1 **Local versus landscape spatial influency on biodiversity: a case study across five European**

2 **industrialized areas**

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9

10 **Abstract**

11

12 Land use change—mostly habitat loss and fragmentation—has been recognized as one of the major drivers

13 of biodiversity loss worldwide. According to the habitat amount hypothesis, these phenomena are mostly

14 driven by the habitat area effect. As a result, species richness is a function of both the extent of suitable

15 habitats and their availability in the surrounding landscape, irrespective of the dimension and isolation of

16 patches of suitable habitat. In this context, we tested how the extent of natural areas, selected as proxies of

17 suitable habitats for biodiversity, influences species richness in highly anthropogenic landscapes. We defined

18 five circular sampling areas of 5 km radius, including both natural reserves and anthropogenic land-uses,

19 centred in five major industrial sites in France, Italy and Germany. We monitored different biodiversity

20 indicators for both terrestrial and aquatic ecosystems, including breeding birds, diurnal butterflies, grassland

21 vegetation, odonata, amphibians, aquatic plants and benthic diatoms. We studied the response of the different

22 indicators to the extent of natural land uses in the sampling area (local effect) and in the surrounding

23 landscape (landscape effect), identified as a peripheral ring encircling the sampling area. Results showed a

24 positive response of 5 out of 7 biodiversity indicators, with aquatic plants and odonata responding positively

25 to the local effect, while birds, vegetation and diatoms showed a positive response to the landscape effect.

26 Diatoms also showed a significant combined response to both effects. We conclude that surrounding

27 landscapes act as important biodiversity sources, increasing the local biodiversity in highly anthropogenic

28 contexts.

29

30 **Keywords:** habitat amount hypothesis, biodiversity indicators, land use, species richness

Introduction

Land use change has been recognized as one of the major drivers of biodiversity loss worldwide (Sala et al. 2000; MEA 2005), causing species loss and biotic homogenization (Hendrickx et al. 2007; Billeter et al. 2008; McKinney 2008; Johnson et al. 2013; Tudesque et al. 2014; Turrini and Knop 2015; Knop 2016). In particular, the conversion of natural areas into agricultural lands, the intensification of agricultural practices and the increase of urban areas are among the most frequent land use changes (Kleijn et al. 2006; Kleijn et al. 2009; Parris 2016). This process also affects freshwater ecosystems since humans live disproportionately near waterways (Sala et al. 2000), consequently altering water quality because of increased nutrient input and chemicals run-off (Foley et al. 2005).

Anthropogenic landscape alteration negatively influences biodiversity through habitat loss—reduction in the proportion of a landscape composed of suitable habitat for focal species—and habitat fragmentation—changes in the arrangement or configuration of the remaining habitat (Chhabra et al. 2006; Vitousek et al. 2008; Smith et al. 2009; Hooke et al. 2012). However, because habitat loss and fragmentation are highly correlated, it is difficult to disentangle the contribution of each process to biodiversity loss (Smith et al. 2009). This constraint is overcome in the context of the "habitat amount hypothesis", which considers these phenomena to be driven by a single underlying process, the habitat area effect (Fahrig 2013). According to this hypothesis, species richness is a function of both the extent and the availability of suitable habitats in the surrounding landscape, irrespective of the dimension and the isolation of patches of suitable habitat. The effects of land use change on biodiversity can thus be measured by focusing on the amount of suitable habitats. In consequence of that, the preservation of natural land use areas, even in small isolated patches, may be considered a key management aspect for the preservation of biodiversity in human-dominated landscapes.

In the present paper, we aim at the identification of general patterns of species richness across different taxa, both terrestrial and aquatic, in order to investigate the habitat amount hypothesis in human-dominated landscapes. We here considered areas with high 'naturalness', i.e. internal characteristics of low local intensity of human disturbance (Kappes et al. 2011), as proxies of suitable habitats in five anthropogenic landscapes across Europe, characterized by the co-occurrence of natural reserves and industrial complexes. In particular, we tested how the extent of natural land use areas in anthropogenic landscapes influences biodiversity measured in terms of species richness of multiple taxa from both terrestrial and aquatic ecosystems. We considered i) the influence of the extent of natural land use on the local biodiversity within a 5 km radius circular sampling area (*local effects*) and whether ii) the local biodiversity was influenced by the extent of natural land use occurring in the surrounding landscape defined as a ring buffering the sampling area (*landscape effects*).

Materials and methods

Sampling design

This work was developed in collaboration with FCA Group and CNH Industrial, in the framework of the Biodiversity Value Index project (BVI), aiming at evaluating the state of biodiversity in five industrial sites in Europe (Fig. 1). We selected five industrial areas (hereinafter study sites), constituted by the aggregation of several industrial buildings: FPT Powertrain Verrone, Magneti Marelli Venaria and IVECO Suzzara in Italy, FPT Industrial Bourbon-Lancy in France and IVECO Magirus Ulm in Germany (Tab. 1). All industrial complexes were located in the nearby (<5 km) of natural reserves within the same biogeographic area (continental), i.e. areas protected according to the national legislation ─ National Natural Reserves ─ or to the European Natura 2000 Network ─ Sites of Community Importance and Special Areas of Conservation.

For each study site, we defined a circular sampling area of 5 km radius, centred in the industrial complex. We chose to work in an area buffering the main source of disturbance in accordance with the guidelines for the environmental implication assessment provided for Natura 2000 Network sites (European Commission Environment 2002). The surface occupied by industrial complexes was always inferior to 4% of the total area. However, other types of anthropogenic land uses were present, i.e. urban and agricultural.

We obtained land cover data from the Corine Land Cover 2006 project (European Environmental Agency 2006, http://www.eea.europa.eu/publications/COR0-landcover). We used Quantum Gis Desktop (Quantum Gis Development Team 2015, software version 2.10.1) to calculate the percentage of coverage of each land use type inside the sampling areas by taking the following steps: i) drawing of the sampling area of 5 km radius around each industrial complex; ii) overlap of the sampling area with the Corine Land Cover data and intersection; iii) calculation of the percentage of each land use.

We differentiated Corine data in artificial land use (urban and industrial), intensive agriculture, extensive agriculture and natural land use (forested classes, wetlands and water bodies). For each land use category, we extrapolated the areas and summed together to obtain a measure of their extent. We focused on natural land use and we expressed the surface data in percentages. The same land use measure was extrapolated for the surrounding landscape identified as a ring of 2.071 km of semi-radius extending around each sampling area. The 2.071 km semi-radius was chosen in order to obtain a surrounding landscape covering the same surface of the sampling area. It is important to notice that the extent of natural land use does not necessarily correspond to the extent of protected reserves, since anthropogenic land uses may be included in protected areas (Fig. 1).

98   Inside the sampling area (i.e. the 5 km radius circle), we considered both terrestrial and aquatic ecosystems.
99   For terrestrial ecosystems, we focused on open field habitats, while for aquatic ecosystems we considered
100 both lentic and lotic habitats. For each habitat, 10 sampling plots located inside the protected reserves
101 included in the sampling area were randomly selected. We chose seven taxonomic groups that proved to be
102 valuable biodiversity indicators according to literature. These are breeding birds, diurnal butterflies and
103 grassland vegetation for open field habitats (Overmars et al. 2014; Manning et al. 2015; Van Swaay et al.
104 2015), odonata, amphibians and aquatic plants for lentic habitats, i.e. ponds (Oertli et al. 2005; Angélibert et
105 al. 2010; Menetrey et al. 2011), and benthic diatoms for the lotic habitats, i.e. rivers, streams and channels
106 (Falasco & Bona 2011; Falasco et al. 2012).

107

108   Data collection

109

110 All biodiversity indicators were identified at the species level and sampled in accordance with standard
111 protocols as follows.

112 *Breeding birds*

113 For the evaluation of the bird community, point counts were performed in accordance with Bibby et al.
114 (2000). In each sampling plot, the operator listened to songbirds and looked for individuals for 10 minutes
115 within a 100 m^2 area. All individuals surveyed or heard were identified and counted. Surveys started few
116 minutes after dawn and ended before 10 AM. Surveys in rainy or windy days were avoided. Bird surveys
117 were conducted during late spring and repeated in early summer, in order to assure that only breeding birds
118 were recorded (Tab. 2).

119 *Diurnal butterflies*

120 We sampled diurnal butterflies along linear transects with a semi-quantitative method: a straight 100 m path
121 was covered at a constant speed, while counting butterflies in an area of 5 m in height and 2.5 m to the right
122 and to the left of the operator (Pollard and Yates, 1993; van Swaay et al. 2012). Surveys were performed
123 during the warmest hours of the day (late morning - early afternoon), when the butterflies are most active,
124 avoiding the collection of data on days with bad weather (strong wind or heavy rain). Surveys were repeated
125 at least three times over the warm season (Tab. 2). Individuals were captured and subsequently released after
126 their identification by means of field characters. When a butterfly could not be identified in the field,
127 specimens were collected and subsequently identified in the laboratory.

128 *Grassland vegetation*

Grassland vegetation was investigated with the method of Braun-Blanquet (1964). For each sampling plot, we defined a 50x50 m square in a homogeneous area, avoiding ecotones in order to have standardized surveys in all sites. The presence of all species inside the square was recorded in order to get a comprehensive list. Surveys were repeated at least three times over the vegetative season (Tab. 2). Species were identified according to Tutin et al. (2001) and Pignatti (1982).

Odonata

Odonata were sampled by visual census, in accordance with Bouwman et al. (2009). The presence of adult specimen was detected along transects on the perimeter of the ponds. Zygopteran and *Sympetrum* species were counted respectively within 2 m from the shore and 3 m from the water; the other species were considered within 5 m from the water. Surveys lasted half an hour and were performed during the warmest hours of the day (late morning - early afternoon) when Odonata are most active, avoiding the collection of data on days with bad weather (strong wind or heavy rain). Flying individuals were identified *in situ*. In each plot, two surveys were performed — a spring session and a summer session (Tab. 2) — in accordance with Angélibert et al. (2010).

Amphibians

The field protocol followed the method by Schmidt (2005). Surveys leasted one hour each and were repeated at least twice over the reproductive season (Tab. 2), under standardized weather conditions, i.e. mild temperatures, with no wind or rain. Surveys after long periods of drought were avoided. Amphibians — adults, subadults, larvae — were surveyed by means of (i) visual census, (ii) identication of calls, and (iii) dip netting. The two species *Rana esculenta* and *Rana lessonae* were considered as one single taxon (green frog complex).

Aquatic plants

We sampled aquatic plants according to the European standard protocol UNI EN 15460:2007. For each sampling plot we defined a sampling transect on the shore along which we compiled an exhaustive list. Surveys were repeated twice during the vegetative season (Tab. 2).

Diatoms

We sampled benthic diatoms following the standard procedure (European Committee for Standardization, 2003; UNI EN 14407:2004) and we performed one sampling session in spring (Tab. 2). Diatom identification was based on several diatom floras and monographies, as well as on recent taxonomic papers (Krammer and Lange-Bertalot 1986-1991a, b; Krammer 1997a, b; Reichardt 1999; Lange-Bertalot 2001; Krammer 2002, 2003; Blanco et al. 2010; Hofmann et al. 2011; Bey and Ector 2013; Falasco and Bona, 2013; Falasco et al. 2013; Ector et al. 2015).

162 Statistical analysis

164 We firstly explored species richness data in accordance with Zuur et al. (2009, 2010). We used Cleveland
165 dotplots and boxplots to assess the presence of extreme values and avoid unusual observations to exert an
166 undue influence on estimated parameters. We evaluated multicollinearity among predictors, namely
167 percentage of surface covered by natural land use in the sampling areas and surrounding landscapes, using
168 Pearson correlation test and variance inflation factors (VIFs) (Zuur et al. 2009). Given their low correlation
169 (r = 0.10, p = 0.06) we include all predictors in the same model.

170 The contribution of the local and landscape effects to biodiversity was tested by means of generalized linear
171 mixed models (GLMMs). Percentage of natural land use in the sampling areas (*local effect*), in the
172 surrounding landscapes (*landscape effect*) and their interaction were used as fixed factors, which were
173 standardized in order to achieve homogenization of their distribution. Given the spatial dependence of the
174 data — 10 sampling plots for each sampling site —, we applied the mixed procedure to include the grouping
175 variable "Site" as a random factor in order to account for the variation it introduced in our samples, rather
176 than to test for its direct effect on the dependent variables. Models were fitted with a Poisson error
177 distribution (link function: log) which is able to deal with count data as recommended in Zuur et al. (2009).
178 Models were tested for over-dispersion and were validated by constructing standard validation plots using
179 the model residuals (Zuur et al. 2009). Statistical models were performed with the package *lme4* (Bates et al.
180 2014) in R environment (R Core Team 2015).

182 **Results**

184 During the surveys, an amount of 190 sampling plots was visited and 340 biological samples were collected.
185 Altogether, we identified 928 species (see ESM_1 for the list of all recorded species). The five study sites
186 showed different values in terms of land use coverage (Fig. 2) as well as of species richness for each
187 biodiversity indicator (Tab. 3). Considering land use, Suzzara (Italy) showed the highest coverage of
188 intensive agriculture, while Ulm (Germany) presented the highest level of industrialization and urbanization.
189 Bourbon-Lancy (France) and Verrone (Italy) showed the highest coverage of extensive agriculture and
190 natural land use respectively.

191 The response to the extent of natural land use in the sampling areas (*local effect*) and in the surrounding
192 landscape (*landscape effect*) differed consistently among biodiversity indicators (Tab. 4).

193   Diurnal butterflies and amphibians did not show any significant response. Species richness of odonata and
194   aquatic plants was positively influenced by the local effect, while grassland vegetation and breeding birds
195   showed a positive response to the landscape effect (Figs. 3 and 4).

196   Diatoms showed a more complex combined response since they were significantly influenced by the
197   landscape effect but also by the interaction of local and landscape effects. In particular, when setting the
198   extent of natural land use in the sampling area at low values, the response to the landscape effect was
199   positive. On the other hand, this response was negative when the extent of natural land use in the sampling
200   area reached high values (Fig. 5).

201

202   **Discussion and conclusions**

203

204   In this work, we showed how biodiversity indicators responded to the extent of natural land use locally and
205   at the landscape level. Aquatic and terrestrial ecosystems were simultaneously analysed at similar spatial
206   scales with a standardized statistical approach in order to shed light on similarities and differences in the
207   response to the land use (Siqueira et al. 2015). In particular, we highlighted a common trend across the
208   different taxonomic groups, since natural land use affected positively species richness in five out of seven
209   biodiversity indicators, both at the local and the landscape level.

210   When focusing on *local effects* (i.e. on the response of biodiversity to the extent of natural areas occurring in
211   a 5 km radius circle areas), we detected a positive influence for aquatic plants and odonata. These results
212   may suggest the positive role played by natural land use at the local scale for maintaining species diversity in
213   human-altered landscapes. Such positive response of aquatic plants is in accordance with Bolpagni and Piotti
214   (2015), who detected high species diversity of aquatic plants in natural lentic habitats. The similar positive
215   response of odonata species richness possibly indicates an indirect relationship with aquatic plants. Indeed,
216   odonata are influenced by the structure of the shoreline vegetation (Buchwald, 1992), which is necessarily
217   more complex and species-rich where ponds are surrounded by natural land use, as suggested by Declerck et
218   al. (2006). More generally, aquatic vegetation is crucial for many aspects of the ecology of the odonata,
219   including habitat heterogeneity required by the larval stages (e.g., protection from predators), emergence
220   supports during metamorphosis, as well as important substrates for oviposition and perching for adult
221   odonata (Corbet and Brooks 2008; Honkanen et al. 2011).

222   When considering *landscape effects* (i.e the response of biodiversity to the extent of natural areas
223   surrounding the 5 km radius circle area), we detected a positive response of the local assemblages of
224   breeding birds and grassland vegetation. These results suggest how surrounding natural areas represent
225   important key factors for preserving biodiversity, especially of terrestrial organisms. These results are in

accordance with literature, where a positive effect of surrounding natural land use has been reported for both birds and grassland vegetation (Wright & Wimberly 2013; Winsa et al. 2015). This positive effect might be due to the possible increase of source of colonists and connectivity. Furthermore, a negative relationship between isolation and bird diversity has been reported, especially for agricultural landscapes (Bailey et al. 2010). This might also have indirect repercussions on vegetation since higher landscape connectivity could guarantee a higher bird-mediated seed dispersal (Herrmann et al. 2016).

A response to the *landscape effect* was also observed for diatoms, which also showed a significant response to the interaction between the local and the landscape effect, i.e. landscape effect became significantly major when the extent of natural land use in the sampling area was low. Given that diatom communities are strongly shaped by water quality (van Dam et al. 1994; Rott et al. 1999; Delgado et al. 2012), we interpreted this result as an indirect top-down cascade effect, which relates land cover to diatoms through the indirect influence of water quality (Tudesque et al. 2014). Indeed, anthropogenic land uses in the surrounding environment may cause nutrients increase in waterbodies, consequently favouring tolerant species and possibly increasing diatom diversity (Blanco et al. 2012). This may explain the negative effect of this interaction, since high naturalness leads towards oligotrophic aquatic environments, which could result in low species richness of diatoms.

A second reason could be that riverine biodiversity indicators integrate the response of the entire upstream area (Tudesque et al. 2014). For these reasons, despite diatoms are widely recognised as effective indicators for measuring water quality (Álvarez-Blanco et al. 2012; Delgado et al. 2012), according to our results diatom species richness proved not to be a reliable metric for detecting the effect of land use on biodiversity in anthropogenic landscapes, in accordance with Blanco et al. (2012).

Surprisingly, diurnal butterflies and amphibians did not show any significant effect to the extent of natural land use both in the sampling areas and in the surrounding landscapes. Concerning amphibians, similar results were obtained in Menetrey et al. (2011), who excluded species richness of amphibians from a multimetric index aimed at the evaluation of pond integrity, since this parameter did not discriminate between different environmental conditions. For diurnal butterflies, Collinge et al. (2003) revealed little influence of landscape composition on butterfly communities. A further issue might be that in our sample amphibians and diurnal butterflies showed the lowest variation in species richness among plots. Consequently, the detected pattern that amphibian and diurnal butterfly species richness are not affected by natural land use must be considered cautiously.

In conclusion, our results are in agreement with the habitat amount hypothesis, which also apply to industrialized and highly anthropogenic contexts.

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475

476 **Figure captions**

477 **Fig. 1** Map representing the location of the five industrial sites and a detailed view of their land use in the
478 sampling area (internal circle, continuous line) and the surrounding landscape (external circle, dashed line). a
479 = Bourbon-Lancy; b = Venaria; c = Verrone; d = Suzzara; e = Ulm

480 **Fig. 2** Percentage of land use coverage calculated for the sampling areas (a) and surrounding landscapes (b).
481 Artificial = percentage of urban and industrial land uses; Intensive = percentage of intensive agriculture;
482 Extensive = percentage of extensive agriculture; Natural = percentage of natural land use

483 **Fig. 3** Predicted values (blue continuous line) and confidence intervals (95%, light grey area) for (a) aquatic
484 plants and (b) odonata against the extent of natural land use in the sampling area (*local effect*)

485 **Fig. 4** Predicted values (blue continuous line) and confidence intervals (95%, light grey area) for (a)
486 breeding birds, (b) grassland vegetation and (c) diatoms against the extent of natural land use in the
487 surrounding landscape (*landscape effect*)

488 **Fig. 5** Predicted species richness of diatoms and the interaction between the extent of natural land use in the
489 sampling areas and surrounding landscapes. Lines represent the *landscape effect* at low (0%, continuous
490 line), intermediate (15%, dashed line) or high (30%, dotted line) cover of natural land use in the sampling
491 area. Major landscape effects are seen at low extent of natural land use in the sampling area, conversely they
492 become negligible at higher extents of natural land use in the sampling area