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HAL Authorization

1 Common bats are more abundant within Natura 2000 areas

2

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17

18 **Abstract**

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

37

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

39 community,

40

41 **Introduction**

42 Among the approaches employed to preserve biodiversity, the establishment and management
43 of statutory protected areas are widely regarded as key strategies (Jackson *et al.*, 2007). This
44 classic protection approach is based on the principle that the designation of areas affords the
45 protection of habitats, species and/or populations that are threatened elsewhere (Gaston *et al.*,
46 2008). In addition to National Nature Reserves, National Parks and Regional Parks that are
47 designated by state members, the European Union (EU) has coordinated the Natura 2000
48 network. This network, which is the most important coordinated supra-national conservation
49 effort at the world level (Evans, 2012), aims at maintaining remarkable biodiversity through
50 the sustainable use of natural resources by establishing a network of areas that must maintain
51 a range of habitat types and wildlife species in a “favorable conservation status” (European
52 Commission, 2000). Over the last 25 years, the EU has built a vast network that represents
53 18% of the EU’s land area (see Supplementary Material S1). In France, this network
54 represents 12.5% of the continental territory. The main objectives of conservation and
55 restoration are stated at the local scale in the Natura 2000 management plan and the French
56 Natura 2000 areas are mainly managed through contracts between land owners and the state.
57 Agri-Environmental Schemes (Caps) are applied in agricultural land, whereas ones in non-
58 agricultural land may receive incentives for measures that favor conservation of biodiversity.
59 Although such actions are defined in each management plan, the field management of Natura
60 2000 has only recently begun (by 2005 only 20% of Natura 2000 sites had endorsed their
61 management plan (Maresca *et al.* 2006) and by 2012 nearly 73%, and establishment of new
62 plan has been slow since 2005, Debain, 2011).

63

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

66 the Natura 2000 network effectively protects species and habitats (Martínez *et al.*, 2006). As
67 Kliejn & van Zuijlen (2004) stated, the observed differences between protected and non-
68 protected areas may result from either the initial state of biodiversity ('designation effect',
69 assessed through a comparison of the biodiversity inside and outside the N2000 sites) or from
70 the protection efficiency *per se* ('protection effect', which involve the comparison of the state
71 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.

77 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
81 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
86 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
88 species (Gaston 2011). Surprisingly, few studies (Devictor *et al.*, 2007, Pellissier *et al.*, 2013)
89 have assessed the importance of classical protection measures on common bird biodiversity,

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 single
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

115 habitat community specialization) were influenced by the Natura 2000 network. This test of
116 Natura 2000 designation effect was achieved through two analyses, one comparing bat
117 activities within and outside Natura 2000 sites and a second approach, in which we assessed
118 the influence of Natura 2000 areas controlling for the main land uses and at different
119 landscape scales. Higher relative abundance within Natura 2000 is expected if the Natura
120 2000 areas exhibit a ‘designation effect’, *i.e.*, the Natura 2000 areas are high-value areas for
121 common species.

122

123 MATERIAL AND METHODS

124 **Car transect survey**

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

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

146

147 **Biological data**

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

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

184

185 **Landscape and Natura 2000 data**

186 Around each of the 1608 transects (i.e. the 2-km road portions), we generated a set of
187 landscape variables using QGIS 2.2. Because the landscape effect on bat activity could
188 change according to the spatial scale considered (Grindal & Brigham, 1999; Lacoeuilhe *et al.*,
189 2016) we calculated these variables at 4 different buffers of 200, 500, 700 and 1000 m. At

190 1000 m landscape scale, there was 25 % overlap between nearby transects belonging to a
191 same road circuit. However, this still allowed the measurement of variation in landscape
192 characteristics within the same road circuit. However, we could not extend the analyses into
193 larger landscape scales, because the overlapping rate between transect buffers would exceed
194 50 %. We assessed the proportion of the Natura 2000 areas included in each buffer (shapes
195 were provided by the Muséum national d'Histoire naturelle (<http://inpn.mnhn.fr>, see
196 Supplementary Material 3). Sites designated for birds (Special Protection Areas, SPA) and/or
197 Habitats directive (Sites of Community Importance, SCI) were considered together as the
198 evaluation deals with the whole network designation effect on non-target bat biodiversity.
199 Note that analyses at the level of each directive (SCI and SPA) did not reveal any obvious
200 difference with the analysis performed at the level of Natura 2000 sites (both together), see
201 Supplementary Material 5, Table S5-3 & S5-4.

202 We also calculated seven land use variables within each buffer, (i) the proportion of intensive
203 agriculture (*Inten. Agri.*, Code 2.1; CORINE Land Cover, including irrigated and non-
204 irrigated arable land such as cereals, leguminous and forage crops.) within each buffer
205 because they represent a major land use pressure for bats (Azam *et al.*, 2016); (ii) the
206 proportion of heterogeneous agriculture (*Heter. agri.*), defined as areas with a complex
207 mosaic of annual and permanent cultures and semi-natural habitats (Code 2.4; CORINE Land
208 Cover); (iii) the proportion of deciduous and mixed forests (*Forests*, Code 3.1; CORINE Land
209 Cover) as they were commonly represented in our buffers and were known to influence bat
210 activity at a landscape scale (Boughey *et al.*, 2011; Frey-Ehrenbold *et al.*, 2013); (iv) the
211 proportion of open habitat (*Open habitat*, code 231 and 32 CORINE Land Cover; (v) the total
212 length of streams and water sources (*Streams*, IGN/2012) because numerous studies have
213 identified aquatic habitat as a favorable where abundance of bat is high (Rainho & Palmeirim
214 2011; Russo & Jones 2003, Salvarina 2016); (vi) the total length of roads (*Roads*, IGN/2012)

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

226

227 **Representativeness of habitats and land uses sampled in the French bat monitoring** 228 **program**

229 This survey protocol resulted in the collection of data along roads chosen by volunteers,
230 which may introduce a bias in the habitat sampling. Preliminary analysis have underlined that
231 the choice of the circuit performed by volunteers plus the randomization introduced by the
232 MNHN limited the bias in habitat representativeness at local and national scales (see
233 Supplementary Material S1, Table S1-1). Because buildings tend to be concentrated along
234 road networks, the main bias observed was with regard to urban areas and, in particular,
235 impervious areas, which comprises 5% of France. However, our survey included 11% at a
236 200 m landscape scale, but note that this bias decreases when landscape scale take into
237 account increase (6% of impervious areas in buffer of 1000 m size). We did not detect any
238 bias for the representativeness of Natura 2000 regardless of the spatial scale considered (see
239 Supplementary Material S1, Table S1-1)

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

255

256 **Statistical analyses**

257 *Model building*

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

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

270

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

273

274 *Check of assumptions of GLMM*

275 Before performing our models, we systematically evaluated the correlations among
276 explanatory variables using Spearman's rho for quantitative variables (Crawley, 2009) to
277 detect obvious correlation (See Supplementary Material 5). Secondly, we performed variance-
278 inflation factors (VIF) on each model (Fox & Monette 1992); all variables had VIF<2
279 indicating no problem of multicollinearity in the explanatory variables of our models.

280 Close points (even if they differ from habitat) are likely to have a similar bat population
281 density due to similar climatic conditions or large-scale landscape compositions and could
282 therefore imply spatial autocorrelation. Thus, we performed a variogram to observe if
283 correlation between neighbors falls off with distance. When we detected such patterns in our
284 data, we added in our GLMM an autocovariate (i.e., a distance-weighted function of
285 neighboring response values; Dormann *et al.* 2007) with the autocov dist function in R
286 (package spdep, Bivand R. *et al.* 2011) to account for spatial autocorrelation.

287

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

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

296

297 [bat activity] ~ *Natura 2000* + *Inten. agri* + *Heter. agri.* + *Forests* + *Open habitat* + *Roads* +
298 *Streams* + *ALAN* + *date of survey* + *wind speed* + *minutes after sunset* + *temperature* + 1|year
299 +1|site

300

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

307

308 **RESULTS**

309 The survey recorded 23164 bat passes, on which analysis was conducted on the most
310 commonly recorded species: *Pipistrellus pipistrellus* (n = 14967), *Eptesicus serotinus* (n =
311 2614), *Pipistrellus kuhlii* (n = 2318), (note that even after checking the identifications, we
312 extrapolated that 8% of *P. kuhlii* calls are not entirely certain and could include *P. nathusius*
313 calls), See Table S1 for details), *Nyctalus leisleri* (n = 1999), *Nyctalus noctula* (755) and
314 *Myotis spp.* (261). The first species represented 65% of the dataset.

315 Without adjusting Natura 2000 effect to take landscape composition into account, *P.*
316 *pipistrellus* and *E. serotinus* exhibited greater relative abundance within Natura 2000 than
317 outside, and *Myotis* spp. have a greater probability to be recorded within Natura 2000, while
318 we detected no significant effect for the three other species (Table 1). In addition, we detected
319 a greater relative abundance, a greater richness and communities are more specialized within
320 Natura 2000 than outside (Table 1).

321 When taking into account the main land uses within landscape and assessing the effect of the
322 amount of Natura 2000 (i.e. areas of Natura 2000 in the neighbor of the sample site and not
323 just being in a site or off site), we detected a similar relationship to the first set of analyses.

324 Among the six species studied, we were able to detect a positive influence of Natura 2000 in
325 the area bordering transects for *P. pipistrellus*, *E. serotinus* and *Myotis* spp. (Table 2).

326 However, the detection of significant effects was scale dependent, and mainly detected at a
327 small spatial scale (i.e. at 200 m, except for *Myotis* spp). In addition, total bat activity and
328 community bat specialization were positively influenced by Natura 2000 areas, whereas no
329 effect was detected for richness (Table 2). Again, we detected more significant effects at a
330 smaller spatial scale.

331 The proportion of intensive agriculture had a significant negative effect on all species (except
332 *N. noctula*). We found same results for all bat community metrics whatever the landscape
333 scale considered. Artificial light at night, heterogeneous agriculture and open habitat, when
334 significant, had a negative effect, whereas streams mainly had significant positive effect
335 (except for *E. serotinus*) but again the effect was scale dependent and varied among species or
336 bat community metrics (Table 2, Fig. 1). Forest effects depend of bat activities measured: for
337 example *P. pipistrellus* was positively influenced by the proportion of forest while *N. lesileri*
338 was negatively influenced, total bat relative abundance positively influenced, while richness
339 and community specialization were negatively influenced by forest (Table 2, Fig. 1).

340

341 **DISCUSSION**

342

343 **'Designation effect' of Natura 2000 areas for bats.**

344 This paper provides evidence that bat activity is globally higher in Natura 2000 sites (+24%;).

345 Three (*P. pipistrellus*, *E. serotinus*, *Myotis* spp) of the six species/genera studied exhibit
346 greater relative abundance in Natura 2000 sites (this increase is noteworthy for *E. serotinus* (x
347 2.1) and *Myotis* spp (x 3.6)) and note that no significant negative effect was detected. The
348 result for *Myotis* spp must be taken with caution; it did not imply that all species of this genus
349 exhibit a similar pattern, but at least one species exhibits a greater bat activity in Natura 2000
350 sites. We also detected a greater richness in Natura 2000 sites (+ 14%). Finally, the bat
351 community specialization, a mean trait community index, is greater in Natura 2000 sites
352 (+9%).

353 In a second step, we evaluated if this Natura 2000 effect was consistent when, in the
354 neighborhood of the survey, the proportion of Natura 2000 was adjusted to the land uses. For
355 *P. pipistrellus*, *E. serotinus* and *Myotis* spp, the increase of Natura 2000 in the surrounding
356 area positively influenced their activity but the effect was spatial scale dependent. Similarly,
357 bat community specialization, increased when Natura 2000 areas increased in the
358 surrounding transects. However, we did not detect any effect of the proportion of Natura 2000
359 areas in the surrounding of transects on species richness. Note that this latter non-detection is
360 possibly linked to the nature of data collected (small sequence of acoustic recordings) which
361 currently did not allow species identification within the *Myotis* group, while this group is very
362 rich in the region considered (39% of the French species, TAXREF, Gargominy *et al.*, 2016-).
363 With the recent arrival on the market of new generation of bat detector-recorders, we could
364 consider recording throughout the entire night (see Newson *et al.*, 2015) and thus expect an

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

368 The positive Natura 2000 effect appears to be relatively small compared to the main
369 land use pressure: intensive agriculture and to a lesser extent heterogeneous agriculture (while
370 this category is known to include more extensive agriculture practice) and light pollution
371 (ALAN) all of them have a deep negative impact. They impact not only the abundance of
372 populations but also the structure of communities, leading to less rich and more generalist
373 communities, thus contributing to a biotic homogenization process (McKinney, 2006, Karp *et*
374 *al.*, 2012). These drivers are known to impact negatively bats (Wickramasinghe *et al.*, 2004;
375 Azam *et al.* 2016) and more broadly biodiversity, both rare and endangered species (Maxwell
376 *et al.*, 2016) as the common species and (plants: moth: Conrad *et al.* 2006; butterflies: Van
377 Dyck *et al.* 2009; bees: Kosior *et al.*, 2007; birds: Donald. 2001) and contribute to large scale
378 biotic homogenization (Le Viol *et al.* 2012). The negative effect of open habitat is mainly due
379 to the requirement of the species studied to avoid large open habitat (Verboom, & Spoelstra,
380 1999) and select wooded edge, woodland or aquatic habitat (Walsh & Harris, 1996). The
381 positive effect of streams is (although detected for many species or bat community measures)
382 in accordance with many studies see the review perform by Salvarina (2016). The contrasting
383 effect of forest areas on the species studied (dominated by aerial hawking species), positive
384 effect on total relative abundance, while negative effect on richness and community
385 specialization, seem to indicate that these land uses, when adjusted for the effect of Natura
386 2000, does not have a broad effect. Zehetmair *et al.*, (2015), in a sample of European beech
387 forest, found that bat activity did not differ significantly between the Natura 2000 and non-
388 Natura 2000 forest on paired comparison of stands with same structural attributes. They
389 concluded that the current management of the Natura 2000 beech forests is almost identical to

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

397 When the road effect is detected, it is surprisingly positive and appears to invalidate this
398 landscape variable as a pressure on bat foraging connectivity. Lisón & Sánchez-Fernández
399 (2017) found that the roadless areas are not significant for the localization of bat hotspots. It
400 should be noted however, that we could not test accurately a “road effect” because our
401 sampling is not representative of national road network: due to security, and transects were
402 mostly located in areas with secondary low-traffic roads. Thus, within this biased sample, we
403 probably detected an effect of availability of hedgerows along roads, where hedgerows are
404 positively selected for by bats (Lacoeuilhe *et al.*, 2016). The increase in relative abundance
405 and degree of specialization may appear limited compared to the negative effect of intensive
406 agriculture (respectively lower by a factor 6 and 2, Table 2). However, it is still notable that
407 for common, widely distributed species, that there is still a Natura 2000 effect, after we
408 control for land use variables like streams that are known to have a positive influence on bats
409 (Salvarina, 2016).

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

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

423

424 **Scale and time effect**

425 When looking at the influence of spatial scale, the positive effect of Natura 2000 tends
426 to be more significant at a smaller scale (*i.e.* 200 m) while some land uses variables remain
427 significant whatever the spatial scale considered (*i.e.* intensive agriculture, ALAN). This
428 finding seems to indicate that Natura 2000 areas do not yet enhance adjacent areas at a large
429 scale for common bats. However, a theoretically and empirically demonstrated reserve effect
430 is the spillover of species richness and community complexity that highlights the important
431 benefit for biodiversity and ecosystems of establishing reserve areas (Roberts *et al.* 2001,
432 Russ & Alcalá, 2011). However, many studies have underlined that reserves could enhance
433 adjacent areas, but that age matters (Claudet *et al.*, 2008, Russ & Alcalá, 2011). It should be
434 emphasized that the purpose of this paper is not to assess the efficiency of the management
435 measures actually occurring in the Natura 2000 areas; here we only assess a designation
436 effect. Indeed, management is probably too recent to be able to produce measurable effects,
437 because during the period studied (2006-2013) only 53% of Natura 2000 sites had endorsed
438 their management plan, and moreover 69% of recordings were collected between 2006 and
439 2009 (see Supplementary Material 1, Fig S1-1). We hypothesize that effect of Natura 2000 at

440 large landscape scale should become more significant in the future with the effects of Natura
441 2000 sites management and the implementation and green infrastructure policies, which aim
442 to connect patches of protected areas. Future research should now focus on comparing
443 biodiversity state before and after the designation of N2000 sites (see Lisón & Sánchez-
444 Fernández, 2017), especially since conservation policy may moderate land-cover
445 transformation (Kallimanis *et al.*, 2015).

446

447 **Car-based bat survey and policy implications**

448 The French bat monitoring program has proven to be a useful tool for evaluating the Natura
449 2000 network owing to its large spatial coverage and its standardized monitoring based on
450 volunteers. This finding emphasizes that public participation in scientific research is a
451 promising approach that can significantly help scientists to address questions relating to
452 biodiversity , as suggested by Bell *et al.*, (2008) and Couvet *et al.*, (2008). Volunteer-based
453 monitoring is likely to have high resilience (Couvet *et al.*, 2008), which is key to maintaining
454 the monitoring program over time and, thus, providing extended time-series (Battersby,
455 2010). However, questions can be raised about more efficient ways to monitor biodiversity of
456 a protected network, between surveying rarer species and habitats or to survey more common
457 and widespread elements not targeted during the designation process. Indeed, rare species
458 (due to lower abundance, lower detection, or their specialization to particular habitats) need
459 focused sampling (Barlow *et al.*, 2015). From a financial point of view, it should be noted that
460 the monitoring of common species does not involve funds dedicated to Natura 2000 surveys.

461

462 **Conclusion and conservation implications**

463 Our study showed a sampling effect induced by the designation process of Natura
464 2000. The species studied are only listed in the Annex IV of the habitat directive (not Annex

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

481 Additionally, most of the Natura 2000 areas designated for the protection of bat
482 species are limited to roosting sites and immediate surroundings. Indeed, for species with a
483 complex life cycle, reproduction is most often considered the most important part of the cycle,
484 and, thus, the protection of reproduction sites is emphasized (Stebbing, 1988). However,
485 whilst sites for bat reproduction are restricted in space, they have a large foraging territory,
486 even during reproduction (Stebbing, 1988). Therefore, protecting hunting territories, which
487 can be quite large and far from roosting sites, is of prime importance from a conservation
488 point of view (Vaughan *et al.*, 1997). In this study, we assessed the influence of Natura 2000
489 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

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498

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711 Zehetmair, T., Müllerb, J., Runkel, V., Stahlschmidt, P., Winter, S., Zharov, A., Grupp, A.,
712 2015. Poor effectiveness of Natura 2000 beech forests in protecting forest-dwelling bats.
713 Journal for Nature Conservation 23, 53–60.

714 Zuur, A.F., Ieno, E.N., Walker, N., Saveliev, A.A., Smith, G.M., 2009. Mixed Effects Models
715 and Extensions in Ecology with R. Statistics for Biology and Health, Springer, New
716 York, USA. 574p.

717

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

720

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

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722

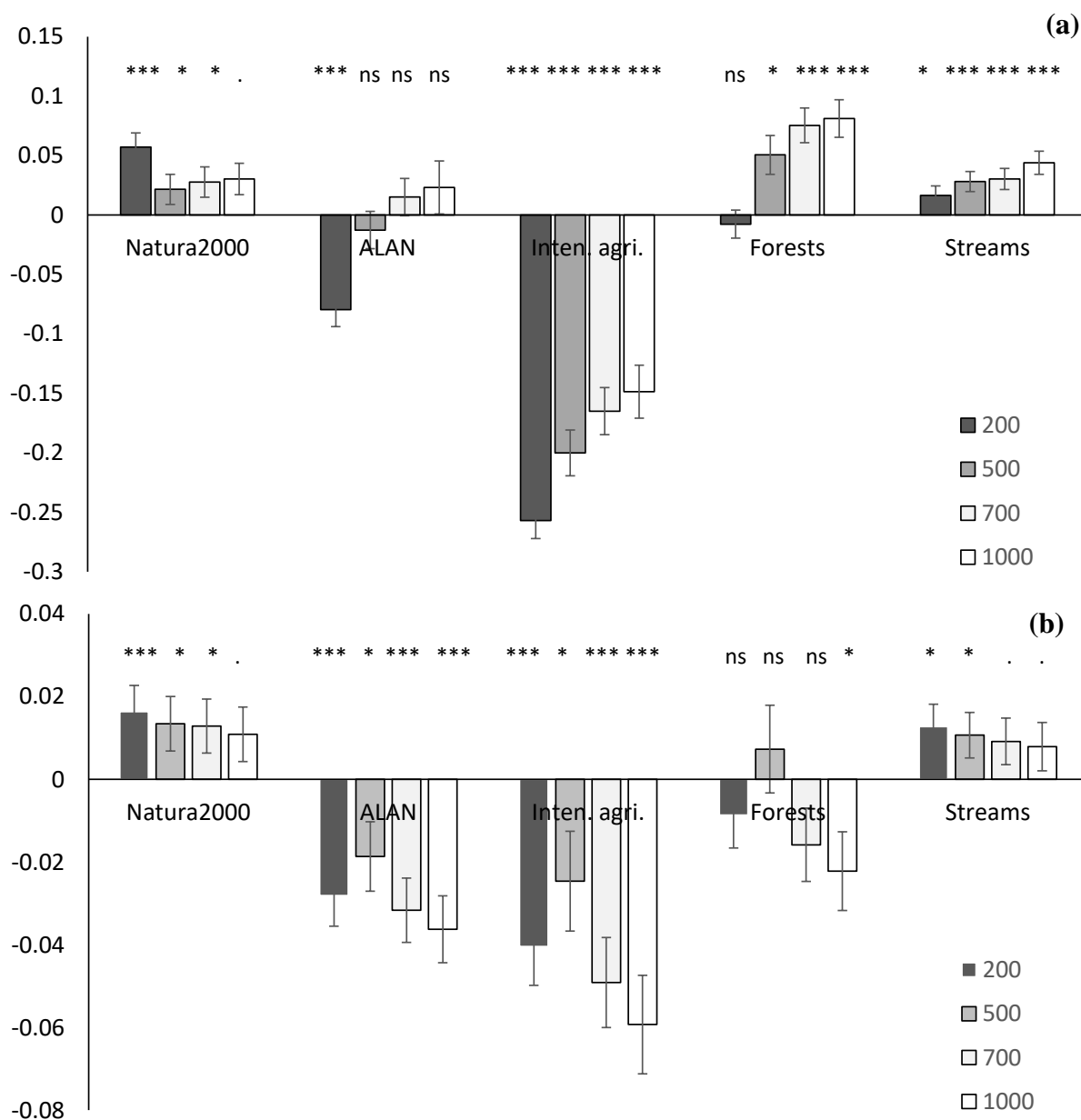
Table 2. Standardized partial regression coefficients from GLMMs model for the 8 landscape effects included in the analysis (i.e. the proportion of the Natura 2000 (Natura 2000), intensive agriculture (*Inten. Agri.*), heterogeneous agriculture (*Heter. agri.*), forests (*Forests*), open habitat (*Open 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	<i>Natura 2000</i>	<i>Inten. agri.</i>	<i>ALAN</i>	<i>Heter. agri.</i>	<i>Forests</i>	<i>Open Habitat</i>	<i>Roads</i>	<i>Streams</i>
<i>Community metrics</i>									
<i>Relative abundance</i>	200	0.057****	-0.257****	-0.079****	-0.081****	-0.008	-0.110****	0.0317****	0.016*
	500	0.022*	-0.200****	-0.013	-0.029*	0.051*	-0.047**	0.029****	0.028****
	700	0.028*	-0.165****	0.015	-0.009	0.075****	-0.028.	0.024**	0.030****
	1000	0.030.	-0.149****	0.023	-0.010	0.081****	-0.033	0.031****	0.044****
<i>Richness</i>	200	0.002	-0.206****	-0.083****	-0.039.	-0.045*	-0.071**	0.010	0.020
	500	-0.006	-0.157****	-0.055*	-0.001	-0.001	-0.026	0.010	0.032*
	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.
<i>CSI</i>	200	0.016****	0.040****	-0.028****	-0.042****	-0.009	-0.030****	0.017*	0.012*
	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
<i>Taxa</i>									
<i>P. pipistrellus</i>	200	0.038**	-0.219***	-0.087***	-0.073***	0.028.	-0.109***	-0.007	0.027*
	500	0.014	-0.175***	-0.046**	-0.021	0.068***	-0.043*	-0.042***	0.034**
	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***
<i>P. kuhlii</i>	200	0.069 .	-0.328***	-0.169***	-0.029	-0.150***	-0.037	0.078***	-0.036
	500	0.009	-0.255***	-0.120 .	0.062	-0.014	0.078	0.072***	-0.016
	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
<i>E. serotinus</i>	200	0.081**	-0.706***	-0.353***	-0.346***	-0.062***	-0.346***	0.089***	-0.121***
	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
<i>N. noctula</i>	200	-0.001	0.220	0.121	-0.486***	-0.235 .	-0.676***	0.025	0.153
	500	-0.001	-0.242	0.043	-0.556***	-0.319 .	-0.807***	0.022	0.204*
	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
<i>N. leisleri</i>	200	0.121	-0.600***	-0.161	-0.463***	-0.286**	-0.408***	0.065	0.198**
	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**
<i>Myotis ssp.</i>	200	0.234***	-0.505***	-0.208*	-0.112	-0.070	-0.150 .	0.084 .	0.092 .
	500	0.214**	-0.526***	-0.109	-0.074	-0.068	-0.078	-0.097	0.130*
	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 S1-1: Characteristics of the protocol and sampling design the two bat surveys

Aim of the program	Monitoring the temporal trends of bat populations at a national scale	
Car transect survey sampling protocol		
<i>Scope</i>	10 km around surveyors' home.	
<i>Circuit length</i>	30 km	
<i>Number of transect per circuit</i>	10	
<i>Transect length</i>	2 km separated of at least 1 km	
<i>Period of sampling</i>	from the 15 th of June to the 31 th of July and 15 August to 31 September	
<i>Weather conditions</i>	no rain, low wind speed (< 7 m/s), temperature > 12°C	
<i>Survey start</i>	as possible 30 minutes after sunset	
Count point survey sampling protocol		
<i>Scope</i>	square of 2km-side randomly chosen (by the Museum) in a radius of 10km from the observer's home, (i.e. on average one square randomly chosen between 80 possible squares).	
<i>Circuit length</i>	square of 2km-side	
<i>Number of point per circuit</i>	10	
<i>Recording duration</i>	6 minutes	
<i>Period of sampling</i>	from the 15 June to 31 July and 15 August to 31 September	
<i>Weather conditions</i>	no rain, low wind speed (< 7 m/s), temperature > 12°C	
<i>Survey start</i>	as possible 30 minutes after sunset	
Bat recording characteristics		
<i>Acoustic detectors</i>	Tranquility Transect Bat detector&D240x	
<i>Intercalibration of detectors</i>	At the MNHN	
Acoustic settings	Tranquility Transect	D240x
<i>Suppliers</i>	Courtpan Design Ltd, UK	Pettersons Elektronik
<i>High pass filter</i>	5 kHz	18 kHz
<i>Frequency</i>	96 000 sample/sec	96 000 sample/sec
<i>Recording device</i>	Zoom H2 digital recorder (Samson technologies, USA)	
<i>File storage format</i>	WAV	
Bat identification		
<i>Software</i>	Syrinx 2.6	
<i>Procedure</i>	<ul style="list-style-type: none"> - Training: 2-day training course+ online self-training courses - Bat first identification: by volunteers - Bat identification validation: by MNHN 	
<i>Taxon identification level</i>	Species level except for <i>Myotis sp.</i> Extensive data expertise evaluated that <i>Pipistrellus kuhlii</i> may potentially include 8% <i>Pipistrellus nathusius</i> , as these two species overlap in their acoustic signatures.	
Meteorological data		
	<ul style="list-style-type: none"> - Temperature (°C) and cloud cover (% in four classes: 0-25%, 25-50%, 50-75%, 75-100%) were recorded by volunteers during the survey. - Wind speed was also recorded by volunteers using Beaufort scale but this empirical categorical measure was not used in the analyses. However we used this field this measure to confirm the appropriateness of the use of the wind speed measure provide by the closest meteorological station. - Wind speed (km/h) and humidity (%) was provided by the closest meteorological station using the public archives the web site Infoclimat [http://www.infoclimat.fr/] 	
Habitat data		
	Volunteers were involved in collecting habitat variable on a detailed and adapted hierarchical system (see hereafter).	

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

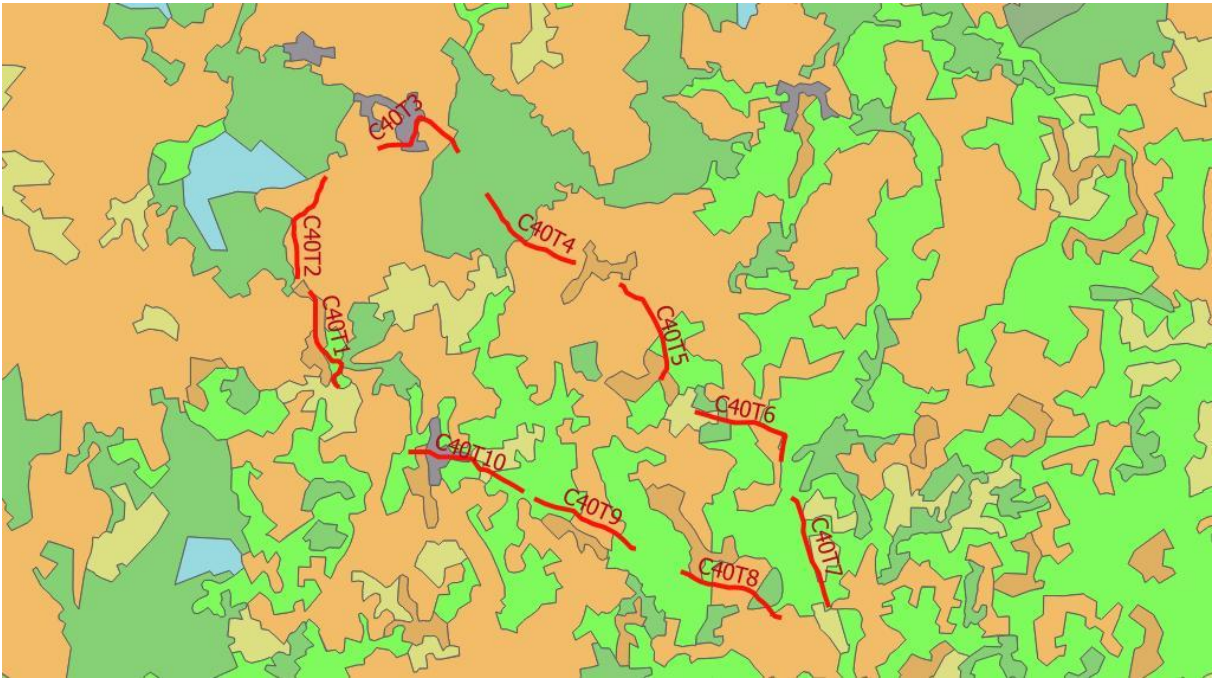
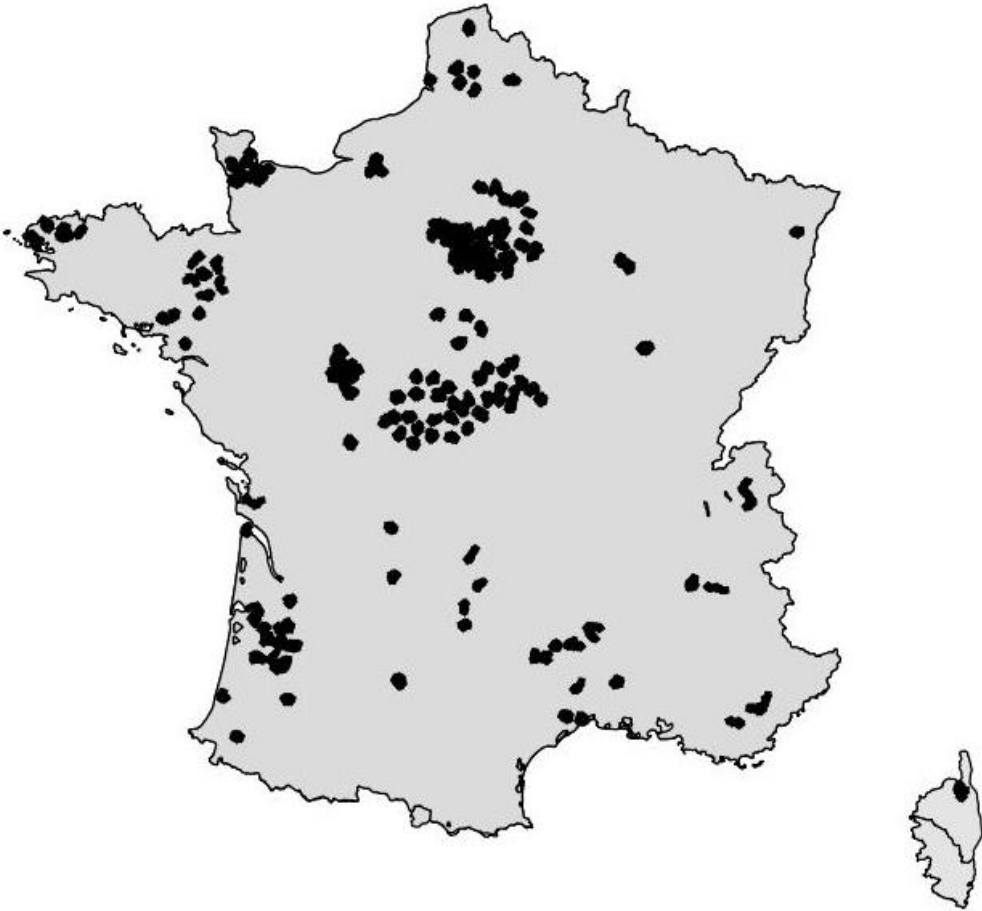


Figure S1-2: Distribution of car transect surveys of the French national bat monitoring program



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%)

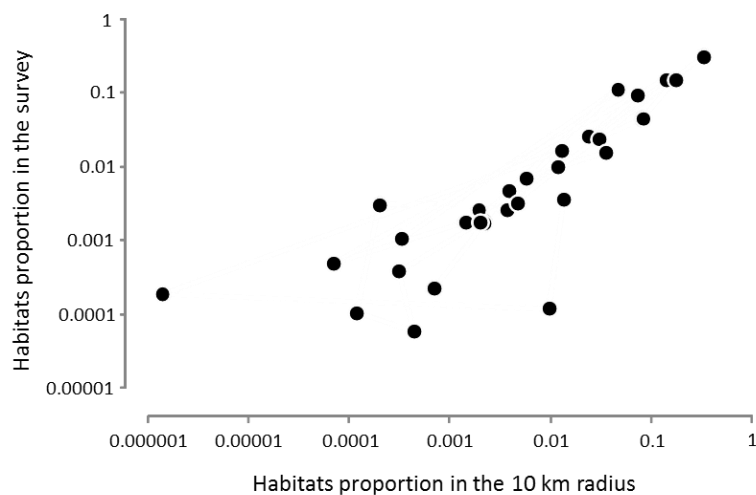


Fig. S1-3: Correlation between habitat proportion at local scale (10 km radius) and proportion sampled in the bat survey. Proportion of Corine Land Cover classes are log transformed.

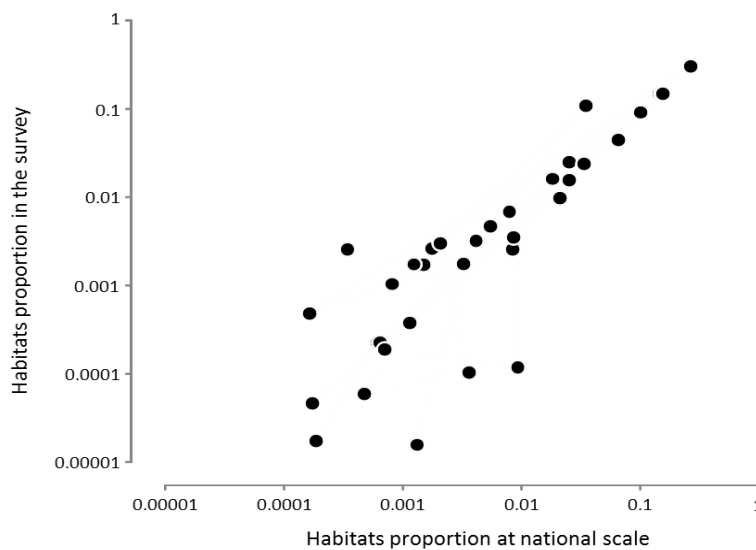


Fig. S1-4: Correlation between habitat proportion at national scale and proportion sampled in the bat survey. Proportion of Corine Land Cover classes are log transformed.

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

No obvious correlation ($\rho \geq 0.5$; 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/relevés-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.

In agricultural habitat, 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
<i>Arable land (n=454)</i>	$\beta=-1.460$	$P<0.0001$	Within Natura 2000 area: 0.09 Without Natura 2000 area: 0.28
<i>Heterogeneous agricultural areas (n=811)</i>	$\beta=-0.850$	$P<0.0001$	Within Natura 2000 area: 0.79 Without Natura 2000 area: 0.90
<i>Pastures (n=643)</i>	$\beta=-0.377$	$P=0.11$	Within Natura 2000 area: 0.85 Without Natura 2000 area: 0.89
<i>Vineyards & orchard (n=63)</i>	$\beta=-0.000$	$P=1$	Within Natura 2000 area: 0.50 Without Natura 2000 area: 0.50

In forest habitat, 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
<i>Broad-leaved forest (n=989)</i>	$\beta=0.465$	$P=0.009$	Within Natura 2000 area: 0.82 Without Natura 2000 area: 0.75
<i>Mixed forest dominant broad-leaved (n=572)</i>	$\beta=-0.603$	$P=0.04$	Within Natura 2000 area: 0.87 Without Natura 2000 area: 0.92
<i>Coniferous forest (n=55)</i>	$\beta=-16.99$	$P=0.9$	Within Natura 2000 area: 0.14 Without Natura 2000 area: 0.66
<i>Mixed forest dominant coniferous (n=263)</i>	$\beta=-1.331$	$P=0.02$	Within Natura 2000 area: 0.86 Without Natura 2000 area: 0.96

3. Device fastener

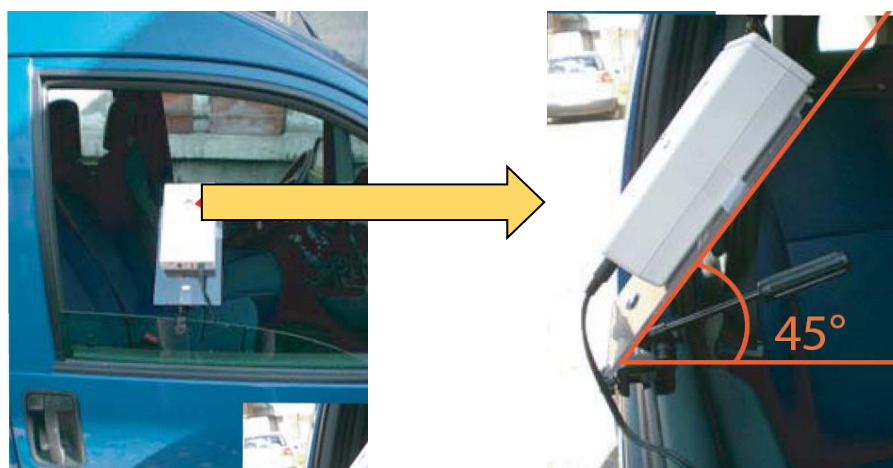


Fig S1-1: arrangement of the detector. The detector was fixed to the passenger-side window at a 45° angle so that the distance to road edges was similar all along the circuits

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

<i>Pipistrellus pipistrellus</i>	1.6 % (\pm 0.5)
<i>Pipistrellus kuhlii</i>	7.8 % (\pm 1.9)
<i>Nyctalus leisleri</i>	6.4 % (\pm 1.5)
<i>Eptesicus serotinus</i>	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	%
<i>Myotis alcaethoe</i>	0.1
<i>Myotis bechsteinii</i>	0.2
<i>Myotis brantii</i>	0.6
<i>Myotis capaccini</i>	0.0
<i>Myotis daubentoni</i>	34.7
<i>Myotis emarginatus</i>	0.5
<i>Myotis myotis</i>	1.8
<i>Myotis mystacinus</i>	1.9
<i>Myotis nattereri</i>	1.8
<i>Myotis puniclus</i>	0.0
<i>Myotis sp</i>	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).**

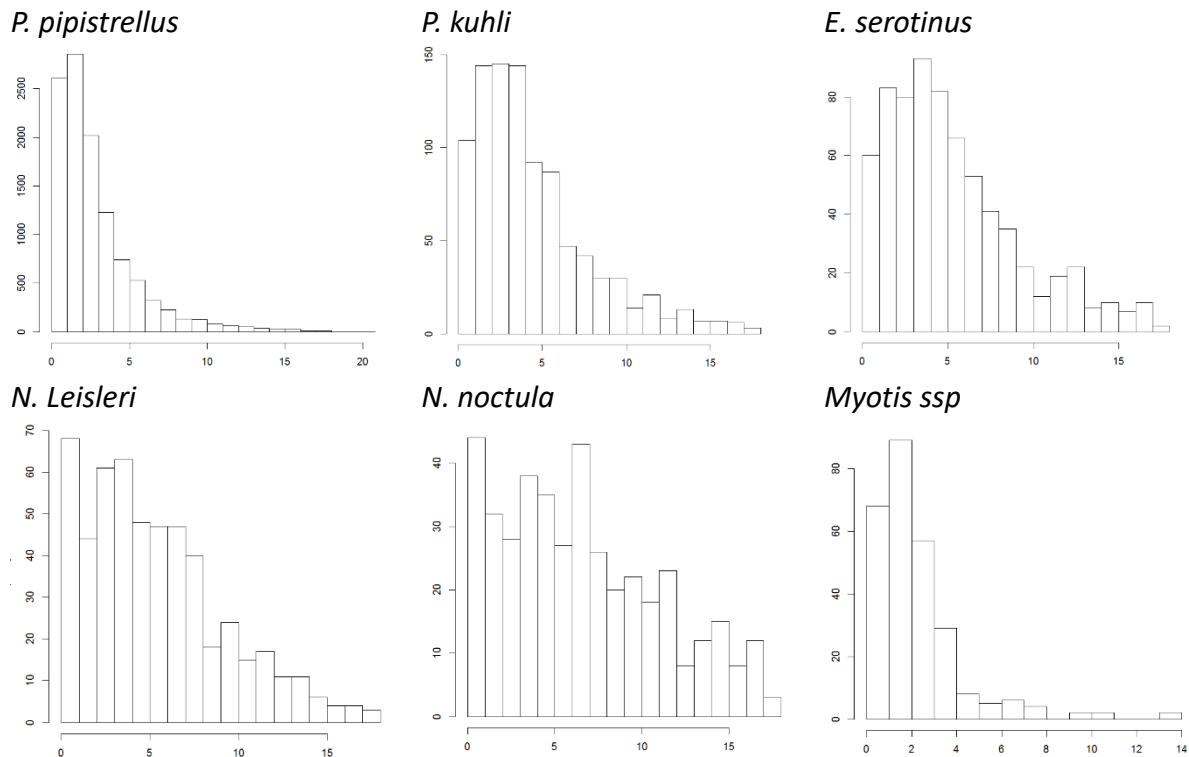
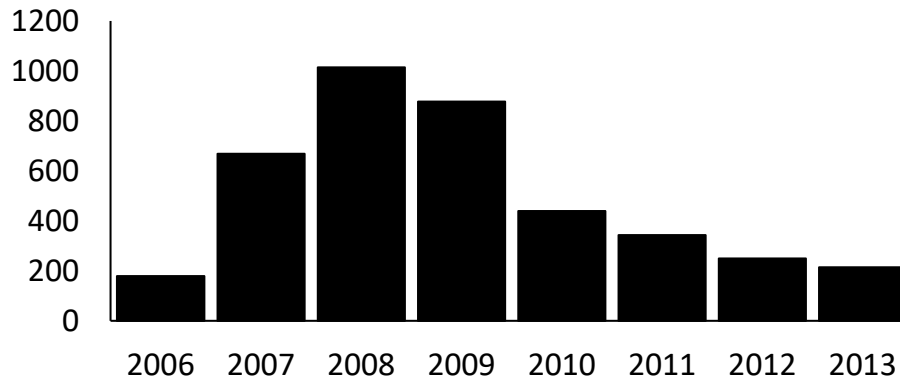


Fig S1-2: Distribution of bat passes duration from the car transect survey (vertical axis are the number of bat pass)

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/relevés-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
<i>Barbastella barbastellus</i>	3.64
<i>Eptesicus serotinus</i>	1.50
<i>Myotis myotis</i>	NA
<i>Myotis daubentonii</i>	3.48
<i>Myotis mystacinus</i>	3.86
<i>Myotis nattereri</i>	2.40
<i>Nyctalus leisleri</i>	0.95
<i>Nyctalus noctula</i>	1.58
<i>Pipistrellus kuhlii</i>	0.75
<i>Pipistrellus nathusii</i>	3.06
<i>Pipistrellus pipistrellus</i>	0.54
<i>Pipistrellus pygmaeus</i>	1.79
<i>Plecotus sp (mainly austriacus)</i>	1.36
<i>Rhinolophus hipposideros</i>	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.

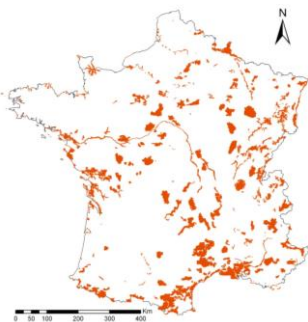


Figure S3-1. Location of the 384 Special Protection Areas (SPA) in France

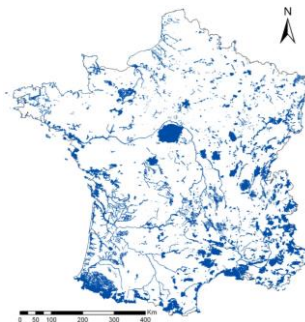


Figure S3-2. Location of the 1369 Sites of Community Interest (SCI) to be designated as Special Areas for Conservation (SACs) by the European Union

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).

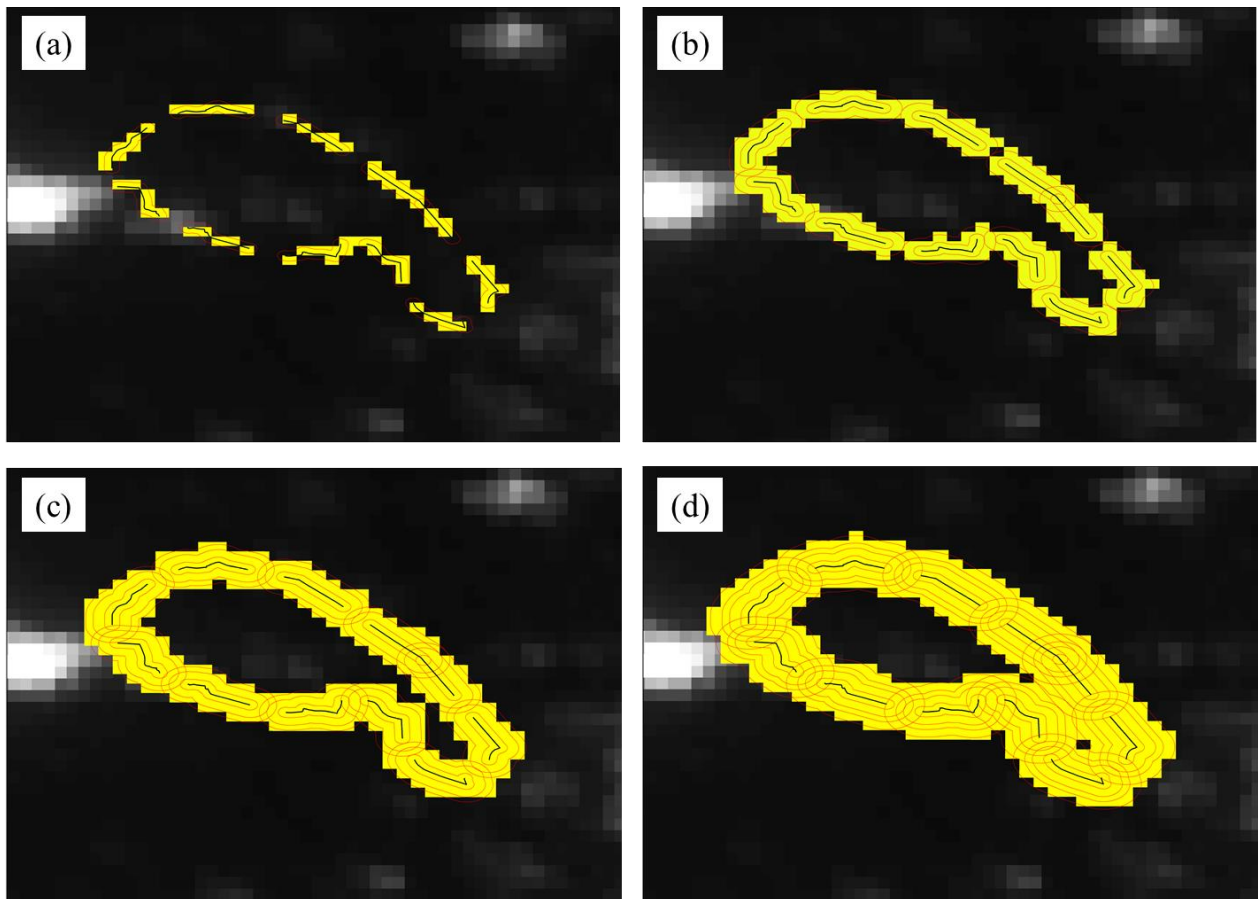


Fig. S1 Demonstration of the procedure done to calculate average radiance within the buffers of (a) 200 m, (b) 500 m, (c) 700 m and (d) 1000 m width from the VIIRS Nighttime Light raster used to. The average radiance was computed by the tool “zonal statistics as Table” from the package “Spatial Analyst” 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.

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

Table S5-2: Selected modelling and error distribution

Bat activity	Model	Error distribution	Non-linear effect included
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	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
<i>P. pipistrellus</i>	GLMM	Poisson	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
<i>P. kuhlii</i>	GLMM	Poisson	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
<i>E. serotinus</i>	GLMM	Poisson	$date\ of\ survey^2 + minutes\ after\ sunset^2 + temperature^2$
<i>N. leislerii</i>	GLMM	zero-inflation, negative binomial	
<i>N. noctula</i>	GLMM	zero-inflation, negative binomial	
<i>Myotis ssp</i>	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 non-linear 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 were 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 < 0.01; ‘*’ P < 0.05; ‘.’ P < 0.1

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

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	<i>Natura 2000</i>	<i>SICs</i>	<i>SPA</i>
<i>Abundance</i>	200	0.057***	0.045 ***	0.045 ***
	500	0.022*	0.024 .	0.025 *
	700	0.028*	0.023	0.036 **
	1000	0.030.	-0.009	0.033 **
<i>Richness</i>	200	0.002	0.022	-0.009
	500	-0.006	0.014	-0.013
	700	-0.009	0.010	-0.016
	1000	-0.013	-0.001	-0.019
<i>CSI</i>	200	0.016***	0.026 ***	0.016 *
	500	0.013*	0.024 ***	0.014 *
	700	0.013*	0.023 ***	0.015 *
	1000	0.011.	0.022 ***	0.013 *
<i>P. pipistrellus</i>	200	0.038**	-0.002	0.032 *
	500	0.014	-0.025	0.001
	700	0.026 .	-0.017	0.021
	1000	0.043**	-0.006	0.038 *
<i>P. kuhlii</i>	200	0.069 .	0.080 *	0.038
	500	0.009	0.066 .	-0.017
	700	-0.027	0.051	0.005
	1000	-0.061	-0.098 *	-0.031
<i>E. serotinus</i>	200	0.081	0.030	0.114 ***
	500	0.036	0.026	0.076 *
	700	0.026	0.021	0.057 .
	1000	0.029	0.034	0.051
<i>N. noctula</i>	200	-0.001	0.090	-0.011
	500	-0.001	0.123	0.006
	700	-0.038	0.114	-0.023
	1000	-0.028	0.142	-0.111
<i>N. leisleri</i>	200	0.121	0.056	0.116
	500	0.133	0.096	0.124
	700	0.117	0.121	0.077
	1000	0.093	0.094	0.050
<i>Myotis spp.</i>	200	0.234***	0.237 ***	0.191 ***
	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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