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Original article

Testing a detection dog to locate bumblebee colonies and estimate nest density*

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Abstract – Bumblebee nests are difficult to find, hampering ecological studies. Effective population size of bumblebees is determined by nest density, so the ability to quantify nest density would greatly aid conservation work. We describe the training and testing of a dog to find bumblebee nests. The dog was trained by the British army, using *B. terrestris* nest material. Its efficacy in finding buried nest material of a range of bumblebee species was 100%, and no false positives were recorded, suggesting that the dog was able to generalize across *Bombus* species. The dog was then used to locate bumblebee nests in four different habitats on the island of Tiree, west Scotland. The dog located 33 nests, and nest densities recorded varied from 0 to 1.86 nests per hectare, according to species and habitat. Habitat preferences appeared to be evident among the bumblebee species, with most *B. muscorum* nests in machair and all of the *B. distinguendus* nests being in dunes. We conclude that the technique has great potential, but note that using a dog to detect nests in more densely vegetated habitats may be less successful.

nest density / nest odour / Hebrides / *Bombus distinguendus* / *Bombus muscorum*

1. INTRODUCTION

Bumblebee nests are difficult to find, often being underground or in dense vegetation (Sladen, 1912; Osborne et al., 2008). Although individual nests of common species are often found serendipitously, they probably represent a small fraction of the nests that are present, so estimating bumblebee nest density is problematic. This is unfortunate since estimation of effective population size requires estimation of nest density (each nest representing one breeding female). Nests of the scarce bumblebee species are very rarely found, so we have a poor idea of their nesting requirements. In the UK, the most detailed descriptions of nesting habitats are provided by Sladen (1912) and for many of our bumblebee species little has been added to his observations since.

These problems have been partially overcome by use of molecular markers to asign worker bees to sibship groups, providing an estimate of the number of nests represented by workers at a particular forage patch (Chapman et al., 2003; Darvill et al., 2004; Knight et al., 2005; Ellis et al., 2006). An alternative technique is to enlist the help of large numbers of volunteers to search for nests (Fussell and Corbet, 1992; Osborne et al., 2008). This approach has provided valuable information on nest density and nesting locations of common bumblebee species, but information is heavily biased towards gardens and common species.

Anecdotal evidence suggests that badgers may be major predators of bumblebee nests, which they dig up at night (Sladen, 1912; Goulson et al., 2002). It seems probable that they detect the nests using olfactory cues. Detection dogs have been used for many years within the armed services, border controls and law enforcement. Detection dogs have also been used in a wide range of

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ecological contexts. For example Wallner and Ellis (1976) trained dogs to locate gypsy moth, *Lymantria dispar*, egg masses with a 99.5% positive identification rate, and dogs have also been used successfully to detect infestations of termites (Brooks et al. 2003). Use of dogs to locate scats has proved to be a valuable tool for studies of scarce mammals such as bobcats (*Lynx rufus*), fishers (*Martes pennanti*) and bush dogs (*Speothos venaticus*), (Long et al., 2007; Dematteo et al., 2009).

Here we test the efficacy of a detection dog in locating bumblebee nests, and then use the dog to detect nests of rare bumblebees and estimate nest densities on the Hebridean island of Tiree (Scotland, UK).

2. METHODS

Dog selection and training were carried out by the Defence Animals Centre at Melton Mowbray, UK, an organisation specializing in the training of drug detection dogs for the prison service and customs. They selected a two year old male English Springer-Spaniel for training which took place between April and June 2006. The techniques used for training and searching are well established and have been described elsewhere (e.g. Long et al., 2007; Helton, 2009). Initial training took place using nest material from a commercially reared *B. terrestris* nest, frozen at –30 °C until needed for training. Continuation training was subsequently performed daily.

2.1. Testing efficacy

In order to test the dog's ability to accurately detect bumblebee nests of different species in field conditions the following method was used. An area of 200 m × 50 m was selected and five round plastic pots (diameter 5 cm, height 3 cm) were buried at random locations within it. Each pot has 6 × 5 mm diameter holes drilled in the lid. The pots contained approximately 7 g of nest material from one of the following:

1. A commercial *B. terrestris* nest
2. A wild *B. terrestris* nest
3. A wild *B. muscorum* nest
4. A wild *B. distinguendus* nest

The fifth pot remained empty as a control. The pots were handled with gloves at all times and they were buried by a third party in the absence of dog or handler. The pots were buried at a depth of 10 cm using a proprietary bulb planter to remove a cylinder of turf and soil, which was then replaced on top of the pot leaving little visible sign of disturbance. Locations and treatments were randomly assigned within the search area. Pots were buried 24 h prior to testing.

The dog was then worked through the area using the standard search technique. This method was repeated at five different locations, in either grazed, short grassland or open woodland habitats, and all located in the Southampton area, southern UK. The number of positive finds, missed pots and false positive finds (either finding the control pot or indicating at inappropriate items) were recorded.

2.2. Locating real bumblebee nests and estimating nest density

During August and September 2006 the dog was used to search twelve 50 m by 500 m transects on the island of Tiree, off the west coast of Scotland. This island contains an unusually high diversity of bumblebee species. Each transect was randomly located within one of four habitat types, upland heath, lowland heath, sand dunes and machair. The latter is a rare habitat confined to west Scotland and Ireland, consisting of a flat coastal plain of species-rich grassland growing on wind-blown shell sand. These four habitats together comprise the majority of the island. The transects were searched using the standard search technique. When nests were located, a GPS reading was taken in order to accurately map their positions.

3. RESULTS

3.1. Testing efficacy of detecting nest material

A 100% efficacy rate was recorded; every pot containing bumblebee nest material was located (20 in total), regardless of the species from which the nest material had been obtained, and there were no false positives (the dog indicated at none of the control pots or at other objects within the search areas). A Fisher's exact test suggests that the success rate in avoiding false positives is unlikely to be due to chance ($P < 0.0001$).

3.2. Locating real bumblebee nests and estimating nest density

In total, the dog located 33 bumblebee nests belonging to four different bumblebee species in the twelve transects on Tiree (a total area of 30 ha) (Tab. I). The species most frequently found was *B. muscorum*, which is the most common species on the island and elsewhere in the Hebrides (Darvill et al., 2006; MacDonald and Nisbet, 2006). Other species found included (in order of declining frequency): *B. distinguendus*, *B. lapidarius* and *B. jonellus*. Numbers are too few for statistical analysis, but there are clear indications of habitat preferences. *B. muscorum* nests occurred in all four habitats searched, but nests were markedly more frequent on machair than elsewhere (14 of the 25 nests found). Sample sizes were small for the remaining species, but it is noteworthy that *B. lapidarius* nests were only recorded on machair and the adjacent dunes, and *B. distinguendus* nests were only found in dunes.

Nest density estimates for each species vary correspondingly; the highest mean density per habitat for *B. muscorum* was 1.86 nests/ha in machair, for *B. distinguendus* was 0.533 nests/ha in dunes, and for *B. lapidarius* was 0.267 nests/ha in machair. The only nest of *B. jonellus* was found in lowland heath (equating to 0.133 nests/ha).

4. DISCUSSION

The test of efficacy using buried nest material clearly indicates that the dog was able to locate bumblebee nest material with a high degree of accuracy, and without any false positives. He was also able to detect nest material from a range of bumblebee species, suggesting that the colonies of different species of bumblebee share a broadly similar smell, or that the particular cues used by the dog are common to all of the bumblebee species used. To our knowledge, there have been no attempts to quantify the chemical composition of the odour produced by bumblebee nests, which is likely to be highly complex. Since the species included in this study span diverse taxonomic lineages within the genus *Bombus* including both pocket-making and pollen-storing species (Cameron et al., 2007), it seems likely that the dog would be able to detect nests of any bumblebee species. To our knowledge the only other attempt to use a dog to detect nests of social insects was by Brooks et al. (2003) who trained dogs to find colonies of the termite *R. flavipes* and found that the dogs were also able to locate colonies of four additional termite species with no further training. It seems probable that detector dogs will generally tend to respond to species that are closely related to that on which they were trained.

When deployed to search for real bumblebee nests on Tiree, the dog proved to be successful, locating 33 wild nests from four species, including those of species such as *B. distinguendus* and *B. muscorum* which are rare and declining across much of Europe (Kosior et al., 2007; Goulson et al., 2008; Goulson, 2010). Thus this approach has obvious value for conservation-related studies of nesting habitat, nest survival etc

Tests of efficacy using buried nest samples suggest that the dog can be 100% effective in finding all of the nests in an area, but we do not know whether this is true when searching for real bumblebee nests. It is notable that some species known to be present on Tiree were not found (*B. ruderarius*, *B. lucorum* complex, *B. hortorum, B. pascuorum*). It is not clear whether this is because the dog did not detect their nests or simply because there were none present in the study areas. It may be that the nesting habits of different bumblebee species influence the ease with which nests can be detected. For example *B. lucorum* often nests deep underground (Sladen, 1912), which may make its nests harder to detect using olfactory cues. We also cannot be sure that there were not more nests of species such as *B. muscorum* that the dog failed to detect. Unfortunately no nest density estimates have ever been made for bumblebees on similar habitat, or anywhere in Scotland, so there are no direct comparisons to be drawn with the figures produced here.

Table I. Numbers of nests located within 50 × 500 m transects in four habitat types on Tiree, Scotland. Mean density per hectare per habitat is also shown.

Habitat/transect		*B. muscorum*	*B. lapidarius*	*B. distinguendus*	*B. jonellus*
Upland heath	1	2	0	0	0
	2	2	0	0	0
	3	0	0	0	0
Density/ha mean ± SE		0.533 ± 0.27	0	0	0
Lowland heath	1	1	0	0	1
	2	0	0	0	0
	3	0	0	0	0
Density/ha mean ± SE		0.133 ± 0.13	0	0	0.133 ± 0.13
Machair	1	0	2	0	0
	2	10	0	0	0
	3	4	0	0	0
Density/ha mean ± SE		1.86 ± 1.16	0.267 ± 0.26	0	0
Dune	1	5	0	3	0
	2	0	1	1	0
	3	1	0	0	0
Density/ha mean ± SE		0.80 ± 0.61	0.133 ± 0.13	0.533 ± 0.35	0

Nonetheless it is informative to compare these densities with those found elsewhere.

Molecular markers have been used to asign workers to sisterhoods and so estimate the number of nests that are represented in samples of workers from particular sites. If multiple nearby sites are included, this approach can also provide estimates of foraging range from the dispersion of sisters across the landscape, and in combination these two measures can be used to estimate nest density. This has only been attempted in lowland sites in mainland England, and both studies took place in July. Darvill et al. (2004) produced estimates of 0.13 nests/ ha for *B. pascuorum* and 1.93 nests/ha for *B. terrestris*. Knight et al. (2005) estimated nest densities of 0.678 nests/ ha for *B. pascuorum*, 0.287 nests/ha for *B. terrestris*, 1.17 nests/ha for *B. lapidarius* and 0.261 nests/ha for *B. pratorum* (2.40 nests/ha for all four species combined). More recently, Osborne et al. (2008) used a markedly different approach. They recruited 719 volunteers from across the UK to search for nests in their gardens and in farmland habitats by 20 minute observations of fixed areas; the premise being that nest traffic would be noticed within this period of time. They estimated that there were approximately 7 bumblebee nests/ha for all bumblebee species combined in June and early July. We would expect the density of nests to fall through the season as some nests succumb to predation or disease.

These estimates of nest density for lowland England are of a similar magnitude to our own for Tiree (summed for all bee species, our nest density estimates are 0.533, 0.267, 2.13 and 1.47 for upland heath, lowland heath, machair and dunes, respectively). It is notable that machair is by far the most floristically rich of these four habitats, which presumably explains the higher density of mature bumblebee nests. Our study was conducted at the end of the nest cycle (August–September), so it is likely that most or all of the nests detected were likely to produce reproductives. We tentatively conclude that the detector dog may well have found most of the nests in the study area, although molecular studies to estimate nest density on Tiree would be very useful to enable a direct comparison.

If the dog is able to find most nests, then this approach could be used to estimate effective population size for rare bumblebee species existing in habitat islands. For example, this study suggests that *B. distinguendus* nests primarily in dunes on Tiree. Multiplying the area of dunes by our nest density estimate could thus produce a crude estimate of the number of nests and hence of the number of breeding females on the island. The National Vegetation Classification (NVC) provides maps of habitat type for the whole of the UK and could be the basis for predicting the density and range of species for a given area.

One caveat must be noted. Testing and field use of our detector dog was all carried out in open habitats. Dense vegetation (hedgerows, shrubs, bramble thickets etc., none of which occur to any significant extent on Tiree) is likely to be far harder for the dog to search, yet is likely to include many of the sites chosen by bumblebees to nest in lowland Europe. This is an aspect of the use of a sniffer dog to detect bumblebee nests which we will address in future studies.

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Essai de localisation des colonies de bourdons et d'estimation de la densité des nids à l'aide d'un chien dressé à la recherche.

***Bombus distinguendus* / *Bombus muscorum* / densité des nids / odeur / méthode de détection / distribution spatiale / îles Hébrides / Écosse**

Zusammenfassung – Ein Test zur Eignung von Spürhunden beim Auffinden von Hummelvölkern und bei der Beurteilung der Nestdichte. Hummelnester sind bekanntermaßen schwierig aufzuspüren und behindern dadurch ökologische Studien, insbesondere bei seltenen Arten, für die kaum Nestbeschreibungen existieren. Da die effektive Populationsgröße für Hummeln über die Nestdichte bestimmt wird, wäre die Möglichkeit zur Bestimmung der Nestdichte eine große Hilfe für Schutzmaßnahmen. Wir beschreiben hier das Training und die Tests mit einem Hund zum Auffinden von Hummelnestern. Der Hund wurde durch die britische Armee mit Nestmaterial von *B. terrestris* trainiert. Seine Erfolgsquote beim Auffinden von verstecktem Nestmaterial verschiedener Hummelarten lag bei 100 %, wobei keine falsch-positiven Fälle vorkamen. Dies lässt vermuten, dass der Hund in der Lage ist, den Duft für verschiedene Hummelarten zu generalisieren. Der Hund wurde dann eingesetzt um Hummelnester zu lokalisieren und um die Nestdichte in vier verschiedenen Habitaten auf der Insel Tiree, West-Schottland, zu quantifizieren. Diese Insel beherbergt mehrere besonders schutzwürdige Hummelarten. Der Hund fand 33 Nester, wobei die erfasste Nestdichte je nach Art und Habitat zwischen 0 und 1,86 Nestern pro Hektar schwankte. Unsere Schätzungen ähneln den einzigen bisher verfügbaren Angaben zu Nestdichten, die vom britischen Festland stammen und vermuten lassen, dass unser Hund einen sehr großen Teil der vorhandenen Nester entdeckt hat. Offensichtlich haben die Hummelarten eine ausgeprägte Präferenz für bestimmte Habitate, wobei die meisten Nester von *B. muscorum* in Machair-Böden vorkamen, während die Nester von *B. distinguendus* ausschließlich in Dünen gefunden wurden. Wir schließen aus unseren Tests, dass diese Erfassungsmethode großes Potential besitzt, dass aber ein Hund in Habitaten mit dichterer Vegetation eventuell weniger erfolgreich ist.

Nestdichte / Nestgeruch / Hebriden / *Bombus distinguendus* / *Bombus muscorum*

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