The hidden ecology of bumblebees: using classical and new methods to explore nest searching, floral resources and badger predation

Submitted by Bethany Rose Roberts to the University of Exeter as a thesis for the degree of Doctor of Philosophy in Biological Sciences in July 2019

I certify that all material in this thesis which is not my own work has been identified and that no material has previously been submitted and approved for the award of a degree by this or any other University.

Signature: ……………………………………………………………..

“If you limit your actions in life to things that nobody can possibly find fault with, you will not do much!”

– Lewis Carroll

General abstract

Bumblebees provide a crucial ecosystem service, and are experiencing worldwide declines due to a number of stressors, such as habitat loss and climate change. Populations are regulated through ‘bottom-up’ (resources) and ‘top-down’ (pesticides, disease and predation) processes. Bumblebees have been widely researched, but there are still aspects of their ecology which are understudied due to the difficulties associated with researching them. This is the case for nest locations and nest predation. This thesis aims to uncover some of these hidden aspects of bumblebee ecology, through the development and implementation of new techniques: thermal cameras as a tool to locate bumblebee nests (**Chapter 3**), artificial nests to study badger predation rates (**Chapter 4 & 5**), and the utilisation of model simulations to look at the potential combined impacts of badger predation and food availability on bumblebee populations (**Chapter 5**). Thermal cameras were found to be unsuccessful, due to their small viewing areas. However the artificial nest method was shown to be a successful way of studying the relative differences in predation rates between habitats and geographic locations, with high predation of bumblebee nests in areas of high badger densities. Model simulations also showed that under high badger predation rates a simulated bumblebee population produced significantly fewer hibernating queens, workers, and colonies. Thus, novel research tools were found to be a useful way of monitoring the effects of top-down and bottom-up effects on bumblebee populations.

Alongside this, classical field techniques were used to study the use of botanical gardens and semi-natural farmland habitats by nest searching queens, and the foraging resources they provide across the flight season (**Chapter 2**). Residential gardens have been shown to be beneficial habitats for bumblebees, especially in urban environments. In **Chapter 2** botanical gardens are looked at in a rural setting, which has not been done previously. Botanical gardens were found to contain a high number of nest searching queens, and experienced a peak of spring floral resources. However semi-natural habitats in farmland provided higher levels of floral resources in early and late summer. Therefore these two habitats may be providing complementary floral resources to pollinators. These results are discussed in terms of the importance of habitat heterogeneity for supporting pollinator populations.

Acknowledgements

To say that this PhD has been a journey would be an understatement. During this time I have changed and grown so much as a person, both because of and in-spite of the process. There are many people who have made this possible and more enjoyable, and I'd like to thank them below.

Firstly, thanks goes to my supervisor Juliet Osborne. She has allowed me to follow my own research passions, and helped me to become an independent scientist. Her support and encouragement have meant a great deal, especially during the final push, of which I had always been scared. She has managed to turn it into a not wholly unenjoyable experience and for that I am forever grateful.

This PhD also would not have happened without the support from the rest of the Bee Group. Pete for teaching me how to mark bumblebees, and for always having questions, Ros for her superior stats knowledge and Grace for her help with fieldwork. Matthias needs a particular mention; his magical mathematical mind has been a true wonder to observe and the speed and patience with which he has helped me has probably not made me any better at maths but was very much appreciated.

I definitely could not have done this without The Bee Team (a.k.a Jess and Ben). Thank you both for always being on hand to answer my questions, stop me from worrying and for reminding me that we are only human. You are both wonderful and are going to do incredible things and I am glad to have basked in the warmth of your shared geniuses.

Now, to all the weirdos I am lucky enough to call my friends. How do I even begin to thank you all? Emily, thank you for introducing me to Lucinda and the lifeline that are her yoga classes. Both you and she are goddesses. Cat, thanks for being the best housemate and for the pre-bedtime Mamma Mia karaoke sessions. Miranda, you will always be my hero, thank you for inspiring me every day and for always making me laugh. Warren, our shared love of organisation and panicking brought us together, thanks for being my musical accomplice, my soul thanks you. To the residents of 3 Norfolk Road who have been and gone, you have all been wonderful and your counsel, laughter and wackiness means I will never forget any of you.

To everyone at work, you are more than just colleagues, and you have made coming in every day an absolute pleasure, even in the darker times when you fear you will never see the light (and the literal dark times due to being in ‘The Cave’). To the lunch crew, who are there at 12 without fail, thank you for always providing interesting conversation, advice and most importantly, cake.

I couldn’t write my acknowledgements without thanking my work husband, Jared. When I found out we would be desk buddies I would never have imagined that you would be such an integral part of my journey. Thank you for always taking off your headphones when I need advice, for putting up with my extraordinary level of untidiness (thank goodness we had the red tape), for telling me when I am being an idiot and for being my friend. Future work husbands have a lot to live up to!

To all the people who have not been mentioned in this by name, but who have been there along the way, thank you. For the laughter, the meals cooked, the adventures, the fishing, the sea swims, the pints, the dancing, the hugs, the crying, the films, the music, the homes, the love. Thank you.

Thanks must also be given to the splann group of women who make up the Mabe Ladies choir and the Celtic Chords. Singing with you each week has replenished my soul; you always make me laugh and smile, I hope I am like all of you when I grow up!

Last, but by no means least, to my family. I’m still not entirely sure you know what I do, and my nan still cannot believe that I haven’t found out all there is to know about bumblebees, but you have all been amazing supports. To my mum, who gave me my passion for nature and taught me all the birds and plants. I bet when you were dragging me around garden centres you never thought I’d actually need to know that information for my thesis! To my dad, who always provides interesting conversation and makes me go running with him, my internal organs are grateful for the bi-annual exercise. To my brother the rap legend, for reminding me I will never be as cool as him. And to my nan, who always tells me about the bumblebees in her garden, and listens patiently over the phone even when I am moaning about things that make no sense to her, you’ve always tried to understand what I do and it means the world. I love you all very much.

Contents

Table and figures

Publications and contributions

- Chapter 3 has been published: Roberts, B. R. and Osborne, J. L. (2019) Testing the efficacy of a thermal camera as a search tool for locating wild bumble bee nests. *Journal of Apicultural Research* DOI: 10.1080/00218839.2019.1614724
    - The experimental design, data collection, data analysis and writing of the manuscript was undertaken by myself. Juliet Osborne assisted with experimental design, commented on manuscripts and approved the final version for submission.
- Chapter 4 has been submitted for publication and is currently under review: Roberts, B. R., Cox, R., Osborne, J. L. (2019) Ecology and Evolution.
    - Experimental design, data collection, data analysis and writing of the manuscript was undertaken by myself. Ruth Cox assisted with data collection and commented on manuscript drafts. Juliet Osborne assisted with experimental design, commented on the manuscript and approved the final version for submission.
- Equations in Chapter 4 were developed by Matthias Becher, who was the creator of the *Bumble*-BEEHAVE model used for simulations

Chapter 1

Introduction

“The story of the life of the humble-bee is largely that of the queen. From start to finish she is the central and dominating personage upon whose genius and energy the existence of the race depends. For she alone survives the winter, and, unaided, founds the colony in which she takes the position of its most important member.”

– Sladen, 1912

Chapter 1: Introduction

1.1 Insect pollinators: their importance and causes of declines

Insects are experiencing worldwide declines (Hallmann *et al.*, 2017; Lister and Garcia, 2018). The main drivers of these declines are climate change (Fox *et al.*, 2014; Lister and Garcia, 2018) and habitat loss and fragmentation (Fox et al., 2014; Nilsson *et al*., 2008; Potts *et al*., 2010); largely through agricultural intensification (Robinson and Sutherland, 2002). Throughout Europe large areas of land are taken up with arable crop fields (Stoate, C *et al.*, 2001), and areas of natural habitats have been lost (Fuller, R. M., 1987). Pollinators are a particularly important group of insects, which provide an important ecosystem service in terms of pollination; 60% of crops require insects for pollination (Klein *et al.*, 2007) and 87% of wildflower species (Ollerton *et al.*, 2011). One study estimated pollination services at a value of over $200 billion in enhanced crop yields (Gallai *et al.*, 2009). Thus, their declines could have implications for global food security. Understanding how pollinator populations are being regulated by both bottom-up (resources) and top-down (predation and disease) factors (**Chapter 4 & 5**), and how these could be acting synergistically is important in further understanding the mechanisms behind their declines.

Bumblebees are a key group of pollinators (Carreck and Williams, 1998; Velthuis and van Doorn, 2006), especially in temperate climates due to their ability to forage at low temperatures and in light rain (Willmer *et al.*, 1994). Globally, *Bombus terrestris* is used as a commercial pollinator, where it is imported to countries outside of its native range to provide pollination services (Velthuis and van Doorn, 2006). They are particularly important for the pollination of crops grown within glasshouses and polythene tunnels, such as tomatoes, peppers and aubergines (Carreck and Williams, 1998; Velthuis and van Doorn, 2006). Supplementing pollination with managed and commercial pollinators can increase fruit weight (Walters and Taylor, 2006), but overall benefits can be mixed (Petersen *et al.*, 2014; Petersen *et al.*, 2013). Commercial *Bombus* species which were imported commercially are now invasive species around the world (Torretta *et al.*, 2006; Velthuis and van Doorn, 2006). The use of commercial and managed pollinators can have negative consequences for native bee and plant species with introduced species often being better foragers and producing more queens (Ings *et al.*, 2006; Velthuis and van Doorn, 2006). Increasing our understanding of wild pollinator ecology, and how they are using remaining habitats in terms of both nesting and foraging (**Chapter 2**), could increase wild populations and lead to less requirement of commercial pollinators. This would benefit wild pollinators, especially where commercial escapees become invasive, and reduce the spread of diseases and competition between commercial and wild pollinators (Goulson *et al.*, 2015; Manley *et al.*, 2015; Potts *et al.*, 2010; Williams and Osborne, 2009). The focus of this thesis is therefore focussed on developing and implementing novel techniques to research and fill some of the remaining knowledge gaps around bumblebee ecology.

1.2 Top-down and bottom-up regulation

Pollinator populations are regulated by both bottom-up and top-down resources. Bottom-up processes act on populations mainly through reducing the growth rate, whereas top-down processes cause direct mortality. The loss of habitats and their fragmentation is a major cause of pollinator declines (Goulson *et al.*, 2015; Potts *et al.*, 2010), having bottom-up effects on insect pollinators; affecting the availability and distribution of nesting and food resources within the landscape (Biesmeijer *et al.*, 2006; Goulson *et al.*, 2008; Potts *et al.*, 2010). Specifically, habitat loss impacts pollinators through reducing the quantities of both food and nest sites, whereas habitat fragmentation leads to these resources being more sparsely distributed within the landscape. This leads to reduced habitat heterogeneity, where there are fewer patches of floral and nesting resources which are smaller and further apart. These can have varying impacts on pollinators, which are discussed below. In order to improve landscapes for pollinators, habitat complexity must be increased to provide sufficient levels of both foraging and nesting resources throughout the flight season.

1.2.1 Bottom-up regulation: habitat loss and fragmentation

Habitat loss has occurred largely due to intensification of agriculture. In Europe, the arable landscape is vast (Stoate *et al.*, 2001) and natural habitats are being lost; for example between 1932 and 1984 the area of unimproved lowland grassland in Britain declined by 90% (Fuller 1987). Declines in semi-natural habitats within this timeframe (1930-1978) led to drastic reductions in nectar availability (Baude *et al.*, 2016). Reduced food availability can affect the growth, lifespan and fecundity of insects (Agarwala *et al.*, 2008; McKay *et al.*, 2016; Olson *et al.*, 2017). Pollinators are especially reliant on nectar and pollen availability. In bumblebees, food availability has been linked to the production of queens and males (Pelletier and Mcneil, 2003; Rotheray *et al.*, 2017) and in solitary bees can impact egg laying and fecundity (Williams and Kremen, 2007). Baude *et al.*, (2016) inferred that in 2007, 50% of nectar provision in the UK was from just four plant species. A lack of floral diversity could have negative consequences for pollinator communities (Fründ *et al.*, 2010), with specialists potentially being more adversely affected (Biesmeijer *et al*., 2006; Roulston and Goodell, 2011).

Habitat fragmentation has also occurred as a result of the large area of land taken up with cropped agricultural fields and populated areas. This has led to foraging and nesting resources which are patchily distributed and confined to semi-natural habitats within the landscape (Söderman *et al.*, 2018). The size of these patches can impact their use by pollinators. Larger patches, and those with clustered floral resources had higher density and diversity of wild bees (Blaauw and Isaacs, 2014; Plascencia and Philpott, 2017), and were also attractive to male and queen bumblebees (Carvell et al., 2015). In contrast, other studied found bumblebees to be unaffected by patch size; being found in greater abundances in floral patches surrounded by higher proportions of arable land (Heard *et al.*, 2007). Bumblebees are relatively large pollinators which will travel several hundred metres to forage (Carvell *et al.*, 2012; Redhead *et al.*, 2016), and are known to fly further to visit highly abundant and diverse floral resources (Jha *et al*., 2013a; Osborne *et al*., 2008a; Redhead *et al*., 2016). Pollinators which have shorter foraging ranges, such as solitary bees (Gathmann and Tscharntke, 2002), or are less mobile such as some butterfly species (Öckinger *et al.*, 2009) are likely to be more adversely affected by habitat fragmentation and isolation from semi-natural habitats. For example, solitary bees produced fewer offspring with lower survival when isolated from semi-natural habitats (Williams and Kremen, 2007). Many pollinators require multiple habitats to complete all stages of their lifecycle (Williams and Osborne, 2009). Therefore, the presence of florally rich habitats within the landscape could act as sources for pollinator populations. In **Chapter 2**, botanical gardens situated within a rural agricultural landscape are studied in terms of their use by nest searching queens, and the floral resources they provide over the bumblebee flight season, in contrast to semi-natural farmland habitats.

1.2.3 Top-down effects: parasites, pathogens, pesticides and predation

As well as bottom-up effects, populations can be regulated through top-down stressors, such as parasites and pathogens, pesticides and other chemicals and predation, all of which can cause direct mortality. Parasites and pathogens are prevalent within the environment (Imhoof and Schmid-Hempel, 1999), and can be spread to wild pollinators via managed and commercial pollinators (Goulson *et al*., 2015; Manley *et al*., 2015; Potts *et al.*, 2010; Williams and Osborne, 2009). This can cause reduced larval survival and increased worker mortality (Graystock *et al.*, 2013). Pesticides such as neonicotinoids can impact learning, foraging and homing abilities of bees (Goulson *et al.*, 2015), with stronger effects observed when pollinators are exposed to a ‘cocktail’ of different pesticides (Gill *et al*., 2012). Herbicides can cause bottom-up effects, through reducing floral availability (Goulson *et al.*, 2015; Williams and Osborne, 2009). Overall, the effects of pesticides and insecticides on pollinators vary, and are often sub-lethal (Goulson *et al.*, 2015; Williams and Osborne, 2009). These stressors are likely having combined effects on pollinators (Goulson *et al*., 2015; Potts *et al*., 2010), which in some cases can cause these sub-lethal effects to become lethal (Goulson *et al.*, 2015). Studies into the combined effects of multiple stressors are limited, due to difficulties of conducting well-replicated studies (Goulson *et al.*, 2015).

Predators of foragers include birds, crab spiders and bee wolves; a predatory wasp which hunt bees (Dukas, 2005; Dukas and Morse, 2003; Goulson *et al.*, 2018a), which can have direct impacts on mortality or indirect effects on behaviour (Preisser *et al.*, 2005). Predators of bumblebee nests include wax moths and badgers. Wax moths are reported as being harmful predators of bumblebee nests, which infest colonies and consume nest material, brood and larvae (Alford, 1975; Pouvreau, 1973; Sladen, 1912). Although found in over 50% of nests, their presence did not appear to affect gyne production (Goulson *et al.*, 2018b). The European badger (*Meles meles*) is another key nest predator of bumblebees in the UK (Goulson *et al.*, 2018b;

Pease, 1898), destroying the whole colony. Only one recent study has recorded evidence of badger predation to bumblebee nests (Goulson *et al.*, 2018b), highlighting it to be the most common predator of bumblebee nests, and therefore in **Chapter 4** an artificial nest method was developed in order to quantify badger predation rates in different habitats. Computer models are a useful tool allowing combined effects of stressors to be tested. In **Chapter 5** the population model *Bumble*-BEEHAVE (Becher *et al.*, 2018) is used to test, for the first time, the top-down effects of badger predation on bumblebee populations under varying food availability.

1.3 Improving landscapes for pollinators

1.3.1 Habitat heterogeneity

Pollinators require a diverse mix of habitats to complete their lifecycle (Williams and Osborne, 2009). Habitats provide differing levels of nesting and floral resources (Cole *et al*., 2017; Osborne *et al.*, 2008b), which can affect the pollinator communities they support. Presence of semi-natural habitats within agricultural areas can increase the species richness and density of pollinators and natural enemies (Öckinger and Smith, 2007; Shackelford *et al.*, 2013), as well as increasing crop visitation rates for a variety of crop systems worldwide (Ricketts *et al.*, 2008). Isolation from semi-natural habitats leads to fewer floral resources, causing reduced offspring production and survival in solitary bees (Williams and Kremen, 2007). A study on the nectar provision of habitats across the UK found woodland and calcareous and neutral grassland habitats provided the highest amount of nectar per unit from the most diverse sources (Baude *et al.*, 2016). Timberlake et al., (2019) showed permanent pasture produce the greatest quantity of farmland nectar in their study, due to the large area they made up. However per unit area, field margins and hedgerows produced the most nectar. Hence it is not only the types of habitats present which is important, but the area they comprise within the landscape. For example Baldock *et al.*, (2019) showed that within urban areas, gardens and allotments contained the highest numbers of pollinators per unit area, but that overall gardens contributed most to the cities pollinator abundance due to making up a larger area. Many studies have looked at the benefits of residential gardens to pollinators (Majewska and Altizer, 2018; Salisbury *et al.*, 2015), and **Chapter 2** builds on this body of research by looking at the floral resources within botanical gardens within a rural agricultural landscape, and compare them to semi-natural farmland habitats. It is predicted that bumblebees within the agricultural landscape will be drawn to the large concentration of floral resources which are expected in the botanical gardens.

1.3.2 Case study: botanical gardens

Residential gardens have been shown to be good habitats for pollinators, providing both foraging and nesting resources. They can improve colony success of bumblebees (Goulson *et al.*, 2002a; Goulson *et al*., 2010), as well as increasing pollination services to agricultural habitats (Cussans *et al.*, 2010; Goulson *et al.*, 2010; Langellotto *et al.*, 2018; Potter and LeBuhn, 2015). Gardens provide a number of nesting sites favoured by bumblebees (Lye *et al.*, 2012; Osborne *et al.*, 2008b) and, alongside linear countryside features (such as fences and hedgerows), contain the highest nesting densities compared to all other habitats (Osborne *et al*., 2008b; Table 1.1). In urban areas, gardens make up a large portion of the urban green space (Baldock *et al.*, 2019; Loram *et al.*, 2007; Salisbury *et al.*, 2015). Botanical gardens are much larger than an individual residential gardens, and are likely to contain a more diverse mix of habitats. This gives them the potential to afford similar benefits to pollinators to those of residential gardens and potentially more. With their key role in plant conservation (Blackmore *et al.*, 2011; Hardwick *et al.*, 2011), botanical gardens contain a number of plant species outside of their native ranges (Pautasso and Parmentier, 2007). Non-native plant species are widely used by pollinators when available (Baldock *et al*., 2019; Hanley *et al*., 2014; Salisbury *et al*., 2015), and provide benefits such as extending the bloom period (Salisbury *et al.*, 2015; Stelzer *et al.*, 2010). Botanical gardens may therefore buffer pollinator populations at times when other habitats, such as farmland, are providing fewer resources. In **Chapter 2**, the use of botanical gardens and semi-natural farmland habitats by nest searching queens, and the floral resources they provide across the flight season is studied within a rural landscape.

1.3.3 Improving agricultural habitats

Agricultural habitats account for 72% of the land use in the UK (Department for Environment, Food and Rural Affairs Department of Agriculture, *et al*., 2018), making up a large area of a pollinators foraging range. Increasing the diversity of habitats within them can therefore provide benefits to pollinators.

This has been largely encouraged through agri-environment schemes (Ansell *et al.*, 2016) which can benefit pollinators through containing greater numbers of colonies (Wood, Holland, Hughes, *et al.*, 2015) and supporting males and queens (Carvell, *et al.*, 2011), but were not found to increase the number of bee and wasp species (Wood, Holland, and Goulson, 2015). Agricultural landscapes can be classified as 'simple', with large crop fields, or 'complex', with smaller fields and a higher amount of semi-natural habitats such as hedgerows and field margins surrounding them (Persson and Smith, 2011; Söderman *et al.*, 2018). Linear features such as hedgerows provide benefits to bumblebees (Kallioniemi *et al.*, 2017) through usage as nesting sites (Osborne *et al.*, 2008b) and for navigation (Cranmer *et al.*, 2012). Complex agricultural landscapes have greater plant species richness (Söderman *et al.*, 2018), and higher abundance and richness of wild bees and hoverflies (Happe *et al.*, 2018; Kennedy *et al.*, 2013; Kleijn and van Langevelde, 2006; Steffan-Dewenter *et al.*, 2002) which can lead to increased fruit set (Chateil and Porcher, 2015). This is potentially due to the provision of nesting resources provided by semi-natural habitats (Greenleaf *et al.*, 2007; Holzschuh *et al.*, 2007), allowing pollinators to complete their lifecycles. In contrast, simple agricultural landscapes contain floral resources which are less abundant and spaced further apart (Persson and Smith, 2011), and receive reduced pollination services of crops (Connelly *et al.*, 2015). In **Chapter 2**, these types of semi-natural habitats within complex farmland are compared to a habitat expected to be beneficial to pollinators; botanical gardens.

1.4 Study species: Bumblebees (*Bombus* spp.)

Bumblebees are central place foragers, with an annual lifecycle. This means that the position of their nest within the landscape affects colony success (Cresswell *et al.*, 2000). In the UK bumblebee populations have experienced declines (Williams and Osborne, 2009), with three species already extinct. This is a concern for the wildflowers and crops which depend on them for pollination (Klein *et al.*, 2007; Ollerton *et al.*, 2011), and this should therefore makes them a priority in terms of conservation and research efforts. Their lifecycle, and its relevance to the chapters in this thesis, is explored below.

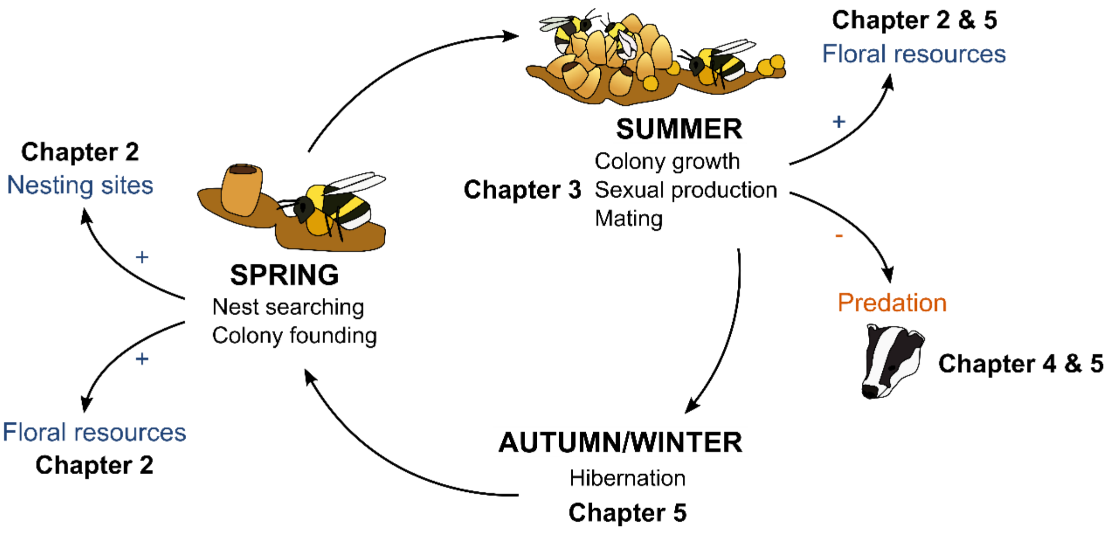

Figure 1.1 Diagram showing the bumblebee colony lifecycle and the aspects that each thesis chapter relates to. The main activities which occur at each stage of the lifecycle are listed, along with the key bottom-up and top-down factors that are studied which could have either a positive (+) or negative (-) impact on colony success and survival.

1.4.1 Queen emergence

The bumblebee lifecycle starts in spring, when mated queens which were produced the previous summer emerge from hibernation. Queens hibernate at varying depths and in a variety of habitats, including at the base of trees and in banks and slopes (Alford, 1975). Their hibernation site is likely to affect their emergence timing, with different species emerging at different times (Alford, 1975; Benton, 2006), however the exact reasons for this are still largely unknown. On emergence, queens require sufficient amounts of nectar and pollen to replenish their depleted fat reserves and to assist ovary development (Vogt *et al.*, 1998). Flowering tree and hedgerow species such as willows (*Salix* spp.) and blackthorn (*Prunus spinosa*), and wildflowers such as red (*Lamium purpureum*) and white dead-nettles (*Lamium album*) are of particular importance (Benton, 2006) during this time. **Chapter 2** explores the floral resources provided by botanical gardens and semi-natural habitats surrounding agricultural crop fields.

1.4.2 Nest searching and nest site preferences

Bumblebees have been found to nest in a variety of habitats, and either: underground, on the surface or above ground. Species may show specific nest site preferences, with some being more generalist nesters, such as *Bombus lucorum* (Svensson *et al.*, 2000) and others more specialist, such as *Bombus pascuorum*, which show strong preferences for grassland habitats (Fussell and Corbet, 1992; Kells and Goulson, 2003; Lye *et al.*, 2012) and are predominantly surface nesters. Other specialists include *Bombus hypnorum* which almost exclusively nest above ground and habitually nest in bird boxes in urban and suburban areas of the UK (Lye *et al.*, 2012). **Chapter 2** compares the numbers of nest searching queen bumblebees in botanical gardens and semi-natural habitats surrounding agricultural fields within a rural setting during the nest searching period and looks at whether nest searching is related to floral availability.

Once a suitable nest site has been found, the queen begins worker production, laying the initial eggs in a brood clump made of pollen (Sladen, 1912). High levels of spring forage are important during this stage, as queens need a lot of energy to incubate the brood to between 31-36˚C (Heinrich, 1974). Once the initial workers are hatched and developed, the queen ceases foraging and spends the rest of the season within the nest laying eggs. Thus begins the colony growth stage.

1.4.3 Colony growth stage

The growth and reproductive success of the colony depends on floral resource availability (Ogilvie and Forrest, 2017; Spiesman *et al.*, 2017). Food availability can affect the sizes of workers (Persson and Smith, 2011), with larger workers being better foragers; collecting more resources (Goulson *et al*., 2002b), having larger foraging ranges (Greenleaf *et al.*, 2007) and potentially a reduced mortality risk from ‘sit and wait’ predators (Romero *et al.*, 2011). This may enable colonies to grow larger, which then increases their chances of producing queens (Williams, *et al.*, 2012). In **Chapter 2**, after exploring queen nest searching choices between the two habitats (botanical gardens and semi-natural farmland habitats) the foraging resources provided by these two habitats across the colony growth stage is studied.

1.5 How are bumblebee populations studied?

For bumblebees, much of what is known about colony dynamics is through monitoring commercial colonies either in the laboratory or in the field (Goulson *et al*., 2002a; Spiesman *et al.*, 2017). These studies have provided useful information on the effects of food availability and surrounding landscape composition on colony growth and success, as well as on the impacts of wax moth infestation. However, it is unknown how transferable these results are to other bumblebee species, and to wild colonies which may respond differently.

The effects of floral abundance and diversity on the abundance and richness of pollinators is well studied (Senapathi *et al.*, 2016; Steffan-Dewenter *et al.*, 2002). Abundance, which for bumblebees is often a count of foraging individuals, is an important and useful measure, as recent studies on invertebrate declines have shown that biomass of certain groups have declined by over 75% (Hallmann *et al.*, 2017), with severe declines in tropical arthropod biomass also seen (Lister and Garcia, 2018). This can have knock-on effects down the food chain. For bumblebees, studying their abundance alone does not provide information on their effective population size. This is due to the colony as a whole being the reproductive unit (Ellis *et al.*, 2006), and thus to determine their effective population sizes research must be conducted into the numbers of colonies different landscapes can support (Darvill *et al*., 2004; Osborne *et al.*, 2008b), and the reproductive success of those colonies particularly in terms of gyne production. This is currently done through the use of genetics, however if more effective nest detection techniques were available (**Chapter 3**) this would allow the study of nests *in situ*.

1.5.1 Use of genetics

Genetics are an extremely useful tool in conservation, to understand the health of populations (DeYoung and Honeycutt, 2005). This can be especially important for small and isolated populations, where inbreeding is likely to occur (Ellis *et al.*, 2006; Goulson *et al.*, 2005). For example in the threatened *Bombus sylvarum*, where habitat loss and fragmentation are leading to reduced genetic drift (Ellis *et al.*, 2006). For bumblebees, microsatellite markers have been used in a number of applications: estimating foraging ranges (Carvell *et al.*, 2012), nesting densities (Darvill *et al.*, 2004; Table 1.1), colony survival (Goulson *et al.*, 2010) and queen dispersal and family lineage survival (Carvell *et al.*, 2017). However, the use of genetics does not give exact nest locations, which is an important step in understanding colony dynamics. **Chapter 3** aimed to fill this gap through testing a thermal camera as a novel bumblebee nest detection tool.

1.5.2 Monitoring of wild nests

Studies which involve the detection and direct observation of wild nests are important. In social insects whose nests are conspicuous, such as ants (Elmes *et al.*, 1991) and wasps (Starr, 1998) this is relatively straightforward. Managed pollinators, such as the honeybee (*Apis mellifera*), provide a useful system where observations can easily be carried out over time, making these the most well studied pollinator species (Vanbergen and Insect Pollinators Initiative, 2013). Bumblebees are also well-studied, however lack of reliable nest detection techniques means studies using wild colonies are rarely achieved. Locating bumblebee nests is difficult, due to their small colony sizes (Prŷs-Jones and Corbet, 2011). Currently the best nest detection method is human searches (O’Connor *et al.*, 2012), particularly using citizen science (Goulson *et al.*, 2018; Lye *et al*., 2012; Osborne *et al.*, 2008b), which can generate data on hundreds of nests. Although citizen science can generate data for large numbers of nests, it does not provide the same quality of data as localised human searches conducted by a researcher would, and in many cases the results are biased towards human-populated areas (Lye *et al.*, 2012; Osborne *et al*., 2008b). Novel nest detection methods include the training of sniffer dogs to locate bumblebee nests (Waters *et al.*, 2011), but this method was still not as effective as human searches (O’Connor *et al.*, 2012). Therefore, a gap was identified for a more effective nest detection technique which could be used in any habitat. In **Chapter 3** the effectiveness of a thermal camera to locate bumblebee nests using heat signatures, rather than visual (human searches) or olfactory (sniffer dog) cues was tested.

1.5.2.1 Case study: Observational study of bumblebee nests by Goulson et al. (2018a, 2018b)

A recent study utilised both human searches and citizen science to monitor bumblebee colonies and look at the effects of predation, wax moth infestation and disease on wild bumblebee colonies (Goulson *et al.*, 2018b, 2018a). Previously much of the information on bumblebee nest predators was old anecdotal records (Alford, 1975; Goulson, *et al.*, 2002; Pouvreau, 1973; Sladen, 1912). In the UK bumblebees do not have a large number of nest predators; these include birds, such as great tits (*Parsus major*), wax moths and the European badger (*M. meles*) (Goulson *et al.*, 2018a). Due to the relatively little known about bumblebee nest predation**,** in **Chapter 4 & 5** I/we develop and implement a novel artificial nest technique to quantify rates of badger predation on bumblebee nests. Using an artificial nest method allows the relative rates of predation in different habitats to be empirically quantified, in the absence of sufficient nest detection methods (**Chapter 3**).

Table 1.1 Nest density estimates for bumblebees (*Bombus*)

Species	**Nest density (nests per km^{-2})†**	**Method of detection**	**Habitat**	**Location**	**Reference**
Bombus distinguendus	31.1	Genetics	Agricultural	Scotland, UK	Charman et al., 2010
	53.3	Sniffer dog	Dunes	Scotland, UK	Waters et al., 2011
Bombus lapidarius	117.2	Genetics	Agricultural	Hertfordshire, UK	Knight et al., 2005
	26.7	Sniffer dog	Coastal	Scotland, UK	Waters et al., 2011
Bombus muscorum	186	Sniffer dog	Coastal	Scotland, UK	Waters et al., 2011
Bombus pascuorum	193	Genetics	Agricultural	Hertfordshire, UK	Darvill et al., 2004
	8	Genetics	Mixed	Lower Saxony, Germany	Herrmann et al., 2007
	67.8	Genetics	Agricultural	Hertfordshire, UK	Knight et al., 2005
	173	Genetics	Agricultural	Hertfordshire, UK	Knight et al., 2009
Bombus pratorum	26.1	Genetics	Agricultural	Hertfordshire, UK	Knight et al., 2005
Bombus terrestris	13	Genetics	Agricultural	Hertfordshire, UK	Darvill et al., 2004
	28.7	Genetics	Agricultural	Hertfordshire, UK	Knight et al., 2005
	89.2	Genetics	Agricultural	Hokkaido, Japan	Nagamitsu & Yamagishi, 2009

Bombus vosnesenskii	17.4	Genetics	Agricultural	Oregon, USA	Rao & Strange, 2012
	1.8	Genetics	Agricultural	Oregon, USA	Rao & Strange, 2012
Bombus spp.	35.9[†]	Citizen science	Gardens	UK wide	Osborne et al., 2008b
	11.4[†]	Citizen science	Grassland <10 cm	UK wide	Osborne et al., 2008b
	14.6[†]	Citizen science	Grassland >10 cm	UK wide	Osborne et al., 2008b
	10.8[†]	Citizen science	Woodland	UK wide	Osborne et al., 2008b
	37.2[†]	Citizen science	Fence line	UK wide	Osborne et al., 2008b
	29.5[†]	Citizen science	Hedgerow	UK wide	Osborne et al., 2008b
	19.9[†]	Citizen science	Woodland edge	UK wide	Osborne et al., 2008b

[†] Nest densities are in hectares

1.6 Thesis overview

The current thesis uses a combination of classical field techniques, novel methods and computer simulations to study some of the hidden aspects of bumblebee ecology. During spring, observations of nest searching queens were conducted in botanical gardens and semi-natural habitats surrounding agricultural crop fields (**Chapter 2**). The floral resources were also quantified during this time, as well as during the early and late summer, to understand how well these two distinct habitats were supporting bumblebees across the season. Novel techniques were then developed, tested and implemented in an attempt to locate wild bumblebee colonies (**Chapter 3**), and then find out what happens to established nests (**Chapter 4 & 5**). Thermal cameras were tested as a novel nest detection tool (**Chapter 3**) but found to be unsuccessful, in the absence of an effective nest detection tool a novel artificial nest technique was developed and used to study badger predation rates in two locations which have differing badger densities (Table B.1): Gloucestershire (**Chapter 4**) and Cornwall (**Chapter 5**). Finally, the population model *Bumble*-BEEHAVE (Becher *et al.*, 2018), was used to, for the first time, explore the combined effects of badger predation rates and food availability on bumblebee populations and colony outputs (**Chapter 5**).

Chapter 2: Botanical gardens and semi-natural farmland habitats provide complementary foraging resources across bumblebees' flight season

Bumblebees are central place foragers, meaning their situation within the foraging landscape affects their access to floral resources. Habitats contain different levels of floral resources throughout the season. This chapter assesses the use of two distinct habitats: botanical gardens and semi-natural farmland habitats by nest searching queens, and records their floral resource provision across the season. These habitats likely differ in the types and amounts of floral resources they contain; with semi-natural farmland habitats containing mostly native flowering plant species and botanical gardens containing large numbers of non-natives. This chapter aims to answer the question: **do botanical**

gardens provide an oases of resources across the season to sustain bumblebees in a rural, agricultural landscape?

Chapter 3: Testing the efficacy of a thermal camera as a search tool for locating wild bumblebee nests

Pollinators are under threat from multiple stressors, and the impacts of these are often explored using commercial bumblebee colonies. These provide insights into the mechanisms of how these stressors affect individuals and the overall colony, but are biased towards a couple of species and may not be representative of wild colonies. Studies of wild colonies are therefore needed, but are currently lacking due to inadequate nest detection methods. Bumblebee colonies are small and inconspicuous, making them difficult to locate and therefore study. Human searches are currently the best nest detection method (O'Connor *et al.*, 2012), but tend to find low numbers in a single season. Sniffer dogs were trained as a new method, but were no better at locating nests than human searches (O'Connor *et al.*, 2012; Waters *et al.*, 2011). These two methods rely on visual and olfactory cues to detect nests, and so a thermal camera was tested in this chapter, to find out: **can thermal cues from bumblebee nest traffic be used to locate wild bumblebee nests?** Thermal cameras reduce the visual complexity of the background, and therefore may be a more effective search tool than those relying on visual and olfactory cues. Published: Roberts, B. R. and Osborne, J. L. (2019) Testing the efficacy of a thermal camera as a search tool for locating wild bumblebee nests. Journal of Apicultural Research https://doi.org/10.1080/00218839.2019.1614724

Chapter 4: Quantifying the relative predation pressure on bumblebee nests by the European badger (*Meles meles*) using artificial nests

Populations can be regulated by both bottom-up (food availability) and top-down (disease and predation) mechanisms. Many studies have looked at bottom-up effects on colonies, showing that stable resources are beneficial (Williams *et al.*, 2012), with reduced food availability affecting queen production

(Rotheray *et al.*, 2017). Badgers are the main predator of bumblebee nests, destroying the whole colony (Goulson *et al.*, 2018b; Pease, 1898). Predation of bumblebee colonies has been recently reported within a wider study on bumblebee colonies (Goulson *et al.*, 2018a), but no studies have empirically attempted to quantify the rates of badger predation to bumblebee nests. Therefore, the current study asked **how does the rate of bumblebee nest predation by badgers differ between grassland and woodland habitats?** This was explored using a novel artificial nest technique in a high density badger population, showing that novel experimental methods can be an effective way of studying top-down stressors to bumblebee populations, in the absence of effective nest detection techniques. Submitted: Roberts, B. R., Cox, R., Osborne, J. L. (2019) Quantifying the relative predation pressure on bumblebee nests by the European badger (*Meles meles*) using artificial nests. Ecology and Evolution.

Chapter 5: Exploring the seasonal effects of badger (*Meles meles*) predation and varying food availability on bumblebee populations

Understanding the factors affecting colony growth and success is important for understanding the long-term impacts on populations. Top-down and bottom-up mechanisms are likely working together to impact colonies of pollinators (Goulson *et al.*, 2015; Potts *et al.*, 2010), but few studies have explored these mechanisms in the field due to the difficulty of designing well replicated studies (Goulson *et al.*, 2015). One way of over-coming these difficulties is through the utilisation of computer models which use realistic inputs about foraging behaviour and colony dynamics to study effects of various factors on bumblebee populations over time. The population model *Bumble*-BEEHAVE (Becher *et al.*, 2018) was used, to ask **how varying badger predation rates affect bumblebee colony outputs under high and low food availability?** This chapter also uses the artificial nest method developed in **Chapter 4** again to determine whether seasonality of badger predation can be detected, in line with natural badger dietary shifts.

Chapter 2

Botanical gardens and semi-natural farmland habitats provide complementary foraging resources across bumblebees' flight season

"*Everybody knows the burly, good-natured bumblebee. Clothed in her lovely coat of fur, she is the life of the gay garden as well as of the modestly blooming wayside as she eagerly hums from flower to flower, diligently collecting her nectar and pollen from the break to the close of day*."

– Sladen, 1912

Chapter 2: Botanical gardens and semi-natural farmland habitats provide complementary foraging resources across bumblebees' flight season

2.1 Abstract

Botanical gardens play a key role in the conservation of plant species, and as such can provide rich floral resources for pollinators, similar to residential gardens. Botanical gardens differ from semi-natural habitats in terms of their floral composition, containing a large number of non-native species. This can provide benefits to pollinators through increasing the floral diversity and extending the flowering season. In contrast, semi-natural habitats within farmland contain mostly native species. Floral resources fluctuate between habitats, with mis-matches often occurring between spring and summer resource peaks. Therefore multiple habitats may be required to fulfil pollinators' nutritional requirements across their flight season. In this chapter the numbers of nest searching queens, and the floral resources provided during spring, early and late summer by botanical gardens and semi-natural habitats surrounding agricultural fields are compared. It is predicted that botanical gardens, similar to residential gardens, will contain greater numbers of nest searching queens and have a higher abundance and diversity of floral resources, compared to semi-natural habitats within farmland. The results partly support this prediction, with higher numbers of nest searching queens recorded in botanical gardens, with them also providing a pulse of spring foraging resources. A non-native *Rhododendron* was an important foraging resource for queen bumblebees during this time. However, during early and late summer semi-natural habitats surrounding agricultural fields provided a higher abundance of floral resources. Despite containing contrasting floral assemblages, with 74% of food plant species being exotics within the botanical gardens, whereas in the semi-natural habitats within farmland 90.3% of food plants are native. There was no significant difference between the total abundance of bumblebees recorded between the two habitats, although the use of exotic and native plant species

differed. These two habitats may therefore be providing complementary resources across the bumblebee flight season.

2.2 Introduction

Botanical gardens play a key role in the conservation of plant species worldwide (Blackmore *et al.*, 2011; Hardwick *et al.*, 2011). Hulme, (2011) provides a definition of botanical gardens as: “an institution holding documented collections of living plants for the purposes of scientific research, conservation, display and education". As such botanical gardens are important habitats not just for species conservation but also for science (Chen and Sun, 2018). Botanical gardens occur worldwide, with over 4 million living plant collections containing over 80,000 species (Heywood, 1995). These types of gardens contain a high number of plant species outside of their native ranges (Pautasso and Parmentier, 2007). Due to this, many of the invasive plant species worldwide originated in botanical gardens (Dawson *et al.*, 2008; Hulme, 2011), which can have negative impacts on native biodiversity (Brown *et al.*, 2002). Non-native plants in botanical gardens also potentially increase the nectar and pollen available to pollinators; through providing access to plants with extended bloom periods (Salisbury *et al.*, 2015), and increasing floral diversity (Pautasso and Parmentier, 2007), which could in turn support more pollinators. In Cornwall, botanical gardens contain an important and unique heritage of plant species which were introduced by ‘plant hunters’ in the mid-1800s, with Cornwall’s mild climate allowing exotic plants such as *Rhododendron*’s, *Magnolia*’s and *Camellia*’s to thrive (Lamb, 2004; Smith and Mulholland, 2008). As well as floral resources, botanical gardens also have the potential to provide increased nesting sites, with bumblebees making use of features typical of gardens and populated areas such as bird boxes, walls, compost heaps and inside buildings (Fussell and Corbet, 1992; Lye *et al*., 2012; Osborne *et al*., 2008b).

Residential gardens have been studied in terms of their benefits to pollinators on a smaller scale (Majewska and Altizer, 2018). Residential gardens contribute a substantial amount to urban ‘green space’ (Baldock *et al.*, 2019; Loram *et al.*, 2007; Salisbury *et al.*, 2015) where they provide benefits to humans through ecosystem service provision (Cameron *et al.*, 2012) and to pollinators through providing key foraging and nesting resources (Baldock *et al*., 2019; Osborne *et al*., 2008b). The presence of gardens in a landscape can increase pollination services to agriculture through spill-over effects (Goulson *et al.*, 2010; Langellotto *et al.*, 2018), leading to increased seed set (Cussans *et al.*, 2010; Potter and LeBuhn, 2015), as well as benefitting bumblebee colonies through increasing survival, abundance and growth (Goulson *et al*., 2002a; Goulson *et al.*, 2010). For example *B. terrestris* colonies located within gardens grew larger than those in other habitats such as farmland (Goulson *et al*., 2002a). Due to their benefits in terms of food and nesting resources, it is hypothesised that botanical gardens, similar to residential gardens, will be better habitats for bumblebees compared to semi-natural farmland habitats; providing increased floral resources throughout the bumblebees' flight season and consequently supporting higher abundances of both nest searching and foraging bumblebees.

Gardens contain a large number of non-native flowering plant species, which can provide benefits to pollinators (Baldock *et al.*, 2019). Certain exotic plant species may be inaccessible to pollinators due to being morphologically incompatible or not providing the right cues to attract native insects (Corbet *et al.*, 2001). Despite this pollinators have been recorded successfully foraging on a range of non-native plant species (Hanley *et al.*, 2014; Salisbury *et al.*, 2015; Sikora *et al.*, 2016). Non-native plant species can benefit pollinators through extending the flowering season (Salisbury *et al.*, 2015); providing nectar and pollen at times of the year when native flowers are low in abundance. For example, the presence of exotic plants enabled *B. terrestris* colonies to successfully over-winter in parts of the UK (Stelzer *et al.*, 2010). When available, pollinators will utilise both native and non-native flowering plants flexibly (Baldock *et al.*, 2019; Salisbury *et al.*, 2015), and may show different preferences depending on their level of feeding specialisation (Hanley *et al.*, 2014; Stouffer *et al.*, 2014). Dietary specialists *Bombus hortorum* and *Bombus pascuorum* were found to prefer non-native plants from within their biogeographic ranges, whereas generalists such as *B. terrestris* and *Bombus* *pratorum* showed preferences for non-native plants from outside of their biogeographic region (Hanley *et al.*, 2014). This ability to utilise exotic resources is likely what has allowed *B. terrestris*, which is imported commercially worldwide for crop pollination (Velthuis and van Doorn, 2006), to be a successful invasive species around the world (Vanbergen *et al.*, 2018). Other studies showed that wild bee abundance and diversity was positively associated with native plant species (Fukase and Simons, 2016; Pardee and Philpott, 2014; Webber and Peterson, 2012). Thus, the relationship between pollinators and native and non-native plants can be complex, leading botanical gardens to be an interesting study site, especially in contrast to semi-natural farmland habitats which will contain mostly native plant species.

Agricultural areas are a prominent habitat within pollinators' foraging ranges, accounting for 72% of the UK landscape (Department for Environment, Food and Rural Affairs, *et al*., 2018) and often providing reduced foraging resources compared to semi-natural habitats (Cole *et al.*, 2017; Williams *et al*., 2012). Complex agricultural landscapes contained increased plant species richness (Söderman *et al.*, 2018), with small-scale habitat features such as uncultivated field margins being of particular importance. At the local scale, the presence of small-scale habitat features within the landscape was found to enhance bee abundance (Kennedy *et al.*, 2013), with features such as ditches able to support the rare bumblebee *Bombus muscorum* (Diekotter *et al.*, 2006). Small-scale features can also provide important early resources (Hannon and Sisk, 2009). Therefore the current study focussed on these small-scale features within the agricultural landscape, due to their importance for nectar provision (Timberlake *et al*., 2019) as well as navigation (Cranmer *et al.*, 2012).

Pollinators require nesting and foraging resources within the landscape in order to build and sustain successful populations. Nesting resources vary between habitats (see Table 1.1), with residential gardens (36 nests per ha^{-1}) and linear countryside habitats such as those used in the current study (20-37 nests per ha^{-1}) estimated to contain the highest nest densities (Osborne *et al*., 2008b). In order to support these nests, sufficient levels of food resources need to be present within pollinators' foraging ranges, and for the entirety of their lifecycles (Menz *et al.*, 2011; Russo *et al.*, 2013; Scheper *et al.*, 2015). Floral resource provision provided by habitats fluctuates seasonally as well as between habitats (Cole *et al*., 2017; Mandelik et al., 2012; Timberlake et al., 2019). Within agricultural landscapes, resource gaps have been observed for early spring, and for early and late summer (Timberlake *et al.*, 2019), at times when mass flowering crops have stopped flowering. Pollinators may respond to these resource fluctuations by utilising different habitats at different time points throughout the season in response to their floral assemblages (Cole *et al.*, 2017; Mandelik *et al.*, 2012). Therefore alternative habitats, such as gardens within these landscapes may buffer pollinator populations against periods of low resources (Goulson *et al.*, 2010).

Mass-flowering crops, such as oilseed rape (*Brassica napus*), provide resource pulses early in the colony lifecycle which can lead to higher bumblebee densities later in the season (Hass *et al.*, 2018; Westphal *et al*., 2003). However, their presence did not lead to increased nest abundance or queen production (Goulson *et al.*, 2010; Westphal *et al.*, 2009). Late season resource pulses provided by red clover were found to attract high abundances of bumblebees, including males and queens (Rundlöf *et al.*, 2014). However the authors did not measure effects at the colony level, and so it is possible that the increased abundance is seen due to bumblebees being highly attracted to abundant mass-flowering crops *(Osborne et al*., 2008a) rather than due to colony benefits. At a local scale, floral dominance of a few key plant species was found to be more important than overall floral abundance for colony growth and reproduction of *B. impatiens* in the US (Spiesman *et al.*, 2017). A similar behaviour was observed in *B. terrestris,* which was found to collect the majority of their pollen from a few abundant plant species (Kämper *et al.*, 2016; Leonhardt and Blüthgen, 2012), with colony growth being more affected by forage availability rather than quality. Whereas *B. pascuorum* foragers were more likely to collect pollen from a mixture of plant species during a single foraging trip (Leonhardt and Blüthgen, 2012) and so may not benefit from resource pulses of single dominant species. Thus, bumblebee species may be affected differently by reduced floral diversity, and habitat heterogeneity is likely necessary for a diverse range of pollinators to achieve their nutritional requirements across the entire nesting season (Hass *et al*., 2018; Jha and Kremen, 2013a).

To understand how bumblebees are utilising two distinct habitats throughout their flight season, seasonal bumblebee and foraging resources in botanical gardens and agricultural landscapes were studied; with a focus on the semi-natural habitats surrounding crop fields. Botanical gardens were predicted to provide a more abundant and diverse floral resource, and plentiful nesting opportunities for bumblebees within an agriculturally dominated landscape, enabling them to support higher abundances and diversities of bumblebees throughout the season. Phenological surveys of bumblebees and plants were conducted during three survey periods: early spring, early summer and late summer, with a specific nest searching queen survey conducted over a 6-week period in the spring. Previous studies focus either solely on nest searching (Kells and Goulson, 2003; Svensson *et al.*, 2000) or floral resources (Timberlake *et al*., 2019). This chapter looks at both of these to build a broader picture of how habitats in a rural landscape can sustain bumblebees throughout the various stages of colony development.

2.3 Methods

2.3.1 Study sites

Data were collected across eight sites in Cornwall, UK (Lat. 50.503632, Long. -4.652498; Fig. 2.1). Each site consisted of a botanical garden and a farmland habitat pair. Botanical gardens used in this study were open to the public, and were sites dedicated to the collection and cultivation of a wide range of usually non-native plant species.

Surveys in botanical gardens were carried out along established paths and often encompassed flower beds. Farmland surveys were carried out along the semi-natural habitats surrounding the agricultural fields, often encompassing Cornish hedgerows, regular hedgerows and grass banks. The agricultural fields mainly consisted of cereals or bare earth, with the only flowering crop being oilseed rape (*Brassica napus*) which was 70% in flower during early spring, reducing to 5% by early summer and had finished flowering completely by late summer. Distances between the botanical garden and farmland habitats at each site ranged from 0.32-1.34 km, and the distance between sites ranged from 2.56-8.30 km. Distances between the sites were large enough that it was likely they were being used by distinct bumblebee colonies. Sufficient distances between the two habitats was important to reduce the chances of recording the same bumblebees during transects, despite it being feasible that bumblebees from the same colonies would have been using the two habitats within each site.

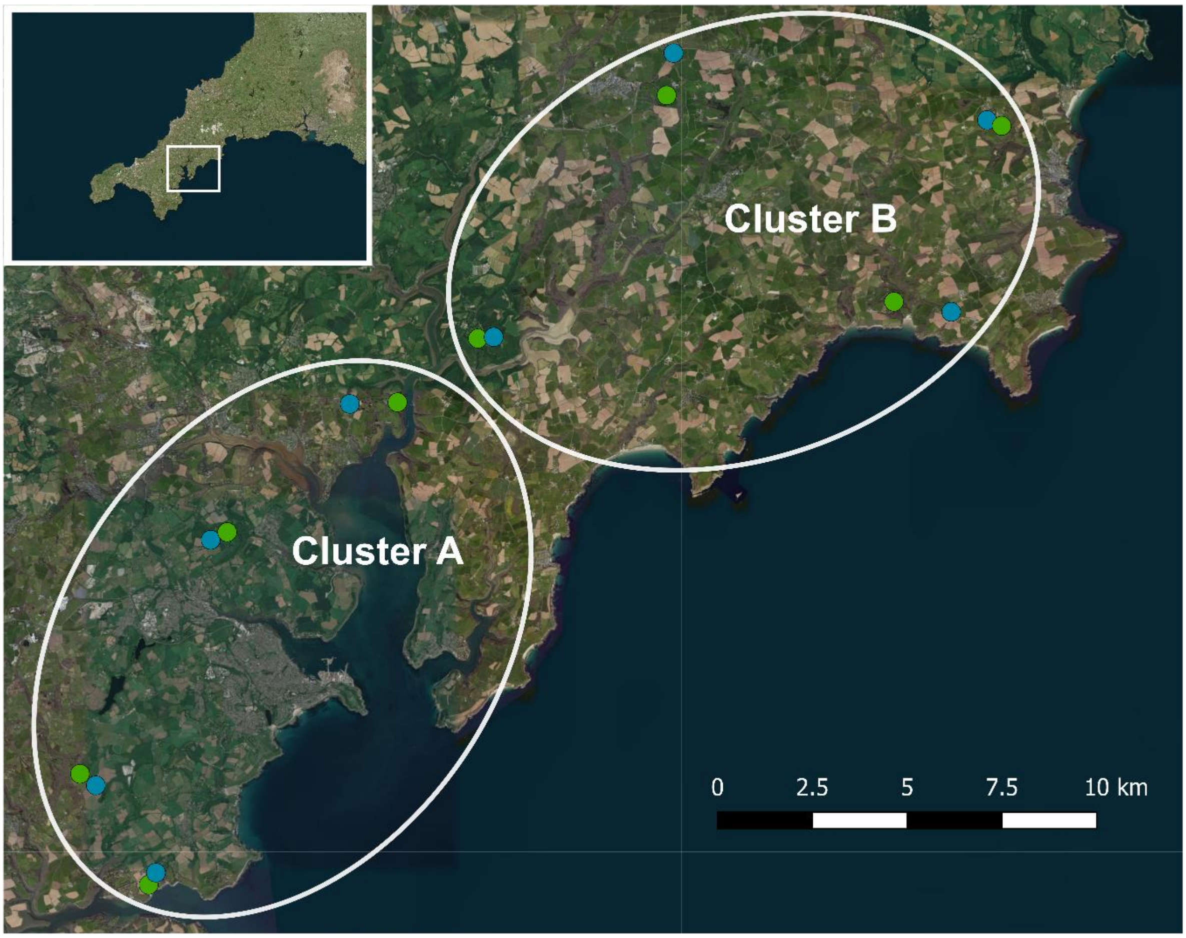

Figure 2.1 Map showing the eight study sites used in the nest searching and phenological surveys. The inset in the top left shows the map of Cornwall, UK with the white box representing the study area which is then enlarged. The botanical garden (green circle) and semi-natural farmland (blue circle) habitats within each site are shown within either Cluster A or Cluster B. Clusters were allocated in order to allow a randomised survey design, with sites within the same cluster always being surveyed together.

2.3.2 Data collection timings

Data was collected over four different survey periods in 2016 (Table 2.1). The first round of phenological surveys, where bumblebees and flowering resources were recorded, were conducted from 21st March-1st April (early spring). Spring nest searching queen surveys were conducted over a 6-week period from 18th April-3rd June. [**Note:** There was a mis-match between the timings of these surveys, due to nest searching surveys initially starting on 15th March but being stopped due to low numbers of nest searching queens. Nest searching queen surveys were not restarted until the 18th April due to poor weather conditions, after the early spring phenological surveys had already been conducted]. The second round of floral surveys were conducted between 6-18th June (early summer) and the final round between 17-23rd August (late summer).

2.3.3 Floral abundance and diversity

When performing floral counts, the number of flower units on each transect was counted for each plant species within the 2 m wide x 2 m high transect area in accordance with Carvell et al., (2004), where one flower 'unit' was classed as an umbel (e.g. *Anthriscus sylvestris*), a head (e.g. *Trifolium repens*) or a capitulum (e.g. *Cirsium arvense*).

All non-horticultural plants were identified to species or genus using Rose and O'Reilly, (2006). They were then classified as either 'native' or 'exotic'; species which were found in the wildflower key but were noted as an introduced species such, as *Pentaglottis sempervirens*, were classified as 'exotic'. For the horticultural species found within botanical gardens, 'exotics' were identified to species and variety where possible through plant labels in the garden, or from photographs and then using the Royal Horticultural Society's plant finder https://www.rhs.org.uk/plants/search-Form. Where exotic plant species could only be identified to genus, different varieties were discerned visually, often by colour or morphological differences, and were given unique number identifiers e.g. '*Rhododendron* spp1'.

During all surveys, plants visited by foraging bumblebees were classified as food plant species. Only data for flowering plants categorised as food plants were used for analysis purposes. This was done due to many horticultural plant species in the botanical gardens providing no benefits to pollinators due to being morphologically unsuitable (Corbet *et al.*, 2001). Only analysing data for plants recorded as food plants allow for more accurate comparison between the two habitats.

2.3.4 Spring nest searching queen surveys

Nest searching queens surveys were conducted to compare their abundance and diversity between the two habitats. Botanical gardens are predicted to support higher abundances and diversity of nest searching queens through containing richer floral resources, and a greater diversity of nest sites based on previous studies in residential gardens (Osborne *et al.*, 2008b; Salisbury *et al*., 2015). Two transects were walked at each site and in each habitat (botanical gardens and farmland); one along a Cornish hedge, a feature made of stone with earth packed into it with hedging or trees planted along the top which is typical of hedgerows in Cornwall, and one along a bank, which included earth banks, grassy banks and flower beds across the sites. These features were chosen as queen bumblebees have been found to preferentially nest search along linear features such as these (Kells and Goulson, 2003; Svensson *et al.*, 2000). Transect lengths ranged between 58.5-100 m depending on the length of the Cornish hedgerow or bank. Transects were walked at a steady pace, with surveys lasting for a total of 20 minutes; 10 minutes walking in one direction and 10 minutes in the other. Transects were walked when wind speed was no more than 5 on the Beaufort scale. Due to the time of year, temperatures ranged from 6-22°C.

Transects were walked once per week for a total of six weeks, between 0830 and 1730 h. The order in which each site, habitat and transect was visited was randomised using an online randomiser (www.random.org). To allow surveys to be performed within the time frame, sites were split into two clusters grouped by geographical location (Fig. 2.1). There were four sites in each cluster, with sites in the same cluster always being surveyed together. During each transect walk, the number of queen bumblebees observed nest searching were counted and identified to species where possible on the wing. Queens observed foraging were also recorded, along with the plant species they were observed visiting. Floral abundance (the number of flower units) and diversity (the number of food plant species) were recorded in accordance with the methods set out in section 2.3.3. Air temperature, wind speed and cloud cover were also recorded.

Table 2.1 Survey design and timings of the two experiments conducted for this study; nest searching and phenological surveys. All surveys were carried out at the same eight sites, within the same two habitats (botanical gardens and semi-natural habitats within farmland).

Date	Survey type	N transect per habitat	N surveys at each site	Transect length
21st March – 1st April	Early spring (phenological survey)	1	1	500 m
18th April – 3rd June	Spring (nest searching survey)	2	6	58.5-100 m*
6-18th June	Early summer (phenological survey)	1	1	500 m
17-23rd August	Late summer (phenological survey)	1	1	500 m

* Transect lengths differ for the nest searching survey as they were conducted along two linear features: a Cornish hedge and a bank, and in some instances the features, and therefore the transect length, were < 100 m. This is taken into account during analysis.

2.3.5 Phenological surveys (early spring, early summer and late summer)

To understand how bumblebees use the two habitats throughout the flight season, and to determine the foraging resources provided by each habitat, phenological surveys were conducted. Bumblebee and flower abundance and diversity were recorded across the eight sites and two habitats over three survey periods during the early spring, early summer and late summer (Table 2.1). Transects were 500 m in length, in the majority of sites transects were set out over a 1 km length, with a 100 m transect walked every other 100 m. For one farmland habitat, the transect was a continuous 500 m due to restricted access. This was done in order to cover a larger area of each site. During each survey period, transects were walked once and were conducted between 0830 and 1800 hours. All bumblebees observed along transects were recorded to species and caste where possible. If they were observed foraging, the flower species they were visiting was recorded. Floral abundance and diversity was recorded in accordance with the methods set out in section 2.3.3. Temperatures ranged from 9-24˚C (early spring: 9-17˚C, early summer: 13-24˚C and late summer: 18-22˚C), cloud cover was between 10-100% and wind speeds were 5 or below on the Beaufort wind scale.

2.3.6 Analysis

2.3.6.1 Spring nest searching queen surveys

2.3.6.1.1 Floral abundance and diversity of bumblebee food plants

Floral abundance and diversity of bumblebee food plants during the nest searching queen surveys were analysed in R (version 3.5.2; R Core Team, 2017). Generalised linear models were built to analyse the effects of 'habitat' and 'week' on floral 'abundance' and 'diversity' of bumblebee food plants. 'Week' was included as a fixed effect in these models, due to floral resources showing weekly variation. For both models, 'transect ID' was nested within 'site' as a random effect, with an observation level random effect 'OLRE' being included in the floral abundance model to account for overdispersion (Browne *et al.*, 2005). 'Transect length' was included as a logged offset to account for varying transect lengths. Both models were fitted with a Poisson family with a square-root link function.

2.3.6.1.2 Nest searching queen abundance and diversity

Analysis was done using the software R (version 3.5.2; R Core Team, 2017). Linear models were built to analyse the effects of 'habitat' and either 'floral abundance' of bumblebee food plants or 'floral diversity' of bumblebee food plants, and their two-way interaction, on nest searching queen 'abundance' and 'diversity'. These were analysed in separate models due to correlation between the two variables. In order to increase the sample sizes, and due to visual observations of the data indicating no differences between the two features surveyed (Cornish hedgerows and banks), data for the response and fixed effects were created by combining data across the two transects conducted in each habitat, as well as across the six weeks. This provided a sum of nest searching queens and an average floral abundance and diversity for each transect. This was necessary, as the average number of queens per transect for each week was only 1.5 and 0.9 for botanical gardens and semi-natural farmland habitats respectively, which meant it was not possible to sufficiently detect any effects. Whereas when summed across weeks the average number of queens per transect were 8.88 and 5.38 for the two habitats respectively.

All models were fitted with a Gaussian distribution, with no random effects due to singularity occurring when 'site' or 'transect ID' were included in the model. Global models with the fixed effects and their interactions were built for all four models (Tables A.3 & A.4). 'Transect length' was included as a logged offset in each of the global models to account for the varying transect lengths, and the floral 'abundance' and 'diversity' variables were scaled accordingly.

2.3.6.3 Phenological surveys (early spring, early summer and late summer)

2.3.6.3.1 Floral abundance and diversity of bumblebee food plants

Floral abundance and diversity of bumblebee food plants from the phenological surveys were analysed in R (version 3.5.2; R Core Team, 2017). Generalised linear models were built to analyse the effects of 'habitat' and 'survey period' and their two-way interaction on floral 'abundance' and 'diversity' of bumblebee food plants. For the 'floral models, 'transect ID' was included as a random effect, along with an observation level random effect 'OLRE' to account for overdispersion (Browne *et al.*, 2005). The floral abundance model was fitted with a Poisson family with a square root link function. For the 'floral diversity' models, only 'site' was included as a random effect. This model was fitted with a Poisson family with a log link function.

2.3.6.3.2 Bumblebee abundance and diversity

Bumblebee abundance (all castes, all species) and diversity (the number of bumblebee species recorded per transect) collected during the phenological surveys were analysed using the statistical software R (version 3.5.2; R Core Team, 2017). Generalised linear models were built to analyse the effects of ‘habitat’, ‘survey period’ and either ‘floral abundance’ or ‘floral diversity’ of bumblebee food plants, and their combined two-way interactions, on bumblebee ‘abundance’ and ‘diversity’. ‘Survey effort’ was included as a logged offset to account for variable survey times for each transect.

For the bumblebee abundance models, an observation level random effect (‘OLRE’) was included to account for over-dispersion (Browne *et al.*, 2005). When ‘floral abundance’ of bumblebee food plants was included in the model as a fixed effect, ‘transect ID’ was nested within ‘site’ as a random effect, and for the bumblebee abundance models with ‘floral diversity’ of bumblebee food plants included as a fixed effect, only ‘transect ID’ was included as a random effect to enable model convergence. Both bumblebee abundance models were fitted with a Poisson family and a square-root link function.

For the bumblebee diversity models which included ‘floral abundance’ of bumblebee food plants as a fixed effect, only ‘site’ was included as a random effect and models were fitted with a Poisson family and square-root link function. For the bumblebee diversity models including ‘floral diversity’ of bumblebee food plants as a fixed effect no random effects were included but instead ‘transect ID’ was included as a fixed effect in the global model. This model was fitted with a Poisson family and a log link function.

2.3.6.4 Model selection

Model selection for both nest searching and phenological surveys was performed using dredge from the MuMIn package (Bartoń, 2017) which compares all possible models to a global model and uses Akaike’s Information Criterion to select the models which best explain the data. Models with a ΔAIC <2 were selected as the top models and their coefficients can be seen in Appendix A (Tables A.1-A.10).

2.3.6.5 Plant-pollinator networks for qualitative analysis

Plant-pollinator networks were created for each habitat and each survey period using data from the phenological surveys. Foragers were grouped by caste and species, and their abundance and the abundance of bumblebee food plant species were used to visualise forager networks and identify foraging preferences. Networks were built using the econullnetr package (Vaughan *et* *al.*, 2018), which builds a null network of plant-pollinator interactions based upon the relative abundance of food plant resources. It then compares this null network to the observed network to identify plant species that are visited significantly more or less frequently than expected from the null model, thereby identifying foraging preferences or resource avoidance. As the phenological data was collected across eight locations, plant and bumblebee data were inputted for each site. This ensures that pollinators are not able to form links with plants from outside of the site they were recorded when building the null networks. Null networks were generated with 1000 simulations, and then visualised within econullnetr using the plotweb function from the bipartite package (Dormann *et al.,* 2019).

2.4 Results

2.4.1 Spring nest searching queen surveys

2.4.1.1 Floral abundance and diversity of bumblebee food plants

During the six weeks of nest searching queen surveys, floral abundance of bumblebee food plants in both habitats was low at the start of the surveys (Fig 2.2). In botanical gardens floral abundance peaked during week three and four before decreasing in week six, whereas in the semi-natural farmland habitats floral abundance increased each week (Fig. 2.2). Floral diversity of bumblebee food plants was relatively consistent across weeks for the two habitats, with farmland plant diversity increasing from an average of 8.14 species in week one and peaking at 13.1 species in week six. Botanical gardens had a higher diversity than farmland throughout with a mean floral diversity of 13.6 species in week one, and peaking at 21.2 species in week five before reducing to 19 species.

Overall, floral abundance of bumblebee food plants was highest in the botanical gardens (z=2.30, p=0.02; Table A.2), and there was a significant interaction between habitat and week. Floral abundance was significantly lower in botanical gardens in week six compared to semi-natural habitats within farmland (z=-4.04, p<0.001; Fig. 2.2, Table A.1).

When looking across the six week survey period, floral abundance was significantly higher in week three, four and five compared to week one (Week three: z=2.55, p=0.01; Week four: z=3.50, p<0.001; Week five: z=4.62, p<0.001; Fig. 2.2, Table A.1). Overall floral diversity was significantly higher in week five compared to week one (z=2.20, p=0.03; Table A.2), and there was no difference between habitats for floral diversity (z=1.01, p=0.314; Table A.2).

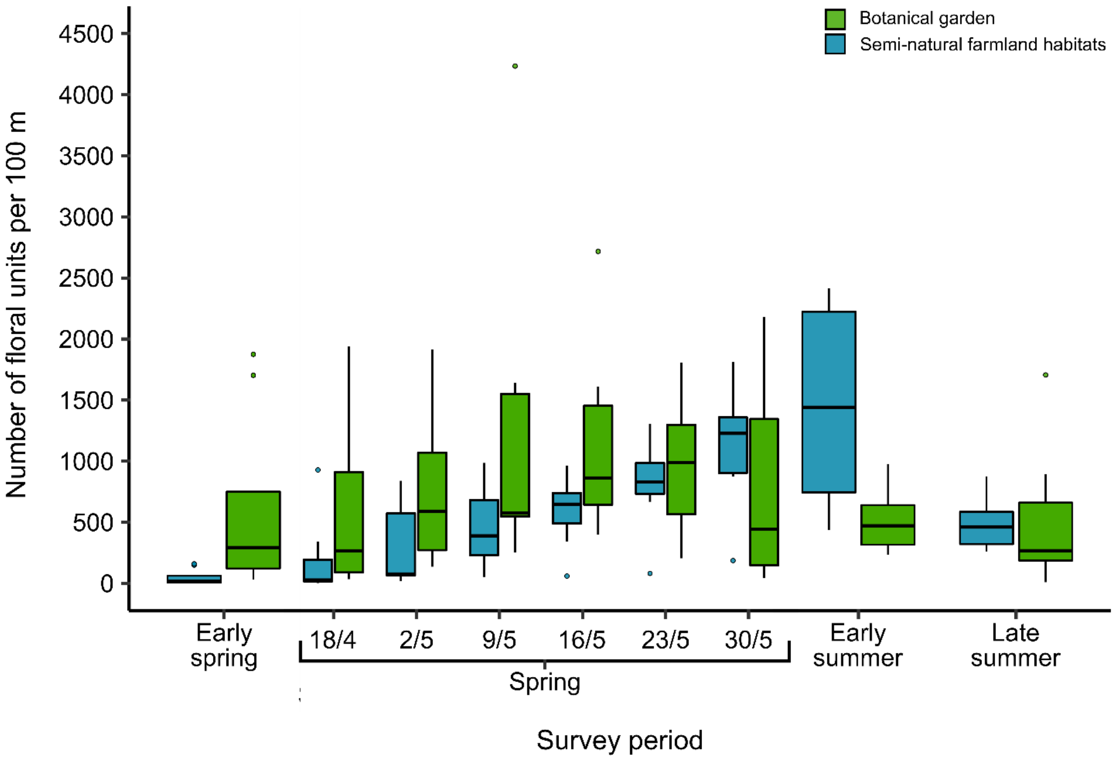

Figure 2.2 Boxplot showing the number of floral units of bumblebee food plants, standardised per 100 m to allow data from both the nest searching (spring) and phenological surveys (early spring, early summer and late summer) to be directly compared. Data from the nest searching survey is slotted into the phenological data, with week one (18th April) through to week six (30th May) being shown.

2.4.1.2 Nest searching queen abundance and diversity

Over the six week survey period 116 nest searching queens of six species were observed; 72 in botanical gardens, and 44 in farmland habitats. *B.*

terrestris accounted for 42% of observations, with *B. pascuorum* and *Bombus lucorum* accounting for a further 36%. The remaining 22% of observations comprised of *Bombus lapidarius*, *Bombus hortorum* and *B. hypnorum*. No *Bombus pratorum* queens were recorded during the study, suggesting that queens of this early emerging species had already finished nest searching by the time the study began.

Significantly higher abundances of nest searching queens were observed in botanical gardens compared to semi-natural areas surrounding agricultural fields, when floral abundance of bumblebee food plants is accounted for (t=2.15; p=<0.01; Fig. 2.3, Table A.3). There was no difference in diversity of nest searching queens between the two habitats (t=1.25; p=0.231; Table A.4). There was also no effect of floral abundance or diversity of bumblebee food plants on nest searching queen abundance or diversity (Queen abundance, floral abundance: t=-1.58, p=0.14 (Table A.3); Queen diversity, floral diversity: t=1.76, p=0.10 (Table A.4)), with it being selected out of the other models.

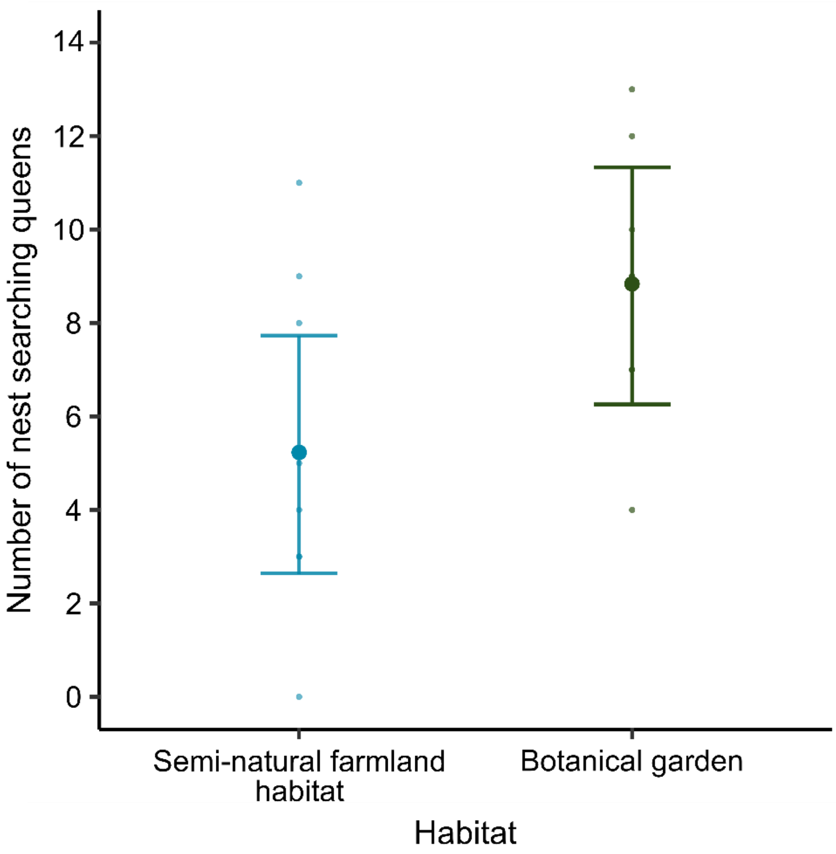

Figure 2.3 Numbers of nest searching queens for each of the eight sites and two habitats summed across the six weeks. Predicted data from the top model is shown with confidence intervals (Table A.3). Raw data are plotted as points.

2.4.2 Phenological surveys across the bumblebee nesting season

2.3.2.1 Floral abundance and diversity

Across the three survey periods and the two habitats (Table 2.1), 225,973 floral units from 442 flowering plant species were recorded. Botanical gardens contained 124,974 floral units from 394 species and farmland contained 100,999 floral units from 109 flowering plant species. The numbers per survey period for each habitat can be seen in Table 2.2.

Table 2.2 The floral abundance and floral diversity for botanical gardens and semi-natural farmland habitats recorded during each of the three survey periods. The total number of floral units and flower species are shown, with the numbers for those classified as bumblebee food plants shown in brackets.

	Botanical gardens		Semi-natural farmland	
Survey period	Floral units	Floral species	Floral units	Floral species
Early spring	31,106 (24,224)	158 (18)	4,727 (1,960)	28 (6)
Early summer	35,846 (20,551)	205 (49)	69,332 (58,852)	62 (17)
Late summer	37,654 (20,455)	130 (40)	26,940 (20,092)	66 (26)

Floral abundance of bumblebee food plants was significantly lower in botanical gardens compared to the semi-natural farmland habitats during both early and late summer (Early summer: z=-5.19, p<0.001; Late summer: z=-2.99, p<0.01; Fig. 2.2, Table A.5). Botanical gardens contained a significantly higher floral diversity of bumblebee food plants (z=2.32, p=0.02; Table A.6), with overall floral diversity being higher during the early and late summer compared to early spring (Early summer: z=6.43, p<0.001; Late summer: z=6.98, p<0.001; Table A.6).

2.4.2.2 Bumblebee abundance and diversity

Across the three survey periods, 467 bumblebees of 9 species were recorded (seven true bumblebees and two ‘cuckoo’ species, *Bombus Psithyrus* spp). 208 of these were recorded in the botanical gardens (early spring 39, early summer: 101, late summer: 68) and 259 were recorded in the semi-natural farmland habitats (early spring 20, early summer: 107, late summer: 132). Across the three survey periods, species composition between the sites differed, although was not tested statistically. *B. pratorum* was the most abundant species during early summer, with three times as many being recorded in botanical gardens compared to farmland (56 and 17 individuals respectively). During late summer *B. pascuorum* was the most abundant species, with almost double the number of individuals occurring in the farmland habitat compared to botanical gardens (85 and 43 individuals respectively).

17 nest searching queens of four species were recorded; nine queens of two species (*B. pratorum* and *B. terrestris*) in botanical gardens (early spring = 8, early summer = 1) and eight queens of three species (*B. pascuorum*, *B. lapidarius* and *B. terrestris*) in farmland during early spring. Due to low numbers, analysis was not conducted on this data.

During the phenological surveys there was no significant difference in bumblebee abundance or diversity between the two habitats (Floral abundance: $z=-1.86$, $p=0.06$ (Table A.7); Floral diversity: $z=1.39$, $p=0.16$ (Table A.8)), with habitat being selected out of models which included floral abundance as a fixed effect. Floral diversity of food plants had a positive effect on both bumblebee abundance and diversity (bumblebee abundance: $z=7.92$, $p<0.001$ (Fig. 2.4b, Table A.7); bumblebee diversity: $z=2.57$, $p=0.01$ (Table A.8)). Floral abundance of food plants had a positive effect on bumblebee diversity only ($z=2.15$, $p=0.03$; Fig. 2.4d, Table A.9).

Early and late summer surveys contained higher abundance and diversity of bumblebees compared to early spring, when abundance of bumblebee food plants was taken into account (Bumblebee abundance: (early summer) $z=5.47$, $p<0.001$, (late summer) $z=5.92$, $p<0.001$ (Fig. 2.4a, Table A.10); Bumblebee diversity: (early summer) $z=4.07$, $p<0.001$, (late summer) $z=3.10$, $p<0.01$ (Fig. 2.4c, Table A.9)).

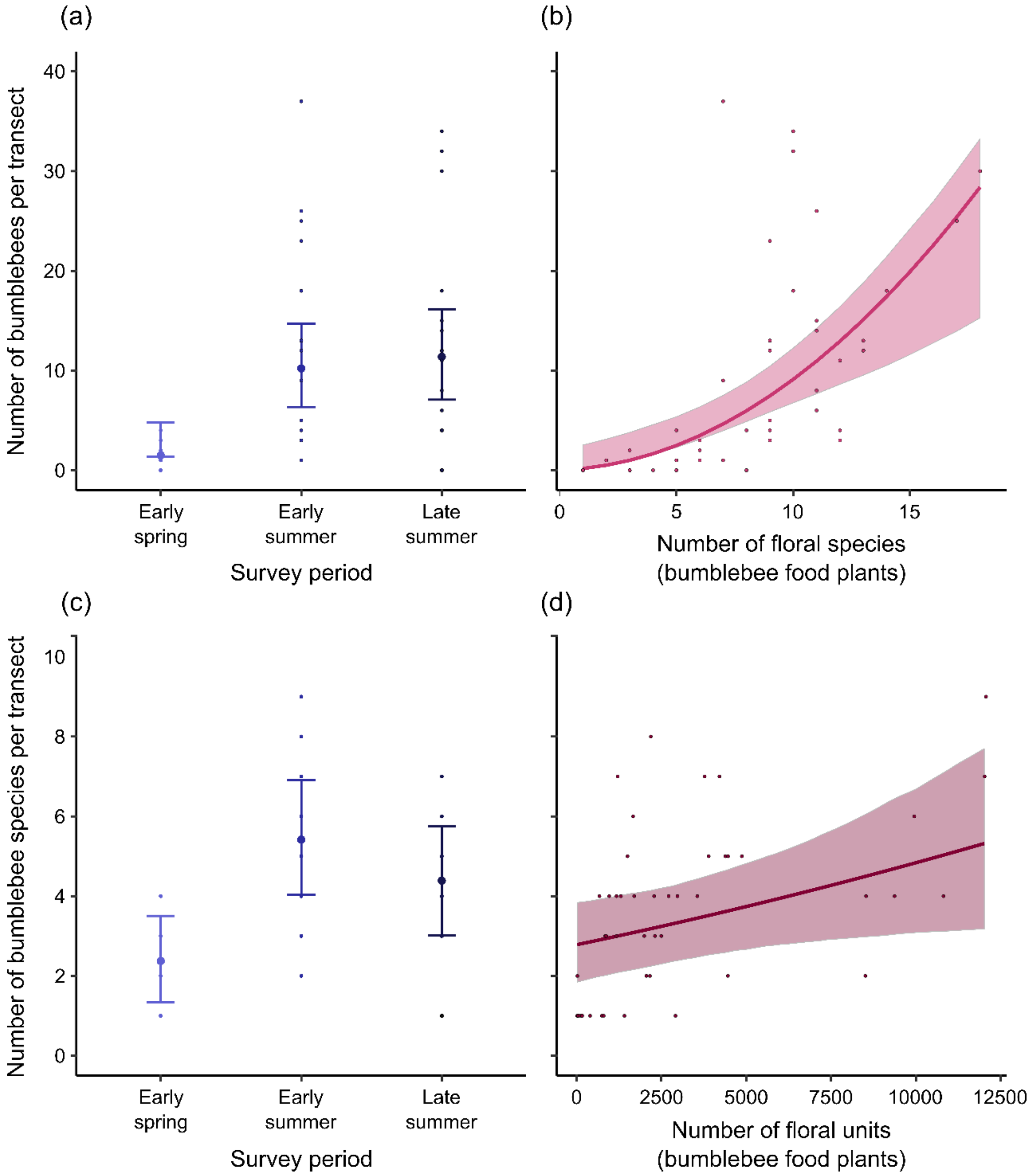

Figure 2.4 Panel showing how bumblebee abundance (a) & (b) and bumblebee diversity (c) & (d) are affected by survey period (a) & (c), floral diversity (b) and floral abundance (d). Predicted data from the top models are shown with confidence intervals, and the raw data are presented as points.

2.4.3 Foraging preferences of bumblebees across the bumblebee nesting period

Within the semi-natural farmland habitats the majority of food plant species were native (90.3%), whereas 74% of food plant species in botanical gardens were non-native and from a variety of geographic ranges (Fig. 2.5). For each of the habitats and survey periods plant-pollinator networks were used to look at which plant species were being visited more or less often than expected, and these results are presented in Fig. 2.6 and Fig. 2.7.

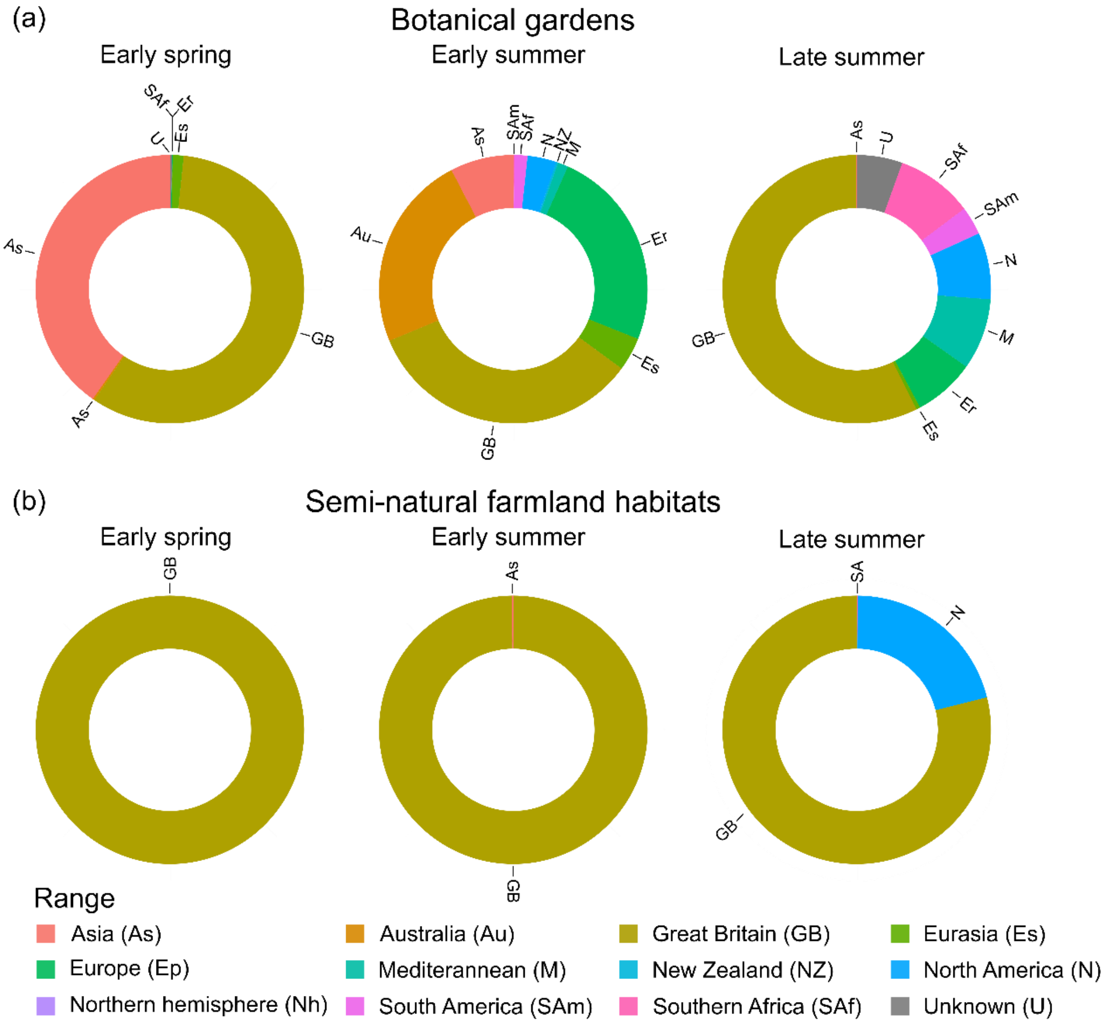

Figure 2.5 Doughnut plot showing the proportion of floral units for bumblebee food plant species within each geographic range, recorded during the phenological surveys in the (a) botanical garden and (b) semi-natural farmland habitats across the three survey periods: early spring, early summer and late summer.

2.4.3.1 Botanical gardens

74% of food plant species recorded in botanical gardens were exotics, with 82 - 90.9% of visits across the three survey periods being to exotic plant species. Despite their high usage, exotic plants accounted for a similar percentage of floral units to native species, with 42.1 - 66.4% of flower units being from exotic plant species across the three survey periods. Plant-pollinator networks showed that *Geranium robertianum* and *Teucrium scorodonia* were the only native species which were visited more often than expected, being favoured by *B. pratorum* males and *B. pascuorum* workers respectively (Fig. 2.6). Exotic species that were visited more often than expected included *Rhododendron* ‘Endsleigh pink’, *Rhododendron obiculare* white and *Iris sibirica* ‘Flight of the butterflies’ in early summer and *Rudbeckia* spp1 and *Hydrangea* spp5 in late summer (Fig. 2.6).

During early spring (21st March – 11th April) 11 bumblebees of four species (*B. hypnorum, B. pascuorum, B. pratorum* and *B. terrestris*) were recorded foraging (Fig. 2.6). Seven plant species were used by bumblebees, out of a total of 157 flowering plant species. Of these, all except one were exotics, with 85.4% of all flowering plant species being made up of exotics during this survey period. The most abundant species visited by bumblebees was *Azalea* spp14 (Flower units = 7,572), which was also the most commonly visited plant species during this survey period (Fig. 2.6).

During early summer (6-18th June) 95 bumblebees of seven species (*B. hortorum*, *B. hypnorum*, *B.lucorum, B. pascuorum*, *B. pratorum*, *B. terrestris* and *B. vestalis*) were recorded foraging (Fig. 2.6). Bumblebees were recorded visiting 31 plant species out of a total of 203 flowering plant species. 71.9% of all flowering plant species, and 87.1% of the plants which were visited by foragers during this survey period were exotics. The most abundant flowering plant species was the native species *Silene dioica* (Flower units = 3,106), which the plant-pollinator network showed to be visited less often than expected (Fig. 2.6). The second most abundant food plant species was *Nepeta* ‘six hills giant’ (Flower units = 2,507) a Mediterranean species, which was also visited less often than expected (Fig. 2.6).

During late summer (17-23rd August) 61 bumblebees of five species (*B. hortorum, B. pascuorum*, *B. pratorum*, *B. terrestris* and *B. terrestris* agg.) were recorded foraging (Fig 2.6). Bumblebees were observed foraging on 27 plant species out of a total of 130 flowering plant species. 63.1% of all flowering plant species were exotics. The most abundant plant species was a native species, *Circaea lutetiana* (Flower units = 10,504) which was only visited by a single *B. terrestris* agg. Worker (Fig. 2.6). The next most abundant species were three exotics with similar abundances to each other: *Crocosmia* x *crocosmiiflora* (Flower units = 1,547), *Geranium wlassovianum* (Flower units = 1,714) and *Nepeta 'six hills giant'* (Flower units = 1,389).

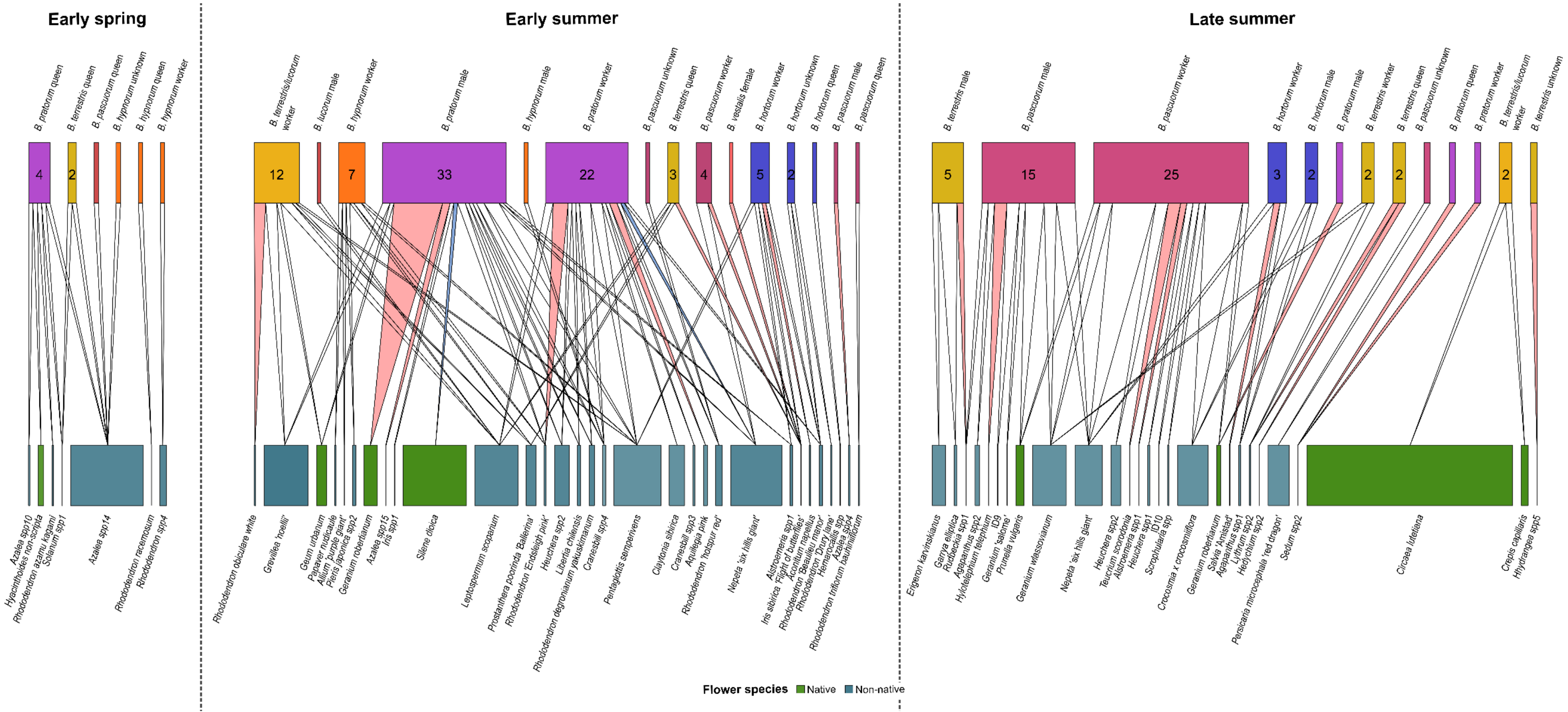

Figure 2.6 Plant-pollinator network for the plant species bumblebees were foraging on in botanical gardens during the early spring, early summer and late summer. The width of the top and bottom bar represents the bumblebee abundance and number of flower units, respectively. Bumblebee species are represented with different colours and are split into castes. The bottom bars are coloured to show native (green) and non-native (blue) plant species. Foraging preferences are shown by the coloured connectors, showing where plants were visited more often than expected (pink) and less often than expected (blue) when compared to the null model using econullnetr (Vaughan *et al.*, 2018). Bumblebee abundances during the early spring, early summer and late summer were 39, 101 and 68 respectively, and the floral abundance of bumblebee food plants were 24,224 units, 20,551 units and 20,455 units respectively.

2.4.3.2 Semi-natural farmland habitats

In the semi-natural farmland habitats, 0.19-21.1% of flower units were made up of exotic species across the three survey periods, with the majority of bumblebee visits being to native species (97.9%). Overall, there were 57.1% fewer bumblebee food plant species found in farmland habitats compared to botanical gardens. Plant-pollinator networks showed that bumblebees were visiting the two most abundant plant species (*S. dioica* and *Epilobium ciliatum*) less often than expected and showed particular preferences for *Cirsium arvense*, *Digitalis purpurea, Geranium robertianum*, *Heracleum sphondylium, Ranunculus repens*, *Rubus fruticosus*, *Sonchus asper* and *Stachys sylvatica* (Fig. 2.7).

During early spring (21st March – 11th April) 20 bumblebees of three species (*B. pascuorum, B. lapidarius* and *B. terrestris*) were recorded, but none were observed foraging. 27 flowering plant species were recorded, four of which were food plant species: *S. dioica* (Flower units = 510), *H. sphondylium* (Flower units = 191), *G. robertianum* (Flower units = 100) and *L. purpureum* (Flower units = 10). The most abundant flowering plant species during this survey period was *Primula vulgaris* (Flower units = 1,093).

During early summer (6-18th June) 93 bumblebees of eight species (*B. hortorum*, *B. hypnorum*, *B. lapidarius*, *B. lucorum*, *B. pascuorum*, *B. pratorum*, *B. sylvestris* and *B. terrestris*) were recorded foraging. Ten plant species, all natives, were visited by bumblebees out of a total of 62 flowering plant species. 4.8% of all flowering plant species were exotics. *S. dioica* was the most abundant flowering plant (Flower units = 41,205), but plant-pollinator networks showed it was visited less often than expected (Fig. 2.7).

During late summer (17-23rd August) 99 bumblebees of six species (*B. hortorum*, *B. hypnorum*, *B. lucorum, B. pascuorum*, *B. pratorum* and *B. terrestris*) were observed foraging. 15 flowering plant species were visited out of a total of 66 species. Only one food plant species was an exotic. Overall exotics made up 4.5% of all flowering plants recorded during this survey period. *S. dioica* continued to be the most abundant flowering plant species (Flower units

= 8,544), followed by an introduced exotic species, *E. ciliatum* (Flower units = 4,217). Both plant species were visited less often than expected (Fig. 2.7).

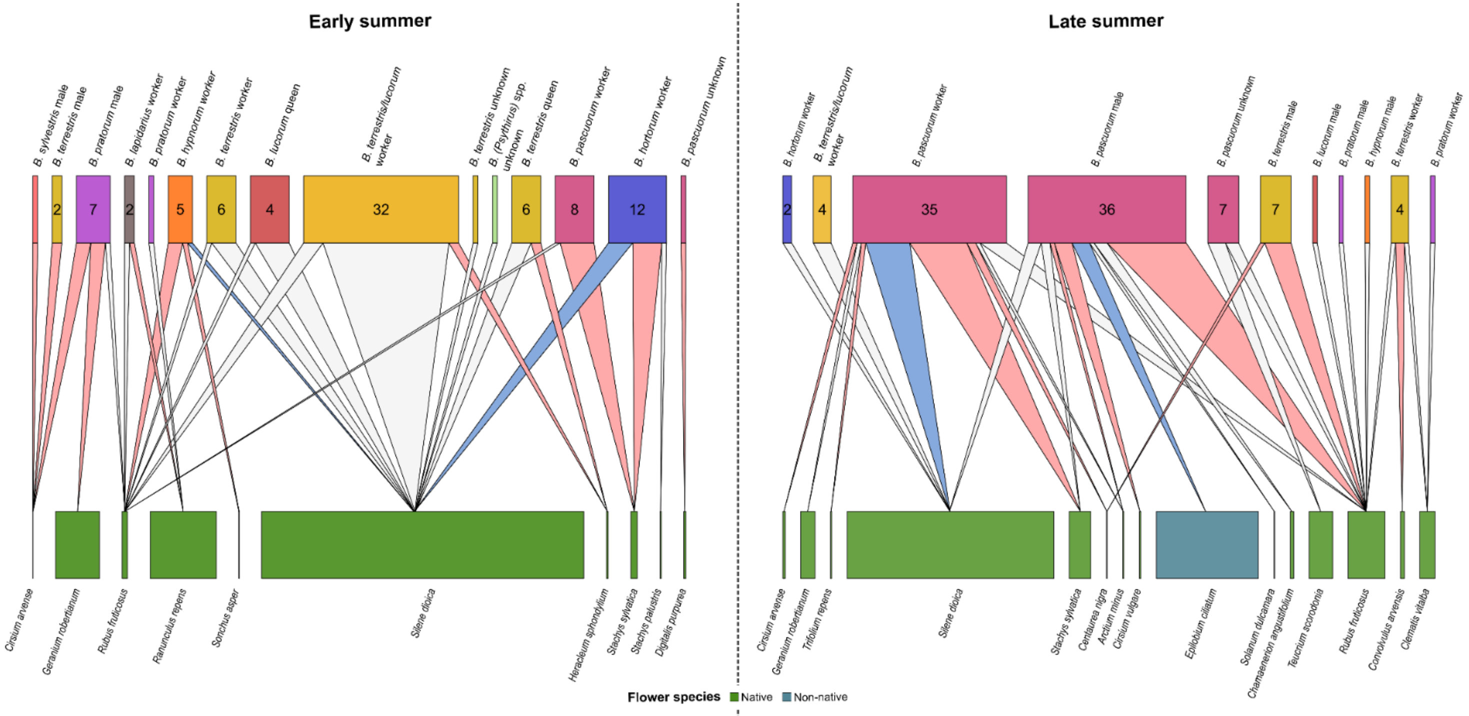

Figure 2.7 Plant-pollinator network for the plant species bumblebees were foraging on in the semi-natural farmland habitats during the early summer and late summer, which is when bumblebees were recorded foraging in this habitat. The width of the top and bottom bar represents the bumblebee abundance and number of flower units, respectively. Bumblebee species are represented with different colours and are split into castes. The bottom bars are coloured to show native (green) and non-native (blue) plant species. Foraging preferences are shown by the coloured connectors, showing where plants were visited more often than expected (pink) and less often than expected (blue) when compared to the null model using econullnetr (Vaughan *et al.*, 2018). Overall bumblebee abundance during the early and late summer were 107 and 132 respectively, and the overall floral abundances were 58,852 units and 20,092 units respectively. A network for the early spring period does not exists due to no foragers being recorded in this habitat.

2.5 Discussion

This study explored the bumblebee and flower phenology within botanical gardens and semi-natural habitats surrounding farmland in Cornwall throughout the bumblebee nesting season; from early spring through to late summer. It was predicted that botanical gardens would be a better habitat for bumblebees; supporting higher numbers of nest searching queens, providing more floral resources throughout the season and in turn having higher abundances of bumblebees. Similar benefits have been seen in residential gardens (Majewska and Altizer, 2018; Salisbury *et al.*, 2015). The results partly support this prediction, with botanical gardens providing a pulse of early spring floral resources and containing higher numbers of nest searching queens. However, semi-natural habitats within farmland then go on to have a higher floral abundance in early and late summer, with similar numbers of bumblebees recorded in both habitats during these survey periods. These findings are discussed in terms of the importance of multiple habitats for supporting pollinators. The potential for botanical gardens to support pollinators within rural agricultural landscapes is also explored.

During the phenological surveys, which covered the early spring and early and late summer, a single transect was walked per site per survey period, resulting in low resolution data. Collecting data over the whole flowering season would have provided detailed data on individual plants flowering times, as well as providing a weekly overview of floral changes, which are seen in the spring nest searching survey data, highlighting any gaps in foraging resources. For example a June resource gap was observed by Timberlake *et al.*, (2019). Collecting data at this resolution is not always possible, especially across a large number of sites, and so collecting data at distinct time points provides an overview of how floral resources and pollinator abundance changes over time. Studies have used this method to observe differences in insect pollinator communities across a variety of semi-natural and agricultural habitats (Cole *et al.*, 2017), and to highlight seasonal discrepancies in forage availability between natural and agricultural habitats (Williams *et al.*, 2012). The findings of this chapter provide useful information on how two distinct habitat types within the rural landscape are supporting pollinators throughout their nesting and foraging season.

Higher numbers of nest searching queens were recorded in botanical gardens, compared to semi-natural farmland habitats, irrespective of floral abundance or diversity. One explanation for this for this could be due to botanical gardens providing greater and/or higher quality nesting sites. Botanical gardens are likely to contain a number of suitable nesting habitats and features similar to those found in residential gardens (Lye *et al*., 2012; Osborne *et al*., 2008b), which would not be present in farmland habitats. However within agricultural landscapes bumblebees are known to nest in the semi-natural habitats such as hedgerow and banks (Kells and Goulson, 2003) which were surveyed in the current study therefore the provision of better nesting resources is unlikely to be the only reason for the greater number of nest searching queens recorded in this habitat. O'connor *et al*., (2017) also found that the number of nests found later in the summer was predicted by the number of nest searching queens. Therefore, botanical gardens in our study may contain higher numbers of nests compared to farmland habitats. However, Osborne *et al.*, (2008) found that linear countryside features similar to those surveyed in our study had similar nest densities to gardens (20-37 ha^{-1} and 36 ha^{-1} respectively). The actual nest densities of the two habitats in the current study are unknown. However, Carvell *et al*., (2017) showed queen bumblebees disperse 1,227 ± 125 m from their natal colonies (Carvell *et al.*, 2017). In our study, the pairs of farmland and botanical gardens were within this distance from each other in five out of the eight sites. Thus, queens would be able to disperse between the two habitats in most of our sites. Little is known about hibernation sites of bumblebees, but due to their heterogeneity and lack of disturbance compared to farmland (Winfree *et al.*, 2009), botanical gardens may contain more hibernating queens and therefore a higher baseline population of queen bumblebees, and thus nest searching queens, come spring.

Botanical gardens provide a pulse of spring foraging resources at a time when semi-natural farmland habitats had low floral abundance (Fig 2.2). This is a time when queen bumblebees are starting to emerge from hibernation and incubate brood (Heinrich, 1974) and so have high energy requirements. An early spring floral resource gap in agricultural habitats was also observed by Timberlake *et al.,* (2019). Early foraging resources, often provided by mass flowering crops, are important for colony growth and success (Parmentier *et al.*, 2014; Westphal *et al.*, 2003; Williams *et al.*, 2012), and so botanical gardens in Cornwall may be filling an important floral resource gap at this time of year in the absence of mass flowering crops during our study (with the exception of oilseed rape being present at one site). The most visited plant species during the early surveys was a *Rhododendron* species originating from Asia, which was foraged on by three queen bumblebees (*B. pratorum*, *B. terrestris* and *B. pascuorum*) and a *B. hypnorum* of unknown caste. Non-native species may therefore play an important role in filling floral resource gaps at times of year when native species are not yet flowering (Stelzer *et al.*, 2010).

Contrary to initial predictions, overall floral resources were higher for semi-natural farmland habitats compared to botanical gardens; although both habitats support a similar abundance and diversity of bumblebees over the whole season. Agricultural landscapes are often homogenous, simple landscapes containing few areas of semi-natural habitat (Söderman *et al.*, 2018). Landscape complexity can impact pollinators through affecting food availability, with floral resources being sparse and less abundant in simpler landscapes with large agricultural fields (Persson and Smith, 2011). In Cornwall, agricultural fields tend to be smaller, with all of the fields used in the current study falling within the 'complex landscape' category defined by Persson and Smith, (2011) as agricultural fields < 15 ha in size. They found that bumblebee colonies produced larger workers in these more complex landscapes (Persson and Smith, 2011). Larger foragers are thought to be more effective at foraging (Goulson et al., 2002b) and may be able to forage further (Greenleaf *et al.*, 2007) which would make them more robust against patchier resources within fragmented landscapes. Complexity of farmland habitats can be increased through the presence of small-scale habitat features such as hedgerows, which can increase flower species richness *(Hannon and Sisk, 2009; Söderman et al.*, 2018; Timberlake *et al.*, 2019). In our study surveys were conducted in the semi-natural habitats surrounding agricultural fields. This, along with the small fields, could explain the high amounts of foraging resources observed. This could have benefits for pollinators with smaller foraging ranges (Greenleaf *et al.*, 2007), such as solitary bees which produced fewer offspring in intensive agricultural landscapes (Williams and Kremen, 2007). The lack of difference in bumblebee abundance between the two habitats support other studies suggesting pollinators such as bumblebees are buffered against habitat fragmentation, due to their longer foraging ranges (Greenleaf *et al.*, 2007; Heard *et al.*, 2007). However this might not translate into higher densities, as the current study did not take into account the vast amount of cropped areas within the farmland habitats, or the lawned and built-up areas within the botanical gardens.

Pollinators can spill-over from semi-natural habitats into agricultural landscapes, supporting pollination services (Goulson *et al.*, 2010; Langellotto *et al.*, 2018). Although not empirically tested in this chapter, it is a potential benefit arising from the presence of botanical gardens within the rural landscape which warrants further research. In residential gardens, pollination of tomatoes was found to be highest in areas of high floral densities (Potter and LeBuhn, 2015) and spill-over of pollinators into crops was found to be highest in the presence of semi-natural habitats at the local scale (Chateil and Porcher, 2015), with pollination services being reduced in agriculturally dominated landscapes (Connelly *et al.*, 2015). Therefore, botanical gardens may act as a beneficial habitat within the agricultural landscape, providing nesting and foraging resources to pollinators. Future research should look at their potential to act as population sources within agriculturally dominated landscapes, and to study the potential for spill-over into surrounding crop fields.

In the botanical gardens, 74% of plant species and 82-90.9% of visits across the three survey periods were made up of exotic plant species (Fig. 2.6). In contrast, 90.3% of plant species in the semi-natural farmland habitat were natives, receiving 97.9% of visits from foragers (Fig. 2.7). Evidence for whether native flowering plants are better than exotics for pollinators is mixed (Majewska and Altizer, 2018). Fukase and Simons, (2016) showed visitation was positively linked to the percentage of native flower species within residential gardens, and higher abundances of pollinators were recorded on native and near-native flower species (Salisbury *et al.*, 2015). Bumblebees have been shown to forage flexibly, visiting both native and non-native plant species when available (Hanley *et al.*, 2014; Salisbury *et al.*, 2015; Sikora *et al.*, 2016), as seen in the current study. The main benefit of exotics is through extending the flowering season (Majewska and Altizer, 2018; Salisbury *et al.*, 2015; Stelzer *et al.*, 2010). This can be observed in the phenological surveys where a *Rhododendron* species is providing an abundant early spring floral resource. Colonies require sufficient nutrients from pollen and nectar for their development (Donkersley *et al.*, 2014; Hass *et al.*, 2018). Few studies have explored the nutritional value of exotic pollen compared to native, although one study found that the amino acid content of exotic plants was similar to natives (Rayner and Langridge, 1985). Future research into the quantities of exotic pollen collected at the whole-colony level, and whether this impacts colony success would provide insight into the colony-level impacts of exotic plant species, and would further strengthen the case for botanical gardens being beneficial habitats for pollinators.

2.6 Conclusion

Botanical gardens and semi-natural farmland habitats are providing important nesting and foraging resources for bumblebees. The prediction that botanical gardens would contain greater numbers of nest searching queens was supported, but botanical gardens were not found to be a better habitat overall, as semi-natural farmland habitats contained a higher floral abundance during early and late summer, and bumblebee abundance across the season was similar across the two habitats. Resource stability is important for bumblebee colony growth and reproduction (Williams *et al.*, 2012), and thus these two habitats may benefit pollinators within the wider landscape through providing resource peaks at different times and thus a more constant level of resources. Botanical gardens provided a pulse of early spring resources, when semi-natural farmland habitats contained few flowering plants. Floral resources in botanical gardens then decline as they increase in semi-natural farmland, peaking in June. Agricultural fields in Cornwall tend to be small, surrounded by semi-natural habitats such as Cornish hedgerows and therefore may provide suitable nesting and foraging resources to support pollinator communities. The benefits of botanical gardens may be more pronounced in intensive agricultural landscapes, where floral resources are sparser. Further research should look into the effects of botanical gardens in areas of high agricultural intensity, to determine whether pollinators are spilling out into the agricultural landscape, and whether gardens support greater numbers of nest searching queens due to having higher baseline populations of hibernating queens, and the resulting impacts of this on nest densities within this, and the surrounding, habitat.

Chapter 3

Testing the efficacy of a thermal camera as a search tool for locating wild bumblebee nests

Published as: Roberts, B.R. and Osborne, J. L., (2019) Testing the efficacy of a thermal camera as a search tool for locating wild bumblebee nests. *Journal of Apicultural Research.* DOI: 10.1080/00218839.2019.1614724

"A nest may be discovered accidentally, the bees having been noticed passing in and out; or we may specially go out to search for the nests, and this in itself is good sport."

– Sladen, 1912

Chapter 3: Testing the efficacy of a thermal camera as a search tool for locating wild bumblebee nests

3.1 Abstract

Research into how bumblebee colonies respond to the stressors affecting their populations are currently studied in the laboratory using commercially reared *Bombus terrestris* colonies. Understanding how these stressors affect wild bumblebee colonies in the field would be a crucial step forward for the conservation of bumblebee species. Currently visual cues are used to locate bumblebee nests, using human searchers looking for the worker nest traffic, but the limitations of this method mean that low numbers of nests are found and so a new method which looks to tackle these limitations is needed. Thermal cameras have been considered as a potential nest searching tool because they reduce the visual complexity of the environment by displaying a homogenised thermal landscape to the searcher. In this study we compare the use of a thermal camera to human searches using two trials: (i) using inexperienced volunteers to search along a transect for a known bumblebee nest and (ii) using an experienced individual to search across a number of novel locations. Thermal cameras were not found to be a better nest detection technique than human searches, having low success rates across both trials. The limitations of thermal cameras as a technique are discussed, along with the potential for technological advancements to improve its potential in the future.

3.2 Introduction

In order to conserve a species it is important to understand how they respond to stressors at both the population and individual level (Goulson, Nicholls, Botías, & Rotheray, 2015). In eusocial species such as bumblebees, population-level effects are those that impact the number of colonies, as it is the colony as a whole which represents one reproductive unit (Ellis *et al.*, 2006).

Understanding the mechanisms driving these population-level changes must be done at the individual colony-level. This is currently not being done in the field due to limitations of current nest detection techniques. Population-level effects are currently explored using genetic markers (Darvill *et al.*, 2004; Herrmann *et al.*, 2007). These have provided insight into broad concepts such as colony survival (Carvell *et al*., 2017; Goulson *et a*l., 2010), foraging distances (Carvell *et al.*, 2012; Knight *et al.*, 2005), queen dispersal distances (Carvell *et al.*, 2017) and a general outline of large-scale habitat preferences (Chapman *et al.*, 2003; Wood, Holland, Hughes, *et al.*, 2015). Studies cannot be conducted at the colony-level using these methods as the analysis of genetic markers in foragers, whilst providing information on relatedness and usage of the landscape by different nests, does not indicate exact nest locations. Colony-level studies are currently conducted in the laboratory, or field, using commercial *Bombus* colonies (Gegear *et a*l., 2006; Gill *et al.*, 2012; Imhoof and Schmid-Hempel, 1999; Stanley *et al.*, 2015). These studies, although useful, are not fully representative of wild bumblebee populations, and are also heavily biased towards *B. terrestris,* and *B. impatiens* in the US, and so it is not clear how transferable the findings in these studies are for other wild bumblebee species. Field studies of wild bumblebee colonies would provide crucial evidence into how stressors affect colony fitness through impacting queen production, worker production, foraging behaviour and queen mortality.

Locating wild bumblebee nests in order to gather such data is challenging, due to the small sizes of bumblebee nests and the lack of advanced nest detection techniques. Bumblebee colony sizes generally range from 50-750 workers depending on the species (Prŷs-Jones and Corbet, 2011). Consequently, nest traffic, the movement of workers in and out of the nest entrance, is low (Goulson, O’Connor, & Park, 2018). This is a limitation of human searches which rely on these visual nest traffic cues (Lye *et al*., 2012; O’Connor *et al.*, 2012; Osborne *et al*., 2008b). Trained sniffer dogs, which rely on olfactory cues to detect nests, were tested as an alternative technique but were no more successful than human searches (O’Connor *et al.*, 2012; Waters *et al.*, 2011). Another key limitation to the human nest detection method is being unable to visually detect the nest traffic against a complex background, largely due to the presence of vegetation (Waters *et al.*, 2011). In the current study a new method which uses heat cues to locate the nest traffic of wild bumblebee nests is tested.

Thermal imaging cameras have been used to study wild animals since 1972 (Graves *et al.*, 1972). The majority of applications for wildlife detection have been for endothermic species, in particular mammals (Cilulko *et al.*, 2013). Invertebrates are not commonly studied using thermal cameras due to many invertebrates being ectothermic and therefore having a small temperature differential between their body and the background. Thermal cameras have the potential to be a good tool for locating wild bumblebee nests as, unlike many invertebrates, social insects such as bumblebees are able to generate and maintain their own body temperatures (Heinrich, 1975; Stabentheiner and Schmaranzer, 1987) often above ambient temperature. Thermal cameras have already been used in some commercial applications to locate invertebrate pests; in the US it is a key tool for locating termite infestations in domestic and commercial buildings (James and Rice, 2002), and it is also used to locate pest infestations in stored food products and tree plantations (Al-doski, Mansor, & Shafri, 2016; Manickavasagan, Jayas, & White, 2008; Nanje Gowda & Alagusundaram, 2013). Thermal cameras have been considered as a potential nest searching tool because they reduce the visual complexity of the environment by displaying a homogenised thermal landscape (Fig. 3.1). Bee behaviour has already been studied using thermal cameras; as tools to monitor the Asian giant honeybees' (*Apis dorsata*) thermal defences against an invading wasp (Kastberger and Stachl, 2003), and recording the thoracic temperature of water-drinking honeybees (*Apis mellifera*) (Kovac and Schmaranzer, 1996). It is also currently being considered as a method for locating the aerial nests of the Asian hornet (*Vespa velutina*) (Keeling *et al.*, 2017) in an attempt to control its spread into the UK and across Europe.

The overall aim of the study is to ascertain whether thermal imaging cameras can be used as a new tool for the detection of wild bumblebee nests through enhanced detection of nest traffic or the nest itself in the case of surface nesting species. There were two main objectives; (i) to test the ability of thermal cameras to locate wild bumblebee nests using inexperienced volunteers and (ii) to test the efficacy of thermal cameras compared to human searches using an experienced individual. The limitations of current thermal camera technology are discussed and recommendations are made for future research.

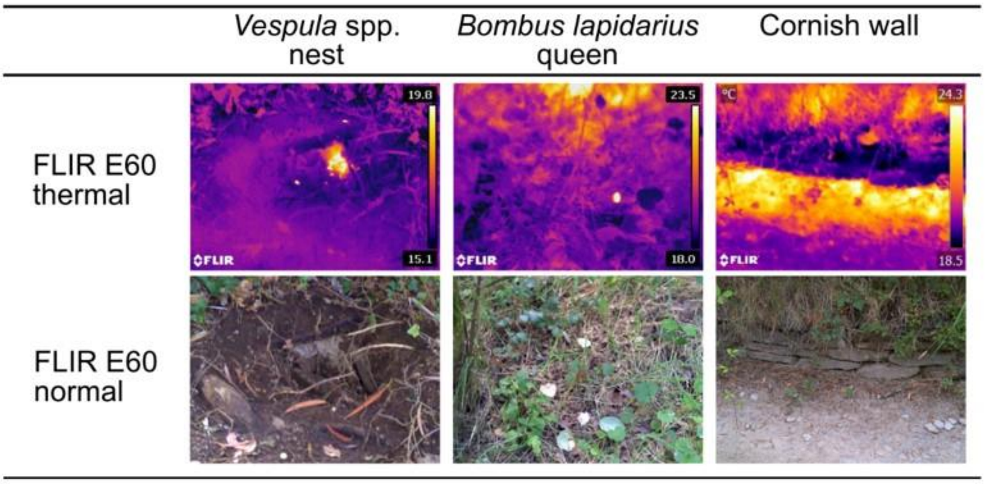

Figure 3.1 Figure showing the differences between the thermal images taken using the FLIR E60 and standard human vision images. From left to right the images show: a *Vespula* spp. nest which has been dug up by a badger (*Meles meles*), a *Bombus lapidarius* queen crawling in the undergrowth, and a Cornish wall; a prominent feature of many of the gardens surveyed during the non-targeted searches.

3.3 Methods

All surveys and experiments for this study took place in south west England (Lat 50°17'N, Long 4°48'W). Experiments were conducted using a FLIR E60 thermal camera (FLIR Systems, E60, 64501-0302). Two types of search method were performed: human unaided visual surveys and thermal camera surveys (Fig. 3.2). Human unaided visual searches consisted of the surveyor walking along the transect and using their own visual observations to search for either the nest traffic emanating from a bumblebee nest, or the nest itself in the case of a surface nest. Thermal camera searches consisted of the surveyor walking the transect in the same way as above but viewing the environment through the viewing screen of the thermal camera. Using this survey method bumblebees were seen as white objects moving on the viewing screen. Once a white object was detected visual observations could then be used to determine if the detected object was a bumblebee. Using the thermal camera, the nest traffic rather than the nest itself was most likely to be detected. Vegetation around the nest may obscure detection of traffic at the entrance, but traffic should still be visible in spite of the vegetation. During both search methods if a bumblebee was seen it was observed until out of sight to determine whether the bumblebee was travelling to a nest.

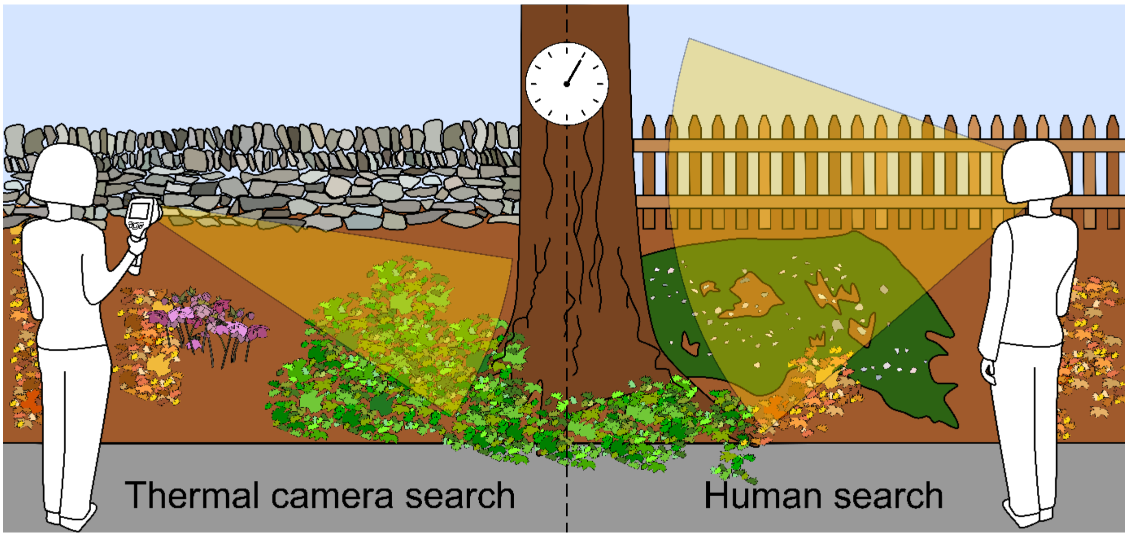

Figure 3.2 Thermal camera and human unaided visual survey method for the non-targeted searches. The individual is shown surveying an area from the path, which contains examples of different habitat features present during both the targeted and the non-targeted searches: dead leaves, vegetation, Cornish walls, fences, trees and mossy banks. The individual would walk slowly along the path whilst continually searching for nests for five minutes. Human and thermal camera searches were performed consecutively. The horizontal field of view for each search method is represented by the yellow beams; 25° for thermal cameras and 60° for human binocular vision. Targeted searches used a similar method, but with a suggested five minute minimum search time and 10-minute cut-off.

3.3.1 Targeted searches using non-experts

This trial was conducted on the 26th and 27th July 2016 in Cornwall, UK (Lat 50°17’N, Long 4°48’W). The aim was to test whether thermal cameras are a more effective tool to detect the nest traffic of a bumblebee nest compared to human unaided visual searches (O’Connor *et al.*, 2012). The 30 m transect used in this trial was located along an established path, where an active *Bombus hortorum* nest had been previously found in a disused rodent hole ~20 m into the transect. The transect was along a bank and encompassed a number of habitat features: leaf litter, exposed soil, natural and planted vegetation and trees.

Members of the public were asked at random to participate in the trial. Participants had no prior training or experience searching for bumblebee nests, and the ages of participants ranged from children to adults, but was not specifically recorded for this study. To ensure all participants had the same basic knowledge level prior to starting their search, they were (i) shown a picture of a bumblebee, (ii) told that bumblebees nest underground (in this case), and (iii) informed they were looking for nest traffic i.e. worker bumblebees coming in and out of the nest. Providing this information ensured that their search effort was spent actively looking for the nest. Those participants using the thermal camera were given a demonstration of how best to use the thermal camera i.e. moving it around to view the transect, and were advised that the nest traffic would appear as small, white moving objects on the cameras viewing screen. Once briefed, participants were taken to the starting point and surveyed the right-hand side of the path.

Participants performed targeted ‘free searches’, which ranged from searching from the path, to climbing onto the bank and searching through the vegetation. In total 25 participants took part, 13 performing thermal camera searches and 12 performing human unaided visual searches. Participants could stop searching at any time, but were advised to search for at least five minutes, and were stopped after 10 minutes if they had not been successful at locating the nest. When a participant found the nest they were asked to raise their hand for confirmation of success, and at this point the timer was either stopped or if the participant had incorrectly identified the nest they were given the option to continue searching until the 10 minute period had elapsed. Total search times of both successful and unsuccessful participants, as well as whether they found the nest or not were all recorded for analysis.

A logistic regression was performed using the statistical programme R (version 3.4.1, (R Core Team, 2017)) with ‘detection success’ as a binary response variable and ‘search method (camera or human)’ as the independent variable, the model was fitted with a binomial family. One thermal camera survey was removed prior to analysis due to the surveyor locating the nest without the thermal camera.

3.3.2 Non-targeted searches using an experienced individual

Between the 25th July and 6th August 2016, searches were conducted to test the effectiveness of using a thermal imaging camera as a searching aid when performing nest surveys across various locations. Both the human searches and thermal camera searches were conducted by myself, as an ‘experienced individual’ as I had undertaken multiple bumblebee surveys prior to the study, and had experience operating thermal cameras. Prior experience is important when testing a novel technique, to reduce the possibilities of ‘false negatives’ which could have occurred if using naïve searchers. Nest surveys were performed across six sites in Cornwall, UK (Lat 50°15’N, Long 5°3’W). Each site was searched for between 92-179 minutes. Surveys consisted of five minute consecutive searches alternating between human unaided visual searches and thermal camera searches. For ease, searches were carried out along already established paths. During both human and thermal camera searches the observer walked along the path at a steady pace, surveying both sides of the path for bumblebee activity. All bumblebees seen were recorded, and their behaviour was classified as: patrolling, foraging, flying, resting or entering and exiting a nest. Where possible caste and species was determined. Bumblebees seen flying were observed until they were out of sight to establish if they were flying to a nest. A hood was added to the thermal camera to prevent the observer using their peripheral vision whilst performing thermal camera searches. This was done for the non-targeted searches only, after an individual in the targeted searches located the nest during a thermal camera survey without using the camera. It also reduced glare on the screen to allow for more optimal use of the camera when searching.

To ascertain whether habitat types affected our ability to detect bumblebees and bumblebee nests using the thermal camera, all habitats and habitat features present during the five-minute searches were recorded. Habitat types and features present during searches included: leaf litter, banks, Cornish hedges, trees, short grassland, flower beds and long grassland. For analysis these were simplified to the number of different features in each search area, giving a crude estimate of habitat complexity.

3.4 Results

3.4.1 Targeted searches using non-experts

Twenty-five searches were conducted by participants, with 24 being used in the final analysis. Human unaided visual searches located the nest 75% of the time, whereas individuals using the thermal camera as a search tool were only able to locate the nest 33.3% of the time, this difference was significant (z_{22}=1.979; p=0.048; Fig. 3.3). Coefficients for the model output can be seen in Table 3.1.

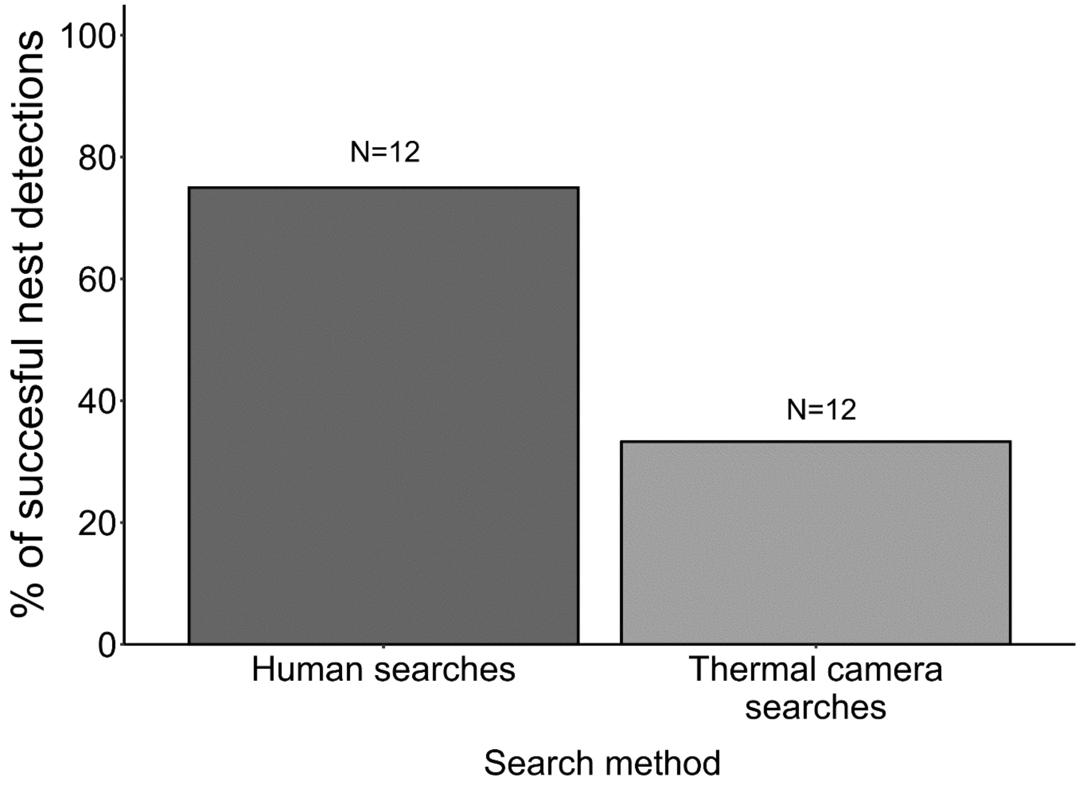

Figure 3.3 The number of times participants successfully located the nest for each search method during the targeted searches, presented as a percentage.

Table 3.1 Coefficients for the best fitting model for the targeted searches. Output generated in the statistical programme R (version 3.4.1, (R Core Team, 2017)) from a logistic regression model fitted with 'detection success' as the response variable, and fitted with a binomial family.

	Estimate±SE	z value	p value
Intercept (Search method (Thermal camera)	-0.69±0.61	-1.13	0.258
	0.69±0.61		
Search method (Human searches)	1.80±0.91	1.98	0.048
Model code: lmer(detection success~search method, family=Binomial)			

3.4.2 Non-targeted searches using an experienced individual

The total search time was 13 hours 48 minutes. Six hours 58 minutes of search time was performed for human unaided visual searches and six hours 48 minutes of search time was performed for thermal camera searches. During searches, five nests of four different species were found (Table 3.2). Only one of these nests was found using the thermal camera, with the other four being found during human unaided visual searches.

The nest detection rate for human unaided visual searches was one nest for every one hour 44 minutes of search time, a much faster rate of detection than thermal camera searches which found one nest for every six hours 48 minutes of search time. Due to the small number of nests found no statistical analysis was performed.

Table 3.2 The location of nests found during the non-targeted searches.

Bumblebee species	Detection method	Number	Habitat
B. hortorum	Human	1	Cornish hedge
B. lapidarius	Human	1	Brick wall
B. terrestris	Human	2	Rodent hole
B. terrestris	Thermal	1	Rodent hole

3.5 Discussion

The use of a thermal imaging camera did not improve the users’ ability to locate bumblebee nests when compared to human searches and therefore it is recommended that further research into other nest detection methods is needed to enable the successful detection of larger numbers of wild bumblebee nests. In all cases it was the nest traffic which led to nests being detected; detection of surface nests due to their thermal signal may be possible without the visual cue from the worker nest traffic, but no such nests were found during the study. The study took place during late-July to early-August at a time when the colonies of some species have reached, or are starting to reach, maximum size (Prŷs-Jones and Corbet, 2011). The majority of colonies in other studies were located within a similar period (Goulson *et al*., 2018). This may lead to a bias towards detection of larger, successful colonies and therefore reduces the ability to monitor how wild colonies are responding to stressors.

One of the limitations of the study was using non-expert individuals to survey for nests, which could have led to ‘false negatives’ due to lack of prior nest searching experience. The use of volunteers to collect data is widely used in citizen science and biological recording projects, with around 70,000 individuals in the UK alone annually submitting species observations to recording programmes (Carvell *et al.*, 2016; Pocock *et al.*, 2015). There are mixed opinions on the validity of using such ‘non-experts’ for scientific research purposes (Cohn, 2008), but many cite this data collection method as a useful tool (Sauermann and Franzoni, 2015), especially for conservation programmes (Johnson *et al.*, 2014). Experience of volunteers was not found to affect their ability to detect wild bumblebee nests when performing human searches (O’Connor *et al.*, 2012). However, when testing a novel nest detection technique, as in our study, the chances of ‘false negatives’ occurring is likely increased due to the combination of the surveyor’s lack of prior nest searching experience and their inexperience of using the novel technology. Small differences in detection success between human and thermal camera survey techniques have been shown in other studies (Graves *et al.*, 1972), likely due to discrepancies between surveyors. Therefore, a single experienced individual was used to conduct the non-targeted searches, in an attempt to reduce both variation between surveyors and the occurrence of ‘false negatives’ when performing bumblebee nest surveys across a range of sites. Using a single experienced individual allowed the efficacy of the two methods for locating nests to be tested more directly, rather than testing the user’s ability to successfully operate the thermal camera, had multiple inexperienced volunteers been used.

The second limitation was with the thermal camera itself. The thermal camera lens had a field of view of 25˚ x 19˚, which is much smaller than the human field of view (190˚ x 135˚) (Fig. 3.4). This reduced field of view meant that the thermal camera user had to move the camera around in order to survey the same area as a human using their natural field of vision. Due to the small colony sizes of bumblebees (Prŷs-Jones and Corbet, 2011), nest traffic emanating from the nest entrance is low. Many bumblebee species nest underground, and so will not give off a thermal signal in the way surface nests may do. It is therefore the nest traffic which will enable detection. The need to move the thermal camera around when searching an area due to its small field of view meant the chances of the camera being trained on a nest entrance when a bumblebee was entering or exiting was low. During human unaided visual searches bumblebees were often seen in the peripheral and monocular field of vision (Fig. 3.4), giving human searches a wider field of vision with which to survey the environment. To address this, a thermal camera with a wider-angle lens could be used e.g. FLIR thermal imaging cameras with a 45˚ field of view are currently available. Although larger than that of the thermal camera used in our study, their field of view is still 15˚ less than a human's binocular vision, and 145˚ less than a humans binocular and peripheral vision combined (Schneck and Dagnelie, 2011). As well as a small viewing area, the ability of a thermal camera to detect warm objects within an environment is affected by a number of other factors such as air temperature, distance from the object and the presence of vegetation (Cilulko *et al.*, 2013). These limitations mean that the detection effectiveness of the thermal camera during the day to detect bumblebee nests was likely reduced. Addressing the current limitations of thermal cameras could improve their use as nest detection tools, for example the use of thermal cameras with wider angled lenses as mentioned above. Further studies would be needed to compare the effectiveness of thermal cameras with a wider field of view to human searches.

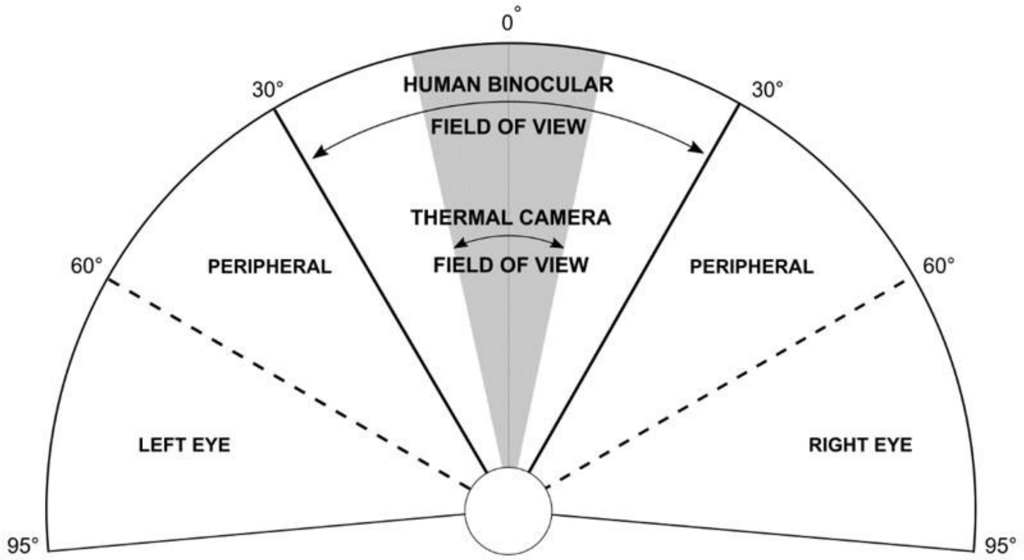

Figure 3.4 The field of vision of a human (white), showing the binocular (60˚), peripheral (120˚) and complete (190˚) vision range. Compared to the field of view of the FLIR E60 thermal imaging camera (25˚) used in this study (grey).

The findings from the current study further support the argument that human searches are currently the best method at locating bumblebee nests. The search methods used during the non-targeted searches were similar to the ’free search’ method used by O’Connor *et al*. (2012) to explore the effectiveness of trained sniffer dogs as a novel bumblebee nest detection method compared to human searches. They recorded a nest detection rate for both humans and sniffer dogs of one nest for every one hour 20 minutes of searching (O’Connor *et al.*, 2012), which is in line with the human search rates found in our own study. The novel detection approach using the thermal camera as a search tool in comparison performed much worse than both of these methods, finding only one nest during six hours 48 minutes of searching. These findings corroborate those of O’Connor *et al*. (2012), showing that human searches are a cost effective method of locating wild bumblebee nests, as although sniffer dogs performed at the same rate as humans, they are more expensive due to the initial training costs and continued upkeep (Mathews *et al.*, 2013; O’Connor *et al.*, 2012). This is also true for thermal cameras which have large upfront costs. There is still a need for a more effective nest detection method to be developed, which would allow larger quantities of nests to be found more quickly in order to fully study wild colonies *in situ*. In the absence of these, other techniques can be developed where necessary and in **Chapters 4 & 5** a novel artificial nest technique is developed and then tested to quantify predation rates to bumblebee nests from the European badger (*M. meles*).

3.6 Conclusion

Due to current lack of efficient detection methods few field studies on the ecology of natural bumblebee nests exist. The current study was unable to establish a novel nest detection method to replace the currently used human searches. Human searches do not find large numbers of nests, and relatively few studies have used nest detection techniques to monitor and study the nests. Without this monitoring of bumblebees at the colony level it is not possible to understand how stressors that affect bumblebees at the population level (Goulson *et al.*, 2015) are actually impacting upon individual colonies, an important area of research due to the continuing decline of bumblebees in the UK (Goulson *et al*., 2008). Studies which have monitored wild nests have provided information on nest predation, survival, disease and gyne production (Goulson *et al*., 2018; Osborne *et al*., 2008b) but further and more longer term monitoring of wild colonies is needed. Future research should focus on better nest detection and monitoring techniques.

Chapter 4

Quantifying the relative predation pressure on bumblebee nests by the European badger (*Meles meles*) using artificial nests

“Badgers are said to be fond of scratching out and eating the nest.”

– Sladen, 1912

Chapter 4: Quantifying the relative predation pressure on bumblebee nests by the European badger (*Meles meles*) using artificial nests

4.1 Abstract

Bumblebee populations are declining. This is likely due to a combination of factors which regulate the populations by affecting the size and success of colonies through limiting resource availability (bottom-up regulation) or factors directly causing mortality, such as the use of pesticides, disease and predation (top-down regulation). Badgers are one of the most damaging nest predators of bumblebees, destroying the whole colony, and are extremely widespread and abundant in the UK. The relative importance of bumblebee nest predation by badgers has not been quantified and so the current study used a novel artificial nest as a proxy for wild bumblebee nests to quantify the relative predation pressure from badgers in two habitats: woodland and grassland, and at two nesting depths: surface and underground, compared to controls. Significantly more artificial nests were dug up compared to empty controls, showing that artificial nests can be successfully used to quantify relative bumblebee nest predation from a large mammalian predator. In an area of high badger densities, predation pressure was greater in woodland than grassland, whereas no difference was observed in relation to nest depth. Understanding the relative impact of badger predation on bumblebee colonies in different habitats and under different densities and seasonal conditions will provide key information on how such top-down regulation affects bumblebee populations.

4.2 Introduction

Understanding the effects of stressors on bumblebee populations is important due to them providing key pollination services (Klein *et al.*, 2007) and to further understand their worldwide decline (Goulson *et al.*, 2008; Potts *et al.*, 2010). The causes of worldwide bumblebee declines are likely due to a combination of factors (Williams and Osborne, 2009), acting on bumblebee colonies from both from the ‘bottom-up’ and ‘top-down’. Bottom-up effects such as resource availability regulate population sizes via limiting the rate of colony growth and success (Ogilvie and Forrest, 2017; Williams *et al.*, 2012). Resource availability can differ between habitats (Baude *et al.*, 2016), and is also influenced by human activity through habitat loss and fragmentation (Goulson *et al.*, 2015; Potts *et al.*, 2010). In contrast, top-down regulation refers to factors that cause direct mortality: these can be influenced by humans, such as effects caused by pesticides (Gill *et al.*, 2012; Rundlöf *et al.*, 2015), or they can be natural population regulators such as disease (Manley *et al.*, 2015) and predation (Goulson *et al.*, 2018a, 2018b). Top-down effects which cause direct mortality will be acting alongside bottom-up regulatory effects, influencing the stability of bumblebee populations.

The degree to which predation of bumblebee nests has an impact on bumblebee populations is relatively unknown. In Europe, the only nest predator which is likely to have substantial negative impacts on bumblebee colonies is the European badger (*Meles meles*), as the whole of the colony is destroyed during a predation event (Goulson *et al.*, 2018b; Pease, 1898). Other nest predators include birds, such as great tits (*Parsus major*) which predate workers entering and exiting the nest (Goulson, *et al.*, 2018b) and wax moths (*Aphomia sociella*) which infest colonies and destroy most of the comb (Alford, 1975; Goulson *et al.*, 2002; Pouvreau, 1973; Sladen, 1912). There is little evidence that either of these two nest predators have negative impacts on colonies in terms of gyne production (Goulson *et al.*, 2018a, 2018b). Other mammals such as foxes (*Vulpes vulpes*), stoats (*Mustela ermine*), moles (*Talpa europaea*) and hedgehogs (*Erinaceus europaeus*) have been anecdotally recorded as nest predators (Alford, 1975; Goulson, *et al.*, 2002; Pouvreau, 1973; Sladen, 1912), but empirical evidence supporting this is lacking. Therefore badgers are likely the most destructive nest predator of bumblebees, with predation pressure likely changing depending on their seasonal diet, habitat use and densities. Although this has not been measured in many contexts due to the difficulty of finding and monitoring wild bumblebee nests (although see Goulson *et al.,* 2018a, 2018b).

Badgers, like bumblebees, are central place foragers (Hipólito *et al.*, 2018), and show individual foraging specialisation (Robertson *et al.*, 2014, 2015). They are seasonal specialists of the earthworm *Lumbricus terrestris* when available (Kruuk and Parish, 1981; Shepherdson *et al.*, 1990), but consume a varied diet of cereals, small vertebrates and invertebrates during times of low earthworm availability (Kruuk and Parish, 1981; Shepherdson *et al.*, 1990), with non-earthworm invertebrate consumption peaking in June-July (Harris, 1984; Kruuk and Parish, 1981; Shepherdson *et al.*, 1990). One study in Ireland found bees and wasps made up 1% of badgers diet from March-September, peaking at 6.5% between June-August (Cleary *et al.*, 2009). Another in Scotland looked specifically at bumblebees, finding they made up 0.8% of badgers annual diet (Kruuk and Parish, 1981). In one of the only studies on bumblebee nest predation, 5.5% of nests over an eight year period were reportedly dug up by badgers (Goulson *et al.*, 2018b); with a peak in June-July. These peaks in invertebrate, and specifically bee and bumblebee consumption coincide with the peak colony sizes of bumblebees (Muller and Schmid-Hempel, 1992), when gynes and males are being produced (Goulson *et al.*, 2018b). Predation pressure will also vary with other factors such as badger densities, which vary across the UK (see Table B.1) and with both badger and bumblebee habitat usage.

Two habitats commonly used by badgers are woodland and grassland. Woodland habitats are the preferred habitat for sett location (Feore and Montgomery, 1999; Harris, 1984), and badgers spend the majority of their time in this habitat (Kruuk, 1978). In contrast, grassland is mainly visited by badgers under wet conditions when foraging for their primary prey item, *L. terrestris* (Kruuk and Parish, 1981; Shepherdson *et al.*, 1990). Bumblebees also utilise these two habitats, for nesting (O'connor *et al*., 2017; O'Connor *et al.*, 2012; Osborne *et al*., 2008b) and foraging (Carvell *et al.*, 2006) . Bumblebees have been estimated to nest at similar densities in woodland and grassland habitats in the UK (see Table 1.1), although other studies have shown bumblebee and pollinator abundance are often negatively impacted by woodland (Diaz-Forero *et al.*, 2012). Thus, it is assumed that badgers are likely to encounter bumblebee nests in the two habitats, although nest detectability may vary.

This study uses a novel technique to quantify the relative predation pressure to bumblebee nests from badgers in woodland and grassland habitats, and compare predation of nests at different soil depths. The artificial nest method used in this study was adapted from that used by Waters *et al.*, (2011) to test the ability of a sniffer dog to locate wild bumblebee nests. They created artificial nests by placing 7 g of nest material in small pots and burying them. After being trained on the artificial nests, dogs successfully located wild bumblebee nests of a variety of *Bombus* species. During training the dogs achieved a 100% detection success, giving no false indications. In woodland, dogs detected 40% of these artificial nests and in grassland this more than doubled to 84% success rate (O’Connor *et al.*, 2012). Badgers, like dogs, have an acute sense of smell and so it is hypothesised that badgers will be able to successfully detect the nest material in the artificial nests used in the current study and will thus be more likely to dig them up than artificial nests that did not contain nest material.

The current study used artificial nests buried at two soil depths: surface (<5 cm underground) and underground (~17 cm underground) to replicate different depths of bumblebee nests. Bumblebee species have specific and differing nesting preferences which may affect their vulnerability to predation. Species such as *Bombus hypnorum* may experience no predation pressure from badgers, as they nest almost exclusively above ground (Lye *et al.*, 2012). Other species, such as *Bombus pascuorum*, preferentially nest in grassland habitats and are surface nesters (Kells and Goulson, 2003; O’connor *et al.*, 2017) and as such may be more vulnerable to predation. More artificial nests are expected to be detected and dug up in woodland, due to badgers spending most of their time in this habitat (Kruuk, 1978) and the stronger scent cues from surface nests means predation is expected to be higher for artificial nests at this depth. The control pots, which contain no nest material are expected to be dug up less often.

4.3 Methods

4.3.1 Study sites

Fieldwork was conducted in two locations with known badger setts; Woodchester Park, Gloucestershire, UK (51˚43’N, 2˚16’E) was the main site, due to it being the location of a long-term badger population monitoring study. Boundary Court, Gloucestershire, UK (51˚43’N, 2˚14’E) was chosen as a secondary site, located ~1.5 km away from the main site and which had previously been used as a badger monitoring study although less regularly or rigorously than the main site. Both sites have similar habitat layouts (Figure 4.1a), with woodland valleys lining the boundaries, and grassland in the centre. Woodchester Park is larger than Boundary Court, covering an area of approximately 7 km^2 with a badger population of around 200 individuals within 22 social groups (Delahay *et al.*, 2000). The grassland is a mixture of grazed and non-grazed fields. Boundary Court is around 3 km^2 with 4 badger social groups (Cheeseman *et al.*, 1981). The sizes of the badger setts within this location are unknown. The grassland was previously cattle grazed pastoral grassland but had been left fallow for at least 2 years. The land-use surrounding both sites is a mixture of residential areas, arable and pastoral agriculture, and grassland. Within both of the study sites, the setts are located within the wooded valley, and the badger territories extend out into the surrounding grassland and arable habitats (Delahay *et al.*, 2006; Fig. 4.1).

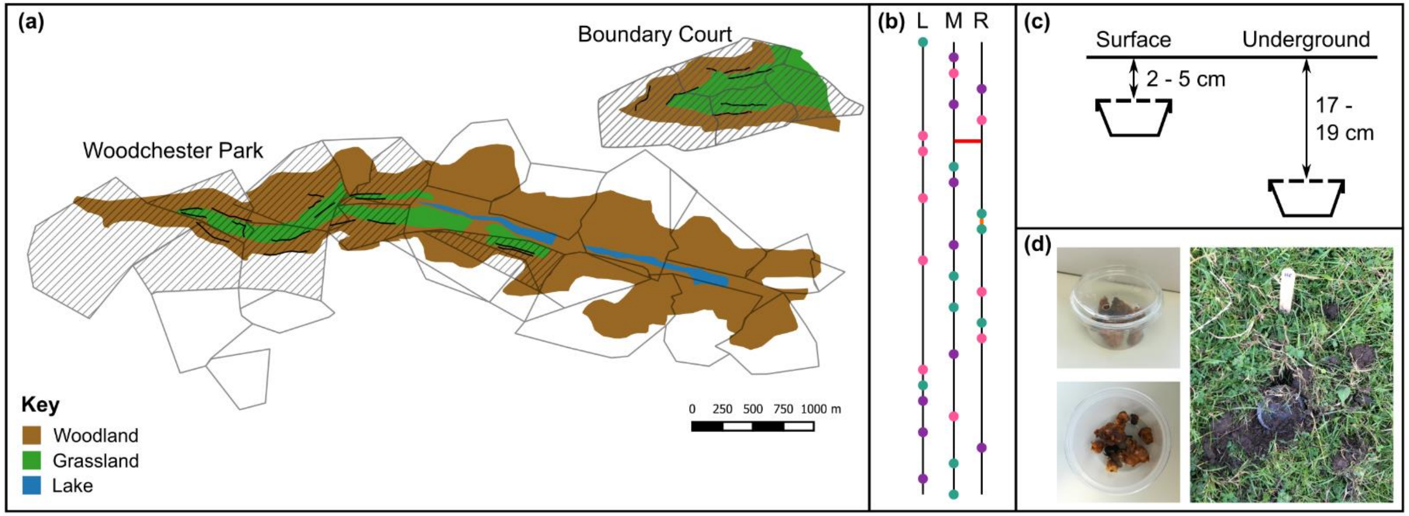

Figure 4.1 Panel figure showing: (a) Map of Woodchester Park and Boundary Court. The foraging boundaries of each sett are shown (polygons), with the setts used in the study represented by dashed polygons. Transects are shown (thick black line). (b) Layout for the transects (solid black line) with the locations of the artificial nests (coloured dots) shown. Each colour represents a different treatment: surface, underground and control to highlight the random way which artificial nests were placed along the transect. The distance between each transect is 2 m (red line) and the distance between each artificial nest is 8 m (orange line). (c) Depth treatments of the artificial nests showing the surface and underground depths. (d) Upper left: An artificial nest filled with 7 g of commercial *Bombus terrestris audax* nest material, showing the five six mm holes drilled into the top. Top and bottom left: An example of the nest material placed into each artificial nest. Right: An artificial nest which has experienced a 'disturbance event' where the soil above the pot has been dug but the pot itself was left in the ground, showing the pot *in situ* with the wooden marker labelled with the position along the transect and the nest depth.

4.3.2 Artificial nests

'Artificial nests' were used to quantify the predation pressure to bumblebee nests by badgers. The artificial nests functioned as proxies for wild nests, providing a scent cue to badgers using commercial nest material, and were placed *in situ* in two different habitats: woodland and grassland, to determine how badger activity within two habitats influences the predation rates. Artificial nests were also buried at two different depths: surface and underground, to test the preferences and detectability of the artificial nests by individual badgers along transects. Empty pots, acting as controls, were placed in the same locations to determine the baseline levels of detection from badgers for a novel object within their territories. Using pots at different depths also gives us a crude measure of whether surface and underground bumblebee nests are experiencing different predation pressures.

The artificial nests were small plastic pots (H: 40 mm, W: 70 mm) with 6 x 5 mm holes drilled into the lids. Each artificial nest was filled with 7 g of nest material (wax, brood cells and bumblebees) from commercially produced *B. terrestris audax* colonies. Gloves were worn at all times during pot handling to minimise the contamination from human scents.

4.3.3 Study design

Sett territories of the badger setts in Woodchester Park, used for artificial nest transects, were determined using data from Delahay *et al.*, (2006) and these can be seen in Fig. 4.1a. Setts were chosen in almost all cases whose territory incorporated both woodland and grassland habitats. Eleven setts were chosen for use in the study; eight setts at Woodchester Park and three at Boundary Court. Of these, only one sett was used that contained only one habitat type (woodland). For the remaining 10 setts, transects were able to be set out in both woodland and grassland habitats. The experimental design was as follows:

Two habitat types were studied:-

- Woodland edges: transects were placed within 10 m of the edge of the woodland.

- Grassland edges: transects were placed within 10 m of the edge of the grassland, which consisted of mostly pastoral grassland ranging from short heavily grazed to longer infrequently grazed fields.

Within each habitat, transects were placed within the badger foraging territories of each sett (Fig. 4.1a). Transects were set up near the edges of the habitats rather than centrally, and along linear features such as a fence lines or a path to replicate the types of features where bumblebees prefer to nest (Kells and Goulson, 2003; Osborne *et al.*, 2008b; Svensson *et al.*, 2000). These features are also used by badgers for moving through habitats and for creating latrines (Balestrieri *et al.*, 2009; Hounsome *et al.*, 2005). Each transect, with the exception of five initial transects which contained only surface and underground nests, consisted of 30 nests; 10 for each of the three artificial nest treatments:

1. Surface nests: pots containing 7 g of commercial *B. terrestris audax* nest material were buried with 1-2 cm of soil covering the lid of the pot to represent surface bumblebee nests (Fig. 4.1c).
2. Underground nests: pots containing 7 g of commercial *B. terrestris audax* nest material were buried at a depth of 17-19 cm and a hole from the pot to the surface was created at an angle to the dug hole to replicate the entrance hole of wild bumblebee nests (Fig. 4.1c).
3. Control nests: empty pots were buried at the same depth as surface nests to represent areas of disturbed ground but which provided no reward to the badgers.

Transect were 4 m wide, with artificial nests being randomly assigned to a position on the transect; either in the centre, or 2 m to the left or right of the central transect (Fig. 4.1b) with a distance of 8 m between each pot. Artificial nest positions, as well as depth along the transect, were assigned at random using an online random list generator (www.random.org). Staggering of nests across the 4 m transect and randomisation was done to increase the effort it takes for badgers to find the pots, and to ensure there was no pattern that badgers might be able to learn during the study. The total length of transects was approximately 232 m. Six transects were discontinuous with <50 m gaps.

Two trail cameras (Bushnell ® Bushnell NatureView Essential HD) set to record 20 seconds of video when triggered by motion were placed along each transect to monitor badger presence and to provide contextual evidence of whether it was only badgers digging up the artificial nests.

The study was carried out over a 4-week period from the 19th July to the 16th August 2017. Artificial nests were buried along the transects by hand during the day and left in place for three consecutive nights. Artificial nest locations were marked with a 15 x 1.7 cm wooden label, with the pot number and nest depth written on. Pot markers were handled using gloves at all times to reduce contamination from human scent, and were placed into the ground approximately 5 cm from the pot location. On the fourth day, transects were re-visited and two variables were recorded:

- Dig up event: a dig up event was classified as when a pot had been removed from the ground. In some cases the nest material had been eaten, but this was not always the case. In instances where the artificial nest could not be found, but visual and physical checks confirmed the pot was not still in the ground, it was recorded as a dig up event.
- Disturbance event: this was classified as where the soil above the artificial nest had visibly been dug but where the artificial nest had either not been reached or had been left in the ground (Fig. 4.1d). This was recorded as a measure of detectability, but was not included in the current analysis.

Artificial nests which remained in the ground after the three nights were retrieved and disposed of. The number of rainfall nights for each transect was calculated using rainfall data from www.glosweather.com, which uses a Davis Instrument Vantage Pro2™ Wireless 6312 console and a Davis Rain Catcher to record rainfall for Gloucestershire, UK. A rainfall night was classed as any evening during the three consecutive nights which received more than 2 mm of rainfall.

4.3.4 Analysis

Analysis was performed using the statistical software R version 3.5.2 (R Core Team, 2017). For analysis, dig up events were categorised as a ‘success’ and disturbance events and artificial nests left in the ground were categorised as a ‘failure’. These terms were used as a combined response variable in the mixed effect model (see Table 4.1). ‘Habitat’, ‘nest depth’ and their two-way interaction were included as fixed effects. ‘Transect ID’ was nested within ‘sett’ as a random effect. The model was fitted with a binomial family.

Model selection was performed using Akaike’s Information Criterion for small sample sizes (AICc) (Bartoń, 2017). Models with a delta AICc <2 when compared to the best fitting model were kept. The coefficients from the best fitting models are reported in Table 4.1.

Camera trap footage was used to verify badger activity at the sites, rather than provide a measurement to be used in analyses. Rainfall was also not included in the analysis due to the consistently high rainfall during the study meaning differences due to varying weather conditions could not be explored.

4.4 Results

In total 125 artificial nests filled with commercial bumblebee nest material were dug up out of a total of 574 nests, equating to 21.8% of nests. The average number of artificial nests dug up for each nest treatment per transect was 3.1 ± 0.4 (mean ± standard error) surface nests and 2.9 ± 0.4 underground nests, compared to 1.4 ± 0.2 controls. The study found that surface and underground artificial nests were dug up significantly more than control pots (Surface: z=4.59, p<0.001; Underground: z=4.11, p=<0.001; Fig. 4.2a, Table 4.1). Significantly more artificial nests were dug up in woodland compared to grassland (z=2.78, p=<0.01; Fig. 4.2b, Table 4.1). There was no significant interaction between the number of underground nests dug up between the two habitats (z=-1.16, p=0.247, Table 4.1), or for surface nests (z=-1.84, p=0.065, Table 4.1) although there were slightly more surface nests dug up in the grassland habitat compared to in woodland, where equal numbers of underground and surface nests were dug up.

Camera trap videos confirmed that badgers were the only species digging up the artificial nests, despite a variety of mammal and bird species being present along the transects. Badgers were captured on the camera traps at eight out of the 11 setts; seven times in the woodland habitat and five in the grassland habitat. Only three transects took place which received one or fewer nights of rainfall, with the remaining 18 transects experiencing two or three nights of rainfall and so rainfall was not used in analysis due to lack of variation.

Table 4.1 Coefficients for the best fitting models for the predation of artificial nests by badgers in different habitats and for different nest treatments. A binomial generalised linear mixed model was fitted with a two-way response matrix for the number of pots per transect which were dug up ‘success’ and the number which were left in the ground ‘fail’.

Model 4.1.1	Estimate ± SE	z value	p value
Intercept (Habitat (Grassland) and Nest treatment (Control))	-3.35 ± 0.53	-6.34	<0.001
Habitat (Woodland)	1.42 ± 0.51	2.78	<0.01
Nest treatment (Surface)	1.53 ± 0.33	2.39	<0.001
Nest treatment (Underground)	1.37 ± 0.33	4.11	<0.001
Model 4.1.2	Estimate±SE	z value	p value
Intercept (Habitat (Grassland) and Nest treatment (Control))	-4.26 ± 0.82	-5.17	<0.001
Habitat (Woodland)	2.56 ± 0.90	2.86	<0.01
Nest treatment (Surface)	2.69 ± 0.76	3.55	<0.01
Nest treatment (Underground)	2.17 ± 0.76	2.86	<0.01
Habitat (Woodland): Nest treatment (Surface)	-1.56 ± 0.85	-1.84	0.065
Habitat (Woodland): Nest treatment (Underground)	-0.98 ± 0.85	-1.16	0.247
Global model code: glmer(cbind(success, fail)~habitat*treatment + (sett/transect ID), family=Binomial)			

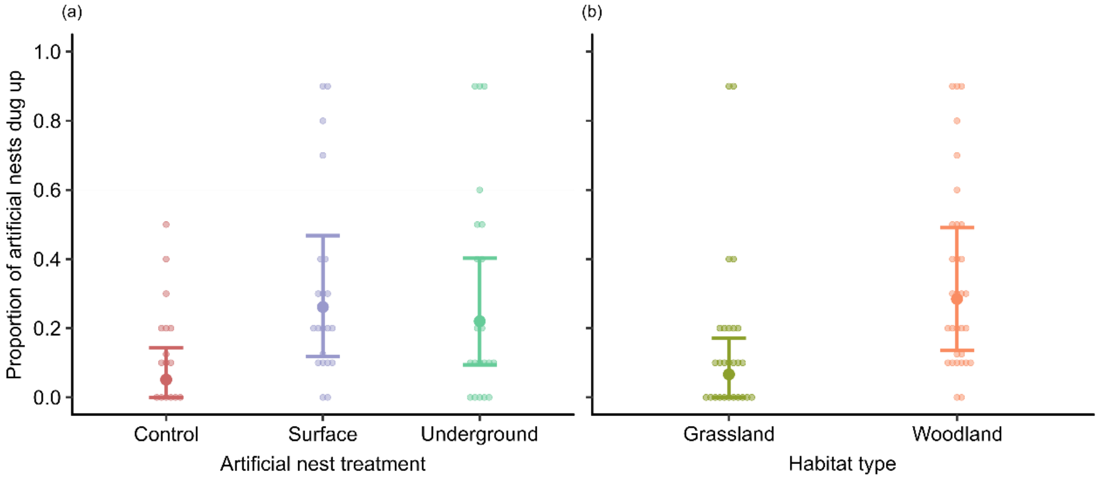

Figure 4.2 The proportion of artificial nests dug up by badgers for (a) each of the three nest treatments: control, surface and underground and (b) the two habitats: woodland and grassland. The raw data is displayed with a beeswarm plot, with the predictions and confidence interval from the best fitting model displayed.

4.5 Discussion

This study has successfully tested a novel technique to estimate the relative predation pressure on bumblebee nests from badgers. Significantly more of the artificial nests were dug up by the badgers than the empty control pots (Fig. 4.2a) and predation pressure was significantly higher in the woodland habitat than in the grassland (Fig. 4.2b). The scent cues provided to the badgers from the artificial nests were likely to be less than those of wild bumblebee nests, as only a small amount of nest material was present in the artificial nests. This suggests badgers are able to detect wild bumblebee nests even when small in size (7 g) which could impact colonies at all stages of their lifecycle. A small proportion of control pots were dug up (Fig. 4.2) despite having no scent profile of bumblebee nests, showing that badgers may have been attracted to the artificial nests due to contamination from human scent, the soil being disturbed or due to the presence of a marker. Even so, there were significantly fewer controls dug up than those filled with nest material. Here, these findings are discussed in the context of how badgers' and bumblebees' utilise the two different habitats for both nesting and foraging.

In a landscape with high badger densities, predation pressure varied between habitats, with greater numbers of artificial nests being dug up in woodland than grassland. It is not possible to distinguish whether this is due to differing levels of badger activity (which will vary between transects) or due to the varying detectability of the artificial nests by the badgers in the different habitats, or both. Badgers utilise the two habitats used in this study differently, with woodland being used for sett locations (Feore and Montgomery, 1999; Harris, 1984) and grassland for earthworm foraging (Da Silva *et al.*, 1993; Kruuk *et al.*, 1979). Kruuk, (1978) found that badgers spend the majority of their time in woodland, the preferred habitat for sett location (Harris, 1984; Smal, 1995), with setts located in woodland being more productive (Feore and Montgomery, 1999). By spending large portions of time in woodland, badgers may have been more likely to come across the artificial nests, explaining the higher relative predation seen in the current study. Badger activity between the two habitats can also vary depending on their foraging activities.

Under wet conditions badgers spend more time foraging for earthworms in grassland habitats (Kruuk, 1978; Shepherdson *et al.*, 1990). During earthworm foraging badgers move slowly through the patch with their nose close to the ground, detecting earthworms on the surface as they pass directly underneath them (Kruuk, 1978; Shepherdson *et al.*, 1990). Their detection ability and preference for artificial nests whilst undertaking this foraging behaviour is likely reduced, due to their slow movement and heightened selectivity for earthworms. In contrast, woodland contains a wider variety of food stuffs (Da Silva *et al.*, 1993), meaning badgers are not showing a preference for one specific food item in this habitat. Badgers also utilise a different foraging technique under dry conditions, moving faster and travelling further (Kruuk, 1978). Shepherdson *et al*., (1990) found that during hotter, drier years when earthworms were less available, insects made up a greater proportion of badgers’ diets (Shepherdson *et al*., 1990). This could mean under future climate change scenarios, which predict that the occurrences and intensities of droughts will increase (Dai, 2013; IPCC, 2014), invertebrates including bumblebees will experience higher levels of predation from badgers. This could increase stress to bumblebee populations at a time when they are already compromised (Phillips *et al.*, 2018). During the current study only three transects took place which received one or fewer nights of rainfall, with the remaining 18 transects experiencing two or three nights of rainfall. It is therefore not possible to draw conclusions about predation under dry conditions. Further studies considering the seasonal effects of badger predation on bumblebee nests would be required to address this.

Studies into badger diets have shown bumblebees only make up small percentages of their overall diet (Goulson *et al.*, 2018b; Kruuk and Parish, 1981), but this could vary with badger densities. In the current study, where badger densities are high, the proportion of artificial nests that were dug up by badgers also seemed high (Fig. 4.2), despite the small amount of nest material (7 g) artificial nests contained. This suggests pressure from this predator on bumblebee nests in the UK could have an effect on bumblebee populations. Woodchester Park, used in the current study, has higher than average badger densities; with 25.3 adults per km^2 (Rogers *et al.*, 1997). Badger densities in other locations range from 0.5 per km^2 in Ireland (Smal, 1995) to 6.2 per km^2 in Staffordshire (Cheeseman *et al.*, 1985); Wytham Woods in Oxford also has extremely high badger densities at 43.6 per km^2 (Noonan *et al.*, 2015). The high badger densities at Woodchester Park allowed the testing of this novel technique, in a scenario where the sett locations and foraging boundaries were known, and where badgers were guaranteed to forage in relatively high numbers; reducing the chances of false negatives. However the high densities are also likely to have affected the predation pressure, therefore further studies in areas with differing badger densities would build a broader picture of the predation risk to bumblebees across the UK.

In **Chapter 5**, the artificial nest method developed in the current chapter is tested in Cornwall, an area of lower than average badger densities (4.7 badgers per km^2 (Cheeseman *et al.*, 1981)). This allows the technique to be tested in a location with badger densities more closely matching other badger densities across the UK (Table B.1). Alongside the artificial nest method, the population model *Bumble*-BEEHAVE (Becher *et al.*, 2018) is used to simulate the combined effects of badger predation and food availability on bumblebee populations over multiple years. Computer models are another way of studying bumblebee populations in the absence of sufficient nest detection techniques (**Chapter 3**) and allows the effects of multiple stressors to be studied over multiple years.

4.6 Conclusion

This study empirically quantified for the first time the relative predation pressure posed to bumblebee nests from badgers. It successfully tested a novel technique using artificial nests as a proxy for wild bumblebee nests, which could be implemented in future studies in a range of habitats, across the nesting season and for a variety of badger densities. This would enable the monitoring of the impacts of predation, a potentially key top-down population regulator, on a vulnerable group of invertebrates that deliver vital ecosystem services. Badgers and bumblebees co-exist in a number of habitats, with woodland being a key shared area. Understanding the long-term consequences of badger predation to bumblebee populations under different badger densities is a key next step in understanding the top down regulation of bumblebee populations, and this is explored in **Chapter 5**.

Chapter 5

Exploring the seasonal effects of badger (*Meles meles*) predation and varying food availability on bumblebee populations

Chapter 5: Exploring the seasonal effects of badger (*Meles meles*) predation and varying food availability on bumblebee populations

5.1 Abstract

Bumblebee populations are regulated by a combination of bottom-up and top-down mechanisms, which may act synergistically. When populations are already under stress, for example when experiencing reduced food availability, the effects of other stressors, such as pesticides, disease and predation can be exacerbated. Badgers are one of the most destructive nest predators of bumblebees, often destroying the whole colony. This chapter builds on the artificial nest method developed in **Chapter 4** to quantify predation rates in an area of lower badger densities (Cornwall). It also aims to determine if the method can be used to detect the seasonality of badger predation, in line with badgers' seasonal dietary shifts. As well as this, the population model *Bumble*-BEEHAVE is used to simulate the combined effects of badger predation and food availability on bumblebee populations over a 20-year period. Using computer models provides a way of monitoring the long-term population effects of multiple stressors, which would be difficult under field or laboratory conditions. High and low predation rates were used in the model to represent the rates in Gloucestershire (high badger density, **Chapter 4**) and Cornwall (low badger density) which is the study location of the current chapter. It is predicted that high levels of predation will have greater impacts on bumblebee colony outputs. This was found to be true, with low badger predation rates having no more impact on bumblebee populations than when predation was not occurring. Under high predation rates the numbers of hibernating queens, workers and colonies produced was significantly lower than under no predation. Food availability was not found to buffer populations against high levels of predation, but low food availability led to significantly fewer queens, workers and colonies being produced compared to high food availability. This reflects wild pollinator populations, which are in decline largely due to loss of foraging resources. These findings are discussed in relation to the different ways that stressors

impact bumblebee colonies, with a view to better understanding the pressures affecting wild bumblebee population dynamics.

5.2 Introduction

Bumblebees are under threat from multiple stressors (Goulson *et al.*, 2015; Williams and Osborne, 2009), acting on populations from both the ‘bottom-up’ (resource availability) and the ‘top-down’ (predation and disease). These two regulatory mechanisms act on populations in contrasting ways; in the first instance by limiting population growth (bottom-up) and in the second by increasing direct mortality (top-down). It can be difficult to look at these regulating forces with wild colonies, with many studies using commercial *Bombus terrestris* colonies, or *Bombus impatiens* in the US (Spiesman *et al.*, 2017). These types of studies provide short-term data on how individual colonies respond to stressors and are biased towards two species. The next step towards conserving bumblebees is to understand how stressors affect populations over a longer period of time, as it is likely that populations under prolonged stress will experience declines (Kitaysky *et al.*, 2007).

Food resources are a key bottom-up regulator of populations (Roulston and Goodell, 2011), and are known to affect the individuals responses to other stressors. Food limitation can affect growth, lifespan and fecundity of various insects (Agarwala *et al.*, 2008; McKay *et al.*, 2016; Olson *et al.*, 2017). It can also exacerbate the impacts of other stressors, such as exposure to diseases and pesticides. Damselfly larvae (*Coenagrion puella*) that had been stressed by both heat and starvation experienced considerable mortality from pesticide exposure compared to unstressed larvae (Dinh *et al.*, 2016). In bumblebees, chronic exposure to a pesticide led to reduced worker sizes, especially when food was limited (Baron *et al.*, 2014). Studies on bumblebee diseases found that when bumblebees were food limited it led to a 50% increase in *Crithidia* infections in *B. terrestris* (Brown *et al.*, 2000) and to bees being 1.6 times more likely to die due to slow bee paralysis virus (Manley *et al.*, 2017). A contrasting study found that food limitation led to reduced *Crithidia* infections (Conroy *et al.*, 2016), but individuals experienced reduced longevity (Sadd, 2011). This highlights the complexity that interacting stressors can have on individuals. When looking at colony level impacts, Rotheray *et al.*, (2017) showed food limited colonies produced fewer queens and males, and were more susceptible to food shortages early in the spring. Similarly, colonies which experienced food supplementation produced a greater number of queens and males (Pelletier and Mcneil, 2003). Food availability is therefore an important aspect of colony development, and more studies which investigate the combined effects of bottom-up and top-down factors are needed to further understand the interplay of these stressors on colonies.

Food availability is a key limiting factor for bumblebee colonies and other pollinators. Habitat loss and fragmentation through agricultural intensification are they key drivers of reduced food availability, leading to lower abundance and increased patchiness of floral resources (Söderman *et al.*, 2018). This has consequences both at the individual and the colony level. Many studies that discuss the importance of floral abundance and diversity to bumblebees focus on their impacts on counts of workers (Senapathi *et al.*, 2016; Steffan-Dewenter *et al.*, 2002). These provide insights into the effects of floral availability on foragers at the individual level, but do not always translate to colony benefits. For example, higher floral abundance has been found to increase the number of workers in a colony, but did not lead to increased queen production (Elliott, 2009; Goulson *et al*., 2002a; Westphal *et al.*, 2009; Williams *et al*., 2012). Floral diversity is also important for supporting diverse pollinator assemblages (Fründ *et al.*, 2010; Jha and Kremen, 2013b). However, some species may be more greatly affected by floral diversity than others. Colony growth and reproduction of *B. impatiens* In the US was most influenced by the floral cover of just a few abundant species (Spiesman *et al.*, 2017). This is also a foraging strategy used by the short-tongued *B. terrestris,* which collected the majority of its pollen from a few abundant plant species, whereas *B. pascuorum* were found to collect pollen from a variety of plant species during a single foraging trip (Kämper *et al.*, 2016). Kriesell *et al*., (2017) showed that the overall amino-acid content of pollen loads for a variety of bumblebee species was similar. Thus, the nutritional values required for successful colony development (Génissel *et al.*, 2002; Tasei and Aupinel, 2008) may be achievable even in low diversity patches, although the composition of pollinators it supports may be affected (Fründ *et al.*, 2010; Jha and Kremen, 2013b). Studying the impacts of stressors under varying food availability scenarios should be a priority, as high-quality food availability may be able to buffer populations against the negative impacts of stressors.

Badgers are the main mammalian predator of bumblebee nests, destroying the whole colony (Goulson, *et al.*, 2018a; Pease, 1898), and are common and widespread throughout the UK. Badger diets vary seasonally (Kruuk and Parish, 1981; Shepherdson *et al.*, 1990); from September to April the bulk of their diet consist of their preferred food source the earthworm (*Lumbricus terrestris*) (Shepherdson *et al.*, 1990), and during this time they can be considered specialist foragers of this species (Balestrieri *et al.*, 2004). In the summer months, when earthworms become less available (Satchell, 1983) badgers consume a more varied diet of, amongst others, insects, small vertebrates, wheat and fruit (Cleary *et al.*, 2009; Harris, 1984; Shepherdson *et al.*, 1990). Insect consumption peaks in June-July (Harris, 1984; Shepherdson *et al.*, 1990), with bee and wasp consumption peaking in June-August (Cleary *et al.*, 2009). A study of bumblebee nest predators also found predation of nests peaked in June-July (Goulson *et al.*, 2018a). These peaks coincide with colonies reaching their maximum sizes (Muller and Schmid-Hempel, 1992), potentially leading to badgers disproportionately predating queen-producing colonies. Badger predation rates of bumblebee nests are predicted to vary seasonally, in line with both reduced earthworm availability and increased detectability of bumblebee nests.

Chapter 4 (Roberts, Cox and Osborne, submitted) successfully tested a novel artificial nest technique for monitoring badger predation of bumblebee nests. This was conducted in an area with high badger densities, of around 16.6 adults per km^2 (see Table B.1). Across the UK badger densities vary, with an estimated average of 8.8 badgers per km^2 (see Table B.1). The study in the current chapter was conducted in Cornwall, which has a badger density of 4.7 badgers per km^2 (Cheeseman *et al.*, 1981). It is predicted that badger density may impact predation rates and therefore lower predation rates are expected in the current study compared to those found in **Chapter 4** (although the comparison is not direct as the studies were done in different years). The current study also aims to determine whether the artificial nest method can be used to detect the seasonality of bumblebee nest predation, through conducting repeated surveys in April, June and August. This is in line with badgers seasonal diet variation and the differing stages of bumblebee colony growth (Benton, 2006; Shepherdson *et al.*, 1990). We then go on to model the impacts of both badger predation and food availability using population models to look at the long-term effects.

Studying the long-term population effects of stressors such as predation is difficult to do experimentally (Muller and Schmid-Hempel, 1992; Wood, Holland, Hughes, *et al.*, 2015). Agent based models are a way that these effects can be explored (Grimm *et al.*, 2017; Henry *et al.*, 2017). A number of bumblebee models have recently been developed (Banks *et al.*, 2017; Bryden *et al.*, 2013a; Cresswell, 2017; Crone and Williams, 2016; Häussler *et al.*, 2017; Olsson *et al.*, 2015). These provide useful insights into how foraging and nesting resources affect colonies (Häussler *et al.*, 2017) and the effects of single stressors (Bryden *et al.*, 2013b; Crone and Williams, 2016) but do not currently have the capacity to monitor the effects of multiple stressors. *Bumble*-BEEHAVE (Becher *et al.*, 2018) is a population model which specifically models the colony dynamics of bumblebees through combining aspects of BEEHAVE (Becher *et al.*, 2014), which models honeybee colony dynamics, and BEESCOUT (Becher *et al.*, 2016), which models foraging bees within the landscape. BEESTEWARD (http://beehave-model.net/download/) is a user-friendly interface for the *Bumble*-BEEHAVE model, and it is used in this chapter to model, for the first time, the effects of badger predation rates and food availability on *B.terrestris* populations and colony outputs. Bumblebee populations are predicted to be most affected by high badger predation rates, and higher levels of food availability may help to buffer populations against the negative effects of predation.

5.3 Methods

5.3.1 Seasonal badger predation of artificial bumblebee nests

5.3.1.1 Study sites

Fieldwork was conducted across ten sites (A-I) in Cornwall (50.2660° N, 5.0527° W; Table 5.1, Fig. 5.1) where badger setts were known to exist, and that contained grassland and woodland habitats within 1 km of each other. Distances between each habitat ranged from 0.073-0.758 km with a mean distance of 0.174 km. This was important, as the exact foraging territories of setts were not known for this study, and so selecting habitats close together increased the chances that they were within the same sett territory. Grassland habitats ranged in size from 2.6-18.8 ha and were a mixture of pastoral (N=5), semi-improved (N=4), and amenity (N=1) grassland (Table 5.1). Woodland habitats ranged in size from 2.5-97.2 ha and varied in their accessibility, with some having complete public access, some limited public access and others being completely private. Most sites were mature, broad leaved woodland, but site H contained a number of laurel species with a few more mature trees mixed within it (Table 5.1). Distances between the sites ranged from 0.99-18.46 km with a mean distance of 5.10 km. This was important to ensure different setts were being studied at each site. Only two sites were within < 1 km of each other, but were separated by a large road and so it is unlikely they are used by the same badger social group.

Table 5.1 Site information for the ten locations used. Sites A-I were all used in the *Bumble*-BEEHAVE simulations, with site J being excluded due to its proximity to a large river estuary, so that much of the landscape on one side of the site was covered by water.

Site	**Habitat**	**Area (Ha)**	**Public access**	**Habitat type**	**Surrounding land-use**
A	Grassland Woodland	4.6 7.4	None None	Pastoral Mixed	Scrub, agricultural (arable) and estuary
B	Grassland Woodland	3.5 10.5	Limited Limited	Amenity Mixed	Woodland and agricultural (pasture and arable)
C	Grassland Woodland	18.8 38.3	Full Full	Fallow Ancient	Woodland and agricultural (pasture)
D	Grassland Woodland	5.7 63.0	Full Limited	Pastoral Deciduous	Woodland, agricultural(arable and pasture) and botanical garden
E	Grassland Woodland	4.8 97.2	Full Full	Semi-improved Deciduous	Woodland, agricultural (arable and pasture) and botanical garden
F	Grassland Woodland	8.5 25.3	Limited None	Amenity Mixed	Golf course, woodland and agricultural (pasture and arable)
G	Grassland Woodland	3.5 41.0	None Limited	Semi-improved Deciduous	Agricultural (pasture)
H	Grassland Woodland	2.6 2.5	Full Limited	Pastoral Deciduous	Agricultural (pasture and arable) and botanical gardens
I	Grassland Woodland	5.3 11.8	None None	Pastoral Mixed	Agricultural (pasture and arable)

J	Grassland Woodland	6.8 9.2	Full Full	Pastoral Deciduous	Agricultural (arable) and estuary

5.3.1.2 Study design

To understand how badger predation rates vary across the season, transects containing artificial nests (developed in **Chapter 4**) were set out in each habitat (woodland and grassland) over three survey periods: spring (3rd – 27th April), early summer (4th June – 3rd July) and late summer (7th – 31st August) in 2018. Detailed methods of how the artificial nests were created, how they were set out along a transect and how transect locations were chosen can be found in **Chapter 4**. In brief, at each site one transect was set up in each habitat. Each transect consisted of 30 artificial nests; 10 surface (buried < 5 cm below the ground), 10 underground (buried 15-17 cm below the ground) and 10 empty controls (buried < 5 cm below the ground). Surface and underground artificial nests were filled with 7 g of commercial *B. terrestris audax* nest material. In contrast to **Chapter 4**, artificial nest locations were marked with a piece of string tied around the nearest piece of vegetation e.g. a long piece of grass, a tree branch or in the adjacent hedgerow. This reduced visual signals which might affect badger predation, but enabled nests to be subsequently found by the surveyor and removed if they had not been dug up by badgers.

Transects were set out at each site once per survey period, with a total of ten woodland and ten grassland transects per survey period; 60 transects in total across the study. Artificial nests were kept in the ground for three consecutive nights, in accordance with the method developed in **Chapter 4**. On the fourth day transects were re-visited and the number of dig up and disturbance events were recorded (see **Chapter 4** for details). Artificial nests which had not been dug up were removed. A break of 50-70 days (mean of 60 days) where no pots were present at the transect occurred at each site between each of the survey periods to reduce the chances of badgers learning the transect locations and associating them with food.

Two camera traps (Bushnell ® Bushnell NatureView Essential HD) were placed along each transect, set to record 20 seconds of video when triggered by motion, to monitor badger presence and to confirm it was only badgers digging up the artificial nests. Rainfall gauges were also put out at each transect to monitor rainfall over the three days.

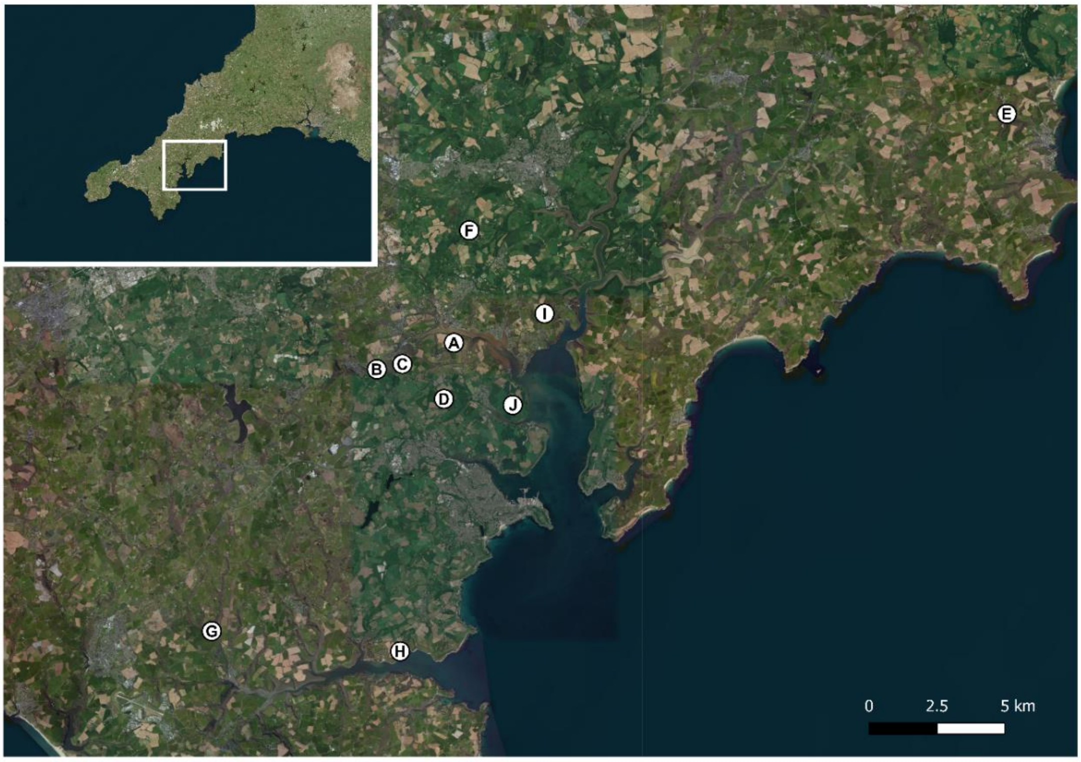

Figure 5.1 Map showing the location of the 10 field sites (white circles marked A-I), with an inset showing the county of Cornwall with the white box representing the specific study area. Satellite image courtesy of Bing Images ©.

5.3.2 Simulating long-term effects of badger predation and floral resource availability on bumblebee populations

5.3.2.1 The *Bumble*-BEEHAVE model

In the model, *B. terrestris* queens randomly emerge from hibernation on 1st April (± 28 days *SD*), following a normal distribution. Once queens emerge, they can nest in any of the available semi-natural habitat types on the map:

woodland, grassland, hedgerows and scrub (Becher *et al.*, 2018). Nest numbers within a landscape are determined by the number of queens which emerge from hibernation and their daily probability to either find a nest site or to die (Becher *et al.*, 2018). Colony growth and reproductive success is then monitored over the season, until newly produced queens are mated and then enter hibernation before re-emerging the following year. The probability of a queen surviving hibernation depends on her weight. The model does not consider immigration or emigration from queens and so all queens which are produced and survive hibernation stay within the landscape.

Habitat maps from realistic landscapes can be used within the model. The foraging landscape is comprised of a number of foraging patches which belong to a certain habitat type. Habitat types within the model are defined by the presence and abundance of a list of 44 flower plant species, and whether they can act as nesting habitats for the bumblebees. Flowers provide nectar and pollen during defined flowering periods, and foraging processes are simulated in great detail (see extensive documentation of the *Bumble*-BEEHAVE model (Becher *et al.*, 2018)). Bumblebees can forage for up to eight hours each day, and the flowering phenology is identical each year.

5.3.2.2 Habitat maps

Habitat maps were created using QGIS (QGIS Development Team, 2018) for each of the Cornwall study sites (Fig. 5.2), excluding Site J (its proximity to a large river estuary meant that much of the landscape on one side of the site was covered by water) giving nine sites as replicates. Maps were made with a 750 m radius which encompassed both the woodland and grassland habitat used during the artificial nest study. Areas of woodland, grassland, scrub and hedgerows were identified and recorded for each map. Habitat types were assigned using the 2007 land cover map data (Morton *et al.*, 2011), as well as ground-truthing during fieldwork. Arable crop fields were not included in the habitat maps and were left as blank space. This is a good representation of much of the arable crop fields in Cornwall which consist mainly of cereals, which provide little or no forage for bumblebees (Easy *et al.*, 2012).

Floral resources for the woodland, scrub and hedgerow habitat were assigned according to *Bumble*-BEEHAVE’s default parameterisation. To simulate high and low food availability the ‘quality’ of food resources in the grassland habitats was varied. For high food availability, the default settings were used, which is representative of species rich grassland. For low food availability, all flower species except dandelion and white clover were removed which is representative of improved grassland (Baude *et al.*, 2016) and in line with grassland habitats found in Cornwall (Knapp *et al.*, 2018). As the area of each habitat type varies for each map, so does the total nectar (l) and pollen (kg) (Fig. 5.3, Table B.2).

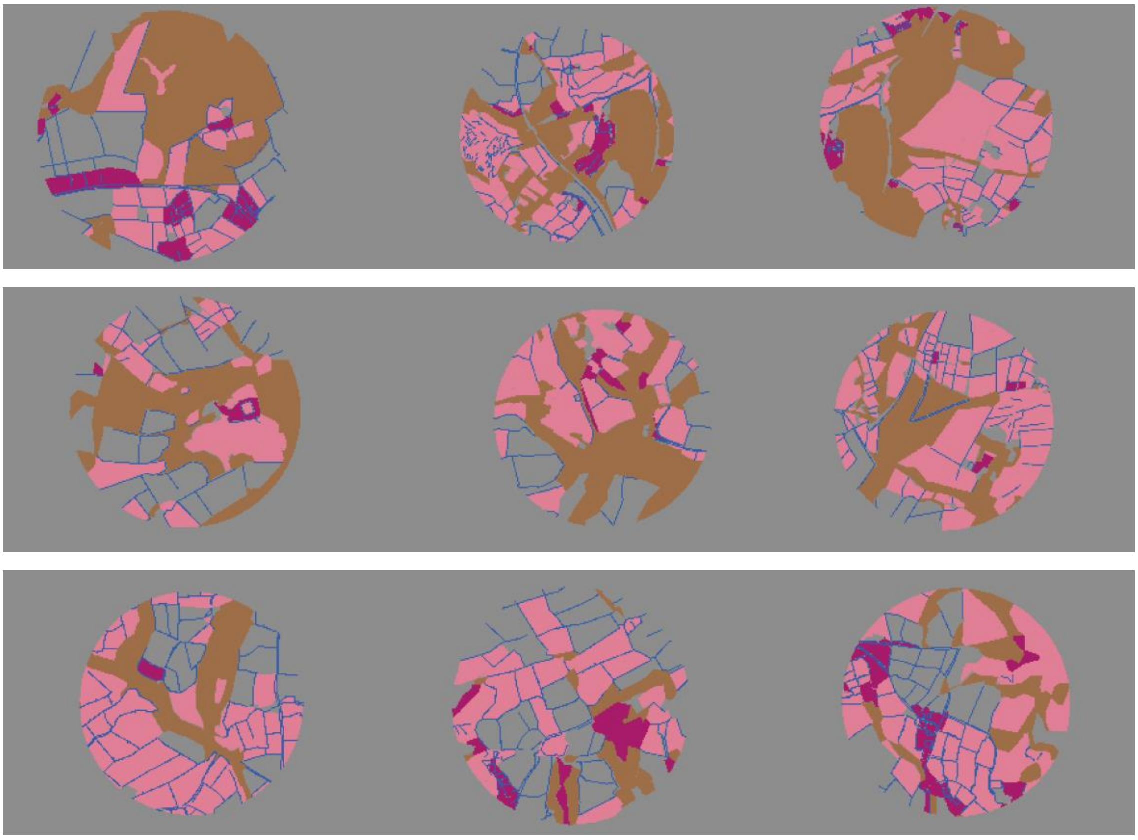

Figure 5.2 The nine habitat maps used for the *Bumble*-BEEHAVE simulations, showing the four habitat types: woodland (brown), grassland (light pink), scrub (dark pink) and hedgerows (blue lines). The blank areas represent agricultural crop fields, which were not included in simulations due to being mainly cereal crops, assumed to have minimal or no forage.

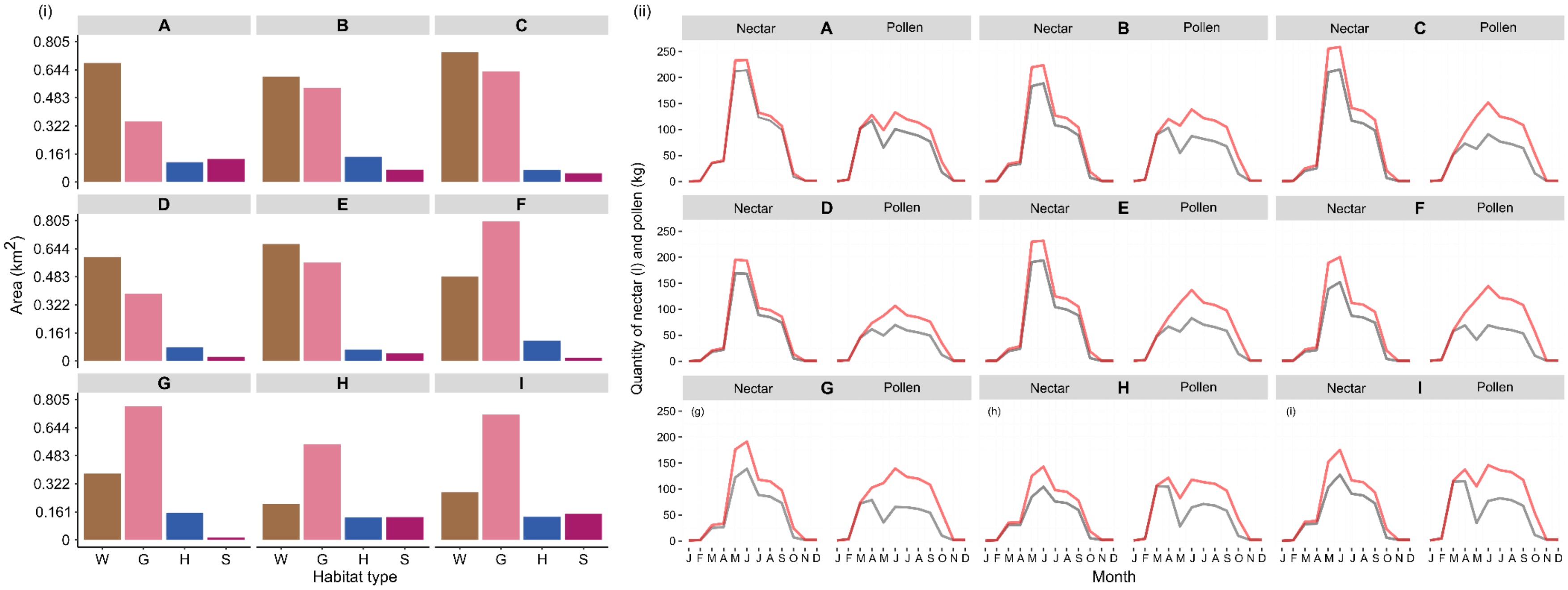

Figure 5.3 Panel showing (i) the area in km^2 of each habitat for each site (A-I); W = woodland, G = grassland, H = hedgerow and S = scrub and (ii) the average simulated quantity of nectar (l) and pollen (kg) per month for each site (A-I) for high (red lines) and low (grey lines) food availability.

5.3.2.3 Setup simulation experiments

Simulations were run for 20 years using *Bumble*-BEEHAVE using the default settings, which seeds the landscape with 500 *B. terrestris* queens on 1st January. To check that this was an appropriate number of queens, simulations were run for 20 years and 20 runs, seeding each map with 500 and 1000 *B. terrestris* queens respectively. Badgers were not included in these test runs. The number of hibernating queens were then averaged across all maps and runs for the 500 and 1000 simulations, and plotted over time to check that the two lines overlap (Fig. 5.4). Both sets of simulations converge after ca. 9 years at about 750 hibernating queens, so 500 initial queens for the badger predation scenarios seemed reasonable and 20 years were a sufficient time for the population to reach equilibrium.

Simulations were run for each habitat map and for all combinations of badger predation rates (none, low and high) and food availability (high and low). The total number of hibernating queens, workers and colonies per day were requested as outputs from the model. To determine the quantities of nectar (l) and pollen (kg) available under high and low food availability, simulations were run for each map over a single year. This provided outputs on the total amounts of nectar (l) and pollen (kg) available each day for each of the habitat types present: grassland, hedgerow, scrub and woodland (Fig 5.3).

For analysis, it was important to choose a year when all landscapes had reached equilibrium (Fig. 5.4). The minimum time landscapes took to reach equilibrium was four years, with a mean convergence time of 6.8 years and a maximum convergence time of 16 years. We therefore chose year 16 as the time point for use in analysis, as all landscapes had converged by this point (Figure B.1).

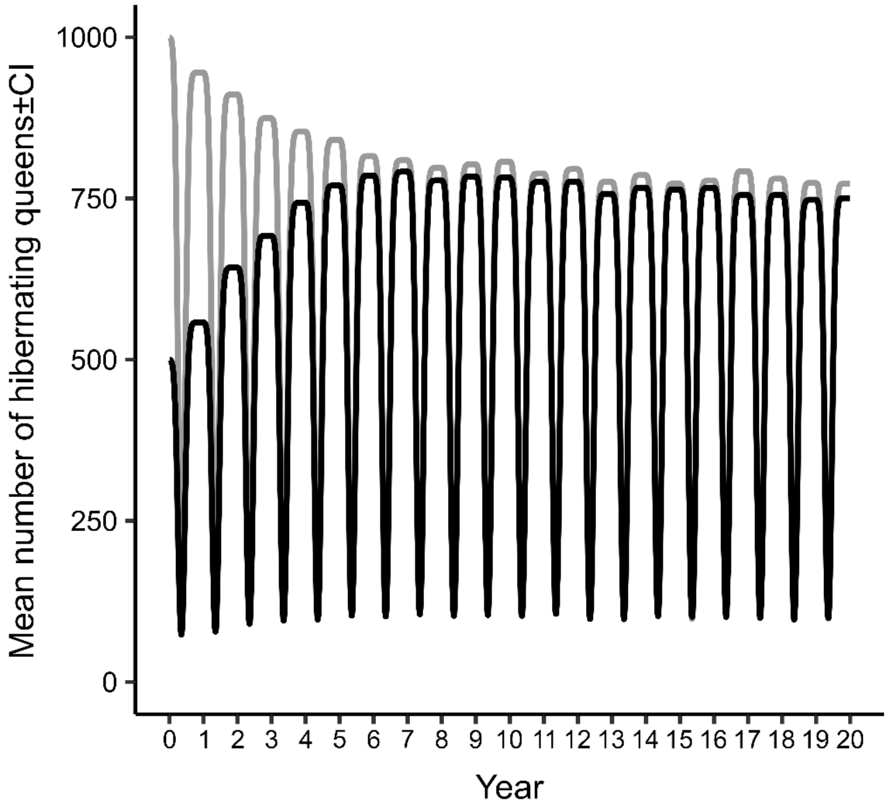

Figure 5.4 *Bumble*-BEEHAVE simulation output over 20 years when seeded with 500 (black line) and 1000 (grey line) *B. terrestris* queens in year 1 (from 20 runs). Populations are seen to be in a state of flux prior to convergence - where the two lines overlap.

5.3.2.4 Parameterisation of badger predation in the model

A unique feature of *Bumble*-BEEHAVE is its inclusion of badger predation within the model. Under default settings, the badgers home range is set to 735 m (ca. 170ha, Kruuk and Parish, 1981), and the probability that a badger comes across a certain nest (‘encounter probability’) is set to 0.19, a value representing the daily range of a badger as a percentage of its total home range (Kowalczyk *et al*., 2006). There is then an arbitrary value for the probability that when a badger encounters a nest it actually digs it up, which under default settings is 0.1. Badger setts can only be located in scrub habitat under default settings but can forage within any of the habitats present in the landscape. The number of badger setts is defined by the user but estimated predation risk assumes only a single active badger per sett. Badgers do not die or reproduce or show any activity other than randomly destroying bumblebee nests within their home range. Under the default settings badgers could only ‘nest’ in scrub habitats, this was updated to include woodland for the current simulations, as woodland is a key habitat for badger sett location (Feore and Montgomery, 1999; Harris, 1984; Smal, 1995).

To simulate the effects of badger predation to bumblebee populations, empirical predation rates were inputted into the model. As the number of badgers or the location of their setts is unknown in the current study, it is assumed that all colonies experience the same predation risk. Hence only a single badger is implemented, but with a foraging range large enough (5000 m) to cover the whole map. A corrected dig up probability was entered into the model based on our own empirical data, and alternative data from the literature (see Section 5.3.2.4.1 to 5.3.2.4.3).

5.3.2.4.1 Daily mortality rate of bumblebee colonies: empirical data

Initial simulations were run using the empirical data collected during the seasonal study in this chapter, which provided daily dig up probabilities of 0.04 for woodland, 0.05 for grassland and an average dig up probability of 0.04 for the hedgerow and scrub habitats, which were the four habitat types present in our habitat maps. These daily rates were calculated from the mean proportion of filled artificial nests which were dug up for each habitat using the following equation:

$$D = 1 - \ (1 - M_{overall\ i})^\wedge{}^{(\frac{1}{d})}$$

Equation 5.1. Equation for calculating the Dig Up probability (D) of bumblebee nests using the empirical data collected in the current study, which was collected over a three-day period. Overall Mortality ($M_{overall}$) is the mean proportion of filled artificial nests which were dug up in each habitat (i) over a period of (d) days.

As we had empirical data for the dig up probabilities, these would have incorporated the natural probability of wild badgers encountering the artificial nests and so in the model the encounter probability was set to 1. Using these predation rates, model simulations showed bumblebee populations collapsed after only 2-13 years (Fig. 5.5). Badgers are extremely widespread across the UK, and densities for Cornwall are relatively low (Table B.1). If badgers were consuming bumblebee colonies at this rate then we would expect the decline of wild bumblebees to be much steeper than has been recorded. As it is, bumblebees are still found in most areas in the UK and so the model was re-parameterised using alternative bumblebee predation rates to see how they compared to our empirical results.

5.3.2.4.2 Daily mortality rate of bumblebee colonies: alternative data

To re-parameterise the model, data from Goulson *et al.*, (2018) were used, which found that over a three year period 50 out of 908 bumblebee nests were dug up by a large mammal, presumed to be the European badger (*M. meles*). Unlike the data collected in our empirical study, observations were collected over multiple years. We parameterised Equation 5.1 with data from Goulson *et al*., (2018), setting ($M_{overall}$) to 0.05507 (= 50 / 908), and (d) to 98, assuming an average colony lifespan (and hence potential exposure do badger predation) of 98 days (Amin *et al.*, 2011; Beekman and Van Stratum, 2000). This results in a dig up probability (D) of 0.00058.

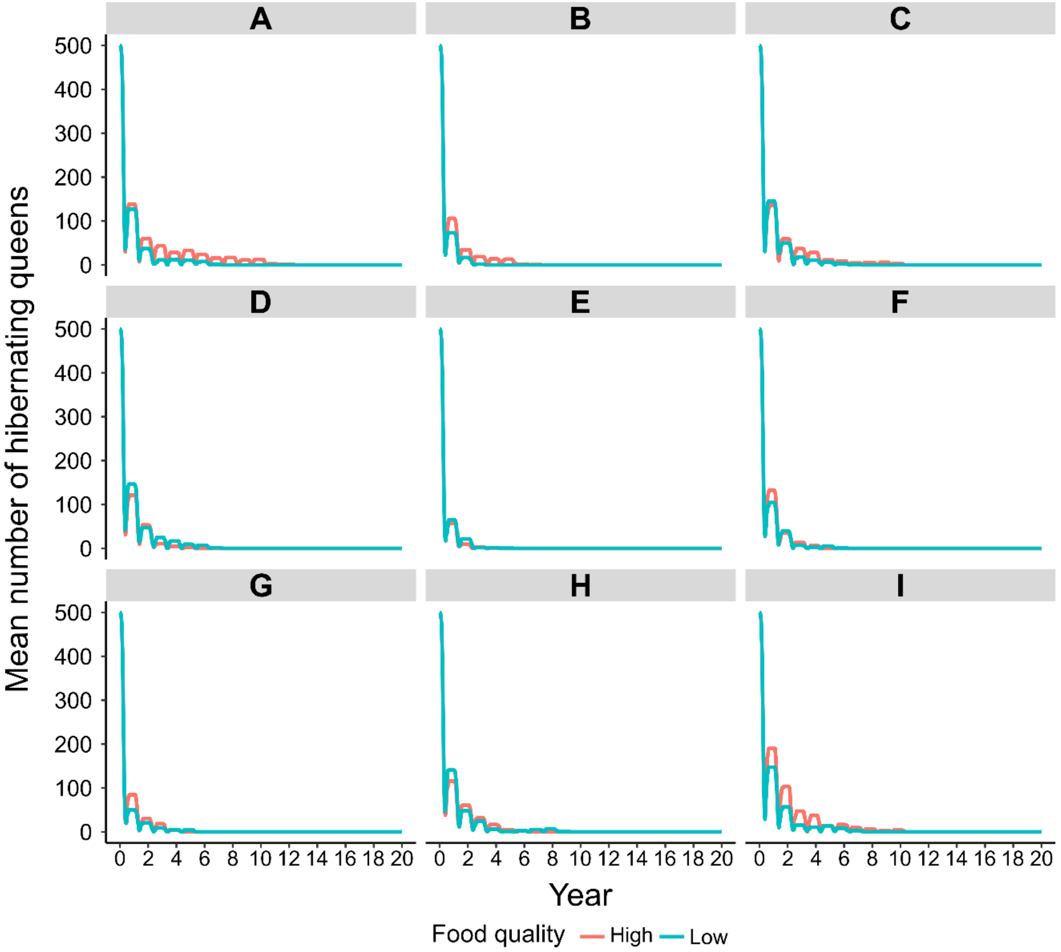

Figure 5.5 *Bumble*-BEEHAVE model simulation outputs showing the number of hibernating queens averaged across the 20 runs and the nine habitat maps for the empirical data collected during the current study. Lines represent the simulations run with high (red) and low (blue) food availability. Using the empirical badger predation rates, bumblebee populations collapsed after 2-13 years, therefore the model was re-parameterised using data from Goulson *et al*., (2018).

5.3.2.4.3 County correction factor

Under the default model settings, multiple badgers can be simulated into the model. This leads to some bumblebee colonies experiencing a magnified predation pressure due to being within the home range of more than one badger. When using values from empirical studies, the presence of multiple badgers would already be taken into account within the data. To explore the effects of high and low badger predation rates on bumblebee colonies, badger densities from the two geographic locations used in **Chapter 4** and the current chapter were used to apply a County correction factor, which is calculated as follows:

$$C_x = B_x \div B_A$$

Equation 5.3. Calculation of the County correction factor (C) for each location (x) using their respective badger densities (B) (Cornwall: 4.7 badgers per km^2; Woodchester Park, Gloucestershire: 16.6 badger per km^2) divided by the average UK badger density (B_A) (8.8 badgers per km^2 (see Table B.1)).

Using this equation a County correction factor (C) of 0.53409 for Cornwall and 1.88068 for Gloucestershire was obtained. These were then inputted into the following equation to calculate the corrected dig up probabilities for high and low badger predation rates:

$$D_{corrected\ x} = D_x \times C_x$$

Equation 5.4. Calculation of the corrected dig up probability for each location ($D_{corrected}$), through combining the dig up probabilities (D) and County correction factor (C) for each location (x).

Using this equation we calculated the corrected Dig up probabilities (D_{Cx}) of 0.00031 to simulate a low predation rate (like in Cornwall) and 0.00109 to simulate a high predation rate (like in Gloucestershire).

5.3.3 Analysis

5.3.3.1 Seasonal badger predation of artificial bumblebee nests

Analysis was performed using the statistical software R (version 3.5.2; R Core Team, 2017). A generalised linear mixed model was built to determine how ‘month’, ‘habitat’, ‘nest treatment’ and their combined three-way interactions affected badger predation rates of artificial bumblebee nests in Cornwall. A global model was built with a two-column response variable with ‘success’ and ‘fail’ i.e. the number of artificial nests which were dug up (‘success’) and the number which were left in the ground (‘fail’) for each nest treatment (control, surface and underground) for each transect (see Table B.3 for model code). The model was then fitted with a Binomial family and a cloglog link to normalise the residuals. ‘Transect ID’ was nested within ‘site’ as a random effect, and an observation level random effect (‘OLRE’) was added to account for over-dispersion (Browne *et al.*, 2005).

Analysis was also performed to determine if the predation rates recorded in the current study were significantly different to those collected in **Chapter 4** (even though data were collected in different years). For this analysis, only the August data from the current study was used to best match the timeframe when the Gloucestershire data were collected. A generalised linear mixed model was built with ‘success’ and ‘fail’ as a combined response variable, denoting whether the artificial nest was dug up or not, and ‘location’ was included as a fixed effect to denote whether the data were collected in Cornwall or Gloucestershire. ‘Transect ID’ was nested within ‘site’ as a random effect. Models were fitted with a Binomial family.

Model selection was performed using the dredge function from the MuMIn package (Bartoń, 2017), which uses Akaike Information Criteria model selection to compare all models to the global model. Models with a ΔAIC <2 were kept as the best fitting models.

5.3.3.2 Bumblebee population survival under varying predation rates and food availability using *Bumble*-BEEHAVE

The results of the *Bumble*-BEEHAVE simulations were analysed using linear models in the statistical software R (version 3.5.2; R Core Team, 2017) to look for effects of badger predation rate and food availability on bumblebee populations. Data from year 16 was used for analysis, as this is when all landscapes had converged (Fig. 5.4). The maximum ‘number of hibernating queens’, ‘number of adult workers’ and ‘number of colonies’ for each map were averaged across the 20 runs. These were then used as the response variables in the models. ‘Badger predation’ (none, low and high) and ‘food availability’ (low and high), and their two-way interaction were included in the global model as fixed effects, except for the maximum queen model which would only converge without the interaction. ‘Site’ was included as a random effect.

To determine whether the nectar (l) and pollen (kg) was significantly different between the two food availability scenarios a generalised linear model was conducted with the total annual ‘nectar (l)’ and ‘pollen (kg)’ as respective response variables, and ‘food availability’ (high and low) as the fixed effect.

‘Site’ was included as a random effect. Models were fitted with a Gamma family and an identity link function.

5.4 Results

5.4.1 Seasonal badger predation of artificial bumblebee nests

In total 177 out of 1772 pots were dug up by badgers during the three survey periods, equating to 10% of total pots. Month by month, 56 out of 598 artificial nests (9.4%) were dug up by badgers in April, 54 out of 584 artificial nests (9.2%) were dug up in June and 67 out of 590 artificial nests (11.4%) were dug up in August. Comparing the two habitats, 76 out of 885 artificial nests (8.6%) were dug up in woodland and 101 out of 887 artificial nests (11.4%) were dug up in grassland habitats. 39 out of 587 (6.6%) control nests were dug up, 73 out of 588 (12.4%) surface nests were dug up and 65 out of 597 (10.9%) underground nests were dug up.

Camera traps confirmed that no other species except badgers interacted with the artificial nests during the study. Badgers were recorded on the camera traps at all nine sites; in nine of the woodlands and seven of the grassland habitats. Across the three survey periods they were recorded at six sites in April and August and eight sites in June.

Surface nests were dug up significantly more than control pots (z=2.23, p=0.026; Table B.3, Fig. 5.6), but there was no significant difference between the number of control and underground nests which were dup up (z=0.79, p=0.429; Table B.3, Fig. 5.6). There was also no significant difference between habitat types, months or their interactions, all of which were selected out of the final model. Predation rates in Gloucestershire (**Chapter 4**) were significantly higher than those in Cornwall (z=2.65, p<0.01; Table B.4).

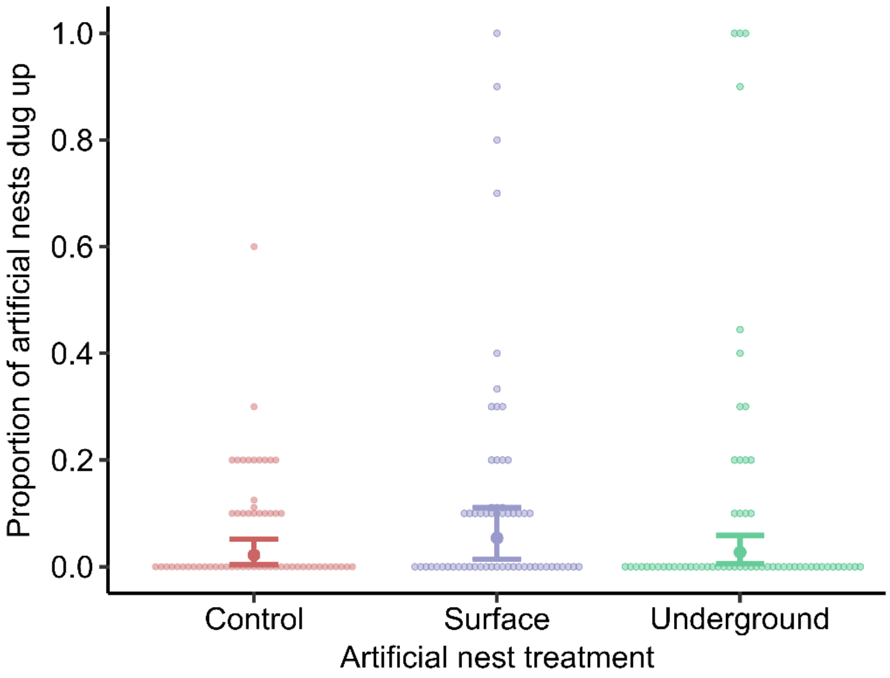

Figure 5.6 Predicted mean and confidence intervals for the proportion of artificial nests dug up by badgers for each treatment are shown, using model predictions from the best fitting model (Table B.3). Raw data is represented with a beeswarm plot.

5.4.2 Bumblebee population survival under varying predation rates and food availability using *Bumble*-BEEHAVE

Analysis confirmed that, there were significantly lower quantities of nectar (l) and pollen (kg) available in the landscapes parameterised to have low food availability compared to those with high food availability (Nectar: t=-14.69, p<0.001; pollen: t=-14.20, p<0.001; Fig 5.3; Table B.5).

Under high badger predation there were significantly fewer hibernating queens, workers and colonies produced compared to no badger predation (Table 5.2) (Hibernating queens: t_{42}=-3.62, p<0.001; workers: t_{40}=-2.50, p=0.017; colonies: t_{40}=-2.50, p=0.017; Fig. 5.7, Table B.6). Under low badger predation there was no difference in the numbers of hibernating queens, workers or colonies produced compared to no predation (Table 5.2) (Hibernating queens: t_{42}=-0.99, p=0.328; workers: t_{40}=-0.63, p=0.530; colonies: t_{40}=-0.49, p=0.626; Fig. 5.7, Table B.6).

Under low food availability there were fewer hibernating queens, workers and colonies produced compared to the high food availability (Table 5.2) (Hibernating queens: t_{42}=-10.04, p<0.001; workers: t_{40}=-5.57, p<0.001; colonies: t_{40}=-5.40, p<0.001; Fig. 5.7, Table B.6). No significant interactions were observed between badger predation rates and food availability.

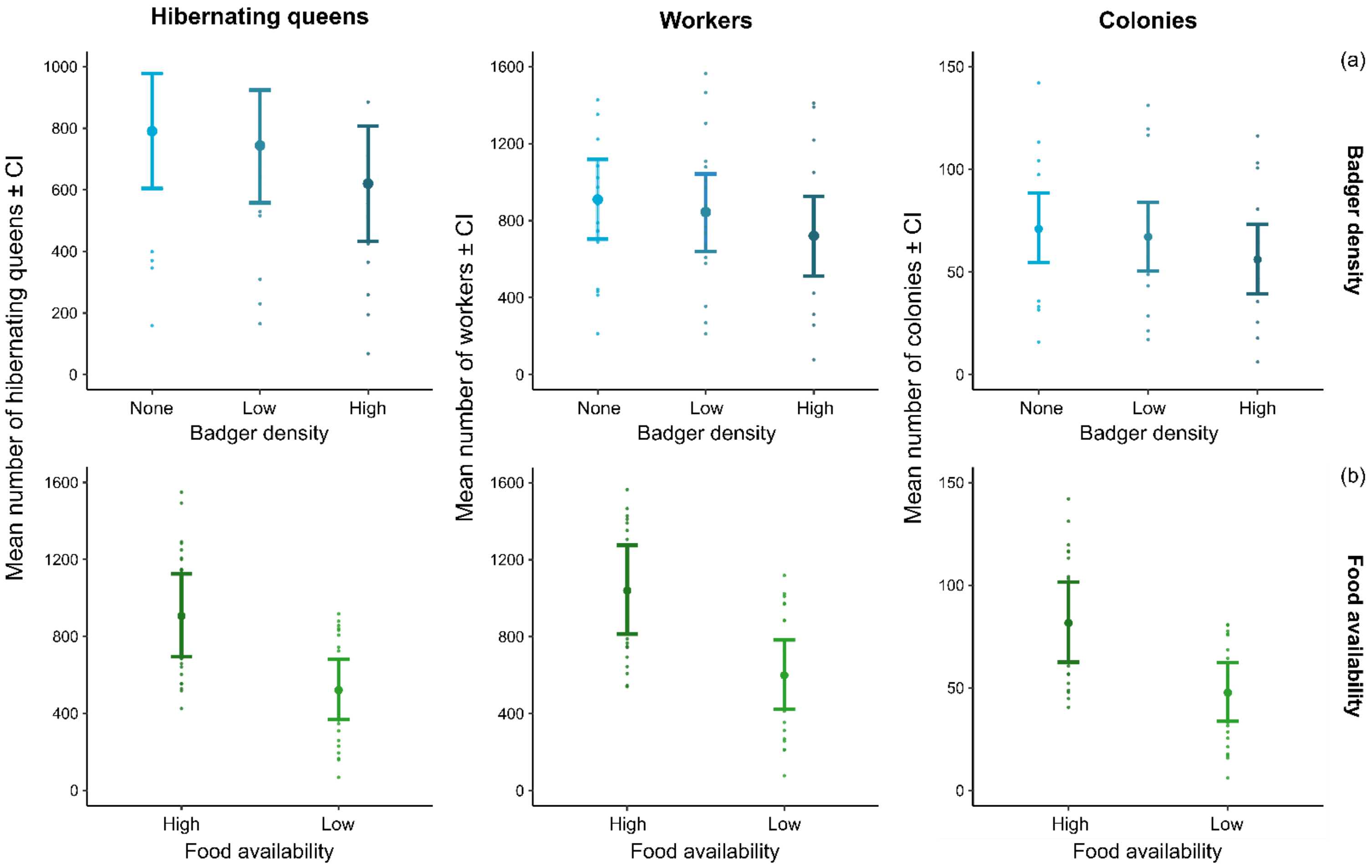

Figure 5.7 *Bumble*-BEEHAVE simulation outputs for the effects of (a) badger predation and (b) food availability on the maximum number of hibernating queens, workers and colonies produced for each landscape (n=9), averaged across the 20 runs. Predicted mean and confidence intervals are shown, generated from the best fitting models for each output (Table B.6), with raw data shown as points.

Table 5.2 The mean ± SE for the average number of hibernating queens, workers and colonies produced per landscape (n = 9) in Year 16 using the Bumble-BEEHAVE simulations.

		Hibernating queens	Workers	Colonies
Badger predation	None	787 ± 47.8	904 ± 54.9	71 ± 4.29
	Low	740 ± 48.3	839 ± 51.8	67.1 ± 4.35
	High	616 ± 44.9	715 ± 50.4	56 ± 3.99
Food quality	High	907 ± 43.7	1039 ± 46.9	81.7 ± 3.93
	Low	521 ± 34.6	599 ± 39.5	47.7 ± 3.13

5.5 Discussion

The current study utilised the novel artificial nest technique developed in **Chapter 4**, to determine whether it was capable of discerning the seasonal effects of badger predation to bumblebee nests reported in the literature (Cleary *et al.*, 2009). No such effects were observed, this is discussed in light of reduced badger activity due to lower badger densities, annual differences in predation and possible low detection due to lack of knowledge of sett foraging territories in the current study. The population model *Bumble*-BEEHAVE (Becher *et al.*, 2018) was then used to simulate, for the first time, the long-term population effects of badger predation of bumblebee colonies, under high and low food availability. Simulations showed that high rates of badger predation negatively impacted bumblebee colony outputs (Table 5.2), even when food availability was high, reducing the numbers of hibernating queens, workers and colonies (Fig 5.7). There was no interaction between predation and food availability, suggesting that populations in low quality habitats are not being more adversely affected by top-down effects such as predation. Food availability alone had the biggest impact on all colony outputs, with significantly more hibernating queens, workers and colonies being produced in landscapes with high food availability (Fig 5.7; Table 5.2). These findings are discussed in relation to the importance of further understanding how top-down and bottom-up regulators may act together to affect populations.

Seasonal predation of bumblebee nests was not detected using the artificial nest method in the current study. Badgers' diets vary seasonally in accordance with prey abundance (Kruuk and Parish, 1981; Shepherdson *et al.*, 1990), with evidence that their consumption of invertebrates, including bees, peaks between June-August (Cleary *et al.*, 2009; Goulson *et al.*, 2018b). The lack of seasonal variation in predation was detected in the current study could have been due to a number of reasons. Firstly, badger diets vary with prey abundance (Kruuk and Parish, 1981; Shepherdson *et al.*, 1990). In the current study, a small and constant amount of nest material was used in the artificial nests (7 g). Using a constant amount of nest material was important for standardisation and consistency, but does not mimic the natural growth of wild bumblebee nests, which would likely affect their detectability by badgers. Colonies tend to reach their maximum sizes in June-July (Muller and Schmid-Hempel, 1992), with queens also being produced around this time. Bumblebee nests are more detectable by humans at this time, shown during citizen science studies (Goulson *et al.*, 2018b; Lye *et al.*, 2012) and this is likely reflected by increased predation and consumption by badgers (Cleary *et al.*, 2009). Alternatively, if badgers were showing an increased *preference* for bumblebee nests during June-August then an increase in the number of artificial nests dug up would have been expected. As this was not seen, it is likely that the artificial nest experiment is providing information representative of badgers' natural foraging behaviour, which is linked to prey *availability*.

The predation rate of artificial nests in the current study was significantly lower than those observed in **Chapter 4**. These two experiments were conducted in different years, and in different geographic locations (Cornwall and Gloucestershire), which could explain some of the observed variation. Certain methodological differences could also have contributed; during the Gloucestershire study in Woodchester Park (**Chapter 4**), the exact sett locations and their foraging boundaries were known (Delahay *et al.*, 2006). This meant that transects at the same site were exposed to the same numbers of badgers. In the current study, which took place in Cornwall, this information was not available. Transects were placed within 1 km of each other at the same site to increase the chances of being within the foraging territory of the same sett but nonetheless transects had the potential to be placed within the foraging territories of different badger setts; thus being exposed to different numbers of badgers. If this was the case, then a bigger difference between the two habitats may have been expected, which was not seen. Therefore, due to the scale of differences observed between the two studies, it is likely that overall badger densities and territory sizes played a role.

Woodchester Park, Gloucestershire where the majority of data were collected in 2017 (**Chapter 4**), has one of the highest badger densities in the UK (average of 16.6 adults per km^2 (Rogers *et al.*, 1997); Table B.1). Cornwall, where the current study was undertaken, has ~5 times lower density in comparison (4.7 badgers per km^2 (Cheeseman *et al.*, 1981); Table B.1). These are both above and below the estimated average badger densities for the UK (8.8 badgers per km^2 (see Table B.1)). As well as having different densities, setts at the two locations differ in their territory sizes, with setts in Gloucestershire having smaller territories than those in Cornwall (Feore and Montgomery, 1999). Despite badgers within the same sett showing individual foraging specialisation (Robertson *et al.*, 2014, 2015), they have overlapping ranges within their territories (Kruuk, 1978; Roper, 2010) and thus in **Chapter 4**, there were likely more badgers exploiting a smaller area which would increase the chances of multiple badgers detecting the artificial nests and therefore could have led to the higher predation rates seen. Due to badgers exploiting individual foraging niches (Robertson *et al.*, 2014, 2015), it may be that within a territory it is specific individuals who are focussing on bumblebee nest predation, however this is not something that the current study was able to determine.

To look at the long-term effects of badger predation on bumblebee nests, the population model *Bumble*-BEEHAVE (Becher *et al.*, 2018) was used. Simulations were run using predation rates from the empirical data collected in the current study, which found average daily predation rates of 0.04. Using this empirical data modelled *B.terrestris* populations went extinct after only 2-13 years. This is unlikely to be a realistic representation of what is happening under natural conditions. Badgers are a widespread and common species within the UK, and if predation of bumblebee nests was causing extinctions at this rate then declines of wild bumblebees would be much steeper than is currently seen. Data on bumblebee nesting densities within the two locations would be useful to determine if badger predation, especially in Woodchester Park where the highest predation rates were observed (**Chapter 4**) are having impacts on actual nesting densities. This would help validate the findings of the model. Reasons for the high extinction rates observed using the empirical data could have been due to a number of reasons both relating to the field study and the model itself.

Firstly, the cost of digging up an artificial nest is likely lower than that of digging up a wild bumblebee nest. This is due to both the physical energy required to extract the artificial nest from the ground, and also the fact that no live bumblebees are present to defend the nest. Bumblebees have been shown to make a hissing noise in response to mammals breath (Kirchner and Röschard, 1999), a defence signal which was enough to deter mice from entering the nest. Other defence mechanisms include the use of honey-daubing by certain species (Fuller and Plowright, 1986), where workers cover intruders with regurgitated liquid. A more common defence mechanism is stinging (Free, 1958). In the absence of these defence mechanisms badgers are less likely to be disturbed or deterred whilst digging up the artificial nests and thus a potentially higher predation rate than under natural conditions is observed. Although, the energetic reward to a badger from a wild bumblebee colony is likely greater than that of the artificial nests due to the larger amount of nest material they would contain and this the predation rate of wild nests by badgers may be lower than those observed in our study but higher than those recorded by Goulson *et al*. (2018).

Secondly, high extinction rates using our empirical predation rates could have been a construct of the model itself. *Bumble*-BEEHAVE (Becher *et al.*, 2018) does not allow the immigration or emigration of bumblebee queens into the modelled landscape. Under natural conditions queens are known to disperse over a kilometre from their natal colony (Carvell *et al.*, 2017). This means that the effects of predation may be buffered by queens immigrating into the area from the surrounding landscape.

High extinction rates caused by the empirical results lead to the model being re-parameterised using data recorded by Goulson *et al.*, (2018). This gave a much lower daily predation rate of 0.00058. When applying the County correction factor (Section 5.3.2.4.3) to take into account the different badger densities in the two locations and to create a high and low badger predation rate, the predicted daily predation probability was 71.6% lower for Cornwall compared to Gloucestershire. This was similar to the 63.6% difference in predation probability between the empirical Cornwall and Gloucestershire data collected in this study and **Chapter 4**. Therefore, although the empirical dig up rates observed using the artificial nest method are inflated compared to real predation events, they provide useful information into the relative differences in predation between geographical locations and habitats and are thus a useful tool for studying the relative effects of badger predation to bumblebee nests.

Simulations using *Bumble*-BEEHAVE showed that under high badger predation we see a significant decrease in the numbers of hibernating queens, workers and colonies produced by the population in each landscape (Fig. 5.7, Table 5.2). Simulated low predation rates had no effect on bumblebee populations or colony outputs. Cornwall has badger densities lower than the UK average but similar to badger densities in many other areas around the UK (Table B.1). These results suggest that badger predation in areas with similar densities to Cornwall are unlikely to have serious implications for bumblebee populations. No significant interaction was observed between predation rate and food availability, suggesting that populations in areas of high floral resources are not buffered against the effects of predation. Predators can impact their prey through direct mortality, or indirectly through causing behavioural changes. Effects of behavioural changes due to predator presence can be similar in strength or stronger than those of direct consumption (Preisser *et al.*, 2005). Bumble bee wolf (*Philanthus bicinctus*) aggregations were found to have negative effects on bumblebee forager densities and seed set (Dukas, 2005), potentially through both direct mortality and avoidance, and crab spider presence on flowers caused reduced visitation and handling times (Dukas and Morse, 2003; Romero *et al.*, 2011). Badgers differ to both these types of predators. Firstly, they are likely foraging opportunistically on nests rather than targeting them specifically; evidenced by the low percentages of bees and wasps seen in badger diets (Cleary *et al.*, 2009; Kruuk and Parish, 1981). Secondly, they target the entire colony rather than individual workers; impacting colonies through direct consumption rather than causing behavioural changes to occur. One of the only ways food availability may cause increased badger predation is through larger nests being potentially more detectable. Colonies in areas of high food availability may therefore be more at risk of predation. This was not taken into account in the current model, and more empirical evidence into the actual mechanisms behind badger predation of bumblebee nests is needed in order to determine if this would be the case.

Food availability had the greatest impact on bumblebee populations. To understand the effects of reduced food availability on bumblebee populations, simulations were run with floristically rich grassland and floristically poor grassland, similar to improved grassland. Grassland is an important natural habitat for pollinators (Öckinger and Smith, 2007), but between 1932 and 1984 the area of 'unimproved grassland' declined by over 90% in Britain (Fuller, 1987). Floristically rich grassland is being lost due being converted to improved grassland for agriculture or for arable cultivation, as well as through the change from hay production to silage (Ridding *et al.*, 2015; Stoate, 1996). Improved grassland is one of the most abundant grassland types in the UK (Haines-Young *et al.*, 2000), but has low floral abundance and diversity (Baude *et al.*, 2016), with the outputs from our simulations showing that landscapes with low quality grassland contained significantly less nectar and pollen compared to the floristically rich grassland. Maps containing floristically rich grassland supported more hibernating queens, workers and colonies compared to landscapes which contained low quality grassland and therefore less food availability. Despite its low floristic richness, due to its large area improved grassland contributes greatly to national nectar resources (Baude *et al.*, 2016), and would therefore benefit from better management to increase its potential for pollinators even further. Our simulations show that reduced food availability can significantly impact bumblebee populations, and support the suggestions by Baude *et al.,* (2016) that managing areas of improved grassland to be more floristically rich would be beneficial for pollinator conservation.

5.6 Conclusion

A combination of field techniques and population models were used to understand the effects of badger predation on bumblebee nests under wild conditions and over multiple years. The artificial nest technique developed in **Chapter 4** was used to determine whether the seasonality of badger predation could be detected using this method. No seasonal effects were found, possibly due to the artificial nests not changing in size across the study as would be the case for wild bumblebee nests. Badger predation peaks during the same time as colonies reach their maximum sizes (Prŷs-Jones and Corbet, 2011) and so badger predation may have greater impacts on queen producing colonies, which in turn could lead to colonies in areas of high food availability experiencing higher predation rates. The current model is not set up to test this, but could be modified in the future to do so if further empirical data are collected. To monitor the combined effects of badger predation rates and food availability the population model *Bumble*-BEEHAVE (Becher *et al.*, 2018) was used. Under high predation rates there were significantly fewer hibernating queens, workers and colonies produced, but overall food availability had the biggest impact on colony outputs (Fig. 5.7). No interaction between food availability and predation was observed. This likely reflects natural scenarios, due to badgers only causing direct mortality to colonies rather than initiating any behavioural avoidance responses as is the case with the presence of predators during foraging trips (Romero *et al.*, 2011). The main way in which food availability could impact badger predation is through larger colonies being more detectable to badgers, which is not taken into account in our simulations. Further studies are needed into the underlying mechanisms driving badger predation of bumblebee nests: is it opportunistic or are individual badgers within setts showing specialisation towards bumblebee nest predation? What role does the size of bumblebee nests have on their detectability and therefore predation rates? The importance of floristically rich grassland to bumblebee populations and colony outputs is also highlighted using the model, further supporting the argument for improved grassland to be managed to produce a floristically rich habitat for pollinators in order to aid their conservation.

Chapter 6

General discussion

Chapter 6: General discussion

6.1 Thesis overview

Firstly, I will summarise my main results, before commenting on the general themes of botanical gardens, badger predation and novel research methods. I will finish by thinking about the future areas of research which would complement and build upon the findings of this thesis. The chapters in this thesis have aimed to fill gaps in our knowledge of bottom-up and top-down regulating factors affecting bumblebees using a mixture of classical field techniques, novel methods and computer models. **Chapter 2** focussed on botanical gardens and semi-natural farmland habitats within a rural setting, exploring the use of these habitats by nest searching queens and foragers across the season. **Chapters 3-5** then went on to develop and implement novel techniques to study some of the hidden aspects of bumblebee ecology: using thermal cameras to locate wild bumblebee nests (**Chapter 3**) and developing and testing an artificial nest technique to quantify rates of badger predation on bumblebee nests (**Chapter 4 & 5**). A population model was then used to look at, for the first time, the effects of badger predation and food availability on bumblebee populations over multiple years (**Chapter 5**). Using novel techniques enabled research which goes beyond the scope of many studies which simply look at the relationship between bees and forage.

6.2 Early season resources and nest searching

In the South West, the landscape is largely agricultural with a focus cattle pasture and some cereals (Easy *et al.*, 2012). Botanical gardens are a historic part of the Cornish landscape (Smith and Mulholland, 2008), providing a unique opportunity to explore how the presence of a large, concentrated area of flowering resources affects bumblebee foraging across the season in a rural agricultural context. **Chapter 2** found that botanical gardens are providing an important early season habitat for bumblebees; containing higher numbers of nest searching queens and providing a pulse of spring resources. In the absence of mass flowering crops, which are a key provider of early season resources in agricultural habitats (Knapp *et al.*, 2018; Westphal *et al.*, 2003), botanical gardens may buffer bumblebee populations within agricultural landscapes during this time of low resource availability. This can be seen in the semi-natural farmland habitats within the current study. Within the botanical gardens, 40.2% of early spring floral units from bumblebee food plants were from a single *Rhododendron* species, and this was the most visited species during this survey period. This highlights the important role exotic species can play in extending the blooming periods for pollinators (Salisbury *et al.*, 2015).

A greater number of nest searching queens were recorded in the botanical gardens, and this was not related to floral resources. O'Connor *et al*. (2017) also found this, and found that the number of nests found later in the season was related to nest searching queen numbers. Therefore, botanical gardens may contain high numbers of bumblebee nests, similar to residential gardens (Osborne *et al.*, 2008b). Suzuki *et al*., (2009) also found that nest searching queens did not respond to floral resources, but that nests themselves were more likely to be located in areas of high floral availability. This makes sense, as queens need a high amount of energy during this period (Heinrich, 1974; Suzuki *et al.*, 2009), and spring forage has been linked to colony growth (Westphal *et al.*, 2009; Williams *et al.*, 2012). Therefore, nest searching queens may be choosing overall habitats based on floral resources, but not the specific features where they choose to nest search. If this is the case, it could explain why higher numbers of nest searching queens are observed in the botanical gardens. The nest searching surveys ended as floral resources in botanical gardens reduced, and those in semi-natural farmland habitats increased (Fig. 2.2). Semi-natural farmland habitats have also been shown to contain high numbers of bumblebee nests (Osborne *et al*., 2008b), and therefore it may be that farmland habitats in the current study are attracting nest searching queens which emerge later. This was outside the scope of the current study, but is an area which future studies should consider.

Alternatively, the higher numbers of nest searching queens seen in the gardens could be because of gardens having better hibernation sites, due to being less disturbed areas compared to agricultural habitats (Kremen *et al.*, 2007) and thus potentially having higher baseline populations of queen bumblebees. In this scenario, botanical gardens would be acting as population 'sources'. Queens are known to disperse between 1,227 ± 125 m from their natal colony (Carvell *et al.*, 2017), but it is not known whether this occurs before or after hibernation, which is potentially important in determining the success of colonies (Fig. 6.1). Both habitats studied in **Chapter 2** have the potential to contain high densities of bumblebee nests (Osborne *et al*., 2008b), and our findings show they supported similar numbers of bumblebees. However, queens which are produced from nests in the botanical gardens may be at a disadvantage, as they have the greatest potential to disperse into a habitat with lower spring forage (Fig. 6.1). Survival of queens from one year to the next was significantly affected by the availability of habitats containing high-value forage (Carvell *et al.*, 2017), therefore queens produced from nests in the botanical gardens may do less well in subsequent years (Fig. 6.1). Whereas queens which are produced in the surrounding agricultural landscape have the propensity to disperse into the botanical gardens, and therefore experience greater benefits (Fig. 6.1). Although as bumblebees can travel further than some smaller-bodied pollinators (Greenleaf *et al.,* 2007), nests which are outside of the botanical gardens are still likely benefitting from the resources within them. Studies into how colonies are utilising the botanical gardens within the landscape would provide further information into this. Thus, the source-sink dynamic between botanical gardens and the surrounding landscape could mean that bumblebee populations in proximity to these habitats are stable. The overall benefit depends on the timing of dispersal by queen bumblebees (Fig. 6.1), which is currently unknown and should therefore is an important area for future research.

Figure 6.1 Flow diagram showing the source-sink dynamics for queens which are produced in the semi-natural farmland habitats and the botanical gardens, depending on whether they disperse before (filled lines) or after (dashed lines) hibernation. The possible benefits (+) and costs (-) of each habitat are also shown. Arrows show what happens to nests in the next year, assuming that they are only dispersing between the two habitats which were studied in **Chapter 2** although in a natural system there would be a wider variety of habitats which they might disperse into.

6.3 Floral resources across the flight season

Chapter 2 found that semi-natural habitats, such as Cornish hedgerows, surrounding agricultural crop fields appear to be good habitats for bumblebees; providing high amounts of floral resources towards the end of spring and early summer, when floral resources had reduced in botanical gardens. Landscape complexity has positive effects on pollinators (Happe *et al.*, 2018; Kennedy *et al.*, 2013; Kleijn and van Langevelde, 2006; Steffan-Dewenter *et al.*, 2002). **Chapter 2** therefore supports the broader concept that complex landscapes, containing small-scale habitat features such as hedgerows, have beneficial effects to pollinator populations. These benefits could be through the provision of nest sites (Osborne *et al*., 2008b), navigational aid (Cranmer *et al.*, 2012) and increased floral resources (Cole *et al.*, 2017; Timberlake *et al*., 2019). This likely explains why the original prediction that botanical gardens would have higher bumblebee abundances, as has been found for residential gardens (Baldock *et al.*, 2019), was not met. The positive effects of botanical gardens may be greater in simple, more intensive agricultural landscapes. Cole *et al.,* (2017) found that arable crop fields contained the lowest floral and pollinator abundance and diversity compared to semi-natural habitats including grassland, woodland, hedgerows and scrub. Bumblebees are more attracted to, and will travel further to visit, patches of high floral availability when the surrounding landscape is florally poor (Heard *et al.*, 2007; Pope and Jha, 2017). Future studies looking into the usage of botanical gardens in different types of landscape, and the numbers of colonies they can support, would help to show how they support pollinators within the wider landscape.

6.4 Locating and monitoring bumblebee colonies

Developing better nest detection techniques would allow sufficient numbers of wild bumblebee nests to be monitored during a single season, allowing the effects of stressors such as disease and predation of different species to be studied more fully. Currently most of our understanding on stressors comes from using commercial colonies of *B. terrestris* or *B. impatiens* in the US (Goulson *et al.*, 2002; Schmid-Hempel and Schmid-Hempel, 1998; Spiesman *et al.*, 2017). Monitoring the impacts of stressors on bumblebee populations in the field can be challenging (Muller and Schmid-Hempel, 1992; Wood, Holland, Hughes, *et al.*, 2015), bumblebee colonies are difficult to detect, due to their small size (Prŷs-Jones and Corbet, 2011). Current nest detection methods involve human searches (O'connor *et al.*, 2017; O'Connor *et al.*, 2012) which rely on visual cues, and sniffer dogs have also been tested (O'Connor *et al.*, 2012; Waters *et al.*, 2011), which rely on olfactory cues, but were found to be ineffective. In **Chapter 3** thermal cues from bumblebee nest traffic was used to test whether thermal cameras were a better nest detection method compared to visual and olfactory cues (Roberts and Osborne, 2019). Thermal cameras homogenise the viewing area, making it less visually complex and thus heat signatures of bumblebees stand out more strongly against the background – partially reducing the difficulty of separating the bees from the vegetation. Thermal cameras, much like sniffer dogs (O'Connor *et al.*, 2012), were found to be a less effective method compared to human searches. This was mainly due to the small viewing area of the thermal camera, suggesting that cameras with larger lenses or wrap-around glasses, enabling the user to utilise their peripheral vision would increase success. In the absence of better nest detection methods, alternative methods must be developed in order to study the effects of stressors on bumblebees, and so **Chapters 4 and 5** aimed to develop and test an artificial nest method to study badger predation.

6.5 What happens to colonies?

Once nests have been established, they can succumb to predation. Due to the difficulty of finding wild nests, as highlighted in **Chapter 3**, a novel artificial nest method was developed to study badger predation of bumblebee nests in two different habitats in two locations (**Chapter 4 & 5**). The method was successful and, in **Chapter 4**, showed that predation rates were higher in woodland than grassland, but did not vary with nest depth. Overall, predation rates were much higher in Gloucestershire (**Chapter 4**) than in Cornwall (**Chapter 5**). These differences may have been due to varying badger densities. Badger densities in Gloucestershire and Cornwall are higher and lower than the UK average (Table B.1), respectively. Badgers also have smaller territories in Gloucestershire compared to Cornwall (Feore and Montgomery, 1999). This means there is an increased chance that a badger will detect an artificial nest, as within the territory badgers ranges overlap (Kruuk, 1978; Roper, 2010). Badgers have seasonal diets (Kruuk and Parish, 1981; Shepherdson *et al.*, 1990), with bumblebee consumption peaking in June-August (Cleary *et al.*, 2009). **Chapter 5** used the artificial nest method to detect this seasonality, but was unsuccessful. Badgers forage on prey items relative to their availability (Kruuk and Parish, 1981). Wild bumblebee nests reach their maximum sizes at a similar time to peak consumption by badgers (Muller and Schmid-Hempel, 1992). Artificial nests were a constant size, and so probably had the same chances of detection across the season, therefore badgers may be predating bumblebee nests depending not only on their availability, but their detectability. The artificial nest method could be developed in order to confirm that badgers do target bumblebee nests depending on their detectability, through adjusting the quantities of nest material within each nest.

6.6 Using simulations to study multiple stressors

The use of models is a common method of exploring overall population effects from a variety of stressors. Computer models provide a unique platform to simulate changes in populations over long time periods, and isolate the effects of specific stressors (Bryden *et al.*, 2013b; Crone and Williams, 2016). This clarity is difficult to attain in the field, due to the variety of biotic and abiotic factors present. In **Chapter 5**, the model *Bumble*-BEEHAVE (Becher, *et al.*, 2018) was used to build on the empirical studies into the effects of badger predation on bumblebee nests (**Chapter 4 and 5**), under high and low food availability. The model was parameterised with data from Goulson *et al.,* (2018), due to the empirical predation rates recorded in **Chapter 4** causing high levels of extinctions within the model.

Reasons for the high levels of extinctions in the simulations using our empirical data were likely threefold: firstly, the artificial nests used would have required much less effort to dig up than a wild bumblebee nest, particularly those underground. There would also have been no defensive behaviour from bumblebees (Free, 1958; Kirchner and Röschard, 1999), meaning there were lower costs compared to when predating a wild bumblebee colony. Empirical values from the artificial nest method may therefore overestimate bumblebee nest predation. Secondly, this could have been an artefact of the model itself which does not allow bumblebee queens to immigrate or emigrate from the modelled landscape. This would happen under natural conditions, potentially leading to populations being buffered against the losses from badger predation. Areas of high badger predation might be acting as sinks for bumblebee populations, with areas of low badger predation acting as sources of bumblebee queens which can re-colonise the sink habitats. Similar occurrences have been seen in areas surrounding beewolf aggregations which reduce the numbers of bumblebee foragers during peak beewolf activity (Dukas, 2005). They do not look at population effects, but discuss the importance of queens moving in from surrounding areas. Finally, the predation rates in our study may have been higher than those reported by Goulson *et al*., (2018) due to methodological differences. Chapter 4 and 5 were conducted in areas of known badger activity, whereas the presence of badgers was not known in all the locations where bumblebee nests were found in the Goulson study. Hence their predation rates are likely to be lower, and perhaps therefore representative of average predation rates across the UK rather than specific predation rates for areas of particular badger densities.

Using *Bumble*-BEEHAVE (Becher, *et al.*, 2018), floral availability was found to significantly impact on bumblebee colony outputs, with low food availability causing significantly fewer hibernating queens, workers and colonies to be produced per landscape (**Chapter 5**). These findings support experimental studies with reduced food availability impacting colony growth and queen production (Crone and Williams, 2016; Rotheray *et al.*, 2017). England and Wales experienced major declines in nectar availability between the 1930s and 1970s (Baude *et al.*, 2016), which stabilised and started to increase by 2007. Food availability in the simulations were adjusted through reducing the quality of grassland from a florally rich habitat to one that only contains two flower species, representative of ‘improved grassland’ found in Cornwall (Knapp *et al.*, 2018). Thus corroborating the suggestion by Baude et al., (2016) that increasing the floral richness of improved grassland will increase the national nectar reserves, due to the vast area of improved grassland within the UK. Carvell, (2002) also showed that grassland management had impacts on bumblebee abundance and species richness, with cattle-grazed grasslands being of most importance, containing high abundance and diversity of flowering plants. Grassland is also a likely important nesting habitat, as although it contained fewer nests than linear countryside features (Osborne *et al.*, 2008b), due to its large area in the UK the numbers of nests per area is likely to be high. Cattle grazed grassland is also a key foraging habitat for badgers (Kruuk, *et al.*, 1979), and so although **Chapter 4** found that badger predation was higher in woodland habitats, the importance of grassland for both badgers and bumblebees makes it a key habitat where high levels of contact are likely occurring. This highlights the importance of further understanding the dynamics of badger predation both empirically, and then utilising models to understand long-term population impacts.

6.7 General themes

6.7.1 Botanical gardens

Throughout this thesis, a key habitat where studies have been performed (**Chapter 2 & 3**) has been botanical gardens. The composition of botanical gardens varies worldwide (Pautasso and Parmentier, 2007), with their main focus being to preserve and conserve plant species, often outside of their native geographic ranges (Blackmore *et al.*, 2011; Chen and Sun, 2018; Hardwick *et al.*, 2011). Cornish gardens historically contain a number of *Camellia* and *Rhododendron* varieties (Smith and Mulholland, 2008). Bumblebees are frequent visitors of *Rhododendron ponticum* (Tiedeken and Stout, 2015), with queen bumblebees likely being particularly important pollinators of these plants (Stout, 2007). The use of *Camellia*s by bumblebees is less studied, with Diptera being the most common and effective pollinator of *Camellia sinensis* in Sri Lanka (Wickramaratne and Vitarana, 1985), and bees making up only a small number of seasonal visitors. In **Chapter 2** bumblebees in botanical gardens were not observed visiting *Camellias*, but 10.1% of all visits were to *Rhododendron*s, with all of these occurring in early spring and early summer. Overall, exotics accounted for over 60% of all flowering plants present in the botanical gardens, which is similar to the percentages found in UK residential gardens (70%) (Loram *et al.*, 2008). In **Chapter 2**, 82.6% of bumblebee visits were to exotic species, with many plants being unique to each garden. However in the semi-natural farmland habitats, 97.9% of visits were to native plant species. This supports the findings of Salisbury *et al*., (2015) that pollinators are attracted to patches with high floral abundance, regardless of whether they are native or exotic. Thus, botanical gardens offer an interesting foraging landscape for pollinators, containing a variety of non-native species with morphologies and floral cues which pollinators may not be used to and a number of which are unusable by pollinators completely (Corbet *et al.*, 2001). However foragers are fairly quick at learning how to handle complex morphologies (Laverty, 1994), and the high visitation to exotic plant species in our study, as well as others (Majewska and Altizer, 2018), suggests pollinators will readily utilise exotics when available, highlighting that the role of exotic plant species in the diets of native pollinators is complex. Future research should look into whether the use of exotic species has impacts on colony development and success.

6.7.2 Badger predation

Chapter 4 and 5 developed and tested an artificial nest method to study the effects of badger predation to bumblebee nests. The population model *Bumble*-BEEHAVE was also used to look at the long-term impacts of badger predation on colony outputs. High rates of badger predation were found to have significant impacts on colony outputs, and the empirical studies showed that the relative predation rates differ between geographic locations. The threat posed by badgers to bumblebee nests may increase with climate change. Climate change in Finland has led to increased badger populations (Bevanger and Lindström, 1995), and a similar pattern can be observed in Wytham Woods, Oxfordshire (Macdonald and Newman, 2002) where milder winters have led to increased earthworm availability which in turn led to increased body weight. Healthier badgers may produce more offspring and thus populations will increase (Delahay *et al.*, 2006). Unfavourable weather conditions, such as summer droughts leads to reduced earthworm availability (Satchell, 1983) and thus badgers will increase their consumption of other food stuffs (Kruuk and Parish, 1981) which could lead to higher predation rates of bumblebee nests. Land use changes seen in Europe, where the area of arable land is vast (Stoate, *et al.*, 2001) and areas of natural habitats have been reduced (Fuller, 1987), meaning that the area which badgers and bumblebees inhabit are increasingly overlapping. Badgers create their setts predominantly in woodland locations (Feore and Montgomery, 1999; Harris, 1984), and although some studies have found negative effects of woodland on bumblebee abundance and species richness (Diaz-Forero *et al.*, 2012), it is probably an important nesting habitat along with grassland (Osborne *et al.*, 2008b) due to the large areas they comprise within the landscape.

6.7.3 Novel methods

Much of what we know about general bumblebee ecology comes from observation studies from the early 20th century (Freeman, 1968; Rau, 1924; Sladen, 1912), where early naturalists went out into the field and made observations of colonies of different species. Later techniques for studying aspects such as foraging behaviour included pollen analysis (Brian, 1951) and mark-recapture (Heinrich, 1976) (see Table 6.1). Early computer models were then built to model the behaviour of bumblebee on the comb (Hogeweg and Hesper, 1983). As technology has developed, the level of detail and the scale at which bumblebees could be studied increased. Use of the harmonic radar allowed bumblebee flight paths to be recorded (Osborne, *et al.*, 1997; Riley *et al.*, 1996), and the use of genetic techniques has provided the biggest increase in our knowledge of bumblebees and how they are affected by the landscape, providing a way to calculate foraging ranges more accurately and to quantify the number of colonies within a landscape (Darvill *et al.*, 2004). However, the use of genetics does not provide an exact nest location, and so **Chapter 3** tested the effectiveness of a thermal camera to locate bumblebee nests (Roberts and Osborne, 2019). Sniffer dogs have also been trained for this purpose (Waters *et al.*, 2011), but much like the thermal camera they did not outperform human searches (O'Connor *et al.*, 2012). Therefore new technologies and methods must be developed.

Despite the enhanced research techniques available, some aspects remain understudied, for example until recently information on nest predators were anecdotal (Alford, 1975; Goulson, *et al.*, 2002; Pouvreau, 1973; Sladen, 1912). Goulson *et al.*, (2018) recently provided an up-to-date study on this, using a combination of human nest searches and citizen science to collect data on hundreds of nest and finding that badgers were the biggest cause of colony predation. However studies like these are biased towards human populated areas, and finding large quantities of wild nests is difficult. Thus, **Chapter 4** developed a novel artificial nest technique to quantify the predation rates to bumblebee nests in woodland and grassland habitats. The population model *Bumble*-BEEHAVE (Becher, *et al.*, 2018) was then used to look at the combined effects of badger predation and food availability on colonies over a 20 year period. Thus the development of technology and novel methodologies are opening up new possibilities in terms of bumblebee research.

Queens are the most important caste, and future research and technological development should focus on filling some of the unanswered gaps surrounding their dispersal, and more detailed tracking during the nest searching stages to determine how they are making decisions about where to nest. Dispersal behaviour has been studied using harmonic radars (Makinson *et al.*, 2019), and genetics (Carvell *et al.*, 2017) but the timings of dispersal (pre- or post-hibernation) still remain unknown. As seen in Fig. 6.1 the timing of dispersal has potential implications for queens and nest success and so is an important area of future research.

Table 6.1 Chronology of the advancements in methodological techniques used to study various aspects of bumblebee behaviour and colony dynamics.

Year	Method	Study aspect	Limitations	Key findings	Reference
1854-1861	Visual observations and flour marking	Male patrolling behaviour	High human effort, low replicability	Males visit the same features during patrols	see Freeman, 1968
1912	Observations (workers and nests)	General bumblebee ecology	Lack of effective nest detection techniques	Provided information on the lifecycle and nesting ecology of bumblebees	Sladen, 1912
1924	Visual observations	Orientation flights	Low level of accuracy	Bees spent time orienting close to and then far from the nest before commencing foraging	Rau, 1924
1924	Homing experiment	Homing ability	Low numbers of workers successfully return	Bumblebees could return home from 1.5 miles	Rau,1924
1951	Pollen analysis	Foraging preferences	Do not know exact foraging locations	Ericaceae and white clover pollen was the most commonly collected	Brian, 1951
1976	Mark-recapture	Foraging trip movements	Low recapture rates	Bumblebee species show individual foraging preferences	Heinrich, 1976

Year	**Method**	**Study aspect**	**Limitations**	**Key findings**	**Reference**
1983	Computer models	Bumblebee behaviour on the comb	Do not exactly replicate natural scenarios	Dominance within the nest can be successfully modelled	Hogeweg and Hesper, 1983
1997	Harmonic radar	Tracking bumblebee flight paths	Signal is obscured by obstacles e.g. vegetation	Successfully tracked bees for up to 600 m	Osborne et al., 1997
1981	Video recording	Caste interactions	Limit to the number of colonies that can be observed	Workers which interact more with the queen go on to lay eggs	van Honk and Hogeweg, 1981
2001	Commercial *B. terrestris* colony	Homing ability	Biased towards a single species	*B. terrestris* workers returned to the colony from distances of 9.8 km	Goulson and Stout, 2001
2003	RFID	Foraging activity	Precise foraging locations not known	Can be used to continually monitor colonies	Streit et al., 2003
2003	Genetics	Foraging behaviour of colonies	Time-intensive	Workers from multiple colonies forage within the same patch	Chapman et al., 2003
2008	Citizen science	Nest densities	Biased towards human populated areas	Gardens and linear countryside features contain highest nest densities	Osborne et al., 2008b

Year	Method	Study aspect	Limitations	Key findings	Reference
2011	Radio-tracking	Flight distances and space use of bumblebees	Tags incur energetic costs to bees	Gardens used more often than expected	Hagen et al., 2011
2011	Sniffer dog	Locate wild bumblebee nests	Expensive and time-consuming to train	Found similar nest numbers to human searches	Waters et al., 2011
2019	Thermal camera	Locate wild bumblebee nests	Small-viewing area	Less effective than human searches	Roberts and Osborne, 2019

6.8 Future Research

6.8.1 Botanical gardens and exotic plants

Chapter 2 showed that botanical gardens are a key spring habitat for bumblebees, being used by more nest searching queens and experiencing a peak in foraging resources. Later in the season, semi-natural habitats surrounding crop fields had high floral resources, and contained similar numbers of bumblebees to botanical gardens. Although if scaled up to the whole landscape, botanical gardens would likely contain more bumblebees per unit area. Future studies should build on these findings by looking at the effects on other pollinators, especially of different sizes. Small bodied pollinators are able to forage over less distance (Greenleaf *et al.*, 2007) and so may be unable to exploit both of these habitats in a landscape; reducing their access to constant floral resources.

Further research into the source-sink dynamics of botanical gardens would also provide further insight into their role within the wider landscape. **Chapter 2** found that nest searching queen abundance was higher in botanical gardens, but that this had no relation to floral resources. Suzuki *et al*., (2009) used a mixture of energy models and physical nest searches to conclude that nests were located in areas of high floral resources, although they observe similar to **Chapter 2** that nest searching queens do not follow this rule. Advanced technologies such as radio-tracking, which has been used previously with limited success (Hagen *et al.*, 2011), would be a useful tool for increasing our knowledge of this. Radio-tracking would also increase our knowledge of queen dispersal behaviour. Queen dispersal ranges have recently been calculated as 1,227 ± 125 m (Carvell *et al.*, 2017), with slight variation between species. When queens choose to disperse, pre- or post-hibernation, is currently unknown. Knowing this, and whether they utilise botanical gardens as hibernating sites would provide insight into whether botanical gardens have higher numbers of nest searching queens due to having a higher baseline of hibernating queens, or whether queens are moving in from the surrounding landscape (Fig. 6.1).

Genetics could be used to determine the number of colonies being supported in landscapes with and without botanical gardens. If botanical gardens contain a higher number of nests than the surrounding landscape, they may be acting as a source of queens to the wider landscape. This could be especially interesting to research in simple vs complex agricultural landscapes (Heard *et al.*, 2007; Persson and Smith, 2011). Botanical gardens are potentially providing a haven of nesting and foraging resources similar to residential gardens (Lye *et al.*, 2012; Salisbury *et al.*, 2015), and thus producing a high number of queens which would disperse into the surrounding landscape. However, if this landscape is poor then this may have negative consequences on queen survival and future nesting success (Carvell *et al.*, 2017; Fig. 6.1).

Despite the presence of native flowering plants in the botanical gardens, **Chapter 2** found that that 82.6% of bumblebee visits in these habitats were to exotic plant species, highlighting that exotic species are utilised by most of our common bumblebee species. Future research could build on this through looking at the usage of exotic pollen at the whole colony level to determine what proportion of pollen resources come from exotic species when nesting within or surrounding botanical gardens. The impacts of high exotic pollen on colony success could also be looked at, to determine whether the presence of botanical gardens is affecting colony success.

6.8.2 Mechanisms underlying badger predation of bumblebee nests

Significantly higher predation rates were recorded in Gloucestershire (**Chapter 4**) compared to Cornwall (**Chapter 5**). The model simulations also found that only high levels of predation had significant impacts on the numbers of hibernating queens, workers and colonies produced. Differences in predation are possibly due to higher badger densities, but the underlying mechanisms driving badger predation of bumblebee nests is still unknown. Future studies could build on the work from the artificial nest method to include genetic estimates of bumblebee populations in the area, combined with better estimates of badger densities to look at whether the numbers of wild bumblebee nests are lower than expected in areas of high badger densities. This would have implications for the source-sink dynamics of populations, with areas of high badger predation being potential population sinks. Through understanding the underpinning mechanisms behind badger predation better, more accurate modelled scenarios could be implemented to look at the effects badger predation could be having across the whole of the UK. Badgers are known to show individual foraging specialisation (Robertson *et al.*, 2014, 2015), and so it may be that only certain badgers within each sett are consuming bumblebee nests. To determine whether this is the case, better video surveillance of nests could be used to recognise if it is the same badgers digging up nests, with individual badgers being recognisable by their tails (Dixon, 2003). This would have implications for model simulations and predation estimates as it could mean that the number of badgers within a sett is not important if not all of the badgers are consuming bumblebee nests.

6.9 Conclusion

This thesis has utilised a combination of classical field techniques, novel techniques and computer models to explore some of the understudied aspects of bumblebee ecology. Thermal cameras were tested as a novel nest detection tool (**Chapter 3**), but were found to be unsuccessful, however technological advancements may increase their potential use in the future. The artificial nest method developed in **Chapter 4** was found to be a successful way of monitoring the relative differences in badger predation rates of bumblebee nests between habitats and geographic locations. Finally, the population model *Bumble*-BEEHAVE (Becher, *et al.*, 2018) was used to look at the combined effects of badger predation under high and low food availability (**Chapter 5**). It showed that high rates of badger predation led to significantly fewer hibernating queens, workers and colonies being produced in each landscape. Food availability did not buffer the negative impacts of predation, but low food availability alone led to a greater reduction in hibernating queens, workers and colonies produced compared to predation. Low food availability was created by reducing the floral diversity of grassland habitats within the models, supporting the recommendation by Baude *et al*., (2016) that increasing the floral richness of semi-improved grassland would have positive effects to pollinator populations.

Chapter 2 showed that botanical gardens are an important spring habitat for bumblebees, with exotic plant species providing a key foraging resource to queen bumblebees during this time. Contrary to initial predictions, semi-natural habitats surrounding agricultural fields provided higher floral resources compared to botanical gardens during early and late summer.

Residential gardens have been found to increase colony numbers and survival (Goulson *et al.*, 2010), and so an important next step in understanding the role of botanical gardens is to explore whether they are providing these same benefits. Due to the complex agricultural landscapes in our study area, it is likely that the beneficial effects of botanical gardens were masked. Given the importance of botanical gardens for plant conservation worldwide (Blackmore *et al.*, 2011; Chen and Sun, 2018; Hardwick *et al.*, 2011), they could play a role in the conservation of pollinators through buffering populations during times of resource fluctuations in other habitats, especially in rural areas. Overall this thesis has shown that some of the hidden aspects of bumblebee ecology remain hidden, but that through the use of classical techniques, novel methods and computer models important steps can be made towards uncovering the previously understudied aspects.

Appendices

Appendix A – Chapter 2

Table A.1 Coefficients for the best fitting model for the floral abundance of bumblebee food plants during the nest searching surveys including habitat and week as fixed effects.

	Estimate ± SE	z value	p value
Intercept (Habitat (Farmland) and Week 1)	8.55 ± 5.08	1.68	0.093
Habitat (Garden)	15.43 ± 6.71	2.30	0.021
Week 2	5.49 ± 5.10	1.08	0.282
Week 3	13.00 ± 5.10	2.55	0.011
Week 4	17.88 ± 5.10	3.50	<0.001
Week 5	23.58 ± 5.11	4.62	<0.001
Week 6	30.47 ± 5.10	5.97	<0.001
Habitat (Garden): Week 2	-0.87 ± 7.20	-0.12	0.904
Habitat (Garden): Week 3	-0.17 ± 7.20	-0.02	0.981
Habitat (Garden): Week 4	-5.32 ± 7.21	-0.74	0.461
Habitat (Garden): Week 5	-14.13 ± 7.21	-1.96	0.050
Habitat (Garden): Week 6	-29.12 ± 7.21	-4.04	<0.001

Global model code:

```
glmer(abundance of bumblebee food plants~habitat*week + offset(log(transect length))) +(1|site/transect ID) + (1|OLRE), family=Poisson(link=”sqrt”)
```

Table A.2 Coefficients for the three best fitting model for the floral diversity of bumblebee food plants during the nest searching surveys including habitat and week as fixed effects.

Model A.2.1	Estimate ± SE	z value	p value
Intercept	-2.99 ± 0.09	-31.76	<0.001
Model A.2.2	Estimate ± SE	z value	p value
Intercept (Habitat (Farmland))	-3.06 ± 0.12	-26.05	<0.001
Habitat (Garden)	0.14 ± 0.14	1.01	0.314
Model A.2.3	Estimate ± SE	z value	p value
Intercept (Week 1)	-3.28 ± 0.15	-21.17	<0.001
Week 2	0.15 ± 0.18	0.83	0.409
Week 3	0.29 ± 0.18	1.60	0.110
Week 4	0.30 ± 0.18	1.61	0.107
Week 5	0.40 ± 0.18	2.20	0.028
Week 6	0.51 ± 0.19	2.74	<0.01

Global model code:

glmer(diversity of bumblebee food plants~habitat*week + offset(log(transect length))) +(1|site/transect ID), family=Poisson(link=”sqrt”)

Table A.3 Coefficients for the two best fitting models for nest searching queen abundance, with mean floral abundance of bumblebee food plants as the fixed effect along with habitat.

Model A.3.1	Estimate ± SE	t value	p value
Intercept (Habitat (Farmland))	-0.84 ± 1.49	0.71	<0.001
Habitat (Garden)	3.61 ± 1.68	2.15	0.050
Model A.3.2			
Intercept (Habitat (Farmland))	0.41 ± 1.16	0.35	0.732
Habitat (Garden)	4.48 ± 1.69	2.65	0.020
Mean floral abundance (bumblebee food plants)	-1.38 ± 0.87	-1.58	0.139
Global model code:			
glm(nest searching queen abundance~habitat*scale(mean floral abundance) + offset(log(transect length)), family=Gaussian)			

Table A.4 Coefficients for the two best fitting models for nest searching queen diversity, with mean floral diversity of bumblebee food plants as the fixed effect along with habitat.

Model A.4.1	Estimate ± SE	t value	p value
Intercept	-1.23 ± 0.33	-3.73	<0.01
Mean floral diversity (bumblebee food plants)	0.60 ± 0.34	1.76	0.100
Model A.4.2	Estimate ± SE	t value	p value
Intercept	-1.24 ± 0.35	-3.50	<0.01
Global model code:			
glm(nest searching queen diversity~habitat*scale(Mean floral diversity) + offset(log(transect length)), family=Gaussian)			

Table A.5 Coefficients for the best fitting model for the floral abundance of bumblebee food plants during the phenological surveys including habitat and survey period as fixed effects.

	Estimate ± SE	z value	p value
Intercept (Habitat (Farmland) and Survey period (Early spring))	12.75 ± 7.14	1.79	0.074
Habitat (Garden)	33.41 ± 10.10	3.31	<0.001
Survey period (Early summer)	69.50 ± 90.4	7.68	<0.001
Survey period (Late summer)	36.22 ± 9.04	4.01	<0.001
Habitat (Garden): Survey period (Early summer)	-66.41 ± 12.80	-5.19	<0.001
Habitat (Garden): Survey period (Late summer)	-38.22 ± 12.79	-2.99	<0.01
Global model code: glmer(abundance of bumblebee food plants~survey period*habitat + (1|transect ID) + (1|OLRE), family=Poisson(link=”sqrt”))			

Table A.6 Coefficients for the best fitting model for the floral diversity of bumblebee food plants during the phenological surveys including habitat and survey period as fixed effects.

	Estimate ± SE	z value	p value
Intercept (Habitat (Farmland) and Survey period (Early spring))	1.19 ± 0.14	8.29	<0.001
Habitat (Garden)	0.24 ± 0.10	2.32	0.02
Survey period (Early summer)	0.97 ± 0.15	6.43	<0.001
Survey period (Late summer)	1.05 ± 0.15	6.98	<0.001
Global model code: glmer(diversity of bumblebee food plants~survey period*habitat + (1|site), family=Poisson(link="log"))			

Table A.7 Coefficients for the three best fitting model for bumblebee abundance during the phenological surveys, with floral abundance of bumblebee food plants, habitat and survey period as fixed effects.

Model A.7.1	Estimate ± SE	z value	p value
Intercept	-2.07 ± 0.23	-9.20	<0.001
Diversity of bumblebee food plants	1.16 ± 0.15	7.20	<0.001
Model A.7.2	Estimate ± SE	z value	p value
Intercept (Habitat (Farmland))	-1.69 ± 0.29	-5.84	<0.001
Habitat (Garden)	-0.76 ± 0.41	-1.86	0.064
Diversity of bumblebee food	1.20 ± 0.15	8.10	<0.001
Model A.7.3	Estimate ± SE	z value	p value
Interecept (Habitat (Farmland))	-1.67 ± 0.29	-5.68	<0.001
Habitat (Garden)	-0.76 ± 0.41	-1.84	0.067
Diversity of bumblebee food	1.34 ± 0.23	5.86	<0.001
Habitat (Garden): Diversity of bumblebee food	-0.24 ± 0.30	-0.81	0.420
Global model code: glmer(abundance of bumblebees~survey period*habitat*scale(diversity of bumblebee food plants) + offset(log(survey effort)) + (1|site/transect ID) + (1|OLRE), family=Poisson(link=”sqrt”))			

Table A.8 Coefficients for the four best fitting models for bumblebee diversity during the phenological surveys, with floral diversity of bumblebee food plants as a fixed effect, along with habitat and survey period.

Model A.8.1	Estimate ± SE	z value	p value
Intercept (Habitat (Farmland) and Survey period (Early spring))	0.58 ± 0.38	1.54	0.124
Habitat (Garden)	0.57 ± 0.41	1.39	0.165
Diversity of bumblebee food plants	0.35 ± 0.14	2.57	0.010
Survey period (Early summer)	0.16 ± 0.41	2.83	<0.01
Survey period (Late summer)	0.63 ± 0.46	1.39	0.165
Habitat (Garden): Survey period (Early summer)	-1.14 ± 0.47	-2.45	0.015
Habitat (Garden): Survey period (Late summer)	-0.91 ± 0.48	-1.89	0.058

Model A.8.2	Estimate ± SE	z value	p value
Intercept (Habitat (Farmland) and Survey period (Early spring))	1.19 ± 0.29	4.15	<0.001
Habitat (Garden)	-0.24 ± 0.18	-1.32	0.188
Diversity of bumblebee food plants	0.65 ± 0.23	2.81	<0.01
Survey period (Early summer)	0.42 ± 0.32	1.33	0.185
Survey period (Late summer)	-0.09 ± 0.37	-0.25	0.801
Habitat (Garden): Diversity of bumblebee food plants	-0.39 ± 0.22	-1.80	0.072

Model A.8.3	Estimate ± SE	z value	p value
Interecept (Habitat (Farmland) and Survey period (Early spring))	1.03 ± 0.27	3.81	<0.001
Habitat (Garden)	-0.28 ± 0.18	-1.58	0.114
Diversity of bumblebee food plants	0.31 ± 0.13	2.39	0.016
Survey period (Early summer)	0.60 ± 0.30	1.99	0.047
Survey period (Late summer)	0.19 ± 0.33	0.56	0.573
Model A.8.4	Estimate ± SE	z value	p value
Intercept (Survey period (Early spring))	0.79 ± 0.22	3.55	<0.001
Diversity of bumblebee food plants	0.21 ± 0.11	1.92	0.055
Survey period (Early summer)	0.75 ± 0.28	2.66	<0.01
Survey period (Late summer)	0.37 ± 0.30	1.21	0.225

Global model code:

glmer(diversity of bumblebees~survey period*habitat + survey period*scale(diversity of bumblebee food plants) + habitat*scale(diversity of bumblebee food plants) + offset(log(survey effort)) + (1|transect ID), family=Poisson(link=”sqrt”))

Table A.9 Coefficients for the best fitting model for bumblebee diversity during the phenological survey, with floral abundance of bumblebee food plants as a fixed effect, along with habitat and survey period.

	Estimate ± SE	z value	p value
Intercept (Survey period (Early spring))	1.38 ± 0.13	10.41	<0.001
Diversity of bumblebee food plants	0.18 ± 0.08	2.15	0.032
Survey period (Early summer)	0.79 ± 0.19	4.07	<0.001
Survey period (Late summer)	0.55 ± 0.18	3.10	<0.01

Global model code:

glmer(diversity of bumblebees~survey period*habitat*scale(abundance of bumblebee food plants) + offset(log(survey effort)) + (1|site), family=Poisson(link=”sqrt”))

Table A.10 Coefficients for the best fitting model for bumblebee abundance during the phenological survey with survey period as a fixed effect.

	Estimate ± SE	z value	p value
Intercept (Survey period (Early spring))	-3.45 ± 0.35	-9.85	<0.001
Survey period (Early summer)	1.97 ± 0.36	5.47	<0.001
Survey period (Late summer)	2.15 ± 0.36	5.92	<0.001

Global model code:

glmer(abundance of bumblebees~survey period*habitat*scale(abundance of bumblebee food plants) + offset(log(survey effort)) + (1|site/transect ID) + (1|OLRE), family=Poisson(link="sqrt"))

Appendix B – Chapter 5

Table B.1 Badger densities for badgers in Great Britain, which were used to calculate a County correction factor for Cornwall and Woodchester Park, by dividing their respective badger densities by the averaged badger density.

Location	Badgers per km^2 (mean)	Reference
Avon, England	4.9	(Cheeseman *et al.*, 1981)
Arndish, Scotland	1.5-2.7 (2.1)	(Kruuk, Hans and Parish, 1982)
Bristol, England	4.4-7.5 (6.0)	(Harris and Cresswell, 1987)
Cornwall, England	4.7	(Cheeseman *et al.*, 1981)
Gloucestershire, England	19.4-19.7 (19.6)	(Cheeseman *et al.*, 1981)
New Deer, Scotland	5.7-6.2 (6.0)	(Kruuk, Hans and Parish, 1982)
Speyside, Scotland	1.1, 1.9, 3.2 (2.1)	(Kruuk, Hans and Parish, 1982)
Staffordshire, England	6.2	(Cheeseman *et al.*, 1985)
Woodchester Park, Gloucestershire, England	7.8-25.3 (16.6)	(Rogers *et al.*, 1997)
Wytham Woods, Oxfordshire, England	8.4, 16.7, 36.4 (20.5)	(Kruuk, Hans and Parish, 1982; Macdonald, D. W. *et al.*, 2009; Woodroffe, 1992)
Average badger density	8.8	

Table B.2 The flower species present in each of the habitat types in the Bumble-BEEHAVE simulations. The flower densities are shown for each of the four habitats: hedgerows (H), woodland (W), scrub (S), high quality grassland (Gh) and low quality grassland (Gl)

	H	W	S	Gh	Gl								
Flower species	Flower density (flower s/m2)					Pollen (g/ flower)	Nectar (ml/ flower)	Prop. of protein in pollen	Sugar conc. on of nectar (mol/l)	Flowering start day	Flowering stop day	Corolla depth (mm)	Nectar flower volume (myl/ flower)
Bugle	0.12	0.30	0.51	0.03	0	0.0007	0.0008	0.0721	0.8247	120	211	10	0.8097
Burdock	0.01	0.10	0.05	0.02	0	0.0004	0.0023	0.1118	0.8865	181	272	3.9	2.2890
Oilseed rape	0.05	0	0.02	0	0	0.0015	0.0210	0.2561	1.4130	120	242	5	21.0324
Giant bindweed	0.05	0	0.03	0	0	0.0009	0.0100	0.2646	0.6646	181	272	0	9.9540
Common knapweed	0.22	0.01	0.56	0.01	0	0.0024	0.0021	0.1590	1.3408	151	272	3	2.1042
Greater knapweed	0	0	0.03	0	0	0.0020	0	0.2978	0	181	272	13.6	0
Rosebay willowherb	0.13	0.02	0.04	0	0	0.0115	0	0.2057	0	181	272	0	0

	H	W	S	Gh	Gl								
Flower species	Flower density (flower s/m2)					Pollen (g/flower)	Nectar (ml/flower)	Prop. of protein in pollen	Sugar conc. on of nectar (mol/l)	Flowering start day	Flowering stop day	Corolla depth (mm)	Nectar flower volume (myl/flower)
Marsh thistle	0	0.02	0.02	0	0	0.0051	0.0006	0.1454	0.8924	181	272	3	0.6393
Spear thistle	0.01	0.01	0.01	0.01	0	0.0031	0.0018	0.1903	1.2903	151	303	6.2	1.8247
Hawthorn	0.44	2.73	0.60	0.62	0	0.0001	0.0019	0.1540	1.0233	120	180	0	1.8753
Foxglove	0.07	0.02	0.09	0	0	0.0216	0.0016	0.2275	0.8243	151	272	7	1.6333
Wild teasel	0.02	0	0.04	0	0	0.0146	0.0098	0.1985	1.0858	181	242	10	9.7611
Vipers bugloss	0.00	0	0.50	0	0	0.0017	0.0009	0.1803	0.6681	151	272	6.7	0.9217
Ground ivy	0.06	0.10	0.10	0.06	0	0.0009	0.0026	0.1907	0.8724	59	150	7	2.6180
Bluebell	0.06	0.60	0.26	0.49	0	0.0019	0	0.3630	0	90	180	0	0
St Johns wort	0	0.01	0.01	0	0	0.0005	0	0.1391	0	151	272	0	0
Field scabious	0.01	0	0.02	0.02	0	0.0089	0	0.1195	0	181	272	0	0
White dead nettle	0.14	0.01	0.17	0.03	0	0.0012	0.0022	0.2280	0.7562	120	364	7.7	2.1677

	H	W	S	Gh	Gl								
Flower species	Flower density (flower s/m2)					Pollen (g/ flower)	Nectar (ml/ flower)	Prop. of protein in pollen	Sugar conc. on of nectar (mol/l)	Flowering start day	Flowering stop day	Corolla depth (mm)	Nectar flower volume (myl/ flower)
Red dead nettle	0.10	0.03	0.03	0.03	0	0.0007	0.0055	0.2280	1.0128	59	303	7	5.4529
Birdsfoot trefoil	0.04	0.04	0.22	0.35	0	0.0010	0.0008	0.3580	0.6976	120	303	9	0.8430
Selfheal	0.04	0.08	0.06	0.11	0	0.0003	0.0006	0.2580	0.6624	151	272	8	0.5824
Blackthorn	0.93	0.33	1.29	0.23	0	0	0.0001	0.2720	0.7797	59	150	0	0.0933
Buttercup	0.38	0.72	0.79	2.16	0	0.0008	0.0002	0.1206	0.7157	120	303	0	0.1972
Dog rose	0.18	0.04	0.06	0	0	0.0007	0	0.0907	0	151	211	0	0
Bramble	0.16	0.42	0.21	0.02	0	0.0005	0.0068	0.1260	0.5004	120	272	0	6.8238
Average Willow	1.28	0	1.21	0	0	0.0103	0.0026	0.2574	1.1345	59	119	0	2.6067
Ragwort	0.02	0	0.06	0.02	0	0.0002	0	0.1550	0	151	303	0	0
Hedge woundwort	0.04	0.04	0.29	0	0	0.0008	0.0015	0.1454	1.0442	181	242	9	1.4646
Comfry	0.04	0	0	0	0	0.0010	0.0043	0.0971	0.9898	120	211	17	4.3178
Dandelion	0.11	0.01	0.11	0.12	0.12	0.0004	0.0005	0.0917	1.2947	1	364	1.2	0.4702

	H	W	S	Gh	Gl								
Flower species	Flower density (flower s/m2)					Pollen (g/ flower)	Nectar (ml/ flower)	Prop. of protein in pollen	Sugar conc. on of nectar (mol/l)	Flowering start day	Flowering stop day	Corolla depth (mm)	Nectar flower volume (myl/ flower)
Red clover	0.05	0.01	0.05	0.25	0.00	0.0005	0.0006	0.2087	0.9860	120	272	10	0.6160
White clover	0.40	0.08	0.76	1.67	1.67	0.0004	0.0007	0.2307	0.9803	151	272	2	0.6666
Tufted vetch	0.05	0	0.03	0.12	0	0.0009	0.0016	0.1296	0	151	242	6.8	1.5871
Common vetch	0.13	0.04	0.01	0.03	0	0.0004	0.0009	0.4280	0.8153	120	272	7	0.8604

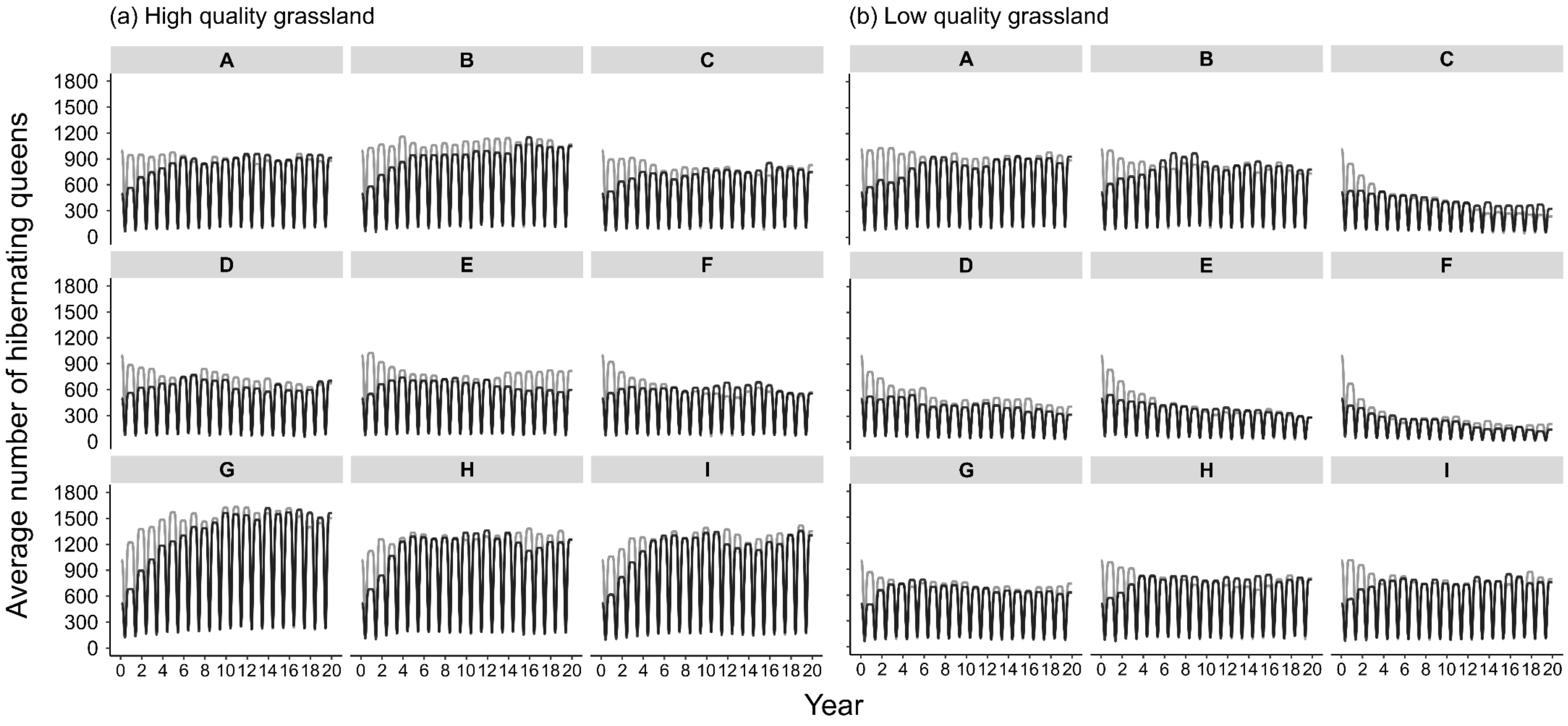

Figure B.1 Simulation outputs from Bumble-BEEHAVE under no badger predation seeded with 500 (black lines) and 1000 (grey lines) B. terrestris queens, for (a) high quality grassland landscapes and (b) low quality grassland landscapes for sites A-I.

Table B.3 Coefficients for the two best fitting models for the empirical badger seasonal data, with nest treatment depth included as a fixed effect.

Model B.3.1	Estimate ± SE	z value	p value
Intercept (Treatment (Control))	-3.64 ± 0.44	-8.23	<0.001
Treatment (Surface)	0.71 ± 0.32	2.23	0.026
Treatment (Underground)	0.27 ± 0.34	0.79	0.429
Model B.3.2	Estimate ± SE	z value	p value
Intercept	-3.31 ± 0.39	-8.39	<0.001
Global model code: glmer(cbind(success, fail)~month*habitat*depth treatment + (1|site/transectID) + (1|OLRE), family=Binomial(link=”cloglog”), control=glmerControl(optimizer=”bobyqa”, optCtrl=list(maxfun=2e4)))			

Table B.4 Coefficients for the best fitting model comparing the predation rates for the empirical data collected in Cornwall (current study) and Gloucestershire (**Chapter 4**).

	Estimate ± SE	z value	p value
Intercept (Location (Cornwall))	-2.99 ± 0.45	-6.67	<0.001
Location (Gloucestershire)	1.56 ± 0.59	2.65	<0.01

Global model:

glmer(cbind(success, fail)~location + (1|site/transect ID), family=Binomial)

Table B.5 Coefficients for the linear models looking at the influence of high and low food availability on the mean quantities of nectar (l) and pollen (kg) available per month.

Model B.5.1 (Mean nectar (l))	Estimate ± SE	t-value	p-value
Intercept (Food availability (High))	849.78 ± 52.11	16.31	<0.001
Food availability (Low)	-165.81 ± 11.29	-14.69	<0.001
Model B.5.2 (Mean pollen (kg))	Estimate ± SE	t-value	p-value
Intercept (Food availability (High))	820.5 ± 38.21	21.47	<0.001
Food availability (Low)	-299.02 ± 21.07	-14.20	<0.001
Global model: lmer(response~food quantity + (1|site))			

Table B.6 Coefficients for the best fitting models for the *Bumble*-BEEHAVE simulation outputs: hibernating queens, workers and colonies with predation rate (none, low and high) and food availability (low and high) included as fixed effects.

Model B.6.1 (Hibernating queens)	Estimate ± SE	t-value	p-value
Intercept (Predation (None) and Food availability (High))	979.68 ± 92.78	10.56	<0.001
Predation (Low)	-46.57 ± 47.07	-0.99	0.328
Predation (High)	-170.33 ± 47.07	-3.62	<0.001
Food availability (Low)	-385.89 ± 38.44	-10.04	<0.001

Model B.6.2 (Workers)	Estimate ± SE	t-value	p-value
Intercept (Predation (None) and Food availability (High))	1120.40 ± 106.99	10.47	<0.001
Predation (Low)	-49.27 ± 77.74	-0.63	0.530
Predation (High)	-193.93 ± 77.74	-2.50	0.017
Food availability (Low)	-432.87 ± 77.74	-5.57	<0.001
Predation (Low):Food availability (Low)	-31.47 ± 109.93	-0.29	0.776
Predation (High):Food availability (Low)	10.07 ± 109.93	0.09	0.928

Model B.6.3 (Colonies)	Estimate ± SE	t-value	p-value
Intercept (Predation (None) and Food availability (High))	87.89 ± 8.78	10.02	<0.001
Predation (Low)	-3.08 ± 6.26	-0.49	0.626
Predation (High)	-15.63 ± 6.26	-2.50	0.017
Food availability (Low)	-33.83 ± 6.26	-5.40	<0.001
Predation (Low):Food availability (Low)	-1.70 ± 8.85	-0.19	0.849
Predation (High):Food availability (Low)	1.38 ± 8.85	0.16	0.877
Global model: lmer(response ~badger predation*food availability + (1|site))			

Bibliography

Agarwala, B. K., Yasuda, H., and Sato, S. (2008) Life history response of a predatory ladybird, *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae), to food stress. *Applied Entomology and Zoology* 43(2): 183–189.

Al-doski, J., Mansor, S. B., and Shafri, H. Z. B. M. (2016) Thermal imaging for pests detecting - a review. *International Journal of Agriculture, Forestry and Plantation* 2: 10–30.

Alford, D. (1975) *Bumblebees*. London: Davis-Poynter.

Amin, M. R., Kwon, Y. J., and Thet, Z. M. (2011) Effect of worker number and diapause duration on colony parameters of the bumblebee, *Bombus terrestris* (Hymenoptera: Apidae). *Journal of Asia-Pacific Entomology* 14(4): 455–458.

Ansell, D., Freudenberger, D., Munro, N., and Gibbons, P. (2016) The cost-effectiveness of agri-environment schemes for biodiversity conservation: A quantitative review. *Agriculture, Ecosystems & Environment* 225: 184–191.

Baldock, K. C. R., Goddard, M. A., Hicks, D. M., Kunin, W. E., Mitschunas, N., Morse, H., Osgathorpe, L. M., Potts, S. G., Robertson, K. M., Scott, A. V, Staniczenko, P. P. A., Stone, G. N., Vaughan, I. P., and Memmott, J. (2019) A systems approach reveals urban pollinator hotspots and conservation opportunities. *Nature Ecology and Evolution* 3(3): 363–373.

Balestrieri, A., Remonti, L., and Prigioni, C. (2004) Diet of the Eurasian Badger (*Meles meles*) in an Agricultural Riverine Habitat (NW Italy). *Hystrix Italian Journal of Mammalogy* 15(2): 3–12.

Balestrieri, A., Remonti, L., and Prigioni, C. (2009) Habitat selection in a low-density badger *Meles meles* population: A Caomparison of radio-tracking and tatrine surveys. *Wildlife Biology* 15(4): 442–448.

Banks, H. T., Banks, J. E., Bommarco, R., Rundlöf, M., and Tillman, K. (2017) Modeling bumble bee population dynamics with delay differential equations. *Ecological Modelling* 351: 14–23.

Baron, G. L., Raine, N. E., and Brown, M. J. F. (2014) Impact of chronic exposure to a pyrethroid pesticide on bumblebees and interactions with a trypanosome parasite. *Journal of Applied Ecology* 51(2): 460–469.

Bartoń, K. (2017) MuMIn: Multi-model inference. R package version 1.40.0. https://CRAN.R-project.org/package=MuMIn’, (1). Available at: https://cran.r-project.org/web/packages/MuMIn/MuMIn.pdf.

Baude, M., Kunin, W. E., Boatman, N. D., Conyers, S., Davies, N., Gillespie, M. A. K., Morton, R. D., Smart, S. M., and Memmott, J. (2016) Historical nectar assessment reveals the fall and rise of floral resources in Britain. *Nature* 530(7588): 85–88.

Becher, M. A, Grimm, V., Thorbek, P., Horn, J., Kennedy, P. J., and Osborne, J. L. (2014) BEEHAVE: a systems model of honeybee colony dynamics and foraging to explore multifactorial causes of colony failure. doi:10.1111/1365-2664.12222.

Becher, M.A., Grimm, V., Knapp, J., Horn, J., Twiston-Davies, G., and Osborne, J. L. (2016) BEESCOUT: A model of bee scouting behaviour and a software tool for characterizing nectar/pollen landscapes for BEEHAVE. *Ecological Modelling* 340: 126–133.

Becher, M. A., Twiston-Davies, G., Penny, T. D., Goulson, D., Rotheray, E. L., and Osborne, J. L. (2018) *Bumble*-BEEHAVE: A systems model for exploring multifactorial causes of bumblebee decline at individual, colony, population and community level. (Beggs, J., Ed.)*Journal of Applied Ecology* 55: 2790–2801.

Beekman, M., and Van Stratum, P. (2000) Does the diapause experience of bumblebee queens *Bombus terrestris* affect colony characteristics? *Ecological Entomology*. doi:10.1046/j.1365-2311.2000.00235.x.

Benton, T. (2006) *Bumblebees*. *Harper Collins*. London, UK.

Bevanger, K., and Lindström, E. R. (1995) Distributional history of the European badger *Meles meles* in Scandinavia during the 20th century. *Annales Zoologici Fennici* 32(1): 5–9.

Biesmeijer, J. C., Roberts, S. P. M., Reemer, M., Ohlemüller, R., Edwards, M., Peeters, T., Schaffers, A. P., Potts, S. G., Kleukers, R., Thomas, C. D., Settele, J., and Kunin, W. E. (2006) Parallel declines in pollinators and insect-pollinated plants in Britain and the Netherlands. *Science* 313: 351–354.

Blaauw, B. R., and Isaacs, R. (2014) Larger patches of diverse floral resources increase insect pollinator density, diversity, and their pollination of native wildflowers. *Basic and Applied Ecology* 15: 701–711.

Blackmore, S., Gibby, M., and Rae, D. (2011) Strengthening the scientific contribution of botanic gardens to the second phase of the *Global Strategy for Plant Conservation. Botanical Journal of the Linnean Society* 166: 267-281

Brian, A. D. (1951) The pollen collected by bumble-bees. *The Journal of Animal Ecology* 20(2): 191–194.

Brown, B., Mitchell, Randall, J., and Graham, Shirley, A. (2002) Competition for pollination between an invasive species (purple loosestrife) and a native congener. *Ecology* 83(8): 2328–2336.

Brown, M. J. F., Loosli, R., and Schmid-Hempel, P. (2000) Condition-dependent expression of virulence in a trypanosome infecting bumblebees. *Oikos* 91(3): 421–427.

Browne, W. J., Subramanian, S. V., Jones, K., and Goldstein, H. (2005) Variance partitioning in multilevel logistic models that exhibit overdispersion. *Journal of the Royal Statistical Society: Series A (Statistics in Society)* 168(3): 599–613.

Bryden, J., Gill, R. J., Mitton, R. A. A., Raine, N. E., and Jansen, V. A. A. (2013a) Chronic sublethal stress causes bee colony failure. *Ecology Letters* 16(12): 1463–1469.

Cameron, R. W. F., Blanuša, T., Taylor, J. E., Salisbury, A., Halstead, A. J., Henricot, B., and Thompson, K. (2012) The domestic garden – Its contribution to urban green infrastructure. *Urban Forestry & Urban Greening* 11(2): 129–137.

Carreck, N., and Williams, I. (1998) The economic value of bees in the UK. *Bee World* 79(3): 115–123.

Carvell, C. (2002) Habitat use and conservation of bumblebees (*Bombus* spp) under different grassland management regimes. *Biological Conservation* 103(1): 33–49.

Carvell, C, Meek, W. ., Pywell, R. ., and Nowakowski, M. (2004) The response of foraging bumblebees to successional change in newly created arable field margins. *Biological Conservation* 118(3): 327–339.

Carvell, C., Roy, D. B., Smart, S. M., Pywell, R. F., Preston, C. D., and Goulson, D. (2006) Declines in forage availability for bumblebees at a national scale. *Biological Conservation* 132(4): 481–489.

Carvell, A. C., Osborne, J. L., Bourke, A. F. G., Freeman, S. N., Pywell, R. F., Applications, E., and Carvell, C. (2011) Bumble bee species' responses to a targeted conservation measure depend on landscape context and habitat quality. *Ecological Applications* 21(5): 1760–1771.

Carvell, C., Jordan, W. C., Bourke, A. F. G., Pickles, R., Redhead, J. W., and Heard, M. S. (2012) Molecular and spatial analyses reveal links between colony-specific foraging distance and landscape-level resource availability in two bumblebee species. *Oikos* 121: 734–742.

Carvell, C., Isaac, N. J. B., Jitlal, M., Peyton, J., Powney, G. D., Roy, D. B., et al. (2016) *Design and testing of a National Pollinator and Pollination Monitoring Framework. Final summary report to the Department for Environment, Food and Rural Affairs (Defra), Scottish Government and Welsh Government: Project WC1101*.

Carvell, C., Bourke, A. F. G., Dreier, S., Freeman, S. N., Hulmes, S., Jordan, W. C., Redhead, J. W., Sumner, S., Wang, J., and Heard, M. S. (2017) Bumblebee family lineage survival is enhanced in high-quality landscapes. *Nature* 543: 547–549.

Chapman, R. E., Wang, J., and Bourke, A. F. G. (2003) Genetic analysis of spatial foraging patterns and resource sharing in bumble bee pollinators. *Molecular Ecology* 12: 2801–2808.

Charman, T. G., Sears, J., Green, R. E., and Bourke, A. F. G. (2010) Conservation genetics, foraging distance and nest density of the scarce Great Yellow Bumblebee (*Bombus distinguendus*). *Molecular Ecology* 19(13): 2661–2674.

Chateil, C., and Porcher, E. (2015) Landscape features are a better correlate of wild plant pollination than agricultural practices in an intensive cropping system. *Agriculture, Ecosystems and Environment* 201: 51–57.

Cheeseman, C. L., Jones, G. W., Gallagher, J., and Mallinson, P. J. (1981) The population structure, density and prevalence of Tuberculosis (*Mycobacterium bovis*) in badgers (*Meles meles*) from four areas in South-West England. *The Journal of Applied Ecology* 18(3): 795–804.

Cheeseman, C. L., Little, T. W. A., Mallinson, P. J., Page, R. J. C., Wilesmith, J. W., and Pritchard, D. G. (1985) Population ecology and prevalence of tuberculosis in badgers in an area of Staffordshire. *Mammal Review* 15(3): 125–135.

Chen, G., and Sun, W. (2018) The role of botanical gardens in scientific research, conservation, and citizen science. *Plant Diversity* 40(4): 181–188.

Cilulko, J., Janiszewski, P., Bogdaszewski, M., and Szczygielska, E. (2013) Infrared thermal imaging in studies of wild animals. *European Journal of Wildlife Research* 59: 17–23.

Cleary, G. P., Corner, L. A. L., O’Keeffe, J., and Marples, N. M. (2009) The diet of the badger *Meles meles* in the Republic of Ireland. *Mammalian Biology* 74(6): 438–447.

Cohn, J. P. (2008) Citizen science: Can volunteers do real research? *BioScience* 58(3): 192.

Cole, L. J., Brocklehurst, S., Robertson, D., Harrison, W., and Mccracken, D. I. (2017) Exploring the interactions between resource availability and the utilisation of semi-natural habitats by insect pollinators in an intensive agricultural landscape. *Agriculture, Ecosystems & Environment* 246: 157–167.

Connelly, H., Poveda, K., and Loeb, G. (2015) Landscape simplification decreases wild bee pollination services to strawberry. *Agriculture, Ecosystems and Environment* 211: 51–56.

Conroy, T. J., Palmer-Young, E. C., Irwin, R. E., and Adler, L. S. (2016) Food limitation affects parasite load and survival of *Bombus impatiens*

(Hymenoptera: Apidae) infected with Crithidia (Trypanosomatida: Trypanosomatidae). *Environmental Entomology* 45(5): 1212–1219.

Corbet, S. A., Bee, J., Dasmahapatra, K., Gale, S., Gorringe, E., Ferla, B. La, Moorhouse, T., Trevail, A., Van Bergen, Y., and Vorontsova, M. (2001) Native or exotic? Double or single? Evaluating plants for pollinator-friendly gardens. *Annals of Botany* 87(2): 219–232.

Cranmer, L., McCollin, D., and Ollerton, J. (2012) Landscape structure influences pollinator movements and directly affects plant reproductive success. *Oikos* 121(4): 562–568.

Cresswell, J. E., Osborne, J. L., and Goulson, D. (2000) An economic model of the limits to foraging range in the central-place foraging with numerical solutions for bumblebees. *Ecological Entomology* 25: 249–255.

Cresswell, J. E. (2017) A demographic approach to evaluating the impact of stressors on bumble bee colonies. *Ecological Entomology* 42(2): 221–229.

Crone, E. E., and Williams, N. M. (2016) Bumble bee colony dynamics: Quantifying the importance of land use and floral resources for colony growth and queen production. *Ecology Letters* 19(4): 460–468.

Cussans, J., Goulson, D., Sanderson, R., Goffe, L., Darvill, B., and Osborne, J. L. (2010) Two bee-pollinated plant species show higher seed production when grown in gardens compared to arable farmland. *PLoS ONE* 5(7): e11753.

Da Silva, J., Woodroffe, R., and Macdonald, D. W. (1993) Habitat, food availability and group territoriality in the European badger, Meles meles. *Oecologia* 95: 558–564.

Dai, A. (2013) Increasing drought under global warming in observations and models. *Nature Climate Change* 3(1): 52–58.

Darvill, B., Knight, M. E., and Goulson, D. (2004) Use of genetic markers to quantify bumblebee foraging range and nest density. *Oikos* 107(3): 471–478.

Dawson, W., Mndolwa, A. S., Burslem, D. F. R. P., and Hulme, P. E. (2008) Assessing the risks of plant invasions arising from collections in tropical botanical gardens. *Biodiversity and Conservation* 17: 1979–1995.

Delahay, R. J., Brown, J. A., Mallinson, P. J., Spyvee, P. D., Handoll, D., Rogers, L. M., and Cheeseman, C. L. (2000) The use of marked bait in studies of the territorial organization of the European Badger (*Meles meles*). *Mammal Review* 30(2): 73–87.

Delahay, R. J., Carter, S. P., Forrester, G. J., Mitchell, A., and Cheeseman, C. L. (2006) Habitat correlates of group size, bodyweight and reproductive performance in a high-density Eurasian badger (*Meles meles*) population. *Journal of Zoology* 270(3): 437–447.

Department for Environment Food and Rural Affairs, Department of Agriculture and Rural Development (Northern Ireland), Welsh Assembly Government, The Department for Rural Affairs and Heritage, The Scottish Government, Rural and Environment Research and Analytical Services (2012) *Agriculture in the United Kingdom 2017*.

DeYoung, R., and Honeycutt, R. L. (2005) The molecular toolbox: Genetic techniques in wildlife ecology and management. *Journal of Wildlife Management* 64(4): 1362–1384.

Diaz-Forero, I., Kuusemets, V., Mänd, M., Liivamägi, A., Kaart, T., and Luig, J. (2012) Influence of local and landscape factors on bumblebees in semi-natural meadows: A multiple-scale study in a forested landscape. *Journal of Insect Conservation* 17: 113–125.

Diekotter, T., Walther-Hellwig, K., Conradi, M., Suter, M., and Frankl, R. (2006) Effects of landscape elements on the distribution of the rare bumblebee species *Bombus muscorum* in an agricultural landscape. *Biodiversity and Conservation* 15: 43–54.

Dinh, K. V., Janssens, L., and Stoks, R. (2016) Exposure to a heat wave under food limitation makes an agricultural insecticide lethal: A mechanistic laboratory experiment. *Global change biology* 22(10): 3361–3372.

Dixon, D. R. (2003) A non-invasive technique for identifying individual badgers *Meles meles*. *Mammal Review* 33(1): 92–94.

Donkersley, P., Rhodes, G., Pickup, R. W., Jones, K. C., Wilson, K., and Philip

Donkersley, C. (2014) Honeybee nutrition is linked to landscape composition. *Ecology and Evolution* 4(21): 4195–4206.

Dukas, R., and Morse, D. H. (2003) Crab spiders affect flower visitation by bees. *Oikos* 101(1): 157–163.

Dukas, R. (2005) Bumble bee predators reduce pollinator density and plant fitness. *Ecology* 86(6): 1401–1406.

Easy, E., Begg, A., Campbell, S., Caudwell, E., Crookes, I., Davie, J., Garvey, K., Karim, A., Keylock, J., Mackley-Ward, H., Ovens, L., Reeder, M., Reid, C., Rubinstein-Baylis, O., Saunders, M., Smith, D., Steward, T., Whiting, J., and Yustisia, W. (2012) *The Future of land use in the South West: Food, water & energy security in the face of environmental change*.

Elliott, S. E. (2009) Surplus nectar available for subalpine bumble bee colony growth. *Environmental entomology* 38(6): 1680–1689.

Ellis, J. S., Knight, M. E., Darvill, B., and Goulson, D. (2006) Extremely low effective population sizes, genetic structuring and reduced genetic diversity in a threatened bumblebee species, *Bombus sylvarum* (Hymenoptera: Apidae). *Molecular Ecology* 15(14): 4375–4386.

Elmes, G. W., Thomas, J. A., and Wardlaw, J. C. (1991) Larvae of Maculinea rebeli, a large-blue butterfly, and their *Myrmica* host ants: Wild adoption and behaviour in ant-nests. *Journal of Zoology* 223(3): 447–460.

Feore, S., and Montgomery, W. I. (1999) Habitat effects on the spatial ecology of the European badger (*Meles meles*). *Journal of Zoology* 247: 537–549.

Fox, R., Oliver, T. H., Harrower, C., Parsons, M. S., Thomas, C. D., and Roy, D. B. (2014) Long-term changes to the frequency of occurrence of British moths are consistent with opposing and synergistic effects of climate and land-use changes. *Journal of Applied Ecology* 51(4): 949–957.

Free, J. B. (1958) The defence of bumblebee colonies. *Behaviour* 12(3): 233–242.

Freeman, R. B. (1968) Charles Darwin on the routes of male humble bees. *Bulletin of the British Museum (Natural History)* 3(6): 177–189.

Fründ, J., Linsenmair, K. E., and Blüthgen, N. (2010) Pollinator diversity and specialization in relation to flower diversity. *Oikos* 119(10): 1581–1590.

Fukase, J., and Simons, A, M. (2016) Increased pollinator activity in urban gardens with more native flora. *Applied Ecology and Environmental Research* 14(1): 297–310.

Fuller, G. A., and Plowright, R. C. (1986) Nest defence by honey-daubing in the bumblebee *Bombus griseocollis* de Geer (Hymenoptera: Apidae). *The Canadian Entomologist* 118(05): 479–480.

Fuller, R. M. (1987) The changing extent and conservation interest of lowland grasslands in England and Wales: A review of grassland surveys 1930-1984. *Biological Conservation* 40(4): 281–300.

Fussell, M., and Corbet, S. A. (1992) The nesting places of some British bumble bees. *Journal of Apicultural Research* 31(1): 32–41.

Gallai, N., Salles, J. M., Settele, J., and Vaissière, B. E. (2009) Economic valuation of the vulnerability of world agriculture confronted with pollinator decline. *Ecological Economics* 68(3): 810–821.

Gathmann, A., and Tscharntke, T. (2002) Foraging ranges of solitary bees. *Journal of Animal Ecology* 71(5): 757–764.

Gegear, R. J., Otterstatter, M. C., and Thomson, J. D. (2006) Bumble-bee foragers infected by a gut parasite have an impaired ability to utilize floral information. *Proceedings of the Royal Society B* 273: 1073–1078.

Génissel, A., Aupinel, P., Bressac, C., Tasei, J.-N., and Chevrier, C. (2002) Influence of pollen origin on performance of *Bombus terrestris* micro-colonies. *Entomologia Experimentalis et Applicata* 104: 329–336.

Gill, R. J., Ramos-Rodriguez, O., and Raine, N. E. (2012b) Combined pesticide exposure severely affects individual- and colony-level traits in bees. *Nature* 491: 105–108.

Goulson, D., and Stout, J. C. (2001) Homing ability of the bumblebee bombus terrestris (hymenoptera: apidae). *Apidologie* 32(1): 105–111.

Goulson, D, Hughes, W. O. H., Derwent, L. C., and Stout, J. C. (2002a) Colony growth of the bumblebee, *Bombus terrestris* in improved and conventional agricultural and suburban habitats. *Stout Source: Oecologia* 130(2): 267–273.

Goulson, D., Peat, J., Stout, J. C., Tucker, J., Darvill, B., Derwent, L. C., and Hughes, W. O. H. (2002b) Can alloethism in workers of the bumblebee, Bombus terrestris, be explained in terms of foraging efficiency? *Animal Behaviour* 64(1): 123–130.

Goulson, D., Hanley, M. E., Darvill, B., Ellis, J. S., and Knight, M. E. (2005) Causes of rarity in bumblebees. *Biological Conservation* 122(1): 1–8.

Goulson, D, Lye, G. C., and Darvill, B. (2008) Decline and conservation of bumble bees. *Annual review of entomology* 53: 191–208.

Goulson, D., Lepais, O., O’Connor, S., Osborne, J. L., Sanderson, R. A., Cussans, J., Goffe, L., and Darvill, B. (2010) Effects of land use at a landscape scale on bumblebee nest density and survival. *Journal of Applied Ecology* 47: 1207–1215.

Goulson, D., Nicholls, E., Botías, C., and Rotheray, E. L. (2015) Bee declines driven by combined stress from parasites, pesticides, and lack of flowers. *Science* 347(6229): 1255957.

Goulson, D., O’Connor, S., and Park, K. J. (2018a) The impacts of predators and parasites on wild bumblebee colonies. *Ecological Entomology* 43(12): 168–181.

Goulson, ., O’Connor, S., and Park, K. J. (2018b) Causes of colony mortality in bumblebees. *Animal Conservation* 21: 45–53.

Graves, H. B., Bellis, E. D., and Knuth, W. M. (1972) Censusing white-tailed deer by airborne thermal infrared imagery. *Wildlife Society* 36(3): 875–884.

Graystock, P., Yates, K., Evison, S. E. F., Darvill, B., Goulson, D., and Hughes, W. O. H. (2013) The Trojan hives: Pollinator pathogens, imported and distributed in bumblebee colonies. *Journal of Applied Ecology* 50(5): 1207–1215.

Greenleaf, S. S., Williams, N. M., Winfree, R., and Kremen, C. (2007) Bee foraging ranges and their relationship to body size. *Oecologia* 153(3): 589–

596.

Grimm, V., Ayllón, D., and Railsback, S. F. (2017) Next-generation individual-based models integrate biodiversity and ecosystems: yes we can, and yes we must. *Ecosystems* 20(2): 229–236.

Hagen, M., Wikelski, M., and Kissling, W. D. (2011) Space use of bumblebees (*Bombus* spp.) revealed by radio-tracking. *PLoS ONE* 6(5): e19997.

Haines-Young, R. H., Barr, C. J., Black, H. I. J., Briggs, D. J., Bunce, R. G. H., Clarke, R. T., et al. (2000) *Accounting for nature: assessing habitats in the UK countryside*. London, DETR.

Hallmann, C. A., Sorg, M., Jongejans, E., Siepel, H., Hofland, N., Schwan, H., Stenmans, W., Müller, A., Sumser, H., Hörren, T., Goulson, D., and De Kroon, H. (2017) More than 75 percent decline over 27 years in total flying insect biomass in protected areas. *PLoS ONE* 12(10).

Hanley, M. E., Awbi, A. J., and Franco, M. (2014) Going native? Flower use by bumblebees in English urban gardens. *Annals of Botany* 113(5): 799–806.

Hannon, L. E., and Sisk, T. D. (2009) Hedgerows in an agri-natural landscape: Potential habitat value for native bees. *Biological Conservation* 142(10): 2140–2154.

Happe, A. K., Riesch, F., Rösch, V., Gallé, R., Tscharntke, T., and Batáry, P. (2018) Small-scale agricultural landscapes and organic management support wild bee communities of cereal field boundaries. *Agriculture, Ecosystems and Environment* 254: 92–98.

Hardwick, K. A., Fiedler, P., Lee, L. C., Pavlik, B., Hobbs, R. J., Aronson, J., et al. (2011) The role of botanic gardens in the science and practice of ecological restoration. *Conservation Biology* 25(2): 265–275.

Harris, S. (1984) Ecology of urban badgers *Meles meles*: Distribution in Britain and habitat selection, persecution, food and damage in the city of Bristol. *Biological Conservation* 28(4): 349–375.

Harris, S., and Cresswell, W. J. (1987) Dynamics of a suburban badger (*Meles meles*) population. *Symposia of the Zoological Society of London* 58: 295–311.

Hass, Annika Louise, Brachmann, L., Batáry, P., Clough, Y., Behling, H., and Tscharntke, T. (2018) Maize-dominated landscapes reduce bumblebee colony growth through pollen diversity loss. *Journal of Applied Ecology* 56: 294–304.

Häussler, J., Sahlin, U., Baey, C., Smith, H. G., and Clough, Y. (2017) Pollinator population size and pollination ecosystem service responses to enhancing floral and nesting resources. *Ecology and Evolution* 7(6): 1898–1908.

Heard, M. S., Carvell, C., Carreck, N. L., Rothery, P., Osborne, J. L., and Bourke, A. F. G. (2007) Landscape context not patch size determines bumble-bee density on flower mixtures sown for agri-environment schemes. *Biology Letters* 3(6): 638–641.

Heinrich, B. (1974) Thermoregulation in bumblebees I. Brood incubation by *Bombus vosnesenskii* queens. *Journal of Comparative Physiology* 88: 129–140.

Heinrich, B. (1975) Thermoregulation in bumblebees II. Energetics of warm-up and free flight. *Journal of Comparative Physiology* 96: 155–166.

Heinrich, B. (1976) The foraging specializations of individual bumblebees. *Ecological Monographs* 46(2): 105–128.

Henry, M., Becher, M. A., Osborne, J. L., Kennedy, P. J., Aupinel, P., Bretagnolle, V., Brun, F., Grimm, V., Horn, J., and Requier, F. (2017) Predictive systems models can help elucidate bee declines driven by multiple combined stressors. *Apidologie* 48(3): 328–339.

Herrmann, F., Westphal, C., Moritz, R. F. A., and Steffan-Dewenter, I. (2007) Genetic diversity and mass resources promote colony size and forager densities of a social bee (*Bombus pascuorum*) in agricultural landscapes. *Molecular Ecology* 16: 1167–1178.

Heywood, V. H. (1995) *Global biodiversity assessment*. *United Nations Environment Programme*. Cambridge University Press, Cambridge.

Hipólito, D., Guedes, D., Cabecinha, D., Serronha, A., Grilo, C., Santos-Reis, M., Monterroso, P., Carvalho, J., Fonseca, C., Pardavila, X., Virgós, E., and Rosalino, L. M. (2018) Drivers of sett site location by European badgers in

Portugal. *Biodiversity and Conservation*: 1–20.

Hogeweg, P., and Hesper, B. (1983) The ontogeny of the interaction structure in bumble bee colonies: A MIRROR model. *Behavioral Ecology and Sociobiology* 12(4): 271–283.

Holzschuh, A., Steffan-Dewenter, I., Kleijn, D., and Tscharntke, T. (2007) Diversity of flower-visiting bees in cereal fields: Effects of farming system, landscape composition and regional context. *Journal of Applied Ecology* 44(1): 41–49.

Hounsome, T. D., Young, R. P., Davison, J., Yarnell, R. W., Trewby, I. D., Garnett, B. T., Delahay, R. J., and Wilson, G. J. (2005) An evaluation of distance sampling to estimate badger (*Meles meles*) abundance. *Journal of Zoology* 266(1): 81–87.

Hulme, P. E. (2011) Addressing the threat to biodiversity from botanic gardens. *Trends in Ecology and Evolution* 26(4): 168–174.

Imhoof, B., and Schmid-Hempel, P. (1999) Colony success of the bumble bee, Bombus terrestris, in relation to infections by two protozoan parasites, *Crithidia bombi* and *Nosema bombi*. *Insectes Sociaux* 46: 233–238.

Ings, T. C., Ward, N. L., and Chittka, L. (2006) Can commercially imported bumble bees out-compete their native conspecifics? *Journal of Applied Ecology* 43(5): 940–948.

IPCC (2014) *Climate change 2014 : synthesis report*. (Pachauri, R. K. and Mayer, L., Eds.). Geneva, Switzerland: IPCC.

James, K., and Rice, D. (2002) Finding termites with thermal imaging. In *Inframation*.

Jha, S., and Kremen, C. (2013a) Resource diversity and landscape-level homogeneity drive native bee foraging. *Proceedings of the National Academy of Sciences of the United States of America* 110(2): 555–8.

Jha, S., Stefanovich, L., and Kremen, C. (2013b) Bumble bee pollen use and preference across spatial scales in human-altered landscapes. *Ecological Entomology* 38(6): 570–579.

Johnson, M. F., Hannah, C., Acton, L., Popovici, R., Karanth, K. K., and Weinthal, E. (2014) Network environmentalism: Citizen scientists as agents for environmental advocacy. *Global Environmental Change* 29: 235–245.

Kallioniemi, E., Åström, J., Rusch, G. M., Dahle, S., Åström, S., and Gjershaug, J. O. (2017) Local resources, linear elements and mass-flowering crops determine bumblebee occurrences in moderately intensified farmlands. *Agriculture, Ecosystems and Environment* 239: 90–100.

Kämper, W., Werner, P. K., Hilpert, A., Westphal, C., Blüthgen, N., Eltz, T., and Leonhardt, S. D. (2016) How landscape, pollen intake and pollen quality affect colony growth in Bombus terrestris. *Landscape Ecology* 31(10): 2245–2258.

Kastberger, G., and Stachl, R. (2003) Infrared imaging technology and biological applications. *Behavior research methods, instruments, & computers* 35(3): 429–439.

Keeling, M. J., Franklin, D. N., Datta, S., Brown, M. A., and Budge, G. E. (2017) Predicting the spread of the Asian hornet (*Vespa velutina*) following its incursion into Great Britain. *Scientific Reports* 7(1): 6240.

Kells, A. R., and Goulson, D. (2003) Preferred nesting sites of bumblebee queens (Hymenoptera: Apidae) in agroecosystems in the UK. *Biological Conservation* 109: 165–174.

Kennedy, C. M., Lonsdorf, E., Neel, M. C., Williams, N. M., Ricketts, T. H., Winfree, R., et al. (2013) A global quantitative synthesis of local and landscape effects on wild bee pollinators in agroecosystems. *Ecology Letters* 16(5): 584–599.

Kirchner, W. H., and Röschard, J. (1999) Hissing in bumblebees: An interspecific defence signal. *Insectes Sociaux* 46(3): 239–243.

Kitaysky, A. S., Piatt, J. F., and Wingfield, J. C. (2007) Stress hormones link food availability and population processes in seabirds. *Marine Ecology Progress Series* 352: 245–258.

Kleijn, D., and van Langevelde, F. (2006) Interacting effects of landscape context and habitat quality on flower visiting insects in agricultural landscapes. *Basic and Applied Ecology* 7(3): 201–214.

Klein, A. M., Vaissière, B. E., Cane, J. H., Steffan-Dewenter, I., Cunningham, S. A., Kremen, C., and Tscharntke, T. (2007) Importance of pollinators in changing landscapes for world crops. *Proceedings of the Royal Society B: Biological Sciences* 274: 303–313.

Knapp, J. L., Becher, M. A., Rankin, C. C., Twiston-Davies, G., and Osborne, J. L. (2018) *Bombus terrestris* in a mass-flowering pollinator-dependent crop: A mutualistic relationship? *Ecology and Evolution* 9: 609–618.

Knight, M. E., Martin, A. P., Bishop, S., Osborne, J. L., Hale, R. J., Sanderson, R. A., and Goulson, D. (2005) An interspecific comparison of foraging range and nest density of four bumblebee (*Bombus*) species. *Molecular ecology* 14(6): 1811–1820.

Knight, M. E., Osborne, J. L., Sanderson, R. A., Hale, R. J., Martin, A. P., and Goulson, D. (2009) Bumblebee nest density and the scale of available forage in arable landscapes. *Insect Conservation and Diversity* 2: 116–124.

Kovac, H., and Schmaranzer, S. (1996) Thermoregulation of honeybees (*Apis mellifera*) foraging in spring and summer at different plants. *Journal of Insect Physiology* 42(11–12): 1071–1076.

Kowalczyk, R., Zalewski, A., and Bogumiła, J. (2006) Daily movement and territory use by badgers *Meles meles* in Białowieża Primeval Forest, Poland. 12(4): 385–391. *https://doi.org/10.2981/0909-6396*

Kremen, C., Williams, N. M., Aizen, M. a., Gemmill-Herren, B., LeBuhn, G., Minckley, R., Packer, L., Potts, S. G., Roulston, T., Steffan-Dewenter, I., Vázquez, D. P., Winfree, R., Adams, L., Crone, E. E., Greenleaf, S. S., Keitt, T. H., Klein, A. M., Regetz, J., and Ricketts, T. H. (2007) Pollination and other ecosystem services produced by mobile organisms: A conceptual framework for the effects of land-use change. *Ecology Letters* 10(4): 299–314.

Kriesell, L., Hilpert, A., and Leonhardt, S. D. (2017) Different but the same: Bumblebee species collect pollen of different plant sources but similar amino acid profiles. *Apidologie* 48(1): 102–116.

Kruuk, H. (1978) Foraging and Spatial Organisation of the European badger, *Meles meles* L . *Behavioral Ecology and Sociobiology* 4(1): 75–89.

Kruuk, H, Parish, T., Brown, C. A. J., and Carrera, J. (1979) The use of pasture by the European badger (*Meles meles*). *Journal of Applied Ecology* 16(2): 453–459.

Kruuk, H., and Parish, T. (1981) Feeding specialisation of the European badger *Meles meles* in Scotland. *Journal of Animal Ecology* 50(3): 773–788.

Kruuk, H., and Parish, T. (1982) Factors affecting population density, group size and territory size of the European badger, *Meles meles*. *Journal of Zoology* 196(1): 31–39.

Lamb, C. (2004) *From the ends of the earth : passionate plant collectors remembered in a Cornish garden*. London, Bene Factum Publishing.

Langellotto, G. A., Melathopoulos, A., Messer, I., Anderson, A., McClintock, N., and Costner, L. (2018) Garden pollinators and the potential for ecosystem service flow to urban and peri-urban agriculture. *Sustainability* 10(6): 2047.

Laverty, T. M. (1994) Costs to foraging bumble bees of switching plant species. *Canadian Journal of Zoology* 72(1): 43–47.

Leonhardt, S. D., and Blüthgen, N. (2012) The same, but different: Pollen foraging in honeybee and bumblebee colonies. *Apidologie* 43(4): 449–464.

Lister, B. C., and Garcia, A. (2018) Climate-driven declines in arthropod abundance restructure a rainforest food web. *Proceedings of the National Academy of Sciences* 115(44): E10397–E10406.

Loram, A., Tratalos, J., Warren, P. H., and Gaston, K. J. (2007) Urban domestic gardens (X): The extent & structure of the resource in five major cities. *Landscape Ecology* 22(4): 601–615.

Loram, A., Warren, P. H., and Gaston, K. J. (2008) Urban domestic gardens (XIV): The characteristics of gardens in five cities. *Environmental Management* 42(3): 361–376.

Lye, G. C., Osborne, J. L., Park, K. J., and Goulson, D. (2012) Using citizen science to monitor *Bombus* populations in the UK: Nesting ecology and relative abundance in the urban environment. *Journal of Insect Conservation* 16(5): 697–707.

Macdonald, D W, and Newman, C. (2002) Population dynamics of badgers (*Meles meles*) in Oxfordshire, U.K.: Numbers, density and cohort life histories, and a possible role of climate change in population growth. *Journal of Zoology* 256(1): 121–138.

Macdonald, D. W., Newman, C., Nouvellet, P. M., and Buesching, C. D. (2009) An analysis of Eurasian badger (*Meles meles*) population dynamics: Implications for regulatory mechanisms. *Journal of Mammalogy* 90(6): 1392–1403.

Majewska, A. A., and Altizer, S. (2018) Planting gardens to support insect pollinators. *Conservation Biology* : 1–11. doi:10.1111/cobi.13271.

Makinson, J. C., Woodgate, J. L., Reynolds, A., Capaldi, E. A., Perry, C. J., and Chittka, L. (2019) Harmonic radar tracking reveals random dispersal pattern of bumblebee (*Bombus terrestris*) queens after hibernation. *Scientific Reports* 9(1): 4651.

Mandelik, Y., Winfree, R., Neeson, T., and Kremen, C. (2012) Complementary habitat use by wild bees in agro-natural landscapes. *Ecological Applications* 22(5): 1535–1546.

Manickavasagan, A., Jayas, D. S., and White, N. D. G. (2008) Thermal imaging to detect infestation by *Cryptolestes ferrugineus* inside wheat kernels. *Journal of Stored Products Research* 44: 186–192.

Manley, R., Boots, M., and Wilfert, L. (2015) Emerging viral disease risk to pollinating insects: Ecological, evolutionary and anthropogenic factors. *Journal of Applied Ecology* 52(2): 331–340.

Manley, R., Boots, M., and Wilfert, L. (2017) Condition-dependent virulence of slow bee paralysis virus in *Bombus terrestris*: Are the impacts of honeybee viruses in wild pollinators underestimated? *Oecologia* 184(2): 305–315.

Mathews, F., Swindells, M., Goodhead, R., August, T. A., Hardman, P., Linton, D. M., and Hosken, D. J. (2013) Effectiveness of search dogs compared with human observers in locating bat carcasses at wind-turbine sites : A blinded randomized trial. *Wildlife Society Bulletin* 37(1): 34–40.

McKay, A. F., Ezenwa, V. O., and Altizer, S. (2016) Consequences of food restriction for immune defense, parasite infection, and fitness in Monarch butterflies. *Physiological and Biochemical Zoology* 89(5): 389–401.

Menz, M. H. M., Phillips, R. D., Winfree, R., Kremen, C., Aizen, M. A., Johnson, S. D., and Dixon, K. W. (2011) Reconnecting plants and pollinators: Challenges in the restoration of pollination mutualisms. *Trends in Plant Science*.

Morton, D., Rowland, C., Wood, C., Meek, L., Marston, C., Smith, G., Wadsworth, R., and Simpson, I. C. (2011) *Final Report for LCM2007 – the new UK land cover map*. *Countryside Survey Technical Report No 11/07 NERC/Centre for Ecology & Hydrology*.

Muller, C., and Schmid-Hempel, P. (1992) Correlates of reproductive success among field colonies of *Bombus lucorum*: The importance of growth and parasites. *Ecological Entomology* 17: 343–353.

Nagamitsu, T., and Yamagishi, H. (2009) Nest density, genetic structure, and triploid workers in exotic *Bombus terrestris* populations colonized Japan. *Apidologie* 40: 429–440.

Nanje Gowda, N. A., and Alagusundaram, K. (2013) Use of thermal imaging to improve the food grains quality during storage. *International Journal of Current Agricultural Research* 1(7): 34–41.

Nilsson, S., Franzén, M., and Jönsson, E. (2008) Long-term land-use changes and extinction of specialised butterflies. *Insect Conservation and Diversity* 1: 197–207.

Noonan, M. J., Markham, A., Newman, C., Trigoni, N., Buesching, C. D., Ellwood, S. A., and MacDonald, D. W. (2015) A new Magneto-Inductive tracking technique to uncover subterranean activity: What do animals do underground? *Methods in Ecology and Evolution* 6: 510–520.

O’Connor, S., Park, K. J., and Goulson, D. (2012) Humans versus dogs; A comparison of methods for the detection of bumble bee nests. *Journal of Apicultural Research* 51(2): 204–211.

O’connor, S., Park, K. J., and Goulson, D. (2017) Location of bumblebee nests is predicted by counts of nest-searching queens. *Ecological Entomology* 42: 731–736.

Öckinger, E., and Smith, H. G. (2007) Semi-natural grasslands as population sources for pollinating insects in agricultural landscapes. *Journal of Applied Ecology* 44(1): 50–59.

Öckinger, E., Franzén, M., Rundlöf, M., and Smith, H. G. (2009) Mobility-dependent effects on species richness in fragmented landscapes. *Basic and Applied Ecology* 10(6): 573–578.

Ogilvie, J. E., and Forrest, J. R. (2017) Interactions between bee foraging and floral resource phenology shape bee populations and communities. *Current Opinion in Insect Science* 21: 75–82.

Ollerton, J., Winfree, R., and Tarrant, S. (2011) How many flowering plants are pollinated by animals? *Oikos* 120(3): 321–326.

Olson, G. L., Myers, J. H., Hemerik, L., and Cory, J. S. (2017) Phylloplane bacteria increase the negative impact of food limitation on insect fitness. *Ecological Entomology* 42(4): 411–421.

Olsson, O., Bolin, A., Smith, H. G., and Lonsdorf, E. V (2015) Modeling pollinating bee visitation rates in heterogeneous landscapes from foraging theory. *Ecological Modelling* 316: 133–143.

Osborne, J. L, Williams, I. H., Carreck, N., Poppy, G. M., Osbome, J. L., Williams, L. H., Carreck, N. L., Poppy, G. M., Riley, J. R., Smith, A. D., Reynolds, D. R., and Edwards, A. S. (1997) Harmonic radar: a new technique for investigating bumblebee and honey bee foraging flight. *Acta Horticulturae* 437: 159–163.

Osborne, J.L., Martin, A. P., Carreck, N. L., Swain, J. L., Knight, M. E., Goulson, D., Hale, R. J., and Sanderson, R. A. (2008a) Bumblebee flight distances in relation to the forage landscape. *The Journal of animal ecology* 77(2): 406–415.

Osborne, J. L., Martin, A. P., Shortall, C. R., Todd, A. D., Goulson, D., Knight, M. E., Hale, R. J., and Sanderson, R. A. (2008b) Quantifying and comparing bumblebee nest densities in gardens and countryside habitats. *Journal of Applied Ecology* 45: 784–792.

Pardee, G. L., and Philpott, S. M. (2014) Native plants are the bee's knees: local and landscape predictors of bee richness and abundance in backyard gardens. *Urban Ecosystems* 17: 641–659.

Parmentier, L., Meeus, I., Cheroutre, L., Mommaerts, V., Louwye, S., and Smagghe, G. (2014) Commercial bumblebee hives to assess an anthropogenic environment for pollinator support: A case study in the region of Ghent (Belgium). *Environmental Monitoring and Assessment* 186(4): 2357–2367.

Pautasso, M., and Parmentier, I. (2007) Are the living collections of the world's botanical gardens following species-richness patterns observed in natural ecosystems? *Botanica Helvetica* 117(1): 15–28.

Pease, A. (1898) The Badger: A Monograph.

Pelletier, L., and Mcneil, J. N. (2003) The effect of food supplementation on reproductive success in bumblebee field colonies. *Source: Oikos* 103(3): 688–694.

Persson, A. S., and Smith, H. G. (2011) Bumblebee colonies produce larger foragers in complex landscapes. *Basic and Applied Ecology* 12(8): 695–702.

Petersen, Jessica D., Reiners, S., and Nault, B. A. (2013) Pollination services provided by bees in pumpkin fields supplemented with either *Apis mellifera* or *Bombus impatiens* or not supplemented. *PLoS ONE* 8(7): e69819.

Petersen, J. D., Huseth, A. S., and Nault, B. A. (2014) Evaluating pollination deficits in pumpkin production in New York. *Environmental Entomology* 43(5): 1247–1253.

Phillips, B. B., Shaw, R. F., Holland, M. J., Fry, E. L., Bardgett, R. D., Bullock, J. M., and Osborne, J. L. (2018) Drought reduces floral resources for pollinators. *Global Change Biology* 24: 3226–3235.

Plascencia, M., and Philpott, S. M. (2017) Floral abundance, richness, and spatial distribution drive urban garden bee communities. *Bulletin of*

Entomological Research 107: 658–667.

Pocock, M. J. O., Roy, H. E., Preston, C. D., and Roy, D. B. (2015) The Biological Records Centre: A pioneer of citizen science. *Biological Journal of the Linnean Society* 115(3): 475–493.

Pope, N. S., and Jha, S. (2017) Seasonal food scarcity prompts long-distance foraging by a wild social bee. *The American Naturalist* 191(1): 45–57.

Potter, A., and LeBuhn, G. (2015) Pollination service to urban agriculture in San Francisco, CA. *Urban Ecosystems* 18(3): 885–893.

Potts, S. G., Biesmeijer, J. C., Kremen, C., Neumann, P., Schweiger, O., and Kunin, W. E. (2010) Global pollinator declines: trends, impacts and drivers. *Trends in Ecology and Evolution* 25(6): 345–353.

Pouvreau, A. (1973) Les ennemis des bourdons. *Apidologie* 4(2): 103–148.

Preisser, E. L., Bolnick, D. I., and Benard, M. E. (2005) Scared to death? The effects of intimidation and consumption in predator-prey interactions. *Ecology* 86(2): 501–509.

Prŷs-Jones, O. E., and Corbet, S. A. (2011) *Bumblebees*. Pelagic Publishing.

Rao, S., and Strange, J. P. (2012) Bumble bee (Hymenoptera: Apidae) foraging distance and colony density associated with a late-season mass flowering crop. *Environmental Entomology* 41(4): 905–915.

Rau, P. (1924) Notes on captive colonies and homing of *Bombus pennsylvanicus* de Geer. *Annals of the Entomological Society of America* 17(4): 368–381.

Rayner, C. J., and Langridge, D. F. (1985) Amino acids in bee-collected pollens from Australian indigenous and exotic plants. *Australian Journal of Experimental Agriculture* 25(3): 722–726.

Redhead, J. W., Dreier, S., Bourke, A. F. G., Heard, M. S., Jordan, W. C., Sumner, S., Wang, J., and Carvell, C. (2016) Effects of habitat composition and landscape structure on worker foraging distances of five bumble bee species. *Ecological Applications* 26(3): 726–739.

Ricketts, T. H., Regetz, J., Steffan-Dewenter, I., Cunningham, S. A., Kremen, C., Bogdanski, A., Gemmill-Herren, B., Greenleaf, S. S., Klein, A. M., Mayfield, M. M., Morandin, L. A., Ochieng', A., Potts, S. G., and Viana, B. F. (2008) Landscape effects on crop pollination services: are there general patterns? *Ecology letters* 11(5): 499–515.

Ridding, L. E., Redhead, J. W., and Pywell, R. F. (2015) Fate of semi-natural grassland in England between 1960 and 2013: A test of national conservation policy. *Global Ecology and Conservation* 4: 516–525.

Riley, J. R., Smith, A. D., Reynolds, D. R., Edwards, A. S., Osborne, J. L., Williams, I. H., Carreck, N. L., and Poppy, G. M. (1996) Tracking bees with harmonic radar. *Nature* 379: 29–30.

Roberts, B. R., and Osborne, J. L. (2019) Testing the efficacy of a thermal camera as a search tool for locating wild bumble bee nests. doi:10.1080/00218839.2019.1614724.

Robertson, A., McDonald, R. A., Delahay, R. J., Kelly, S. D., and Bearhop, S. (2014) Individual foraging specialisation in a social mammal: the European badger (*Meles meles*). *Oecologia* 176(2): 409–421.

Robertson, A., McDonald, R. A., Delahay, R. J., Kelly, S. D., and Bearhop, S. (2015) Resource availability affects individual niche variation and its consequences in group-living European badgers *Meles meles*. *Oecologia* 178: 31–43.

Robinson, R. A., and Sutherland, W. J. (2002) Post-war changes in arable farming and biodiversity in Great Britain. *Journal of Applied Ecology* 39(1): 157–176.

Rogers, L. M., Cheeseman, C. L., Mallinson, P. J., and Clifton-Hadley, R. (1997) The demography of a high-density badger (*Meles meles*) population in the west of England. *Journal of Zoology* 242: 705–728.

Romero, G. Q., Antiqueira, P. A. P., and Koricheva, J. (2011) A meta-analysis of predation risk effects on pollinator behaviour. *PLoS ONE* 6(6): e20689

Roper, T. (2010) *Badger*. London, UK: Collins.

Rose, F., and O'Reilly, C. (2006) *The wild flower key : how to identify wild flowers, trees and shrubs in Britain and Ireland*. Frederick Warne.

Rotheray, E. L., Osborne, J. L., and Goulson, D. (2017) Quantifying the food requirements and effects of food stress on bumble bee colony development. doi:10.1080/00218839.2017.1307712.

Roulston, T. H., and Goodell, K. (2011) The role of resources and risks in regulating wild bee populations. *Annual Review of Entomology* 56(1): 293–312.

Rundlöf, M., Persson, A. S., Smith, H. G., and Bommarco, R. (2014) Late-season mass-flowering red clover increases bumble bee queen and male densities. *Biological Conservation* 172: 138–145.

Rundlöf, M., Andersson, G. K. S., Bommarco, R., Fries, I., Hederström, V., Herbertsson, L., Jonsson, O., Klatt, B. K., Pedersen, T. R., Yourstone, J., and Smith, H. G. (2015) Seed coating with a neonicotinoid insecticide negatively affects wild bees. *Nature* 521: 77–80.

Russo, L., Debarros, N., Yang, S., Shea, K., and Mortensen, D. (2013) Supporting crop pollinators with floral resources: network-based phenological matching. *Ecology and Evolution* 3(9): 3125–3140.

Sadd, B. M. (2011) Food-environment mediates the outcome of specific interactions between a bumblebee and its trypanosome parasite. *Evolution* 65(10): 2995–3001.

Salisbury, A., Armitage, J., Bostock, H., Perry, J., Tatchell, M., and Thompson, K. (2015) Enhancing gardens as habitats for flower-visiting aerial insects (pollinators): Should we plant native or exotic species? *Journal of Applied Ecology* 52: 1156–1164.

Satchell, J. E. (1983) *Earthworm ecology : from Darwin to vermiculture*. Chapman and Hall.

Sauermann, H., and Franzoni, C. (2015) Crowd science user contribution patterns and their implications. *Proceedings of the National Academy of Sciences* 112(3): 679–684.

Scheper, J., Bommarco, R., Holzschuh, A., Potts, S. G., Riedinger, V., Roberts, S. P. M., Rundl€ Of, M., Smith, H. G., Steffan-Dewenter, I., Wickens, J. B., Wickens, V. J., and Kleijn, D. (2015) Local and landscape-level floral resources explain effects of wildflower strips on wild bees across four European countries. *Journal of Applied Ecology* 52: 1165–1175.

Schmid-Hempel, R., and Schmid-Hempel, P. (1998) Colony performance and immonocompetence of a social insect, *Bombus terrestris*, in poor and variable environments. *Functional Ecology* 12(1): 22–30.

Schneck, M. E., and Dagnelie, G. (2011) Prosthetic vision assessment. In *Visual Prosthetics: Physiology, Bioengineering, Rehabilitation*. Springer doi:10.1007/978-1-4419-0754-7_20.

Senapathi, D., Goddard, M. A., Kunin, W. E., and Baldock, K. C. R. (2016) Landscape impacts on pollinator communities in temperate systems: evidence and knowledge gaps. doi:10.1111/1365-2435.12809.

Shackelford, G., Steward, P. R., Benton, T. G., Kunin, W. E., Potts, S. G., Biesmeijer, J. C., and Sait, S. M. (2013) Comparison of pollinators and natural enemies: A meta-analysis of landscape and local effects on abundance and richness in crops. *Biological Reviews* 88(4): 1002–1021.

Shepherdson, D. J., Roper, T. J., and Lüps, P. (1990) Diet, food availability and foraging behaviour of badgers (*Meles meles* L.) in southern England. *Z. Säugetierkunde* 55: 81–93.

Sikora, A., Michołap, P., and Kelm, M. (2016) Flowering Plants Preferred by Bumblebees (Bombus Latr.) in the Botanical Garden of Medicinal Plants in Wrocław. *Journal of Apicultural Science* 60(2): 59–68.

Sladen, F. W. L. (1912) *The humble-bee, its life-history and how to domesticate it*. London, UK: Macmillan and co.

Smal, C. (1995) *The badger and habitat survey of Ireland: The abundance and distribution of the badger Meles meles in Ireland, with especial reference to habitat surveys*. Dublin.

Smith, R., and Mulholland, B. (2008) Use of micropropagation for the conservation of rare Cornish garden plants at risk from *Phytophthora ramorum*©. *Combined Proceedings International Plant Propagators' Society* 58: 456–460.

Söderman, A. M. E., Irminger Street, T., Hall, K., Olsson, O., Prentice, H. C., and Smith, H. G. (2018) The value of small arable habitats in the agricultural landscape: Importance for vascular plants and the provisioning of floral resources for bees. *Ecological Indicators* 84: 553–563.

Spiesman, B. J., Bennett, A., Isaacs, R., and Gratton, C. (2017) Bumble bee colony growth and reproduction depend on local flower dominance and natural habitat area in the surrounding landscape. *Biological Conservation* 206: 217–223.

Stabentheiner, A., and Schmaranzer, S. (1987) Thermographic determination of body temperatures in honey bees and hornets: Calibration and applications. *Thermology* 2: 563–572.

Stanley, D. A., Garratt, M. P. D., Wickens, J. B., Wickens, V. J., Potts, S. G., and Raine, N. E. (2015) Neonicotinoid pesticide exposure impairs crop pollination services provided by bumblebees. *Nature* 528(7583): 548–50.

Starr, C. K. (1998) The nesting association of the social wasps *Mischocyttarus immarginatus* and *Polybia* spp. in Costa Rica. *Biotropica* 20(2): 171–173.

Steffan-Dewenter, I., Münzenberg, U., Bürger, C., Thies, C., and Tscharntke, T. (2002) Scale-dependent effects of landscape context on three pollinator guilds. *Ecology* 83(5): 1421–1432.

Stelzer, R. J., Chittka, L., Carlton, M., and Ings, T. C. (2010) Winter active bumblebees (*Bombus terrestris*) achieve high foraging rates in urban Britain. *PLoS ONE* 5(3): e9559.

Stoate, C. (1996) The changing face of lowland farming and wildlife part 2 1945-1995. *British Wildlife* 7(3): 162–172.

Stoate, C, Boatman, N. D., Borralho, R. J., Carvalho, C. R., De Snoo, G. R., and Eden, P. (2001) Ecological impacts of arable intensification in Europe. *Journal of Environmental Management* 63(4): 337–365.

Stouffer, D. B., Cirtwill, A. R., and Bascompte, J. (2014) How exotic plants integrate into pollination networks. *Journal of Ecology* 102(6): 1442–1450.

Stout, J. C. (2007) Pollination of invasive *Rhododendron ponticum* (Ericaceae) in Ireland. *Apidologie* 38(2): 198–206.

Streit, S., Bock, F., Pirk, C. W. W., and Tautz, J. (2003) Automatic life-long monitoring of individual insect behaviour now possible. *Zoology* 106: 169–171.

Suzuki, Y., Kawaguchi, L. G., Munidasa, D. T., and Toquenaga, Y. (2009) Do bumble bee queens choose nest sites to maximize foraging rate? Testing models of nest site selection. *Behavioral Ecology and Sociobiology* 63(9): 1353–1362.

Svensson, B., Lagerlof, J., and Svensson, G. (2000) Habitat preferences of nest-seeking bumble bees (Hymenoptera: Apidae) in an agricultural landscape. *Agriculture, Ecosystems & Environment* 77: 247–255.

Tasei, J.-N., and Aupinel, P. (2008) Nutritive value of 15 single pollens and pollen mixes tested on larvae produced by bumblebee workers (*Bombus terrestris*, Hymenoptera: Apidae). *Apidologie* 39(4): 397–409.

QGIS Development Team (2018) QGIS Geographic Information System. Open Source Geospatial Foundation Project. Available at: http://qgis.osgeo.org.

R Core Team. (2017) R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing. Available at: https://www.r-project.org/.

Tiedeken, E. J., and Stout, J. C. (2015) Insect-flower interaction network structure is resilient to a temporary pulse of floral resources from invasive *Rhododendron ponticum*. *PLoS ONE* 10(3): 1–19.

Timberlake, T. P., Vaughan, I. P., and Memmott, J. (2019) Phenology of farmland floral resources reveals seasonal gaps in nectar availability for bumblebees. *Journal of Applied Ecology* 00: 1–12. doi:10.1111/1365-2664.13403.

Torretta, J. P., Medan, D., and Abrahamovich, A. H. (2006) First record of the invasive bumblebee *Bombus terrestris* (L.) (Hymenoptera, Apidae) in Argentina. *Transactions of the American Entomological Society* 132(3): 285–289.

van Honk, C., and Hogeweg, P. (1981) The ontogeny of the social structure in a captive *Bombus terrestris* colony. *Behavioral Ecology and Sociobiology* 9(2): 111–119.

Vanbergen, A. J., Espíndola, A., and Aizen, M. A. (2018) Risks to pollinators and pollination from invasive alien species. *Nature Ecology & Evolution* 2.

Vanbergen, A. J., and Insect Pollinators Initiative, T. (2013) Threats to an ecosystem service: Pressures on pollinators. *Frontiers in Ecology and the Environment* 11(5): 251–259.

Vaughan, I., Gotelli, N. J., Memmott, J., Pearson, C. E., Woodward, G., and Symondson, W. O. (2018) econullnetr: An R package using null models to analyse the structure of ecological networks and identify resource selection. *Methods in Ecology and Evolution* 9(3): 728–733

Velthuis, H. H. W., and van Doorn, A. (2006) A century of advances in bumblebee domestication and the economic and environmental aspects of its commercialization for pollination. *Apidologie* 37(4): 421–451.

Vogt, F. D., Heinrich, B., and Plowright, C. (1998) Ovary development in bumble bee queens: The influence of abdominal temperature and food availability. *Canadian Journal of Zoology* 76(11): 2026–2030.

Walters, S. A., and Taylor, B. H. (2006) Effects of honey bee pollination on pumpkin fruit and seed yield. *HortScience* 41(2): 370–373.

Waters, J., O’Connor, S., Park, K. J., and Goulson, D. (2011) Testing a detection dog to locate bumblebee colonies and estimate nest density. *Apidologie* 42: 200–205.

Webber, C., and Peterson, A. (2012) Native and exotic flower visitors in the Christchurch Botanic Gardens and their contrasting plant preferences. *New Zealand Natural Sciences* 37: 37–49.

Westphal, C., Steffan-Dewenter, I., and Tscharntke, T. (2009) Mass flowering oilseed rape improves early colony growth but not sexual reproduction of bumblebees. *Journal of Applied Ecology* 46(1): 187–193.

Westphal, C., Steffan-Dewenter, I., and Tscharntke, T. (2006) Foraging trip duration of bumblebees in relation to landscape-wide resource availability. *Ecological Entomology* 31(4): 389–394.

Westphal, C., Steffan-Dewenter, I., and Tscharntke, T. (2003) Mass flowering crops enhance pollinator densities at a landscape scale. *Ecology Letters* 6: 961–965.

Wickramaratne, M. R. T., and Vitarana, S. I. (1985) Insect pollination of tea, *Camellia sinensis*, in Sri Lanka. *Tropical Agriculture* 62(3): 243–247.

Williams, N. M., and Kremen, C. (2007) Resource distributions among habitats determine solitary bee offspring production in a mosaic landscape. *Ecological Applications* 17(3): 910–921.

Williams, N. M., Regetz, J., and Kremen, C. (2012) Landscape-scale resources promote colony growth but not reproductive performance of bumble bees. *Ecology* 93(5): 1049–1058.

Williams, P. H. P. H., and Osborne, J. L. (2009) Bumblebee vulnerability and conservation world-wide. *Apidologie* 40(3): 367–387.

Willmer, P. G., Bataw, A. A. M., and Hughes, J. P. (1994) The superiority of bumblebees to honeybees as pollinators: Insect visits to raspberry flowers. *Ecological Entomology* 19(3): 271–284.

Winfree, R., Aguilar, R., Vázquez, D. P., LeBuhn, G., and Aizen, M. A. (2009) A meta-analysis of bees' responses to anthropogenic disturbance. *Ecology* 90(8): 2068–76.

Wood, T. J., Holland, J. M., and Goulson, D. (2015) Pollinator-friendly management does not increase the diversity of farmland bees and wasps. *Biological Conservation* 187: 120–126.

Wood, T. J., Holland, J. M., Hughes, W. O. H., and Goulson, D. (2015) Targeted agri-environment schemes significantly improve the population size of common farmland bumblebee species. *Molecular Ecology* 24: 1668–1680.

Woodroffe, R. (1992) *Factors affecting reproductive success in the European badger, Meles meles L.* PhD Thesis, University of Oxford, Oxford.