

Common bats are more abundant within Natura 2000 areas

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1 Common bats are more abundant within Natura 2000 areas

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Abstract

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The Natura 2000 network is the largest conservation effort in Europe. However, despite the known importance of conserving common and widespread biodiversity, criteria used to designate a Natura 2000 site are oriented toward rare and/or emblematic biodiversity. In this study, we took advantage of the fact that the management of Natura 2000 is just beginning to assess whether the five most common bat (Chiroptera) species and one genus in France exhibit a greater relative abundance within rather than outside Natura 2000 boundaries, and three bats communities index: total relative abundance, species richness, Community Habitat specialization index. We model the relative abundance of each taxa and indices using data from a nationwide volunteer-based acoustic survey. We found that three of the six taxa studied exhibit greater relative abundance within Natura 2000 sites (this increase is noteworthy for E. serotinus (x 2.1) and Myotis ssp (x 3.6)). We also provide evidence that total relative abundance of bat activity and richness are globally higher in Natura 2000 sites (respectively +24% and + 14%) and on average communities are more specialized within Natura 2000 sites. In addition, when the effect of Natura 2000 is adjusted to the main land use types, a significant positive effect of Natura 2000 remains for most metrics. The positive Natura 2000 effect appears relatively small compared to the main land use pressure: intensive agriculture and artificial light at night. However, Natura 2000 has a comparable sized effect as habitat widely recognized as having a positive impact on bats, such as streams.

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Keywords: bats relative abundance, common biodiversity, Natura 2000; mean trait community,

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Introduction

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Among the approaches employed to preserve biodiversity, the establishment and management of statutory protected areas are widely regarded as key strategies (Jackson et al., 2007). This classic protection approach is based on the principle that the designation of areas affords the protection of habitats, species and/or populations that are threatened elsewhere (Gaston et al., 2008). In addition to National Nature Reserves, National Parks and Regional Parks that are designated by state members, the European Union (EU) has coordinated the Natura 2000 network. This network, which is the most important coordinated supra-national conservation effort at the world level (Evans, 2012), aims at maintaining remarkable biodiversity through the sustainable use of natural resources by establishing a network of areas that must maintain a range of habitat types and wildlife species in a "favorable conservation status" (European Commission, 2000). Over the last 25 years, the EU has built a vast network that represents 18% of the EU's land area (see Supplementary Material S1). In France, this network represents 12.5% of the continental territory. The main objectives of conservation and restoration are stated at the local scale in the Natura 2000 management plan and the French Natura 2000 areas are mainly managed through contracts between land owners and the state. Agri-Environmental Schemes (Caps) are applied in agricultural land, whereas ones in nonagricultural land may receive incentives for measures that favor conservation of biodiversity. Although such actions are defined in each management plan, the field management of Natura 2000 has only recently begun (by 2005 only 20% of Natura 2000 sites had endorsed their management plan (Maresca et al. 2006) and by 2012 nearly 73%, and establishment of new plan has been slow since 2005, Debain, 2011).

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Identification of the Natura 2000 areas is now broadly completed (Note however, that Member States can regularly add new sites), and attention is turning toward assessing whether

the Natura 2000 network effectively protects species and habitats (Martínez et al., 2006). As 66 67 Kliejn & van Zuijlen (2004) stated, the observed differences between protected and nonprotected areas may result from either the initial state of biodiversity ('designation effect'. 68 69 assessed through a comparison of the biodiversity inside and outside the N2000 sites) or from the protection efficiency per se ('protection effect', which involve the comparison of the state 70 of biodiversity before and after the designation of N2000 sites). Thus, the French case 72 provides a good opportunity to assess the first of these effects ('designation effect'), i.e., 73 differences in the initial biodiversity state between areas inside and outside the Natura 2000 74 network, because within the studied period half of Natura 2000 sites had just endorsed their 75 management plan and thus, management actions were too recent to already have an effect on 76 the state of the biodiversity. A growing number of studies have been carried out to evaluate the effectiveness of the Natura 78 2000 network for threatened species (Trochet & Schmeller, 2013) or species or habitats 79 targeted by Natura 2000 (i.e. listed under priority Annex, vegetation: Rosati et al., 2008, bats: 80 Lisón et al., 2015). Some studies like Donald et al., (2007) have found a strong positive correlation between Member States' percentage land cover of Natura 2000 and population 82 trends of species targeted by the EU Birds Directive (EC79/409). 83 Common biodiversity is of particular importance in ecosystem functioning (Smith & Knapp, 84 2003; Solan et al., 2004), but there are few protection measures directed toward common 85 species. While some common species are legally protected (common bats are listed in Annex IV Habitat Directive), there are no protected areas designated specifically at the benefice of 87 these species, as protected areas are not considered as the appropriate tool for these kinds of species (Gaston 2011). Surprisingly, few studies (Devictor et al., 2007, Pellissier et al., 2013) 88 89 have assessed the importance of classical protection measures on common bird biodiversity,

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90 showing that common bird species have actually benefited from protection measures directed 91 toward other target species. 92 In addition, among the studies focused on the evaluation of the designation effectiveness of 93 the Natura 2000 network, the overwhelming majority of them were gap analyses based on 94 species distribution (vertebrates: Kukkala et al., 2016, invertebrates: Sánchez-Fernández et 95 al., 2013, amphibians and reptiles: Abellán & Sánchez-Fernández, 2015, Bat: Lisón et al., 96 2013, 2015, all vertebrates: Kukkala et al., 2016, all taxa: Trochet & Schmeller, 2013), while 97 ecosystem functioning also requires the consideration of species abundance and community 98 scale (Loreau et al., 2001). In addition, a review of studies dealing with Natura 2000 perform 99 by Orlikowska et al., (2016) concluded that a majority of studies were performed in singe 100 Natura 2000 site, and suggest that more studies will need to encompass large spatial scale. 101 Monitoring based on skilled volunteers counting common species (birds, amphibians and 102 reptiles, mammals, butterflies and plants) has already provided reliable abundance data and 103 unbiased results (Genet & Sargent, 2003; Schmeller et al., 2009) regarding the effects of 104 climate change (Julliard et al., 2004) or the effectiveness of protection measures (Devictor et 105 al., 2007; Jiguet, 2011). Using volunteer bat-workers, the French National History Museum 106 initiated a bat monitoring program (Vigie Nature 2011) to assess the current state and trends 107 of common bat population in France (Kerbiriou et al., 2010). Most of the European bat 108 populations, including common species, have experienced dramatic declines in the last 109 century (Voigt & Kingston, 2016) mostly because of the habitat loss and fragmentation 110 induced by agricultural intensification (Wickramasinghe et al., 2004, Azam et al., 2016), 111 urbanization (Kurta & Teramino, 1992; Azam et al., 2016) and intensive forest management 112 (O'Donnell, 2000) 113 Using the French bat monitoring data, we tested, mostly on non-target species, whether the 114 relative abundance of species or community metrics (total relative abundance, richness and habitat community specialization) were influenced by the Natura 2000 network. This test of Natura 2000 designation effect was achieved through two analyses, one comparing bat activities within and outside Natura 2000 sites and a second approach, in which we assessed the influence of Natura 2000 areas controlling for the main land uses and at different landscape scales. Higher relative abundance within Natura 2000 is expected if the Natura 2000 areas exhibit a 'designation effect', *i.e.*, the Natura 2000 areas are high-value areas for common species.

MATERIAL AND METHODS

Car transect survey

The data were provided by the French bat monitoring program (FBMP) (http://vigienature.mnhn.fr/page/vigie-chiro), a citizen-science program running since 2006 and coordinated by the French National Museum of Natural History (NMNH). Volunteers recorded bat activity while driving at a constant low-speed (25 ± 5 km/h) along a 30 km road circuit within a 10 km buffer around their home. Surveyors were asked to design their road circuit so that it proportionally crossed, as much as possible, the different land-cover types and it remained on low-traffic roads for security reasons. After final validation of the circuit outline, program coordinators randomly selected the starting point of the survey. Each circuit was then divided into 10 road transects (length of transect 2 km) where bats were recorded, separated by 1 km road portions where recording was not carried out (Supplementary Material 1). Such a sampling design resulted in a survey of habitats that are quite representative of French land cover (see Supplementary Material 1). We used data from surveys carried out every year from the 15th of June to the 31st of July, corresponding to a seasonal peak in bat activity, a period during which bat females typically give birth to and feed their offspring. Surveys started 30 min after sunset and lasted approximately 1.5 h during

the period of bat activity, and were only carried out when weather conditions were favorable (*i.e.*, no rain, low wind speed of <7 m/s, temperature >12 °C). This sampling design is quite similar to that used by many European bat monitoring schemes (Roche *et al.*, 2011). At total of 160 different car-based circuits were surveyed during the study period (2006 to 2013) (Supplementary Material 1), corresponding to 1608 independent 2-km transects, and each circuit was sampled an average of 2.4 ± 0.04 times.

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Biological data

Along transects, volunteer surveyors recorded bat echolocation calls with ultrasound detectors connected to digital recorder (See Supplementary Material 1 for more details about technical points). Once the recording was performed, the observers were asked to identify the species detected in each transect using Syrinx software version 2.6 (Burt, 2006) and spectrogram analyses. The majority of volunteers involved in the bat monitoring participated in training courses organized by the MNHN, allowing a high level of homogeneity of the identification criteria. In a second step, the identifications were validated by MNHN experts. Bat calls were identified at the species level except for species of the Myotis genus. Due to the lack of general consensus on the acoustic criteria to distinguish some Myotis species and some overlap between their acoustic repertoires, it was not possible to assign the exact species with certainty for all calls; thus, we constructed a *Myotis spp* group (see Supplementary Material S1). Note that according to their foraging trait, bat species of the genus *Myotis* within the study area could be considered as a group of slow-flying species including mainly "narrow space gleaning foragers" and "trawling bat" compared to the group of the fast-flying species including mainly "open or space aerial foragers" (i.e. Pipistrellus spp, Eptesicus serotinus and Nyctalus spp) species that mainly hunt for airbone prey in open space or in edge space (Dezinger & Schnitzler, 2013). Indeed, an analysis based on a group composed of species of

the same genus (i.e. *Myotis spp*) could lead to non-significant results due to opposite effects among species included such a group. However, when significant effects are detected, this involves that at least one species of this group was enough sensitive to the variable tested. As it is impossible to distinguish individual bats from echolocation calls. For this reason, we produced for each 2-km road transect sampled, and for each year, a bat activity index (name hereafter "relative abundance") defined as a number of bat pass per species (a bat pass corresponds to a trigger of the bat detector in time expansion, see Supplementary Material 1). The response variables were (i) species relative abundance (i.e. number of bat passes); (ii) total relative abundance (this measure was corrected by species distance of detection (Barataud 2015), species bat activity was weighted by the multiplicative inverse of the distance of detection squared, thus a species with a great distance of detection contributes less for total relative abundance than a species with a small distance of detection; (iii) species richness, (iv) Community Habitat specialization index (CSI). CSI is a mean trait community index based on weighted average of habitat specialization of each species present in the community, for more detail on CSI calculation see Supplementary Material 2. The recorded species are all protected species listed in annex IV of the Habitats Directive but they are not target species of the Natura 2000 network (non annex II species, except within the group Myotis spp which may include some species included in annex II but mainly in very slight proportion in our data collection see Supplementary Material 1, Table S1-6).

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Landscape and Natura 2000 data

Around each of the 1608 transects (i.e. the 2-km road portions), we generated a set of landscape variables using QGIS 2.2. Because the landscape effect on bat activity could change according to the spatial scale considered (Grindal & Brigham, 1999; Lacoeuilhe *et al.*, 2016) we calculated these variables at 4 different buffers of 200, 500, 700 and 1000 m. At

1000 m landscape scale, there was 25 % overlap between nearby transects belonging to a same road circuit. However, this still allowed the measurement of variation in landscape characteristics within the same road circuit. However, we could not extend the analyses into larger landscape scales, because the overlapping rate between transect buffers would exceed 50 %. We assessed the proportion of the Natura 2000 areas included in each buffer (shapes were provided by the Muséum national d'Histoire naturelle (http://inpn.mnhn.fr, see Supplementary Material 3). Sites designated for birds (Special Protection Areas, SPA) and/or Habitats directive (Sites of Community Importance, SCI) were considered together as the evaluation deals with the whole network designation effect on non-target bat biodiversity. Note that analyses at the level of each directive (SCI and SPA) did not reveal any obvious difference with the analysis performed at the level of Natura 2000 sites (both together), see Supplementary Material 5, Table S5-3 & S5-4. We also calculated seven land use variables within each buffer, (i) the proportion of intensive agriculture (Inten. Agri., Code 2.1; CORINE Land Cover, including irrigated and nonirrigated arable land such as cereals, leguminous and forage crops.) within each buffer because they represent a major land use pressure for bats (Azam et al., 2016); (ii) the proportion of heterogeneous agriculture (*Heter. agri.*), defined as areas with a complex mosaic of annual and permanent cultures and semi-natural habitats (Code 2.4; CORINE Land Cover); (iii) the proportion of deciduous and mixed forests (*Forests*, Code 3.1; CORINE Land Cover) as they were commonly represented in our buffers and were known to influence bat activity at a landscape scale (Boughey et al., 2011; Frey-Ehrenbold et al., 2013;); (iv) the proportion of open habitat (Open habitat, code 231 and 32 CORINE Land Cover; (v) the total length of streams and water sources (Streams, IGN/2012) because numerous studies have identified aquatic habitat as a favorable where abundance of bat is high (Rainho & Palmeirim 2011; Russo & Jones 2003, Salvarina 2016); (vi) the total length of roads (*Roads*, IGN/2012)

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within each buffer because effect of road network can be considered as a land use pressure, especially regarding landscape connectivity. However, this pressure could not accurately be tested in our study because transects were mostly located in areas with secondary low-traffic roads, therefore, we only took into account the total length of roads as a potential confounding factor of the urbanization. In addition, roads often included hedgerows, which could have a positive effect on bat activity (Verboom & Huitema, 1997; Lacoeuilhe *et al.*, 2016); and (vii) artificial light at night (*ALAN*) defined as the radiant flux (i.e., radiant power) reflected or emitted by a given surface., (see supplementary material 4 for details on the computation of this variable). We used light pollution data as a proxy of the urbanization process as it encompasses both the effects of impervious surfaces and urban-induced pollution that propagates beyond urban areas (Azam *et al.* 2016)

Representativeness of habitats and land uses sampled in the French bat monitoring program

This survey protocol resulted in the collection of data along roads chosen by volunteers, which may introduce a bias in the habitat sampling. Preliminary analysis have underlined that the choice of the circuit performed by volunteers plus the randomization introduced by the MNHN limited the bias in habitat representativeness at local and national scales (see Supplementary Material S1, Table S1-1). Because buildings tend to be concentrated along road networks, the main bias observed was with regard to urban areas and, in particular, impervious areas, which comprises 5% of France. However, our survey included 11% at a 200 m landscape scale, but note that this bias decreases when landscape scale take into account increase (6% of impervious areas in buffer of 1000 m size). We did not detect any bias for the representativeness of Natura 2000 regardless of the spatial scale considered (see Supplementary Material S1, Table S1-1)

A second expected type of bias concerns the presence of hedgerows or forest edges along the roads, which could result in an overall increase in insect availability, as aerial insects tend to concentrate on the lee side of hedgerows (*e.g.*, Lewis & Stephenson, 1966). Therefore, the observation of an over-representation of linear elements in the Natura 2000 areas could indicate a biased prediction of bat activity. Indeed, some bat species tend to forage along these linear elements (Verboom & Huitema, 1997) and fly along such features when commuting from roosts to foraging areas (Limpens & Kapteyn, 1991). Surprisingly, we detected a slight trend for there to be fewer hedgerows within the Natura 2000 area (See Supplementary Material 1, table S1-2, table S1-3). In general, habitats along roads are not a random subset of all the available habitats in the wider countryside. In fact, some rare habitats are most likely underrepresented; in addition, with such road sampling, we mainly sampled edge habitats instead of core habitats, which could result in a bias for some *Myotis* species that preferentially forage in closed environments (see Russo & Jones, 2003; Rainho, 2007) but is most likely to not a major problem for the five other species studied here, which do not select closed environments (Russ & Montgomery, 2002; Rainho, 2007).

Statistical analyses

257 Model building

Initially, we assessed if bat activity measures along transects differ from site to site within or outside a Natura 2000 site (*Natura 2000_PA*, a binomial variable) using Generalized Linear Mixed Model (GLMM). The response variable was either species relative abundance (n=6 taxa) or bat community metrics (total relative abundance, richness or CSI). We add as covariables *the date of survey, the wind speed, the time elapsed after sunset* and *the temperature* because we assumed that bat activity might be affected by weather conditions (Ciechanowski *et al.*, 2007). Note however, that the protocol is performed only when weather conditions are

generally favorable (see Supplementary Material 1, Table S1-1). As these co-variables might have non-linear effects on these co-variables, we included quadratic form terms for the variables (see Supplementary Material 5, S5-1). According to the hierarchical structure of our sampling design (same sites were sampled year to year), we treated *site* and *year* as random effects (Zuur *et al.*, 2009). Thus, our models were structured in the following way:

[bat activity] ~ Natura 2000_PA + date of survey + wind speed + minutes after sunset + temperature+ 1|year +1|site

Check of assumptions of GLMM

Before performing our models, we systematically evaluated the correlations among explanatory variables using Spearman's rho for quantitative variables (Crawley, 2009) to detect obvious correlation (See Supplementary Material 5). Secondly, we performed variance-inflation factors (VIF) on each model (Fox & Monette 1992); all variables had VIF<2 indicating no problem of multicollinearity in the explanatory variables of our models. Close points (even if they differ from habitat) are likely to have a similar bat population density due to similar climatic conditions or large-scale landscape compositions and could therefore imply spatial autocorrelation. Thus, we performed a variogram to observe if correlation between neighbors falls off with distance. When we detected such patterns in our data, we added in our GLMM an autocovariate (i.e., a distance-weighted function of neighboring response values; Dormann *et al.* 2007) with the autocov dist function in R (package spdep, Bivand R. *et al.* 2011) to account for spatial autocorrelation.

In a second step, using the same approach and same model validation (check of spatial autocorrelation, multicollinearity), we assessed the effect of the proportion of Natura 2000

areas at a landscape scale on bat activity taking into account the main land uses. Thus, in addition to Natura 2000 areas, we included seven other landscape variables as fixed effects. These analyses were performed at four scales: 200, 500, 700 and 1000 m. The fixed effects were centered and standardized so that the regression coefficients were comparable in magnitude and their effects were biologically interpretable (Schielzeth 2010). Thus, our models were structured in the following way:

[bat activity] ~ Natura 2000 + Inten. agri + Heter. agri. + Forests + Open habitat + Roads + Streams + ALAN + date of survey + wind speed + minutes after sunset + temperature + 1|year + 1|site

According to the nature of the response variable (bat count) and potential over-dispersion, we chose the best error distribution among Poisson, a negative binomial distribution and zero inflation models with a negative binomial or Poisson error distribution (Zuur *et al.* 2009), and when abundance modelling did not fit well (i.e. *Myotis ssp*), we transformed the response variable (i.e. bat relative abundance) into a presence/absence variable and used a binomial error distribution, (see Supplementary Material 5, Table S5-1).

RESULTS

The survey recorded 23164 bat passes, on which analysis was conducted on the most commonly recorded species: *Pipistrellus pipistrellus* (n = 14967), *Eptesicus serotinus* (n = 2614), *Pipistrellus kuhlii* (n = 2318), (note that even after checking the identifications, we extrapolated that 8% of *P. kuhlii* calls are not entirely certain and could include *P. nathusius* calls), See Table S1 for details), *Nyctalus leisleri* (n = 1999), *Nyctalus noctula* (755) and *Myotis* spp. (261). The first species represented 65% of the dataset.

Without adjusting Natura 2000 effect to take landscape composition into account, P. pipistrellus and E. serotinus exhibited greater relative abundance within Natura 2000 than outside, and Myotis spp. have a greater probability to be recorded within Natura 2000, while we detected no significant effect for the three other species (Table 1). In addition, we detected a greater relative abundance, a greater richness and communities are more specialized within Natura 2000 than outside (Table 1). When taking into account the main land uses within landscape and assessing the effect of the amount of Natura 2000 (i.e. areas of Natura 2000 in the neighbor of the sample site and not just being in a site or off site), we detected a similar relationship to the first set of analyses. Among the six species studied, we were able to detect a positive influence of Natura 2000 in the area bordering transects for *P. pipistrellus*, *E. serotinus* and *Myotis* spp. (Table 2). However, the detection of significant effects was scale dependent, and mainly detected at a small spatial scale (i.e. at 200 m, except for *Myotis* spp). In addition, total bat activity and community bat specialization were positively influenced by Natura 2000 areas, whereas no effect was detected for richness (Table 2). Again, we detected more significant effects at a smaller spatial scale. The proportion of intensive agriculture had a significant negative effect on all species (except N. noctula). We found same results for all bat community metrics whatever the landscape scale considered. Artificial light at night, heterogeneous agriculture and open habitat, when significant, had a negative effect, whereas streams mainly had significant positive effect (except for E. serotinus) but again the effect was scale dependent and varied among species or bat community metrics (Table 2, Fig. 1). Forest effects depend of bat activities measured: for example P. pipistrellus was positively influenced by the proportion of forest while N. lesileri was negatively influenced, total bat relative abundance positively influenced, while richness and community specialization were negatively influenced by forest (Table 2, Fig. 1).

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'Designation effect' of Natura 2000 areas for bats.

This paper provides evidence that bat activity is globally higher in Natura 2000 sites (+24%;). Three (P. pipistrellus, E. serotinus, Myotis spp) of the six species/genera studied exhibit greater relative abundance in Natura 2000 sites (this increase is noteworthy for E. serotinus (x 2.1) and Myotis ssp (x 3.6)) and note that no significant negative effect was detected. The result for Myotis spp must be taken with caution; it did not imply that all species of this genus exhibit a similar pattern, but at least one species exhibits a greater bat activity in Natura 2000 sites. We also detected a greater richness in Natura 2000 sites (+ 14%). Finally, the bat community specialization, a mean trait community index, is greater in Natura 2000 sites (+9%).In a second step, we evaluated if this Natura 2000 effect was consistent when, in the neighborhood of the survey, the proportion of Natura 2000 was adjusted to the land uses. For P. pipistrellus, E. serotinus and Myotis spp, the increase of Natura 2000 in the surrounding area positively influenced their activity but the effect was spatial scale dependent. Similarly, bat community specialization, increased when Natura 2000 areas increased in grea surrounding transects. However, we did not detect any effect of the proportion of Natura 2000 areas in the surrounding of transects on species richness. Note that this latter non-detection is possibly linked to the nature of data collected (small sequence of acoustic recordings) which currently did not allow species identification within the *Myotis* group, while this group is very rich in the region considered (39% of the French species, TAXREF, Gargominy et al., 2016-). With the recent arrival on the market of new generation of bat detector-recorders, we could consider recording throughout the entire night (see Newson et al., 2015) and thus expect an

increase in the probability of contacting and identifying *Myotis* species. A protocol using this new technology is under development by the FBMP (Bas *et al.*, 2017), thus a more accurate assessment of Natura 2000 effect on richness could be expected in the future.

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The positive Natura 2000 effect appears to be relatively small compared to the main land use pressure: intensive agriculture and to a lesser extent heterogeneous agriculture (while this category is known to include more extensive agriculture practice) and light pollution (ALAN) all of them have a deep negative impact. They impact not only the abundance of populations but also the structure of communities, leading to less rich and more generalist communities, thus contributing to a biotic homogenization process (McKinney, 2006, Karp et al., 2012). These drivers are known to impact negatively bats (Wickramasinghe et al., 2004; Azam et al. 2016) and more broadly biodiversity, both rare and endangered species (Maxwell et al., 2016) as the common species and (plants: moth: Conrad et al. 2006; butterflies: Van Dyck et al. 2009; bees: Kosior et al., 2007; birds: Donald. 2001) and contribute to large scale biotic homogenization (Le Viol et al. 2012). The negative effect of open habitat is mainly due to the requirement of the species studied to avoid large open habitat (Verboom, & Spoelstra, 1999) and select wooded edge, woodland or aquatic habitat (Walsh & Harris, 1996). The positive effect of streams is (although detected for many species or bat community measures) in accordance with many studies see the review perform by Salvarina (2016). The contrasting effect of forest areas on the species studied (dominated by aerial hawking species), positive effect on total relative abundance, while negative effect on richness and community specialization, seem to indicate that these land uses, when adjusted for the effect of Natura 2000, does not have a broad effect. Zehetmair et al., (2015), in a sample of European beech forest, found that bat activity did not differ significantly between the Natura 2000 and non-Natura 2000 forest on paired comparison of stands with same structural attributes. They concluded that the current management of the Natura 2000 beech forests is almost identical to

that of non-Natura 2000 commercial forests, and thus, the Natura 2000 status has not led to an increase of bat-relevant habitat variables yet. By comparing similar beech forest habitats, Zehetmair *et al.*, (2015) may have missed the fact that the Natura 2000 network may also include a higher proportion of high quality habitats than usual non Natura 2000 forests. On the contrary, our study design (including a great number of sites and, in turn, a wide range of forest types) enables us to test for a more global Natura 2000 designation effect, which remains even after taking landscape variables into account.

When the road effect is detected, it is surprisingly positive and appears to invalidate this landscape variable as a pressure on bat foraging connectivity. Lisón & Sánchez-Fernández (2017) found that the roadless areas are not significant for the localization of bat hotspots. It should be noted however, that we could not test accurately a "road effect" because our sampling is not representative of national road network: due to security, and transects were mostly located in areas with secondary low-traffic roads. Thus, within this biased sample, we probably detected an effect of availability of hedgerows along roads, where hedgerows are positively selected for by bats (Lacoeuilhe *et al.*, 2016). The increase in relative abundance and degree of specialization may appear limited compared to the negative effect of intensive agriculture (respectively lower by a factor 6 and 2, Table 2). However, it is still notable that for common, widely distributed species, that there is still a Natura 2000 effect, after we control for land use variables like streams that are known to have a positive influence on bats (Salvarina, 2016).

It appears that the areas designated as Natura 2000 areas exhibit a 'designation effect' and providing evidence that such areas are most likely to be of better quality for bats with regard to their foraging activity. Similar results have been found by Lisón *et al.*, (2015) in Spain, a study involving bat distribution and not bat abundance. Indeed, the Natura 2000 areas contain less intensive agriculture areas (Supplementary material Table S1-3) and more semi-

natural areas and harbor more common biodiversity than non-Natura 2000 areas (Pellissier *et al.*, 2013). This non-random habitat composition of Natura 2000 sites is part of the raw "designation effect" of this network. Interestingly, when this habitat "bias" is partially taken into account because the effect of Natura 2000 was adjusted to the main land use types, a significant positive effect of Natura 2000 remains for most metrics. One possible interpretation is that Natura 2000 have, on average, better structural and more micro-habitats features or food webs important for bats than the same broad habitat types outside the network.

Scale and time effect

When looking at the influence of spatial scale, the positive effect of Natura 2000 tends to be more significant at a smaller scale (i.e. 200 m) while some land uses variables remain significant whatever the spatial scale considered (i.e. intensive agriculture, ALAN). This finding seems to indicate that Natura 2000 areas do not yet enhance adjacent areas at a large scale for common bats. However, a theoretically and empirically demonstrated reserve effect is the spillover of species richness and community complexity that highlights the important benefit for biodiversity and ecosystems of establishing reserve areas (Roberts *et al.* 2001, Russ & Alcala, 2011). However, many studies have underlined that reserves could enhance adjacent areas, but that age matters (Claudet *et al.*, 2008, Russ & Alcala, 2011). It should be emphasized that the purpose of this paper is not to assess the efficiency of the management measures actually occurring in the Natura 2000 areas; here we only assess a designation effect. Indeed, management is probably too recent to be able to produce measurable effects, because during the period studied (2006-2013) only 53% of Natura 2000 sites had endorsed their management plan, and moreover 69% of recordings were collected between 2006 and 2009 (see Supplementary Material 1, Fig S1-1). We hypothesize that effect of Natura 2000 at

large landscape scale should become more significant in the future with the effects of Natura 2000 sites management and the implementation and green infrastructure policies, which aim to connect patches of protected areas. Future research should now focus on comparing biodiversity state before and after the designation of N2000 sites (see Lisón & Sánchez-Fernández, 2017), especially since conservation policy may moderate land-cover transformation (Kallimanis *et al.*, 2015).

Car-based bat survey and policy implications

The French bat monitoring program has proven to be a useful tool for evaluating the Natura 2000 network owing to its large spatial coverage and its standardized monitoring based on volunteers. This finding emphasizes that public participation in scientific research is a promising approach that can significantly help scientists to address questions relating to biodiversity, as suggested by Bell *et al.*, (2008) and Couvet *et al.*, (2008). Volunteer-based monitoring is likely to have high resilience (Couvet *et al.*, 2008), which is key to maintaining the monitoring program over time and, thus, providing extended time-series (Battersby, 2010). However, questions can be raised about more efficient ways to monitor biodiversity of a protected network, between surveying rarer species and habitats or to survey more common and widespread elements not targeted during the designation process. Indeed, rare species (due to lower abundance, lower detection, or their specialization to particular habitats) need focused sampling (Barlow *et al.*, 2015). From a financial point of view, it should be noted that the monitoring of common species does not involve funds dedicated to Natura 2000 surveys.

Conclusion and conservation implications

Our study showed a sampling effect induced by the designation process of Natura 2000. The species studied are only listed in the Annex IV of the habitat directive (not Annex

II), despite the fact that they benefit from a strict protection regime, they are not target species for the designation of Natura 2000 areas. In addition, in the meta analysis performed in Spain, Lisón et al., (2017) found that bats were not "emblematic" species for the managers, especially the bat species of Annex IV. However, they remain policy relevant because they are species of European concern and it is a highly valuable feature the Natura 2000 network benefits common biodiversity. This result is congruent with many studies that assert that the establishment and management of statutory protected areas are widely regarded as key strategies and efficient tool for safeguarding key biodiversity areas (Jackson et al., 2009; Watson et al., 2014). Although gap analysis have clearly demonstrated that the global protected area network is still far from complete for encompassing broader biodiversity (Rodrigues et al., 2004), our results suggest that Natura 2000 is able to encompass nontargeted species, at higher level of relative abundance than outside the same landscapes. As this network is the most important conservation effort in Europe (Evans, 2012), it is important to recognize that its biodiversity coverage extends beyond the rare or threatened targeted species and habitats and potentially promote management measures towards this more common part of biodiversity and its functional role.

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Additionally, most of the Natura 2000 areas designated for the protection of bat species are limited to roosting sites and immediate surroundings. Indeed, for species with a complex life cycle, reproduction is most often considered the most important part of the cycle, and, thus, the protection of reproduction sites is emphasized (Stebbings, 1988). However, whilst sites for bat reproduction are restricted in space, they have a large foraging territory, even during reproduction (Stebbings, 1988). Therefore, protecting hunting territories, which can be quite large and far from roosting sites, is of prime importance from a conservation point of view (Vaughan *et al.*, 1997). In this study, we assessed the influence of Natura 2000 areas on measures of foraging activity for six common Chiroptera species and three measures

490 of community. Given the trophic level occupied by all the Chiroptera species in Europe (top 491 predators), the implications of our results may extend beyond the taxa considered and have 492 important implications for the conservation of biodiversity as a whole. 493 494 Acknowledgments 495 We deeply thank the Vigie Nature volunteers for data collection. We also thank Gino 496 Nearns and Julie Pauwels for an earlier reading of our manuscript and two anonymous 497 referees for their relevant and useful comments. 498 499 **REFERENCES** 500 Azam, C., Le Viol, I., Julien, J.-F., Bas, Y., Kerbiriou, C. 2016. Disentangling the relative 501 effect of light pollution, impervious surfaces and intensive agriculture on bat activity 502 with a national-scale monitoring program. Landscape Ecol. 31, 2471–2483. 503 http//dx.doi.org/10.1007/s10980-016-0417-3 504 Barataud, M. 2015. Acoustic Ecology of European Bats, Species identification, study of their 505 habitat and foraging behavior. Biotope, Mèze; Muséum national d'histoire naturelle, 506 Paris (Inventaires et biodiversité Series), 352 p. 507 Barlow, K.E., Briggs, P.A., Haysom, K.A., Hutson, A.M., Lechiara, N.L., Racey, P.A., 508 Walsh, A.L., Langton, S.D., 2015. Citizen science reveals trends in bat populations: 509 The National Bat Monitoring Program in Great Britain. Biol. Conserv. 182, 14-26. 510 Bas, Y., Bas, D., Julien, J.F. 2017. Tadarida: A Toolbox for Animal Detection on Acoustic 511 Recordings. Journal of Open Research Software 5, 6. http://dx.doi.org/10.5334/jors.154 512 Battersby, J. 2010. Guidelines for surveillance and monitoring of European bats. EUROBATS 513 Publication Series, 51–95. 514 Bell, S., M. Marzano, J. Cent, H. Kobierska, D. Podjed, D. Vandzinskaite, H. Reinert, A. 515 Armaitiene, M. Grodzińska-Jurczak, and R. Muršič. 2008. What counts? Volunteers and 516 their organisations in the recording and monitoring of biodiversity. Biodivers. Conserv.

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Table 1. Partial regression coefficient of the raw Natura 2000 effect for the 3 community metrics and 6 taxa.

Bat activity metric	Natura 2000 effect		
	Estimate	P-value	Factor of variation
Relative abundance	$\beta = 0.2195 \pm 0.035$	P < 0.001	x 1.24 number of bat passes/2km transect
Richness	$\beta = 0.130 \pm 0.052$	P = 0.010	x 1.14 number of species/2km transect
CSI	$\beta = 0.061 \pm 0.020$	P = 0.002	x 1.09 the degree of community specialization
P. pipistrellus	$\beta = 0.186 \pm 0.041$	P < 0.001	x 1.14 number of passes/2km transect
P. kuhlii	$\beta = 0.113 \pm 0.128$	P = 0.378	
E. serotinus	$\beta = 0.721 \pm 0.090$	P < 0.001	x 2.05 number of passes/2km transect
N. noctula	β = -0.346 ± 0.387	P = 0.371	
N. leisleri	$\beta = 0.272 \pm 0.254$	P = 0.280	
Myotis spp.	$\beta = 1.372 \pm 0.209$	P < 0.001	x 3.68 occurrence of contact/2km transect

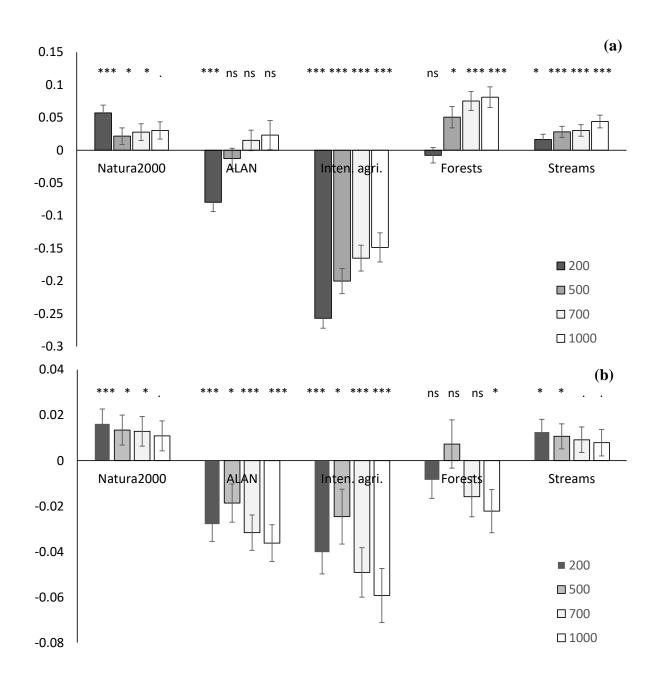
Table 2. Standardized partial regression coefficients from GLMMs model for the 8 landscape effects included in the analysis (i.e. the proportion of the Natura 2000 (Natura 2000), intensive agriculture (*Inten. Agri.*), heterogeneous agriculture (*Heter. agri.*), forests (*Forests*), open habitat (*Open habitat*) the total length of primary roads (*Roads*) and streams (*Streams*) and the average luminance (ALAN)), for the 3 community metrics and 6 taxa and at 4 landscape scales considered. P-value level of significance associated are '*** P < 0.001; '** P < 0.01; '*' P < 0.05; '.' P < 0.1

	Scale	Natura 2000	Inten. agri.	ALAN	Heter. agri.	Forests	Open Habitat	Roads	Streams
Community	metrics	7							
	200	0.057***	-0.257***	-0.079***	-0.081***	-0.008	-0.110***	0.0317***	0.016*
Relative	500	0.022*	-0.200***	-0.013	-0.029*	0.051*	-0.047**	0.029***	0.028***
abundance	700	0.028*	-0.165***	0.015	-0.009	0.075***	-0.028.	0.024**	0.030***
10	1000	0.030.	-0.149***	0.023	-0.010	0.081***	-0.033	0.031***	0.044***
	200	0.002	-0.206***	-0.083***	-0.039.	-0.045*	-0.071**	0.010	0.020
n: I	500	-0.006	-0.157***	-0.055*	-0.001	-0.001	-0.026	0.010	0.032*
Richness	700	-0.009	-0.204***	-0.080***	-0.039.	-0.048*	-0.064**	0.005	0.034*
	1000	-0.013	-0.217***	-0.089***	-0.047*	-0.061*	0.076**	0.015	0.031.
	200	0.016***	0.040***	-0.028***	-0.042***	-0.009	-0.030***	0.017*	0.012*
CSI	500	0.013*	-0.025*	-0.019*	-0.029**	0.007	0.019.	0.011*	0.010*
	700	0.013*	-0.049***	-0.031***	-0.0503***	-0.016.	-0.039***	0.015***	0.009.

	1000	0.011.	-0.059***	-0.036***	-0.053***	-0.022*	-0.047***	0.017***	0.008
Taxa									-
	200	0.038**	-0.219***	-0.087***	-0.073***	0.028.	-0.109***	-0.007	0.027*
D	500	0.014	-0.175***	-0.046**	-0.021	0.068***	-0.043*	-0.042***	0.034**
P. pipistrellus	700	0.026 .	-0.152***	-0.027.	-0.004	0.078***	-0.028	0.004	0.031**
	1000	0.043**	-0.174***	-0.027.	-0.021	0.050*	-0.072**	0.009	0.048***
	200	0.069 .	-0.328***	-0.169***	-0.029	-0.150***	-0.037	0.078***	-0.036
P. kuhlii	500	0.009	-0.255***	-0.120 .	0.062	-0.014	0.078	0.072***	-0.016
Р. кипш	700	-0.027	-0.300***	-0.148*	0.019	-0.022	0.048	0.052***	0.007
	1000	-0.061	-0.370***	-0.204**	-0.006	-0.030	0.021	0.045*	0.037
	200	0.081**	-0.706***	-0.353***	-0.346***	-0.062***	-0.346***	0.089***	-0.121***
E. serotinus	500	0.036	-0.436***	-0.140*	-0.190***	0.207***	-0.182***	0.116 *	-0.061*
	700	0.026	-0.703***	-0.411***	-0.357***	0.007	-0.345***	0.077***	-0.059*
	1000	0.029	-0.722***	-0.297***	-0.397***	0.013	-0.441***	0.099***	-0.023
	200	-0.001	0.220	0.121	-0.486***	-0.235 .	-0.676***	0.025	0.153
N. maatula	500	-0.001	-0.242	0.043	-0.556***	-0.319 .	-0.807***	0.022	0.204*
N. noctula	700	-0.038	-0.197	0.0001	-0.678***	-0.204	-0.812***	0.175 .	0.212*
	1000	-0.028	-0.312	-0.060	-0.830***	-0.297	-0.996***	0.203*	0.167
	200	0.121	-0.600***	-0.161	-0.463***	-0.286**	-0.408***	0.065	0.198**
N. leisleri	500	0.133	-0.417**	-0.027	-0.390***	0.098	-0.329*	0.004	0.188**
	700	0.117	-0.571***	-0.137	-0.585***	-0.277*	-0.475***	0.067***	0.190**

	1000	0.093	-0.598***	-0.153	-0.638***	-0.320**	-0.551***	0.106 .	0.207**
	200	0.234***	-0.505***	-0.208*	-0.112	-0.070	-0.150 .	0.084 .	0.092 .
Mustia aan	500	0.214**	-0.526***	-0.109	-0.074	-0.068	-0.078	-0.097	0.130*
Myotis ssp.	700	0.225**	-0.527***	-0.081	-0.076	-0.067	-0.077	0.093 .	0.114**
	1000	0.213**	-0.553***	-0.062	-0.064	-0.056	-0.049	0.106 .	0.056

Fig.1 Standardized partial regression coefficients and associated standard errors from GLMMs model for the proportion of the Natura 2000 ($Natura\ 2000$), intensive agriculture ($Inten.\ Agri.$), forests (Forests), streams (Streams) and average luminance (ALAN) for the total relative abundance (a) and Community Specialization Index (b). The gray gradients correspond to the different buffer sizes. P-value level of significance associated are '*** P < 0.001; '** P < 0.01; '** P < 0.05; '.' P < 0.1.



Supplementary Material 1: Detailed information on the French Bat Monitoring Program (FBMP)

Table C1 1. Characteristics of the protocol	and sampling design the two bat surveys
Table 31-1. Characteristics of the brotocol	i aliu saliibiilig uesigii tile two bat sui vevs

Aim of the program	the protocol and sampling desi	ds of bat populations at a nationa				
Ailli of the program	scale	is of bat populations at a flationa				
Car transect survey sampling pro						
Scope	10 km around surveyors' home.					
Circuit length	30 km					
Number of transect per circuit	10					
Transect length	2 km separated of at least 1 km					
Period of sampling	from the 15 th of June to the 31 th of July and 15 August to 3					
. eea oj eapg	September					
Weather conditions	no rain, low wind speed (< 7 m/s), temperature > 12°C					
Survey start	as possible 30 minutes after sunset					
Count point survey sampling pro	-	•				
Scope		sen (by the Museum) in a radius o				
300pc	10km from the observer's home, (i	.e. on average one square randomly				
	chosen between 80 possible square	es).				
Circuit length	square of 2km-side					
Number of point per circuit	10					
Recording duration	6 minutes					
Period of sampling	from the 15 June to 31 July and 15	=				
Weather conditions	no rain, low wind speed (< 7 m/s), temperature > 12°C					
Survey start	as possible 30 minutes after sunset					
Bat recording characteristics						
Acoustic detectors	Tranquility Transect Bat detector&	D240x				
Intercalibration of detectors	At the MNHN					
Acoustic settings	Tranquility Transect	D240x				
Suppliers	Courtpan Design Ltd, UK	Pettersson Elektronik				
High pass filter	5 kHz	18 kHz				
Frequency	96 000 sample/sec	96 000 sample/sec				
Recording device	Zoom H2 digital recorder (Samson technologies, USA)					
File storage format	WAV					
Bat identification						
Software	Syrinx 2.6					
Procedure	 Training: 2-day training course Bat first identification: by vol 	unteers				
	- Bat identification validation:					
Taxon identification level	Species level except for <i>Myotis sp.</i> Extensive data expertise evaluated					
	that <i>Pipistrellus kuhlii</i> may potentially include 8 %					
	Pipistrellus nathusius, as these two species overlap in their acoustic					
	signatures.					
Meteorological data	- Temperature (°C) and cloud cov 50%, 50-75%, 75-100%) were survey.	ver (% in four classes: 0-25%, 25 recorded by volunteers during the				
		ure was not used in the analyses				
	However we used this field	this measure to confirm the				
	appropriateness of the use of	of the wind speed measure provid-				
	by the closest meteorological stat	tion.				
		he public archives the web sit				
	Infoclimat [http://www.infoclima					
Habitat data	Volunteers were involved in					
	a detailed and adapted hiera	rchical system (see hereafter)				

Figure S1-1: Example of one road circuit composed of 10 transects

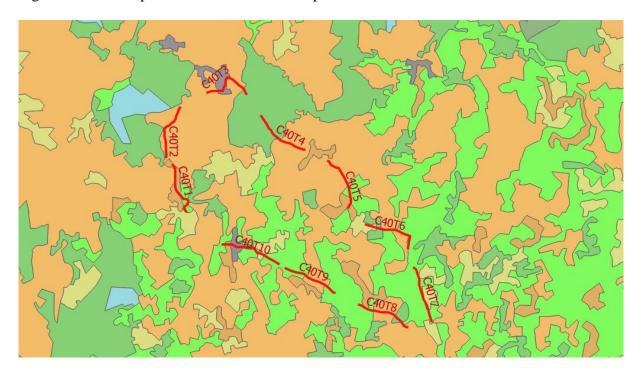
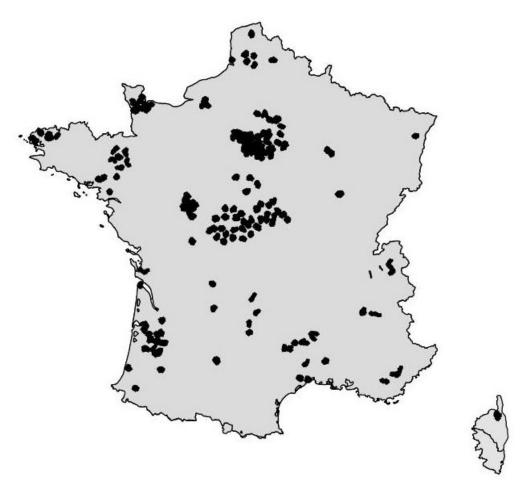


Figure S1-2: Distribution of car transect surveys of the French national bat monitoring program



1. Assessment of potential biases in habitat sampling

We estimated biases occurring in our monitoring scheme at local and national scales. First, we compared the proportion of different habitats in the 10 km radius around observers houses in relation to the proportion within 200 meters buffer around each 2 km transect. Second, we compared to the proportion of different habitats at the French scale to the proportion in 200 meters buffer around each 2 km transect We used Corine Land Cover data base. With one such sampling design, habitats surveyed are representative of the habitat at the local scale (R^2 =0.96; Fig 2-S1) and the national scale (R^2 =0.95; Fig 3-S1). The correlation is quite good especially for the commonest habitat and as expected the major difference occurred with rare habitats. The main bias is towards urban areas and particularly discontinuous artificial surfaces which in a 10 km radius around volunteer's house, represented 5% of habitat while in the sampled transect the proportion of this habitat reached 10%. The difference is similar when comparing the national habitat proportion to sampled habitat (3% vs 10%)

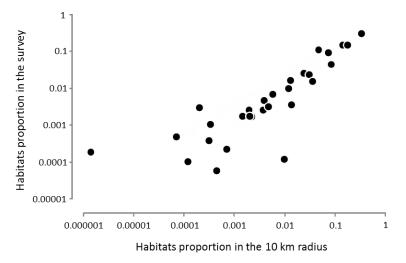


Fig. S1-3: Correlation between habitat proportion at local scale (10 km radius) and proportion sampled in the bat survey. Proportion of Corine Land Cover classes are log transformed.

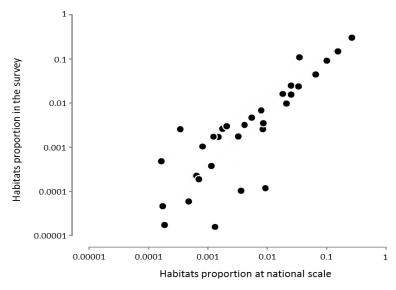


Fig. S1-4: Correlation between habitat proportion at national scale and proportion sampled in the bat survey. Proportion of Corine Land Cover classes are log transformed.

The coverage of our 160 road circuits representing 1608 different transects was largely representative of French landcover (Table S1-2).

Table S1-2: The land-cover variables surrounding each transect at a range of spatial scales (200–1000 m), as well as their representativeness in mainland France Landscape. The variables are the proportions of Natura 2000 (*Natura 2000*), intensive agriculture (*Inten. agri.*), heterogeneous agriculture (*Heter. agri.*), deciduous forests (*Forests*), *Open Habitat* and mean length per hectare of roads (*Roads*) and streams (*Streams*) and the Artificial Light At Night (ALAN), The range represents the minimum and the maximum values encountered for each variable around transects at 1000m landscape-scale (similar range were observed for the 3 other landscape scales tested). Proportion of Natura 2000 were similar among landscape-scale, however a slight significant positive trend were detected (*P*=0.03).

Landscape	Continental	Transects	Transects	Transects	Transects	Range
Variables	France	(200 m)	(500 m)	(700 m)	(1000 m)	(min-max)
	12.5	12.1	12.7	13.1	13.3	0-100
Inten. agri. (%)	28.1	32.5	34.0	34.3	34.9	0-100
Heter. agri. (%)	11.0	12.9	11.5	11.8	11.4	0-92
Forests (%)	19.9	18.6	24.7	20.7	21.2	0-100
Open habitat (%)	18.5	15.9	15.7	15.7	16.3	0-99
Imper. surf. (%)	5.2	11.3	8.74	7.3	6.5	0–92
Roads (m)	1.92	12.8	6.8	5.4	4.3	0-9391
Streams (m)	6.67	12.9	10.5	9.8	9.1	0-
streams (m)	0.07	12.9	10.5	9.0	9.1	22262
ALAN	3.2	2.2	2.2	2.1	2.1	0.1-35

No obvious correlation (rho|≥0.5; Freckleton, 2002) was detected between landuse variables and Natura2000 (Table S1-3)

2. Assessment of potential bias of habitat characteristics between sampling within and outside Natura 2000 areas.

Because foraging activity of bats is not only influenced by the type of natural habitat type but also by small scale habitat characteristics in the agricultural landscape, such as the presence of linear elements like hedges (Limpens *et al.*, 1989; Krusic *et al.*, 1996; Verboom & Huitema, 1997; Downs & Racey, 2006), we have attempt to evaluate possible bias between sampling areas surveyed within and outside Natura 2000 areas. In forest habitat, we evaluated potential bias linked with the recording position: recording along forest paths or at forest edge.

2.1. Methods

2.1.1. Data

In the French national bat monitoring volunteers were involved in collecting habitat variable on a detailed and adapted basis. Habitat information is recorded on the first reconnaissance visit to the circuit. The appropriate habitat codes are chosen from an established hierarchical system which is very similar to widely use habitat code in bird monitoring schemes (see Crick 1992 for the UK and Barnagaud *et al.*, 2012 for the French monitoring), but adapted to take account of bat foraging specifics and particularly linear element (for more details about the French Bat monitoring see the website [http://vigienature.mnhn.fr/page/releves-d-habitats]).

Volunteers are invited to record habitat characteristic every 400m along 2km transects. Currently more than 55% of circuits have been described by volunteers.

2.2.2. Statistical analysis

We assess the presence of potential sampling bias among areas within Natura 2000 designation or without using Generalized Linear Model (GLM) with a quasi-binomial error distribution.

<u>In agricultural habitat</u>, we test the possible difference of linear element within or without Natura 2000 areas according to agricultural habitat classes (Heterogeneous agricultural areas, Arable land, Pastures, Vineyards & orchard). We detect a slight trend of lower proportion of hedgerow within an Natura 2000 area (Table S1-3), not however that except for the Arable land the differences are quite weak.

Table S1-3: Probability to encounter a hedgerow, in the collected samples, according the fact being within or without a Natura 2000 area.

	Effect	P-value	Probability
Arable land (n=454)	β=-1.460	P<0.0001	Within Natura 2000 area: 0.09
			Without Natura 2000 area: 0.28
Heterogeneous agricultural areas	β=-0.850	P<0.0001	Within Natura 2000 area: 0.79
(n=811)	·		Without Natura 2000 area: 0.90
Pastures (n=643)	β=0.377	P=0.11	Within Natura 2000 area: 0.85
			Without Natura 2000 area: 0.89
Vineyards & orchard (n=63)	β=0.000	P=1	Within Natura 2000 area: 0.50
,	•		Without Natura 2000 area: 0.50

<u>In forest habitat</u>, we evaluate the existence of potential bias of recording position (in forest path or in forest edge) among areas within Natura 2000 designation or without. This effect was adjusted to forest habitat classes (Broad-leaved forest, Coniferous forest, Mixed forest dominant broad-leaved, Mixed forest dominant coniferous). Except for the Coniferous forest (note the small sample) the differences are quite weak, even if statistically significant (table S1-4).

Table S1-4: Probability to being located in a forest path instead a forest edge, in the collected samples, according the fact being within or without a Natura 2000 area.

	Effect	P-value	Probability
Broad-leaved forest (n=989)	β=0.465	P=0.009	Within Natura 2000 area: 0.82 Without Natura 2000 area: 0.75
Mixed forest dominant broad- leaved (n=572)	β=-0.603	P=0.04	Within Natura 2000 area: 0.87 Without Natura 2000 area: 0.92
Coniferous forest (n=55)	β=-16.99	P=0.9	Within Natura 2000 area: 0.14 Without Natura 2000 area: 0.66
Mixed forest dominant coniferous (n=263)	β=-1.331	P=0.02	Within Natura 2000 area: 0.86 Without Natura 2000 area: 0.96

3. Device fastener

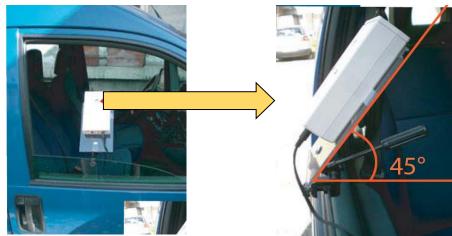


Fig S1-1: arrangement of the detector. The detector was fixed to the passenger-side window at a 45° angle so that the distance to road edges was similar all along the circuits

Recording characteristic

Bats calls were detected using a Tranquility Transect Bat detector (Courtpan Design Ltd, UK) and D240x (Pettersson Elektronik) and recorded on a Zoom H2 digital recorder (Samson technologies, USA). Before used all detector are calibrated at the French National Museum History in order to obtain the same level of sensibility. Signal from the condenser microphone of the bat detector was internally amplified and high pass filtered with a 5 kHz corner frequency and fed to the Zoom H2 which recorded it at 96 000 sample/s as PCM files. Sound was stored on Secured Card in Waveform Audio File Format more commonly known as WAV format, a format without compression. Tranquility Transect set to record 0.32 s of sound at x10 time expansion. Each recorded 0.32 s is followed by 3.2 s of playback, during which time the detector does not record. For the D240x the most similar storage time is 0.1 seconds of sound instead of 0.32, thus we performed a posteriori elimination of time expansion sequences that were spaces of less than 3.2 seconds. All ultrasound detectors were previously calibrated by the French National Museum of Natural History in order to have the trigger level.

Bat identification

Species acoustic identification was done by volunteer surveyors using Syrinx software version 2.6 (Burt 2006), followed by an identification validation by Museum experts. This approach allows identification difficulties to be highlighted, also volunteers become expert through training sessions (each year the museum offers free training for volunteers) and through some kind of self-training thanks to the corrected identification forms we send back.

Table S1-6: Identification error rate of volunteers after one training session

Pipistrellus pipistrellus	1.6 % (± 0.5)	
Pipistrellus kuhlii	7.8 % (± 1.9)	
Nyctalus leisleri	6.4 % (± 1.5)	
Eptesicus serotinus	$9.7 \% (\pm 2.7)$	

Identifications were analyzed call by call and in the case of ambiguous call patterns, we encourage volunteers to use Scan'R (Binary Acoustic Technology, 2010) to isolate each bat vocalization and automate the measurement of relevant parameters (Russ 1999; Barataud

2012; Gannon *et al.*, 2004; Obrist *et al.*, 2004). The main relevant parameters used were: call duration (msec), time elapsed since between calls (msec), maximum frequency detected (Fmax, KHz), minimum frequency detected (Fmin, KHz), total bandwidth [Fmax - Fmin] (KHz), frequency at strongest sound pressure level (KHz), location of dominant Frequencies (% of total duration), high end of characteristic (KHz) [Similar to Fk], low end of characteristic (KHz) [Similar to Fc], global slope of the call (KHz per msec), time of the heel or High Fc (percent of duration), upper slope [start to High Fc] (KHz per msec), lower slope [High Fc to Low Fc] (KHz per msec), fundamental frequency (KHz), 2nd harmonic frequency (KHz), curvature measurement as a way to characterize the shape of bat calls, curve fit error parameter; a measurement of how much error exists between the curvature model and the actual shape of the call.

Each contact was assigned to the species level, however due to the lack of general consensus for some species about the acoustic criteria and some overlap between acoustic repertoires, it was not possible to assign the exact species with certainty for all calls; thus

- (i) we constructed a group for species from the *Myotis* genus. Note that some species included in the same group may have different ecology, however, from a foraging behavior perspective, these groups have some convergences. The *Myotis* group primarily included species considered to be gleaners, which capture the majority of their prey from substrates in cluttered environments (Arlettaz *et al.*, 2001). These species eat mainly diurnal brachyceran Diptera and non-volant arthropod such as weevils, lepidopteran larvae, harvestmen and spiders (Dietz *et al.*, 2007). In addition note that the group *Myotis* exhibit avoidance behavior to artificial light points such street lamp (Lacoeuilhe *et al.* 2014; Azam *et al.*, 2015). Among this *Myotis* ssp. group, the majority of *Myotis* identified at the species level were *Myotis daubentoni*.
- (ii) we estimate that among *P. kuhlii* may potentially include some *P. nathusii*, indeed these two species have some overlap between acoustic repertoires. Extensive validations with experts on more than 58% of the whole data indicate that probably less than 8% of the *P. kuhlii* identifications are not entirely certain and that it resides a small probability that it can be alternatively be *P. nathusius*. Note that these two species forage mostly on flying prey in open spaces (Dietz *et al.*, 2007) and are "attracted" by artificial light points such street lamp (Lacoeuilhe *et al.* 2014, Azam *et al.* 2015).

Table S1-7: composition of the *Myotis* spp category

Species	%
Myotis alcathoe	0.1
Myotis bechsteinii	0.2
Myotis brantii	0.6
Myotis capaccini	0.0
Myotis daubentoni	34.7
Myotis emarginatus	0.5
Myotis myotis	1.8
Myotis mystacinus	1.9
Myotis nattereri	1.8
Myotis puniclus	0.0
Myotis sp	58.3

Bat activity measure

We stored the same bat signal with the two possible outputs of the detector: time expansion (t_{exp}) and high frequency (t_{hf}) . Time expansion is the format widely used from the beginning of the FBMP and bat activity index (name in this study relative abundance) is built with this format. It is often possible to discern echolocation pulses from more than one individual bat in one sound interval, based for example on differing frequencies of maximum energy (F_{max}) and inter-pulse intervals between echolocation calls. Time expansion is used from the Bat Ireland monitoring (Roche et al. 2011) and in numerous countries through the *ibats* programs. With time expansion and with our setting each recorded 0.32 s is followed by 3.2 s of playback, during which time the detector does not record. High frequency outputs, is a continuous sound recording that allow considers the duration of bat call passes. Thus this measure is certainly a best descriptor of bat activity, however identification on this format it is very time consuming and require greater expert knowledge. Volunteers are not requested to identify high frequency format and thus very few identification of the FBMP come from this format. This format was used to assess bat pass duration (Fig S1-2).

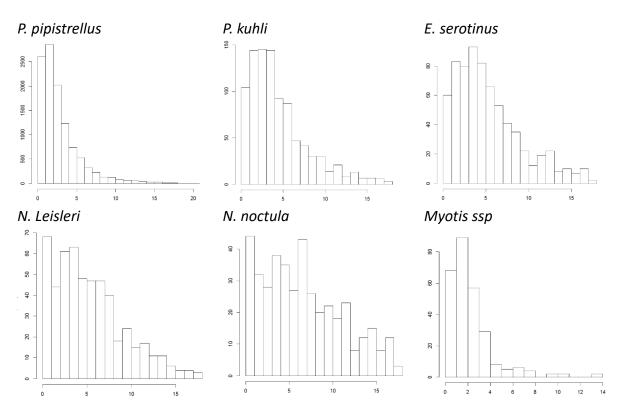
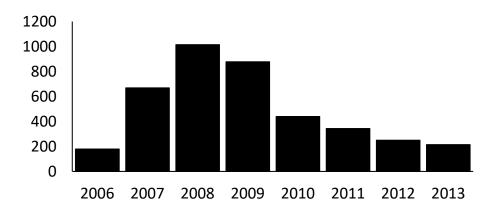


Fig S1-2: Distribution of bat passes duration from the car transect survey (vertical axis are the number of bat pass)

With the bat detector setting, (each recorded 0.32 s is followed by 3.2 s), a detector is able to detect the following contact only 3.52s after the previous contact. This following contact could be either the same individual or another individual. However, according that (i) the car rolls at 25Km/h at the time of the recordings, 3.52 s mean that car is 25meters away (ii) the bat pass duration distribution (see Fig S1-2) and (iii) the distance of detection of bat (see Barataud 2015), we hypothesize that multiple counts of the same individual may be rare and concern mainly *Nyctalus spp* and E. serotinus.

Variation of the sampling between years

Fig S1-3 .Number of available transects surveyed per years



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Supplementary material 2: Detailed information on the calculation of specialization indices

In the French national bat monitoring volunteers were involved in recording habitat. Habitat information is recorded on the first reconnaissance visit to the circuit. The appropriate habitat codes are chosen from an established hierarchical system allowing describe more than 950 habitat type, which is very similar to widely use habitat code in birds monitoring schemes (see Crick 1992 for the UK and Barnagaud *et al.*, 2012 for the French monitoring), but adapted to take into account bat foraging specificities and particularly linear element (for more details about the French Bat monitoring see the website [http://vigienature.mnhn.fr/page/releves-d-habitats]). Habitat classes are collected in a radius of 100m around the sampled point and grouped into 18 classes in order to obtain sufficient number of sample per class (for more methodological information see Julliard *et al.*, 2006; DeVictor *et al.*, 2008; Kerbiriou *et al.*, 2010).

Specialization species index (SSI)

We quantified the species specialization index (SSI), which is the degree of habitat specialization for a species, as the coefficient of variation (SD/mean) of its densities across habitats following Julliard *et al.*'s approach (2006). SSI is thus independent to species habitat preferences. However, it is also possible to assess the species specializations to forest habitats by calculating the average density of a species inventoried in forest habitats divided by the average density in non-forest habitats.

Table S2-1: Habitat Specialization Index, NA indicates that data are insufficient for this species to allow a robust estimate of SSI.

Bat species	Habitat Specialization Index
Barbastella barbastellus	3.64
Eptesicus serotinus	1.50
Myotis myotis	NA
Myotis daubentonii	3.48
Myotis mystacinus	3.86
Myotis nattereri	2.40
Nyctalus leisleri	0.95
Nyctalus noctula	1.58
Pipistrellus kuhlii	0.75
Pipistrellus nathusii	3.06
Pipistrellus pipistrellus	0.54
Pipistrellus pygmaeus	1.79
Plecotus sp (mainly austriacus)	1.36
Rhinolophus hipposideros	NA

Assessment of bat community specializations indices

The Community Specialization Index (CSI) is calculated as the arithmetic mean of the species specialization index (SSI) of the species detected, weighted by the abundances (Julliard et al., 2006).

$$CSI_{j} = \frac{\sum_{i=1}^{n} a_{ij} (SSI_{i})}{\sum_{i=1}^{n} a_{ij}}$$

where n is the total number of species recorded, a_{ij} is the abundance of individuals of species i (with a SSI specialization index) in segment j (Devictor et al., 2008).

The CSI reflects the relative abundance of more or less specialized species in local assemblages and is therefore expected to decrease following the relative declines of specialists (species with a high SSI). For the analyses, we kept only site with a non-null abundance because a null CSI does not describe a generalist community.

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Supplementary material 3: Description of the Natura 2000 network in France

The Natura 2000 network is regulated by two directives: following the Bird Directive of the European Union (79/409/EC), 384 Special Protection Areas (thereafter SPA, Fig.S1) were designated in France to protect endangered bird species, from 1986 onwards. The other Natura 2000 areas are Special Areas of Conservation (thereafter SAC, Fig.S2) which are defined following the Habitat Directive of the European Union (92/43/EEC) to protect non-bird animals and plant species and habitats listed in the Annexes I and II of this directive. These areas are identified as Sites of Community Importance (thereafter SCI, Fig.S2) and after approval of the European Union designated as SAC. To date, in France, *circa* the half of the 1369 SCI were further designated as SAC and thus effectively protected, since 2005. As a whole (SCI/SAC and SPA combined), the Natura 2000 networks represents 12.5% of the continental territory of France. After the designation phase occurring in 2000/2005, the Natura 2000 network is now engaged in a growing management process, based on management plans, voluntary measures and impact assessment prior to new human activities.

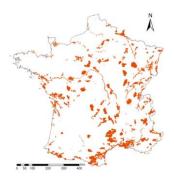


Figure S3-1. Location of the 384 Special Protection Areas (SPA) in France



Figure S3-2. Location of the 1369 Sites of Community Interest (SCI) to be designated as Special Areas for Conservation (SACs) by the European Union

Supplementary material 4: Detailed information on the variable Artificial Light at Night (ALAN)

We used radiance as a measure of ALAN as it is defined as the radiant flux (i.e., radiant power) reflected or emitted by a given surface. We used the VIIRS nighttime lights (2012) which is a 2-months composite raster of radiance data (in nW/ cm-2 sr) collected by the Suomi NPP-VIIRS Day/ Night Band during 2 time-periods in 2012 (20 nights in total) on cloud-free nights with zero moonlight (Baugh et al. 2013). We then computed the average radiance within each buffer with the tool "Zonal statistics as Table" from the package "Spatial Analyst" (Fig. S1, see also Azam et al. 2016).

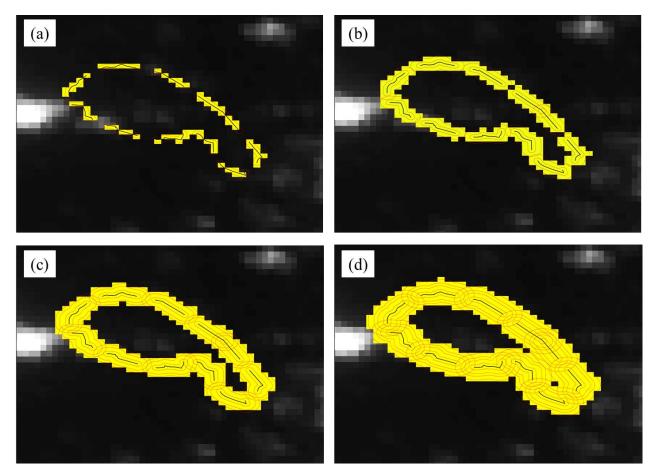


Fig. S1 Demonstration of the procedure done to calculate average radiance within the buffers of (a) 200 m, (b) 500 m, (c) 700 m and (d) 1000 m width from the VIIRS Nighttime Light raster used to. The average radiance was computed by the tool "zonal statistics as Table" from the package "Spatial Analyst" of ArcGIS 10.2 which proceed by creating a raster (yellow) from the polygon shapefile input (red) before applying an internal resampling so that input raster resolution match with the value raster resolution.

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Supplementary material 5: Detailed information on statistical analysis

Table S5-1 Correlation coefficients of the 8 landscape variables included in the analysis (i.e. The variables are the proportions of Natura 2000 (*Natura 2000*), intensive agriculture (*Inten. agri.*), heterogeneous agriculture (*Heter. agri.*), deciduous forests (*Forests*), *Open Habitat* and mean length per hectare of roads (*Roads*) and streams (*Streams*) and the Artificial Light At Night (ALAN), at the 1000 m landscape-scale which showed the strongest the correlation coefficients between landscape variables.

	ALAN	Inten. agri	Heter. agri	Forests	Open Habitat	Roads	Streams
Natura 2000	-0.25	-0.36	0.004	0.14	0.32	0.09	0.19
ALAN		-0.10	-0.14	-0.07	-0.39	0.13	0.02
Inten. agri.	/		-0.32	-0.46	-0.58	0.04	-0.30
Heter. agri.	/		/	-0.25	0.23	-0.007	0.09
Forest	/		/	/	-0.15	-0.02	-0.007
Open Habitat	-		_	-	-	0.06	0.33
Roads	/		/	/	-	/	-0.009

Table S5-2: Selected modelling and error distribution

Bat activity	Model	Error	Non-linear effect included
		distribution	
Abundance	GLMM	Poisson	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
Richness	GLMM	Poisson	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
CSI	GLMM	Gaussian	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
P. pipistrellus	GLMM	Poisson	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
P. kuhlii	GLMM	Poisson	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
E. serotinus	GLMM	Poisson	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
N. leislerii	GLMM	zero-inflation,	
		negative binomial	
N. noctula	GLMM	zero-inflation,	
		negative binomial	
Myotis ssp	GLMM	Quasibinomial	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$

GLMM were performed using R package *lme4* (function *glmer*, *glmer.nb*); (²) indicated that we use a quadratic effect to fit nonlinear relationships. We found similar result when nonlinear effects were performed with Generalized Additive Mixed Models (GAMM; Wood, 2006, R package mgcv) to handle nonlinear relationships between the response and some predictive variables; Zero-inflated model were performed using R package glmmADMB (function glmmadmb); quasibinomial were performed using R package MASS (function *glmmPQL*). Choice and validation of model where based on a multi-criteria approach (residual pattern, AIC, pseudo R²) following Zuur et al. (2009).

Specific analyses according to the European Union Directives

Natura 2000 sites include two network: special protection area (SPA, i.e. sites designated under the European Union Directive on the Conservation of Wild Birds) and Sites of Community Importance, (SCI), i.e. sites designated under the European Union's Habitats Directive, also known as the Directive on the Conservation of Natural Habitats and of Wild Fauna and Flora), we performed modelling at three level SPA, SIC and Natura 2000 sites (both together). SPA, SIC modelling were similar than performed in Table 1 and Table 2

Table S5-3: Partial regression coefficient of the raw Natura 2000 effect for the 3 community metrics and 6 taxa. P-value level of significance associated are "*** P < 0.001; "*" P < 0.05; "." P < 0.05; "." P < 0.01

Bat activity metric	Natura 2000	SICs	SPA
Abundance	$\beta = 0.220 \pm 0.035 ***$	$\beta = 0.196 \pm 0.02 ***$	$\beta = 0.142 \pm 0.040 ***$
Richness	$\beta = 0.130 \pm 0.052 **$	$\beta = 0.154 \pm 0.036 ***$	$\beta = 0.070 \pm 0.058$
CSI	$\beta = 0.061 \pm 0.020 **$	$\beta = 0.060 \pm 0.013 ***$	$\beta = 0.066 \pm 0.020 **$
P. pipistrellus	$\beta = 0.186 \pm 0.041 ***$	$\beta = 0.097 \pm 0.026 ***$	$\beta = 0.137 \pm 0.046 **$
P. kuhlii	$\beta = 0.113 \pm 0.128$	$\beta = 0.181 \pm 0.062 **$	$\beta = -0.278 \pm 0.153$.
E. serotinus	$\beta = 0.721 \pm 0.090 ***$	$\beta = 0.346 \pm 0.056 ***$	$\beta = 0.703 \pm 0.101 ***$
N. noctula	$\beta = -0.346 \pm 0.387$	$\beta = -0.105 \pm 0.235$	$\beta = -0.357 \pm 0.002$
N. leisleri	$\beta = 0.272 \pm 0.254$	$\beta = 0.488 \pm 0.159 **$	$\beta = 0.187 \pm 0.269$
Myotis spp.	$\beta = 1.372 \pm 0.209 ***$	$\beta = 0.694 \pm 0.141 ***$	$\beta = 1.368 \pm 0.233 ***$

Table S5-4: Standardized partial regression coefficients from GLMMs model for the proportion of SPA, SACs and Natura 2000 sites for the three community metrics and 6 taxa and at 4 landscape scales considered. P-value level of significance associated are '***' P < 0.001; '**' P < 0.01; '*' P < 0.05; '.' P < 0.1

	Scale	Natura 2000	SICs	SPA
	200	0.057***	0.045 ***	0.045 ***
A hum dan aa	500	0.022*	0.024 .	0.025 *
Abundance	700	0.028*	0.023	0.036 **
	1000	0.030.	-0.009	0.033 **
	200	0.002	0.022	-0.009
Di alamana	500	-0.006	0.014	-0.013
Richness	700	-0.009	0.010	-0.016
	1000	-0.013	-0.001	-0.019
	200	0.016***	0.026 ***	0.016 *
CCI	500	0.013*	0.024 ***	0.014 *
CSI	700	0.013*	0.023 ***	0.015 *
	1000	0.011.	0.022 ***	0.013 *
	200	0.038**	-0.002	0.032 *
D	500	0.014	-0.025	0.001
P. pipistrellus	700	0.026 .	-0.017	0.021
	1000	0.043**	-0.006	0.038 *
	200	0.069 .	0.080 *	0.038
D 1 11	500	0.009	0.066 .	-0.017
P. kuhlii	700	-0.027	0.051	0.005
	1000	-0.061	-0.098 *	-0.031
	200	0.081	0.030	0.114 ***
<i>r</i> .:	500	0.036	0.026	0.076 *
E. serotinus	700	0.026	0.021	0.057.
	1000	0.029	0.034	0.051
	200	-0.001	0.090	-0.011
N	500	-0.001	0.123	0.006
N. noctula	700	-0.038	0.114	-0.023
	1000	-0.028	0.142	-0.111
	200	0.121	0.056	0.116
N/ 1-:-1:	500	0.133	0.096	0.124
N. leisleri	700	0.117	0.121	0.077
	1000	0.093	0.094	0.050
	200	0.234***	0.237 ***	0.191 ***
Myotis ssp.	500	0.214**	0.230 ***	0.220 **
	700	0.225**	0.280 ***	0.231 **
	1000	0.213**	0.229 **	0.255 ***

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