is now on the wane (Nasr 2010).

Plant derived pesticides such as essential oils are less likely to lead to resistance as there are a range of chemicals present rather than just a single compound. They tend to be less effective than other chemical methods however (Goodwin 2010). After investigating a number of essential oils for effectiveness against varroa and low bee mortality, Lindberg *et al* (2000) found that whilst a range of essential oils might be useful as part of a wider management strategy, they were unlikely to be of use as a standalone treatment.

For example, thymol, derived from *Thymus vulgaris*, is one of the most widely used of the essential oils for varroa control. It has been found to not move from beeswax into honey to the same extent as most synthetic acaracides and any residues will be at very low levels and thus considered safe, however it may affect the taste of honey with long-term use (Bogdanov 2005). The effectiveness of thymol is very variable however, although because it tends to work better in warmer weather it might turn out to be a useful acaracide in Australia (Nasr 2010). It should also be kept in mind that the effectiveness of plant derived essential oils is always likely to be variable due to natural variation in the source material (Maggie *et al* 2011).

A novel organic control strategy using a solution of propolis is proposed by Simone-Finstrom & Spivak (2010). As this is already part of the bees defence system it is safe to use in hives and, consisting of tree resin and wax, it is also safe for humans to handle. Laboratory assays showed that mites are very sensitive to propolis, even at a 10% dilution, and the use of such a solution trickled into hives is a mite control strategy is under investigation. Whilst this is safe and environmentally acceptable solution, it might prove to be hard to collect and process propolis in the necessary amounts for large scale use.

Physical and cultural controls

There are a range of non-chemical measures that an apiarist can take to reduce the likelihood of infestation in their hives, or to reduce economic losses if one occurs.

One critical cultural control measure is to remove any weak or abandoned hives as soon as possible. Abandoned hives will attract SHB (Neumann & Elzen 2004) and robber bees from other hives on which varroa mites can travel.

Maintaining strong, healthy colonies is also important to reduce the impact of both honeybee pests. Stronger colonies have been found to be able to cope with SHB better than weak colonies, despite the fact that more beetles are often present in strong hives (Annand 2011), and weak colonies should be removed or combined to minimise the impact of SHB (DPI 2003; Annand 2011).

As SHB is capable of independent flight, it can invade other colonies relatively easily. As the bees' alarm pheromone one of things that attract SHB to a colony is, it follows that apiarists should minimise disturbance to the bees when working hives, in particular to avoid squashing bees (Annand 2011).

As should be the case for preventing a range of pests and diseases, apiarists must also be very vigilant in using equipment such as tools, supers or frames in multiple hives. Because of SHB susceptibility to cold to is possible to treat infected hives and equipment in a freezer for 6 hours or coolroom for 12 days (DPI, 2003). Larvae leaving the frames are attracted to light so having a light in the corner of the honey shed enables the larvae to be collected easily (DPI, 2003).

Modifying an entrance to a hive is another control measure that can help the bees protect the hive from SHB. For instance, Ellis *et al* (2002c) found that replacing the entrance hole with a 2 cm PVC pipe about 10 cm above the bottom board of a hive can significantly reduce SHB numbers compared with controls, however the modified hives had trouble with water drainage, impaired thermoregulation and increased floor debris. More investigations are thus needed to investigate the optimum entrance size, however due to its role in hive thermoregulation, this is likely to be quite climate and season specific.

Placing hives in the sun may help reduce SHB as the beetle has shown a preference for entering traps (Arbogast *et al* 2009a) and hives (Annand 2011) that are in the shade. This may not be a suitable control measure in summer in some parts of Australia however.

SHB traps containing chemicals have already been discussed, however there are other trapping options that do not require chemicals. Buchholz *et al* (2009) investigated the use of lime and diatomaceous earth to kill SHB in traps. Diatomaceous earth was found to be particularly effective, killing all SHB within the traps and reducing the SHB in hives by 58% under field conditions. This study was only preliminary and did not investigate the effects on bee health or the most suitable trap construction but points the way for further study.

Torto *et al* (2007) found that the addition of baits can increase the effectiveness of a trap. They used a pollen mix inoculated with the *K. ohmeri* yeast, known to attract SHB, and captured significantly more beetles than unbaited traps. Although they describe this technique as a useful tool for monitoring SHB levels, they also found that the traps came close to eliminating SHB in some hives. There was no discussion however on the effects that this might have on the bee colonies; the volatiles released by the yeast mimic the alarm signals of the bees, thus having this scent present in the hive may potentially create communication or physiological problems through stress responses.

Hood & Miller (2003) investigated a range of liquids for use in traps that could attract and kill SHB: vinegar, alcohol, beer, mineral oil, glycerol and honey. It was found that in in-hive traps cider vinegar was the most effective at killing SHB. By contrast, laboratory studies of the substances found that mineral oil was the most lethal to SHB, whilst cider vinegar showed low levels of lethality. This highlights the need for field trials of possible control techniques, as results may well differ from laboratory studies.

Oil traps placed in the bottom of hives have been used to control SHB, however they must be protected to stop bees falling in (Annand 2008). Another problem is the need for the hive to be perfectly level to stop spillages. A down-to earth Australian variation gets around this problem by using a small plastic fishing tackle box with a lid and many small compartments ensuring that the oil is in smaller, less spillable amounts. Small holes punched in the top allow SHB to enter but not the bees (Annand 2008).

Because varroa is not free living, there are fewer opportunities for traps or cultural techniques to control the mite. Providing bees with smaller cell comb foundations on which to build brood cells has been used for controlling varroa, however this has been found not to be an effective control (Ellis *et al* 2009; Goodwin 2010). Powdered sugar sprinkled into the hive can help with varroa control. This works by two mechanisms: it encourages grooming behaviour in bees which leads to bees dislodging and damaging mites on their bodies and the sugar sticks to the pads on the mites' feet making them lose their grip (DPI 2003).

Since the varroa mite has a greater chance of reproducing in drone cells, regular removal of drone brood is also a possible control measure. This was found by Calderone (2005) to be effective, reducing varroa numbers and not adversely effecting bee health or honey production. It is labour intensive however, so might not be a suitable strategy for large scale apiaries, and the long term effects having fewer drones in the gene pool has not been studied.

Biological controls

A range of fungal controls have been tried for the control of both SHB and varroa. Leemon & McMahon (2009) found that whilst various strains of *Metarhizium* and *Beauveria* fungal pathogens were effective in killing SHB in laboratory assays, adding loose spores to hives did not achieve SHB control. It was found that in strong healthy hives, worker bees removed spores before they had a significant effect on the SHB, and weak hives were overrun by SHB despite the spores being present. They suggest that an effective way to apply the fungal spores would be to place them in in-hive traps that only SHB could enter. Encouragingly, they also found that the spores did not survive in honey and there were no lasting effects on bees form the presence of the fungal strains.

Metarhizium strains have also been shown to be a useful control measure for varroa. Kanga *et al* (2003) found that the introduction of *Metarhizium* was as effective as standard chemical treatments against varroa, particularly when no brood is present in the hive, and is safe for bees. Like Leemon & McMahon (2009) they found the sprinkling of spores directly into the hive was not effective, but they found success using strips of spores stuck to the frames in the hives.

Spooner-Heart (2008) investigated possible biological controls for SHB at the ground stage when larvae have left the hive to pupate. They found that the application of *Metarhizium* to the soil was not an effective control for SHB at the soil dwelling stage, however positive results were found for predatory nematode species, particularly *Heterorhabditis bacteriophora*, which achieved a greater than 90% increase in SHB mortality. These nematodes are already registered for use within Australia for the control of soil dwelling insect pests, so their environmental and human safety have had already been established. Whilst this study was conducted in laboratory rather than in the field, the effectiveness of the nematodes at the commercially recommended doses indicates that this is a possible sustainable and affordable control measure for SHB.

Another natural control for pests and parasites in bee hives is pseudoscorpions. These are predators of varroa in India, keeping hives relatively clear of the mite (Donovan & Paul 2006). Donovan & Paul further suggest that one of the reasons that *A. cerana* can coexist with varroa without drastic losses might be due to the presence of pseudoscorpions. Whilst the introduction of these natural predators to areas such as Australia would be too risky ecologically, and unacceptable under quarantine standards, there are over 150 species of pseudoscorpions in Australia (Harvery 2009) and it would be worth investigating feral beehives to see if they can be found in cohabitation. In other countries where pseudoscorpions have been known to occur in bee hives, even if they are not now, their reintroduction might be a useful control strategy. Europe for instance has a species of pseudoscorpion that used to inhabit bee hives but due to modern beekeeping techniques have not been seen in them for 60 years (Donovan & Paul 2006).

Breeding A. mellifera for resistance

Various breeding programs are in place to select for lines of bees that can resist varroa infestations. Breeding for 'hygienic behaviour', where bees are able detect and remove brood that are infested with disease or parasites, has been shown to aid in varroa control (Ibrahim & Spivak 2006; Reuter *et al* 2007). Likewise, increased grooming behaviour, where bees show a tendency to remove mites from other adult bees, is another line of selection in breeding programs (Ibrahim *et al* 2007).

Breeding for colonies that have a reduced amount of drone brood is another strategy. Like the removal of drone combs, has the disadvantage of a potential reduction in the local populations of drones available for breeding, reducing the genetic diversity of the local hives.

The inconsistent guarding behaviour towards SHB shown in European bee colonies points to a possible focus for breeding programs for the control of SHB (Ellis *et al* 2003c). The fact that some colonies exhibit successful guarding behaviour means that the genetics are already present in the gene pool and that colonies exhibiting this behaviour are not at a disadvantage.

Developing sustainable control strategies

As the above discussion shows, there is no one effective, long-term control measure for the control of SHB or varroa in honeybee hives. Whilst there are some effective treatments, such as the use of in-hive traps for the control of SHB, these often require more work for the apiarist so may not be economically feasible.

The developing resistance of varroa to a range of chemicals has increased interest in Integrated Pest Management (IMP) for control of the mite. Keogh *et al* (2010) summarises IPM from varroa control as consisting of four common components: close monitoring of mite numbers to determine treatment thresholds; selective use of chemicals; the application of diverse approaches to reduce populations and the rate at which they increase; and the replacement of colonies when mite burden makes them uneconomic to treat. This could be seen as a good base from which to build a control strategy for SHB too.

Monitoring mite infestation levels is difficult as the mites are so small and are often encased in the brood cells. If there are drone pupae in the brood some of these should be removed with tweezers and examined for mites (DPI 2003). A commonly used monitoring technique in the US involves collecting bees in a jar with holes in the lid, sprinkling them with icing sugar, rotating the jar to coat the bees, then shaking the jar allowing the sugar and any possible mites to fall out the holes (DPI 2003).

Although SHB are bigger, they can be just as hard to monitor numbers, as they move fast and avoid the light, and frequently move in and out of the hive. It is particularly hard to work out the numbers of SHB in any one hive when there is a lot of migration between hives. For instance Levot (2008) found more SHB were killed in a test treatment than were present at the start of the experiment. Traps can provide useful monitoring tools for SHB, particularly if they can be baited with yeast inoculated pollen, or utilise the SHBs tendency to seek out small, dark hides (Arbogast *et al* 2007).

With the widespread development of resistance, at least in varroa, Keogh's suggested 'selective use of chemicals' is becoming more difficult, and there must be more emphasis on the 'application of diverse approaches to reduce populations'. Diverse approaches include the cultural and physical controls discussed above, as well as large scale breeding programs. Many of these approaches will be time costly for apiary management however and must be subject to careful cost-benefit analysis for each individual situation (Goodwin 2010). It is also worth noting that the efficacy of any control measures is very dependent on such factors as the health and even location of the hive (Sammataro *et al* 2004).

Beyond IPM

Both SHB and varroa exist in balance with honeybee species in their native habitats, and in some places feral populations of honeybees have been found to cope with introduced varroa with minimal harm to colonies. Such populations have been noted in North America (Seeley 2007; Villa *et al* 2008), South America (Rozencranz 1999; Fries & Bommarco 2007) and Europe (Fries & Bommarco 2007; Le Conte *et al* 2007). This shows that the honeybees, and the varroa-virus complex and are able to adapt for coexistence. The ultimate aim for the control of varroa and SHB thus might be the development of adaptations that allow it and *A. mellifera* to coexist, thus reducing the need for chemical controls and time-costly management practices.

Breeding programs aim to establish lines of honeybee that can coexist with varroa and SHB, but such breeding programs only work with one side of the partnership: the bees. Under standard chemical management practices, the other part of the partnership, the varroa-virus complex, is incidentally being selected for hardiness and resistance to chemical controls. In different unmanaged populations, adaptations for coexistence have been shown to occur in both the honeybee (Fries & Bommarco 2007) and varroa (Seeley 2007). Furthermore, deliberate breeding programs tend to focus on single resistance mechanisms, whilst naturally developed coexistence often involves a wide variety of mechanisms, as described in Uruguayan and Mexican bees (Rosencrantz 1999).

Adaptation to coexistence with mites does not necessarily have to be a long process – Villa *et al* (2008) found that feral honeybee colony longevity and swarming decreased dramatically at the introduction of varroa but rebounded to pre-varroa levels within 5 years, and Fries *et al* (2007) found that after 7 years of isolation, honeybee colonies on a Nordic island were coexisting with varroa. However in other cases, this has been a much longer process, for instance in France the first resistant colony was noted 25 years after the introduction of varroa (Le Conte *et al* 2007).

On varroa becoming established in Australia, leaving bee colonies without treatment may in fact be the best long-term solution, allowing for honeybee adaptations, and reduced virulence of varroa, to develop as quickly as possible. This would be unlikely to be either economically or politically feasible however. Crop losses, as well as direct losses to apiarists, would amount to millions of dollars and negatively impact Australia's food security. Many commentators regard the use of chemicals as necessity in managing varroa (for example, Keogh *et al* 2010; Goodwin 2010), and the persistence of feral colonies as a problem, due to harbouring the mite, rather than a solution (Keogh *et al* 2010).

Given the large population of feral honeybees in Australia, it may be the case that two different lines of defence against varroa will eventuate once it becomes established: one in managed hives where chemicals and other strategies are used, and the other in feral populations that will be developing adaptations to coexist. Ideally, it might be possible for these two strategies to work in complement – control of varroa in managed hives could preserve pollination services whilst feral populations re-establish. It is thus important that feral bee colonies are not deliberately destroyed in an effort to control varroa, except perhaps in small areas in the initial establishment phase when eradication might be a possibility.

Whilst the SHB has been established in Australia for close to a decade, there appears to have been no investigations into its effect on feral honeybee colonies. Undertaking such investigations should be a priority as it would give an indication as to whether coexistence mechanisms have become established for honeybees and SHB, and thus whether this might be a possibility for varroa too.

However, even if a coexistence between these pests and honeybees in Australia can develop, it may come at a cost - selection for resistance to these pests might create unwanted honeybee behaviours in other areas. As an illustration of this, it has been suggested that some of the African honeybee's negative attributes, such as aggressive behaviour and propensity to abscond, are possible adaptations to coexistence with SHB (Ellis & Hepburn 2006). The emergence of similar behaviours in European honeybees, and consequent increased difficulties in hive management, might be the trade-off for coexistence. Likewise, modern honeybee strains have been selected for the reduced use of propolis as it interferes with hive management, however as it is a useful tool in the bees' arsenal against various pests and diseases, coexistence may favour its greater use (Simone-Finstrom & Spivak 2010).

Conclusion

The huge economic losses, and devastating effects on honeybee populations associated with the introduction of the SHB and varroa mite outside their respective native territories have ensured that there has been much research on effective control measures for these pests. Chemical control methods cannot be relied upon due to widespread resistance, in the case of synthetic chemicals and formic acid, or to variable results, in the case of plant-derived oils. Furthermore, they can inhibit the long-term development of a stable coexistence between honeybees and these pests. Cultural and physical control methods at the level of hive or apiary have higher associated costs but are more sustainable into the long-term. Such control measures in will be useful in maintain pollination industries whilst still allowing for adaptations for coexistence to arise. Deliberate honeybee breeding programs may compliment these strategies. It is likely that Australian honeybees and their pests can develop adaptations that will allow them to successfully coexist, however this process is likely to be delayed by the use of chemical controls.

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