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1 A diagnosis-based approach to assess specific risks of river degradation in a multiple pressure  
2 context: insights from fish communities

3

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1 ABSTRACT

2 In the context of increasing pressure on water bodies, many fish-based indices have been  
3 developed to evaluate the ecological status of rivers. However, most of these indices suffer  
4 from several limitations, which hamper the capacity of water managers to select the most  
5 appropriate measures of restoration. Those limitations include: (i) being dependent on  
6 reference conditions, (ii) not satisfactorily handling complex and non-linear biological  
7 responses to pressure gradients, and (iii) being unable to identify specific risks of stream  
8 degradation in a multi-pressure context. To tackle those issues, we developed a diagnosis-  
9 based approach using Random Forest models to predict the impairment probabilities of river  
10 fish communities by 28 pressure categories (chemical, hydromorphological and biological). In  
11 addition, the database includes the abundances of 72 fish species collected from 1527 sites in  
12 France, sampled between 2005 and 2015; and fish taxonomic and biological information.  
13 Twenty random forest models provided at least good performances when evaluating  
14 impairment probabilities of fish communities by those pressures. The best performing models  
15 indicated that fish communities were impacted, on average, by  $7.34 \pm 0.03$  abiotic pressure  
16 categories (mean  $\pm$  SE), and that hydromorphological alterations ( $5.27 \pm 0.02$ ) were more  
17 often detected than chemical ones ( $2.06 \pm 0.02$ ). These models showed that alterations in  
18 longitudinal continuity, and contaminations by Polycyclic Aromatic Hydrocarbons were  
19 respectively the most frequent hydromorphological and chemical pressure categories in  
20 French rivers. This approach has also efficiently detected the functional impact of invasive  
21 alien species. Identifying and ranking the impacts of multiple anthropogenic pressures that  
22 trigger functional shifts in river biological communities is essential for managers to prioritize  
23 actions and to implement appropriate restoration programmes. Actually implemented in an R  
24 package, this approach has the capacity to detect a variety of impairments, resulting in an  
25 efficient assessment of ecological risks across various spatial and temporal scales.

- 26 Keywords: ecological risk assessment, functional trait, hydromorphology, machine learning,  
27 invasive alien species, water chemistry

## 28 INTRODUCTION

29 By the second half of the twentieth century, the international community substantiated water-  
30 related issues, and started to evaluate the ecological status of surface and ground waters  
31 (Cooley et al. 2013). The assessment of this ecological status has been based on the  
32 monitoring of biological quality elements (BQEs; i.e., fishes, benthic invertebrates,  
33 phytobenthos, macrophytes, phytoplankton) and of supporting environmental conditions (e.g.  
34 physico-chemical and hydromorphological parameters). Reaching the “good” ecological  
35 status for all the water bodies has then been the overarching goal of many environmental  
36 legislations and water policies worldwide, including the Water Framework Directive in  
37 Europe (WFD; EC, 2000). This objective has required to (I) evaluate the ecological status of  
38 water bodies, (II) identify the potential causes of deviation from the “good” status by  
39 considering multiple anthropogenic pressure categories and (III) implement appropriate  
40 monitoring, management and restoration programmes. This challenging goal has stimulated a  
41 flourishing scientific literature, which aims at designing biotic indices capable of translating  
42 river conditions into simple measures summarizing the complexity of ecosystems. For  
43 instance, Birk et al. (2012) provided an overview of about three hundred methods based on  
44 the main BQEs to implement the WFD. Yet, a recent report from the European Environment  
45 Agency indicated that only 40% of surface waters were at least in “good” ecological status  
46 (EEA, 2018) suggesting that if step I is fulfilled for most of the EU members, steps II and III  
47 require further collective efforts.

48

49 In the context of stream BQE-based evaluation, fish communities have received particular  
50 attention, probably because they offer several advantages. They are widely distributed in lotic  
51 ecosystems. They are relatively easy to identify at the species level and a substantial but  
52 scattered amount of information is available on their functional and life history characteristics.

53 Their potential number of species is limited in Europe (< 250 species). They exhibit longer  
54 lifespan compared to other BQEs, integrating environmental changes over longer periods.  
55 Last, they are sensitive to biological (Gallardo et al., 2016; Sagouis et al., 2015), physico-  
56 chemical and hydromorphological pressures (Azimi and Rocher, 2016; Schinegger et al.,  
57 2012). In particular, weirs and dams are severely impairing fish assemblages (Branco et al.,  
58 2014; Mims and Olden, 2013), diadromous species (Drouineau et al., 2018; Fuller et al.,  
59 2015; Lasne et al., 2015), native fish species (Meador and Carlisle, 2012), and their  
60 environment via habitat modifications and disruption in the ecological continuity of rivers, at  
61 both local and large spatial scales (Ali et al., 2019; Kuriqi et al., 2019). To move efficiently  
62 beyond the mere assessment of ecological status (step I), fish-based indices present, however,  
63 at least one of three limitations that need to be overcome.

64 1) Fish-based indices must rely upon the debated notion of reference conditions, which are  
65 often difficult to evaluate and an unrealistic target for the management of heavily modified  
66 water bodies (Dufour and Piégay, 2009; Tweedley et al., 2017).

67 2) Despite growing evidence that fish communities may exhibit non-linear responses to  
68 pressure gradients (Taylor et al., 2014), most fish-based indices assume linear relationships  
69 (but see Bhagat et al., 2007).

70 3) Fish-based indices are unable to identify specific risks of stream degradation in a multi-  
71 pressure context and to disentangle the effects of co-occurring pressures acting across various  
72 spatial and temporal scales (Reyjol et al., 2014). This tremendously limits the capacity of  
73 water managers to opt for the most relevant restoration measures, for a given water body.

74

75 To overcome these limitations, we here developed a robust and flexible diagnosis-based  
76 approach, able to identify individual pressures involved in fish community impairment under  
77 multiple pressure scenarios. This approach has been grounded upon the fundamental notion

78 that functional species traits - the physiological, morphological and behavioural  
79 characteristics of organisms - integrate the spatial and temporal variability of the environment  
80 (Southwood, 1977; Townsend and Hildrew, 1994). Various natural environmental filters,  
81 acting at different nested scales from ecoregions to reaches, filter functional traits.  
82 Anthropogenic pressures are then able to modify this environmental filtering, leading to  
83 different combinations of traits that are often specific to a given pressure category (Desrosiers  
84 et al., 2019). Hence, according to our hypothesis, substantial modifications in the relative  
85 frequencies of a large set of functional traits (i.e. biological metrics) within communities  
86 should specifically reflect biological responses to one or more environmental pressures.  
87  
88 Random forest (RF) models have been successfully applied in many environmental and  
89 ecological studies including - but not limited to - air quality modeling (Choubin et al., 2020),  
90 flood susceptibility assessment (Hosseini et al., 2020), groundwater nitrate modeling  
91 (Rahmati et al., 2019), earth fissure modeling (Choubin et al., 2019), risk assessment for  
92 invasive species (Keller et al., 2011; Philibert et al., 2011), and stream impairment modeling  
93 under multi-pressure scenario (Larras et al., 2017; Mondy and Usseglio-Polatera, 2013). The  
94 advantage of RF models, over other uni- and multi-variate methods, is their ability to handle  
95 non-linear relationships between predictors and response variables, to integrate complex  
96 interactions among a large number of predictors and to generate good predictions with their  
97 associated probabilities (Prasad et al., 2006; Rahmati et al., 2019). The proposed diagnosis-  
98 based approach is a generalization of the use of RF models to estimate probabilities of  
99 community impairments, in a multi-pressure context, using complex combinations of  
100 taxonomic and biological metrics. We tested this flexible approach with river fish  
101 communities by: 1) compiling fish functional traits *sensu stricto* and any relevant information  
102 on how fish assemblages may interact with their environment (e.g. functional guilds, life

103 history strategies, taxonomic structure), 2) identifying and describing abiotic and biotic  
104 pressure categories to which fishes are potentially sensitive at various spatial and temporal  
105 scales, and 3) using RF models to calculate probabilities of community impairment by any  
106 given pressure category. Based on fish communities, this study aims at demonstrating the  
107 efficiency of the flexible diagnosis-based approach for enhancing our understanding of shifts  
108 in ecosystem structure and functions triggered by given levels of various anthropogenic  
109 pressures (step II), and for facilitating decision-making processes by environmental managers  
110 (step III).

111

## 112 MATERIAL & METHODS

### 113 *Fish and environmental data*

114 This approach is based on three large datasets gathering information on i) the spatial and  
115 temporal distribution of French stream fish communities (“Fish record dataset”), ii) the  
116 taxonomic, biological, and ecological characteristics of fish species (“Fish characteristic  
117 dataset”), and iii) the levels of chemical, hydromorphological, and biological pressures  
118 impairing sites at each sampling event (“Pressure dataset”).

119

120 The Fish record dataset contains electrofishing samples performed from 2005 to 2015 in 1527  
121 sites belonging to the French Water Framework Directive’s surveillance monitoring, covering  
122 the mainland France (c.a. 550 000 km<sup>2</sup>). The standardized electrofishing protocol followed the  
123 recommendations of the European Committee for Standardisation’s standard (CEN, 2003;  
124 Marzin et al., 2014). This protocol was conducted during low-flow periods (from May to  
125 October) with a sampling design that depended on river width and depth. Fishes were  
126 identified to the species level, counted and then released back into the river. On average, the  
127 sites were sampled  $5.5 \pm 0.1$  times (Mean  $\pm$  SE; min = 1; max = 11) over the 2005-2015



128 period. The Fish record dataset was thus composed of the abundances of 72 fish species  
129 distributed in 8529 sampling events (site x year; Table A).  
130  
131 The Fish characteristic dataset gathered information about 56 fish characteristics (260  
132 categories) compiled from different sources of information (Table B). Hereafter, we  
133 considered ‘fish characteristics’ as any descriptive information related to the physiology,  
134 morphology and behaviour (i.e. functional traits *sensu stricto*; N = 53), life history strategy  
135 (lifespan; N = 1), and taxonomy (family and order levels; N = 2) of fish. Fifty-five fish  
136 characteristics were coded using a full disjunctive system (i.e. presence/absence of various  
137 categories). One fish characteristic was structured using a fuzzy-coding technique (‘food’;  
138 Table B). The number of categories defined per characteristic was rather low (between 2 and  
139 10), except for the characteristics related to taxonomic classification (N = 20  
140 families/categories; N = 13 orders/categories).  
141  
142 The Fish record and Fish characteristic datasets were then used to calculate 1576 metrics for  
143 each of 8529 fish sampling events. These metrics covered: (i) the relative frequency of the  
144 log-transformed abundances of organisms using each of the 260 categories belonging to the  
145 56 fish characteristics (260 metrics), (ii) taxonomy-based diversity indices calculated at the  
146 scale of each trait category (e.g. the proportion of piscivorous taxa, the Shannon diversity of  
147 rheophiles; 1255 metrics), (iii) Rao functional diversity indices calculated at the scale of each  
148 characteristic (Rao, 1982; 56 metrics), (iv) the level of diet specialization (Mondy and  
149 Usseglio-Polatera, 2014; one metric) and (v) taxonomy-based richness or diversity indices  
150 (e.g. the species richness, the Shannon diversity index; four metrics; Table B).  
151

152 The Pressure dataset was obtained by defining 28 pressure categories that describe the  
153 impairment of water quality (WQ: chemistry; 13 categories; 188 individual parameters taken  
154 into account; Table C), the impairment of habitat (HD: hydromorphology; 14 categories; 29  
155 individual parameters; Table D), and the contamination of local fish assemblages by invasive  
156 alien species (IAS: 1 category; 1 individual parameter; Table A), at each site for each  
157 sampling date. These pressure categories were selected and defined according to available  
158 information and their potential relevance for fish communities. Each pressure category was  
159 defined by one to several individual parameters, each of which was described by two pressure  
160 levels (“low” vs. “significant”). Then, the pressure level allocated to a given site for a given  
161 pressure category was the worst pressure level over all the individual parameters taken into  
162 account for characterizing this pressure category (i.e., following the “one out, all out”  
163 aggregation rule; Gottardo et al., 2011).

164

165 For parameters related to WQ pressure categories, pressure levels were assessed by  
166 comparing the mean value of individual parameters over a given period with the  
167 “good/moderate” quality class boundary of the French water quality assessment system (i.e.  
168 SEQ-Eau V2.0; Oudin and Maupas, 2003), allowing to distinguish “low” (high/good) from  
169 “significant” (moderate/poor/bad) pressure levels. Four of the 14 HD pressure categories were  
170 defined according to the literature (i.e. ‘Transportation facilities’, ‘Urbanization’,  
171 ‘Hydrological instability’, ‘Straightening’; Larras et al., 2017; Mondy et al., 2012; Mondy and  
172 Usseglio-Polatera, 2013; Villeneuve et al., 2015), whereas the remaining categories and  
173 associated parameters were newly described in this study after preliminary tests to define the  
174 low-significant boundaries of pressure levels (Table D). For the IAS pressure category, we  
175 have considered as “significant” the effect of the presence of at least one invasive alien  
176 species in the fish assemblage. The list of invasive alien species, including 17 species, was

177 established by combining the information from three data sources: the French National  
178 Museum of Natural History, the IUCN Red list of threatened species in France (those  
179 considered as “alien species”) and Fishbase (those considered as “Potential pest”) (more  
180 details in Table A). We used the Cohen’s kappa coefficient to assess non-random co-  
181 occurrences of pressure categories; i.e., after removing random co-occurrences (“low” vs.  
182 “significant”; *psych*-package in R).

183

#### 184 *Random forest models*

185 Random forests (RF) are a particular type of machine learning methods able to investigate  
186 complex and potentially non-linear relationships (Breiman, 2001). Although several other  
187 types of machine learning methods exist (k-nearest neighbour, support vector machine), RF  
188 models were selected here because they can handle a large number of predictive variables.  
189 The importance of the contribution of those predictors to model outputs (probabilities) can be  
190 easily quantified. Moreover, RF models provide good predictive performances (Rahmati et  
191 al., 2019). One RF model was built per pressure category taking the pressure level (“low” vs.  
192 “significant”) as the response variable and the fish metrics as the predictive variables. We  
193 tested five periods (from one to five years before fish sampling) to identify the most likely  
194 integration time of fish communities regarding WQ for two reasons: fish have potentially long  
195 life cycles (from one year to several decades) and the water samples (taken for chemical  
196 analyses) are punctual (i.e., several times a year) and were not necessarily obtained the same  
197 day as fish samples. Since all the HD pressures were defined once for the period covered by  
198 fish sampling, we did not test several integration periods as we did for WQ pressures. The  
199 period yielding the best model performance (see below) was selected independently for each  
200 WQ pressure. Among all the pressures, the one induced by the invasive alien species is  
201 particular because it is the only biotic pressure where a subset of the fish assemblage (IAS)

202 may impact the other subset (native species). To this end, the associated RF model was built  
203 differently. All the fish metrics were calculated again without the invasive alien species  
204 (predictive variables) while the presence (= “significant pressure”) versus absence (= “low  
205 pressure”) of invasive alien species was considered as the response variable. Although the  
206 *ecodiag*-package presented in this study offers ways to tune RF models and can easily be used  
207 in a cross-validation procedure, we have only presented the results of models parametrized  
208 following published strategies (e.g., 1500 classification trees, random selection of 50 fish  
209 metrics at each node in individual trees; (Larras et al., 2017). Based on the fish metric values,  
210 each pressure-specific model therefore provided, for each sampling event (n = 8529), a mean  
211 impairment probability calculated over all the trees defining a given forest. In order to identify  
212 pressure syndromes (i.e. combinations of pressures often impairing fish communities) over  
213 the entire studied area (mainland France), we also performed a Principal Component Analysis  
214 (PCA) on the impairment probabilities predicted by the abiotic and biotic models exhibiting at  
215 least “good” performances, considering all the 8529 sampling events.

216

### 217 *Model performances*

218 To assess model performances, each RF model was built on a learning dataset and then tested  
219 against a test dataset (Larras et al., 2017; Mondy and Usseglio-Polatera, 2013). Learning and  
220 test datasets corresponded to independent subsamples of the input database, i.e., the metric  
221 and pressure data measured for all the sampling events. To take into account the temporal  
222 correlations among sampling events, the sites were randomly allocated either to the learning  
223 or to the test dataset (75% and 25% of the sites, respectively). Model performance was then  
224 evaluated with the Area Under the Curve (AUC). The AUC values indicate “good” model  
225 performances if they are equal to or higher than 0.70 (i.e., maximizing both sensitivity and  
226 specificity; Pearce and Ferrier, 2000). We used this AUC criterion to assess model

227 performances using mainly the test ( $AUC_{\text{test}}$ ) datasets, and to a lower extent the learning  
228 ( $AUC_{\text{learn}}$ ) one. Evaluating both  $AUC_{\text{test}}$  and  $AUC_{\text{learn}}$  represent a trade-off to minimize  
229 prediction errors in test datasets and to avoid over-fitting in learning datasets. We then  
230 investigated spatial and temporal changes in RF model outputs (i.e., impairment probability  
231 by any given pressure) using models with the best performances.

232

### 233 *Metric importance and ecological validation*

234 Our approach is based upon the assumption that community responses to environmental  
235 pressures manifest through changes in the relative utilization frequency of functional traits  
236 (trait-based metrics). Trait-based metrics may not contribute equally, however, to this  
237 community response. To our knowledge, there are currently eight methods that can evaluate  
238 the importance of each fish-based metric in the community response to each pressure category  
239 (i.e., RF model output): “anova.test”, “auc”, “chi.squared”, “gain.ratio”, “information.gain”,  
240 “kruskal.test”, “ranger.impurity”, and “ranger.permutation”. Those methods rank metrics  
241 from the most to the least important metric in each RF model. Since those methods may  
242 provide different ranking values for a given metric, the ranks provided by each method were  
243 first scaled in the range [0,1], and then averaged across all the methods (Congalton, 1991).  
244 This mean scaled rank was considered as the final measure of metric importance. The  
245 ecological validation of our approach was then performed by investigating the relationship  
246 between the fish metric values and the impairment probabilities generated by each RF model.  
247 This relationship was illustrated for the most important metrics (based on their mean scaled  
248 ranks) and the most accurate abiotic and biotic models ( $AUC > 0.7$ ). The strength of those  
249 relationships was assessed using non-parametric Spearman correlations. The diagnosis-based  
250 approach was implemented in the *ecodiag*-package in R (R Core Team, 2018) and is currently  
251 hosted on GitHub (Table E).

252

## 253 RESULTS

### 254 *Model performances*

255 The integration period over which the WQ pressures were assessed had a pressure-dependent  
256 effect on the performances of the corresponding RF models and varied from three to five  
257 years (Table 1; Table F). For example, the best model performances were obtained using an  
258 integration period of three years for “Nitrogen compounds” whereas they were obtained using  
259 an integration period of five years for “Phosphorous compounds”. Due to an insufficient  
260 amount of data corresponding to significantly impaired conditions regarding “Acidification”,  
261 this WQ pressure was discarded from further analyses.

262

263 All the HD, WQ and IAS models had  $AUC_{\text{learn}}$  values higher than 0.75, except for “Mineral  
264 micropollutants” ( $AUC_{\text{learn}} = 0.67$ ; Table 1). In contrast, some models exhibited “poor” (four  
265 models) or only “fair” (three models) performances with the test dataset. Most of the models,  
266 however, displayed good performances with both the learning and test datasets ( $AUC > 0.70$ ;  
267 HD: 10/14; WQ: 9/12; IAS: 1/1), resulting in clearly distinct distributions of impairment  
268 probabilities across pressure levels (Fig. A). For the 19 abiotic models, we found no  
269 significant differences in  $AUC_{\text{test}}$  between the ten HD and the nine WQ pressure categories  
270 (Wilcoxon rank sum test:  $W = 55.5$ ,  $p\text{-value} = 0.41$ ; similar results were obtained with  
271  $AUC_{\text{learn}}$ :  $W = 29.5$ ,  $p = 0.22$ ).

272

### 273 *Impairment probabilities across spatial scales*

274 The proposed diagnosis-based approach applied to fish communities allowed us to investigate  
275 pressure patterns at different spatial scales. At a large spatial scale (mainland France), the RF  
276 models identified the hydromorphological pressures as the most frequently significant

277 pressures (from 26 to 81% of the sampling events) detected at any given site compared to  
278 water quality pressures (mainly from 8 to 22%; except for “Nitrates” [48%] and “PAH”  
279 [54%]) or invasive alien species pressure (43% of the sampling events; Fig. 1A). Overall, RF  
280 models predicted that a given sampling event was significantly impacted, on average, by  $5.27$   
281  $\pm 0.02$  HD (min. = 0, max. = 10) and  $2.06 \pm 0.02$  WQ (min. = 0, max. = 9) pressure types  
282 (impairment probabilities  $> 0.50$ ). The first two PCA axes explained 50% of the total variance  
283 in impairment probabilities (Fig. 1B). The first axis discriminated two hydromorphological  
284 pressures related to the river continuity (HD12 = ‘Longitudinal continuity’; HD14 =  
285 ‘Proximal continuity’; negatively correlated to Axis 1) from all the other pressure categories  
286 (more or less positively correlated to Axis 1). The second axis separated the two broad types  
287 of pressure categories with most of the HD and WQ pressure impairment probabilities being,  
288 respectively, positively (except HD07) and negatively (except WQ9 and WQ11) correlated to  
289 this axis (Fig. 1B). The IAS pressure was positively – but lowly - correlated to the first PCA  
290 axis. Note that overall the non-random co-occurrence of pressure categories (RF models’  
291 inputs) was very low (mean Cohen’s Kappa coefficient =  $0.07 \pm 0.01$ ) except for 11 of the  
292 190 pairs of pressures (Table G).

293

294 The proposed diagnosis-based approach also highlighted broad spatial patterns in stream  
295 degradation. For each site and each of four pressure categories, impairment probabilities were  
296 averaged across sampling events (over the 2005-2015 period) to remove the temporal effect in  
297 the data, and then plotted on a map displaying the main French drainage basins (Fig. 2). We  
298 provided this spatial distribution of impairment probabilities for the two HD and WQ models  
299 exhibiting the highest performances. The regional patterns of impairment probabilities were  
300 different among pressure categories. The risk of fish community impairment by ‘Longitudinal  
301 continuity’ alteration was lower (mean impairment probability  $< 0.50$ ) for rivers located near

302 the Atlantic and Mediterranean coasts, than those located further inland (Fig. 2A). The effects  
303 of modifications in 'Stream width', a proxy of stream channeling and bank modifications, on  
304 fish communities, were the most important in sites located on the main large rivers (the Loire,  
305 the Rhone, the Seine, and the Adour-Garonne rivers; Fig. 2B) and their lowland tributaries. In  
306 addition, 'Nitrates' and 'Nitrogen compounds' (other than nitrates) exhibited contrasting  
307 impairment patterns: the former had a pronounced, large-scale impact in the northwestern part  
308 of France, and to a lower extent, in the Adour-Garonne basin (South-west; Fig. 2C) whereas  
309 the latter had rather local effects on communities and mainly in areas exhibiting the most  
310 intensive agricultural activity (Fig. 2D). The impact of invasive alien species on native fish  
311 assemblages, was also the most important in sites located on main large rivers (the Loire, the  
312 Rhone, the Adour-Garonne, and to a lower extent, the Seine rivers) and their lowland  
313 tributaries (Fig. B). Finally, we assessed the global impairment probability of the whole fish  
314 communities (including native and invasive alien species) over the decade, by averaging the  
315 impairment probabilities of the 19 HD and WQ models with at least good performances  
316 ( $AUC_{test} > 0.70$ ; Table 1). Least impacted sites were mainly located within or near the main  
317 mountain ranges (e.g., Pyrenees, Alps, Massif Central mountains, and to a lower extent the  
318 Vosges, and Jura Mountains; Fig. 2E).

319

#### 320 *Impairment probabilities across temporal scales*

321 This approach has also allowed assessing broad temporal patterns in stream degradation at  
322 large spatial scale. Figure 3 depicts changes in the distribution of impairment probabilities at  
323 all the sites sampled in both 2005 and 2015 ( $n = 543$ ). These trends showed pressure-specific  
324 patterns. There was only a significant decreasing risk of community impairment due to  
325 'Nitrogen compounds' and to 'Phosphorous compounds' in 2015 compared to 2005  
326 (Wilcoxon sign-rank test for paired data =  $9.5 \cdot 10^4$  and  $1.0 \cdot 10^5$ , both p-values  $< 0.0001$ ,



327 respectively; Fig. 3D and F). Differences in the total impairment probabilities (i.e. mean  
328 impairment probabilities of the 19 HD and WQ models with  $AUC_{\text{test}} > 0.70$ ; Table 1) between  
329 2005 and 2015, tended to decrease, indicating a weak improvement in overall site condition  
330 over the decade (Wilcoxon sign-rank test for paired data =  $1.0 \cdot 10^5$ , p-value < 0.0001; Fig.  
331 3G). There was also a significant decrease in the impairment probabilities induced by the  
332 invasive alien species (Wilcoxon sign-rank test for paired data =  $8.7 \cdot 10^4$ , p-value = 0.0004;  
333 Fig. 3H).

334

335 Finally, the diagnosis-based approach allowed detecting temporal patterns in ecological  
336 recovery or ecological degradation at a local spatial scale (i.e. site level). Two sites, A and B  
337 (located on the Fig. 2F), were sampled between 2005 and 2015, and characterized using  
338 outputs of the three most efficient WQ ('Nitrates', 'Nitrogen compounds', 'Phosphorous  
339 compounds') and HD ('Longitudinal continuity', 'Stream width', 'Transportation facilities')  
340 models. Site A displayed low impairment probabilities for all the six pressure types over the  
341 whole decade (impairment probabilities far lower than 0.50; Fig. 4A) with no clear variation  
342 in temporal pattern. In contrast, site B exhibited rather high impairment probabilities  
343 (impairment probabilities  $\geq 0.50$ ) for all the six pressure categories in 2005. However, for this  
344 site, the model results seem to indicate an improvement over the decade regarding the three  
345 nutrient-related pressures: nitrates and, during the more recent years, phosphorous and other  
346 nitrogen compounds (Fig. 4B).

347

#### 348 *Ecological validation*

349 The relationships between impairment probabilities and observed metric values were explored  
350 for the five most contributive metrics to each model (among 1576 tested metrics) and each of  
351 the 19 most accurate RF models ( $AUC_{\text{test}} > 0.70$ ; Table 1). For example, the probability of

352 impairment in longitudinal continuity between a given site and the sea increased with a  
353 decreasing proportion of individuals requiring more than four years to reach sexual maturity  
354 in the fish assemblage (i.e., Anguillidae, sea and river lampreys; Fig. 5A). Conversely, the  
355 proportion of eurythermal individuals in the assemblage was positively correlated to the  
356 probability of impairment by hydrological instability (Fig. 5B). There was a negative and non-  
357 linear relationship between the relative abundance of individuals with a long period of  
358 reproduction and the probability of significant impairment of fish communities by nitrates  
359 (Fig. 5C). Finally, a negative, linear relationship was also detected between the proportion of  
360 lithophilous spawners and the probability of impairment by fungicides (Fig. 5D). The  
361 remaining relationships and associated correlation coefficients are listed in Table H.

362

## 363 DISCUSSION

### 364 *Reliability of the diagnosis-based approach*

365 Combining a diverse array of taxonomic and functional information on fish communities has  
366 enabled us to build efficient models predicting community impairment by individual  
367 anthropogenic pressure categories in a multi-pressure context. Compared to many other  
368 studies on fish assemblages, we have constructed an unparalleled fish characteristic database  
369 by gathering and harmonizing information from various published functional trait databases,  
370 enriched by additional information on life-history strategies (e.g., longevity) and taxonomy (at  
371 family and order levels) of fish (Table B). We have taken into account a large set of pressure  
372 categories related to water quality (n = 13), habitat degradation (n = 14), and invasive alien  
373 species (n = 1), unlike most of previous fish studies investigating the effects of pressures  
374 related to WQ (Azimi and Rocher, 2016), HD (Adamczyk et al., 2017) or both WQ and HD  
375 but targeting less than 15 pressure categories (Pont et al., 2006; Schinegger et al., 2018, 2012;  
376 but see Allan et al., 2013). The majority of the pressure-specific models built here exhibited

377 very good ( $AUC_{\text{test}} \geq 0.80$ ) or good performances ( $0.70 \leq AUC_{\text{test}} < 0.80$ ). Using the 20 best  
378 performing models, we have detected a higher proportion of sampling events impaired by HD  
379 than WQ or IAS pressures (Fig. 1A). This result is consistent with recent literature that has  
380 considered that hydromorphological alterations (e.g. river discontinuities) may be the most  
381 frequent pressures acting on fish communities (Poikane et al., 2017; Schinegger et al., 2012)  
382 and on European surface waters, in general (EEA, 2018). Although we must be careful when  
383 comparing studies with not strictly similar definitions of pressure types, the obvious trend of a  
384 greater effect of HD pressures on fish communities suggests (i) more widespread  
385 anthropogenic modifications in HD than WQ conditions of streams, and/or (ii) higher  
386 sensitivity of fishes to changes in HD conditions than to changes in WQ conditions. Because  
387 we have found (i) a higher proportion of fish communities impaired by HD than WQ pressure  
388 categories (Fig. 1A) and (ii) no significant differences in the capacity of RF models to detect  
389 HD or WQ pressure categories, the first hypothesis seems more plausible.

390

### 391 *Disentangling multiple co-occurring pressures*

392 In this diagnosis-based approach, a RF model is built for each pressure category to detect  
393 bio/ecological shifts in fish communities. However, biological communities are often  
394 simultaneously impacted by multiple pressures (Allan et al., 2013; Poikane et al., 2017;  
395 Schinegger et al., 2018; this study). We found that fish communities were significantly  
396 impacted by, on average,  $7.34 \pm 0.03$  abiotic pressure categories (mean  $\pm$  SE; impairment  
397 probability  $> 0.50$ ; min. = 0, max. = 17). In a multi-pressure context, RF models are robust  
398 methods to detect the main impairment probabilities of fish communities by individual  
399 pressure categories using various combinations of biological and taxonomic metrics that  
400 respond to those pressures (Table H, Fig. 5). However, there are two main reasons explaining  
401 why the combined effects of co-occurring pressures on fish communities (e.g. direct, indirect,

402 nested, additive, synergistic or antagonistic) may have reduced the efficiency of some RF  
403 models in their capacity to detect the effects of individual pressure categories (Townsend et  
404 al., 2008; Villeneuve et al., 2018). First, the intensity of a given pressure may not be strong  
405 enough to provide a distinguishable signal in the functional trait profiles of fish communities.  
406 Second, the pressure gradient was not wide enough - i.e., provided not enough contrasting  
407 situations (“low” vs. “significant”) - to be efficiently discriminated by RF models. In those  
408 cases, comparing the variations in trait profiles of fish communities in mono- vs. multi-  
409 pressure contexts may help to improve the performance of poorly accurate RF models (Larras  
410 et al., 2017) and to understand the underlying effects of multiple pressures on ecosystem  
411 functions (Poikane et al., 2017).

412

#### 413 *Ecological validation and interpretation*

414 The analysis of the contribution of the most important biological metrics to the model  
415 impairment probabilities as well as the analysis of the response patterns of those metrics along  
416 an increasing gradient of impairment probability, allowed us gaining insights into the  
417 potential mechanisms explaining the community responses - linear or non-linear - to multiple  
418 pressures (Fig. 5, Table H). Yet, mechanisms of whole community responses to multiple  
419 pressures may be more complex than mere pressure-response relationships. For example, HD  
420 and WQ pressures may have direct lethal or sublethal effects on fish and indirect functional  
421 effects that percolate - more or less rapidly - through food webs. A combination of those  
422 direct and indirect effects may explain why the best RF model performances were obtained  
423 using different integration periods for the different WQ pressure categories. Therefore, the  
424 different integration periods should be interpreted as the period of time over which individual  
425 WQ parameters need to be taken into account to observe significant shifts in the functional  
426 composition of fish communities. A comprehensive ecological interpretation of RF model

427 outputs and underlying mechanisms would thus require answering the following questions i)  
428 “*which combinations of fish metrics respond to which pressures?*” and ii) “*which ones are the*  
429 *most (or least) responsive?*”. Although we have provided a few examples (Fig. 5, Table H),  
430 with more than 1500 fish metrics taken into account in this study, this task is challenging and  
431 beyond the scope of the present study. Addressing this task along with evaluating functional  
432 trait profiles in mono- vs. multi-pressure context would represent a significant addition to the  
433 research in the field of ecological risk assessment.

434

#### 435 *Impairment probabilities across spatial and temporal scales*

436 Based on the large spatial-temporal extent of our datasets, we have been able to investigate  
437 spatial and temporal patterns in fish community responses to anthropogenic pressures, via RF  
438 model results. The spatial patterns of river degradation found in this study were consistent  
439 with those observed in recently published papers; e.g. the widespread impact of longitudinal  
440 continuity impairment (Fig. 2A) on European (EEA, 2018; Schinegger et al., 2018) and North  
441 American fish communities (Cooper et al., 2016). In addition, Schinegger et al. (2012)  
442 showed that, overall, both WQ and HD pressures impaired lowland rivers whereas HD  
443 pressures dominantly impaired mountain and headwater streams, resulting in lowland streams  
444 being prone to a higher risk of degradation than mountain streams. Our analyses have  
445 confirmed this observation as the mean impairment probability (IP) of the 19 best performing  
446 abiotic RF models was negatively correlated with the site altitude (Spearman rank correlation  
447 coefficient = -0.59, p-value < 0.0001; see also Fig. 2E).

448

449 The temporal patterns in river conditions indicate that risks of river impairments, by at least  
450 two WQ variables, have significantly decreased between 2005 and 2015 (e.g., for nitrogen  
451 (except nitrates) and phosphorous compounds; Fig. 3D and F). These results are in line with

452 recent reductions in nutrient pollutions within freshwater ecosystems after many EU  
453 environmental policies to prevent nutrient losses (EEA, 2018; Flourey et al., 2013; Latli et al.,  
454 2017). Similar decreasing trends were observed for the overall risk of river degradation (Fig.  
455 3E). At the site scale, the diagnosis-based approach was also able to detect a decrease in  
456 impairment risks due to two WQ pressures ('Nitrates' and 'Nitrogen compounds') over a ten-  
457 year period (Fig. 4B). Overall, a powerful advantage of this approach is that various  
458 combinations of spatial (from site to catchment area) and temporal (from years to decades)  
459 scales can be investigated in order to optimize the selection of the most appropriate  
460 restoration measures, considering the pressure categories providing the highest impairment  
461 probabilities.

462

#### 463 *Effects of invasive alien species*

464 Our diagnosis-based approach using a wide variety of taxonomic and functional information  
465 has demonstrated high performance for evaluating the risk of community impairment by  
466 invasive alien species ( $AUC_{test} = 0.893$ ). With 43% of all the sampling events being  
467 significantly impacted by invasive alien species (impairment probability  $> 0.5$ ; Fig. 1A; Fig.  
468 B), our results have indicated that this pressure category is widespread across the French  
469 metropolitan territory. Those results are in line with many other recent studies from other  
470 locations worldwide (e.g., Europe, North America, New Zealand) and dealing with various  
471 invasive alien species across the tree of life (e.g., fish, reptile, birds, fungi, plants; Gallardo et  
472 al., 2016; Peoples and Goforth, 2017a, 2017b; Peoples and Midway, 2018; Strubbe et al.,  
473 2015).

474

475 Our results have indicated, however, that impairment probabilities due to invasive alien  
476 species globally decreased significantly over time in the sites that were sampled in both 2005

477 and 2015 (i.e., over a decade; Fig. 3H). Assuming that the risk of impairment by invasive  
478 alien species reflects the vulnerability of the native assemblage, and then the state of the  
479 surrounding ecosystem, we would expect to have higher probabilities of impairment by  
480 invasive alien species in disturbed sites compared to less disturbed sites (Gallardo et al.,  
481 2016). We have tested this hypothesis using two distinct, but complementary ways. First, we  
482 have checked the most important fish metrics contributing to the model predictions and found  
483 that the “proportion of omnivorous fish species in the native assemblage” was one of these  
484 metrics, and was highly ( $\rho = 0.85$ ) and positively correlated to the probability of impairment  
485 by IAS (Table H). This first result is supporting our hypothesis since omnivorous and  
486 generalist species are usually found in disturbed aquatic systems with high turbidity, organic  
487 matter and nutrient load (Gallardo et al., 2016). Second, we have evaluated the correlation  
488 between the mean impairment probabilities over the 19 abiotic pressure categories—a proxy  
489 of the overall ecosystem state—and the impairment probabilities resulting from the IAS RF  
490 model. Interestingly, we have found a positive and significant correlation (Fig. C), suggesting  
491 that the significant decrease in the impairment risk of native assemblages by invasive alien  
492 species was most likely related to the significant improvement in the ecosystem state over the  
493 studied decade (i.e., significant decrease in the mean total impairment probability; Fig. 3G).

494

495 Although our diagnosis-based approach was efficient at detecting the impairments induced by  
496 the invasive alien species and provided encouraging results, we acknowledge that an  
497 exhaustive evaluation of their effects on native communities is a more challenging task than  
498 simply considering their presence/absence (Jeschke et al., 2014; Schlaepfer, 2018). First,  
499 establishing a list of IAS was not a trivial task since this status could change across locations  
500 (local scale vs. regional scale), expert knowledge and institutions. This is the reason why we  
501 have chosen to combine the information from three main data sources (Table A). Second,

502 along with the presence/absence of invasive alien species, we could integrate their taxonomic  
503 richness and composition (i.e, species identities and relative abundances) as well as key  
504 functional metrics (e.g., the proportion of invasive alien predators, the maximum weight or  
505 fecundity of invasive alien species) within a multi-parametric “invasive alien species”  
506 pressure category. In this sense, our data indicate that, when present in the assemblage (i.e.,  
507 41% of the sampling events using abundance data), the IAS represented on average  $6.1 \pm 0.2$   
508 % of the total fish abundances within the assemblages. Third, we have only considered the  
509 impairment of native assemblages by invasive alien fish species whereas recent meta-analyses  
510 have demonstrated that the expansion of invasive alien macrophytes (Gallardo et al., 2016) or  
511 benthic macroinvertebrates (Latli et al., 2017; Otjacques et al., 2016) could trigger  
512 disproportionate adverse effects on native fish communities. Integrating those suggestions in  
513 future studies would most likely help to improve our understanding of the impacts of invasive  
514 alien species on native fish communities.

515

### 516 *Caveats*

517 We admit that the definition of multi-parametric “pressure categories” has required the  
518 aggregation of miscellaneous pressure parameters (from 1 to more than 70; described at  
519 various spatial and temporal scales). Each parameter may influence the fish assemblages and  
520 the fish-based metrics in different ways. The use of multi-parametric pressure categories was,  
521 however, a robust way to reduce models’ complexity and to deal with heterogeneous  
522 environmental data (e.g., parameters with missing data). Indeed, the performance of this  
523 predictive approach relies on the accuracy and availability of data used to characterize  
524 anthropogenic pressures. For instance, the ‘Acidification’ model was dropped since the  
525 associated pressure gradient could not be properly described (i.e., lack of significantly  
526 impacted sampling events). In addition, the amount of data available for describing the



527 intensity of WQ pressure categories was more variable among sampling events compared to  
528 HD ones (i.e., more missing data in WQ than in HD pressure categories; Tables C and D).  
529 Yet, the performances of WQ and HD models were not significantly different (i.e., no  
530 significant difference in  $AUC_{test}$  between the ten HD and nine WQ models exhibiting at least  
531 ‘good’ performances; see ‘Results’ section), thus demonstrating the robustness of this  
532 diagnosis-based approach. We therefore advocate that future studies on risk assessment  
533 should seek to establish standardized definitions of pressure categories based on (i) data  
534 availability and accuracy, and (ii) a broad range of individual pressure parameters, including,  
535 but not limited to, climatic factors (e.g. modifications in temperature and precipitation  
536 patterns), and overharvesting (Schinegger et al., 2013).

537

538 A recurrent issue in ecological risk assessment studies is to discriminate the individual effects  
539 of correlated pressure categories. This issue, however, can be addressed with our diagnosis-  
540 based approach by evaluating the most contributive fish metrics to each RF model of the  
541 correlated pressure categories. The hypothesis behind is that two strongly correlated pressures  
542 should be translated into similar functional trait profiles within communities. Although we  
543 found that the non-random co-occurrence of pressure categories was low overall (mean  
544 Cohen’s Kappa coefficient =  $0.07 \pm 0.01$ ), a few pairs of pressure categories exhibited rather  
545 high co-occurrences (Table G). Interestingly, correlated pressure categories shared some  
546 similar fish metrics among the five most important ones to each model thus confirming our  
547 hypothesis (Tables G and H).

548

#### 549 *Implications of the diagnosis-based approach*

550 The present work has proposed an approach to help scientists and environmental managers for  
551 identifying the individual impacts of various anthropogenic pressures potentially impairing

552 stream fish communities, both taxonomically and functionally (step II). We have designed the  
553 *R ecodiag*-package to allow assessment of pressure-specific impairment risks and to  
554 investigate the responses of communities to contrasting pressure levels. This R package  
555 produces convenient visualizations of model outputs (radar charts), enhancing the  
556 simultaneous evaluation of multiple pressures and the comparison of impairment risks over  
557 time. This last point is particularly useful for stream managers to rank the effects of co-  
558 occurring pressures for prioritizing management measures and implementing the most  
559 appropriate ones, for restoring river integrity (step III). Although we mainly presented results  
560 at the country scale (mainland France), this approach can also be applied at the site or local  
561 scale, which is, to date, the main spatial scale of most management measures. For instance, in  
562 the situation of Fig. 4B, stream managers could focus their efforts on reducing the effects of  
563 ‘Phosphorous compounds’, ‘Transportation facilities’ and anthropogenic modifications of  
564 ‘Stream width’, by acting on the individual parameters defining these pressure categories (e.g.  
565 total phosphorous concentrations, presence of roads or railways near stream banks and areas  
566 of intensive farming, respectively). The other significant pressures could be more difficult to  
567 manage over a short period (e.g. the cumulative effects of barriers between the studied site  
568 and sea; pressure category ‘Longitudinal continuity’) or did not represent serious threats  
569 (impairment probability lower than 0.50; ‘Nitrates’, and ‘Nitrogen compounds’).

570

### 571 *Conclusion*

572 The current application of the diagnosis-based approach does not aim at replacing existing  
573 fish-based indices. The major goal of fish-based indices is to provide an efficient evaluation  
574 of the global impact of anthropogenic pressures impairing the monitored water bodies. This  
575 approach is rather complementary to fish-based indices for more precisely identifying the  
576 nature of the anthropogenic pressures significantly impairing each of these water bodies (here,

577 river reaches) and for helping managers to select the best strategy for restoring water and  
578 habitat quality in these water bodies (e.g. reaching the ‘good’ ecological status). Out of the 28  
579 abiotic and biotic pressure categories tested, the random forest models have efficiently  
580 detected the impairments due to nine chemical, ten hydromorphological and one biological  
581 pressures. The best performing models also indicated that fish communities were impacted,  
582 on average, by seven abiotic pressure categories but the overall impairment probabilities of  
583 the whole fish assemblages (including native and invasive alien species) decreased  
584 significantly between 2005 and 2015. This diagnosis-based approach is robust and flexible  
585 enough to assess specific risks of river degradation under multi-pressure scenarios using  
586 distinct biological quality elements (i.e., macroinvertebrates, diatoms, and now fishes). It is  
587 also capable to handle complex pressure-response relationships (linear and non-linear  
588 relationships), able to diagnose various water bodies (from headwaters to lowland rivers), and  
589 to simultaneously consider a variety of pressure categories (hydrological, chemical and  
590 biological ones). *In fine*, merging the information provided by distinct biological quality  
591 elements (e.g., macroinvertebrates, diatoms, fishes, macrophytes) should pave the way for a  
592 holistic and efficient understanding of the structure and functions of stream ecosystems, that  
593 will help water managers worldwide in the decision-making process in relation to river  
594 restoration.

595

## 596 SUPPORTING INFORMATION

597 Table A: List of fish species

598 Table B: List of fish characteristics

599 Table C: Description of water quality pressure categories

600 Table D: Description of hydromorphological pressure categories

601 Table E: *ecodiag*-package

602 Table F: Results of RF models for water quality pressure categories and associated integration  
603 periods

604 Table G: Cohen's kappa coefficients evaluating the non-random co-occurrence of pressure  
605 categories

606 Table H: List of the most contributive biological metrics to RF models

607 Figure A: Distribution of impairment probabilities across pressure levels for RF models  
608 exhibiting at least 'good' performances

609 Figure B: Time-integrated map of fish assemblage impairment probabilities (IP) by the  
610 invasive alien species.

611 Figure C: Correlation between the mean total IP and the mean IP by the invasive alien  
612 species.

613

#### 614 AUTHORS' CONTRIBUTION

615 PUP and CM designed methodology; CM developed the *ecodiag*-package; LV, AC and KK  
616 helped with environmental data; JB and M-LM worked on biological and environmental data  
617 related to migratory fish species; SB provided morphological trait data; SD and YR brought  
618 insights regarding WFD implications and ecology of fish communities; OD contributed to  
619 research design and ideas, compiled all biological and environmental data, performed  
620 analyses, and led the writing of the manuscript. All authors contributed critically to the drafts  
621 and gave final approval for publication.

622

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634

#### 635 DATA ACCESSIBILITY

636 Data will be made available in the form of a data paper or through public repositories.

637

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867 Table and figure captions

868 Table 1: List of pressure categories and associated codes, integration period (for chemical  
869 pressures only; expressed in number of years - before sampling events - taken into account for  
870 pressure description), and AUC of models built on the learning datasets ( $AUC_{learn}$ ) and tested  
871 against new data ( $AUC_{test}$ ). Within hydromorphological (HD) and chemical (WQ) pressure  
872 categories, individual models are sorted in decreasing order of  $AUC_{test}$ . Shades of grey denote  
873 model performances: dark grey = at least “good”; light grey = “fair”; blank = “poor”.

874

875 Figure 1: A/ Relative proportions of sampling events with impairment probabilities higher  
876 (black bars; “significant” impact) than 0.5 for each of the 20 hydromorphological (HD), water  
877 quality (WQ), and biological (IAS) models with  $AUC_{test} > 0.7$ ; B/ Principal Component  
878 Analysis performed on impairment probabilities from the 19 models corresponding to the HD  
879 (dashed lines and triangles) and WQ (solid lines and circles) pressure categories ( $n = 8529$   
880 sampling events). Correlations of the probabilities provided by the 19 models with the two  
881 first principal components. Due to the particularity of the IAS model (see Material and  
882 Method section), associated impairment probabilities were projected onto the existing  
883 multivariate space (as a supplementary variable) and correlations recalculated (dotted line and  
884 square). See Table 1 for the list of codes and labels.

885

886 Figure 2: Time-integrated map of fish assemblage impairment probabilities (IP) at each of the  
887 1527 sites. For each site, IPs were averaged across sampling events for the two  
888 hydromorphological (A/ and B/), and chemical (C/ and D/) pressure categories corresponding  
889 to RF models with the highest  $AUC_{test}$ . In E/ a mean IP was calculated for each site by  
890 averaging IPs across the 19 HD and WQ pressure categories with  $AUC_{test} > 0.70$  and over the

891 2005-2015 period. Lines and shaded areas on maps represent the boundaries of drainage  
892 basins (B1 = Adour-Garonne; B2 = Loire; B3 = Seine; B4 = Escaut-Somme; B5 = Meuse; B6  
893 = Rhine; B7 = Rhone; B8 = Corse) and the locations of mountain ranges (M1 = Pyrenees; M2  
894 = Massif Central; M3 = Alps; M4 = Jura Mountains; M5 = Vosges), respectively. Panels A-D  
895 are on a different color scale (presented in panel A) compared to panel E. The locations of  
896 Site A and B are indicated (cf. Fig. 4).

897

898 Figure 3: Temporal changes in impairment probability distributions (IP), at the sites that were  
899 sampled in both 2005 and 2015 ( $n = 543$ ). Results are provided for the three  
900 hydromorphological (HD; A/, C/, and E/) and chemical (WQ; B/, D/ and F/) pressure  
901 categories corresponding to the RF models with the highest  $AUC_{test} (> 0.80)$  and for the 19  
902 best performing HD and WQ models ( $AUC_{test} > 0.70$ ; G/). Since abiotic and biotic models  
903 were built differently, temporal changes in IP due to invasive alien species are shown in the  
904 last panel (H/) and were not included in the calculation of the 'Total IP'. (\*\*\*) indicates a  
905 highly significant decrease in mean IP values (black triangles) in 2015 compared to 2005  
906 (Wilcoxon sign-rank test for paired data; all  $p$ -values  $< 0.0001$ ), whereas (\*\*) indicates a  
907 significant decrease in IP between the two dates ( $0.0001 < p$ -value  $< 0.001$ ).

908

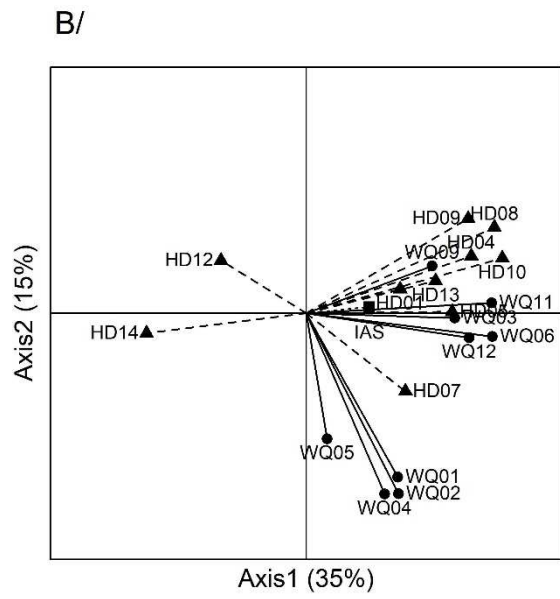
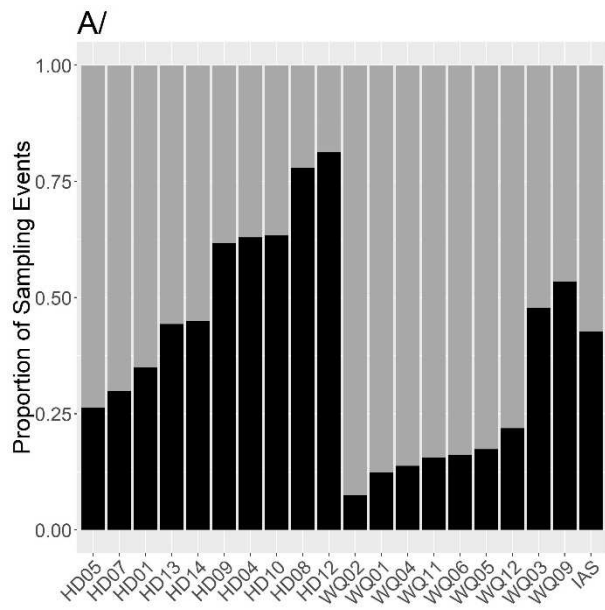
909 Figure 4: Radar charts based on site-specific and inter-annual changes in impairment  
910 probabilities (IPs) for the three hydromorphological (HD) and the three chemical (WQ)  
911 pressure categories corresponding to the RF models with the highest  $AUC_{test} (> 0.80)$ . A/  
912 example of a site (the Loup river, at Tourrette-sur-Loup) with stable and low IP ( $< 0.50$ ; i.e.  
913 the dashed blue line) for all the sampling events; B/ example of another site (the Gier river, at  
914 Givors) with a partial ecological recovery for three pressures (Nitrogen compounds, Nitrates,  
915 and Phosphorous compounds), whereas IPs remained stable and high (i.e.  $IP > 0.50$ ) for the



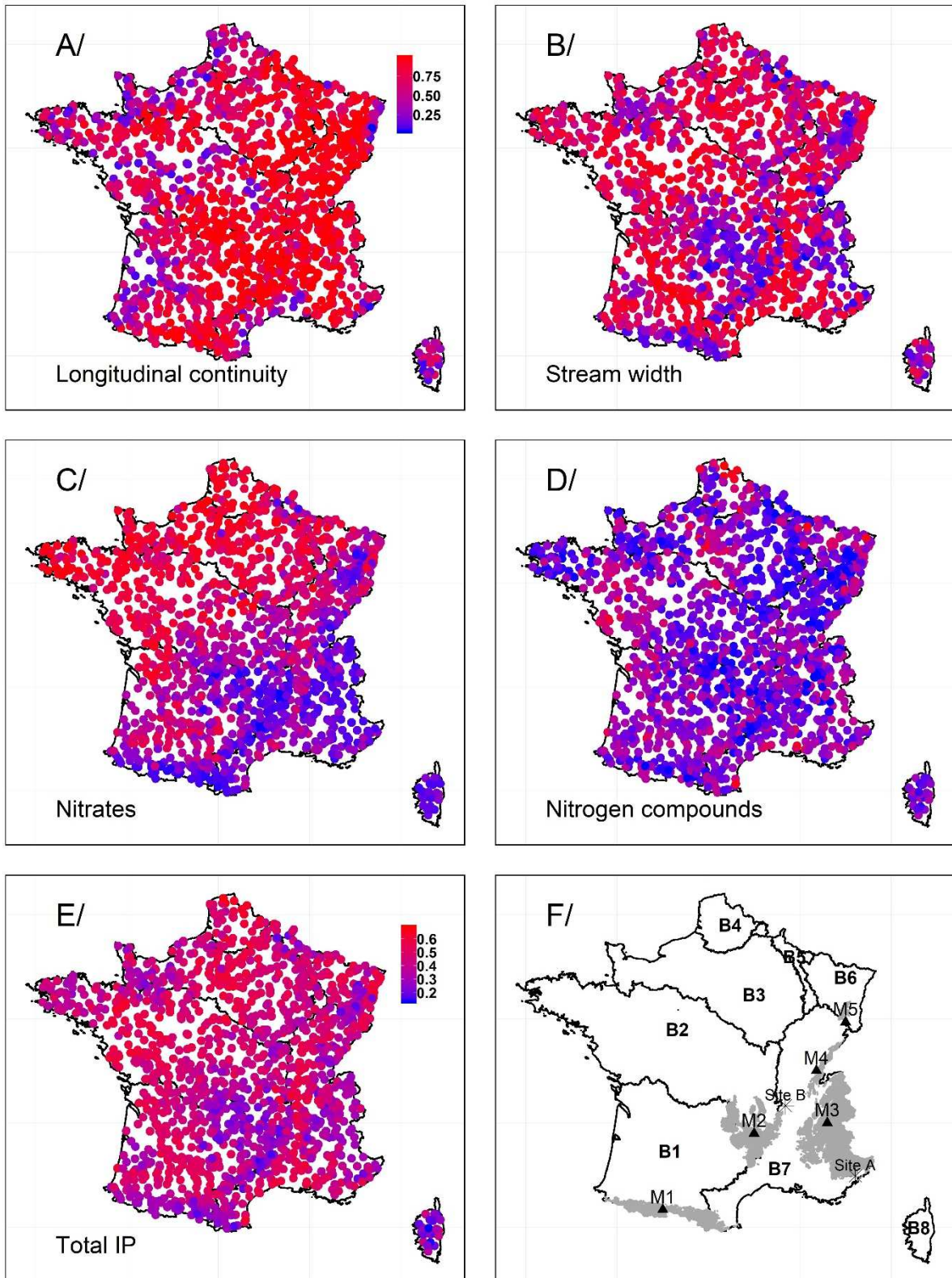
916 three other pressure categories over the same period. “Long. continuity” = Longitudinal  
917 continuity; “N. comp.” = Nitrogen compounds (except nitrates); “P. comp.” = Phosphorous  
918 compounds. See Fig. 2F for the site locations in France.

919

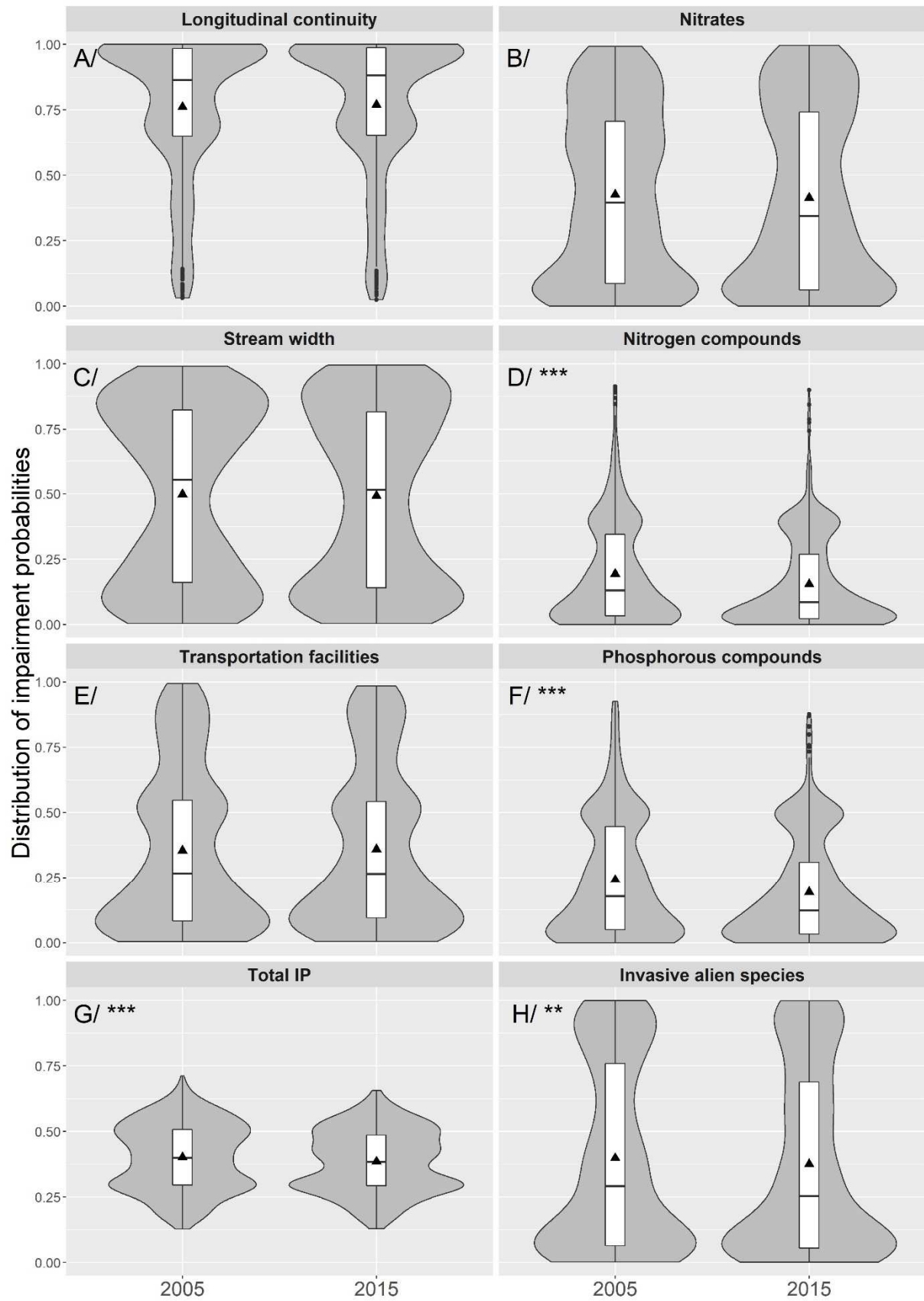
920 Figure 5: Relationships between observed metric values (x-axis) and impairment probabilities  
921 (y-axis) predicted by two hydromorphology-related (A/, B/) and two water quality-related (C/,  
922 D/) RF models. rho = Spearman rank correlation coefficient (all p-values < 0.0001). We also  
923 explored the relationships between the impairment probabilities of the 19 best performing RF  
924 models (AUC<sub>test</sub> > 0.70; Table 1) and the observed metric values of the five most  
925 contributive metrics to the predictions of those models (Table H).



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Figure 1

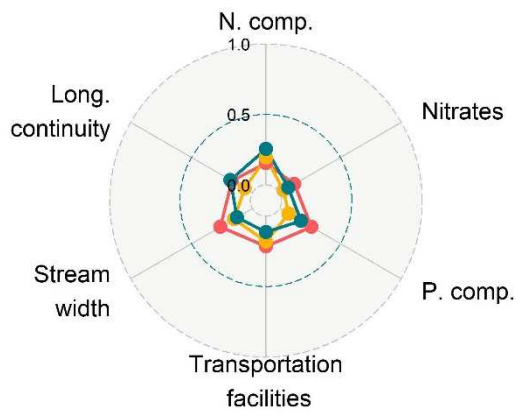


4  
 5 Figure 2  
 6

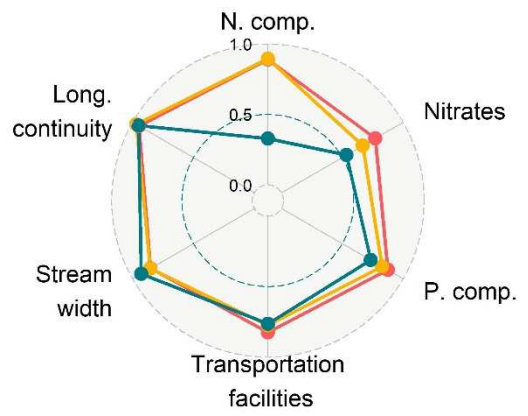


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Figure 3

A/ Site A



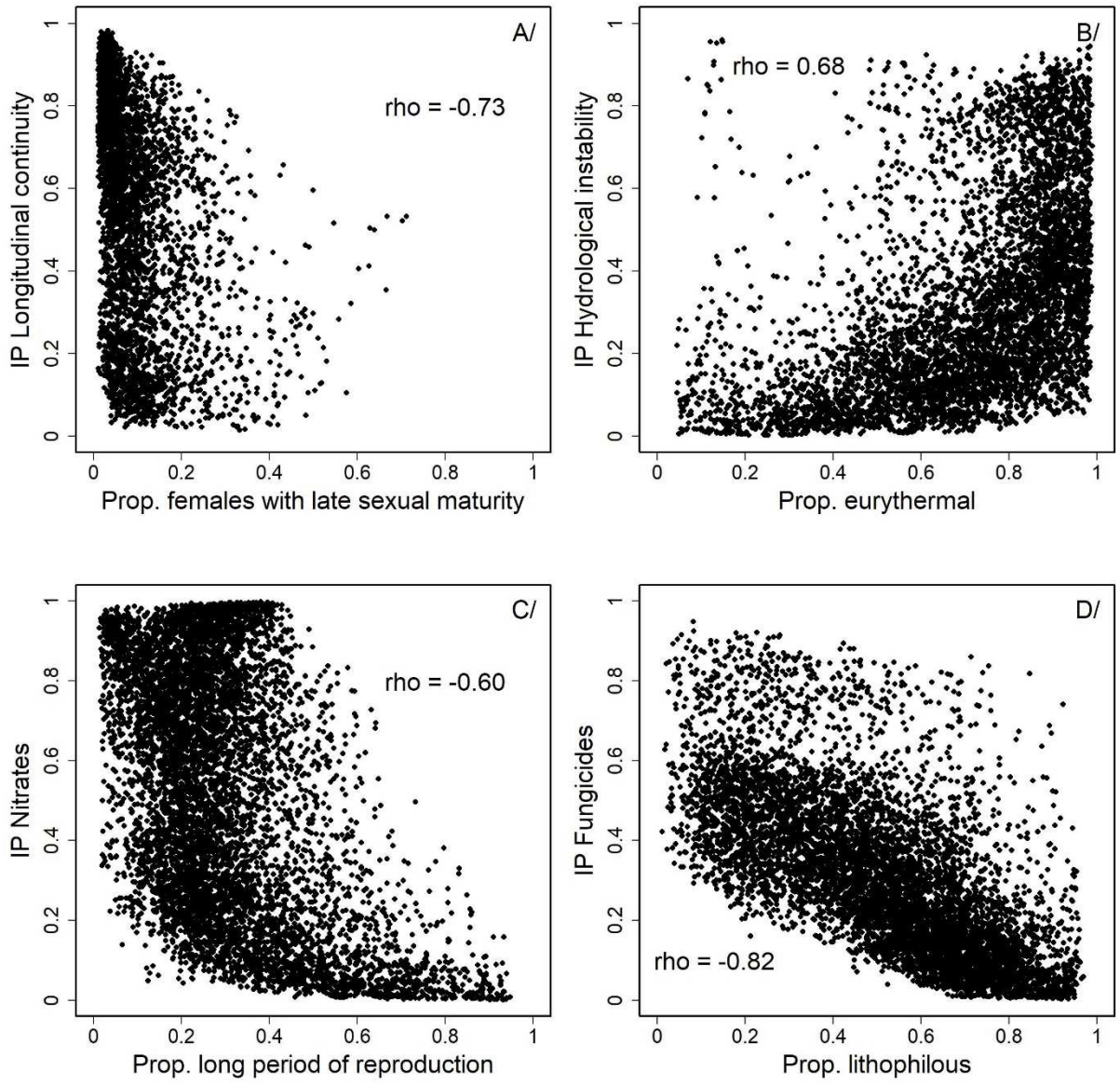
B/ Site B



Years:  
—●— 2005  
—●— 2010  
—●— 2015

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Figure 4

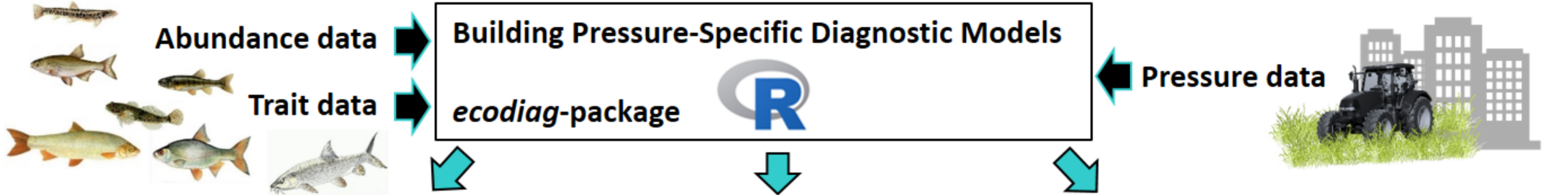


15  
16 Figure 5

1 Table 1

Code	Pressure category	Year	AUC <sub>train</sub>	AUC <sub>test</sub>
<b>Chemical:</b>				
WQ03	Nitrates	4	0.914	0.878
WQ02	Nitrogen compounds (except nitrates)	3	0.897	0.871
WQ04	Phosphorous compounds	5	0.896	0.849
WQ01	Organic matter	4	0.894	0.847
WQ06	Fungicides	5	0.825	0.789
WQ05	Suspended particles	5	0.803	0.766
WQ12	PCB	3	0.774	0.760
WQ09	PAH	4	0.778	0.748
WQ11	Insecticides	3	0.745	0.719
WQ08	Herbicides	5	0.767	0.678
WQ10	Organic micropollutants (other)	5	0.792	0.655
WQ07	Mineral micropollutants	3	0.674	0.637
<b>Hydromorphological:</b>				
HD12	Longitudinal continuity	-	0.924	0.858
HD10	Stream width	-	0.895	0.852
HD01	Transportation facilities	-	0.896	0.819
HD08	Flow types	-	0.925	0.787
HD04	Clogging risk	-	0.861	0.774
HD09	Sediments	-	0.863	0.761
HD07	Catchment anthropization	-	0.854	0.755
HD05	Hydrological instability	-	0.861	0.748
HD13	Lateral continuity	-	0.851	0.728
HD14	Proximal continuity	-	0.852	0.703
HD11	Stream depth	-	0.852	0.696
HD02	Riverine vegetation	-	0.857	0.694
HD03	Urbanization	-	0.820	0.613
HD06	Straightening	-	0.805	0.597
<b>Biological:</b>				
IAS	Invasive alien species	-	0.897	0.893

2



**Non-linear ecological responses**      **Impairment Probabilities (IP) across spatial and temporal scales**

