

ANGLIA RUSKIN UNIVERSITY

FACULTY OF SCIENCE AND ENGINEERING

**THE NESTING ECOLOGY OF FOSSORIAL SOLITARY BEES**

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A thesis in partial fulfilment of the requirements of Anglia Ruskin University  
for the degree of Doctor of Philosophy

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**Abstract**

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In the UK, fossorial solitary bees account for approximately 52% of bee species, but their nesting ecology has received little attention in the academic literature. This research gap is largely a result of logistical barriers, which centre around the difficulties associated with locating nests and the time resources required to collect quality data. It is further compounded by the lack of standardised methodologies associated with this type of analysis. This PhD used three methodological approaches to overcome these obstacles, investigate the efficacy of the methods and shed light on the ecological requirements of fossorial solitary bees.

The three principal methods enlisted in this research were a web-based citizen science project, which was linked with a field-based observational study of nest sites and finally a manipulative field experiment. All three of these methods were found to provide important insights into solitary bee nesting ecology and the field study benefitted significantly from being linked with the citizen science project, which provided accurate locations of active nesting sites.

The citizen science data demonstrated the capacity of *Andrena fulva*, *Andrena cineraria*, *Halictus rubicundus* and *Colletes hederæ* to nest within a broad range of environmental conditions including slope and ground cover, while the field-based study indicated that high-density nesting relies on the presence of specific environmental characteristics, such as bare ground and sandy soil. Significant interspecific differences in nesting characteristics were also identified indicating the need for further study of individual species. The field experiment found that the creation of bare ground and the maintenance of vegetation at a low level increased the nesting density of fossorial species six-fold.

This research has explored and interrogated the efficacy of these three methodological approaches. Furthermore, it has elucidated some of the important environmental considerations for the protection and provision of suitable solitary bee nesting sites.

**Keywords:** Nesting, solitary bees, citizen science, conservation

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Stephanie Maher, March 2019





# 1. Introduction

## 1.1 Pollinators

Bees (Hymenoptera: Apoidea: Anthophila) first evolved roughly 100 million years ago in the early Cretaceous period (Poinar and Danforth, 2006) and can be found on all continents except Antarctica. They have a (largely) mutualistic relationship with Angiosperms (flowering plants), whose rapid diversification and radiation can be attributed to the evolution of animal-mediated pollen dispersal (Danforth *et al*, 2006). Of all the pollinators, bees are the largest and most important group with more than 20,000 extant species (IPBES, 2016; discoverlife.org). The Angiosperms provide a nectar source; a substance high in sugar for energy, pollen; a high protein substance which bees use to feed their young and resins; which bees use to feed and medicate larvae. In return bees transport pollen from male to female plants of the same species thereby facilitating germination and the persistence of the plants' progeny through time. Many bee species are polylectic meaning that they are generalist species and will feed from many different plant species, while others are more specialised to particular plant types and are considered oligolectic.

Bee diversity (Biesmeijer *et al*, 2006) and insect biomass (Hallmann *et al*, 2017) have been shown to be in decline, while anthropogenic change has led to increased rates of pollinator species extinction (Ollerton *et al*, 2014). Although much of the evidence of these patterns comes from only a handful of geographical locations (predominantly the UK, Western Europe, North America). There is agreement that the most important drivers of these changes are habitat loss, agricultural intensification, pesticide use, invasive species and climate change (Brown *et al*, 2016, Potts *et al*, 2010). Despite this consensus, experts also agree that there remain significant gaps in our knowledge of pollinators, both in terms of their ecology and their geographic location, and calls for further research to address these shortcomings are both numerous and frequent (Hallmann *et al*, 2017, Ollerton, 2017, Vanbergen *et al*, 2012; Breeze *et al*, 2011).

## 1.2 Pollinator ecosystem services

Pollinators are a crucial part of the global ecosystem, not just because they are highly speciose but also because they influence the productivity of approximately 75% of crop species (Klein *et al*, 2007). These pollination

services are worth an estimated \$235 – 577 billion to the global economy annually (IPBES, 2016). Worldwide the number of managed honeybee hives is estimated to have grown by approximately 45% over the period 1961 to 2009 (Aizen and Harder, 2009). However, with an increasing reliance on honeybees to pollinate agricultural crops, these same crops become highly vulnerable to the threats currently facing honeybee population stability (Potts *et al*, 2010). Wild bees are highly speciose with diverse ecological and physical traits, meaning their inclusion in any pollination system greatly improves the system's resilience (Breeze *et al*, 2011). This exemplifies the lack of resilience that is inherent in any system largely reliant on one component and compounds the importance of diversifying with wild bees. Furthermore, honeybees alone are not sufficient to provide quality pollination services. A recent study of UK apple orchards (a highly valuable crop) found that of the £92.1m per annum that insect pollinators contribute to the economic outputs of orchards; £51.4m can be attributed to solitary bees (Garratt *et al*, 2016). In Denmark, the bee communities at strawberry farms were found to primarily constitute ground-nesting solitary bee species (Ahrenfeldt *et al*, 2019), while in Canadian strawberry crops, honeybee pollination was shown to result in lower yields than wild bee pollination (MacInnis and Forrest, 2019).

Solitary bees constitute roughly 90% of the approximately 275 wild bee species in the UK, but to date have largely been absent from the research and policy agenda (Wood *et al*, 2016). Not only do solitary bees represent a significant portion of UK biodiversity, they are also important and highly efficient pollinators (Garratt *et al*, 2016). Solitary bees can be loosely divided into three groups on the basis of their nesting strategy. There are: aerial nesting species (nest in pre-existing cavities in the landscape); fossorial species (construct a nest subterraneously by digging down into the ground) and; parasitic species (do not build a nest but parasitise the nests of others). This thesis focuses on fossorial solitary bees, which represent the majority of UK solitary bee species and of which, seven are classified as UK BAP priority species (jncc.gov.uk). These bees are informally known as “miners” and include all species of Andrenidae and Meilittidae (Potts *et al*, 2005). Most species of Halictidae, Colletidae and Anthorophoridae are also categorised as mining bees.

### 1.3 The resource requirements of fossorial solitary bees

Fossorial solitary bees require two principal resources to be present in a landscape in order to survive and thrive: forage and a nest site or, in other words, food and a home. However, there exists a significant shortfall in current knowledge regarding both the importance and characteristics of nesting resources. England's National Pollinator Strategy ([gov.co.uk/government/publications/national-pollinator-strategy-2014-2024-implementation plan](http://gov.co.uk/government/publications/national-pollinator-strategy-2014-2024-implementation-plan)) places a heavy emphasis on the provision of flower-rich habitats for pollinator conservation as there is little quantitative data regarding supply or demand of nesting resources (Dicks *et al*, 2015). Furthermore, there is a widely accepted assumption that nest sites are not a limiting resource, particularly with regard to ground-nesting species, however there is a lack of evidence to support this conclusion (Roulston and Goodell, 2011). The absence of empirical research surrounding nesting preferences and their interaction with landscape features is particularly compelling (Sardinas and Kremen, 2014). This research deficit may be attributable both to the difficulty associated with finding nests (Wood *et al*, 2016) and the complex task of unravelling the impacts of nest site variables from local forage resources (Roulston and Goodell, 2011). As a result of these complications, the community of foraging bees at a particular site has been accepted as being representative of the species nesting there (Potts *et al*, 2005). While there is no significant body of empirical evidence to either support or debunk this claim, one recent study that sampled both foraging and nesting bees on hybrid sunflower fields in the Sacramento Valley found that all nesting bees were represented in the foraging community (Sardinas *et al*, 2015). However the foraging community represented more than double the species richness of the nesting community (six nesting species versus fourteen foraging species). Whilst this was a relatively small scale study, it does indicate that using the foraging community as a proxy for the nesting community is not justified. Furthermore, foraging surveys may underestimates the number of species benefitting from the use of an area as a nest site by omitting species of cuckoo bee, which may be more often found in the vicinity of the hosts' nests than on forage.

### 1.4 Conservation of solitary bees

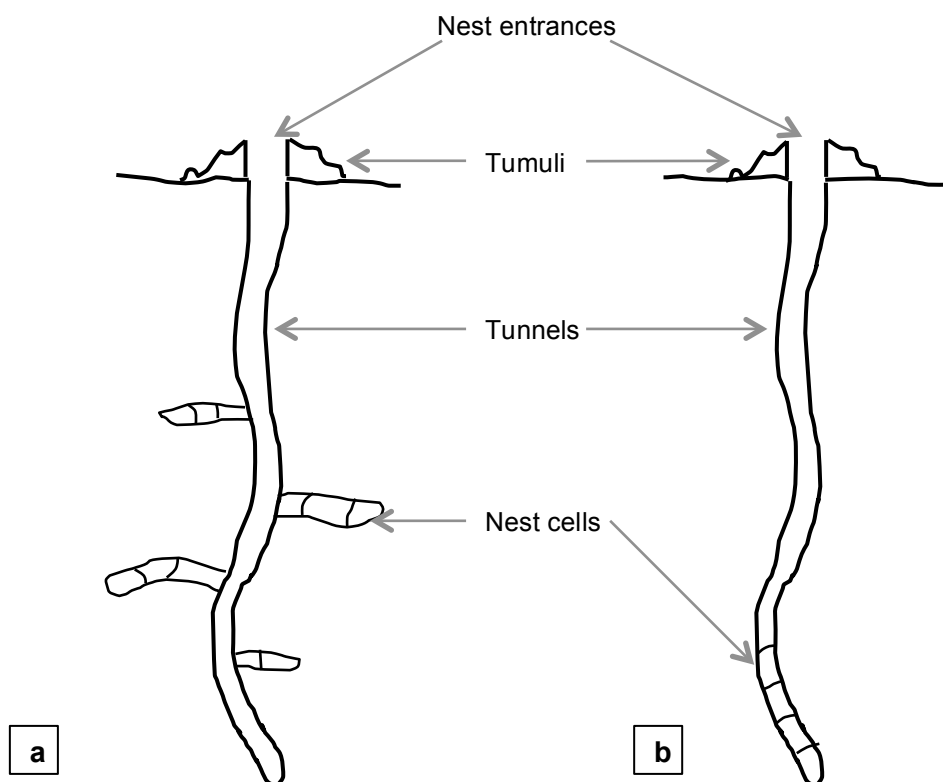
While the energy and attention that has been put into pollinator conservation has greatly improved, there remain some shortfalls in current conservation practice with regard to solitary bees. The sown flower agri-environment

options, which are designed to enhance pollinator species richness and abundance rarely include plant species that flower during March and April (Wood *et al*, 2016), which is a crucial period for some solitary bee species. It has recently been reported that only 34.7% of solitary bee species present on agricultural land in south east England actually utilise the sown flowering resources to any meaningful degree (Wood *et al*, 2016). There have been many calls for more fundamental and ecological research of solitary bees (Everaars and Dormann, 2014) both to advance our ecological knowledge and to better inform policy and management strategies. Furthermore, this need for more basic ecological research of pollinators to manage and reduce the factors threatening their survival was a key recommendation of a 2012 workshop report on linking research and policy (Vanbergen *et al*, 2012). Looking to the future, it is important that we establish how to protect, conserve and provide for this group. In order to do this, we must first establish more thorough understanding of the fundamental needs of these important insects. This approach has the potential to lead to meaningful impact and policy amendments in terms of agri-environment schemes, conservation practices, urban planning and land management.

A substantial knowledge gap exists around solitary bee nesting ecology. At the most fundamental level, more data on solitary bee nest site characteristics are required. Closing this knowledge gap will allow us to understand the impacts of current conservation actions and planning policies on the availability of suitable nest sites and help us to better plan for the future. Management practices such as tilling (Williams *et al*, 2010), pesticide use (Godfray *et al*, 2014) and irrigation (Sardinas *et al*, 2016) could all theoretically undermine the suitability of a site for nesting, however, until we better understand the fundamental ecology of nest sites, it is difficult to study or model the management impacts in any meaningful way. Furthermore, we cannot hope to design best practice guidelines for land managers or effective policy to help conserve and protect this large group of species without empirical understanding of their basic ecology. Empirical nesting data would have the potential to improve modelling studies, conservation actions and urban planning by facilitating these endeavours to have a basis in empirical science. The implications for these data extend to the local, regional and national levels. Much of the empirical evidence of solitary bee nesting ecology that exists comes from outside the UK in climates, ecosystems and with species assemblages substantially different from what exists here (Sardinas *et al*, 2016; Sardinas *et al*, 2015; Cane, 2015; Wuellner, 1999).

Therefore, this research has the potential to provide clear and relevant data specific to the British context.

The subterranean structure of fossorial bee nests is variable, but most structures are minor variants of the generic structure shown in Fig. 1.1, adapted from Cane and Neff (2011). The nest entrance is surrounded by a pile of soil excavated from the ground known as a tumulus. A tunnel extends down into the soil and nest cells are created. These cells may be constructed in branches off the main tunnel (Fig.1.1a) or within the tunnel itself (Fig.1.1b). Each nest cell is provisioned with a ball of pollen, which a single larva will feed on until it is ready to emerge as an adult.



**Figure 1.1:** Diagram depicting the two most common structures of fossorial bee nests. **a** represents a branching nest in which nest cells are created in branches off the main tunnel. **b** depicts a non-branching nest, wherein the nest cells are situated in the main tunnel itself. Adapted from Cane and Neff (2011)

### 1.5 Top-down statistical modelling of bee populations

Although little energy has been focussed on empirically describing nest sites, there has been a growing interest in, and reliance upon, top-down modelling studies to inform and improve conservation policy. Many of these have attempted to model wild bee distribution and vulnerability across landscapes (Lonsdorf, 2009; Olsson *et al*, 2015; Everaars and Dormann, 2014). However, when it comes to nesting preferences these studies lack the basic empirical evidence to support their assumptions, relying on expert opinion in the most robust cases, or proxies and major oversimplifications in others (Sardinas *et al*, 2015). The Lonsdorf model was the first quantitative model to attempt to predict pollinator abundance in a landscape (Lonsdorf, 2009). Although the model was well validated in two of the study sites (explaining approximately 80% of the variation) it failed to predict a significant amount of variation in the third site, which the authors attributed to a lack of fine scale empirical data regarding foraging and nesting resources. Olsson *et al*, (2015) made some improvement to the Lonsdorf model by incorporating behavioural components, specifically, central place foraging theory was brought in to extend the model so that it could be used more effectively in more complex landscape systems. However, they did not expand on nesting variables and so there remained a shortfall in this approach. Everaars and Dormann (2014) attempted to model the way wild solitary bees respond to landscape configuration using individual based simulation models (IBMs). Whilst the authors tried to build nesting preferences into the model, they simplified the nesting categories to above ground or subterranean sites. Furthermore, subterranean nests were modelled as equally likely to occur anywhere in the foraging habitat while aerial nests had a built in preference for field margins. These assumptions are potentially leading to poor prediction because recent evidences indicates that soil nesting bees also prefer to nest in margins and almost certainly diverge from long held beliefs about where they will and will not site a nest (Sardinas *et al*, 2016). Other parameters in this experiment, particularly those relating to foraging behaviour, were based on proxies derived from body size. Although this approach has value (Greenleaf *et al*, 2007), what actually happens in situ has been shown to be far more complex (Zurbuchen *et al*, 2010). One of the most robust, holistic and recent top-down models of pollination ecosystem services was published by Häussler *et al*, (2017). This model explicitly integrates the preferential use of higher quality nesting resources, although it focuses primarily on

bumblebees. To truly determine the validity of this or any other model, a comparison with empirical data is required. In the long run, even top-down approaches would benefit from better fundamental understanding of how solitary bees operate in the physical environment.

One significant issue with modelling studies is that they can create the illusion that they are adding to the body of evidence surrounding conservation management practices. Keitt (2009) in their modelling study recommended providing nesting habitat without any indication of what nesting habitat is. This kind of generic recommendation is insufficient in the practical management of species and can lead to a one size fits all approach in which only the most robust species are provided for. Modelling studies have enormous potential to help both understand ecology and to plan conservation actions. However, studies which attempt to address bee distribution or pollination services could be greatly improved if there existed a wider knowledge base concerning the ecological requirements of the solitary bees.

### *1.6 Thesis aims*

The aim of this thesis is to better understand what constitutes suitable nesting sites for aggregate-nesting, fossorial solitary bees in the UK and to begin to understand how to provide suitable nesting habitat using empirical approaches. As some of the barriers to fossorial bee nesting research are the temporal and logistical issues associated with gathering empirical data on nesting ecology, particular attention is paid in this thesis to the designing, trialling and validating of various methods. I aimed to use these methods to contribute to bridging the knowledge gap relating to solitary bee nesting ecology, and as such, there are two central themes to this work: the evaluation of methods and the environmental characteristics of nests sites. Figure 1.2 illustrates how these themes are dealt with in this thesis.

### 1.7 Structure of thesis

The following is a brief summary of each of the following chapters and their contents.

**Chapter 2** uses a citizen science approach to gather data on the characteristics of solitary bee nest sites. The research questions addressed in this chapter are:

- Can citizen science be used to examine the nesting aggregations of solitary bee species?
- Do *Andrena fulva* (Müller in Allioni, 1766), *Andrena cineraria* (Linnaeus, 1758), *Halictus rubicundus* (Christ, 1791) or *Colletes hederæ* (Schmidt and Westrich, 1993) associate with particular environmental characteristics in choosing a nest site?
- Do broad scale environmental variables affect the nest aggregation size of these four solitary bees?

**Chapter 3** explores the spatial differences in nesting density between and within fossorial bee nest sites. The research questions addressed in this chapter are:

- Do differences in the environmental characteristics of a site influence the nesting density of *A.fulva*, *A.cineraria* and *C.hederæ* among aggregations?
- Do differences in environmental characteristics within a nesting aggregation contribute to the clumped spatial distribution of aggregations for these species?
- Are there interspecific differences in the environmental characteristics of where nest sites occur?

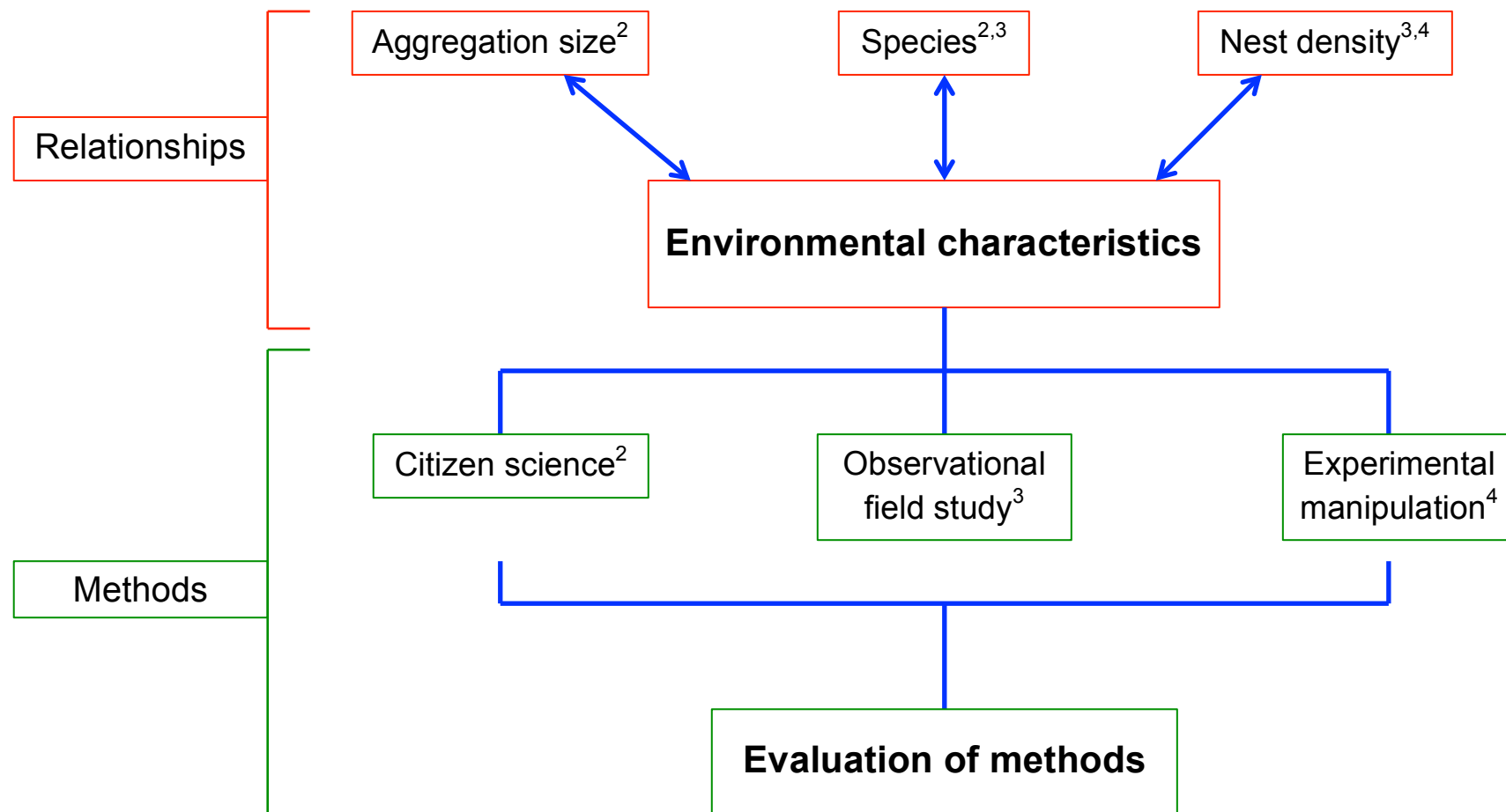
**Chapter 4** describes a field experiment, which explores how vegetation management practices impact fossorial bee and wasp nesting. The research questions addressed in this chapter are:

- Does vegetation cover impact solitary bee nesting?



- How do four different vegetation management strategies impact the nesting density of solitary bees and wasps?
- Can *Halictus rubicundus* return to a nesting site from which it has been excluded, within the first year of management change?

**Chapter 5** brings together the preceding chapters to establish the novel contributions of this work and its implications for wildlife management and conservation.



**Figure 1.2:** Summary of thesis structure. Numbers in superscript indicate chapters where the relationship or method is addressed. The environmental characteristics and evaluation of methods are the principal themes running through the thesis.

## **2: The Solitary Bee Project: Using citizen science to examine the nesting ecology of fossorial solitary bees**

### **2.1 Introduction**

#### *2.1.1 Solitary bees*

Solitary bees constitute roughly 250 species of wild bee in the UK, but there remains significant knowledge gaps with regard to their ecology (Wood *et al*, 2016). This group, which encompasses many important pollinators, including those of high value crops such as apples (Garratt *et al*, 2016), has faced diversity loss (Biesmeijer *et al*, 2006) declines in relative abundance (Bartomeus *et al*, 2013) and extinctions (Ollerton *et al*, 2014). There are two principal resources that solitary bee populations require in order to survive and proliferate: suitable and robust nest sites for their offspring to successfully mature in, and sufficient forage material both for their own survival and to provision their young. The foraging requirements of bees have been well studied (Strickler, 1979; MacIvor *et al*, 2014; Dicks *et al*, 2015; Wood *et al*, 2016) as they are believed to be the principal limiting resource. However, within a landscape, Gathmann and Tscharnkte (2002) have speculated that nest sites may be a limiting factor for solitary bees more often than forage, although it is clear that these resources must work in tandem. There is a lack of empirical research regarding the nesting requirements of fossorial solitary bees in the UK (but see Potts and Willmer, 1997). Although there are multiple reasons for this gap, the difficulties associated with finding sufficient nest sites in the field and the lack of robust experimental methods are two of the most significant barriers. These issues are further compounded by the short flight periods of many UK solitary bees, which result in only a small window of time when active nest sites can be discovered and examined.

#### *2.1.2 Solitary bee nesting ecology*

There have been two main types of empirical study performed on ground nesting bees' nesting ecology; those studies that focus on single species (Potts and Willmer, 1997; Wuellner, 1999; Julier and Roulston, 2009; Xie *et al*, 2013) and those that examine the effects of nest site suitability on wider metrics such as species richness, abundance and community composition (Grundel *et al*, 2010; Sardinas and Kremen, 2014; Martins *et al*, 2018). A mixed picture emerges when the species-specific studies are taken together, indicating that

there may be significant interspecific diversity in solitary bee nest site preferences. Within the UK, Potts and Willmer (1997) empirically examined *Halictus rubicundus* (Christ, 1791) nest aggregations and identified a preference for softer soils with a moderate slope and southern aspect. Wuellner (1999) showed that the North American bee *Dieunomia triangulifera* (Vachel, 1897) preferentially nests in areas of bare, compacted soil with a warm soil surface temperature and close to visual landmarks. A study in Chinese *Camellia oleifera* (Abel) orchards found that *Andrena camellia* (Wu, 1977) preferred loose, moist and low temperature soil conditions (Xie *et al*, 2013). The abundance of *Peponapis pruinosa* (Say, 1837), a specialist pollinator of pumpkin in the USA, has been shown to be negatively related to soil clay content and positively related to soil irrigation (Julier and Roulston, 2009). In all of these studies, nest temperature seems to be an important characteristic and comes through either directly, in that individuals have been observed nesting in areas with particular soil temperatures (Wuellner, 1999; Xie *et al*, 2013), or indirectly, where they are observed nesting at sites whose physical characteristics seem to confer some thermal benefits (Potts and Willmer, 1997). Although the direction of preference seems to be somewhat mixed. There is a similar pattern when it comes to the physical characteristics of the soil, where these attributes appear to be uniformly important but the specifics of what is preferred seem to vary on a species to species basis.

Across geographical regions the availability of ground nesting resources has been shown to be a strong predictor of bee abundance (Potts *et al*, 2003; Sardinias and Kremen, 2014), species richness (Grundel *et al*, 2010) and community composition (Potts *et al*, 2005; Grundel *et al*, 2010). A recent study from Quebec, Canada showed that bee diversity in apple orchards, blueberry and raspberry fields was influenced by the presence of suitable nesting resources (Martins *et al*, 2018). Again, the important characteristics of the nesting resource varies, but the availability of bare ground (Potts *et al*, 2005), sloped terrain (Sardinias and Kremen, 2014), sandy soils (Cane, 1991) and soils with low organic matter content (Grundel *et al*, 2010) are factors that have proven to be important in at least some contexts.

Taken together, these studies show that nesting resources, as related to ground nesting species, are important for the survival and proliferation of bees in general. However, due to geographical differences in community composition and interspecific differences in nest site preferences, a 'one size fits all' approach will not be sufficient in this context. This conclusion was also reached

by Kim *et al* (2006) who found that across agricultural landscapes, ground nesting bee density was impacted by proximity to semi-natural habitat and edaphic factors of the individual sites. However, species differed in their response to the specific measured variables.

### 2.1.3 Citizen science approaches

In this study, I used a citizen science approach to circumvent the logistical and temporal issues associated with in-situ solitary bee nesting research and to attempt to identify and examine nesting aggregations on a national scale. Citizen science is an effective method of up-scaling research projects, both temporally and spatially, by capturing far more data than could be achieved by one individual (Pocock and Evans, 2014). It is also an excellent way to engage and educate the public about their local wildlife and scientific research (Kremen *et al*, 2011). Citizen scientists can be defined as members of the public who assist with scientific research on a voluntary basis (Cohn, 2008) and the UK boasts a long history of highly engaged amateur naturalists recording wildlife. The State of Nature reports are an example of the power of public recording. The most recent publication reported that 56% of the 8000 UK species studied, declined over the last fifty years (Hayhow *et al*, 2016). Many of these data were a result of the British public observing and recording wildlife voluntarily and although these population trends are discouraging, we would not have the evidence to show they exist without this level of public engagement. However, the success of citizen science projects, even in this country, is not guaranteed and there is much to be learned from both the successes and difficulties of previous endeavours.

Hypothesis-led projects use citizen science as a tool to test hypotheses defined by professional scientists. This differs from traditional citizen science approaches which simply require participants to observe and record wildlife without a specific goal. Although the hypothesis-led approach has gained momentum in the last few years, its potential remains largely unharnessed (Roy *et al*, 2016). The Big Bumblebee Discovery was a one-year UK citizen science project that targeted schools and aimed to assess the multi-level impact of the landscape on bumblebee abundance and diversity (Roy *et al*, 2016). The participants (pupils aged 7-11) were asked to count the bumblebees that visited a lavender plant in a five minute time periods and then identify the bees according to six colour types. This study also investigated the accuracy of the submitted data through photo

verification. This process revealed a high rate of species misidentification, meaning the data could not be used to formally test the original hypotheses. A North American citizen science project by Delaney *et al* (2008) had more success both in terms of hypothesis testing and data quality. This study used volunteers to assess the presence of native and non-native crab species in the intertidal zones at sites in the North East USA and participants were found to be able to identify the species with 95% accuracy. The authors partly attributed this result to the fact that they narrowed the study to just three crab species, thereby making the protocol easier for participants to learn and implement. Pocock and Evans (2014) had similar success with a UK based project on *Cameraria ohridella* (Deschka and Dimic, 1986) (the horse chestnut leaf miner). Also a hypothesis-led study, this project assessed the rapid range expansion of this insect by having participants report instances of the distinctive damage it causes to the leaves of horse chestnut trees. It also attempted to assess larval parasitism levels, although the data accuracy for this portion of the study was low as it required participants to rear the larvae at home. A similar observation was made by Birkin and Goulson (2015) that asking participants to make a large time investment risks undermining volunteer engagement.

#### *2.1.4 Data quality in nature-based citizen science projects*

The debate around data accuracy in citizen science projects and how to optimise it remains contentious (Aceves-Bueno *et al*, 2017; Specht and Lewandowski, 2018) and there is no guaranteed formula or template to adhere to when designing citizen science projects that will guarantee high data quality. Aceves-Bueno *et al* (2017) attempted to quantitatively assess the capacity of volunteers to record accurate data in line with professionals by adopting an approach similar to metaanalysis, where the results of multiple, similar studies are pooled and analysed. They found that in 62% of the cases studied, citizen science data did not differ significantly from professionally-collected data. Based on their analysis they made two recommendations for the design of citizen science projects. Firstly, they emphasised employing quality assurance methods such as the collection of reference data and/or monitoring the activity of the citizens closely. Secondly, they advise designing the project with the skill of the citizen in mind. In other words, simplify the tasks and provide training where possible. They also recommend recruiting citizens with either an economic

or health stake in the project as they are more motivated to collect quality data. In summary, this analysis took a fairly pessimistic view in terms of achieving high data quality from citizen science. However, the methodology of this study and therefore its results have been questioned. Specht and Lewandowski (2018) argued that this type of analysis is inappropriate in this context for three principal reasons 1. Many studies that attempt to assess citizen data quality use professionally collected data as the reference sample. However, research has shown that professional is not always accurate and the more robust method would be to assess both citizen and professionally collected data against a common reference. The authors also point out that in terms of precision, citizens and professionals have been shown to perform to roughly the same standard. 2. There is a high degree of methodological variability between studies that attempt to assess citizen data quality. In terms of the species being studied, the tasks the citizens are required to action, the level of training, participant characteristics and so on. Therefore these studies are not suitable to be aggregated for the purpose of metaanalysis or summary statistics. 3. When the data from multiple participants are viewed only in aggregate and not individually, and compared to a single professional reference, a biased and pessimistic view of data quality from the whole group can form. Following these criticisms, Specht and Lewandowski (2018) made three fundamental recommendations for evaluating citizen science data quality which can be summarised as 1. Assess both the accuracy and precision of citizen and professionally collected data in the same way. 2. Only aggregate data when the first condition is met and when the assessed metrics represent a single unified concept. 3. Pay close attention to what is driving any variation in citizen data quality and adjust the method based on those drivers. To summarise, the data quality of citizen science data cannot be aggregated and quantitatively assessed to provide a quantitative framework of best practice until enough appropriate studies (as outlined by Specht and Lewandowski, 2018) exist.

As no empirical framework exists for the design of robust citizen science projects, I used previous studies as a grounding for project design (Delaney *et al*, 2008; Pocock and Evans, 2014; Birkin and Goulson, 2015; Roy *et al*, 2016). A random forest algorithm was used to analyse the data. This powerful statistical method has been used in a variety of ecological studies due to its capacity to cope with nonlinear data with predictor variables that are often part of complex interactions (Olden *et al*, 2008). It has been used to model the habitat of

endangered crayfish in Japan (Usio, 2007) and even to estimate solitary bee populations from images of ground nests in New Zealand (Hart and Huang, 2011).

I propose that citizen science has the potential to be an effective tool for the study of fossorial bee nesting ecology and can provide valuable insights into this knowledge gap, difficult to achieve via more traditional survey approaches.



### 2.1.5 Research questions and hypotheses

This project set out to address three **research questions**:

1. Can citizen science be used to examine the nesting aggregations of solitary bee species?
2. Do *Andrena fulva*, *Andrena cineraria*, *Halictus rubicundus* or *Colletes hederæ* associate with particular environmental characteristics in choosing a nest site?
3. Do broad scale environmental variables affect the nest aggregation size of these four solitary bees?

#### **Hypotheses:**

1. Citizen science can be used to examine the nesting aggregations of solitary bee species and will generate substantial data for the four species included in the study. The number of accurate records submitted by participants will be equal between species.
2. All four species will exhibit similar preferences for sloped, bare ground in open sun. These preferences will be particularly strong for *A. fulva* and *A. cineraria* as these species are active in early spring and will require their nest sites to be thermally optimised due to low temperatures at this point in the season. *Colletes hederæ* will similarly exhibit strong preferences for these conditions as its flight period is in late autumn.
3. Environmental variables do affect the size of solitary bee nesting aggregations. Nesting aggregations will be significantly larger in areas of lawn or bare, sloped ground that are not shaded to optimise thermal conditions, digging conditions and nest stability.

## 2.2 Methods

### 2.2.1 Optimisation of record accuracy

Based on the citizen science literature, a number of control measures were taken when designing The Solitary Bee Project to optimise the chances of receiving high quality data. These included:

1. Narrowing the study to four species: This decision stemmed from the high data accuracy recorded by Delaney *et al* (2008) in their study focussed on three crab species.
2. Simplifying the tasks: The study by Roy *et al* (2016) on bumblebees found that participants had difficulty matching described colour forms to what they observed in the field. Therefore, this study used simplified methods, only requiring participants to answer multiple choice questions, in order to circumvent this potential issue.
3. Not requiring repeated visits or a large time commitment: Birkin and Goulson (2015) and Pocock and Evans (2014) have recommended reducing participant time investment. Both struggled to get sufficient data from activities that required substantial participant time and commitment in their respective projects.

### 2.2.2 Species selection

A process of elimination protocol was used to choose four solitary bee species to build the project around. This selection process was based on a number of criteria designed to make the project both scientifically sound and to improve the chances of receiving accurate records. Fundamentally, this approach aimed to identify species that are already recorded by the public and have been regularly observed in the East of England (based on records submitted to iRecord) and nest in aggregations (established based on Bees, Wasps and Ants Recording Society (BWARS) species accounts and Falk (2015)) (see Tables 2.1 and 2.2 for a detailed breakdown of the process). The publicly submitted solitary bee iRecord records for the period October 2015 - October 2016 across the UK were analysed and the following ten species were pulled out: *Colletes hederæ*, *Colletes succinctus* (Linnaeus, 1758), *Andrena clarkella* (Kirby, 1802), *Andrena fulva*, *Andrena cineraria*, *Halictus rubicundus*, *Halictus tumulorum* (Linnaeus, 1758), *Dasypoda hirtipes* (Fabricius, 1793), *Lasioglossum morio* (Fabricius, 1793), *Anthophora bimaculata* (Kirby, 1802) (Tables 2.1 and

2.2). From this list of ten, four species of scientific interest and trait diversity were chosen on the basis of the following reasoning: of the four species, two were early season emergers, being on the wing from February or March (*A. fulva* and *A. cineraria*), one was a mid-season emerger (*H. rubicundus*) and is active from June or July, and one was a late season emerger (*C. hederæ*), becoming active in late August or September. *Halictus rubicundus* is the one species for which we have empirical, U.K. based data on their nest site preferences and so this species was chosen to build upon that knowledge. *Halictus rubicundus* also has an interesting biology, as it has a relatively long flight period and exhibits both solitary and eusocial behaviour in different parts of its range (Eickwort *et al*, 1995). *Colletes hederæ* is already the subject of a fairly large scale citizen science project run by BWARS (bwars.com). This highly successful project has monitored the spread of this species across the U.K. since its first sighting here in 2001 (BWARS, 2018). There were two major benefits of including this species in the study; from a citizen science perspective, some members of the public are already accustomed to submitting records for this species, with more than 600 sightings submitted to iRecord between Oct 2015 and Oct 2016. From a scientific perspective, this is a species of great interest. Having first appeared in the U.K. in 2001 after spreading beyond its continental European native range, it has spread rapidly across the country (Roberts and Vereecken, 2010). Adding to the knowledge base of this species contributes not only to fundamental solitary bee knowledge but also to our knowledge base concerning non-native species. Finally, all four of these species are morphologically distinct and thus, possible for the public to identify.

### 2.2.3 Brief biology of the four study species

*Andrena fulva* is a species native to the UK. It is very common in southern England and is increasing in the Scottish part of its range, although it remains rare in Ireland (Falk, 2015). It is a univoltine species and does much of its foraging on spring-flowering shrubs such as hawthorn and fruit trees. Larvae develop underground and overwinter as adults before emerging the following spring. *Andrena cineraria* is also a native species and is widespread across the UK and Ireland. It is univoltine and forages on a similar range of plants to *A. fulva*. *Andrena cineraria* also overwinters as

adults in the natal cells (bwars.com, 2019<sup>1</sup>). *Halictus rubicundus* is a native species, common throughout Britain and Ireland. It is highly polylectic and females can be seen on the wing from March through to October (Falk, 2015). Peak activity for this species occurs from June, when males become active. *Halictus rubicundus* is reported as solitary in the northern parts of its range, but eusocial in the south (Soucy, 2002). *Colletes hederæ* is the only non-native species included in this study and was first recorded in the UK in Dorset in 2001 (bwars.com, 2019<sup>2</sup>). It is the last solitary bee to emerge each year in the UK and principally utilises ivy as both a pollen and nectar source (Falk, 2015).

#### 2.2.4 Study design

The study was designed to measure variables that are accessible to non-experts (Roy *et al*, 2016). Participation in the project did not require any specialist equipment or training, nor did it ask volunteers for a major time commitment. Each individual could submit as few or as many records as they wished, with no obligation for repeated visits or monitoring of the site. The participants were requested to upload a photo of the bee and the site, so an expert could compare the photographs to the submitted data for validation purposes, although this was not made compulsory. This approach was favoured as many of the most knowledgeable and experienced potential participants would be disinclined to photograph specimens, particularly common species such as those in this study, as they are confident in their identification skills. A project-specific website (thesolitarybeeproject.org) (Appendix 1) was designed and launched both as an information tool and a portal through which participants could submit records.

The secondary aim of The Solitary Bee Project was to use it as a tool to identify nest sites for fine scale ecological analysis. This was attempted in the summer of 2016 without the aid of public records but not enough nest sites could be identified for robust analysis due to their cryptic nature. As participants in the citizen science project needed to record location data, this information was used for a separate but linked analysis (Chapter 3).

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<sup>1</sup> <http://www.bwars.com/bee/andrenidae/andrena-cineraria>. Date accessed: 16/07/19

<sup>2</sup> <http://www.bwars.com/bee/colletidae/colletes-hederæ>. Date accessed: 16/07/19

Participants were asked to answer the following simple questions regarding the nesting aggregation. These questions were designed with regard to the recommendations outlined by previous citizen science projects, detailed in section 2.2.1:

1. How many nests were there?
  - a. 1-10
  - b. 11-30
  - c. 31-50
  - d. 51+
2. How sloped was the ground?
  - a. Flat
  - b. Sloped
  - c. Vertical
3. How much shade was the aggregation in?
  - a. Aggregation was completely in the open
  - b. Aggregation was adjacent to trees or buildings that could provide shade
  - c. Aggregation was completely shaded by trees or other structure
4. What was the ground cover like?
  - a. Bare earth
  - b. Lawn
  - c. Under mulch
  - d. Other

**Table 2.1:** Protocol for choosing species included in citizen science project. Public records examined were those submitted to iRecord between 10/10/2015 and 10/10/2016.

Group	UK species	Records	Top two most recorded species in group OR all species with 20 or more records	Species	Records
<i>Colletes</i>	9	697		<i>C. hederæ</i>	614
				<i>C. succinctus</i>	22
<i>Hylaeus</i>	12	70		<i>H. communis</i>	19
				<i>H. hyalinatus</i>	17
<i>Andrena</i>	67	2063		<i>A. hattorfiana</i>	38
				<i>A. clarkella</i>	35
				<i>A. fulva</i>	34
				<i>A. haemorrhoa</i>	28
				<i>A. cineraria</i>	20
<i>Panurgus</i>	2	16		<i>P. banksianus</i>	12
				<i>P. calcaratus</i>	4
<i>Halictus</i>	8	108		<i>H. rubicundis</i>	72
				<i>H. tumulorum</i>	33
<i>Lasioglossum</i>	34	290		<i>L. calceatum</i>	58
				<i>L. morio</i>	43
<i>Dasypoda</i>	1	26		<i>D. hirtipes</i>	26
<i>Melitta</i>	4	21		<i>M. tricincta</i>	11
				<i>M. haemorrhoidalis</i>	7
<i>Macropis</i>	1	11		<i>M. europaea</i>	11
<i>Anthophora</i>	5	807		<i>A. plumipes</i>	664
				<i>A. furcata</i>	51
				<i>A. bimaculata</i>	32
<i>Eucera</i>	2	31		<i>E. longicornis</i>	31

**Table 2.2:** Protocol for choosing species included in citizen science project. Cells highlighted in green were considered for inclusion. Those in red were dropped from consideration when they failed to meet a selection criterion.

Species	>20 records?	Nests consistently in ground aggregations?	East England records?
<i>C. hederæ</i>	✓	✓	✓
<i>C. succinctus</i>	✓	✓	✓
<i>H. communis</i>	X		
<i>H. hyalinatus</i>	X		
<i>A. hattorfiana</i>	✓	X	
<i>A. clarkella</i>	✓	✓	✓
<i>A. fulva</i>	✓	✓	✓
<i>A. haemorrhoea</i>	✓	X	
<i>A. cineraria</i>	✓	✓	✓
<i>P. banksianus</i>	X		
<i>P. calcaratus</i>	X		
<i>H. rubicundis</i>	✓	✓	✓
<i>H. tumulorum</i>	✓	✓	✓
<i>L. calceatum</i>	✓	X	
<i>L. morio</i>	✓	✓	✓
<i>D. hirtipes</i>	✓	✓	✓
<i>M. tricineta</i>	X		
<i>M. haemorrhoidalis</i>	X		
<i>M. europaea</i>	X		
<i>A. plumipes</i>	✓	X	
<i>A. furcata</i>	✓	X	
<i>A. bimaculata</i>	✓	✓	✓
<i>E. longicornis</i>	✓	✓	X

### *2.2.5 Data validation*

Records received were validated by Stephanie Maher using a traffic light system where records coded as 'red' were immediately rejected, 'green' records were immediately accepted and 'yellow' records were examined more closely before being assigned either 'red' or 'green' status. Ultimately, records were rejected if the species identification was incorrect, the bee had not been seen at the nest site or if there were missing data.

### *2.2.6 Participant recruitment*

Immediately preceding and following the launch of the project, it was advertised through BWARS and through the personal contacts of those involved in the project. This resulted in the project being 'target marketed' either to those with a professional interest e.g. university contacts, or amateurs who are members of a wildlife recording society and are accustomed to wildlife recording. At the beginning of June 2017, three months into the project, a press release was circulated to local and national media outlets by the university press office. This press release resulted in a number of local and national radio and television appearances and newspaper articles about The Solitary Bee Project. In this way, the project was marketed to a much wider group of potential participants from this point on.

### *2.2.7 Data analysis*

In the case of the data generated by The Solitary Bee Project, general linear modelling was not appropriate as the dependent variable (size of aggregation) was measured on a categorical scale and therefore the data did not meet the assumptions of normality or unboundedness. Instead, a random forest algorithm was developed as a classification model (500 trees) (Breiman, 2001) to try to predict the number of nests in an aggregation (categorical outcome of 1-10; 11-30; 31-50 or 51+ nests) based on the level of shade, slope and type of ground cover. A random variable, consisting of values between 0 and 100, was added to the model so that the relative contributions of the other variables could be examined. Random forest is a machine learning approach that iteratively uses different subsets of the data to create many decision trees. Every tree is then tested against the data that has not been used to make that tree. The error associated with each of these iterations is then averaged to produce the 'Out of Bag Error' or the prediction error of the random forest. In this way, random



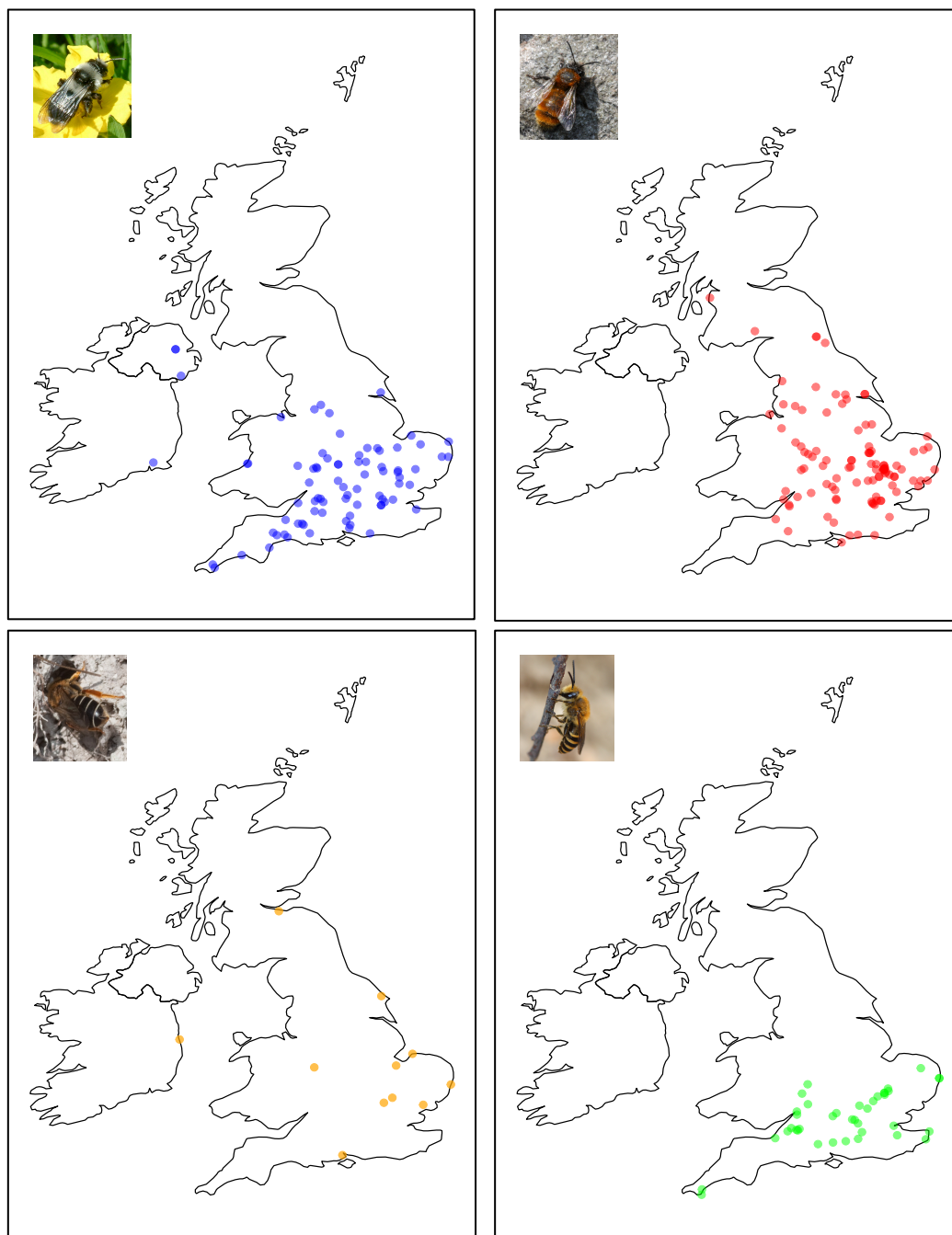
forest combines many classifications trees in order to produce more accurate classifications and is a robust method of examining variable importance and modelling interactions between variables (Cutler *et al*, 2007).

All analyses were carried out in R version 3.5.1 (R Core Team, 2015). The R package 'randomForest' was used for the random forest analysis (Liaw and Wiener, 2002).

## 2.3 Results

### 2.3.1 Summary of records

In total, The Solitary Bee Project collected 396 records from across the UK and Ireland in 2017. Two hundred and thirty six of these records were assessed as being accurate and indicative of an active nesting aggregation (Fig. 2.1). There was substantial variability in the number of accurate records submitted per species. Close to ten times more accurate records were received for *A. fulva* compared with *H. rubicundus* (105 and 11 accurate records respectively). There were also marked interspecific differences in the proportion of accurate records submitted, where again *A. fulva* had the highest proportion (74.5% of records were accurate) and *H. rubicundus* the lowest (20.8% of records were accurate) (Table 2.3).



**Figure 2.1:** Distribution maps for the records submitted for each species: *A. cineraria* – blue dots (n=81); *A. fulva* – red dots (n=105); *H. rubicundus* – orange dots (n=11); *C. hederæ* – green dots (n=39). Darker pigments indicate overlapping records. Photos of bees by Dr. Thomas Ings.

**Table 2.3:** Summary of records by species

Species	Total records	Accurate records
<i>A. fulva</i>	141	105 (74.5%)
<i>A. cineraria</i>	127	81 (63.8%)
<i>H. rubicundus</i>	53	11 (20.8%)
<i>C. hederæ</i>	71	39 (54.9%)

### 2.3.2 Solitary bee nest sites

Records were used to build a picture of the areas in which the four species were nesting in terms of their ground cover (Table 2.4), shade (Fig. 2.2), and slope (Fig. 2.3). Standard statistical tests, such as chi square which can be used to examine preferences, were not appropriate for use in this context. This is because these statistical analyses rely on the assumption that every option that is being tested was always available to every individual. This was not the case in this study as the four species of bee were active at different times in the season, therefore, no statistical test was used to interrogate interspecific differences. For individual species, this assumption was not met as individuals may have existed in fragmented habitats where they could not access areas which represented all levels of each variable.

#### ***Andrena fulva***

The ground cover of *A. fulva* aggregations was reported to be grass in 56% of aggregations and bare in 34% of aggregations (Table 2.4). Eighty two percent of records reported this species to be nesting in flat ground with no slope. The records also indicated that *A. fulva* has a broad tolerance for shade with 68% of aggregations reported to be experiencing at least partial shade. That said just 10% of aggregations were reported be in full shade.

#### ***Andrena cineraria***

53% of *A. cineraria* aggregations were reported to be fully in the open and 44% were reported to experience some shade (Table 2.4). Just 3% of aggregations were reported as fully shaded. Sixty seven percent of *A. cineraria* aggregations occurred on flat ground and 30% were sloped. Sixty four percent of aggregations

were reported to be in a grassy area and 30% occurred in bare ground. This reflects a very similar pattern to *A. fulva*.

### ***Halictus rubicundus***

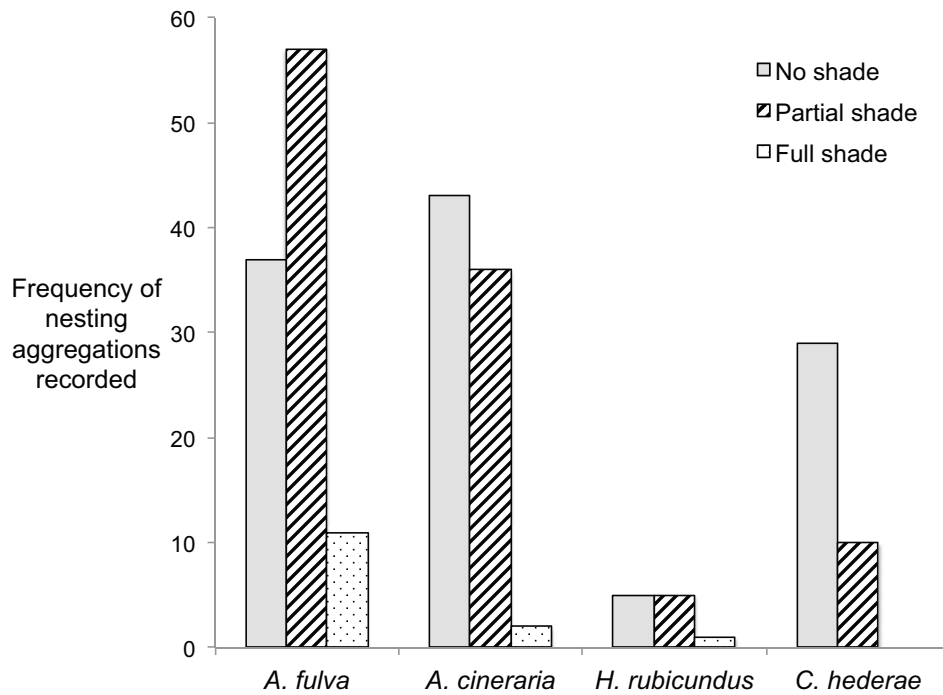
From the 11 accurate records of *H. rubicundus* aggregations, 54% occurred on flat ground and 36% on sloped ground (Table 2.4). Most records (74%) specified that the ground was bare. No strong trend manifested in terms of shade with 45% of records specifying the aggregation was in the open and a further 45% specifying the aggregation to be in partial shade.

### ***Colletes hederæ***

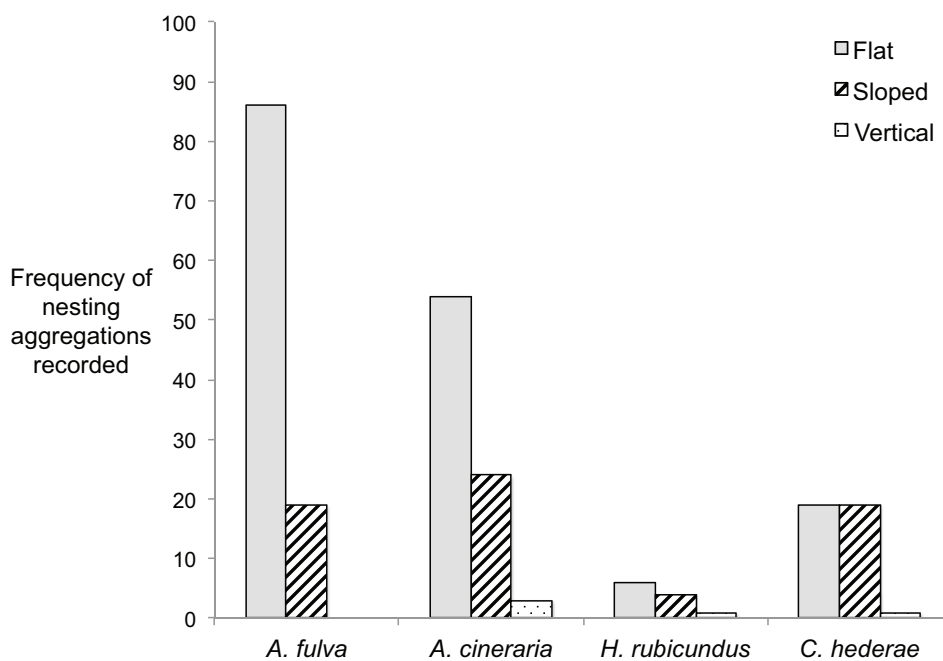
A majority (74%) of *C. hederæ* aggregations were reported to be subject to no shade at all and no aggregations were reported to be fully shaded (Table 2.4). Aggregations occurred equally on on flat ground and on sloped ground. Sixty two percent of recorded aggregations reported grass as the primary ground cover, with 36% recorded as bare.

**Table 2.4:** Species summaries of proportion of records received in each category for the three environmental variables.

	<b>Shade</b>	<b>Slope</b>	<b>Ground cover</b>
<i>A. fulva</i> (n=105)	No shade: 36% Part shade: 54% Full shade: 10%	Flat: 82% Sloped: 18% Vertical: -	Bare: 34% Grass: 56% Other: 10%
<i>A. cineraria</i> (n=81)	No shade: 53% Part shade: 44% Full shade: 3%	Flat: 67% Sloped: 30% Vertical: 3%	Bare: 30% Grass: 64% Other: 6%
<i>H. rubicundus</i> (n=11)	No shade: 45% Part shade: 45% Full shade: 10%	Flat: 54% Sloped: 36% Vertical: 10%	Bare: 74% Grass: 18% Other: 8%
<i>C. hederæ</i> (n=39)	No shade: 74% Part shade: 26% Full shade: -	Flat: 49% Sloped: 49% Vertical: 2%	Bare: 36% Grass: 62% Other: 2%



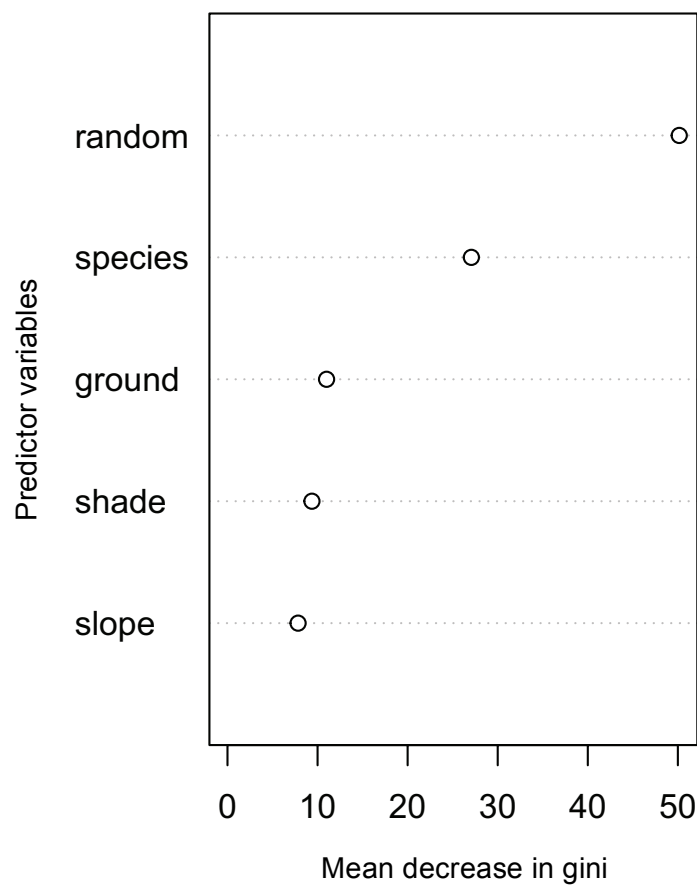
**Figure 2.2:** Frequency distribution of the number of nesting aggregations in shade, partial shade and full shade for each of the four study species.



**Figure 2.3:** Frequency distribution of the number of nesting aggregations in flat, sloped and vertical terrain for each of the four study species.

### 2.3.3 Predicting the aggregation size

The random forest algorithm could not successfully predict the size of an aggregation and had an 'out of the bag' error rate of 57.6%. The variable importance plot shows that the random variable inserted into the model was the main driver of the model (Fig. 2.4). Therefore, none of the measured variables are considered to be useful predictors of the number of nests in an aggregation.



**Figure 2.4:** Variable importance plot. This plot illustrates a list of the most predictive variables in descending order. In this case, the random variable, which is listed first, contributes the most to the model.

#### 2.3.4 Participant reach

The media attention three months into the project resulted in what might be considered a 'high quantity, low accuracy' scenario where in the first three months of the project, a total of 89 records were received, of which 82% were verified as accurate and 70% included one or more photographs. However, the records submitted post media were of a substantially lower quality. Of the records submitted in the three months post media, only 49% were validated and taken through for data analysis. Not only were many of the records removed from the analysis, the percentage of records with accompanying images dropped to 33%.

#### 2.3.5 Site recruitment

The project also set out to identify active nesting aggregations for fine scale analysis. This aim was met and 27 active nesting aggregations were sampled. The interspecific differences in the number of records submitted was in some ways reflected in this portion of the analysis (Table 2.5). For example, no *H. rubicundus* nest sites were successfully sampled, which is reflective of the low success rate in obtaining accurate records for this species. However, this pattern did not hold for *A. fulva*, which was the most well-recorded species in the main but had the second lowest number of sites in the fine scale analysis. *Colletes hederæ*, conversely, received the second lowest number of record submissions at 71 with a 54.9% accuracy rate but had the highest number of sampled sites in the fine scale analysis.

**Table 2.5:** Summary of the number of sites surveyed at a fine scale, per species

Species	No. of sites
<i>A. fulva</i>	7
<i>A. cineraria</i>	9
<i>H. rubicundus</i>	0
<i>C. hederæ</i>	11



## 2.4 Discussion

The Solitary Bee Project collected 236 accurate records of solitary bee nest site locations of four species from across the UK. Although the resolution of the data was not fine enough to identify any influence of environmental factors on aggregation size, the project did reveal some interesting trends in terms of where these four species nest in a landscape. All species were found to have broad tolerances for the measured environmental characteristics but they did exhibit some differences in their nest site characteristics. Furthermore, these results suggest that interspecific differences may be reduced when species are closely related and have similar flight seasons, although more rigorous testing is required to substantiate this theory. This study took a novel approach to overcoming the logistical barriers associated with solitary bee nesting research and showed that citizen science can be an effective tool in this context, although data resolution would benefit from some methodological changes in the future. There was substantial engagement with the project and the near 400 submitted nest site records reflect the engagement the public had with this lesser known but important group of bees.

### 2.4.1 Shade

Nest sites that are in full sun are understood to be attractive to ground nesting species as they experience increased soil temperature (Brockmann, 1979; Potts and Willmer, 1997). *Colletes hederæ* followed this trend and was most often reported from sites that experienced no shade (Fig. 2.2). The Solitary Bee Project was based on broad categories of shade (none, some, complete) and so could not pull apart the level of shade the aggregations in the “some” category experienced throughout the day. However, there was evidence that all species could tolerate at least some level of shade, including *H. rubicundus* which has previously been found to show a strong preference for sites in full sun (Potts and Willmer, 1997). In fact, *A. fulva* was found to nest in open areas in very few cases (Fig 2.2), which indicates that there may be benefits to nesting in a shaded site that outweigh the disadvantages. In this study, a number of participants reported *A. fulva* to be nesting near and in some cases, directly beneath, trees. Although *A. fulva* is polylectic, a study of their pollen loads in Cardiff, Wales, found that females were primarily foraging on flowering trees such as maple and cherry (Paxton, 1991), and so the tendency of *A. fulva* to nest in shaded areas may be a function of their decision to nest in close proximity to forage material. As *A. fulva* is active in early spring, when flowering

plants are scarce, there may be more pressure on it than summer flying species to site a nest close to abundant foraging resources. Trees can also alter the soil environment in ways that may facilitate nesting. For example, fossorial insects do not nest in hard-packed soil (Gliński *et al*, 2011; Srba and Heneberg, 2012) but tree roots can break up compact or hardened soil layers, which may make the area habitable. Trees also help to mitigate extremes in soil surface temperature (Edmondson *et al*, 2016), which may be beneficial to larvae developing underground (Potts and Willmer, 1997).

#### 2.4.2 Slope

Sloped, south-facing ground experiences higher soil temperatures and this is thought to confer significant benefit to species that nest in these areas (Potts and Willmer, 1997). In this study, the majority of *C. hederæ* nesting sites were reported to be on sloped ground, in line with previous findings for this species (Bischoff *et al*, 2004). Conversely, most *A. fulva* nesting sites occurred on flat ground (Fig 2.3), a tendency that has previously been noted for other *Andrena* species that occur in North America (Youssef and Bohart, 1968). Neither *H. rubicundus* nor *A. cineraria* demonstrated a trend for any particular terrain gradient. This mixed picture is not unusual. When Srba and Heneberg (2012) examined the nest sites of five species of digger wasp (four Sphecids and one Crabronid), they also identified interspecific differences in the slope of the nesting areas, noting that there were preferences for both sloped and flat terrain. Potts and Willmer's (1997) study of *H. rubicundus* nesting aggregations found that across sites studied there was no correlation between slope and nesting density, however, within a site this species nested at higher densities in areas with steeper slopes. So, although these species possess the capacity to nest in variously sloped ground, these may not represent optimal nest sites and further study is required to untangle this relationship.

#### 2.4.3 Ground cover

Seventy four percent of *H. rubicundus* nest sites were found in bare ground, but for the other three species (*A. fulva*, *A. cineraria* and *C. hederæ*), grass was reported as the primary ground cover of the aggregation in the majority of records. Bare ground has been shown to encourage solitary bee nesting (Gregory and Wright, 2005; Dicks *et al*, 2010), but the results of the current study show the capacity of some species to nest in grassy areas. For *A. fulva* and *A. cineraria*, many participants reported the aggregations to be in mown

lawn or grazed grass, indicating that these species can tolerate significant anthropogenic disturbance to the surface area of the nest site. This is supported by the fact that many of the aggregations recorded in bare ground occurred on footpaths with frequent pedestrian or vehicular traffic. Although little work has been done on the impact of disturbance on solitary bee nest sites, Ullmann *et al* (2016) found that tilling the soil to a depth of 40cm delayed the emergence of the squash bee *P. pruinosa* in an agricultural landscape, but roughly 50% of the bees survived this management. This suggests that disturbed landscapes in both rural and urban contexts, where the level of disturbance is not so extreme, could potentially contribute to the persistence of ground nesting solitary bee populations. This may indicate that these species have a broad tolerance for the surface characteristics of the ground they nest in and that other factors are more important for optimising their nest site location. Srba and Heneberg (2012) studied the nest site characteristics of five species of digger wasp and found a general trend of preference for low vegetation cover with some species more strongly selecting for this than others. However, they noted that one species showed no preference for vegetation cover and dug nests in areas with up to 80% cover. The authors did identify strong interspecific differences in the wasps' soil requirements. Evidence shows that edaphic factors such as soil particle size (Cane, 1991) and organic matter content (Grundel *et al*, 2010) can drive the presence and density of fossorial bees and wasps and so it is possible that the subterranean environment is a more dominant feature compared to surface characteristics for some species (this will be explored further in Chapter 3).

#### 2.4.4 Interspecific similarities and differences

Overall, *A. fulva* and *A. cineraria* exhibited similar trends in terms of their environmental characteristics. Furthermore, these two species were sometimes reported to be nesting together, a scenario I observed personally. This, taken with previous work (Youssef and Bohart, 1968) suggests that these closely related species with strongly overlapping flight seasons favour similar nesting conditions. This pattern has also been identified in the Colletidae family, where steep, south-facing, sandy slopes are preferred (Bischoff *et al*, 2004). This tentatively suggests that phylogenetic relatedness could potentially play a role in defining the nest site requirements of a species, but more research with the specific aim of testing this hypothesis is required.

There seemed to be some inter-familial differences, with the majority of *C. hederæ* and *A. fulva* nest sites having values for slope and shade in opposite

directions. Furthermore, *H. rubicundus* was the only species for which bare soil was identified as the primary ground cover in the majority of records, although this is based on a small sample. Summarising previous research on species from different families demonstrates some divergences in their preferred nest site characteristics (Potts and Willmer, 1997; Wuellner, 1999; Julier and Roulston, 2009; Xie *et al*, 2013). These differences may have originally manifested for many reasons. Although all species have a common goal of finding a nest site that is suitable for larval development and subsequent survival of offspring, they may differ significantly in what constitutes optimal soil for digging and what is the optimal position for forage. There is a high degree of morphological diversity within the ground nesting solitary bees, particularly with regards to body size. Smaller bees are likely to have shorter foraging ranges (Greenleaf *et al*, 2007) and so may be more restricted in requiring a nest site in close proximity to suitable food resources. Furthermore, oligolectic species may be constrained by the presence of their food plants. Body size may also impact the ability of an individual to excavate nests in harder soils. Indeed, Cane (1991) found that larger bees tend to nest in soils with higher clay content, and nesting in these conditions results in higher energy costs (Srba and Heneberg, 2012). Species' flight periods may also play a role. Spring flying species such as *A. fulva* and *A. cineraria* have to cope with lower soil temperatures which will alter the digging conditions (Xie *et al*, 2013). For example, soils with larger particles, such as sand, freeze more easily than those with a higher proportion of fine particles such as clay. In this way clay rich soils may be easier to dig in the spring, but in the summer months sandy soils represent a lower energy excavation. The closely related digger wasps *Ammophila pubescens* (Curtis, 1836) and *A. sabulosa* (Linnaeus, 1758) have been shown to preferentially nest in areas where the soil has a low gravel content (Srba and Heneberg, 2012).

Although there is evidence to suggest that phylogenetic relatedness can help determine nesting requirements, Cane (1991), found that ground nesting bees exhibit substantial variability in their preferences for soil grain size. They also noted that the species with more similar preferences did not necessarily align along taxonomic relationships. The resolution may be that in certain families e.g. Colletidae and Andrenidae, taxonomic relatedness does help to delineate nesting preferences but this is not true for all fossorial families. Looking ahead, significantly more research would be required to gain a clear understanding of how these factors interact.

#### 2.4.5 Aggregation size

The data collected did not distinguish any differences in environmental conditions for different sized aggregations. This may have been due to the broad nature of the questions asked, which is an inherent risk when designing hypothesis-led citizen science projects. There is a careful balance to be struck between establishing complexity in the methods for scientific robustness but maintaining simplicity in order to encourage participation and ensure accurate reporting. The categorical nature of the questions seemed to have a negative effect on data resolution as the categories were constrained. This was especially apparent with the *C. hederae* data for which 77% of records reported the nest site to consist of more than 51 nests (the highest category). A separate, fine scale study of a subset of the aggregations revealed a wide range in the mean number of nests per square metre (Chapter 3). For example, *A. fulva* had a mean highest nest density of eight nests per square metre and *C. hederae* 49 nests per square metre, and so measuring the number of nests in an aggregation on a continuous scale would likely have improved the resolution.

#### 2.4.6 Participant recruitment

There have been inherent and long standing difficulties with the communication of science in mainstream media (Bell, 1994; Barron and Brown, 2012; Mehr, 2015) and the drop in accurate records following the national media attention of this project demonstrates some of the issues that can arise. However, the project's appearance in the media did, overall, result in a higher number of accurate records being submitted in absolute terms. The impact of media coverage can possibly be tempered by careful planning of the project, for example by simplifying the actions and by having clear, concise and readily available instructions. Social media has been acknowledged as an important tool for citizen science endeavours for engaging participants and creating a sense of community around a project (Stafford *et al*, 2010; Dickinson *et al*, 2012). I found that exploiting social media platforms as forums in which potential participants could ask questions and clarify instructions with researchers directly was of great help, although this may not be sustainable for large schemes. Whether or not records are accurate, greater engagement represents more people learning about, and becoming aware of, solitary bees, which is in itself an important function of citizen science.

## 2.5 Conclusion

This study represents the first instance of using citizen science as a tool to examine solitary bee nesting ecology and illustrated the efficacy of a citizen science approach in this context. On the whole, this study demonstrated the capacity of the four target species to tolerate a broad range of environmental variables, although questions remain around whether these conditions impact the nesting density of bees. Srba and Heneberg (2012) in their study of digger wasp nesting found that some variables may be used to identify usable nesting areas and that others are important for determining nest density at a suitable site and the results from this study may be a reflection of this process. Most *C. hederæ* nest aggregations occurred at sloped sites in full sun, whereas the majority of *A. fulva* sites were found in flat, shaded areas, although further experimental study would help to reinforce these findings. The distance to forage and phylogenetic relatedness may play a role in determining the nest site requirements of a ground nesting species, but more empirical research is required. Despite not being able to discriminate the effects of the measured environmental variables on aggregation size, I believe that by calculating the total number of nests or nest density on a continuous scale the data resolution required for this analysis could be achieved. The location, size and nesting density of solitary bee aggregations are likely determined by complex interactions between the abiotic environment, foraging resources, phylogeny, parasite load and natal nest site fidelity and more empirical studies are needed to elucidate these forces. A better understanding of solitary bee nesting ecology will assist us in developing robust conservation practices and policy going forward as, in order to protect anything, we must first understand what needs protecting.

### 3. Analysis of the spatial differentiation in fossorial bee nesting densities

#### 3.1 Introduction

This chapter uses the spatial differences both among and within nesting aggregations as a mechanism to elucidate physical characteristics that contribute to an optimal nesting environment. The approach of this study follows Potts and Willmer (1997), where higher nesting density is used as an indicator of a higher quality nesting site. In terms of nest density, nesting aggregations can differ on two levels: first, at the level of the nest site, there can be differences in maximum nest density among aggregations; secondly, within aggregations, patches of high density nesting and low density nesting occur.

##### *3.1.1 Why do nesting aggregations form and why is this important for conservation?*

Within a nesting aggregation there is no cooperation between individuals and each female excavates her nest and provisions her eggs solitarily (Linsley, 1958), although, there are some species of solitary bee that do demonstrate loose eusocial behaviour (e.g. *H. rubicundus*) (Eickwort *et al*, 1996).

Fundamentally, in order for nesting aggregations to form and be maintained, the benefits associated with aggregate nesting must outweigh the significant costs of increased intraspecific competition, increased risks of predation and parasitism and enhanced pathogen transmission (Rosenheim, 1990). Holistically speaking, there is no definitive explanation for why bees nest in aggregations and how they overcome these disadvantages, but there are a number of theories that have been investigated to varying degrees.

Natal philopatry (the tendency of an animal to habitually return to its natal site) is a significant force acting on the nesting behaviour and decision making of solitary bees (Steffan-Dewenter and Schiele, 2004; Yanega, 1990). However, in order for this strategy to develop there must first be significant costs and/or benefits associated with nesting in a particular location. Therefore natal nest site fidelity cannot drive the formation of aggregations but simply helps to maintain them over generations.

Solitary bee nesting aggregations can be heavily parasitized and many species are specific hosts to cleptoparasitic species (Linsley, 1958). These interactions

influence the behaviour and life histories of the host species (Lienhard *et al*, 2009) and since a significant level of parasitism occurs at the nest, parasitism has the potential to impact the nesting strategies of solitary bees. The evidence to support the hypothesis that parasitism drives the formation of aggregations is mixed. Steffan-Dewenter and Schiele (2008), whom carried out experiments with the aerial nesting solitary species *Osmia bicornis* (Linnaeus, 1758), found that population growth was not impacted by natural enemies and concluded that it was most likely driven by nest site availability. Furthermore, parasites that end up in solitary bee nests do not always get there directly. The meloid beetle *Tricrania stansburyi* (Haldeman) is a cleptoparasite of the gregarious, aerial nesting bee *Osmia lignaria* (Say) (Torchio and Bosch, 1992) that is transferred to its hosts' nests by phoresis. However, a significant amount of nest parasitism does occur at the nest site (Potts, 1995; Wcislo and Cane, 1996) and so may play a role in the spatial configuration of aggregations.

One of the most popular theories in this context is that nesting aggregations form as selfish herds (Hamilton, 1971). In this way, nests at the centre of the aggregation would experience less parasitism than those on the periphery, conferring enough benefit to successful individuals to drive the formation of aggregations. Studies, which have found that nest parasitism decreases as nesting density increases within an aggregation, have been used as evidence to support this theory (see Wcislo, 1984) who studied the digger wasp *Crabro cribrellifer* and Larsson (1986) who studied the digger wasp *Bembix rostrata* and its nest parasite *Metopia leucocephala*. Although these results have been interpreted to show that aggregations as a whole behave as selfish herds, in the strictest sense, they do not. Hamilton (1971) in his definitive paper of a selfish herd pointed out that a nest site in the centre of a colony does not behave in the same manner as an individual at the centre of a herd because it is not mobile and, more importantly, there is an assumption that the parasitic animal is likely to approach from the periphery of the colony. Therefore, in order for the aggregation to act as a selfish herd the parasites must enlist a peripheral approach wherein the nests on the outside of the aggregation are parasitized first and most heavily, but there is no evidence for this.

Rosenheim (1990) pointed out that "nest density, rather than geometric position relative to the centre of the aggregation appears to be the critical factor." Aggregations are not regularly spaced and do not follow a pattern of increasing nest density towards the geometric centre. Instead aggregations clump into



areas of high and low density, on a continual gradient, and so while individual patches may be considered to be proximate selfish herds, the aggregation as a whole, is not. In terms of the spatial distribution of nests within aggregations, parasite pressure may be an important factor, as the density dependence of parasite load demonstrates (Wcislo, 1984; Larsson, 1986). This relationship may be the demonstration of a Turing pattern in nature, whereby the bee nests are acting as the activators and the parasites the inhibitors (Turing, 1952). As the activators, the bee nests stimulate production of more bee nests and more parasites and the parasites, acting as the inhibitors, slow down the production of more bee nests and more parasites. As parasitic species require multiple nests to parasitise, their range expands faster than the range of bee nests, resulting in high density patches of nests with lower parasite load than the low density areas surrounding them. But this is just a theory and requires empirical investigation. The literature demonstrates that parasitism does impact the spatial structures of nesting aggregations, certainly at the within-aggregation scale (Wcislo, 1984; Larsson, 1986) although more research is required to elucidate the impacts of parasitism on the formation of aggregations as a whole. Closely related aggregate and individual nesting species offer a potential opportunity to investigate these dynamics further and fully elucidate how these forces work in tandem.

Rosenheim (1990) proposed that the information centre hypothesis might have importance for aggregate nesting solitary bees. This mechanism was first described for cliff swallows where individuals that have an unsuccessful foraging trip return to the colony, identify a successful individual and follow them to the food resource (Brown, 1986). This strategy has not been specifically investigated for solitary bees and so it cannot be substantiated (Wcislo and Cane, 1996). Theoretically, this mechanism could confer significant benefit to individuals in an aggregation who must quickly locate appropriate forage in their proximity after emergence. Local enhancement (the attraction of a foraging animal to other feeding conspecifics) may also contribute to foraging efficiency for solitary bees, as has been shown for some eusocial (Slaa *et al*, 2003) and social (Avarguès-Weber and Chittka, 2014) species. However this strategy operates at forage and so individuals would not need to nest gregariously in order to exploit this mechanism.

The opportunity to reuse old nests may encourage aggregate nesting (Myers and Loveless, 1976) as, theoretically, this could confer significant

benefit. In this scenario, females would not have to expend significant and valuable energy on finding and excavating a nest and instead focus their efforts on reproducing and providing food for their developing broods. There is evidence that this is an important mechanism in the nest site dynamics of the digger wasp *Cerceris arenaria* (Linnaeus, 1758) (Polidori *et al*, 2006) but, to date, no studies have explicitly validated this strategy for soil-nesting bees. Eickwort *et al* (1977) proposed that aggregations might form as a result of individual females using the presence of conspecifics at a site as an indicator of high quality nesting habitat (Rudolf and Rödel, 2005). However, this hypothesis relies on two further assumptions. First, the assumption that not all habitats are created equal and, that different patches within a landscape represent varying quality in terms of nest site suitability. Second, there must be a considerable cost associated with finding a suitable nest site independently if the costs of aggregate nesting (intraspecific competition and increased predation being the two most significant) are to be outweighed. Both of these assumptions rely on the fact that high quality nesting habitat is rare, suggesting that specific physical properties are required of a potential nest site. These fundamental assumptions form what is referred to as the 'limited substrate hypothesis'.

It is unlikely that all habitats have the same capacity to support populations of nesting bees (Grundel *et al*, 2010) and the 'limited substrate hypothesis' postulates that there are restricted areas within a landscape with the necessary physical characteristics to be suitable nest sites for bees. As I have established in Chapter 2, the tendency of fossorial bees to nest in a location with particular physical or edaphic attributes has been well documented and it has been determined that these factors do influence the nesting behaviour of bees. For example, Potts and Willmer (1997) found that the patterns of nest site selection of *H. rubicundus* supported the 'limited substrate hypothesis' as females chose to nest in areas that conferred thermal stability to the nest by establishing themselves in sloped, south facing ground. The hypothesis was reinforced by the observation that within aggregations females nested in areas with softer soils that required less energy to excavate and avoided areas with harder substrates. Despite this evidence, there remain significant knowledge gaps around the extent of the validity of the 'limited substrate hypothesis' and how it relates to the size and density of nesting aggregations. There is also little clarity around whether there are species-specific requirements of the physical attributes of

a nest site or whether the important features are common across all fossorial species. This is an important aspect of nesting biology to understand as the physical landscapes both in the UK and around the world have gone through monumental change over the last one hundred years, with the proliferation of intensive agricultural practices and the expansion of urban developments. If we can establish the importance of the abiotic environment and understand how to provide nesting habitat for soil-nesting species then, theoretically, we can help to protect and provide additional opportunities for these populations in changing environments. Examining spatial differences both within and among aggregations of individual species and comparing them to other species can help to answer these questions.

The principal focus of this chapter is to understand what constitutes suitable nesting habitat for fossorial solitary bees so that these areas may be protected and, if necessary, provided. In terms of the spatial distribution of the nests within the aggregations, these patterns may help identify preferences for certain environmental characteristics. These spatial dynamics are highly likely to be impacted by other forces, including nest site parasitism. Although these mechanisms may also be important for spatial patterning, the scope of this study is to analyse spatial patterning only in terms of differences in physical attributes.

### *3.1.2 Methodological considerations*

This study follows Michener's (1974) definition of a nesting aggregation as grouped nests in a restricted area, where each female makes her own nest. I define an active nest aggregation as one where females are present and exhibiting nest building behaviour or nest provisioning behaviour. This is an important distinction as artefacts of nests or emergence holes are often seen in the field but do not correspond with current conditions nor can they be explicitly linked to a species. As I have established in Chapters 1 and 2, there is a limited number of empirical studies of fossorial solitary bee nesting ecology. One consequence of this is a current lack of standardised methods for surveying solitary bee nesting aggregations. The traditional methods used to measure bee species richness and abundance at a site (i.e. pan traps and sweep netting on forage) are not appropriate in this setting as they fail to link the specimens with their nest sites. Therefore, the survey methods for this study were developed using the work of Potts and Willmer (1997) and Potts *et al*, (2005) as guidance.

Locating a sufficient number of nesting aggregations in the field for *in situ* study of their ecology presents a significant logistical barrier and nesting studies are historically poor at reporting how study nests were located. Some completely omit this information (Cane, 1991; Potts and Willmer, 1997), while others seem to rely heavily on local knowledge of the study site (Potts *et al*, 2003 and 2005), an approach, which cannot be repeated at other locations. Recently, the use of emergence traps has been enlisted in an attempt to standardise survey methods (Sardinas and Kremen, 2014; Sardinas *et al*, 2016). Although this method can be effective in terms of estimating the nesting species richness at a site (Sardinas and Kremen, 2014) it would probably not suffice as a method for examining nest ecology or estimating abundance as nests are commonly aggregated. That said, this approach might be useful for identifying and quantifying the ecology of inconspicuous nests (such as those constructed by *Lasioglossum* species), which are difficult to spot by eye. I circumvented this challenge by enlisting the help of the public under the umbrella of my citizen science project, The Solitary Bee Project (Chapter 2).

### 3.1.3 Research questions and hypotheses

This chapter set out to address three **research questions**:

1. Do differences in the environmental characteristics of a site influence the nesting density of *A. fulva*, *A. cineraria* and *C. hederæ* among aggregations?
2. Do differences in environmental characteristics within a nesting aggregation contribute to the clumped spatial distribution of aggregations for these species?
3. Are there interspecific differences in the environmental characteristics of where nest sites occur?

### Hypotheses

1. The environmental characteristics of the nesting sites will influence nesting density. Factors that will lead to increased nesting density will be: availability of bare ground, sloped terrain, sandy soil and open sites. Lower density aggregations will occur in areas that are shaded, flat, have high clay content in the soil and and experience little direct sunlight.
2. Environmental characteristics will not impact spatial distribution of nests within the aggregation. There will be no differences in the physical characteristics of high and low density nest patches within the aggregations for any species.

3. Interspecific differences will exist between *C. hederæ* and the two *Andrena* species. No interspecific differences will be identified between the *Andrena* species. *Colletes hederæ* will occur more frequently on sloped ground compared to the two *Andrena* species, which will most often be found on flat terrain.

## 3.2 Methods

### 3.2.1 *Study species' and field sites*

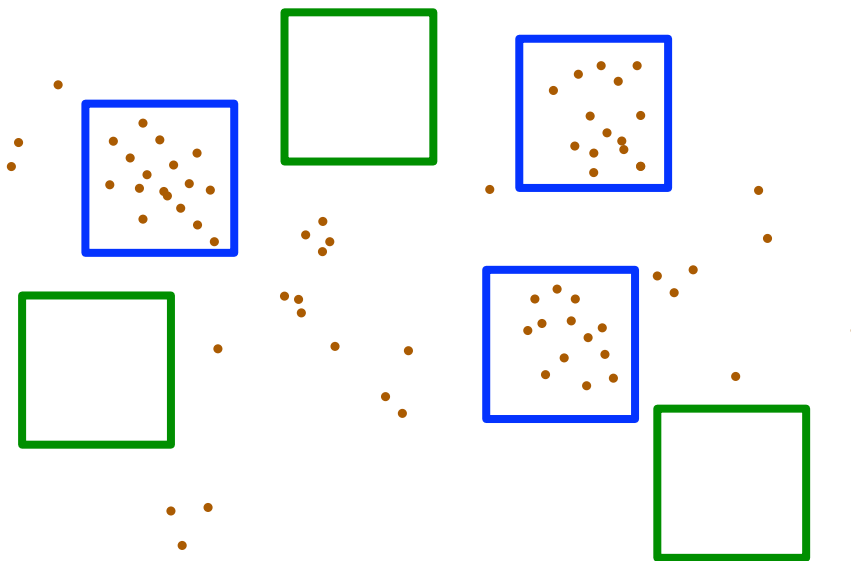
The species chosen for this study were based on those in the citizen science project. A detailed explanation of how and why these focal species were chosen can be found in the methods section of chapter two. The Solitary Bee Project asked members of the public to submit sightings of the nesting aggregations of four solitary bee species: *A. fulva*, *A. cineraria*, *H. rubicundus* and *C. hederæ*. This study used those records to identify and sample active nesting aggregations (Table 3.1).

**Table 3.1:** Summary of field sites that were used to study nesting aggregations

Site	Site name	Coordinates	Date sampled	Quadrats	Species
1	Cambridge Uni Botanic Gardens	TL 45495719	30/03/17	6	<i>Andrena fulva</i>
2	Jesus College, Cambridge University	TL 45425893	30/03/17	6	<i>Andrena fulva</i>
3	Mill Road Cemetery, Cambridge	TL 462582	03/04/17	6	<i>Andrena fulva</i>
4	Natural History Museum, London	TQ 26627898	08/04/17	6	<i>Andrena fulva</i>
5	Wandlebury Country Park, Cambridge	TL 498532	12/04/17	6	<i>Andrena fulva</i>
6	Holland Park, London	TQ 247797	08/04/17	6	<i>A. fulva/ A. cineraria</i>
7	Barnack Hills and Holes NNR	TF 077044	15/04/17	6	<i>Andrena cineraria</i>
8	Tyntesfield House, Bristol	ST 5012871678	16/04/17	6	<i>Andrena cineraria</i>
9	Tyntesfield House, Bristol	ST 5068971192	16/04/17	4	<i>Andrena cineraria</i>
10	Nailsea Tesco, Bristol	ST 4789770821	16/04/17	2	<i>Andrena cineraria</i>
11	Paxton Pits NR, St. Neots	TL 192623	04/05/17	6	<i>Andrena cineraria</i>
12	Pitsford Reservoir, Northamptonshire	SP 774711	11/05/17	6	<i>Andrena cineraria</i>
13	Calke Abbey, Derby	SK 371230	24/05/17	6	<i>Andrena cineraria</i>
14	Dartmouth Park, West Bromwich	SP 013915	24/05/17	6	<i>Andrena cineraria</i>
15	Cambridge Uni Botanic Gardens	TL 453571	18/09/17	6	<i>Colletes hederæ</i>
16	Wandlebury Country Park, Cambridge	TL 496533	19/09/17	6	<i>Colletes hederæ</i>
17	Cranwich Heath, Norfolk	TL 775941	21/09/17	6	<i>Colletes hederæ</i>
18	Beeston Common, Norfolk	TG 163419	28/09/17	6	<i>Colletes hederæ</i>
19	Ladybird Nurseries, Suffolk	TM 387588	29/09/17	3	<i>Colletes hederæ</i>
20	Argal Reservoir, Cornwall	SW 761328	11/10/17	6	<i>Colletes hederæ</i>
21	Loe Bar, Cornwall	SW 641242	11/10/17	6	<i>Colletes hederæ</i>
22	Little Dartmouth, Devon	SX 883496	12/10/17	6	<i>Colletes hederæ</i>
23	Red Lodge Heath, Suffolk	TL 695700	26/10/17	6	<i>Colletes hederæ</i>
24	Lackford Lakes, Suffolk	TL 801706	07/11/17	6	<i>Colletes hederæ</i>

### 3.2.2 Sampling the nesting aggregation

This study aimed to evaluate both ‘among aggregation variation’ and ‘within aggregation variation’ in nesting density. Among aggregation variation refers to the relationship between the environmental characteristics of the aggregations and the number of nests in the highest nest density patches. Within aggregation variation is the differences in environmental factors between patches of highest density nesting and lowest density nesting within aggregations. Each aggregation was first examined to ensure that it was an active site (females present and exhibiting either nest building behaviour or nest provisioning behaviour) and to identify the nesting species. The entire aggregation was surveyed and the three patches with the highest nesting densities were identified subjectively, by eye. A 1m<sup>2</sup> quadrat was laid down in every high density patch (Fig. 3.1) and a number of measurements were taken (explained below). The quadrat was then moved to the nearest patch of ground which had no nests (or the lowest density patches where this was not possible) and the measurements were repeated. Low density quadrats were used to evaluate within aggregation variation. Three pairs of quadrats were used in all aggregations except for sites nine and ten, which had to be sampled under time pressure, and site nineteen. The aggregation at site nineteen occurred in a small earth bank against a large greenhouse and all areas of the bank were occupied by nests, therefore only high density quadrats were sampled at this site.



**Figure 3.1:** Illustrative example of placement of the three pairs of quadrats at each aggregation. Brown dots indicate nests. Blue squares indicate high density quadrats and green squares indicate low density quadrats.



### 3.2.3 Quadrat measurements

Within each quadrat, the following variables were measured:

1. Number of nests.
2. Soil hardness - measured at the centre of the quadrat using a hand-held penetrometer (ELE International, UK).
3. Slope and aspect - measured at the centre of the quadrat using a clinometer.
4. Ground cover; the amount and type of vegetation in the quadrat- estimated as a percentage.
5. Shade; an estimation of the level of shade that part of the aggregation experienced throughout the day, based on surrounding structures.

A standard soil corer (LaMotte handheld soil sampler; 25cm length, 2.5cm core diameter) was used to obtain a soil sample (approx. 100g) from the centre of the quadrats to 12 cm depth. This depth roughly correlates to the depth at which brood cells are situated. The samples were sealed in plastic bags and refrigerated at 4°C to maintain soil moisture until they were analysed. These samples were used to analyse soil composition. Soil moisture content was established via laboratory analysis. This was attempted in the field using a soil moisture probe but I reasoned that taking a soil sample would be more informative as probes only measure moisture levels at approximately three centimetres below the surface. Solitary bee brood cells are deeper in the soil and so a soil sample measured at 12cm would be more likely to represent moisture level at the critical depth. When nest searching solitary bee females will perform test digs at the soil surface before choosing a nest site. However, the key factors for brood survival occur at brood cell depth. Following standard laboratory procedures for establishing soil moisture content, the samples were weighed, dried in an oven at 105°C for 24h, and then reweighed (Standards Association of Australia, 1990). Using a pestle and mortar, the sample was ground for 45 seconds to break down any soil aggregates and then passed through a 2mm aperture sieve for two minutes to separate the gravel fraction, which was weighed. Thirty-five grams of the remaining non-gravel fraction of each sample was placed in a furnace at 550°C for 3h, cooled and re-weighed for loss on ignition analysis. This sample mass, furnace temperature and duration were chosen in accordance with the guidelines laid out in Hoogsteen *et al*, (2015). The mass loss indicated the mass of the organic matter present in the sample. There are two principal methods for determining soil texture, the hydrometer method and the sieving method. The hydrometer method is based on Stokes law which states that particles fall out of suspension at different rates,

based on their size. For this method, soil particles are dispersed and then agitated. After dispersion, the proportion of each particle group (sand, silt, clay) is determined using a hydrometer, which measures the amount of particles in suspension. The sieving method is similarly based on particle size but instead a de-aggregated soil sample is passed through sieves of varying sizes to determine the amount of each particle fraction in the sample. The sample is sieved for two minutes through 250µm and 63µm aperture sieves to derive the sand, silt and clay component fractions. Both of these methods were trialled for this analysis but the sieving method was pursued as there was precedent for using it in the context of soil analysis of solitary bee nests (Potts and Willmer, 1997). Particulate fractions and size classes used were in accordance with British Standard System (BSS): gravel (> 2mm in diameter); sand (250µm to 2mm); silt (63–250µm); and clay (< 63µm).

#### 3.2.4 Spatial analysis of sites

Principal Components Analysis is often used to summarise and visualise environmental data (House and Spellerberg, 1983). It is an ordination method and transforms data into fewer dimensions thereby simplifying the complexity in high-dimensional data (Lever and Krzywinski, 2017), which often occurs in environmental studies. Ordination is the term used for techniques wherein a multi-dimensional dataset is adapted, such that when it is projected onto two dimensional space, any inherent patterns in the data become apparent visually (Pielou, 1984). In this study, there are high dimensional data as every nesting aggregation was measured on the basis of a large range of environmental features. PCA was thus used to visualise the environmental variation between sites for all species on the basis of a smaller number of derived variables or principal components. This analysis was performed using the R package 'vegan' (Oksanen *et al*, 2009) using standardised, centred data. The mean values of the environmental characteristics for the high density quadrats from each site was used in this analysis. The specific variables used in the PCA were:

- Slope of the ground
- Soil penetration
- Percentage sand present in soil
- Percentage silt present in soil
- Percentage clay present in soil
- Percentage gravel present in soil
- Percentage organic matter content of the soil
- Soil moisture content
- Proportion of the nest site covered in grass

- Proportion of the nest site that was bare ground
- The approximate amount of shade experienced by the nest site

The slope variable was log transformed so that the PCA was not dominated by its skewedness. Nest sites were also investigated according to their soil texture and visualised on the soil texture triangle. The soil texture triangle was developed by Davis and Bennett (1927) and is a graphical representation of the twelve soil types. It can be used to determine soil type based on the proportions of sand, silt and clay particles in the sample.

### *3.2.5 Interspecific variation in nest density*

The data were investigated to identify any interspecific differences in nesting density. The mean nest density value for all high density quadrats for each species was used in this analysis. This analysis followed the procedures used in Srba and Henberg (2012), where differences in the nest site characteristics of multiple Sphecidae species were investigated. The data were analysed using a one-way ANOVA. Tukey's test was used for post hoc analysis and eta squared to identify effect size. The density data were not normally distributed and so were transformed using a log transformation to conform to the assumptions of normality.

### *3.2.6 Among aggregation variation in nest density for each species*

Due to the low sample sizes for each species relative to the number of predictor variables in this dataset, logistic regression approaches were not appropriate. Therefore, multivariate methods were again explored as an alternative analytical approach. The linearity of the environmental data for each species was investigated using detrended correspondence analysis where a gradient of less than two indicated a linear relationship (Šmilauer and Lepš, 2014). Linear relationships were established for all variables (all gradients < 2) and so the Redundancy Analysis (RDA) method was employed. RDA is a constrained ordination method wherein the capacity of the constrained variable(s) to explain the variation in the response variables is examined. In this study, the analysis was performed in 'reverse' fashion, where the variation in nesting density was used as the predictor, or constrained variable, to determine the amount of variation it explained in the environmental characteristics. In this way, insights could be gained into the relationships between nest density and environmental characteristics, but the issues with small sample sizes and large groups of predictor variables were overcome. Each species was analysed individually due

to the interspecific differences in nesting density. For this analysis, the values from the high nest density quadrats were used, where the values for each site were determined by taking a mean of the values measured in the three quadrats at that site. Post RDA, the marginal testing method was used to test for significance as per Legendre *et al*, (2011). Marginal tests are ANOVA-like permutation tests for the effects of constraints, consisting of 999 permutations. RDA analyses were performed using the R package 'vegan' (Oksanen *et al*, 2009), marginal tests were performed in 'sdat' (Zhang and Laber, 2015).

### 3.2.7 Within aggregation variation for all species

Variation in nesting density within aggregations was analysed using the data from all six quadrats measured at each site. For each site the measurements for the environmental variables (shade, slope, soil texture, soil gravel content, soil moisture, soil penetration, proportion of bare ground, proportion of grass, height of vegetation) from the three high density quadrats were averaged to obtain values for high density nest patches. The same procedure was applied to the three low density quadrats. The response variable was binary and coded as either 'high' or 'low'. In the first instance, a generalized linear mixed model with site as a random effect and all other variables as fixed effects was run. Site was found to have no significant effect and so was removed from the model, according to model simplification methods (Crawley, 2013). The data were subsequently modelled using a generalized linear model with binomial error distribution as the outcome variable had two levels: high density and low density. Model fit was evaluated using Akaike Information Criterion (AIC) values. Comparison of AIC is a useful method for distinguishing between two competing models, where a lower AIC value is indicative of superior goodness of fit (Murtaugh, 2014). Selection of variables for the final model was done using a backward selection procedure. The final model was evaluated using the following parameters: sensitivity (proportion of observed positive outcomes (high density) that were predicted to be positive), specificity (proportion of observed negative outcomes (low density) that were predicted to be negative), overall classification and goodness of fit. Sensitivity, specificity and overall classification were calculated using a threshold cut-off value of 0.5. This value is the probability that a prediction is true and is a trade-off between the false positives and false negatives. 0.5 was chosen as it represented the best balance between specificity, sensitivity and overall classification (Table 3.2). This method of threshold selection is known as Max SSS and has been shown to be the most robust method of threshold selection for presence-only data (Liu *et al*, 2013).

**Table 3.2:** Changes in specificity, sensitivity and overall classification across the full spectrum of cut-off threshold values

<b>Cut-off threshold</b>	<b>Specificity</b>	<b>Sensitivity</b>	<b>Overall</b>
0.9	100	0	48
0.8	100	12	54
0.7	87	36	60
0.6	78	52	65
0.5	78	64	71
0.4	52	68	60
0.3	0	100	48
0.2	0	100	48
0.1	0	100	48

### 3.2.8 Interspecific variation in nest site characteristics

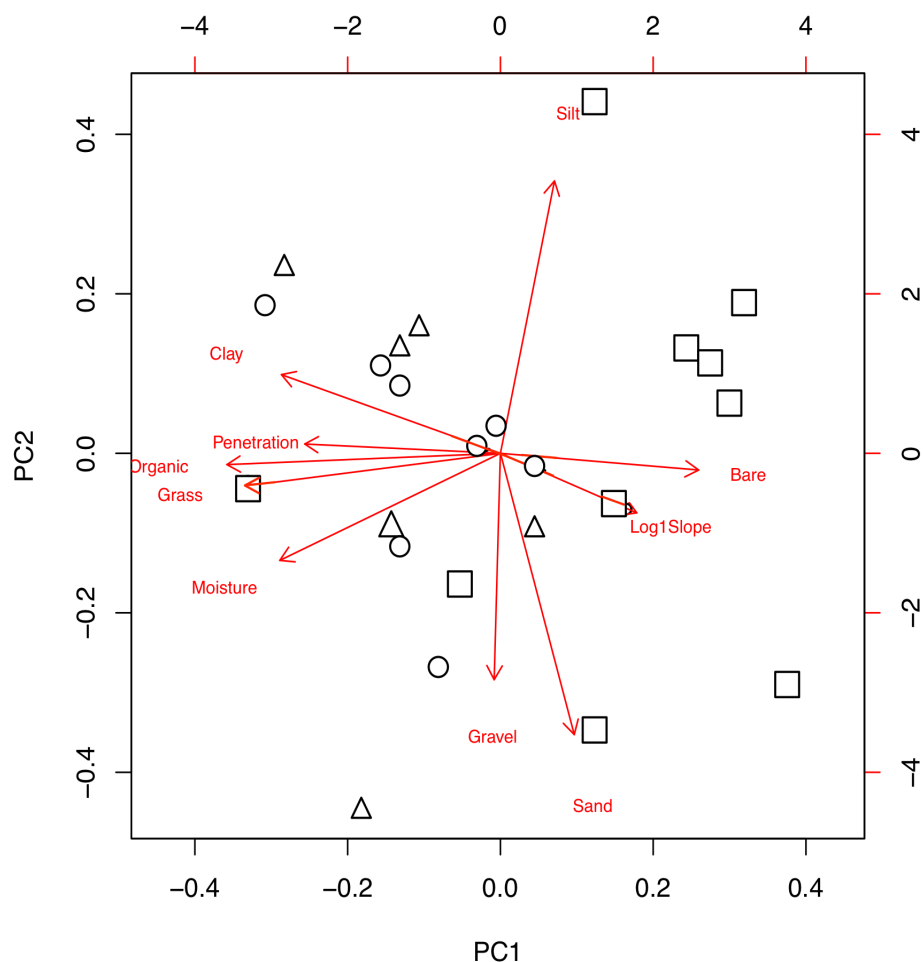
The data were investigated to identify whether nesting characteristics varied between species and were analysed using the procedures from Srba and Heneberg (2012) (as with differences in nest density). Shapiro tests were used to check the data for normality and Bartlett's test for equal variances. Variables which could not be transformed to exhibit a normal distribution and equal variances (silt and shade) were analysed using kruskal-wallis tests and Dunn's test for post-hoc analysis. Epsilon squares were used to calculate effect sizes (Tomczak & Tomczak, 2014). Normally distributed variables with equal variances were analysed using one-way ANOVAs with Tukey's test for post hoc analysis. The variable 'slope' was log transformed to meet these assumptions. Eta squared was subsequently used to measure effect size. Data were not collected for vegetation height at *A. fulva* nest sites as this was added to the protocol after these sites had been surveyed. Therefore, difference between *A. cineraria* and *C. hederæ* was investigated using Mann-Whitney U test as the data were not normally distributed.

All analyses were performed using R version 3.5.1 (R core team, 2015).

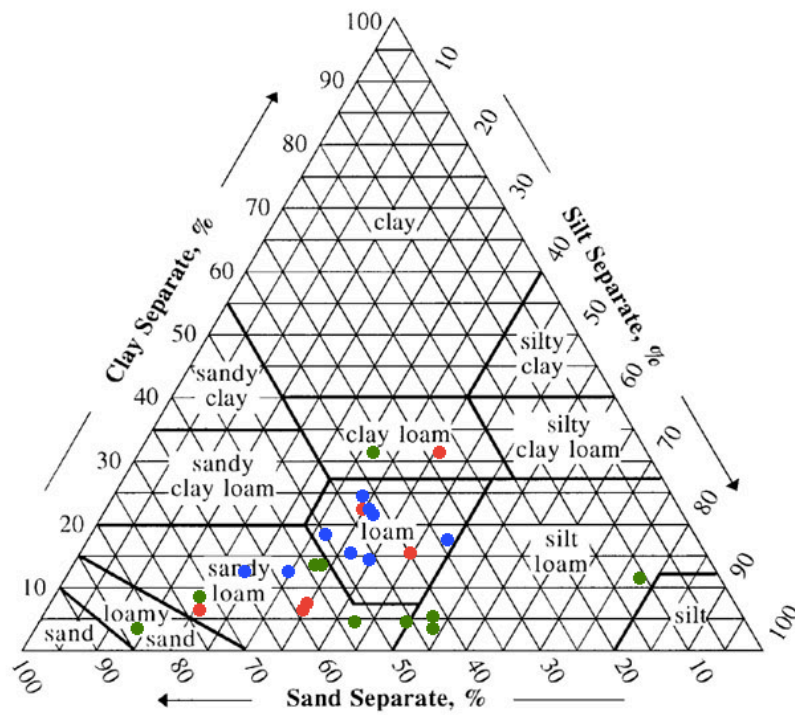
### 3.3 Results

#### 3.3.1 Spatial analysis of sites.

In total, 132 quadrats were measured as part of this study, 69 from high density nesting areas and 66 from low density nesting areas. The PCA analysis considered the data from the high density nesting areas and found that the first two components explained 61% of the environmental variation between field sites (eigenvalues: axis 1 = 0.38, axis 2 = 0.23). The analysis showed that most *C.hederae* nesting sites were similar to each other in terms of environmental characteristics and distinct from the *Andrena* nest sites (Fig. 3.2). In terms of soil analysis, the majority of sites in this study fell into the 'loam' or 'sandy loam' categories of soil texture (Fig. 3.3).



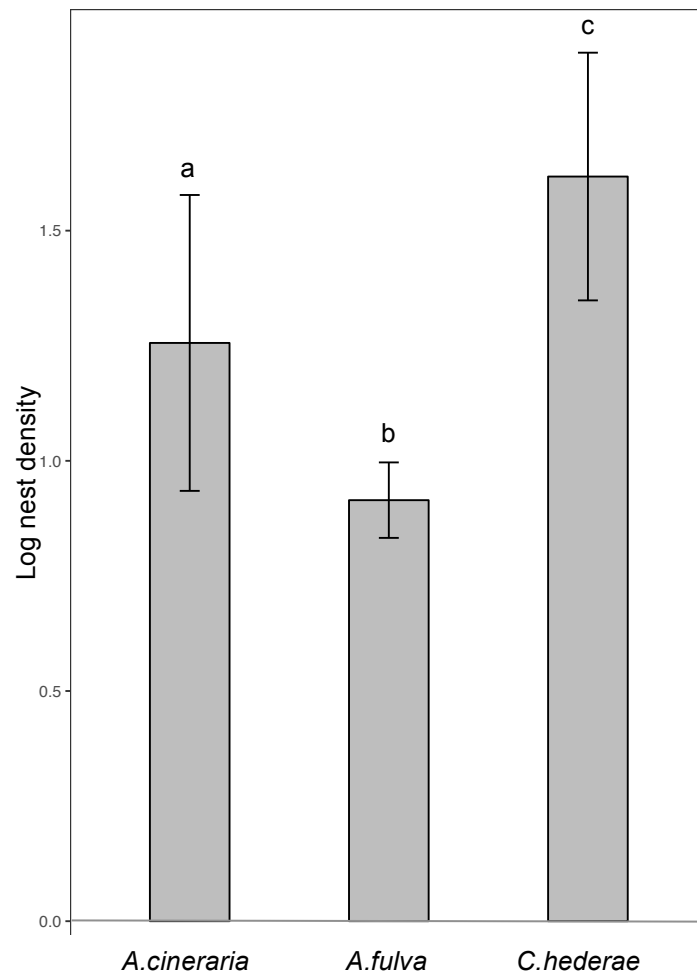
**Figure 3.2:** Biplot of principal components analysis for all sites and environmental characteristics showing the relationships between all variables. Positively correlated variables are grouped together, whereas negatively correlated variables are on opposite sides of the plot origin. The length of the arrow indicates the strength of the correlation between the variable and the principal component. Shapes indicate the species present at the site (*A. fulva*: triangle; *A. cineraria*: circle; *C. hederae*: square).



**Figure 3.3:** Soil texture triangle (Davis and Bennett, 1927) showing the position of the soil type of every sample site. *A. fulva* sites: red (n=6); *A. cineraria* sites: blue (n=9); *C. hederæ* sites: green (n=10).

### 3.3.2 Interspecific variation in nest density

Nesting density differed among all three species (ANOVA:  $F_{2,22} = 13.89$ ,  $p = 0.00013$ ; Tukey:  $p < 0.05$ ), with highest densities occurring in *C. hederæ* (Fig. 3.4). The effect size of this difference was medium (eta squared = 0.56).



**Figure 3.4:** Barplot showing difference in mean log nest density between species with standard deviation. Letters indicate post hoc groupings and show that all three groups differ from each other ( $p < 0.05$ ).

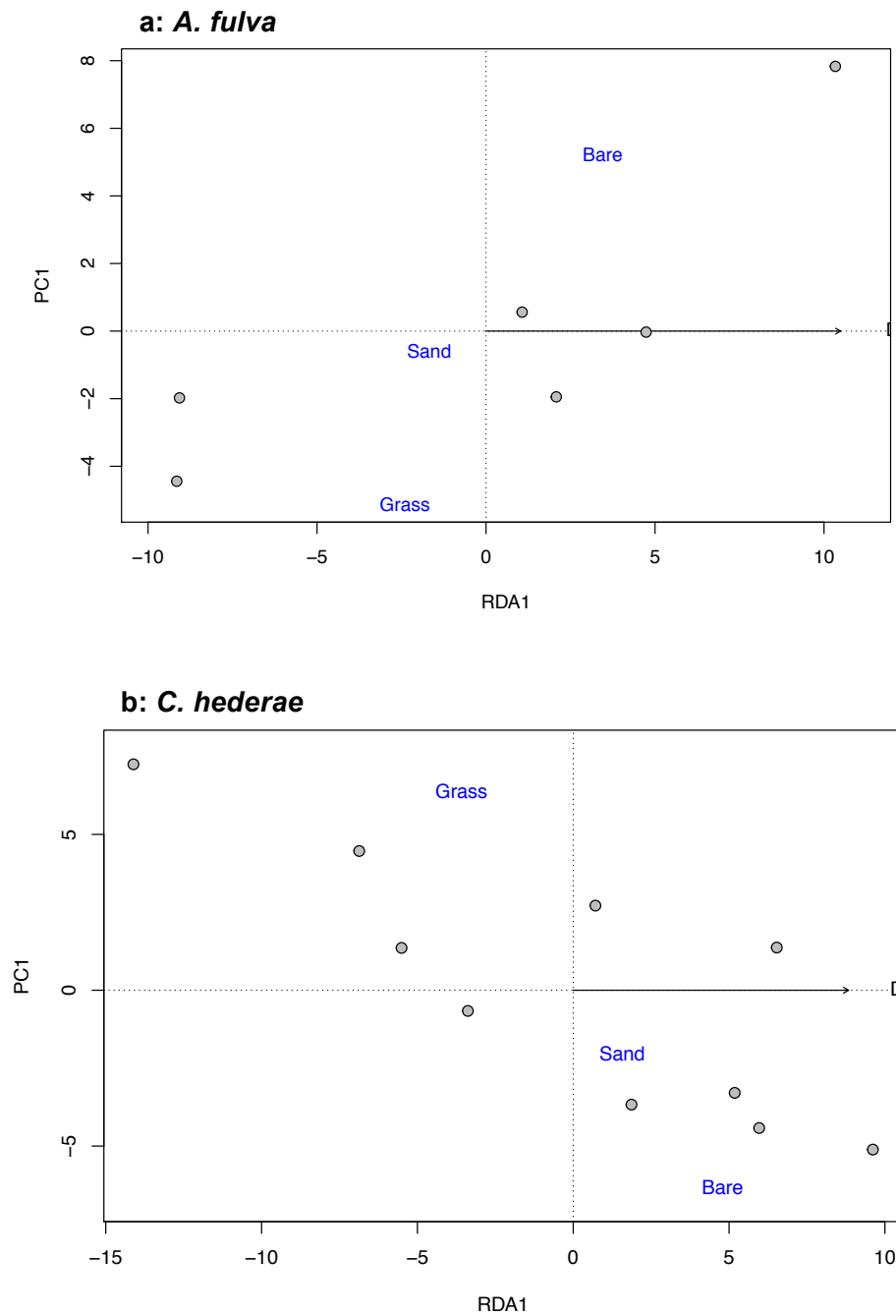


### 3.3.3 Redundancy analyses for each species with nesting density constrained

In the case of *A. fulva*, nesting density explained 23% of the variation in environmental characteristics, compared to just 4% for *A. cineraria* (Fig. 3.5). Twenty five percent of the variation in environmental characteristics was explained by nesting density for *C. hederæ*. The variation in environmental characteristics was not significantly related to nesting density for any of the species (Fig 3.5; *A. fulva* - marginal test:  $F_{1,4} = 1.19$ ,  $p = 0.32$ ; *A. cineraria* – marginal test:  $F_{1,6} = 0.24$ ,  $p = 0.8$ ; *C. hederæ* - marginal test:  $F_{1,8} = 2.65$ ,  $p = 0.09$ ). For both *C. hederæ* and *A. fulva* the variables that were best explained by nest density were the proportion of bare ground, grass and sand, as these variables were most highly loaded on the first axis of the RDA (Table 3.3). For both species, nest density was positively associated with bare ground and negatively associated with grass. For *C. hederæ* sand was positively associated with nest density but the opposite relationship was identified for *A. fulva*.

**Table 3.3:** Summary of the loading values for the three most important variables on the first axis of the RDAs for both *A. fulva* and *C. hederæ*.

Variable	<i>A. fulva</i> loadings	<i>C. hederæ</i> loadings
Bare ground	4.8	3.46
Proportion of sand	1.57	-1.68
Proportion of grass	-3.59	-2.4



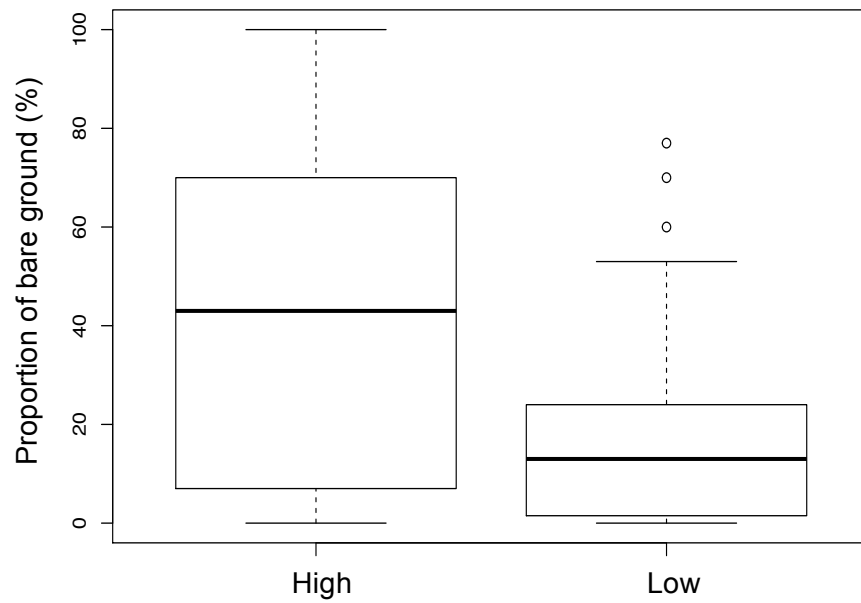
**Figure 3.5:** Redundancy analyses for *A. fulva* (a) and *C. hederæ* (b) nest sites with nesting density constrained. The variables percentage sand, percentage grass and percentage bare ground were best explained by variation in nest density. Grey circles represent sites and nesting density increases with the black arrow.

### 3.3.4 Within aggregation variation in nest density

The pattern of both high and low density patches of nests occurring within aggregations was significantly influenced only by the proportion of bare ground (Table 3.4). High density nesting was more likely to occur in areas with more bare earth (Fig. 3.6). The final model parameters were: specificity = 78%; sensitivity = 64%; overall classification = 71%. The model was well fitted (Hosmer-Lemeshow goodness-of-fit test:  $\chi^2_8 = 7.46$ ,  $p = 0.49$ ), but explained just 18% of the variation in nesting density (Nagelkerke  $R^2 = 0.18$ ). This indicates that while the relationship between the proportion of bare ground and high or low density nesting within aggregations is small, it is reliable.

**Table 3.4:** Summary of the binary logistic regression analysis of factors affecting high and low density nesting.

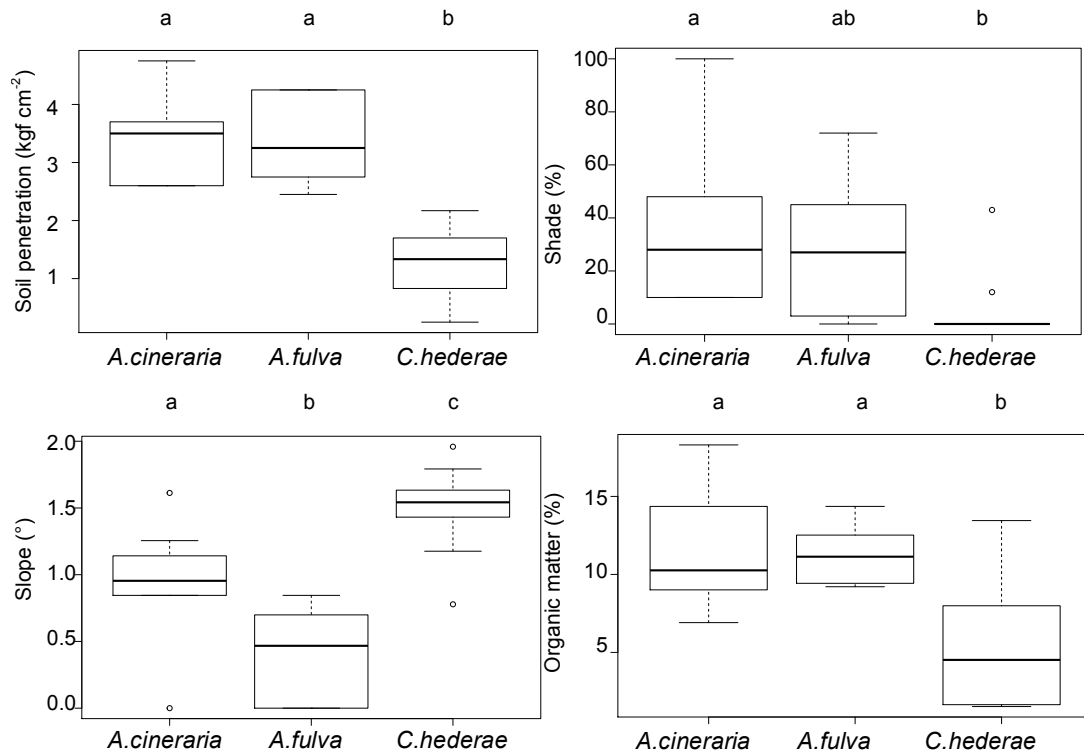
	Estimate	Std. Error	z value	P	95% C.I. for Exp(Est)	
					Lower	Upper
(Intercept)	-0.78	0.46	-1.71	0.09	-1.67	0.11
Bare	0.03	0.01	2.43	0.01	0.01	0.05



**Figure 3.6:** Boxplot showing the proportion of bare ground at high nest density and low nest density patches for all species. Data are given as medians (lines in the boxes), 25th and 75th quartiles percentile (boxes), 10th and 90th percentile ranges (whiskers), and outliers (circles).

### 3.3.5 Interspecific differences

Soil penetration, slope, percentage shade and soil organic matter content differed between the nest sites of the three species (Table 3.5; Fig. 3.7). This effect was largest for soil penetration, with an effect size of 0.71. The only factor for which all three species were different from each other was slope (Tukey HSD:  $p < 0.05$ ; Fig. 3.7). In this case, *A. fulva* was found to nest in flat terrain with a mean slope angle of  $2.6^\circ$ , whereas *A. cineraria* nest sites had a mean slope of  $9.3^\circ$ . *Colletes hederæ* occurred in terrain with the highest mean slope of  $37.2^\circ$ . With regard to soil penetration (Tukey HSD:  $p < 0.05$ ), shade (Dunn:  $p < 0.05$ ) and organic matter content (Tukey HSD:  $p < 0.05$ ) the differences were inter-familial, occurring between *C. hederæ* and the *Andrena* species (Fig. 3.7). The mean values for soil properties (soil penetration and organic matter content) at *C. hederæ* sites, were approximately half that measured at the *Andrena* sites (Table 3.6). For percentage shade, the gap was larger with the *Andrena* nest sites experiencing more than five times the amount of shade as *C. hederæ* nesting aggregations (Table 3.6).



**Figure 3.7:** Boxplots showing the differences in soil penetration, shade, slope and organic matter content between *A.cineraria*, *A.fulva* and *C.hederæ* nest sites. Letters indicate post hoc groupings. Data shown are as explained in Figure 3.6 legend.

**Table 3.5:** Summary of statistical tests investigating interspecific differences in nest site characteristics. Significant results ( $p < 0.05$ ) are highlighted in bold.

	Test	Test statistic	p-value	Effect size
LogSlope (°)	One way ANOVA	<b>F = 15.49</b>	<b><math>6.32 \times 10^{-5}</math></b>	<b>0.58</b>
Shade (%)	Kruskal Wallis	<b><math>\chi^2 = 10.877</math></b>	<b>0.004</b>	<b>0.45</b>
Soil penetration (kgf cm <sup>-2</sup> )	One way ANOVA	<b>F = 26.31</b>	<b><math>1.46 \times 10^{-6}</math></b>	<b>0.71</b>
Soil moisture (%)	One way ANOVA	F = 0.729	0.494	
Gravel (%)	One way ANOVA	F = 0.367	0.697	
Organic matter (%)	One way ANOVA	<b>F = 9.196</b>	<b>0.00125</b>	<b>0.46</b>
Sand (%)	One way ANOVA	F = 0.059	0.943	
Silt (%)	Kruskal Wallis	$\chi^2 = 0.37$	0.83	
Clay (%)	One way ANOVA	F = 2.58	0.1	
Bare (%)	One way ANOVA	F = 1.116	0.345	
Grass (%)	One way ANOVA	F = 2.199	0.135	
Veg height (cm)	Mann-Whitney	W = 30	0.394	

**Table 3.6:** Mean and standard deviation values of environmental characteristics for the three species at sites occupied by high density nesting (n=25).

	<i>A. cineraria</i>	<i>A. fulva</i>	<i>C. hederæ</i>
Sand (%)	49.2 ± 10.7	50.5 ± 15.9	50.4 ± 19.7
Silt (%)	33.1 ± 7.2	34.4 ± 8.7	40.4 ± 19
Clay (%)	17.7 ± 5.8	15.1 ± 9.8	9.42 ± 8.5
Gravel (%)	8.4 ± 8.9	12.7 ± 11.9	12.6 ± 7.1
Organic matter (%)	11.7 ± 3.9	11.4 ± 2.7	5.3 ± 4.2
Soil penetration (kgf cm <sup>-2</sup> )	3.7 ± 1	3.4 ± 1.3	1.3 ± 0.7
Soil moisture (%)	15.5 ± 6.3	16.9 ± 5.4	12.8 ± 8.3
Shade (%)	37.3 ± 28.6	29.7 ± 30.8	5.5 ± 14.3
Slope (°)	9.3 ± 6.2	2.6 ± 4.2	37.2 ± 25.8
Bare (%)	36.8 ± 32.5	38.6 ± 30	54.2 ± 36.1
Grass (%)	43.2 ± 25	57 ± 29.6	27.1 ± 33.4
Veg height (cm)	2.5 ± 1.3	N/A	4.2 ± 4.4

### 3.4. Discussion

This study has found that there is a relationship between the environmental characteristics of a site and the differences in nesting densities observed both between and within nesting aggregations of *A. cineraria*, *A. fulva* and *C. hederæ*. Species was also found to play an important role, both in defining what represented 'high nesting density' and in determining what environmental attributes led to the optimisation of a site as nesting habitat. The findings from this study support the 'limited substrate hypothesis' as high-density nesting was associated with areas that had particular abiotic qualities.

#### 3.4.1 Spatial analysis of differences between nesting sites

The PCA visualised the differences between nest sites and demonstrated a clear split between *C. hederæ* sites and sites of both *Andrena* species (Fig. 3.2). Only two *C. hederæ* sites did not fall into this group; site 20, which had the mean lowest nesting density of all *C. hederæ* sites and site 16. This suggests that there are potentially differences in the nesting requirements of Colletidae species and Andrenidae species, but this cannot be established by the present study and more targeted research is required.

#### 3.4.2 Nest density

The findings from this study indicate that these three species nest in different densities in the UK. *Colletes hederæ* nested at significantly higher densities than both *A. fulva* and *A. cineraria* and *A. cineraria* nested in higher densities than *A. fulva*. This finding meant that it was not valid to examine these three species in a combined analysis with nesting density as the response variable.

*Colletes hederæ* was first recorded in the UK in 2001, but despite its recent appearance, the largest recorded populations of this species occur in Southern England, where they nest in the tens of thousands (Dellicour *et al*, 2014). At the time of writing, *C. hederæ* lacks specific natural enemies in this country that are present in the more southern parts of its range (species such as the cleptoparasitic bee *Epolus fallax* (Morawitz, 1872) and the meloid beetle *Stenoria analis* (Schaum)) and this may be driving its capabilities to nest in much higher densities in the UK (Dellicour *et al*,



2014). This situation does represent an excellent opportunity to study the influence of parasites on the nesting dynamics of fossorial solitary bees by comparing *C.hederae* aggregations in the UK to aggregations elsewhere in its geographical range. In comparison, the native species *A. fulva* has been found to experience significant parasitism from specific natural enemies (Paxton and Pohl, 1999). At one nesting site in Wales, the parasitic bee *Nomada panzeri* (Lepeletier, 1841) was found to have replaced 18% of the host's offspring (Paxton and Pohl, 1999).

Dellicour *et al* (2014) in their analysis of the rapid range expansion of *C. hederae* into the UK also pointed to forage as an important driver of its success. *Colletes hederae* utilises *Hedera helix* as its principal foraging plant. Because of this, they likely experience minimal competition for locally abundant forage that can have a long flowering period (6-8 weeks) (Dellicour *et al*, 2014). This can lead to females provisioning a lot more brood cells than other, closely related, species. In contrast, both *Andrena* species are active in spring and early summer when there is likely increased competition for locally abundant forage (such as flowering trees), which do not flower for as long as ivy.

#### 3.4.3 Among aggregation variation

For *C. hederae*, changes in nesting density accounted for 25% of the variation in environmental conditions between sites. Three factors were particularly well explained: the amount of bare ground and grass, and the proportion of sand in the soil (Fig. 3.5). Levels of bare ground and sand increased with increasing nesting density. For *A. fulva*, changes in nest density explained 23% of the environmental variation in the sites but in the case of *A. cineraria* only 4% of the variation in environmental characteristics was explained by nest density. This may have been due to outliers in the *A. cineraria* dataset that did not exist in the *A. fulva* dataset. In the case of *A. fulva*, nest density best explained differences in the amount of bare ground and grass and, the proportion of sand in the soil (Fig. 3.5), although this result was not significant. Higher nest densities occurred at sites with more bare ground and lower sand content, while low nesting densities occurred at sites with more grass. Sandy soils allow for easier digging conditions as these soils generally do not clump into aggregates and have better drainage (Cane, 1991). The current literature establishes bare ground as one of the most important predictors of nesting

occurrence (Potts *et al*, 2005; Sardinias and Kremen, 2014), although some evidence does exist to the contrary (Pane and Harmon-Threatt, 2017). This paper suggested that nesting studies may be biased toward sampling in areas with bare ground as, anecdotally, this feature is thought to be an important driver of nest site occurrence. However, given the spectrum of independent scientists who have reached this conclusion, over significant timescales and using multiple scientific approaches, this determination appears improbable (Wuellner, 1999; Potts *et al*, 2005). That said, it is likely that as the body of research on nesting ecology expands, primary assumptions will be challenged, but this is a symptom of the empirical knowledge base being so small and not necessarily any indicator of bias.

#### *3.4.4 Within aggregation variation*

This study has found that the environmental characteristics of the nest site do influence the spatial distribution of the nests within the aggregation. Higher density nesting was found to occur in areas where there was more bare ground. The importance of bare ground for the facilitation of solitary bee nesting has already been established (Wuellner, 1999; Potts *et al*, 2005). Although it is clear that bare ground is not a requirement for nesting, it does seem to encourage high density nesting and therefore likely improves the quality of a potential nesting site. There are three potential reasons why bare ground is preferred. The presence of bare ground indicates the absence of vegetation and this may facilitate easier nest digging. The less vegetation there is at a site, the less obstruction there will be in the ground for straightforward nest construction in the earth. Furthermore, a lack of vegetation means that nests are not obscured and makes navigation back to the nest after foraging bouts more efficient. Finally, it has been demonstrated that soil temperatures increase with decreasing vegetation cover (Song *et al*, 2013) and this could confer significant benefit to developing larvae. There are few clear drawbacks to establishing nests in bare patches of ground. The most obvious is that the location of the nest is not at all obscured and therefore may be easier for parasites to locate. This may reinforce the motivation of females to nest in high densities in bare ground as this may dilute the effects of their particular nest being easier to find.

It is unlikely that the proportion of bare ground alone determines the spatial distribution of nests in an aggregation. As the findings from this study

accounted for less than 20% of the within-aggregation variation in nest density, the hypothesis that other forces are more important drivers of the spatial distribution is supported. Evidence that parasite load decreases with increasing nest density (Wcislo, 1984; Larsson, 1986) indicates that these forces may work in tandem to create the clumped spatial distribution characteristic of solitary bee nesting aggregations. An empirical study that took into account both the physical characteristics and parasite dynamics of nesting aggregations would likely be very revealing in this context.

#### 3.4.5 Interspecific differences

Soil penetration, organic matter content, shade and slope were all found to differ significantly between the nesting sites of the three species (Fig 3.7; Tables 3.5 and 3.6). The results for both slope and shade are aligned with the findings of Chapter 2. Significant interspecific differences in slope were identified between all three species in this study. *Colletes hederæ* occurred at sites with the steepest slopes and *A. fulva* nested at sites with the flattest. In terms of shade, *C. hederæ* aggregations were found to experience significantly less shade than both *A. fulva* and *A. cineraria*. Further discussion on the implications of these findings can be found in Chapter 2.

Soil penetration was significantly higher at *A. fulva* and *A. cineraria* sites than at *C. hederæ* sites, with this relationship exhibiting the largest effect size of any environmental variable. Harder soils likely confer greater stability to the nesting burrow, which may be why the *Andrena* species exhibited a preference for this characteristic. Although previous work has shown that some fossorial species prefer softer soils as they require less energetic investment for excavation (Potts and Willmer, 1998). A study of the nesting densities of *A. camellia* at sites with homogenous and abundant foraging resource availability, Xie *et al* (2013), found that this species nested in significantly higher densities where the soil was loose (low penetration resistance), moist and experienced low temperatures. They placed particular emphasis on the importance of low soil compactness explaining that low soil temperatures could lead to increased soil moisture, in part due to reduced water transpiration and subsequently looser soil that requires less energetic investment for digging. The difference in preference between *C. hederæ* and the *Andrena* species could be explained by differences in their brood cell linings. The lining produced by *Colletes*

species is far more robust than that produced by the *Andrena* species, therefore *Andrena* species may rely more heavily on the stability offered by harder soils than *Colletes*.

Organic matter content was found to be significantly lower in the soils of *C. hederæ* nest sites than in the nest sites of both *Andrena* species. The organic matter content of the soil is the sum of decomposing plant and animal material along with soil organisms and the material they produce. The high organic matter contents at the *Andrena* sites may be due to the fact that all seven *A. fulva* sites and many *A. cineraria* sites were urban green areas such as parks and cemeteries, where the aggregations often occurred in grassy spaces adjacent to trees. This high presence of vegetation in the vicinity of the nest sites can lead to higher organic matter content in the soil. Therefore, this may be a consequence of the females' decision to nest near trees, which can provide a significant food source when in flower. Although this study found no significant difference between the nest sites of the two *Andrena* species in terms of organic matter content, a study of *A. vaga* (Panzer, 1799) nesting ecology in Cologne, Germany, reported the organic matter content of the soil at their study site to be 3.1% (Bischoff, 2003), substantially lower than the mean values for the two *Andrena* species recorded in this study (11.7% and 11.4%). This discrepancy could be attributable to multiple factors, but significantly, the *A. vaga* site in question is singular in its size and nesting density (approximately 10,000 nests in one location) and so is likely a poor candidate for comparison. Very few nesting studies have reported values for organic matter content and none could be identified for the species particular to this study. One UK study found that *H. rubicundus* nest sites had a mean organic matter content of 0.06% (Potts and Willmer, 1997) and while this is substantially smaller than the values reported here, they also reported that organic matter content did not vary significantly within or between *H. rubicundus* aggregations. This pattern of stability between aggregations of the same species does reconcile with the results of this study. Going beyond species-specific studies, organic matter content was shown to be a significant predictor of bee community composition at sites in Northwest Indiana, USA (Grundel *et al*, 2010). The percentage of soil nesting bees at a site was negatively correlated with soil organic matter content within 150m of the sampling transect. While soil organic matter content may not impact the nesting density of a species across nest sites or

within them, it may act as an environmental filter that can exclude species from certain sites and restricts them to particular areas (Kraft *et al*, 2015).

It should be noted that interspecific differences in the environmental attributes of nesting sites do not necessarily indicate differences in habitat preference between species. These differences may exist because certain characteristics are not available to all three species for a variety of reasons. These reasons may include temporal variation in the landscape between flight seasons or because the absence of suitable forage excludes a species from certain sites. What these differences do establish is that suitable nesting area is not the same for all fossorial solitary bees and that more detailed study of the ecological requirements of this group is required at the species level.

#### 3.4.6 Why do aggregations form?

Synthesising the current literature on this topic, the findings from this study and the findings of the citizen science project (Chapter 2), I hypothesise that aggregations form, and are spatially distributed in high density patches because of the interaction between three principal driving forces: abiotic properties of the site, proximity to forage and parasite pressure. As discussed previously, the physical location of the nest site is important because its physical characteristics such as its edaphic attributes and slope of the ground confer advantages including architectural stability, optimal digging conditions and thermal properties. Second, and moving beyond the scope of this study, these abiotic factors must exist at a site, which is within flight range of abundant foraging material. This theory is supported by findings that bees forage within just one per cent of their potential range in areas of floral abundance (Sardinas *et al*, 2015) and studies of specialist bees that show they preferentially nest in proximity to their host plant species (Julier and Roulston, 2009). I propose that bees nest in aggregations because suitable nest sites, both in terms of physical properties and proximity to forage are highly restricted within a landscape. This can also explain why some species of solitary bee, which are active at similar times and have similar foraging strategies, are frequently observed nesting together in one aggregation. An example of this are the three *Andrena* species *A. fulva*, *A. cineraria* and *A. nitida* (Müller, 1776), which I have observed nesting together on multiple occasions. This phenomenon has also been observed in *Trigona* species (Eltz *et al*, 2001). Parasite pressure then acts at the level of the aggregation along with the finer scale differences in the physical characteristic

of the site i.e. the amount of bare ground available. Together, these forces result in the spatial patterning of nests within aggregations into high and low density patches.

### 3.5 Conclusion

It has been established that the availability of suitable nesting habitat in a landscape can contribute to the long term stability of ground-nesting bee populations (Lopez-Urbe *et al*, 2015). With this in mind, it is imperative that we have a clear understanding of what constitutes a suitable nest site for the many species of fossorial solitary bee. This study represents a step towards that goal and has found that the environmental characteristics of a nesting site can influence nesting density. In terms of surface characteristics, both *A. fulva* and *C. hederæ* nesting densities exhibited a positive relationship with bare ground and a negative relationship with grass, a finding that could potentially be used in practice for the creation of high quality nesting habitat. Within aggregations, across all three species, spatial patterning of nests was also related to the proportion of bare ground, reinforcing the importance of this factor. However, it is likely that other forces (such as parasitism) also contribute heavily to the spatial configuration of nesting aggregations. There were significant interspecific differences in nest site characteristics, with most divergence occurring between *C. hederæ* and both *Andrena* species, indicating a potential role of taxonomic relatedness, flight period or differences in brood cell linings. Irrespective of mechanism, these findings indicate that one suite of physical attributes cannot be used to create optimal nesting habitat for all fossorial species. The results from both the nest density analyses and the characterisation of interspecific differences are in line with previous studies of both fossorial wasps (Srba and Heneberg, 2012; Bonte, 2005) and bees (Potts *et al*, 2003, 2005), which have found that while the limitation of suitable nest sites can impact nest density, species respond differently to abiotic characteristics.

## **4: The effects of vegetation management on fossorial bee and wasp nesting: a field experiment**

### **4.1 Introduction**

Solitary bees constitute the majority of bee species in the UK, they are important pollinators, and there is some evidence that they are experiencing declines in both occupancy and species richness in some parts of the world (IPBES, 2016). It is therefore crucial that we begin to establish quality guidance for the protection and creation of suitable nesting sites. While the previous two chapters of this thesis have focussed on adding to the knowledge base of what constitutes suitable nesting habitat, this chapter moves the research into practical application and will focus on testing the efficacy of potential management actions for the creation of suitable nesting habitat.

#### *4.1.1 Fossorial bees and wasps*

*Halictus rubicundus* was the focal species for this experiment as it had been recorded nesting successfully at the experimental site in the 1990s (Potts, 1995), but had left the site in the interim years (K.Wilson, pers. comm.). *Halictus rubicundus* is one of the largest species of halictine bee in the UK, it is highly polylectic and is widely distributed throughout the country (Collins and Roy, 2018). It exhibits interesting social behaviour and is reported to be eusocial in the warmer, southern parts of its range but solitary in the north (Potts and Willmer, 1997; Soucy, 2002). In a eusocial setting, mated queens found new nests in the spring and produce workers. The colony then produces males and females at the end of the summer. *Halictus rubicundus* is an aggregate nesting species and is one of the few fossorial bees for which empirical studies of its nesting environment have been carried out in the UK (Potts and Willmer, 1997).

Bees are not the only group of hymenopteran that nests solitarily in the ground as there are many species of wasp that also utilise this nesting strategy. These species are commonly known as digger wasps and there are more than 110 species present in Britain (bwars.com, 2019. About bees wasps and ants. Available at: <http://www.bwars.com>. [Accessed 15 August 2019]). As predatory species, they do not have the same requirement for flowering plants to be present in the landscape as solitary bees, but they

still face pressure to optimise their nesting sites. In this way, the provision of suitable nesting areas has the potential to attract not just multiple species of bee, but also wasps. Therefore, as a conservation action, the improvement/provision of nesting sites may represent high return on investment in terms of biodiversity.

#### 4.1.2 Providing nesting habitat

It has been demonstrated for one species of fossorial bee, that they can potentially prosper when the physical landscape is managed in their favour. The US native ground nesting bee *Nomia melanderi* (Cockerell, 1906, family: Halictidae), is the most successful managed fossorial bee in the world (Cane, 2008) and is singular in its capacity to thrive in artificially created nesting environments. This species is a highly effective pollinator of alfalfa (*Medicago sativa*) and is routinely translocated to agricultural contexts for the purposes of crop pollination. The principal reason for this species' success as an intensively managed animal is the combination of its unique biology and nesting requirements, which are fully elucidated. *Nomia melanderi* creates shallow nests in silty damp nesting soils (Cane, 2008), which means that it is amenable to being physically removed from a nesting site in soil blocks and transferred to a suitably prepared site. These prepared sites are cleared of vegetation, periodically sealed with salt and supplementally subirrigated to maintain a favourable nesting environment for the bees. *Nomia melanderi*'s foraging requirements are also easily met by the presence of the alfalfa crop, whose flowering season coincides with *N. melanderi*'s flight season. This mechanism of management is not feasible for the majority of fossorial species as the physical characteristics of their nesting sites (i.e heavily sandy soils) cannot be easily translocated intact. Furthermore, there is significantly less clarity around how to provide and maintain a suitable nesting site for most fossorial bee species. However, the success of *N. melanderi* confirms that fossorial species can profit significantly from sympathetic landscape management.

Much of the current, available advice for creating solitary bee nesting habitat outside of agri-environment schemes, focuses on the provision of nests for aerial nesting species (Wildlife Trusts: [wildaboutgardens.org.uk](http://wildaboutgardens.org.uk), RSPB: [rspb.org.uk](http://rspb.org.uk), Friends of the Earth: [friendsoftheearth.uk](http://friendsoftheearth.uk). Date accessed: 01/02/2019). Of the organisations that have published recommendations for ground nesting habitat, almost all advise the creation



of bare patches of soil in sunny, well-drained locations (Buglife: [buglife.org.uk](http://buglife.org.uk), The Xerces Society: [xerces.org](http://xerces.org), Penn State: [ento.psu.edu](http://ento.psu.edu). Date accessed: 01/02/2019), although none cite the sources of evidence for these actions. Conservation Evidence, a UK-based organisation that aims to provide evidence-based guidance for the conservation of wildlife, also recommends the creation of bare patches of ground for soil-nesting bees (Dicks *et al*, 2010), but this recommendation is formed on the basis of five studies. The first, and most robust, was a replicated controlled trial from Germany that found that fossorial bees and wasps preferred to nest in areas free of vegetation, where the soil had been dug and raised (Wesserling & Tscharntke, 1995). The four smaller trials, which were not replicated and controlled, also showed that the presence of bare ground had a positive impact on fossorial bee and/or wasp nesting (Severns, 2004; Gregory and Wright, 2005; Edwards, 1998; Edwards, 1996). This is the best guidance available at this time but even these actions have only been robustly tested by one experimental study, although the four smaller trials do help indicate that manipulating the environment can successfully increase nesting density.

In the absence of an appropriate evidence base detailing the physical nesting requirements of all fossorial species and the efficacy of potential management actions, very little guidance for the provision and maintenance of nesting habitat exists within policy guidelines and that which does exist is not always fit for purpose. For example, the Green, Low-carbon, Agri-environment Scheme (GLAS) is the agri-environment scheme in place in the Republic of Ireland for 2014-2020. Within this policy document there is an action for the provision of solitary bee nesting habitat on farmland (GLAS, 2015). The 'conservation of solitary bees' is a tier three general action for which participants are paid 45 euro per habitat per year for the 'sand' portion of this action. The objective is 'to improve biodiversity in the farming landscape and replace habitats lost through changes in farming practice' and requires participants to 'create a bee habitat by placing 1 tonne of builders' sand in a mound in a LPIS (Land Parcel Identification System) parcel or field and fence it from livestock by the 31st May'. The detail of this action includes the following: 'The fenced off area around the bee habitat must be strimmed annually and throughout the GLAS contract to keep shading vegetation under control. Pesticides cannot be applied to the habitat.' However, there is no evidence that this management yields

any results and confers any significant benefit to solitary ground nesting bee populations in agricultural contexts.

While we may not have a full and complete understanding of solitary bee nesting ecology, it is important that potential management actions are empirically tested for efficacy and practicality, or, in the absence of evidence, that expert opinion is sought. In this way effective recommendations can be made, although we may still lack intimate understanding of why these strategies work. Furthermore, a significant issue associated with conservation policy that is not grounded in evidence is that conservation as a whole may be seen as an unjustifiable expenditure if the policies that are in place do not yield results (Sutherland and Wordley, 2017). Therefore, in order to protect and conserve fossorial solitary bee and wasp populations for the long term robust, evidence-informed policy is required.

#### *4.1.3 Nesting experiments*

Roulston and Goodell (2011) laid out some common issues with solitary bee nesting studies in general and I attempt to address these in this experiment. They are particularly critical of the fact that of the few nesting studies that exist, none (or very few) have manipulated the nesting resources. They argue that ‘nest site limitation should be demonstrated by associating an increase in nesting resources with a subsequent increase in population sizes of bees, without changing other important variables.’

This chapter describes a replicated controlled trial, which set out to test the effects of both surface characteristics and anthropogenic disturbance on the success of a location as a nest site for fossorial, solitary bees and wasps. In terms of surface characteristics, bare ground has been found to be an important factor for solitary bee and wasp nesting (Wuellner, 1999; Potts *et al*, 2005), but how much more suitable bare ground is compared to other ground cover has not yet been quantified experimentally. Two of the experimental treatments in this study resulted in the creation of bare ground, by mechanical means (digging) and chemical means (spraying with herbicide). The aim of this approach was to understand whether both the presence of bare ground itself and the mechanism by which it is created influence the occurrence of nests. The other two treatments did not create bare ground directly, but manipulated the height of the vegetation in the

plots over the experimental period. Chapter 2 found that bees had the capacity to nest in areas that are vegetated and the aim of this approach was to more clearly understand the impacts of both the presence of vegetation and the height of vegetation on nesting density. There is some evidence that fossorial bees can withstand anthropogenic disturbance to the nest site and will nest in areas that experience frequent disturbance to the ground surface (Ullman *et al*, 2016), but again, this has not been quantified experimentally. This replicated controlled trial included fifty experimental plots in total, with ten repeats of five treatments. Anderson and Harmon-Threatt (2016) tested the effects of three different seed mixes on the occurrence of bee nests in a short, three day field experiment. They allocated four 0.3ha plots per treatment, but reported low statistical power. Wessering and Tschardtke (1995) used five experimental fields per treatment in their similar study of fossorial bee nesting. This experiment ran for 12 weeks and the number of nests in the plots was counted every two weeks. This included both holes with tumuli and those without, which may have represented emergence holes. These were included as solitary bees have been found to reuse emergence nests in this way (Cane, 2003) and Cane (2008) found that in *N. melanderi* nest aggregations, holes without tumuli were being used as nests just as frequently as holes with tumuli. Although population level effects could not be detected by this experiment, as it ran for only one year, it can show whether manipulating the environment in a certain way will lead to the local population of fossorial bees and wasps utilising that site as a nesting area. This experiment was also restricted to one site, meaning that landscape level effects, including foraging resources, did not change between plots and so the impact of nesting resource availability could be isolated.

#### 4.1.4 Research questions and hypotheses

This experiment set out to address three **research questions**:

1. Does vegetation cover impact solitary bee nesting?
2. How do four different vegetation management strategies impact the nesting density of solitary bees and wasps?
3. Can *H. rubicundus* return to a nesting site from which it has been excluded, within the first year of management change?

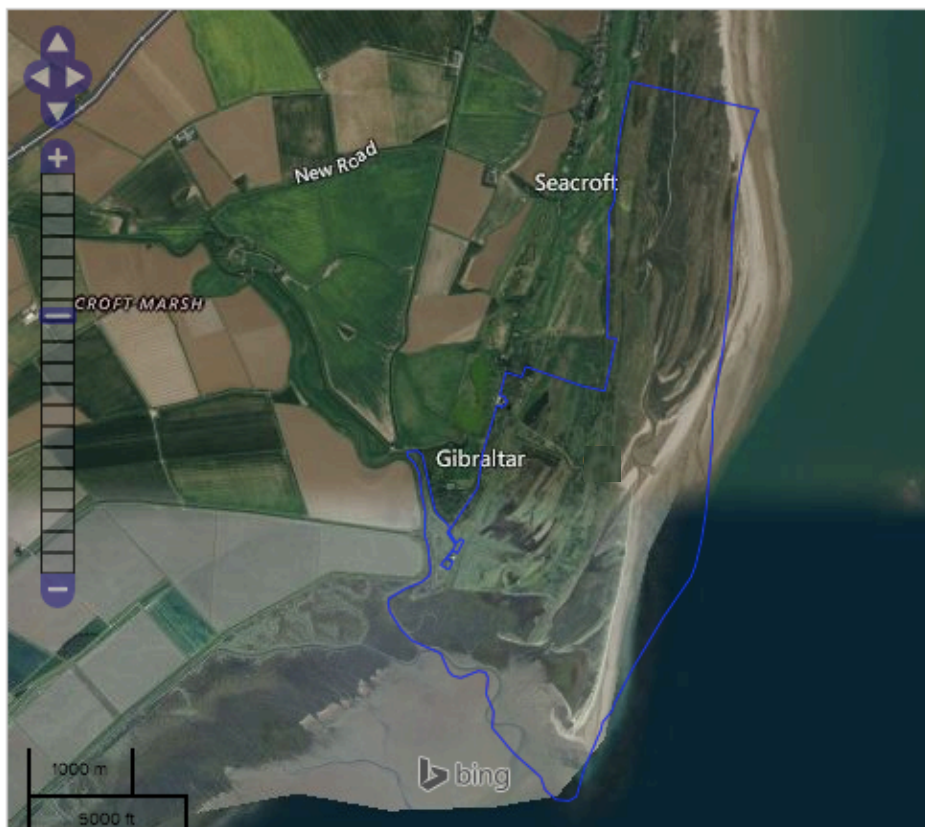
**Hypotheses:**

1. Vegetation cover does impact solitary bee nesting and differences will be exhibited between the treatments. Plots with lower vegetation cover will support high nesting densities.
2. Through the creation of bare earth, the uprooting and chemical spraying treatments will increase the number of bee and wasp nests. Plots that are strimmed once will not see an increase in nests due to the lack of bare earth and plots that are strimmed regularly will also be unattractive due to the regular disturbance to the surface of the ground.
3. *Halictus rubicundus* will return to plots where bare ground is available.

## 4.2 Methods

### 4.2.1 Site

Gibraltar Point National Nature Reserve is an area of approximately 414ha in Lincolnshire, England (coordinates: 53°06'N 00°20'E) (Fig. 4.1), which comprises two parallel ridges of sand dunes, separated by approximately half a kilometre of salt marsh. Key habitats at this site include sand dunes, salt marshes, freshwater habitats and open water. The site is used for both recreation and grazing and supports a variety of vegetation including ferns, sea holly and sea campion. It is also a significant site for some important bird species including grey plover, bar-tailed godwit and brent goose. In the mid 1990s a 600m eroded path along the west dunes at this site housed a large (roughly 500 nests) nesting aggregation of the solitary bee *H. rubicundus* (Potts, 1995). However, a survey carried out in July 2017 and observations by the site ranger over previous years revealed that the species may no longer nest in this area. In the interim years, the former nesting area has become covered in vegetation due to a decline in the rabbit population and a lack of vegetation management in this area of the site (K. Wilson pers. comm.).

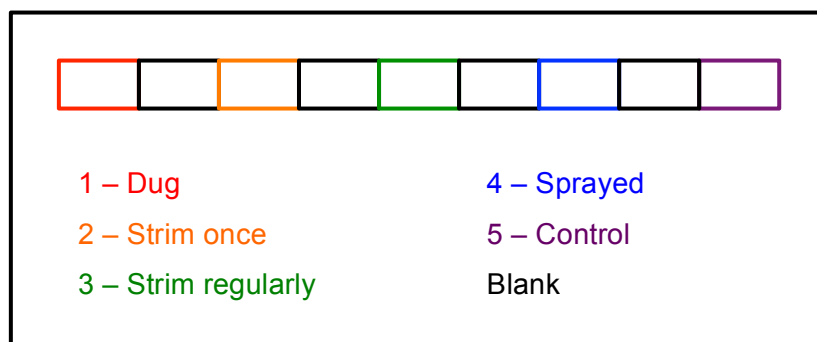


**Figure 4.1:** Map of Gibraltar Point NNR. Blue outline shows boundaries of the reserve. Map retrieved from ramsar.org 2019.

#### 4.2.2 *Experimental set up*

This experiment enlisted ten replicates of each treatment to try to achieve robust results with high statistical power. This decision was validated by a post hoc power analysis of control and bare ground plots, based on the means and the theta value (0.905) observed in this study. The relationship between control plots and plots where bare ground was created was investigated because it represented the most biologically interesting on the basis of what is known about solitary bee nesting i.e. that bare ground is important. This analysis determined that a sample size of seven plots per treatment would be the minimum needed to obtain statistical power at the recommended 0.80 level (Cohen, 1988) and was carried out using the power analysis test for negative binomial models in the MKmisc R package (Kohl, 2018).

In 2018, the experimental plots were established at the interface of sand dune and salt marsh habitat (Fig. 4.2) along a 360 x 1.5m (540m<sup>2</sup>) stretch of land. Each plot measured 3 x 1.5m (4.5m<sup>2</sup>). The size of the plots was constrained by the area available for experimental manipulation, which was bounded by fencing on one side and a public footpath on the other. The fifty plots were segregated into ten experimental blocks, within which each treatment was represented once (Fig 4.2). This was done to ensure that the experimental treatments were sufficiently spread along the full length of the experimental area. In this way if local populations of fossorial species emerged in close proximity to the experimental area, their choice to establish nest sites within certain treatment plots would not be constrained by their potentially limited flight ranges. The configuration of plot treatments within each block was assigned randomly, using a random number generator. Each experimental plot was buffered by a 'blank' plot, which had the same spatial configuration. This prevented positive spatial autocorrelation. This scenario can occur when experimental plots that are similar are directly adjacent to one another, thereby potentially confounding an individual's choice to nest in a plot with its spatial proximity to another plot instead of the experimental treatment. The blocks were not physically separated in space anymore than plots were. The idea of blocks was only used to ensure even spread of treatments across the experimental area.



**Figure 4.2:** Experimental setup for a field experiemnt testing the effects of vegetation management on solitary bee nesting. The map at the top indicates the area where the fifty experimental plots were established. The diagram below represents one unit of the experimental area. There were ten repeats of this unit with the order of the treatments within each unit randomly generated.

Each of the fifty experimental plots was assigned to one of five experimental treatments (four manipulations and one control), which manipulated the resident vegetation. Plot treatments were designed to mimic natural spatial variation in habitat, but critically, the treatments reflected the different strategies by which land managers currently manage vegetation. In this way, the outputs from this research could offer insights into how familiar practices could potentially be optimised for the creation of solitary bee and wasp nesting habitat. Each plot was subject to one treatment only to clearly examine the impacts of each potential management action in isolation.

The following is a description of how the experimental treatments were applied and includes their shortened labels by which they will be referred to, from this point on. All experimental manipulations were carried out in May 2018 and had been completed by the beginning of the survey period, unless otherwise stated:

#### **Treatment one**

##### **Label: Dug**

Plots subject to this treatment had all resident vegetation uprooted and the resulting bare ground compacted (Fig. 4.3a). Vegetation was dug out manually using a spade, ensuring that the plant and associated roots were removed. The bare ground was then stamped down underfoot so the soil surface was not loose.

#### **Treatment two**

##### **Label: Sprayed**

Plots subject to this treatment were sprayed twice, ten days apart, at the beginning of the experimental period (Fig. 4.3b). Plots received two sprays to fully remove the vegetation and create bare ground. A glyphosate-based herbicide spray was used (roundup pro biactive, manufactured by Monsanto). This chemical was already in use for vegetation management on the site. Glyphosate is the active ingredient in this herbicide and, as per product guidance, the dosage was at the rate of 250ml/10l of water (as per label standard) using a standard nozzle, giving a surface of 5l/ha. Spraying was carried out by a member of reserve staff in calm conditions. The correct nozzle size was used to avoid any fine



spray drift that might cause damage to non-target areas. Dead material was removed using a rake.

### **Treatment three**

#### **Label: Strimmed once**

Plots subject to this treatment were strimmed once, in the middle of May 2018, two weeks before the survey period commenced (Fig. 4.3c). Strimming was done to take the vegetation to as short a height as possible (approximately 2cm) and all strimming, for this treatment and treatment four was carried out by one member of reserve staff.

### **Treatment four**

#### **Label: Strimmed repeatedly**

Plots subject to this treatment were strimmed as described in treatment three, but this management was repeated once every two weeks over the entire experimental period of twelve weeks (Fig. 4.3c).

### **Treatment five**

#### **Label: Controls**

Control plots were not subject to any management of the resident vegetation (Fig. 4.3d).

Plots were monitored for fossorial bee and wasp nests every two weeks from June 5<sup>th</sup> to August 25<sup>th</sup> 2018. Each plot was investigated systematically, the observer started at the bottom left corner and systematically surveyed the entirety of the plot, until the top right corner had been reached. Plots were examined for approximately three minutes by the same individual at every survey to try to prevent bias in sampling effort. Although search efficiency was likely reduced in plots with more vegetation. At each survey, all holes within the plots were counted. Any individuals that were present and exhibiting nest-building or nest-provisioning behaviour were identified in the field or sampled for laboratory identification. A hand net was used to collect individual insects, which were killed immediately using ethyl acetate infused specimen pots. To demonstrate that the effects of the treatments were maintained through to the final survey, percentage vegetation cover (estimated by eye) was estimated for each experimental plot at the final survey.

Empirical measurements of soil texture, soil moisture, soil penetration and slope were taken from across the experimental area. Measurements were taken from every tenth plot. A 100g soil sample was taken for soil analysis. Procedures for the measurement of slope and soil analysis were as described in Chapter 3.



a: Dug plot



b: Sprayed plot (one spray)



c: Strimmed plot



d: Plots before treatment (control)



e: Most highly vegetated sprayed plot at final survey

**Figure 4.3:** Images of example treatment plots.

#### *4.2.3 Data analysis*

This dataset was split into six surveys for nesting activity. Because the experiment was not concerned with the effects of time on nesting (this is well established as species' have differing flight seasons), the data from each survey were analysed independently. Survey six was excluded from analysis, as this survey returned only two nests.

The experimental setup for this project included the separation of treatments into 'blocks'. This blocking was carried out, not to control for any differences in environmental characteristics across the experimental area, but to ensure satisfactory separation of the different treatments along the full length of the experimental area. This area was largely homogenous in its physical attributes. This was substantiated by empirical measurements of soil texture, soil moisture, soil penetration and slope taken from across the experimental area. The experimental area was south east facing. Mean soil moisture content was  $16.2\% \pm 0.03$ . The soil had a mean organic matter content of  $12.5\% \pm 0.1$  and gravel proportion  $10.2\% \pm 0.1$ . Soil texture was loam with mean sand content of  $47.1\% \pm 0.1$ , silt  $44.7\% \pm 0.1$  and clay  $8.2\% \pm 0.02$ . Individual measurements can be found in Table 4.1. Landscape level effects were not a confounding variable, as noted previously, this experiment occurred on one site and so its scale precluded it from suffering from landscape level heterogeneity. For these reasons, treatment was not nested within the block variable and instead block and treatment were included as independent variables in the analyses. This way any magnitude of difference between blocks could be elucidated.

The data from each survey were tested for overdispersion using the dispersion test from R package 'AER' (Kleiber and Zeileis, 2008) and were found to be overdispersed ( $c > 1$ ). Overdispersion occurs when the variance in the data has a value that is greater than the mean value. Overdispersion often occurs in count data as they can not include negative values and frequently include zero counts. Data with these characteristics fail the assumption of homogeneity of variance (variance=mean) on which most linear regression models rely.

The data were subsequently modelled using negative binomial regression. Negative binomial regression has the capacity to analyse overdispersed data and has been used in a number of similar studies including Martins *et al* (2018) study of the relationship between bee abundance and landscape features. All analyses were carried out in R version 3.5.1 (R core team, 2015).

**Table 4.1:** Measurements of site characteristics at five points (within every tenth plot) along the experimental area

	A	B	C	D	E
Soil penetration (kgf cm <sup>-2</sup> )	1.3	1.3	1.5	2.1	2.1
Soil moisture (%)	20	13	15	15	18
Slope (°)	12	12	20	8	10
Gravel (%)	9	2	5	20	12
Sand (%)	45.5	31.6	45.4	61.2	51.6
Silt (%)	47.9	58.8	49.1	30.6	37
Clay (%)	6.6	9.6	5.4	8.2	11.4
Organic matter (%)	14	18	11	8	12

## 4.3 Results

### 4.3.1 *The effects of vegetation treatment on nesting*

In all five survey periods that were analysed, the vegetation management significantly impacted the number of fossorial bee and wasp nests in the experimental area (Table 4.2). For all five surveys, plots that were trimmed regularly had higher numbers of nests compared to control plots (Table 4.2, Fig. 4.4), as did plots that were dug, although no difference was identified at the fifth survey for this treatment. Across the entire experimental period, the mean number of nests per plot for both regularly trimmed and dug plots was thirteen nests per plot. In total, plots with these treatments supported 133 and 121 nests respectively. This is in comparison to control and sprayed plots which housed 15 and 24 nests respectively and had a mean value of two nests per plot. Plots that were trimmed once supported 32 nests with a mean of three. Control plots had a mean nesting density of two nests per plot, despite having significant vegetation coverage. Nests in these plots were observed in naturally occurring, bare earth patches within the plot, reinforcing the importance of bare earth for colonisation. No significant differences between control plots and sprayed or trimmed once plots were found at any survey date (Table 4.2, Fig. 4.4). The blocks had an effect on the number of nests in surveys one, three and five, indicating that there was a spatial effect operating (Table 4.2).

Survey six, on the 14th of August, was not included in the analysis as it returned only two nests from the entire experimental area. The survey of vegetation cover found that this characteristic still varied between the treatments, establishing that an effect was still acting on this date (Table 4.3). The low return of active nests on this date was likely due to the fact that there are few species of fossorial bee and wasp that are active at this point in the season. This effect of seasonality was apparent across the three months of the experiment within the plots that had been subject to the most successful treatments (Fig. 4.5). Survey two, on the 16th of June, was the most productive survey, returning 105 nests from the fifty plots.

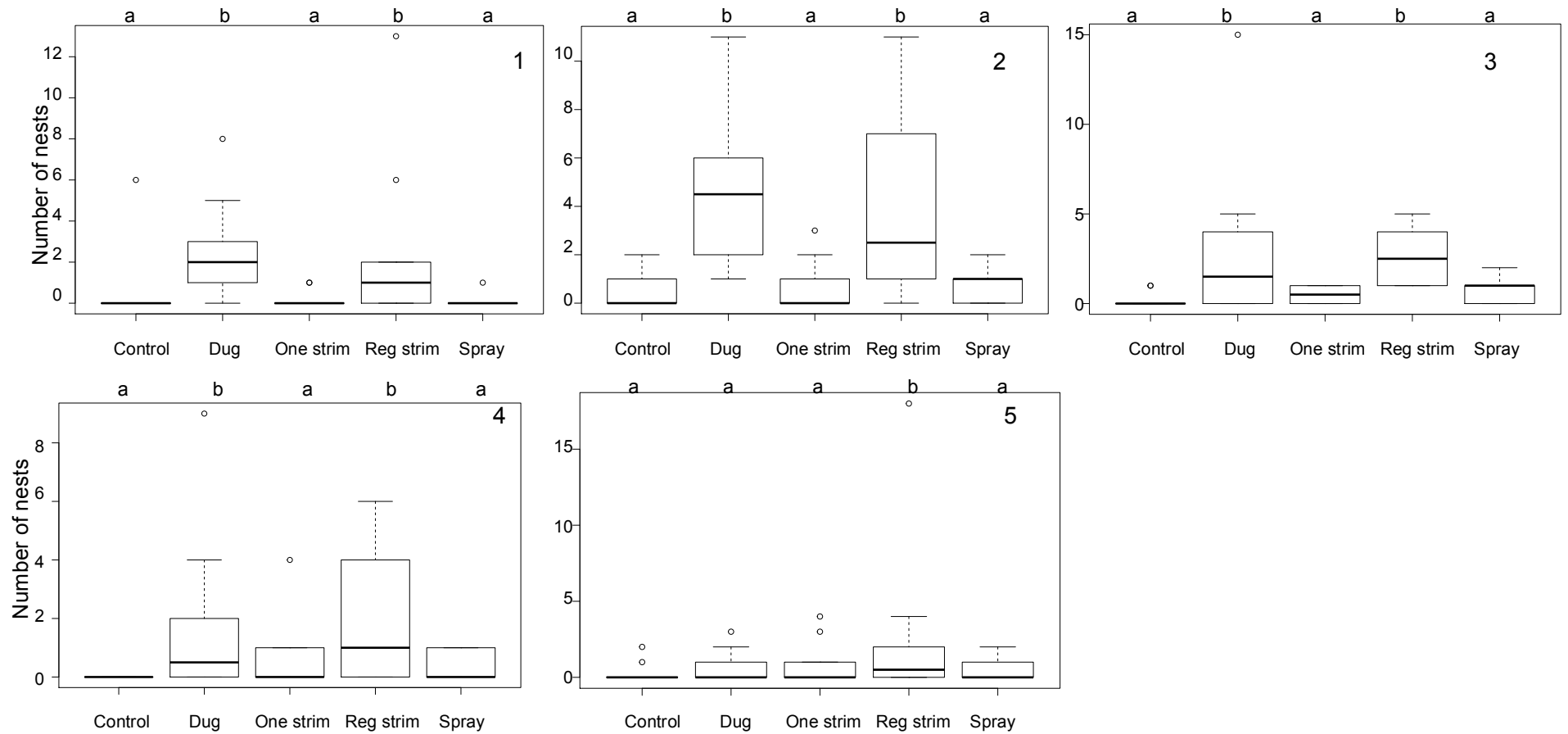


**Table 4.2:** Summary statistics for the five negative binomial models of the effects of vegetation treatment on the number of nests for each survey. (N=50 for each model). Control plots were defined as the reference level for treatment and block one for the blocks. Results in bold indicate a significant result.

	<b>Survey One</b>	<b>Survey Two</b>	<b>Survey Three</b>	<b>Survey Four</b>	<b>Survey Five</b>
Dug	<b>Est: 2.33</b> <b>z: 3.07</b> <b>p: 0.002</b>	<b>Est: 2.5</b> <b>z: 4.6</b> <b>p: 4.8x10<sup>-6</sup></b>	<b>Est: 2.63</b> <b>z: 3.47</b> <b>p: 0.0005</b>	<b>Est: 2.81</b> <b>z: 2.66</b> <b>p: 0.008</b>	Est: 0.79 z: 0.92 p: 0.36
Strimmed once	Est: -0.41 z: -0.41 p: 0.68	Est: 0.58 z: 0.9 p: 0.37	Est: 0.921 z: 1.05 p: 0.29	Est: 1.7 z: 1.53 p: 0.13	Est: 0.87 z: 1.02 p: 0.31
Strimmed regularly	<b>Est: 1.88</b> <b>z: 2.43</b> <b>p: 0.02</b>	<b>Est: 2.22</b> <b>z: 4.02</b> <b>p: 5.9x10<sup>-5</sup></b>	<b>Est: 2.62</b> <b>z: 3.46</b> <b>p: 0.0005</b>	<b>Est: 2.94</b> <b>z: 2.8</b> <b>p: 0.005</b>	<b>Est: 1.89</b> <b>z: 2.37</b> <b>p: 0.02</b>
Sprayed	Est: -1.5 z: 1.33 p: 0.26	Est: 0.72 z: 1.13 p: 0.26	Est: 1.41 z: 1.73 p: 0.08	Est: 1.11 z: 0.94 p: 0.35	Est: 0.31 z: 0.34 p: 0.73
Block 2	<b>Est: -2.44</b> <b>z: -2.37</b> <b>p: 0.02</b>	Est: 0.75 z: 1.27 p: 0.21	Est: 0.08 z: 0.12 p: 0.9	Est: 1.75 z: 2.15 p: 0.03	Est: 0.35 z: 0.34 p: 0.73
Block 3	Est: -1.42 z: -1.61 p: 0.11	Est: 0.85 z: 1.44 p: 0.15	Est: -0.54 z: -0.76 p: 0.45	Est: 1.29 z: 1.53 p: 0.13	Est: 0.01 z: 0.01 p: 0.99
Block 4	<b>Est: -1.5</b> <b>z: 1.33</b> <b>p: 0.03</b>	<b>Est: 1.17</b> <b>z: 2.07</b> <b>p: 0.04</b>	Est: 0.06 z: 0.09 p: 0.93	Est: -0.86 z: -0.68 p: 0.5	Est: -1.15 z: -0.82 p: 0.41
Block 5	Est: -0.92 z: -1.1 p: 0.27	Est: 0.98 z: 1.7 p: 0.09	Est: -0.59 z: -0.81 p: 0.42	Est: 0.62 z: 0.68 p: 0.5	<b>Est: 1.86</b> <b>z: 2.03</b> <b>p: 0.04</b>
Block 6	<b>Est: -3.5</b> <b>z: -2.6</b> <b>p: 0.01</b>	Est: 0.23 z: 0.36 p: 0.72	Est: 0.28 z: 0.47 p: 0.64	Est: -0.85 z: -0.67 p: 0.5	Est: -0.36 z: -0.31 p: 0.76
Block 7	Est: -1.64 z: -1.82 p: 0.07	Est: 1.03 z: 0.57 p: 0.07	Est: 0.89 z: 1.59 p: 0.11	Est: 1.52 z: 1.83 p: 0.07	Est: 0.25 z: 0.24 p: 0.81
Block 8	<b>Est: -1.83</b> <b>z: -1.97</b> <b>p: 0.048</b>	Est: 0.37 z: 0.59 p: 0.56	Est: 0.04 z: 0.06 p: 0.96	Est: -0.36 z: 0 p: 1	Est: 0.97 z: 0.99 p: 0.32
Block 9	<b>Est: -1.84</b> <b>z: -1.98</b> <b>p: 0.048</b>	Est: 0.36 z: 0.57 p: 0.57	Est: -1.21 z: -1.41 p: 0.16	Est: -0.17 z: -0.16 p: 0.87	Est: -36 z: 00 p: 1
Block 10	<b>Est: -3.5</b> <b>z: -2.6</b> <b>p: 0.009</b>	Est: 0.06 z: 0.09 p: 0.93	Est: -0.05 z: -0.08 p: 0.93	Est: 0.48 z: 0.51 p: 0.61	Est: 0.06 z: 0.06 p: 0.95

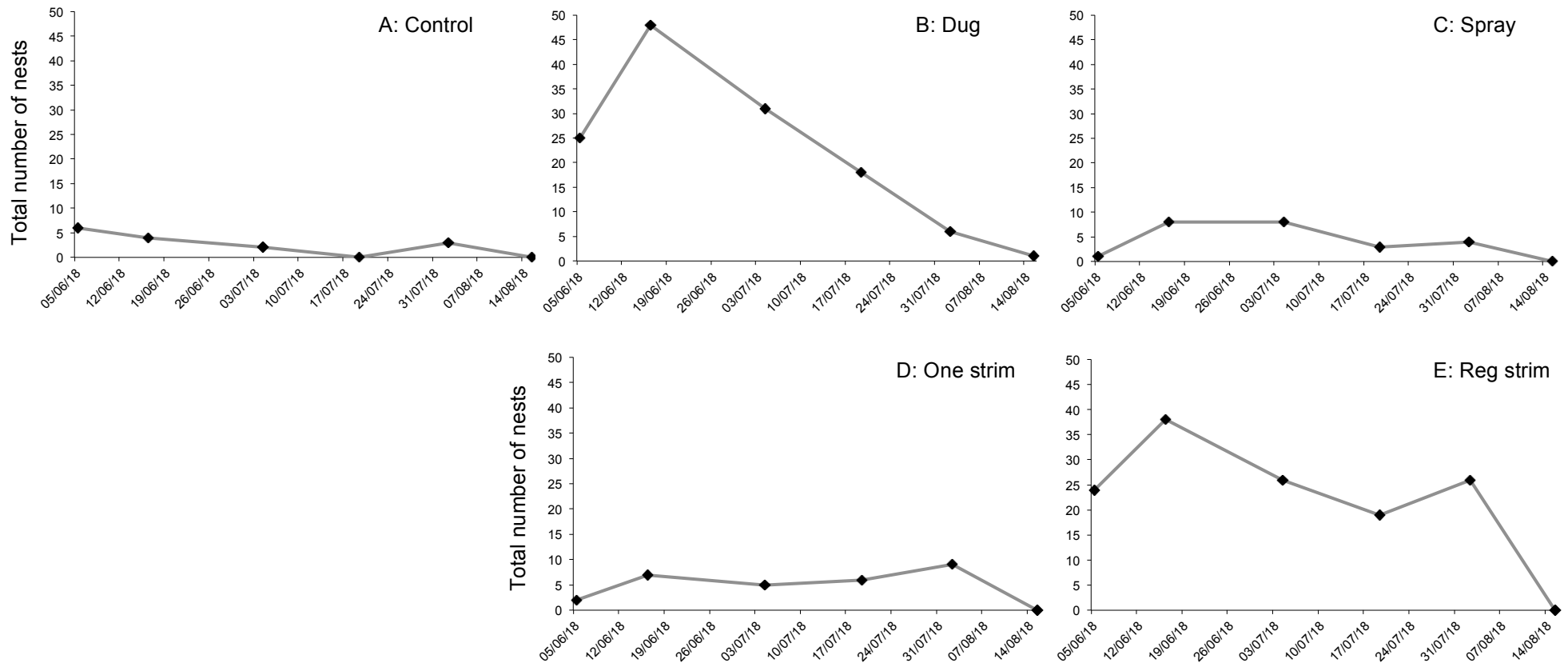
**Table 4.3:** Mean and standard deviation of percentage vegetation cover for all five treatments. This was calculated as a mean of the ten relevant plots for the treatment.

<b>Treatment</b>	<b>Veg cover (%)</b>
Dug	3 ± 2
Strim once	65 ± 29
Strim regularly	41 ± 24
Spray	16 ± 26
Control	98 ± 3



**Figure 4.4:** Boxplots illustrating the differences in the number of nests between treatments. Numbers represent the survey period. Letters indicate post hoc groupings for each treatment compared to control. Data shown are as explained in Figure 3.6 legend.





**Figure 4.5:** Line graphs showing the changes in the total number of nests over time for each treatment. Each value represents the combined number of nests from every replicate of the treatment (n=10) on the date of a survey.

#### 4.3.2 *Halictus rubicundus*

*Halictus rubicundus* was observed nesting in two adjacent plots in early July, specimens were collected and confirmed as this species. One nest was identified in a plot that had been strimmed once and fifteen active nests were found in a plot that had been dug. There were further nests outside of the experimental plots but in close proximity to them, in bare earth formed by an infrequently used footpath.

#### 4.3.3 *Other significant species identified in experimental plots*

In plots that had been dug and bare earth was created the following wasp species were identified exhibiting nest building or provisioning behaviour or, in the case of parasitic species, investigating nests: *Cerceris rybyensis* (Linnaeus, 1771) (a species of digger wasp of the Sphecid family), another Sphecid species; and an *Arachnospila* species.

## 4.4 Discussion

This experiment has shown that the creation of suitable nesting areas leads to an increase in the number of solitary bee and wasp nests. The creation of bare ground through digging and compacting the soil, and regularly strimming the resident vegetation were the two mechanisms by which suitable nesting habitat was established. Seventy seven percent of the 331 nests identified in this experiment were found in plots that were subject to one of these two treatments (now designated successful treatments).

### 4.4.1 Successful treatments

In line with previous findings (Wesserling & Tschardtke 1995; Severns, 2004; Gregory and Wright, 2005) and the accepted wisdom, plots where firm, bare ground was created were successful at attracting nesting species. In Oxfordshire, where nesting habitat was also created by digging out the ground, the managed areas experienced rapid colonisation by fossorial bees and wasps in the first year (Gregory and Wright, 2005). Three years post management, 80 species had been recorded nesting in the area. In Oregon, USA, the clearance of vegetation from small 1m<sup>2</sup> plots for the plantation of a rare plant, Kincaid's lupine, led to similarly rapid colonisation of the site by native fossorial bee species, in particular *Lasioglossum anhypops* (McGinley, 1986) (Severns, 2004). Therefore, the support grows for the establishment of bare ground to benefit fossorial bees and wasps.

A novel finding from this research is that plots where the vegetation was strimmed repeatedly were as successful as the dug plots, indicating that individuals can tolerate high levels of physical disturbance to the surface vegetation around the nest. This is encouraging and suggests that highly disturbed urban lawned areas, such as gardens and parks, may represent suitable nesting sites for fossorial species. This theory is supported by the fact that ground-nesting bee species can dominate bee communities in urban settings (Sirohi *et al*, 2015). That said, in tests of garden verses non-garden sites, greater ground-nesting bee species richness has been recorded at non-garden sites, where more spring-blooming trees and possibly, better nesting sites, were available (Langellotto, 2017). Similarly, a study of grassland habitats that were intensively grazed and extensively (unintensively) grazed found that while differences in vegetation

characteristics had little impact, insects at sites with high intensity grazing suffered from the disruption to plant-insect interactions (Kruess and Tschamntke, 2002). Again, the importance of combining suitable nest sites with sufficient forage material is clear. These studies, combined with the results from this experiment, seem to indicate that while fossorial species can potentially tolerate significant disturbance to the surface of the nest site, it is important that this disturbance does not undermine their access to sufficient foraging material in the surrounding habitat.

#### 4.4.2 Other treatments

Plots that were strimmed just once did not successfully attract nesting bees and wasps. This may be because the height of the vegetation in these plots over the course of the experiment did not offer sufficient access to the surface area of the soil for individuals to dig a nest. Furthermore, ground nesting bees use geographical landmarks to assist them when returning to the nest (Brünnert *et al*, 1994). High vegetation may make it more difficult for individuals to navigate back to the nest if landmarks cannot be easily distinguished from an appropriate distance. The temperature of the soil also decreases as the height of the surrounding vegetation increases (Song *et al*, 2013) adding to the unsuitability of the area as a nest site. The findings from this treatment indicate that a suitable nesting site must be easily accessible and not heavily obscured by surrounding structures or tall vegetation that may also reduce the soil temperature.

Sprayed plots did not provide suitable nesting habitat, despite the fact that these plots did offer patches of bare earth. After initial spraying there was some dead vegetation left on the ground, which may have impeded digging initially, but by the time the second survey occurred this had naturally dissipated and by the end of the experiment, there were just a few areas of vegetation within the plots. Figure 4.3d depicts the most highly vegetated sprayed plot at the final experimental survey. Despite the bare earth, these plots were not exploited as nesting sites as frequently as dug plots, with only 24 nests being found in sprayed plots over the entire experimental period, contrasting sharply with the 121 found in dug plots. There are multiple potential explanations for why these plots were unattractive to nesting females and further work is required to conclusively elucidate this. There may have been some dead vegetation left on the plots after spraying and raking that deterred females from nesting there. Glyphosate is

biodegraded both aerobically and anaerobically by soil microorganisms (Zhan *et al*, 2018) and so should dissipate away after a few days, although it's possible that glyphosate residues remained in the soil throughout the experimental period and that these had a repellent effect. More detailed analyses would be required to explicitly determine why these plots were unattractive, but it may be that the chemical spray alters the environment in such a way that nesting species avoid it. Recent work has begun to show that glyphosate has a lethal effect on developing stingless bee larvae, which come into contact with this compound in their larval food (Seide *et al*, 2018). Furthermore, Chan *et al*, (2019) found that direct exposure of solitary bees to insecticide residues in soil can be lethal and so there may be a selection pressure on bees to be able to actively avoid areas with high levels of pesticide. It has been established for other invertebrate species that they can express avoidance behaviour when pesticides are detected in soil (Louriero *et al*, 2005), although this has not been shown for glyphosate. Contrarily, experimental work has found that honeybees and bumblebees cannot detect and avoid neonicotinoid pesticides in sucrose solution (Kessler *et al*, 2015). Although it is unclear what mechanism is acting here, this result does tell us that it is not just the end product that matters but the process that was used to create the product that can significantly impact the outcome. For future management this research highlights that while the creation of bare ground is important and can encourage soil nesting bees and wasps into an area, this habitat should be created through physical manipulation of the environment and not by chemical means. Additionally, while the results from this experiment indicate that managed lawned area may have the potential to support nesting species, these findings also suggest that if these areas are exposed to pesticides, this may deter bees and wasps from establishing at these sites.

#### *4.4.3 Nest sites in the landscape*

The importance of bare ground for soil-nesting species has come through strongly in this experiment but looking at the bigger picture and where sites with plentiful bare ground currently exist is concerning. Brownfield sites may represent important refugia in the landscape for fossorial species due to their high proportion of bare earth but with this in mind, the current trend of loss of brownfield sites and their use for urban development (Macadam and Bairner, 2012) may have significant detrimental consequences for this group. That said, the species in this study seem to be able to exploit small

fragments of bare soil in an otherwise highly vegetated environment. This indicates that the provision of patches of open ground may be enough to positively impact fossorial bee and wasp populations over time. Whether patch size affects the quality of a nest site and bigger patches have the capacity to support higher nesting densities remains unknown. Again, as discussed in chapters two and three, the availability of forage in the surrounding landscape is likely to be an important factor in determining these dynamics.

To reiterate its importance, a reminder that the findings from this experiment do not represent a population effect, just that individuals from the local, existing population moved into the nesting sites when they were created. A longer term experiment over multiple seasons would be required to establish whether increasing the amount of suitable nesting area in a landscape has a positive impact on fossorial bee and wasp populations.

#### *4.4.4 Halictus rubicundus and other sampled specimens*

*Halictus rubicundus* re-established itself in the experimental area, marking this management as a success at attracting the local population into the newly created plots. Fifteen of the sixteen *H. rubicundus* nests identified in this study occurred in a plot that had been returned to bare earth so the females exhibited a strong preference for this treatment. The preference of *H. rubicundus* females to nest in bare ground has been demonstrated in a number of studies, both in Europe (Potts *et al*, 2005) and the USA (Cane, 2015; Soucy, 2002). Furthermore, there is evidence to suggest that bare earth nest sites are enhanced by the presence of pebbles or stones (Potts and Willmer, 1997; Soucy, 2002; Cane, 2015). This phenomenon may be driven by the thermal benefits that the presence of stones confers to the adjacent soil (Potts and Willmer, 1997) or because they represent useful landmarks when navigating back to the nest (Brunnert *et al*, 1994). Although the exact mechanisms of this preference is unknown, it has been shown that even in the case where bare earth is available, females will preferentially nest next to pebbles (Cane, 2015). Therefore the plots at Gibraltar Point may benefit from the addition of this material to the bare ground plots. *H. rubicundus* nests in sandy loam soil (Cane, 2015), which was the soil type present at the experimental site. This demonstrates that the site had the edaphic capacity to support this nesting species, but was not being utilised due to the surface characteristics. This is important

because it shows that managing the vegetation and creating bare ground at a site may not be enough to support nesting fossorial species if the soil conditions at that site are not favourable. Therefore, any plans to create nesting habitat must start with an investigation of the soil environment, the importance of this has also been demonstrated by the findings from Chapter three.

Not all nests could be associated with a specific species as individuals were not always present at the nest at the time of the survey. Of the individuals that were identified, three wasp species were found nesting in the dug plots. *Cerceris rybyensis* or the ornate-tailed digger wasp is a member of the Crabronidae family and is a common and widely distributed species in the UK. Similar to the closely related species *C. halone* (Banks, 1912), *C. rybyensis* nests in sandy soils in areas where there is little vegetation (Byers, 1978). The second specimen was of the genus *Arachnospila* which is a member of the *Pompilidae* or spider wasp family and many of the species in this group are fossorial. The final specimen was from the *Sphecidae* or digger wasp family but could not be identified further than this. Fossorial wasps have very little profile with the wider public, despite their importance as predators of pest species and their contribution to pollination, but it is clear from this experiment that managing a site to favour fossorial bees can also benefit fossorial wasps.

#### **4.5 Conclusion**

The sympathetic management of vegetation at a site with favourable soil conditions can encourage local solitary bee and wasp species to utilise that site for nesting. Sympathetic management involves the creation of bare ground or patches of bare ground through physical manipulation of the vegetation i.e. digging and strimming. Regular strimming of vegetation does not appear to have a negative impact on the success of an area as a nesting site, as long as foraging resources remain present in the surrounding landscape. Vegetation that is of a height to obscure nest entrances and/or impede digging may discourage nesting. Furthermore, the use of the chemical glyphosate to clear vegetation deters individuals from nesting at a site. Similar management strategies can be successful for both solitary bee and wasp species and so can be highly effective methods of increasing biodiversity and potentially safeguarding the future of

populations of these species. The findings of this replicated, controlled experiment substantiate previous findings and taken together, these results provide an appropriate evidence base for the advocacy for the creation of bare ground as a conservation action for fossorial bees and wasps.



## **5: General discussion and concluding statement**

### **5.1 General discussion**

Looking to the future, as the global environment continues to experience unprecedented change, Murray *et al* (2009) put it well when they stated ‘the sensitivity of particular bee species to the wide range of natural and anthropogenic drivers remains largely unknown. It is therefore essential to understand the basic ecology of bees in order to predict how they respond to environmental change and how these changes can be mitigated against.’ This thesis addresses one particular gap in our understanding of basic bee ecology and as such, has contributed to our potential to appropriately protect and conserve solitary bees in the future.

#### *5.1.1 Solitary bee nest site characteristics*

The citizen science project showed that while the four species of solitary bee examined had the capacity to nest within a range of environmental characteristics, there were indications that differences between the species existed. These findings were further developed by the fine scale studies of Chapter 3, which described the similarities and differences between the nest sites of the solitary bee species and demonstrated that particular environmental characteristics could lead to higher density nesting.

The importance of bare ground for the facilitation of fossorial bee nesting came through strongly, in all three data chapters. This finding builds on previous work (Wuellner, 1999; Potts *et al*, 2005) and emphasises the ubiquity of this requirement across fossorial species. However, this conclusion raises concerns as the provision of bare ground is rarely, if ever, included in conservation actions or land management plans. Bare ground is important not just for solitary bee nesting but for the proliferation of other insect groups, such as Carabid beetles (New, 2007; Cameron and Leather, 2011). This thesis supports the notion that the provision of bare earth is important in terms of insect conservation and should be included in more conservation management actions. However, as the findings from Chapter 4 demonstrated, it is important to consider how the bare ground is produced and physical manipulation of the environment is favoured over chemical approaches. Brownfield sites in particular can contain vast quantities of this important site characteristic and their future protection should be safeguarded (Eyre and Woodward, 2003).

In order to explicitly define the nesting requirements of solitary bees, more experimental study is called for. This thesis has demonstrated the efficacy of field experiments in this context and there would be significant value in repeating and up-scaling this approach. Furthermore, laboratory experimental analysis has the potential to provide substantial insights due to its capacity to control for all confounding variables. While there is some published information on how to take wild larval specimens into the laboratory for further development (Sommeijer, 2012), more guidance is required.

#### *5.1.2 Vegetation management and conservation of solitary bees*

Both the field studies and field experiment found that regularly trimmed or mown vegetation or grass can support sizeable populations of fossorial bees, a finding that contradicts some of the accepted wisdom that intensive management of a site will have a negative impact on bee populations. However, it is important to stress that while nest sites themselves can occur in managed grasslands, the bee populations will still require access to suitable forage material in close proximity (Dicks *et al*, 2015). Therefore, in terms of land management, it is important to note that while some lawned areas can be tolerated, they must co-exist with habitat that offers foraging opportunities such as, wildflowers. As has been recommended many times before (Williams and Kremen, 2007; Mandelik *et al*, 2012), the creation and curation of mosaic habitats is likely to be most beneficial for fossorial bee and wasp populations. In terms of nesting sites, Chapter 4 demonstrated that the provision of even small areas of bare ground can benefit fossorial bees and wasps. Therefore this represents a simple, low investment way of improving sites for bee and wasp conservation.

#### *5.1.3 Methodological considerations*

This thesis has explored three methodological approaches for the assessment of fossorial bee nesting ecology. The most novel approach was the citizen science project, as hypothesis-led citizen science is, in itself, a nascent field and citizen science approaches have never before been used to collect data on solitary bee nest sites. Although this approach proved successful and has the potential to be a very useful tool in this area

of research, there are some considerations to be taken into account for future endeavours.

The use of the citizen science project to recruit field sites for the fine scale study proved successful and this represents a potential framework for overcoming the logistical issue of identifying active nest sites quickly for the purposes of analysis. However, in this case, the recruitment of sites was, in a way, biased by a lack of ethical approval to gather personal data (such as names and email addresses) from citizen scientists. This led to the exclusion of any nesting aggregations that were recorded on private land as potential field sites, as site permissions could not be obtained. This was a particular issue for *A. fulva*, for which many records were reported from gardens. This also explains why, despite being the most successful species in terms of the number of records submitted (141) only seven *A. fulva* field sites were sampled. For future work, the collection of participant contact information would facilitate access to these sites and reduce the associated biases.

Another bias that likely presented itself in the citizen science data was that of remote versus populated areas. Nest sites were probably more likely to be reported if they occurred in areas where participants were likely to find them e.g. public places with lots of footfall. In this way, nest sites that were present in more remote areas with little or no public access were probably excluded from the study. This dynamic may also have presented itself in the results of the study. For example, although the majority of records from the citizen science project reported *A. fulva* to be nesting in grassy areas, the fine scale study of Chapter three found that increasing bare ground favours increasing nest density for this species. These results may indicate a bias in the citizen data towards lawned areas such as gardens and parks, which participants encountered more frequently. It may also be demonstrating that *A. fulva* is frequently nesting sub-optimally due to a lack of high quality nesting sites within dispersal distance, but this would require further study.

Future research would benefit from the continuation of the use of citizen science to understand solitary bee nesting ecology. One potential mechanism would be to build a recording scheme for solitary bee nest sites into established monitoring networks, such as iRecord. The availability of a current database of active nesting sites could potentially facilitate a lot more research in this area as it would negate the requirement of the researcher

to locate nest sites at the outset of any study. The Solitary Bee Project focussed on four charismatic solitary bee species, that can be identified in the field. The same level of success could probably not be achieved for species that are more difficult to identify, however, records of nesting sites, even if the specific species nesting there is unknown, are still valuable. Additionally, if photographs of the species are included in the record, then in some cases these could be identified by experts, at least to genus level.

#### *5.1.4 Environmental filtering*

Environmental filtering is a metaphor for the way in which the characteristics of an environment only permit species with certain traits or phenotypes to establish and be maintained there (Lailberte *et al*, 2014). Kraft *et al* (2015) expanded this definition and developed a framework (summarised in Fig. 5.1) for the consideration of how environmental filtering may act within an environment in light of species coexistence theory. Although these theories are typically discussed in terms of the community assembly at a site, it is interesting to view the presence of nesting bees and the density of those nests through the lens of these concepts. And so I use this framework to examine the findings from this thesis with regard to the nesting ecology of fossorial solitary bees.

The first level of this framework is **dispersal limitation**. Dispersal limitation is a separate mechanism to environmental filtering and occurs when there is no capacity for a species to arrive at a suitable site as it too far from the source site, in this case, the natal nest site. Therefore, the first step of evaluating an observed pattern in terms of environmental filtering is to discount the potential effects of dispersal limitation. Part of the reason why presence/absence studies were not used as part of the observational studies in this thesis was because the absence of nest sites does not indicate that the abiotic environment is unsuitable for nesting, instead, dispersal limitation may be acting. Therefore, the patterns observed in this research can not be attributed to dispersal limitation.

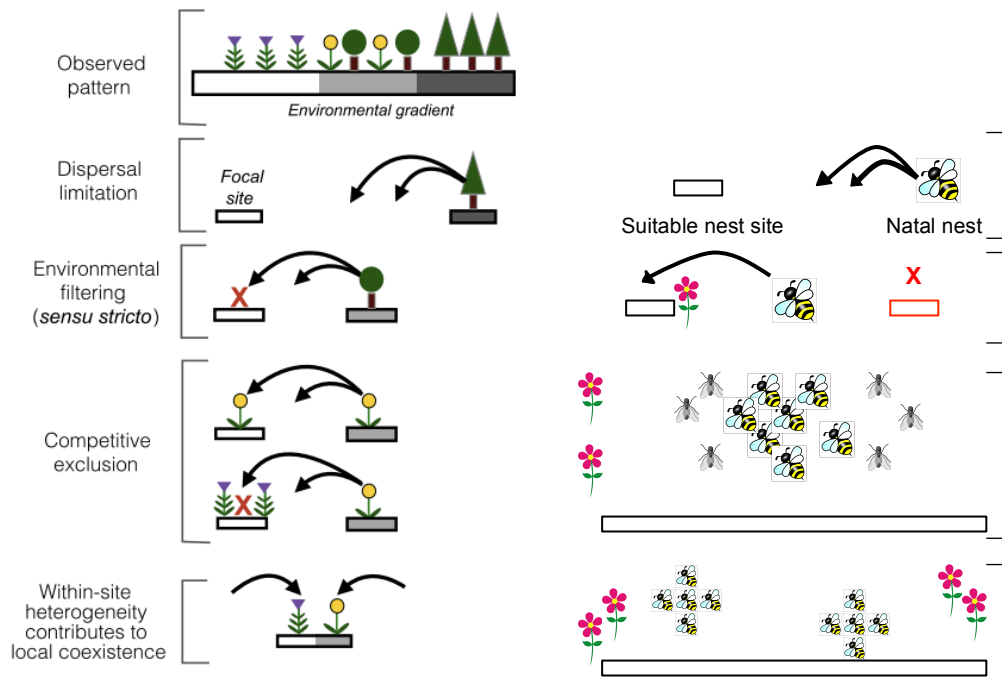
Once dispersal limitation has been excluded, **environmental filtering** may subsequently be considered. In the context of this research, this step is represented by the abiotic attributes that must be present at a site in order to facilitate nesting. This would include fundamental characteristics, such as, the ground must be diggable, through to attributes that exist on a

spectrum, including those most closely considered by this thesis such as soil texture and surface characteristics. Other features, including the presence of suitable forage within reasonable proximity, would also act at this level of the framework.

**Competitive exclusion** is the next level of the framework and refers to the effects of competitors on the establishment of species. The significant interaction at this level is the interaction with parasitic species and how that drives the organisation of nests within an aggregation.

The final level of the framework is **within-site heterogeneity**, which considers finer scale differences in the abiotic environment. Applying this to nesting ecology, it could potentially include considerations such as the availability of bare ground within an aggregation or the maintenance of nest architectural integrity in areas of high density nesting.

This thesis did not set out to test the theory of environmental filtering and the data collected cannot be used to either support or contradict any of the associated theories. However, the findings from this study and the theories presented, when viewed in light of environmental filtering, as defined by Kraft *et al* (2015) (Fig. 5.1) are interesting. The presence of fossorial bees at a site and the spatial dynamics of the nesting sites may be well encapsulated by this framework. While more species specific studies are required to elucidate any specific ecological needs, if this framework was empirically validated, it could offer a more global understanding of solitary bee nesting that would be particularly useful for informing top-down modelling studies of bee populations.



**Figure 5.1:** The diagram on the left shows the environmental filtering framework taken from and defined by Kraft *et al*, (2015). The diagram on the right demonstrates how solitary bee nesting dynamics may fit that framework.

#### *5.1.5 Future research directions and potential policy implications*

There are a number of research questions that have been raised by this work. First, it would be useful to better understand the interaction between the provision of suitable nesting sites and the provision of foraging resources. For example, how far will different species travel from the nest site to access forage? By better understanding this relationship more robust conservation strategies can be designed wherein all critical ecological resources are provided within suitable proximity. On the question of the provision of nesting resources, this thesis has described some of the important physical attributes of nesting sites and the next step is to decipher whether artificial nesting sites with these particular characteristics would be utilised by ground nesting bees. This research would be particularly useful in informing some of the ineffective recommendations that are currently part of the Irish agri-environment scheme (discussed in Chapter 4). Finally, this thesis has focussed on the nesting ecology of aggregate nesting species of solitary bee, but many species do not nest in these formations and instead establish nests individually, away from conspecifics. Future research could focus on this group of species, although there would be significant difficulties associated with carrying out this work due to the highly cryptic and ephemeral nature of these nests. Insights from this thesis go some way to overcoming these issues. By using our findings to provide suitable nesting sites, potentially attracting non-aggregate nesting species into these sites and monitoring them over multiple years, we could begin to develop an understanding of the nesting requirements of non-aggregate nesting species.

## 5.2 Concluding statement

This thesis has made significant, original contributions to scientific knowledge both in terms of fundamental ecological knowledge and in the exploration of methodological approaches, thereby potentially enhancing our capacity to protect and conserve wild bees.

With regard to ecological knowledge, this thesis represents the first empirical body of work examining the nesting ecology of fossorial bees in Britain for more than twenty five years. It is the first piece of research to empirically describe the nest site characteristics of the native species *A. fulva* and *A. cineraria* and the non-native species *C. hederæ* in the UK context. It is also the first in the UK to empirically examine nest site management strategies for the conservation of fossorial bees and wasps and make evidence-informed recommendations for the implementation of these strategies in the future.

With regard to research methods, this thesis has presented the first instance of the use of citizen science to gather data on fossorial bee nest sites. It has validated this approach and demonstrated its efficacy in overcoming some of the innate difficulties with gathering fossorial bee nesting data. By linking this approach with a field observational study, this thesis has shown how traditional approaches can be successful when a database of active nest sites is available to the researcher. Finally, this thesis has demonstrated that experimental studies are highly effective in the context of nesting research and represent an approach by which great gains can be made in both the understanding of fossorial bee and wasp nesting and in developing and validating management practices for the protection and conservation of this important group.



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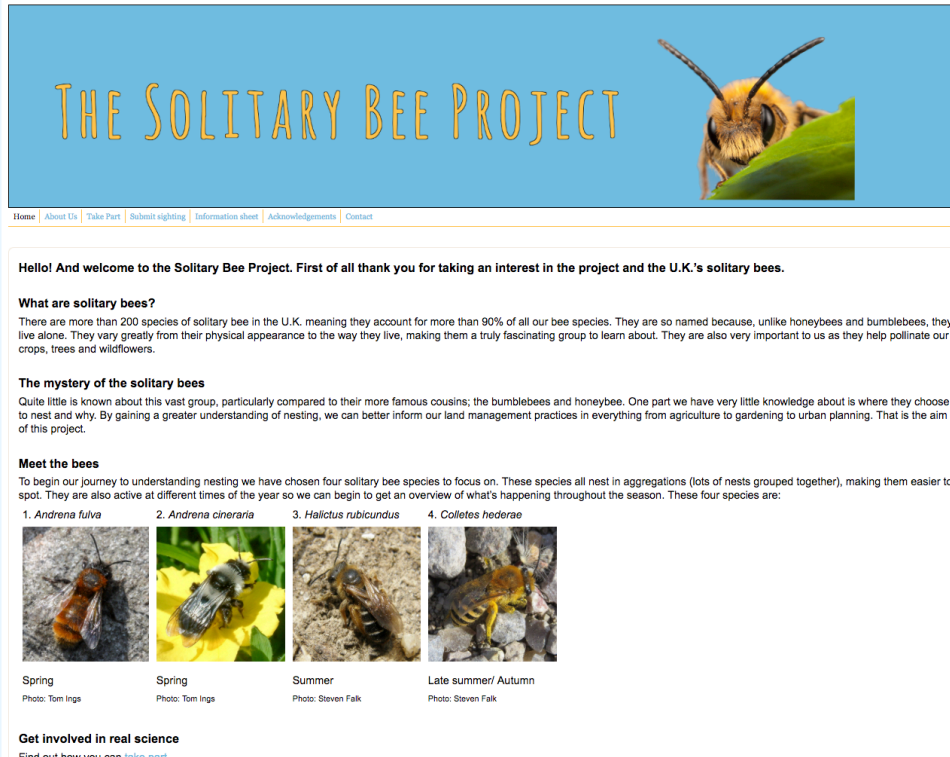
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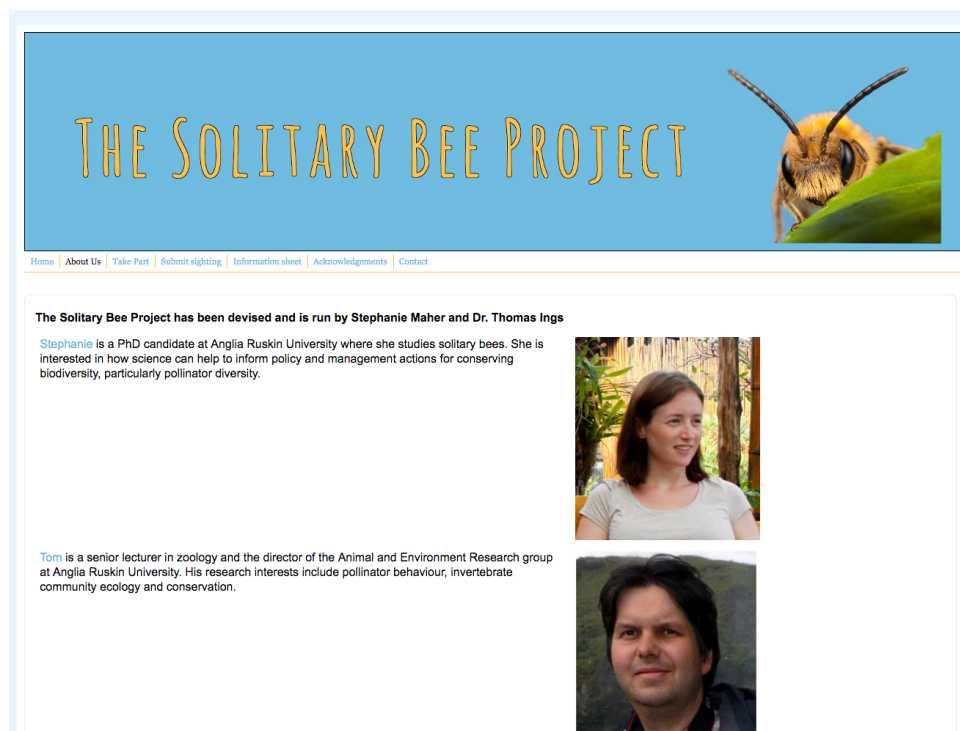
## Appendices

### Appendix 1: Screenshots from the project website: Thesolitarybeeproject.org

#### Home page




#### About us



## Take part

# THE SOLITARY BEE PROJECT






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We just need you to find aggregations and tell us about it. Maybe you have already seen nesting aggregations in previous years, in your garden or your local area, well we'd love you to take a closer look at those. Or maybe you're willing to look for the nests, just spend a little time searching the ground on your usual walking route or daily commute, you may be surprised by what you see!

[Click here](#) for a printable/ downloadable copy of the questions we'd like you to answer. You can either print it off and take it out with you or just download it to your phone. Either way we ask that you come back here and submit your recording through the website. [Click here](#) for our information sheet, which will give you some more guidance on how to identify the bees and answer the questions.

All of the submitted records will be used in research to better understand the needs of our solitary bees, the results of which we hope to publish in the future. Keep an eye on this page and our social media accounts for updates!

**Here are some photos of solitary bee nests to help you get started:**




There are some other animals, such as ants, that have nests in the ground so be careful not to get confused. Remember, it's important that you see a bee near the nest, or preferably, using the nest so you're absolutely sure. If in doubt, snap a picture or video and send it to us to have a look at, we're always happy to help.

[solitarybeesproject.org](http://solitarybeesproject.org)

## Submit sighting

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What Did You See? | When did you see it? | Where did you see it?

Please enter the species you saw and any other information about them. If you see more than one species in the aggregation please submit each separately.




Photo: Tom Ings




Photo: Tom Ings




Photo: Steven Falk




Photo: Steven Falk

Species:

☒ *Andrena fulva* (tawny mining bee)

☐ *Andrena cineraria* (ashy mining bee)

☐ *Halictus rubicundus*

☐ *Colletes hederæ* \*

Photos

[Add photo](#)

How many nests were there?:

Select option for nest

How shady was the aggregation?:

How sloped was the ground the aggregation was on?:


Was most of the ground::

Overall comment:

The page uses an Indicia Data Warehouse supported by the Biological Records Centre

## Contact Us

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### Contact

Your name \*

Your e-mail address \*


Subject \*

Message \*

Send message

## Acknowledgements




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**This project has been funded by Anglia Ruskin University.**

This website uses an [Indicia](#) Data Warehouse hosted by the [Biological Records Centre](#) (a group within the NERC [Centre for Ecology and Hydrology](#)).



The many beautiful photos on this site are being used with the kind permission of the artists who created them. You can see more of their work in the following places:

[Steven Falk](#), whose Flickr account I would implore anyone with an interest in wildlife to explore.

If you're interested in bee identification and taxonomy then Steven's excellent field guide can be found [here](#)

The photograph on our banner is by [Antony](#)

We also thank Max Kerney and Fin Oates for their help with design.