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

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

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

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

## INTRODUCTION

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

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

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

1) Fish-based indices must rely upon the debated notion of reference conditions, which are often difficult to evaluate and an unrealistic target for the management of heavily modified water bodies (Dufour and Piégay, 2009; Tweedley et al., 2017).
2) Despite growing evidence that fish communities may exhibit non-linear responses to pressure gradients (Taylor et al., 2014), most fish-based indices assume linear relationships (but see Bhagat et al., 2007).
3) Fish-based indices are unable to identify specific risks of stream degradation in a multipressure context and to disentangle the effects of co-occurring pressures acting across various spatial and temporal scales (Reyjol et al., 2014). This tremendously limits the capacity of water managers to opt for the most relevant restoration measures, for a given water body.

To overcome these limitations, we here developed a robust and flexible diagnosis-based approach, able to identify individual pressures involved in fish community impairment under multiple pressure scenarios. This approach has been grounded upon the fundamental notion
that functional species traits - the physiological, morphological and behavioural characteristics of organisms - integrate the spatial and temporal variability of the environment (Southwood, 1977; Townsend and Hildrew, 1994). Various natural environmental filters, acting at different nested scales from ecoregions to reaches, filter functional traits. Anthropogenic pressures are then able to modify this environmental filtering, leading to different combinations of traits that are often specific to a given pressure category (Desrosiers et al., 2019). Hence, according to our hypothesis, substantial modifications in the relative frequencies of a large set of functional traits (i.e. biological metrics) within communities should specifically reflect biological responses to one or more environmental pressures.

Random forest (RF) models have been successfully applied in many environmental and ecological studies including - but not limited to - air quality modeling (Choubin et al., 2020), flood susceptibility assessment (Hosseini et al., 2020), groundwater nitrate modeling (Rahmati et al., 2019), earth fissure modeling (Choubin et al., 2019), risk assessment for invasive species (Keller et al., 2011; Philibert et al., 2011), and stream impairment modeling under multi-pressure scenario (Larras et al., 2017; Mondy and Usseglio-Polatera, 2013). The advantage of RF models, over other uni- and multi-variate methods, is their ability to handle non-linear relationships between predictors and response variables, to integrate complex interactions among a large number of predictors and to generate good predictions with their associated probabilities (Prasad et al., 2006; Rahmati et al., 2019). The proposed diagnosisbased approach is a generalization of the use of RF models to estimate probabilities of community impairments, in a multi-pressure context, using complex combinations of taxonomic and biological metrics. We tested this flexible approach with river fish communities by: 1) compiling fish functional traits sensu stricto and any relevant information on how fish assemblages may interact with their environment (e.g. functional guilds, life
history strategies, taxonomic structure), 2) identifying and describing abiotic and biotic pressure categories to which fishes are potentially sensitive at various spatial and temporal scales, and 3) using RF models to calculate probabilities of community impairment by any given pressure category. Based on fish communities, this study aims at demonstrating the efficiency of the flexible diagnosis-based approach for enhancing our understanding of shifts in ecosystem structure and functions triggered by given levels of various anthropogenic pressures (step II), and for facilitating decision-making processes by environmental managers (step III).

## MATERIAL \& METHODS

## Fish and environmental data

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

The Fish record dataset contains electrofishing samples performed from 2005 to 2015 in 1527 sites belonging to the French Water Framework Directive's surveillance monitoring, covering the mainland France (c.a. $550000 \mathrm{~km}^{2}$ ). The standardized electrofishing protocol followed the recommendations of the European Committee for Standardisation's standard (CEN, 2003; Marzin et al., 2014). This protocol was conducted during low-flow periods (from May to October) with a sampling design that depended on river width and depth. Fishes were identified to the species level, counted and then released back into the river. On average, the sites were sampled $5.5 \pm 0.1$ times ( Mean $\pm$ SE; $\min =1 ; \max =11$ ) over the 2005-2015
period. The Fish record dataset was thus composed of the abundances of 72 fish species distributed in 8529 sampling events (site x year; Table A).

The Fish characteristic dataset gathered information about 56 fish characteristics (260 categories) compiled from different sources of information (Table B). Hereafter, we considered 'fish characteristics' as any descriptive information related to the physiology, morphology and behaviour (i.e. functional traits sensu stricto; $\mathrm{N}=53$ ), life history strategy (lifespan; $\mathrm{N}=1$ ), and taxonomy (family and order levels; $\mathrm{N}=2$ ) of fish. Fifty-five fish characteristics were coded using a full disjunctive system (i.e. presence/absence of various categories). One fish characteristic was structured using a fuzzy-coding technique ('food'; Table B). The number of categories defined per characteristic was rather low (between 2 and 10), except for the characteristics related to taxonomic classification ( $\mathrm{N}=20$ families/categories; $\mathrm{N}=13$ orders/categories).

The Fish record and Fish characteristic datasets were then used to calculate 1576 metrics for each of 8529 fish sampling events. These metrics covered: (i) the relative frequency of the log-transformed abundances of organisms using each of the 260 categories belonging to the 56 fish characteristics (260 metrics), (ii) taxonomy-based diversity indices calculated at the scale of each trait category (e.g. the proportion of piscivorous taxa, the Shannon diversity of rheophiles; 1255 metrics), (iii) Rao functional diversity indices calculated at the scale of each characteristic (Rao, 1982; 56 metrics), (iv) the level of diet specialization (Mondy and Usseglio-Polatera, 2014; one metric) and (v) taxonomy-based richness or diversity indices (e.g. the species richness, the Shannon diversity index; four metrics; Table B).

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

For parameters related to WQ pressure categories, pressure levels were assessed by comparing the mean value of individual parameters over a given period with the "good/moderate" quality class boundary of the French water quality assessment system (i.e. SEQ-Eau V2.0; Oudin and Maupas, 2003), allowing to distinguish "low" (high/good) from "significant" (moderate/poor/bad) pressure levels. Four of the 14 HD pressure categories were defined according to the literature (i.e. 'Transportation facilities, 'Urbanization', 'Hydrological instability', 'Straightening'; Larras et al., 2017; Mondy et al., 2012; Mondy and Usseglio-Polatera, 2013; Villeneuve et al., 2015), whereas the remaining categories and associated parameters were newly described in this study after preliminary tests to define the low-significant boundaries of pressure levels (Table D). For the IAS pressure category, we have considered as "significant" the effect of the presence of at least one invasive alien species in the fish assemblage. The list of invasive alien species, including 17 species, was
established by combining the information from three data sources: the French National Museum of Natural History, the IUCN Red list of threatened species in France (those considered as "alien species") and Fishbase (those considered as "Potential pest") (more details in Table A). We used the Cohen's kappa coefficient to assess non-random cooccurrences of pressure categories; i.e., after removing random co-occurrences ("low" $v s$. "significant"; psych-package in R).

## Random forest models

Random forests (RF) are a particular type of machine learning methods able to investigate complex and potentially non-linear relationships (Breiman, 2001). Although several other types of machine learning methods exist ( k -nearest neighbour, support vector machine), RF models were selected here because they can handle a large number of predictive variables. The importance of the contribution of those predictors to model outputs (probabilities) can be easily quantified. Moreover, RF models provide good predictive performances (Rahmati et al., 2019). One RF model was built per pressure category taking the pressure level ("low" vs. "significant") as the response variable and the fish metrics as the predictive variables. We tested five periods (from one to five years before fish sampling) to identify the most likely integration time of fish communities regarding WQ for two reasons: fish have potentially long life cycles (from one year to several decades) and the water samples (taken for chemical analyses) are punctual (i.e., several times a year) and were not necessarily obtained the same day as fish samples. Since all the HD pressures were defined once for the period covered by fish sampling, we did not test several integration periods as we did for WQ pressures. The period yielding the best model performance (see below) was selected independently for each WQ pressure. Among all the pressures, the one induced by the invasive alien species is particular because it is the only biotic pressure where a subset of the fish assemblage (IAS)
may impact the other subset (native species). To this end, the associated RF model was built differently. All the fish metrics were calculated again without the invasive alien species (predictive variables) while the presence (= "significant pressure") versus absence (="low pressure") of invasive alien species was considered as the response variable. Although the ecodiag-package presented in this study offers ways to tune RF models and can easily be used in a cross-validation procedure, we have only presented the results of models parametrized following published strategies (e.g., 1500 classification trees, random selection of 50 fish metrics at each node in individual trees; (Larras et al., 2017). Based on the fish metric values, each pressure-specific model therefore provided, for each sampling event ( $\mathrm{n}=8529$ ), a mean impairment probability calculated over all the trees defining a given forest. In order to identify pressure syndromes (i.e. combinations of pressures often impairing fish communities) over the entire studied area (mainland France), we also performed a Principal Component Analysis (PCA) on the impairment probabilities predicted by the abiotic and biotic models exhibiting at least "good" performances, considering all the 8529 sampling events.

## Model performances

To assess model performances, each RF model was built on a learning dataset and then tested against a test dataset (Larras et al., 2017; Mondy and Usseglio-Polatera, 2013). Learning and test datasets corresponded to independent subsamples of the input database, i.e., the metric and pressure data measured for all the sampling events. To take into account the temporal correlations among sampling events, the sites were randomly allocated either to the learning or to the test dataset ( $75 \%$ and $25 \%$ of the sites, respectively). Model performance was then evaluated with the Area Under the Curve (AUC). The AUC values indicate "good" model performances if they are equal to or higher than 0.70 (i.e., maximizing both sensitivity and specificity; Pearce and Ferrier, 2000). We used this AUC criterion to assess model
performances using mainly the test ( $\mathrm{AUC}_{\text {test }}$ ) datasets, and to a lower extent the learning $\left(\mathrm{AUC}_{\text {learn }}\right)$ one. Evaluating both $\mathrm{AUC}_{\text {test }}$ and $\mathrm{AUC}_{\text {learn }}$ represent a trade-off to minimize prediction errors in test datasets and to avoid over-fitting in learning datasets. We then investigated spatial and temporal changes in RF model outputs (i.e., impairment probability by any given pressure) using models with the best performances.

## Metric importance and ecological validation

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

## RESULTS

## Model performances

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

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

## Impairment probabilities across spatial scales

The proposed diagnosis-based approach applied to fish communities allowed us to investigate pressure patterns at different spatial scales. At a large spatial scale (mainland France), the RF models identified the hydromorphological pressures as the most frequently significant
pressures (from 26 to $81 \%$ of the sampling events) detected at any given site compared to water quality pressures (mainly from 8 to $22 \%$; except for "Nitrates" [48\%] and "PAH" [54\%]) or invasive alien species pressure ( $43 \%$ of the sampling events; Fig. 1A). Overall, RF models predicted that a given sampling event was significantly impacted, on average, by 5.27 $\pm 0.02 \mathrm{HD}(\min .=0, \max .=10)$ and $2.06 \pm 0.02 \mathrm{WQ}(\min .=0, \max .=9)$ pressure types (impairment probabilities > 0.50). The first two PCA axes explained $50 \%$ of the total variance in impairment probabilities (Fig. 1B). The first axis discriminated two hydromorphological pressures related to the river continuity $(\mathrm{HD} 12=$ 'Longitudinal continuity'; $\mathrm{HD} 14=$ 'Proximal continuity'; negatively correlated to Axis 1) from all the other pressure categories (more or less positively correlated to Axis 1 ). The second axis separated the two broad types of pressure categories with most of the HD and WQ pressure impairment probabilities being, respectively, positively (except HD07) and negatively (except WQ9 and WQ11) correlated to this axis (Fig. 1B). The IAS pressure was positively - but lowly - correlated to the first PCA axis. Note that overall the non-random co-occurrence of pressure categories (RF models' inputs) was very low (mean Cohen's Kappa coefficient $=0.07 \pm 0.01$ ) except for 11 of the 190 pairs of pressures (Table G).

The proposed diagnosis-based approach also highlighted broad spatial patterns in stream degradation. For each site and each of four pressure categories, impairment probabilities were averaged across sampling events (over the 2005-2015 period) to remove the temporal effect in the data, and then plotted on a map displaying the main French drainage basins (Fig. 2). We provided this spatial distribution of impairment probabilities for the two HD and WQ models exhibiting the highest performances. The regional patterns of impairment probabilities were different among pressure categories. The risk of fish community impairment by 'Longitudinal continuity' alteration was lower (mean impairment probability $<0.50$ ) for rivers located near
the Atlantic and Mediterranean coasts, than those located further inland (Fig. 2A). The effects of modifications in 'Stream width', a proxy of stream channeling and bank modifications, on fish communities, were the most important in sites located on the main large rivers (the Loire, the Rhone, the Seine, and the Adour-Garonne rivers; Fig. 2B) and their lowland tributaries. In addition, 'Nitrates' and 'Nitrogen compounds' (other than nitrates) exhibited contrasting impairment patterns: the former had a pronounced, large-scale impact in the northwestern part of France, and to a lower extent, in the Adour-Garonne basin (South-west; Fig. 2C) whereas the latter had rather local effects on communities and mainly in areas exhibiting the most intensive agricultural activity (Fig. 2D). The impact of invasive alien species on native fish assemblages, was also the most important in sites located on main large rivers (the Loire, the Rhone, the Adour-Garonne, and to a lower extent, the Seine rivers) and their lowland tributaries (Fig. B). Finally, we assessed the global impairment probability of the whole fish communities (including native and invasive alien species) over the decade, by averaging the impairment probabilities of the 19 HD and WQ models with at least good performances $\left(\mathrm{AUC}_{\text {test }}>0.70\right.$; Table 1$)$. Least impacted sites were mainly located within or near the main mountain ranges (e.g., Pyrenees, Alps, Massif Central mountains, and to a lower extent the Vosges, and Jura Mountains; Fig. 2E).

## Impairment probabilities across temporal scales

This approach has also allowed assessing broad temporal patterns in stream degradation at large spatial scale. Figure 3 depicts changes in the distribution of impairment probabilities at all the sites sampled in both 2005 and $2015(\mathrm{n}=543)$. These trends showed pressure-specific patterns. There was only a significant decreasing risk of community impairment due to 'Nitrogen compounds' and to 'Phosphorous compounds' in 2015 compared to 2005 (Wilcoxon sign-rank test for paired data $=9.510^{4}$ and $1.010^{5}$, both p-values $<0.0001$,
respectively; Fig. 3D and F). Differences in the total impairment probabilities (i.e. mean impairment probabilities of the 19 HD and WQ models with $\mathrm{AUC}_{\text {test }}>0.70$; Table 1) between 2005 and 2015, tended to decrease, indicating a weak improvement in overall site condition over the decade (Wilcoxon sign-rank test for paired data $=1.010^{5}$, p -value $<0.0001$; Fig. 3G). There was also a significant decrease in the impairment probabilities induced by the invasive alien species (Wilcoxon sign-rank test for paired data $=8.710^{4}$, p -value $=0.0004$; Fig. 3H).

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

## Ecological validation

The relationships between impairment probabilities and observed metric values were explored for the five most contributive metrics to each model (among 1576 tested metrics) and each of the 19 most accurate RF models $\left(\mathrm{AUC}_{\text {test }}>0.70\right.$; Table 1). For example, the probability of
impairment in longitudinal continuity between a given site and the sea increased with a decreasing proportion of individuals requiring more than four years to reach sexual maturity in the fish assemblage (i.e., Anguillidae, sea and river lampreys; Fig. 5A). Conversely, the proportion of eurythermal individuals in the assemblage was positively correlated to the probability of impairment by hydrological instability (Fig. 5B). There was a negative and nonlinear relationship between the relative abundance of individuals with a long period of reproduction and the probability of significant impairment of fish communities by nitrates (Fig. 5C). Finally, a negative, linear relationship was also detected between the proportion of lithophilous spawners and the probability of impairment by fungicides (Fig. 5D). The remaining relationships and associated correlation coefficients are listed in Table H .

## DISCUSSION

## Reliability of the diagnosis-based approach

Combining a diverse array of taxonomic and functional information on fish communities has enabled us to build efficient models predicting community impairment by individual anthropogenic pressure categories in a multi-pressure context. Compared to many other studies on fish assemblages, we have constructed an unparalleled fish characteristic database by gathering and harmonizing information from various published functional trait databases, enriched by additional information on life-history strategies (e.g., longevity) and taxonomy (at family and order levels) of fish (Table B). We have taken into account a large set of pressure categories related to water quality $(\mathrm{n}=13)$, habitat degradation $(\mathrm{n}=14)$, and invasive alien species ( $\mathrm{n}=1$ ), unlike most of previous fish studies investigating the effects of pressures related to WQ (Azimi and Rocher, 2016), HD (Adamczyk et al., 2017) or both WQ and HD but targeting less than 15 pressure categories (Pont et al., 2006; Schinegger et al., 2018, 2012; but see Allan et al., 2013). The majority of the pressure-specific models built here exhibited
very good ( $\left.\mathrm{AUC}_{\text {test }} \geq 0.80\right)$ or good performances $\left(0.70 \leq \mathrm{AUC}_{\text {test }}<0.80\right)$. Using the 20 best performing models, we have detected a higher proportion of sampling events impaired by HD than WQ or IAS pressures (Fig. 1A). This result is consistent with recent literature that has considered that hydromorphological alterations (e.g. river discontinuities) may be the most frequent pressures acting on fish communities (Poikane et al., 2017; Schinegger et al., 2012) and on European surface waters, in general (EEA, 2018). Although we must be careful when comparing studies with not strictly similar definitions of pressure types, the obvious trend of a greater effect of HD pressures on fish communities suggests (i) more widespread anthropogenic modifications in HD than WQ conditions of streams, and/or (ii) higher sensitivity of fishes to changes in HD conditions than to changes in WQ conditions. Because we have found (i) a higher proportion of fish communities impaired by HD than WQ pressure categories (Fig. 1A) and (ii) no significant differences in the capacity of RF models to detect HD or WQ pressure categories, the first hypothesis seems more plausible.

## Disentangling multiple co-occurring pressures

In this diagnosis-based approach, a RF model is built for each pressure category to detect bio/ecological shifts in fish communities. However, biological communities are often simultaneously impacted by multiple pressures (Allan et al., 2013; Poikane et al., 2017; Schinegger et al., 2018; this study). We found that fish communities were significantly impacted by, on average, $7.34 \pm 0.03$ abiotic pressure categories (mean $\pm \mathrm{SE}$; impairment probability $>0.50 ; \min .=0, \max .=17$ ). In a multi-pressure context, RF models are robust methods to detect the main impairment probabilities of fish communities by individual pressure categories using various combinations of biological and taxonomic metrics that respond to those pressures (Table H, Fig. 5). However, there are two main reasons explaining why the combined effects of co-occurring pressures on fish communities (e.g. direct, indirect,
nested, additive, synergistic or antagonistic) may have reduced the efficiency of some RF models in their capacity to detect the effects of individual pressure categories (Townsend et al., 2008; Villeneuve et al., 2018). First, the intensity of a given pressure may not be strong enough to provide a distinguishable signal in the functional trait profiles of fish communities. Second, the pressure gradient was not wide enough - i.e., provided not enough contrasting situations ("low" vs. "significant") - to be efficiently discriminated by RF models. In those cases, comparing the variations in trait profiles of fish communities in mono- vs. multipressure contexts may help to improve the performance of poorly accurate RF models (Larras et al., 2017) and to understand the underlying effects of multiple pressures on ecosystem functions (Poikane et al., 2017).

## Ecological validation and interpretation

The analysis of the contribution of the most important biological metrics to the model impairment probabilities as well as the analysis of the response patterns of those metrics along an increasing gradient of impairment probability, allowed us gaining insights into the potential mechanisms explaining the community responses - linear or non-linear - to multiple pressures (Fig. 5, Table H). Yet, mechanisms of whole community responses to multiple pressures may be more complex than mere pressure-response relationships. For example, HD and WQ pressures may have direct lethal or sublethal effects on fish and indirect functional effects that percolate - more or less rapidly - through food webs. A combination of those direct and indirect effects may explain why the best RF model performances were obtained using different integration periods for the different WQ pressure categories. Therefore, the different integration periods should be interpreted as the period of time over which individual WQ parameters need to be taken into account to observe significant shifts in the functional composition of fish communities. A comprehensive ecological interpretation of RF model
outputs and underlying mechanisms would thus require answering the following questions i) "which combinations of fish metrics respond to which pressures?" and ii) "which ones are the most (or least) responsive?". Although we have provided a few examples (Fig. 5, Table H), with more than 1500 fish metrics taken into account in this study, this task is challenging and beyond the scope of the present study. Addressing this task along with evaluating functional trait profiles in mono- vs. multi-pressure context would represent a significant addition to the research in the field of ecological risk assessment.

## Impairment probabilities across spatial and temporal scales

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

The temporal patterns in river conditions indicate that risks of river impairments, by at least two WQ variables, have significantly decreased between 2005 and 2015 (e.g., for nitrogen (except nitrates) and phosphorous compounds; Fig. 3D and F). These results are in line with
recent reductions in nutrient pollutions within freshwater ecosystems after many EU environmental policies to prevent nutrient losses (EEA, 2018; Floury et al., 2013; Latli et al., 2017). Similar decreasing trends were observed for the overall risk of river degradation (Fig. 3E). At the site scale, the diagnosis-based approach was also able to detect a decrease in impairment risks due to two WQ pressures ('Nitrates' and 'Nitrogen compounds') over a tenyear period (Fig. 4B). Overall, a powerful advantage of this approach is that various combinations of spatial (from site to catchment area) and temporal (from years to decades) scales can be investigated in order to optimize the selection of the most appropriate restoration measures, considering the pressure categories providing the highest impairment probabilities.

## Effects of invasive alien species

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

Our results have indicated, however, that impairment probabilities due to invasive alien species globally decreased significantly over time in the sites that were sampled in both 2005
and 2015 (i.e., over a decade; Fig. 3H). Assuming that the risk of impairment by invasive alien species reflects the vulnerability of the native assemblage, and then the state of the surrounding ecosystem, we would expect to have higher probabilities of impairment by invasive alien species in disturbed sites compared to less disturbed sites (Gallardo et al., 2016). We have tested this hypothesis using two distinct, but complementary ways. First, we have checked the most important fish metrics contributing to the model predictions and found that the "proportion of omnivorous fish species in the native assemblage" was one of these metrics, and was highly (rho $=0.85$ ) and positively correlated to the probability of impairment by IAS (Table H). This first result is supporting our hypothesis since omnivorous and generalist species are usually found in disturbed aquatic systems with high turbidity, organic matter and nutrient load (Gallardo et al., 2016). Second, we have evaluated the correlation between the mean impairment probabilities over the 19 abiotic pressure categories-a proxy of the overall ecosystem state-and the impairment probabilities resulting from the IAS RF model. Interestingly, we have found a positive and significant correlation (Fig. C), suggesting that the significant decrease in the impairment risk of native assemblages by invasive alien species was most likely related to the significant improvement in the ecosystem state over the studied decade (i.e., significant decrease in the mean total impairment probability; Fig. 3G).

Although our diagnosis-based approach was efficient at detecting the impairments induced by the invasive alien species and provided encouraging results, we acknowledge that an exhaustive evaluation of their effects on native communities is a more challenging task than simply considering their presence/absence (Jeschke et al., 2014; Schlaepfer, 2018). First, establishing a list of IAS was not a trivial task since this status could change across locations (local scale vs. regional scale), expert knowledge and institutions. This is the reason why we have chosen to combine the information from three main data sources (Table A). Second,
along with the presence/absence of invasive alien species, we could integrate their taxonomic richness and composition (i.e, species identities and relative abundances) as well as key functional metrics (e.g., the proportion of invasive alien predators, the maximum weight or fecundity of invasive alien species) within a multi-parametric "invasive alien species" pressure category. In this sense, our data indicate that, when present in the assemblage (i.e., $41 \%$ of the sampling events using abundance data), the IAS represented on average $6.1 \pm 0.2$ \% of the total fish abundances within the assemblages. Third, we have only considered the impairment of native assemblages by invasive alien fish species whereas recent meta-analyses have demonstrated that the expansion of invasive alien macrophytes (Gallardo et al., 2016) or benthic macroinvertebrates (Latli et al., 2017; Otjacques et al., 2016) could trigger disproportionate adverse effects on native fish communities. Integrating those suggestions in future studies would most likely help to improve our understanding of the impacts of invasive alien species on native fish communities.

## Caveats

We admit that the definition of multi-parametric "pressure categories" has required the aggregation of miscellaneous pressure parameters (from 1 to more than 70; described at various spatial and temporal scales). Each parameter may influence the fish assemblages and the fish-based metrics in different ways. The use of multi-parametric pressure categories was, however, a robust way to reduce models' complexity and to deal with heterogeneous environmental data (e.g., parameters with missing data). Indeed, the performance of this predictive approach relies on the accuracy and availability of data used to characterize anthropogenic pressures. For instance, the 'Acidification' model was dropped since the associated pressure gradient could not be properly described (i.e., lack of significantly impacted sampling events). In addition, the amount of data available for describing the
intensity of WQ pressure categories was more variable among sampling events compared to HD ones (i.e., more missing data in WQ than in HD pressure categories; Tables C and D ). Yet, the performances of WQ and HD models were not significantly different (i.e., no significant difference in $\mathrm{AUC}_{\text {test }}$ between the ten HD and nine WQ models exhibiting at least 'good' performances; see 'Results' section), thus demonstrating the robustness of this diagnosis-based approach. We therefore advocate that future studies on risk assessment should seek to establish standardized definitions of pressure categories based on (i) data availability and accuracy, and (ii) a broad range of individual pressure parameters, including, but not limited to, climatic factors (e.g. modifications in temperature and precipitation patterns), and overharvesting (Schinegger et al., 2013).

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

## Implications of the diagnosis-based approach

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

## Conclusion

The current application of the diagnosis-based approach does not aim at replacing existing fish-based indices. The major goal of fish-based indices is to provide an efficient evaluation of the global impact of anthropogenic pressures impairing the monitored water bodies. This approach is rather complementary to fish-based indices for more precisely identifying the nature of the anthropogenic pressures significantly impairing each of these water bodies (here,
river reaches) and for helping managers to select the best strategy for restoring water and habitat quality in these water bodies (e.g. reaching the 'good' ecological status). Out of the 28 abiotic and biotic pressure categories tested, the random forest models have efficiently detected the impairments due to nine chemical, ten hydromorphological and one biological pressures. The best performing models also indicated that fish communities were impacted, on average, by seven abiotic pressure categories but the overall impairment probabilities of the whole fish assemblages (including native and invasive alien species) decreased significantly between 2005 and 2015. This diagnosis-based approach is robust and flexible enough to assess specific risks of river degradation under multi-pressure scenarios using distinct biological quality elements (i.e., macroinvertebrates, diatoms, and now fishes). It is also capable to handle complex pressure-response relationships (linear and non-linear relationships), able to diagnose various water bodies (from headwaters to lowland rivers), and to simultaneously consider a variety of pressure categories (hydrological, chemical and biological ones). In fine, merging the information provided by distinct biological quality elements (e.g., macroinvertebrates, diatoms, fishes, macrophytes) should pave the way for a holistic and efficient understanding of the structure and functions of stream ecosystems, that will help water managers worldwide in the decision-making process in relation to river restoration.

## SUPPORTING INFORMATION

Table A: List of fish species
Table B: List of fish characteristics
Table C: Description of water quality pressure categories
Table D: Description of hydromorphological pressure categories
Table E: ecodiag-package

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

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

Table H: List of the most contributive biological metrics to RF models
Figure A: Distribution of impairment probabilities across pressure levels for RF models exhibiting at least 'good' performances

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

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

## AUTHORS' CONTRIBUTION

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

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## DATA ACCESSIBILITY

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

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Table and figure captions

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

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

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

2005-2015 period. Lines and shaded areas on maps represent the boundaries of drainage basins $(\mathrm{B} 1=$ Adour-Garonne; B2 $=$ Loire; B3 $=$ Seine; $\mathrm{B} 4=$ Escaut-Somme; B5 $=$ Meuse; B6 $=$ Rhine; $\mathrm{B} 7=$ Rhone; $\mathrm{B} 8=$ Corse $)$ and the locations of mountain ranges $(\mathrm{M} 1=$ Pyrenees; M 2 $=$ Massif Central; M3 = Alps; M4 = Jura Mountains; M5 = Vosges), respectively. Panels A-D are on a different color scale (presented in panel A) compared to panel E. The locations of Site A and B are indicated (cf. Fig. 4).

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

Figure 4: Radar charts based on site-specific and inter-annual changes in impairment probabilities (IPs) for the three hydromorphological (HD) and the three chemical (WQ) pressure categories corresponding to the RF models with the highest $\mathrm{AUC}_{\text {test }}(>0.80) . \mathrm{A} /$ example of a site (the Loup river, at Tourrette-sur-Loup) with stable and low IP (<0.50; i.e. the dashed blue line) for all the sampling events; B/ example of another site (the Gier river, at Givors) with a partial ecological recovery for three pressures (Nitrogen compounds, Nitrates, and Phosphorous compounds), whereas IPs remained stable and high (i.e. IP $>0.50$ ) for the
three other pressure categories over the same period. "Long. continuity" = Longitudinal continuity; "N. comp." = Nitrogen compounds (except nitrates); "P. comp." = Phosphorous compounds. See Fig. 2F for the site locations in France.

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


Figure 1


Figure 2


Figure 3

A/ Site A
Long.
continuity
Stream
width

## B/ Site B



Figure 4


Figure 5

Table 1

| Code | Pressure category | Year | AUC $_{\text {train }}$ | AUC $_{\text {test }}$ |
| :--- | :--- | :--- | :--- | :--- |
| Chemical: | Nitrates |  |  |  |
| WQ03 | Nitrogen compounds (except nitrates) | 4 | 0.914 | 0.878 |
| WQ02 | 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 |
| Hydromorphological: |  |  |  |  |
| 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 | - | 0.851 | 0.748 |
| HD13 | Lateral continuity | - | 0.852 | 0.703 |
| HD14 | Proximal continuity | - | 0.852 | 0.696 |
| HD11 | Stream depth | - | 0.857 | 0.694 |
| HD02 | Riverine vegetation | - | 0.820 | 0.613 |
| HD03 | Urbanization | 0.805 | 0.597 |  |
| HD06 | Straightening |  | 0.897 | 0.893 |
| Biological: | Invasive alien species |  |  |  |

2


