Acta Chiropterologica, 22(2): 257–269, 2020
PL ISSN 1508-1109 © Museum and Institute of Zoology PAS
doi: 10.3161/15081109ACC2020.22.2.003

Moths consumed by the barbastelle *Barbastella barbastellus* require larval host plants that occur within the bat's foraging habitats

Andrew Carr[1, 4], Andrew Weatherall[2], Penelope Fialas[1], Matt R. K. Zeale[1], Elizabeth L. Clare[3], and Gareth Jones[1]

[1]*School of Biological Sciences, University of Bristol, Life Sciences Building, 24 Tyndall Avenue, Bristol, BS8 1TQ, United Kingdom*
[2]*National School of Forestry, University of Cumbria, Ambleside, Cumbria, LA22 9BB, United Kingdom*
[3]*School of Biological and Chemical Sciences, Queen Mary University of London, London, E1 4NS, United Kingdom*
[4]*Corresponding author: E-mail: andrew.carr@carrecology.uk*

Loss of foraging habitat and reductions in insect prey are key factors responsible for declines in bat populations. Identifying important bat foraging habitats and the ecological requirements and conservation status of prey provides evidence for appropriately targeted conservation management strategies. We examined the foraging habits of the barbastelle, *Barbastella barbastellus*, at the northern edge of its European range using a combination of radio tracking, to determine home range use and habitat selection, and DNA metabarcoding, to identify prey items consumed. Riparian vegetation and broadleaved woodland were the habitat types most strongly selected by foraging bats. Hedgerows within pastoral habitats were also important foraging features within the landscape. We identified 120 different prey items within faecal pellets, the majority of which were moths (97.5%). The large majority (97%) of prey items consumed have larval stages dependent on host plants that are typically found within habitats selected by foraging *B. barbastellus*. Almost half of all moth prey species identified have undergone considerable population declines in recent decades. We present the most comprehensive list to date of prey consumed by *B. barbastellus* and provide evidence that conservation management policies should target the protection and enhancement of key *B. barbastellus* foraging habitats within a 6.5 km sustenance zone around maternity roost sites. Riparian habitat, broadleaved woodland and linear landscape features such as hedgerows should be managed to improve their value to foraging *B. barbastellus* as well as the developmental stages of their preferred moth prey.

Key words: conservation, bats, diet, radio tracking, trophic relationships

Introduction

Loss of foraging habitat and reductions in insect prey numbers have resulted in population declines of insectivorous bat species (Wickramasinghe *et al*., 2004; Rainho and Palmeirim, 2011; Muylaert *et al*., 2016). For a population of bats to remain stable or to increase in number, productive foraging habitats are required. Foraging requirements of insectivorous bats are complex and require detailed understanding. For example, foraging areas must (i) provide a suitable amount of prey, (ii) be structured to allow successful capture of those prey, and (iii) be within an area that bats can reach from roost sites, both in terms of distance and the ability of the bats to traverse the landscape (Schnitzler *et al*., 2003).

An animal with a specialised diet may be negatively affected by a reduction in a single or small number of prey species. Investigating the conservation status and population trends of an animal's prey can identify current or perceived future impacts of changes in food resources. Furthermore, the prey of insectivorous bats may have more than one life stage (i.e. via metamorphosis) and their habitat requirements may differ during each life stage. For example, the Mediterranean horseshoe bat (*Rhinolophus euryale*) consumes adult moths, many of which have larvae that occupy habitats outside of the bats' primary foraging habitats (Arrizabalaga-Escudero *et al*., 2015), and differential use of larvae and adult stages of aquatic insects occurs in two species of trawling *Myotis* bats (Krüger *et al*., 2014). Hence, research on habitat use of life stages of prey items that are not necessarily eaten by bats, but which are vital for the viability and productivity of the prey species,

is potentially important (Arrizabalaga-Escudero *et al*., 2015).

DNA barcoding (Herbert *et al*., 2003), combined with high-throughput sequencing, has been successfully used to identify consumed prey from bat faecal pellets with greater resolution than is possible using traditional microscopic diet analysis (Razgour *et al*., 2011; Hope *et al*., 2014; Salinas-Ramos *et al*., 2015). It is possible to identify prey in digested bat faecal pellets to genus or species using molecular methods (Clare *et al*., 2009; Zeale *et al*., 2011), providing conservation managers with evidence to target the protection of specific prey species favoured by bats. Long-term monitoring of lepidopteran species in Britain has allowed monitoring of population trends (Conrad *et al*., 2004; Fox *et al*., 2013; Macgregor *et al*., 2019). Although large proportions of moth species are experiencing severe population declines, some are increasing in numbers (Fox *et al*., 2013).

The barbastelle, *Barbastella barbastellus* (Schreber, 1774), is an insectivorous bat with population declines throughout its known distribution (Piraccini, 2016). *Barbastella barbastellus* has echolocation adaptations and hunting strategies that are highly specialised (Goerlitz *et al*., 2010; Zeale *et al*., 2011). The species consumes adult moths captured on the wing using stealth echolocation and is considered to have a narrow dietary breadth relative to other insectivorous bats (Goerlitz *et al*., 2010; Zeale *et al*., 2011). When foraging, the bat appears to target habitats associated with abundant moth prey, such as riparian vegetation and broadleaved woodland (Zeale *et al*., 2012; Ancillotto *et al*., 2015; T. Kokurewicz, G. Apoznański, S. Pettersson, S. Sánchez-Navarro, and J. Rydell, in lit.) and can travel up to 20 km to exploit these habitats (Zeale *et al*., 2012). Although *B. barbastellus* typically expresses high inter-annual fidelity to roosting sites, returning to the same breeding site each year (Hillen *et al*., 2009), variation in selected foraging patches within and between seasons is apparent, possibly as a response to changes in prey availability (Greenaway, 2008).

Although the foraging behaviour of *B. barbastellus* has been explored in several studies (Hillen *et al*., 2009; Zeale *et al*., 2012; T. Kokurewicz, G. Apoznański, S. Pettersson, S. Sánchez-Navarro, and J. Rydell, in lit.) consideration of diet and conservation status of their insect prey within a foraging context has not. *B. barbastellus* feeds largely on moths as does the Mediterranean horseshoe bat (*R. euryale*). Some *R. euryale* prey species are known to have larval habitat requirements outside the bats' foraging habitat (Arrizabalaga-Escudero *et al*., 2015). Whether this is true for *B. barbastellus* prey is unknown.

Using a combination of radio tracking of bats and DNA metabarcoding of prey sequences extracted from bat faeces, we test the hypothesis that *B. barbastellus* feeds on prey whose larval food plants occur primarily within bats' core foraging areas. We identify if those prey are experiencing long-term population declines, which is a threat to *B. barbastellus* conservation. We provide new insights into the diet of *B. barbastellus* and new evidence to enhance practical conservation measures.

Materials and Methods

Data Collection

Bats were captured at six woodland sites located throughout southern and eastern England (Supplementary Table S1), between April 2014 and September 2015, using a combination of mist nets, harp traps and acoustic lures [see Carr *et al*. (2018) for a detailed procedure]. Ranging and foraging behaviour were studied by radio tracking at one of these sites, in Devon (3°43'35''W, 50°35'41''N), between May and July 2015. Bats were captured and radio tagged under licence from Natural England (2014/SCI/0429 and 2016-20013-SCI-SCI-1). Faecal pellets were collected at all six trapping sites by placing caught bats in sterilised hessian bags until they defecated (< 30 minutes). One faecal pellet from each bat was placed in a sterile collection tube, dampened with 96% ethanol, labelled and stored at -18°C. We obtained additional faecal pellets from the floor below a maternity roost inside an uninhabited barn in Norfolk (0°04'31"W, 51°59'34"N). These samples were collected monthly, between April and August 2015, and within 24 hours of being deposited by bats in the roost. To minimise the chance of pellets being from other bat species any old pellets were swept from the floor before a white collection sheet was placed on the ground beneath the colony. Pellet collections were made by a surveyor who has in-depth knowledge of this *B. barbastellus* maternity colony (Jane Harris), and was confident that the droppings were from the target species by monitoring their location within the barn during each collection period.

Radio tracked bats were fitted with lightweight radio transmitter tags [Pip3, 0.45 g; Biotrack Ltd., Wareham, UK — see Carr *et al*. (2018) for a detailed procedure] and tracked continuously throughout the night using the homing in method with r-1000 telemetry receivers (Communications Specialists Inc, Orange, USA) and three-element Yagi antennae (Wildlife Materials Inc, Murphysboro, USA — White and Garrot, 1990; Jones and Morton, 1992; Duvergé, 1996). A bat's position was recorded every five to ten minutes throughout the night to ensure the full nightly movements of the bat was recorded. Any night on which a signal was lost for a period of time that resulted in less than 95% contact time with the bat was removed from analysis. Bat fix locations were estimated using surveyor location, distance of signal (gain) and the direction of peak

signal from the surveyor. Activity type was determined by the nature of the signal. A rapidly moving directional signal was classified as a commuting bat, a fluctuating signal within a defined area was classified as a foraging bat and a static persistent signal was classified as a stationary roosting bat (Russo *et al*., 2004; Zeale *et al*., 2012).

Analysis of Foraging Behaviour

We digitised radio tracking fixes using Quantum GIS software 2.8.1 (Quantum GIS Development Team 2015, Wien) and the distance/azimuth Python plug-in 0.9.1 (Paulo and Laplante, Technology One). Digitised fixes were analysed using Ranges 7 (Anatrack Ltd., Dorset, UK) to calculate minimum convex polygon (MCP) home range areas and cluster core foraging areas (Zeale *et al*., 2016). We used 100% MCPs to represent the total area covered by individual bats, using all fixes obtained from the bat concerned. The colony home range area was represented by an 100% MCP using all fixes from all tracked bats. Foraging areas were defined using a cluster analysis of fix locations for each individual bat (Zeale *et al*., 2016). Analysis of utilisation distribution discontinuities in intervals of 5% found that 10% of fixes from each bat disproportionately increased the range size. Examination of these fixes identified that they were predominantly from bats categorised as commuting. Ninety percent cluster cores were therefore used to describe core foraging areas.

We examined habitat selection by comparing the composition of habitats within areas used for foraging (cluster cores) with the composition of habitats available to bats (individual MCPs). Compositional analysis was used to determine whether habitats were used in proportion to availability, or if selection was occurring, and to determine the ranking of habitat types (Compositional Analysis Plus Microsoft Excel Tool 6.2 — Smith Ecology Ltd., Wales, UK) (Boitani and Fuller, 2001; Zeale *et al*., 2016). To satisfy the criteria that the number of habitat categories be one less than the number of animals tracked (Aebischer *et al*., 1993), six broad habitat types were designated, whereby each habitat type comprised one or more Phase 1 habitat type. Phase 1 habitat categories are used within the UK for assigning habitat categories in accordance with guidance provided by the Joint Nature Conservation Committee (JNCC, 2010).

The six habitat types included in compositional analysis were: (i) Arable (J1.1) — cropland, horticultural land, freshly-ploughed land and recently reseeded grassland; (ii) Moorland (D) — vegetation dominated by ericoids or dwarf gorse species, as well as 'heaths' dominated by lichens and bryophytes, dwarf forbs, *Carex bigelowii* or *Juncus trifidus*; (iii) Pasture (B1-4 and J1.2) — improved, unimproved and semi-improved acidic, neutral and calcareous grassland. Managed amenity grassland was also included; (iv) Urban (J3) — built-up areas including bare ground, buildings and caravan sites; (v) Wetland (F1, F2.1, E and G) — bog, swamp, marginal, inundation (all narrow strips of emergent vegetation occurring on the margins of watercourses) and open water habitat and; (vi) Woodland (A1-3) — broadleaved, coniferous and mixed woodland that was either semi-natural or plantation. Scrub woodland (seral or climax community).

We created digital habitat maps in QGIS using data provided by the Woodland Trust and by Defra (MAGIC map; DEFRA, London; updated in 2014), as well as aerial photographs dated between 2012 and 2016 (Google Earth, version 6.2.2.6613). Habitat maps were ground-validated in 2016.

Analysis of Faecal Pellets

Prey DNA was extracted from faecal pellets using a QIAamp DNA Stool Mini Kit (Cat No./ID: 51504 — Qiagen Ltd., UK) and amplified by Polymerase Chain Reaction (PCR) at Queen Mary University London (QMUL) using a standardised approach following the technique used by Salinas-Ramos *et al*. (2015). CO1 primers ZBJ-ArtF1c and ZBJ-ArtR2c (Zeale *et al*., 2011) were used for high-throughput sequencing modified with adaptors for Ion Torrent, as these primers have been shown to be effective at amplifying arthropod prey while avoiding amplification of non-target DNA such as bat, bacteria and fungi (Zeale *et al*., 2011).

The obtained molecular sequences were analysed using the Galaxy platform (Afgan *et al*., 2016). Reads were separated by forward and reverse MIDs (Multiplex Identifiers) with a maximum of two mismatches allowed (Salinas-Ramos *et al*., 2015). All sequences shorter than 147 base pairs (bp) or longer than 167 bp (target amplicon length was 157 bp) were filtered out. Reads were collapsed so that identical sequences are represented by a single haplotype. Parameter selection was based on relative effect for the sequencing platform (Clare *et al*., 2016).

To identify prey consumed, representative sequences of each haplotype were run through the Basic Local Alignment Search Tool (BLAST; http://blast.ncbi.nlm.nih.gov/Blast.cgi) using a bit score of 281 and a threshold of 98.5% sequence similarity for prey identification based on mean sequence divergence values estimated by Zeale *et al*. (2011) for the amplified COI region. We investigated a taxonomic hierarchy using MEGAN6 software (Huson *et al*., 2016). Even with this conservative approach some sequences were assigned to more than one taxon with equal sequence similarity. To address this problem, we adopted a procedure used by Razgour *et al*. (2011) and Hope *et al*. (2014) which includes additional criteria to create identification confidence levels as follows: 1. Solid match (> 98.5%) to one species — species-level assignment, or match (> 98.5%) to more than one species, all belonging to the same genus — genus-level assignment; 2. Match (> 98.5%) to more than one species belonging to different genera, only one of which was a UK species — species-level assignment to UK species; 3. Match (> 98.5%) to several species of different genera within the same family or to reference sequences only identified to the family-level — family level assignment.

A sequence assigned to species, genus or family level using the above criteria was documented as a prey item (i.e., a haplotype(s) confirmed to match a reference in a molecular database). Frequency of occurrence was calculated as the number of faecal pellets that contained a particular prey item (i.e., if a moth species was identified in eight faecal pellets it had a frequency of occurrence of 8. If a moth was identified in only one faecal pellet is was given a frequency of occurrence of 1).

Prey Larval Habitat, Conservation Status and Population Trends

To establish the source habitat requirements of prey larvae, we searched for the host plants of the larvae of identified prey moth species in the Natural History Museum (London)'s

database of the World's lepidopteran host plants (HOSTS) (Robinson *et al*., 2010), the UK Moths website (https://www.ukmoths. org.uk/) and reference books. The larvae of moth species were placed in categories adapted from Arrizabalaga-Escudero *et al*. (2015) as follows: 1. Foraging habitat: > 60% of the larval host plants appear in *B. barbastellus* foraging habitat; 2. Non-foraging habitat: > 60% of the larval host plants appear in *B. barbastellus* non-foraging habitat; 3. Generalists: where neither of the previous criteria is fulfilled, i.e. > 60% threshold was not reached.

Barbastella barbastellus foraging and non-foraging habitat was determined though compositional analysis, as described in the Analysis of Foraging Behaviour section, above. Foraging habitats were those most strongly selected by radio tracked bats. Non-foraging habitats were those most weakly selected by radio tracked bats.

The conservation statuses of consumed prey were identified using the IUCN Red List of Threatened Species (http://www.iucnredlist.org/). Population trends were determined using the Rothamsted Insect Survey by Rothamsted Research and Butterfly Conservation that uses an extensive Britain-wide network of trap data, operational since 1968 (Conrad *et al*., 2004; Fox *et al*., 2013). Where trend data were not available, a species was classified as 'data deficient' and not included in a category. Species were categorised as having population trends that have (i) significantly declined, (ii) significantly increased, or (iii) been stable between 1968 and 2007 (Fox *et al*., 2013). Only macro-moths (typical wingspan >20 mm) were analysed as data for most micromoth taxa are limited. All values of central tendency are provided as means ± SD, unless otherwise stated, depending on the distribution of data.

Results

Ranging Behaviour and Foraging Habitat Selection

Seven female *B. barbastellus* [including pregnant ($n = 3$), lactating ($n = 3$) and nulliparous ($n = 1$) bats] were radio tracked to foraging grounds for an average of 2.7 days (range = 2−3 days). The number of fixes per individual was 139 ± 38. Bats showed considerable variation in MCP size (56.9−1293.3 ha) and maximum range span (1.2−8.3 km) (Supplementary Table S2). Weather conditions during tracking nights were consistently mild and mostly dry; no extreme weather events occurred. Minimum night temperature remained above 5°C throughout the tracking period.

Core foraging areas represented only 5.8 ± 3.7% of MCP home range areas (Fig. 1 and Supplementary Table S2). Foraging areas were predominantly outside of the woodland in which bats roosted during the day, except for a pregnant female, which foraged almost exclusively within the roost (home) wood. Lactating females returned to the home wood throughout the night (range = 2−3 times per night). Pregnant bats returned less often (range = 1−2 times per night) and not on every night that they were tracked. Bats showed high fidelity to foraging sites over the period in which they were tracked ($\bar{x}$ = 6 days, range = 3−8 days).

The composition of habitats within core foraging areas (90% cluster cores) was significantly different from that available within home range areas (individual bat MCPs) (weighted mean Wilk's lambda = 0.0123, $\chi^2_5 = 30.78$, $P < 0.01$). Selection of habitats, in order of strongly to weakly selected, was wetland > woodland > pasture > arable > urban >>> moorland (where > was selected to that immediately following and where >>> shows significant selection between the two adjacent habitat categories) (Supplementary Table S3).

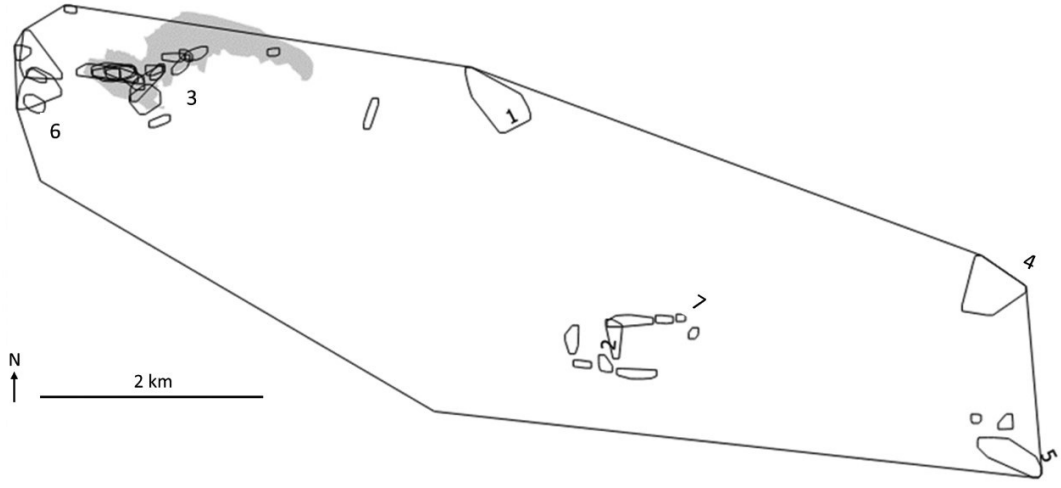

Fig. 1. Individual *B. barbastellus* core foraging areas (smaller polygons) and combined home range area (large delimiting polygon) for seven individual bats, radio tracked from the colony roost woodland (grey shaded polygon). Numbers correspond to a bat IDs (Supplementary Table S2)

Diet Composition

Prey DNA was successfully extracted from 33 faecal samples, including 28 from caught bats and five from collections made within the maternity roost in Norfolk. Faecal pellets obtained from caught bats included samples from male bats (n = 8) as well as pregnant (n = 6), lactating (n = 6), post lactating (n = 1) and non-breeding (n = 7) female bats. A total of 20,497 unique haplotypes were recovered from 33 samples (not including singletons). A representative of each unique haplotype was run through BLAST. The mean number of haplotypes per faecal pellet was 621 ± 381. Filtering (using a bit score of 281 or above and 98.5% sequence similarity to referenced sequences) identified 120 prey items (multiple haplotypes can match to a single prey item, some haplotypes had no match meeting this criteria) belonging to the orders Diptera (n = 2), Lepidoptera (n = 117), and Neuroptera (n = 1), from 18 families (Table 1 and Fig. 2). The number of prey items (i.e. a haplotype(s) confirmed to match a reference in a molecular database) per faecal pellet collected from captured bats (n = 28) was 5.9 ± 3.2 and from maternity colony samples (n = 5) was 10.6 ± 4.9. Most prey items appeared in only one or two faecal samples.

Most prey items (97.5 %) were moths. Dipteran flies and lacewings were identified although their frequency of occurrence within faecal pellets was low (n = 3). Moth prey varied in size from large macromoths (e.g. *Noctua pronuba*, wingspan 45−55 mm) to small micromoths (e.g. *Eudonia mercurella*, wingspan 16–19 mm) although a large proportion of moths belonged to families Geometridae and Noctuidae that are mostly medium-sized (wingspan 20−45 mm) (Scoble, 1992). Of the 117 confirmed moth species less than half were identified in more than one faecal pellet (n = 41). Prey items appearing in three or more faecal pellets (n = 21) comprised only 18% of prey items identified across all samples. Moth species frequently consumed (confirmed in ≥ five faecal pellets) included *N. pronuba* (n = 9 pellets), *Agrotis exclamationis* (n = 7 pellets), *Autographa gamma* (n = 6 pellets), *Eupithecia abbreviata* (n = 6 pellets), *Agrotis segetum* (n = 5 pellets), *Idaea biselata* (n = 5 pellets), *Phlogophora meticulosa* (n = 5 pellets), and *Xanthorhoe montanata* (n = 5 pellets).

Prey Larval Habitat

Only species within the order Lepidoptera (in this case only moths) were included. One hundred of the 117 Lepidoptera prey items that we identified

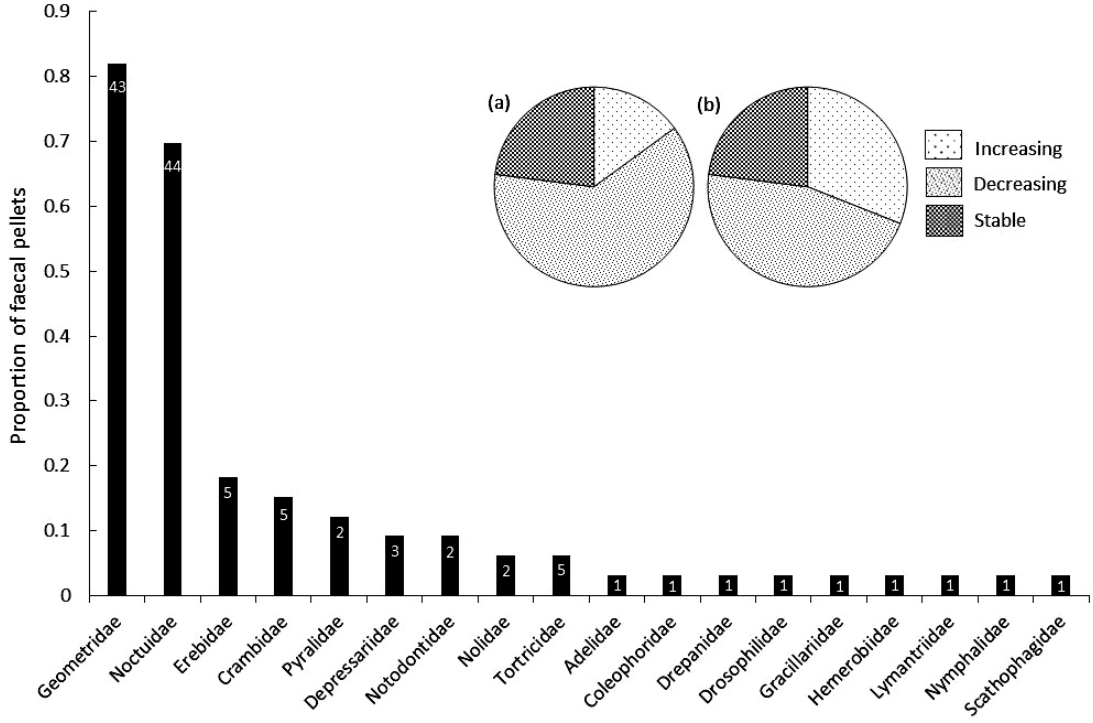

FIG. 2. Proportion of faecal pellets (n = 33) containing prey DNA from each of the 18 different insect families identified within the diet of *B. barbastellus*. Numbers within each column (white text) are the number of species within that family of the total confirmed prey species (n = 120); and population trends for (a) all monitored British macromoths (n = 1086) (Fox *et al*., 2013) and (b) all consumed *B. barbastellus* macromoth prey with population trend data (n = 75)

TABLE 1. List of prey (n = 120) identified in 33 *B. barbastellus* faecal samples using high-throughput molecular sequencing. Confidence levels follow Razgour *et al*. (2011) and Hope *et al*. (2014) based on the BOLD identification system, whereby confidence level 1 = solid match (> 98.5%) to one species or match (> 98.5%) to more than one species, all belonging to the same genus; level 2 = match (> 98.5%) to more than one species belonging to different genera, only one of which was a UK species; and level 3 = match to several species of different genera, or to reference sequences only identified to family (> 98.5%). Presence of ears (†) indicates prey items from families known to possess hearing-based defences against echolocating bats (Roeder, 1974; Fullard, 1982; Scoble, 1992). Sequence similarity scores are rounded to the nearest decimal point

Order/Family	Species	Confidence level	Sequence similarity	Frequency of occurrence (n faecal pellets)
Lepidoptera				
Adelidae	*Nematopogon swammerdamella*	1	100	1
Coleophoridae	*Coleophora kuehnella*	1	99.6	1
Crambidae†	*Eudonia lacustrata*	2	99	2
	E. mercurella	1	99.6	2
	Eurrhypara hortulata	1	100	1
	Scoparia basistrigalis	1	99	3
	Udea lutealis	1	100	1
Depressariidae	*Agonopterix heracliana*	1	99.6	1
	Carcina quercana	1	100	1
	Depressaria pastinacella	1	100	1
Drepanidae†	*Polyploca ridens*	1	100	1
	Thyatira batis	1	100	1
Erebidae	*Herminia grisealis*	1	99.6	2
	H. tarsipennalis	1	100	1
	Lymantria monacha	1	100	1
	Scoliopteryx libatrix	1	100	2
	Spilarctia luteum	1	100	1
Geometridae†	*Aethalura punctulata*	1	100	1
	Alcis repandata	1	99.6	1
	Apocheima pilosaria	1	100	1
	Asthena albulata	1	100	2
	Biston betularia	2	99	1
	Cabera pusaria	1	100	2
	Catarhoe rubidata	2	99	1
	Chloroclysta miata	2	99	1
	C. v-ata	1	99.6	1
	Cosmorhoe ocellata	1	100	1
	Cyclophora annularia	1	99	1
	Dysstroma truncata	1	100	3
	Ectropis crepuscularia	1	100	1
	Electrophaes corylata	1	99.6	1
	Epirrhoe alternata	1	99.6	4
	Eulithis prunata	1	100	1
	Eupithecia abbreviata	1	99	6
	E. absinthiata	1	99.6	1
	E. exiguata	1	99.6	1
	E. plumbeolata	2	99	1
	E. subfuscata	1	100	1
	E. vulgata	1	99	1
	Gymnoscelis rufifasciata	1	100	2
	Hemithea aestivaria	1	100	1
	Hydriomena furcata	1	100	1
	H. impluviata	1	100	1
	Idaea aversata	1	99.6	2
	I. biselata	1	100	5
	Lampropteryx suffumata	1	99	1
	Lomaspilis marginata	1	100	2
	Lomographa bimaculata	1	100	1
	Odontopera bidentata	1	100	4
	Opisthograptis luteolata	1	100	2
	Peribatodes rhomboidaria	2	99	1
	Perizoma affinitatum	2	99	1
	Petrophora chlorosata	1	99	2
	Scopula floslactata	1	100	2
	Selenia dentaria	1	99	3
	Trichopteryx carpinata	1	100	1
	Xanthorhoe designata	1	99	1

TABLE 1. Continued

Order/Family	Species	Confidence level	Sequence similarity	Frequency of occurrence (*n* faecal pellets)
	X. fluctuata	1	99	2
	X. montanata	1	100	5
Gracillariidae	*Cameraria ohridella*	2	99.6	1
Lymantriidae†	*Euproctis similis*	1	100	1
Noctuidae†	*Abrostola tripartita*	1	99.6	2
	Agrotis exclamationis	1	100	7
	A. ipsilon	1	99	1
	A. segetum	1	99.6	5
	Amphipyra pyramidea	1	100	3
	Anaplectoides prasina	1	99	1
	Apamea monoglypha	1	99	4
	A. sordens	1	99.6	1
	Atethmia centrago	1	100	1
	Autographa gamma	1	100	6
	Caradrina clavipalpis	1	100	1
	C. kadenii	2	99.6	1
	C. morpheus	1	100	1
	Cerastis leucographa	1	99.6	1
	C. rubricosa	1	99	1
	Charanyca trigrammica	1	100	1
	Conistra vaccinii	1	99	1
	Cosmia trapezina	1	100	3
	Cryphia domestica	1	99.6	1
	Cucullia chamomillae	1	100	2
	Diarsia rubi	1	99	1
	Hoplodrina ambigua	1	100	3
	H. blanda	1	99.6	1
	H. octogenaria	1	100	1
	Hypena proboscidalis	1	100	1
	Lithophane socia	1	99	1
	Luperina testacea	1	99.6	1
	Mamestra brassicae	1	100	1
	Mesapamea secalis/didyma	3	99	2
	Mythimna impura	2	99.6	2
	Noctua comes	1	100	1
	N. fimbriata	1	100	1
	N. janthe	1	100	2
	N. pronuba	1	100	9
	Ochropleura plecta	1	99.6	3
	Oligia strigilis/versicolor	3	99.6	1
	Orthosia gothica	1	99.6	1
	O. gracilis	1	99.6	1
	O. incerta	2	99	2
	Phlogophora meticulosa	2	99	5
	Rivula sericealis	1	99.6	1
	Spaelotis ravida	1	99.6	1
	Xestia c-nigrum	1	100	3
	X. xanthographa	1	100	3
Nolidae	*Nola confusalis*	1	99.6	1
	Nycteola revayana	1	100	1
Notodontidae†	*Notodonta ziczac*	1	99.6	1
	Pheosia gnoma	1	100	2
Nymphalidae	*Maniola jurtina*	1	100	1
Pyralidae†	*Cryptoblabes bistriga*	1	100	1
	Endotricha flammealis	1	100	3
Tortricidae†	*Archips podana*	2	99.6	1
	A. xylosteana	1	100	1
	Pammene fasciana	1	100	1
	Pandemis cerasana	1	99.6	1
	Pseudargyrotoza conwagana	1	99.6	1
Diptera				
Drosophilidae	*Drosophila melanogaster*	1	100	1
Scathophagidae	*Scathophaga stercoraria*	2	99	1
Neuroptera				
Hemerobiidae	Hemerobiidae spp.	3	100	1

were categorised as having their larval host plants in habitats associated with either (i) *B. barbastellus* foraging habitat, (ii) *B. barbastellus* non-foraging habitat or (iii) are considered generalists (Supplementary Table S4). The larvae of 17 species were not associated with a habitat due to having a food source other than plants (e.g. fungi, mosses, insects, leaf-litter). Most prey items (79%) were categorised as having a larval stage dependent on host plants that were within *B. barbastellus* foraging habitats (i.e. wetland, woodland and pastural habitats). Only 3% of prey items were found to feed on larval host plants located mainly outside *B. barbastellus* foraging grounds (i.e. arable, urban and moorland habitats). These included the flounced rustic (*Luperina testacea*) and large yellow underwing (*Noctua pronuba*) that feed on cultivated plants [although *N. pronuba* is known to commonly feed on wild plants and grasses (Robinson *et al*., 2010)], and the brimstone (*Opisthograptis luteolata*) that specialises on plum trees (*Prunus* spp.). For the remaining 18% of prey items, larval host plants are located within *B. barbastellus* foraging and non-foraging habitats and so the larvae were considered generalists. Of the 100 categorised prey items, 67% of the larvae were associated with host plants located in hedgerows.

Prey Conservation Status and Population Trends

Of the total number of moth species (n = 117) identified in faecal pellets, six were classified as Vulnerable by the IUCN Red List of Threatened Species (http://www.iucnredlist.org/) including, *Atethmia centrago*, *Caradrina morpheus*, *Diarsia rubi*, *Hoplodrina blanda*, *Orthosia gracilis* and *Spilarctia luteum*, with each showing population reductions by more than 70% between 1968 and 2007 (Fox *et al*., 2013). In contrast, seven species including, *Abrostola tripartita*, *Chloroclystis v-ata*, *Lymantria monacha*, *Noctua pronuba*, *Noctua janthe*, *Nola confusalis* and *Trichopteryx carpinata* increased in abundance by more than 100% between the same period (Fox *et al*., 2013).

Seventy-five *B. barbastellus* moth prey species have had their population trends monitored since 1968 (Fox *et al*., 2013; Macgregor *et al*., 2019). Of these 31% have significantly increased, 46% have significantly decreased, and 23% have stable populations. Moths with increasing population trends were more frequent in the diet of *B. barbastellus* (31%) than in all monitored macromoths (15%). Moths with decreasing population trends were less frequent in the diet of *B. barbastellus* (47%) than in all monitored macromoths (62%). Most prey species consumed, belonged to the families Geometridae and Noctuidae (Fig. 2).

Discussion

Ranging Behaviour and Foraging Habitat Selection

Radio tracked bats showed considerable variation in individual minimum convex polygon (MCP) areas and foraging distances. This agrees with other studies on *B. barbastellus* (Hillen *et al*., 2009; Zeale *et al*., 2012; Ancillotto *et al*., 2015; T. Kokurewicz, G. Apoznański, S. Pettersson, S. Sánchez-Navarro, and J. Rydell, in lit.) and highlights (i) the likelihood of needing a large core sustenance zone (6.5 km radius) around maternity sites, relative to other bats species (Bat Conservation Trust, 2016), and (ii) the ability of *B. barbastellus* to travel relatively long distances from roosting sites to foraging grounds on occasion (Russo *et al*., 2010; Zeale *et al*., 2012) and is likely influenced by the distance of productive foraging grounds (Ancillotto *et al*., 2015).

Although variable, we found that MCP size was smaller than documented in other radio tracking studies of *B. barbastellus* (Hillen *et al*., 2009; Zeale *et al*., 2012; T. Kokurewicz, G. Apoznański, S. Pettersson, S. Sánchez-Navarro, and J. Rydell, in lit.), which included or exclusively involved post-lactating females and male bats. Our study mainly included pregnant and lactating bats, which is most likely the reason for the comparatively small size of individual MCPs, although availability of preferred foraging habitats (Ancillotto *et al*., 2015) and seasonal differences in prey abundance may also influence findings. High wing loading during pregnancy and the requirement among lactating bats to return to roost sites repeatedly during the night to suckle pups probably constrain range spans during these periods (Henry *et al*., 2002; Womack *et al*., 2013). Despite differences in range sizes among studies, our results demonstrate that adult female *B. barbastellus* can travel long distances even under periods of reproductive stress and appear to have limited barriers to movement when traversing a landscape, suggesting that the species is able to exploit fragmented landscapes. This contrasts with less mobile species such as *Myotis bechsteinii* that rarely cross barriers in the landscape, such as roads (Kerth and Melber, 2008).

Core foraging areas represented a small fraction of home range areas, and there was little overlap of

foraging areas among individual bats. This spatial organisation of foraging areas in the landscape appears to be common among colonies of *B. barbastellus* throughout Europe (Hillen *et al*., 2009; Zeale *et al*., 2012; Ancillotto *et al*., 2014). We found wetland, woodland and pasture habitat types were favoured over others and that selection of foraging habitat type seems to be consistent throughout the breeding period (Hillen *et al*., 2009; Zeale *et al*., 2012). Wetland and woodland habitats are associated with high insect abundance, particularly riparian vegetation and broadleaved woodland, which support diverse populations of moths (Salsamendi *et al*., 2012; Highland *et al*., 2013; Ancillotto *et al*., 2014; Lintott *et al*., 2014). As such, the protection and restoration of bankside vegetation, wet meadows, wet woodland, and broadleaved woodland are considered important conservation measures for *B. barbastellus*.

Pasture, which was dominated by improved grassland, may contain relatively few potential prey yet the habitat was strongly selected for foraging. This can be explained by the presence of features at boundaries such as hedgerows providing suitable foraging structure (Zeale *et al*., 2012) and abundance of potential prey (Merckx *et al*., 2012). Bats that we radio tracked at these habitats were indeed observed foraging along hedgerows and minor tree lines.

Diet Composition

Given that a diverse range of moth species identified in the diet, both taxonomically and by body size, it seems appropriate to suggest that *B. barbastellus* probably consumes moth prey largely in line with availability rather than preferentially selecting certain moth species over others. Although this suggestion is in agreement with recent research that bats do consume prey following availability (Arrizabalaga-Escudero *et al*., 2019), all frequently consumed moths belonged to Geometridae and Noctuidae families. More research is required to test if *B. barbastellus* consumes prey in line with availability or targets prey with certain attributes such as size.

Barbastella barbastellus uses a stealth echolocation strategy to trump moth auditory defences, modulating the intensity of search-phase and approach calls to avoid eliciting evasive flight manoeuvres in moths (Goerlitz *et al*., 2010; Lewanzik and Goerlitz, 2018). As such, *B. barbastellus* is able to capture and consume tympanate moths more frequently than other sympatric aerial-hawking bat species (Goerlitz *et al*., 2010; Lewanzik and Goerlitz, 2018). We found that 74% of the insects consumed by *B. barbastellus* were in families known to have simple tympanic ears (Roeder, 1974; Fullard, 1982; Scoble, 1992).

The primers used to amplify DNA extracted from faecal pellets (Zeale *et al*., 2011) are arguably less suited to amplifying coleopteran DNA barcodes than those of other insect orders (Brandon-Mong *et al*., 2015). While this may bias findings in some diet studies, where insectivores consume beetles alongside other insect orders, traditional microscopic analyses of faecal material from *B. barbastellus* show presence of beetles in the diet is extremely rare (Rydell *et al*., 1996; Zeale *et al*., 2011). Indeed, undigested moth fragments dominate *B. barbastellus* faecal pellets when observed under a microscope, corroborating findings based on DNA-based approaches (Zeale *et al*., 2011). The method used to generate identifications of prey has been widely used in the literature but can result in false positive identifications at the species level given the small size of the fragment and taxonomic biases (Alberdi *et al*., 2017). While other regions and methods have been explored (Alberdi *et al*., 2017; Clare *et al*., 2019) we focus on this method to make our data comparable to other work on *B. barbastellus* (Goerlitz *et al*., 2010; Zeale, 2011) however we suggest that species level designations be treated as likely but with some caution.

Prey Larval Habitat

The importance of prey source habitat in bat conservation was studied at a breeding colony of *R. euryale* in the northern Iberian Peninsula (Arrizabalaga-Escudero *et al*., 2015). *Rhinolophus euryale* is a moth specialist and eats adult Lepidoptera which have larval host plants found mostly outside the bats' preferred foraging grounds, although seasonal variation was evident (Arrizabalaga-Escudero *et al*., 2015). In our study, we found that 79% of moth species consumed by *B. barbastellus* require larval host plants mainly within the species' preferred foraging habitats, and a further 18% of moth species were generalists (i.e. had larval host plants that are abundant in habitats used by foraging *B. barbastellus* and in habitats that they use rarely), leaving only 3% of consumed prey as mostly dependent on host plants within habitats not selected by foraging *B. barbastellus*. In contrast to *R. euryale*, conservation measures for *B. barbastellus* could

focus primarily on conserving and enhancing habitats selected as foraging sites by bats without the need to necessarily enhance other habitats occupied by the larval stages of some prey items. Importantly, the larvae of 67% of moth prey identified in faecal pellets feed on host plants found in hedgerows, reinforcing the value of targeting this habitat feature within conservation management plans for *B. barbastellus* and their prey.

An estimated loss of 50% of hedgerows within Britain since the 1940s (Robinson and Sutherland, 2002) is concerning as these marginal features are key habitats for bats and their insect prey. The plantation of new hedgerows in the agroecosystems of the European Union is encouraged by the Common Agricultural Policy (CAP) (European Commission, 2020). It is unclear whether incentives for land managers to retain and reinstate hedgerows will be in place when the UK is no longer an EU member state. Our findings demonstrate the importance of conserving hedgerows for the conservation of protected species.

Prey Conservation Status and Population Trends

Long-term monitoring (1968−2007) of British macromoths has shown that overall abundance has decreased by 28%, with the largest declines (up to 40%) recorded in the south of England (Fox *et al*., 2013). More than half of all monitored moths were found to be declining. Wing span has shown to be the best predictor for population status with larger moths more likely to be decreasing in number (Coulthard *et al*., 2019). We find *B. barbastellus* consumed mostly medium-sized moths (Geometridae and Noctuidae) that appear to be less likely to be in decline relative to large bodied moths (Coulthard *et al*., 2019).

Almost half of moth species consumed by *B. barbastellus* are declining in number throughout Britain. Given the considerable specialisation of *B. barbastellus* for feeding on moths, reductions in moth abundance are expected to have strong negative consequences for the species. In addition, more moth species are declining in southern, compared with northern Britain (Mattila *et al*., 2008; Fox *et al*., 2013). This is of concern as *B. barbastellus* distribution is restricted mostly to southern areas of England and Wales.

Conclusion

Our findings provide evidence that can be used to inform conservation management of *B. barbastellus* in fragmented landscapes, dominated by agricultural habitats. *Barbastella barbastellus* is highly dependent on moths and declines in moth prey populations within Britain and throughout Europe is cause for considerable concern. *B. barbastellus* consumes medium-sized moths most frequently that, as larvae and adults, require the same habitats as those selected by foraging bats. As such, creating, restoring and conserving habitats preferred by foraging *B. barbastellus* will help to conserve moth prey during all ontogenetic life stages. Conservation management should focus on the protection and enhancement of foraging habitats within a 6.5 km sustenance zone around maternity roost sites, including bankside vegetation, wet meadows, wet woodland, broadleaved woodland and marginal habitat such as hedgerows. Hedgerows may be particularly important as they can support a diverse range of flora, if managed sensitively, that are required as food plants by moth larvae.

Supplementary Information

Contents: Supplementary Tables: Table S1. List of key woodland characteristics of study sites 1–6 from which faecal pellets were taken from captured *B. barbastellus*; Table S2. List of radio tracked female *B. barbastellus* ($n = 7$) showing tracking start date, number of days tracked, reproductive status, maximum convex polygons (MCP), foraging cores, and mean maximum range travelled; Table S3. Simplified ranking matrix for radio tracked *B. barbastellus* ($n = 7$) comparing proportions of habitat within used habitat (90% cluster cores) and available habitat (maximum convex polygons (MCPs)) showing selection for each category on every row compared to the corresponding habitat in each column; Table S4. List of identified prey species confirmed within the faecal pellets of *B. barbastellus* showing prey host plant habitat types and Lepidoptera population trends since 1968. Supplementary Information is available exclusively on BioOne.

Acknowledgements

We thank Ian Davidson-Watts of Davidson-Watts Ecology and Jane Harris for providing bat faecal samples. We thank the many volunteers that participated in data collection. We thank Moth Broyles and Andy Wakefield for help with insect identification. This work was supported by the Scottish Forestry Trust (59 George Street, Edinburgh, UK), The Forestry Commission (231 Corstorphine Road, Edinburgh, UK), and the Woodland Trust (as part of the Moor than Meets the Eye Heritage Lottery Fund partnership) (Kempton Way, Grantham, UK).

Literature Cited

Aebischer, N. J., P. A. Robertson, and R. E. Kenward. 1993. Compositional analysis of habitat use from animal radio tracking. Ecology, 74: 1313–1325. http://dx.doi.org/10.2307/1940062.

Afgan, E., D. Baker, M. van den Beek, D. Blankenberg, D. Bouvier, M. Čech, J. Chilton, D. Clements, N. Coraor, C. Eberhard, *et al.* 2016. The Galaxy platform for accessible, reproducible and collaborative biomedical analyses: 2016 update. Nucleic Acids Research, 44: W3−W10. https://doi.org/10.1093/nar/gkw343.

Alberdi, A., O. Aizpurua, M. T. P. Gilbert, and K. Bohmann. 2017. Scrutinizing key steps for reliable metabarcoding of environmental samples. Methods in Ecology and Evolution, 2017: 1–14. http://doi.org/10.1111/2041-210X.12849.

Ancillotto, L., L. Cistrone, F. Mosconi, G. Jones, L. Boitani, and D. Russo, 2015. The importance of non-forest landscapes for the conservation of forest bats: lessons from barbastelles (*Barbastella barbastellus*). Biodiversity and Conservation, 24: 171–185. https://doi.org/10.1007/s10531-014-0802-7.

Arrizabalaga-Escudero, A., I. Garin, J. L. García-Mudarra, A. Alberdi, J., Aihartza, and U. Goiti. 2015. Trophic requirements beyond foraging habitats: the importance of prey source habitats in bat conservation. Biological Conservation, 191: 512–519. https://doi.org/10.1016/j.biocon.2015.07.043.

Arrizabalaga-Escudero, A., T. Merckx, G. García-Baquero, N. Wahlberg, O. Aizpurua, I. Garin, U. Goiti, and J. Aihartza. 2019. Trait-based functional dietary analysis provides a better insight into the foraging ecology of bats. Journal of Animal Ecology, 88: 1587–1600. https://doi.org/10.1111/1365-2656.13055.

Bat Conservation Trust. 2016. Core sustenance zones: determining zone size. Bat Conservation Trust, London, UK, 7 pp.

Boitani, L., and T. K. Fuller. 2001. Research techniques in animal ecology: controversies and consequences, Columbia University Press, New York, 464 pp.

Brandon-Mong, G. J., H. M. Gan, K. W. Sing, P. S. Lee, P. E. Lim, and J. J. Wilson. 2015. DNA metabarcoding of insects and allies: an evaluation of primers and pipelines. Bulletin of Entomological Research, 105: 717–727. https://doi.org/10.1017/S0007485315000681.

Carr, A., M. R. K. Zeale, A. Weatherall, J. S. P., Froidevaux, and G. Jones. 2018. Ground-based and LiDAR-derived measurements reveal scale-dependent selection of roost characteristics by the rare tree-dwelling bat *Barbastella barbastellus*. Forest Ecology and Management, 417: 237–246. https://doi.org/10.1016/j.foreco.2018.02.041.

Clare, E. L., E. E. Fraser, H. E. Braid, M. B. Fenton, and P. D. N. Hebert. 2009. Species on the menu of a generalist predator, the eastern red bat (*Lasiurus borealis*): using a molecular approach to detect arthropod prey. Molecular Ecology, 18: 2532–2542. https://doi.org/10.1111/j.1365-294X.2009.04184.x.

Clare, E. L., F. J. J. Chain, J. E. Littlefair, and M. E. Cristescu, 2016. The effects of parameter choice on defining molecular operational taxonomic units and resulting ecological analyses of metabarcoding data. Genome, 59: 981–990. https://doi.org/10.1139/gen-2015-0184.

Clare, E. L., A. J. Fazekas, N. V. Ivanova, R. M. Floyd, P. D. N. Hebert, A. M. Adams, J. Nagel, R. Girton, S. G. Newmaster, and M. B. Fenton. 2019. Approaches to integrating genetic data into ecological networks. Molecular Ecology, 2018: 1–17. https://doi.org/10.1111/mec.14941.

Conrad, K. F., I. P. Woiwod, M. Parsons, R. Fox, and M. S. Warren. 2004. Long-term population trends in widespread British moths. Journal of Insect Conservation, 8: 119–136. https://doi.org/10.1023/B:JICO.0000045810.36433.c6.

Coulthard, E., J. Norrey, C. Shortall, and W. E. Harris. 2019. Ecological traits predict population changes in moths. Biological Conservation, 233: 213–219. https://doi.org/10.1016/j.biocon.2019.02.023.

Duvergé, P. L. 1996. Foraging activity, habitat use, development of juveniles, and diet of the greater horseshoe bat (*Rhinolophus ferrumequinum* — Schreber 1774) in Southwest England. Ph.D. Thesis, University of Bristol, Bristol, UK, 310 pp.

European Commission. 2020. Common agricultural policy. European Commission. https://ec.europa.eu/info/food-farming-fisheries/key-policies/common-agricultural-policy_en. Accessed 24 May 2020.

Fox, R., M. Parsons, and J. Chapman. 2013. The state of Britain's larger moths 2013. Butterfly Conservation and Rothamsted Research, Wareham, UK, 32 pp.

Fullard, J. H. 1982. Echolocation assemblages and their effects on moth auditory systems. Canadian Journal Zoology, 60: 2572–2576. https://doi.org/10.1139/z82-330.

Goerlitz, H. R., H. M. Ter Hofstede, M. R. K. Zeale, G. Jones, and M. W. Holderied. 2010. An aerial-hawking bat uses stealth echolocation to counter moth hearing. Current Biology, 20: 1568–1572. https://doi.org/10.1016/j.cub.2010.07.046.

Greenaway, F. 2008. Barbastelle bats in the Sussex West Weald 1997–2008. Sussex Wildlife Trust, West Weald Landscape Partnership, Sussex, UK, 38 pp.

Henry, M. L., D. W. Thomas, R. Al Vaudry, and M. Carrier. 2002. Foraging distances and home range of pregnant and lactating little brown bats (*Myotis lucifugus*). Journal of Mammalogy, 83: 767–774. https://doi.org/10.1644/1545-1542(2002)083%3C0767:FDAHRO%3E2.0.CO;2.

Herbert, P. D., A. Cywinska, S. J. Ball, and J. R. deWaard. 2003. Biological identifications through DNA barcodes. Proceedings of the Royal Society Biological Science, 270B: 313–321. https://doi.org/10.1098/rspb.2002.2218.

Highland, S. A., J. C. Miller, and J. A. Jones. 2013. Determinants of moth diversity and community in a temperate mountain landscape: vegetation, topography, and seasonality. Ecosphere, 4: 129. https://doi.org/10.1890/ES12-00384.1.

Hillen, J., A. Kiefer, and M. Veith. 2009. Foraging site fidelity shapes the spatial organisation of a population of female western barbastelle bats. Biological Conservation, 142: 817–823. https://doi.org/10.1016/j.biocon.2008.12.017.

Hope, P. R., K. Bohmann, M. T. P., Gilbert, M. L. Zepeda-Mendoza, O., Razgour, and G. Jones. 2014. Second generation sequencing and morphological faecal analysis reveal unexpected foraging behaviour by *Myotis nattereri* (Chiroptera, Vespertilionidae) in winter. Frontiers in Zoology, 11: 39. https://doi.org/10.1186/1742-9994-11-39.

Huson, D. H., S. Beier, I. Flade, A. Górska, M. El-Hadidi, S. Mitra, H. J. Ruscheweyh, and R. Tappu. 2016. MEGAN community edition — interactive exploration and analysis of large-scale microbiome sequencing data. PLoS Computer Biology, 12: e1004957. https://doi.org/10.1371/ journal.pcbi.1004957.

JNCC [Joint Nature Conservation Committee]. 2010. Handbook for Phase 1 Habitat Survey: A technique for environmental audit. Joint Nature Conservation Committee, Peterborough, UK, 83 pp.

Jones, G., and M. Morton. 1992. Radio tracking studies and habitat use by greater horseshoe bats (*Rhinolophus ferrumequinum*). Pp. 521–537, *in* Wildlife telemetry: remote monitoring and tracking of animals (I. G. Priede and S. M. Swift, eds.). Ellis Horwood, Chichester, UK, 708 pp.

Kerth, G., and M. Melber. 2008. Species-specific barrier effects of a motorway on the habitat use of two threatened forest-living bat species. Biological Conservation, 142: 270–279. https://doi.org/10.1016/j.biocon.2008.10.022.

Krüger, F., E. L. Clare, S. Greif, B. M. Siemers, W. O. C. Symondson, and R. S. Sommer. 2014. An integrative approach to detect subtle trophic niche differentiation in the sympatric trawling bat species *Myotis dasycneme* and *Myotis daubentonii*. Molecular Ecology, 23: 3657–3671. https://doi.org/10.1111/mec.12512.

Lewanzik, D., and H. R. Goerlitz. 2018. Continued source level reduction during attack in the low- amplitude bat *Barbastella barbastellus* prevents moth evasive flight. Functional Ecology, 32: 1251–1261. https://doi.org/10.1111/1365-2435.13073.

Lintott, P. R., N. Bunnefeld, E. Fuentes-Montemayor, J. Minderman, L. M. Blackmore, D. Goulson, and K. J. Park. 2014. Moth species richness, abundance and diversity in fragmented urban woodlands: implications for conservation and management strategies. Biodiversity and Conservation, 23: 2875–2901. http://dx.doi.org/10.1007/s10531-014-0753-z.

Macgregor, C. J., J. H. Williams, J. R. Bell, and C. D. Thomas. 2019. Moth biomass increases and decreases over 50 years in Britain. Nature Ecology and Evolution, 3: 1645–1649. https://doi.org/10.1038/s41559-019-1028-6.

Mattila, N., J. S. Kotiaho, V. Kaitala, and A. Komonen. 2008. The use of ecological traits in extinction risk assessments: a case study on geometrid moths. Biological Conservation, 141: 2322–2328. https://doi.org/10.1016/j.biocon.2008.06.024.

Merckx, T., L. Marini, R. E. Feber, and D. W. Macdonald. 2012. Hedgerow trees and field margins enhance macromoth diversity: implication for management. Journal of Applied Ecology, 49: 1396–1404. http://dx.doi.org/10.1111%2Fj.1365-2664.2012.02211.x.

Muylaert, R. L., R. D. Stevens, and M. C. Ribeiro. 2016. Threshold effect of habitat loss on bat richness in cerrado-forest landscapes. Ecological Application, 26: 1854–1867. https://doi.org/10.1890/15-1757.1.

Piraccini, R. 2016. *Barbastella barbastellus*. The IUCN Red List of Threatened Species 2016. International Union for the Conservation of Nature. https://www.doi:10.2305/IUCN.UK.2016-2. Accessed 12 November 17.

Rainho, A., and J. M. Palmeirim. 2011. The importance of distance to resources in the spatial modelling of bat foraging habitat. PLoS ONE, 6: e19227. https://doi.org/10.1371/journal.pone.0019227.

Razgour, O., E. L. Clare, M. R. K. Zeale, J. Hanmer, I. B. Schnell, M. Rasmussen, T. P. Gilbert, and G. Jones. 2011. High-throughput sequencing offers insight into mechanisms of resource partitioning in cryptic bat species. Ecology and Evolution, 1: 556–570. https://dx.doi.org/10.1002%2Fece3.49.

Robinson, G. S., P. R. Ackery, I. J. Kitching, G. W. Beccaloni, and L. M. Hernández. 2010. HOSTS: a database of the World's lepidopteran hostplants. Natural History Museum, London, UK. http://www.nhm.ac.uk/hosts. Accessed 15.05.2018.

Robinson, R., and W. J. Sutherland. 2002. Post-war changes in arable farming and biodiversity in Great Britain. Journal of Applied Ecology, 39: 157–176. https://doi.org/10.1046/j.1365-2664.2002.00695.x.

Roeder, K. D. 1974. Responses of the less sensitive acoustic sense cells in the tympanic organs of some noctuid and geometrid moths. Journal of Insect Physiology, 20: 55–66. https://doi.org/10.1016/0022-1910(74)90123-1.

Russo, D., L. Cistrone, G. Jones, and S. Mazzoleni. 2004. Roost selection by barbastelle bats (*Barbastella barbastellus*, Chiroptera: Vespertilionidae) in beech woodlands of central Italy: consequences for conservation. Biological Conservation, 117: 73–81. https://doi.org/10.1016/S0006-3207(03)00266-0.

Russo, D., L. Cistrone, A. P. Garonna, and G. Jones. 2010. Reconsidering the importance of harvested woodlands for the conservation of tree-dwelling bats. Biodiversity and Conservation, 19: 2501–2515. http://dx.doi.org/10.1007/s10531-010-9856-3.

Rydell, J., G. Natuschke, A. Theiler, and P. E. Zingg. 1996. Food habits of the barbastelle bat *Barbastella barbastellus*. Ecography, 19: 62–66. https://doi.org/10.1111/j.1600-0587.1996.tb00155.x.

Salinas-Ramos, V. B., L. G. Herrera Montalvo, V. León-Regagnon, A. Arrizabalaga-Escudero, and E. L. Clare. 2015. Dietary overlap and seasonality in three species of mormoopid bats from a tropical dry forest. Molecular Ecology, 24: 5296–5307. https://doi.org/10.1111/mec.13386.

Salsamendi, E., I. Arostegui, J. Aihartza, D. Almenar, U. Goiti, and I. Garin. 2012. Foraging ecology in Mehely's horseshoe bats: influence of habitat structure and water availability. Acta Chiropterologica, 14: 121–132. https://doi.org/10.3161/150811012X654330.

Schnitzler, H.-U., C. F. Moss, and A. Denzinger. 2003. From spatial orientation to food acquisition in echolocating bats. Trends in Ecology & Evolution, 18: 386–394. https://doi.org/10.1016/S0169-5347(03)00185-X.

Scoble, M. J. 1992. The Lepidoptera. Form, function and diversity. Oxford University Press, Oxford, UK, 404 pp.

White, G. C., and G. C. Garrott. 1990. Analysis of wildlife radio tracking data. Academic Press, San Diego, California, USA, 383 pp.

Wickramasinghe, L. P., S. Harris, G. Jones, and N. V. Jennings. 2004. Abundance and species richness of nocturnal insects on organic and conventional farms: effects of agricultural intensification on bat foraging. Conservation Biology, 18: 1283–1292. https://doi.org/10.1111/j.1523-1739. 2004.00152.x.

Womack, K. M., S. K. Amelon, and F. R. Thompson. 2013. Summer home range size of female Indiana bats (*Myotis Sodalis*) in Missouri, USA. Acta Chiropterologica, 15: 423–429. https://doi.org/10.3161/150811013X679044.

Zeale, M. R. K. 2011. Conservation biology of the barbastelle (*Barbastella barbastellus*): applications of spatial modelling, ecology and molecular analysis of diet. Ph.D. Thesis, University of Bristol, Bristol, UK, 145 pp.

Zeale, M. R. K., R. K. Butlin, G. L. A. Barker, D. C. Lees, and G. Jones. 2011. Taxon-specific PCR for DNA barcoding arthropod prey in bat faeces. Molecular Ecology Resources, 11: 236–244. https://doi.org/10.1111/j.1755-0998.2010.02920.x.

Zeale, M. R. K., I. Davidson-Watts, and G. Jones. 2012. Home range use and habitat selection by barbastelle bats (*Barbastella barbastellus*): implications for conservation. Journal of Mammalogy, 93: 1110–1118. https://doi.org/10.1644/11-MAMM-A-366.1.

Zeale, M. R. K., E. Bennitt, S. E. Newson, C. Packman, W. J. Browne, S. Harris, G. Jones, and E. Stone. 2016. Mitigating the impact of bats in historic churches: the response of Natterer's bats *Myotis nattereri* to artificial roosts and deterrence. PLoS ONE, 11: e0146782. https://doi.org/10.1371/journal.pone.0146782.

Received 24 March 2020, accepted 24 July 2020

Associate Editor: Wiesław Bogdanowicz