Ecological niche modelling of two cryptic bat species calls for a reassessment of their conservation status

THOMAS SATTLER*, FABIO BONTADINA*, ALEXANDRE H. HIRZEL*†
and RAPHAËL ARLETTAZ*

Zoological Institute, Division of Conservation Biology, University of Bern, Baltzerstrasse 6, CH–3012 Bern, Switzerland; and †Department of Ecology and Evolution, University of Lausanne, CH–1015 Lausanne, Switzerland

Summary

1. It is difficult to establish conservation priorities for cryptic species when their ecological requirements are confounded by problems with species identification. In some cases, such as Chiroptera, cryptic taxa may actually consist of both widespread, abundant species and localized, rare species. Discrimination between these species may be facilitated by phenotypic, species-specific traits such as echolocation calls. Echolocation studies supported by genetic data have revealed that one of the most abundant bat species in Europe actually consists of two cryptic species: *Pipistrellus pipistrellus* and *P. pygmaeus*.

2. We recorded echolocation calls from both species along road transects in Switzerland to study their distribution and abundance. Using Ecological Niche Factor Analysis and discriminant analysis, we characterized species-specific habitat requirements, built habitat suitability maps and examined interspecific differences in niche parameters.

3. The presence of *P. pygmaeus* was associated with landscape matrices comprising large rivers and lakes, human settlements and open woodland. *P. pipistrellus* utilized similar habitat matrices but was far more tolerant to deviations from its optimal habitat. *P. pygmaeus* occupied a much narrower ecological niche, encompassed mainly within that of its sister taxon.

4. *Synthesis and applications. P. pipistrellus* is ranked as 'not threatened' in Switzerland. The results from this study indicate an abundance approximately 30 times higher than that of *P. pygmaeus*. In contrast, *P. pygmaeus* is distributed patchily and occurs at comparatively low densities. We recommend reclassification of *P. pygmaeus* as 'rare and potentially threatened'. Conservation of *P. pygmaeus* should focus on the management of riparian woodland in areas with a high probability of occurrence. This study emphasizes the need to recognize the potential existence of cryptic taxa so that effective conservation management of rare species can be put into place before they are seriously endangered.

Key-words: Boyce Index, Ecological Niche Factor Analysis (ENFA), habitat suitability, niche differentiation, *Pipistrellus pipistrellus*, *P. pygmaeus*, red list status, Switzerland.

Introduction

Species are classified into Red List categories indicating the risk of extinction on the basis of a number of criteria including population size, population trend and distribution range (IUCN 2001). Red Lists classify species into categories from 'least concerned' to 'extinct' on either global or regional scale. Defining the status of a species is crucial for prioritizing conservation action but appropriate data are often difficult to collect. Taxonomic confusion may complicate the task further: even in well-known orders such as mammals (Jones 1997) and amphibians (Vallan, Vences & Glaw 2003), new cryptic species are still being found, making it difficult to establish conservation status accurately.

Correspondence and current address: Thomas Sattler, Swiss Federal Research Institute WSL, Research Unit Ecosystem Boundaries, Zürcherstrasse 111, 8903 Birmensdorf, Switzerland (e-mail: thomassattler@gmx.net).

Cryptic species, although reproductively isolated, are morphologically alike (Mayr 1977). New molecular tools allow taxonomists to distinguish between cryptic species on the basis of genetic data. For instance, during the 10 years between the first two editions of *Mammals of the World* (Honacki, Kinmam & Koeppl 1982; Wilson & Reeder 1993) 459 new species have been described, of which 63% are based on molecular evidence (Medellín & Soberón 1999). Among lemurs of the genus *Microcebus*, the number of recognized species has doubled recently based on molecular analysis (Yoder *et al.* 2000).

The recognition of cryptic species within taxonomic groups may necessitate a complete reappraisal of conservation status. Furthermore, the failure to recognize cryptic species may have dramatic consequences: in a captive breeding programme for the highly endangered Brazilian monkeys *Brachyteles arachnoides* and *B. hypoxanthus*, the numerous hybrids obtained after reproduction in captivity could not be used for restocking natural populations (Brito 2004). In addition, once cryptic species are distinguished, there is still a need to collect basic ecological data in order to draw up appropriate conservation guidelines (Arlettaz 1999).

Cryptic species are numerous in the order Chiroptera (Mayer & von Helversen 1999; Hulva *et al.* 2004). Selective forces have led to acoustic divergence, with specializations arising for different foraging tactics, microhabitats and/or food types (Jones 1997). In Europe, where bats are well studied, the discovery of cryptic species has increased the number of recorded species from 30 to 39 in a few decades (Dietz & von Helversen 2004). Recently, additional species have been proposed to exist as cryptic species in Europe (Ibáñez *et al.* 2006; Mayer, Dietz & Kiefer 2007). One of the most astonishing cases of cryptic diversity is the pipistrelle bat complex *Pipistrellus pipistrellus* sensu lato (Schreber 1774). *P. pipistrellus* was once considered the most abundant and best-known bat species in Europe (Stebbings 1988; Jones 1999), but it actually consists of two cryptic species (Jones & van Parijs 1993; Barratt *et al.* 1997). Differences in search call frequency between two forms of sympatric pipistrelles (45 kHz vs. 55 kHz) had been recorded previously within this species complex (Zingg 1990), but it is only through refined ecological and genetic research that the existence of a second species, *P. pygmaeus* (Leach 1825), could be proved (Barlow 1997; Barlow & Jones 1997; Barratt *et al.* 1997; Vaughan, Jones & Harris 1997; Mayer & von Helversen 1999).

According to current knowledge, the distribution of *P. pipistrellus* ranges from the British Isles to North Africa and the Middle East (Jones 1999). The coexistence of *P. pygmaeus* is confirmed in many countries within these limits, but its abundance often remains unknown. Bat surveys in England and Ireland revealed that the two species occur at similar densities (Vaughan, Jones & Harris 1997; Russ & Montgomery 2002),

contrary to the situation in Central Europe where *P. pygmaeus* appears to be much more localized (Häussler *et al.* 2000; Wicht *et al.* 2003). The ecological reasons for this difference in density depending on geographical area are still poorly understood; new investigations of the ecological niche of the two pipistrelles are therefore needed, particularly outside the United Kingdom. The present study analyses the distribution of the two species in Switzerland and links their geographical pattern of occurrence with habitat features. We used Geographical Information Systems (GIS) and Ecological Niche Factor Analysis (ENFA) to prepare species-specific optimal habitat profiles, to compare respective ecological niche parameters and to predict habitat suitability on a nation-wide scale (Buckland & Elston 1993; Manel *et al.* 1999; Vayssières, Plant & Allen-Diaz 2000). We opted for the environmental envelope approach because absence of evidence cannot be equated with evidence of absence when recording bats (Guisan & Zimmermann 2000; Jaberg & Guisan 2001; Hirzel & Guisan 2002; Hirzel *et al.* 2002; Rushton, Ormerod & Kerby 2004). The main aims of this study were: (1) to assess the current range of the two cryptic species *P. pipistrellus* and *P. pygmaeus* in the six biogeographical regions of Switzerland and to draw predictive distribution maps; (2) to re-evaluate their national conservation status; and (3) to investigate their species-specific ecological habitat requirements as a basis for proposing tailored conservation guidance.

Methods

STUDY AREA

The study was carried out in Switzerland and Liechtenstein in an area of approximately 26 000 km². As the altitudinal distribution of reproductive *P. pipistrellus* in Central Europe is mainly below 1500 m (Arlettaz *et al.* 1997a), we did not consider higher elevations. Switzerland was divided into six biogeographical regions according to Gonseth *et al.* (2001; Fig. 1). The Jura Mountains are a hilly system under an oceanic climate regime (yearly precipitation of 1000–2000 mm). They are dominated mainly by beech *Fagus sylvatica* L. and coniferous forests interspersed with pastures. The relatively flat Swiss midlands (300–700 m a.s.l.) are heavily urbanized and industrialized, devoted to intensive agriculture (arable land in the West, dairy farming and meat production in the East), but harbours numerous lakes and rivers. Forests are of limited extent. The four bioregions in the Alps coincide with major watersheds. The southern Alps are warm and wet with cool winters. In the northern Alps, precipitation is high whereas the climate becomes drier in the central Alps in the interior. The western Alps are even warmer and drier, which allows intensive fruit production, including vineyards on the south-exposed slopes. Dairy farming dominates in the northern and eastern Alps, and the valley floors are
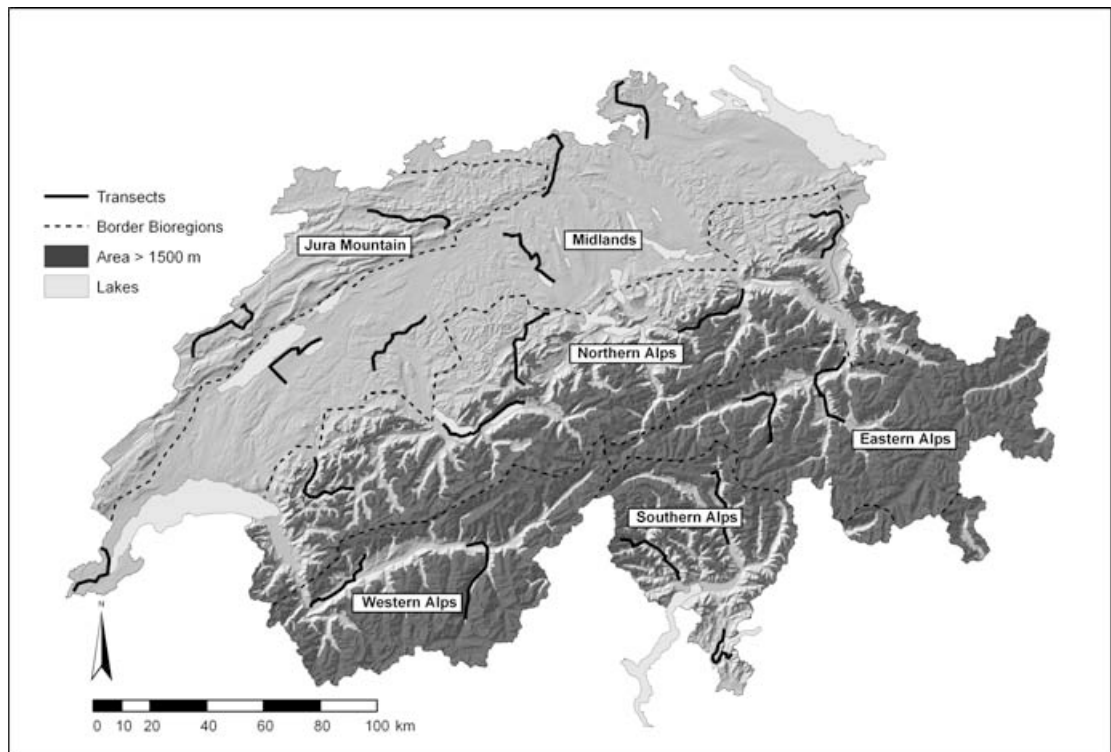
Fig. 1. Biogeographical map of Switzerland. Bright grey: area < 1500 m (= study area), dark grey: area > 1500 m. Dotted lines indicate boundaries of biogeographical regions. The 20 transects (continuous lines) were stratified to biogeographical region according to their fraction on total area of Switzerland < 1500 m: Jura Mountain (two transects), Midlands (six), Northern Alps (five), Western Alps (two), Southern Alps (three), Eastern Alps (two).

farmed intensively. The steep slopes are covered with forests, but meadows and pasture occur on plateaux and gentler slopes.

PRESENCE DATA COLLECTION

Both study species are insectivorous and forage in semicluttered areas. From June to August females aggregate to form maternity roosts, which are found mainly in crevices of buildings and below rooftops.

TRANSECTS

Bat presence was surveyed by recording echolocation calls from a vehicle along twenty 40-km-long road transects. The transects were selected following a stratified sampling assigned to the six biogeographical regions (Hirzel & Guisan 2002), with the proportion of transects in a given region depending on the area relative to the whole reference area (Switzerland) below 1500 m elevation (Fig. 1). Transects were chosen along 6-m-wide paved roads and mirrored a progressive altitudinal gradient, making sure that habitat types available locally below 1500 m altitude were represented. Each transect was surveyed twice, first during the breeding period (1 June–31 July 2002) and secondly during the post-breeding period (1 August–30 September 2002) so that a total of 1600 km was covered. A single transect per night was surveyed, starting 1 h after sunset. The timing was chosen to avoid bats commuting from

roosts and to concentrate on peak foraging activity (Barlow & Jones 1997). During echolocation recordings, vehicle speed was kept around 25 km h^{-1}. Survey nights were constrained by weather conditions (no rainfall, no strong wind, ambient temperature ≥ 10 C) and low moon intensity (no recording at full moon ± one night).

RECORDING OF ECHOLOCATION CALLS

Bats were recorded using a Pettersson D980 ultrasound detector (Pettersson Electronic AB, Uppsala, Sweden). Audio sequences of 3 s duration were recorded and time-expanded (10×) with a sampling frequency of 350 kHz (resolution: 8 bits) onto metal dioxide tapes using a Sony WM-D6C Stereo Cassette-Recorder (Sony, Tokyo, Japan). Geographic location of recordings was documented with a Global Positioning System (Garmin eTrex, Olathe, KA, USA). To avoid recording the same individual bat twice, we ignored subsequent bat calls at the same, species-specific frequency of maximum energy (FMAXE; either 45 for *P. pipistrellus* or 55 kHz for *P. pygmaeus*) within a radius of 500 m from a previous recording site. This threshold was robust enough to ensure data independence for the purpose of the selected method of analysis (Hirzel, Helfer & Metral 2001; Hirzel *et al*. 2002). In order to limit any bias due to spatial autocorrelation that might arise from two single surveys along a transect, presence data were corrected as follows: whenever two detections of pipistrelles were < 1 km apart, one of the two records

was discarded randomly. Note that our two thresholds of 500 and 1000 m correspond to the radius of a circle which would represent average nightly home ranges of 109 ha in *P. pygmaeus* and 91·9 ha in *P. pipistrellus* (Davidson-Watts & Jones 2006).

SOUND ANALYSIS

Bat audio sequences were analysed in the laboratory with the program CANARY version 1·2·4 (Charif, Mitchell & Clark 1995). Sampling frequency was set to 440 kHz, with 8 bits per sample, and 512 points Fast-Fourier transformations with a Blackman window analysis were performed. For spectrograms, a resolution of 861·3 Hz was applied. We used a linear discriminant analysis developed by Zingg (1990) based on calls of bats recorded in Switzerland to identify species. The classification function (Appendix S1, Supplementary material) includes five call parameters: duration (D), start frequency, end frequency, frequency at D/2 and FMAXE (frequencies measured in the centre of call width). If no clear attribution was achieved based on the classification function or if FMAXE was between 50 and 51 kHz, the call sequence was not assignable and was therefore discarded. Between one and 11 echolocation calls from a single sequence were needed to achieve species identification.

ADDITIONAL DATA

For ENFA models, Hirzel, Helfer & Metral (2001) have shown that prediction reliability regarding distribution maps based on habitat suitability may be affected by the number of presence points. As the audio sample of *P. pygmaeus* was small we added additional presence points, or locations, resulting from casual recordings obtained between 1984 and 2003 by regional bat experts and ourselves, to the data set. The number of locations was selected to maintain the same relative proportion as in the stratified design of the transects in the six biogeographical regions. Presence points were grouped into clusters so that locations in each cluster were not separated by more than 30 km. One location was selected at random from each cluster so that the total number of locations maintained the proportions of our stratified design in each biogeographical region. If more locations were needed to maintain the proportions, additional presence points were selected at random from the cluster containing most observations. We ensured that no pair of locations was separated by less than 1 km.

ENVIRONMENTAL PREDICTORS

The study area was modelled as a raster map (1 ha per pixel) based on the Swiss Coordinate System (plane projection, $n = 2\,470\,145$ cells). Four types of environmental descriptor classes were included (Geostat, Swiss Federal Office of Statistics): (i)

topographic variables (continuous), including elevation, slope, and aspect; (ii) anthropogenic variables (boolean data indicating presence–absence) including human settlements; (iii) habitat variables comprising land use information such as forest types and agricultural use (boolean data); and (iv) hydrological data for rivers and lakes (boolean data). Boolean eco-geographical variables (EGVs) had to be rendered quantitative following the procedure described in Hirzel *et al.* (2004). Maps indicating the distance from the focal cell to a cell with presence of the concerned variable were calculated using the module 'Distance' of the program IDRISI version 32·01 (Eastman 2002). The module 'CircAn' of BIOMAPPER version 3·2 (Hirzel, Hausser & Perrin 2005) was used to compute the frequency of occurrence of the focal feature within a 1-km circular window, corresponding roughly to species' home range (Davidson-Watts & Jones 2006). A preliminary analysis was performed with 58 variables. Based on quality information given by cross-validation indices (see below), 23 EGVs were retained for the final model (Table 1). The distributions of the EGVs were normalized by the Box–Cox algorithm (Sokal & Rohlf 1994).

ENFA

The principles and procedure of ENFA, based on the niche concept by Hutchinson (1957) and implemented in a multivariate statistical framework, have been described in detail in previous papers (Hirzel *et al.* 2002). ENFA summarizes the overall information under the form of two types of indices. The first index is termed 'marginality': it maximizes the multivariate distance of the EGVs between the cells occupied by the species and the cells within the whole reference area. Marginality values for each EGV are also used to calculate the relative difference between the transect data set and the enhanced data set of *P. pygmaeus*. The second and subsequent factors are termed 'specialization(s)'; they account for the decreasing residual variance after removal of upper-ranked explanatory factors, and denote to which extent the species' EGVs distribution is narrow with respect to the overall distribution of the EGVs in the whole reference area. The inverse of specialization is therefore a measure of species' tolerance. Marginality and specialization are uncorrelated factors, with the major information contained within the first factors (Hirzel *et al.* 2002). A global marginality factor close to 1 means that the species lives in a very particular habitat relative to the reference set. A randomly chosen set of cells is expected to have a tolerance of 1, i.e. any value below 1 indicates some form of specialization.

HABITAT SUITABILITY MAPS

Habitat suitability maps are calculated by the median algorithm based on the first factors obtained by the ENFA. The number of factors included results from a

Table 1. Correlation between the ENFA factors and the ecogeographical variables for *P. pygmaeus*. Factor 1 explains 100% of the marginality. The percentages indicate the amount of specialization accounted for by the factor

	Factor 1[1] (9%)	Factor 2[2] (23%)	Factor 3[2] (14%)	Factor 4[2] (8%)	Factor 5[2] (7%)	Factor 6[2] (6%)	Factor 7[2] (5%)
Alpine meadow frequency	0	0	*	***	**	**	***
Single building frequency	+ +	*	0	**	****	*	*
Distance to single buildings	−[3]	*	0	*	**	0	*
Bushes and scrubland frequency	+ +	*	0	0	*	0	*
Altitude	−	********	*	*	***	*****	***
Bushy forest frequency	+	0	*	*	**	*	*
Dense forest frequency	−	*	******	**	0	***	0
Open forest frequency	+	0	*	******	**	*	*
Grove frequency	+ +	*	***	*	***	*	0
Grass frequency	0	*	**	0	0	**	*
Distance to small streams	−[3]	0	*	0	***	*	*
Distance to wide rivers	− −[3]	*	*	**	0	0	**
Distance to lake-borders	−[3]	*	**	0	***	*	0
Meadow frequency	0	****	***	0	****	****	0
Pasture frequency	−	0	0	***	**	***	***
Riparian forest frequency	+ +	*	***	***	*	0	******
Riparian vegetation frequency	+ +	*	0	*	*	0	*
Slopes	−	*	**	****	**	***	***
Town frequency	+ +	0	**	**	*	0	0
Distance to towns	−[3]	*	**	**	*	**	**
Village frequency	+	*	**	**	*	*	***
Distance to villages	−[3]	0	*	*	*	*	**
Vineyard frequency	0	***	**	*	**	***	*

[1]Marginality factor. Positive values mean that the species was found in locations with higher values than the average cell. Negative values mean the reverse. The greater the number of symbols, the higher the correlation; 0 indicates a very weak correlation.
[2]Specialization factor. Any number > 0 means the species was found occupying a narrower range of values than available. The greater the number of symbols, the narrower the range; 0 indicates a very low specialization. [3]Avoidance of an increasing distance to the variable may be understood as 'preference of proximity' to the habitat mentioned.

comparison of factors' eigenvalues based on a MacArthur's broken-stick distribution (Jackson 1993; Hirzel *et al.* 2002). On one factor axis, calculation is based on a count of all cells from the species distribution that lay at least as far apart from the median as the focal cell. This procedure is repeated for each factor included in the habitat suitability calculation. Overall habitat suitability for each cell is calculated by combining the score of each factor. Habitat suitability varies from 0 (worst habitat) to 100 (best habitat) and indicates how the environmental combination of a single cell suits the requirements of the focal species.

EVALUATION

We evaluated the habitat suitability model accuracy by means of 10-fold cross-validation (Fielding & Bell 1997): the presence data set was partitioned evenly but randomly into 10 partitions. Each partition was used in turn to evaluate the predictions computed by a model calibrated on the other nine partitions. This process provided 10 values for each evaluation measure, summarized by their mean and standard deviation. We computed three recent presence-only evaluation measures. First, the Absolute Validation Index (AVI) is the proportion of the evaluation partition with habitat suitability greater than 50; it indicates how well the model discriminates high-suitability from low-suitability areas (Hirzel & Arlettaz 2003; Hirzel *et al.* 2004); AVI varies from 0 to 1. Secondly, the Contrast Validation Index (CVI; equals the AVI minus the AVI of a null model which would predict suitable habitat at random) indicates how much the AVI differs from what would have been obtained with a random model (Hirzel & Arlettaz 2003; Hirzel *et al.* 2004); it varies from 0 to AVI. On the basis of an arbitrary threshold (habitat suitability = 50), these two measures determine how good the model is at discriminating between presence and absence. Thirdly, and by contrast, the Boyce index *B4* (Boyce *et al.* 2002) provides a more continuous assessment of model predictive power (Hirzel *et al.* 2006). We defined four classes of habitat suitability (hereafter called unsuitable, marginal, suitable and optimal habitat) and counted how many presence points of the evaluation partition fell into each. Combined with the total area covered by each class in the study area, this provided a predicted-to-expected frequency (PE, Boyce's area-adjusted frequency) of presence for each of them. The Boyce index *B4* was finally computed by the Spearman's rank between the PE and the class rank, varying from –1 to 1, with 0 indicating a random model (Boyce *et al.* 2002; Hirzel *et al.* 2006).

The three habitat suitability class boundaries used to delineate the four classes were set as follows: habitat suitability values with no presence points (PE = 0)

denotes unsuitable habitat; habitat suitability values for which presences are less frequent than expected by chance ($0 < PE \leq 1$) defines marginal habitat; suitable and optimal habitat shared habitat suitability values for which presences were more frequent than expected by chance ($PE > 1$), the boundary being placed so as to maximize the PE difference between them (Hirzel *et al.* 2006).

HABITAT NICHE DIFFERENTIATION BETWEEN THE TWO SPECIES

We performed a discriminant analysis to compare the ecological niches of *P. pygmaeus* and *P. pipistrellus* (Legendre & Legendre 1998). Like the ENFA, this multivariate analysis works in the space defined by the predictors but it uses both species' distributions simultaneously. It computes the factor that maximizes the interspecific variance while minimizing the intraspecific variance. In other words, the discriminant factor is the direction along which the two species differ the most, i.e. it is correlated with the variables on which they are most differently distributed. This common factor allowed us to further investigate the relationships between the two niches. We used the discriminant factor as an integrative variable on which to compare the niches and compute their overlap. We computed niche breadth by means of the standardized Levins' index B* and Hurlbert's index B′ (Hurlbert 1978). To analyse how much the niches of the two species were overlapping we used the Lloyd's asymmetric overlap index (Hurlbert 1978) computed on the discriminant factor. These computations are integrated in BIOMAPPER 3·2 (Hirzel, Hausser & Perrin 2005).

Results

PRESENCE DATA

Overall, 1811 sequences of bat calls were recorded along the 20 road transects each monitored twice. Of those, 236 calls (13%) were either of bad quality or could not be identified reliably and were discarded, giving an overall density of 0·98 observations per km of road ($1\cdot13$ km^{-1} for total call sequences). Of the 1575 identified call sequences, Zingg's classification function (Zingg 1990) attributed 29 to *P. pygmaeus* and 951 to *P. pipistrellus*. The remaining 595 call sequences were attributed to other bat species. Observations per survey varied greatly for both species: *P. pygmaeus* was recorded between no and five times per 40 km survey (mean = 0·73), while *P. pipistrellus* was identified between three and 62 times (mean = 23·8) per survey. The overall ratio of observation of *P. pipistrellus* to *P. pygmaeus* on transects was 32·8 to 1. While *P. pipistrellus* was detected on all 20 transects, *P. pygmaeus* was only detected on six transects. No difference in numbers of bat passes between surveys in the breeding and the post-breeding period were found for either species (matched-pair *t*-test, *P. pipistrellus*

$t = 0\cdot23$, d.f. = 19, $P = 0\cdot05$; *P. pygmaeus*, $t = 0\cdot64$, d.f. = 19, $P = 0\cdot05$). The percentages of observations of *P. pygmaeus* (first value in brackets, $n = 29$) and *P. pipistrellus* (second value in brackets, $n = 595$) with respect to region were: Jura Mountains (0% vs. 9·4%), Midlands (27·5% vs. 21·2% records), Northern Alps (13·8% vs. 29·3%), Eastern Alps (55·2 vs. 10%), Southern Alps (3·4 vs. 18·8%) and Western Alps (0% vs. 11·2%). After correcting for spatial autocorrelation (see Methods), 20 presence points for *P. pygmaeus* and 375 presence points for *P. pipistrellus* could be retained for further analysis (hereafter 'transect data').

Further screening of 85 locations, where 129 casual records of *P. pygmaeus* had been obtained previously, resulted in the retention of 43 locations (see justification in the Methods). This procedure yielded a total of 63 presence locations for ENFA modelling of *P. pygmaeus* (hereafter 'enhanced data'). The ENFA of the transect data and the enhanced data showed a marginal, negligible variation in marginality and specialization indices. The ENFA additionally revealed differences in EGVs of less than 13% for the 23 explanatory variables, with regard to patterns of habitat preference/avoidance. As the 'enhanced data set' performed better for all three evaluation indices implemented in BIOMAPPER (AVI, CVI, Boyce Index) we considered that sample in subsequent statistical treatment.

ENFA

A high global marginality value of 1·2 for *P. pygmaeus* indicates that this species foraged in very particular habitats compared to their availability in the reference area, mainly near wide rivers and lakes (Table 1). Occurrence also correlated positively with proximity and availability of buildings, towns and villages. In addition, *P. pygmaeus* preferred open woodland (such as groves and open forests), bushes and scrubland, especially when these were adjacent to rivers and lakes, whereas this species avoided foraging along roads in dense forests. A tolerance of 0·68 indicates a certain tolerance towards deviations from its optimal habitat. *P. pygmaeus* chose low altitudes [600 ± 194 m (mean ± SD), compared to 784 ± 321 m for the whole reference area] and was not tolerant towards changes in altitude (high value of the first specialization factor for that EGV). *P. pygmaeus* avoided open land such as pasture. Based on seven factors totalling 87% of overall information, the habitat suitability map (Fig. 2) indicates that favourable habitats are sparse and distributed patchily in Switzerland, with wide rivers and lakes being essential components.

A comparatively much lower marginality of 0·74, combined with an extremely high tolerance index of 0·99 (1 indicates no specialization at all) indicates that the ecology of *P. pipistrellus* differed markedly from that of *P. pygmaeus*. Applying MacArthurs' broken-stick rule, 10 factors explaining 82·6% of the information were retained for calculating habitat suitability.
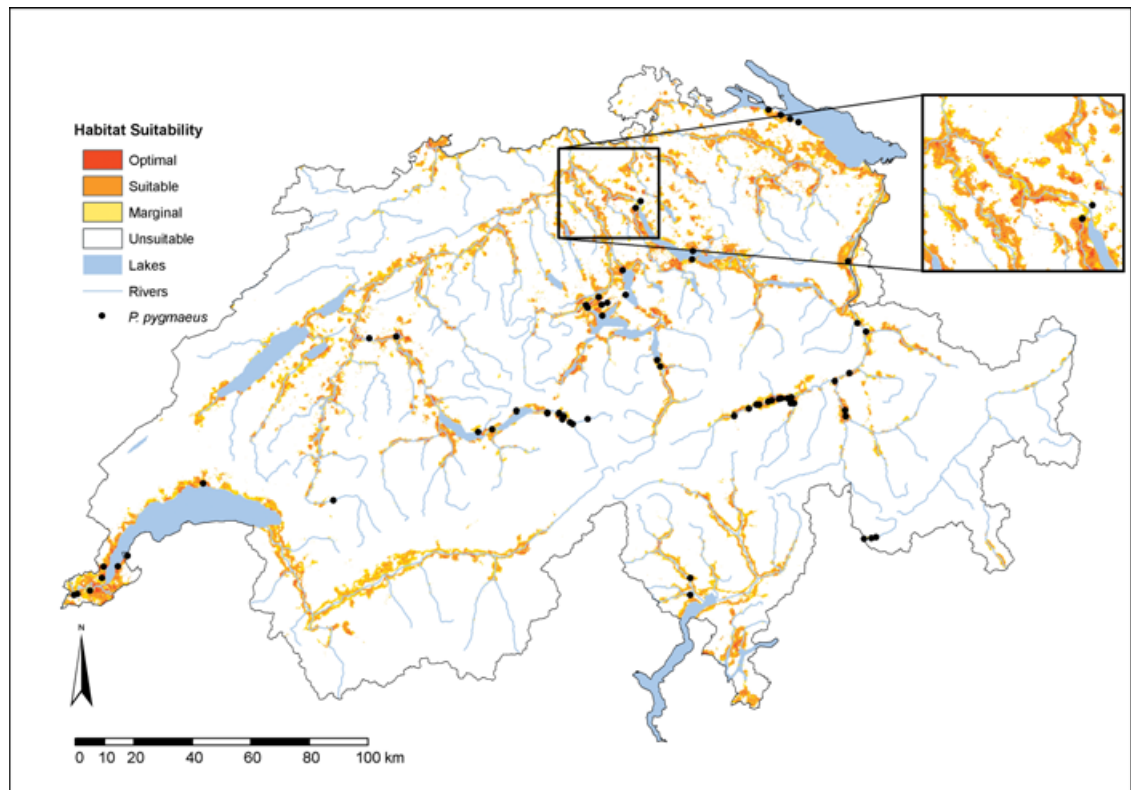
Fig. 2. *P. pygmaeus*: habitat suitability map and presence points used for ENFA (*n* = 63). Habitat quality is demonstrated by visualizing the four bin classes optimal (0·8% of total study area), suitable (11·4%), marginal (5·4%) and unsuitable (82·4%) habitat.

P. pipistrellus preferred proximity and availability of villages and buildings (Table 2) and the species was neutral regarding towns. Additionally, strongly preferred habitat types were wide rivers, riparian vegetation, vineyards and groves. *P. pipistrellus* occurred predominantly in lowlands, but tolerated higher elevations than *P. pygmaeus*. Dense forests were avoided and the species exhibited a narrow niche with regard to this predictor. In contrast to *P. pygmaeus*, favourable habitat for *P. pipistrellus* seems to be distributed widely in Switzerland, from the Midlands into the main Alpine valleys (Fig. 3). It is only at high elevation in the Jura Mountains and in the Northern Alps that suitable habitats seem to be lacking. Despite the fact that *P. pipistrellus* showed a preference for wide rivers, it also colonized areas far from water.

The three indices used for evaluating the habitat suitability models were all higher for *P. pygmaeus* than for *P. pipistrellus* (Table 3). While slightly lower, the AVI for *P. pipistrellus* was similar to that of *P. pygmaeus*, indicating that the fractions of correctly classified presence points and of the evaluation partition were comparable. A mean CVI (which indicates to what extent a suitability map differs from a purely random model) of 0·59 for *P. pygmaeus* indicated an appropriate map, whereas a CVI of 0·11 for *P. pipistrellus* denoted a somewhat inaccurate map. This means that the modelling using the selected EGVs had difficulty distinguishing the specific habitat preferred by *P. pipistrellus* from the overall habitat available in the

reference area. However, a low CVI is to be expected with such a tolerant species as *P. pipistrellus*. The Boyce index B4 for *P. pygmaeus* (0·98 ± 0·06) was near its theoretical maximum of 1, attesting to very good predictive power. The mean B4 of *P. pipistrellus* was also quite high (0·84 ± 0·25), but the large standard deviation is a symptom of low robustness.

HABITAT DIFFERENTIATION BETWEEN *P. PYGMAEUS* AND *P. PIPISTRELLUS*

The distribution range of species observations along the first discriminant axis shows that the niche of *P. pipistrellus* is wide and encompasses most of the narrow niche of *P. pygmaeus* (Fig. 4). The discriminant axis does not really separate the two species, although the discriminant function indicates on which variables they differ most markedly (Table 4). A high frequency of vineyards and open habitats such as meadow and pasture, as well as increasing distances away from lakes, wide rivers and single buildings, correlates only with the occurrence of *P. pipistrellus*. On the other hand, a high frequency of riparian forests, single buildings as well as bushes and scrubland favours *P. pygmaeus*. The standardized Levins' niche breadth index B* and the Hurlbert's niche breadth index B' indicate that the habitat niche for *P. pygmaeus* (B* = 0·19; B' = 0·11) is substantially smaller than that of *P. pipistrellus* (B* = 0·42; B' = 0·59). The Lloyd's asymmetric overlap of the ecological niche of *P. pipistrellus* on the niche of

Table 2. Correlation between the ENFA factors and the ecogeographical variables for *P. pipistrellus*. Factor 1 explains 100% of the marginality. The percentages indicate the amount of specialization accounted for by the factor

	Factor 1[1] (6%)	Factor 2[2] (11%)	Factor 3[2] (8%)	Factor 4[2] (7%)	Factor 5[2] (6%)	Factor 6[2] (6%)	Factor 7[2] (6%)	Factor 8[2] (5%)	Factor 9[2] (5%)	Factor 10[2] (5%)
Alpine meadow frequency	0	0	*	*	**	0	0	*	**	*****
Single building frequency	+ +	*	0	**	**	***	*	*****	*	0
Distance to single buildings	–[3]	*	0	0	0	*	*	**	0	*
Bushes and scrubland frequency	+ +	*	*	*	**	*	0	*	**	***
Altitude	–	*	0	***	****	***	**	**	***	**
Bushy forest frequency	+	*	*	**	*	*	**	0	*	*
Dense forest frequency	–	******	****	*	***	*****	*	****	*	*
Open forest frequency	+ +	**	*	0	0	**	0	0	***	**
Grove frequency	+ + +	0	******	*	**	***	*	0	**	*
Grass frequency	+ +	**	*	*	**	0	*	0	*	*
Distance to small streams	–[3]	*	*	*	0	*	*	0	**	*
Distance to wide rivers	–[3]	0	*	**	0	0	*	0	**	**
Distance to lake-borders	–[3]	0	*	0	**	***	0	*	***	0
Meadow frequency	0	*****	**	*	*****	****	**	****	0	**
Pasture frequency	–	**	*	****	***	**	0	**	*	**
Riparian forest frequency	+	*	******	*****	*	0	*	0	**	*
Riparian vegetation frequency	+ + +	0	*	0	0	*	*	0	*	*
Slopes	–	***	***	**	**	*	0	***	**	*
Town frequency	+	*	*	****	*	*	***	****	***	*****
Distance to towns	0	**	*	****	**	**	****	**	***	****
Village frequency	+ + +	**	*	**	***	**	*******	**	***	**
Distance to villages	–[3]	*	0	*	0	*	***	*	**	*
Vineyard frequency	+ + +	*	*	*	0	**	*	**	0	0

[1]Marginality factor. The symbol + means that the species was found in locations with higher values than average. The symbol – means the reverse. The greater the number of symbols, the higher the correlation; 0 indicates a very weak correlation. [2]Specialization factor. The symbol * means the species was found occupying a narrower range of values than available. The greater the number of asterix, the narrower the range; 0 indicates a very low specialization. [3]Avoidance of an increasing distance to the variable may be understood as 'preference of proximity' to the habitat mentioned.
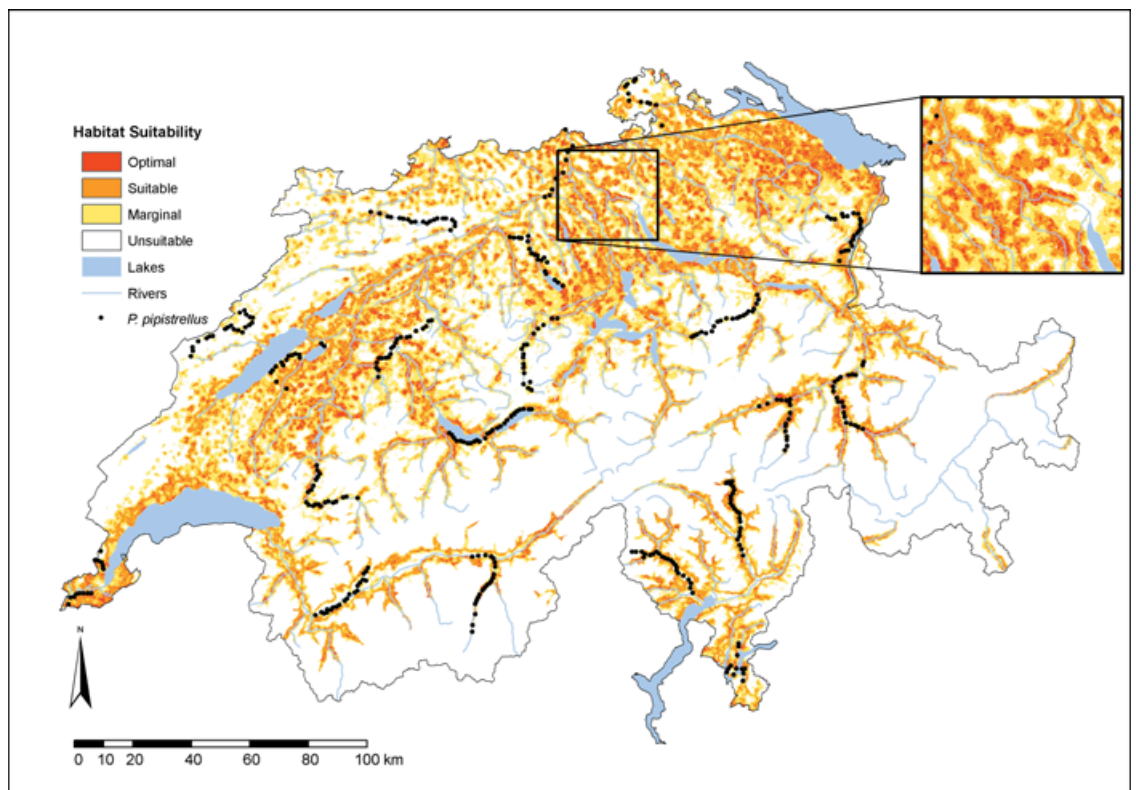
Fig. 3. *P. pipistrellus*: habitat suitability map and presence points used for ENFA (*n* = 375). Habitat quality is demonstrated by visualizing the four bin classes optimal (5·2% of total study area), suitable (28·5%), marginal (33·8%) and unsuitable (32·5%) habitat.

Table 3. Model evaluation indices for the habitat suitability maps of *P. pygmaeus* and *P. pipistrellus*, computed with 10-fold cross-validation. High mean values indicate a high consistency with evaluation data sets. The lower the standard deviation (SD), the more robust the prediction

	Absolute validation index[1]	Contrast validation index[2]	Boyce index[3]
P. pygmaeus			
Mean	0·77	0·59	0·98
SD	0·17	0·17	0·06
P. pipistrellus			
Mean	0·68	0·11	0·84
SD	0·08	0·08	0·25

[1]AVI varies from 0 to 1. [2]CVI varies from 0 to AVI. [3]Boyce's index varies from –1 to 1, with 0 indicating a random model.

P. pygmaeus is 15·6, whereas the reciprocal overlap is only 2·4. These findings indicate that the habitat width of *P. pipistrellus* almost completely encompasses the habitat width of *P. pygmaeus* (Fig. 4).

Discussion

Our results show that *P. pipistrellus* is encountered frequently along Swiss roads: 60·3% of all bat acoustic recordings were of this species, whereas *P. pygmaeus* comprised only 1·84% of observations in these sampling conditions. Previous acoustic studies have shown that *P. pipistrellus* is not detected more easily or more frequently than *P. pygmaeus* along road networks, nor is *P. pipistrellus* a road specialist (Sattler 2003). The present findings thus suggest that *P. pygmaeus* is much less widespread and much less abundant than *P. pipistrellus* in Switzerland. This contrasts with road transect observations in Ireland, where *P. pipistrellus* and *P. pygmaeus* contributed to 43% and 28%, respectively, of all bat observations along road transects close to linear habitat features (Russ & Montgomery 2002). In England, these proportions were 30% and 34%, respectively (Vaughan, Jones & Harris 1997).

The distribution of foraging *P. pygmaeus* in Switzerland is limited to a narrow range of habitats. *P. pipistrellus*, in contrast, occurs in a wide range of habitats. Additionally, *P. pygmaeus* is very specific regarding its habitat requirements and remains in its optimal habitats, whereas *P. pipistrellus* is tolerant to deviations from its optimum habitat. These quantitative findings corroborate results from England, Ireland, Germany and Italy: *P. pygmaeus* is frequently found near water bodies in all areas studied to date; in Ireland and Italy, open deciduous forests constitute other suitable habitats (Vaughan, Jones & Harris 1997; Häussler *et al.* 2000; Russ & Montgomery 2002; Russo & Jones 2003; Davidson-Watts & Jones 2006).

The comparison of observation maps with habitat suitability maps show that *P. pygmaeus* was not detected in some areas where its occurrence would be predicted a posteriori (e.g. Western Alps, corresponding to canton of Valais). It is unlikely that the species has been overlooked in this area because intensive bat monitoring has been performed continuously by regional bat experts since the 1980s (Arlettaz *et al.* 1997a; unpublished data). Historical colonization processes could explain this regional absence, however, especially in the least accessible Alpine valleys.

Evaluation indices, especially a low CVI, suggest that the habitat suitability model for *P. pipistrellus* – despite a high number of presence points – does not describe accurately the distribution pattern of the species. Several studies have shown that it is easier to predict habitat suitability maps for marginal than for widespread and common species, for purely methodological reasons (Stockwell & Peterson 2002; Segurado & Araújo 2004). The interspecific differences in the accuracy of our habitat suitability models also illustrate that a higher number of presence locations does not improve model precision and reliability *per se* (Zaniewski, Lehmann & Overton 2002).

Predictive distribution models such as ENFA represent empirical models which sacrifice generality for precision and reality (Guisan & Zimmermann
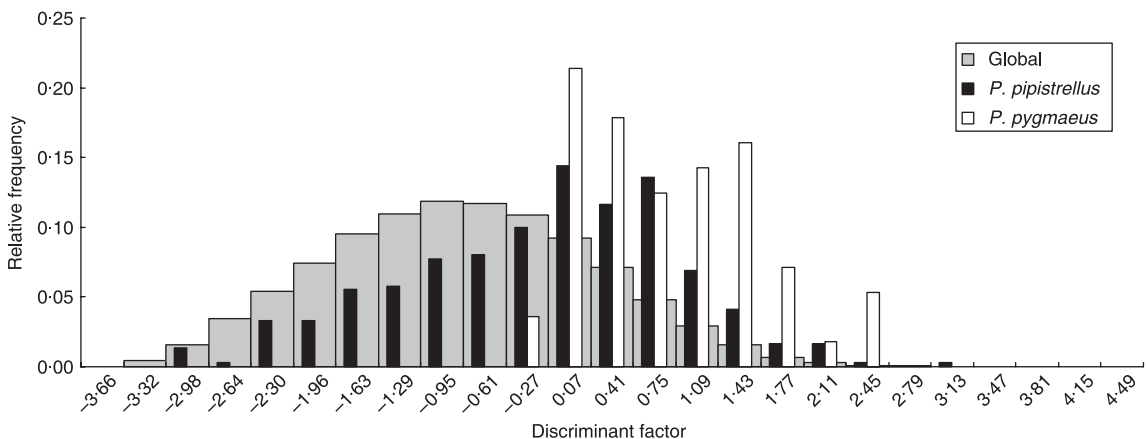
Fig. 4. Niche characteristics for *P. pygmaeus* (white) and *P. pipistrellus* (black) in relation to global distribution of all cells of the study area (grey) along the discriminant factor.

Table 4. Coefficients of the first discriminant factor between the habitat characteristics of *P. pygmaeus* and *P. pipistrellus*. Positive values indicate variables that favour *P. pygmaeus*, whereas the distribution of *P. pipistrellus* covers the whole range (Fig. 4)

Coefficients	Value
Vineyard frequency	−0·319
Distance to lake-borders	−0·31
Distance to wide rivers	−0·289
Meadow frequency	−0·275
Distance to single buildings	−0·261
Dense forest frequency	−0·217
Altitude	−0·215
Grass frequency	−0·203
Pasture frequency	−0·18
Slopes	−0·082
Riparian vegetation frequency	−0·075
Distance to villages	−0·066
Alpine meadow frequency	−0·042
Town frequency	−0·028
Village frequency	−0·028
Distance to small streams	−0·025
Bushy forest frequency	0·091
Grove frequency	0·105
Open forest frequency	0·118
Distance to towns	0·164
Bushes and scrubland frequency	0·269
Single building frequency	0·334
Riparian forest frequency	0·378

2000). It is often contended that presence-only models are of limited use and validity because of two main reasons: unknown sampling bias due to non-systematic data samples and habitat suitability maps yielding over-optimistic predictions (Zaniewski, Lehmann & Overton 2002). In this study we avoided unknown sampling bias through a systematic sampling design applying a proportional stratified sampling regime. Based on this sampling scheme, the data set was enhanced in a systematic way for *P. pygmaeus* with additional observations not arising from our standard road transects. Supplementary analysis for *P. pygmaeus* with data obtained unsystematically yielded similar results as the systematic approach for both habitat selection and habitat suitability maps, corroborating previous simulations with modelled virtual species distributions which showed high robustness in the ENFA approach (Hirzel, Helfer & Metral 2001). The present study applies cross-validated predicted-to-expected frequency curves so that the effect of too optimistic predictions is minimized. This new method (Hirzel *et al.* 2006) allowed us to define objectively four suitability classes. These classes were based on the actual data and not on arbitrary thresholds. Moreover, if the objective of the habitat suitability model is to protect rare or endangered species, over-estimating distribution areas might be preferable to under-estimating species presence (Fielding & Bell 1997).

It has been suggested that habitat suitability modelling should take interspecific competition into consideration (Guisan & Zimmermann 2000). Unfortunately, we cannot draw any conclusions concerning competition or stable coexistence between *P. pygmaeus* and *P. pipistrellus*, as we were able to compare only respective habitat breadth and differentiation. As morphological resemblance is thought to reflect niche similarity, some authors have predicted only small niche differentiation in morphologically similar bats (Aldridge & Rautenbach 1987; Findley 1993). In practice, however, substantial niche separations between cryptic species have been found, including microhabitat and food partitioning (Arlettaz, Perrin & Hausser 1997b; Arlettaz 1999). Fine-grained microhabitat and diet differentiation are expected to occur in the two pipistrelle bats in Switzerland, as has been found in the United Kindom (Barlow 1997).

In Switzerland, *P. pipistrellus* is widespread and abundant; it can therefore continue to be classified safely as 'not threatened' (Duelli 1994). In contrast, *P. pygmaeus*, which was confused until recently with *P. pipistrellus*, is a rare bat with a patchy distribution which qualifies for the category 'near threatened'. These findings suggest that an amendment to Swiss bat conservation policy is appropriate. First, it will be essential to put more emphasis upon the protection of *P. pygmaeus* roosts. Secondly, conservation managers need to recognize that the distribution of *P. pygmaeus* is confined to a tiny range of habitat types and that large rivers bordered by riparian woodland are a crucial habitat for this species. During the last 150 years, most Swiss streams have been dammed and canalized, with riparian forests being destroyed systematically (Ewald 1978). Therefore, the present fragmented distribution of *P. pygmaeus* may result from a dramatic drop in habitat availability. The protection of the remaining riparian forests and the restoration of wide stream beds to support natural vegetation dynamics would thus be essential steps for ensuring the persistence of *P. pygmaeus*.

World-wide, there are around 1116 recognized bat species (Simmons 2005), but it is likely that more cryptic species will be discovered in the near future, especially in more remote biomes (Jones 1997). Species protection will necessitate further taxonomic and ecological investigations as only reliable status assessments and evidence-based protection guidance will enable development and/or readjustment of effective international and national bat conservation policies. Spatially explicit methods such as ENFA can provide some decisive assistance in the delicate task of determining species basic ecological needs.

References

Aldridge, H.D.J.N. & Rautenbach, I.L. (1987) Morphology, echolocation and resource partitioning in insectivorous bats. *Journal of Animal Ecology*, **56**, 763–778.

Arlettaz, R. (1999) Habitat selection as a major resource partitioning mechanism between the two sympatric sibling bat species *Myotis myotis* and *Myotis blythii*. *Journal of Animal Ecology*, **68**, 460–471.

Arlettaz, R., Lugon, A., Sierro, A. & Desfayes, M. (1997a) Les chauves-souris du Valais (Suisse): statut, zoogeographie et ecologie [The bats of Valais (Switzerland): status, zoo-geography and ecology]. *Le Rhinolophe*, **12**, 1–42.

Arlettaz, R., Perrin, N. & Hausser, J. (1997b) Trophic resource partitioning and competition between the two sibling bat species *Myotis myotis* and *Myotis blythii*. *Journal of Animal Ecology*, **66**, 897–911.

Barlow, K.E. (1997) The diets of two phonic types of the bat *Pipistrellus pipistrellus* in Britain. *Journal of Zoology*, **243**, 597–609.

Barlow, K.E. & Jones, G. (1997) Function of pipistrelle social calls: field data and a playback experiment. *Animal Behaviour*, **53**, 991–999.

Barratt, E.M., Deaville, R., Burland, T.M., Bruford, M.W., Jones, G., Racey, P.A. & Wayne, R.K. (1997) DNA answers the call of pipistrelle bat species. *Nature*, **387**, 138–139.

Boyce, M.S., Vernier, P.R., Nielsen, S.E. & Schmiegelow, F.K.A. (2002) Evaluating resource selection functions. *Ecological Modelling*, **157**, 281–300.

Brito, D. (2004) Lack of adequate taxonomic knowledge may hinder endemic mammal conservation in the Brazilian Atlantic Forest. *Biodiversity and Conservation*, **13**, 2135–2144.

Buckland, S.T. & Elston, D.A. (1993) Empirical models for the spatial distribution of wildlife. *Journal of Applied Ecology*, **30**, 478–495.

Charif, R.A., Mitchell, S. & Clark, C.W. (1995) *Canary 1·2 User's Manual*. Cornell Laboratory of Ornithology, Ithaca, NY.

Davidson-Watts, I. & Jones, G. (2006) Differences in foraging behaviour between *Pipistrellus pipistrellus* (Schreber, 1774) and *Pipistrellus pygmaeus* (Leach, 1825). *Journal of Zoology*, **298**, 55–62.

Dietz, C. & von Helversen, O. (2004) *Illustrated Identification Key to the Bats of Europe*, 1st edn, *Tuebingen & Erlangen.*. Electronic publication. Available at: www.uni-tuebingen.de/tierphys/Kontakt/mitarbeiter_seiten/dietz.htm (accessed June 2005).

Duelli, P. (1994) *Rote Liste der Gefährdeten Tierarten in der Schweiz* [Red list of endangered animals of Switzerland]. Bundesamt für Umwelt, Wald und Landschaft (BUWAL) [Federal Office for the Environment FOEN], Bern.

Eastman, J.R. (2002) *IDRISI 32·22*. Clark Laboratories, The Idrisi Project, Worcester MA.

Ewald, K.C. (1978) *Der Landschaftswandel. Zur Veränderung Schweizerischer Kulturlandschaften im 20. Jahrhundert* [Landscape change. On the alteration of Swiss cultural landscapes in the 20th century]. Eidgenössische Anstalt für das forstliche Versuchswesen [Swiss Federal Research Institute WSL], Birmensdorf.

Fielding, A.H. & Bell, J.F. (1997) A review of methods for the assessment of prediction errors in conservation presence/absence models. *Environmental Conservation*, **24**, 38–49.

Findley, J.S. (1993) *Bats: a Community Perspective*. Cambridge University Press, Cambridge.

Gonseth, Y., Wohlgemuth, T., Sansonnens, B. & Buttler, A. (2001) Die biogeographischen Regionen der Schweiz, Erläuterungen und Einteilungsstandard [Biogeographic regions of Switzerland. Explanations and standards of classification]. Bundesamt für Unwelt, Wald und Landschaft (BUWAL) [Federal Office for the Environment FOEN], Bern.

Guisan, A. & Zimmermann, K. (2000) Predictive habitat distribution models in ecology. *Ecological Modelling*, **135**, 147–186.

Häussler, U., Nagel, A., Braun, M. & Arnold, A. (2000) External characters discriminating sibling species of European pipistrelles, *Pipistrellus pipistrellus* (Schreber, 1774) and *P. pygmaeus* (Leach, 1825). *Myotis*, **37**, 27–40.

Hirzel, A.H. & Arlettaz, R. (2003) Modelling habitat suitability for complex species distributions by the environmental-distance geometric mean. *Environmental Management*, **32**, 614–623.

Hirzel, A. & Guisan, A. (2002) Which is the optimal sampling strategy for habitat suitability modelling. *Ecological Modelling*, **157**, 331–341.

Hirzel, A., Hausser, J., Chessel, D. & Perrin, N. (2002) Ecological-niche factor analysis: how to compute habitat-suitability maps without absence data? *Ecology*, **83**, 2027–2036.

Hirzel, A., Hausser, J. & Perrin, N. (2005) *Biomapper 3.2. Laboratory of Conservation Biology*. Department of Ecology and Evolution. University of Lausanne, Lausanne. Available at: http://www.unil.ch/biomapper (accessed May 2005).

Hirzel, A., Helfer, V. & Metral, F. (2001) Assessing habitat suitability models with a virtual species. *Ecological Modelling*, **145**, 111–121.

Hirzel, A.H., Le Lay, G., Helfer, V., Randin, C. & Guisan, A. (2006) Evaluating the ability of habitat suitability models to predict species presences. *Ecological Modelling*, **199**, 142–152.

Hirzel, A., Posse, B., Oggier, P.-A., Crettenand, Y., Glenz, C. & Arlettaz, R. (2004) Ecological requirements of reintro-duced species and the implications for release policy: the case of the bearded vulture. *Journal of Applied Ecology*, **41**, 1103–1116.

Honacki, J.H., Kinmam, K.E. & Koeppl, J.W. (1982) *Mammal Species of the World: a Taxonomic and Geographic Reference*, 1st edn. Allen Press, Inc. & Association of Systematic Collections, Lawrence, Kansas.

Hulva, P., Horacek, I., Strelkov, P.P. & Benda, P. (2004) Molecular architecture of *Pipistrellus pipistrellus Pipistrellus pygmaeus* complex (Chiroptera: Vespertilionidae): further cryptic species and Mediterranean origin of the divergence. *Molecular Phylogenetics and Evolution*, **32**, 1023–1035.

Hurlbert, S.H. (1978) Measurement of niche overlap and some relatives. *Ecology*, **59**, 67–77.

Hutchinson, G.E. (1957) Concluding remarks. *In Cold Spring Harbour Symposium on Quantitative Biology*, **22**, 415–427.

Ibáñez, C., García-Mudarra, J.L., Ruedi, M., Stadelmann, B. & Juste, J. (2006) The Iberian contribution to cryptic diversity in European bats. *Acta Chiropterologica*, **8**, 277–297.

International Union for the Conservation of Nature and Natural Resources (IUCN) (2001) *IUCN Red List Categories and Criteria*: version 3·1. IUCN, Gland, Switzerland and Cambridge, UK.

Jaberg, C. & Guisan, A. (2001) Modelling the distribution of bats in relation to landscape structure in a temperate mountain environment. *Journal of Applied Ecology*, **38**, 1169–1181.

Jackson, D.A. (1993) Stopping rules in principal components analysis: a comparison of heuristical and statistical approaches. *Ecology*, **74**, 2204–2214.

Jones, G. (1997) Acoustic signals and speciation: the roles of natural and sexual selection in the evolution of cryptic species. *Advances in the Study of Behavior*, **26**, 317–354.

Jones, G. (1999) *Pipistrellus pipistrellus* (Schreber 1774). *The Atlas of European Mammals* (eds A.J. Mitchell-Jones, G. Amori, W. Bogdanowicz, B. Krystufek, P.J. Reijnders, F. Spitzenberger, M. Stubbe, J.B. Thissen, V. Vohralik & J. Zima), pp. 126–127. Poyser Natural History, London.

Jones, G. & van Parijs, S.M. (1993) Bimodal echolocation in pipistrelle bats – are cryptic species present. *Proceedings of the Royal Society of London, Series B, Biological Sciences*, **251**, 119–125.

Legendre, L. & Legendre, P. (1998) *Numerical Ecology*, 2nd edn. Elsevier Science PV, Amsterdam.

Manel, S., Dias, J.M., Buckton, S.T. & Ormerod, S.J. (1999) Alternative methods for predicting species distribution: an illustration with Himalayan river birds. *Journal of Applied Ecology*, **36**, 734–747.

Mayer, F., Dietz, C. & Kiefer, A. (2007) Molecular species identification boosts bat diversity. *Frontiers in Zoology*, **4**, 4. DOI: 10.1186/1742-9994-4-4.

Mayer, F. & von Helversen, O. (1999) Cryptic species and genetic diversity in European bats. *Zoology*, **102** (Suppl. II), Abstract: 8.

Mayr, E. (1977) *Populations, Species, and Evolution*. Harvard University Press, Cambridge, MA.

Medellín, R.A. & Soberón, J. (1999) Predictions of mammal diversity on four land masses. *Conservation Biology*, **13**, 143–149.

Rushton, S.P., Ormerod, S.J. & Kerby, G. (2004) New paradigms for modelling species distributions? *Journal of Applied Ecology*, **41**, 193–200.

Russ, J.M. & Montgomery, W.I. (2002) Habitat associations of bats in Northern Ireland: implications for conservation. *Biological Conservation*, **108**, 49–58.

Russo, D. & Jones, G. (2003) Use of foraging habitats by bats in a Mediterranean area determined by acoustic surveys: conservation implications. *Ecography*, **26**, 197–209.

Sattler, T. (2003) *Ecological factors affecting the distribution of the sibling species* Pipistrellus pygmaeus *and* Pipistrellus pipistrellus *in Switzerland*. Masters thesis, University of Bern, Bern.

Segurado, P. & Araújo, M.B. (2004) An evaluation of methods for modelling species distributions. *Journal of Biogeography*, **31**, 1555–1568.

Simmons, N.B. (2005) Order Chiroptera. *Mammal Species of the World: a Taxonomic and Geographic Reference* (eds D.E. Wilson, D.M. Reeder & M.D. Baltimore), pp. 312–529. Johns Hopkins University Press, Baltimore and London.

Sokal, R.R. & Rohlf, F.J. (1994) *Biometry: the Principles and Practice of Statistics in Biological Research*, 3rd edn. W. H. Freeman, New York.

Stebbings, R.E. (1988) *Conservation of European Bats*. Christopher Helm, London.

Stockwell, D.R.B. & Peterson, A.T. (2002) Effects of sample size on accuracy of species distribution models. *Ecological Modelling*, **148**, 1–13.

Vallan, D., Vences, M. & Glaw, F. (2003) Two new species of the *Boophis mandraka* complex (Anura, Mantellidae) from the Andasibe region in eastern Madagascar. *Amphibia–Reptilia*, **24**, 305–319.

Vaughan, N., Jones, G. & Harris, S. (1997) Habitat use by bats (Chiroptera) assessed by means of a broad-band acoustic method. *Journal of Applied Ecology*, **34**, 716–730.

Vayssières, M.P., Plant, R.E. & Allen-Diaz, B.H. (2000) Classification trees: an alternative non-parametric approach for predicting species distributions. *Journal of Vegetation Science*, **11**, 679–694.

Wicht, B., Moretti, M., Preatoni, D., Tosi, G. & Martinoli, A. (2003) The presence of Soprano pipistrelle *Pipistrellus pygmaeus* in Switzerland: first molecular and bioacoustic evidences. *Revue Suisse de Zoologie*, **110**, 411–426.

Wilson, D.E. & Reeder, D.M. (1993) *Mammal Species of the World: a Taxonomic and Geographic Reference*, 2nd edn. Smithsonian Institution Press, Washington, DC.

Yoder, A.D., Rasoloarison, R.M., Goodman, S.M., Irwin, J.A., Atsalis, S., Ravosa, M.J. & Ganzhorn, J.U. (2000) Remarkable species diversity in Malagasy mouse lemurs (primates, *Microcebus*). *Proceedings of the National Academy of Sciences USA*, **97**, 11325–11330.

Zaniewski, A.E., Lehmann, A. & Overton, J.M.C. (2002) Predicting species spatial distributions using presence-only data: a case study of native New Zealand ferns. *Ecological Modelling*, **157**, 261–280.

Zingg, P.E. (1990) Akustische Artidentifikation von Fledermäusen (Mammalia: Chiroptera) in der Schweiz [Acoustic Species Identification of Bats (Mammalia, Chiroptera) in Switzerland]. *Revue Suisse de Zoologie*, **97**, 263–294.

Supplementary material

The following supplementary material is available for this article.

Appendix S1 Classification function for bat search calls.

This material is available as part of the online article from: http://www.blackwell-synergy.com/doi/full/10.1111/j.1365-2664.2007.01328.x
(This link will take you to the article abstract.)

Please note: Blackwell Publishing is not responsible for the content or functionality of any supplementary materials supplied by the authors. Any queries (other than missing material) should be directed to the corresponding author for the article.