

Review

Mitigating the anthropogenic spread of bee parasites to protect wild pollinators



Dave Goulson *, William O.H. Hughes

School of Life Sciences, University of Sussex, Falmer, Brighton BN1 9QG, United Kingdom

ARTICLE INFO

Article history:

Received 13 February 2015
 Received in revised form 5 June 2015
 Accepted 15 June 2015
 Available online xxxx

Keywords:

Apis mellifera
Bombus impatiens
Bombus terrestris
 Commercial bumble bees
 Disease screening
 Emerging diseases
 Pollination

ABSTRACT

Bees naturally suffer from a broad range of parasites, including mites, protozoans, bacteria, fungi and viruses. Some appear to be host-specific, but most appear able to infect multiple bee species, and some are found in insects outside of the Hymenoptera. The host range, natural geographic range and virulence in different hosts are poorly understood for most bee parasites. It is of considerable concern that the anthropogenic movement of bees species for crop pollination purposes has led to the accidental introduction of bee parasites to countries and continents where they do not naturally occur, exposing native bees to parasites against which they may have little resistance. In at least one instance, that of the South American bumble bee *Bombus dahlbomii*, this has led to a catastrophic population collapse. The main bees that are moved by man are the western honeybee, *Apis mellifera*, and two species of bumble bee, the European *Bombus terrestris* and the North American *Bombus impatiens*. We propose a range of mitigation strategies that could greatly reduce the risk of further impacts of the commercial bee trade on global bee health, including stricter controls on international movement of bees and improved hygiene and parasite screening of colonies before and after shipping.

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* Corresponding author.
 E-mail address: d.goulson@sussex.ac.uk (D. Goulson).

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1. Introduction

The ecological and economic importance of bees and other insect pollinators is well known. Although the major human food crops (rice, wheat, barley, maize) are not reliant on them, insect pollinators benefit the production of 75% of crop species, providing a global pollination service estimated to be \$215 billion p.a. (Gallai et al., 2009). The importance of bees for the production of many crops has led to the domestication and management of some species. The best known and most widespread managed pollinator is the western honey bee *Apis mellifera*. However, bumble bees (*Bombus* spp.) are more efficient pollinators of certain crops and several species of bumble bee are now produced commercially in factories for the pollination of a variety of fruit and vegetable crops in greenhouses, polytunnels and open fields, with over a million bumble bee colonies p.a. being produced and exported on a global scale. For a similar reason, certain solitary bees are also produced commercially for crop pollination, notably the alfalfa bee (*Megachile rotundata*) and various mason bees (*Osmia* spp.) (Delaplane et al., 2000).

The devastating impacts that non-native organisms have wreaked on native ecosystems surely ought to have taught us a lesson as to the risks of allowing release of alien species. The introduction of Nile perch to Lake Victoria, and the introduction of cane toads, prickly pear, rabbits, foxes, and cats amongst numerous others to Australia are well-known examples, but these are just the tip of the iceberg; for example Australia alone has nearly 3000 non-native species established in the wild (Alexander, 1996). A strong case can be made that alien species represent the biggest threat to global biodiversity after habitat loss (with climate change perhaps set to displace both) (Pimm et al., 1995; Ricciardi, 2007). The risks posed by non-native species have long been widely understood (Vila et al., 2010), and are reflected in various legal restrictions on the importation of such species to most countries (Pyke et al., 2008). However, there appears to have been a reluctance to regard bees as potential invasive species, presumably because of their widely-appreciated beneficial role as pollinators (Goulson, 2003). Hence deliberate and sometimes indiscriminate transportation and release of honeybees, bumble bees and various other bee species to new countries and regions began thousands of years ago and continues to recent times. As we shall see, this global transportation of bee species may pose one of the biggest threats to bee diversity worldwide, threatening the vital ecosystem service that they provide to crops and wildflowers. Mitigating this threat whilst still maintaining the valuable pollination services that managed bees provide is the challenge facing conservationists, policy makers, farmers and bee producers today. It is likely that some of these stakeholders are not even aware of this threat at present.

This paper is not a systematic review in that the studies included were not included based on preselected criteria. They were instead included based on the authors' knowledge of the subject area and online searches of Web of Science and Google Scholar.

2. A brief chronology of bee introduction events

The honeybee *A. mellifera*, thought to be native to Africa, western Asia, and southeast Europe, was domesticated in pre-history and has since been deliberately introduced to every continent except Antarctica (Michener, 1979). Some of the most significant landmarks in their spread include their shipment to the Americas in about 1620 (Buchmann and Nabhan, 1996), to Australia in 1826 (Doull, 1973) and to New Zealand in 1839 (Hopkins, 1911). The honeybee is now arguably the most widespread species on Earth, after man. Four bumble bee species, *Bombus*

hortorum, *Bombus terrestris*, *Bombus subterraneus* and *Bombus ruderatus*, were introduced from the UK to New Zealand in 1885 and 1906 to pollinate red clover (Hopkins, 1914). Numerous *Megachile* spp. and *Osmia* spp. were introduced to North America from Europe and Asia during the twentieth century, often for reasons that are unclear (reviewed in Goulson, 2003). *B. ruderatus* was introduced from the naturalized population in New Zealand to Chile in 1982 and 1983 for pollination of red clover (Arretz and Macfarlane, 1986) and by 1994 had spread to Argentina (Abrahamovich et al., 2001).

In the mid 1980's, commercial rearing of the European species *B. terrestris* began, primarily to supply pollination for glasshouse tomatoes, and this quickly became a global trade (Velthuis and van Doorn, 2006) which sparked a new wave of bee introductions. In the early 1990's *B. terrestris* became established in Japan, having escaped from commercial glasshouses (Inoue et al., 2008; Nagamitsu et al., 2007). The species arrived in Tasmania in 1992 from New Zealand, though the mechanism of transport remains unknown (Buttermore, 1997; Stout and Goulson, 2000). In 1998, *B. terrestris* was deliberately introduced to Chile (from Europe rather than New Zealand or Tasmania), despite the presence of native *Bombus* species. It has since spread to Argentina and continues to advance both north and south in South America (Schmid-Hempel et al., 2014). The extent of exportation of *B. terrestris* from Europe is considered commercially sensitive and so is hard to establish, but it is likely that they are currently being exported to many other countries where they have not yet become established. In North America the commercial bumble bee trade focussed on *Bombus impatiens*, a species native to the east of the continent but which was moved outside its native range, as far afield as Mexico where it has established in the wild (Vergara, 2008).

These introductions pose a number of risks, including: competition with native species; hybridisation with native species; disruption of plant–pollinator interactions; improved pollination of non-native plants; and the spread of parasites to native species (reviewed in Goulson, 2003). This last threat is arguably the most serious. Emergent parasites represent one of the most significant threats to biodiversity and spillover of parasites from introduced organisms to native species can be particularly damaging, either because novel species or strains of parasite are introduced or because the increased density of hosts leads to higher prevalence (Daszak et al., 2000; Cunningham et al., 2003).

3. An overview of bee parasites

Bees naturally suffer from a broad range of parasitoids and parasites, the later including protozoans, fungi, bacteria and viruses. Because of their commercial importance, by far the majority of research has focussed on those associated with honey bees and to a lesser extent with bumble bees, with very little known about the parasites of other wild bee species (Goulson, 2003). Some bee parasites, such as Deformed Wing Virus (DWW) and *Nosema ceranae*, have broad host ranges and are able to infect both honey bees and bumble bees whilst others, such as *Crithidia bombi* or *Paenibacillus larvae*, are seemingly specific to one or the other (Genersch et al., 2006; Genersch, 2010; Graystock et al., 2013a). Natural parasites undoubtedly play an important but poorly-understood role in influencing the population dynamics of their bee hosts, but invasion by non-native parasites has the potential to lead to more dramatic effects since we would expect their novel hosts to have little resistance (Daszak et al., 2000; Rosenkranz et al., 2010). The risk is likely to be greatest when the natural host(s) of the parasite is closely

related to potential new hosts (Perlman and Jaenike, 2003). Impact on novel hosts will be determined by transmission rates (which will in turn depend upon the biology of both the host and the parasite) and the virulence of the parasite within the novel hosts (Meeus et al., 2011).

Unfortunately, our knowledge of some basic aspects of the biology of bees and their parasitic organisms is frequently lacking, rendering predictions as to the likely impacts of non-native parasites problematic. Firstly, we do not have good long-term population data sets for any wild bee species in the world, so we are poorly able to detect changes in abundance. Declines are usually only detected when a bee becomes absent from a region, which is rather late to implement any conservation strategy. Secondly, we have a very poor grasp of the natural host range of many bee parasites. For the vast majority of the world's approximately 22,000 known bee species, there has not been a single study on their parasites. No doubt many new parasites (and hosts) remain to be discovered. We know that some parasites that were first identified from honeybees are by no means restricted to *Apis* species. For example, Singh et al. (2010) detected DWV, black queen cell virus, sacbrood virus, Israeli acute paralysis virus and Kashmir bee virus in bumble bees collected near honeybee apiaries. Symptomatic infections of DWV and acute bee paralysis virus have both been reported in bumble bees (Bailey and Gibbs, 1964; Genersch et al., 2006), and the *N. ceranae* microsporidian has jumped host to infect bumble bees (Li et al., 2012; Graystock et al., 2013a,b; 2014; Fürst et al., 2014). DWV is known to also occur in wasps, whilst other parasites such as the fungus *Ascosphaera* and microsporidians have been detected in a broad range of bees, wasps and hoverflies (Evison et al., 2012). Inevitably, because of the huge diversity of species within the Insecta, most insects have not been screened for parasites, either known or unknown. Given this ignorance, we inevitably have a very poor understanding of the natural population dynamics of the many insect parasites which have multiple hosts. Thirdly, we have scant knowledge of the natural geographic distribution of bee parasites. Many were no doubt redistributed around the globe in historical times with movement of domesticated honeybees, long before we had any knowledge of most parasitic organisms. It is now very difficult to establish which bee parasites naturally occur in, for example, the Americas or Australia (or to know what impacts these early bee and parasite introductions had on the native pollinator fauna, which was largely unstudied at the time). Thus one of the most basic challenges we face is detecting when a non-native bee parasite has arrived.

4. Anthropogenic spread of non-native parasites of bees

The best-documented examples of invasions by non-native parasites are in the honeybee (Table 1). The spread of most honey bee parasites has occurred inadvertently as a result of transporting honey bees long distances around the globe. Much of this happened in historic times, long before there was any awareness of the risks posed by non-native species, and before many bee parasites had been discovered. However, it has continued to the present, despite some improvements in quarantine procedures. The best-known example is the mite *Varroa destructor*, originally associated with the Asian honey bee *Apis cerana*, which has jumped hosts to the European honey bee *A. mellifera*, a naive host which has little resistance. Since the 1960s it has spread from Asia to Europe, the Americas and New Zealand (Rosenkranz et al., 2010). The mite vectors pathogens such as DWV, and the combined effect of the mite and such diseases is a major contributor to honey bee colony losses in North America and Europe (Rosenkranz et al., 2010; Nazzi et al., 2012). Fortunately, the mite appears unable to survive on bees outside the genus *Apis*.

A strikingly similar series of events have also seen the microsporidian *N. ceranae* jump from *A. cerana* to *A. mellifera* and in the last 20 years it has spread to Europe and the Americas, where it is now prevalent at high frequency (Klee et al., 2007; Chen et al., 2008), and can have debilitating effects upon colonies (Botias et al., 2013). It has also been detected in wild

bumble bees in Europe, China and South America (Li et al., 2012; Fürst et al., 2014), and in the lab it appears to have higher virulence in bumble bees than it does in honey bees, causing significant mortality (Graystock et al., 2013a), though the impact it has had on wild populations is not known.

Another parasite which is causing considerable concern to beekeepers is the African honey bee parasite *Aethina tumida* (small hive beetle) which recently invaded North America, Egypt, Australia and Europe, where it has caused significant economic harm to apicultural operations (Spiewok et al., 2007). Its host range is not confined to honeybees, for it has been found to attack commercial *B. impatiens* colonies where it causes considerable damage (Spiewok and Neumann, 2006; Hoffmann et al., 2008). It seems highly likely that it also attacks other wild bumble bee species that are not so readily cultured and therefore have not been studied in this respect.

Bee parasites are also being redistributed around the globe by the commercial trade in bumble bee colonies (Table 1). Unfortunately, it does not seem possible yet to rear bumble bee colonies that are free of parasites, not least because the bees are reared on honey bee collected pollen, providing a route of exposure to many bee diseases. Commercial colonies of *B. terrestris* are commonly infected with one or more parasites that are infectious and virulent to native bumble bees and to honeybees, including *Nosema bombi*, *N. ceranae*, *Apicystis bombi*, the mite *Locustacarus buchneri* and DWV (Whittington and Winston, 2003; Gegear et al., 2005; Colla et al., 2006; Goka et al., 2006; Otterstatter and Thomson, 2007; Manson et al., 2010; Singh et al., 2010; Meeus et al., 2011; Murray et al., 2013; Graystock et al., 2013b). Indeed, one might argue that commercial rearing, in which hundreds of thousands of bumble bee nests are reared alongside one another in large factories, are likely to provide ideal conditions for parasite replication unless very efficient sanitation measures are in operation. Such conditions may also promote the evolution of higher virulence by rendering horizontal transmission a more effective strategy than vertical transmission (Pulkkinen et al., 2010; Meeus et al., 2011).

The first documented spread of a non-native parasite with commercial bumble bees was of the tracheal mite *L. buchneri*. *B. terrestris* imported from Europe to Japan in the 1990s were frequently infested with the tracheal mite (Goka et al., 2001). Although this mite also occurs in Japan, the European race is genetically distinct. In addition to importing *B. terrestris*, queens of a Japanese bumble bee, *B. ignitus*, were sent to Europe to establish a commercial stock, and the established nests re-imported back to Japan have been found to be infected with the European race of the mite. In laboratory studies these mite are able to infest various Japanese bumble bee species, and by 2001 mites of the European haplotype were identified in native Japanese bees (Goka et al., 2006). It seems likely that the worldwide trade in bumble bees has led to a global redistribution of various strains of the mite (Goka et al., 2006).

It is not known whether these imported tracheal mite strains inflict more harm on native hosts than do the native parasite strains, but there is strong circumstantial evidence that other parasites associated with commercial bumble bee colonies are having devastating impacts on wild bumble bee populations. In North America, the accidental importation of a non-native strain of the parasite *N. bombi* via commercial bumble bees has been implicated in the dramatic decline of five bumble bee species (*Bombus affinis*, *Bombus franklini*, *Bombus occidentalis*, *Bombus pensylvanicus* and *Bombus terricola*) (Thorp, 2005; Thorp and Shepherd, 2005; Winter et al., 2006a,b). Several of these were once amongst the most widespread and abundant bumble bees in North America, and all rapidly disappeared from >90% of their native range; *B. franklini* went extinct (Cameron et al., 2011). These declines occurred soon after the commercial trade in bumble bees began, and it has been suggested that bee stocks were sent back and forward between Europe and North America (Winter et al., 2006a,b), providing a route for transfer of parasites, but documentation of such movements is lacking, and convincing causal evidence remains elusive (Cameron et al., 2011;

Table 1
Bee parasites for which there is evidence of anthropogenic spread to wild bees.

Parasite	Parasite taxa	Host	Spread to	Pathology	Refs
<i>Apicystis bombi</i>	Neogregarine	Bumblebees	Honeybees?	Parasitizes adult bumblebees. Faecal–oral transmission. Degrades fat body, has neurological effects and can cause mortality. Spillover from commercial bees to wild bumblebees; implicated in bumblebee declines in Argentina. Can infect honeybees.	Liu et al. (1974), Plischuk and Lange (2009), Plischuk et al. (2011), Arbetman et al. (2013), Graystock et al. (2013b), Maharramov et al. (2013), Graystock et al. (2014)
<i>Crithidia bombi</i>	Trypanosome	Bumblebees	Bumblebees	Parasitizes adult bees. Faecal–oral transmission and context-dependent virulence. Appears unable to infect honeybees. Spillover from commercial bumblebees to wild bumblebees; implicated in bumblebee declines in Argentina.	Schmid-Hempel (2001), Brown et al. (2003), Graystock et al. (2014), Schmid-Hempel et al. (2014)
<i>Nosema bombi</i>	Microsporidian	Bumblebees	Bumblebees	Parasitizes adult bees. Faecal–oral transmission, reducing worker survival and colony fitness. Spillover from commercial bumblebees implicated in bumblebee declines in North America.	Colla et al. (2006), Otti and Schmid-Hempel (2007), Cameron et al. (2011)
<i>Nosema ceranae</i>	Microsporidian	Honeybees	Bumblebees	Emerging disease of adult bees. Natural parasite of <i>Apis cerana</i> , jumped host to <i>Apis mellifera</i> , and then to bumblebees. Spillover from honeybees and commercial bumblebees to wild bumblebees. Can lead to mortality.	Plischuk et al. (2009), Fries (2010), Graystock et al. (2013a), Fürst et al. (2014), Graystock et al. (2014)
<i>Locustacarus buchneri</i>	Mite	Bumblebees	Bumblebees	Tracheal mite, feeding on haemolymph of adult bumblebees. May cause lethargy, altered foraging behaviour and reduced lifespan. Spillover has taken place from commercial bumblebees to wild bumblebees in Japan.	Goka et al. (2000), Otterstatter and Whidden (2004), Otterstatter et al. (2005), Goka et al. (2006)
Deformed wing virus (DWW)	Iflaviridae	Honeybees	Bumblebees Solitary bees?	Parasite of brood and adults. In honeybees, infected brood may develop into adults with deformed wings; infection in adults has neurological effects and can cause mortality. Has been detected in wild and commercially produced bumblebees. In bumblebees, infections of brood can also lead to adults with deformed wings, and infections of adults can cause mortality. Has been detected in solitary bees, but infectivity or pathology unknown	Genersch et al. (2006), Chen and Siede (2007), de Miranda and Genersch (2010), Evison et al. (2012), Fürst et al. (2014), Manley et al. (2015), McMahon et al. (2015)
Slow bee paralysis virus (SBPV)	Iflaviridae	Honeybees	Bumblebees?	Causes paralysis of adult honeybees. Has been detected in wild bumblebees, but infectivity or pathology unknown.	Chen and Siede (2007), McMahon et al. (2015)
Israeli acute paralysis virus (IAPV)	Dicistroviridae	Honeybees	Bumblebees	Causes paralysis and mortality in adult honeybees. Has been detected in wild bumblebees. Reduces brood production in bumblebees.	Chen and Siede (2007), Singh et al. (2010), Meeus et al. (2014)
Acute bee paralysis virus (ABPV)	Dicistroviridae	Honeybees	Bumblebees	Causes paralysis and mortality in adult honeybees. Has been detected in wild bumblebees. Also causes paralysis in bumblebees	Bailey and Gibbs (1964), Meeus et al. (2010), McMahon et al. (2015)
Kashmir bee virus (KBV)	Dicistroviridae	Honeybees	Bumblebees	Causes mortality in adult honeybees. Has been detected in wild bumblebees. Delays oviposition and reduces brood production in bumblebees.	Chen and Siede (2007), Meeus et al. (2014)
Black queen cell virus (BQCV)	Dicistroviridae	Honeybees	Bumblebees? Solitary bees?	Causes mortality of queen larvae. Has been detected in wild bumblebees and solitary bees, but infectivity or pathology unknown.	Chen and Siede (2007), McMahon et al. (2015), Manley et al. (2015)
Sacbrood virus (SBV)	Dicistroviridae	Honeybees	Bumblebees? Solitary bees?	Lethal disease of honeybee larvae. Has been detected in wild bumblebees and solitary bees, but infectivity or pathology unknown.	Chen and Siede (2007), Manley et al. (2015), McMahon et al. (2015)
<i>Ascospaera</i> spp.	Fungus	Honeybees	Bumblebees	Lethal, specialist brood disease (chalkbrood). Also infects solitary bees. Detected in wild bumblebees. Infections reported from adult bumblebees.	Aronstein and Murray (2010), Evison et al. (2012), Maxfield-Taylor et al. (2015)
<i>Aethina tumida</i>	Small hive beetle	Honeybees	Bumblebees	Emerging parasite, spreading from Africa to America, Australasia and Asia over last decade. Larvae feed on honey and pollen, and can destroy colonies. Can parasitize bumblebees	Spiewok and Neumann, 2006; Hoffmann et al. (2008)

Meeus et al., 2011). Most recently, Cordes et al. (2012) screened a large number of samples of 36 North American bumble bee species for *Nosema* and *Crithidia*, and demonstrated that surviving populations of the declining species exhibit higher prevalence of *N. bombi* than do stable species. However, this does not prove that *N. bombi* is the cause of declines.

The evidence from South America is a little clearer; here, the arrival of the European *B. terrestris* appears to have led to the rapid local extinction of the native *Bombus dahlbomii* (Schmid-Hempel et al., 2014). *B. terrestris* is spreading in South America at a speed of approximately 200 km per year, far more rapidly than has been recorded in other invasions, so that is now occupies a region in excess of 2,400 km north to south (Goulson, 2003; Schmid-Hempel et al., 2014). *B. dahlbomii*

disappears in the season that *B. terrestris* arrives, so that the two species are rarely observed together, a speed that can only plausibly be explained by pathogen spillover (the two species are unlikely to compete strongly for floral resources since they have markedly different tongue lengths and floral preferences). However, the identity of the pathogen remains the subject of debate. Plischuk and Lange (2009) and Arbetman et al. (2013) report that the neogregarine *A. bombi* seems to have arrived in Argentina by 2009, along with *B. terrestris*. No infections were reported in samples of *B. dahlbomii* or in *B. ruderatus* before *B. terrestris* arrived (Arbetman et al., 2013), and the haplotype now found in Argentina is identical to the most abundant haplotype in Europe (Maharramov et al., 2013), suggesting that it may be the culprit. In addition, Schmid-Hempel et al. (2014) report high

prevalence of the trypanosome *C. bombi* in the invading *B. terrestris* population. It is unclear whether this parasite was introduced to the region with *B. ruderatus* when it was introduced in 1982 or was always present, but the arrival of *B. terrestris* corresponds with a dramatic increase in genetic diversity of the parasite, strongly suggesting that new strains of the parasite arrived with *B. terrestris* (Schmid-Hempel et al., 2014). It has previously been shown that *C. bombi* has higher virulence in hosts which have not previously encountered that particular strain of the pathogen (Imhoof and Schmid-Hempel, 1998).

Either parasite remains a plausible candidate for the demise of *B. dahlbomii*, or indeed it may be due to a third, or a combination of more than one; as in North America, the causal agent for declines has yet to be established beyond doubt. The phenomenon is difficult to study because *B. dahlbomii* have never been reared in the laboratory, and the species is now in imminent danger of extinction. The extent to which *B. terrestris* will spread in South America, and the effects this may have on other native South American species which live further north remains unknown. There is a clear parallel with the devastating impact that the arrival of European diseases had on the native American human population 500 years ago, and it is unfortunate that we did not learn lessons from that.

5. Spillover of native parasites from managed bee stocks

Even when managed bees do not carry non-native species or strains of parasites, they can still impact on native pollinators in two ways. First, if managed bees carry native parasites then they may artificially increase the parasite population in the area. Second, even if managed bees are entirely free of parasites on arrival at a site, they may become infected with parasites from wild bees, and these parasites may then spillback into wild bees. Managed bees interact with wild bees and other pollinators during shared flower use either when the managed bees forage away from the target crop on wildflowers, or when wild bees visit the crop. Such shared flower use can be a major mechanism for the transmission and dispersal of parasites (Durrer and Schmid-Hempel, 1994). Managed bees are often kept at high density, providing suitable conditions for the rapid multiplication of parasites (Whitehorn et al., 2013) which can then spill over into wild populations via shared flower use (Colla et al., 2006; Otterstatter and Thomson, 2008; Yoneda et al., 2008). There is clear evidence that commercial bees do not confine themselves to foraging on the target crop, even when used in glasshouses (Whittington et al., 2004; Kraus et al., 2011; Foulis and Goulson, 2014). In Canada, Colla et al. (2006) found *C. bombi* in wild bumble bees of four different species sampled close to glasshouses containing commercial *B. impatiens*, and no infections in wild bumble bees samples at remote sites (though we now know that *C. bombi* does occur at low prevalence in wild populations of at least 15 North American bumble bee species, Cordes et al., 2012). A combination of field observations and modelling suggest that waves of *C. bombi* infection can travel outwards from glasshouses containing commercial bumble bees (Otterstatter and Thomson, 2008). Predictions suggested that waves of infection can spread at ~2 km per week, with up to 100% of wild bees within the spreading radius contracting an infection, although this has not been clearly demonstrated in the field (Otterstatter and Thomson, 2008). Whitehorn et al. (2013) found no evidence of such spillover from commercial bumble bees in the UK, but Graystock et al. (2014) found *A. bombi*, *C. bombi* and *N. ceranae* to all be higher in the UK near greenhouses using commercially produced bumble bees than near greenhouses that were not.

More recently, Fürst et al. (2014) examined geographic patterns of prevalence of *N. ceranae* and DWV in honeybees and wild bumble bees in the UK and found strong evidence that pathogens regularly transmit between the two but that the majority of flow is from the managed honeybees (which tend to be more numerous) into the wild bumble bees. This was corroborated by studies of genetic variation in DWV which demonstrated that sympatric populations of honeybees and

bumble bees tended to share the same parasite strains. Clearly, the health of wild pollinator populations ought to be a consideration in the management of commercial bees, regardless of whether they are native.

6. Interactions between parasites and pesticides

Several recent studies indicate that interactive effects between parasites and pesticides could be especially harmful for bees (Alaux et al., 2010; Vidau et al., 2011; Pettis et al., 2012; Aufauvre et al., 2012, but note that Baron et al. (2014) found no significant synergy between pyrethroids and *C. bombi* in bumble bees). For instance, developmental exposure to neonicotinoid insecticides renders honey bees more susceptible to the impact of the alien pathogen *N. ceranae* (Wu et al., 2012). The neonicotinoid imidacloprid can act synergistically with *Nosema* spp. by increasing the prevalence of *Nosema* infections in hives (Pettis et al., 2012) and increasing *Nosema*-induced mortality (Alaux et al., 2010). Similarly, Aufauvre et al. (2012) demonstrated that mortality of honey bees was greater when bees were exposed to the insecticide fipronil and also infected by *N. ceranae* than when only a single stress factor was present. There is evidence that exposure to pesticides may impair the immune function of insects, which would explain these effects (James and Xu, 2012; Nazzi et al., 2012; Brunner et al., 2013). For example, Di Prisco et al. (2013) recently showed that exposure to neonicotinoids (clothianidin or imidacloprid) leads to immunosuppression in honey bees, which, in turn, promotes the replication of the deformed wing virus in insects bearing covert infections. This effect was found at very low concentrations, well below those that bees are likely to encounter in the field. In general, it seems likely that there is a metabolic trade-off between production of detoxification enzymes (for example the cytochrome P450 gene family members, Scott and Wen, 2001), and up-regulation of immune functions. Overall, it seems likely that low level exposure to pesticides such as is likely to routinely occur in agricultural environments may exacerbate parasite problems in bees.

7. Mitigation strategies

The importance of preventing the anthropogenic spread of bee parasites is therefore clear. It is also clear that achieving this for parasites that can be difficult to detect and generally impossible to cure is not straightforward. However, there are nevertheless many potential strategies that can be employed to reduce the anthropogenic contributions to the spread of bee parasites in the future (Fig. 1).

7.1. Mitigation measures in preventing entry to the factory or apiary

The best way of stopping the anthropogenic spread of bee parasites is by ensuring that commercially managed bees are free from parasites, and the most straightforward way to achieve that is to ensure that no parasites enter into bee production factories or apiaries. There are three main routes by which parasites can enter commercial facilities, all of which can be controlled.

7.1.1. Bees

New bumble bee queens are periodically introduced into factories that commercially produce bumble bees (Velthuis and van Doorn, 2006), and queens or swarms are similarly introduced periodically into honeybee apiaries. Ensuring that new bees are free of parasites when they are received would therefore be a good start. In addition, quarantining as far as possible any bees that enter production facilities or apiaries until they have been confirmed, by screening or long-term monitoring, to be free of parasites would be prudent. Such quarantining of new queens would be relatively straightforward in factories producing bumble bee colonies, but is also possible to a more limited extent in honeybee apiaries, e.g. by placing hives containing new queens or bees

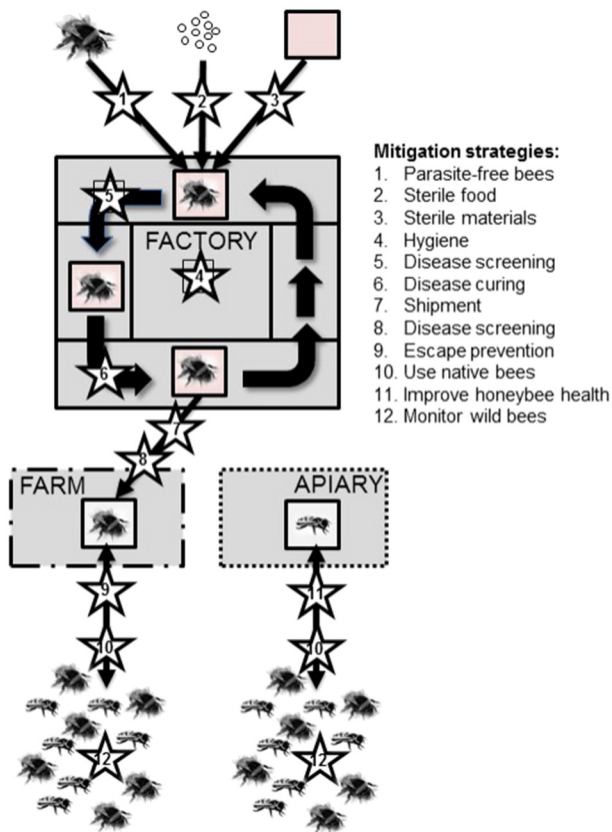


Fig. 1. Schematic illustrating the many opportunities for mitigating the impacts of parasites associated with managed bumblebee and honeybee colonies on wild pollinators.

far enough away from existing colonies to avoid drifting (movement of workers between hives).

7.1.2. Food

Commercially produced bumble bees are reared on sugar syrup and honeybee-collected pollen (Velthuis and van Doorn, 2006), whilst honeybees are also commonly fed sugar syrup and sometimes pollen substitute. Accidentally feeding parasite-contaminated food to bees is obviously not conducive to producing colonies that are parasite-free. The preparation of parasite-free sugar syrup is unproblematic, although care still needs to be taken to avoid contamination of the containers used to hold or dispense the syrup. Minimising the levels of viable parasites in pollen, without harming the nutritional value of the pollen, is more difficult. Honeybee pollen is probably always contaminated with a wide diversity of parasites, including both those that infect honeybees and also those that do not infect honeybees but do infect other bees (Singh et al., 2010; Graystock et al., 2013b). Some commercial producers of bumble bees now routinely treat honeybee pollen with gamma radiation before feeding it to their bumble bees in an attempt to reduce the levels of parasites (Graystock et al., submitted for publication). This appears to be effective at substantially, but not always completely, reducing the viability of a variety of parasites (Meeus et al., 2014; Graystock et al., submitted for publication), and therefore appears to be a sensible first step. It may be that the irradiation method can be refined to produce a more complete reduction in parasites, or alternatively a complete solution would be the development of a hygienic, nutritionally equivalent pollen substitute to replace the use of honeybee pollen for the commercial rearing of bumble bees. Pollen substitutes are already widely available for feeding to honeybees (Crane, 1990), and the development of equivalents for bumblebees should be a matter of priority.

7.1.3. Other materials

The risks of introducing parasite-carrying bees into a factory or apiary, or feeding bees parasite-contaminated food, are relatively obvious. However, these are not the only materials that are introduced into factories and apiaries, and a scrupulous care is needed with all materials. The spores of many parasites are extremely resilient and persistent, and can contaminate hive boxes or other materials equipment (Morse, 1990; Dobbelaere et al., 2001; Aronstein and Murray, 2010). Avoiding the reuse of hive boxes and other hive materials, or sterilising thoroughly any materials that are reused, is therefore sensible. Many bee parasites are hard to detect, so this strategy should be employed even when material is from colonies that are believed to be free of parasites.

7.2. Mitigation measures in the factory or apiary: breaking the infection cycle

Preventing the entry of parasites into bee production facilities or apiaries is an essential step, but it is unrealistic to expect it to always be fully effective. The high transmissibility of many bee parasites means that any parasites that enter a factory or apiary will often have the potential to spread rapidly and widely within it. Protocols therefore need to be in place to break the infection cycle in order to prevent this.

7.2.1. Hygiene

The ease with which parasites spores can contaminate hive boxes, equipment and even clothing (Morse, 1990; Dobbelaere et al., 2001; Aronstein and Murray, 2010), means that compartmentalisation of colonies into different rooms or apiaries, and strict hygienic procedures during the production and management of colonies, are essential to prevent the spread of any parasites and potentially allow their eradication before the entire operation is infected. Thorough sterilisation of equipment and changing of clothing when moving between rooms or apiaries needs to accompany this to prevent the accidental carry-over of parasites when the beekeeper or factory worker moves between areas.

7.2.2. Parasite screening

Regardless of what measures are in place to prevent the entry and dispersal of parasites in factories or apiaries, the routine screening of bees is essential to allow the early detection and eradication of parasites. Although some parasites of honeybees can cause very obvious symptoms (Morse, 1990), it is unfortunately the case that most bee parasites are hard to detect. Screening of faeces or gut samples by microscopy is laborious, can easily miss low intensity infections of protozoan or fungal parasites even when carried out rigorously by well trained personnel, and cannot detect virus infections (Morse, 1990). The only solution that allows the reliable detection of all parasites, including low intensity infections, is the destructive screening of bees using sensitive PCR and RT-PCR molecular methods. There are now suitable protocols for most known bee parasites (Chen et al., 2005; Meeus et al., 2010; Graystock et al., 2014), and they can be a relatively cheap, quick and sensitive method of screening large numbers of bees for parasite infection. With current protocols, screening for many parasites requires many reactions, so these methods could be improved further by the development of more complex multiplex protocols to allow the detection of very many parasites in a single PCR or RT-PCR reaction.

7.2.3. Curing

Arguably the most important, but most commonly forgotten, fact with regards to the spread of parasites from commercially produced bumble bees is that there are no methods currently available for curing any bumble bee parasite. There are precious few cases where a disease in human societies, animal production or agriculture has been eradicated without a method being available to cure the disease, so developing such methods clearly needs to be a priority. Methods have been developed successfully to cure honeybee colonies of a wide diversity of

parasites, including acaricides, antibiotics, fungicides and RNA interference (RNAi) (Morse, 1990; Maori et al., 2009), so development of suitable approaches to cure bee diseases is possible. The development of similar strategies to control bumble bee parasites, and of strategies to cover a broader range of honeybee parasites, is now needed. In lieu of a method of curing parasites, the rapid elimination of any colonies found to be carrying parasites is the best alternative.

7.3. Mitigation measures from the factory to the farm

The challenge of minimising the anthropogenic spread of bee parasites does not end when bees leave the production apiary or factory, and measures are needed after shipment and arrival to ensure that the benefits of good practice in the production apiary or factory are realised.

7.3.1. Shipment

Bumble bee colonies are produced in a small number of factories and then transported internationally to the end-user farms. Honeybee colonies too are sometimes shipped over considerable distances. The shipment of animals in confined and often environmentally suboptimal conditions has long been recognised to cause stress that can lower the animal's resistance to parasites, resulting in the phenomenon known as 'shipping fever' (Barham et al., 2002). A similar effect has been suggested to play a role in the large-scale losses of honeybee colonies in North America that are often shipped by truck over long distances (Oldroyd, 2007). Minimising the time from shipment to arrival on farms by shipping colonies rapidly and with minimal storage after shipment will therefore help reduce the susceptibility of colonies either to parasites they may be carrying or any they encounter during or after arrival. In addition, ensuring colonies have adequate nutritional resources and minimal stress during shipment, including ensuring that colonies are treated as fragile goods and not exposed to high or low temperatures, is likely to be worthwhile to minimise the stress the bees are placed under and reduce the possibility of 'shipping fever'.

7.3.2. Parasite screening on arrival

Although most countries have some form of checks of honeybee colonies for notifiable parasites, and some national and supranational policy-making organisations require that imported commercially produced bumble bee colonies are free of parasites, the level of regulation is extremely variable, generally restricted to a limited subset of potential parasites, sometimes involves only checks by veterinarians that are unable to detect most parasite infections, and often limited to compliance records of parasite screening carried out by the producers of colonies themselves (European Commission, 1992; HM Government, 2006; Velthuis and van Doorn, 2006; Winter et al., 2006a,b; The Food and Environment Research Agency, 2012; Natural England, 2012). Investigation of parasite levels in commercially produced bumble bee colonies has produced a clear contrast in results: on the one hand, the commercial producers believe that their bumble bee colonies are free of any parasites; on the other hand, numerous independent studies of colonies after importation have found colonies to very frequently be carrying parasites on arrival (Whittington and Winston, 2003; Gegeer et al., 2005; Colla et al., 2006; Goka et al., 2006; Otterstatter and Thomson, 2007; Manson et al., 2010; Singh et al., 2010; Meeus et al., 2011; Murray et al., 2013; Graystock et al., 2013b). Relying on producers to self-certify the parasite-free status of their colonies clearly raises a conflict of interest. It would seem sensible for relevant authorities to carry out independent screening for parasites of random colonies on arrival at farms, using sensitive PCR-based methods. This could perhaps be funded by a 'honesty' levy on producers or via punitive fines on producers when their colonies are found to carry parasites, which would avoid the end-user farmers bearing the costs (as would be the case if, for example, parasite carrying colonies were destroyed). Ultimately, robust, independent verification of the parasite-free status of colonies would be in the interests of producers as well as enabling policy-

making and conservation organisations to be confident that no parasite-carrying colonies are being imported.

7.3.3. Escape prevention

When bee colonies are used in open landscapes then there is little which can be done to prevent their workers interacting with wild bees during shared flower use. However, excluders can be placed on the entrances of hives to prevent the exit of queens from bumble bee colonies, and the use of excluders or good swarm management practice can prevent the exit of swarms from honeybee colonies (Hooper, 2010). In addition, commercially produced bumble bee colonies are most commonly used in greenhouses or polytunnels, and preventing the escape of bees is more feasible in these settings. Mesh netting can be placed over vents, windows and doors to prevent the exit of bees from, or the entry of bees into, greenhouses or polytunnels. Such an approach is already employed in Japan (Meeus et al., 2011). Farmers have a justifiable concern about netting causing an increase in humidity and mould, but this should not be an intractable problem to solve, e.g. wind-powered, mesh covered fans could be incorporated in place of open (or meshed) vents.

7.4. Mitigation measures in policy

Although there are therefore many direct mitigation measures which can be taken by producers, farmer end-users, or conservation organisations, there is also the need for policy-makers to take action to help mitigate the risks of anthropogenic spread of bee parasites.

7.4.1. Encourage the use of native bees where possible in preference to importing non-native species

The production, export and import on a global scale of non-native bee species for pollination represents an obvious danger, not only in terms of the possible co-introduction of parasites but also because of the other ecological threats that non-native species can pose to ecosystems (Vila et al., 2010). The importance of imported bees for crop pollination is likely to preclude banning the importation of non-native bees in most cases unless there is an economically viable alternative. A more realistic solution is therefore to prioritise the development of native bee taxa for commercial use, and to then legislate against the importation of non-native taxa as soon as a native alternative is available. A successful example of such a strategy is the UK in which relatively temperate actions by policy makers encouraged the market to shift over several years from the importation of non-native bumble bees to the use of the native *Bombus terrestris audax* subspecies, to the point at which banning the importation and use of non-native bumble bees became possible without harming the farmer end-users (Natural England, 2014). The development of native bumble bee taxa for commercial use will often not be as straightforward as in this example, but it nevertheless demonstrates what can be achieved when industry, farmers and policy makers pull in the same direction. Of course, it is probable that factories will continue to produce non-native bees alongside native taxa, so the success of this mitigation strategy requires there to be strict procedures in place to prevent any interactions between the different species or subspecies, particularly, but not limited to, cross-breeding between taxa in the factory. In order for regulators and consumers to be confident in the provenance of the bees that are imported, it will also be necessary for there to be some level of independent checking of bees on arrival to confirm that they are the species and subspecies claimed. Both for this, and for the industry to be able to check the rigour of its own production procedures, there is therefore a need for the development of accurate and cost-effective methods to identify the species and subspecies of bees, which may in some cases need the development of suitable genetic markers (Estoup et al., 1996; Widmer et al., 1998; Williams et al., 2012).

7.4.2. Reduce the spillover of parasites from honeybees by improving their health and restricting their use near areas with vulnerable populations of rare species of wild bee

Given the evidence for spillover of parasites from honeybees to wild bees (e.g. Graystock et al., 2013b; Fürst et al., 2014; Manley et al., 2015), any improvements in the health of honeybees is likely to benefit wild bees too. Management of honeybee parasites is very variable within and between countries, and is usually left to individual beekeepers with varying levels of support and guidance (Crane, 1990; Neumann and Carreck, 2010). Better guidance and assistance to beekeepers, including strategies that aim to manage a broad range of parasites rather than a very small set that are considered of greatest risk, is therefore worthwhile. Currently, apicultural practices are concerned only with diseases that threaten the health of the honeybee colonies in the apiary, and apiculturists frequently tolerate parasite infections that do not directly represent a serious threat to their colonies. Apicultural practices need to move beyond this by recognising that even parasite infections that have a limited impact on the honeybee colonies themselves are likely to be spilling over to and negatively affecting wild bees. For responsible environmental stewardship, apiculturists need to apply much stricter approaches to eliminate as far as possible all parasites from their honeybee colonies for the benefit of the surrounding wild bee community. In addition, there is scope in some cases to reduce the risk of anthropogenic spillover of parasites by minimising interactions between managed bees and wild bees. In most cases this is of course impractical, but in some targeted cases it may be worth considering. For example, preventing the placement of honeybees hives or the use of commercially produced bumble bees in protected areas which support populations of rare wild bees would seem prudent.

7.4.3. Monitor the numbers and prevalence of parasites in wild bees so we can identify and address a problem before it becomes too late

At present we do not have good population data for any wild pollinator species, and hence we have only a limited idea of which species are declining, where, or how rapidly. If a disease epizootic were to strike, it is likely that it would only be noticed if it had dramatic and rapid consequences, as in the case of the disease-associated decline towards extinction of *B. dahlbomii* in Argentina (Arbetman et al., 2013; Schmid-Hempel et al., 2014). A reliable, long-term monitoring programme is essential if we are to detect problems at an early stage or quantify the effectiveness of any strategies that we implement to conserve wild pollinators. Ideally, such long-term monitoring should include the screening of representative samples for parasites using sensitive molecular methods, in order to develop a sound understanding of parasite prevalence, how it correlates with population dynamics, and then to be able to detect any changes in it.

8. Conclusions

The anthropogenic movement of managed bees has led to irreversible introductions of bee parasites to new regions of the globe, with significant adverse effects on health of both wild and managed species. We should learn from our past mistakes, and take steps to prevent further redistribution of bee parasites in the future. We propose a range of mitigation strategies targeted at all stages of the rearing, distribution and on-farm management of commercial bees which, if properly implemented, would greatly reduce the likelihood of further adverse impacts on both wild and managed pollinators.

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