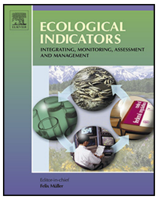
Ecological Indicators 74 (2017) 19–27

Contents lists available at ScienceDirect

Ecological Indicators

journal homepage: www.elsevier.com/locate/ecolind



Evaluating the use of *Myotis daubentonii* as an ecological indicator in Mediterranean riparian habitats

Adrià López-Baucells[a,b,*], Laura Casanova[a,**], Xavier Puig-Montserrat[c], Anna Espinal[d], Ferran Páramo[a], Carles Flaquer[a]

[a] Granollers Museum of Natural Sciences, Bat Research Group, Palaudàries, 102. Jardins Antoni Jonch Cuspinera, 08402 Granollers, Catalonia, Spain
[b] Center for Ecology, Evolution and Environmental Changes, Faculdade de Ciências da Universidade de Lisboa, Edifício C2, Campo Grande, 1749-016 Lisbon, Portugal
[c] Galanthus Association, Carretera de Juià 46, 17460 Celrà, Catalonia, Spain
[d] Servei d'Estadística Aplicada, Autonomous University of Barcelona, Campus UAB, Edifici CM7, 08193 Cerdanyola del Vallès, Barcelona, Catalonia, Spain

ARTICLE INFO

Article history:
Received 2 June 2016
Received in revised form 1 September 2016
Accepted 5 November 2016

Keywords:
Bioindicators
Ecological indicators
Conservation
Bats
Chiroptera
Citizen science

ABSTRACT

In recent years, interest and concern regarding biodiversity conservation have grown remarkably not only among conservationists but also amongst a wider public beyond scientific institutions. The monitoring of fauna and flora over long periods of time has been satisfactorily proven to be a viable tool for quantifying how environmental changes affect natural communities. Some bat species are regarded as good bioindicators, mainly due to their longevity and high sensitivity to environmental changes. *Myotis daubentonii* is one of the species most closely associated with riparian habitats in the north-east Iberian Peninsula, and is used as an ecological indicator in specific monitoring programs such as the Waterway Survey (United Kingdom) and the QuiroRius (Spain). Nonetheless, there is still great controversy as to whether *M. daubentonii* is a good biological indicator or not. While some authors accept it as a bioindicator, others point to the studies carried out in the U.K., Poland, Switzerland and Germany that show a remarkable increase in the numbers of this bat when pollution increases in canalized rivers, which suggest that it is in fact a generalist species.

Due to the lack of information regarding habitat-quality requirements in Daubenton's bats in the Mediterranean region and the species' potential as a bioindicator in riparian habitats, we aimed to 1) examine how QuiroRius data match other well-established biological indicators (IBMWP for invertebrates and QBR for riparian forests); 2) analyse how environmental variables at both local and landscape scales affect the presence of *M. daubentonii*; and 3) describe how environmental traits influence the relative abundance of *M. daubentonii*.

A total of 104 streams below 1000 m a.s.l. were simultaneously sampled using bat, macroinvertebrate and vegetation bioindicators. Despite having similar conservation aims, these three bioindicators did not provide consistent images of overall ecosystem quality and thus a multidisciplinary approach is necessary for a full analysis of the health of these riparian ecosystems. *M. daubentonii* were found more frequently in wide rivers with well-structured native riparian forests; on the other hand, landscape composition at broader scales and altitude had no influence on bat presence/abundance.

Thus, we suggest that QuiroRius could be used as a complementary bioindicator for analysing riparian forest quality but cannot be used alone as a tool for evaluating correctly overall riparian ecosystem health. Both relative abundance and/or presence/absence could be used as bioindicator surrogates given that the effect of microhabitat environmental predictors had similar impact on both these measures.

---

* Corresponding author at: Flat 5, 14 Sandy Grove, Manchester, M68QX Manchester, United Kingdom.
** Equally contributing co-first author.

E-mail addresses: adria.baucells@gmail.com (A. López-Baucells), lauracasanovabatlles@gmail.com (L. Casanova), xavierpuigm@gmail.com (X. Puig-Montserrat), Anna.Espinal@uab.cat (A. Espinal), ferranparamo@gmail.com (F. Páramo), cflaquer@ajuntament.granollers.cat (C. Flaquer).

http://dx.doi.org/10.1016/j.ecolind.2016.11.012
1470-160X/

1. Introduction

Since society became more aware of how human activities affect the natural environment, interest and concern regarding biodiversity conservation have grown remarkably among both conservationists and the general public, far beyond scientific institutions. Concern about how natural resources are being exploited and the impact such activity is having upon the natural environment is now part of everyday life. Thus, quantifying and monitoring ecosystem health has become a priority for conservationists as a way of understanding and minimizing as many environmental impacts as possible. Monitoring fauna and flora over time has been satisfactorily proven to be a viable conservation tool as it is essential to quantify how environmental changes affect natural communities (Castro-Luna et al., 2007; Barlow et al., 2015). In fact, certain species act as ecological or environmental indicators due to their sensitivity to a wide range of environmental stressors and to their predictable reactions to them (Jones et al., 2009). Some species are known to be sensitive to ecosystem changes such as shifts in water quality and increased eutrophication and pollution (Jones et al., 2009; Barlow et al., 2015). Thus, riparian ecosystems are often key habitats in monitoring programs as they are sensitive to the direct and notable effect of surrounding human settlements and to the accumulative effect of catchment areas.

In Europe, a number of bat species are considered to be good bioindicators (Flaquer and Puig-Montserrat, 2012) due to the rich trophic diversity present in this group (highly adapted to different prey such as spiders, moths, beetles, mosquitoes and even vertebrates); in addition, they sometimes provide pest control as a supplementary ecosystem service (Jones et al., 2009; Kasso and Balakrishnan, 2013; Barlow et al., 2015; Puig-Montserrat et al., 2015). Bat populations may be indirectly affected by water pollution, as some metals and organoclorines from contaminated river sediments have been found in Chironomidae flies, a common prey item amongst insectivorous bats (Kalcounis-Rueppell et al., 2007). Certain bats have degrees of response to habitat degradation that correlate closely to responses in other taxa (Jones et al., 2009). However, what makes bats good potential biological indicators for detecting past disturbance events is their slow reproductive rates. This means that population declines can be rapid, but also that take a long time to recover from declines. Bats need a constant healthy environment to rise in population numbers, and thus, past population declines can be easily detected and accurately assessed through a long-term monitoring programs (Jones et al., 2009; Barlow et al., 2015).

Myotis daubentonii and *M. capaccinii* are the only two trawling bat species (both closely associated with riparian habitats) found in the north-east Iberian Peninsula and the only species that are used as ecological indicators in specific monitoring programs such as the Waterway Survey in the UK. Daubenton's bat monitoring began in the UK during the 1990s as part of the National Bat Monitoring Program (NBMP), which was subsequently adapted in 2007 by the Granollers Museum of Natural Sciences and the Galanthus Association in Catalonia (NE Spain) to create a local protocol known as QuiroRius. In general, insectivorous trawling bat species are top predators on riparian insects, which is why they are widely considered to be good species models for understanding the effects of water quality at high trophic levels (Kalcounis-Rueppell et al., 2007). It is commonly assumed that the foraging activity of bats is directly related to insect abundance and also to the quality of riparian zones (Scott et al., 2010). Although both species are protected by current legislation, only *M. capaccinii* is classified as Endangered in Catalonia (*Decret legislatiu* 2/2008) and Spain (*Real Decreto* 139/2011). Thus, given this species' rarity, these monitoring programs use only data on Daubenton's bat (Flaquer and Puig-Montserrat, 2009). Roost segregation is well studied in both species and, whereas *M. capaccinii* mainly roosts in caves or similar underground tunnels, *M. daubentonii* can be found in urban environments such as in buildings or under bridges. Clear sexual elevational segregation has been reported in Daubenton's bat, with females recorded mainly up to around 900 m a.s.l. and males commoner at higher altitudes (Russo, 2002).

Great controversy exists as to whether *M. daubentonii* can be considered to be a good biological indicator. In some countries such as the United Kingdom it is accepted as a bioindicator (Abbott et al., 2009; Lintott et al., 2015), even though certain studies performed in that country, as well as in Poland, Switzerland and Germany, do show that there is a remarkable increase in this bat's numbers when pollution increases in canalized rivers, thereby suggesting that it is a more generalist species (Kokurewicz, 1995; Vaughan et al., 1996; Racey et al., 1998; Downs and Racey, 2006). Studies that show that *M. daubentonii* prefers upstream stretches of river support the hypothesis that this species could be affected by organic pollution accumulated downstream (Abbott et al., 2009). Data from bat monitoring programs in Britain show that *M. daubentonii* is more active in less polluted rivers and is associated with greater insect biodiversity (Abbott et al., 2009). Nevertheless, unlike other bat species in Europe, *M. daubentonii* has recently increased in number (Barlow et al., 2015), a finding attributed by some researchers to the increase in water pollution that leads to more eutrophic surface waters (with the consequent dramatic decrease in guild richness) and an increase in the availability of Chironomidae species (Abbott et al., 2009).

To our knowledge, no articles exist that report habitat quality requirements for Daubenton's bats in the Mediterranean region. In this study we aimed:

1) To compare how data for *M. daubentonii* compares to data generated by other well-established biological indicators (IBMWP and QBR) as a means of evaluating its potential as a biological indicator;
2) To analyse the effect of environmental variables at both local and landscape scales on the presence of *M. daubentonii*;
3) To describe how these environmental traits influence the relative abundance of *M. daubentonii* in the localities in which it is present.

2. Material and methods

The study was conducted in the NE Iberian Peninsula, a Mediterranean coastal region with a climate classified as 'dry-summer' or 'Mediterranean' according to Köppen's classification. This region is thus characterized by hot dry summers and mild rainy winters (www.eoearth.org). Bat sampling localities were homogeneously stratified along the upper, middle and lower reaches of rivers (Fig. 1) on 18 different Mediterranean rivers. Of these localities, 26 (=104 sampling points) were simultaneously and additionally sampled for macroinvertebrate, bat and plant biological indicators in August and September 2014. In order to ensure normal levels for nitrates (5–20 mg/L), pH (7–8), dissolved oxygen (40–80%) and temperature (16–24.1 °C), all these measurements were checked at every monitoring point on every sampling occasion.

2.1. Study species

Myotis daubentonii and *M. capaccinii* hunt almost exclusively over open water by 'trawling', a technique that consists of flying over and very close to the water surface in order to gaff emerging or floating prey, or catch insects just above the water surface (Warren et al., 2000; Abbott et al., 2009; Akasaka et al., 2009). Both are the only bat species that make figure-of-eight turns when flying over open water (Abbott et al., 2009). Daubenton's bats usually forage over smooth calm waters bordered by well-developed riparian vegetation along rivers over 5 m in width (Warren et al., 2000), and seems to prefer trees on both river banks and sites with high water quality. Many European populations of Daubenton's bats that declined during the past century due to their sensitivity to habitat change (Scott et al., 2010) are currently stable or even display positive trends (Barlow et al., 2015). These improvements could be driven by the influence of legal protection and greater awareness of the importance of bat conservation, and/or by changes in climate and agricultural practices (Barlow et al., 2015). However, global analysis does still indicate that Daubenton's bat is an especially vulnerable species due to its limited habitat preferences and dependence on non-polluted feeding areas (Warren et al., 2000).

2.2. Bat activity (QuiroRius index)

Long-term data on bat activity was taken from the QuiroRius database (2007–2014). This volunteer monitoring program was created by the Granollers Museum of Natural Sciences and the Galanthus Association with the principal aim of obtaining data on the relative abundance of Daubenton's bat in the riparian habitats in the region. In the QuiroRius monitoring methodology – adapted

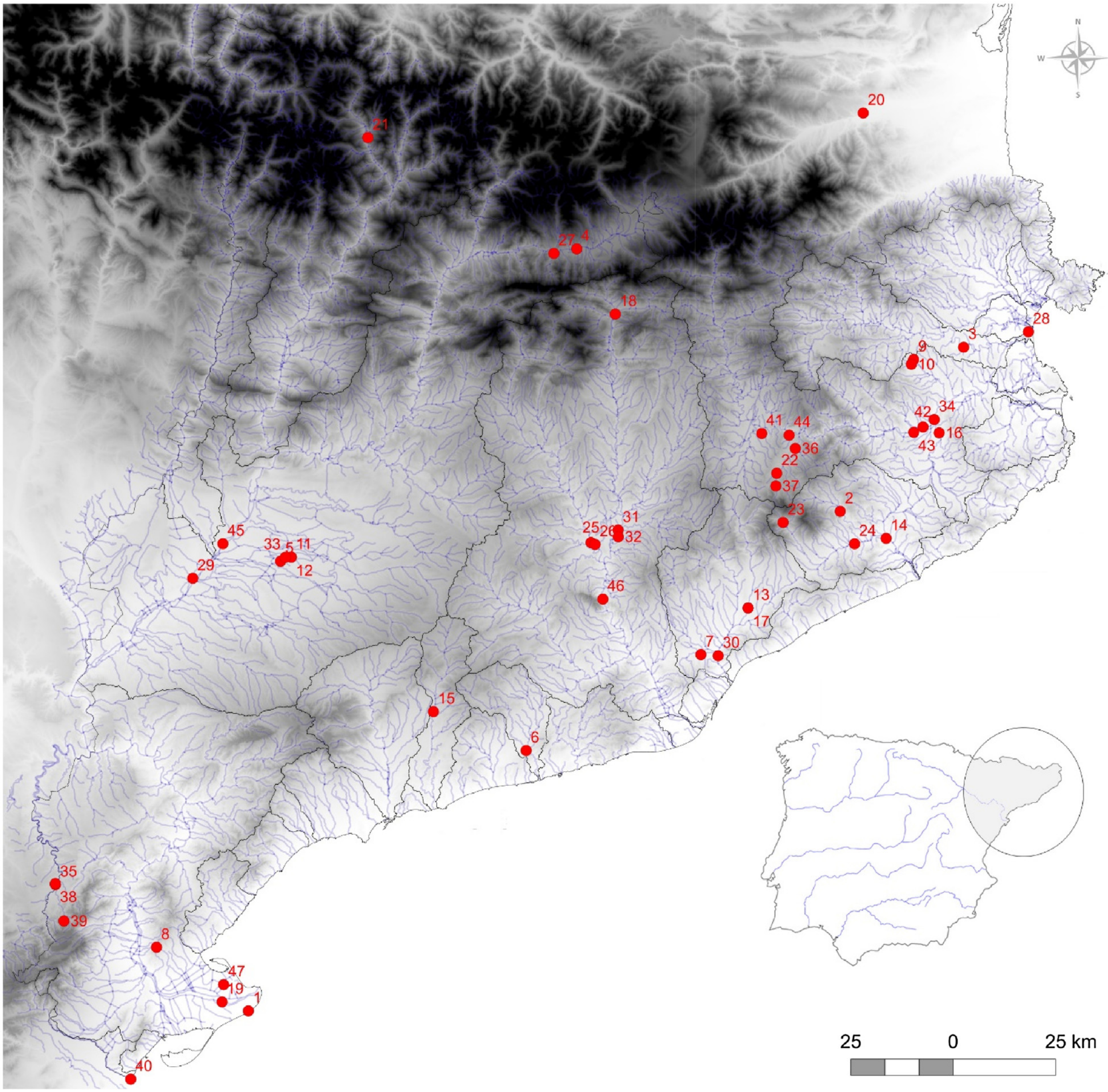


Fig. 1. Monitoring points used in this study in Catalonia (NE Iberian Peninsula). 1 Alfacada; 2 Arbucies; 3 Bàscara; 4 Basses de Gallissar; 5 Canal Urgell a Vila-sana; 6 Castellet; 7 Cerdanyola del Vallès; 8 Ciutat de Tortosa; 9 Estany Banyoles Nord; 10 Estany Banyoles Sud; 11 Estany d'Ivars i Vila-sana; 12 Estany d'Ivars; 13 Figaró; 14 Fogars de Tordera; 15 Gaià; 16 Girona Est; 17 Granollers; 18 Guardiola de Berguedà; 19 Illa del Riu; 20 Ille Sur Têt; 21 La Mollera de Guingueta d'Àneu; 22 Les Cabrades Guilleries; 23 Llavina Montseny; 24 Llobateres Sant Celoni; 25 Manresa Parc Fluvial el Pont Nou; 26 Manresa Passeig del Riu; 27 Martinet de Cerdanya; 28 Mig dos Rius; 29 Mitjana de Lleida; 30 Montcada; 31 Navarcles Oest; 32 Navarcles Sud; 33 Oest Estany d'Ivars i Vila-sana; 34 Indoor swimming pool GEIEG, Avinguda de França roundabout; 35 Pont de la Vall de Safan Pont de Lledó; 36 Pont de Malafogassa; 37 Riera de Sant Segimon; 38 Riu Algars Pont de Lledó; 39 Riu Estrets; 40 Mouth of river Sènia; 41 Roda de Ter; 42 Salt Est AP7; 43 Salt Oest AP7; 44 Sau; 45 Sot del Fuster; 46 Sud de Monistrol; 47 Tora del Mig.

in 2007 from the British NBMP – each volunteer is responsible for an independent sampling station, which consists of a 1-km long transect that includes four sampling points. Each point is characterized by the following parameters – width, depth, section width, channel structure and water speed – and is surveyed for 10 min/night, one hour after the sunset, twice per year in August (with a minimum of 10 days between the first and the second sampling nights). Bat passes at each point are quantified (by direct observation) in terms of the number of events (an event is when a bat crosses the light beam). A light beam is shone perpendicular to the river and a heterodyne detector with microphone is placed at a 45° angle to the water surface (Flaquer and Puig-Montserrat, 2009). The detector, tuned to 40 kHz, allows the observer to hear the characteristic calls of *M. daubentonii*. When a Daubenton's bat approaches, the observers wait until it passes through the light before counting it; identification must always be confirmed by observation of this bat's characteristic flight pattern (Abbott et al., 2009). Bat activity is calculated as the number of bat passes/night. This monitoring protocol is highly biased to female and juvenile foraging activity in stations below an altitude of 900 m a.s.l. (Russo, 2002).

Bat activity data have been collected since 2007 by trained volunteers at 180 monitoring points and 47 stations. The activity index is measured by the number of bat passes/min and is taken in August-September after the parturition period. In 2014, 26 additional localities (four sampling points per station, sampled twice per year, giving a total of eight replicates at 104 monitoring points) were simultaneously sampled by specialists for bats, macroinvertebrates (IBMWP) and vegetation (QBR).

2.2.1. Macroinvertebrates (IBMWP index)

Data for macroinvertebrates were collected using the IBMWP (Iberian Biological Monitoring Working Party) protocol (Tafur et al., 2010). This index evaluates the quality of rivers by considering the organic pollution tolerance of the invertebrate groups present at the sampled sites. Pollution-intolerant invertebrates such as *Tricoptera* generally score higher than those that are less sensitive. The Average Score per Taxon (ASTP) can be calculated by dividing the IBMWP by the number of observed families at each monitoring point. This measurement provides a value for the balance between pollution-tolerant and pollution-sensitive families and uses invertebrate presence/absence data instead of abundance (Scott et al., 2010; Abbott et al., 2009)

2.2.2. Riparian vegetation (QBR index)

Data for riparian vegetation was obtained using the QBR index (Index of Riparian Quality; Munné et al., 1998). This index uses a combination of the 'total riparian cover', 'cover structure', 'cover quality' and 'river channel naturalness' to quantitatively evaluate the quality of the riparian vegetation (Fornells et al., 1998; Munné et al., 1998; Suárez et al., 2002; Colwell and Hix, 2008). These variables must be evaluated and quantified according to the guidelines and questionnaires in the QBR index. 'Total riparian cover' measures the percentage of cover of any kind of plant except annuals. The vegetation structure is not considered, only the total cover. The score for 'cover structure' measures the complexity of the vegetation system and depends on the percentage of cover that is forest or, if trees are absent, that is shrubs and other low vegetation. Linear arrangements (mostly plantations) or disconnected patches may lower the initial value, while helophytes in the channel or the presence of shrubs below the forest increase the score. 'Cover quality' evaluates the number of species of true riparian trees and the geomorphology of the river. Both a tunnel disposition of trees and gallery structure of vegetation increase the score in terms of the percentage of cover. Allochthonous species, on the other hand, lower the index score. Finally, 'River channel naturalness' quantifies morphological changes produced in the alluvial terraces, including channel reduction due to agricultural activities, the elimination of meanders and the straightening of river courses.

2.2.3. Landscape composition

The effect of landscape composition on *M. daubentonii* foraging activity was quantified using satellite images based on the 1:50,000-scale (30 × 30 m resolution) Catalan habitat cartography (Departament de Medi Ambient i Habitatge, 2005). Five environmental variables were included in the analysis: altitude, forest cover, urban cover, shrub cover and riparian cover. These environmental variables were calculated by reclassifying the original cover layers in the local cartography (Mapa d'Hàbitats i Model Digital d'Elevacions (DEM) de la Generalitat de Catalunya) with QQIS v. 2.0.1 Dufour (Germany), combined with the R packages "maptools" v. 0.8-37 (Bivand and Lewin-Koh, 2013), "rgdal" v. 1.1-3 (Bivand et al., 2013), "raster" v. 2.5-2 (Hijmans and Van Etten, 2013) and "rgeos" v. 0.3-15 (Bivand and Rundel, 2013). Landscape metrics were calculated in buffers with radii of 250, 1000 and 5000 m around the stations. The largest buffer was chosen by taking into account the mean foraging distance for this species during the breeding season (Dietz et al., 2006). For these analyses, all localities available in the monitoring database were included.

2.2.4. Statistical methods

1) In order to test the capacity of the QuiroRius data to act as a bioindicator index in riparian habitats, Spearman's rank order correlations were carried out between the bat activity index (as a continuous variable) and the IBMWP and QBR indices. Mann Whitney *U* tests comparing the results of the indices were also performed between localities with and without bats. Measures for all biological indices were standardized (1–5 for all indices; Appendix A: Table A1) to be able to test for agreement between results (considering sampling stations as study units and points as replicates). Kappa statistics were used to test the degree of agreement between different classifications. This calculation is based on the difference between the proportion of agreement present in our data and the agreement expected only by chance. Kappa provides a standardized measure between −1 and 1, where 1 is perfect agreement, 0 is directly related to chance, while negative values indicate less agreement than would be expected by chance (Viera and Garrett, 2005). Weighted Kappa statistics were used as they assign less weight to agreement if categories are further apart in order to also include the proximity of results in the tests.
2) To analyse the effect of covariates in certain rivers when *Myotis daubentonii* is present, the bat activity index was categorized as a binary presence/absence variable. Two different generalized linear models (GLMs) for a binary response were established, the first with several microhabitat covariables: all QBR components (total cover, cover naturalness, cover structure, cover quality) and river width; and the second including only landscape variables: land cover and altitude. The results of these models are presented using the corresponding odds ratio (OR) and their confidence intervals (Hosmer and Lemeshow, 2000).
3) Generalized linear models (GLMs) were used to evaluate how environmental variables (at both landscape and microhabitat scales) affect bat activity at the localities in which *M. daubentonii* occurs. Following Burnham and Anderson (2003), the most parsimonious models were selected using Akaike's Information Criterion corrected for small samples sizes (AICc). The best models were obtained selecting models with an AICc difference from the best model (Δi) <2, using the R packages "bestglm" v. 0.34 (McLeod and Changjiang, 2014).

To avoid multicollinearity, the correlation between the predictors in the models was calculated using the Corrplot package

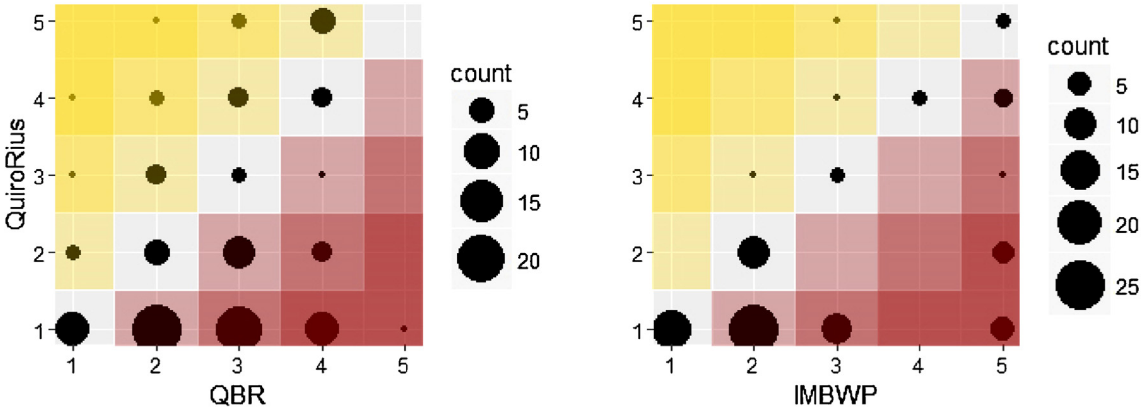


Fig. 2. Comparison between the river health classifications at the same sampling locations generated by the QuiroRius and QBR bioindicators (weighted kappa statistic = 0.43), and between QuiroRius and IBMWP (weighted kappa statistic = 0.39). Standardized classifications ranging from 1 to 5 (Appendix A). Size of circles: N° of sampled locations with each classification; Orange: Daubenton's bioindicator overestimating ecosystem quality; Red: Daubenton's bioindicator underestimating ecosystem quality.

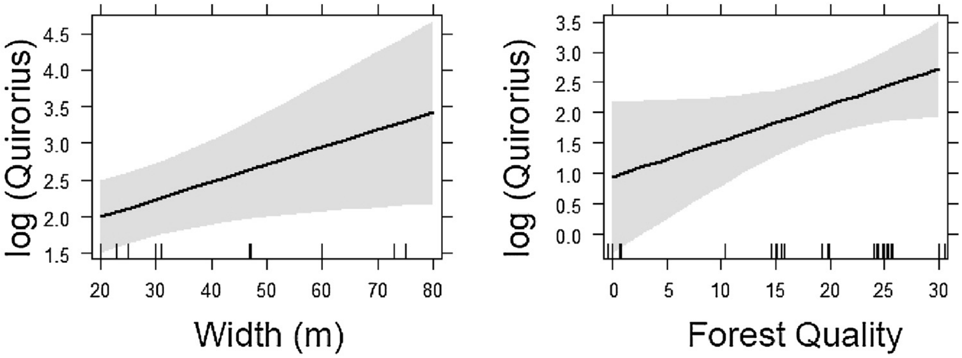


Fig. 3. Effect of the width and the Riparian Forest Quality on the probability of the presence of *Myotis daubentonii* modelled with a logistic generalized linear model and plotted while compensating for the effect of the other environmental predictors.

(Wei, 2013); all predictors correlated with others with r > 0.8 were excluded. Additionally, the Variance Inflation Factor (VIF) of each predictor was calculated to avoid autocorrelation between predictors. All predictors with VIFs <3 were included (Neter et al., 1990).

All statistical analyses were carried out using R software, version 3.2.4. (R Foundation for Statistical Computing); significance levels were fixed at 0.05.

3. Results

1) Agreement between different ecological indicators

In general, sample rivers with the presence of Daubenton's bat had higher riparian vegetation quality (QBR) than those where the species is absent (W-value = 946.5, p = 0.0327, n = 102). The IBMWP index for all these localities, however, showed no significant differences in the quality of macroinvertebrate communities.

In order to test the potential of the QuiroRius data as an ecological indicator for riverine habitats, we computed the linear correlation coefficient between bat activity and QBR, and between bat activity and IBMWP. In all cases, a very low correlation coefficient was obtained (Adj. R-squared for QBR: 0.032 and IMBWP: 0.020). Nevertheless, for the standardized final values of the three indices (Appendix A Table A1), the results for QuiroRius were closer to the IBMWP results than to the QBR results. At most sampling locations the agreement between the three indices was low and usually underestimated the quality of the sampled ecosystem (Fig. 2). The indices actually provided significantly contrasting results, as shown by the weighted kappa statistics, which were 0.39 between IBMWP-Quirorius and 0.43 between QBR-Quirorius. In any two of the paired-index comparisons, a 'moderate' agreement was detected (set as 0.57 by Viera and Garrett, 2005). In terms of bat activity, QBR weakly corresponded, while IBMWP was never statistically correlated (Fig. 3).

2) Which environmental factors determine the presence of Daubenton's bat in a given river?

The best model to explain the effect of environmental variables on Daubenton's bat presence included 'river width' and 'riparian forest quality' as predictor variables. In fact, we found that, of all local environment variables, both of these variables had a statistically significant effect on bat presence at a microhabitat scale (Table 1)—i.e. bats were more often present in forests with a higher quality ranking and wider waterways. We expected for a one-unit increase in forest quality, double the odds of detecting Daubenton's bats; and 3 times of odds for the river width. Thus, a combination of complex gallery and/or tunnel-stratified vegetation without too many allochthonous plant species in wide rivers favours the presence of Daubenton's bat more than either the total cover, the structure or the naturalness of the channel. On the other hand, when considering landscape composition, we found that in both the 250-m and 1000-m buffers no environmental variable had a statistically significant effect on species presence. Yet, in larger areas (5000-m buffers), forest cover did begin to affect presence (Table 1) and Daubenton's bats were more often found in well-forested areas. We found that a one-unit increase in forest cover increase in almost

Table 1
Summary of the environmental factors influencing *Myotis daubentonii* presence based on the selected models as per Burnham and Anderson (2003).

Model for *Myotis daubentonii* presence at microhabitat scale

Bat presence ~ River width + Riparian forest quality, family = binomial

	Estimate	Std. Error	z value	Pr(>|z|)
(Intercept)	−0.4941	0.2396	−2.062	0.039179*
River Width	1.0846	0.3286	3.301	0.000965*
Riparian Forest Quality	0.7030	0.2737	2.569	0.010210*

	OR	2.5%	97.5%
Intercept	0.610	0.376	0.972
River width	2.958	1.685	6.274
Riparian Forest Quality	2.019	1.219	3.608

Model for *Myotis daubentonii* presence at landscape scale in a 5000-m buffer

Bat presence ~ Forest cover + Urban cover, family = binomial

	Estimate	Std. Error	z value	Pr(>|z|)
(Intercept)	1.7181	0.5226	3.288	0.00101
Forest cover	1.1737	0.6137	1.912	0.05584*
Urban cover	−0.5582	0.3483	−1.603	0.10902

	OR	2.5%	97.5%
Intercept	5.573	2.326	19.776
Forest cover	3.233	1.159	13.718
Urban cover	0.572	0.270	1.122

3 times the odds of detecting Daubenton's bats. The selected model included both urban and forest cover as predictors.

3) How do environmental factors influence the relative abundance and levels of foraging activity in Daubenton's bat?

When considering only localities where this bat occurs, 'Cover quality' and 'Width' again had a statistically significant effect at microhabitat scale upon relative bat abundance in all selected models (Table 2). Bats thus prefer to forage along wide rivers, which leads to an increase in overall bat activity levels wherever riparian forests hold a certain number of native species, tree cover is tunnel-like in structure and there is a complex disposition of gallery vegetation. At landscape scale (250-, 1000- and 5000-m buffers), we were unable to detect any environmental predictor – either for land cover or altitude – that had a statistically significant effect on relative bat abundance.

4. Discussion

1) Agreement between ecological indicators

Despite having similar conservation aims, the three bioindicator indices that we tested did not afford consistent images of ecosystem quality: QBR corresponded only weakly to activity in Daubenton's bat, while IBMWP was never statistically correlated to bat activity. Furthermore, we found no significant correlation between QBR and IBMWP. Given that *M. daubentonii* activity levels as a continuous quantitative biological indicator are only weakly supported by QBR and IBMWP, we recommend a multi-faceted approach when trying to evaluate ecosystem health. According to the other biological indices, fewer bat passes are not always related to poor-quality riparian forests. Nevertheless, greater levels of bat activity could help identify a good-quality ecosystem as higher abundances usually reflect high-quality riparian forests.

The fact that trawling bats are supposed to basically feed on pollution-tolerant *Chironomidae* that withstand low oxygen levels (Scott et al., 2010) apparently contradicts their preference for high quality waters. It is generally affirmed that, as an aquatic habitat specialist, any change in water quality negatively affecting its prey base (i.e. macroinvertebrates) will harm *M. daubentonii* populations (Kalcounis-Rueppell et al., 2007). However, no significant relationship was detected between the invertebrate index and bat activity, which could be explained by the fact that IBMWP only takes into account larval forms that live in the water and ignores the flying forms that constitute the vast proportion of the available feeding resources for bats (Flavin et al., 2001). As bats are highly mobile, they can switch foraging areas quite rapidly, thereby making presence/absence relationships difficult to analyse. Although we expected that the predominance of specific insect taxa would determine the relative abundance of Daubenton's bat, our results showed no significant relationship between any invertebrate taxon and bat activity. Thus, the IBMWP index does not provide the same information and results for ecosystem health as QuiroRius and so both indices should be considered individually and interpreted separately in specific management cases.

The literature provides evidence that *M. daubentonii* feeds on several prey types besides Chironomidae (Warren et al., 2000; Biscardi et al., 2007; Abbott et al., 2009). For instance, 80% of the diet of Daubenton's bat in Ireland consisted of Chironomidae/Ceratopogonidae (24%), Nematocera (21%), other Diptera (10%) and Trichoptera (26%) (Flavin et al., 2001). Totals of 44% and 30% Trichoptera have been found in diet items for this bat in Scotland and Ireland, respectively. They mainly feed on Chironomidae early in the year (April) and at the end of the year (August) when adult Trichoptera are available and seem to positively select prey items from this group of insects (Abbott et al., 2009). Further diet studies should be performed to confirm whether or not *M. daubentonii* positively selects Chironomidae as prey in the Mediterranean region.

2) Which environmental traits determine the presence of Daubenton's bat along a given river?

The literature suggests that well-developed riparian vegetation on riverbanks with trees on both sides is essential for the presence of this bat (Warren et al., 2000; Biscardi et al., 2007). This need for trees on both banks may be related to the distribution of insects that feed close to trees and hedges since many invertebrates use this type of vegetation as protection from the wind (Warren et al., 2000; Wickramasinghe et al., 2004). In fact, river meanders are known to create marginal habitats where an increase in aquatic larvae enhances prey availability for bats (Ober and Hayes, 2008;

Table 2

Summary of environmental factors influencing *Myotis daubentonii* abundance or foraging activity levels based on selected models as per Burnham and Anderson (2003).

Model for *Myotis daubentonii* abundance at microhabitat scale

Bat mean abundance ~ River width +Riparian Forest Quality, family = Gaussian

Coefficients:

	Estimate	Std. Error	t value	Pr(>|t|)
(Intercept)	2.0995	0.2366	8.873	1.76e-10***
River width	0.6068	0.2581	2.351	0.0245*
Riparian Forest Quality	0.5329	0.2581	2.064	0.0464*

Akasaka et al., 2009). Additionally, this bat has been reported to use riverbank vegetation as corridors, which confirms the importance of rivers as environmental connectors. This species tends to use channels with trees that form continuous lines (preferably with both banks vegetated) to be able to commute between feeding habitats and roosts (Warren et al., 2000; Biscardi et al., 2007; Scott et al., 2010). In our study area, the environmental variables that most influenced bat presence were 'riparian forest quality' and 'width', which fully agrees with previous findings and the available literature, and suggests that complex forests with native trees and tunnel-structured vegetation are more likely to harbour bat populations, above all in wider rivers. At landscape scale, forest cover only weakly (but positively) affected bat presence at larger scales (buffer with radius of 5 km).

The fact that all bat surveys were conducted below 1000 m a.s.l. and were strongly biased by the presence of large Daubenton's maternity colonies feeding along rivers (females and juveniles) suggests that between sea-level and 1000 m a.s.l. altitude has no significant effect on the establishment of female congregations. This finding is clearly supported by Russo (2002), who described only a strong segregation between males and females at around only 900 m a.s.l.

Due to the long time required for bats to recover declining populations, the absence of *M. daubentonii* from a certain river could be due to historical factors (such as earlier pollution events along the river or in the surrounding human settlements), which would explain why so many apparently relevant environmental variables for this bat species do not have any influence, and why we found that some ostensibly 'healthy' rivers had no Daubenton's bats. Those events could have led to local extinctions from which this bat has not yet recovered; alternatively, the species may not forage in a particular area due to historical memory. However, long-term monitoring programs are needed to better explore these speculations.

3) How do environmental traits influence levels of foraging activity in Daubenton's bat?

Results concerning microhabitat characteristics confirmed the expected hypothesis and concurs with the information in the literature on this subject: bats clearly prefer wide rivers bordered by well-structured and native riparian forests (Biscardi et al., 2007; Warren et al., 2000). According to the literature, Daubenton's bat prefers a 5–10 m or >10 m inter-bank distance to narrow channels; the inter-bank distance influences their foraging activity, while narrow stretches of rivers affect their manoeuvrability and reduce the available foraging area (Warren et al., 2000; Biscardi et al., 2007; Scott et al., 2010). Well-structured forests provide enough space in which to fly and a greater complexity of vegetation that maximizes insect diversity (Ober and Hayes, 2008). It is known that Daubenton's bat tends to avoid cluttered environments and fast waters because rapids may interfere and hamper prey detection by echolocation (Rydell et al., 1999; Warren et al., 2000; Biscardi et al., 2007), while more complex structures may help increase insect availability. As previously reported by other studies, good stretches of river for Daubenton's bat should have limited but well-structured vegetation cover that is accessible for feeding (Vindigni et al., 2009). Additionally, bats are thought to use lines of tree as navigational aids when commuting (Scott et al., 2010) and so the cover structure may also play an important role by providing bats with clues and corridors when commuting.

At landscape scale, our results did not detect any relationship between bats' foraging activity and any particular land cover, which means that Daubenton's foraging activity will rarely depend on large natural areas but is, rather, highly influenced by the quality of the local microhabitat and of the riparian forest along the river.

5. Conclusions

We provide new and unique data from a *Myotis daubentonii* long-term monitoring program in the Mediterranean that can be used to decipher the controversy about the potential use of this species as an ecological indicator. Our analyses were based on a new perspective that considers the evaluation of riparian forest condition as our primary ecosystem condition baseline in addition to water quality and the availability of invertebrates.

Kokurewicz (1995) suggested that this species would benefit from polluted and eutrophic waters and the increase of *Chironomidae* insects (Poland), as opposed to those who consider *M. daubentonii* to be a viable biological indicator, which may explain the increases of the bat in populations in some European regions. Some support for this hypothesis was provided by Racey et al. (1998) and Vaughan et al. (1996) using data from Scotland and England, respectively. However, most of the support for this opportunistic feeding behaviour was explained by investigating diet composition (e.g. an increased abundance of pollution-tolerant insects), which has been proven to vary across regions and seasons (Abbott et al., 2009). In fact, Flavin et al. (2001), among other authors, contradict the theory that *M. daubentonii* benefits from polluted areas by suggesting that it positively selects other taxa to predate upon. As Abbott et al. (2009) pointed out, the divergence of results, which was controversial among chiropterologists, might actually be a result of sample size bias; as some authors used small data sets from localized regions (Racey et al., 1998; Vaughan et al., 1996). When data is obtained from a large-scale monitoring project, *M. daubentonii* is shown to be more active in less polluted rivers or, as highlighted in our study-case, rivers with well-conserved riparian forests.

We suggest that *M. daubentonii* can be used as a biological indicator but only in very specific areas, at a local or micro-habitat scale, where the quality of riparian forests is high. Despite all having similar aims in the context of biological conservation, the three tested biological indicators do not provide consistent images of global ecosystem quality. Results must be carefully examined and interpreted. Of these three biological indicators, QuiroRius is useful as a biological indicator providing complementary information on riparian forest quality but cannot be used alone to fully evaluate riparian ecosystem health. Additionally, both relative bat abundance and presence/absence could be used as surrogate bioindicators for riparian forest quality as the effect of microhabitat environmental predictors have a similar impact on bat activity levels as on their presence. Our results indicate that wide rivers with well-structured native riparian forests are the best habitat for females (and probably for the presence of maternity colonies) and also that landscape composition and altitude may be disregarded when analysing the possibilities of using *Myotis daubentonii* as a biological indicator (only between 0 and 1000 m) as its presence is mainly influenced by local riparian characteristics.

Nonetheless, general research on the responses of *M. daubentonii* to habitat change and degradation has yet to publish any robust conclusions and tends to embrace biological knowledge gaps about how to extrapolate the results. The effects of pollution and global environmental degradation on bats may be more convoluted than previously indicated in the literature. Further research at a continental scale is required to understand the complex relationship between water eutrophication, riparian forest quality, insect availability and the abundance of Daubenton's bats.

Acknowledgements

We would like to thank Toni Arrizabalaga for all the logistic support, Miriam Carrero for her valuable contribution during the fieldwork, Eva de Lecea from the Associació Hàbitats for providing field equipment and logistic support, and Constantí Stefanescu, who disinterestedly contributed to the project by reviewing the manuscript and providing constructive suggestions that greatly improved the quality of the results. We also want to extend our gratitude to James Kemp for proofreading the manuscript. This work could never have been performed without the invaluable contribution from all the volunteers and collaborators with the project. This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Appendix A.

Table A1

Standardized values for the three tested biological indicators: *Myotis daubentonii* foraging activity (QuiroRius), macroinvertebrates (IBMWP, Iberian Biological Monitoring Working Party) and the riparian vegetation (QBR, Quality del Bosc de Ribera).

QuiroRius	IBMWP	QBR	Score
Natural >100	Natural	Riparian habitat in natural condition $\geq$95	5
Good quality 21–100	Good	Some disturbance, good quality 75–90	4
Fair quality 6–20	Fair	Significant disturbance, fair quality 55–70	3
Bad quality 1–5	Bad	Serious alteration, bad quality 30–50	2
Very bad quality 0	Very bad	Extreme degradation, very bad quality $\leq$25	1

References

Abbott, I.M., Sleeman, D.P., Harrison, S., 2009. Bat activity affected by sewage effluent in Irish rivers. Biol. Conserv. 142 (12), 2904–2914, http://dx.doi.org/10.1016/j.biocon.2009.07.012.

Akasaka, T., Nakano, D., Nakamura, F., 2009. Influence of prey variables, food supply, and river restoration on the foraging activity of Daubenton's bat (*Myotis daubentonii*) in the Shibetsu River, a large lowland river in Japan. Biol. Conserv. 142 (7), 1302–1310, http://dx.doi.org/10.1016/j.biocon.2009.01.028.

Barlow, K.E., Briggs, P.A., Haysom, K.A., Hutson, A.M., Lechiara, N.L., Racey, P.A., Walsh, A.L., Langton, S.D., 2015. Citizen science reveals trends in bat populations: the national bat monitoring programme in Great Britain. Biol. Conserv. 182, 14–26, http://dx.doi.org/10.1016/j.biocon.2014.11.022.

Biscardi, S., Russo, D., Casciani, V., Cesarini, D., Mei, M., Boitani, L., 2007. Foraging requirements of the endangered long-fingered bat: the influence of micro-habitat structure, water quality and prey type. J. Zool. 273 (4), 372–381, http://dx.doi.org/10.1111/j.1469-7998.2007.00337.x.

Bivand, R., Lewin-Koh, N., 2013. maptools: Tools for reading and handling spatial objects. R package version 0. 8–27.

Bivand, R., Rundel, C., 2013. Rgeos: interface to geometry engine-Open source (GEOS). R package version 0. 3–2.

Bivand, R., Keitt, T., Rowlingson, B., 2013. rgdal: Bindings for the geospatial data abstraction library. R package version 0. 8–10.

Burnham, K.P., Anderson, D.R., 2003. Model Selection and Inference: A Practical Information-Theoretic Approach. Springer, New York.

Castro-Luna, A.A., Sosa, V.J., Castillo-Campos, G., 2007. Quantifying phyllostomid bats at different taxonomic levels as ecological indicators in a disturbed tropical forest. Acta Chiropterologica 9 (1), 219–228, http://dx.doi.org/10.3161/1733-5329(2007)9[219:QPBADT]2.0.CO;2.

Colwell, S.R., Hix, D.M., 2008. Adaptation of the QBR index for use in riparian forests of central Ohio. In: Jacobs Douglass, F., Michler Charles, H. (Eds.), Proceedings of the 16th Central Hardwoods Forest Conference. West Lafayette, Newtown Square, PA: U.S. Department of Agriculture, Forest Service, Northern Research Station, pp. 331–340.

Dietz, M., Encarnação, J.A., Kalko, E.K., 2006. Small scale distribution patterns of female and male Daubenton's bats (*Myotis daubentonii*). Acta Chiropterologica 8 (2), 403–415, http://dx.doi.org/10.3161/1733-5329(2006)8[403:SSDPOF]2.0.CO;2.

Downs, N.C., Racey, P.A., 2006. The use by bats of habitat features in mixed farmland in Scotland. Acta Chiropterologica 8 (1), 169–185, http://dx.doi.org/10.3161/1733-5329(2006)8[169:TUBBOH]2.0.CO;2.

Flaquer, C., Puig-Montserrat, X., 2009. Seguiment de ratpenats aquàtics de Catalunya. Granollers Museum of Natural Sciences, Granollers, Spain.

Flaquer, C., Puig-Montserrat, X., 2012. Proceedings of the International Symposium on the Importance of Bats as Bioindicators , Granollers Museum of Natural Sciences Edicions, Granollers, Spain.

Flavin, D.A., Biggane, S.S., Shiel, C.B., Smiddy, P., Fairley, J.S., 2001. Analysis of the diet of Daubenton's bat *Myotis daubentonii* in Ireland. Acta Theriol. 46 (1), 43–52, http://dx.doi.org/10.1007/BF03192415.

Fornells, N.P., Solá, C., Munné, A., 1998. QBR: Un índice rápido para la evaluación de la calidad de los ecosistemas de ribera. Tecnología del Agua 175, 20–39.

Hijmans, R.J., Van Etten, J., 2013. Raster: geographic data analysis and modeling. R package version 2. 1–49.

Hosmer, D.W., Lemeshow, S., 2000. Introduction to the logistic regression model. In: Applied Logistic Regression, second edition., pp. 1–30, http://dx.doi.org/10.1002/0471722146.ch1.

Jones, G., Jacobs, D.S., Kunz, T.H., Willig, M.R., Racey, P.A., 2009. Carpe noctem: the importance of bats as bioindicators. Endanger. Species Res. 8 (1–2), 93–115, http://dx.doi.org/10.3354/esr;1;00182.

Kalcounis-Rueppell, M.C., Payne, V.H., Huff, S.R., Boyko, A.L., 2007. Effects of wastewater treatment plant effluent on bat foraging ecology in an urban stream system. Biol. Conserv. 138 (1), 120–130, http://dx.doi.org/10.1016/j.biocon.2007.04.009.

Kasso, M., Balakrishnan, M., 2013. Ecological and economic importance of bats (Order Chiroptera). ISRN Biodivers. 2013, http://dx.doi.org/10.1155/2013/187415.

Kokurewicz, T., 1995. Increased population of Daubenton's bat (*Myotis daubentonii* (Kuhl, 1819)) (Chiroptera: Vespertilionidae) in Poland. Myotis 32–33, 155–161.

Lintott, P.R., Bunnefeld, N., Park, K.J., 2015. Opportunities for improving the foraging potential of urban waterways for bats. Biol. Conserv. 191, 224–233, http://dx.doi.org/10.1016/j.biocon.2015.06.036.

McLeod, A.I., Changjiang, X., 2014. bestglm: best subset GLM.-R package ver. 0.34.

Munné, A., Solà, C., Rieradevall, M., Prat N., 1998. Índex QBR. Mètode per a l'avaluació de la qualitat dels ecosistemes de ribera. Estudis de la Qualitat Ecológica deis Rius. Diputació de Barcelona, Área de Medí Ambient. Barcelona, Spain.

Neter, J., Wasserman, W., Kutner, M.H., 1990. Applied Linear Statistical Models: Regression, Analysis of Variance, and Experimental Designs. Irwin, New Jersey.

Ober, H.K., Hayes, J.P., 2008. Influence of vegetation on bat use of riparian areas at multiple spatial scales. J. Wildl. Manage. 72 (2), 396–404, http://dx.doi.org/10.2193/2007-193.

Puig-Montserrat, X., Torre, I., López-Baucells, A., Guerrieri, E., Monti, M.M., Ràfols-García, R., Ferrer, X., Gisbert, D., Flaquer, C., 2015. Pest control service provided by bats in Mediterranean rice paddies: linking agroecosystems structure to ecological functions. Mamm. Biol. 80 (3), 237–245, http://dx.doi.org/10.1016/j.mambio.2015.03.008.

Racey, P.A., Swift, S.M., Rydell, J., Brodie, L., 1998. Bats and insects over two Scottish rivers with contrasting nitrate status. Anim. Conserv. 1, 195–202, http://dx.doi.org/10.1111/j.1469-1795.1998.tb00029.x.

Russo, D., 2002. Elevation affects the distribution of the two sexes in Daubenton's bats *Myotis daubentonii* (Chiroptera: Vespertilionidae) from Italy. Mammalia 66 (4), 543–552, http://dx.doi.org/10.1515/mamm.2002.66.4.543.

Rydell, J., Miller, L.A., Jensen, M.E., 1999. Echolocation constraints of Daubenton's bat foraging over water. Funct. Ecol. 13 (2), 247–255, http://dx.doi.org/10.1046/j.1365-2435.1999.00304.x.

Scott, S.J., McLaren, G., Jones, G., Harris, S., 2010. The impact of riparian habitat quality on the foraging and activity of pipistrelle bats (Pipistrellus spp.). J. Zool. 280 (4), 371–378, http://dx.doi.org/10.1111/j.1469-7998.2009.00670.x.

Suárez, M.L., Vidal-Abarca Gutiérrez, M.R., Sánchez-Montoya, M.D.M., Alba Tercedor, J., Álvarez, M., Avilés, J., Bonada i Caparrós, N., Casas, J., Jáimez-Cuéllar, P., Munné i Torras, T., Pardo, I., 2002. Las riberas de los ríos mediterráneos y su calidad: el uso del índice QBR. Limnetica 21 (3–4), 135–148.

Tafur, C.M., Revilla, M.H., Ruiz, W.P., Aguilar, R.G., Guzmán, I.A., 2010. El indice Biological Monitoring Working Party (BMWP), modificado y adaptado a tres microcuencas del Alto Chicama. La Libertad. Perú. 2008. Sciendo 13 (2), 5–20.

Vaughan, N., Jones, G., Harris, S., 1996. Effects of sewage effluent on the activity of bats (Chiroptera: Vespertilionidae) foraging along rivers. Biol. Conserv. 78, 337–348, http://dx.doi.org/10.1016/S0006-3207(96)00009-2.

Viera, A.J., Garrett, J.M., 2005. Understanding interobserver agreement: the kappa statistic. Fam. Med. 37 (5), 360–363.

Vindigni, M.A., Morris, A.D., Miller, D.A., Kalcounis-Rueppell, M.C., 2009. Use of modified water sources by bats in a managed pine landscape. For. Ecol. Manage. 258 (9), 2056–2061, http://dx.doi.org/10.1016/j.foreco.2009.07.058.

Warren, R.D., Waters, D.A., Altringham, J.D., Bullock, D.J., 2000. The distribution of Daubenton's bats (*Myotis daubentonii*) and pipistrelle bats (*Pipistrellus pipistrellus*) (Vespertilionidae) in relation to small-scale variation in riverine habitat. Biol. Conserv. 92 (1), 85–91, http://dx.doi.org/10.1016/S0006-3207(99)00062-2.

Wei, T., 2013. corrplot: Visualization of a correlation matrix. R package version 0.73.

Wickramasinghe, L.P., Harris, S., Jones, G., Vaughan Jennings, N., 2004. Abundance and species richness of nocturnal insects on organic and conventional farms: effects of agricultural intensification on bat foraging. Conserv. Biol. 18 (5), 1283–1292, http://dx.doi.org/10.1111/j.1523-1739.2004.00152.x.