

**Occurrence and population density of wild-living honey bees  
in Europe and the impact of different habitat types on their  
foraging and overwintering success**

Vorkommen und Populationsdichte von wild lebenden Honigbienen  
in Europa und die Auswirkungen unterschiedlicher Habitattypen  
auf ihr Sammelverhalten und den Überwinterungserfolg



Doctoral thesis for a doctoral degree  
at the Graduate School of Life Sciences,  
Julius-Maximilians-Universität Würzburg,  
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Würzburg, 2022



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“Holy Mother Earth,  
the trees and all nature,  
are witnesses of your  
thoughts and deeds.”

Winnebago’s wise saying



Winnebago family by Seth Eastman





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Due to their cryptic way of life high above the ground, wild-living honey bee colonies were so far not systematically studied in Europe. This thesis contains the first systematic surveys of wild-living colonies in Germany and Spain. Photo taken by Dimi Dumortier.

## SUMMARY

The original habitat of native European honey bees (*Apis mellifera*) is forest, but currently there is a lack of data about the occurrence of wild honey bee populations in Europe. Prior to being kept by humans in hives, honey bees nested as wild species in hollow trees in temperate forests. However, in the 20<sup>th</sup> century, intensification of silviculture and agriculture with accompanying losses of nesting sites and depletion of food resources caused population declines in Europe. When the varroa mite (*Varroa destructor*), an invasive ectoparasite from Asia, was introduced in the late 1970s, wild honey bees were thought to be eradicated in Europe. Nevertheless, sporadic, mostly anecdotal, reports from ornithologists or forest ecologists indicated that honey bee colonies still occupy European forest areas. In my thesis I hypothesize that near-natural deciduous forests may provide sufficient large networks of nesting sites representing refugia for wild-living honey bees. Using two special search techniques, i.e. the tracking of flight routes of honey bee foragers (the “beelining” method) and the inspection of known cavity trees, I collected for the first time data on the occurrence and density of wild-living honey bees in forest areas in Germany (CHAPTER 3). I found wild-living honey bee colonies in the Hainich national park at low densities in two succeeding years. In another forest region, I checked known habitat trees containing black woodpecker cavities for occupation by wild-living honey bee colonies. It turned out that honey bees regularly use these cavities and occur in similar densities in both studied forest regions, independent of the applied detection method. Extrapolating these densities to all German forest areas, I estimate several thousand wild-living colonies in Germany that potentially interact in different ways with the forest environment. I conclude that honey bees regularly colonize forest areas in Germany and that networks of mapped woodpecker cavities offer unique possibilities to study the ecology of wild-living honey bees over several years.

While their population status is ambiguous and the density of colonies low, the fact that honey bees can still be found in forests poses questions about food supply in forest environments. Consequently, I investigated the suitability of woodlands as a honey bee foraging habitat (CHAPTER 4). As their native habitat, forests are assumed to provide important pollen and nectar sources for honey bee colonies. However, resource supply

might be spatially and temporally restricted and landscape-scale studies in European forest regions are lacking. Therefore, I set up twelve honey bee colonies in observation hives at locations with varying degree of forest cover. Capitalizing on the unique communication behaviour, the waggle dance, I examined the foraging distances and habitat preferences of honey bees over almost an entire foraging season. Moreover, by connecting this decoded dance information with colony weight recordings, I could draw conclusions about the contribution of the different habitat types to honey yield. Foraging distances generally increased with the amount of forest in the surrounding landscape. Yet, forest cover did not have an effect on colony weight. Compared to expectations based on the proportions of different habitats in the surroundings, colonies foraged more frequently in cropland and grasslands than in deciduous and coniferous forests, especially in late summer when pollen foraging in the forest is most difficult. In contrast, colonies used forests for nectar/honeydew foraging in early summer during times of colony weight gain emphasizing forests as a temporarily significant source of carbohydrates. Importantly, my study shows that the ecological and economic value of managed forest as habitat for honey bees and other wild pollinators can be significantly increased by the continuous provision of floral resources, especially for pollen foraging.

The density of these wild-living honey bee colonies and their survival is driven by several factors that vary locally, making it crucial to compare results in different regions. Therefore, I investigated a wild-living honey bee population in Galicia in north-western Spain, where colonies were observed to reside in hollow electric poles (CHAPTER 5). The observed colony density only in these poles was almost twice as high as in German forest areas, suggesting generally more suitable resource conditions for the bees in Galicia. Based on morphometric analyses of their wing venation patterns, I assigned the colonies to the native evolutionary lineage (M-lineage) where the particularly threatened subspecies *Apis mellifera iberiensis* also belongs to. Averaged over two consecutive years, almost half of the colonies survived winter (23 out of 52). Interestingly, semi-natural areas both increased abundance and subsequent colony survival. Colonies surrounded by more semi-natural habitat (and therefore less intensive cropland) had an elevated overwintering probability, indicating that colonies need a certain amount of semi-natural habitat in the landscape to survive. Due to their ease of access these power poles in Galicia are, ideally suited to assess the population demography of wild-living Galician honey bee colonies through a long-term monitoring.

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In a nutshell, my thesis indicates that honey bees in Europe always existed in the wild. I performed the first survey of wild-living bee density yet done in Germany and Spain. My thesis identifies the landscape as a major factor that compromises winter survival and reports the first data on overwintering rates of wild-living honey bees in Europe. Besides, I established methods to efficiently detect wild-living honey bees in different habitat. While colonies can be found all over Europe, their survival and viability depend on unpolluted, flower rich habitats. The protection of near-natural habitat and of nesting sites is of paramount importance for the conservation of wild-living honey bees in Europe.



A black woodpecker cavity in the Swabian Alb that is occupied by a honey bee colony. In this region, around ten percent of the mapped black woodpecker cavities are dwelling places for honey bee colonies, offering the unique possibility to study them over several years. Photo taken by Ingo Arndt.

## ZUSAMMENFASSUNG

Das ursprüngliche Habitat der Westlichen Honigbiene (*Apis mellifera*) ist der Wald, doch derzeit fehlt es an Daten über das Vorkommen von wilden Honigbienenpopulationen in Europa. Bevor die Honigbiene von Menschen in künstlichen Behausungen gehalten wurde, nistete sie in den gemäßigten Breiten in hohlen Bäumen als wild lebende Art. Doch die Intensivierung der Forst- und Landwirtschaft, der damit einhergehende Verlust von Nistplätzen und die Verschlechterung der Nahrungsressourcen führten zu einem Rückgang der Honigbienenpopulationen im 20. Jahrhundert. Nachdem die Varroa-Milbe (*Varroa destructor*), ein invasiver Ektoparasit, in den späten 1970er-Jahren aus Asien eingeschleppt wurde, nahm man an, dass wilde Honigbienen in Europa ausgestorben seien. Nichtsdestotrotz gaben sporadische, hauptsächlich anekdotische Berichte von Ornithologen oder Waldökologen Anlass zur Vermutung, dass Honigbienenvölker immer noch in europäischen Wäldern zu finden seien.

In meiner vorliegenden Dissertation stelle ich die Hypothese auf, dass naturnahe Laubwälder ein ausreichend großes Netz von Nistplätzen bieten und als Zufluchtsorte für wild lebende Honigbienen fungieren können. Mit Hilfe zweier spezieller Suchtechniken – dem Nachverfolgen der Flugrouten von Honigbienen-Sammlerinnen (die ‚Bee-Lining‘-Methode) und der Inspektion bekannter Baumhöhlen – habe ich erstmalig Daten über das Vorkommen und die Populationsdichte von wild lebenden Honigbienen in deutschen Waldgebieten gesammelt (CHAPTER 3). In zwei aufeinanderfolgenden Jahren habe ich wild lebende Honigbienenvölker im Hainich Nationalpark entdeckt, wobei die Populationsdichten gering waren. In einem anderen Waldgebiet habe ich kartierte Habitatbäume mit Höhlen des Schwarzspechts auf ihre Besiedlung mit Honigbienenvölker hin überprüft. Es stellte sich heraus, dass Honigbienen diese Schwarzspechthöhlen regelmäßig nutzen und in ähnlich niedrigen Dichten in beiden untersuchten Waldgebieten vorkommen. Mittels Extrapolation schätze ich die Zahl der wild lebenden Bienenvölker in allen deutschen Waldgebieten auf mehrere Tausend, die auf vielfältige Weise mit der Waldumgebung interagieren können. Zusammenfassend zeigte sich, dass Honigbienen regelmäßig deutsche Waldgebiete bewohnen und dass Daten über kartierte Spechthöhlen



eine einmalige Möglichkeit bieten, die Ökologie der Honigbienen als Wildtier mittels eines Langzeitmonitorings zu untersuchen.

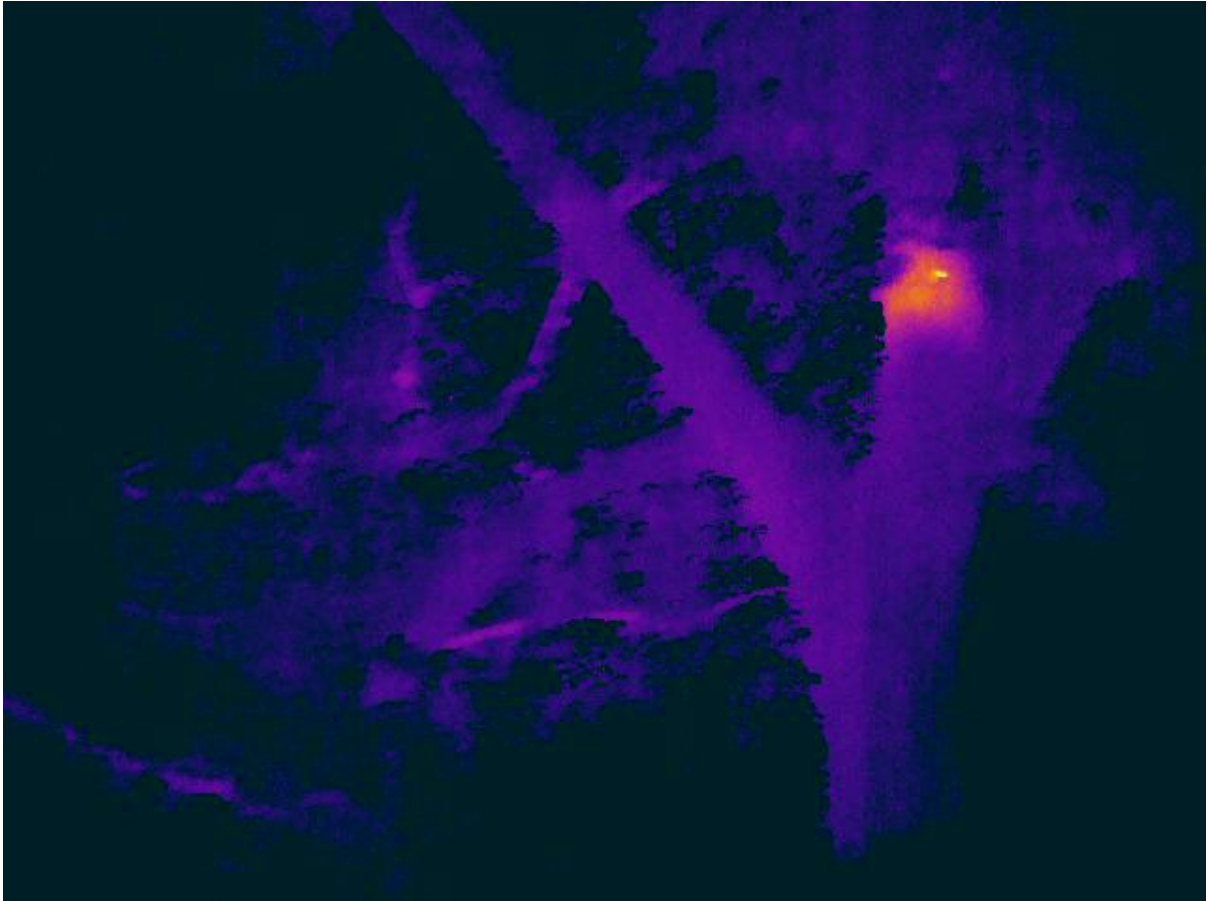
Auch wenn der Populationsstatus noch ungeklärt und die Populationsdichte gering ist, wirft die Existenz wild lebender Honigbienen Fragen bezüglich der Nahrungsversorgung im Wald auf. Folglich habe ich untersucht, ob eine ausreichende Futtersversorgung für Honigbienen in Wäldern gegeben ist (CHAPTER 4). Wälder gelten als der ursprüngliche Lebensraum der Westlichen Honigbiene und man nimmt an, dass sie wichtige Pollen- und Nektarquellen für Honigbienenvölker liefern. Das Nahrungsangebot könnte jedoch räumlich und zeitlich begrenzt sein, wobei hierzu bislang Studien in europäischen Waldregionen fehlen. Daher habe ich zwölf Honigbienenvölker in Beobachtungsstöcken, jeweils an Orten mit unterschiedlichem Waldanteil, aufgestellt. Indem ich mir das einzigartige Kommunikationsverhalten – den Schwänzeltanz – zu Nutzen machte, untersuchte ich Sammeldistanzen und Habitatpräferenzen von Honigbienen über fast eine ganze Bienensaison hinweg. Darüber hinaus konnte ich durch die Verknüpfung der entschlüsselten Tanzinformationen mit Gewichtsaufzeichnungen der Bienenvölker Rückschlüsse auf den Beitrag der verschiedenen Habitattypen zum Honigertrag der Völker ziehen. Die Entfernungen bei der Nahrungssuche nahmen grundsätzlich mit dem Waldanteil in der umgebenden Landschaft zu. Obwohl Bienenvölker, die tiefer im Wald stationiert waren, weiter fliegen mussten, war ihre Gewichtszunahme nicht reduziert. Im Vergleich zu den Erwartungen, die sich aus den flächenmäßigen Anteilen der verschiedenen Habitate in der Umgebung ergeben, sammelten die Völker häufiger in Acker- und Grasland als in Laub- und Nadelwald, wobei der Spätsommer die schwierigste Zeit für die Pollenversorgung im Wald war. Auf die Phase im Frühsommer von Mitte Mai bis Mitte Juli bezogen, in der die Völker an Gewicht zunahmen, wurde der Wald zum Sammeln für Nektar/Honigtau beinahe erwartungsgemäß genutzt. Das unterstreicht die Bedeutung des Waldes als wichtige Quelle für Kohlenhydrate während eines kurzen Zeitraums im Jahr. Meine Untersuchungen zeigen, dass der ökologische und ökonomische Wert von Wirtschaftswald als Lebensraum für Honigbienen und andere Bestäuber durch die kontinuierliche Versorgung von Blütenressourcen, insbesondere in Bezug auf Pollen, erheblich gesteigert werden kann.

Die Dichte wild lebender Honigbienenvölker und deren Überleben ist durch mehrere Faktoren bestimmt die lokal variieren, weshalb es äußerst wichtig ist, die Ergebnisse



hinsichtlich verschiedener Regionen zu vergleichen. Im Zuge dieser Arbeit habe ich daher zusätzlich noch eine wild lebende Honigbienenpopulation in Galicien im Nordwesten Spaniens untersucht, wo die Bienenvölker in hohlen Strommasten nisteten (CHAPTER 5). Die beobachtete Völkerdichte war allein in diesen Strommasten fast doppelt so hoch wie in deutschen Waldgebieten, was auf grundsätzlich geeignetere Bedingungen für Bienen in Galicien schließen lässt. Anhand morphometrischer Analysen der Flügeläderung habe ich die Bienenvölker der einheimischen Evolutionslinie (M-Linie) zugeordnet, zu der auch die besonders bedrohte Unterart *Apis mellifera iberiensis* gehört. In zwei aufeinander folgenden Jahren überlebte im Durchschnitt fast die Hälfte der Bienenvölker den Winter (23 von 52). Interessanterweise waren in naturnahen Gebieten sowohl die Häufigkeit als auch das Überleben der Bienenvölker höher. Kolonien, die von mehr naturnahen Lebensräumen (und damit weniger intensiv genutzten Ackerflächen) umgeben waren, wiesen eine höhere Überwinterungswahrscheinlichkeit auf, was darauf hindeutet, dass die Kolonien einen gewissen Anteil an naturnahem Lebensraum in der Landschaft zum Überleben benötigen. Diese Strommasten in Galicien sind aufgrund ihrer leichten Zugänglichkeit ideal geeignet, um die Populationsdemografie der dortigen wild lebenden Honigbienen durch ein Langzeit-Monitoring zu untersuchen.

Zusammenfassend lässt sich sagen, dass Honigbienen wohl ununterbrochen als wild lebende Spezies in Europa existierten. Im Zuge meiner Doktorarbeit habe ich die erste quantitative Untersuchung wild lebender Honigbienen in Deutschland und Spanien durchgeführt. Meinen Ergebnissen zufolge ist die Landschaft ein entscheidender Faktor, der das Winterüberleben beeinflusst. Zudem beinhaltet meine Arbeit die ersten Daten über Überwinterungsraten von wild lebenden Honigbienen in Europa. Weiters habe ich Methoden entwickelt, um wild lebende Honigbienen in verschiedenen Lebensräumen zuverlässig und schnell zu finden. Alle drei Studien meiner Dissertation betonen, wie wichtig es ist, naturnahe Gebiete für den Schutz von wild lebenden Honigbienen zu erhalten. Zwar sind wild lebende Bienenvölker überall in Europa zu finden, doch ihre Überlebensfähigkeit hängt von blütenreichen, nicht mit Pestiziden belasteten Lebensräumen ab. Der Schutz von Lebensräumen und Nistplätzen ist für die Erhaltung der wild lebenden Honigbienen in Europa von größter Bedeutung.



A thermal image that shows a honey bee colony in the Ringpark in Würzburg residing in a tree cavity. I took the picture in late autumn when the colony was still rearing brood, but outside temperature in the night was already low.

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## CHAPTER 1: GENERAL INTRODUCTION

### LACK OF DATA ABOUT WILD-LIVING HONEY BEE POPULATIONS IN EUROPE

Today, the Western honey bee *Apis mellifera* is commonly seen as a domesticated insect, and it is widely believed that wild-living<sup>1</sup> colonies went extinct in Europe (Moritz et al., 2007; De la Rúa et al., 2009; Meixner et al., 2010, 2015). Originally endemic to Eurasia and Africa (Ruttner, 1988; Whitfield et al., 2006; Han et al., 2012; Dogantzis et al., 2021), Western honey bees in Central Europe were already heavily influenced by humans since the Middle Ages (Crane, 1999), therefore exhibiting a dual nature as managed and wild species for a long time. Following the devastating global spread of the ectoparasitic mite *Varroa destructor* from Asia in the late 1970s (Anderson and Trueman, 2000), beekeepers in Europe treat their colonies several times a year with various acaricides to control mite populations. Hence, it is generally accepted that honey bees need human intervention to survive (Rosenkranz et al., 2010), contributing to the assumption that wild colonies do no longer exist in Europe (Moritz et al., 2007; De la Rúa et al., 2009; Meixner et al., 2010, 2015).

Contrary to many other pollinators, there is a lack of data about the presence of wild honey bee populations in Europe (Moritz et al., 2005; Dall’Olio and Garrido, 2021). In fact, the honey bee is currently listed as “data deficient” on the International Union for Conservation of Nature’s Red List for Threatened Species (Nieto et al., 2014), probably due to the fact that honey bees are considered exclusively as domesticated insects rather than wild species. That there is no knowledge about wild honey bees is all the more surprising, since global decline of pollinator populations has raised concerns about food security (Klein et al., 2007; Potts et al., 2010; Ollerton et al., 2011; Leonhardt et al., 2013; Breeze et al., 2014; Aizen et al., 2019) and consequently spurred considerable scientific attention and conservation efforts (Biesmeijer et al., 2006; Neumann and Carreck, 2010; Burkle et al., 2013; Dietemann et al., 2013; Dainese et al., 2019). Therefore, we urgently need an inventory on the occurrence of wild-living colonies in Europe. This would allow the assessment of the

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<sup>1</sup> In this thesis I use the term “wild-living” honey bees to describe colonies living outside of apiaries. This includes honey bee colonies that are truly “wild” (descended from colonies that never inhabited a beehive), “feral” (having swarmed from a beekeeper) and colonies whose population status is not yet clear.

status of remaining wild populations which is crucial for the conservation of honey bee diversity (De la Rúa et al., 2009).

#### THREATS TO HONEY BEE COLONIES AND THEIR LIMITING FACTORS

Nowadays, the Western honey bee comprises more than 30 subspecies (Fontana et al., 2018) and is the most frequent flower visitor in both agricultural and semi-natural habitats (Garibaldi et al., 2011; Henry and Rodet, 2018; Hung et al., 2018; Stanley et al., 2020). While the monetary importance for honey and wax production is small, the honey bee's value as a key pollinator aiding human society has been estimated to exceed 153 billion dollar (Gallai et al., 2009). To secure pollination in agriculturally productive regions, migratory beekeepers are paid to move their colonies to crop fields (Alger et al., 2018; Martínez-López et al., 2022). Furthermore, honey bee breeders ship queens and whole colonies across the globe. However, this global trade of bees leads to an exchange and spill-over of pathogens and parasites (Mutinelli, 2011; Beaurepaire et al., 2020). Also, it jeopardizes the genetic diversity of the honey bee (De la Rúa et al., 2009, 2013; Espregueira Themudo et al., 2020; Kükner et al., 2021; Tanasković et al., 2021, 2022 but see Parejo et al., 2020 or Harpur et al., 2012) with potentially detrimental effects on colonies that are regionally well adapted to climate, vegetation, pests, and pathogens (Michener, 1974; Strange et al., 2007; Büchler et al., 2014; Meixner et al., 2015; Tribe et al., 2017). The Varroa mite that switched host (from Eastern (*Apis cerana*) to Western honey bee colonies) with the help of migratory beekeepers is currently considered the major threat to apiculture (Dietemann et al., 2012; Mondet et al., 2020; Le Conte et al., 2020). Nevertheless, the worldwide distribution of *Apis mellifera*, large sums of research money spent, and widespread husbandry by beekeepers made the honey bee not only the best studied insect but also a species with low chances to go extinct (Ghazoul, 2005; Iwasaki and Hogendoorn, 2021; Colla, 2022). From a global perspective, the number of managed colonies even increased steadily (Aizen and Harder, 2009; Moritz and Erler, 2016; Mashilingi et al., 2022), mainly because of the introduction of *Apis mellifera* to Asia for industrial honey production (Pirk et al., 2017; Osterman et al., 2021). Conversely, wild-living colonies, often still consisting of threatened native subspecies, are indeed very much at risk and should receive more scientific attention.

## VARROA RESISTANT BEES DUE TO NATURAL SELECTION AND DIFFERENT LIVING CONDITIONS

Despite the global spread of Varroa (Martin et al., 2012; Wilfert et al., 2016), it has been stated that honey bee colonies are still abundant in large temperate woodlands in North America and in the Southern Ural (Seeley, 2007; Ilyasov et al., 2015). Also, there are reports of European populations of managed colonies naturally surviving Varroa infestation for several years without mite control (Rinderer et al., 2001; Le Conte et al., 2007; Locke and Fries, 2011; Locke et al., 2012; Kefuss et al., 2015; Locke, 2016; Oddie et al., 2017; Kruitwagen et al., 2017; Panziera et al., 2017; McMullan, 2018; Muñoz-Colmenero et al., 2020). These examples illustrate that colonies surviving without human intervention can evolve through natural selection (Neumann and Blacquièrè, 2017; Brosi et al., 2017; Blacquièrè et al., 2019; Seeley, 2019; van Alphen and Fernhout, 2020). Contrariwise, managed bees are being treated with organic acids or other acaricides which leads to resistant mites and reduces chances for bees and mites to adapt to one another (Milani, 1999; Fries and Camazine, 2001; Martin, 2004; Pettis, 2004; de Mattos et al., 2017; Schmid-Hempel, 2021). This is aggravated by breeding programs for *Apis mellifera* selecting specific behavioural traits that are favourable for honey production but ultimately might hamper the bee's fitness (Neumann and Blacquièrè, 2017). Hence, it is likely that besides the globally documented cases of honey bee colonies showing resistance mechanisms to Varroa in Africa and South America (Rosenkranz, 1999; Brettell and Martin, 2017; Martin et al., 2020; Grindrod and Martin, 2021), also in some European regions colonies might have been surviving in the wild.

Wild-living populations are under the same human-mediated pressure since they are in most places surrounded by managed populations (Jones, 2004; Jaffé et al., 2010; Requier et al., 2020) with which they hybridize and together form one biological population (Randi, 2008; Ropars et al., 2021). Even if there are no genetic differences, the disparities in living conditions of wild-living and managed honey bees could result in fitness differences. Technically, the life-history strategy of wild-living colonies makes them less vulnerable to parasites than beekeepers' colonies (Bailey, 1958; Goodwin et al., 1994; Seeley and Smith, 2015; Loftus et al., 2016; Seeley, 2019). Naturally nesting colonies often stay smaller, rear less brood and swarm frequently, all of which reduces the reproductive potential of the Varroa mites in the brood (Loftus et al., 2016). Beekeepers, however, usually prevent swarming and provide unnaturally spacious hives, causing large colonies with continuous

brood rearing activity where the brood parasite proliferates. Furthermore, crowded colonies in apiaries next to each other favour drifting behaviour of returning foragers and the subsequent spread of diseases between colonies at the same location (Frey and Rosenkranz, 2014; Forfert et al., 2015; Seeley and Smith, 2015; Nolan and Delaplane, 2017; Peck and Seeley, 2019; but see Peck et al., 2016 for floral transmission of *Varroa* mites). In a nutshell, both subjection to natural selection and different living conditions potentially lead to more resilient bees that can survive in the wild. However, first and foremost the bees need suitable dwelling places and ample food resources.

#### THE SUITABILITY OF FORESTS AS A HABITAT FOR WILD-LIVING HONEY BEES IS UNKNOWN

While wild pollinators face multiple threats including agricultural intensification with heavy pesticide use or novel parasites and pathogens, they primarily suffer from human-induced landscape changes and loss of flower-rich habitats (Steffan-Dewenter, 2003; Naug, 2009; Winfree et al., 2009; Le Féon et al., 2010; Kennedy et al., 2013; Vanbergen and Initiative, 2013; Tonietto and Larkin, 2018; Dolezal et al., 2019). It is conceivable that forests, the original habitat of the Western honey bee, still provide a sufficient large network of cavities for viable populations of wild-living honey bees. But beside nesting sites, colonies need food resources throughout the year to prevail (Schellhorn et al., 2015). In fact, pioneering studies of a feral honey bee population in the Arnot forest in the US showed that colony mortality is especially high in the first year (i.e. for founder colonies) (Seeley, 1978, 1985, 2017), establishing nutrition as a crucial factor that limits the survival of wild-living colonies. Regarding European forests, it is unclear, how much the forage situation changed through intensification of forest management in the last centuries. For example it was shown that reduction of game and absence of light in today's forests hinders the establishment of an otherwise flower-rich understory (Proctor et al., 2012; Williams and Winfree, 2013; Eckerter et al., 2019; Braun-Reichert et al., 2021) thus shifting species composition towards more shade-tolerant and nutrient-demanding species (Verheyen et al., 2012). Therefore, resource supply by forests in Europe might be spatially and temporally restricted.

Fortunately, honey bees offer researchers unique insights into their food choices via the famous waggle dance used to inform nestmates about locations of profitable food sources (Frisch et al., 1967). Their communication signal can be decoded and thus represents real-

time information on how colonies allocate foragers across a landscape (Visscher and Seeley, 1982; Waddington, 1982; Steffan-Dewenter and Kuhn, 2003; Young et al., 2021). This presents the possibility to study honey bee food supply and foraging patterns in different landscapes and to identify temporal shortages that might reduce honey bee abundances (Di Pasquale et al., 2016; Requier et al., 2017). However, most studies on honey bee foraging inferred by waggle dance decoding have focused on crops, rural or urban lands (e.g. Steffan-Dewenter and Kuhn, 2003; Couvillon et al., 2014; Garbuzov et al., 2015; Danner et al., 2016; Sponsler et al., 2017; Bänsch et al., 2020; Samuelson et al., 2021). Landscape-scale studies which investigate the suitability of Central European forests as foraging habitat for potential wild-living honey bees and managed colonies are currently lacking.

#### THIS THESIS

Conducting the first systematic survey of wild-living colonies in Europe and studying the impact of different habitat types on their foraging and overwintering success were the main objectives of my thesis. By investigating wild-living honey bees in Germany and Spain, this thesis will set the groundwork for future studies exploring the lives of honey bees in the wild and will present ways to foster them through improved foraging conditions in forest landscapes.





The author of this thesis inspects a wild-living honey bee colony in a black woodpecker cavity. Photo taken by Ingo Arndt.



## CHAPTER 2: OBJECTIVES AND METHODS

My PhD research represents the first systematic investigation of wild-living colonies in Europe, thus detection methods had to be tested and adapted to European forests, especially because of the dense beech forest canopy. As it is known that honey bees prefer cavities far from the ground (Seeley and Morse, 1978; Seeley, 2019), the discovery of honey bee nests in the forest by chance is very unlikely. For a census in Germany, I therefore applied “beelining”, a specific craft, that was proposed and used as a research technique for pioneering studies of feral honey bee colonies in North America (Visscher and Seeley, 1989; Wenner et al., 1992) (CHAPTER 3). Beelining relies on the bees’ habit of returning in a straight line to the hive from a foraging spot and is applicable in any type of habitat.

Anecdotal reports indicated that honey bees use black woodpecker cavities (Meyer and Meyer, 2001; Sikora, 2008; Sikora et al., 2016) and hollow electric poles (A. Machado, personal communication) as nesting sites. Hence, I examined a multitude of these black woodpecker cavities in a forested region in Germany (CHAPTER 3) and hollow electric power poles in a mixed agricultural landscape in Spain (CHAPTER 5). The choice of the two regions was an opportunistic one, as in both study areas wild-living honey bees have been encountered occasionally. The fact that honey bee regularly used these natural and anthropogenic cavities makes them interesting for researching population demography parameters of wild-living colonies via long-term monitoring projects.

The suitability of European forest landscapes for wild-living honey bees is mainly unknown. To assess food preferences of honey bees in forest landscapes, I decoded waggle dances that contain filtered information about the distance and direction of attractive food resources and complemented this information with colony weight recordings (CHAPTER 4). While honey bee waggle dance decoding is a rapidly growing field of research with increasing applications in ecological studies on landscape-scale, the combination of honey bee habitat preferences inferred by decoded dances and weight recordings is to my knowledge novel. It enabled me to directly infer the contribution of different habitat types to the colonies’ honey yield, thereby posing interesting perspectives for future ecological studies with honey bee colonies in other landscapes.

The aims of my thesis were to evaluate whether honey bee colonies still occur in German forest areas and to investigate the temporal and spatial foraging patterns of such wild-living colonies in forest landscapes (CHAPTERS 3 & CHAPTER 4). Moreover, I determined densities and survival rates of native honey bee colonies living wildly in hollow power poles in Galicia, Spain (CHAPTER 5). In the following I give a short description of the objectives and methods of the studies included in this thesis.

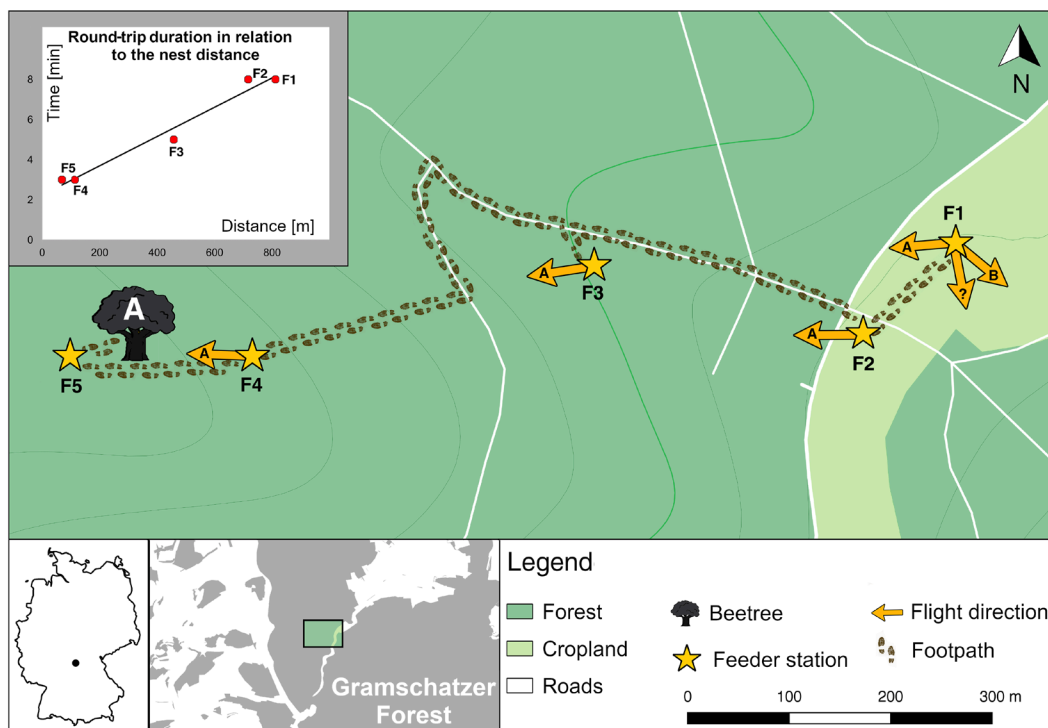
### *CHAPTER 3: OCCURRENCE OF WILD-LIVING HONEY BEE COLONIES IN GERMAN FORESTS*

In this chapter I describe the attempts to determine whether colonies still inhabit German forest areas and if so, at what densities. Due to their protection status, near-natural forests exhibit significantly more nesting cavities than managed forests and could potentially provide a network of nest sites, large enough for wild-living honey bee populations to persist. As the density of honey bee colonies in temperate woodlands might naturally be very low and nests extremely cryptic, I used two specific searching techniques to localize colonies: the tracing of nest sites based on forager flight routes (“beelining”), and the direct inspection of trees with black woodpecker cavities.

I performed beelining in the Hainich national park in Central Germany where beekeeping inside the park boundaries is forbidden. While the beelining method was described in the context of honey hunting in Hungarian beech forests (Gunda, 1968), it has not been used for scientific purpose in Europe yet. Independent of the cavity type used by the bees and whether cavity entrance of the colony is visible for humans, beelining represents an unbiased method for the detection of wild-living colonies in any habitat from scratch. The method takes advantage of the bees foraging site-fidelity and their recruiting behaviour to attractive food sources when forage is scarce. The workflow is the following: Some foraging honey bees are caught on flowers with a special box (the beelining box) and fed with a high-concentrated, scented sugar solution that ensures rapid recruitment. After a few round trips, the bees use straight flight paths (so called “beelines”) between the nest and the feeding location. Using paint marks on the bees’ thoraxes and timing their round-trip times, the distance between feeder and hive can be estimated. The minimum round-trip duration of the fastest individuals is translated into a maximum distance to the honey bee colony by the following formula:  $distance (m) = 243 \times t (min) - 627.75$ , where  $t$  is the round-trip time of the fastest bees (Seeley, 2016).

By visually determining the direction of their homeward flights, the route to the bees' nest can be obtained and subsequently the feeder can be moved step-by-step nearer to the colony until the colony is spotted or the location is narrowed down to a very small area (see Fig. 1 for an exemplary bee hunt in the Gramschatzer forest near Würzburg or Fig. S2 in CHAPTER 3).

Usually, the actual density of wild-living colonies is likely to be higher compared with the density inferred by beelining. This has two reasons: Firstly, only colonies can be found whose foragers are caught on flowers at the sampling locations. Secondly, if the nest cavities cannot be found (as it was the case in the Hainich national park (CHAPTER 3)), colonies in the vicinity of each other cannot be distinguished. Therefore, the density of honey bee colonies inferred by beelining must be regarded as a lower limit. A short videoclip displaying a bee hunt in Germany can be watched here: <https://www.swrfernsehen.de/natuerlich/sensation-wilde-honigbienen-im-wald-entdeckt-100.html>



**Figure 1:** Exemplary bee hunt in the Gramschatzer forest near Würzburg. At the first feeder station (F1) three distinct flight directions of bees caught on pumpkin flowers could be observed (“A”, “B” and “?”). Direction A was pursued, and round-trip times and directions were recorded for the feeders F1-F5. Finally, the colony was found in a tree cavity (beetree A). During another bee hunt a couple of weeks later we found another colony in the direction B. The subfigure in the upper left corner shows the empirically determined relationship between the distances between feeder and nest and the round-trip times.

To simulate realistic dispersal events from managed hives and determine dispersal distance, I set up swarms in three locations around the Hainich forest (see Fig. 2) and decoded their nest site dances to study how far swarms from beekeeper-managed hives would potentially move into the forest. These dispersal distances can be compared to the distances from the detected colonies to the nearest beekeepers to assess whether the colonization in one dispersal event was likely or not.



**Figure 2:** Swarm observation mount to assess dispersal distances of a honey bee colony.

The fact that there was no beekeeping activity allowed inside the park boundaries and that nesting cavities are more abundant in comparison to managed forests made the national park ideally suited for a first census of colonies living in the wild. However, beelining is a tedious process, and it usually takes several days to locate a colony. Therefore, I examined

another, more efficient, technique, that builds on the knowledge of tree cavities in a forest area.

Cavity trees are in some parts of Germany protected by the law, and it was reported that social insects occasionally use woodpecker cavities as nest sites (Johnsson et al., 1993; Meyer and Meyer, 2001; Sikora, 2008; Sikora et al., 2016). Protected woodpecker cavities are due to the GPS coordinates easy to locate and represent some of the most spacious cavities in managed forests. To test whether black woodpecker cavities are used by honey bees and whether they can be utilized for a long-term monitoring of honey bee colonies, I inspected a subset (N=98) of the known black woodpecker cavities in the biosphere reserve Swabian Alb in Southern Germany with binoculars from the ground during weather conditions suitable for honey bee foraging.

The discovery of wild-living honey bees in German forests is described in CHAPTER 3. While the results were intriguing and opened a new field of research, they also pointed out the urgent need to determine the suitability of European forests for honey bee colonies in terms of food supply (CHAPTER 4).

#### *CHAPTER 4: HONEY BEE FORAGING IN CENTRAL EUROPEAN FOREST LANDSCAPES*

Wild-living honey bee colonies still regularly occur in German forests, even though at low densities. One reason for these low densities might be insufficient or temporally restricted food supply in forest areas. To explore this, I set up twelve honey bee colonies in the Steigerwald (Lower Franconia) in locations with varying degree of forest cover. Capitalizing on the unique communication behaviour, the waggle dance, where collecting bees communicate the locations of profitable food sources to nestmates (von Frisch, 1967), I investigated honey bee foraging in the forest during almost a whole season, simulating wild-living colonies as closely as possible. Filming of honey bee waggle dances was facilitated by a glass window on one side of the observation hive box. Later, I screened the recorded movies of an observation day for each colony in such a way that the sampled waggle dances were dispersed over the entire videorecorded period.

As it was shown that honey bee colonies exhibit dance dialects between different species (Lindauer, 1956; Puchihiwewa et al., 1985; Kohl et al., 2020), between different subspecies (Gould, 1982) and in different terrain (Tautz et al., 2004; George et al., 2021), we determined the increase of waggle dance duration with increasing foraging distance in the

study region by a feeder experiment and trained the bees to locations with known distances in the forest (Kohl and Rutschmann, 2021). By decoding dances of marked individuals one can obtain a calibration curve to infer natural foraging locations from waggle dances.

The following two formulas (Kohl and Rutschmann, 2021, Arra, Rutschmann, Kohl, unpublished data) were used to translate the mean circuit duration  $t$  per dance (in seconds) into distance ( $d$ , in meters) depending on the value of  $t$ :

- $t \leq 4.439852$ :  $d = \ln(-0.2253179 * t + 1.3305467) * (-1534.086) - 84.98$
- $t > 4.439852$ :  $d = 1242.854 * t - 3903.066$

Further, as dance decoding is afflicted by several types of noise (Schürch et al., 2013, 2016; Tautz, 2022), we measured the spatial deviation from the actual location during the feeder experiment and used the data to produce an uncertainty buffer for each inferred dance location for natural food sources.

Honey bee colonies regulate their foraging activity in order to maximize the ratio of energy gained versus energy spent per foraging trip and will always fly as short as possible (Seeley et al., 1991; Seeley, 1994). Therefore, the distance at which honey bees forage can be seen as a proxy for forage availability: near advertised distances signal abundant forage in the surrounding, longer foraging distance can be interpreted as periods of food scarcity (Couvillon et al., 2014; Garbuzov et al., 2015, 2020). Furthermore, when considering both distance and direction signal of waggle dances, one can infer the approximate foraging location and make geographic maps of the bees' feeding sites to study their habitat preferences and determine whether forests were used at the expected level, given the proportional occurrence in the landscape (calculated from Weigand et al., 2020). Additionally, I recorded colony weight changes with weight scales. The setup along the forest gradient allowed me to investigate whether colonies deep inside the forest accumulated more or less weight (which is mainly correlated to the nectar influx of a colony (McLellan, 1977; Meikle et al., 2008)) than colonies at the forest edge. The simultaneous use of habitat preferences and colony weight changes permitted me to determine the contribution of different habitat types to colony weight gain and subsequent honey yield.

While most dance decoding studies so far compared proportional habitat use with the amount of different landcover types inside a certain circular buffer area (mostly 2 km radius e.g. Steffan-Dewenter and Kuhn, 2003; Danner et al., 2016), I established an empirical

visitation probability of a foraging patch depending on the distance between the foraging patch and the colony. For each patch, the area was multiplied by the visitation probability. Then, the values for each of the different land cover types were summed up over all patches and divided by the total (to normalize it). In this way, I evaluated the landscape according to the surrounding habitats and its distance to the colony, without overestimating patches in the near surrounding of the colony (Supplementary material of CHAPTER 4).

My specific objectives of CHAPTER 4 were the following:

- Does the proportion of forest cover surrounding a colony affect the colony's foraging distances?
- Do honey bees use woodland at the expected level?
- What is the contribution of different habitat types to colony weight gain and does forest cover have an influence on it?

Understanding and comparing the drivers of the wild-living honey bee colony density in different regions and habitat types will help to answer the question how frequent honey bees would naturally be and what determines viability of wild-living honey bee populations. Hence, I investigated a mixed, agricultural landscape in Galicia, Spain for the occurrence of wild-living honey bees.

#### CHAPTER 5: WILD-LIVING HONEY BEES IN AN AGRICULTURAL LANDSCAPE IN SPAIN

On the Iberian Peninsula the native honey bee population is still mostly free of introgression, and little affected by selective breeding (Cánovas et al., 2011; Chávez-Galarza et al., 2015, 2017). There were anecdotal reports of bees residing in hollow power poles in Galicia, Spain (A. Machado, personal communication). In a first step, I surveyed an area of 136 km<sup>2</sup> surrounding the city of Xinzo de Limia in the province Ourense, Galicia for hollow power poles by driving all major roads with a car (Fig. S2 in CHAPTER 5). Thus, I inspected this pool of 214 hollow power poles from the ground with binoculars looking for poles occupied by bees. Pollen import or regular directional flight traffic of honey bees were indicators of an active honey bee nest. To determine the winter survival rate, I re-examined all power poles that were occupied in autumn (October) again in spring (March). To assess the influence of the surrounding land, I furthermore quantified the proportions of different habitat types around every pole (calculated from Pflugmacher et al., 2019). As 80

% of honey bee foraging occurs within two kilometres (Danner et al., 2014, 2017, own data CHAPTER 4), I chose this radius for the landscape classification. A principal component analysis (PCA) showed that cropland and semi-natural areas are inversely correlated to each other in the study region, thereby representing a land-use intensity gradient from intensive agricultural land to near-natural shrubland and forests.

I set out to answer the following questions:

- What is the density of honey bee colonies living in power poles in Galicia?
- Is the occupation rate of poles higher in locations with larger proportions of semi-natural habitat?
- What is the winter survival of the colonies and does the amount of semi-natural habitat influence it?

To determine whether the colonies residing in the electric power poles are members of the native evolutionary lineage or likely to descend from imported honey bee stocks by beekeepers, I took bee samples and utilized morphological identification via wing venation (Oleksa and Tofilski, 2015). From the sampled colonies I prepared the right forewings of ten workers per colony between two glass slides and took digital images. Then I used the freely available software identify 1.3 (Nawrocka et al., 2017) to measure 19 landmarks that I manually placed on each image. This assessment of the evolutionary lineage is important as locally adapted colonies showed better performance than imported stocks in different regions in Europe (Büchler et al., 2014; Meixner et al., 2015) and native subspecies are crucial for the conservation of honey bee's genetic diversity in Europe (De la Rúa et al., 2009; Oleksa et al., 2013b).

Further, specific study sites, bee keeping procedures, materials and statistical analyses are specific to the individual experiments and are at large described in the respective chapters (CHAPTER 3, CHAPTER 4 & CHAPTER 5). Each of the following three chapters is an independent manuscript that is either already published (CHAPTER 3 & CHAPTER 4), or that is submitted to a peer-reviewed journal (CHAPTER 5). Spelling of all manuscripts was adapted to English orthography. Honey bees are true bees, and as such I spelled them as two words throughout the whole thesis. Data and scripts that have not been published yet are available from <https://github.com/brutschmann/thesis>.







Beelining relies on the tracing of nest sites based on forager flight routes. It represents an unbiased method for the detection of wild-living colonies in any habitat from scratch. The picture shows a wooden beelining box with a honey bee comb filled with a high concentrated, scented sugar solution. Two bees are arriving at the feeder, two nestmates are already filling their honey stomach on the comb. Photo taken by Ingo Arndt.

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## CHAPTER 3: OCCURRENCE OF WILD-LIVING HONEY BEE COLONIES IN GERMAN FORESTS

This chapter is published as:

Kohl PL & **Rutschmann B** (2018) The neglected bee trees: European beech forests as a home for feral honey bee colonies. *PeerJ* 6:e4602.

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<https://doi.org/10.7717/peerj.4602>

### Abstract

It is a common belief that feral honey bee colonies (*Apis mellifera* L.) were eradicated in Europe through the loss of habitats, domestication by man and spread of pathogens and parasites. Interestingly, no scientific data are available, neither about the past nor the present status of naturally nesting honey bee colonies. We expected near-natural beech (*Fagus sylvatica* L.) forests to provide enough suitable nest sites to be a home for feral honey bee colonies in Europe. Here, we made a first assessment of their occurrence and density in two German woodland areas based on two methods, the tracing of nest sites based on forager flight routes (beelining technique), and the direct inspection of potential cavity trees. Further, we established experimental swarms at forest edges and decoded dances for nest sites performed by scout bees in order to study how far swarms from beekeeper-managed hives would potentially move into a forest. We found that feral honey bee colonies regularly inhabit tree cavities in near-natural beech forests at densities of at least 0.11–0.14 colonies/km<sup>2</sup>. Colonies were not confined to the forest edges; they were also living deep inside the forests. We estimated a median distance of 2,600 m from the bee trees to the next apiaries, while scout bees in experimental swarms communicated nest sites in close distances (median: 470 m). We extrapolate that there are several thousand feral honey bee colonies in German woodlands. These have to be taken in account when assessing the role

of forest areas in providing pollination services to the surrounding land, and their occurrence has implications for the species' perception among researchers, beekeepers, and conservationists. This study provides a starting point for investigating the life-histories and the ecological interactions of honey bees in temperate European forest environments.

### **Introduction**

The habitat of the western honey bee (*Apis mellifera* L.) in temperate Europe was deciduous or mixed forest, with trees providing both nest cavities and food resources (Seeley and Morse, 1976; Seeley, 1985; Ruttner, 1988; Crane, 1999). However, since the Middle Ages, the life of honey bees in Central Europe has considerably been affected by man. When people started to take colonies to their homes and manage them in beekeeping hives, the habitat of honey bees shifted from woodland areas to the agro-urban space (Crane, 1999; Banaszak, 2009). The logging of old-grown forests and an increase in land-use intensity further reduced the habitat for wild honey bee colonies (De la Rúa et al., 2009; Küster, 2010; Potts et al., 2010). Advances in beekeeping technique allowed for a better control over honey bee reproduction, altering their life-history and limiting natural selection (Loftus et al., 2016; Neumann and Blacquièrre, 2017; Brosi et al., 2017). Lastly, the trade in selected honey bee strains by bee breeders across Europe has led to genetic admixture and spread of pathogens and invasive parasites (De la Rúa et al., 2009; Mutinelli, 2011) challenging the health of these important pollinators (Neumann and Carreck, 2010; Meixner et al., 2015). Today, among the most harmful pests is the ectoparasitic mite *Varroa destructor* (Anderson and Trueman, 2000), which is infecting virtually all honey bee colonies across Europe (Le Conte et al., 2010; Rosenkranz et al., 2010; Nazzi and Le Conte, 2016). There is consensus among researchers and beekeepers that colonies not treated against the mite will usually die within a few years due to both direct damage and associated virus infections (Sammataro et al., 2000; Moritz et al., 2010; Neumann and Carreck, 2010; Rosenkranz et al., 2010; Martin et al., 2012; van Dooremalen et al., 2012). The question is: what does that mean for feral honey bee colonies, which nest naturally with no human interference? It is widely assumed that since the arrival of the mite, feral honey bee colonies were all but wiped out (Moritz et al., 2007; De la Rúa et al., 2009; Meixner et al., 2015). However, it has been reported that naturally nesting honey bees colonies are still abundant in large temperate woodlands in North America and the Southern Ural (Seeley, 2007; Ilyasov et al., 2015), and they regularly occupy cavity trees along rural avenues in Northern

Poland (Oleksa et al., 2013b). There are two reasons why we should reconsider the common perception about the status of honey bee colonies living wildly in Central Europe: different host-parasite ecology, and a simple lack of data.

In predicting the fate of feral colonies, we usually consult knowledge about honey bee-parasite relationships that was gathered in the context of managed apiaries. It is long recognized, however, that feral colonies generally experience a reduced parasite pressure due to ecological factors (Bailey, 1958). The crowding of honey bee colonies at bee yards increases the risk of erroneous homing by workers (drifting), which boosts epidemic spread among hives and individual colonies' parasite loads (Fries and Camazine, 2001; Forfert et al., 2015; Seeley and Smith, 2015). Parasite loads were also found to be positively correlated with the colony density at the landscape scale (Frey and Rosenkranz, 2014; Forfert et al., 2016; Nolan and Delaplane, 2017). Because people tend to have their bee yards near settlements, beekeeping activity is not distributed evenly across space, resulting in high local colony densities. A study on parasite pressure of honey bees along an urbanization and management gradient in the Eastern United States, indeed found that parasite burden was both higher in urban as compared to rural locations, and in managed as compared to feral hives (Youngsteadt et al., 2015). In contrast, worker bees of feral colonies in the UK had higher virus infestations than workers of managed colonies (Thompson et al., 2014). However, the sampling was potentially biased towards locations in urban areas with high overall colony densities. The density of naturally nesting colonies in remote woodlands is low and colonies nest individually in widely separated tree cavities (Visscher and Seeley, 1982; Seeley et al., 2015; Seeley, 2017). In addition, the life-history strategy of feral colonies makes them less vulnerable to parasites than beekeepers' colonies. Naturally nesting colonies stay smaller, rear less brood and swarm frequently, all of which reduces the reproductive potential of *Varroa* mites. Beekeepers, however, usually prevent swarming and provide unnaturally spacious hives, resulting in large colonies with continuous brood rearing activity (Loftus et al., 2016; Seeley, 2017). Both empirical and theoretical studies suggest that these factors alone – the spacing of colonies, and their life-history – can reduce the infection levels of *V. destructor* considerably (Loftus et al., 2016; DeGrandi-Hoffman et al., 2017; Seeley, 2017). Further, it is predicted that under certain conditions, populations of honey bee colonies not treated against parasites will evolve resistance mechanisms against these pests through natural selection (Neumann and Blacquière, 2017). Selection for resistance against *Varroa* was indeed documented in

several studies of both closed and open managed populations that did not receive treatment against the mites (Rinderer et al., 2001; Le Conte et al., 2007; Locke and Fries, 2011; Locke et al., 2012; Kefuss et al., 2015; Locke, 2016; Oddie et al., 2017). So far, however, only one population of feral colonies has been studied with respect to its survival and host-parasite ecology: although the new parasite and its associated viruses evidently led to an initial drop in the population sizes of feral honey bee colonies in North America (Kraus and Page, 1995; Mikheyev et al., 2015), a population of honey bees living in the deciduous forests of New York State was found to be stable over decades (Seeley, 2007; Mikheyev et al., 2015; Seeley et al., 2015; Seeley, 2017). This population exhibits genomic signatures of selection after the arrival of *Varroa* (Mikheyev et al., 2015) and individual bees were found to show intense hygienic and biting behaviour against the mites (D. Peck & T. D. Seeley, 2018, personal communication). Interestingly however, a high winter mortality of founder colonies in new nests suggests that the population is more limited by food supply than by parasites (Seeley, 1978, 1985, 2017).

The fact that we owe our knowledge about forest dwelling temperate honey bees almost exclusively to a feral population outside its natural range, from the deciduous forests of New York State (Seeley, 1985), points to the second problem: there are actually no scientific data available about the ecology of feral honey bee colonies in Europe, neither before nor after the arrival of the *Varroa* mite (Geldmann and González-Varo, 2018). It is often quoted that feral honey bee colonies have always been rare (Ruttner, 1973), but there is no information about their actual abundance and distribution at earlier times. Further, the density of honey bee colonies in temperate woodlands might naturally be very low and nests are extremely cryptic (Seeley and Morse, 1976; Visscher and Seeley, 1989): discovering a honey bee nest by chance and without using specific searching techniques is hence very unlikely. To overcome the difficulty in directly detecting honey bee nests, genotyping of sampled drones or queen offspring with microsatellite markers was used to estimate honey bee colony densities in different regions across their native range (Moritz et al., 2007, 2008; Jaffé et al., 2010). At sample sites in Europe, inferred colony densities resembled those of known managed colonies, so it was concluded that wild or feral honey bees do not contribute to the population as a whole (Moritz et al., 2007; Jaffé et al., 2010). However, it is probable that the resolution of the method is not precise enough to detect low densities of feral colonies in regions where many more colonies live in managed hives (Arundel et al., 2012, 2013).

Getting precise information about the presence of feral honey bee colonies is important: they need to be considered when estimating overall wild pollinator abundances or when assessing the role of forest areas in providing pollination services to the surrounding land (Tschardt et al., 2005; Jaffé et al., 2010; Mitchell et al., 2014; Decocq et al., 2016). The occurrence of feral honey bee colonies in woodlands would also have special implications for the species itself, for example its legal protection status and its perception among bee researchers, forest ecologists and conservationists. Although feral honey bee colonies can be a rich source for studying the natural interaction of honey bees with the forest environment (Seeley, 1985) and they can represent an important reservoir of genetic diversity (Oleksa et al., 2013b), so far, they have got little attention in Europe.

In most parts of Central Europe, beech dominated forests (*Fagus sylvatica* L.) represent the major type of natural vegetation (Bohn et al., 2003; Tinner and Lotter, 2006; Bolte et al., 2007), so that increasing efforts are being devoted to the assessment of biodiversity in these forests and to their conservation (Brunet et al., 2010; Scherfose et al., 2007). Little is known about the role of beech forests as a home for pollinating insects (Sobek et al., 2009). At first glance, they actually appear to provide limited nectar resources: beech trees are wind pollinated, and mature stands form dense canopies, which limit light transmission to the forest floor and hence the growth of shrubs and other tree species (Packham et al., 2012). However, the wild bee fauna of the Hainich, an ancient beech forest in central Germany, was found to be quite diverse (Sobek et al., 2009). Further, there are historical reports about people hunting for honey in the Carpathians, a region where today's last primeval beech forests are situated (Gunda, 1968), and honey bees were repeatedly reported to occupy old black woodpecker cavities (*Dryocopus martius* L.) in beech trees (Meyer and Meyer, 2001; Sikora, 2008; Sikora et al., 2016).

We assumed that near-natural deciduous forests in Germany could provide enough suitable nest sites to be a home for feral honey bee colonies. Here, we made a first assessment of their occurrence and density by using two methods, the tracing of nest sites based on forager flight routes (beelining technique), and the direct inspection of potential cavity trees. Further, we established experimental swarms at forest edges and decoded dances for nest sites performed by scout bees in order to study how far swarms from beekeeper-managed hives would potentially move into a forest.

## Methods

### *Study sites*

We conducted our study in two different beech forest areas in Germany, the Hainich and the forests of the Swabian Alb.

### *Hainich*

The Hainich forest in central Thuringia (51.08, 10.43) is one of the largest non-fragmented stretches of deciduous forest in Central Europe. It lies at about 400 m a.s.l. and its climate is characterized by a mean annual temperature of 7.5 °C and a mean annual precipitation of 630 mm (Gauer and Aldinger, 2005). Since 1964, its southern part served as a military training ground, and was therefore not disturbed much over the last 60 years. Seven thousand six hundred ha from a total wooded area of 16,000 ha were designated National Park in 1997 and became a World Heritage Site as part of the primeval beech forests of the Carpathians and other regions in Europe (Knapp et al., 2007). The dominant tree species are beech (*F. sylvatica* L., 65 %), ash (*Fraxinus excelsior* L., 25 %) and maple (*Acer pseudoplatanus* L., *Acer platanoides* L. and *Acer campestre* L., 7 %) in an uneven age distribution (1–250 years) (Kutsch et al., 2010). Beside these, there are also lime trees (*Tilia cordata* Mill. and *Tilia platyphyllos* Scop.) which, like maple trees, are rich nectar and pollen sources for honey bees and other pollinating insects (Tofilski and Oleksa, 2013). Due to the regulations of the authorities it is not allowed to keep managed honey bee colonies inside the National Park.

### *Swabian Alb*

The Biosphere Reserve Swabian Alb is a characteristic, low-mountain cultural landscape in the southwest of Germany (48.41, 9.54). While the foothills at about 300 m a.s.l. have a mild climate with an annual mean temperature of 8 °C, the Alb plateau at about 800 m a.s.l. is considerably cooler with an annual mean temperature of 6.7 °C. Precipitation amounts to 750–1,050 mm annually. The Biosphere Reserve, which encompasses 85,000 ha around the former army training ground Münsingen, was formed in 2008 and designated as UNESCO Biosphere Reserve in May 2009. Large areas of the reserve consist of extensively used species-rich meadows, juniper heaths and fruit orchards, but there are also richly structured forests with many hollow trees that serve as shelter for birds, small mammals, bats, and insects. The dominating tree species is beech (*F. sylvatica* L.), but ash (*F.*



*excelsior* L.), maple (*A. pseudoplatanus* L.), elm (*Ulmus glabra* Huds.), lime (*T. cordata* Mill. and *T. platyphyllos* Scop.), hornbeam (*Carpinus betulus* L.), fir (*Picea abies* (L.), H. Karst) and spruce (*Abies alba* Mill.) are also common (Jooß, 2013).

#### *Occurrence and density of feral honey bee colonies in the Hainich forest*

The fact that foraging honey bees have high site fidelity and recruit nest mates to rich food sources can be used to trace feral colonies when their nest sites are difficult to find (Visscher and Seeley, 1989; Wenner et al., 1992; Seeley, 2016). “Beelining,” the tracking of honey bees based on their flight paths is actually an ancient method, which has been tested and proven by honey hunters for centuries (Gunda, 1968; Crane, 1999).

In September 2016, June 2017, and September 2017, we spent a total of 10 days searching for feral honey bee colonies in the Hainich forest. We worked during dearth seasons, when nectar was scarce and honey bees readily accepted our artificial feeding stations. On meadows inside or near the Hainich forest, we caught foraging honey bees with a special “beelining box.” Inside this dark box, we fed the bees highly concentrated sugar syrup (approximately 2.0 mol/l) flavoured with anise essence. After release, most foragers oriented themselves, flew away and came back some minutes later. By means of different shellac colour paints, we individually marked the bees and recorded their round-trip times. Bees of the same nest can vary considerably in round trip duration, because they have different propensities to dance and spend different amounts of time inside the nest (Arnold et al., 2002). Therefore, to estimate the maximum distance of the bee tree, we considered the trip duration of the fastest bees. After a few round trips, the bees usually use an almost direct flight path (the “beeline”) between the feeder and their nest, so that their flight direction can be recorded. To calculate maximum nest distances, we used the following empirical formula which accounts for the homeward flight, the landing and unloading at the nest, and the rushing back and landing at the feeder:  $distance (m) = 243 \times t (min) - 627.75$ , where  $t$  is the round trip time of the fastest bee (Seeley, 2016). Based on a pilot beelining test in a forest area near Würzburg, we knew that the formula was generally suitable in our terrain. We used the average flight direction and the inferred maximum nest distance to make a prediction of the nest location. Then we trapped a bunch of foragers in the box again and released them at a new feeding site a couple of hundred meters in the direction of the nest. We repeated this procedure until the fastest bees only needed around three minutes for a round trip, which is when the bee tree is no more than 100 m away

(Seeley, 2016). We then started searching the trees in the area for the nest cavity with binoculars. In order to get information about the locations of apiaries around the Hainich National Park, we spoke to local beekeepers. To estimate the minimum density of feral colonies in the Hainich forest, we assumed that by catching foraging bees at a site with flowers in bloom, we would have obtained information about nests in an area of about 2 km around that site, as this corresponds to the area honey bee colonies usually forage (Visscher and Seeley, 1982; Steffan-Dewenter and Kuhn, 2003; Couvillon et al., 2015; Danner et al., 2016). We sampled slightly different but overlapping areas in the two years, an estimated 30.58 and 43.23 km<sup>2</sup> in 2016 and 2017 respectively, totalling 49.50 km<sup>2</sup>.

#### *Occupation of old woodpecker cavities by honey bee colonies in the Swabian Alb*

Beelining is a useful method for exploring the general occurrence and density of feral colonies in a woodland from scratch. However, it can be difficult to exactly localize the bees' nesting sites, a prerequisite for many ecological studies. An alternative way to determine the locations of feral honey bee colonies is to directly inspect suitable cavity trees in the region of interest (Oleksa et al., 2013b). A cavity for honey bees needs a volume of at least 20 l in order to allow them to hoard enough honey to overwinter (Seeley, 1985). The black woodpecker (*D. martius* L.) is the largest woodpecker in the Palearctic and excavates spacious nest cavities (Johnsson et al., 1993; Blume, 1996), making it a key candidate for creating nest sites that are suitable for honey bee colonies in managed forests. In the forest clusters of the Biosphere Reserve Swabian Alb, there is a long record of trees worked by black woodpeckers (Sikora et al., 2016). Because of the high ecological value of microhabitat structures in managed beech forests, such trees get permanently marked and are protected as a measure of biodiversity promotion (Bütler et al., 2013). In September 2017, we inspected 98 trees from a list of 282 trees in a forest area of 180 km<sup>2</sup> that are known to contain woodpecker cavities older than 10 years (Sikora et al., 2016). The selection of sites was based on logistic considerations. We found the tree locations with the aid of GPS coordinates. We inspected cavities with binoculars from the ground during weather conditions suitable for honey bee foraging. A cavity was considered occupied by a feral colony when there was regular flight traffic of foraging worker honey bees and pollen import. To make an estimate of the colony density, we calculated the density of all 282 habitat trees in the beech forest clusters and multiplied it with the occupation rate of the inspected cavities in fall 2017. Note that this density estimate must be regarded as a

lower limit because we only considered honey bee colonies nesting in old woodpecker cavities.

*Potential dispersal distances of honey bee swarms taking off from apiaries near the Hainich forest*

In the context of honey bees colonizing woodlands, an interesting question is how far swarms from beekeeper-managed colonies would usually move into the forests. When swarming bees leave their old nest, they first settle in close vicinity and form a hanging swarm cluster, typically on a tree branch. Scout bees then search the area for suitable nesting sites and report the location of their finds back at the swarm cluster via the waggle dance. Other bees that attend these dances may fly to the advertised sites, and—if they consider one appropriate—join in the dancing. Individual bees cease dancing after some time, regardless of the quality of the advertised site, so information about the best nesting sites accumulates over time through tradition, while information about inferior sites gets gradually lost. After hours or days, the outcome is a consensus about where to go, which is followed by the swarm's lift-off and move towards the new home (Lindauer, 1955; Seeley, 2010). As scout bees report their findings publicly on the swarm cluster, a human observer can easily obtain information about a swarm's potential nesting sites by observing and reading the bees' dances (Lindauer, 1955; Seeley and Morse, 1977).

In August 2017, we investigated the potential dispersal behaviour of swarms taking off near the Hainich forest, by decoding the nest site dances of three experimental swarms set up around the National Park. To prepare swarms, we captured the queen of a source colony and put her into a small cage of meshwork. Then, we swept 850–950 g worker bees (7,000–8,000 bees) off their combs into a screen cage. Together with their queens, the swarms were kept in a cool and dark place for >72 h and fed 50 % sucrose solution ad libitum, provided through a gravity feeder bottle. It is known that artificial swarms prepared like this behave like natural ones in that they start searching for nest sites and eventually move to a new home (Morse and Boch, 1971; Seeley and Morse, 1977). To simulate realistic dispersal events from managed hives, we set up the swarms at sites around the Hainich forest, which were near settlements or actually close to an apiary: in Weberstedt (northeast of the Hainich, 10.49, 51.11, near an apiary, distance to forest: 870 m), in Craula (southeast of the Hainich, 10.47, 51.06, distance to forest: 450 m), and at Mallinde (southwest of the Hainich, 10.40, 51.05, distance to forest: 170 m). We used a board of plywood (50 × 45 cm) vertically fixed

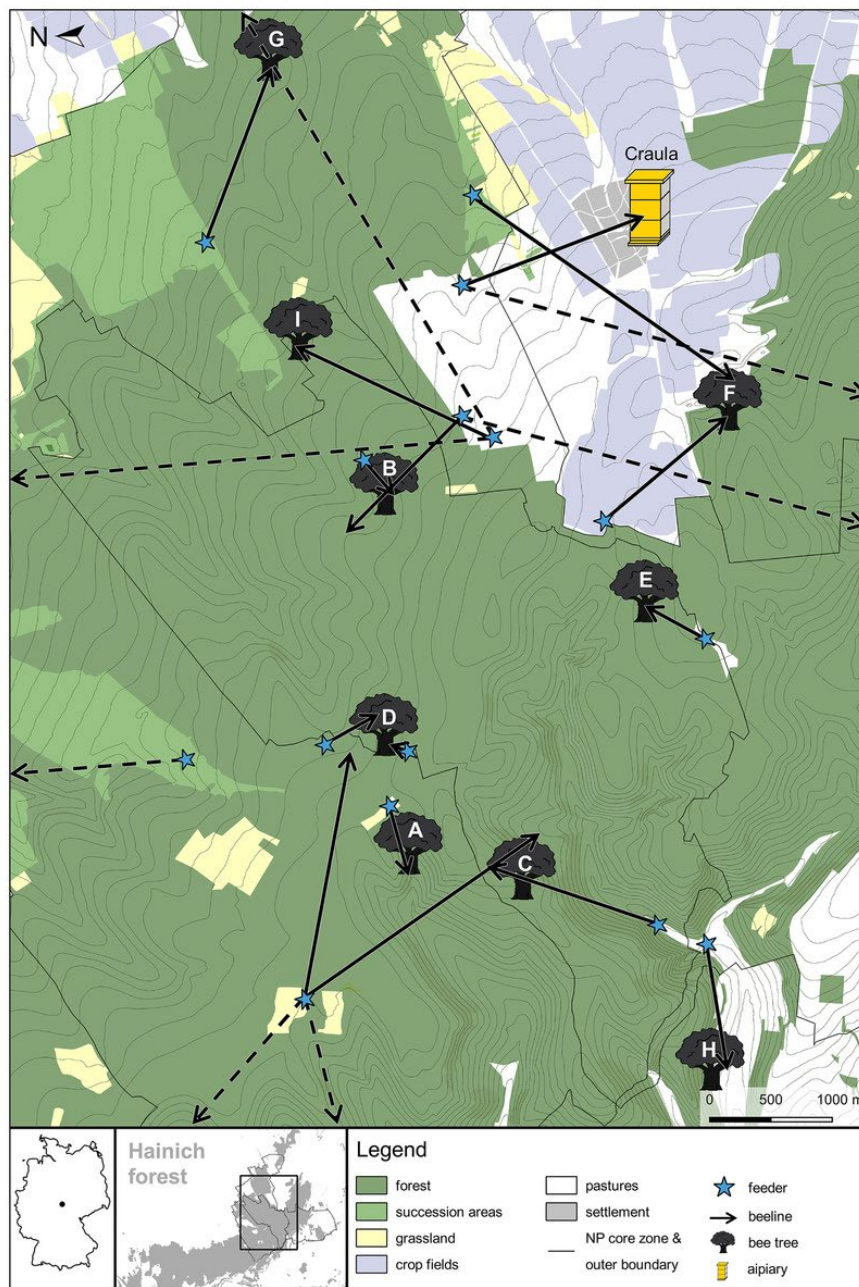
to a wooden stand as a swarm observation mount. We positioned the caged queen at a gap in the centre of the board and let the workers out of the screen cage so that they could assemble on the board around their queen. When established, we started video recording the swarms for a later analysis of nest site dances. In order to avoid having bees dancing for nectar sources, we continued to feed the swarms sucrose during the experiments (Seeley and Buhrman, 1999). We worked during optimal weather conditions for swarming, on a sunny day with temperatures  $>20$  °C. As the swarms' queens were confined to a cage at any time, we prevented the swarms from actually moving to a new nest cavity. Thus, scout bees could search for nest sites, dance and reach a consensus about where to go, but lifted swarms would always have to come back to the swarm mount and their queens. On all three swarms we observed dances soon after the setup was established, and two swarms (in Craula and in Weberstedt) actually tried to move after some hours. In the evening after the observation, we swept the bees off the swarm mount into the screen cage again and brought the colonies back to their original hives.

For decoding the nest site dances, we played the videos until a dance commenced. Then we measured the duration of around six consecutive dance circuits per dance to calculate an average circuit duration for each dance and aligned a digital protractor to obtain the dance angle in relation to the vertical. We translated dance circuit duration into nest site distance based on the original data of Von Frisch (1967). The distance-dance circuit duration relationship is well described by two linear regression equations, with a breaking point at about 1,000 m distance. We used  $distance (m) = 466.5495 \times circuit\ duration (s) - 675.0336$  for circuit durations up to 3.15 s, and  $distance (m) = 1102.7328 \times circuit\ duration (s) - 2666.6256$  for circuit durations  $>3.15$  s (see Fig. S1). To get to know the direction of the advertised nest sites, we calculated the solar azimuth at the time of each dance using the AstroWin 32 software (Strickling, 2012) and added the solar azimuth angles to the dance angles. We could not decode all dances performed by the scout bees, as these did not dance exclusively on the surface of the swarm. Some opportunistically used the wooden board as a dance floor and communicated nest sites covered by the swarm cluster out of our sight. However, as the observed dances were most likely a random portion of all dances, they should well represent the overall range of nest site distances and preferences by the swarms.

## Results

### *Occurrence and density of feral honey bee colonies in beech forests*

In total, we detected nine bee trees in the Hainich forest (Fig. 1). All nest sites were located in areas with dense tree canopies. This hampered the determination of exact flight directions near the nest sites and locating the actual cavity tree was infeasible. We therefore determined the approximate location of each bee tree by taking into account the round-trip durations of bees foraging at the last feeder and the information obtained at the previous feeding sites (for detailed examples of how we inferred the bee tree locations see Fig. S2). In 2016, we inferred the bee tree locations A, B, C and D. In 2017, we sampled a slightly different area and detected bee tree location D again plus the bee tree locations E, F, G, H, and I. This corresponds to an estimated average colony density of 0.13 colonies per square kilometre (0.13 col/km<sup>2</sup> in 2016, 0.14 col/km<sup>2</sup> in 2017). While five bee trees lay within the core zone of the National Park, another two lay within its boundaries, and two just outside the boundaries of the National Park. We got to know that in every village around the National Park there is at least one apiary with around 10 managed colonies each. In one case, the inferred location of a potential bee tree (location H) was near the forest edge, so we specifically searched that area for beekeepers' hives to be sure that the tracked bees came from a feral colony. The approximate mean  $\pm$  SD distances of the bee trees to the forest border and to the nearest apiary were  $980 \pm 830$  and  $2,660 \pm 1,080$  m respectively (Table 1).



**Figure 1:** Map of the bee tree locations inferred from beelining data in the Hainich forest in the years 2016 and 2017.

Arrows represent recorded beelines (putative flight vectors of foragers from artificial feeders to their nests), with an arrow tip predicting a bee tree location. The vector lengths give the maximum distances of the nests based on round trip times of the fastest bees, and the vector angles give the direction of the nest based on average flight directions. Dashed arrows represent beelines to bee trees or apiaries that probably lay outside of the map because the respective bees had long round-trip durations. Note, however, that we might have overestimated nest distances when the observed bees spent long times inside their nests between foraging flights (e.g., due to recruitment dancing activity) (Germany map created in R with package “maps” (R Core Team, 2016; Brownrigg, 2017). Other map data © Mapbox, © OpenStreetMap).

**Table 1:** Distances of the inferred bee tree locations in the Hainich forest to the forest edge and to the next apiary.

<b>Bee tree</b>	<b>Distance to forest edge (m)</b>	<b>Distance to next apiary (m)</b>
<b>A</b>	2,350	4,300
<b>B</b>	700	2,600
<b>C</b>	1,700	3,600
<b>D</b>	2,250	4,000
<b>E</b>	550	2,850
<b>F</b>	150	1,500
<b>G</b>	500	1,500
<b>H</b>	<50	1,150
<b>I</b>	550	2,400
<b>Median</b>	550	2,600
<b>Mean</b>	980	2,660
<b>SD</b>	830	1,080

In the beech forests of the Biosphere Reserve Swabian Alb we inspected 98 habitat trees with old black woodpecker cavities for the presence of honey bee colonies (see Fig. S3 in supplemental information for a map of the region). Many of the habitat trees contained more than one cavity (Fig. 2). Seven trees (7.1 %) were found to be occupied by honey bees. Based on the density of mapped woodpecker cavity trees in the region, we estimated the density of feral honey bees to be at least 0.11 colonies per square kilometre.





**Figure 2:** Bee trees in the Swabian Alb.

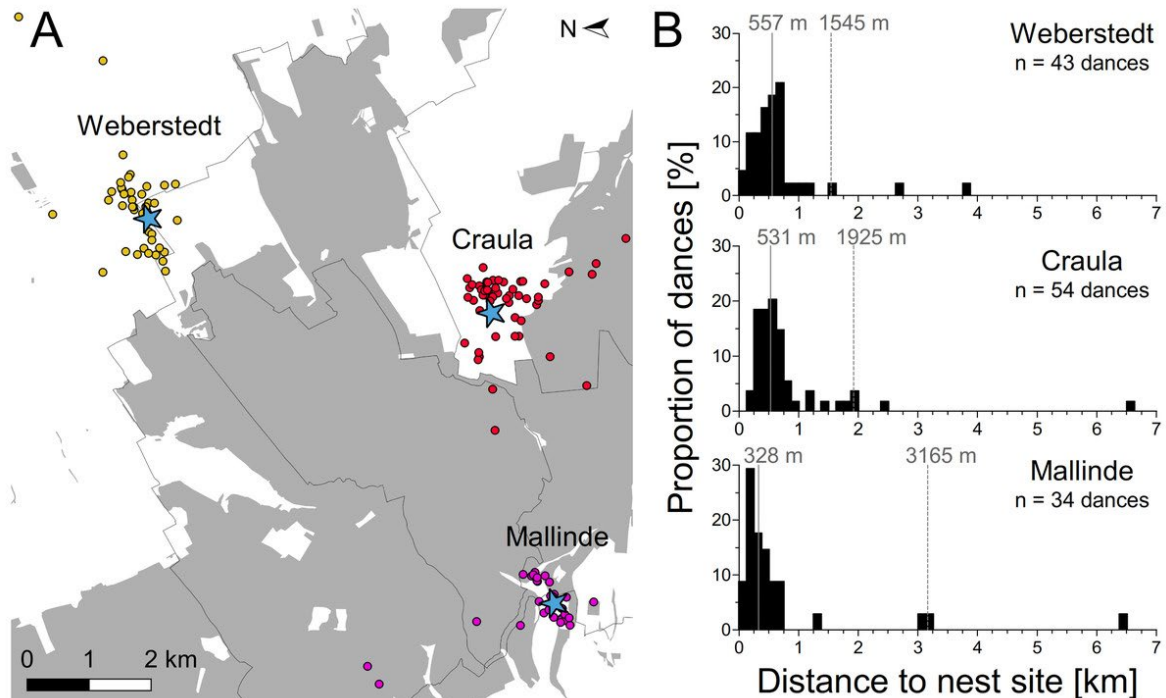
(A) Portrait of a bee tree with three cavity openings made by the black woodpecker. (B) close-up of an old woodpecker cavity occupied by a honey bee colony. Both trees are beech (*Fagus sylvatica* L.) (Germany map created in R with package “maps” (R Core Team, 2016; Brownrigg, 2017). Photo credit: Patrick Laurenz Kohl).

#### *Potential dispersal distances of honey bee swarms*

Nest site scouts of three experimental swarms set up around the Hainich forest apparently covered a large area, dancing for nest sites up to 6.5 km away (Fig. 3). However, the majority of dances indicated sites within a few hundred meters around the swarm mounts. Bees of the swarms in Craula and at Mallinde communicated nest sites located in the Hainich forest, but those of the swarm in Weberstedt did not. Potential nest site hot spots (sites which were advertised repeatedly by scout bees) of the swarms in Weberstedt and Craula lay outside the forest, while the swarm at Mallinde showed preferences for nest site at the forest margin. Average median and 95-percentile nest site range for the three swarms were 472 and 2,112 m, respectively. On the swarm at Mallinde, which was located closest to the forest, nest site scouts danced for nearer sites than those of the other swarms (Kruskal-



Wallis test:  $\chi^2 = 10.92$ ,  $P = 0.0049$ ; Dunn's multiple comparison: Mallinde vs. Weberstedt ( $P < 0.05^*$ ), Mallinde vs. Craula ( $P < 0.01^{**}$ ), Craula vs. Weberstedt ( $P > 0.05$ )).



**Figure 3:** Potential dispersal distances of honey bee swarms.

(A) Overall pattern of nest sites found by the experimental swarms set up around the Hainich forest (grey). Blue asterisks depict the positions of the three swarms and dots give the position of advertised nest sites as inferred by decoding the nest site dances of these swarms in Weberstedt (yellow dots), Craula (red dots) and at Mallinde (violet dots). One advertised nest site of each swarm in Craula and at Mallinde lay outside the range of the presented map (Map data © Mapbox, © OpenStreetMap). (B) Frequency distribution of potential nest site distances as indicated by dancing bees on the experimental swarms. Solid lines indicate the median and dashed lines give the 95th percentile range of nest site distances.

## Discussion

We set out to address a simple question: are there naturally nesting honey bee colonies living in German beech forests? Our results show that the answer is yes, but at low densities. Through the use of beelining technique in the Hainich National Park, where there is no beekeeping activity, we got a picture of the distribution and density of feral colonies in a near-natural beech forest. We found that honey bee colonies were not confined to the forest edges; they were also living deep inside the wood. We inferred a density of around 0.13 colonies per square kilometre. This estimate must be regarded as conservative because we

assumed that we would have kept track of any colony in a large area of 2 km around each sampling site. Especially in the core zone of the National Park, there were no opportunities to sample foraging worker bees, so that we might not have sampled the area exhaustively. Further, as we could not exactly locate the actual cavity trees, there might have been more than one bee tree at their inferred locations. Our direct survey of cavity trees in the beech forests of the Swabian Alb corroborated the findings from the Hainich forest that honey bee colonies are regularly inhabiting old tree cavities in beech forests. Our estimates of the lower limit for the colony densities in both woodland areas are similar and are in accordance with the feral colony density estimated in Northern Poland (0.1 colonies/km<sup>2</sup>) (Oleksa et al., 2013b). However, it is quite low in comparison to the population density of feral honey bee colonies found in the temperate forests of New York State (1.0 colonies/km<sup>2</sup>) (Seeley, 2007; Seeley et al., 2015).

It is clear that our data present a “snapshot,” without information about the life-histories of individual honey bee colonies. However, we think that they provide some valuable information:

1. The occurrence of feral honey bee colonies in near-natural beech forests is the norm rather than an exception. We found several honey bee nests in both of the two independent woodlands, and in the Hainich forest there were occupied bee trees in both study years. In a pilot beelining tests in a forest area near Würzburg in 2016, we also found feral honey bee colonies.
2. It is clear that the density of feral colonies in a near-natural deciduous forest in Germany is much lower than the density of beekeeper-managed colonies in rural and urban areas, which is generally between one and three colonies per square kilometre (Jones, 2004). However, when using our estimates to extrapolate the total population size of feral honey bee colonies living in German forest stands older than 80 years, we infer a size of 4,400–5,600 colonies, which we think is a noteworthy number.
3. The data gathered in the beech forests of the Swabian Alb provide valuable information for future studies and conservation. We hypothesize that the abundance and activity of black woodpeckers is a key factor determining the density of cavities that are large enough for honey bee colonies in managed forests. Beech trees with a diameter at breast height of about 40 cm, which start to be attractive for the woodpecker (Taux, 1976), still have intact sapwood and bark. However, the bird is

capable of identifying fungal activity in a tree's central heartwood and can make cavities in trees that would otherwise lack any microhabitat structures (Zahner et al., 2012). Interestingly, the black woodpecker reached its present day population size in Central Europe not before the 1950s, probably as a response to the recovering forests (Cuisin, 1985; Blume, 1996). This suggests that protecting large habitat trees in managed forests not only helps charismatic forest birds but can also promote the occurrence and abundance of feral honey bee colonies.

4. We found that scout bees of swarms set out around the Hainich forest advertised several nest sites within short distances, mostly outside the forest. On the swarm, which was set up closest to the forest edge, scout bees advertised nest sites at closer distances than those of the other swarms, possibly because there were even more suitable nest sites nearby. Prior studies documented similarly short dispersal distances (modal value: 450 m) (Lindauer, 1955; Seeley and Morse, 1977; Seeley, 2010), suggesting that honey bee swarms will always make the most economical decision and move little when suitable nest sites are close by (Seeley, 2010). Our data suggest that there are suitable nesting sites for honey bee colonies in the agro-urban space, and that a beech forest like the Hainich provides ample, but not necessarily better nesting opportunities. Given that the median distance of advertised nest sites in our swarms was only 470 m, the bee trees in the Hainich forest, which were located at a median distance of 2,600 m from the next apiaries, were probably not colonized by swarms from beekeepers' hives in single dispersal events.

Forest dwelling feral honey bee colonies live in another environment than managed colonies and are subjected to natural selection: they nest in widely spaced tree cavities, no one treats them against parasites, nor are they fed in times of nectar scarcity. There needs to be a certain network of tree cavities for a noteworthy number of feral colonies to persist in any region. This basic requirement seems to be at least partially fulfilled in near-natural or ecologically managed deciduous forests or where there are networks of forest fragments and rural avenues lined with old trees (Oleksa et al., 2013b). It will be interesting to study the life-history traits of feral honey bee colonies in European landscapes and to pose questions such as the following: What is the average longevity of feral colonies and what is the lifetime reproductive success of their queens? Is the number of feral colonies actually limited by parasites in the first place? Or is it the availability of suitable cavities, or resource limitation that keeps feral honey bee colonies at low densities? Understanding and

comparing the drivers of the feral honey bee colony density in different woodland areas will also help to answer the question how frequent honey bees would naturally be in temperate Europe.

Irrespective of the fate of individual colonies, the regular nesting of honey bees in woodlands has ecological impacts. Beekeeping activity is not distributed equally across the country but concentrated in rural and urban areas. Managed hives are usually only occasionally moved into the forests (Banaszak, 2009). Consequently, this leads to an uneven distribution of foraging worker honey bees in the landscape, regardless of differences in resource availability among habitats. However, when there are feral colonies living in a forest, the density of foraging workers in that area will increase, with possible consequences for the pollinator community composition of forest dwelling plants (Steffan-Dewenter and Tscharnke, 2000; Mallinger et al., 2017; Hung et al., 2018). This is also relevant if the density of feral colonies is low. Based on their colony size and the proportion of foraging workers, it must be assumed that a single honey bee colony sends out up to 5,000 foraging bees daily (Seeley, 1985). When such colonies live in forest fragments surrounded by arable land, they may also contribute to the pollination of nearby crops (Mitchell et al., 2014; Decocq et al., 2016). Another point is that feral honey bee colonies will directly or indirectly interact with other organisms that live in hollow trees. They certainly compete for tree cavities that are also attractive to other animals (Johnsson et al., 1993; Sikora et al., 2016). Further, as they transport nutrients to their nests, honey bee colonies are hotspots of accumulated biomass: individuals can directly be preyed upon and consumed by birds, mammals and other insects (Morse and Nowogrodzki, 1990). And through the drop of organic matter like wax, pollen, or dead bees, honey bee nests indirectly nourish a whole cosmos of organisms that live from organic detritus inside tree cavities. As tree cavities with dead organic matter are generally rare in managed forests, the presence of honey bees could have a positive effect on the abundance of specialized arthropod communities in European beech forests (Möller, 2009; Müller et al., 2014).

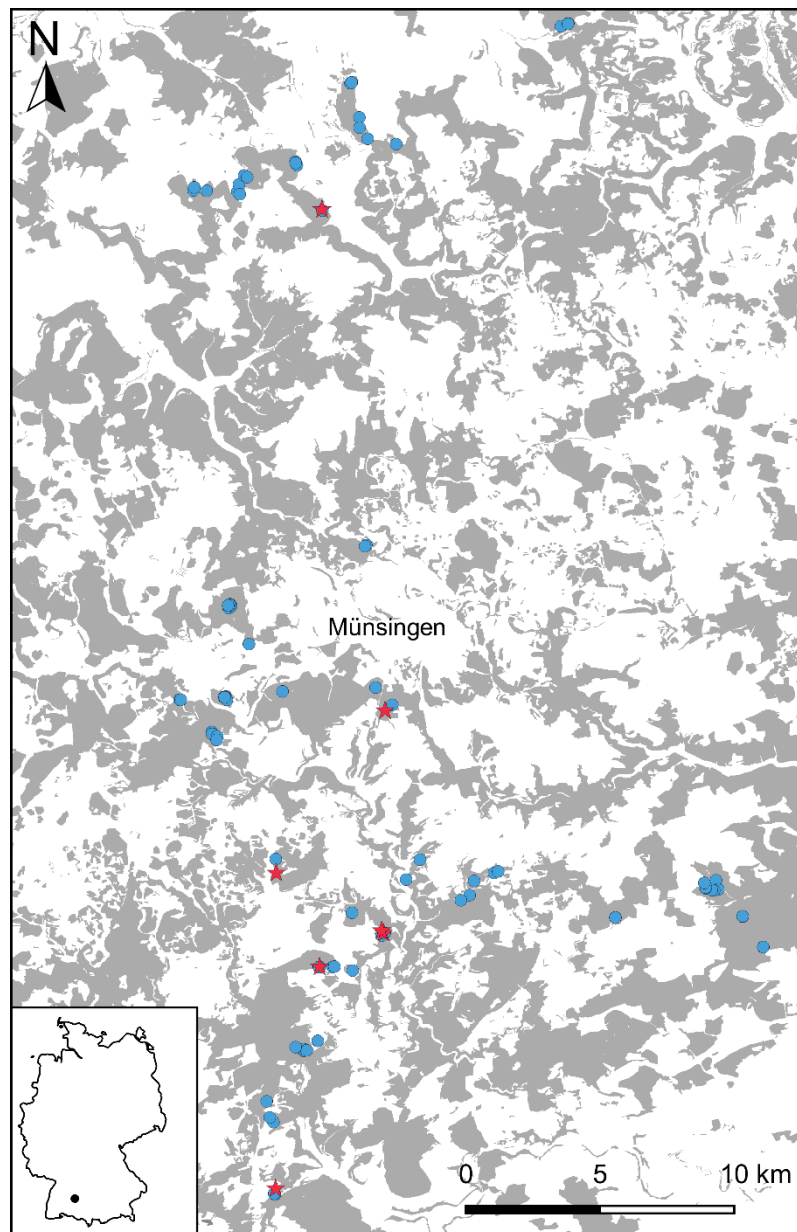
### **Conclusion**

Our study showed that honey bee colonies nesting naturally in tree cavities are more common than it is generally assumed. When assessing the impact of honey bees on ecosystems, or when evaluating the need for their conservation, one should differentiate between managed colonies aggregated in apiaries and feral colonies living singly in natural

nests. In Europe, *A. mellifera* is treated as a domesticated animal and feral honey bee colonies lack any legal protection. There is a lot to be investigated about the life of feral honey bees and their interactions with the (forest) environment. However, changing the perspective and recognizing that the honey bee can also live as a wild animal is a prerequisite for achieving a full understanding of its biology.

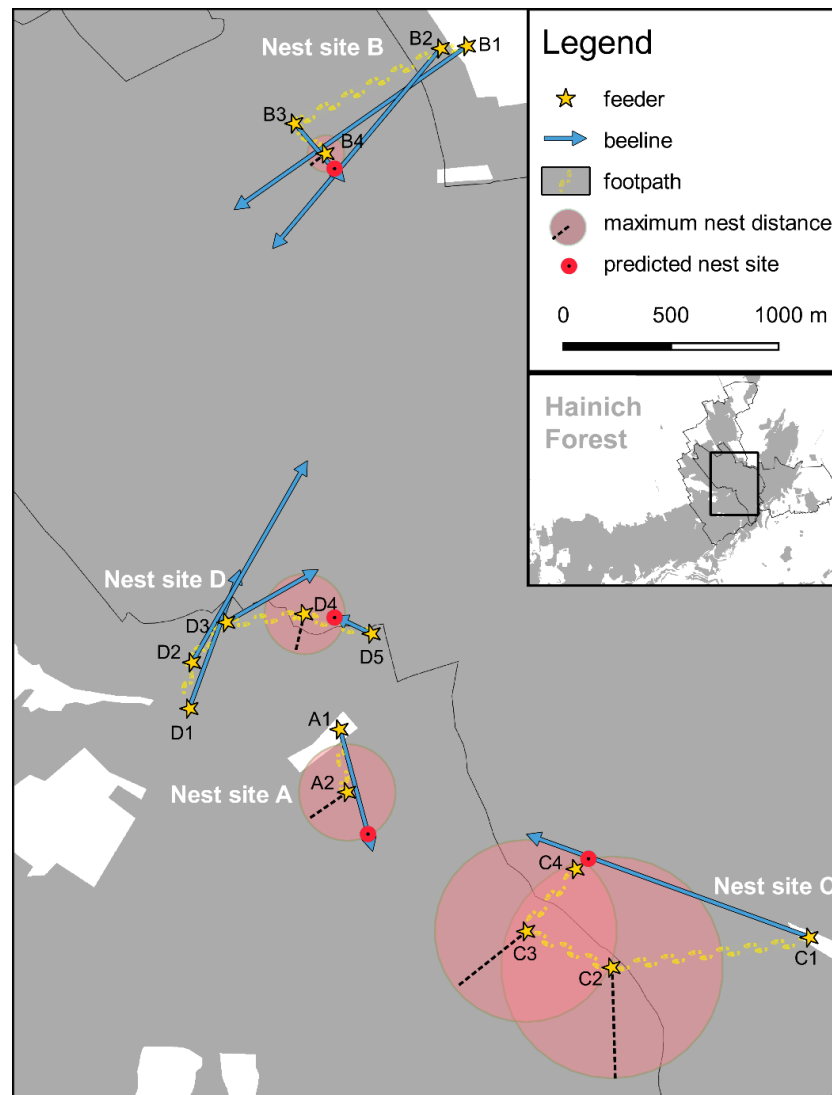
### **Acknowledgements**

We thank the administration of the Hainich National Park for the possibility of conducting our study in the core zone of the National Park. We are grateful to Luis G. Sikora who kindly provided the GPS coordinates of habitat trees and helped us to find and inspect these trees. We also thank the Deutsche Wildtierstiftung for sharing the habitat tree data and the administration of the Biosphere Reserve Swabian Alb for enabling us to work there. For their valuable comments on the manuscript, we thank Harald Detering, Fabienne Maihoff, Manuel Püttmanns, Sereina Rutschmann and Michael Tölle-Nelken. Last but not least, we thank Jürgen Tautz and Ingolf Steffan-Dewenter for their suggestions how to improve this paper and for supporting this study.

**Supplementary material**

**Figure S1:** Map of the Swabian Alb.

Map of the 98 inspected cavity trees (blue dots) in the Biosphere Reserve Swabian Alb with the 7 cavity trees occupied by honey bees (red asterisks) (Map data © Mapbox, © OpenStreetMap).



**Figure S2:** Detailed picture describing four bee hunts with beelining technique.

When we had problems observing vanishing bearings beneath the dense tree canopy, we used the minimum round-trip time to calculate the maximum distance from the feeder to the nest (red transparent circles). Nest site A was predicted to be on the beeline from feeder A1 and inside the maximum nest range obtained at feeder A2. Nest site B was predicted to be at the crossing of the beelines from the feeders B2 and B3, and within the maximum nest range inferred from feeder B4. Nest site C was predicted to be on the beeline from feeder C1 and very close to the feeder C4 where the round-trip time was shortest (Note that the predicted nest site for C lies also within the maximum nest ranges inferred from the feeders C2 and C3). Nest site D was predicted based on the beeline from feeder D5 and the maximum nest range inferred from feeder D4 (Map data © Mapbox, © OpenStreetMap).

Further supplementary data on dance circuit duration of waggle dancing bees in relation to flight distances, beelining-raw data, and data on advertised nest sites can be found online at <https://doi.org/10.7717/peerj.4602>.





Before canopy closure in spring, mixed deciduous forests in Germany offer abundant food resources (here wild garlic (*Allium ursinum*) is in bloom). However, studies during the whole year about the suitability of Central European forest landscapes as a habitat for honey bees are lacking. Photo taken by Ingo Arndt.



## CHAPTER 4: HONEY BEE FORAGING IN CENTRAL EUROPEAN FOREST LANDSCAPES

This chapter is submitted as:

**Rutschmann B**, Kohl PL, Steffan-Dewenter I (submitted) Foraging distances, habitat preferences and seasonal colony performance of honey bees in Central European forest landscapes.

### Abstract

1. Woodland is considered as native habitat of the Western honey bee (*Apis mellifera*) and is assumed to provide important pollen and nectar sources. However, resource supply might be spatially and temporally restricted and landscape-scale studies in European forest regions are currently lacking.
2. Capitalizing on the unique communication behaviour, the waggle dance, we investigated honey bee foraging in a deciduous forest region in southern Germany. We analysed 2022 waggle dances performed by bees of twelve colonies placed at locations with varying degree of forest cover (50-99 % at a 2 km radius) from March–August 2019, thereby identifying foraging distances and habitat preferences over almost an entire foraging season. By connecting dance information with colony weight recordings, we estimated the contribution of the different habitat types to colony weight gain.
3. Foraging distances generally increased with the amount of forest in the surrounding landscape. Yet, this effect strongly depended on the season and was more pronounced for pollen than for nectar foraging. Even though colonies in forest-dominated landscapes had to fly further, colony weight was not affected by forest cover.
4. Compared to expectations based on the proportions of different habitats, colonies foraged more frequently in grasslands and cropland than in deciduous and coniferous

forests, with late summer being an especially difficult period for pollen foraging in forests. During a phase of colony weight gain in early summer, the use of forests for nectar/honeydew foraging was close to the expectation, highlighting forests as an important source of carbohydrates during short periods of the year.

5. *Policy implication.* The ecological and economic value of managed forest as habitat for honey bees and other wild pollinators could be significantly increased by the continuous provision of floral resources, especially for pollen foraging. We therefore recommend diversifying forest stands with insect-pollinated trees, permitting secondary successions in forest gaps, and at larger scales, creating forest landscapes with high habitat diversity.

## **Introduction**

The original habitat of native European honey bees is forest (Zander, 1949; Ruttner, 1988; Crane, 1999), but the quality, quantity, and seasonal availability of food resources in different forest types is mainly unknown. Overuse of timber and further changes in forest management paved the way to structurally different forests during the last centuries (Küster, 2003; Brunet et al., 2010; Hanula, 2015). Subsequently, beekeeping shifted to agricultural landscapes where mass-flowering crops provide resources and pollination is needed (Crane, 1999). Nowadays, the density of wild-living honey bee colonies in European forests is low (Kohl and Rutschmann, 2018; Requier et al., 2020), and professional beekeepers move their hives only temporally inside forests to exploit ephemeral nectar flows of individual tree species or the production of honeydew by tree aphids (Ruppertshofen, 1995; Liebig, 1999; Prešern et al., 2019). Nevertheless, forests still cover more than one third of the land area in Europe (Raši, 2020), and may have properties beneficial for bees (Hanula et al., 2016). For example, woodlands provide resources across different layers from the understory to the canopy, offer a rich flora at roadside corridors, forest clearings or edges and shelter colonies from agrochemicals, gusty winds and searing summer heat (Zander, 1949; Ulyshen et al., 2010; Hanula et al., 2016; Bentrup et al., 2019). However, reforestation, fire exclusion, the reduction of game and the exclusion of livestock resulted in unnaturally dense stands with closed canopies (Hanula, 2015), hampering the establishment of an otherwise flower-rich understory in today's forests (Williams and Winfree, 2013; Hanula, 2015; Eckert et al., 2022; Braun-Reichert et al., 2021) and shifting species composition towards more shade-tolerant and nutrient-demanding species (Verheyen et al., 2012). As

colonies require a sustained supply of carbohydrates and pollen to maintain colony functioning, landscape composition is seen as a critical determinant of honey bee health, colony performance and overwintering success (Di Pasquale et al., 2013; Sponsler and Johnson, 2015; Alaux et al., 2017; Rutschmann et al., 2022), potentially leading to decreased densities of honey bee colonies and other flower-visiting insects. Therefore, there is a growing interest in optimizing forest management practices for pollinator conservation (Eckerter et al., 2022), including the identification of temporal shortages of feeding resources in different forest types.

Honey bees offer researchers unique insights into their food choices via the waggle dance which they use to inform nestmates about the locations of profitable food sources (Frisch et al., 1967). The distance and direction signal of waggle dances can be decoded to infer the bees' approximate foraging locations and habitat preferences (Visscher and Seeley, 1982; Beekman and Ratnieks, 2000; Steffan-Dewenter and Kuhn, 2003; Young et al., 2021). Yet, most studies on honey bee foraging inferred by waggle dance decoding have focused on crops, rural or urban lands (e.g. Couvillon et al., 2014; Garbuzov et al., 2015; Danner et al., 2016; Sponsler et al., 2017; Bänsch et al., 2020; Samuelson et al., 2021). Landscape-scale studies which investigate the suitability of Central European forests as foraging habitat for honey bees are currently lacking.

To close this knowledge gap, we experimentally placed twelve honey bee colonies at locations with varying degree of forest cover and decoded their waggle dances at regular intervals from early spring to late summer. Reducing beekeeping management of the colonies to a bare minimum, we closely simulated wild-living colonies inhabiting a Central European deciduous and mixed forest. Assuming that changes in the availability of attractive resources inside the forest or in adjacent fields would be indicated by changes in the foraging distances, we asked (1) whether the proportion of forest cover would affect foraging distances and whether this effect would differ between seasons and forage types (nectar/honeydew versus pollen). Furthermore, by analysing the approximate foraging locations of honey bees with geographic land use maps, we tested (2) whether honey bees use woodland at the expected level given the landscape-wide availability of forests. Finally (3), via connecting waggle dance information with hive weight recordings, we directly estimated the contribution of different habitat types to the net weight gain and subsequent honey yield of honey bee colonies.

## Methods

### *Study region and site selection*

We conducted this study in the years 2018 and 2019 in the Steigerwald, a low mountain forest region in southern Germany. Most of the forested area (around 165 km<sup>2</sup>) is managed by the Bavarian state-owned forest enterprise which aims to reconcile commercial timber production and biodiversity conservation e.g. by setting aside islands of old-grown forests (Mergner and Kraus, 2020). With a share of 44 % of the forest area, beech (*Fagus sylvatica*) is the dominant tree species, followed by oak (*Quercus spp.*, 21 %), Scots pine (*Pinus sylvestris*, 13 %), spruce (*Picea abies*) and hornbeam (*Carpinus betulus*) (Mergner and Kraus, 2020). Being part of the warm temperate zone (BayFORKLIM, 1996), the study area has a mean annual temperature of 7.5 °C, and a mean annual rainfall of about 850 mm (Mergner and Kraus, 2020).

We intersected the state forest area with a recently published pan-European land cover data set representing five major habitat types (Weigand et al., 2020) using GIS software (QGIS Development Team, 2009). We chose twelve sites according to the proportion of forest within two-kilometres circular buffer areas to represent a gradient from locations at the forest edge to locations surrounded only by woodland (Fig. 1A). Besides, the proportions of grassland and cropland within the buffer areas were as balanced as possible, and two neighbouring locations were at least around two kilometres apart to ensure spatial independence (distance to nearest neighbour: mean +/- s.d.: 2643 m +/- 505 m; range: 1972 m – 3920 m).

### *Observation hives and honey bee colonies*

We used custom-built observation hives that were based on single Zander hive boxes (volume approximately 45 l) with ten frames each, allowing to house normally sized honey bee colonies (Fig 1B). Each hive had a glass window for filming the waggle dancing bees on the outer side of the first comb. In July 2018, we transported twelve equally sized colonies of *A. mellifera carnica* to the selected sites in the forest. The mere beekeeping manipulation was that we treated colonies against varroosis with formic acid in August 2018. While using the rest of the honey bee flight season 2018 to test the methodology, the main data collection started in March 2019 with the colonies having over-wintered in the forest. Further methodological details are given in the supplementary material (SM).

*Video recording and dance screening*

On nine observation days between March and September 2019 with good weather conditions, we simultaneously recorded dancing bees in all twelve study colonies using digital camcorders. For every colony, we screened the videos of each observation day and selected dances in such a way that the samples were distributed across the whole videorecorded periods (see also SM for camera setup and screening scheme). Numerous dances were recorded except on the last observation day in September 2019 when only foragers of one colony danced (thus, the date was discarded for the analysis. In case pollen was visible on the hind legs, we identified them as dances for pollen, all other dances were classified as nectar/honeydew dances.

*Decoding waggle dances to infer foraging distances and habitat use*

Back in the hive, honey bee foragers perform a figure eight-shaped dance on the comb, consisting of alternating waggle phases and return phases (Frisch et al., 1967). The bearing of the dancing bee during the waggle run and the duration of the waggle phase encode the direction and distance of the advertised resource in the field. We inferred approximate foraging distances from circuit duration (duration of one waggle phase plus one return phase) since circuit duration is faster to time than waggle duration and provides distance information sufficiently accurate for forage mapping (Arra et al., unpublished data). To obtain an average circuit duration of a dance, we timed the duration of several consecutive dance circuits (mean 6.2 +/- 2.1 circuits). We measured the dance angle (relative to a vertical) by aligning a digital protractor with the dancer's body orientation during the waggle phases and obtained the direction of the resource in the field by adding the current solar azimuth angle. The measurements were manually performed on a computer using the program *utilius fairplay 5* from *ccc* software.

To derive foraging distance from circuit duration we used our own calibration curve that is based on a feeder training experiment performed in the same forest region and with the same bees as this study (Kohl and Rutschmann, 2021); Arra et al., unpublished data). We applied the following two formulas to translate the mean circuit duration  $t$  per dance (in seconds) into distance ( $d$ , in meters) depending on the value of  $t$ :

- $t \leq 4.439852$ :  $d = \ln(-0.2253179 * t + 1.3305467) * (-1534.086) - 84.98$
- $t > 4.439852$ :  $d = 1242.854 * t - 3903.066$

To infer the geographic foraging locations indicated by dances we combined distance and angle in the field for every decoded dance.

We directly obtained information on the error of our decoding method from the feeder training experiment (Fig. S1): We back-decoded the dances for the known feeder positions and determined for each dance-indicated location the spatial deviation (in meters) from the actual location of the respective feeder. We use these data to produce an uncertainty buffer ( $b$ , in meters) for each inferred dance location which includes 75 % of all distance deviations for a given foraging distance  $d$ :

$$- \quad b = 33.7335867 + 0.3301049 * d$$

Regarding the habitat analysis, the proportional amount of different habitat types in the uncertainty buffer was calculated for every dance. Further details and R codes can be found in the SM.

#### *Calculating the expected habitat use of honey bee colonies in the study landscapes*

The information of realized habitat is especially insightful when related to the actual frequencies of the different habitat types in the study landscape. Bees travel only as far as necessary during food collection to minimize energy expenditure and mortality risk (Frisch et al., 1967; Seeley, 1994). As a result, patches close to the hive are of greater importance and will be visited more frequently than patches of similar quality further from the colony. In calculating expected habitat use we therefore used a distance correction: based on the observed honey bee foraging distance distribution, patches further away from the hive location were considered to contribute less to the expected habitat use than patches in close vicinity (Fig. S2). This allowed us to identify which habitat types foragers preferred without overestimating the importance of habitats occurring proximate to the hives (Henry et al., 2012; Couvillon et al., 2014). The workflow for the habitat analysis is described in the SM. With this method, the twelve study landscapes had forest cover values (both deciduous and coniferous combined) ranging from 52 % to 96 % (Fig. 1C).

#### *Weight analysis*

We used eleven weight scales (Capaz BEE HIVE SCALE GSM 200; one location had no GSM signal) with a 100 g precision, equipped with a GSM module, and set the hive scales to record hourly weights during the whole experimentation period. To extract meaningful

information, we manually took care of artefacts during the preliminary data analysis (caused by scales malfunction, manipulations during the videorecording setup or by swarms leaving the colony in spring). For the analysis we chose to use the corrected midnight weight when all foragers were back in their nest.

### *Statistics*

#### Foraging distances and colony weight changes

A linear mixed effect model (Bates et al., 2015) was built with foraging distance as response variable, forest cover (both deciduous and coniferous combined and scaled), observation day and forage type (nectar/honeydew versus pollen) as categorical explanatory variables with all the two-way-interactions and the three-way-interaction. Colony location was modelled as a random factor (Harrison et al., 2018). Due to non-normality, we box cox transformed the model. An anova (Kenward-Roger, Type II) from the stats package in R was used for the inference.

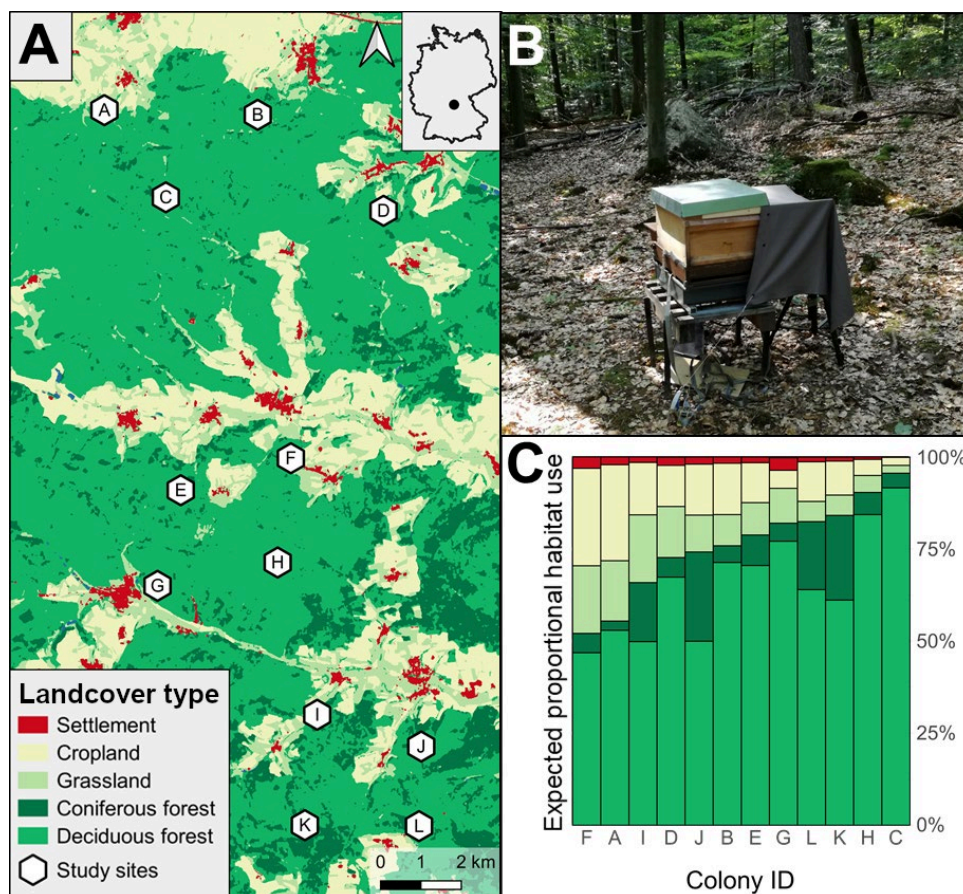
Likewise, we modelled the influence of forest cover on daily colony weight changes with a linear mixed effect model with forest cover (both deciduous and coniferous combined and scaled) as the explanatory variable and colony location as a random factor. For inference, an anova (Kenward-Roger, Type II) was used again.

#### Habitat use

We built a generalized linear mixed effect model using the glmmTMB package (Bolker, 2016; Magnusson et al., 2017) to analyse whether the observed number of dances per habitat category deviated from the expected number of dances based on the distance-corrected habitat proportions in the landscapes surrounding the colonies. The number of observed dances for a habitat type was the response variable. We modelled the ratio between observed dances and expected dances for a habitat by including “expected number of dances” (habitat proportion\*number of dances per colony, observation day and forage type) as an offset term in the model, thereby also accounting for different number of dances of different colonies and dates (Bolker, 2016). A generalized Poisson distribution (“genpois”) was chosen for the response variable (Brooks et al., 2019). Colony locations were implemented as a random factor (Harrison et al., 2018). We followed an information theoretic approach to select the best among several models that included different combinations of the four explanatory variables, habitat type (deciduous forest, coniferous

forest, grassland, cropland, and settlement), forage type (nectar/honeydew versus pollen), observation day, forest cover (both deciduous and coniferous combined and scaled), and their interactions. Model comparisons were based on AIC (“AICtab” from the package *bbmle*). For inference we used the *Anova.glmTMB*-command from the *glmmTMB* package (Chi-Square test, type II anova) (Magnusson et al., 2017). To visualize the results of habitat use analyses, we plotted observed habitat use for every colony and observation date (in per cent) next to the respective expected habitat use.

For all models, model assumptions were checked and verified with the package *DHARMA* (Hartig and Hartig, 2017).



**Figure 1:** (A) Landcover map of the study region based on Weigand et al., 2020. (B) Observation hive in the forest on a weight scale during dance recording (C) Expected proportional habitat use of the twelve colonies across the forest cover gradient.



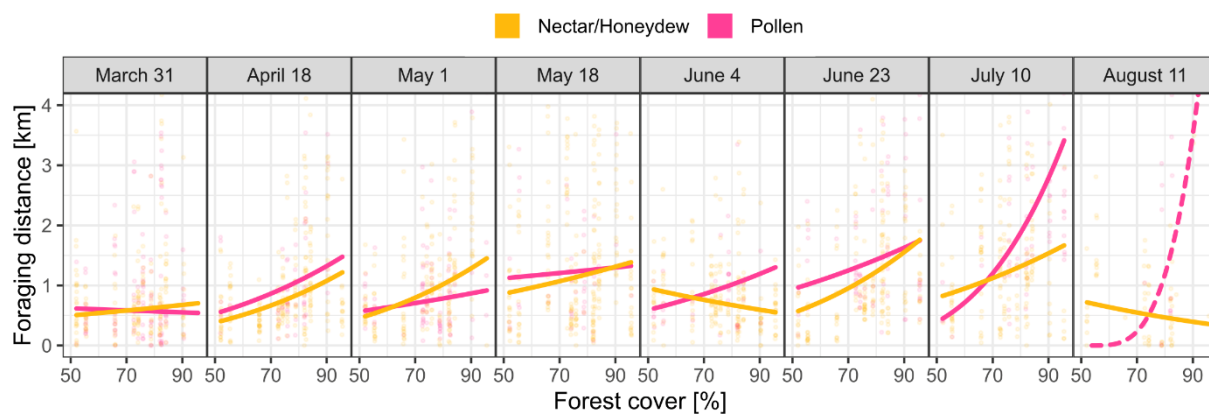
## Results

### *Foraging distances in relation to season, forage type, and forest cover*

We observed and decoded a total of 2022 waggle dances performed by the twelve colonies residing in the forest (mean dances analysed per observation day 253 +/- 72, range: 82-307). While most dances (1497 dances, 74 %) were performed for carbohydrate sources (nectar/honeydew), 525 dances (26 %) were performed for pollen. 81 % of the advertised food locations were less than two kilometres away from the colonies. The overall median, mean and maximum foraging distances, as inferred from waggle dances, were 861 m, 1257 m, and 9060 m, respectively, meaning that colonies potentially foraged in areas up to 82 km<sup>2</sup> around their hives. Foraging distances varied considerably between observation days ( $p < 0.001$ ; table 1 and Fig S3): They were shortest in early spring (median distance on March 31<sup>st</sup>: 592 m) and largest in midsummer (median distance on July 10<sup>th</sup>: 1466 m). Overall, foraging distance distributions for nectar/honeydew and pollen sources significantly differed, i.e. distances for nectar were significantly shorter than those for pollen sources, implying more reliable nectar than pollen supply in the vicinity (median distance for nectar/honeydew: 817 m, median distance for pollen: 988 m;  $p < 0.01$ , Fig. S4). When considering all analysed dances and all observation days together, foraging distances increased with the amount of forest in the surrounding landscape ( $p = 0.043$ ; Fig. 2): from 626 m (mean) in colonies with 52 % forest to 1215 m (mean) in colonies with 96 % forest. Importantly, the effect of forest cover on foraging distance varied strongly depending on the season and on the forage type considered (three-way-interaction between forest cover, forage type and observation day:  $p < 0.01$ ). Prior to the closing of the canopy (and therefore reduced light falling on the understory), on March 31<sup>st</sup>, forest cover had no effect on foraging distances, and colonies generally foraged in their closer surroundings for both pollen and nectar. On five of the remaining seven observation days (April 18<sup>th</sup>, May 1<sup>st</sup>, May 18<sup>th</sup>, June 23<sup>rd</sup>, July 10<sup>th</sup>), foraging distances for both pollen and nectar/honeydew increased with forest cover, signifying resources outside the forest or at the forest edges. However, on June 4<sup>th</sup> and August 11<sup>th</sup>, forest cover differentially affected foraging distances for pollen and carbohydrate sources: while pollen foraging distances increased with forest cover, we observed a trend for shorter nectar/honeydew foraging distances in hives with more forest in the surroundings, indicating carbohydrate sources but not pollen availability inside the forest.

**Table 1:** Effects of season (observation day), forage type (nectar/honeydew versus pollen) and forest cover on foraging distances as inferred from waggle dances. Results of a three-way ANOVA (Type II with Kenward-Roger's method) with two-way- and three-way-interactions are given (N=2022 dances).

factor	F value	d.f.	p value	
<b>main effects</b>				
observation day	29.00	7	< 2.2e-16	***
forage type	8.21	1	0.0042	**
forest cover	5.34	1	0.0431	*
<b>interactions</b>				
observation day × forage type	3.49	7	0.0010	**
observation day × forest cover	7.00	7	2.9e-08	***
forage type × forest cover	0.05	1	0.8267	
observation day × forage type × forest cover	3.27	7	0.0018	**



**Figure 2:** Influence of forest cover on foraging distances on different dance observation days for the different forage types, nectar/honeydew and pollen. Predictions from the model are given as coloured lines. On August 11<sup>th</sup>, the prediction curve for pollen is dashed as only four colonies were dancing for pollen sources.

#### *Habitat preferences in relation to forage type, season, and forest cover*

We identified a model with the explanatory variable habitat type and its interactions with forest cover, forage type and observation day as the best model to describe the spatial foraging patterns (see SM, table S1, for an AIC-based comparison of different models). Bees significantly preferred certain habitat types over others (main effect of habitat type on the relative number of advertised food locations per unit area,  $p < 0.001$ , table 2). Habitat type preferences were more pronounced during pollen foraging than during nectar foraging (two-way interaction between habitat type and forage type,  $p < 0.01$ , Fig. 3A and 3B). While the majority of dances pointed to deciduous forests, these areas were less often advertised by the bees than expected given their landscape wide proportion (expected use for nectar:

64.0 %, observed use for nectar: 57.2 %, here and in the following, the reported proportional habitat use values are median values over all colonies, Fig 3A). Especially when looking at pollen collection, deciduous forests were less frequented than expected (expected use for pollen: 65.7 %, observed use for pollen: 45.2 %, Fig. 3B). Coniferous forests were about as attractive for both nectar and pollen collection as expected and were only slightly under-used (expected use for nectar: 6.0 %, observed use for nectar: 5.9 %; expected use for pollen: 6.0 %, observed use for pollen: 5.3 %). Both croplands and grasslands were visited much more often than expected (expected cropland use for nectar: 11.0 %, observed cropland use for nectar: 14.7 %; expected cropland use for pollen: 11.0 %, observed cropland use for pollen: 18.5 %; expected grassland use for nectar: 9.5 %, observed grassland use for nectar: 11.8 %; expected grassland use for pollen: 9.6 %, observed grassland use for pollen: 16.3 %). Settlements were under-used during both nectar and pollen collection, but they only made up a small fraction of the landscapes surrounding our study colonies.

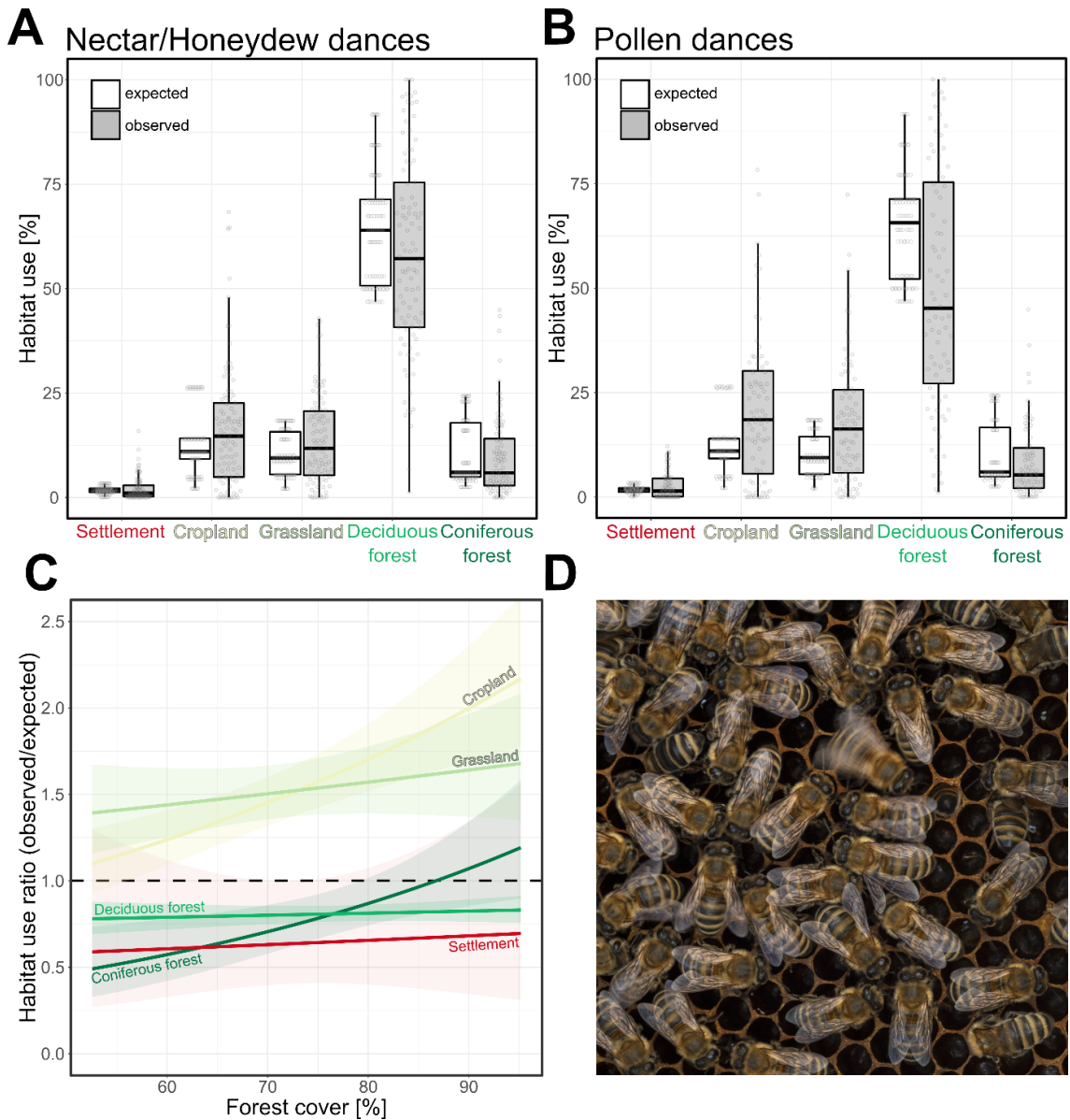
**Table 2:** Effects of habitat type, forage type (nectar/honeydew versus pollen), season (observation day) and forest cover on the relative number of foraging sites per unit area as communicated by waggle dances. Results of three-way ANOVA (Type II Wald chi-square tests) with two-way- and three-way-interactions are given (N=1938 dances).

<b>factor</b>	<b>Chi-sq</b>	<b>d.f.</b>	<b>p value</b>	
<b>main effects</b>				
<b>habitat type</b>	204.91	4	<2.2e-16	***
<b>interactions</b>				
<b>habitat type × forage type</b>	16.08	4	0.0029	**
<b>habitat type × observation day</b>	102.37	28	2.0e-10	***
<b>habitat type × forest cover</b>	15.62	4	0.0036	**

Taking the different observation days into account, we found strong spatio-temporal shifts in habitat use (interaction between habitat type and observation date,  $p < 0.001$ , Fig. S5). In spring, colonies first shifted foraging preferences from forests (March 31<sup>st</sup>) to grassland and cropland (April 18<sup>th</sup> and May 1<sup>st</sup>), and then shifted back to forage in forests at the expected level (May 18<sup>th</sup>). In summer, colonies continued foraging for nectar/honeydew in forests at the expected frequency (on June 4, June 23<sup>rd</sup> and on August 11<sup>th</sup>), but clearly under-used deciduous forests for pollen collection from June onwards (Fig. S5). In August, pollen foragers also disproportionately danced for settlements, indicating that pollen sources from residential areas were attractive late in the season when pollen was scarce in other habitats. Importantly, forest cover itself also had a significant influence on the colonies' habitat use

( $p < 0.01$ , Fig. 3C): the frequency of foraging in cropland, grassland and coniferous forests increased with higher proportions of forest cover in the surroundings, highlighting again the importance of crop- and grasslands, but also of coniferous forest, for colonies residing in forest-dominated landscapes.

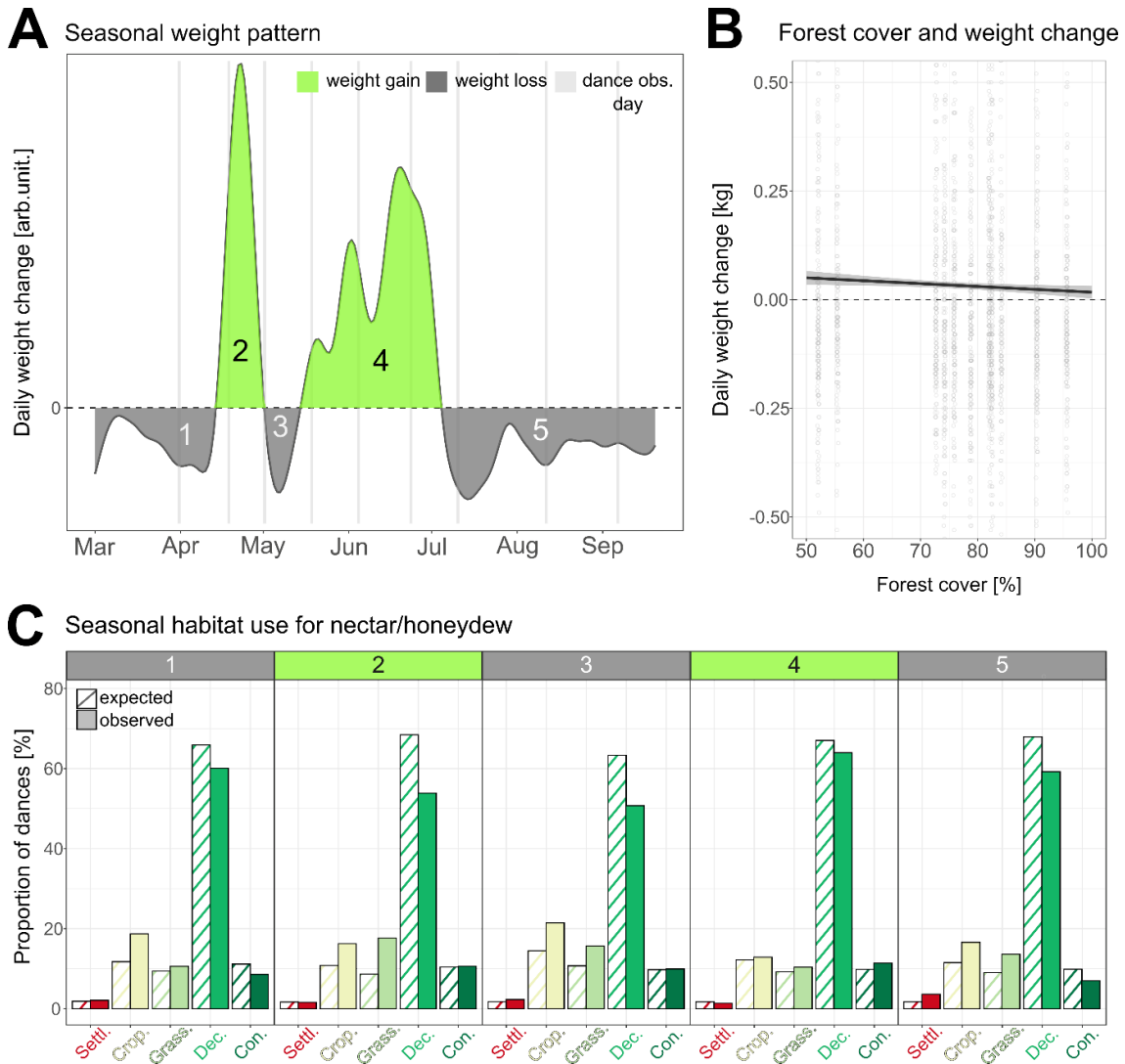
**Figure 3:** Expected (white) and observed (grey) habitat use of the five major landcover types in the study



region during the foraging season (March to August) for nectar/honeydew dances (N=1441) (A) and for pollen dances (N=497) (B). (C) Influence of forest cover on the bee's habitat preference for all decoded dances. Coloured regression lines show model predictions and 95 % confidence intervals. (D) Forager performing a waggle dance inside the observation hive (upper right quarter, bee with blurred abdomen). Photo taken by Ingo Arndt.

### **Contribution of different habitat types to colony weight gain**

Hive scale recordings revealed five distinct phases of colony weight change (phases 1–5 in Fig. 4A and 4C). The two phases of colony weight accumulation (net nectar/honeydew influx into the colonies) occurred between mid-April and beginning of May and between mid-May and the beginning of July, respectively. Forest cover had no effect on daily colony weight changes ( $p=0.28$ , Fig. 4B and Fig. S6). Combining the spatial information from waggle dances for nectar/honeydew sources and colony weight change data, we estimated the proportional contribution of the different habitat types to the carbohydrate intake for each of the five weight change phases (Fig 4C). During the first two-week phase of net weight gain, between mid-April and beginning of May (phase 2), colonies clearly over-used agricultural areas and grasslands (including orchards) suggesting that they foraged on fruit trees and oilseed rape. While the average daily weight increase in these two weeks was highest (0.39 kg/day), the contribution to the total yield in this phase was 32 %. In the second phase of net weight gain, between mid-May and July (phase 4), 68 % of the total yield was accumulated, albeit the average daily weight gain was only 0.23 kg/day. Interestingly, in this phase the visitation of the different habitat types was almost as expected, with coniferous forest areas now being slightly over-used. In the three phases of net weight loss (phases 1, 3 and 5) nectar/honeydew foraging was clearly biased towards crop- and grassland, suggesting that times of general carbohydrate scarcity might be more severe in landscapes with high forest cover. From July on (phase 5), the bees also showed a preference for residential areas which were generally further away from the colonies than any other habitat type.



**Figure 4:** (A) Seasonal pattern of colony weight changes. We averaged the values from all colonies and used generalized additive modelling (GAM) to identify times of major nectar/honeydew gain (and loss) in the whole region. Positive values represent times of weight gain (highlighted in green), negative values (dark grey) show phases of colony weight loss. Dance observation days are marked as light grey vertical lines. Phase 3 represents a period of unfavourable weather conditions. (B) Daily colony weight changes across the foraging season in relation to forest cover. (C) Habitat use for carbohydrate intake inferred from dances for nectar/honeydew sources for the five phases. The striped bar plots represent the expected proportion of dances for a certain habitat and phase. The filled bar plots illustrate the observed proportions.

## Discussion

In this study we analysed honey bee foraging and colony performance in central European forest landscapes. Colonies foraged over larger distances in landscapes with higher forest cover, showed stronger preferences for remaining non-forest habitats, and were more

limited in pollen than carbohydrate resources. Seasonal variation in foraging distances, and long periods with colony weight losses revealed that managed forest lack key resources for wild-living honey bee colonies, thereby underpinning the need for more pollinator-friendly management of temperate forest ecosystems.

Previous studies decoding waggle dances of honey bees placed in mixed European agricultural landscapes demonstrated that the bees need to fly larger distances to collect pollen in simple compared to complex landscapes (Steffan-Dewenter and Kuhn, 2003), that they preferentially forage in semi-natural habitat and on mass-flowering crop fields (Bänsch et al., 2020; Danner et al., 2016; Couvillon et al., 2014), and that flower-rich urban areas can provide better bee forage than rural land (Samuelson et al., 2021). We here complement this knowledge by exploring the value of woodland as foraging habitat for honey bees.

While the overall mean foraging distance of 1257 m in the Steigerwald forest region was within reported limits (from 740 m to 1526 m; (Bänsch et al., 2020; Danner et al., 2016; Samuelson et al., 2021; Couvillon et al., 2015; Steffan-Dewenter and Kuhn, 2003), the average foraging distance of colonies located deep inside forests was twice as large as that of colonies in landscapes with only about 50 % forest cover. Furthermore, mapping dance-communicated foraging locations revealed that bees foraged significantly less frequently in forests than expected given their landscape-wide availability. This shows that beech-dominated forests in Central Europe generally provide inferior forage resources for honey bees than open habitats. However, we also found that one needs to differentiate between time of the year and forage type when evaluating the value of forests for honey bees.

Early in the season (in March), colonies foraged locally and used forests at the expected level for both pollen and nectar collection, indicating that spring-flowering plants on the forest floor offer important resources for honey bees after hibernation. After tree canopy closure in April, however, foraging distances increased, forest cover affected foraging distances and forests were less frequented than expected, indicating scarcity of forage in woodlands (Heinrich, 1976; Ginsberg, 1983; Mandelik et al., 2012). In line with studies conducted in agricultural landscapes (Couvillon et al., 2015; Garbuzov et al., 2020), we found that bees had to fly furthest in summer (here: July), and a steep increase in foraging distances with forest cover indicated that the lack of forage during the ‘summer blues’ is even exacerbated in forests compared to open habitat.

Beyond the effect of season, the use of forest strongly depended on forage type. Distinct distance distributions of pollen and nectar/honeydew foraging locations suggested that the two forage types were collected at different habitat patches. Clearly, the overall larger foraging distances for pollen and the significantly stronger preference against forests during pollen foraging than carbohydrate foraging showed that especially pollen forage is scarce in forests. While nectar and honeydew are stored in large quantities in the hive, pollen stores last no longer than a few days and thus need to be supplied continuously (Seeley, 1995). Therefore, the lack of pollen supply in forests might be notably detrimental in late summer when colonies require protein to raise winter bees (Winston, 1991) but face difficulties finding it.

The bees' under-use of forests was slightly less pronounced during nectar foraging. In two occasions, nectar foraging distances even decreased with increasing forest cover, suggesting nectar availability: In the beginning of June, blooming trees like lime, black locust and chestnut or shrubs like raspberry might have produced nectar, and the woolly beech aphid (*Phyllaphis fagi*) could have been a source of honeydew (Schmutterer, 1952, personal observations). Likewise, in August, honey bees probably collected honeydew produced by aphids on coniferous tree species. Although these phases of forest-borne honey flow were temporally restricted, the connection of dance information and colony weight recordings revealed that their contribution to the overall colony weight gain was substantial. While the first phase of net weight gain in April was clearly attributable to mass-flowering oilseed rape and fruit trees, during the second weight gain phase, from mid-May to July, 75 % of nectar dances communicated forest locations, and this phase contributed to an estimated 65 % of the total weight yield of the colonies. It is this early summer nectar flow that explains why colony performance, measured as the average daily weight gain, was not negatively affected by forest cover. Since beekeepers know about the melliferous potential of forest, they seasonally monitor plant-sucking insects and move their hives into forests to exploit temporal honey dew flows (Ruppertshofen, 1995; Liebig, 1999; Prešern et al., 2019). These insights also show that a certain amount of forest in the landscape will be beneficial for bees due to the supply of complementary carbohydrate resources during short phases of the year, even though forests do not continuously provide resources (Mitchell et al., 2014; Eeraerts et al., 2021). We show that combining dance decoding with daily weight recordings offers the unique opportunity to evaluate the quality of different habitats and



directly assess resource availability and identify bottlenecks or interruptions in food supply in different landscapes (Schellhorn et al., 2015).

Large beech-dominated forests in Central Europe may be a suboptimal habitat for honey bees and other flower-visiting insects when considering the whole foraging season. In fact, the lack of continuous forage might partly explain why the densities of wild-living honey bee colonies in Central European forests are low (Kohl and Rutschmann, 2018; Requier et al., 2020) compared to those in species-rich forests in the Northeast USA (1 colony km<sup>-2</sup>, Seeley, 2019), or in historic tree beekeeping regions in Russia (0.5 colonies km<sup>-2</sup>, Galton, 1971). However, our data imply that simple interventions could have great potential for releasing resource limitations in managed forests. The distinct under-use of forests after canopy closure indicates that the main driver of food scarcity in the studied forests is low light permeability, lack of open areas and the dominance of a few, mainly wind-pollinated tree species. Therefore, if it is the goal to promote populations of bees and other pollinators within managed forests, we recommend supplementation of forest stands by native insect-pollinated trees (e.g., willows, linden, maple, cherry, alder buckthorn, mountain ash and sweet or horse chestnut) and identifying and promoting forest-dwelling plant species of the herb and shrub layer, especially those that produce pollen in late summer. Moreover, pollinator-friendly management should focus on transitional structures with open areas (e.g., clearings and roadside corridors) in closed forest landscapes. At larger scales, we propose the creation of landscapes with a high diversity of different forest types and open habitats.

### **Acknowledgments**

We thank Ulrich Mergner and the state forest department in Ebrach for permitting us to do this research in the Steigerwald forest. We thank Jürgen Tautz for providing beekeeping material and other resources. We are indebted to Gaby Läbisch, who helped managing the study colonies prior to the experimentation period. We are grateful to Johann Kaderschabek for manufacturing the observation hives. We are obliged to Susanne Schiele and Gemma Villagomez for their support with the honey bee queens and together with Thomas Igerst for their help with the weight scales. We thank Sebastian Roth and Judith Kleiß who gave valuable comments on the manuscript. We are aware of having negatively impacted the environment through carbon emissions associated with driving between the hives during

observation days. Hence, one of the authors has privately compensated the estimated 2 tons of CO<sub>2</sub> emissions.

## Supplementary material

### *Observation hives and honey bee colonies*

We used custom-built observation hives (Fig. 1B) with a glass window on one side. The entrance of the hive was constantly narrowed down to about 3 cm width and a wedge behind the entrance guided homecoming bees to the window side of the first comb. Outside observation hours, this window was covered by a wooden board. If necessary, any holes between the observation comb and its frame were plugged with little wax pieces right before each video recording session. These measures hindered the bees from easily switching sides near the entrance to regions that could not be filmed. Hence, we were able to record most waggle dances. During video recording, we used a light-proof cloth spread out over the hive to prevent direct sunlight from entering the colony (Fig. 1B). Four colonies at three locations were replaced before or during the main experimentation period in 2019: two after winter due to small colony sizes (colonies at locations A and C) and two after the swarming season due to the absence of a laying queen (colony E in May and colony C in August).

### *Setup of the cameras and dance sampling scheme*

We simultaneously recorded dancing bees in all twelve study colonies using digital camcorders (Sony HDR-CX240E, Panasonic HC-X929 and Panasonic HC-V707; frame rate: 25 Hz). For this purpose, we drove from one study site to the other by car and set up the tripods and cameras for recording. A metal chain attached to the observation hive within the view of the cameras served as a vertical reference for the later video analysis. The internal clock of the cameras was set before filming and the actual time was spoken on the video when we started recording. Filming lasted around six hours per colony and observation day until the external power banks were empty. We reversed the order of setting up the cameras at the sites between observation days.

For every colony, we screened the videos of an observation day in such a way that the sampled dances were distributed across the whole videorecorded period (time of observed dances: between 09:04-17:20). Except for two camcorders, all cameras split the recorded videos into segments of 12 min and 35 seconds. For these ten cameras the following scheme was applied: we started the screening for dances with the second recorded video segment. The first five dances from different foraging bees in this video segment were analysed. We then skipped two video segments (~27 minutes) and screened the fifth segment of the day

for the next five dances. In case not enough dances could be found in a focal segment, we also screened the subsequent video segments that would have been skipped otherwise. Screening was stopped when around 25 dances were analysed per colony and observation day. An equivalent temporal sampling scheme was applied for colonies recorded with the two cameras that used a different video segmenting. We could not completely exclude the possibility that one individual bee was analysed twice in different videos on the same observation day. However, since a colony has foragers in the range of thousands, repeated sampling should have rarely occurred and therefore we treated every dance as independent.

#### *Notes on forage mapping*

Dance decoding is afflicted by several types of noise. Errors stem from both the variability in behaviour of the bees and the decoding method (Schürch et al., 2013, 2016; Tautz, 2022). Therefore, individual dances indicating locations very close to the hives could be translated into negative foraging distances. Negative foraging distances were set to a foraging distance of 1 meter to avoid successive problems with non-positive values in the models. Also, we avoided including the first and the last circuit of a dance due to the increased variability of waggle runs therein (Couvillon et al., 2012).

Since the error in forage mapping increases with the foraging distance (Fig S1), we only considered dances indicating foraging distances up to 4 km from the hives for the analyses of habitat use (N=1938 out of 2022 dances).

#### *Calculating distance-corrected land cover and expected habitat use*

So far, most ecological studies on honey bee foraging considered circular landscapes surrounding colonies with a fixed radius (often two kilometres, e.g. (Steffan-Dewenter and Kuhn, 2003; Danner et al., 2016; Samuelson and Leadbeater, 2018) to quantify the proportion of different land use types that might affect foraging patterns and to determine expected habitat use. However, this method neglects that different land cover types often have an unbalanced spatial distribution within the considered landscapes, and that habitat patches close to the hive are more important in influencing foraging patterns than areas far from the hive. Recently, some dance decoding studies implemented a distance correction to determine the most profitable habitat types based on waggle dance decoding without overestimating the importance of habitat types that are, by coincidence, proximate to the hive (Henry et al., 2012; Couvillon et al., 2014; Stange et al., 2017; Samuelson et al., 2021).

We chose a similar but novel approach to calculate distance-corrected land cover proportions and the expected habitat use. From the overall distribution of foraging distances based on all waggle dances decoded in our study (Fig. S2A) we first derived a three-parameter log-logistic function that describes the visitation probability of a given habitat patch in relation to its distance from the hive (Fig. S2B). Since we sampled dances from multiple colonies placed in differently structured landscapes and at different times of the season, this visitation probability distribution should closely predict how a colony would distribute its foragers in a hypothetical landscape in which equal, average-quality food patches are homogeneously distributed. In such a landscape, the distribution of foragers would merely be the result of the scout bees' innate search behaviour (which determines the probability of finding a food patch in relation to its distance from the hive), the distance-dependent probability of recruits to find an advertised patch, and the distance-dependent net energetic return during foraging (a forage patch closer to the hive provides higher net profit and thus is visited more frequently than an equal patch further from the hive). We used this visitation probability function to differently weight the contribution of habitat patches in the study landscapes based on their distance from the hive so that patches further away contributed less to the expected habitat use.

To classify the study landscapes, we used a recently published pan-European land cover data set derived from satellite images with a ten meters resolution (Weigand et al., 2020). The land cover types present in our study area were deciduous forests, coniferous forests, cropland, grassland, settlements, open soil, and water bodies (the latter two habitat types represented less than 0.2 % and were neglected for the final analysis). We divided the landscapes into equally sized square raster cells of 80m<sup>2</sup> ("st\_make\_grid" command from sf package) and calculated for each raster cell within an area of 4 km from the hive the proportional area of each of the five major land cover types. The distances from the hive to the centroids of the raster cells were taken to calculate the visitation probabilities for each raster cell using our visitation probability function (Fig. S2B). For each raster cell the proportional area of each land cover type was multiplied by the visitation probability. Then, the values for each of the five land cover types were summed up over all raster cells of a landscape and divided by the total (to normalize it). In this way we obtained for each study landscape a proportion of each of the five land cover types that summed up to one. These distance-corrected proportions can be considered as the effective land cover proportions for the central place foraging honey bees. They also represent the expected contribution of each

land cover type to honey bee colonies' food intake under the null hypothesis that all land cover types provide the same forage (Fig. 1C). Our approach of directly implementing a distance-correction into the calculation of expected habitat use has the advantage that expected and observed habitat use can be compared and any deviations are directly interpretable as preferences for/against a certain landscape type.

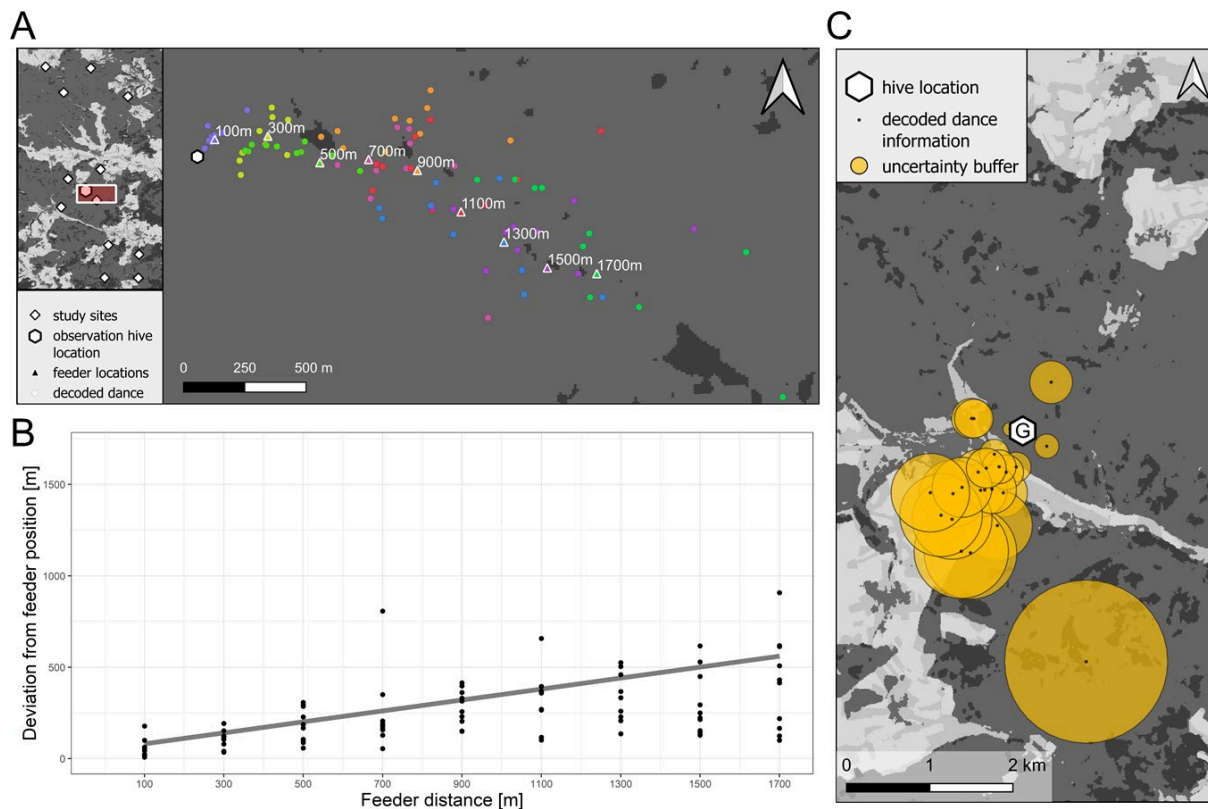
R packages used for these analyses include: `sf` (Pebesma, 2018), `sp` (Pebesma and Bivand, 2005; Pebesma et al., 2012), `raster` (Hijmans et al., 2015), `exactextractr` (Baston, 2020), `data.table` (Dowle et al., 2019). The tidyverse “architecture” (Wickham, 2017) was used for data transformation, `ggplot2` (Wickham, 2016) and `patchwork` (Pedersen, 2020) to create the figures. Individual changes to the plots were done with Inkscape 1.0.2-2 (Inkscape Project, 2020) and `gimp` 2.10.18 (The GIMP Development Team, 2020). All statistical analyses were performed in RStudio (RStudio Team, 2020) with R 4.0.3 (R Core Team, 2016).

**Table S1:** Selection of the best model of habitat use based on AIC. The models are ordered according to their  $\Delta$ AIC values. Models with factor combinations that did not converge were not considered.

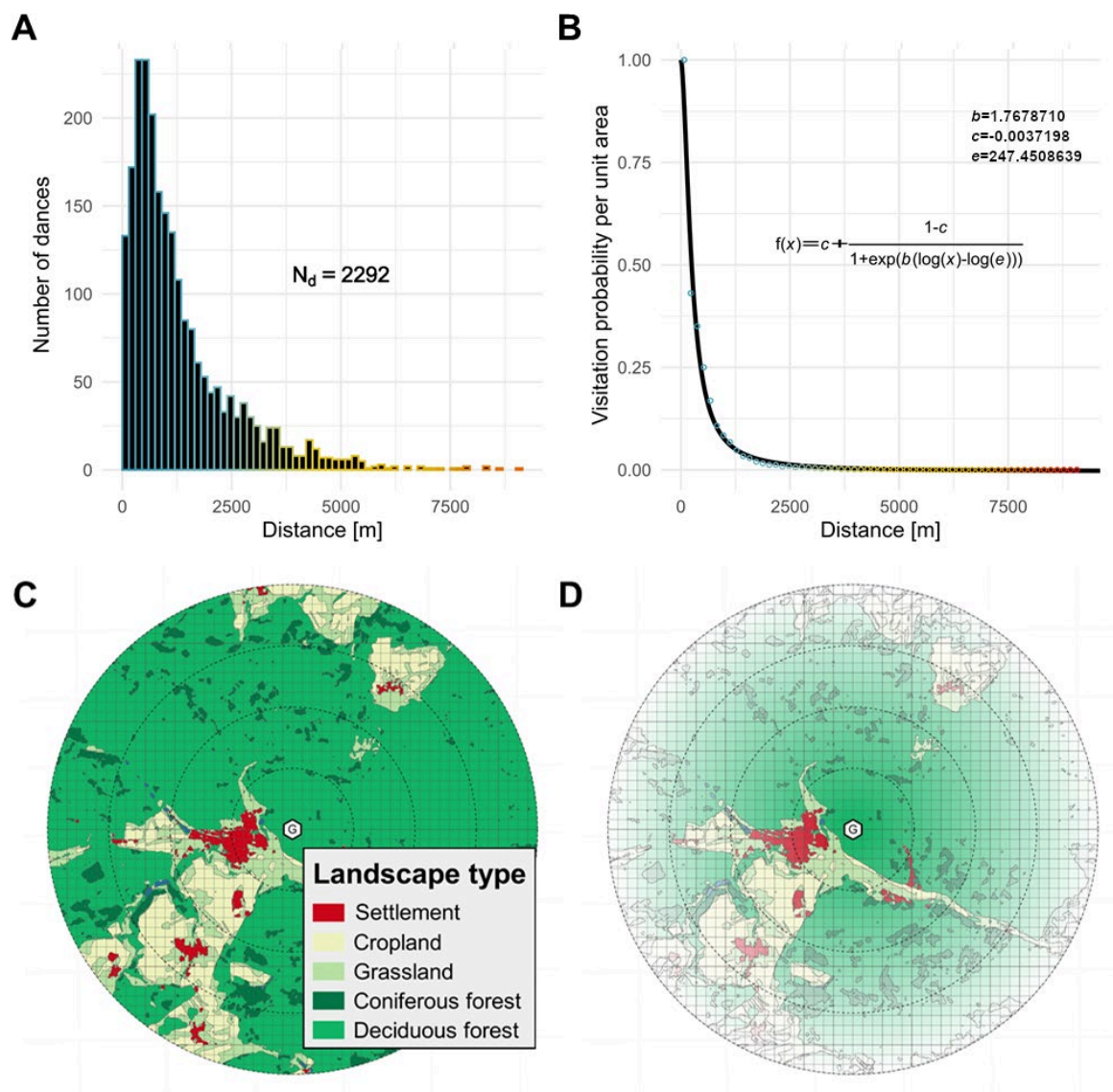
Model	df	$\Delta$ AIC
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:observation_day + habitat_type:forage_type	52	0.0
habitat_type + forest_cover + observation_day + forage_type + habitat_type:observation_day + habitat_type:forage_type	48	7.3
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:observation_day + habitat_type:forage_type + habitat_type:forest_cover:forage_type	57	7.8
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:observation_day + habitat_type:forage_type + habitat_type:forage_type:forest_cover	57	7.8
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:observation_day + habitat_type:forage_type + habitat_type:forest_cover:observation_day	87	9.7
habitat_type + forest_cover + observation_day + forage_type + habitat_type:observation_day	44	16.1
habitat_type + forage_type + observation_day	47	16.6
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:observation_day + habitat_type:forage_type + habitat_type:forest_cover: observation_day + habitat_type:forest_cover:forage_type	92	17.9
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:observation_day + habitat_type:forage_type + habitat_type:forest_cover:observation_day + habitat_type:forest_cover:forage_type	92	17.9
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:observation_day + habitat_type:forage_type + habitat_type:forest_cover:observation_day + habitat_type:observation_day:forage_type	122	19.5
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:observation_day + habitat_type:forage_type + habitat_type:forest_cover:observation_day + habitat_type:observation_day:forage_type	122	19.5
habitat_type + forest_cover + forage_type + habitat_type:forest_cover + habitat_type:forage_type	17	23.3
habitat_type * observation_day	42	24.0

habitat_type + forage_type + observation_day	43	26.0
habitat_type + forest_cover + forage_type + habitat_type:forage_type	13	26.6
habitat_type * forest_cover	12	27.1
habitat_type + forest_cover + forage_type + habitat_type:forest_cover	13	29.0
habitat_type + forest_cover + forage_type + habitat_type:forest_cover + habitat_type:forage_type + habitat_type:forest_cover:forage_type	22	30.4
habitat_type + forest_cover + forage_type	9	32.9
habitat_type + forage_type + observation_day	82	34.1
habitat_type + forage_type + observation_day	82	34.1
habitat_type * forage_type	12	34.3
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover + habitat_type:forage_type	24	37.0
habitat_type + forest_cover + forage_type + habitat_type:forest_cover + habitat_type:forest_cover:forage_type	18	37.8
habitat_type	7	39.0
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forage_type	20	40.3
habitat_type + forest_cover + observation_day + forage_type + habitat_type:forest_cover	20	42.7
habitat_type + forest_cover + observation_day	15	44.7
habitat_type + forest_cover + observation_day + forage_type	16	46.6
habitat_type + forage_type + observation_day + habitat_type:forage_type	19	48.1
habitat_type + forage_type + observation_day	15	54.9
habitat_type + forest_cover + observation_day + habitat_type:forest_cover + habitat_type:forest_cover:observation_day	54	57.9

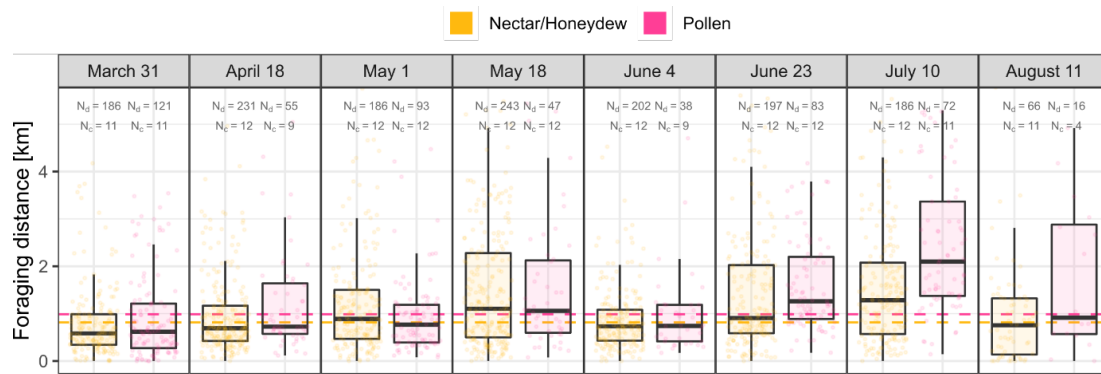




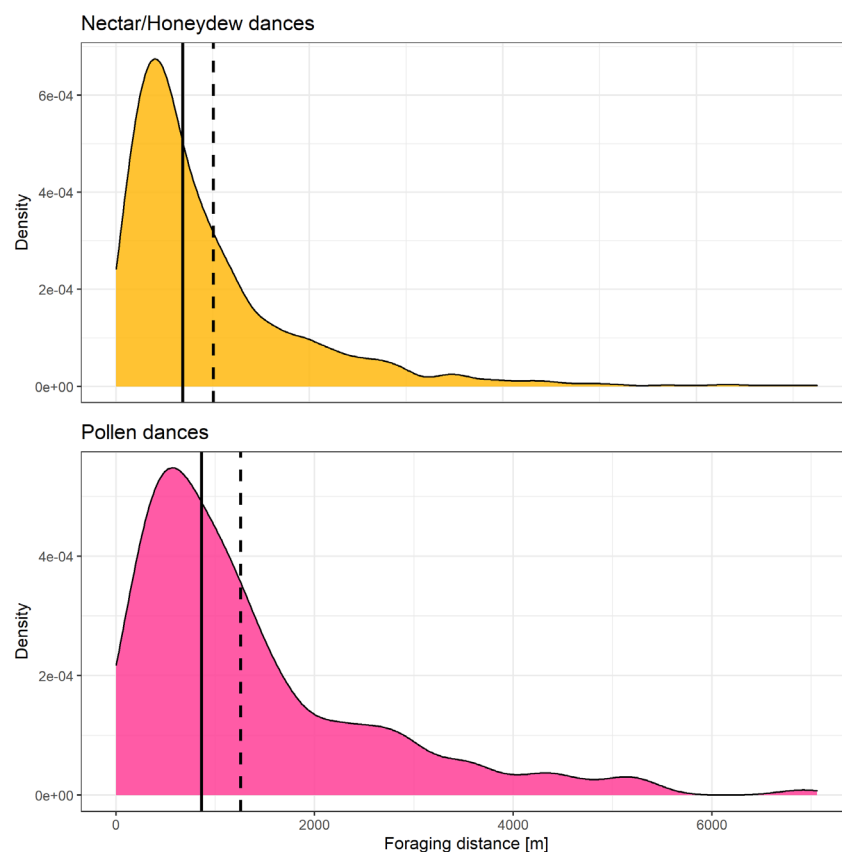
**Figure S1:** (A) Map overview of the feeder training experiment performed to calibrate the distance-related increase in waggle and return phase duration of the bees used in this study (Kohl and Rutschmann 2021). The observation hive used for the feeder experiment is illustrated by a hexagon and the study sites used for this study are depicted by diamond symbols (map overview, left). The feeders (triangles) were located along a forest road at distances between 100 m and 1.7 km from the observation hive (right). The dots denote locations communicated by the waggle dances performed by the bees visiting the feeder stations (dance locations are in the same colour as the corresponding feeding location). The dark grey map background illustrates deciduous (brighter) and coniferous forest areas (darker). (B) Spatial deviation (in meters) between the foraging locations indicated by dances and the actual locations of the feeders for nine feeders placed at increasing distances from the hive. The line fits the 75 percentiles of all the distance deviations (quantile regression produced using function “rq” from the “quantreg” package in R (Koenker et al., 2018)). We used this regression function to calculate uncertainty buffers for dance-inferred natural foraging locations. (C) Example of a forage map (study colony G on 18<sup>th</sup> of May 2019, 25 dances). Back dots are the dance-decoded foraging locations, and the yellow areas are the respective uncertainty buffers. The radii of the buffers increase linearly with the distance to the hive. We considered the average proportions of land cover types within the buffer areas as the observed proportional habitat use.



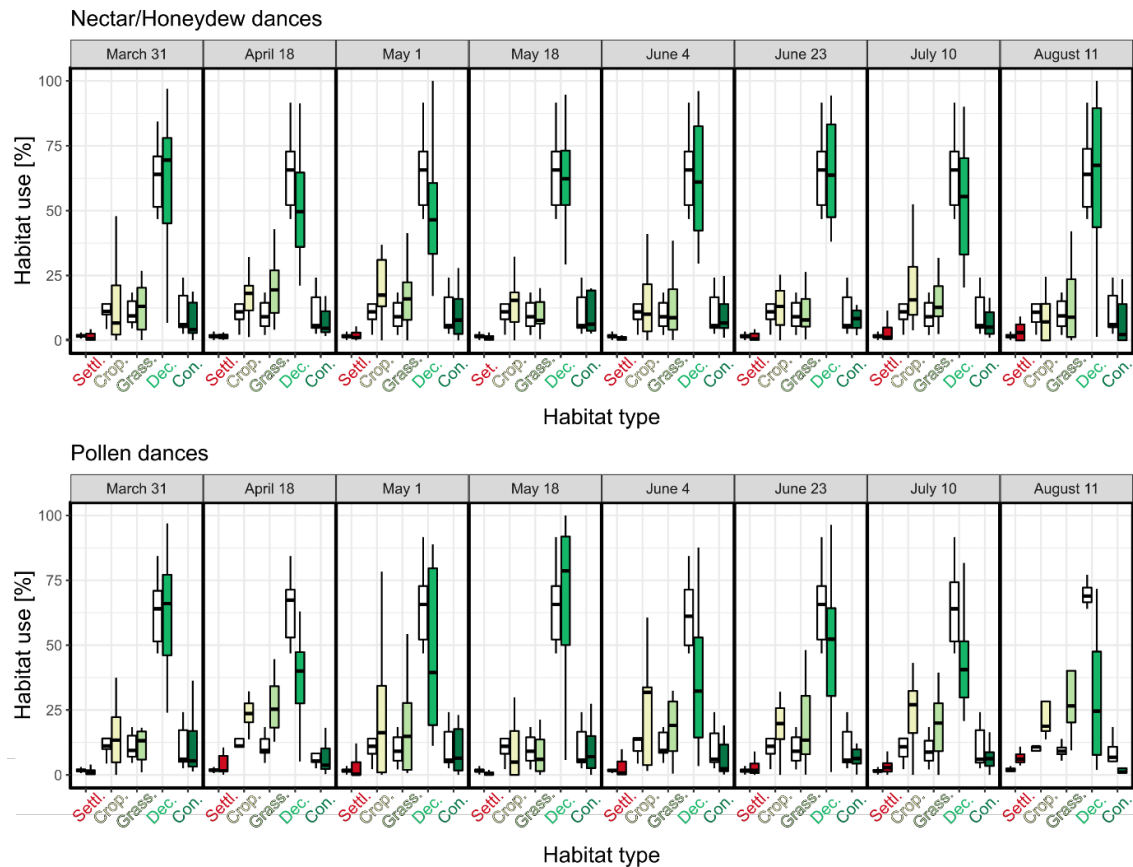
**Figure S2:** (A) Histogram of all foraging distances inferred from dances performed by bees in the Steigerwald in 2018 and 2019 (bar width=150 m). For each distance class (bar) in the histogram, the corresponding foraging area was calculated (area within the distance of the right margin minus the area within the distance of the left margin of a bar). (B) We derived a distribution of visitation probability depending on distance from the hive by dividing the number of dances per distance class by the area corresponding to that distance class and rescaling to obtain values between 0 and 1 (open dots, colours match the colours of histogram bars in A). A three-parameter log-logistic function fits the visitation probability as a function of distance well (black line). (C) Example of a landscape surrounding an observation hive (study colony G). Dashed circles display 1, 2, 3 and 4 km buffers around the colony. (D) Graphical representation of a “distance-corrected landscape” (again, study colony G). The more transparent a raster cell, the less it contributes to the expected habitat use of the colony in the centre of the landscape due to the greater distance from the hive.



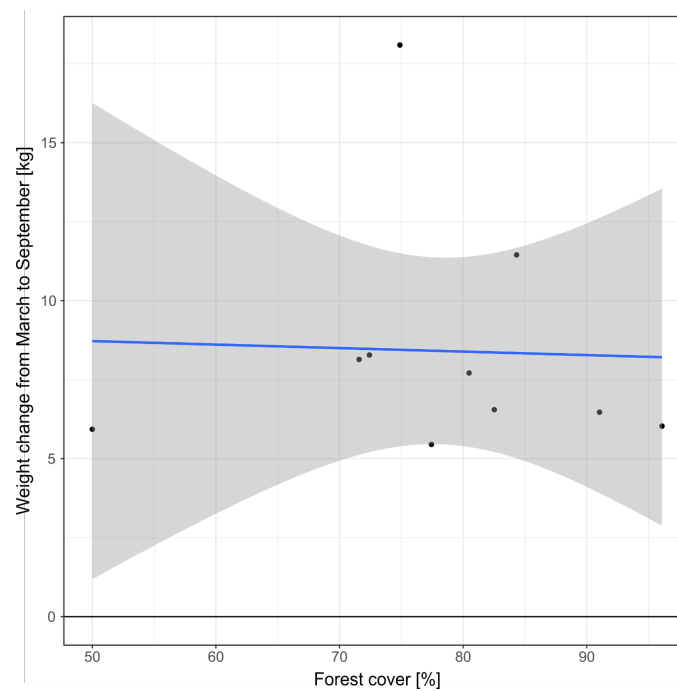
**Figure S3:** Nectar/honeydew and pollen foraging distances across the season. Nectar/honeydew is colour-coded in yellow, pollen in pink. Dashed lines give median values for each forage type calculated for the whole foraging season. Raw data are depicted in the background (extreme values exceed the y-axis range). The number of the dances (Nd) for the different experimentation days and forage types are given (both for nectar/honeydew and pollen) as well the number of colonies (Nc) from which these dances were analysed.



**Figure S4:** Advertised foraging distance distributions for nectar/honeydew and pollen. Black lines depict the median distance, dashed lines give the mean value. The means (linear mixed effect model, factor forage type,  $p=0.0042$ ) as well as the distributions of foraging distances differ significantly (Two-sample Kolmogorov-Smirnov test,  $p=0.015$ ).



**Figure S5:** Expected (white box plots) and observed (coloured in five individual colours) habitat use of the five major land cover types on the different observation days for nectar/honeydew foraging (N=1441 dances) (top) and for pollen foraging (N=497 dances) (bottom). The boxplots allow easy comparison of observed and expected habitat use.



**Figure S6:** Overall weight change from March to September 2019 for 10 colonies in relation to forest cover.





Honey bee colony residing in a hollow electric power pole. Such poles are spacious inside and provide maximum protection from predators as they are virtually indestructible. This makes them interesting subjects for the conservation of wild-living honey bees. Photo taken by Dimi Dumortier.

## CHAPTER 5: WILD-LIVING HONEY BEES IN AN AGRICULTURAL LANDSCAPE IN SPAIN

This chapter is published as:

**Rutschmann B**, Kohl PL, Machado A, Steffan-Dewenter I (2022) Semi-natural habitats promote winter survival of wild-living honey bees in an agricultural landscape. *Biological Conservation* 266: 109450.

Elsevier kindly grants the use and share of this works for scholarly purposes.

The original article is available at:

<https://doi.org/10.1016/j.biocon.2022.109450>

### Abstract

The diversity of endemic honey bee subspecies and ecotypes is at risk in Europe because modern apiculture promotes only a small number of honey bee strains. A crucial step for the conservation of honey bee diversity is the assessment of the status of remaining wild populations and their limiting factors. Here we present a two-year census of native, wild-living honey bees inhabiting power poles in an intensive agricultural landscape in Galicia, NW Spain. The autumn colony densities were at least 0.22 and 0.17 colonies/km<sup>2</sup> and winter survival rates were 59 % and 26 % for the years 2019 ( $N = 29$ ) and 2020 ( $N = 23$ ), respectively. Both the initial occurrence and the subsequent winter survival of the colonies were positively correlated with increasing proportions of wood- and shrubland in the surroundings in both study years. These observations highlight the importance of semi-natural habitats for the conservation of wild-living honey bees.

## Introduction

The abundance of insect pollinators is in decline in several European countries, posing a risk to the stability of the important ecosystem function of pollination (Aizen et al., 2019; Potts et al., 2010). In this context, losses of managed Western honey bee (*Apis mellifera*) colonies have gained disproportionately high attention (Garibaldi et al., 2013; Geldmann and González-Varo, 2018), although they have largely different causes than the decline in wild pollinators. While wild pollinators primarily suffer from reduction and fragmentation of habitat and land use intensification (Steffan-Dewenter, 2003; Winfree et al., 2009; Kennedy et al., 2013; Tonietto and Larkin, 2018), managed honey bees receive critical resources from beekeepers. The latter are rather threatened by novel pathogens and parasites which easily spread, aided by migratory beekeeping and the global trade of the bees and their products (Genersch, 2010; Meixner et al., 2015; Brosi et al., 2017; Beaurepaire et al., 2020). Furthermore, it has been shown that trends in managed honey bee stocks strongly depend on socioeconomic factors and vary between regions (Smith et al., 2013; Moritz and Eler, 2016; Herrera, 2020). For example, while managed honey bee populations are in decline in temperate European countries since the 1960s, they are strongly increasing in many southern European countries in the Mediterranean basin (Herrera, 2020). Given that the concentration of large numbers of commercial honey bee colonies in apiaries can negatively affect wild pollinator populations *via* competition for food (Lindström et al., 2016; Herbertsson et al., 2016; Magrach et al., 2017; Mallinger et al., 2017; Henry and Rodet, 2018; Herrera, 2020 but see Steffan-Dewenter and Tschardtke, 2000) or the dissemination of infectious diseases (Fürst et al., 2014; Graystock et al., 2015; McMahon et al., 2015; Tehel et al., 2016; Alger et al., 2019; Manley et al., 2019; Tehel et al., 2020), managed honey bees could actually exacerbate wild pollinator decline in some regions (Geldmann and González-Varo, 2018).

Ironically, the pollinators who are potentially most affected by increasing managed honey bee densities are native honey bees that are still maintained in small scale traditional apiaries or that live as wild colonies (Requier et al., 2019). This is on the one hand, because modern intensive apiculture promotes only a small number of honey bee strains, which are often allochthonous. As a consequence, introgression threatens the geographic diversity of honey bee subspecies and ecotypes with their potentially adaptive genetic variation (Muñoz, 2020). On the other hand, current apicultural practices commonly involve large-



scale transhumance aiding the dissemination of pests and parasites. Obviously, pathogens introduced with managed hives should primarily affect other honey bees (prior to other wild bees and other organisms) in any given region.

In contrast with the practices used in modern intensive apiculture, current works suggest that a key to the long-term conservation of honey bee diversity and to sustainable beekeeping, is to foster natural selection (Neumann and Blacquière, 2017; Blacquière et al., 2019; Seeley, 2019) and to promote bees of local origin (Strange et al., 2007; Büchler et al., 2014; Meixner et al., 2015). Populations of colonies which are not treated against parasites and which are allowed to vary in fitness (as opposed to survival and reproduction being dependent on the beekeepers' actions) are expected to evolve resistance against emerging pests and pathogens (Neumann and Blacquière, 2017; Blacquière et al., 2019). Furthermore, when selection pressure is high, maladaptive introgressions by foreign bees should be less likely to spread in the population (Requier et al., 2019). While the approach taken so far is to leave the bees in a quasi-wild manner (Darwinian beekeeping) (Seeley, 2019), few attempts have been made to assess the extant status of potential wild honey bee populations, probably because it is widely believed that wild or feral colonies do not exist, or because they are not easy to locate (but see (Kohl and Rutschmann, 2018; Oleksa et al., 2013b; Seeley, 2019; Browne et al., 2020).

Landscape scale variation in flower resource provision and land-use intensity has an effect on various measures of colony performance in social bees (Steffan-Dewenter et al., 2002; Steffan-Dewenter and Kuhn, 2003; Decourtye et al., 2010; Donkersley et al., 2014; Sponsler and Johnson, 2015; Lecocq et al., 2015; Alaux et al., 2017; Alburaki et al., 2017; Muñoz-Colmenero et al., 2020). Throughout the year, a honey bee colony requires a sustained supply of nectar and pollen to maintain colony functioning and to strengthen immune defences (Schmehl et al., 2014). In fact, nutrition has been identified as a crucial point in honey bee health (DeGrandi-Hoffman et al., 2010) and temporal shortages of feeding resources can decrease abundances of honey bees and other flower-visiting insects in a region (Di Pasquale et al., 2016; Requier et al., 2017). In this context, the loss of natural habitats is a major threat to pollinators (Naug, 2009; Dolezal et al., 2019). Studies with managed honey bee colonies identified semi-natural habitat and areas with diverse floral resources as predictors for colony overwintering success (Döke et al., 2019; Kuchling et

al., 2018). However, such investigations are currently lacking for wild honey bees, presumably due to their cryptic way of life (Kohl and Rutschmann, 2018).

Despite high numbers of managed hives and an intensification of apiculture in the last decades (Jones, 2004; Herrera, 2020), the Iberian Peninsula still harbours a large population of the native honey bee subspecies *A. m. iberiensis* (Pinto et al., 2012). The population is still mostly free of introgression and little affected by selective breeding (Cánovas et al., 2011; Chávez-Galarza et al., 2015, 2017) but at present, no data are available on the occurrence and number of wild-living colonies. Here we report on the population density and overwinter survival rate of wild-living honey bee colonies from Galicia based on censuses of hollow power poles before and after the winters of two successive years. Specifically, we asked: (Q1) is the occupation rate of the surveyed power poles higher in locations with larger proportions of semi-natural habitat; (Q2) does the amount of semi-natural habitat promote the subsequent winter survival of these wild-living colonies.

## Methods

### *Study region*

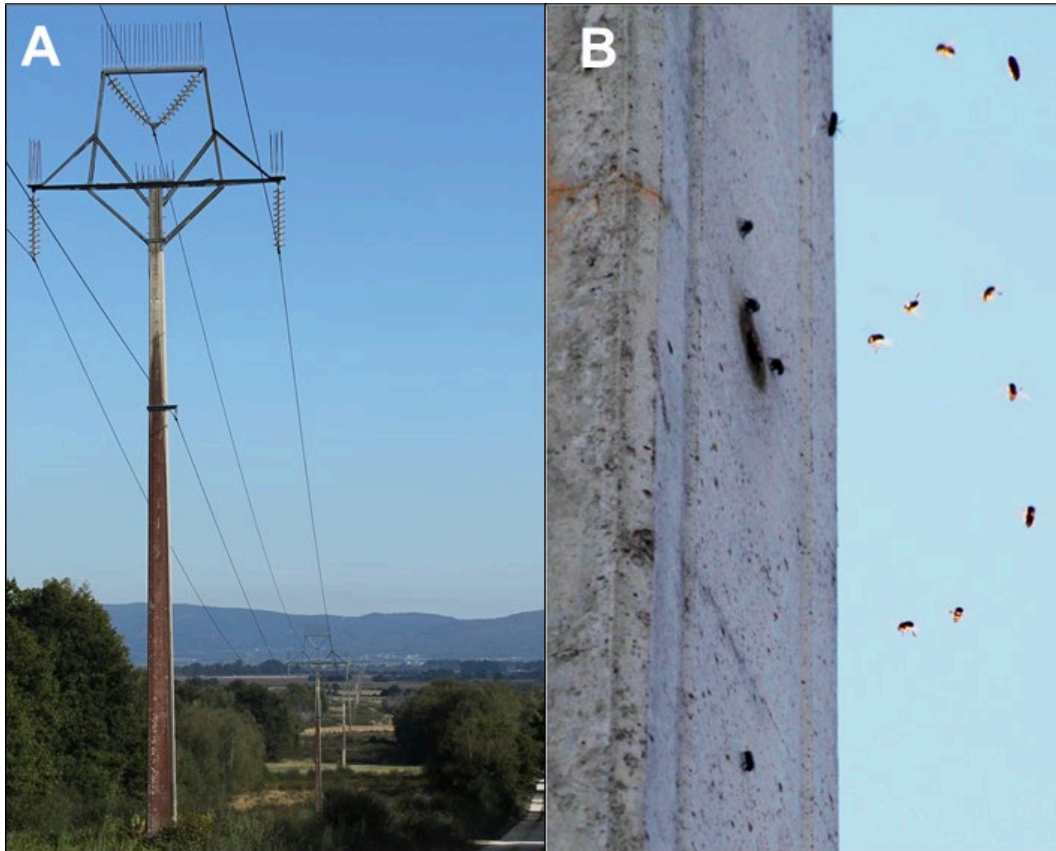
We conducted our census in the surroundings of the city of Xinzo de Limia, in the province of Ourense in Galicia, NW Spain. The region has a warm-summer Mediterranean climate with westerly winds extending the impact from the ocean. It is characterized by a yearly average temperature of 12.2 °C and a rainfall of 1133 mm, which is significantly higher than for a typical Mediterranean climate (<https://de.climate-data.org/europa/spanien/galicien/xinzo-de-limia-44326/>).

The mountainous land of the province exhibits a rich flora with elements of both the Eurosiberian and the Mediterranean floristic regions, the dominant forms of vegetation being shrubland (45 %) and forests (37 %) (see supplementary material Fig. S1). The areas not used for intensive agriculture provide ample supply of flower resources to honey bees; the most important forage plants include *Castanea sativa*, *Rubus spp.*, *Calluna vulgaris*, and *Erica*, *Cytisus*, *Ulex*, and *Genista* species (Seijo and Jato, 1998; Díaz Losada et al., 1997; Díaz-Losada, 2002; Rodríguez-Castiñeira et al., 2015). While industrial agriculture generally only makes up for a very small part of the land-use in the province (<4 %, see supplementary material Fig. S1), we worked in a flat area around the former Laguna de Antela, which was drained in 1958 and is now part of the largest coherent area of intensively

farmed land of the province (Soto et al., 2011). The main crop produced in this area is potato.

*Power pole search and honey bee colony censuses*

One of us (AM) repeatedly observed that honey bee colonies chose hollow power poles as their dwelling places. The type of poles that could serve as nesting sites was about 7 to 13 m tall and made of concrete. Since they were hollow inside and contained small entrance holes (18 mm cross-section) they offered a protected cavity of more than 1000 l; (see [http://www.prefabricadoshormigon-prephor.es/images/Pdfs/Concrete\\_Poles/technical\\_data\\_concrete\\_poles\\_for\\_overhead\\_power\\_lines\\_hv\\_and\\_hvh-20131203131141.pdf](http://www.prefabricadoshormigon-prephor.es/images/Pdfs/Concrete_Poles/technical_data_concrete_poles_for_overhead_power_lines_hv_and_hvh-20131203131141.pdf) and Fig. 1) We took advantage of the fact that these poles were frequent in the region and easy to locate. In October 2019, we surveyed the study area by driving all major roads with a car and scanning the surroundings. During good weather conditions with temperatures exceeding 15 °C, we inspected all detected poles from the ground with binoculars. Regular directional flight traffic of honey bees and pollen import were indicative of an active honey bee nest. To make an estimate of the minimum wild-living honey bee colony density, we recorded our searching track with a GPS device equipped with OpenStreetMap (OSM) and calculated the examined area based on the assumption that we were able to spot poles up to 250 m away from both sides of the road (see supplementary material Fig. S2). In March 2020 we re-examined all power poles that were occupied in the preceding autumn to determine the wild-living colonies' winter survival rate. The autumn power pole census and the subsequent test for winter survival were repeated in October 2020 and March 2021, respectively.



**Figure 1:** (A) Electricity corridor with hollow power poles comprising suitable nest sites for honey bees. (B) Close-up of a honey bee nest entrance in a power pole.

#### *Evolutionary lineage identification*

Due to a significant correlation between genetic information and wing venation patterns, wing morphometry can be used as a cost-effective and reliable identification measure for honey bees (Oleksa and Tofilski, 2015; Nawrocka et al., 2017; Henriques et al., 2020). To see whether the discovered wild-living colonies were members of the honey bee subspecies native to the study region or likely to descent from allochthonous honey bee stocks imported by beekeepers, we determined the bees' evolutionary lineages based on morphometric analyses of their wing venation patterns. In October 2019 we collected bee samples from a subset of the colonies and analysed the wings of 10 workers per colony using standard methods (Nawrocka et al., 2017). Further information on the morphometric analyses is given in the supplementary material.

*Influence of landscape on pole occupation and winter survival*

To test the relationship between landscape context and the occupation rate of power poles as well as the winter survival of colonies, we first imported the coordinates of the power pole locations into QGIS version 3.16.2 (QGIS Development Team, 2009) and quantified the landscapes within circular buffer areas with a radius of 2 km around every pole. This radius was chosen as it reflects around 80 % of the range over which temperate *A. mellifera* forage (Danner et al., 2014; Couvillon et al., 2015), and because earlier studies showed that the landscape has an effect on honey bee colony performance at this scale (Danner et al., 2016). The circular areas were intersected with a recently published pan-European land cover data set (Pflugmacher et al., 2019). Land cover types present in our study area were grouped into the five categories “semi-natural areas” (consisting of broadleaved, coniferous, and mixed forests, and shrubland), cropland, grassland, urban areas, and water bodies. The relatively broad category “semi-natural areas” was created because different forests, heaths and other shrubland types were generally spatially intertwined in the mountainous areas and their occurrence was highly inter-correlated (see Fig. 1A and supplementary material Fig. S3). The category “grassland” was not included into this category since the intensity of grassland use vastly differed depending on the location within the study region. The proportions of the five land cover types were quantified for all the circular buffer areas around the colonies. A PCA analysis of these circular areas revealed that the landscape variation was largely driven by the contrasting proportions of cropland and semi-natural areas (see supplementary material Fig. S3). To assess the influence of the surrounding land on the occurrence (pole occupation rate) and winter survival of wild-living colonies we used a generalized linear model and a generalized linear mixed effect model, respectively. First, we modelled the occupation of the power poles by honey bees (as a binomial factor, “occupied” = 1, “not occupied” = 0) as a function of the percentage of semi-natural habitat surrounding the colonies and the factor “year” (function `glm` of the `stats` package). A type II-anova with Kenward-Roger approximation (`anova` function from the `car` package (Fox et al., 2012)) was used for inference. Second, we modelled winter survival as a binomial factor (“overwinter success” = 1, “overwinter failure” = 0) in relation to the percentage of semi-natural habitat surrounding a colony and the factor “year” (function `glmmTMB` of the package `glmmTMB` (Bolker, 2016; Magnusson et al., 2017)). Here, we accounted for the possibility that nests had the same outcomes of winter survival simply because they were close to each other and thus shared

the same landscape (spatial autocorrelation) by including the spatial covariance structure among power poles as an additional factor (Kristensen, 2019). Moran's I tests were separately applied to each year (function `testSpatialAutocorrelation` from the DHARMA package; Hartig and Hartig, 2017), but showed no signs of spatial autocorrelation (see R-script in the supplementary material). To make inferences for the effect of semi-natural areas after the potential effect of between-year variation on winter survival, we performed a type II-anova (`Anova.glmmTMB` function from the `glmmTMB` package). Residuals of the models were inspected with DHARMA package (Hartig and Hartig, 2017); no significant deviations from model assumptions were detected.

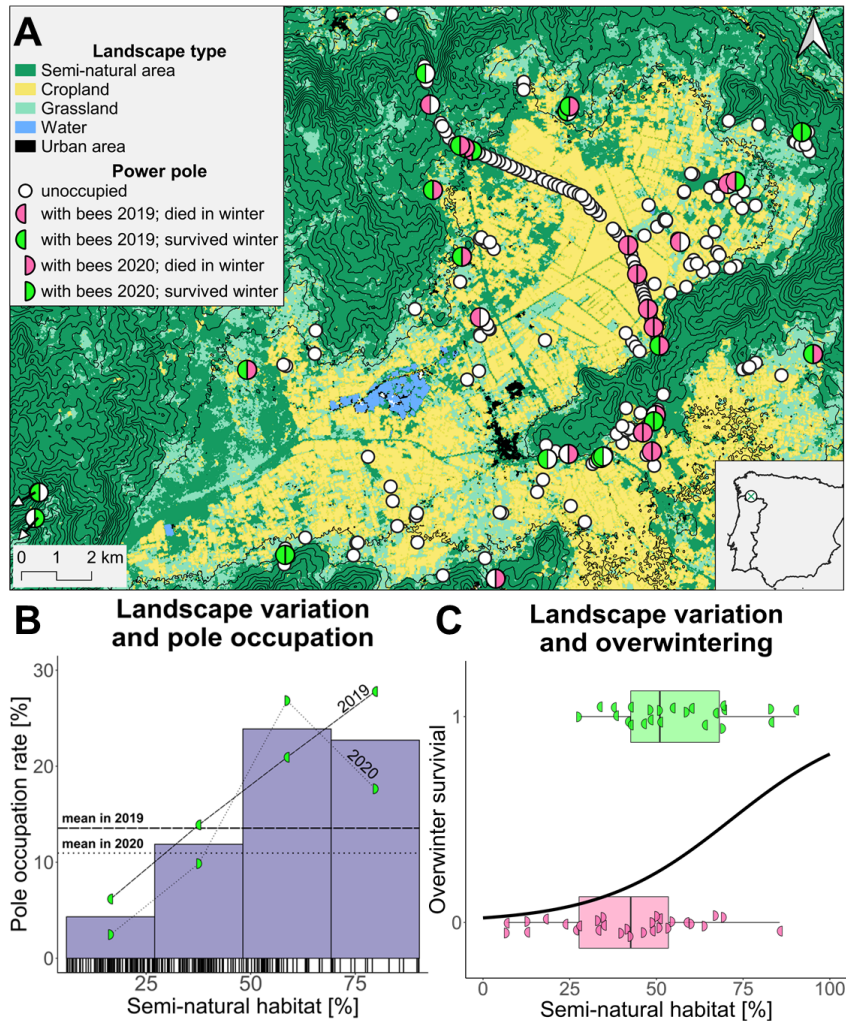
All statistical analyses were performed in RStudio (RStudio Team, 2020) with R 4.0.3 (R Core Team, 2016). The tidyverse “architecture” (Wickham, 2017) was used for data management, and `ggplot2` (Wickham, 2016) was used to create the figures. Individual annotations to the plots were performed with Inkscape 1.0.2-2 (Inkscape Project, 2020) and `gimp` 2.10.18 (The GIMP Development Team, 2020).

## Results

In October 2019, we searched an area of 136 km<sup>2</sup> for electric power poles suited as nest sites for honey bees. We found 214 poles (1.6 poles/ km<sup>2</sup>), from which 29 poles were occupied by honey bee colonies (occupation rate: 13.6 %). This corresponded to a minimum density of 0.22 colonies per km<sup>2</sup>. Wing morphometric analyses of bees sampled from 16 of the 29 colonies revealed that all belonged to the evolutionary lineage including the Iberian honey bee, *Apis mellifera iberiensis*, (lineage M) so that it can be assumed that the focal bees were native to the study region and did not decent from imported honey bee stocks (see supplementary material). At the resurvey of the 29 colonies in March 2020 we found that 17 colonies had survived the winter (59 %) (Fig. 2A). In October 2020, we re-inspected 210 power poles and found that 23 were occupied (occupation rate: 10.9 %), corresponding to a minimum density of 0.17 colonies per km<sup>2</sup>. From these 23 colonies, six overwintered successfully (26 %), resulting in an average overwintering rate of 42 % for the two consecutive years.

The landscape analyses revealed that pole occupation was skewed towards semi-natural areas in both years (`glm anova`,  $F(1, 421) = 17.6$ ,  $p < 0.001$ ) (Fig. 2B). While the average occupation rate of power poles surrounded by landscape with less than 25 % semi-natural

habitat was only 4 %, occupation of poles with 25–50 % semi-natural habitat was around 13 % and occupation of poles in landscapes with more than 50 % semi-natural habitat was more than 21 %. Furthermore, the subsequent winter survival of honey bee colonies was positively correlated with semi-natural habitat (glmmTMB anova:  $\chi^2(1) = 5.9, p = 0.015$ ) (Fig. 2C). Taken both years together, among the colonies living in landscapes with less than 25 % semi-natural habitat no colony out of 6 (0 %) survived, colonies with 25–50 % semi-natural habitat survived in 10 out of 22 cases (45 %) and among the colonies in poles surrounded by more than 50 % semi-natural habitat 13 out of 24 (54 %) survived the winter. Our model further predicted an almost 7-fold increase in winter survival rates from 8 % in landscapes with less than 25 % semi-natural habitat to 54 % in landscapes with more than 75 % semi-natural habitat.



**Figure 2:** (A) Landcover map of the study area around Xinzo de Limia in Galicia, NW Spain showing the locations of power poles and honey bee nests. Note that two poles with bees that survived the winter are shown although their locations would be outside the map (indicated by the arrows). Occupied poles have different symbols for the different years and different colours depending on the overwintering success. (B) Occupation rate of the surveyed power poles for distinct groups with similar landscape variation. The bar chart shows the average occupation rate across the two years; values for the individual years are depicted with a dashed line (October 2019) and dotted line (October 2020). The mean occupation rates in the year 2019 (13.6 %) and in 2020 (11.0 %) are depicted as dashed and dotted horizontal lines, respectively. The distribution of power poles along the semi-natural gradient is indicated on the x-axis with black marks representing individual poles. Note that while the distinct groups with similar landscape variation span the same range, the amount of power poles that fall in the different groups is highly skewed to poles with lower proportions of semi-natural habitat (C) Relationship between percentage of semi-natural habitats and honey bee colony winter survival. Raw data are on a binomial scale with “1” denoting “winter survived” and “0” denoting “winter not survived” with half-circles identifying the two consecutive years (see legend 2A). Horizontal box plots additionally describe the distribution of sites with overwintered and non-overwintered colonies along a semi-natural habitat gradient. The black line is the model of survival probability across the two years (see text for statistics).



## Discussion

The wild-living honey bee colony density we inferred for an agricultural landscape in Spain by examining power poles (0.17–0.22 colonies/km<sup>2</sup>) must be regarded as a lower limit since there were probably some additional nesting opportunities in urban areas and forests (see Kohl and Rutschmann, 2018; Albouy, 2019; Requier et al., 2020; Browne et al., 2020 or the BEEtree-Monitor ([www.beetrees.org](http://www.beetrees.org))). Nevertheless, our estimates exceed the feral colony densities reported for rural avenues in Northern Poland (0.1 colonies/km<sup>2</sup>) (Oleksa et al., 2013b) and for near-natural beech forests in Germany (0.11–0.14 colonies/km<sup>2</sup>) (Kohl and Rutschmann, 2018). This difference could be due to the climate and the resource conditions being more favourable for honey bees, and/or the bees being better adapted to a life without beekeeping management in Galicia due to few commercial breeding efforts. Alternatively, the higher density of wild-living colonies could simply be the result of more swarms escaping from managed hives which are generally more numerous in Spain than in Poland and Germany (Jones, 2004). Given that in any region, feral/wild-living and managed colonies of the same honey bee species belong to one biological population and that migrations occur between both groups (swarms escape from apiaries and beekeepers can catch wild swarms), population demographic parameters need to be measured over several years to safely determine whether a given population is self-sustaining or not (Seeley, 1978; Oldroyd et al., 1997; Seeley, 2017). While the population status of the wild-living honey bee colonies in Galicia is still ambiguous, our present study points to a key promoting factor.

Both the initial occurrence and the subsequent winter survival of the colonies were positively associated with the amount of semi-natural habitat (wood- and shrubland) in the landscapes surrounding the power poles. When offered two equally suited cavities as a new home, a dispersing honey bee swarm chooses the one that is closer to the parental colony (Lindauer, 1955; Seeley and Morse, 1977; Jaycox and Parise, 1980, 1981; Schmidt, 1995; Kohl and Rutschmann, 2018). Hence the non-random occupation of power poles observed in autumn suggests that semi-natural zones act as a source habitat of honey bee swarms. It is conceivable that beekeepers prefer to place their managed hives in such areas (Geldmann and González-Varo, 2018), which would lead to landscape variation in the number of escaping swarms and explain the observed pattern. However, since the distribution of wild-living colonies was even more biased towards semi-natural areas after the winter (and thus

before the swarming season), the landscape-dependent occupation of power poles might be readily explained by the landscape-dependent survival of wild-living colonies alone.

What our correlative analysis cannot clarify is the underlying cause of the landscape effect. On the one hand, most bee forage plants in the region are abundant in the semi-natural areas but rare in the highly agricultural zones. On the other hand, shrublands and forests are little contaminated, but the intensively farmed croplands of the study area are highly exposed to nitrogen and pesticides (López-Pérez et al., 2006; Rodes and Mejuto, 2018). Hence, it is conceivable that the landscape effect was due to the positive influence of flower resources, the negative effect of pollutants or a combination of both (see for example Seibold et al., 2019; Rundlöf et al., 2015; Goulson et al., 2015 or Brown et al., 2016). Managed honey bee colonies are not critically limited by habitat quality since beekeepers can feed sugar to keep their hives well-nourished. However, our data suggest that the survival of wild-living honey bee colonies is indeed highly dependent on flower-rich, unpolluted areas, which are often only found in natural, protected areas. Conversely, provided that wild-living honey bee colonies are less affected by pests and parasites than managed colonies due to different life histories and infection probabilities (frequent swarming and spatial dispersion of nests (Seeley and Smith, 2015; Loftus et al., 2016; Nolan and Delaplane, 2017)) or higher genetic diversity and associated immunocompetence (Youngsteadt et al., 2015; López-Urbe et al., 2017), our observations suggest that wild-living honey bee colonies might have chances to form viable populations in Europe if suitable habitat is available.

Our choice to use hollow electric poles as a monitoring tool for wild honey bees was mainly based on practical reasons: finding power poles in the open area was much easier than finding hollow trees in the forest. However hollow power poles also combine several features that make them ideally suited as honey bee nest sites: they are very spacious inside and provide maximum protection from predators as they are virtually indestructible (except due to human intervention; see supplementary material Fig. S4) and have very small nest entrances high above the ground. In our study area they can be considered as true back-up nest sites because in these intensive agricultural landscapes they probably represented the major nesting opportunity. With thousands of electric power poles all over rural areas in Spain and other parts of Europe, we should consider their potential role as important nest sites for wild-living honey bees and other cavity nesting insects.

## **Conclusion**

In Europe, the Western honey bee is regarded as the domesticated insect species managed for honey production and crop pollination whose wild ancestors have long gone extinct. However, this study contributes to the increasing body of evidence that honey bees still exist in the wild in Europe and gives hope that honey bees are still able to survive on their own if conditions are suitable. We suggest that a distinction should be made between wild-living and commercially managed honey bee colonies, especially in the context of conservation. More studies are needed that identify regions with possible native wild populations of *Apis mellifera* and that investigate the conditions under which they are viable.

## **Acknowledgements**

We thank Xosé Santos Otero from Forestal do colectivo Amigas das Arbores de Ourense for sharing his invaluable knowledge about the study region with us. Also, we are indebted to Magnus Lickfett for assistance in the pole inventory and to Cristina Ganuza and the anonymous reviewers for their valuable comments on the manuscript. We are aware of having negatively impacted the environment through carbon emissions associated with mobility for this study. Hence, one of the authors has privately compensated the estimated 6 tons of CO<sub>2</sub> emissions.

## Supplementary material

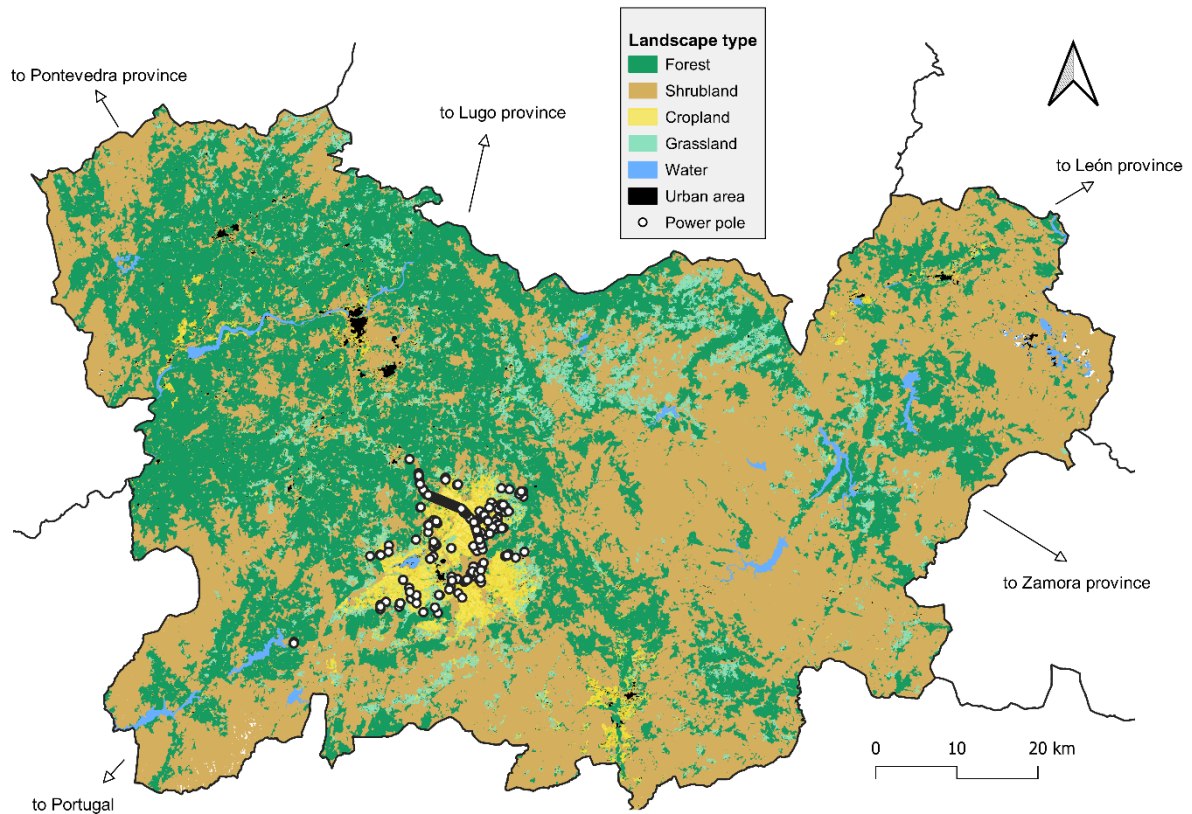
### *Morphological survey of the honey bee samples*

By taking samples in October 2019 from a subset of the colonies (N=16) we could classify these colonies by wing analysis (Oleksa and Tofilski, 2015). Not all colonies in that year could be sampled due to the risk of getting an electric shock in some locations. From the sampled colonies the right forewings of ten workers were prepared between two glass slides. We took digital images using a Motic Moticam 2300 which was mounted on a microscope with the software Motic Image Plus 2.0. We used the freely available software identify 1.3 (Nawrocka et al., 2017) to measure nineteen landmarks that were manually placed by the same person on each image. Colony averages were compared with the reference samples in identify, allowing the calculation of relative assignment probabilities to different lineages (Nawrocka et al., 2017). The honey bee subspecies native to the Iberian Peninsula, *Apis mellifera iberiensis*, belongs to the endangered evolutionary lineage of the West and Northern European honey bees (lineage M). There are two other evolutionary lineages in Europe, which are dominating modern apiaries (lineage C and lineage O). Currently, reference data for M-lineage honey bees utilized by the software identify is restricted to samples of the sister-subspecies *A. m. mellifera*. Therefore, we could only identify the heritage of our samples on the level of the evolutionary lineage and not on the level of subspecies.

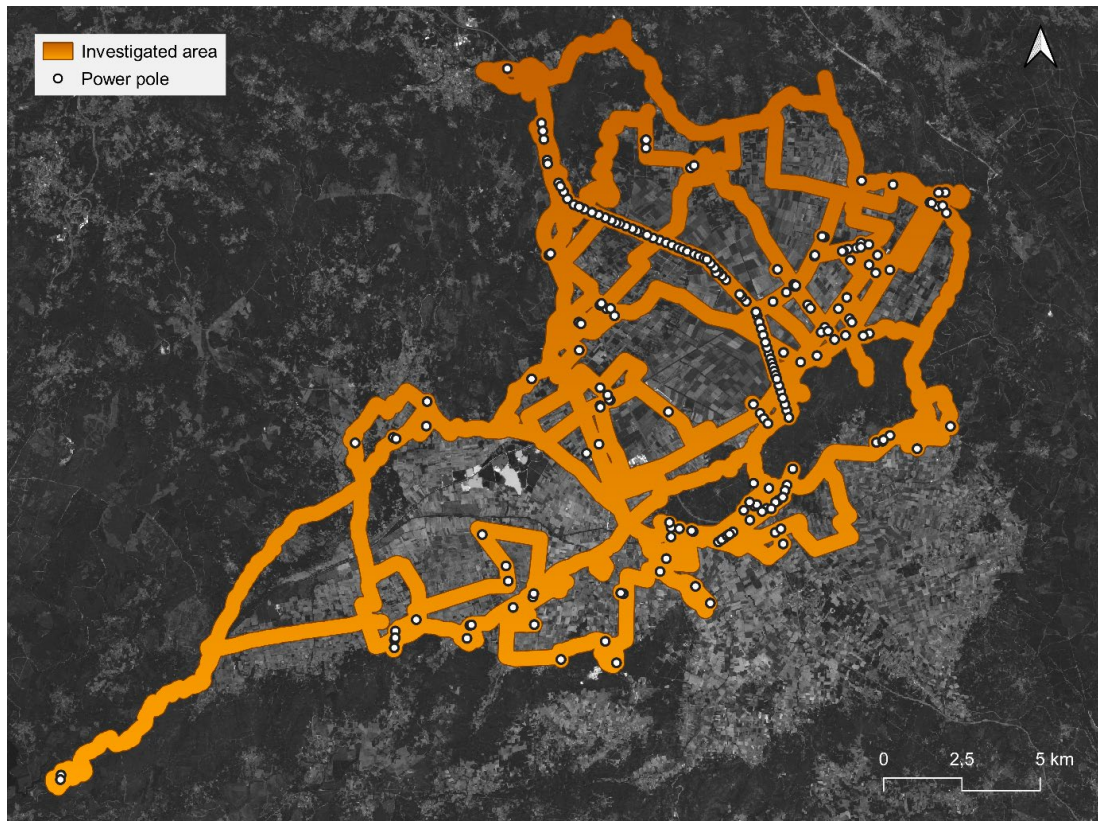
**Table S1:** A taxonomic wing morphometric analysis of bees from the wild-living colonies for which we were able to get samples (N=16) confirmed that all colonies belong to the evolutionary lineage including *Apis mellifera iberiensis* (lineage M). The relative assignment probabilities to a different lineage (M/A, M/C, M/O) are also shown. Absolute probabilities of M-lineage classification are low since the reference data currently only include samples of the sister subspecies of *A. m. iberiensis*.

Pole_Nr	classification	probability	M/A	M/C	M/O
1	M	0.000225	1.42E+13	1.42E+13	1.42E+13
2	M	1.14E-06	7.27E+10	7.42E+10	7.09E+10
3	M	5.30E-08	3.39E+09	3.40E+09	3.36E+09
6	M	1.23E-06	7.70E+10	7.67E+10	7.37E+10
9	M	1.35E-05	8.55E+11	8.16E+11	8.58E+11
10	M	6.92E-13	4.30E+04	4.48E+04	4.32E+04
12	M	9.97E-07	6.24E+10	6.76E+10	6.05E+10
13	M	1.10E-06	7.10E+10	6.79E+10	6.84E+10
14	M	8.39E-07	5.32E+10	5.08E+10	5.23E+10
18	M	2.36E-08	1.49E+09	1.49E+09	1.55E+09
20	M	1.00E-07	6.35E+09	6.62E+09	6.55E+09

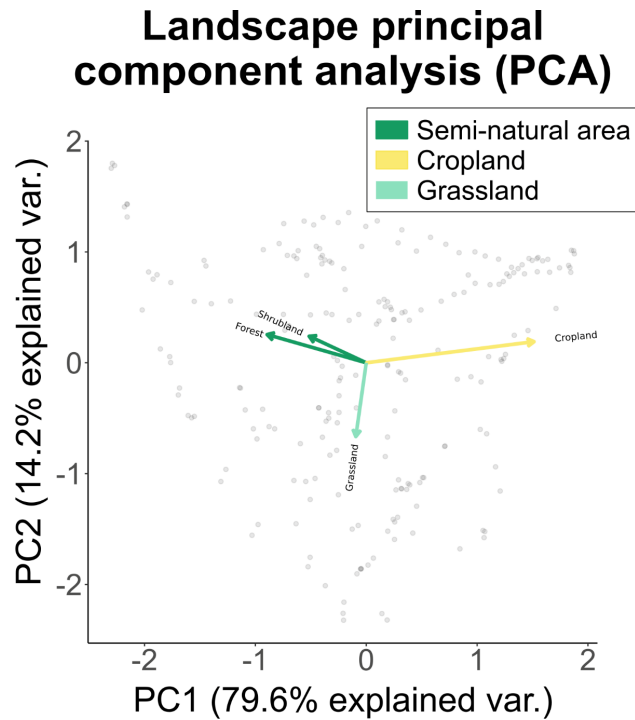
21	M	2.71E-08	1.66E+09	1.77E+09	1.76E+09
23	M	2.27E-07	1.44E+10	1.55E+10	1.40E+10
24	M	1.08E -02	6.84E+14	6.77E+14	6.70E+14
27	M	4.29E-07	2.81E+10	2.82E+10	2.78E+10
28	M	2.78E-08	1.80E+09	1.84E+09	1.77E+09



**Figure S1:** Overview of the landscape types in the Ourense province based on the classification of Pflugmacher et al., 2019. The study region with the observed power poles is located around the biggest patch of industrial cropland in the centre of the map.



**Figure S2:** Surveyed area for the power pole inventory. The tracked GPS route is shown with a 250m buffer area to each side of the driven road representing the assumed distance limit to spot a power pole. Map data: contains modified Copernicus Sentinel data 2019/2020 processed by Sentinel Hub.



**Figure S3:** Principal component analysis (PCA; using the `prcomp` function in R from the package `ggbiplot` (Vu, 2011)) of the variation in land cover within 2 km buffer zones around the power poles (the individual pole sites are indicated by grey dots). Large parts of the variation were explained by the first principal component axis, as it captured 79.6 % of the variation and was negatively correlated with semi-natural areas (forests and shrubland) and positively correlated with cropland. The landscape types "urban area" and "water" are not shown as their loading scores are too small to be visible.



**Figure S4:** Power pole with closed entrances probably due to maintenance work of the electricity company. The bees cannot enter the hollow pole anymore to build a nest inside the cavity.





Power line in Galicia, Spain. This picture shows the striking landscape contrasts. From large areas of intensive potato cultivation with high pesticide and fertiliser inputs in the foreground to semi-natural areas with no or traditional, extensive use in the background. In Galicia I empirically identified landscape composition as one important driver for viable populations of wild-living honey bees. Therefore, the protection of near-natural habitat is of paramount importance for the conservation of wild-living honey bees in Europe. Photo taken by Alejandro Machado.

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## CHAPTER 6: GENERAL DISCUSSION

While researchers and bee scientists were generally unaware of the existence of wild-living honey bee colonies, local beekeepers, ornithologists, or forest rangers knew about them but were at the same time not familiar with the importance of this phenomenon. Until recently, the cryptic nature of wild-living honey bee colonies, e.g. living secretly in trees or power poles high above ground, has prevented them from being systematically studied in Europe. In fact, we owed our knowledge about honey bees living wildly in temperate zones almost exclusively to two populations outside their natural range in a forest in New York State (Seeley, 2007; Mikheyev et al., 2015; Seeley et al., 2015; Seeley, 2017, 2019) and in the Wyperfield National Park in southeast Australia (Oldroyd et al., 1997). Additional to the studies in the US, Oleksa et al. (2013b) described wild-living colonies in Polish rural avenues that were initially discovered as a by-catch while investigating hermit beetles (*Osmoderma eremita*) (Oleksa et al., 2013a). Two other groups surveyed parasite burden of wild-living colonies in the UK and Poland (Thompson et al., 2014; Łopieńska-Biernat et al., 2017). However, systematic studies about occurrence, density, survival rates, and food resources were missing in Europe. Thus, in my thesis I performed the first quantitative surveys of wild-living honey bees in Germany and Spain and due to their occurrence studied wild-living honey bee colonies in both countries.

After it had been assumed by many for the last 40 years that wild-living honey bees in Europe were “all but wiped out” (Meixner et al., 2015), the findings of my thesis open up many new research questions, e.g. the following:

- How long do wild-living colonies survive and what is the average longevity of these colonies?
- What is the lifetime reproductive success of their queens?
- Do these colonies present a risk to the managed honey bee population by harbouring disease agents and re-infecting managed stocks? Or is the transfer of pathogens and parasites rather happening from managed to wild populations via human-mediated dissemination?
- What are the limiting factors of wild-living populations in Europe? Is the number of wild-living colonies limited by parasites in the first place? Or is it the availability of suitable cavities, or resource limitation that keeps wild-living honey bee population density low?

- Are wild-living honey bees threatened by the same stressors as beekeepers' colonies? And how can we foster wild-living populations and create conditions under which they are viable?

#### THE STUDIES ON WILD-LIVING HONEY BEE COLONIES PRESENTED IN THIS THESIS

My thesis shows that wild-living honey bee colonies can still be found in various habitats and regions in Europe and as an important reservoir of genetic variation, their population status should be clarified. Given that wild-living and managed colonies belong to the same biological population in any given region, and that both groups migrate (swarms escape from apiaries and beekeepers can catch wild swarms causing a constant gene flow between wild-living and human-kept populations), population demographic parameters must be measured over several years to safely determine whether a given population is self-sustaining (Seeley, 1978; Oldroyd et al., 1997; Seeley, 2017). So far, it is still unclear if the survival and reproductive rate of the wild-living colonies in the two surveyed areas allow stable populations or whether they still depend on the recurring emigration of swarms from managed apiaries. Since there are thousands of woodpecker cavities and hollow poles in Europe being regularly occupied by honey bee colonies, it is possible to use them to study population demography parameters of honey bees in the wild.

While the survival of colonies residing in black woodpecker cavities in German forests is low (Kohl, Rutschmann & Steffan-Dewenter, submitted), preliminary results on the survival rates of colonies in power poles in Galicia are in the range of what is expected for a self-sustaining population in the first two years. Even though colonies were present in both countries at low densities, the colony density in the Spanish mixed agricultural landscape almost doubles the one in German forest areas, implying better foraging conditions or more suitable nest sites. Importantly, based on morphometric analyses of their wing venation patterns, the colonies in Galicia belong to the endangered evolutionary lineage of the West and Northern European honey bees (M-lineage). Future studies to identify the genetic identity with more scrutiny could make use of new methods that are evolving including rapid innovations in single-nucleotide polymorphism (SNP) array technology and high-throughput sequencing (Parejo et al., 2018).

Interestingly, in the mixed landscape in Spain, colonies surrounded by more semi-natural habitat had an increased overwintering probability and I empirically identified landscape composition as one important driver for viable populations of wild-living honey bees. While the intensively farmed croplands of the study area are heavily contaminated with nitrogen and

pesticides, shrublands and woods are mostly unpolluted and forage plants like *Castanea sativa*, *Rubus* spp., *Calluna vulgaris*, and *Erica*, species might provide crucial resources in near-natural areas for the bee's survival. Unravelling whether the intensive agricultural land or the semi-natural areas (or a combination of both) drive the difference in hibernation success should be the subject of further studies.

Concern emerged that high densities of managed honey bee colonies may have unintended negative effects on local wild bees populations via resource competition (for reviews see Mallinger et al., 2017 or Wojcik et al., 2018) or dissemination of infectious diseases (see Goulson and Hughes, 2015; Graystock et al., 2016; Piot et al., 2022), and could actually exacerbate wild pollinator decline in some regions (Geldmann and González-Varo, 2018). Regarding wild-living colonies, their density is clearly lower than the one of beekeeper-managed colonies (Jones, 2004; Jaffé et al., 2010; Requier et al., 2020). Also, they are more equally distributed across the landscape, thereby reducing potential spread of diseases or competition for food. This stands in contrast to managed colonies since beekeeping activity is concentrated in rural and urban areas (Lorenz and Stark, 2015; Ropars et al., 2019). Furthermore, due to the conditions under which wild-living colonies exist (e.g. frequent swarming and small colonies (Seeley, 2019)), they should be less affected by pests and parasites than managed colonies and therefore do realistically not threaten other pollinators in Europe.

Overall, extrapolating the entire population size of wild-living honey bee colonies residing in German forests yields several thousand colonies (or over 80'000 wild-living colonies in European forests (Requier et al., 2020)). They need to be considered when estimating overall wild pollinator abundances or when assessing the role of forest areas in providing pollination services to the surrounding land (Tscharntke et al., 2005; Jaffé et al., 2010; Mitchell et al., 2014; Decocq et al., 2016). Moreover, knowledge about differences in resource availability among different habitats and different types of forests are needed. Understanding and comparing the drivers of the wild-living honey bee colony density in differently managed areas (Hinson et al., 2015) will help to answer the question how frequent honey bees would naturally be in temperate Europe. Compared to densities of one feral colony per square kilometre in the species-rich Arnot forests in North-eastern United States (Seeley, 2019) or one feral colony per two square kilometre in historic tree beekeeping regions in Russia (Galton, 1971), the density of wild-living honey bee colonies in Europe is low, suggesting mediocre foraging or nesting conditions.

Lack of resources might partly explain the low densities of wild-living honey bee colonies in Central European forests. My study presented in CHAPTER 4 is the first to compare a European forest landscape in terms of its suitability as refugium for pollinating insects. Investigating colonies located in forests, the original habitat of the honey bee in temperate Europe, I identified periods that lack key resources for wild-living honey bee colonies. While spring-flowering plants on the forest floor offered important resources for honey bees after hibernation, the shortage of pollen supply in forests in late summer is especially harmful, as colonies need protein to raise winter bees. Nevertheless, contribution to overall weight gain was substantial during short periods of the year and even though forests do not provide resources on a constant basis, a certain quantity of forest in the landscape will benefit bees by providing additional carbohydrate supplies (probably mainly honeydew) during short periods of the year (Mitchell et al., 2014; Eeraerts et al., 2021). In fact, simple interventions could have great potential for releasing resource limitations in managed forests. I highlight strategies to improve the suboptimal forage situation in today's managed forests, e.g. creating more open areas, thereby improving light permeability (as a main driver of food scarcity) or introducing more insect-pollinated tree species. Importantly, measures aiming at increasing pollen sources inside the forest that flower late in the season are urgently required.

In my thesis I established new methods to the research foraging ecology of wild-living honey bees. My approach to combine waggle dance decoding and colony weight recordings at the same time gave fundamental insights not only into the seasonal foraging patterns but also into colony performance of a social bee species in forest dominated landscape. This combination allows to infer habitat quality, immediately assess resource availability, and identify food supply bottlenecks or interruptions in any habitat (Schellhorn et al., 2015). Furthermore, I used an innovative methodology for evaluating the landscape for its expected ability to feed the bees. From the overall distribution of foraging distances (based on all waggle dances decoded in the study) I derived a function that describes the visitation probability of a given habitat patch in relation to its distance from the hive. According to this visitation probability the surrounding patches of the colony are weighted with further patches contributing less to the expected habitat use. This approach has the advantage of directly incorporating a distance-correction into the computation of expected habitat use. Expected and observed habitat usage can be easily compared, and any discrepancies may be immediately interpreted as preferences for or against a specific habitat type. Lastly, while the waggle phase is the actual distance signal used by the bees, I show that inferring approximate foraging distances from measured dance circuit duration

provides distance information sufficiently accurate for forage mapping, especially when large numbers of dances (here  $N=2292$ ) are being decoded. Nevertheless, dance decoding is very noisy (Schürch et al., 2013, 2016; Tautz, 2022). Therefore, I used a regression function to calculate uncertainty buffers for dance-inferred natural foraging locations based on empirical data from a feeder experiment (Kohl and Rutschmann, 2021). Based on the results of the feeder training experiment it became evident that the error in forage mapping increases linearly with foraging distance. Especially for advertised distances far from the hive this uncertainty should be taken into account.

#### NEW WAVE OF STUDIES EXAMINING WILD-LIVING HONEY BEE COLONIES IN THEIR NATIVE RANGE

Although *Apis mellifera* is an important and natural part of the local fauna and biodiversity in Europe, knowledge about wild-living colonies is only about to emerge. Contrary to Europe, wild honey bee populations were described and investigated in great detail outside their native range (in the US: e.g. Gambino et al., 1990; Morse et al., 1990; Kraus and Page, 1995; Baum et al., 2005; Loper et al., 2006; Baum et al., 2008; Rangel et al., 2016; Seeley, 2019; in Australia: e.g. Oldroyd et al., 1997; Cunningham et al., 2022; in South America: Rosenkranz, 1999; Brettell and Martin, 2017; Martin et al., 2020). Similarly, when reviewing documented evidence of wild *Apis mellifera* nesting sites to assess possible impact of honey bees on other cavity-using species, Saunders et al. (2021) found only six (out of 27) studies carried out in their native range and only two studies stemming from Europe (including CHAPTER 3 of this thesis). Consequently, much of the applied research on wild-living honey bee colonies, e.g. on parasite loads in feral colonies (Youngsteadt et al., 2015; López-Urbe et al., 2017; Rangel et al., 2020; Hinshaw et al., 2021), forager survival times and life spans (Ward et al., 2022), or population growth rate and virulence of *Varroa destructor* collected from feral and managed colonies (Dynes et al., 2020), was undertaken in the New World where the honey bee was introduced by settlers.

After CHAPTER 3 of this thesis was published, a handful of further studies, both research and opinion paper, on the subject of wild-living colonies in Europe were released. Requier et al. (2019) emphasized the value of native honey bee colonies and encouraged integrated conservation planning in natural areas to foster wild populations. Blacquière and Panziera highlighted the importance of leaving room for nature in the colonies reproduction (i.e. natural selection) to enhance resilience and genetic diversity in managed and wild colonies (Blacquière and Panziera, 2018; Blacquière et al., 2019; Panziera et al., 2022). Requier et al. (2020)

analysed the tree cavity densities of 106 forest areas across Europe and estimated that more than 80'000 wild-living honey bee colonies could be sustained in European forests. Importantly, cavity density was highest in unmanaged broadleaved forests, a category most German national parks belong to. Regarding data-driven studies, most reports documenting the occurrence of honey bee colonies nesting wildly in Europe used help from the public to gather information about the bees' locations. Following a nationwide appeal in a newspaper, 182 colonies, largely comprised of pure *Apis mellifera*, were described in Ireland (Hassett et al., 2018; Browne et al., 2020). Unfortunately, the authors do not clarify whether the reported survival times refer to colony lifespans or to the number of consecutive years a nest site was inhabited. Moro et al. (2021) recounted 48 European reports from wild-living colonies via a targeted citizen science approach without any information on their life-histories. A case of a population of wild-living honey bees was found in the Serbian capital Belgrade, consisting of 460 opportunistic reports of wild-living colonies for the years 2011-2017 (Bila Dubaić et al., 2021). Similarly, the BEEtree-Monitor illustrates the clustering of wild-living colonies in densely populated, urban areas in Germany (for a map of wild-living colonies see [www.beetrees.org](http://www.beetrees.org)). The downside using crowdsourcing to gather honey bee locations is the bias towards urban areas and often the compromised data quality.

An increasing number of books give examples of Varroa resistant stocks and emphasize new ways to keep honey bees (Heaf, 2021; Powell, 2019; Mittl, 2022) or address the ecology of wild-living colonies (Seeley, 2019; Albouy, 2019). The cultural heritage of tree beekeeping was inscribed in 2020 on the Representative List of the Intangible Cultural Heritage of Humanity in Poland and Belarus (<https://ich.unesco.org/en/RL/tree-beekeeping-culture-01573> or see Madras-Majewska and Skonieczna, 2021). Furthermore, the method of beelining was recently used to a greater extent in several European locations by different individuals or associations (in Germany (Sebastian Roth, personal communication, [waldbienen.eu](http://waldbienen.eu)), in an Austrian wilderness area (Oberreiter et al., 2021), in Switzerland ([freethebees.ch](http://freethebees.ch)) or in the UK (<https://www.theguardian.com/environment/2021/nov/07/no-one-knew-they-existed-wild-heirs-of-lost-british-honeybee-found-at-blenheim>)). All these developments buttressed the view that the honey bee also occurs in Europe as a wild species and I pledge that research should be undertaken to identify additional regions with native populations of *Apis mellifera* potentially thriving without human intervention. Pan-European monitoring projects should be set in place to determine the factors that affect the viability of wild-living honey bee populations in different regions and climates. Independent of their population status, their occurrence has

implications for the species' perception among researchers, beekeepers, and conservationists and one should differentiate between managed colonies aggregated in apiaries and colonies living singly in natural nests. From a conservational point of view, it is evident that, due to their low densities, wild-living colonies do not threaten other pollinators. On the contrary, as they undergo a process of natural selection they can be an important asset to foster locally adapted wild honey bee populations and deserve protection. Furthermore, there is an urgent need for ecological research of the honey bee in nature e.g. to investigate the suitability of different cavity types for colony wellbeing (Mitchell, 2016; Erdoğan, 2019; Seeley, 2019), the ecological interaction and potential competition for cavities or the interaction with honey bee predators (Saunders et al., 2021).

In conclusion, the findings of my thesis suggest that the availability of semi-natural habitats is an important driver of wild-living colonies. Particularly, if sufficient suitable habitat is available, colonies may have a chance to develop sustainable populations in Europe. Therefore, the protection and floral enrichment of habitats and safeguarding tree cavities and other sorts of nesting sites are of paramount importance for the conservation of wild-living honey bees in Europe. Merely reducing the level of intensification in forestry and agriculture could improve flower density and cavity abundance and subsequently can give honey bees the chance to resettle in European landscapes as a wild species.



There is high competition for tree cavities in today's forests. Here a young black woodpecker (*Dryocopus martius*) and a honey bee colony (located in different cavities) share the same habitat tree. Photo taken by Luigi Marchesi.



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## REFERENCES

- Aizen, M.A., Aguiar, S., Biesmeijer, J.C., Garibaldi, L.A., Inouye, D.W., Jung, C., Martins, D.J., Medel, R., Morales, C.L., Ngo, H., Pauw, A., Paxton, R.J., Sáez, A., Seymour, C.L., 2019. Global agricultural productivity is threatened by increasing pollinator dependence without a parallel increase in crop diversification. *Global Change Biology* 25, 3516–3527. <https://doi.org/10.1111/gcb.14736>
- Aizen, M.A., Harder, L.D., 2009. The Global Stock of Domesticated Honey Bees Is Growing Slower Than Agricultural Demand for Pollination. *Current Biology* 19, 915–918. <https://doi.org/10.1016/j.cub.2009.03.071>
- Alaux, C., Allier, F., Decourtye, A., Odoux, J.-F., Tamic, T., Chabirand, M., Delestra, E., Decugis, F., Le Conte, Y., Henry, M., 2017. A ‘Landscape physiology’ approach for assessing bee health highlights the benefits of floral landscape enrichment and semi-natural habitats. *Sci Rep* 7, 40568. <https://doi.org/10.1038/srep40568>
- Albouy, V., 2019. Abeilles mellifères à l’état sauvage: Une histoire naturelle, Alternatives apicoles. Editions de Terran.
- Alburaki, M., Steckel, S.J., Williams, M.T., Skinner, J.A., Tarpy, D.R., Meikle, W.G., Adamczyk, J., Stewart, S.D., 2017. Agricultural Landscape and Pesticide Effects on Honey Bee (Hymenoptera: Apidae) Biological Traits. *Journal of Economic Entomology* 110, 835–847. <https://doi.org/10.1093/jee/tox111>
- Alger, S.A., Burnham, P.A., Boncristiani, H.F., Brody, A.K., 2019. RNA virus spillover from managed honeybees (*Apis mellifera*) to wild bumblebees (*Bombus* spp.). *PLOS ONE* 14, e0217822. <https://doi.org/10.1371/journal.pone.0217822>
- Alger, S.A., Burnham, P.A., Lamas, Z.S., Brody, A.K., Richardson, L.L., 2018. Home sick: impacts of migratory beekeeping on honey bee (*Apis mellifera*) pests, pathogens, and colony size. *PeerJ* 6, e5812. <https://doi.org/10.7717/peerj.5812>
- Anderson, D.L., Trueman, J.W.H., 2000. *Varroa jacobsoni* (Acari: Varroidae) is more than one species. *Experimental & Applied Acarology* 24, 165–189. <https://doi.org/10/c2263v>
- Arnold, G., Quenet, B., Papin, C., Masson, C., Kirchner, W.H., 2002. Intra-Colonial Variability in the Dance Communication in Honeybees (*Apis mellifera*). *Ethology* 108, 751–761. <https://doi.org/10/cztcjg>
- Arundel, J., Oldroyd, B.P., Winter, S., 2013. Modelling estimates of honey bee (*Apis* spp.) colony density from drones. *Ecological Modelling* 267, 1–10. <https://doi.org/10/f5bq94>
- Arundel, J., Oldroyd, B.P., Winter, S., 2012. Modelling honey bee queen mating as a measure of feral colony density. *Ecological Modelling* 247, 48–57. <https://doi.org/10/f4kczs>
- Bailey, L., 1958. Wild Honeybees and Disease. *Bee World* 39, 92–95. <https://doi.org/10/gcm44j>
- Banaszak, J., 2009. Pollinating insects (Hymenoptera: Apoidea, Apiformes) as an example of changes in fauna. *Fragmenta Faunistica* 52, 105–123.
- Bänsch, S., Tschardt, T., Ratnieks, F.L.W., Härtel, S., Westphal, C., 2020. Foraging of honey bees in agricultural landscapes with changing patterns of flower resources. *Agriculture, Ecosystems & Environment* 291, 106792. <https://doi.org/10.1016/j.agee.2019.106792>
- Baston, D., 2020. exactextractr: Fast extraction from raster datasets using polygons. R package version 0.2 1.

- Bates, D., Mächler, M., Bolker, B., Walker, S., 2015. Fitting Linear Mixed-Effects Models Using lme4. *Journal of Statistical Software* 67, 1–48. <https://doi.org/10.18637/jss.v067.i01>
- Baum, K.A., Rubink, W.L., Pinto, M.A., Coulson, R.N., 2005. Spatial and Temporal Distribution and Nest Site Characteristics of Feral Honey Bee (Hymenoptera: Apidae) Colonies in a Coastal Prairie Landscape. *Environ Entomol* 34, 610–618. <https://doi.org/10.1603/0046-225X-34.3.610>
- Baum, K.A., Tchakerian, M.D., Thoenes, S.C., Coulson, R.N., 2008. Africanized honey bees in urban environments: A spatio-temporal analysis. *Landscape and Urban Planning* 85, 123–132. <https://doi.org/10.1016/j.landurbplan.2007.10.005>
- BayFORKLIM, B.K., 1996. *Klimaatlas von Bayern*. Lindner, München.
- Beaurepaire, A., Piot, N., Doublet, V., Antunez, K., Campbell, E., Chantawannakul, P., Chejanovsky, N., Gajda, A., Heerman, M., Panziera, D., Smagghe, G., Yañez, O., de Miranda, J.R., Dalmon, A., 2020. Diversity and Global Distribution of Viruses of the Western Honey Bee, *Apis mellifera*. *Insects* 11, 239. <https://doi.org/10.3390/insects11040239>
- Beekman, M., Ratnieks, F.L.W., 2000. Long-range foraging by the honey-bee, *Apis mellifera* L. *Funct Ecology* 14, 490–496. <https://doi.org/10.1046/j.1365-2435.2000.00443.x>
- Bentrup, G., Hopwood, J., Adamson, N.L., Vaughan, M., 2019. Temperate Agroforestry Systems and Insect Pollinators: A Review. *Forests* 10, 981. <https://doi.org/10.3390/f10110981>
- 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., Kunin, W.E., 2006. Parallel Declines in Pollinators and Insect-Pollinated Plants in Britain and the Netherlands. *Science* 313, 351–354. <https://doi.org/10.1126/science.1127863>
- Bila Dubaić, J., Simonović, S., Plečaš, M., Stanisavljević, L., Davidović, S., Tanasković, M., Četković, A., 2021. Unprecedented Density and Persistence of Feral Honey Bees in Urban Environments of a Large SE-European City (Belgrade, Serbia). *Insects* 12, 1127. <https://doi.org/10.3390/insects12121127>
- Blacquièrè, T., Boot, W., Calis, J., Moro, A., Neumann, P., Panziera, D., 2019. Darwinian black box selection for resistance to settled invasive *Varroa destructor* parasites in honey bees. *Biological Invasions* 21, 2519–2528. <https://doi.org/10.1007/s10530-019-02001-0>
- Blacquièrè, T., Panziera, D., 2018. A Plea for Use of Honey Bees' Natural Resilience in Beekeeping. *Bee World* 95, 34–38. <https://doi.org/10.1080/0005772X.2018.1430999>
- Blume, D., 1996. *Schwarzspecht, Grauspecht, Grünspecht*. Westarp, die neue Brehm Bücherei.
- Bohn, U., Neuhäusl, R., Gollub, G., Hettwer, C., Neuhäuslová, Z., Raus, T., Schlüter, H., Weber, H., 2003. Map of the natural vegetation of Europe. Scale 1: 2500000. *Landwirtschaftsverlag*, Münster.
- Bolker, B., 2016. *Getting started with the glmmTMB package*. Vienna, Austria: R Foundation for Statistical Computing. software.
- Bolte, A., Czajkowski, T., Kompa, T., 2007. The north-eastern distribution range of European beech—a review. *Forestry: An International Journal of Forest Research* 80, 413–429. <https://doi.org/10/c579j7>
- Box, G.E., Cox, D.R., 1964. An analysis of transformations. *Journal of the Royal Statistical Society: Series B (Methodological)* 26, 211–243.

- Braun-Reichert, R., Rubanschi, S., Poschlod, P., 2021. The importance of small natural features in forests—How the overgrowth of forest gaps affects indigenous flower supply and flower-visiting insects and seed sets of six *Campanula* species. *Ecology and Evolution* 11, 11991–12002. <https://doi.org/10.1002/ece3.7965>
- Breeze, T.D., Vaissière, B.E., Bommarco, R., Petanidou, T., Seraphides, N., Kozák, L., Scheper, J., Biesmeijer, J.C., Kleijn, D., Gyldenkerne, S., Moretti, M., Holzschuh, A., Steffan-Dewenter, I., Stout, J.C., Pärtel, M., Zobel, M., Potts, S.G., 2014. Agricultural Policies Exacerbate Honeybee Pollination Service Supply-Demand Mismatches Across Europe. *PLoS ONE* 9, e82996. <https://doi.org/10.1371/journal.pone.0082996>
- Brettell, L.E., Martin, S.J., 2017. Oldest *Varroa* tolerant honey bee population provides insight into the origins of the global decline of honey bees. *Sci Rep* 7, 45953. <https://doi.org/10.1038/srep45953>
- Brooks, M.E., Kristensen, K., Darrigo, M.R., Rubim, P., Uriarte, M., Bruna, E., Bolker, B.M., 2019. Statistical modeling of patterns in annual reproductive rates. *Ecology* 100. <https://doi.org/10.1002/ecy.2706>
- Brosi, B.J., Delaplane, K.S., Boots, M., de Roode, J.C., 2017. Ecological and evolutionary approaches to managing honeybee disease. *Nat Ecol Evol* 1, 1250–1262. <https://doi.org/10.1038/s41559-017-0246-z>
- Brown, M.J.F., Dicks, L.V., Paxton, R.J., Baldock, K.C.R., Barron, A.B., Chauzat, M.-P., Freitas, B.M., Goulson, D., Jepsen, S., Kremen, C., Li, J., Neumann, P., Pattermore, D.E., Potts, S.G., Schweiger, O., Seymour, C.L., Stout, J.C., 2016. A horizon scan of future threats and opportunities for pollinators and pollination. *PeerJ* 4, e2249. <https://doi.org/10.7717/peerj.2249>
- Browne, K.A., Hassett, J., Geary, M., Moore, E., Henriques, D., Soland-Reckeweg, G., Ferrari, R., Mac Loughlin, E., O'Brien, E., O'Driscoll, S., Young, P., Pinto, M.A., McCormack, G.P., 2020. Investigation of free-living honey bee colonies in Ireland. *Journal of Apicultural Research* 1–12. <https://doi.org/10.1080/00218839.2020.1837530>
- Brownrigg, R., 2017. Original S code by Richard A. Becker and Allan R. Wilks. R version by Ray Brownrigg. Enhancements by Thomas P Minka and Alex Deckmyn. maps: Draw Geographical Maps.
- Brunet, J., Fritz, O., Richnau, G., 2010. Biodiversity in European beech forests—a review with recommendations for sustainable forest management. *Ecological Bulletins* 53.
- Büchler, R., Costa, C., Hatjina, F., Andonov, S., Meixner, M.D., Conte, Y.L., Uzunov, A., Berg, S., Bienkowska, M., Bouga, M., Drazic, M., Dyrba, W., Kryger, P., Panasiuk, B., Pechhacker, H., Petrov, P., Kezić, N., Korpela, S., Wilde, J., 2014. The influence of genetic origin and its interaction with environmental effects on the survival of *Apis mellifera* L. colonies in Europe. *Journal of Apicultural Research* 53, 205–214. <https://doi.org/10.3896/IBRA.1.53.2.03>
- Burkle, L.A., Marlin, J.C., Knight, T.M., 2013. Plant-Pollinator Interactions over 120 Years: Loss of Species, Co-Occurrence, and Function. *Science* 339, 1611–1615. <https://doi.org/10.1126/science.1232728>
- Bütler, R., Lachat, T., Larrieu, L., Paillet, Y., 2013. 2.1 Habitat trees: key elements for forest biodiversity, in: Kraus, D., Krumm, F. (Eds.), *Integrative Approaches as an Opportunity for the Conservation of Forest Biodiversity*. Germany: European Forest Institute, pp. 84–91.
- Cánovas, F., de la Rúa, P., Serrano, J., Galián, J., 2011. Microsatellite variability reveals beekeeping influences on Iberian honeybee populations. *Apidologie* 42, 235–251. <https://doi.org/10.1007/s13592-011-0020-1>
- Chávez-Galarza, J., Garnery, L., Henriques, D., Neves, C.J., Loucif-Ayad, W., Jonhston, J. S., Pinto, M. A., 2017. Mitochondrial DNA variation of *Apis mellifera iberiensis*: further insights from a large-scale

- study using sequence data of the tRNA<sub>Leu</sub>-cox2 intergenic region. *Apidologie* 48, 533–544.  
<https://doi.org/10.1007/s13592-017-0498-2>
- Chávez-Galarza, J., Henriques, D., Johnston, J.S., Carneiro, M., Rufino, J., Patton, J.C., Pinto, M.A., 2015. Revisiting the Iberian honey bee (*Apis mellifera iberiensis*) contact zone: maternal and genome-wide nuclear variations provide support for secondary contact from historical refugia. *Mol Ecol* 24, 2973–2992. <https://doi.org/10.1111/mec.13223>
- Colla, S.R., 2022. The potential consequences of ‘bee washing’ on wild bee health and conservation. *International Journal for Parasitology: Parasites and Wildlife* 18, 30–32.  
<https://doi.org/10.1016/j.ijppaw.2022.03.011>
- Couvillon, M.J., Riddell Pearce, F.C., Accleton, C., Fensome, K.A., Quah, S.K.L., Taylor, E.L., Ratnieks, F.L.W., 2015. Honey bee foraging distance depends on month and forage type. *Apidologie* 46, 61–70.  
<https://doi.org/10/f6vnmc>
- Couvillon, M.J., Riddell Pearce, F.C., Harris-Jones, E.L., Kuepfer, A.M., Mackenzie-Smith, S.J., Rozario, L.A., Schurch, R., Ratnieks, F.L.W., 2012. Intra-dance variation among waggle runs and the design of efficient protocols for honey bee dance decoding. *Biology Open* 1, 467–472.  
<https://doi.org/10.1242/bio.20121099>
- Couvillon, M.J., Schürch, R., Ratnieks, F.L.W., 2014. Waggle Dance Distances as Integrative Indicators of Seasonal Foraging Challenges. *PLoS ONE* 9, e93495. <https://doi.org/10.1371/journal.pone.0093495>
- Couvillon, M.J., Schürch, R., Ratnieks, F.L.W., 2014. Dancing Bees Communicate a Foraging Preference for Rural Lands in High-Level Agri-Environment Schemes. *Current Biology* 24, 1212–1215.  
<https://doi.org/10.1016/j.cub.2014.03.072>
- Crane, E., 1999. *The world history of beekeeping and honey hunting*. Routledge.
- Cuisin, M., 1985. Range-expansion of the black woodpecker in western Europe. *British Birds* 78, 184–187.
- Cunningham, S.A., Crane, M.J., Evans, M.J., Hingee, K.L., Lindenmayer, D.B., 2022. Density of invasive western honey bee (*Apis mellifera*) colonies in fragmented woodlands indicates potential for large impacts on native species. *Sci Rep* 12, 3603. <https://doi.org/10.1038/s41598-022-07635-0>
- Dainese, M., Martin, E.A., Aizen, M.A., Albrecht, M., Bartomeus, I., Bommarco, R., Carvalheiro, L.G., Chaplin-Kramer, R., Gagic, V., Garibaldi, L.A., Ghazoul, J., Grab, H., Jonsson, M., Karp, D.S., Kennedy, C.M., Kleijn, D., Kremen, C., Landis, D.A., Letourneau, D.K., Marini, L., Poveda, K., Rader, R., Smith, H.G., Tschardtke, T., Andersson, G.K.S., Badenhausser, I., Baensch, S., Bezerra, A.D.M., Caballero-Lopez, B., Cavigliasso, P., Classen, A., Cusser, S., Dudenhöffer, J.H., Ekroos, J., Fijen, T., Franck, P., Freitas, B.M., Garratt, M.P.D., Gratton, C., Hipólito, J., Holzschuh, A., Hunt, L., Iverson, A.L., Jha, S., Keasar, T., Kim, T.N., Kishinevsky, M., Klatt, B.K., Klein, A.-M., Krewenka, K.M., Krishnan, S., Larsen, A.E., Lavigne, C., Liere, H., Maas, B., Mallinger, R.E., Pachon, E.M., Martínez-Salinas, A., Meehan, T.D., Mitchell, M.G.E., Molina, G.A.R., Nesper, M., Nilsson, L., O’Rourke, M.E., Peters, M.K., Ple, M., Ramos, D. de L., Rosenheim, J.A., Rundlöf, M., Rusch, A., Sáez, A., Scheper, J., Schleuning, M., Schmack, J.M., Sciligo, A.R., Seymour, C., Stanley, D.A., Stewart, R., Stout, J.C., Sutter, L., Takada, M.B., Taki, H., Tamburini, G., Tschumi, M., Viana, B.F., Westphal, C., Willcox, B.K., Wratten, S.D., Yoshioka, A., Zaragoza-Trello, C., Zhang, W., Zou, Y., Steffan-Dewenter, I.,

2019. A global synthesis reveals biodiversity-mediated benefits for crop production. *SCIENCE ADVANCES* 14. <https://doi.org/10.1126/sciadv.aax0121>
- Dall'Olio, R., Garrido, C., 2021. Free-Living Colonies: Trick or Treat? *Bee World* 1–3. <https://doi.org/10.1080/0005772X.2021.1885792>
- Danner, N., Härtel, S., Steffan-Dewenter, I., 2014. Maize pollen foraging by honey bees in relation to crop area and landscape context. *Basic and Applied Ecology* 15, 677–684. <https://doi.org/10.1016/j.baae.2014.08.010>
- Danner, N., Keller, A., Härtel, S., Steffan-Dewenter, I., 2017. Honey bee foraging ecology: Season but not landscape diversity shapes the amount and diversity of collected pollen. *PLoS ONE* 12, e0183716. <https://doi.org/10.1371/journal.pone.0183716>
- Danner, N., Molitor, A.M., Schiele, S., Härtel, S., Steffan-Dewenter, I., 2016. Season and landscape composition affect pollen foraging distances and habitat use of honey bees. *Ecological Applications* 26, 1920–1929. <https://doi.org/10.1890/15-1840.1>
- De la Rúa, P., Jaffé, R., Dall'Olio, R., Muñoz, I., Serrano, J., 2009. Biodiversity, conservation and current threats to European honeybees. *Apidologie* 40, 263–284. <https://doi.org/10/dn4ksd>
- De la Rúa, P., Jaffé, R., Muñoz, I., Serrano, J., Moritz, R.F.A., Kraus, F.B., 2013. Conserving genetic diversity in the honeybee: Comments on Harpur *et al.* (2012). *Mol Ecol* 22, 3208–3210. <https://doi.org/10.1111/mec.12333>
- de Mattos, I.M., Soares, A.E.E., Tarpy, D.R., 2017. Effects of synthetic acaricides on honey bee grooming behavior against the parasitic *Varroa destructor* mite. *Apidologie* 48, 483–494. <https://doi.org/10.1007/s13592-017-0491-9>
- Decocq, G., Andrieu, E., Brunet, J., Chabrierie, O., De Frenne, P., De Smedt, P., Deconchat, M., Diekmann, M., Ehrmann, S., Giffard, B., Mifsud, E.G., Hansen, K., Hermy, M., Kolb, A., Lenoir, J., Liira, J., Moldan, F., Prokofieva, I., Rosenqvist, L., Varela, E., Valdés, A., Verheyen, K., Wulf, M., 2016. Ecosystem Services from Small Forest Patches in Agricultural Landscapes. *Current Forestry Reports* 2, 30–44. <https://doi.org/10/f94d2h>
- Decourtye, A., Mader, E., Desneux, N., 2010. Landscape enhancement of floral resources for honey bees in agro-ecosystems. *Apidologie* 41, 264–277. <https://doi.org/10.1051/apido/2010024>
- DeGrandi-Hoffman, G., Ahumada, F., Graham, H., 2017. Are Dispersal Mechanisms Changing the Host–Parasite Relationship and Increasing the Virulence of *Varroa destructor* (Mesostigmata: Varroidae) in Managed Honey Bee (Hymenoptera: Apidae) Colonies? *Environmental Entomology* 46, 737–746. <https://doi.org/10/b623>
- DeGrandi-Hoffman, G., Chen, Y., Huang, E., Huang, M.H., 2010. The effect of diet on protein concentration, hypopharyngeal gland development and virus load in worker honey bees (*Apis mellifera* L.). *Journal of Insect Physiology* 56, 1184–1191. <https://doi.org/10.1016/j.jinsphys.2010.03.017>
- Di Pasquale, G., Alaux, C., Le Conte, Y., Odoux, J.-F., Pioz, M., Vaissière, B.E., Belzunces, L.P., Decourtye, A., 2016. Variations in the Availability of Pollen Resources Affect Honey Bee Health. *PLoS ONE* 11, e0162818. <https://doi.org/10.1371/journal.pone.0162818>

- Di Pasquale, G., Salignon, M., Le Conte, Y., Belzunces, L.P., Decourtye, A., Kretzschmar, A., Suchail, S., Brunet, J.-L., Alaux, C., 2013. Influence of Pollen Nutrition on Honey Bee Health: Do Pollen Quality and Diversity Matter? *PLoS ONE* 8, e72016. <https://doi.org/10.1371/journal.pone.0072016>
- Díaz Losada, E., González Porto, A., Saa Otero, M., 1997. Estudio melisopalínológico en Galicia (NW de España). *Orsis* 12, 027–038.
- Díaz-Losada, E., 2002. Método de determinación del origen geográfico del polen apícola comercial. *Lazaroa* 23, 25–34.
- Dietemann, V., Ellis, J.D., Neumann, P., 2013. The COLOSS BEEBOOK Volume II: Standard methods for *Apis mellifera* pest and pathogen research. International Bee Research Association IBRA.
- Dietemann, V., Pflugfelder, J., Anderson, D., Charrière, J.-D., Chejanovsky, N., Dainat, B., de Miranda, J., Delaplane, K., Dillier, F.-X., Fuch, S., Gallmann, P., Gauthier, L., Imdorf, A., Koeniger, N., Kralj, J., Meikle, W., Pettis, J., Rosenkranz, P., Sammataro, D., Smith, D., Yañez, O., Neumann, P., 2012. *Varroa destructor*: research avenues towards sustainable control. *Journal of Apicultural Research* 51, 125–132. <https://doi.org/10.3896/IBRA.1.51.1.15>
- Dogantzis, K.A., Tiwari, T., Conflitti, I.M., Dey, A., Patch, H.M., Muli, E.M., Garnery, L., Whitfield, C.W., Stolle, E., Alqarni, A.S., Allsopp, M.H., Zayed, A., 2021. Thrice out of Asia and the adaptive radiation of the western honey bee. *Sci. Adv.* 7, eabj2151. <https://doi.org/10.1126/sciadv.abj2151>
- Döke, M.A., McGrady, C.M., Otieno, M., Grozinger, C.M., Frazier, M., 2019. Colony Size, Rather Than Geographic Origin of Stocks, Predicts Overwintering Success in Honey Bees (Hymenoptera: Apidae) in the Northeastern United States. *Journal of Economic Entomology* 112, 525–533. <https://doi.org/10.1093/jee/toy377>
- Dolezal, A.G., St. Clair, A.L., Zhang, G., Toth, A.L., O’Neal, M.E., 2019. Native habitat mitigates feast–famine conditions faced by honey bees in an agricultural landscape. *Proc Natl Acad Sci USA* 116, 25147–25155. <https://doi.org/10.1073/pnas.1912801116>
- Donkersley, P., Rhodes, G., Pickup, R.W., Jones, K.C., Wilson, K., 2014. Honeybee nutrition is linked to landscape composition. *Ecol Evol* 4, 4195–4206. <https://doi.org/10.1002/ece3.1293>
- Dowle, M., Srinivasan, A., Gorecki, J., Chirico, M., Stetsenko, P., Short, T., Lianoglou, S., Antonyan, E., Bonsch, M., Parsonage, H., others, 2019. Package ‘data.table.’ Extension of ‘data.frame.’
- Dynes, T.L., Berry, J.A., Delaplane, K.S., de Roode, J.C., Brosi, B.J., 2020. Assessing virulence of *Varroa destructor* mites from different honey bee management regimes. *Apidologie* 51, 276–289. <https://doi.org/10.1007/s13592-019-00716-6>
- Eckerter, T., Braunisch, V., Pufal, G., Klein, A.M., 2022. Small clear-cuts in managed forests support trap-nesting bees, wasps and their parasitoids. *Forest Ecology and Management* 509, 120076. <https://doi.org/10.1016/j.foreco.2022.120076>
- Eckerter, T., Buse, J., Förschler, M., Pufal, G., 2019. Additive positive effects of canopy openness on European bilberry (*Vaccinium myrtillus*) fruit quantity and quality. *Forest Ecology and Management* 433, 122–130. <https://doi.org/10.1016/j.foreco.2018.10.059>
- Eeraerts, M., Van Den Berge, S., Proesmans, W., Verheyen, K., Smaghe, G., Meeus, I., 2021. Fruit orchards and woody semi-natural habitat provide complementary resources for pollinators in agricultural landscapes. *Landscape Ecol* 36, 1377–1390. <https://doi.org/10.1007/s10980-021-01220-y>

- Erdoğan, Y., 2019. Comparison of colony performances of honeybee (*Apis Mellifera* L.) housed in hives made of different materials. Italian Journal of Animal Science 18, 934–940.  
<https://doi.org/10.1080/1828051X.2019.1604088>
- Espregueira Themudo, G., Rey-Iglesia, A., Robles Tascón, L., Bruun Jensen, A., da Fonseca, R.R., Campos, P.F., 2020. Declining genetic diversity of European honeybees along the twentieth century. Sci Rep 10, 10520. <https://doi.org/10.1038/s41598-020-67370-2>
- Fontana, P., Costa, C., Prisco, G.D., Ruzzier, E., Annoscia, D., Battisti, A., Caoduro, G., Carpana, E., Contessi, A., Dal, A., Dall, R., Cristofaro, A.D., Felicioli, A., Floris, I., Gardi, T., Lodesani, M., Malagnini, V., Manias, L., Manino, A., Marzi, G., Massa, B., Mutinelli, F., Nazzi, F., Pennacchio, F., Porporato, M., Stoppa, G., Tormen, N., Valentini, M., Segrè, A., 2018. Appeal for biodiversity protection of native honey bee subspecies of *Apis mellifera* in Italy (San Michele all’Adige declaration) 16.
- Forfert, N., Natsopoulou, M.E., Frey, E., Rosenkranz, P., Paxton, R.J., Moritz, R.F.A., 2015. Parasites and Pathogens of the Honeybee (*Apis mellifera*) and Their Influence on Inter-Colonial Transmission. PLOS ONE 10, e0140337. <https://doi.org/10/f77wmd>
- Forfert, N., Natsopoulou, M.E., Paxton, R.J., Moritz, R.F.A., 2016. Viral prevalence increases with regional colony abundance in honey bee drones (*Apis mellifera* L). Infection, Genetics and Evolution 44, 549–554. <https://doi.org/10/f84p4c>
- Fox, J., Weisberg, S., Adler, D., Bates, D., Baud-Bovy, G., Ellison, S., Firth, D., Friendly, M., Gorjanc, G., Graves, S., others, 2012. Package ‘car.’ Vienna: R Foundation for Statistical Computing 16.
- Frey, E., Rosenkranz, P., 2014. Autumn Invasion Rates of *Varroa destructor* (Mesostigmata: Varroidae) Into Honey Bee (Hymenoptera: Apidae) Colonies and the Resulting Increase in Mite Populations. Journal of Economic Entomology 107, 508–515. <https://doi.org/10/f56kx5>
- Fries, I., Camazine, S., 2001. Implications of horizontal and vertical pathogen transmission for honey bee epidemiology. Apidologie 32, 199–214. <https://doi.org/10.1051/apido:2001122>
- Fürst, M.A., McMahon, D.P., Osborne, J.L., Paxton, R.J., Brown, M.J.F., 2014. Disease associations between honeybees and bumblebees as a threat to wild pollinators. Nature 506, 364–366.  
<https://doi.org/10.1038/nature12977>
- Gallai, N., Salles, J.-M., Settele, J., Vaissière, B.E., 2009. Economic valuation of the vulnerability of world agriculture confronted with pollinator decline. Ecological Economics 68, 810–821.  
<https://doi.org/10.1016/j.ecolecon.2008.06.014>
- Galton, D., 1971. Survey of a thousand years of beekeeping in Russia.
- Gambino, P., Hoelmer, K., Daly, H.V., 1990. Nest sites of feral honey bees in California, USA. Apidologie 21, 35–45. <https://doi.org/10.1051/apido:19900105>
- Garbuzov, M., Balfour, N.J., Shackleton, K., Al Toufaily, H., Scandian, L., Ratnieks, F.L.W., 2020. Multiple methods of assessing nectar foraging conditions indicate peak foraging difficulty in late season. Insect Conserv Divers icad.12420. <https://doi.org/10.1111/icad.12420>
- Garbuzov, M., Schürch, R., Ratnieks, F.L.W., 2015. Eating locally: dance decoding demonstrates that urban honey bees in Brighton, UK, forage mainly in the surrounding urban area. Urban Ecosyst 18, 411–418.  
<https://doi.org/10.1007/s11252-014-0403-y>

- Garibaldi, L.A., Steffan-Dewenter, I., Kremen, C., Morales, J.M., Bommarco, R., Cunningham, S.A.,  
Carvalho, L.G., Chacoff, N.P., Dudenhöffer, J.H., Greenleaf, S.S., Holzschuh, A., Isaacs, R.,  
Krewenka, K., Mandelik, Y., Mayfield, M.M., Morandin, L.A., Potts, S.G., Ricketts, T.H.,  
Szentgyörgyi, H., Viana, B.F., Westphal, C., Winfree, R., Klein, A.M., 2011. Stability of pollination  
services decreases with isolation from natural areas despite honey bee visits: Habitat isolation and  
pollination stability. *Ecology Letters* 14, 1062–1072. <https://doi.org/10.1111/j.1461-0248.2011.01669.x>
- Garibaldi, L.A., Steffan-Dewenter, I., Winfree, R., Aizen, M.A., Bommarco, R., Cunningham, S.A., Kremen, C.,  
Carvalho, L.G., Harder, L.D., Afik, O., Bartomeus, I., Benjamin, F., Boreux, V., Cariveau, D.,  
Chacoff, N.P., Dudenhoffer, J.H., Freitas, B.M., Ghazoul, J., Greenleaf, S., Hipolito, J., Holzschuh, A.,  
Howlett, B., Isaacs, R., Javorek, S.K., Kennedy, C.M., Krewenka, K.M., Krishnan, S., Mandelik, Y.,  
Mayfield, M.M., Motzke, I., Munyuli, T., Nault, B.A., Otieno, M., Petersen, J., Pisanty, G., Potts, S.G.,  
Rader, R., Ricketts, T.H., Rundlof, M., Seymour, C.L., Schuepp, C., Szentgyorgyi, H., Taki, H.,  
Tschardt, T., Vergara, C.H., Viana, B.F., Wanger, T.C., Westphal, C., Williams, N., Klein, A.M.,  
2013. Wild Pollinators Enhance Fruit Set of Crops Regardless of Honey Bee Abundance. *Science* 339,  
1608–1611. <https://doi.org/10.1126/science.1230200>
- Gauer, J., Aldinger, E., 2005. Waldökologische Naturräume Deutschlands–Forstliche Wuchsgebiete und  
Wuchsbezirke. *Mitteilungen des Vereins für Forstliche Standortskunde und Forstpflanzenzüchtung* 43,  
1–324.
- Geldmann, J., González-Varo, J.P., 2018. Conserving honey bees does not help wildlife. *Science* 359, 392–393.  
<https://doi.org/10.1126/science.aar2269>
- Genersch, E., 2010. Honey bee pathology: current threats to honey bees and beekeeping. *Applied Microbiology  
and Biotechnology* 87, 87–97. <https://doi.org/10.1007/s00253-010-2573-8>
- George, E.A., Thulasi, N., Kohl, P.L., Suresh, S., Rutschmann, B., Brockmann, A., 2021. Distance estimation by  
Asian honey bees in two visually different landscapes. *Journal of Experimental Biology* 224.  
<https://doi.org/10.1242/jeb.242404>
- Ghazoul, J., 2005. Buzziness as usual? Questioning the global pollination crisis. *Trends in Ecology & Evolution*  
20, 367–373. <https://doi.org/10.1016/j.tree.2005.04.026>
- Ginsberg, H.S., 1983. Foraging Ecology of Bees in an Old Field. *Ecology* 64, 165–175.  
<https://doi.org/10.2307/1937338>
- Goodwin, R.M., Houten, A.T., Perry, J.H., 1994. Incidence of American foulbrood infections in feral honey bee  
colonies in New Zealand. *New Zealand Journal of Zoology* 21, 285–287.  
<https://doi.org/10.1080/03014223.1994.9517996>
- Gould, J.L., 1982. Why do honey bees have dialects? *Behav Ecol Sociobiol* 10, 53–56.  
<https://doi.org/10.1007/BF00296395>
- Goulson, D., Hughes, W.O.H., 2015. Mitigating the anthropogenic spread of bee parasites to protect wild  
pollinators. *Biological Conservation* 191, 10–19. <https://doi.org/10.1016/j.biocon.2015.06.023>
- Goulson, D., Nicholls, E., Botias, C., Rotheray, E.L., 2015. Bee declines driven by combined stress from  
parasites, pesticides, and lack of flowers. *Science* 347, 1255957–1255957.  
<https://doi.org/10.1126/science.1255957>



- Graystock, P., Blane, E.J., McFrederick, Q.S., Goulson, D., Hughes, W.O.H., 2016. Do managed bees drive parasite spread and emergence in wild bees? *International Journal for Parasitology: Parasites and Wildlife* 5, 64–75. <https://doi.org/10.1016/j.ijppaw.2015.10.001>
- Graystock, P., Goulson, D., Hughes, W.O.H., 2015. Parasites in bloom: flowers aid dispersal and transmission of pollinator parasites within and between bee species. *Proceedings of the Royal Society B: Biological Sciences* 282, 20151371. <https://doi.org/10.1098/rspb.2015.1371>
- Grindrod, I., Martin, S.J., 2021. Parallel evolution of *Varroa* resistance in honey bees: a common mechanism across continents? *Proc. R. Soc. B.* 288, 20211375. <https://doi.org/10.1098/rspb.2021.1375>
- Gunda, B., 1968. Bee-hunting in the Carpathian area. *Acta Ethnographica Academiae Scientiarum Hungaricae* 17, 1–62.
- Han, F., Wallberg, A., Webster, M.T., 2012. From where did the Western honeybee (*Apis mellifera*) originate? *Ecol Evol* 2, 1949–1957. <https://doi.org/10.1002/ece3.312>
- Hanula, J.L., 2015. Have changing forests conditions contributed to pollinator decline in the southeastern United States? *Forest Ecology and Management* 11. <https://doi.org/10.1016/j.foreco.2015.03.044>
- Hanula, J.L., Ulyshen, M.D., Horn, S., 2016. Conserving Pollinators in North American Forests: A Review. *Natural Areas Journal* 36, 427–439. <https://doi.org/10.3375/043.036.0409>
- Harpur, B.A., Minaei, S., Kent, C.F., Zayed, A., 2012. Management increases genetic diversity of honey bees via admixture: GENETIC DIVERSITY IN THE HONEY BEE. *Molecular Ecology* 21, 4414–4421. <https://doi.org/10.1111/j.1365-294X.2012.05614.x>
- Harrison, X.A., Donaldson, L., Correa-Cano, M.E., Evans, J., Fisher, D.N., Goodwin, C.E.D., Robinson, B.S., Hodgson, D.J., Inger, R., 2018. A brief introduction to mixed effects modelling and multi-model inference in ecology. *PeerJ* 6, e4794. <https://doi.org/10.7717/peerj.4794>
- Hartig, F., Hartig, M.F., 2017. Package ‘DHARMA.’
- Hassett, J., Browne, K.A., McCormack, G.P., Moore, E., Society, N.I.H.B., Soland, G., Geary, M., 2018. A significant pure population of the dark European honey bee (*Apis mellifera mellifera*) remains in Ireland. *Journal of Apicultural Research* 57, 337–350. <https://doi.org/10.1080/00218839.2018.1433949>
- Heaf, D., 2021. Treatment-Free Beekeeping.
- Heinrich, B., 1976. Flowering Phenologies: Bog, Woodland, and Disturbed Habitats. *Ecology* 57, 890–899. <https://doi.org/10.2307/1941055>
- Henriques, D., Chávez-Galarza, J., S. G. Teixeira, J., Ferreira, H., J. Neves, C., Francoy, T.M., Pinto, M.A., 2020. Wing Geometric Morphometrics of Workers and Drones and Single Nucleotide Polymorphisms Provide Similar Genetic Structure in the Iberian Honey Bee (*Apis mellifera iberiensis*). *Insects* 11, 89. <https://doi.org/10.3390/insects11020089>
- Henry, M., Fröchen, M., Maillat-Mezeray, J., Breyne, E., Allier, F., Odoux, J.-F., Decourtye, A., 2012. Spatial autocorrelation in honeybee foraging activity reveals optimal focus scale for predicting agro-environmental scheme efficiency. *Ecological Modelling* 225, 103–114. <https://doi.org/10.1016/j.ecolmodel.2011.11.015>
- Henry, M., Rodet, G., 2018. Controlling the impact of the managed honeybee on wild bees in protected areas. *Sci Rep* 8, 9308. <https://doi.org/10.1038/s41598-018-27591-y>

- Herbertsson, L., Lindström, S.A.M., Rundlöf, M., Bommarco, R., Smith, H.G., 2016. Competition between managed honeybees and wild bumblebees depends on landscape context. *Basic and Applied Ecology* 17, 609–616. <https://doi.org/10.1016/j.baae.2016.05.001>
- Herrera, C.M., 2020. Gradual replacement of wild bees by honeybees in flowers of the Mediterranean Basin over the last 50 years. *Proceedings of the Royal Society B: Biological Sciences* 287, 20192657. <https://doi.org/10.1098/rspb.2019.2657>
- Hijmans, R.J., Van Etten, J., Cheng, J., Mattiuzzi, M., Sumner, M., Greenberg, J.A., Lamigueiro, O.P., Bevan, A., Racine, E.B., Shortridge, A., others, 2015. Package ‘raster.’ R package 734.
- Hinshaw, C., Evans, K.C., Rosa, C., López-Urbe, M.M., 2021. The Role of Pathogen Dynamics and Immune Gene Expression in the Survival of Feral Honey Bees. *Front. Ecol. Evol.* 8, 594263. <https://doi.org/10.3389/fevo.2020.594263>
- Hinson, E.M., Duncan, M., Lim, J., Arundel, J., Oldroyd, B.P., 2015. The density of feral honey bee (*Apis mellifera*) colonies in South East Australia is greater in undisturbed than in disturbed habitats. *Apidologie* 46, 403–413. <https://doi.org/10.1007/s13592-014-0334-x>
- Hung, K.-L.J., Kingston, J.M., Albrecht, M., Holway, D.A., Kohn, J.R., 2018. The worldwide importance of honey bees as pollinators in natural habitats. *Proceedings of the Royal Society of London B: Biological Sciences* 285. <https://doi.org/10.1098/rspb.2017.2140>
- Ilyasov, R.A., Kosarev, M.N., Neal, A., Yumaguzhin, F.G., 2015. Burzyan Wild-Hive Honeybee *A.M. mellifera* in South Ural. *Bee World* 92, 7–11. <https://doi.org/10/gcm45m>
- Inkscape Project, 2020. Inkscape.
- Iwasaki, J.M., Hogendoorn, K., 2021. How protection of honey bees can help and hinder bee conservation. *Current Opinion in Insect Science* 46, 112–118. <https://doi.org/10.1016/j.cois.2021.05.005>
- Jaffé, R., Dietmann, V., Allsopp, M.H., Costa, C., Crewe, R.M., Dall’Olio, R., De la Rúa, P., El-Niweiri, M.A.A., Fries, I., Kezic, N., Meusel, M.S., Paxton, R.J., Shaibi, T., Stolle, E., Moritz, R.F.A., 2010. Estimating the Density of Honeybee Colonies across Their Natural Range to Fill the Gap in Pollinator Decline Censuses. *Conservation Biology* 24, 583–593. <https://doi.org/10/dpkc7v>
- Jaycox, E.R., Parise, S.G., 1981. Homesite selection by swarms of black-bodied honey bees, *Apis mellifera caucasica* and *A. m. carnica* (Hymenoptera: Apidae). *Journal of the Kansas Entomological Society* 697–703.
- Jaycox, E.R., Parise, S.G., 1980. Homesite selection by Italian honey bee swarms, *Apis mellifera ligustica* (Hymenoptera: Apidae). *Journal of the Kansas Entomological Society* 171–178.
- Johnsson, K., Nilsson, S.G., Tjernberg, M., 1993. Characteristics and utilization of old Black Woodpecker *Dryocopus martius* holes by hole-nesting species. *Ibis* 135, 410–416. <https://doi.org/10/cqhntz>
- Jones, R., 2004. European beekeeping in the 21st century: strengths, weaknesses, opportunities, threats. *Bee World* 85, 77–80. <https://doi.org/10.1080/0005772X.2004.11099637>
- Jooß, R., 2013. Schwäbische Alb (Swabian Alb) Biosphere Reserve. *eco. mont-Journal on Protected Mountain Areas Research* 5, 43–48.
- Kefuss, J., Vanpoucke, J., Bolt, M., Kefuss, C., 2015. Selection for resistance to *Varroa destructor* under commercial beekeeping conditions. *Journal of Apicultural Research* 54, 563–576. <https://doi.org/10.1080/00218839.2016.1160709>

- Kennedy, C.M., Lonsdorf, E., Neel, M.C., Williams, N.M., Ricketts, T.H., Winfree, R., Bommarco, R., Brittain, C., Burley, A.L., Cariveau, D., Carvalheiro, L.G., Chacoff, N.P., Cunningham, S.A., Danforth, B.N., Dudenhöffer, J.-H., Elle, E., Gaines, H.R., Garibaldi, L.A., Gratton, C., Holzschuh, A., Isaacs, R., Javorek, S.K., Jha, S., Klein, A.M., Krewenka, K., Mandelik, Y., Mayfield, M.M., Morandin, L., Neame, L.A., Otieno, M., Park, M., Potts, S.G., Rundlöf, M., Saez, A., Steffan-Dewenter, I., Taki, H., Viana, B.F., Westphal, C., Wilson, J.K., Greenleaf, S.S., Kremen, C., 2013. A global quantitative synthesis of local and landscape effects on wild bee pollinators in agroecosystems. *Ecology Letters* 16, 584–599. <https://doi.org/10.1111/ele.12082>
- Klein, A.-M., Vaissière, B.E., Cane, J.H., Steffan-Dewenter, I., Cunningham, S.A., Kremen, C., Tscharntke, T., 2007. Importance of pollinators in changing landscapes for world crops. *Proc. R. Soc. B.* 274, 303–313. <https://doi.org/10.1098/rspb.2006.3721>
- Knapp, H., Nickel, E., Plachter, H., 2007. Buchenwälder—ein europäischer Beitrag zum Waldarbeitsprogramm der CBD. *Natur und Landschaft* 9, 386–390.
- Koenker, R., Portnoy, S., Ng, P.T., Zeileis, A., Grosjean, P., Ripley, B.D., 2018. Package ‘quantreg.’ Reference manual available at R-CRAN: <https://cran.rproject.org/web/packages/quantreg/quantreg.pdf>.
- Kohl, P.L., Rutschmann, B., 2021. Honey bees communicate distance via non-linear waggle duration functions. *PeerJ* 9, e11187. <https://doi.org/10.7717/peerj.11187>
- Kohl, P.L., Rutschmann, B., 2018. The neglected bee trees: European beech forests as a home for feral honey bee colonies. *PeerJ* 6, e4602. <https://doi.org/10.7717/peerj.4602>
- Kohl, P.L., Thulasi, N., Rutschmann, B., George, E.A., Steffan-Dewenter, I., Brockmann, A., 2020. Adaptive evolution of honeybee dance dialects. *Proc. R. Soc. B.* 287, 20200190. <https://doi.org/10.1098/rspb.2020.0190>
- Kraus, B., Page, Jr., Robert E., 1995. Effect of *Varroa jacobsoni* (Mesostigmata: Varroidae) on Feral *Apis mellifera* (Hymenoptera: Apidae) in California. *Environmental Entomology* 24, 1473–1480. <https://doi.org/10/gcm439>
- Kristensen, K., 2019. Covariance structures with glmmTMB. URL: <https://cran.rproject.org/web/packages/glmmTMB/vignettes/covstruct.html>.
- Kruitwagen, A., van Langevelde, F., van Dooremalen, C., Blacquière, T., 2017. Naturally selected honey bee (*Apis mellifera*) colonies resistant to *Varroa destructor* do not groom more intensively. *Journal of Apicultural Research* 56, 354–365. <https://doi.org/10.1080/00218839.2017.1329797>
- Kuchling, S., Kopačka, I., Kalcher-Sommersguter, E., Schwarz, M., Crailsheim, K., Brodschneider, R., 2018. Investigating the role of landscape composition on honey bee colony winter mortality: A long-term analysis. *Sci Rep* 8, 12263. <https://doi.org/10.1038/s41598-018-30891-y>
- Kükreer, M., Kence, M., Kence, A., 2021. Honey Bee Diversity Is Swayed by Migratory Beekeeping and Trade Despite Conservation Practices: Genetic Evidence for the Impact of Anthropogenic Factors on Population Structure. *Front. Ecol. Evol.* 9, 556816. <https://doi.org/10.3389/fevo.2021.556816>
- Küster, H., 2010. *Geschichte der Landschaft in Mitteleuropa: Von der Eiszeit bis zur Gegenwart*. CH Beck.
- Küster, H., 2003. *Geschichte des Waldes: von der Urzeit bis zur Gegenwart*. CH Beck.

- Kutsch, W.L., Persson, T., Schrupf, M., Moyano, F.E., Mund, M., Andersson, S., Schulze, E.-D., 2010. Heterotrophic soil respiration and soil carbon dynamics in the deciduous Hainich forest obtained by three approaches. *Biogeochemistry* 100, 167–183. <https://doi.org/10/bsddsc>
- Le Conte, Y., De Vaublanc, G., Crauser, D., Jeanne, F., Rousselle, J.-C., Bécard, J.-M., 2007. Honey bee colonies that have survived *Varroa destructor*. *Apidologie* 38, 566–572. <https://doi.org/10.1051/apido:2007040>
- Le Conte, Y., Ellis, M., Ritter, W., 2010. *Varroa* mites and honey bee health: can *Varroa* explain part of the colony losses?\*. *Apidologie* 41, 353–363. <https://doi.org/10/dm495b>
- Le Conte, Y., Meixner, M.D., Brandt, A., Carreck, N.L., Costa, C., Mondet, F., Büchler, R., 2020. Geographical Distribution and Selection of European Honey Bees Resistant to *Varroa destructor*. *Insects* 11, 873. <https://doi.org/10.3390/insects11120873>
- Le Féon, V., Schermann-Legionnet, A., Delettre, Y., Aviron, S., Billeter, R., Bugter, R., Hendrickx, F., Burel, F., 2010. Intensification of agriculture, landscape composition and wild bee communities: A large scale study in four European countries. *Agriculture, Ecosystems & Environment* 137, 143–150. <https://doi.org/10.1016/j.agee.2010.01.015>
- Lecocq, A., Kryger, P., Vejsnæs, F., Bruun Jensen, A., 2015. Weight Watching and the Effect of Landscape on Honeybee Colony Productivity: Investigating the Value of Colony Weight Monitoring for the Beekeeping Industry. *PLOS ONE* 10, e0132473. <https://doi.org/10.1371/journal.pone.0132473>
- Leonhardt, S.D., Gallai, N., Garibaldi, L.A., Kuhlmann, M., Klein, A.-M., 2013. Economic gain, stability of pollination and bee diversity decrease from southern to northern Europe. *Basic and Applied Ecology* 14, 461–471. <https://doi.org/10.1016/j.baae.2013.06.003>
- Liebig, G., 1999. Die Waldtracht: Entstehung, Beobachtung, Prognose; 16 Tabellen. G. Liebig.
- Lindauer, M., 1956. Über die Verständigung bei indischen Bienen. *Zeitschrift für vergleichende Physiologie* 38, 521–557.
- Lindauer, M., 1955. Schwarmbienen auf Wohnungssuche. *Zeitschrift für vergleichende Physiologie* 37, 263–324. <https://doi.org/10/dsgwtx>
- Lindström, S.A.M., Herbertsson, L., Rundlöf, M., Bommarco, R., Smith, H.G., 2016. Experimental evidence that honeybees depress wild insect densities in a flowering crop. *Proceedings of the Royal Society B: Biological Sciences* 283, 20161641. <https://doi.org/10.1098/rspb.2016.1641>
- Locke, B., 2016. Natural *Varroa* mite-surviving *Apis mellifera* honeybee populations. *Apidologie* 47, 467–482. <https://doi.org/10/f3tpnp>
- Locke, B., Conte, Y.L., Crauser, D., Fries, I., 2012. Host adaptations reduce the reproductive success of *Varroa destructor* in two distinct European honey bee populations. *Ecology and evolution* 2, 1144–1150. <https://doi.org/10.1002/ece3.248>
- Locke, B., Fries, I., 2011. Characteristics of honey bee colonies (*Apis mellifera*) in Sweden surviving *Varroa destructor* infestation. *Apidologie* 42, 533–542. <https://doi.org/10.1007/s13592-011-0029-5>
- Loftus, J.C., Smith, M.L., Seeley, T.D., 2016. How Honey Bee Colonies Survive in the Wild: Testing the Importance of Small Nests and Frequent Swarming. *PLOS ONE* 11, e0150362. <https://doi.org/10/f8vmd5>

- Loper, G.M., Sammataro, D., Finley, J., Cole, J., 2006. Feral honey bees in southern Arizona 10 years after *Varroa* infestation. *American Bee Journal* 146, 521–524.
- López-Pérez, G.C., Arias-Estévez, M., López-Periago, E., Soto-González, B., Cancho-Grande, B., Simal-Gándara, J., 2006. Dynamics of Pesticides in Potato Crops. *Journal of Agricultural and Food Chemistry* 54, 1797–1803. <https://doi.org/10.1021/jf0525737>
- López-Urbe, M.M., Appler, R.H., Youngsteadt, E., Dunn, R.R., Frank, S.D., Tarpy, D.R., 2017. Higher immunocompetence is associated with higher genetic diversity in feral honey bee colonies (*Apis mellifera*). *Conserv Genet* 18, 659–666. <https://doi.org/10.1007/s10592-017-0942-x>
- Łopieńska-Biernat, E., Sokół, R., Michalczyk, M., Żółtowska, K., Stryński, R., 2017. Biochemical status of feral honey bees (*Apis mellifera*) infested with various pathogens. *Journal of Apicultural Research* 56, 606–615. <https://doi.org/10.1080/00218839.2017.1343020>
- Lorenz, S., Stark, K., 2015. Saving the honeybees in Berlin? A case study of the urban beekeeping boom. *Environmental Sociology* 1, 116–126. <https://doi.org/10.1080/23251042.2015.1008383>
- Madras-Majewska, B., Skonieczna, L., 2021. Diversification of Morphological Features of the Dark European Honey Bee of the ‘Augustow M’ Line. *Animals* 11, 1156. <https://doi.org/10.3390/ani11041156>
- Magnusson, A., Skaug, H., Nielsen, A., Berg, C., Kristensen, K., Maechler, M., van Benthem, K., Bolker, B., Brooks, M., Brooks, M.M., 2017. Package ‘glmmTMB.’ R Package Version 0.2. 0.
- Magrach, A., González-Varo, J.P., Boiffier, M., Vilà, M., Bartomeus, I., 2017. Honeybee spillover reshuffles pollinator diets and affects plant reproductive success. *Nature Ecology & Evolution* 1, 1299–1307. <https://doi.org/10.1038/s41559-017-0249-9>
- Mallinger, R.E., Gaines-Day, H.R., Gratton, C., 2017. Do managed bees have negative effects on wild bees?: A systematic review of the literature. *PLOS ONE* 12, e0189268. <https://doi.org/10.1371/journal.pone.0189268>
- Mandelik, Y., Winfree, R., Neeson, T., Kremen, C., 2012. Complementary habitat use by wild bees in agri-natural landscapes. *Ecological Applications* 22, 1535–1546. <https://doi.org/10.1890/11-1299.1>
- Manley, R., Temperton, B., Doyle, T., Gates, D., Hedges, S., Boots, M., Wilfert, L., 2019. Knock-on community impacts of a novel vector: spillover of emerging DWV-B from *Varroa* -infested honeybees to wild bumblebees. *Ecology Letters* ele.13323. <https://doi.org/10.1111/ele.13323>
- Martin, S.J., 2004. Acaricide (pyrethroid) resistance in *Varroa destructor*. *Bee World* 85, 67–69. <https://doi.org/10.1080/0005772X.2004.11099632>
- Martin, S.J., Hawkins, G.P., Brettell, L.E., Reece, N., Correia-Oliveira, M.E., Allsopp, M.H., 2020. *Varroa destructor* reproduction and cell re-capping in mite-resistant *Apis mellifera* populations. *Apidologie* 51, 369–381. <https://doi.org/10.1007/s13592-019-00721-9>
- Martin, S.J., Highfield, A.C., Brettell, L., Villalobos, E.M., Budge, G.E., Powell, M., Nikaido, S., Schroeder, D.C., 2012. Global Honey Bee Viral Landscape Altered by a Parasitic Mite. *Science* 336, 1304. <https://doi.org/10/f32k9x>
- Martínez-López, V., Ruiz, C., De la Rúa, P., 2022. “Migratory beekeeping and its influence on the prevalence and dispersal of pathogens to managed and wild bees.” *International Journal for Parasitology: Parasites and Wildlife* S2213224422000499. <https://doi.org/10.1016/j.ijppaw.2022.05.004>

- Mashilingi, S.K., Zhang, H., Garibaldi, L.A., An, J., 2022. Honeybees are far too insufficient to supply optimum pollination services in agricultural systems worldwide. *Agriculture, Ecosystems & Environment* 335, 108003. <https://doi.org/10.1016/j.agee.2022.108003>
- McLellan, A.R., 1977. Honeybee Colony Weight as an Index of Honey Production and Nectar Flow: A Critical Evaluation. *The Journal of Applied Ecology* 14, 401. <https://doi.org/10.2307/2402553>
- McMahon, D.P., Fürst, M.A., Caspar, J., Theodorou, P., Brown, M.J.F., Paxton, R.J., 2015. A sting in the spit: widespread cross-infection of multiple RNA viruses across wild and managed bees. *Journal of Animal Ecology* 84, 615–624. <https://doi.org/10.1111/1365-2656.12345>
- McMullan, J., 2018. Adaptation in Honey Bee (*Apis mellifera*) Colonies Exhibiting Tolerance to *Varroa destructor* in Ireland. *Bee World* 95, 39–43. <https://doi.org/10.1080/0005772X.2018.1431000>
- Meikle, W.G., Rector, B.G., Mercadier, G., Holst, N., 2008. Within-day variation in continuous hive weight data as a measure of honey bee colony activity. *Apidologie* 39, 694–707. <https://doi.org/10.1051/apido:2008055>
- Meixner, M.D., Costa, C., Kryger, P., Hatjina, F., Bouga, M., Ivanova, E., Büchler, R., 2010. Conserving diversity and vitality for honey bee breeding. *Journal of Apicultural Research* 49, 85–92. <https://doi.org/10.3896/IBRA.1.49.1.12>
- Meixner, M.D., Kryger, P., Costa, C., 2015. Effects of genotype, environment, and their interactions on honey bee health in Europe. *Social Insects \* Vectors and Medical and Veterinary Entomology* 10, 177–184. <https://doi.org/10/gcm437>
- Mergner, U., Kraus, D., 2020. Learning from nature: Integrative forest management in Ebrach, Germany. pp. 196–213.
- Meyer, W., Meyer, B., 2001. Bau und Nutzung von Schwarzspechthöhlen in Thüringen. *Abhandlungen und Berichte aus dem Museum Heineanum* 5, 121–131.
- Michener, C.D., 1974. *The social behavior of the bees: a comparative study*. Harvard University Press.
- Mikheyev, A.S., Tin, M.M.Y., Arora, J., Seeley, T.D., 2015. Museum samples reveal rapid evolution by wild honey bees exposed to a novel parasite. *Nature Communications* 6, 7991. <https://doi.org/10.1038/ncomms8991>
- Milani, N., 1999. The resistance of *Varroa jacobsoni* Oud. to acaricides. *Apidologie* 30, 229–234. <https://doi.org/10.1051/apido:19990211>
- Mitchell, D., 2016. Ratios of colony mass to thermal conductance of tree and man-made nest enclosures of *Apis mellifera*: implications for survival, clustering, humidity regulation and *Varroa destructor*. *Int J Biometeorol* 60, 629–638. <https://doi.org/10.1007/s00484-015-1057-z>
- Mitchell, M.G.E., Bennett, E.M., Gonzalez, A., 2014. Forest fragments modulate the provision of multiple ecosystem services. *Journal of Applied Ecology* 51, 909–918. <https://doi.org/10/f6kj96>
- Mittl, S., 2022. Nachhaltig Imkern mit gesunden Honigbienen: Aus Vergangenheit und Gegenwart für die Zukunft lernen. Haupt Verlag, Bern, Switzerland.
- Möller, G., 2009. Struktur- und Substratbindung holzbewohnender Insekten, Schwerpunkt Coleoptera - Käfer (PhD Thesis). Freie Universität Berlin.
- Mondet, F., Beaurepaire, A., McAfee, A., Locke, B., Alaux, C., Blanchard, S., Danka, B., Le Conte, Y., 2020. Honey bee survival mechanisms against the parasite *Varroa destructor*: a systematic review of

- phenotypic and genomic research efforts. *International Journal for Parasitology* 50, 433–447.  
<https://doi.org/10.1016/j.ijpara.2020.03.005>
- Moritz, R.F.A., de Miranda, J., Fries, I., Le Conte, Y., Neumann, P., Paxton, R.J., 2010. Research strategies to improve honeybee health in Europe\*. *Apidologie* 41, 227–242. <https://doi.org/10/fjzd2m>
- Moritz, R.F.A., Dietemann, V., Crewe, R., 2008. Determining colony densities in wild honeybee populations (*Apis mellifera*) with linked microsatellite DNA markers. *Journal of Insect Conservation* 12, 455–459.  
<https://doi.org/10/fkww3q>
- Moritz, R.F.A., Erler, S., 2016. Lost colonies found in a data mine: Global honey trade but not pests or pesticides as a major cause of regional honeybee colony declines. *Agriculture, Ecosystems & Environment* 216, 44–50. <https://doi.org/10.1016/j.agee.2015.09.027>
- Moritz, R.F.A., Härtel, S., Neumann, P., 2005. Global invasions of the western honeybee (*Apis mellifera*) and the consequences for biodiversity. *Écoscience* 12, 289–301. <https://doi.org/10.2980/i1195-6860-12-3-289.1>
- Moritz, R.F.A., Kraus, F.B., Kryger, P., Crewe, R.M., 2007. The size of wild honeybee populations (*Apis mellifera*) and its implications for the conservation of honeybees. *Journal of Insect Conservation* 11, 391–397. <https://doi.org/10/dzjfxr>
- Moro, A., Beaurepaire, A., Dall’Olio, R., Rogenstein, S., Blacquièrre, T., Dahle, B., de Miranda, J.R., Dietemann, V., Locke, B., Licón Luna, R.M., Le Conte, Y., Neumann, P., 2021. Using Citizen Science to Scout Honey Bee Colonies That Naturally Survive *Varroa destructor* Infestations. *Insects* 12, 536.  
<https://doi.org/10.3390/insects12060536>
- Morse, R.A., Boch, R., 1971. Pheromone Concert in Swarming Honey Bees (Hymenoptera: Apidae)1. *Annals of the Entomological Society of America* 64, 1414–1417. <https://doi.org/10/gcm45n>
- Morse, R.A., Camazine, S., Ferracane, M., Minacci, P., Nowogrodzki, R., Ratnieks, F.L.W., Spielholz, J., Underwood, B.A., 1990. The Population Density of Feral Colonies of Honey Bees (Hymenoptera: Apidae) in a City in Upstate New York. *Journal of Economic Entomology* 83, 81–83.  
<https://doi.org/10.1093/jee/83.1.81>
- Morse, R.A., Nowogrodzki, R., 1990. *Honey bee pests, predators, and diseases*. Cornell University Press, Ithaca, NY.
- Müller, J., Jarzabek-Müller, A., Bussler, H., Gossner, M., 2014. Hollow beech trees identified as keystone structures for saproxylic beetles by analyses of functional and phylogenetic diversity. *Animal Conservation* 17, 154–162. <https://doi.org/10.1111/acv.12075>
- Muñoz, I., De la Rúa, P., 2021. Wide genetic diversity in Old World honey bees threaten by introgression. *Apidologie* 52, 200–217. <https://doi.org/10.1007/s13592-020-00810-0>
- Muñoz-Colmenero, M., Baroja-Careaga, I., Kovačić, M., Filipi, J., Puškadija, Z., Kezić, N., Estonba, A., Büchler, R., Zarraonaindia, I., 2020. Differences in honey bee bacterial diversity and composition in agricultural and pristine environments – a field study. *Apidologie*. <https://doi.org/10.1007/s13592-020-00779-w>
- Mutinelli, F., 2011. The spread of pathogens through trade in honey bees and their products (including queen bees and semen): overview and recent developments. *Revue scientifique et technique (International Office of Epizootics)* 30, 257–271. <https://doi.org/10/gcm455>

- Naug, D., 2009. Nutritional stress due to habitat loss may explain recent honeybee colony collapses. *Biological Conservation* 142, 2369–2372. <https://doi.org/10.1016/j.biocon.2009.04.007>
- Nawrocka, A., Kandemir, İ., Fuchs, S., Tofilski, A., 2017. Computer software for identification of honey bee subspecies and evolutionary lineages. *Apidologie*. <https://doi.org/10.1007/s13592-017-0538-y>
- Nazzi, F., Le Conte, Y., 2016. Ecology of *Varroa destructor*, the Major Ectoparasite of the Western Honey Bee, *Apis mellifera*. *Annual Review of Entomology* 61, 417–432. <https://doi.org/10/gcm436>
- Neumann, P., Blacquière, T., 2017. The Darwin cure for apiculture? Natural selection and managed honeybee health. *Evolutionary Applications* 10, 226–230. <https://doi.org/10/f9zfs>
- Neumann, P., Carreck, N.L., 2010. Honey bee colony losses. *Journal of Apicultural Research* 49, 1–6. <https://doi.org/10/fkbh2w>
- Nieto, A., Roberts, S., Kemp, J., Rasmont, P., Kuhlmann, M., García Criado, M., Biesmeijer, J., Bogusch, P., Dathe, H., De la Rúa, P., others, 2014. European red list of bees. Publication Office of the European Union, Luxembourg. Europe 2.
- Nolan, M.P., Delaplane, K.S., 2017. Distance between honey bee *Apis mellifera* colonies regulates populations of *Varroa destructor* at a landscape scale. *Apidologie* 48, 8–16. <https://doi.org/10/f9nj9>
- Oberreiter, H., Dünser, A., Schlagbauer, J., Brodschneider, R., 2021. Das Wildnisgebiet Dürrenstein – ein Lebensraum für wildlebende Bienenvölker? 19.
- Oddie, M.A., Dahle, B., Neumann, P., 2017. Norwegian honey bees surviving *Varroa destructor* mite infestations by means of natural selection. *PeerJ* 5, e3956. <https://doi.org/10.7717/peerj.3956>
- Oldroyd, B.P., Thexton, E.G., Lawler, S.H., Crozier, R.H., 1997. Population demography of Australian feral bees (*Apis mellifera*). *Oecologia* 111, 381–387. <https://doi.org/10.1007/s004420050249>
- Oleksa, A., Chybicki, I.J., Gawroński, R., Svensson, G.P., Burczyk, J., 2013a. Isolation by distance in saproxylic beetles may increase with niche specialization. *J Insect Conserv* 17, 219–233. <https://doi.org/10.1007/s10841-012-9499-7>
- Oleksa, A., Gawroński, R., Tofilski, A., 2013b. Rural avenues as a refuge for feral honey bee population. *Journal of Insect Conservation* 17, 465–472. <https://doi.org/10/f4xf44>
- Oleksa, A., Tofilski, A., 2015. Wing geometric morphometrics and microsatellite analysis provide similar discrimination of honey bee subspecies. *Apidologie* 46, 49–60. <https://doi.org/10.1007/s13592-014-0300-7>
- Ollerton, J., Winfree, R., Tarrant, S., 2011. How many flowering plants are pollinated by animals? *Oikos* 120, 321–326. <https://doi.org/10.1111/j.1600-0706.2010.18644.x>
- Osterman, J., Aizen, M.A., Biesmeijer, J.C., Bosch, J., Howlett, B.G., Inouye, D.W., Jung, C., Martins, D.J., Medel, R., Pauw, A., Seymour, C.L., Paxton, R.J., 2021. Global trends in the number and diversity of managed pollinator species. *Agriculture, Ecosystems & Environment* 322, 107653. <https://doi.org/10.1016/j.agee.2021.107653>
- Packham, J.R., Thomas, P.A., Atkinson, M.D., Degen, T., 2012. Biological Flora of the British Isles: *Fagus sylvatica*. *Journal of Ecology* 100, 1557–1608. <https://doi.org/10/f4csz3>
- Panziera, D., Requier, F., Chantawannakul, P., Pirk, C.W.W., Blacquière, T., 2022. The Diversity Decline in Wild and Managed Honey Bee Populations Urges for an Integrated Conservation Approach. *Front. Ecol. Evol.* 10, 767950. <https://doi.org/10.3389/fevo.2022.767950>



- Panziera, D., van Langevelde, F., Blacqui re, T., 2017. *Varroa* sensitive hygiene contributes to naturally selected *varroa* resistance in honey bees. *Journal of Apicultural Research* 56, 635–642.  
<https://doi.org/10.1080/00218839.2017.1351860>
- Parejo, M., Henriques, D., Pinto, M.A., Soland-Reckeweg, G., Neuditschko, M., 2018. Empirical comparison of microsatellite and SNP markers to estimate introgression in *Apis mellifera mellifera*. *Journal of Apicultural Research* 57, 504–506. <https://doi.org/10.1080/00218839.2018.1494894>
- Parejo, M., Wragg, D., Henriques, D., Charri re, J.-D., Estonba, A., 2020. Digging into the Genomic Past of Swiss Honey Bees by Whole-Genome Sequencing Museum Specimens. *Genome Biology and Evolution* 12, 2535–2551. <https://doi.org/10.1093/gbe/evaa188>
- Pebesma, E., 2018. Simple Features for R: Standardized Support for Spatial Vector Data. *The R Journal* 10, 439–446. <https://doi.org/10.32614/RJ-2018-009>
- Pebesma, E., Bivand, R., Pebesma, M.E., RColorBrewer, S., Collate, A., 2012. Package ‘sp.’ The Comprehensive R Archive Network.
- Pebesma, E., Bivand, R.S., 2005. S classes and methods for spatial data: the sp package. *R news* 5, 9–13.
- Peck, D.T., Seeley, T.D., 2019. Mite bombs or robber lures? The roles of drifting and robbing in *Varroa* destructor transmission from collapsing honey bee colonies to their neighbors. *PLoS ONE* 14, e0218392. <https://doi.org/10.1371/journal.pone.0218392>
- Peck, D.T., Smith, M.L., Seeley, T.D., 2016. *Varroa* destructor Mites Can Nimbly Climb from Flowers onto Foraging Honey Bees. *PLoS ONE* 11, e0167798. <https://doi.org/10.1371/journal.pone.0167798>
- Pedersen, T.L., 2020. patchwork: The Composer of Plots.
- Pettis, J.S., 2004. A scientific note on *Varroa destructor* resistance to coumaphos in the United States. *Apidologie* 35, 91–92. <https://doi.org/10.1051/apido:2003060>
- Pflugmacher, D., Rabe, A., Peters, M., Hostert, P., 2019. Mapping pan-European land cover using Landsat spectral-temporal metrics and the European LUCAS survey. *Remote Sensing of Environment* 221, 583–595. <https://doi.org/10.1016/j.rse.2018.12.001>
- Pinto, M.A., Mu oz, I., Ch vez-Galarza, J., De la R a, P., 2012. The Atlantic side of the Iberian Peninsula: a hot-spot of novel African honey bee maternal diversity. *Apidologie* 43, 663–673.  
<https://doi.org/10.1007/s13592-012-0141-1>
- Piot, N., Schweiger, O., Meeus, I., Y nez, O., Straub, L., Villamar-Bouza, L., De la R a, P., Jara, L., Ruiz, C., Malmstr m, M., Mustafa, S., Nielsen, A., M nd, M., Karise, R., Tlak-Gajger, I.,  zg r, E., Keskin, N., Di vart, V., Dalmon, A., Gajda, A., Neumann, P., Smagghe, G., Graystock, P., Radzevi iut , R., Paxton, R.J., de Miranda, J.R., 2022. Honey bees and climate explain viral prevalence in wild bee communities on a continental scale. *Sci Rep* 12, 1904. <https://doi.org/10.1038/s41598-022-05603-2>
- Pirk, C.W.W., Crewe, R.M., Moritz, R.F.A., 2017. Risks and benefits of the biological interface between managed and wild bee pollinators. *Funct Ecol* 31, 47–55. <https://doi.org/10.1111/1365-2435.12768>
- Potts, S.G., Biesmeijer, J.C., Kremen, C., Neumann, P., Schweiger, O., Kunin, W.E., 2010. Global pollinator declines: trends, impacts and drivers. *Trends in Ecology & Evolution* 25, 345–353.  
<https://doi.org/10/cmb34c>
- Powell, J., 2019. The Tree Beekeeping Field Guide. Natural Beekeeping Trust.

- Prešern, J., Mihelič, J., Kobal, M., 2019. Growing stock of nectar- and honeydew-producing tree species determines the beekeepers' profit. *Forest Ecology and Management* 448, 490–498.  
<https://doi.org/10.1016/j.foreco.2019.06.031>
- Proctor, E., Nol, E., Burke, D., Crins, W.J., 2012. Responses of insect pollinators and understory plants to silviculture in northern hardwood forests. *Biodivers Conserv* 21, 1703–1740.  
<https://doi.org/10.1007/s10531-012-0272-8>
- Punchihewa, R.W.K., Koeniger, N., Kevan, P.G., Gadawski, R.M., 1985. Observations on the Dance Communication and Natural Foraging Ranges of *Apis Cerana*, *Apis Dorsata* and *Apis Florea* in Sri Lanka. *Journal of Apicultural Research* 24, 168–175. <https://doi.org/10.1080/00218839.1985.11100667>
- QGIS Development Team, 2009. QGIS Geographic Information System. Open Source Geospatial Foundation.
- R Core Team, 2016. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.
- Randi, E., 2008. Detecting hybridization between wild species and their domesticated relatives. *Molecular Ecology* 17, 285–293. <https://doi.org/10.1111/j.1365-294X.2007.03417.x>
- Rangel, J., Giresi, M., Pinto, M.A., Baum, K.A., Rubink, W.L., Coulson, R.N., Johnston, J.S., 2016. Africanization of a feral honey bee (*Apis mellifera*) population in South Texas: does a decade make a difference? *Ecol Evol* 6, 2158–2169. <https://doi.org/10.1002/ece3.1974>
- Rangel, J., Traver, B., Stoner, M., Hatter, A., Trevelline, B., Garza, C., Shepherd, T., Seeley, T.D., Wenzel, J., 2020. Genetic diversity of wild and managed honey bees (*Apis mellifera*) in Southwestern Pennsylvania, and prevalence of the microsporidian gut pathogens *Nosema ceranae* and *N. apis*. *Apidologie* 51, 802–814. <https://doi.org/10.1007/s13592-020-00762-5>
- Raši, R., 2020. State of Europe's forests 2020, in: Proceedings of the Ministerial Conference on the Protection of Forests in Europe, Bratislava, Slovakia. pp. 14–15.
- Requier, F., Garnery, L., Kohl, P.L., Njovu, H.K., Pirk, C.W.W., Crewe, R.M., Steffan-Dewenter, I., 2019. The Conservation of Native Honey Bees Is Crucial. *Trends in Ecology & Evolution* 34, 789–798.  
<https://doi.org/10.1016/j.tree.2019.04.008>
- Requier, F., Odoux, J.-F., Henry, M., Bretagnolle, V., 2017. The carry-over effects of pollen shortage decrease the survival of honeybee colonies in farmlands. *J Appl Ecol* 54, 1161–1170.  
<https://doi.org/10.1111/1365-2664.12836>
- Requier, F., Paillet, Y., Laroche, F., Rutschmann, B., Zhang, J., Lombardi, F., Svoboda, M., Steffan-Dewenter, I., 2020. Contribution of European forests to safeguard wild honeybee populations. *CONSERVATION LETTERS* 13. <https://doi.org/10.1111/conl.12693>
- Rinderer, T.E., de Guzman, L.I., Delatte, G., Stelzer, J., Lancaster, V., Kuznetsov, V., Beaman, L., Watts, R., Harris, J., 2001. Resistance to the parasitic mite *Varroa destructor* in honey bees from far-eastern Russia. *Apidologie* 32, 381–394. <https://doi.org/10.1051/apido:2001138>
- Ripley, B., Venables, B., Bates, D.M., Hornik, K., Gebhardt, A., Firth, D., Ripley, M.B., 2013. Package 'mass.' *Cran r* 538, 113–120.
- Rodes, J.M.A., Mejuto, M.F., 2018. Las aguas subterráneas en España: hacia la sostenibilidad del recurso, in: Congreso Nacional Del Agua Orihuela: Innovación y Sostenibilidad. Universitat d'Alacant/Universidad de Alicante, pp. 1229–1254.

- Rodríguez-Castiñeira, J., Armesto-Baztán, S., De Sá-Otero, M.P., 2015. Origen botánico y contenido en proteína de mieles artesanales procedentes de colmenares de Galicia (NO de España). *Botanica Complutensis* 39, 105–114. [https://doi.org/10.5209/rev\\_BOCM.2015.v39.49139](https://doi.org/10.5209/rev_BOCM.2015.v39.49139)
- Ropars, L., Affre, L., Geslin, B., 2021. Morphometric identification of honey bee subspecies reveals a high proportion of hybrids within a Mediterranean protected area. *Journal of Apicultural Research* 60, 871–874. <https://doi.org/10.1080/00218839.2021.1950973>
- Ropars, L., Dajoz, I., Fontaine, C., Muratet, A., Geslin, B., 2019. Wild pollinator activity negatively related to honey bee colony densities in urban context. *PLoS ONE* 14, e0222316. <https://doi.org/10.1371/journal.pone.0222316>
- Rosenkranz, P., 1999. Honey bee (*Apis mellifera* L.) tolerance to *Varroa jacobsoni* Oud. in South America. *Apidologie* 30, 159–172. <https://doi.org/10.1051/apido:19990206>
- Rosenkranz, P., Aumeier, P., Ziegelmann, B., 2010. Biology and control of *Varroa destructor*. *Journal of Invertebrate Pathology* 103, S96–S119. <https://doi.org/10/dm3hk6>
- RStudio Team, 2020. RStudio: Integrated Development Environment for R. RStudio, PBC., Boston, MA.
- Rundlöf, M., Andersson, G.K.S., Bommarco, R., Fries, I., Hederström, V., Herbertsson, L., Jonsson, O., Klatt, B.K., Pedersen, T.R., Yourstone, J., Smith, H.G., 2015. Seed coating with a neonicotinoid insecticide negatively affects wild bees. *Nature* 521, 77–80. <https://doi.org/10.1038/nature14420>
- Ruppertshofen, H., 1995. *The buzzing forest*. Ehrenwirth.
- Rutschmann, B., Kohl, P.L., Machado, A., Steffan-Dewenter, I., 2022. Semi-natural habitats promote winter survival of wild-living honeybees in an agricultural landscape. *Biological Conservation* 266, 109450. <https://doi.org/10.1016/j.biocon.2022.109450>
- Ruttner, F., 1988. *Biogeography and Taxonomy of Honeybees*. Springer Berlin Heidelberg.
- Ruttner, F., 1973. Races of bees, in: *The Hive and the Honey Bee*. Dadant & Sons, Hamilton, Illinois, USA, pp. 20–38.
- Sammataro, D., Gerson, U., Needham, G., 2000. Parasitic Mites of Honey Bees: Life History, Implications, and Impact. *Annual Review of Entomology* 45, 519–548. <https://doi.org/10/csdcs5>
- Samuelson, A.E., Leadbeater, E., 2018. A land classification protocol for pollinator ecology research: An urbanization case study. *Ecol Evol* 8, 5598–5610. <https://doi.org/10.1002/ece3.4087>
- Samuelson, A.E., Schürch, R., Leadbeater, E., 2021. Dancing bees evaluate central urban forage resources as superior to agricultural land. *J Appl Ecol* 1365–2664.14011. <https://doi.org/10.1111/1365-2664.14011>
- Saunders, M.E., Goodwin, E.K., Santos, K.C., Sonter, C.A., Rader, R., 2021. Cavity occupancy by wild honey bees: need for evidence of ecological impacts. *Front Ecol Environ* fee.2347. <https://doi.org/10.1002/fee.2347>
- Schellhorn, N.A., Gagic, V., Bommarco, R., 2015. Time will tell: resource continuity bolsters ecosystem services. *Trends in Ecology & Evolution* 30, 524–530. <https://doi.org/10.1016/j.tree.2015.06.007>
- Scherföse, V., Hoffmann, A., Jeschke, L., Panek, N., Riecken, U., Ssymank, A., 2007. Threat and protection of beech forests in Germany. *Natur und Landschaft* 82, 416–422.
- Schmehl, D.R., Teal, P.E.A., Frazier, J.L., Grozinger, C.M., 2014. Genomic analysis of the interaction between pesticide exposure and nutrition in honey bees (*Apis mellifera*). *Journal of Insect Physiology* 71, 177–190. <https://doi.org/10.1016/j.jinsphys.2014.10.002>

- Schmid-Hempel, P., 2021. *Evolutionary parasitology: the integrated study of infections, immunology, ecology, and genetics*. Oxford University Press.
- Schmidt, J.O., 1995. Dispersal Distance and Direction of Reproductive European Honey Bee Swarms (Hymenoptera: Apidae). *Journal of the Kansas Entomological Society* 68, 320–325.
- Schmutterer, H., 1952. Zur Kenntnis der Buchenblattlaus *Phyllaphis fagi* (L.) (Homoptera, Aphidoidea), einer wichtigen Honigtauerzeugerin auf Buche. *Anzeiger für Schädlingskunde* 25, 1–5.  
<https://doi.org/10.1007/BF02276343>
- Schürch, R., Couvillon, M.J., Burns, D.D.R., Tasman, K., Waxman, D., Ratnieks, F.L.W., 2013. Incorporating variability in honey bee waggle dance decoding improves the mapping of communicated resource locations. *J Comp Physiol A* 199, 1143–1152. <https://doi.org/10.1007/s00359-013-0860-4>
- Schürch, R., Ratnieks, F.L.W., Samuelson, E.E.W., Couvillon, M.J., 2016. Dancing to her own beat: honey bee foragers communicate via individually calibrated waggle dances. *J Exp Biol* 219, 1287–1289.  
<https://doi.org/10.1242/jeb.134874>
- Seeley, T.D., 2019. *The Lives of Bees: The Untold Story of the Honey Bee in the Wild*. Princeton University Press.
- Seeley, T.D., 2017. Life-history traits of wild honey bee colonies living in forests around Ithaca, NY, USA. *Apidologie* 48, 743–754. <https://doi.org/10/gcm438>
- Seeley, T.D., 2016. *Following the Wild Bees: The Craft and Science of Bee Hunting*. Princeton University Press.
- Seeley, T.D., 2010. *Honeybee Democracy*. Princeton University Press.
- Seeley, T.D., 2009. *The wisdom of the hive: the social physiology of honey bee colonies*. Harvard University Press.
- Seeley, T.D., 2007. Honey bees of the Arnot Forest: a population of feral colonies persisting with *Varroa destructor* in the northeastern United States. *Apidologie* 38, 19–29. <https://doi.org/10/bwg8b8>
- Seeley, T.D., 1994. Honey bee foragers as sensory units of their colonies. *Behav Ecol Sociobiol* 34, 51–62.  
<https://doi.org/10.1007/BF00175458>
- Seeley, T.D., 1985. *Honeybee Ecology: A Study of Adaptation in Social Life: A Study of Adaptation in Social Life*, Princeton Legacy Library. Princeton University Press.
- Seeley, T.D., 1978. Life history strategy of the honey bee, *Apis mellifera*. *Oecologia* 32, 109–118.  
<https://doi.org/10/fk89x7>
- Seeley, T.D., Buhrman, S.C., 1999. Group decision making in swarms of honey bees. *Behavioral Ecology and Sociobiology* 45, 19–31. <https://doi.org/10/c3q9mm>
- Seeley, T.D., Morse, R.A., 1978. Nest site selection by the honey bee, *Apis mellifera*. *Ins. Soc* 25, 323–337.  
<https://doi.org/10.1007/BF02224297>
- Seeley, T.D., Morse, R.A., 1977. Dispersal behavior of honey bee swarms. *Psyche* 84, 199–209.
- Seeley, T.D., Morse, R.A., 1976. The nest of the honey bee (*Apis mellifera* L.). *Insectes Sociaux* 23, 495–512.  
<https://doi.org/10/b76nb3>
- Seeley, T.D., Smith, M.L., 2015. Crowding honeybee colonies in apiaries can increase their vulnerability to the deadly ectoparasite *Varroa destructor*. *Apidologie* 46, 716–727. <https://doi.org/10/f7zghj>

- Seeley, T.D., Tarpay, D.R., Griffin, S.R., Carcione, A., Delaney, D.A., 2015. A survivor population of wild colonies of European honeybees in the northeastern United States: investigating its genetic structure. *Apidologie* 46, 654–666. <https://doi.org/10/f7k9vd>
- Seeley, T.D., Camazine, S., Sneyd, J., 1991. Collective decision-making in honey bees: how colonies choose among nectar sources. *Behav Ecol Sociobiol* 28. <https://doi.org/10.1007/BF00175101>
- Seibold, S., Gossner, M.M., Simons, N.K., Blüthgen, N., Müller, J., Ambarlı, D., Ammer, C., Bauhus, J., Fischer, M., Habel, J.C., Linsenmair, K.E., Naus, T., Penone, C., Prati, D., Schall, P., Schulze, E.-D., Vogt, J., Wöllauer, S., Weisser, W.W., 2019. Arthropod decline in grasslands and forests is associated with landscape-level drivers. *Nature* 574, 671–674. <https://doi.org/10.1038/s41586-019-1684-3>
- Seijo, M.C., Jato, M.V., 1998. Palynological characterization of honeys from Galicia (Northwest Spain). *Grana* 37, 285–292. <https://doi.org/10.1080/00173139809362680>
- Sikora, L.G., 2008. Entwicklung von Schwarzspechthöhlen im östlichen Schurwald zwischen 1997 und 2007. *Ornithologische Jahreshefte für Baden-Württemberg* 24.
- Sikora, L.G., Schnitt, D., Kinser, A., 2016. Folgeuntersuchung Von Schwarzspecht-Höhlenbäumen Im Biosphärengebiet Schwäbische Alb. *Deutsche Wildtier Stiftung*.
- Smith, K.M., Loh, E.H., Rostal, M.K., Zambrana-Torrel, C.M., Mendiola, L., Daszak, P., 2013. Pathogens, Pests, and Economics: Drivers of Honey Bee Colony Declines and Losses. *EcoHealth* 10, 434–445. <https://doi.org/10.1007/s10393-013-0870-2>
- Sobek, S., Tschardt, T., Scherber, C., Schiele, S., Steffan-Dewenter, I., 2009. Canopy vs. understory: Does tree diversity affect bee and wasp communities and their natural enemies across forest strata? *Forest Ecology and Management* 258, 609–615. <https://doi.org/10.1016/j.foreco.2009.04.026>
- Soto, M.F., García, A.F., Cuesta, G.F., Prieto, J.R.F., 2011. La desecación de la laguna de Antela. *BAGE: Boletín de la Asociación de Geógrafos Españoles* 295–312.
- Sponsler, D.B., Johnson, R., 2015. Honey bee success predicted by landscape composition in Ohio, USA. *PeerJ* 3, e838. <https://doi.org/10.7717/peerj.838>
- Sponsler, D.B., Matcham, E.G., Lin, C.-H., Lanterman, J.L., Johnson, R.M., 2017. Spatial and taxonomic patterns of honey bee foraging: A choice test between urban and agricultural landscapes. *Journal of Urban Ecology* 3. <https://doi.org/10.1093/jue/juw008>
- Stange, E., Zulian, G., Rusch, G., Barton, D., Nowell, M., 2017. Ecosystem services mapping for municipal policy: ESTIMAP and zoning for urban beekeeping. *OE* 2, e14014. <https://doi.org/10.3897/oneeco.2.e14014>
- Stanley, D.A., Msweli, S.M., Johnson, S.D., 2020. Native honeybees as flower visitors and pollinators in wild plant communities in a biodiversity hotspot. *Ecosphere* 11. <https://doi.org/10.1002/ecs2.2957>
- Steffan-Dewenter, I., 2003. Importance of Habitat Area and Landscape Context for Species Richness of Bees and Wasps in Fragmented Orchard Meadows. *Conservation Biology* 17, 1036–1044. <https://doi.org/10.1046/j.1523-1739.2003.01575.x>
- Steffan-Dewenter, I., Kuhn, A., 2003. Honeybee foraging in differentially structured landscapes. *Proceedings of the Royal Society of London B: Biological Sciences* 270, 569–575. <https://doi.org/10.1098/rspb.2002.2292>

- Steffan-Dewenter, I., Münzenberg, U., Bürger, C., Thies, C., Tschardtke, T., 2002. Scale-dependent effects of landscape context on three pollinator guilds. *Ecology* 83, 1421–1432. [https://doi.org/10.1890/0012-9658\(2002\)083\[1421:SDEOLC\]2.0.CO;2](https://doi.org/10.1890/0012-9658(2002)083[1421:SDEOLC]2.0.CO;2)
- Steffan-Dewenter, I., Tschardtke, T., 2000. Resource overlap and possible competition between honey bees and wild bees in central Europe. *Oecologia* 122, 288–296. <https://doi.org/10/bj7dwz>
- Strange, J.P., Garnery, L., Sheppard, W.S., 2007. Persistence of the Landes ecotype of *Apis mellifera mellifera* in southwest France: confirmation of a locally adaptive annual brood cycle trait. *Apidologie* 38, 259–267. <https://doi.org/10.1051/apido:2007012>
- Strickling, W., 2012. AstroWin 32, available at <http://www.strickling.net/software.htm>.
- Tanasković, M., Erić, P., Patenković, A., Erić, K., Mihajlović, M., Tanasić, V., Kusza, S., Oleksa, A., Stanisavljević, L., Davidović, S., 2022. Further Evidence of Population Admixture in the Serbian Honey Bee Population. *Insects* 13, 180. <https://doi.org/10.3390/insects13020180>
- Tanasković, M., Erić, P., Patenković, A., Erić, K., Mihajlović, M., Tanasić, V., Stanisavljević, L., Davidović, S., 2021. MtDNA Analysis Indicates Human-Induced Temporal Changes of Serbian Honey Bees Diversity. *Insects* 12, 767. <https://doi.org/10.3390/insects12090767>
- Tautz, J., 2022. Communication between honeybees, 1st ed. Springer Nature, Cham, Switzerland.
- Tautz, J., Zhang, S., Spaethe, J., Brockmann, A., Si, A., Srinivasan, M., 2004. Honeybee Odometry: Performance in Varying Natural Terrain. *PLoS Biol* 2, e211. <https://doi.org/10.1371/journal.pbio.0020211>
- Taux, K., 1976. Über Nisthöhlenanlage und Brutbestand des Schwarzspechtes im Landkreis Oldenburg/Oldb. *Vogelkundliche Berichte Niedersachsen* 8.
- Tehel, A., Brown, M.J., Paxton, R.J., 2016. Impact of managed honey bee viruses on wild bees. *Current Opinion in Virology* 19, 16–22. <https://doi.org/10.1016/j.coviro.2016.06.006>
- Tehel, A., Streicher, T., Tragust, S., Paxton, R.J., 2020. Experimental infection of bumblebees with honeybee-associated viruses: no direct fitness costs but potential future threats to novel wild bee hosts. *Royal Society Open Science* 7, 200480. <https://doi.org/10.1098/rsos.200480>
- The GIMP Development Team, 2020. GIMP.
- Thompson, C.E., Biesmeijer, J.C., Allnutt, T.R., Pietravalle, S., Budge, G.E., 2014. Parasite pressures on feral honey bees (*Apis mellifera* sp.). *PLOS ONE* 9, e105164. <https://doi.org/10.1371/journal.pone.0105164>
- Tinner, W., Lotter, A.F., 2006. Holocene expansions of *Fagus silvatica* and *Abies alba* in Central Europe: where are we after eight decades of debate? *Quaternary Science Reviews* 25, 526–549. <https://doi.org/10/dmpr67>
- Tofilski, A., Oleksa, A., 2013. There are still Bee Trees in Europe. *Bee World* 90, 18–19. <https://doi.org/10/gcm452>
- Tonietto, R.K., Larkin, D.J., 2018. Habitat restoration benefits wild bees: A meta-analysis. *Journal of Applied Ecology* 55, 582–590. <https://doi.org/10.1111/1365-2664.13012>
- Tribe, G., Tautz, J., Sternberg, K., Cullinan, J., 2017. Firewalls in bee nests—survival value of propolis walls of wild Cape honeybee (*Apis mellifera capensis*). *Sci Nat* 104, 29. <https://doi.org/10.1007/s00114-017-1449-5>

- Tscharntke, T., Klein, A.M., Kruess, A., Steffan-Dewenter, I., Thies, C., 2005. Landscape perspectives on agricultural intensification and biodiversity – ecosystem service management. *Ecology Letters* 8, 857–874. <https://doi.org/10/d98cvz>
- Ulyshen, M.D., Soon, V., Hanula, J.L., 2010. On the vertical distribution of bees in a temperate deciduous forest. *Insect Conservation and Diversity*. <https://doi.org/10.1111/j.1752-4598.2010.00092.x>
- van Alphen, J.J.M., Fernhout, B.J., 2020. Natural selection, selective breeding, and the evolution of resistance of honeybees (*Apis mellifera*) against *Varroa*. *Zoological Lett* 6, 6. <https://doi.org/10.1186/s40851-020-00158-4>
- van Dooremalen, C., Gerritsen, L., Cornelissen, B., van der Steen, J.J.M., van Langevelde, F., Blacquière, T., 2012. Winter Survival of Individual Honey Bees and Honey Bee Colonies Depends on Level of *Varroa* destructor Infestation. *PLOS ONE* 7, e36285. <https://doi.org/10/f32knz>
- Vanbergen, A.J., Initiative, the I.P., 2013. Threats to an ecosystem service: pressures on pollinators. *Frontiers in Ecology and the Environment* 11, 251–259. <https://doi.org/10.1890/120126>
- Verheyen, K., Baeten, L., De Frenne, P., Bernhardt-Römermann, M., Brunet, J., Cornelis, J., Decocq, G., Dierschke, H., Eriksson, O., Hédli, R., Heinken, T., Hermy, M., Hommel, P., Kirby, K., Naaf, T., Peterken, G., Petřík, P., Pfadenhauer, J., Van Calster, H., Walther, G.-R., Wulf, M., Verstraeten, G., 2012. Driving factors behind the eutrophication signal in understorey plant communities of deciduous temperate forests: Drivers of change in forest understorey vegetation. *Journal of Ecology* 100, 352–365. <https://doi.org/10.1111/j.1365-2745.2011.01928.x>
- Visscher, P.K., Seeley, T.D., 1989. Bee-lining as a research technique in ecological studies of honey bees. *American Bee Journal* 129, 536–539.
- Visscher, P.K., Seeley, T.D., 1982. Foraging Strategy of Honeybee Colonies in a Temperate Deciduous Forest. *Ecology* 63, 1790–1801. <https://doi.org/10/ddzxwh>
- von Frisch, K., 1967. *The dance language and orientation of bees*, Belknap Press. Harvard University Press.
- Vu, V.Q., 2011. ggbiplot: A ggplot2 based biplot. R package version 0.55 755.
- Waddington, K.D., 1982. Honey bee foraging profitability and round dance correlates. *J. Comp. Physiol.* 148, 297–301. <https://doi.org/10.1007/BF00679014>
- Ward, K., Cleare, X., Li-Byarlay, H., 2022. The Life Span and Levels of Oxidative Stress in Foragers Between Feral and Managed Honey Bee Colonies. *Journal of Insect Science* 22, 20. <https://doi.org/10.1093/jisesa/ieac002>
- Weigand, M., Staab, J., Wurm, M., Taubenböck, H., 2020. Spatial and semantic effects of LUCAS samples on fully automated land use/land cover classification in high-resolution Sentinel-2 data. *International Journal of Applied Earth Observation and Geoinformation* 88, 102065. <https://doi.org/10.1016/j.jag.2020.102065>
- Wenner, A.M., Alcock, J.E., Meade, D.E., 1992. Efficient hunting of feral colonies. *BeeScience* 2, 64–70.
- Whitfield, C.W., Behura, S.K., Berlocher, S.H., Clark, A.G., Johnston, J.S., Sheppard, W.S., Smith, D.R., Suarez, A.V., Weaver, D., Tsutsui, N.D., 2006. Thrice Out of Africa: Ancient and Recent Expansions of the Honey Bee, *Apis mellifera*. *Science* 314, 642–645. <https://doi.org/10.1126/science.1132772>
- Wickham, H., 2017. *The tidyverse*. R package ver 1, 836.
- Wickham, H., 2016. *ggplot2: Elegant Graphics for Data Analysis*. Springer-Verlag New York.

- Wilfert, L., Long, G., Leggett, H.C., Schmid-Hempel, P., Butlin, R., Martin, S.J.M., Boots, M., 2016. Deformed wing virus is a recent global epidemic in honeybees driven by Varroa mites. *Science* 351, 594–597. <https://doi.org/10.1126/science.aac9976>
- Williams, N.M., Winfree, R., 2013. Local habitat characteristics but not landscape urbanization drive pollinator visitation and native plant pollination in forest remnants. *Biological Conservation* 160, 10–18. <https://doi.org/10.1016/j.biocon.2012.12.035>
- Winfree, R., Aguilar, R., Vázquez, D.P., LeBuhn, G., Aizen, M.A., 2009. A meta-analysis of bees' responses to anthropogenic disturbance. *Ecology* 90, 2068–2076. <https://doi.org/10.1890/08-1245.1>
- Winston, M.L., 1991. *The biology of the honey bee*. harvard university press.
- Wojcik, V.A., Morandin, L.A., Davies Adams, L., Rourke, K.E., 2018. Floral Resource Competition Between Honey Bees and Wild Bees: Is There Clear Evidence and Can We Guide Management and Conservation? *Environmental Entomology* 47, 822–833. <https://doi.org/10.1093/ee/nvy077>
- Young, A.M., Kohl, P.L., Rutschmann, B., Steffan-Dewenter, I., Brockmann, A., Dyer, F.C., 2021. Temporal and spatial foraging patterns of three Asian honey bee species in Bangalore, India. *Apidologie* 52, 503–523. <https://doi.org/10.1007/s13592-020-00839-1>
- Youngsteadt, E., Appler, R.H., López-Uribe, M.M., Tarpy, D.R., Frank, S.D., 2015. Urbanization increases pathogen pressure on feral and managed honey bees. *PLOS ONE* 10, e0142031. <https://doi.org/10.1371/journal.pone.0142031>
- Zahner, V., Sikora, L., Pasinelli, G., 2012. Heart rot as a key factor for cavity tree selection in the black woodpecker. *Forest Ecology and Management* 271, 98–103. <https://doi.org/10/f3xfr8>
- Zander, E., 1949. *Beitrage zur Herkunftsbestimmung bei Honig Band IV, Band IV*. Franz Ehrenwirth Verlag, München.



## STATEMENT OF INDIVIDUAL AUTHOR CONTRIBUTIONS

### Statement of individual author contributions and of legal second publication rights

<b>Publication</b> (complete reference):					
Kohl PL, <b>Rutschmann B</b> (2018) The neglected bee trees: European beech forests as a home for feral honey bee colonies. <i>PeerJ</i> 6:e4602. <a href="https://doi.org/10.7717/peerj.4602">https://doi.org/10.7717/peerj.4602</a>					
<b>Participated in</b>	<b>Author Initials, Responsibility decreasing from left to right</b>				
Study Design	BR & PLK				
Methods Development	BR & PLK				
Data Collection	BR	PLK			
Data Analysis and Interpretation	BR & PLK				
Manuscript Writing					
Writing of Introduction	PLK	BR			
Writing of Materials & Methods	BR	PLK			
Writing of Discussion	PLK	BR			
Writing of First Draft	PLK & BR				

Explanations (if applicable):

<b>Publication</b> (complete reference):					
<b>Rutschmann B</b> , Kohl PL, Steffan-Dewenter I (submitted) Foraging distances, habitat preferences and seasonal colony performance of honey bees in Central European forest landscapes.					
<b>Participated in</b>	<b>Author Initials, Responsibility decreasing from left to right</b>				
Study Design	BR	ISD	PLK		
Methods Development	BR	PLK	ISD		
Data Collection	BR	PLK			
Data Analysis and Interpretation	BR	PLK	ISD		
Manuscript Writing					
Writing of Introduction	BR	PLK	ISD		
Writing of Materials & Methods	BR	PLK	ISD		
Writing of Discussion	BR	PLK	ISD		
Writing of First Draft	BR	PLK	ISD		

Explanations (if applicable):

<b>Publication</b> (complete reference):					
<b>Rutschmann B</b> , Kohl PL, Machado A, Steffan-Dewenter I (2022) Semi-natural habitats promote winter survival of wild-living honey bees in an agricultural landscape. <i>Biological Conservation</i> 266, 109450. <a href="https://doi.org/10.1016/j.biocon.2022.109450">https://doi.org/10.1016/j.biocon.2022.109450</a>					
<b>Participated in</b>	<b>Author Initials</b> , Responsibility decreasing from left to right				
Study Design	BR	PLK	AM		
Methods Development	BR	PLK			
Data Collection	BR	PLK	AM		
Data Analysis and Interpretation	BR	PLK	ISD		
Manuscript Writing					
Writing of Introduction	PLK & BR	ISD	AM		
Writing of Materials & Methods	BR	PLK	AM	ISD	
Writing of Discussion	BR & PLK	ISD	AM		
Writing of First Draft	BR & PLK	ISD	AM		

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Benjamin Rutschmann      26.05.2022, Würzburg

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Doctoral Researcher's Name      Date      Place      Signature

Ingolf Steffan-Dewenter      24.05.2022, Würzburg

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Primary Supervisor's Name      Date      Place      Signature

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<b>Publication</b> (complete reference):					
Kohl PL, <b>Rutschmann B</b> (2018) The neglected bee trees: European beech forests as a home for feral honey bee colonies. <i>PeerJ</i> 6:e4602. <a href="https://doi.org/10.7717/peerj.4602">https://doi.org/10.7717/peerj.4602</a>					
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S2	BR				

Explanations (if applicable): Figure 2 consists of two photos.

<b>Publication</b> (complete reference):					
<b>Rutschmann B</b> , Kohl PL, Steffan-Dewenter I (submitted) Foraging distances, habitat preferences and seasonal colony performance of honey bees in Central European forest landscapes.					
Figure	Author Initials, Responsibility decreasing from left to right				
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S1	BR	PLK			
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S5	BR				
S6	BR				

Explanations (if applicable):

<b>Publication</b> (complete reference):					
<b>Rutschmann B</b> , Kohl PL, Machado A, Steffan-Dewenter I (2022) Semi-natural habitats promote winter survival of wild-living honey bees in an agricultural landscape. <i>Biological Conservation</i> 266, 109450. <a href="https://doi.org/10.1016/j.biocon.2022.109450">https://doi.org/10.1016/j.biocon.2022.109450</a>					
<b>Figure</b>	<b>Author Initials, Responsibility decreasing from left to right</b>				
1	PLK	AM	BR		
2	BR				
S1	BR				
S2	BR				
S3	BR				
S4	AM				

Explanations (if applicable): Figure 1 and figure S4 consists of photos.

I also confirm my primary supervisor's acceptance.

Benjamin Rutschmann      26.05.2022, Würzburg

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Doctoral Researcher's Name    Date    Place    Signature

## ACKNOWLEDGEMENTS

First and foremost, I would like to thank my three supervisors and my colleague Patrick Kohl. My very special thank goes to Patrick Kohl. Inspired by the work of Thomas Seeley, the two of us set up this research project on wild-living honey bee colonies in Germany and Spain. Thank you for the incredible journey we started back then in 2016, for all the help, for all the discussions on the topic and for your friendship.

I want to thank my primary supervisor Ingolf Steffan-Dewenter for welcoming me into the Zoology III department. He always was a very supportive supervisor and gave me the opportunity to develop and pursue my own research interests and ideas. Furthermore, he gave me valuable and motivating feedback for designing field experiments and during manuscript writing.

I would like to thank my second thesis supervisor Jürgen Tautz, who made it possible that I could come to Würzburg and follow my dream and study honey bees. He always had an open ear and gladly, he supported my shift from the topic of RFID technology in bee research into the Graduate School of Life Sciences (GSLs) to study the fascinating, neglected topic of wild-living honey bee colonies in Europe.

I warmly thank my external supervisor Axel Brockmann, who was my first mentor in the scientific community when I joined his lab in Bangalore before starting my PhD in Würzburg. The continuous visits in India allowed me to work with the mesmerizing Asian honey bee species, importantly also to study their behaviour as wild, undomesticated species.

I am thankful to the GSLs for granting me a Career Development Fellowship. Furthermore, I received generous funding from the GSLs to visit two international conferences and a spring summer school. The whole GSLs team is working hard every day to make our life as PhD students easier. My thanks go especially to Irina Pleines-Meinhold, Heike Schrenk, Sebastian Michel and all the other current and former members of the team. Moreover, I would like to thank BayIND for supporting my research stays in India to study Asian honey bees, BAYHOST for funding my project in Krakow with a mobility grant, the Unibund Würzburg for contributing to the lab analysis costs, and last but not least beegut for supporting our research on wild-living honey bees generously (thanks Samuel, Hannes, Martina, and the whole team!).

This work would also not have been possible without the help of our technical and administrative personal. I am extremely grateful to the whole HOBOS team: Anneli Kiessling, Gaby Läubisch, Konrad Öchsner, Gerhard Vonend, Markus Kiunke, Hartmut Vierle, Kristina Vonened, Marina Kretzschmar and Florian Schimpf. Furthermore, I am grateful to Birgit Bünger, Michaela Jäger, Susanne Schiele, Johann Kaderschabek, Norbert Schneider, Thomas Igerst, Gudrun Grimmer, Beate Krischke for their help and support regarding all imaginable issues. Also, thank you Amy/Jie Zhang for helping me with my first steps in GIS, something that became a passion.

I would like to thank Heinrich Demmel, who was also keeping bees at the Gut Gieshügel.

Thanks to the working students Stefan Fallert, Kerstin Hikel, Jonathan Heinze and Maria Scherrer for help with various projects.

Thanks to Sandra and Sonja who shared the office with me. I was often on field work but with you guys it was always pleasant.

I am grateful to my PhD fellows of whom many became good friends. In particular, I thank Cristina, Mehdi, Justine, Patrick, Federico, Giulia, Phil, Gemma, Daniel, Elena, Sebbo, Nicola, Fabienne, Birte and all other members of the Zoology III group, a group full of vibrant, enthusiastic ecology students from all parts of the world. Thanks also to the many Post-Docs: Mark Otieno, Sarah Redlich, Fabrice Requier, Douglas Sponsler, Sam Boff Vieira, Zsolt Kárpáti, Antonia Mayr and all others.

Thanks also to the gardening team. On Hubland, we grow the most delicious organic tomatoes in Lower Frankonia!

I would like to thank all forest managers, rangers, and administrative staff in the forest regions where we worked. Andreas Henkel and Manfred Großmann from the Hainich national park; Rüdiger Jooß from the biosphere reserve Swabian Alb; Ulrich Mergner from the state forest Ebrach and Johannes Wurm from the Nürnberger Reichswald.

I spent a wonderful time in Simon's Town, South Africa. Thank you, Karin Sternberg and Jenny Cullinan, for taking me with you on the hunt for wild *Apis mellifera capensis* colonies. Thanks, Geoff Tribe, for the trip to Porterville, chasing banded bee pirates. The work of you three inspired me to look at honey bees differently as holistic creatures. Thanks Duncan and Diana Reekie, staying at your place was a magic experience.

I would like to warmly thank Adam Tofilski: you are such inspiring scientist and mentor. Thank you and your whole team for hosting and supporting me during my research stay in Krakow.

I would like to express my gratitude that I was able to take part in two woodpecker conferences and getting the opportunity to present the results, namely the occupation of black woodpecker cavities by honey bee colonies in the wonderful place of Białowieża, Poland. I also thank my friend Dror Noe for travelling there with me. I am indebted to Luis Sikora, Norbert Wimmer, Volker Zahner, and many other ornithologists and woodpecker researchers for the help and support for our projects.

I want to thank my Master student Kristin Garner, a curious and very talented bee enthusiast. The field work in the Kellerwald and the Hainich national parks was great.

I am deeply thankful to my colleagues and friends Alejandro Machado and Xosé Santos in Galicia.

Marianne Beck supported us with a charming, reliable car for our field work. “Gertrud Kunigunde” never let us down.

I would like to thank my dear friends from Innsbruck; it broke my heart to leave back then in 2016. Thank you, Verena, Sara, Burks, Berni, Wolfi, Matze, Dani, Georgi, Serge, Jambi, and many more. Also, many thanks to you Janine, for pushing me to pursue my fascination about honey bees in a scientific way. One of my biggest thanks goes to my “Imkerpate” in Axams, Reinhard Hetzenauer.

I would like to thank my flatmates during my time in Gerbrunn. Especially Karo, Tobi, Margaritari, Celine, Elisa, Pia, Meike, Alex, Lilly and last but not least Berat, who was also a great climbing buddy.

Julie, merci d'avoir cru en moi et de m'avoir toujours soutenue.

Before the pandemic I moved to Würzburg, Würzburg city. During the pandemic, Sartoriusstr. 14 became my home. Thanks Clemens, Judith and Johanna for the wonderful time, the friendship, your presence and all the support.

I would like to thank Ricarda Scheiner, Laura Değirmenci, Felix Schilcher: we had a fruitful scientific exchange due to our RFID projects.

I am thankful for the inspiring time with journalists and reporters: Christoph Würzburger for making a wonderful little reportage; Jean-Baptiste Pouchain for documenting our trip in the forests in Coburg; Dimi Dumortier and Toon Angeli for accompanying us to many of our field trips in the Swabian Alb and spending some memorable Karaoke night together with us; Andreas, Frank, Lea for shooting parts of “Kielings wilde Welt” with us in the forest; Iris Gesang for adapting the script and making a scientifically sound story out of it; Ingo Arndt for coming to the field with us and providing us with the most wonderful pictures about honey bees.

Sebastian Roth and Felix Remter from Munich – thank you for all the fertile discussions, your comments on my work, and on our common interest on wild-living honey bees. Also, thanks to André Wermelinger, Francis Cordillot, Frank Krumm, and many more for lively discussions on the topic in Würzburg and other places.

I had the opportunity to exchange thoughts and ideas at many national and international conferences. The groups from Halle, Bern, and many more were so much fun. Thanks Oleg, Delphine, Silvio, Laura, Jana, and all the others.

Thanks to all the people in NCBS: Hema, Aridni, Divya, Elea, Prabhu, Tejal, Hinal, Hanumanta, Manal, Helena, Rikesh, Aroumoy, Sophia, Franziska, Friedrich, Mahesh, Allison, Sruthi, Ebi, Ben, Neethu and all the other students, especially also the ones in Parijata.

I thank Sebastian Spiewok who motivated us to write some articles for the beekeeping audience.

Judith, I could not have finished this thesis without your support. You had so much patience with me. Your motivation, and incredible support gave me strength. I am deeply thankful for that and for your enduring love.

I am truly indebted to my parents, my siblings, and my grandparents. I am very thankful that you always encouraged me and believed in me especially when I had doubts. Mam & Pa, Andri, Sereina, Nani & Grospi, Grosmami & Grospapi thank you so much for always being there for me.



## LIST OF PUBLICATIONS

### Published in peer reviewed journals

- Rutschmann B\***, Kohl PL\*, Machado A, Steffan-Dewenter I (2022) Semi-natural habitats promote winter survival of wild-living honeybees in an agricultural landscape." *Biological Conservation* 266 (2022): 109450. <https://doi.org/10.1016/j.biocon.2022.109450> \*These authors contributed equally to this work.
- Schilcher F, Hilsmann L, Rauscher L, Değirmenci L, Krischke M, Krischke B, Ankenbrand M, **Rutschmann B**, Mueller MJ, Steffan-Dewenter I, Scheiner R (2021) In Vitro Rearing Changes Social Task Performance and Physiology in Honeybees. *Insects* 13.1 (2021): 4. <https://doi.org/10.3390/insects13010004>
- George EA, Thulasi N, Kohl PL, Suresh S, **Rutschmann B**, Brockmann A (2021) Distance estimation by Asian honey bees in two visually different landscapes. *Journal of Experimental Biology*. <https://doi.org/10.1242/jeb.242404>
- Kohl, PL, **Rutschmann B** (2021) Honey bees communicate distance via non-linear waggle duration functions. *PeerJ* 9: e11187 <https://doi.org/10.7717/peerj.11187>
- Young AM\*, Kohl PL\*, **Rutschmann B\***, Steffan-Dewenter I, Brockmann A, Dyer FC (2021) Temporal and spatial foraging patterns of three Asian honey bee species in Bangalore, India. *Apidologie* 52.2: 503-523 <https://doi.org/10.1007/s13592-020-00839-1> \*These authors contributed equally to this work.
- Otieno M, Joshi NK, **Rutschmann B** (2021) Flower visitors of *Streptocarpus teitensis*: implications for conservation of a critically endangered African violet species in Kenya. *PeerJ* 9: e10473. <https://doi.org/10.7717/peerj.10473>
- Requier F, Paillet Y, Laroche F, **Rutschmann B**, Zhang J, Lombardi F, Svoboda F, Steffan-Dewenter I (2020) Contribution of European forests to safeguard wild honeybee populations. *Conservation Letters*. 13:e12693. <https://doi.org/10.1111/conl.12693>
- Kohl PL, Thulasi N, **Rutschmann B**, George EA, Steffan-Dewenter I, Brockmann A (2020) Adaptive evolution of honeybee dance dialects. *Proc. R. Soc. B*. 28720200190. <http://doi.org/10.1098/rspb.2020.0190>
- Kablau A, Berg S, **Rutschmann B**, Scheiner R (2020) Short-term hyperthermia at larval age reduces sucrose responsiveness of adult honeybees and can increase life span. *Apidologie* 51, 570–582. <https://doi.org/10.1007/s13592-020-00743-8>
- Kohl PL\*, **Rutschmann B\*** (2018) The neglected bee trees: European beech forests as a home for feral honey bee colonies. *PeerJ* 6:e4602. <https://doi.org/10.7717/peerj.4602> \*These authors contributed equally to this work.

Submitted to peer reviewed journals

**Rutschmann B**, Kohl PL, Steffan-Dewenter I (submitted) Foraging distances, habitat preferences and seasonal colony performance of honeybees in Central European forest landscapes.

Kohl PL, **Rutschmann B**, Steffan-Dewenter I (submitted) Population demography of feral honeybee colonies in Central European forests.

Non-refereed national publications

**Rutschmann B**, Kohl PL, Roth S (2018) Beelining – wie man wild lebende Honigbienen findet (Beelining – how to find wild honey bee colonies). *German Beekeeping Journal*.

Kohl PL and **Rutschmann B** (2018) Versteckt und unerforscht: Wild lebende Honigbienen in unseren Wäldern (Hidden and poorly studied: wild honey bee colonies in our forests). *German Beekeeping Journal*.

Service as reviewer

*Oecologia, Journal of Apicultural Research, Apidologie, Behavioral Ecology and Sociobiology*

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## **CURRICULUM VITAE**





## Affidavit

I hereby confirm that my thesis entitled “Occurrence and population density of wild-living honey bees in Europe and the impact of different habitat types on their foraging and overwintering success” is the result of my own work. I did not receive any help or support from commercial consultants. All sources and / or materials applied are listed and specified in the thesis.

Furthermore, I confirm that this thesis has not yet been submitted as part of another examination process neither in identical nor in similar form.

Würzburg, 26.05.2022  
Place, Date

Signature

## Eidesstattliche Erklärung

Hiermit erkläre ich an Eides statt, die Dissertation „Vorkommen und Populationsdichte von wild lebenden Honigbienen in Europa und die Auswirkungen unterschiedlicher Habitattypen auf ihr Sammelverhalten und den Überwinterungserfolg” eigenständig, d.h. insbesondere selbständig und ohne Hilfe eines kommerziellen Promotionsberaters, angefertigt und keine anderen als die von mir angegebenen Quellen und Hilfsmittel verwendet zu haben.

Ich erkläre außerdem, dass die Dissertation weder in gleicher noch in ähnlicher Form bereits in einem anderen Prüfungsverfahren vorgelegen hat.

Würzburg, 26.05.2022  
Ort, Datum

Unterschrift